

# **NUTRIENTS and MICROBES**

**PROFESSIONAL DEVELOPMENT COURSE**  
**2.4 CEUs, 24 PDHs, 24 Training Hours or Contact Hours**



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### **Contributing Editors**

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Eric Pearce S.M.E., chemistry and biological review.

Pete Greer S.M.E., retired biology instructor.

Jack White, Environmental, Health, Safety expert, City of Phoenix. Art Credits.

## **Important Information about this Manual**

This manual has been prepared to educate employees in the general awareness of dealing with complex wastewater treatment procedures and regulatory requirements for safely handling hazardous and toxic materials. The scope of the problem is quite large, requiring a major effort to bring it under control. Employee health and safety, as well as that of the public, depend upon careful application of safe treatment procedures. The manner in which we deal with such hazards will affect the earth and its inhabitants for many generations to come.

This manual will cover general laws, regulations, required procedures and generally accepted policies relating to wastewater treatment and wastewater sampling. It should be noted, however, that the regulation of wastewater treatment, sampling and other hazardous materials is an ongoing process and subject to change over time. For this reason, a list of resources is provided to assist in obtaining the most up-to-date information on various subjects.

This manual is not a guidance document for employees who are involved with pollution control or wastewater treatment. It is not designed to meet the requirements of the United States Environmental Protection Agency (EPA) or Department of Labor-Occupational Safety and Health Administration (OSHA) or state environmental or health departments.

This course manual will provide general educational awareness guidance of activated sludge. This document is not a detailed wastewater treatment textbook or a comprehensive source book on occupational safety and health.

Technical Learning College or Technical Learning Consultants, Inc. makes no warranty, guarantee or representation as to the absolute correctness or appropriateness of the information in this manual and assumes no responsibility in connection with the implementation of this information. It cannot be assumed that this manual contains all measures and concepts required for specific conditions or circumstances. This document should be used for educational guidance and is not considered a legal document.

Individuals who are responsible for the treatment of wastewater, wastewater sampling or the health and safety of workers at wastewater sites should obtain and comply with the most recent federal, state, and local regulations relevant to these sites and are urged to consult with OSHA, EPA and other appropriate federal, state, health and local agencies.

## **Technical Learning College's Scope and Function**

Technical Learning College (**TLC**) offers affordable continuing education for today's working professionals who need to maintain licenses or certifications. TLC holds approximately 80 different governmental approvals for granting of continuing education credit.

TLC's delivery method of continuing education can include traditional types of classroom lectures and distance-based courses or independent study. Most of TLC's distance based or independent study courses are offered in a print-based format and you are welcome to examine this material on your computer with no obligation. Our courses are designed to be flexible and for you to finish the material at your leisure. Students can also receive course materials through the mail. The CEU course or e-manual will contain all your lessons, activities and assignments. Most CEU courses allow students to submit lessons using e-mail or fax, however some courses require students to submit lessons by postal mail (See the course description for more information). Students have direct contact with their instructor—primarily by e-mail. TLC's CEU courses may use such technologies as the World Wide Web, e-mail, CD-ROMs, videotapes and hard copies (See the course description). Make sure you have access to the necessary equipment before enrolling, i.e., printer, Microsoft Word and/or Adobe Acrobat Reader. Some courses may require proctored exams, depending upon your state requirements.

### **Flexible Learning**

At TLC, there are no scheduled online sessions you need contend with, nor are you required to participate in, learning teams or groups designed for the "typical" younger campus-based student. You will work at your own pace, completing assignments in time frames that work best for you. TLC's method of flexible individualized instruction is designed to provide each student the guidance and support needed for successful course completion.

We will beat any other training competitor's price for the same CEU material or classroom training. Student satisfaction is guaranteed.

### **Course Structure**

TLC's online courses combine the best of online delivery and traditional university textbooks. Online you will find the course syllabus, course content, assignments, and online open book exams. This student-friendly course design allows you the most flexibility in choosing when and where you will study.

### **Classroom of One**

TLC Online offers you the best of both worlds. You learn on your own terms and your own time, but you are never on your own. Once you enrolled, you will be assigned a personal student service representative who works with you on an individualized basis throughout your program of study. Course-specific faculty members are assigned at the beginning of each course, providing the academic support you need to successfully complete each course.

### **Satisfaction Guaranteed**

Our iron-clad, risk-free Guarantee ensures you will be another satisfied TLC student.

We have many years of experience, dealing with thousands of students. We assure you, our customer satisfaction is second to none. This is one reason we have taught more than 10,000 students.

Our administrative staff is trained to provide the best customer service in town. Part of that training is knowing how to solve most problems on the spot with an exchange or refund.

**TLC Continuing Education Course Material Development**

TLC's continuing education course material development was based upon several factors; extensive academic research, advice from subject matter experts, data analysis, task analysis and training needs assessment process information gathered from other states.

Please fax or e-mail the answer key to TLC  
Western Campus Fax (928) 272-0747.

**Rush Grading Service**

If you need this assignment graded and the results mailed to you within a 48-hour period, prepare to pay an additional rush service handling fee of \$50.00. This fee may not cover postage costs. If you need this service, simply write RUSH on the top of your Registration Form. We will place you in the front of the grading and processing line.

For security purposes, please fax or e-mail a copy of your driver's license and always call us to confirm we've received your assignment and to confirm your identity.

Thank you...

## **CEU COURSE DESCRIPTION**

### **NUTRIENTS AND MICROBES CEU TRAINING COURSE**

This CEU course is a review of nitrogen and phosphorus control technologies and techniques currently applied and emerging at municipal wastewater treatment plants (WWTP). This course is general in nature and not state specific but will contain different wastewater treatment methods, policies and ideas. You will not need any other materials for this course.

This CEU course is intended for Wastewater Treatment, Collections and Pretreatment/ Industrial Waste Inspectors. The target audience for this course is the person interested in working in a wastewater treatment or collections facility and wishing to maintain CEUs for a certification license or to learn how to do the job safely and effectively, and/or to meet education needs for promotion.

**This course will cover some of the key challenges faced by wastewater treatment professionals today including:**

- \* Many of the wastewater treatment and collection facilities are now old and worn, and require further improvement, repair or replacement to maintain their useful life;
- \* The character and quantity of contaminants presenting problems today are far more complex than those that presented challenges in the past;
- \* Population growth is taxing many existing wastewater treatment systems and creating a need for new plants and new technologies;
- \* Farm runoff (CAFO) and increasing urbanization provide additional sources of pollution not controlled by wastewater treatment; and
- \* One third of new development is served by decentralized systems (e.g., septic systems) as population migrates further from metropolitan areas.

#### **CEU Course Learning Objectives and Timed Outcomes**

1. Examine and understand various WWT Treatment processes and results. 110 minutes.
2. Understanding the basic system components of a wastewater treatment facility. 135 minutes
3. Understand various Effects of WWT Pollutants. 35 minutes.
4. Understand various Effects of WWT Nutrients and Nutrient Removal processes. 45 minutes.
5. Examine and describe various describe the results and various types of Nitrogen. 70 minutes.
6. Understand and describe various forms and problems relating to Phosphorus. 60 minutes.
7. Overview and understanding of different activated sludge processes. 175 minutes
8. A detailed understanding of the Preliminary Treatment operation. 115 minutes
9. A detailed understanding of the operations and components of clarifiers including Secondary clarification. 115 minutes.
10. Examine and diagnose various problems found in the clarifiers and possible corrective measures. 130 minutes.
11. Examine and describe various the operations and components of processing solids. 115 minutes.
12. Examine and describe Wastewater Treatment Water Quality Criteria including microorganisms (bugs) used along with the terminology and formulas to determine their performance. 120 minutes
13. Examine and describe Wastewater Treatment Water Quality Criteria including Microlife, Microorganisms in Lagoons, Nitrification, Algae Groups and formulas to determine their performance. 135 minutes
14. Examine and describe various EPA Wastewater Rules and Regulations. 25 minutes.
15. Examine and describe various Wastewater Analyses and other Laboratory Procedures. 125 minutes.

**This course will cover various wastewater treatment methods including:**

- \* Best conventional pollutant control technology (BCT) for conventional pollutants and applicable to existing dischargers.
- \* Best practicable control technology currently available (BPT) for conventional, toxic and non-conventional pollutants and applicable to existing dischargers.
- \* Best available technology economically achievable (BAT) for toxic and non-conventional pollutants and applicable to existing dischargers.
- \* New source performance standards (NSPS) for conventional pollutants and applicable to new sources.

**Prerequisites:** None

**Final Examination for Credit**

Opportunity to pass the final comprehensive examination is limited to three attempts per course enrollment.

**Flexible Learning**

At TLC, there are no scheduled online sessions you need contend with, nor are you required to participate in learning teams or groups designed for the typical younger campus-based student. You will work at your own pace, completing assignments in time frames that work best for you. TLC's method of flexible individualized instruction is designed to provide each student the guidance and support needed for successful course completion.

We will beat any other training competitor's price for the same CEU material or classroom training. Student satisfaction is guaranteed.

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**Classroom of One**

TLC offers you the best of both worlds. You learn on your own terms and your own time, but you are never on your own. Once enrolled, you will be assigned a personal student service representative who works with you on an individualized basis throughout your program of study. Course specific faculty members are assigned at the beginning of each course providing the academic support you need to successfully complete each course.

**Written Assignment Instructions**

The Nutrients and Microbes CEU training course uses a multiple choice style answer key. You can write your answers in this manual or type out your own answer key. TLC would prefer that you type out and fax or e-mail the final assignment to TLC, but it is not required.

**Course Procedures for Registration and Support**

All of TLC's correspondence courses have complete registration and support services offered. Delivery of services will include, e-mail, web site, telephone, fax and mail support. TLC will attempt immediate and prompt service.

When a student registers for a correspondence course, he or she is assigned a start date and an end date. It is the student's responsibility to note dates for assignments and keep up with the course work. If a student falls behind, he or she must contact TLC and request an end date extension in order to complete the course. It is the prerogative of TLC to decide whether to grant the request. All students will be tracked by their social security number or a unique number assigned to the student.

**Feedback Mechanism (examination procedures)**

Each student will receive a feedback form as part of study packet. You will be able to find this form in the rear of the course or lesson.

**Security and Integrity**

All students are required to do their own work. All lesson sheets and final exams are not returned to the student to discourage sharing of answers. Any fraud or deceit and the student will result in forfeiture of all fees and the appropriate agency will be notified.

**Grading Criteria**

TLC will offer the student either pass/fail or a standard letter grading assignment. If TLC is not notified, you will only receive a pass/fail notice. For security purposes, please fax or e-mail a copy of your driver's license and always call us to confirm we've received your assignment and to confirm your identity. TLC offers students the option of either pass/fail or assignment of a standard letter grade. If a standard letter grade is not requested, a pass/fail notice will be issued.

Final course grades are based on the total number of possible points. The grading scale is administered equally to all students in the course. Do not expect to receive a grade higher than that merited by your total points. No point adjustments will be made for class participation or other subjective factors. If TLC is not notified, you will only receive a pass/fail notice. In order to pass your final assignment, you are required to obtain a minimum score of 70% on your assignment.

**Required Texts**

The Nutrients and Microbes CEU training course will not require any other materials. This course comes complete.

**Environmental Terms, Abbreviations, and Acronyms**

TLC provides a glossary that defines, in non-technical language, commonly used environmental terms appearing in publications and materials. It also explains abbreviations and acronyms used throughout the EPA and other agencies. You can find the glossary in the rear of the manual.

**Recordkeeping and Reporting Practices**

TLC will keep all student records for a minimum of seven years. It is your responsibility to give the completion certificate to the appropriate agencies. TLC will mail a copy to Indiana, Texas, or any other State that requires a copy from the Training Provider.

**ADA Compliance**

TLC will make reasonable accommodations for persons with documented disabilities. Students should notify TLC and their instructors of any special needs. Course content may vary from this outline to meet the needs of this particular group. Alternative assignment is available.

Note to students: Final course grades are based on the total number of possible points. The grading scale is administered equally to all students in the course. Do not expect to receive a grade higher than that merited by your total points. No point adjustments will be made for class participation or other subjective factors.

**Credit/no credit option (P/Z) - None Available**

Note to students: Keep a copy of everything you submit. If your work is lost you can submit your copy for grading. If you do not receive your certificate of completion or quiz results within two or three weeks after submitting it, please contact your instructor.

We expect every student to produce his or her original, independent work. Any student whose work indicates a violation of the Academic Misconduct Policy (cheating, plagiarism) can expect penalties as specified in the Student Handbook, which is available through Student Services; contact them at (928) 468-0665.

A student who registers for a Distance Learning course is assigned a "start date" and an "end date." It is the student's responsibility to note due dates for assignments and to keep up with the course work. If a student falls behind, she or he must contact the instructor and request an extension of her or his end date in order to complete the course. It is the prerogative of the instructor to decide whether or not to grant the request. You will have 90 days from receipt of this manual to complete it in order to receive your Continuing Education Units (CEUs) or Professional Development Hours (PDHs). A score of 70% or better is necessary to pass this course. If you should need any assistance, please email all concerns and the final test to [info@tlch2o.com](mailto:info@tlch2o.com).

**Course Objective:** To provide 24 hours of continuing education training in effective and efficient wastewater treatment methods pertaining to wastewater nutrients and microbe identification and control, and generally accepted wastewater treatment methods.

**Educational Mission**

**The educational mission of TLC is:**

To provide TLC students with comprehensive and ongoing training in the theory and skills needed for the environmental education field,

To provide TLC students opportunities to apply and understand the theory and skills needed for operator certification,

To provide opportunities for TLC students to learn and practice environmental educational skills with members of the community for the purpose of sharing diverse perspectives and experience,

To provide a forum in which students can exchange experiences and ideas related to environmental education,

To provide a forum for the collection and dissemination of current information related to environmental education, and to maintain an environment that nurtures academic and personal growth.

**When the Student finishes this course...**

**At the conclusion of this course:**

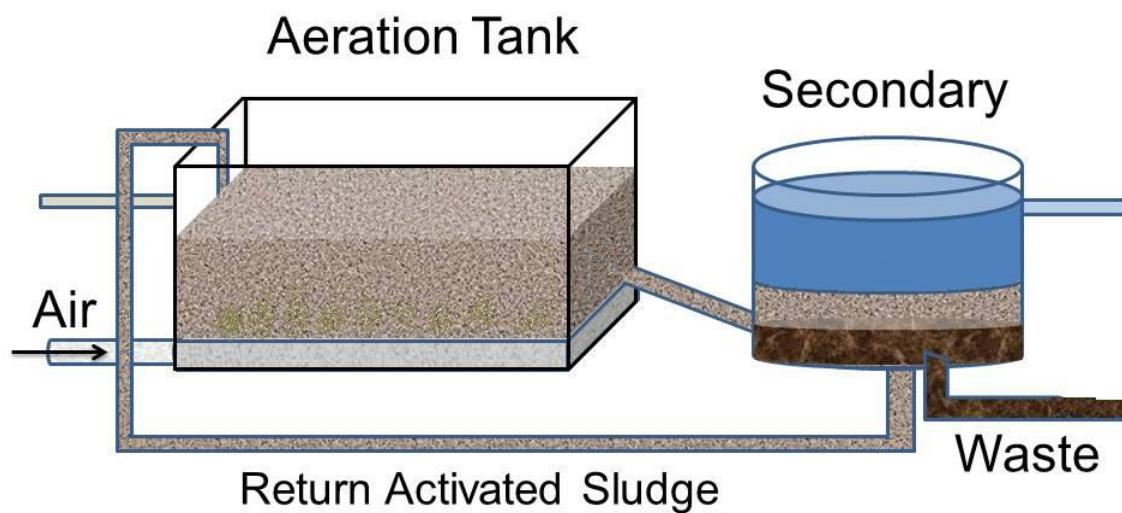
At the finish of this course, you (the student) should be able to explain and describe various treatments and methods of dealing with Phosphorus and Nitrogen including activated sludge methods and explain wastewater treatment sampling techniques and biological monitoring, microbe (bug) identification and microorganism control methods.

Upon completion of this course, the student will obtain 24 hours of continuing education relating to wastewater treatment and nitrogen and phosphorus control technologies and techniques currently applied and emerging at municipal wastewater treatment plants (WWTP).

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## Common Wastewater Acronyms and Terms

### Acronyms and Abbreviations

**A/E Contract:** Architectural and Engineering Contracts  
**A/O:** Pho-redox  
**AMSA:** Association of Metropolitan Sewerage Agencies  
**AOB:** Ammonia Oxidizing Bacteria  
**ASM:** Activated Sludge Model  
**AT3:** Aeration Tank 3  
**BABE:** Bio-Augmentation Batch Enhanced  
**BAF:** Biological Aerated Filter  
**BAR:** Bio-Augmentation Regeneration/Reaeration  
**BCFS:** Biological Chemical Phosphorus and Nitrogen Removal  
**bDON:** Biodegradable Fraction of Dissolved Organic Nitrogen  
**BHRC:** Ballasted High Rate Clarification Processes  
**BNR:** Biological Nutrient Removal  
**BOD:** Biochemical Oxygen Demand  
**BOD5:** Biochemical Oxygen Demand (5-day)  
**BPR:** Biological Phosphorus Removal  
**COD:** Chemical Oxygen Demand  
**CSO:** Combined Sewer Overflow  
**CWA:** Clean Water Act  
**CWSRF:** Clean Water State Revolving Fund  
**D&D:** Drying and Dewatering Facility  
**DAF:** Dissolved Air Flotation  
**DNR:** Department of Natural Resources  
**DO:** Dissolved Oxygen  
**DON:** Dissolved Organic Nitrogen  
**E1:** Estrone  
**E2:17  $\beta$ -estradiol**  
**EBPR:** Enhanced Biological Phosphorus Removal  
**EDC :** Endocrine Disrupting Chemicals  
**EDTA:** Ethylene Diamine Tetraacetic Acid  
**EE2:** 17 $\alpha$ -ethynylestradiol  
**EPA:** U.S. Environmental Protection Agency  
**EPA or USEPA:** United States Environmental Protection Agency  
**FFS:** Fixed-film Systems  
**FWPCA :** Federal Water Pollution Control Act  
**FWS:** Free Water Surface  
**GAO:** Glycogen Accumulating Organism  
**GIS:** Geographic Information System  
**HHWP:** Household Hazardous Waste Collection Program  
**HRSD:** Hampton Roads Sanitation District  
**HRT:** Hydraulic Retention Time  
**I&C:** Instrumentation and Control System  
**I/I:** Infiltration and Inflow  
**iDON:** Inert Dissolved Organic Nitrogen  
**ISF:** Intermittent Sand Filter  
**ISS:** Inline Storage System  
**IWA:** International Water Association

**IWPP:** Industrial Waste Pretreatment Program  
**JHB:** Johannesburg  
**LIMS:** Laboratory Information Management Systems  
**MAUREEN:** Mainstream Autotrophic Recycle Enhanced N-removal  
**MBBR:** Moving-Bed Biofilm Reactor  
**MBDT:** Minority Business Development and Training  
**MBE:** Minority Business Enterprise  
**MBR:** Membrane Bioreactor  
**MGD:** Million Gallons per Day  
**MLE:** Modified Ludzak Ettinger  
**MUCT :** Modified University of Capetown  
**N:** Nitrogen  
**NOAA :** National Oceanic and Atmospheric Administration  
**NOB :** Nitrite Oxidizing Bacteria  
**NPDES :** National Pollutant Discharge Elimination System  
**NTT:** Nitrogen Trading Tool  
**ORD:** EPA Office of Research and Development  
**ORP:** Oxidation Reduction Potential  
**OWASA :** Orange Water and Sewer Authority  
**OWM :** EPA Office of Wastewater Management  
**P:** Phosphorus  
**P2:** Pollution Prevention Initiative  
**PAH:** Polycyclic Aromatic Hydrocarbons  
**PAO:** Phosphate Accumulating Organism  
**PHA:** Polyhydroxyalkanoates  
**PHB:** Poly-B-hydroxy-butyrate  
**PHV:** Poly-hydroxy valerate  
**POTW :** Publicly Owned Treatment Works  
**PPCPs :** Pharmaceuticals and Personal Care Products  
**QA/QC:** Quality Assurance and Quality Control  
**RAS:** Return Activated Sludge  
**RBC:** Rotating Biological Contactor  
**rbCOD:** Readily Biodegradable Chemical Oxygen Demand  
**rDON:** Recalcitrant Dissolved Organic Nitrogen  
**RO:** Reverse Osmosis  
**RSF:** Recirculating Sand Filters  
**S/W/MBE:** Small, Women's, Minority Business Enterprise  
**SAV:** Submerged Aquatic Vegetation  
**SBR :** Sequencing Batch Reactors  
**SHARON :** Single Reactor High-activity Ammonia Removal Over Nitrite  
**SND :** Simultaneous Nitrification-Denitrification  
**SRT :** Solids Retention Time  
**SSES:** Sewer System Evaluation Survey  
**SSO:** Sanitary Sewer Overflow  
**STAC:** Chesapeake Bay Program Scientific and Technical Advisory Committee  
**SWIS:** Subsurface Wastewater Infiltration System  
**TAT:** Technical Advisory Team  
**TDS:** Total Dissolved Solids  
**TKN:** Total Kjeldahl Nitrogen  
**TMDL :** Total Maximum Daily Loads  
**TN:** Total Nitrogen

**TP**: Total Phosphorus

**TSS** : Total Suspended Solids

**TUDP**: Bio-P Model of the Delft University of Technology

**UCT**: University of Capetown

**USDA**: U.S. Department of Agriculture

**USGS**: U.S. Geological Survey

**VFA**: Volatile Fatty Acids

**VIP**: Virginia Initiative Plant

**VSS**: Volatile Suspended Solids

**WAS**: Waste Activated Sludge

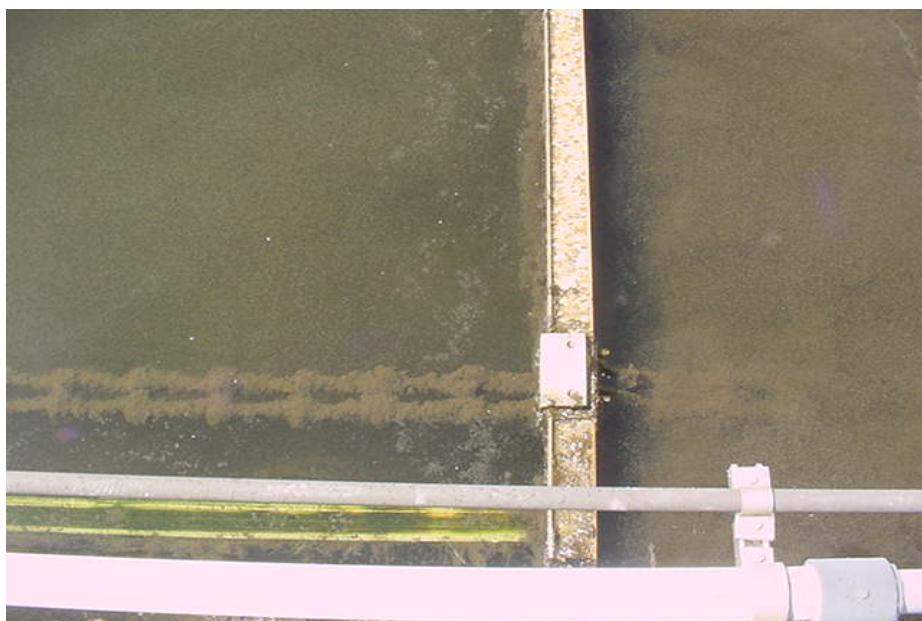
**WEF**: Water Environment Federation

**WERF**: Water Environment Research Foundation

**WPAP**: Water Pollution Abatement Program

**WQS**: Water Quality Standard

**WWTP**: Wastewater Treatment Plant



Rectangular clarifier mechanism, flights, and chains.



Here is one of TLC's professors Marcos Aparecido Silva Bueno showing microscopic views of commonly found MO's in a classroom setting. Professor Marcos Aparecido Silva Bueno is a world renowned microbiological expert.



## Key Words

This glossary includes a collection of terms used in this course and an explanation of each term.

### **Act or “the Act” [40 CFR §403.3(b)]**

The Federal Water Pollution Control Act, also known as the Clean Water Act, as amended, 33 USC 1251 et.seq.

### **Approval Authority [40 CFR §403.3(c)]**

The Director in an NPDES State with an approved State Pretreatment Program and the appropriate EPA Regional Administrator in a non-NPDES State or State without an approved pretreatment program.

### **Approved POTW Pretreatment Program or Program [40 CFR §403.3(d)]**

A program administered by a POTW that meets the criteria established in 40 CFR Part 403 and which has been approved by a Regional Administrator or State Director.

### **Approved State Pretreatment Program**

A program administered by a State that meets the criteria established in 40 CFR §403.10 and which has been approved by a Regional Administrator

### **Approved/Authorized State**

A State with an NPDES permit program approved pursuant to section 402(b) of the Act and an approved State Pretreatment Program.

### **Baseline Monitoring Report (BMR) [paraphrased from 40 CFR §403.12(b)]**

A report submitted by categorical industrial users (CIUs) within 180 days after the effective date of an applicable categorical standard, or at least 90 days prior to commencement of discharge for new sources, which contains specific facility information, including flow and pollutant concentration data. For existing sources, the report must also certify as to the compliance status of the facility with respect to the categorical standards.

### **Best Available Technology Economically Achievable (BAT)**

A level of technology based on the best existing control and treatment measures that are economically achievable within the given industrial category or subcategory.

### **Best Management Practices (BMPs)**

Schedules of activities, prohibitions of practices, maintenance procedures, and other management practices to prevent or reduce the pollution of waters of the U.S. BMPs also include treatment requirements, operating procedures and practices to control plant site runoff, spillage or leaks, sludge or waste disposal, or drainage from raw material storage.

### **Best Practicable Control Technology Currently Available (BPT)**

A level of technology represented by the average of the best existing wastewater treatment performance levels within an industrial category or subcategory.

### **Best Professional Judgment (BPJ)**

The method used by a permit writer to develop technology-based limitations on a case-by-case basis using all reasonably available and relevant data.

### **Blowdown**

The discharge of water with high concentrations of accumulated solids from boilers to prevent plugging of the boiler tubes and/or steam lines. In cooling towers, blowdown is discharged to reduce the concentration of dissolved salts in the recirculating cooling water.

### **Bypass [40 CFR §403.17(a)]**

The intentional diversion of waste streams from any portion of an Industrial User's treatment facility.

### **Categorical Industrial User (CIU)**

An industrial user subject to National categorical pretreatment standards.

### **Categorical Pretreatment Standards [40 CFR § 403.6 and 40 CFR Parts 405-471]**

Limitations on pollutant discharges to POTWs promulgated by the EPA in accordance with Section 307 of the Clean Water Act, that apply to specific process wastewater discharges of particular industrial categories.

**Chain of Custody (COC)**

A record of each person involved in the possession of a sample from the person who collects the sample to the person who analyzes the sample in the laboratory.

**Chronic**

A stimulus that lingers or continues for a relatively long period of time, often one-tenth of the life span or more. Chronic should be considered a relative term depending on the life span of an organism. The measurement of chronic effect can be reduced growth, reduced reproduction, etc., in addition to lethality.

**Clean Water Act (CWA)**

The common name for the Federal Water Pollution Control Act. Public law 92-500; 33 U.S.C. 1251 et seq.; legislation which provides statutory authority for both NPDES and Pretreatment Programs.

**Code of Federal Regulations (CFR)**

A codification of Federal rules published annually by the Office of the Federal Register National Archives and Records Administration. Title 40 of the CFR contains the regulations for *Protection of the Environment*.

**Combined Sewer Overflow (CSO)**

A discharge of untreated wastewater from a combined sewer system at a point prior to the headworks of a publicly owned treatment works. CSOs generally occur during wet weather (rainfall or snowfall). During periods of wet weather, these systems become overloaded, bypass treatment works, and discharge directly to receiving waters.

**Combined Wastestream Formula (CWF) *[paraphrased from 40 CFR §403.6(e)]***

Procedure for calculating alternative discharge limits at industrial facilities where a regulated wastestream from a categorical industrial user is combined with other waste streams prior to treatment.

**Compliance Schedule**

A schedule of remedial measures included in a permit or an enforcement order, including a sequence of interim requirements (for example, actions, operations, or milestone events) that lead to compliance with the CWA and regulations.

**Composite Sample**

Sample composed of two or more discrete samples. The aggregate sample will reflect the average water quality covering the compositing or sample period.

**Concentration-based Limit**

A limit based upon the relative strength of a pollutant in a wastestream, usually expressed in mg/l.

**Continuous Discharge**

A discharge that occurs without interruption during the operating hours of a facility, except for infrequent shutdowns for maintenance, process changes or similar activities.

**Control Authority *[paraphrased from 40 CFR § 403.12(a)]***

A POTW with an approved pretreatment program or the approval authority in the absence of a POTW pretreatment program.

**Conventional Pollutants**

BOD, TSS, fecal coliform, oil and grease, and pH

**Daily Maximum Limitations**

The maximum allowable discharge of pollutants during a 24-hour period. Where daily maximum limitations are expressed in units of mass, the daily discharge is the total mass discharged over the course of the day. Where daily maximum limitations are expressed in terms of a concentration, the daily discharge is the arithmetic average measurement of the pollutant concentration derived from all measurements taken that day.

**Detection Limit**

The minimum concentration of an analyte (substance) that can be measured and reported with a 99% confidence that the analyte concentration is greater than zero as determined by the procedure set forth in 40 CFR Part 136, Appendix B.

**Development Document**

Detailed report of studies conducted by the U.S. EPA for the purpose of establishing effluent guidelines and categorical pretreatment standards.

**Dilute Wastestream** *[paraphrased from 40 CFR §403.6(e)(1)(i)]*

For purposes of the combined wastestream formula, the average daily flow (at least a 30-day average) from : (a) boiler blowdown streams, non-contact cooling streams, storm water streams, and demineralized backwash streams; provided, however, that where such streams contain a significant amount of a pollutant, and the combination of such streams, prior to treatment, with an industrial user's regulated process wastestream(s) will result in a substantial reduction of that pollutant, the Control Authority, upon application of the industrial user, may exercise its discretion to determine whether such stream(s) should be classified as diluted or unregulated. In its application to the Control Authority, the industrial user must provide engineering, production, sampling and analysis, and such other information so the control authority can make its determination; or (b) sanitary wastestreams where such streams are not regulated by a categorical pretreatment standard; or (c) from any process wastestreams which were, or could have been, entirely exempted from categorical pretreatment standards pursuant to paragraph 8 of the NRDC v. Costle Consent Decree (12 ERC 1833) for one or more of the following reasons (see Appendix D of 40 CFR Part 403):  
a. the pollutants of concern are not detectable in the effluent from the industrial user (paragraph(8)(a)(iii));  
b. the pollutants of concern are present only in trace amounts and are neither causing nor likely to cause toxic effects (paragraph (8)(a)(iii));  
c. the pollutants of concern are present in amounts too small to be effectively deduced by technologies known to the Administrator (paragraph (8)(a)(iii)); or  
d. the wastestream contains only pollutants which are compatible with the POTW (paragraph (8)(b)(I)).

**Effluent Limitations Guideline**

Any effluent limitations guidelines issued by the EPA pursuant to Section 304(b) of the CWA. These regulations are published to adopt or revise a national standard prescribing restrictions on quantities, rates, and concentrations of chemical, physical, biological, and other constituents which are discharged from point sources, in specific industrial categories (e.g., metal finishing, metal molding and casting, etc.).

**Enforcement Response Plan** *[paraphrased from 40 CFR §403.8(f)(5)]*

Step-by-step enforcement procedures followed by Control Authority staff to identify, document, and respond to violations.

**Existing Source**

Any source of discharge, the construction or operation of which commenced prior to the publication by the EPA of proposed categorical pretreatment standards, which will be applicable to such source if the standard is thereafter promulgated in accordance with Section 307 of the Act.

**Federal Water Pollution Control Act (FWPCA)**

The title of Public law 92-500; 33 U.S.C. 1251 et seq., also known as the Clean Water Act (CWA), enacted October 18, 1972.

**Flow Weighted Average Formula (FWA)** *[paraphrased from 40 CFR §403.6(e)]*

A procedure used to calculate alternative limits where wastestreams regulated by a categorical pretreatment standard and nonregulated wastestreams combine after treatment but prior to the monitoring point.

**Flow Proportional Composite Sample**

Combination of individual samples proportional to the flow of the wastestream at the time of sampling.

**Fundamentally Different Factors** *[paraphrased from 40 CFR §403.13]*

Case-by-case variance from categorical pretreatment standards based on the factors considered by the EPA in developing the applicable category/subcategory being fundamentally different than factors relating to a specific industrial user.

**General Prohibitions** *[40 CFR §403.5(a)(1)]*

No user shall introduce into a POTW any pollutant(s) which cause pass through or interference.

**Grab Sample**

A sample which is taken from a wastestream on a one-time basis with no regard to the flow of the wastestream and without consideration of time. A single grab sample should be taken over a period of time not to exceed 15 minutes.

**Indirect Discharge or Discharge [40 CFR §403.3(g)]**

The introduction of pollutants into a POTW from any non-domestic source regulated under section 307(b), (c), or (d) of the Act.

**Industrial User (IU) or User [40 CFR §403.3(h)]**

A source of indirect discharge.

**Industrial Waste Survey**

The process of identifying and locating industrial users and characterizing their industrial discharge.

**Inhibition Concentration**

Estimate of the toxicant concentration that would cause a given percent reduction (e.g., IC25) in a nonlethal biological measurement of the test organisms, such as reproduction or growth.

**Interference [paraphrased from 40 CFR §403.3(i)]**

A discharge which, alone or in conjunction with a discharge or discharges from other sources, both: (1) inhibits or disrupts the POTW, its treatment processes or operations, or its sludge processes, use or disposal; and (2) therefore is a cause of a violation of any requirement of the POTW's NPDES permit (including an increase in the magnitude or duration of a violation) or of the prevention of sewage sludge use or disposal in compliance with ... [applicable] statutory provisions and regulations or permits issued there under (or more stringent State or local regulations)

**Local Limits [paraphrased 40 CFR § 403.5(c)]**

Specific discharge limits developed and enforced by POTWs upon industrial or commercial facilities to implement the general and specific discharge prohibitions listed in 40 CFR §§403.5(a)(1) and (b).

**Monthly Average**

The arithmetic average value of all samples taken in a calendar month for an individual pollutant parameter. The monthly average may be the average of all grab samples taken in a given calendar month, or the average of all composite samples taken in a given calendar month.

**National Pollutant Discharge Elimination System (NPDES)**

The national program for issuing, modifying, revoking and reissuing, terminating, monitoring and enforcing discharge permits from point sources to waters of the United States, and imposing and enforcing pretreatment requirements, under sections 307, 402, 318, and 405 of the CWA.

**National Pretreatment Standard or Pretreatment Standard or Standard**

[40 CFR §403.3(j)] Any regulation containing pollutant discharge limits promulgated by the EPA in accordance with section 307(b) and (c) of the Act, which applies to Industrial Users. This term includes prohibitive discharge limits established pursuant to §403.5.

**New Source [40 CFR §403.3(k)]**

Any building, structure, facility or installation from which there is or may be a discharge of pollutants, the construction of which commenced after the publication of proposed Pretreatment Standards under section 307(c) of the Act which will be applicable to such source if such standards are thereafter promulgated in accordance with that section *provided that*:

- (a) The building, structure, facility or installation is constructed at a site at which no other discharge source is located; or
- (b) The building, structure, facility or installation totally replaces the process or production equipment that causes the discharge of pollutants at an existing source; or
- (c) The production or wastewater generating processes of the building, structure, facility, or installation are substantially independent of an existing source at the same site. In determining whether these are substantially independent, factors such as the extent to which the new facility is integrated with the existing plant, and the extent to which the new facility is engaged in the same general type of activity as the existing source, should be considered.

Construction on a site at which an existing source is located results in a modification rather than a new source if the construction does not create a new building, structure, facility, or installation meeting the criteria of paragraphs (k)(1)(ii), or (k)(1)(iii) of this section but otherwise alters, replaces, or adds to existing processor production equipment.

Construction of a new source, as defined under this paragraph has commenced if the owner or operator has:

- (i) Begun, or caused to begin as part of a continuous onsite construction program:

- (A) Any placement, assembly, or installation of facilities or equipment; or
- (B) Significant site preparation work including clearing, excavation, or removal of existing buildings, structures, or facilities which is necessary for the placement, assembly, or installation of new source facilities or equipment, or
- (C) Entered into a binding contractual obligation for the purchase of facilities or equipment which are intended to be used in its operation within a reasonable time. Options to purchase or contracts which can be terminated or modified without substantial loss, and contracts for feasibility, engineering, and design studies do not constitute a contractual obligation under this paragraph.

**90-Day Final Compliance Report [40 CFR §403.12(d)]**

A report submitted by categorical industrial users within 90 days following the date for final compliance with the standards. This report must contain flow measurement (of regulated process streams and other streams), measurement of pollutants, and a certification as to whether the categorical standards are being met.

**Nonconventional Pollutants**

Any pollutant that is neither a toxic pollutant nor a conventional pollutant (e.g., manganese, ammonia, etc.)

**Non-Contact Cooling Water**

Water used for cooling which does not come into direct contact with any raw material, intermediate product, waste product, or finished product. The only pollutant contributed from the discharge is heat.

**Non-Regulated Wastestream**

Unregulated and dilute wastestreams (not regulated by categorical standards).

**Pass Through [40 CFR §403.3(n)]**

A discharge which exits the POTW into waters of the United States in quantities or concentrations which, alone or in conjunction with a discharge or discharges from other sources, is a cause of a violation of any requirement of the POTW's NPDES permit (including an increase in the magnitude or duration of a violation).

**Periodic Compliance Report [paraphrased from 40 CFR §403.12(e) & (h)]**

A report on compliance status submitted by categorical industrial users and significant noncategorical industrial users to the control authority at least semiannually (once every six months).

**Point Source [40 CFR 122.2]**

Any discernible, confined, and discrete conveyance, including but not limited to any pipe, ditch, channel, tunnel, conduit, well, discrete fixture, container, rolling stock concentrated animal feeding operation vessel, or other floating craft from which pollutants are or may be discharged.

**Pollutant [40 CFR 122.2]**

Dredged spoil, solid waste, incinerator residue, filter backwash, sewage, garbage, sewage sludge, munitions, chemical wastes, biological materials, radioactive materials (except those regulated under the Atomic Energy Act of 1954, as amended (42 U.S.C. 2011 et seq.)), heat, wrecked or discarded equipment, rock, sand, cellar dirt, and industrial, municipal and agricultural waste discharged into water.

**Pretreatment [paraphrased from 40 CFR §403.3(q)]**

The reduction of the amount of pollutants, the elimination of pollutants, or the alteration of the nature of pollutant properties in wastewater prior to or in lieu of discharging or otherwise introducing such pollutants into a POTW.

**Pretreatment Requirements [40 CFR §403.3(r)]**

Any substantive or procedural requirement related to Pretreatment, other than a National Pretreatment Standard, imposed on an Industrial User.

**Pretreatment Standards for Existing Sources (PSES)**

Categorical Standards and requirements applicable to industrial sources that began construction prior to the publication of the proposed pretreatment standards for that industrial category. (see individual standards at 40 CFR Parts 405-471.)

**Pretreatment Standards for New Sources (PSNS)**

Categorical Standards and requirements applicable to industrial sources that began construction after the publication of the proposed pretreatment standards for that industrial category. (see individual standards at 40 CFR Parts 405-471.)

**Priority Pollutant**

Pollutant listed by the Administrator of the EPA under Clean Water Act section 307(a). The list of the current 126 Priority Pollutants can be found in 40 CFR Part 423 Appendix A.

**Process Wastewater**

Any water which, during manufacturing or processing, comes into contact with or results from the production or use of any raw material, intermediate product, finished product, byproduct, or waste product.

**Production-Based Standards**

A discharge standard expressed in terms of pollutant mass allowed in a discharge per unit of product manufactured.

**Publicly Owned Treatment Works (POTW) [40 CFR §403.3(o)]**

A treatment works as defined by section 212 of the Act, which is owned by a State or municipality (as defined by section 502(4) of the Act). This definition includes any devices or systems used in the storage, treatment, recycling, and reclamation of municipal sewage or industrial wastes of a liquid nature. It also includes sewers, pipes or other conveyances only if they convey wastewater to a POTW Treatment Plant.

The term also means the municipality as defined in section 502(4) of the Act, which has jurisdiction over the Indirect Discharges to and the discharges from such a treatment works.

**Regulated Wastestream**

For purposes of applying the combined wastestream formula, a wastestream from an industrial process that is regulated by a categorical standard.

**Removal Credit [paraphrased from 40 CFR §403.7]**

Variance from a pollutant limit specified in a categorical pretreatment standard to reflect removal by the POTW of said pollutant.

**Representative Sample**

A sample from a wastestream that is as nearly identical as possible in composition to that in the larger volume of wastewater being discharged and typical of the discharge from the facility on a normal operating day.

**Sanitary Sewer Overflow (SSO)**

Untreated or partially treated sewage overflows from a sanitary sewer collection system.

**Self-Monitoring**

Sampling and analyses performed by a facility to ensure compliance with a permit or other regulatory requirements.

**Sewer Use Ordinance (SUO)**

A legal mechanism implemented by a local government entity which sets out, among others, requirements for the discharge of pollutants into a publicly owned treatment works.

**Significant Industrial User (SIU) [paraphrased from 40 CFR §403.3(t)]**

(1) All users subject to Categorical Pretreatment Standards under 40 CFR 403.6 and 40 CFR chapter I, subchapter N; and (2) Any other industrial user that: discharges an average of 25,000 gallons per day or more of process wastewater to the POTW (excluding sanitary, noncontact cooling and boiler blowdown wastewater); contributes a process wastestream which makes up 5 percent or more of the average dry weather hydraulic or organic capacity of the POTW treatment plant; or is designated as such by the Control Authority as defined in 40 CFR 403.12(a) on the basis that the industrial user has a reasonable potential for adversely affecting the POTW's operation or for violating any pretreatment standard or requirement (in accordance with 40 CFR 403.8(f)(6)].

**Significant Noncompliance (SNC) [40 CFR §403.8(f)(2)(vii)]**

Industrial user violations meeting one or more of the following criteria:

- 1) Chronic violations of wastewater discharge limits, defined here as those in which sixty-six percent or more of all of the measurements taken during a six month period exceed (by any magnitude) the daily maximum limit or the average limit for the same pollutant parameter;
- 2) Technical Review Criteria (TRC) violations, defined here as those in which thirty-three percent or more of all of the measurements for each pollutants parameter taken during a six-month period equal or exceed the product of the daily maximum limit or the average limit multiplied by the applicable TRC (TRC=1.4 for BOD, TSS, fats, oil, and grease, and 1.2 for all other pollutants except pH);
- 3) Any other violation of a pretreatment effluent limit (daily maximum or longer-term average) that the Control Authority determines has caused, alone or in combination with other dischargers, interference or pass through (including endangering the health of POTW personnel or the general public);
- 4) Any discharge of a pollutant that has caused imminent endangerment to human health, welfare or to the environment or has resulted in the POTW's exercise of its emergency authority under paragraph (f)(1)(vi)(B) of this section to halt or prevent such a discharge;
- 5) Failure to meet, within 90 days after the schedule date, a compliance schedule milestone contained in a local control mechanism or enforcement order for starting construction, completing construction, or attaining final compliance;
- 6) Failure to provide, within 30 days after the due date, required reports such as baseline monitoring reports, 90-day compliance reports, periodic self-monitoring reports, and reports on compliance with compliance schedules;
- 7) Failure to accurately report noncompliance;
- 8) Any other violation or group of violations which the Control Authority determines will adversely affect the operation or implementation of the local pretreatment program.

**Slug Discharge [40 CFR §403.8(f)(2)(v)]**

Any discharge of a non-routine, episodic nature, including but not limited to, an accidental spill or a non-customary batch discharge.

**Specific Prohibitions [40 CFR §403.5(b)]**

The following pollutants shall not be introduced into a POTW:

- 1) Pollutants which create a fire or explosion hazard in the POTW, including but not limited to, wastestreams with a closed cup flashpoint of less than 140 degrees Fahrenheit or 60 degrees Centigrade using the test methods specified in 40 CFR Part 261.21;
- 2) Pollutants which will cause corrosive structural damage to the POTW, but in no case discharges with pH lower than 5.0, unless the works is specifically designed to accommodate such discharges;
- 3) Solid or viscous pollutants in amounts which will cause obstruction to the flow in the POTW resulting in interference;
- 4) Any pollutant, including oxygen-demanding pollutants (BOD, etc.) Released in a discharge at a flow rate and/or concentration which will cause interference with the POTW;
- 5) Heat in amounts which will inhibit biological activity in the POTW resulting in interference, but in no case heat in such quantities that the temperature at the POTW treatment plant exceeds 40°C (104°F) unless the Approval Authority, upon request of the POTW, approves alternative temperature limits;
- 6) Petroleum oil, nonbiodegradable cutting oil, or products of mineral oil origin in amounts that will cause interference or pass through;
- 7) Pollutants which result in the presence of toxic gases, vapors, or fumes within the POTW in a quantity that may cause acute worker health and safety problems;
- 8) Any trucked or hauled pollutants, except at discharge points designated by the POTW.

**Standard Industrial Classification (SIC)**

A system developed by the U.S. Office of Management and Budget that is used to classify various types of business entities. Effective in 1998, the SIC scheme is replace by the North American Industry Classification System (**NAICS**), although the EPA has not yet implemented this change.

**Storm Water**

Rain water, snowmelt, and surface runoff and drainage.

**Time Proportional Composite Sample**

A sample consisting of a series of aliquots collected from a representative point in the discharge stream at equal time intervals over the entire discharge period on the sampling day.

**Toxic Pollutant**

Any pollutant listed as toxic under section 307(a)(1) of the CWA, or in the case of sludge use or disposal practices, any pollutant identified in regulations implementing section 405(d) of the CWA.

**Toxicity Reduction Evaluation**

A site-specific study conducted in a stepwise process designed to identify the causative agent(s) of effluent toxicity, isolate the sources of toxicity, evaluate the effectiveness of toxicity control options, and then confirm the reduction in effluent toxicity.

**Toxicity Test**

A procedure to determine the toxicity of a chemical or an effluent using living organisms. A toxicity test measures the degree of effect on exposed test organisms of a specific chemical or effluent.

**Toxicity Identification Evaluation**

Set of procedures to identify the specific chemicals responsible for effluent toxicity.

**Unregulated Wastestream**

For purposes of applying the combined wastestream formula, a wastestream not regulated by a categorical standard nor considered a dilute wastestream.

**Upset [paraphrased from 40 CFR §403.16(a)]**

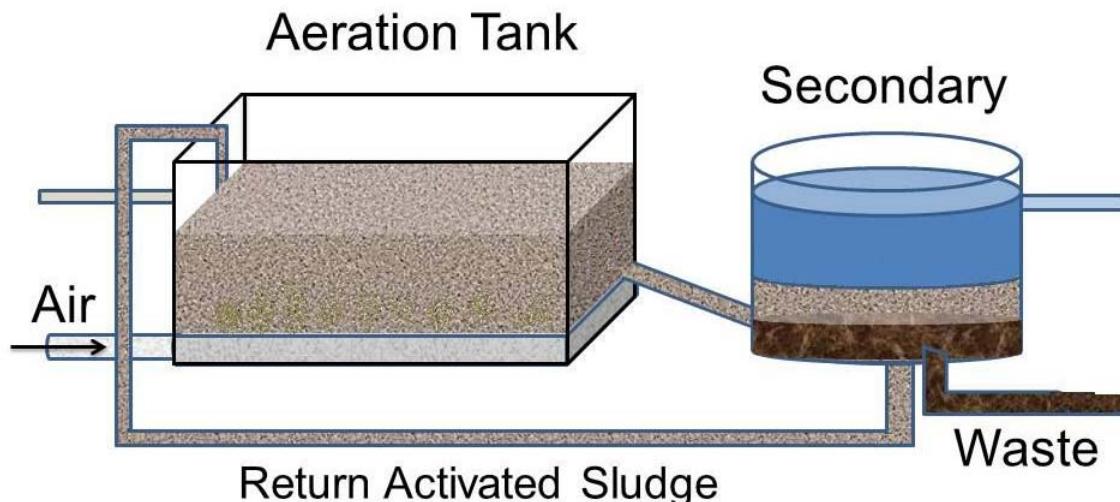
An exceptional incident in which there is unintentional and temporary noncompliance with categorical Pretreatment Standards because of factors beyond the reasonable control of the Industrial User. An Upset does not include noncompliance to the extent caused by operational error, improperly designed treatment facilities, inadequate treatment facilities, lack of preventative maintenance, or careless or improper operation.

**Water Quality Criteria**

Comprised of both numeric and narrative criteria. Numeric criteria are scientifically derived ambient concentrations developed by EPA or States for various pollutants of concern to protect human health and aquatic life. Narrative criteria are statements that describe the desired water quality goal.

**Water Quality Standard**

A statute or regulation that consists of the beneficial designated use or uses of a waterbody, the numeric and narrative water quality criteria that are necessary to protect the use or uses of that particular waterbody, and an antidegradation statement.



# Clean Water Act

## What is Wastewater Treatment?

Wastewater treatment is the process of cleaning used water and sewage so it can be returned safely to our environment. Wastewater treatment is the last line of defense against water pollution. If you envision the water cycle as a whole, you can clean water produced by wastewater treatment is the same water that eventually ends up back in our lakes and rivers, where we get our drinking water.

## Why Are Wastewater Treatment Plants Important?

Wastewater treatment plants are vital to our communities. They protect public health by eliminating disease-causing bacteria from water. By protecting water quality, wastewater treatment plants make it possible for us to safely enjoy the recreational use of clean oceans, lakes, streams and rivers.

## 33 U.S.C. s/s 1251 et seq. (1977)

The Clean Water Act is a 1977 amendment to the Federal Water Pollution Control Act of 1972, which set the basic structure for regulating discharges of pollutants to waters of the United States.

The law gave the EPA the authority to set effluent standards on an industry basis (technology-based) and continued the requirements to set water quality standards for all contaminants in surface waters. The CWA makes it unlawful for any person to discharge any pollutant from a point source into navigable waters unless a permit (**NPDES**) is obtained under the act.

The 1977 amendments focused on toxic pollutants. In 1987, the PCA was reauthorized and again focused on toxic substances, authorized citizen suit provisions, and funded sewage treatment plants (**POTW's**) under the Construction Grants Program. The CWA provides for the delegation by the EPA of many permitting, administrative, and enforcement aspects of the law to state governments. In states with the authority to implement CWA programs, the EPA still retains oversight responsibilities. In 1972, Congress enacted the first comprehensive national clean water legislation in response to growing public concern for serious and widespread water pollution. The Clean Water Act is the primary federal law that protects our nation's waters, including lakes, rivers, aquifers, and coastal areas.



Lake Erie was dying. The Potomac River was clogged with blue-green algae blooms that were a nuisance and a threat to public health. Many of the nation's rivers were little more than open sewers and sewage frequently washed up on shore. Fish kills were a common sight. Wetlands were disappearing at a rapid rate.

Today, the quality of our waters has improved dramatically as a result of a cooperative effort by federal, state, tribal and local governments to implement the pollution control programs established in 1972 by the Clean Water Act. The Clean Water Act's primary objective is to restore and maintain the integrity of the nation's waters. This objective translates into two fundamental national goals:

- eliminate the discharge of pollutants into the nation's waters, and
- achieve water quality levels that are fishable and swimmable.

The Clean Water Act focuses on improving the quality of the nation's waters. It provides a comprehensive framework of standards, technical tools and financial assistance to address the many causes of pollution and poor water quality. This includes municipal and industrial wastewater discharges, polluted runoff from urban and rural areas, and habitat destruction. For example, the Clean Water Act requires major industries to meet performance standards to ensure pollution control; charges states, and tribes with setting specific water quality criteria appropriate for their waters and developing pollution control programs to meet them; provides funding to states and communities to help them meet their clean water infrastructure needs; protects valuable wetlands and other aquatic habitats through a permitting process that ensures development, and other activities are conducted in an environmentally sound manner. After 25 years, the act continues to provide a clear path for clean water and a solid foundation for an effective national water program.

#### **In 1972**

Only a third of the nation's waters were safe for fishing and swimming. Wetlands losses were estimated at about 460,000 acres annually. Agricultural runoff resulted in the erosion of 2.25 billion tons of soil and the deposit of large amounts of phosphorus and nitrogen into many waters. Sewage treatment plants served only 85 million people.

#### **Today**

Two-thirds of the nation's waters are safe for fishing and swimming. The rate of annual wetlands losses is estimated at about 70,000-90,000 acres according to recent studies. The amount of soil lost due to agricultural runoff has been cut by one billion tons annually, and phosphorus and nitrogen levels in water sources are down. Modern wastewater treatment facilities serve 173 million people.

#### **The Future**

All Americans will enjoy clean water that is safe for fishing and swimming. We will achieve a net gain of wetlands by preventing additional losses and restoring hundreds of thousands of acres of wetlands. Soil erosion and runoff of phosphorus and nitrogen into watersheds will be minimized, helping to sustain the nation's farming economy and aquatic systems. The nation's waters will be free of effects of sewage discharges.



Rotifer, an excellent MO or Microorganism or Indicator Organism.

## **Wastewater Treatment WWT Introduction**

During the early days of our nation's history, people living in both the cities and the countryside used cesspools and privies to dispose of domestic wastewater. Cities began to install wastewater collection systems in the late nineteenth century because of an increasing awareness of waterborne disease and the popularity of indoor plumbing and flush toilets.

The use of sewage collection systems brought dramatic improvements to public health, further encouraging the growth of metropolitan areas. In the year 2000 approximately 208 million people in the U.S. were served by centralized collection systems.

### **Wastewater Treatment**

In 1892, only 27 American cities provided wastewater treatment. Today, more than 16,000 publicly-owned wastewater treatment plants operate in the United States and its territories. The constructions of wastewater treatment facilities blossomed in the 1920s and again after the passage of the CWA in 1972 with the availability of grant funding and new requirements calling for minimum levels of treatment. Adequate treatment of wastewater, along with the ability to provide a sufficient supply of clean water, has become a major concern for many communities.

### **What is in Wastewater?**

Wastewater is mostly water by weight. Other materials make up only a small portion of wastewater, but can be present in large enough quantities to endanger public health and the environment. Because practically anything that can be flushed down a toilet, drain, or sewer can be found in wastewater, even household sewage contains many potential pollutants. The wastewater components that should be of most concern to homeowners and communities are those that have the potential to cause disease or detrimental environmental effects.

### **Basic Wastewater Treatment Processes**

#### **Physical**

Physical processes were some of the earliest methods to remove solids from wastewater, usually by passing wastewater through screens to remove debris and solids. In addition, solids that are heavier than water will settle out from wastewater by gravity. Particles with entrapped air float to the top of water and can also be removed. These physical processes are employed in many modern wastewater treatment facilities today.

#### **Biological**

In nature, bacteria and other small organisms in water consume organic matter in sewage, turning it into new bacterial cells, carbon dioxide, and other by-products. The bacteria normally present in water must have oxygen to do their part in breaking down the sewage. In the 1920s, scientists observed that these natural processes could be contained and accelerated in systems to remove organic material from wastewater. With the addition of oxygen to wastewater, masses of microorganisms grew and rapidly metabolized organic pollutants.

Any excess microbiological growth could be removed from the wastewater by physical processes. Activated Sludge is a suspended growth process for removing organic matter from sewage by saturating it with air and microorganisms that can break down the organic matter. Advanced Treatment involves treatment levels beyond secondary treatment.



### **Chemical**

Chemicals can be used to create changes in pollutants that increase the removal of these new forms by physical processes. Simple chemicals such as alum, lime or iron salts can be added to wastewater to cause certain pollutants, such as phosphorus, to floc or bunch together into large, heavier masses which can be removed faster through physical processes. Over the past 30 years, the chemical industry has developed synthetic inert chemicals known as polymers to further improve the physical separation step in wastewater treatment. Polymers are often used at the later stages of treatment to improve the settling of excess microbiological growth or biosolids.

### **Organisms**

Many different types of organisms live in wastewater and some are essential contributors to treatment. A variety of bacteria, protozoa, and worms work to break down certain carbon-based (organic) pollutants in wastewater by consuming them. Through this process, organisms turn wastes into carbon dioxide, water, or new cell growth.

Bacteria and other microorganisms are particularly plentiful in wastewater and accomplish most of the treatment. Most wastewater treatment systems are designed to rely in large part on biological processes.

### **Pathogens**

Many disease-causing viruses, parasites, and bacteria also are present in wastewater and enter from almost anywhere in the community. These pathogens often originate from people and animals are infected with or are carriers of a disease. Graywater and blackwater from typical homes contain enough pathogens to pose a risk to public health. Other likely sources in communities include hospitals, schools, farms, and food processing plants.

Some illnesses from wastewater-related sources are relatively common. Gastroenteritis (shown below) can result from a variety of pathogens in wastewater, and cases of illnesses caused by the parasitic protozoa *Giardia lamblia* and *Cryptosporidium* are not unusual in the U.S. Other important wastewater-related diseases include hepatitis A, typhoid, polio, cholera, and dysentery. Outbreaks of these diseases can occur as a result of drinking water from wells polluted by wastewater, eating contaminated fish, or recreational activities in polluted waters. Some illnesses can be spread by animals and insects that come in contact with wastewater.

Even municipal drinking water sources are not completely immune to health risks from wastewater pathogens. Drinking water treatment efforts can become overwhelmed when water resources are heavily polluted by wastewater. For this reason, wastewater treatment is as important to public health as drinking water treatment.



### **Ciliate**

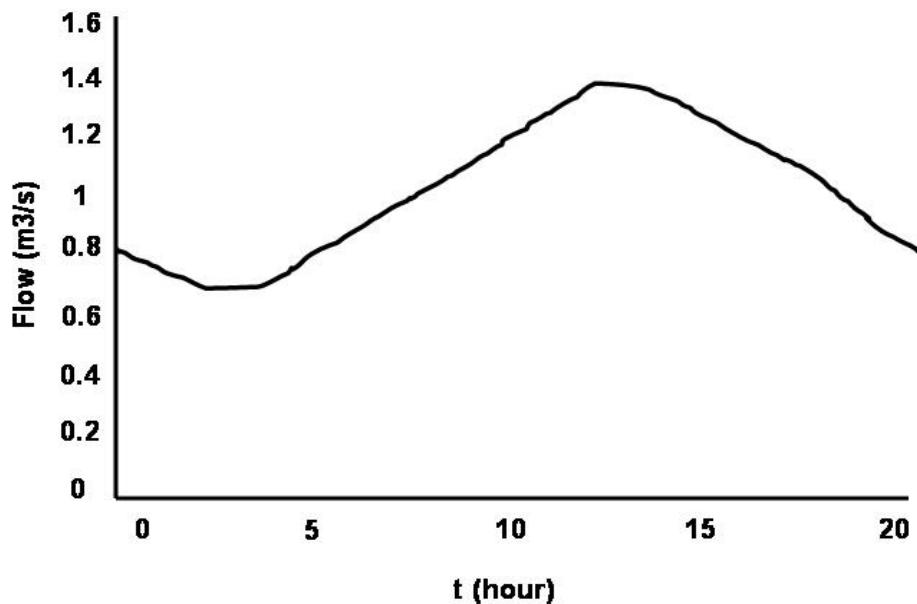
*Vorticella* is a stalked ciliate. There are at least a dozen species found in activated sludge ranging in length from about 30 to 150  $\mu\text{m}$ . These organisms are oval to round shaped, have a contractile stalk, a domed feeding zone, and a water vacuole located near the terminal end of the feeding cavity.

## Domestic Wastewater Characteristics

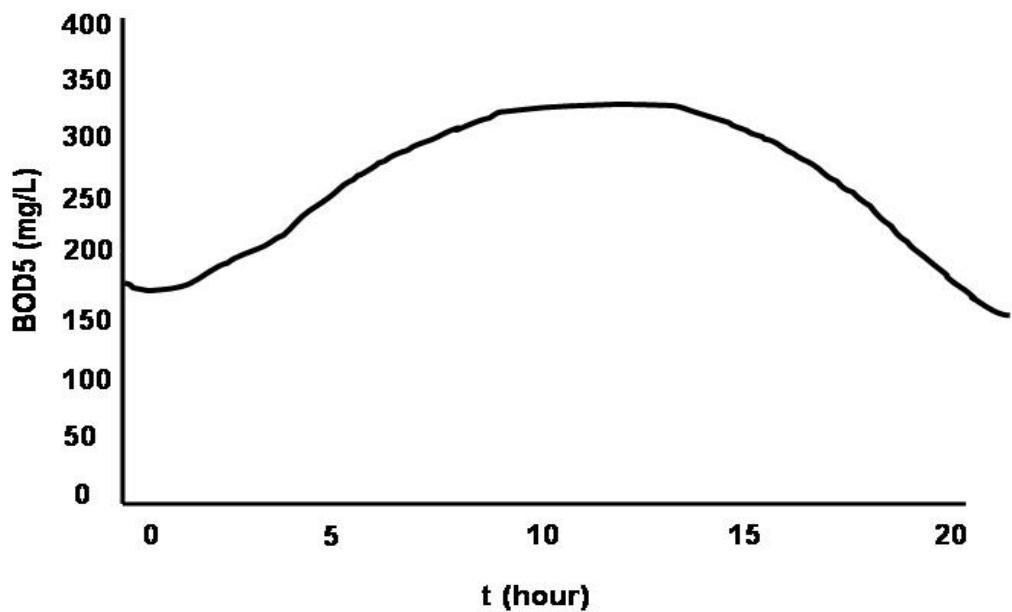
Typical major pollutant characteristics of domestic wastewater

Type	Pollutant	Conc. (mg/L)
<b>Physical</b>	Total Suspended Solids	300
	Volatile Suspended Solids	240
	Fixed Suspended Solids	60
	Total Dissolved Solids	440
	Volatile Suspended Solids	175
	Fixed Suspended Solids	265
	Temperature	10 - 25 °C
	Color	Grey - Black
<b>Chemical</b>	BOD <sub>5</sub>	250
	COD	500
	TOC	160
	Total N	40
	Organic N	15
	Free ammonia N	25
	Nitrite N	0
	Nitrates N	0
	Total P	9
	Organic P	4
	Inorganic P	5
	Alkalinity	100
	Fats, oil and grease (FOG)	100
<b>Microbiological</b>	Total coliform	10 <sup>8</sup> - 10 <sup>9</sup> MPN/L
	Fecal coliform	10 <sup>7</sup> - 10 <sup>8</sup> MPN/L
	Non-fecal coliform	9x10 <sup>7</sup> - 9x10 <sup>8</sup> MPN/L
	Total viruses	1,000-10,0000 infectious units/L

Typical Flow Rate of Domestic Wastewater



Typical BOD5 Variation of Domestic Wastewater



# Effects of Wastewater Pollutants

## Effect of BOD

- Depletes dissolved oxygen from streams, lakes and oceans.
- May cause death of aerobic organisms (fish kills, etc.).
- Increases anaerobic properties of water.

## Effect of TSS

- **Increases turbidity**
  - Less light - reduced photosynthesis.
  - Causes fish's gills to get plugged up.
- **Increases silting**
  - Reduces lifetime of lakes.
  - Changes benthic (i.e., bottom) ecology.

## Effects of Phosphorous and Nitrogen

- **Increases algal photosynthesis (eutrophication)**
  - Increased plant life on surface.
  - Reduces light in lower levels.

## Additional Effects of Nitrogen

- Organic nitrogen and ammonia are converted to nitrates in water.
- Nitrates are converted to nitrites in digestive system.
- Nitrites are assimilated into blood stream where they are converted by respiration to nitrates.
- May cause suffocation (blue baby syndrome).

## Effect of pH

- Organisms are very susceptible to acids and bases.
- Recommended to have near neutral conditions (6.5 - 8.5).

## Effect of Pathogens

### May infect:

- Humans
- Animals



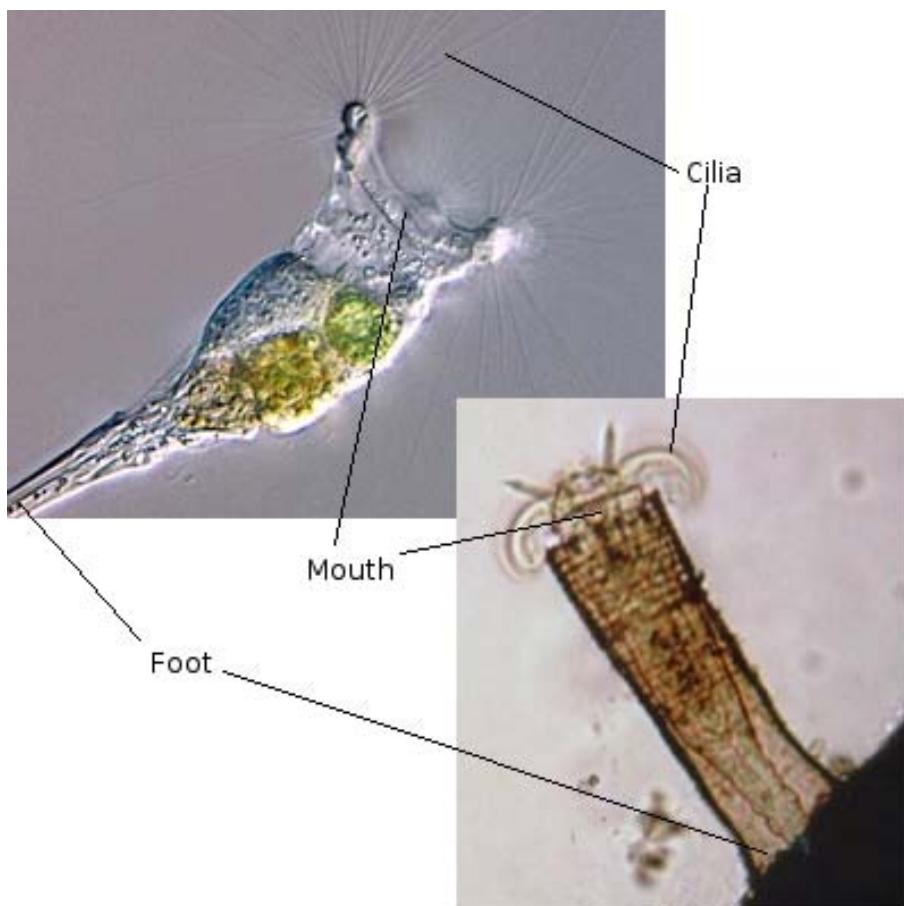
Domestic waste overflow at the head works. Yes, incredibly headworks do overflow, usually due to rags, grease and debris or operator error. We do not like to see this happening and are very careful about letting the public and state regulatory agencies see this activity. One activity the State does not want to see but will happen during a rain storm is bypassing untreated waste to the outfall.



Vorticella



Ciliate



Rotifer

## Organic Matter

Organic materials are found everywhere in the environment. They are composed of the carbon-based chemicals that are the building blocks of most living things. Organic materials in wastewater originate from plants, animals, or synthetic organic compounds, and enter wastewater in human wastes, paper products, detergents, cosmetics, foods, and from agricultural, commercial, and industrial sources.

Organic compounds normally are some combination of carbon, hydrogen, oxygen, nitrogen, and other elements. Many organics are proteins, carbohydrates, or fats and are biodegradable, which means they can be consumed and broken down by organisms. However, even biodegradable materials can cause pollution. In fact, too much organic matter in wastewater can be devastating to receiving waters.

Large amounts of biodegradable materials are dangerous to lakes, streams, and oceans, because organisms use dissolved oxygen in the water to break down the wastes. This can reduce or deplete the supply of oxygen in the water needed by aquatic life, resulting in fish kills, odors, and overall degradation of water quality. The amount of oxygen organisms need to break down wastes in wastewater is referred to as the biochemical oxygen demand (**BOD**) and is one of the measurements used to assess overall wastewater strength.

Some organic compounds are more stable than others and cannot be quickly broken down by organisms, posing an additional challenge for treatment. This is true of many synthetic organic compounds developed for agriculture and industry.

In addition, certain synthetic organics are highly toxic. Pesticides and herbicides are toxic to humans, fish, and aquatic plants and often are disposed of improperly in drains or carried in stormwater. In receiving waters, they kill or contaminate fish, making them unfit to eat. They also can damage processes in treatment plants. Benzene and toluene are two toxic organic compounds found in some solvents, pesticides, and other products. New synthetic organic compounds are being developed all the time, which can complicate treatment efforts.

## Oil and Grease

Fatty organic materials from animals, vegetables, and petroleum also are not quickly broken down by bacteria and can cause pollution in receiving environments. When large amounts of oils and greases are discharged to receiving waters from community systems, they increase BOD and they may float to the surface and harden, causing aesthetically unpleasing conditions. They also can trap trash, plants, and other materials, causing foul odors, attracting flies and mosquitoes and other disease vectors. In some cases, too much oil and grease causes septic conditions in ponds and lakes by preventing oxygen from the atmosphere from reaching the water.

Onsite systems also can be harmed by too much oil and grease, which can clog onsite system drainfield pipes and soils, adding to the risk of system failure. Excessive grease also adds to the septic tank scum layer, causing more frequent tank pumping to be required. Both possibilities can result in significant costs to homeowners.

Petroleum-based waste oils used for motors and industry are considered hazardous waste and should be collected and disposed of separately from wastewater.

## Inorganics

Inorganic minerals, metals, and compounds, such as sodium, potassium, calcium, magnesium, cadmium, copper, lead, nickel, and zinc are common in wastewater from both residential and nonresidential sources. They can originate from a variety of sources in the community including industrial and commercial sources, stormwater, and inflow and infiltration from cracked pipes and leaky manhole covers. Most inorganic substances are relatively stable, and cannot be broken down easily by organisms in wastewater.

Large amounts of many inorganic substances can contaminate soil and water. Some are toxic to animals and humans and may accumulate in the environment. For this reason, extra treatment steps are often required to remove inorganic materials from industrial wastewater sources. For example, heavy metals which are discharged with many types of industrial wastewaters are difficult to remove by conventional treatment methods. Although acute poisonings from heavy metals in drinking water are rare in the U.S., potential long-term health effects of ingesting small amounts of some inorganic substances over an extended period of time are possible.

## Nutrients

Wastewater often contains large amounts of the nutrients nitrogen and phosphorus in the form of nitrate and phosphate, which promote plant growth. Organisms only require small amounts of nutrients in biological treatment, so there normally is an excess available in treated wastewater. In severe cases, excessive nutrients in receiving waters cause algae and other plants to grow quickly depleting oxygen in the water, deprived of oxygen, fish and other aquatic life die, emitting foul odors. Nutrients from wastewater have also been linked to ocean "red tides" that poison fish and cause illness in humans. Nitrogen in drinking water may contribute to miscarriages and is the cause of a serious illness in infants called methemoglobinemia or "blue baby syndrome."

## Solids

Solid materials in wastewater can consist of organic and/or inorganic materials and organisms. The solids must be significantly reduced by treatment or they can increase BOD when discharged to receiving waters and provide places for microorganisms to escape disinfection. They also can clog soil absorption fields in onsite systems.

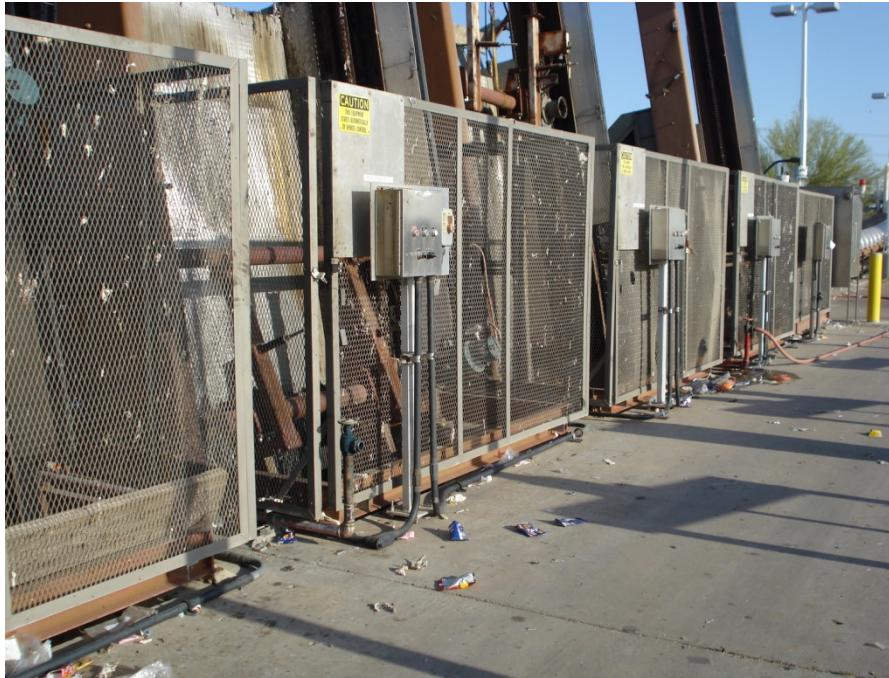
**Settleable solids:** Certain substances, such as sand, grit, and heavier organic and inorganic materials settle out from the rest of the wastewater stream during the preliminary stages of treatment. On the bottom of settling tanks and ponds, organic material makes up a biologically active layer of sludge that aids in treatment.

**Suspended solids:** Materials that resist settling may remain suspended in wastewater. Suspended solids in wastewater must be treated, or they will clog soil absorption systems or reduce the effectiveness of disinfection systems.

**Dissolved solids:** Small particles of certain wastewater materials can dissolve, like salt in water. Some dissolved materials are consumed by microorganisms in wastewater, but others, such as heavy metals, are difficult to remove by conventional treatment. Excessive amounts of dissolved solids in wastewater can have adverse effects on the environment.

## Gases

Certain gases in wastewater can cause odors, affect treatment, or are potentially dangerous. Methane gas, for example, is a byproduct of anaerobic biological treatment and is highly combustible. Special precautions need to be taken near septic tanks, manholes, treatment plants, and other areas where wastewater gases can collect.



Mechanical Bar Screens. Operators are necessary to pick up trash that is blown off the rakes.



Here is a grinder pump that is installed after the bar screens. This debris is sent to the landfill.



Caked grease stuck on weir.



Floating scum in primary clarifier.



Maintenance on a circular clarifier should be performed annually.



Scum Box



Scraping mechanism inside a clarifier.



Scum rake collecting oil and other floating particles.

## Hydrogen Sulfide and Ammonia

The gases hydrogen sulfide and ammonia can be toxic and pose asphyxiation hazards. Ammonia as a dissolved gas in wastewater also is dangerous to fish. Both gases emit odors, which can be a serious nuisance. Unless effectively contained or minimized by design and location, wastewater odors can affect the mental well-being and quality of life of residents. In some cases, odors can even lower property values and affect the local economy.

Hydrogen sulfide or  $H_2S$  problems are very common in the collection and wastewater system. There are many chemicals used to help or treat this problem. Here are a few used in the treatment of hydrogen sulfide problems: Salts of zinc, lime, hydrogen peroxide, chlorine and magnesium hydroxide. Hydrogen sulfide production in collection systems can cause a number of problems such as corrosion of the pipes, manholes, and creation of hazardous atmospheres and foul odors. The best method of controlling hydrogen sulfide is to eliminate its habitat or growth area by keeping sewers cleaner, this will harbor fewer slime bacteria. Here are some important statements regarding the reduction of hydrogen sulfide: Salts of zinc and iron may precipitate sulfides, lime treatments can also kill bacteria which produce hydrogen sulfide, but this creates a sludge disposal problem and chlorination is effective at reducing the bacteria which produce hydrogen sulfide. Hydrogen sulfide conditions occur in the sewer system because of the lack of oxygen.

### **Pollutants, Oxygen-Demanding Substances**

Dissolved oxygen is a key element in water quality that is necessary to support aquatic life. A demand is placed on the natural supply of dissolved oxygen by many pollutants in wastewater. This is called biochemical oxygen demand, or BOD, and is used to measure how well a sewage treatment plant is working. If the effluent, the treated wastewater produced by a treatment plant, has a high content of organic pollutants or ammonia, it will demand more oxygen from the water and leave the water with less oxygen to support fish and other aquatic life. Organic matter and ammonia are "oxygen-demanding" substances. Oxygen-demanding substances are contributed by domestic sewage and agricultural and industrial wastes of both plant and animal origin, such as those from food processing, paper mills, tanning, and other manufacturing processes. These substances are usually destroyed or converted to other compounds by bacteria if there is sufficient oxygen present in the water, but the dissolved oxygen needed to sustain fish life is used up in this break down process.

### **Pathogens**

Disinfection of wastewater and chlorination of drinking water supplies has reduced the occurrence of waterborne diseases such as typhoid fever, cholera, and dysentery, which remain problems in underdeveloped countries while they have been virtually eliminated in the infectious micro-organisms, or pathogens, may be carried into surface and groundwater by sewage from cities and institutions, by certain kinds of industrial wastes, such as tanning and meat packing plants, and by the contamination of storm runoff with animal wastes from pets, livestock and wild animals, such as geese or deer. Humans may come in contact with these pathogens either by drinking contaminated water or through swimming, fishing, or other contact activities. Modern disinfection techniques have greatly reduced the danger of waterborne disease.

### **Nutrients**

Carbon, nitrogen, and phosphorus are essential to living organisms and are the chief nutrients present in natural water. Large amounts of these nutrients are also present in sewage, certain industrial wastes, and drainage from fertilized land. Conventional secondary biological treatment processes do not remove the phosphorus and nitrogen to any substantial extent. They may convert the organic forms of these substances into mineral form, making them more usable by plant life. When an excess of these nutrients over-stimulates the growth of water plants, the result causes unsightly conditions, interferes with drinking water treatment processes, and causes unpleasant and disagreeable tastes and odors in drinking water.

The release of large amounts of nutrients, primarily phosphorus but occasionally nitrogen, causes nutrient enrichment which results in excessive growth of algae. Uncontrolled algae growth blocks out sunlight and chokes aquatic plants and animals by depleting dissolved oxygen in the water at night. The release of nutrients in quantities that exceed the affected waterbody's ability to assimilate them results in a condition called eutrophication or cultural enrichment.

### Inorganic and Synthetic Organic Chemicals

A vast array of chemicals is included in this category. Examples include detergents, household cleaning aids, heavy metals, pharmaceuticals, synthetic organic pesticides and herbicides, industrial chemicals, and the wastes from their manufacture. Many of these substances are toxic to fish and aquatic life and many are harmful to humans. Some are known to be highly poisonous at very low concentrations. Others can cause taste and odor problems, and many are not effectively removed by conventional wastewater treatment.

### Thermal

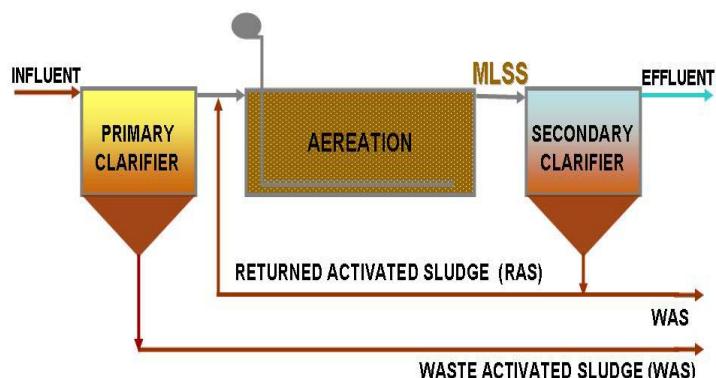
Heat reduces the capacity of water to retain oxygen. In some areas, water used for cooling is discharged to streams at elevated temperatures from power plants and industries. Even discharges from wastewater treatment plants and storm water retention ponds affected by summer heat can be released at temperatures above that of the receiving water, and elevate the stream temperature. Unchecked discharges of waste heat can seriously alter the ecology of a lake, a stream, or estuary.

### Primary Treatment

The initial stage in the treatment of domestic wastewater is known as primary treatment. Coarse solids are removed from the wastewater in the primary stage of treatment. In some treatment plants, primary and secondary stages may be combined into one basic operation. At many wastewater treatment facilities, influent passes through preliminary treatment units before primary and secondary treatment begins. One of the most common forms of pollution control in the United States is **wastewater treatment**. The country has a vast system of collection sewers, pumping stations, and treatment plants. Sewers collect the wastewater from homes, businesses, and many industries, and deliver it to plants for treatment. Most treatment plants were built to clean wastewater for discharge into streams or other receiving waters, or for reuse.

Years ago, when sewage was dumped into waterways, a natural process of purification began. First, the sheer volume of clean water in the stream diluted wastes. Bacteria and other small organisms in the water consumed the sewage and other organic matter, turning it into new bacterial cells; carbon dioxide and other products. Today's higher populations and greater volume of domestic and industrial wastewater require that communities give nature a helping hand. The basic function of wastewater treatment is to speed up the natural processes by which water is purified.

There are **two basic stages** in the treatment of wastes, **primary** and **secondary**. In the primary stage, solids are allowed to settle and removed from wastewater. The secondary stage uses biological processes to further purify wastewater. Sometimes, these stages are combined into one operation.



## Preliminary Treatment

The Preliminary Treatment is purely physical stage consisting of Coarse Screening, Raw Influent Pumping, Static Fine Screening, Grit Removal, and Selector Tanks. The raw wastewater enters from the collection system into the Coarse Screening process. After the wastewater has been screened, it may flow into a grit chamber where sand, grit, cinders, and small stones settle to the bottom.

Removing the grit and gravel that washes off streets or land during storms is very important, especially in cities with combined sewer systems. Large amounts of grit and sand entering a treatment plant can cause serious operating problems, such as excessive wear of pumps and other equipment, clogging of aeration devices, or taking up capacity in tanks that is needed for treatment.

In some plants, another finer screen is placed after the grit chamber to remove any additional material that might damage equipment or interfere with later processes. The grit and screenings removed by these processes must be periodically collected and trucked to a landfill for disposal or are incinerated.

### Collected Grit



The Coarse Screening consists of a basket shaped bar screen which collects larger debris (several inches in diameter) prior to the Raw Influent Pumping. This debris is removed and placed into a dumpster for disposal into the landfill.

The wastewater then passes into the Raw Influent Pumping process that consists of submersible centrifugal pumps. These influent pumps operate under a principal termed prerotation, which allows them to vary their pump rate hydraulically without the use of complex and expensive electronics.

### Manual and Mechanical Bar Screens

The flow then passes into the Static Fine Screening process which consists of two stationary (or static) screens which remove finer debris not captured by the coarse screens. This screened debris is then dewatered and collected in hoppers for disposal into a landfill.

The wastewater then passes into the Grit Removal process which consists of two vortex grit separators which produce a whirlpool action to force the finest debris to the outside perimeter for subsequent collection. This debris is then collected in hoppers, dewatered, and disposed into a landfill. The screened and de-gritted wastewater then enters into Primary Sedimentation.

### Fine Screening



### Primary Sedimentation

With the screening completed and the grit removed, wastewater still contains dissolved organic and inorganic constituents along with suspended solids. The suspended solids consist of minute particles of matter that can be removed from the wastewater with further treatment such as sedimentation or gravity settling, chemical coagulation, or filtration.



Primary Clarifier

Pollutants that are dissolved or are very fine and remain suspended in the wastewater are not removed effectively by gravity settling. When the wastewater enters a sedimentation tank, it slows down and the suspended solids gradually sink to the bottom. This mass of solids is called primary sludge. Various methods have been devised to remove solids, newer plants have some type of mechanical equipment to remove the settled solids and some plants remove solids continuously while others do so at intervals.

### Secondary Treatment

After the wastewater has been through Primary Treatment processes, it flows into the next stage of treatment called secondary. Secondary treatment processes can remove up to 90 percent of the organic matter in wastewater by using biological treatment processes. The two most common conventional methods used to achieve secondary treatment are attached growth processes and suspended growth processes.

The Secondary Treatment stage consists of a biological process such as **Oxidation Ditches** and a physical process, **Secondary Clarification**. The Preliminary Treatment stage removed as much solids as possible using physical processes, however, very fine solids are still present that cannot be removed physically.

The wastewater enters from Preliminary Treatment into the Oxidation Ditches process which is a biological process consisting of two large oval shaped basins which are capable of removing these finer solids. This is accomplished by maintaining a population of microorganisms within the oxidation basins which consume the very fine solids (which are primarily organic) and also adhere to the solids themselves. By consuming and adhering to these finer solids they form larger and heavier aggregates that can be physically separated. Thus, after this process has taken place within the Oxidation Ditches the wastewater then enters Secondary Clarification process which can provide this physical separation.



# Nitrogen and Phosphorus Removal Technologies

## Introduction

This section provides information on a number of different technologies that can reduce nitrogen and phosphorus levels. The actual technology selected will be site-specific and dependent on many factors including soil conditions, influent water quality, required effluent levels, disposal options, availability of land, cost, etc. In some cases, a combination of technologies may be necessary to effectively remove all the contaminants of concern. Small system owners and operators should work closely with their state onsite and decentralized program staff as well as engineers to ensure that the technologies selected will work effectively in combination to achieve the effluent goals.

## Nutrient Removal Technologies

### Fixed-film systems - Aerobic/anaerobic trickling filter package plant

Fixed-film systems (FFSs) are biological treatment processes that employ a medium such as rock, plastic, wood, or other natural or synthetic solid material that will support biomass on its surface and within its porous structure (USEPA, 2008c). Trickling filter FFSs are typically constructed as beds of media through which wastewater flows. Oxygen is normally provided by natural or forced ventilation. Commercial on-site systems use synthetic media and receive wastewater from overlying sprayheads for aerobic treatment and nitrification. Nitrified effluent returns to the anoxic zone to mix with either septic tank contents or incoming septic tank effluent for denitrification. A portion of the denitrified effluent is discharged for disposal or further treatment. Aerobic tanks are available in residential or small community sizes. Typical trickling filters systems currently available are capable of producing effluent BOD and TSS concentrations of 5 to 40 mg/L. Nitrogen removal typically varies from 0 to 35 percent although removal percentages as high as 65% have been demonstrated through USEPA's Environmental Technology Verification (ETV) program. Phosphorus removal is typically 10 to 15 percent.

Higher removal occurs at low loading rates in warm climates. Systems can be configured for single-pass use where the treated water is applied to the trickling filter once before being disposed of, or for multi-pass use where a portion of the treated water is cycled back to the septic tank and re-treated via a closed loop.

Multi-pass systems result in higher treatment quality and assist in removing Total Nitrogen (TN) levels by promoting nitrification in the aerobic media bed and denitrification in the anaerobic septic tank. Factors affecting performance include influent wastewater characteristics, hydraulic and organic loading, medium type, maintenance of optimal DO levels, and recirculation rates.

### Sequencing batch reactor (SBR)

The SBR process is a sequential suspended growth (activated sludge) process in which all major steps occur in the same tank in sequential order (USEPA, 2008d). The SBR system is typically found in packaged configurations for onsite and small community or cluster applications. The major components of the package include the batch tank, aerator, mixer, decanter device, process control system (including timers), pumps, piping, and appurtenances. Aeration may be provided by diffused air or mechanical devices. SBRs are often sized to provide mixing as well and are operated by the process control timers.

Mechanical aerators have the added value of potential operation as mixers or aerators. The decanter is a critical element in the process. Several decanter configurations are available, including fixed and floating units. At least one commercial package employs a thermal processing step for the excess sludge produced and wasted during the "idle" step. The key to the SBR process is the control system, which consists of a combination of level sensors, timers, and microprocessors which can be configured to meet the needs of the system.

SBRs can be designed and operated to enhance removal of nitrogen, phosphorus, and ammonia, in addition to removing TSS and BOD. Package plant SBRs are suitable for areas with little land, stringent treatment requirements, and small wastewater flows such as RV parks or mobile homes, campgrounds, construction sites, rural schools, hotels, and other small applications. These systems are also useful for treating pharmaceutical, brewery, dairy, pulp and paper, and chemical wastes (USEPA, 2000d).

### **Intermittent sand filters (ISF)**

ISF is used to describe a variety of packed-bed filters of sand or other granular materials available on the market (USEPA, 2008g). Sand filters provide advanced secondary treatment of settled wastewater or septic tank effluent. They consist of a lined (e.g., impervious PVC liner on sand bedding) excavation or structure filled with uniform washed sand that is placed over an underdrain system. The wastewater is directed onto the surface of the sand through a distribution network and allowed to percolate through the sand to the underdrain system. The underdrain system collects the filter effluent for further processing or discharge.

Sand filters are aerobic, fixed-film bioreactors. Bioslimes from the growth of microorganisms develop as films on the sand particle surfaces. The microorganisms in the slimes capture soluble and colloidal waste materials in the wastewater as it percolates over the sand surfaces. The captured materials are metabolized into new cell mass or degraded under aerobic conditions to carbon dioxide and water. Most biochemical treatment occurs within approximately 6 inches of the filter surface. Other treatment mechanisms that occur in sand filters include physical processes, such as straining and sedimentation, to remove suspended solids within the pores of the media. Most suspended solids are strained out at the filter surface.

Chemical adsorption can occur throughout the media bed. Adsorption sites in the media are usually limited, however. The capacity of the media to retain ions depends on the target constituent, the pH, and the mineralogy of the media. Phosphorous is one element of concern in wastewater that can be removed in this manner, but the number of available adsorption sites is limited by the characteristics of the media.

Sand filters can be used for a broad range of applications, including single-family residences, large commercial establishments, and small communities. Sand filters are frequently used to pretreat septic tank effluent prior to subsurface infiltration onsite where the soil has insufficient unsaturated depth above ground water or bedrock to achieve adequate treatment. They are also used to meet water quality requirements (with the possible exception of fecal coliform removal) before direct discharge to surface water. Sand filters are used primarily to treat domestic wastewater, but they have been used successfully in treatment trains to treat wastewaters high in organic materials such as those from restaurants and supermarkets. Single-pass ISF filters are most frequently used for smaller applications and sites where nitrogen removal is not required. However, they can be combined with anoxic processes to significantly increase nitrogen removal.

### **Recirculating sand filters (RSF)**

Recirculating filters using sand, gravel, or other media provide advanced secondary treatment of settled wastewater or septic tank effluent (USEPA, 2008h). They consist of a lined (e.g., impervious PVC liner on sand bedding) excavation or structure filled with uniform washed sand that is placed over an underdrain system. The wastewater is directed onto the surface of the sand through a distribution network and allowed to percolate through the sand to the underdrain system. The underdrain system collects and recycles the filter effluent to the recirculation tank for further processing or discharge.

The basic components of recirculating filters include a recirculation/dosing tank, pump and controls, distribution network, filter bed with an underdrain system, and a return line. The return line or the underdrain must split the flow to recycle a portion of the filtrate to the recirculation/dosing tank. A small volume of wastewater and filtrate is dosed to the filter surface on a timed cycle 1 to 3 times per hour.

Recirculation ratios are typically between 3:1 and 5:1. In the recirculation tank, the returned aerobic filtrate mixes with the anaerobic septic tank effluent before being reapplied to the filter. RSFs can be used for a broad range of applications, including single-family residences, large commercial establishments, and small communities. They produce a high quality effluent with approximately 85 to 95 percent BOD and TSS removal. In addition, almost complete nitrification is achieved.

Denitrification also has been shown to occur in RSFs. Depending on modifications in design and operation, 50 percent or more of applied nitrogen can be removed (USEPA, 1999). To enhance this capability, they can be combined with a greater supply of biodegradable organic carbon, time, and mixing than is normally available from the conventional recirculation tank.

### **Natural Systems**

The natural systems described here include constructed wetlands and floating aquatic plant treatment systems. Wetland systems are typically described in terms of the position of the water surface and/or the type of vegetation grown. Most natural wetlands are free water surface (FWS) systems where the water surface is exposed to the atmosphere; these include bogs (primary vegetation mosses), swamps (primary vegetation trees), and marshes (primary vegetation grasses and emergent macrophytes) (USEPA, 2000e). subsurface flow (SF) wetlands are specifically designed to treat or polish wastewater and are typically constructed as a bed or channel containing appropriate media.

Constructed wetlands treat wastewater by bacterial decomposition, settling, and filtering. As in tank designs, bacteria break down organic matter in the wastewater, aerobically, anoxically and anaerobically. Oxygen for aerobic decomposition is supplied by the plants growing in the wetland. Solids are filtered and finally settle out of the wastewater within the wetland. After about two weeks in the wetland, effluent is usually discharged by gravity to an unlined wetland bed. If these systems discharge effluent to surface ditches, they require a NPDES permit.

The submerged plant roots do provide substrate for microbial processes. However, the amount of oxygen that emergent macrophytes can transmit from the leaves to their roots is negligible compared to the oxygen demand of wastewater. Therefore subsurface flow wetlands are devoid of oxygen. The lack of oxygen in these subsurface flow systems means that ammonia oxidation via biological nitrification will not occur without the use of an additional unit process, such as a gravel trickling filter for nitrification of the wastewater ammonia. Vertical flow wetland beds are a modification of subsurface flow wetlands which contain gravel or coarse sand and are loaded intermittently at the top surface. Unlike ammonia oxidation, nitrate removal in a subsurface flow wetland can be rapid and effective because the anoxic conditions and carbon sources necessary to support the treatment reactions occur naturally in these systems.

FWS wetlands with long detention times can remove minor amounts of phosphorus through plant uptake, adsorption, complexation, and precipitation. However, removal via plant uptake is limited to phosphorus retained in plant litter that is buried by sediments before plant decomposition occurs (i.e. peat building process). Phosphorus removal is typically greater in the first year or two because of soil absorption and rapidly expanding vegetation but decreases when the system reaches equilibrium, and unburied plant litter releases phosphorus back into the water as it decomposes. Phosphorus removal is also possible with the use of an addition process, such as chemical addition and mixing prior to a final deep settling pond.

Aquatic systems using duckweed have been used for a number of years to treat wastewater for various purposes (WEF, 2001). Duckweed (*Lemna spp.*) are floating macrophytes. Duckweed fronds can double their mass in two days under ideal conditions of nutrient availability, sunlight, and temperature. Although duckweed can be found in most regions, the rate of growth is optimal at 20 to 30°C and they grow best in a pH range of 3.5 to 8.5.

Duckweed can grow about six months per year in most U.S. climates. High levels of BOD and TSS removal have been observed from duckweed systems. To achieve secondary treatment most duckweed systems are coupled with either facultative or aerated ponds. Nitrogen is removed by plant uptake and harvesting, by denitrification, or a combination of the two. Typically less than 1 mg/L of phosphorus can be removed by plant uptake and harvest. If significant phosphorus removal is required, chemical precipitation with alum, ferric chloride, or other chemicals used in a separate treatment step is necessary. The major disadvantage of duckweed systems is the large amount of biomass produced by the rapidly growing plants, which creates a solids handling requirement similar to handling sludge at an aerobic wastewater treatment facility.

### **Proprietary Filters/Improved and Emerging Technologies**

A number of companies have developed proprietary nitrogen and phosphorus removal technologies that can be used at centralized wastewater treatment facilities as well as at onsite, decentralized systems. This section provides a general description of some of these technologies without mentioning specific trade names.

### **Sustainable Nutrient Recovery**

While the U.S. is primarily addressing nutrient removal concerns through development of WQSSs and treatment at centralized wastewater facilities, a number of European countries including Switzerland, Sweden, and the Netherlands are conducting research on innovative sustainable nutrient recovery systems. The concept behind these new technologies is to separate and treat toilet waste before it leaves the home or building and mixes with the larger waste stream to be carried to WWTPs.

Recent studies have shown that about 80 percent of the nitrogen and 50 percent of the phosphorus in wastewater are derived from urine although urine makes up only 1 percent of the volume of wastewater (Larsen and Leinert, 2007). Separating the urine from wastewater could offer various advantages: WWTPs could be built on a smaller scale, water bodies will be better protected from nitrogen and phosphorus pollution, nutrients could be recycled for agricultural use, and various constituents of concern including hormones and pharmaceutical compounds could be removed before being mixed with wastewater and released to the environment. A major benefit would be reduced energy consumption at WWTPs as a result of reduced treatment requirements for nitrogen. Also, separating 50 to 60 percent of urine could reduce in-plant nitrogen gas discharges and result in fewer impurities in methane captured from sludge digestion.

Organizations such as the Swiss Federal Institute of Aquatic Science and Technology (Eawag) are currently experimenting with the development and application of “NoMix technology” to separate urine from solid waste at the toilet bowl. While similar in size and shape to current toilets, this new technology has two waste pipes – a small front one that collects and diverts urine into a storage tank, and a larger rear waste pipe that operates like a standard toilet. The first of these toilets were installed in two “eco-villages” in Sweden in 1994 and since then have spread to other locations throughout the country and to Denmark, the Netherlands, and Switzerland. The concept is now taking hold in Austria and Germany. While the pollutant-free urine, or “urevit,” can be spray-applied directly onto agricultural fields; in the Netherlands, a company called Grontmij trucks stored urine to a special treatment plant where the phosphate is precipitated out as a mineral called struvite and used as a fertilizer.

Novaquatis, a branch of Eawag is experimenting with extracting nitrogen and potassium from urine that can be sprayed directly onto crops. Eawag is also experimenting with a pilot decentralized basement sewage plant where domestic wastewater is treated in a MBR so it can be reused for flushing the toilets or watering the garden and the sewage sludge is composted. While still experimental, some of these technologies may have practical future applications if widely applicable low-cost solutions can be found for urine transport, or stable and cost-effective technologies can be developed for decentralized treatment.

While studies of consumer attitudes and acceptance appear to be positive, technological improvements are still needed to prevent clogging in pipes, to identify best treatment options that can be applied in practice; and to identify how and where to convert urine to fertilizer.

Sustainability concerns are also driving the wastewater treatment industry to start looking at sludge as a renewable resource. Historically, agricultural use has been the traditional approach for disposal of municipal sludge due to its high nutrient content for fertilizing crops, and its low cost approach. As scientific advances detect smaller and smaller quantities of contaminants (i.e., heavy metals, pathogenic microorganisms, pharmaceuticals, and personal care products), the public, farming organizations, and the food industry are raising concerns about continuing this practice. As noted above, researchers are discovering that valuable products can be generated from sewage treatment byproducts such as energy extracted from anaerobic digestion, construction materials such as bricks, and nutrients such as phosphorus that can be extracted from sludge and used as fertilizer.

In February 2008, the non-profit Global Water Research Coalition, an international water research alliance formed by 12 world-leading research organizations, released a report titled, *State of Science Report: Energy and Resource Recovery from Sludge* (Kalogo and Monteith, 2008). The report focuses on:

- The international situation of energy and resource recovery from sludge
- How the use of different sludge treatment processes affects the possibility of recovering energy and/or materials from the residual sludge
- The influence of market and regulatory drivers on the fate of the sludge end-product
- The feasibility of energy and resource recovery from sludge
- The social, economic, and environmental performance (triple bottom line or TBL assessment) of current alternatives technologies

**Four market drivers are identified and discussed including:**

- Sustainability and environmental concerns, such as the threat of soil pollution, global warming and resource depletion
- Rising energy costs and the need of more electricity and heat to operate the plants
- Requirements for high quality of resources for industrial applications, such as calcium phosphate for the phosphate industry
- Regulation as a factor stimulating the development of new technologies

In the report, energy recovery technologies are classified into sludge-to-biogas processes, sludge-to-syngas processes, sludge-to-oil processes, and sludge-to-liquid processes. The technologies available for resource recovery discussed in the report include those to recover phosphorus, building materials, nitrogen, and volatile acids. The report, which covers both established as well as emerging technologies, will be used as the basis for development of the coalition's future strategic research plan on energy and recovery from sludge. As a technical resource, it provides a valuable overview of sludge disposal practices in various countries such as the U.S., the Netherlands, the United Kingdom, Germany, Sweden, Japan, and China; and presents a number of treatment processes for resource recovery.

Other groups have looked at recovering phosphorus from the supernatant from anaerobic digestion. Several different processes have been proposed that rely on precipitation of the phosphorus as either struvite or calcium phosphate. Work is underway on projects in Italy, Germany, the Netherlands, and Canada (SCOPE, 2004).

## **Nutrient Removal for Small Communities and Decentralized Wastewater Treatment Systems**

Approximately 25 percent of the U.S. population is served by onsite septic or decentralized systems. Onsite septic systems treat and dispose of effluent on the same property that produces the wastewater, whereas decentralized treatment refers to onsite or cluster systems that are used to treat and dispose of relatively small volumes of wastewater, generally from dwellings and businesses that are located relatively close together. In many cases, wastewater from several homes is pretreated onsite by individual septic tanks before being transported through alternative sewers to an offsite decentralized treatment unit that is relatively simple to operate and maintain.

The remaining 75 percent of the population is served by centralized wastewater treatment facilities, which collect and treat large volumes of wastewater. There is, in fact, a growing movement toward decentralized or clustered wastewater treatment systems to reduce cost, to provide groundwater recharge near the source, and for speed and ease in siting since they are generally located underground. The use of residential cluster development is gaining in popularity across the U.S. as a means to permanently protect open space, preserve agricultural land, and protect wildlife habitat (Mega et al., 1998). As part of these developments, wastewater systems such as community drainfields, irrigation systems, and package plants are being installed to reduce infrastructure investment and minimize adverse environmental impacts. Additional alternatives that include aerobic tanks, sand filters, and constructed wetlands can be used to reduce nutrient pollution; particularly in sensitive coastal areas or over sensitive, unconfined aquifers used for drinking water (Anderson and Gustafson, 1998).

### **Phosphorus Removal**

Few phosphorus removal processes are well developed for onsite wastewater systems application (USEPA, 2008e). The controlled addition of chemicals such as aluminum, iron, and calcium compounds with subsequent flocculation and sedimentation has had only limited success because of inadequate operation and maintenance of mechanical equipment and excessive sludge production. Most notable successes have come with special filter materials that are naturally high in their concentration of the above chemicals, but their service lives are finite. Studies of high-iron sands and high-aluminum muds indicate that 50 to 95 percent of the phosphorus can be removed. However, the life of these systems has yet to be determined, after which the filter media will have to be removed and replaced. Use of supplemental iron powder mixed with natural sands is also being researched. Aside from specialized filter media, the most likely phosphorus-reduction systems are iron-rich intermittent sand filter (ISF) media and SBRs. These are discussed in more detail below.

### **Nitrogen Removal**

Processes that remove 25 to 50 percent of total nitrogen include aerobic biological systems and media filters, especially recirculating filters (USEPA, 2008f). The vast majority of on-site and cluster nitrogen-removal systems employ nitrification and denitrification biological reactions. Most notable of these are recirculating sand filters (RSFs) with enhanced anoxic modifications, SBRs, and an array of aerobic nitrification processes combined with an anoxic/anaerobic process to perform denitrification. Some of the combinations are proprietary. A few recently developed highly instrumented systems that utilize membrane solids separation following biological nitrification and denitrification are capable of removing total nitrogen down to very low concentrations (i.e. 3 – 4 mg/L TN). Nitrogen removal systems generally are located last in the treatment train prior to subsurface wastewater infiltration system (SWIS) disposal or surface water disposal, in which case a disinfection step is typically required. Usually, the minimum total nitrogen standard that can be regularly met is about 10 mg/L. These technologies can be either above ground or below ground.

## Secondary Clarification Process

The Secondary Clarification process consists of four rectangular tanks which provide quiescent (or calm) conditions which allow the larger aggregates of solids and microorganisms to settle out for collection. The clear overflow (or upper layer) is collected at the end of the tank and passed onto the Tertiary process for additional treatment if available.

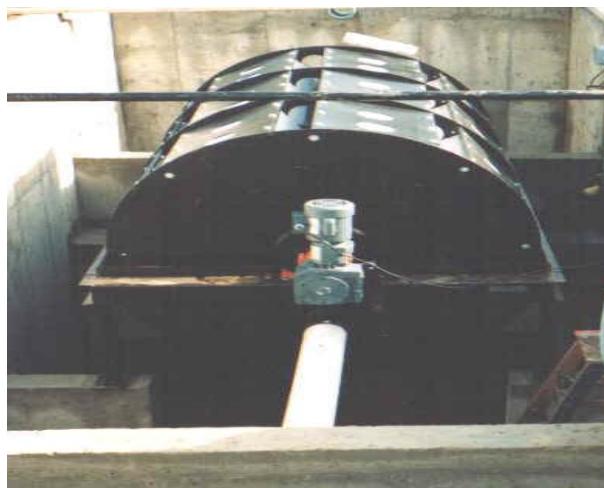
The majority of microorganism-rich underflow (or lower layer) is re-circulated to Tanks as Return Sludge to help sustain the microorganism population in the Oxidation Ditches process. However, if all the underflow was returned the plant would soon become overloaded with solids, therefore, a small portion of this mixture termed Waste Sludge is removed from the system for disposal. The Waste Sludge is transported into the Solids Handing process for disposal.



Secondary Clarifier

### Fixed Film Systems

Fixed film systems grow microorganisms on substrates such as rocks, sand or plastic. The wastewater is spread over the substrate, allowing the wastewater to flow past the film of microorganisms fixed to the substrate. As organic matter and nutrients are absorbed from the wastewater, the film of microorganisms grows and thickens. Trickling filters, rotating biological contactors, and sand filters are examples of fixed film systems.



Empty RBC

### Suspended Film Systems

Suspended film systems stir and suspend microorganisms in wastewater. As the microorganisms absorb organic matter and nutrients from the wastewater, they grow in size and number. After the microorganisms have been suspended in the wastewater for several hours, they are settled out as sludge. Some of the sludge is pumped back into the incoming wastewater to provide "seed" microorganisms. The remainder is wasted and sent on to a sludge treatment process. Activated sludge, extended aeration, oxidation ditch, and sequential batch reactor systems are all examples of suspended film systems.

### Lagoon Systems

Lagoon systems are shallow basins which hold the waste-water for several months to allow for the natural degradation of sewage. These systems take advantage of natural aeration and microorganisms in the wastewater to renovate sewage.

### **Other Important Wastewater Characteristics**

In addition to the many substances found in wastewater, there are other characteristics system designers and operators use to evaluate wastewater. For example, the color, odor, and turbidity of wastewater give clues about the amount and type of pollutants present and treatment necessary. The following are some other important wastewater characteristics that can affect public health and the environment, as well as the design, cost, and effectiveness of treatment.



**Sampling Industrial Waste**, in this photograph, the Inspector or Sampler is shaking the sample to make sure that the sample is mixed-up before pouring off a smaller sample into the smaller sample bottles on the ground. Normally, these Inspectors or Samplers will work in pairs. Get used to having wastewater and/or industrial waste/odors all over your clothes. But other than that, spiders, grease, confined spaces, irate customers, the interesting odors and dangerous Hydrogen Sulfide gas; this is a good job to have, a secure and well-paying job.

#### **Temperature**

The best temperatures for wastewater treatment probably range from 77 to 95 degrees Fahrenheit. In general, biological treatment activity accelerates in warm temperatures and slows in cool temperatures, but extreme hot or cold can stop treatment processes altogether. Therefore, some systems are less effective during cold weather and some may not be appropriate for very cold climates.

Wastewater temperature also affects receiving waters. Hot water, for example, which is a byproduct of many manufacturing processes, can be a pollutant. When discharged in large quantities, it can raise the temperature of receiving streams locally and disrupt the natural balance of aquatic life.

#### **pH**

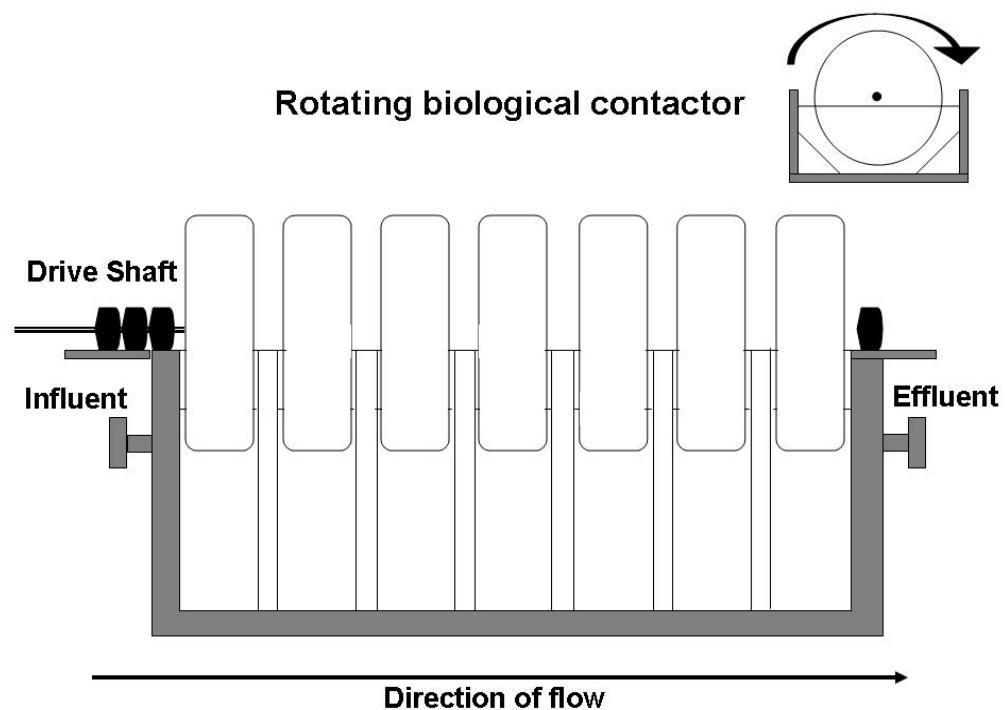
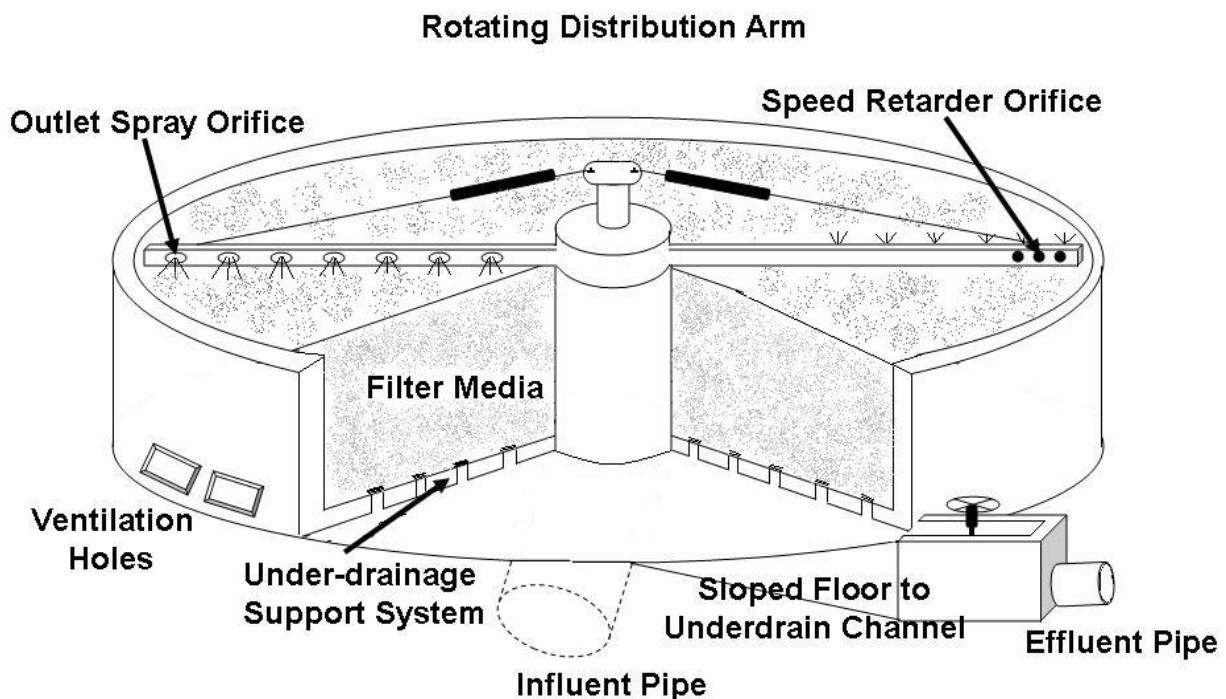
The acidity or alkalinity of wastewater affects both treatment and the environment. Low pH indicates increasing acidity while a high pH indicates increasing alkalinity (a pH of 7 is neutral). The pH of wastewater needs to remain between 6 and 9 to protect organisms. Acids and other substances that alter pH can inactivate treatment processes when they enter wastewater from industrial or commercial sources.



This operator is splitting the sample for bacteriological analysis. Always wear gloves for your and others' safety. We've all seen the operator holds a sandwich in one hand while working in the lab, or the operator does not wear gloves at all. I personally like the operator who can smoke a cigarette or eat while sampling. Hey, why don't you just drink the sample when you are finished? You've already got a free taste of sample. We all get a free-taste-sample working in this industry. Believe or not, I had some terrible experiences, and so might you. I've have been soaked head to toe with mixed liquor, but after a few dozen times, I got used to it. Wastewater samples will make you resistant to any known disease, well, you have to pay the price and get sick for a short period, but after the first 6 months, you can endure almost anything.

My advice is to know how to protect yourself and be careful. Get your hepatitis shots, wear PPE, and don't freak out with a spill or mishap. Always be prepared for the worst. Bring extra clothes to work and soap too.

Free-taste-sample is one of the less known benefits for working at a wastewater facility. Tasty for sure! Guaranteed to curl your toes, you'll never complain again about anything else in life.



## Total Dissolved Solids

Water is a good solvent and picks up impurities easily. Pure water is tasteless, colorless, and odorless and is often called the universal solvent. Dissolved solids refer to any minerals, salts, metals, cations or anions dissolved in water. Total dissolved solids (TDS) comprise inorganic salts (principally calcium, magnesium, potassium, sodium, bicarbonates, chlorides and sulfates) and some small amounts of organic matter that are dissolved in water.

TDS in drinking-water originate from natural sources, sewage, urban run-off, industrial wastewater, and chemicals used in the water treatment process, and the nature of the piping or hardware used to convey the water, i.e., the plumbing. In the United States, elevated TDS has been due to natural environmental features such as: mineral springs, carbonate deposits, salt deposits, and sea water intrusion, but other sources may include: salts used for road de-icing, anti-skid materials, drinking water treatment chemicals, stormwater and agricultural runoff, and point/non-point wastewater discharges.

In general, the total dissolved solids concentration is the sum of the cations (positively charged) and anions (negatively charged) ions in the water. Therefore, the total dissolved solids test provides a qualitative measure of the amount of dissolved ions, but does not tell us the nature or ion relationships.

In addition, the test does not provide us insight into the specific water quality issues, such as: Elevated Hardness, Salty Taste, or Corrosiveness. Therefore, the total dissolved solids test is used as an indicator test to determine the general quality of the water.

### Total Solids

The term "total solids" refers to matter suspended or dissolved in water or wastewater, and is related to both specific conductance and turbidity.

Total solids (also referred to as total residue) are the term used for material left in a container after evaporation and drying of a water sample. Total Solids includes both total suspended solids, the portion of total solids retained by a filter and total dissolved solids, the portion that passes through a filter (American Public Health Association, 1998).

Total solids can be measured by evaporating a water sample in a weighed dish, and then drying the residue in an oven at 103 to 105° C. The increase in weight of the dish represents the total solids. Instead of total solids, laboratories often measure total suspended solids and/or total dissolved solids.



### **Total Suspended Solids (TSS)**

Total Suspended Solids (TSS) are solids in water that can be trapped by a filter. TSS can include a wide variety of material, such as silt, decaying plant and animal matter, industrial wastes, and sewage. High concentrations of suspended solids can cause many problems for stream health and aquatic life.

High TSS can block light from reaching submerged vegetation. As the amount of light passing through the water is reduced, photosynthesis slows down. Reduced rates of photosynthesis causes less dissolved oxygen to be released into the water by plants. If light is completely blocked from bottom dwelling plants, the plants will stop producing oxygen and will die. As the plants are decomposed, bacteria will use up even more oxygen from the water. Low dissolved oxygen can lead to fish kills.



**Sampling downstream from a wastewater plant's discharge point.**

High TSS can also cause an increase in surface water temperature, because the suspended particles absorb heat from sunlight. This can cause dissolved oxygen levels to fall even further (because warmer waters can hold less DO), and can harm aquatic life in many other ways, as discussed in the temperature section. (The decrease in water clarity caused by TSS can affect the ability of fish to see and catch food.

Suspended sediment can also clog fish gills, reduce growth rates, decrease resistance to disease, and prevent egg and larval development. When suspended solids settle to the bottom of a water body, they can smother the eggs of fish and aquatic insects, as well as suffocate newly hatched insect larvae. Settling sediments can fill in spaces between rocks which could have been used by aquatic organisms for homes.

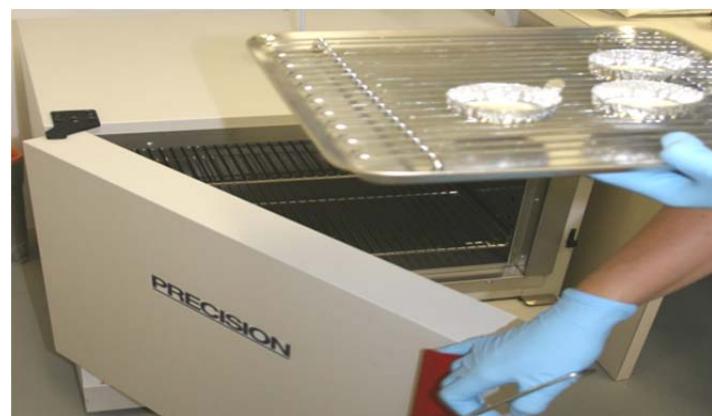


**Dead fish in lake using reclaimed water.**

High TSS in a water body can often mean higher concentrations of bacteria, nutrients, pesticides, and metals in the water. These pollutants may attach to sediment particles on the land and be carried into water bodies with storm water. In the water, the pollutants may be released from the sediment or travel farther downstream. High TSS can cause problems for industrial use, because the solids may clog or scour pipes and machinery.

### Measurement of Total Suspended Solids

To measure TSS, the water sample is filtered through a pre-weighed filter. The residue retained on the filter is dried in an oven at 103 to 105° C until the weight of the filter no longer changes. The increase in weight of the filter represents the total suspended solids. TSS can also be measured by analyzing for total solids and subtracting total dissolved solids.



**Total Dissolved Solids (TDS)** are solids in water that can pass through a filter (usually with a pore size of 0.45 micrometers). TDS is a measure of the amount of material dissolved in water. This material can include carbonate, bicarbonate, chloride, sulfate, phosphate, nitrate, calcium, magnesium, sodium, organic ions, and other ions. A certain level of these ions in water is necessary for aquatic life. Changes in TDS concentrations can be harmful because the density of the water determines the flow of water into and out of an organism's cells (Mitchell and Stapp, 1992). However, if TDS concentrations are too high or too low, the growth of many aquatic lives can be limited, and death may occur.

Similar to TSS, high concentrations of TDS may also reduce water clarity, contribute to a decrease in photosynthesis, combine with toxic compounds and heavy metals, and lead to an increase in water temperature. TDS is used to estimate the quality of drinking water, because it represents the amount of ions in the water. Water with high TDS often has a bad taste and/or high water hardness, and could result in a laxative effect.



The TDS concentration of a water sample can be estimated from specific conductance if a linear correlation between the two parameters is first established. Depending on the chemistry of the water, TDS (mg/l) can be estimated by multiplying specific conductance (micromhos/cm) by a factor between 0.55 and 0.75. TDS can also be determined by measuring individual ions and adding them up.



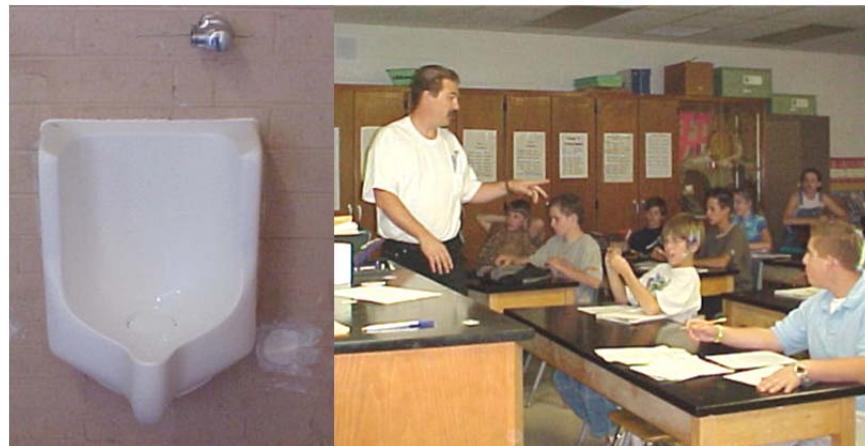
Conductivity Meter

## Flow

Whether a system serves a single home or an entire community, it must be able to handle fluctuations in the quantity and quality of wastewater it receives to ensure proper treatment is provided at all times. Systems that are inadequately designed or hydraulically overloaded may fail to provide treatment and allow the release of pollutants to the environment.

To design systems that are both as safe and as cost-effective as possible, engineers must estimate the average and maximum (peak) amount of flows generated by various sources. Because extreme fluctuations in flow can occur during different times of the day and on different days of the week, estimates are based on observations of the minimum and maximum amounts of water used on an hourly, daily, weekly, and seasonal basis. The possibility of instantaneous peak flow events that result from several or all water-using appliances or fixtures being used at once also is taken into account.

The number, type, and efficiency of all water-using fixtures and appliances at the source is factored into the estimate (for example, the number and amount of water normally used by faucets, toilets, and washing machines), as is the number of possible users or units that can affect the amount of water used (for example, the number of residents, bedrooms, customers, students, patients, seats, or meals served).



Waterless urinals are reducing water use but are concentrating the wastestream. Water conservation education is now taught at schools and this too is affecting our flow dynamics and MO's. Anything new always affects the bugs and no one cares but us.

According to studies, water use in many homes is lowest from about midnight to 5 a.m., averaging less than one gallon per person per hour, but then rises sharply in the morning around 6 am to a little over 3 gallons per person per hour. During the day, water use drops off moderately and rises again in the early evening hours. Weekly peak flows may occur in some homes on weekends, especially when all adults work during the week. In U.S. homes, average water use is approximately 45 gallons per person per day, but may range from 35 to 60 gallons or more.

Peak flows at stores and other businesses typically occur during business hours and during meal times at restaurants. Rental properties, resorts, and commercial establishments in tourist areas may have extreme flow variations seasonally. Estimating flow volumes for centralized treatment systems is a complicated task, especially when designing a new treatment plant in a community where one has never existed previously.

Engineers must allow for additional flows during wet weather due to inflow and infiltration of extra water into sewers. Excess water can enter sewers through leaky manhole covers and cracked pipes and pipe joints, diluting wastewater, which affects its overall characteristics. This can increase flows to treatment plants sometimes by as much as three or four times the original design load.



Grout is used to prevent infiltration into manholes.

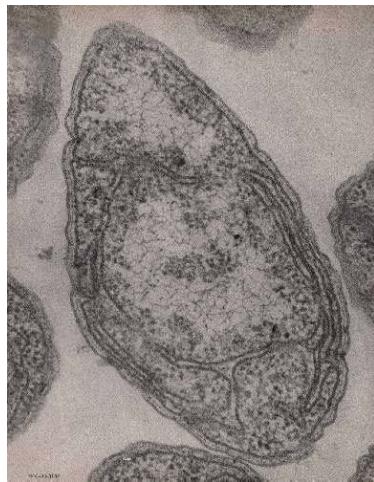
The main focus of wastewater treatment plants is to reduce the BOD and COD in the effluent discharged to natural waters, meeting state and federal discharge criteria. Wastewater treatment plants are designed to function as "microbiology farms," where bacteria and other microorganisms are fed oxygen and organic waste. Treatment of wastewater usually involves biological processes such as the activated sludge system in the secondary stage after preliminary screening to remove coarse particles and primary sedimentation that settles out suspended solids. These secondary treatment steps are generally considered environmental biotechnologies that harness natural self-purification processes contained in bioreactors for the biodegradation of organic matter and bioconversion of soluble nutrients in the wastewater.

#### **Application Specific Microbiology**

Each wastewater stream is unique, and so too are the community of microorganisms that process it. This "application-specific microbiology" is the preferred methodology in wastewater treatment affecting the efficiency of biological nutrient removal. The right laboratory prepared bugs are more efficient in organics removal if they have the right growth environment. This efficiency is multiplied if microorganisms are allowed to grow as a layer of biofilm on specifically designed support media. In this way, optimized biological processing of a waste stream can occur. To reduce the start-up phase for growing a mature biofilm one can also purchase "application specific bacterial cultures" from appropriate microbiology vendors.



Draining Biofilm



*Nitrosomonas europaea*

#### **Advanced Methods of Wastewater Treatment**

As our country and the demand for clean water have grown, it has become more important to produce cleaner wastewater effluents, yet some contaminants are more difficult to remove than others. The demand for cleaner discharges has been met through better and more complete methods of removing pollutants at wastewater treatment plants, in addition to pretreatment and pollution prevention which helps limit types of wastes discharged to the sanitary sewer system.

Currently, nearly all WWTPs provide a minimum of secondary treatment. In some receiving waters, the discharge of secondary treatment effluent would still degrade water quality and inhibit aquatic life. Further treatment is needed.



Discharge point from a wastewater plant into a wetlands project.

#### **Advanced Treatment Technologies**

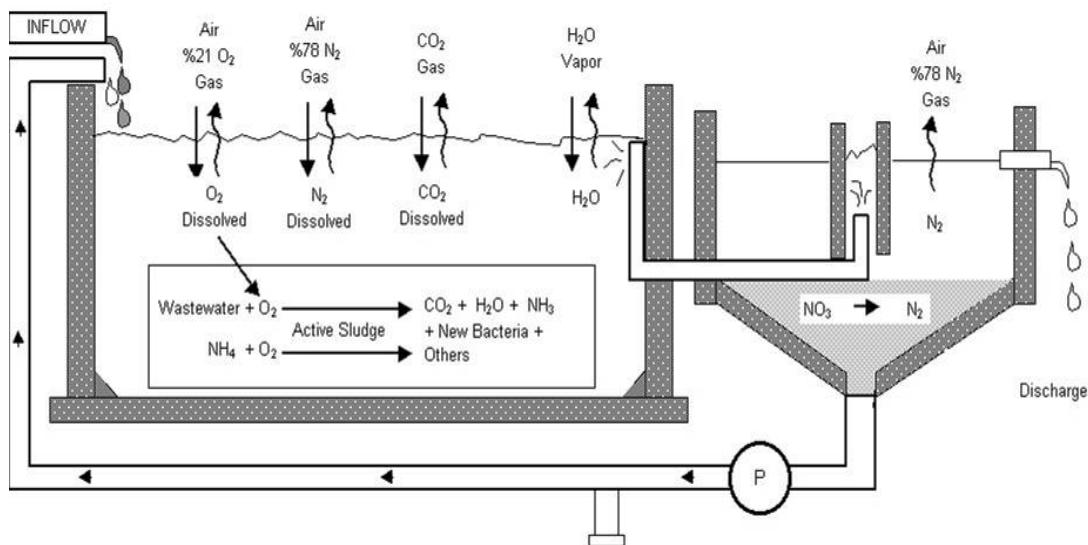
Treatment levels beyond secondary are called advanced treatment. Advanced treatment technologies can be extensions of conventional secondary biological treatment to further stabilize oxygen-demanding substances in the wastewater, or to remove nitrogen and phosphorus. Advanced treatment may also involve physical-chemical separation techniques such as adsorption, flocculation/precipitation, membranes for advanced filtration, ion exchange, and reverse osmosis. In various combinations, these processes can achieve any degree of pollution control desired. As wastewater is purified to higher and higher degrees by such advanced treatment processes, the treated effluents can be reused for urban, landscape, and agricultural irrigation, industrial cooling and processing, recreational uses and water recharge, and even indirect augmentation of drinking water supplies.

## Nitrogen Control

Nitrogen in one form or another is present in municipal wastewater and is usually not removed by secondary treatment. If discharged into lakes and streams or estuary waters, nitrogen in the form of ammonia can exert a direct demand on oxygen or stimulate the excessive growth of algae. Ammonia in wastewater effluent can be toxic to aquatic life in certain instances. By providing additional biological treatment beyond the secondary stage, nitrifying bacteria present in wastewater treatment can biologically convert ammonia to the non-toxic nitrate through a process known as nitrification. The nitrification process is normally sufficient to remove the toxicity associated with ammonia in the effluent. Since nitrate is also a nutrient, excess amounts can contribute to the uncontrolled growth of algae. In situations where nitrogen must be completely removed from effluent, additional biological process can be added to the system to convert the nitrate to nitrogen gas. We will cover this in much more detail in a few more pages.

### Conversion of Nitrate to Nitrogen Gas

The conversion of nitrate to nitrogen gas is accomplished by bacteria in a process known as denitrification. Effluent with nitrogen in the form of nitrate is placed into a tank devoid of oxygen, where carbon-containing chemicals, such as methanol, are added or a small stream of raw wastewater is mixed in with the nitrified effluent. In this oxygen free environment, bacteria use the oxygen attached to the nitrogen in the nitrate form, releasing nitrogen gas. Because nitrogen comprises almost 80 percent of the air in the earth's atmosphere, the release of nitrogen into the atmosphere does not cause any environmental harm.



### Biological Phosphorus Control

Like nitrogen, phosphorus is also a necessary nutrient for the growth of algae. Phosphorus reduction is often needed to prevent excessive algal growth before discharging effluent into lakes, reservoirs and estuaries. Phosphorus removal can be achieved through chemical addition and a coagulation-sedimentation process discussed in the following section. Some biological treatment processes called biological nutrient removal (BNR) can also achieve nutrient reduction, removing both nitrogen and phosphorus.

Most of the BNR processes involve modifications of suspended growth treatment systems so that the bacteria in these systems also convert nitrate nitrogen to inert nitrogen gas and trap phosphorus in the solids that are removed from the effluent.

### Coagulation-Sedimentation Process

A process known as chemical coagulation-sedimentation is used to increase the removal of solids from effluent after primary and secondary treatment. Solids heavier than water settle out of wastewater by gravity. With the addition of specific chemicals, solids can become heavier than water and will settle.

Alum, lime, or iron salts are chemicals added to the wastewater to remove phosphorus. With these chemicals, the smaller particles 'floc' or clump together into large masses. The larger masses of particles will settle faster when the effluent reaches the next step the sedimentation tank. This process can reduce the concentration of phosphate by more than 95 percent.

Although used for years in the treatment of industrial wastes and in water treatment, coagulation-sedimentation is considered an advanced process because it is not routinely applied to the treatment of municipal wastewater. In some cases, the process is used as a necessary pretreatment step for other advanced techniques. This process produces a chemical sludge, and the cost of disposing of this material can be significant.



### Carbon Adsorption

Carbon adsorption technology can remove organic materials from wastewater that resist removal by biological treatment. These resistant, trace organic substances can contribute to taste and odor problems in water, taint fish flesh, and cause foaming and fish kills. Carbon adsorption consists of passing the wastewater effluent through a bed or canister of activated carbon granules or powder which remove more than 98 percent of the trace organic substances. The substances adhere to the carbon surface and are removed from the water. To help reduce the cost of the procedure, the carbon granules can be cleaned by heating and used again.



Granular Carbon

### The Use or Disposal of Wastewater Residuals and Biosolids

When pollutants are removed from water, there is always something left over. It may be rags and sticks caught on the screens at the beginning of primary treatment. It may be the solids that settle to the bottom of sedimentation tanks. Whatever it is, there are always residuals that must be reused, burned, buried, or disposed of in some manner that does not harm the environment.

The utilization and disposal of the residual process solids is addressed by the CWA, Resource Conservation and Recovery Act (RCRA), and other federal laws. These Federal laws re-enforce the need to employ environmentally sound residuals management techniques and to beneficially use biosolids whenever possible. We will cover this in much more detail in a few more pages.

## Processed Wastewater Solids

Biosolids are processed wastewater solids ("sewage sludge") that meet rigorous standards allowing safe reuse for beneficial purposes. Currently, more than half of the biosolids produced by municipal wastewater treatment systems are applied to land as a soil conditioner or fertilizer and the remaining solids are incinerated or landfilled.



Large solids treatment facility

### **Ocean Dumping**

Ocean dumping of these solids is no longer allowed.

### **Biosolids Stabilization**

Prior to utilization or disposal, biosolids are stabilized to control odors and reduce the number of disease-causing organisms. Sewage solids, or sludge, when separated from the wastewater, still contain around 98 percent water. They are usually thickened and may be dewatered to reduce the volume to be transported for final processing, disposal, or beneficial use.

### **Dewatering Processes**

Dewatering processes include drying beds, belt filter presses, plate and frame presses, and centrifuges. To improve dewatering effectiveness, the solids can be pretreated with chemicals such as lime, ferric chloride, or polymers to produce larger particles which are easier to remove.



Centrifuge



Filter Press

### **Digestion**

Digestion is a form of stabilization where the volatile material in the wastewater solids can decompose naturally and the potential for odor production is reduced. Digestion without air in an enclosed tank (anaerobic solids digestion) has the added benefit of producing methane gas which can be recovered and used as a source of energy. Stabilization of solids may also be accomplished by composting, heat treatments, drying or the addition of lime or other alkaline materials. After stabilization, the biosolids can be safely spread on land.

### **Land Application**

In many areas, biosolids are marketed to farmers as fertilizer. Federal regulation (40 CFR Part 503) defines minimum requirements for such land application practices, including contaminant limits, field management practices, treatment requirements, monitoring, recordkeeping, and reporting requirements. Properly treated and applied biosolids are a good source of organic matter for improving soil structure and help supply nitrogen, phosphorus, and micronutrients that are required by plants.

Biosolids have also been used successfully for many years as a soil conditioner and fertilizer, and for restoring and re-vegetating areas with poor soils due to construction activities, strip mining or other practices. Under this biosolids management approach, treated solids in semi liquid or dewatered form are transported to the soil treatment areas. The slurry or dewatered biosolids, containing nutrients and stabilized organic matter, is spread over the land to give nature a hand in returning grass, trees, and flowers to barren land.

Restoration of the countryside also helps control the flow of acid drainage from mines that endangers fish and other aquatic life and contaminates the water with acid, salts, and excessive quantities of metals.

### **Incineration**

Incineration consists of burning the dried solids to reduce the organic residuals to an ash that can be disposed of or reused. Incinerators often include heat recovery features. Undigested sludge solids have significant fuel value as a result of their high organic content. However, the water content must be greatly reduced by dewatering or drying to take advantage of the fuel potential of the biosolids. For this reason, pressure filtration dewatering equipment is used to obtain biosolids which are sufficiently dry to burn without continual reliance on auxiliary fuels. In some cities, biosolids are mixed with refuse or refuse derived fuel prior to burning. Generally, waste heat is recovered to provide the greatest amount of energy efficiency.

### **Beneficial Use Products from Biosolids**

Heat dried biosolids pellets have been produced and used extensively as a fertilizer product for lawn care, turf production, citrus groves, and vegetable production for many years. Composting of biosolids is also a well-established approach to solids management that has been adopted by a number of communities. The composted peat-like product has shown particular promise for use in the production of soil additives for re-vegetation of topsoil depleted areas, and as a potting soil amendment.

Effective pretreatment of industrial wastes prevents excessive levels of unwanted constituents, such as heavy metals (i.e. cadmium, mercury, and lead) and persistent organic compounds from contaminating the residuals of wastewater treatment and limiting the potential for beneficial use.

Effective stabilization of wastewater residuals and their conversion to biosolid products can be costly. Some cities have produced fertilizers from biosolids which are sold to help pay part of the cost of treating wastewater. Some municipalities use composted, heat dried, or lime stabilized biosolid products on parks and other public areas.

### **Decentralized (Onsite and Cluster) Systems**

A decentralized wastewater system treats sewage from homes and businesses that are not connected to a centralized wastewater treatment plant. Decentralized treatment systems include onsite systems and cluster systems. An onsite system is a wastewater system relying on natural processes, although sometimes containing mechanical components, to collect, treat, disperse or reclaim wastewater from a single dwelling or building. A septic tank and soil adsorption field is an example of an onsite system.

A wastewater collection and treatment system under some form of common ownership that collects wastewater from two or more dwellings or buildings and conveys it to a treatment and dispersal system located on a suitable site near the dwellings or buildings is a cluster system.

Decentralized systems include those using alternative treatment technologies like media filters, constructed wetland systems, aerobic treatment units, and a variety of soil dispersal systems. Soil dispersal systems include pressure systems such as low pressure pipe and drip dispersal systems. These systems treat and disperse relatively small volumes of wastewater, and are generally found in rural and suburban areas.

While septic tanks and soil absorption systems have significant limitations, decentralized systems can effectively protect water quality and public health from groundwater and surface water contamination if managed properly (i.e. properly sited, sized, designed, installed, operated, and maintained). Nitrate concentrations in groundwater that exceed the drinking water standards can cause health problems.

### **Onsite Treatment**

Onsite wastewater systems contain three components: a treatment unit which treats water prior to dispersal into the environment; a soil dispersal component which assures that treated water is released into the environment at a rate which can be assimilated; and a management system which assures proper long term operation of the complete system.

Disinfection of the treated effluent may be provided prior to dispersal. A typical onsite system consists of a septic tank followed by an effluent distribution system. Alternative treatment systems include aerobic treatment and sand filtration systems. We will cover this in much more detail in a few more pages.

### **Conventional Septic Tanks**

A septic tank is a tank buried in the ground used to treat sewage without the presence of oxygen (anaerobic). The sewage flows from the plumbing in a home or small business establishment into the first of two chambers, where solids settle out. The liquid then flows into the second chamber. Anaerobic bacteria in the sewage break down the organic matter, allowing cleaner water to flow out of the second chamber. The liquid typically discharges through a subsurface distribution system. Periodically, the solid matter in the bottom of the tank, referred to as septage, must be removed and disposed of properly.

### **Aerobic Treatment Units**

Aerobic treatment units are also used to provide onsite wastewater treatment. They are similar to septic tanks, except that air is introduced and mixed with the wastewater inside the tank. Aerobic (requiring oxygen) bacteria consume the organic matter in the sewage. As with the typical septic system, the effluent discharge from an aerobic system is typically released through a sub-surface distribution system or may be disinfected and discharged directly to surface water. Aerobic treatment units also require the removal and proper disposal of solids that accumulate in the tank.

### **Media Filters**

Media filters are used to provide further treatment of septic tank effluent, and provide high levels of nitrification. They can be designed to pass the effluent once or multiple times through the media bed. Media, such as sand, acts as a filter. The media is placed two to three feet deep above a liner of impermeable material such as plastic or concrete. Septic tank effluent is applied to the filter surface in intermittent doses and is further treated as it slowly trickles through the media. In most media filters, wastewater is collected in an underdrain then either pumped back to the filter bed or to other types of treatment. We will cover this in much more detail in a few more pages.

### **Dispersal Approaches**

Traditional onsite systems include treatment units followed by a drainfield or absorption field. Wastewater from the treatment unit is dispersed through a suitable soil layer where it receives additional treatment by the soil microorganisms and filtering properties of the soil. If the soil is unsuitable for the installation of a soil absorption field, alternative methods can be used to further treat or distribute the treated effluent. The most common alternative dispersal systems include low-pressure pipe, mounds, drip disposal, and evapotranspiration beds.

#### **Absorption Field**

When soil conditions permit, the most common method to disperse septic tank or aerobic system effluent is an absorption field consisting of a series of perforated parallel pipes laid in trenches on gravel or crushed stone or as a direct discharge to the soil through trenches.

Typically, effluent flows into the absorption field from a distribution box which maintains an even flow of effluent to the absorption field. From there, the effluent drains through the stone and into the soil which provides further treatment.

#### **Mound System**

When the soil is not conducive to percolation or when the groundwater level is high, a mound system is commonly used. A mound system is a distribution system constructed above the original ground level by using granular material such as sand and gravel to receive the septic tank effluent before it flows to the native soil below. The effluent flows to a dosing tank that is equipped with a pump. Here the effluent is stored until there is sufficient liquid. Once the liquid is pumped out, it moves evenly throughout the mound before reaching less permeable soil or groundwater. The granular material acts as a treatment medium and improves the removal of pollutants in ways that may not be provided by substandard native soils.

#### **Drip Dispersal System**

Where soils are very thin or have reduced permeability, drip dispersal systems can be utilized. The typical drip system operates like drip irrigation at a moderately high pressure. The components of a drip system include filters to remove solids, a network of drip tubes to disperse liquid into soil, tanks to hold liquid, and controllers to regulate the flow to the drip system.

#### **Evapotranspiration Beds**

Evapotranspiration (ET) bed is an onsite dispersal system where pretreated wastewater evaporates from the soil surface or is transpired by plants into the atmosphere. Usually, ET beds are used in arid climates and there is no discharge either to surface or groundwater. Vegetation is planted on the surface of the sand bed to improve the transpiration process and landscaping enhances the aesthetics of the bed.

## Management of Decentralized Systems

Ensuring performance of decentralized wastewater treatment systems is an issue of national concern because these systems are a permanent component of our nation's wastewater infrastructure. Twenty five percent of households nationwide and one-third of the new homes being constructed are served by onsite systems. Many of the existing systems do not perform adequately due to a lack of management. Therefore, the EPA promotes the sustained management of decentralized wastewater systems to enhance their performance and reliability. The EPA strongly encourages communities to establish management programs for the maintenance of onsite systems in addition to improving local requirements for onsite system siting and system design.

Communities benefit from effective onsite system management programs by enjoying improved protection of public health and local surface water and groundwater resources, preserving rural areas, protecting property owners' investments through increased system service life, and avoiding the need to finance costly central wastewater collection and treatment systems.

### Dispose of Household Hazardous Wastes Safely

Many household products are potentially hazardous to people and the environment and never should be flushed down drains, toilets, or storm sewers. Treatment plant workers can be injured and wastewater systems can be damaged as a result of improper disposal of hazardous materials. Other hazardous chemicals cannot be treated effectively by municipal wastewater systems and may reach local drinking water sources. When flushed into septic systems and other onsite systems, they can temporarily disrupt the biological processes in the tank and soil absorption field, allowing hazardous chemicals and untreated wastewater to reach groundwater.

Some examples of hazardous household materials include motor oil, transmission fluid, antifreeze, paint, paint thinner, varnish, polish, wax, solvents, pesticides, rat poison, oven cleaner, and battery fluid. Many of these materials can be recycled or safely disposed of at community recycling centers.

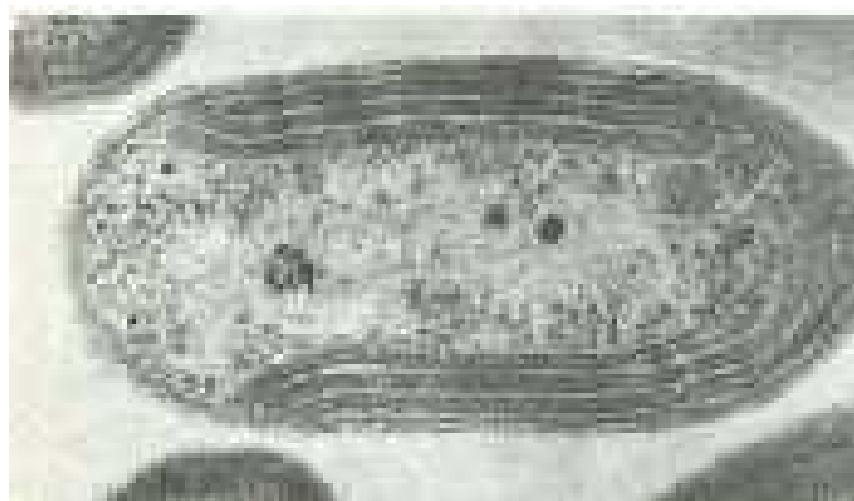


A drive-thru household hazardous waste collection site, trying to keep the toxic material out of the sewer system. These workers usually get to keep lots of goodies and take the materials home while at the same time keeping the bad stuff from upsetting the plant. I worked a day at one of these facilities and I was amazed with the chemicals that people keep around the home, example, 1 pound of liquid Mercury and another had a bottle of Sodium Cyanide.

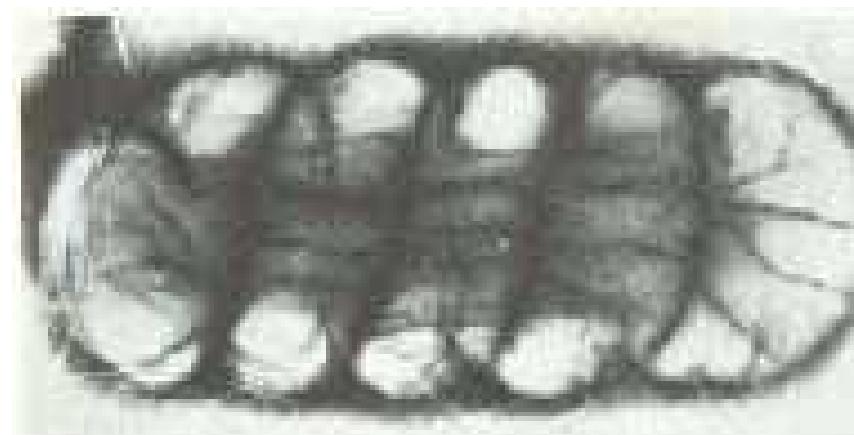


Top photograph, a clarifier's raking mechanism.  
Bottom, scum armature equipment.





*Nitrobacter winogradskyi*



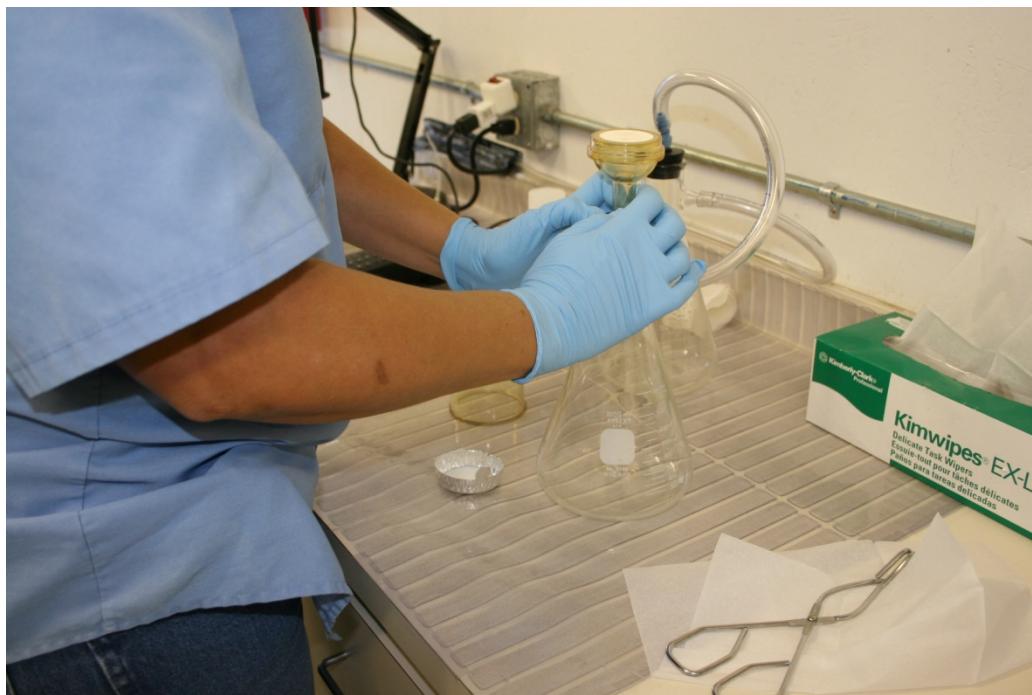
*Nitrospira gracilis*

Of all biological waste treatment methods, aerobic digestion is the most widespread process used throughout the world (more than 95%).

Nature gives, takes and does everything in-between. Nowhere is this better exemplified than the biological solution it offers to mankind's waste problems. An illustration of nature's work is its influence on the constant cycle of biological waste treatment. Microorganisms, like all living things, require food for growth.

Biological sewage treatment consists of many different microorganisms, mostly bacteria, carrying out a stepwise, continuous, sequential attack on the organic compounds found in wastewater and upon which the microbes feed.

Aerobic digestion of waste is the natural biological degradation and purification process in which bacteria that thrive in oxygen-rich environments break down and digest the waste. During this oxidation process, pollutants are broken down into carbon dioxide ( $\text{CO}_2$ ), water ( $\text{H}_2\text{O}$ ), nitrates, sulfates and biomass (micro-organisms). By optimizing the oxygen supply with so called aerators the process can be significantly accelerated.



Total dissolved solids - The weight per unit volume of all volatile and non-volatile solids dissolved in a water or wastewater after a sample has been filtered to remove colloidal and suspended solids.



## Water Quality Criteria

Many types of microscopic plants and animals, such as plankton, water beetles, and insects that live in or on the water, serve as food for small fish. Small fish are eaten by larger fish which, in turn, are consumed by even larger fish. These large fish may ultimately be consumed by humans. All life along the food chain is dependent on the water environment and it is for this reason that the quality of the nation's surface waters must be protected.

The Clean Water Act directs the EPA to develop criteria for water quality that accurately reflect the latest scientific knowledge about the effects of pollutants on aquatic life and human health. In developing these criteria, the EPA examines the effects of specific pollutants on plankton, fish, shellfish, wildlife, plant life, aesthetics, and recreation in any body of water. This includes specific information on the concentration and dispersal of pollutants through biological, physical, and chemical processes as well as the effects of pollutants on biological communities as a whole.

States may use the criteria that are developed by the EPA to help set water quality standards that protect the uses of their waters or they may develop their own water quality criteria. The EPA publishes human health and aquatic life criteria and is currently developing sediment and biological criteria. These criteria are complementary; each is designed to protect specific types of living organisms or ecological systems from the adverse effects of pollution.

### Human Health Criteria

People can potentially be exposed to water pollutants when they drink untreated surface water or eat fish, shellfish, or wildlife that have been contaminated by pollutants in surface waters. To reduce the risk to humans from these sources, the EPA scientists research information to determine the levels at which specific chemicals are not likely to adversely affect human health. The EPA publishes these levels as human health criteria that the states use, along with other information, to set allowable concentrations of pollutants in their water quality standards. In this way, the EPA and the states work together to protect people from exposure to harmful pollutants in surface waters.

For an in-depth look visit:

<http://epa.gov/waterscience/criteria/humanhealth/>

### Aquatic Life Criteria

Aquatic life criteria provide protection for plants and animals that are found in surface waters. The EPA develops these criteria as numeric limits on the amounts of chemicals that can be present in river, lake, or stream water without harm to aquatic life. Aquatic life criteria are designed to provide protection for both freshwater and saltwater aquatic organisms from the effects of acute (short term) and chronic (long term) exposure to potentially harmful chemicals. Aquatic life criteria are based on toxicity information and are developed to protect aquatic organisms from death, slower growth, reduced reproduction, and the accumulation of harmful levels of toxic chemicals in their tissues that may adversely affect consumers of such organisms.

For an in-depth look visit:

<http://epa.gov/waterscience/criteria/aqlife.html>

### Sediment Quality Criteria Guidance

In a healthy aquatic community, sediments provide a habitat for many living organisms. Worms, plants, and tiny microorganisms living in or on the sediment sustain the fish and shellfish that, in turn, nourish larger fish, wildlife, and man.

### **Pollutants in the Sediment**

Controlling the concentration of pollutants in the sediment helps to protect bottom dwelling species and prevents harmful toxins from moving up the food chain and accumulating in the tissue of animals at progressively higher levels. This is particularly important at the lower levels of the food chain because the concentration of many pollutants may increase at each link in the food chain. A pollutant level in the sediment that does not harm snails or small fish may bioaccumulate in the food chain and become very harmful to larger fish, birds, mammals, wildlife, and people.

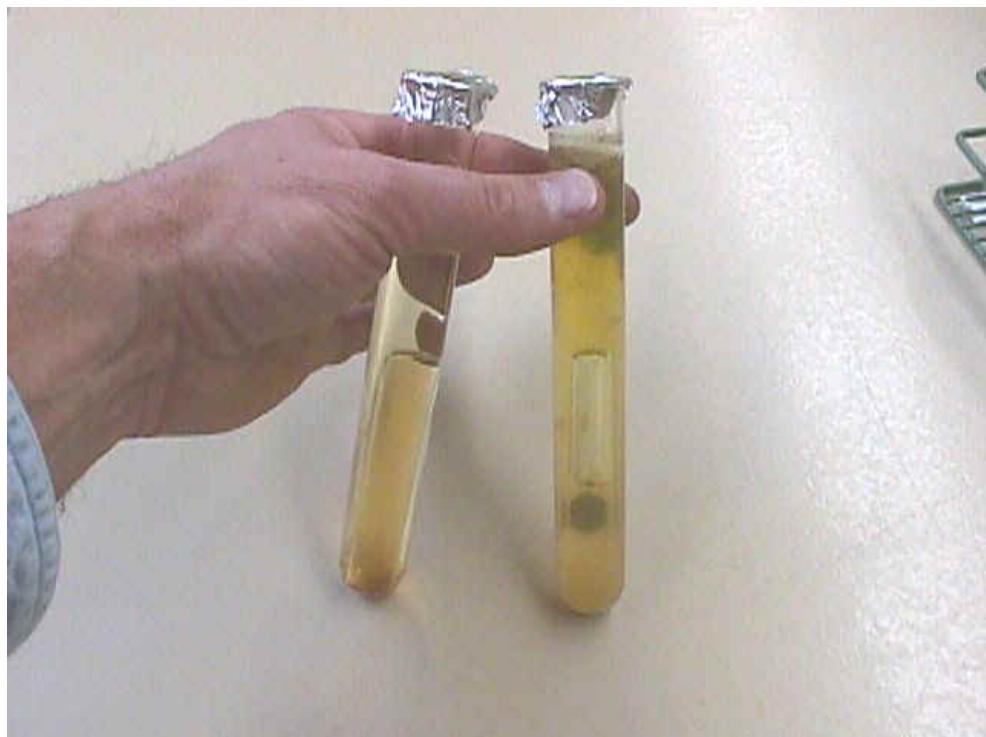
The EPA develops sediment quality criteria guidance on the concentrations or amounts of individual chemicals that can be present in river, lake, or stream sediments and still protect sediment-dwelling organisms and ultimately animals higher in the food chain from the harmful effects of toxic pollutants.

For an in-depth look visit:

<http://epa.gov/waterscience/criteria/sediment/>

### **Biological Criteria**

A water body in its natural condition is free from the harmful effects of pollution, habitat loss, and other negative stressors. It is characterized by a particular biological diversity and abundance of organisms. This biological integrity--or natural structure and function of aquatic life--can be dramatically different in various types of water bodies in different parts of the country. Because of this, the EPA is developing methodologies that states can use to assess the biological integrity of their waters and, in so doing, set protective water quality standards. These methodologies will describe scientific methods for determining a particular aquatic community's health and for maintaining optimal conditions in various bodies of water.



Standard Total Coliform Fermentation Technique

## **Summary**

The goal of all biological wastewater treatment systems is to remove the non-settling solids and the dissolved organic load from the effluents by using microbial populations. Biological treatments are generally part of secondary treatment systems. The microorganisms used are responsible for the degradation of the organic matter and the stabilization of organic wastes. With regard to the way in which they utilize oxygen, they can be classified into aerobic (require oxygen for their metabolism), anaerobic (grow in absence of oxygen) and facultative (can proliferate either in absence or presence of oxygen although using different metabolic processes). Most of the micro-organisms present in wastewater treatment systems use the organic content of the wastewater as an energy source to grow, and are thus classified as heterotrophes from a nutritional point of view. The population active in a biological wastewater treatment is mixed, complex and interrelated.

## **Genera**

By example, in a single aerobic system, members of the genera *Pseudomonas*, *Nocardia*, *Flavobacterium*, *Achromobacter* and *Zooglea* may be present, together with filamentous organisms (*Beggiaota* and *Spaerotilus* among others). In a well-functioning system, protozoas and rotifers are usually present and are useful in consuming dispersed bacteria or non-settling particles. More extensive description and treatment of the microbiology of wastewater treatment systems are given elsewhere (Stanier, 1976).

The organic load present is incorporated in part as biomass by the microbial populations, and almost all the rest is liberated as gas (carbon dioxide ( $\text{CO}_2$ ) if the treatment is aerobic, or carbon dioxide plus methane ( $\text{CH}_4$ ) if the process is anaerobic) and water. In fisheries wastewaters the non- biodegradable portion is very low.

Unless the cell mass formed during the biological treatment is removed from the wastewater (e.g., by sedimentation or other treatment described in the previous section), the treatment is largely incomplete, because the biomass itself will appear as organic load in the effluent and the only pollution reduction accomplished is that fraction liberated as gases.

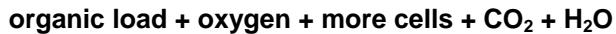
The biological treatment processes used for wastewater treatment are broadly classified as aerobic in which aerobic and facultative micro-organisms predominate or anaerobic which use anaerobic micro-organism.

If the microorganisms or Bugs are suspended in the wastewater during biological operation, the operations are "called suspended growth processes", while the micro-organisms that are attached to a surface over which they grow are called "attached growth processes".

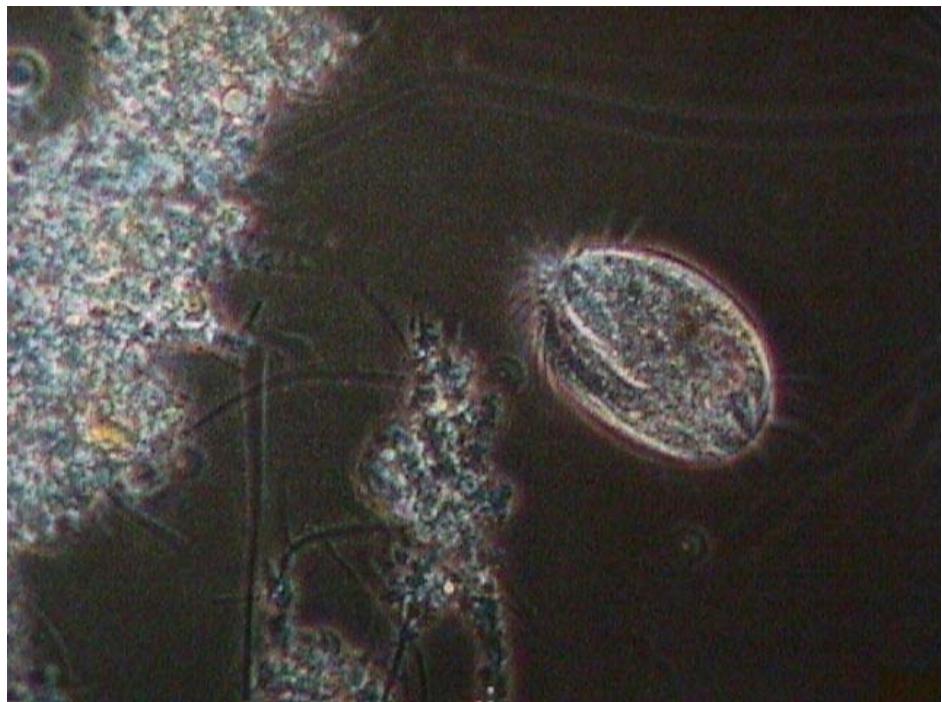
This section explains the principles and main characteristics of the most common processes in each case.

## **Aerobic Processes**

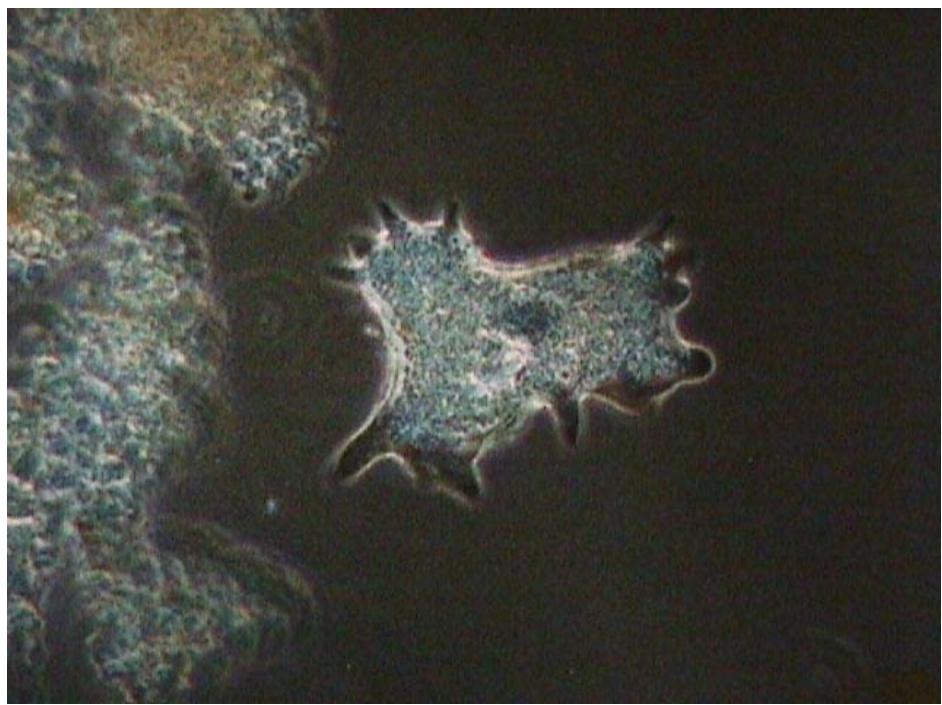
In these, the reactions occurring can be summarized as:



In fisheries wastewaters, the need for addition of nutrients (the most common being nitrogen and phosphorus) seldom appears, but an adequate provision of oxygen is essential for successful operation of the systems. The most common aerobic processes are: activated sludge systems, lagoons, trickling filters and rotating disk contactors. These aerobic processes are described, together with the devices used for aeration.



Ciliate above, Amoeba, below.



## The Microlife or the Microorganisms

We talked about the basic components and designs of wastewater treatment now let's look at the main "Team Players". Your process will respond to whatever direction you give it. You can run your plant (the team) to always try for the better or be content with the way it is. To get the best, it takes work!

Most activated sludge processes are used to degrade carbonaceous BOD. It is also possible to design and/or operate the basic system to oxidize ammonia (nitrification).

Many plants are now designed to achieve nitrification. Other system modifications include phosphorus removal and biological denitrification. Activated sludge plants are usually designed from pilot plant and laboratory studies.

From this approach, it is possible to design a process based on the amount of time the sludge spends in the system, generally termed mean cell residence time (MCRT), or on the amount of food provided to the bacteria in the aeration tank (the food-to-microorganism ratio, F/M). What does this mean?

Suppose a person ate 10 pounds of hot dogs (BOD) and weighed 200 pounds (MLSS).

What is the ratio of food to weight?

It would be 10 lbs. to 200 lbs. If we divide 200 into 10, the ratio is .05 or 5%.



Activated Sludge Aeration Basin, you can tell by the bubbles.

## F/M and MCRT

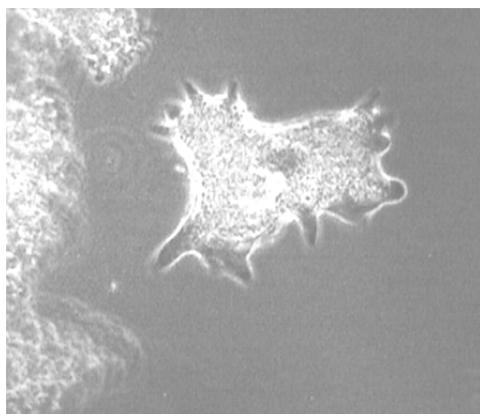
The following are some general statements about F/M and MCRT assuming that the environmental conditions are properly controlled.

- a. The optimum operating point of either helps obtain the desired effluent concentration.
- b. Both provide a means for maintaining the best effluent and sludge quality.
- c. Both techniques attempt to regulate rate of growth, metabolism, and stabilization of food matter.
- d. Both techniques indicate the solids level needed to stabilize the food and attain sludge quality.
- e. The desired solids level is controlled by wasting.
  - 1. To maintain – waste amount of net daily
  - 2. To increase – decrease waste rate
  - 3. To decrease – increase waste rate
- f. They are interrelated so changing one control changes the other.
- g. Once the control point is set, it should remain constant until change in effluent or sludge quality requires a change.

The operating control point is that point when the best effluent and sludge quality is obtained for the existing conditions.



Ciliate



Amoeba

## Microorganisms in Lagoons

Before we look at the bugs themselves, let's look at eating habits. Have you ever met a person who was a picky eater?

You have people who will put their noses up at some things and others who would eat anything. Predators typically eat from a narrow set of prey, while omnivores and scavengers eat from a broader food selection.

- Swimming and gliding ciliates engulf bacteria or other prey.
- Stalked ciliates attach to the biomass and vortex suspended bacteria into their gullets, while crawlers break bacteria loose from the floc surface.
- Predators feed mostly on stalked and swimming ciliates. The omnivores, such as most rotifers, eat whatever is readily available, while the worms feed on the floc or prey on larger organisms. Microorganisms are directly affected by their treatment environment.
- Changes in food, dissolved oxygen, temperature, pH, total dissolved solids, sludge age, presence of toxins, and other factors create a dynamic environment for the treatment organisms.

Food (organic loading) regulates microorganism numbers, diversity, and species when other factors are not limiting. The relative abundance and occurrence of organisms at different loadings can reveal why some organisms are present in large numbers while others are absent.

### Aerobic Bacteria

The aerobic bacteria that occur are similar to those found in other treatment processes such as activated sludge. Three functional groups occur: freely dispersed, single bacteria; floc-forming bacteria; and filamentous bacteria. All function similarly to oxidize organic carbon (BOD) to produce CO<sub>2</sub> and new bacteria (new sludge).

Many bacterial species that degrade wastes grow as single bacteria dispersed in the wastewater. Although these readily oxidize BOD, they do not settle and hence often leave the system in the effluent as solids (TSS). These tend to grow in lagoons at high organic loading and low oxygen conditions. More important are the floc-forming bacteria, those that grow in a large aggregate (floc) due to exocellular polymer production (the glycocalyx).

This growth form is important as these flocs degrade BOD and settle at the end of the process, producing a low TSS effluent.



A number of filamentous bacteria occur in lagoons, usually at specific growth environments. These generally do not cause any operational problems in lagoons, in contrast to activated sludge where filamentous bulking and poor sludge settling is a common problem.

Most heterotrophic bacteria have a wide range in environmental tolerance and can function effectively in BOD removal over a wide range in pH and temperature. Aerobic BOD removal generally proceeds well from pH 6.5 to 9.0 and at temperatures from 3-4°C to 60-70°C (mesophilic bacteria are replaced by thermophilic bacteria at temperatures above 35°C). BOD removal generally declines rapidly below 3-4°C and ceases at 1-2°C. A very specialized group of bacteria occurs to some extent in lagoons (and other wastewater treatment systems) that can oxidize ammonia via nitrite to nitrate, termed nitrifying bacteria. These bacteria are strict aerobes and require a redox potential of at least +200 mV (Holt et al., 1994).

## Aerated lagoons

The aerated lagoons are basins, normally excavated in earth and operated without solids recycling into the system. This is the major difference with respect to activated sludge systems. Two types are the most common: the completely mixed lagoon (also called completely suspended) in which the concentration of solids and dissolved oxygen are maintained fairly uniform and neither the incoming solids nor the biomass of microorganisms settle, and the facultative (aerobic-anaerobic or partially suspended) lagoons. In the facultative lagoons, the power input is reduced causing accumulation of solids in the bottom which undergo anaerobic decomposition, while the upper portions are maintained aerobic. The main operational difference between these lagoons is the power input, which is in the order of 2.5-6 Watts per cubic meter ( $\text{W/m}^3$ ) for aerobic lagoons while the requirements for facultative lagoons are of 0.8-1  $\text{W/m}^3$ . Being open to the atmosphere, the lagoons are exposed to low temperatures which can cause reduced biological activity and eventually the formation of ice. This can be partially alleviated by increasing the depth of the basin. These units require a secondary sedimentation unit, which in some cases can be a shallow basin excavated in earth, or conventional settling tanks can be used.

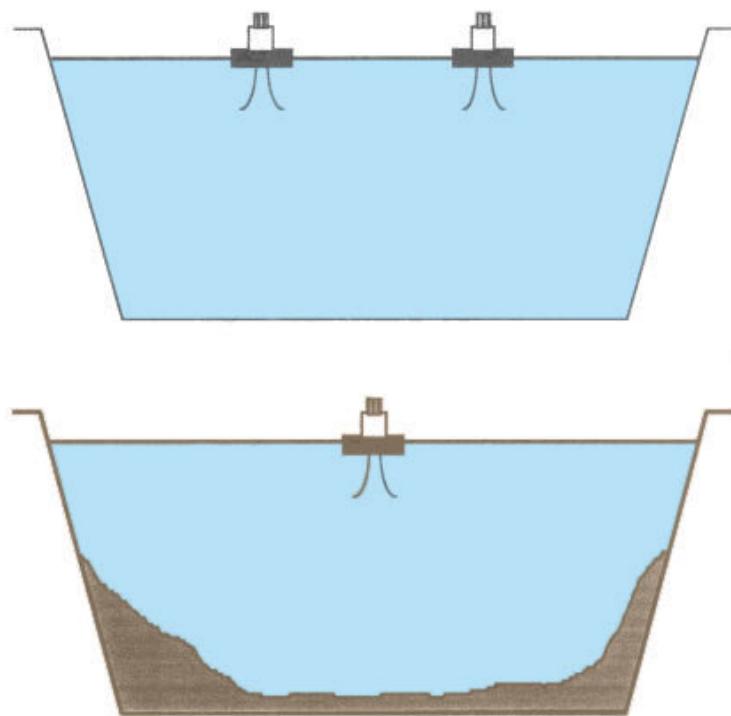


Diagram of aerobic (top) and facultative (bottom) aerated lagoons.

If excavated basins are used for settling, care should be taken to provide a residence time long enough for the solids to settle, and there should also be provision for the accumulation of sludge. There is a very high possibility of offensive odor development due to the decomposition of the settled sludge, and algae might develop in the upper layers contributing to an increased content of suspended solids in the effluent. Odors can be minimized by using minimum depths of up to 2 m, while algae production is reduced with liquid retention time of less than two days. The solids will also accumulate, all along the aeration basins in the facultative lagoons and even in corners, or between aeration units in the completely mixed lagoon. These accumulated solids will, on the whole, decompose in the bottom, but since there is always a non-biodegradable fraction, a permanent deposit will build up. Therefore, periodic removal of these accumulated solids becomes necessary. We will cover this in much more detail in a few more pages.

## Nitrification

It was once thought that only two bacteria were involved in nitrification: *Nitrosomonas europaea*, which oxidizes ammonia to nitrite, and *Nitrobacter winogradskyi*, which oxidizes nitrite to nitrate. It is now known that at least 5 genera of bacteria oxidize ammonia and at least three genera of bacteria oxidize nitrite (Holt et al., 1994). Besides oxygen, these nitrifying bacteria require a neutral pH (7-8) and substantial alkalinity (these autotrophs use CO<sub>2</sub> as a carbon source for growth). This indicates that complete nitrification would be expected at pond pH values between pH 7.0 and 8.5. Nitrification ceases at pH values above pH 9 and declines markedly at pH values below 7. This results from the growth inhibition of the nitrifying bacteria. Nitrification, however, is not a major pathway for nitrogen removal in lagoons. Nitrifying bacteria exists in low numbers in lagoons. They prefer attached growth systems and/or high MLSS sludge systems.

### Anaerobic Bacteria

Anaerobic, heterotrophic bacteria that commonly occur in lagoons are involved in methane formation (acid-forming and methane bacteria) and in sulfate reduction (sulfate reducing bacteria). Anaerobic methane formation involves three different groups of anaerobic bacteria that function together to convert organic materials to methane via a three-step process. General anaerobic degraders - many genera of anaerobic bacteria hydrolyze proteins, fats, and poly saccharides present in wastewater to amino acids, short-chain peptides, fatty acids, glycerol, and mono- and di-saccharides. These have a wide environmental tolerance in pH and temperature.

### Photosynthetic Organisms

Acid-forming bacteria - this diverse group of bacteria converts products from above under anaerobic conditions to simple alcohols and organic acids such as acetic, propionic, and butyric. These bacteria are hardy and occur over a wide pH and temperature range.

Methane forming bacteria - these bacteria convert formic acid, methanol, methylamine, and acetic acid under anaerobic conditions to methane. Methane is derived in part from these compounds and in part from CO<sub>2</sub> reduction. Methane bacteria are environmentally sensitive and have a narrow pH range of 6.5-7.5 and require temperatures > 14° C.

Note that the products of the acid formers (principally acetic acid) become the substrate for the methane producers. A problem exists at times where the acid formers overproduce organic acids, lowering the pH below where the methane bacteria can function (a pH < 6.5). This can stop methane formation and lead to a buildup of sludge in a lagoon with a low pH. In an anaerobic fermenter, this is called a "stuck digester". Also, methane fermentation ceases at cold temperature, probably not occurring in most lagoons in the wintertime in cold climates. A number of anaerobic bacteria (14 genera reported to date (Bolt et al., 1994)) called sulfate reducing bacteria can use sulfate as an electron acceptor, reducing sulfate to hydrogen sulfide.

This occurs when BOD and sulfate are present and oxygen is absent. Sulfate reduction is a major cause of odors in ponds. Anaerobic, photosynthetic bacteria occur in all lagoons and are the predominant photo-synthetic organisms in anaerobic lagoons. The anaerobic sulfur bacteria, generally grouped into the red and green sulfur bacteria and represented by about 28 genera (Ehrlich, 1990), oxidize reduced sulfur compounds (H<sub>2</sub>S) using light energy to produce sulfur and sulfate. Here, H<sub>2</sub>S is used in place of H<sub>2</sub>O as used by algae and green plants, producing SO<sub>4</sub><sup>-</sup> instead of O<sub>2</sub>. All are either strict anaerobes or microaerophilic. Most common are *Chromatium*, *Thiocystis*, and *Thiopedia*, which can grow in profusion and give a lagoon a pink or red color. Finding them is most often an indication of organic overloading and anaerobic conditions in an intended aerobic system.

Conversion of odorous sulfides to sulfur and sulfate by these sulfur bacteria is a significant odor control mechanism in facultative and anaerobic lagoons, and can be desirable.

## Major Algae Groups



Blue-green algae are the slimy stuff. Its cells lack nuclei and its pigment is scattered. Blue-green algae are not actually algae, they are bacteria.



Green algae cells have nuclei and the pigment is distinct. Green algae are the most common algae in ponds and can be multicellular.



Euglenoids are green or brown and swim with their flagellum, too. They are easy to spot because of their red eye. Euglenoids are microscopic and single celled.



Dinoflagellates have a flagella and can swim in open waters. They are microscopic and single celled.



Diatoms look like two shells that fit together. They are microscopic and single celled.

## Algae

Algae are aerobic organisms that are photosynthetic and grow with simple inorganic compounds  $\text{CO}_2$ ,  $\text{NH}_3$ ,  $\text{NO}_3$ , and  $\text{PO}_4$  using light as an energy source. (\*\*Note that algae produce oxygen during the daylight hours and consume oxygen at night.)

Algae are desirable in lagoons as they generate oxygen needed by bacteria for waste stabilization. Three major groups occur in lagoons, based on their chlorophyll type: brown algae (diatoms), green algae, and red algae. The predominant algal species at any given time is dependent on growth conditions, particularly temperature, organic loading, oxygen status, nutrient availability, and predation pressures. A fourth type of "algae" common in lagoons is the cyanobacteria or blue-green bacteria.

These organisms grow much as the true algae, with the exception that most species can fix atmospheric nitrogen. Blue-green bacteria often bloom in lagoons and some species produce odorous and toxic by-products.

### Blue-Green Bacteria

Blue-green bacteria appear to be favored by poor growth conditions including high temperature, low light, low nutrient availability (many fix nitrogen) and high predation pressure. Common blue-green bacteria in waste treatment systems include **Aphanothece, Microcystis, Oscillatoria and Anabaena**.

Algae can bloom in lagoons at any time of the year (even under the ice); however, a succession of algae types occurs over the season. There is also a shift in the algal species present in a lagoon through the season, caused by temperature and rotifer and Daphnia predation. Diatoms usually predominate in the wintertime at temperatures  $<60^{\circ}\text{F}$ . In the early spring, when predation is low and lagoon temperatures increase above  $60^{\circ}\text{F}$ , green algae such as Chlorella, Chlamydomonas, and Euglena often predominate in waste treatment lagoons. The predominant green algae change to species with spikes or horns such as Scenedesmus, Micractinium, and Ankistrodesmus later in the season when Rotifers and Daphnia are active (these species survive predation better).

Algae grow at warmer temperatures, longer detention time, and when inorganic minerals needed for growth are in excess. Alkalinity (inorganic carbon) is the only nutrient likely to be limiting for algal growth in lagoons. Substantial sludge accumulation in a lagoon may become soluble upon warming in the spring, releasing algal growth nutrients and causing an algal bloom. Sludge resolution of nutrients is a major cause of high algal growth in a lagoon, requiring sludge removal from the lagoon for correction.



Algae on the Secondary clarifier, this is not a good sign.

## Treatment Lagoon

The pH at a treatment lagoon is determined by the various chemical species of alkalinity that are present. The main species present are carbon dioxide ( $\text{CO}_2$ ), bicarbonate ion ( $\text{HCO}_3^-$ ), and carbonate ion ( $\text{CO}_3^{2-}$ ). Alkalinity and pH can affect which species will be present. High amounts of  $\text{CO}_2$  yield a low lagoon pH, while high amounts of  $\text{CO}_3^{2-}$  yield a high lagoon pH.

Bacterial growth on BOD releases  $\text{CO}_2$  which subsequently dissolves in water to yield carbonic acid ( $\text{H}_2\text{CO}_3$ ). This rapidly dissociates to bicarbonate ion, increasing the lagoon alkalinity. Bacterial oxidation of BOD causes a decrease in lagoon pH due to  $\text{CO}_2$  release.

Algal growth in lagoons has the opposite effect on lagoon pH, raising the pH due to algal use for growth of inorganic carbon ( $\text{CO}_2$  and  $\text{HCO}_3^-$ ). Algal growth reduces the lagoon alkalinity which may cause the pH to increase if the lagoon alkalinity (pH buffer capacity) is low. Algae can grow to such an extent in lagoons (a bloom) that they consume all of the  $\text{CO}_2$  and  $\text{HCO}_3^-$  present for photosynthesis, leaving only carbonate ( $\text{CO}_3^{2-}$ ) as the pH buffering species. This causes the pH of the lagoon to become alkaline. pH values of 9.5 or greater are common in lagoons during algal blooms, which can lead to lagoon effluent pH violations (in most states this is pH = 9). It should be noted that an increase in the lagoon pH caused by algal growth can be beneficial. Natural disinfection of pathogens is enhanced at higher pH.

Phosphorus removal by natural chemical precipitation is greatly enhanced at pH values greater than pH = 8.5. In addition, ammonia stripping to the atmosphere is enhanced at higher pH values ( $\text{NH}_3$  is strippable, not  $\text{NH}_4^+$ ).

### Protozoans and Microinvertebrates

Many higher life forms (animals) develop in lagoons. These include protozoans and microinvertebrates such as rotifers, daphnia, annelids, chironomids (midge larvae), and mosquito larvae (often termed the zooplankton). These organisms play a role in waste purification by feeding on bacteria and algae and promoting flocculation and settling of particulate material.

Protozoans are the most common higher life forms in lagoons with about 250 species identified in lagoons to date (Curds, 1992). Rotifers and daphnia are particularly important in controlling algal overgrowth and these often "bloom" when algal concentrations are high.

These microinvertebrates are relatively slow growing and generally only occur in systems with a detention time of >10 days. Mosquitoes grow in lagoons where shoreline vegetation is not removed, possibly causing a nuisance and public health problem.

*Culex tarsalis*, the vector of Western Equine Encephalitis in the western U.S., grows well in wastewater lagoons (USEPA, 1983). The requirement for a minimum lagoon bank slope and removal of shoreline vegetation by most regulatory agencies is based on the public health need to reduce mosquito vectors.



## Activated Sludge Methods

We have some wastewater treatment plants that grow the microorganisms (Bugs) in large tanks. To have enough oxygen in the tanks we add oxygen by blowing air into the tank full of wastewater and microorganisms. The air is bubbled in the water and mixes “the bugs,” food and oxygen together. When we treat wastewater this way, we call it the activated sludge method. With all of this food and air, the microbes grow and multiply very rapidly.

Pretty soon the population of bugs gets too large and some of them need to be removed to make room for new bugs to grow. We remove the excess bugs by sedimentation in the same kind of tanks used for primary treatment. In the tank, the bugs sink to the bottom and we remove them. The settled bugs are also called waste activated sludge. The waste sludge is treated separately, and the remaining wastewater is now much cleaner. In fact, after primary and secondary treatment, about 85% or more of all pollutants in the wastewater has been removed and it goes on to Disinfection. These systems originated in England in the early 1900's and earned their name because a sludge (mass of microbes) is produced which aerobically degrades and stabilizes the organic load of a wastewater. Below diagram shows the layout of a typical activated sludge system.

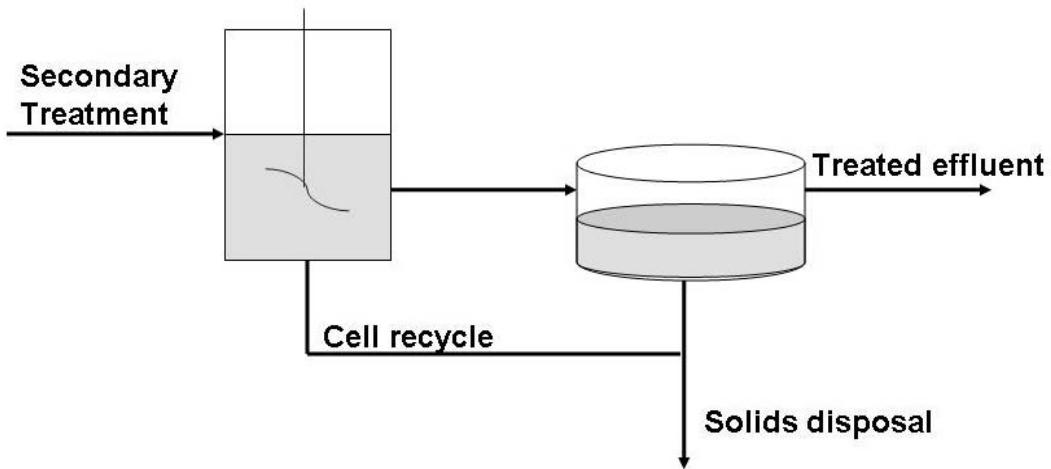


Diagram of a simple activated sludge system.

For larger systems, especially when high variability is expected, the design involves the use of multiple aeration tanks and multiple settling tanks. The number of units employed depends on the flow of wastewater being generated.

### Organic Load

The organic load (generally coming from primary treatment operations such as settling, screening or flotation) enters the reactor where the active microbial population (activated sludge) is present. The reactor must be continuously aerated. The mixture then passes to a secondary settling tank where the cells are settled. The treated wastewater is generally discharged after disinfection while the settled biomass is recycled in part to the aeration basin. The cells must be recycled in order to maintain sufficient biomass to degrade the organic load as quickly as possible. The amount that is recirculated depends on the need to obtain a high degradation rate and on the need for the bacteria to flocculate properly so that the secondary settling separates the cells satisfactorily. As the cells are retained longer in the system, the flocculating characteristics of the cells improve since they start to produce extra cellular slime which favors flocculating.

### Common Types

The most common types of activated sludge are the conventional and the continuous flow stiffer tank, in which the contents are completely mixed. In the conventional process, the wastewater is circulated along the aeration tank, with the flow being arranged by baffles in plug flow mode. The oxygen demand for this arrangement is maximum at the inlet as is the organic load concentration.

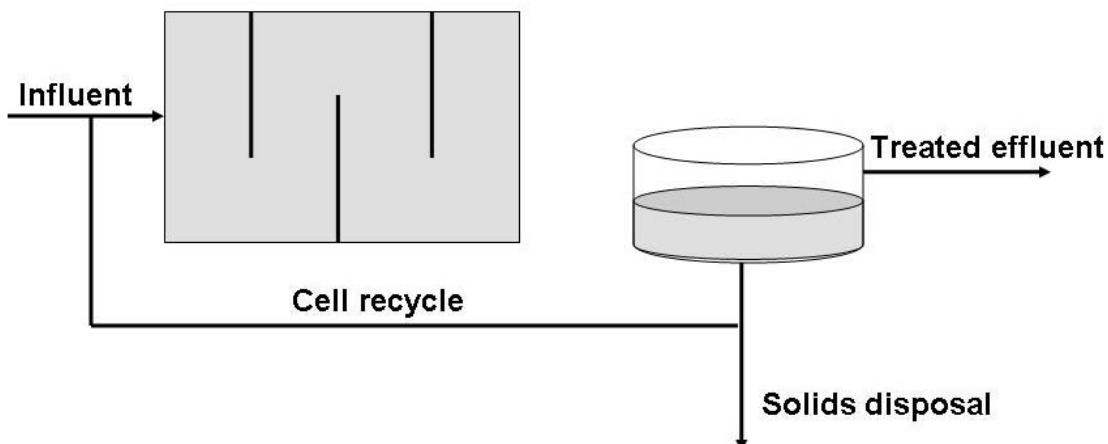


Diagram of a conventional activated sludge process.

In the completely mixed process the inflow streams are usually introduced at several points to facilitate the homogeneity of the mixing; if the mixing is complete, the properties are constant throughout the reactor. This configuration is inherently more stable to perturbations because mixing causes the dilution of the incoming stream into the tank. In fisheries wastewaters the perturbations that may appear are peaks of concentration of organic load or flow peaks. The flow peaks can be damped in the primary treatment tanks. The conventional configurations would require less reactor volume if smooth plug flow could be assured, which usually does not occur.

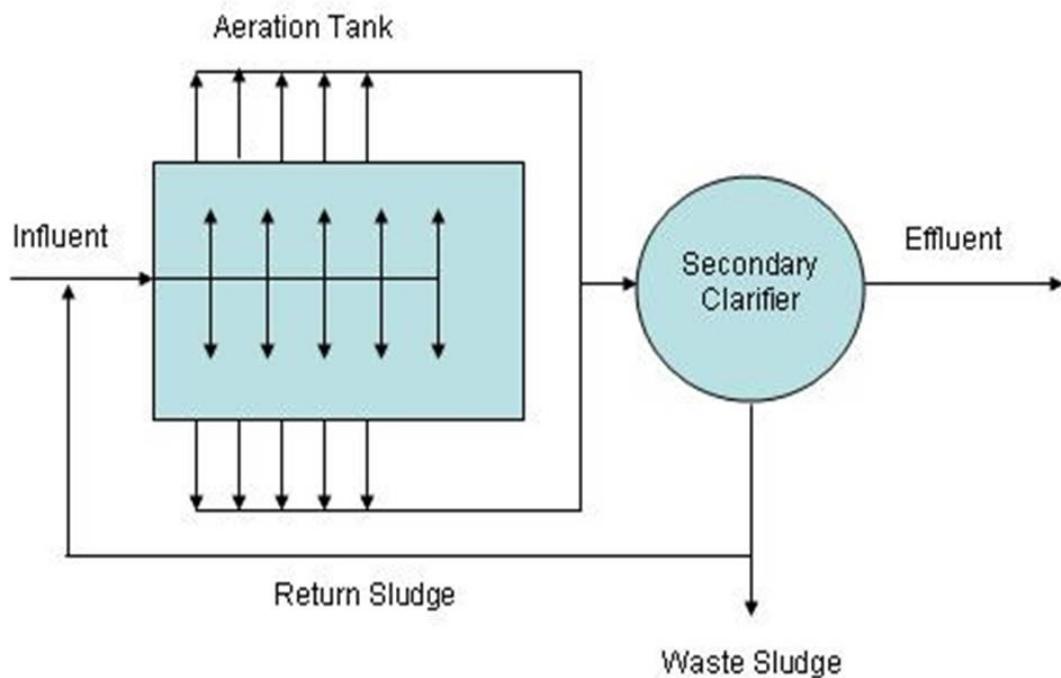
Other versions of activated sludge systems (e.g., extended aeration, contact stabilization, step aeration and pure oxygen processes) are used in other kinds of wastewaters but are not known to be applied to treat fisheries wastewaters. They are discussed elsewhere (Metcalf and Eddy Inc., 1979; Eckenfelder, 1980). In all activated sludge systems, the cells are separated from the liquid and partially returned to the system to have a relatively high concentration of cells that degrade the organic load in a relatively short time.

Therefore two different resident times are characteristic:

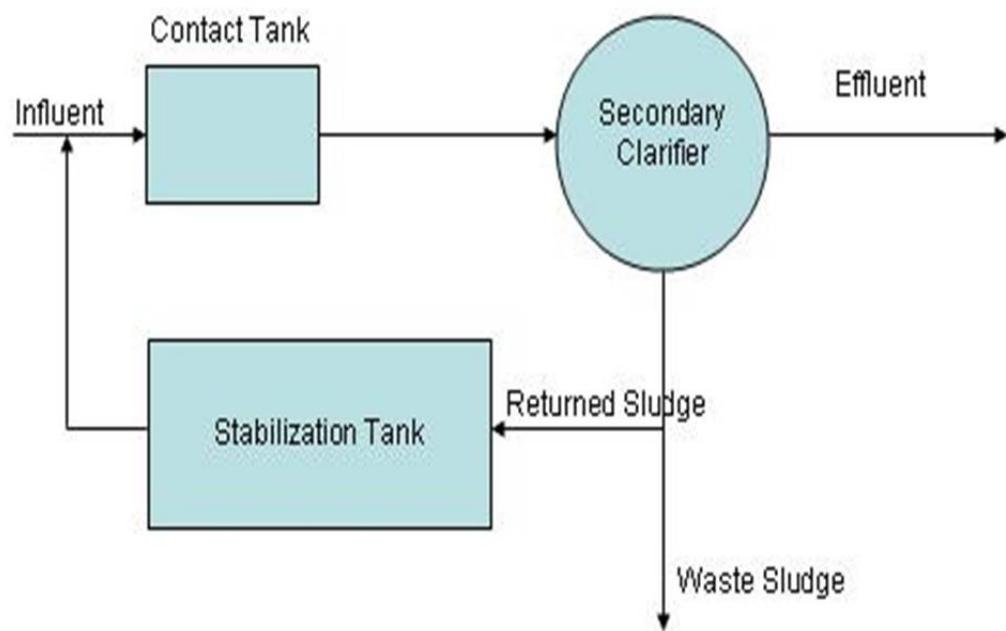
the hydraulic residence time ( $\theta_H$ ) given by the ratio of reactor volume (V) to flow of wastewater (Q):

$$\theta_H = V/Q$$

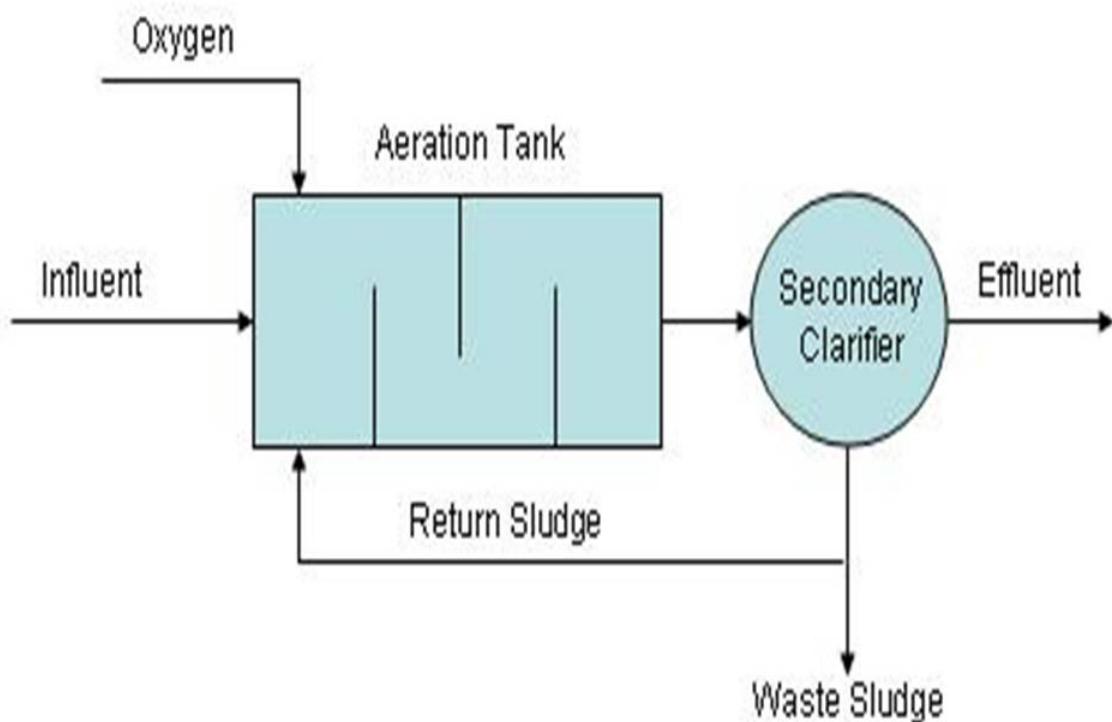
and the cell residence time ( $\theta_c$ ) given by the ratio of cells present in the reactor to the mass of cells wasted per day. Typical  $\theta_H$  values are in the order of 3-6 hours, while  $\theta_c$  fluctuates between 3 and 15 days. Such difference in resident times is obtained by discharging the clarified effluent but wasting only a small fraction of the sludge. This in turn can be accomplished by discarding a portion of the sludge from the settling tank or by wasting a fraction of the outlet of the reactor before entering the settling tank. In activated sludge systems, organic load removals of 85-95% are the most common. A key factor in the success of these systems is its proper operation, which requires trained manpower. Although used by some large fisheries which operate on a year-round basis, activated sludge may not prove to be economical or feasible for small seafood processors who operate seasonally because of the need to have a fairly constant supply of wastewater to maintain the micro-organisms.



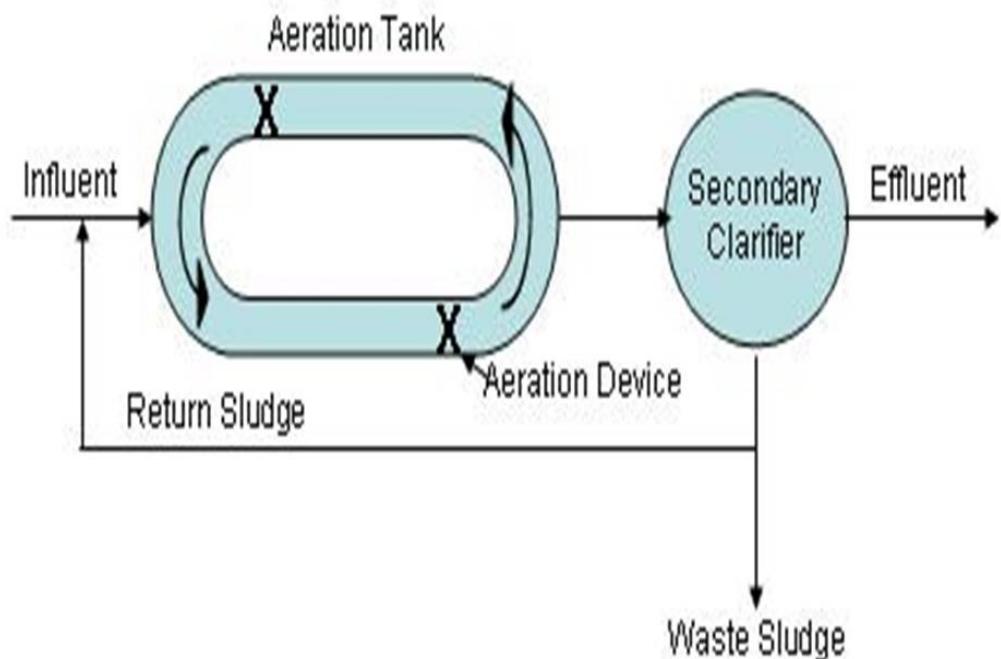
## Complete Mix Activated Sludge Process



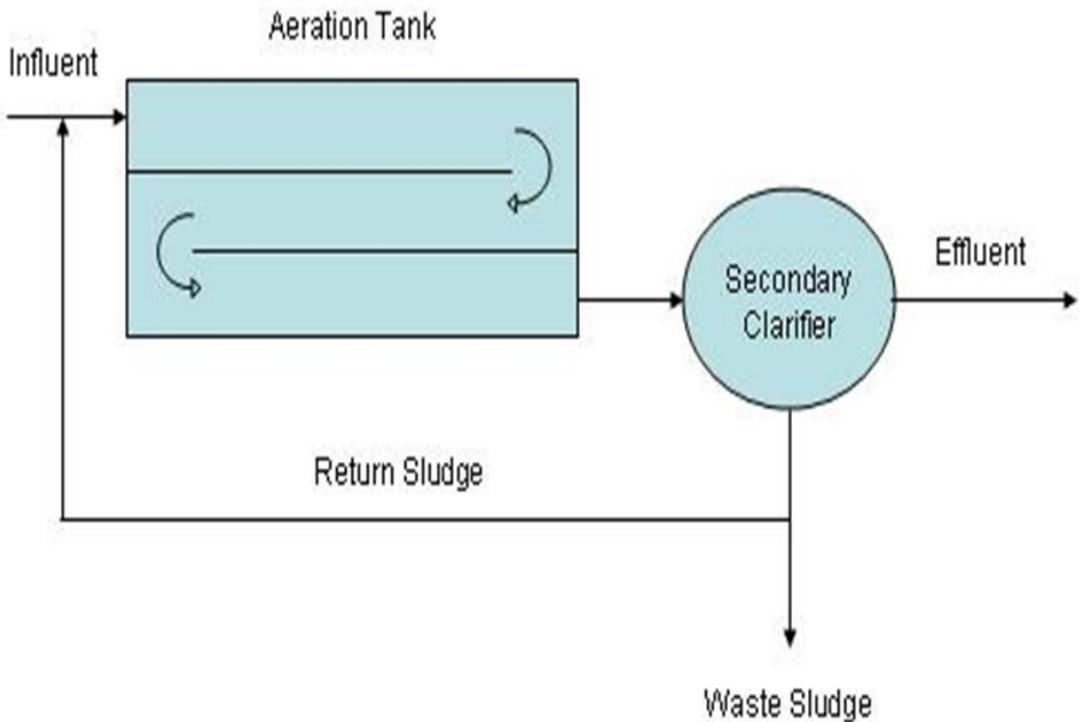
## Contact Stabilization Activated Sludge



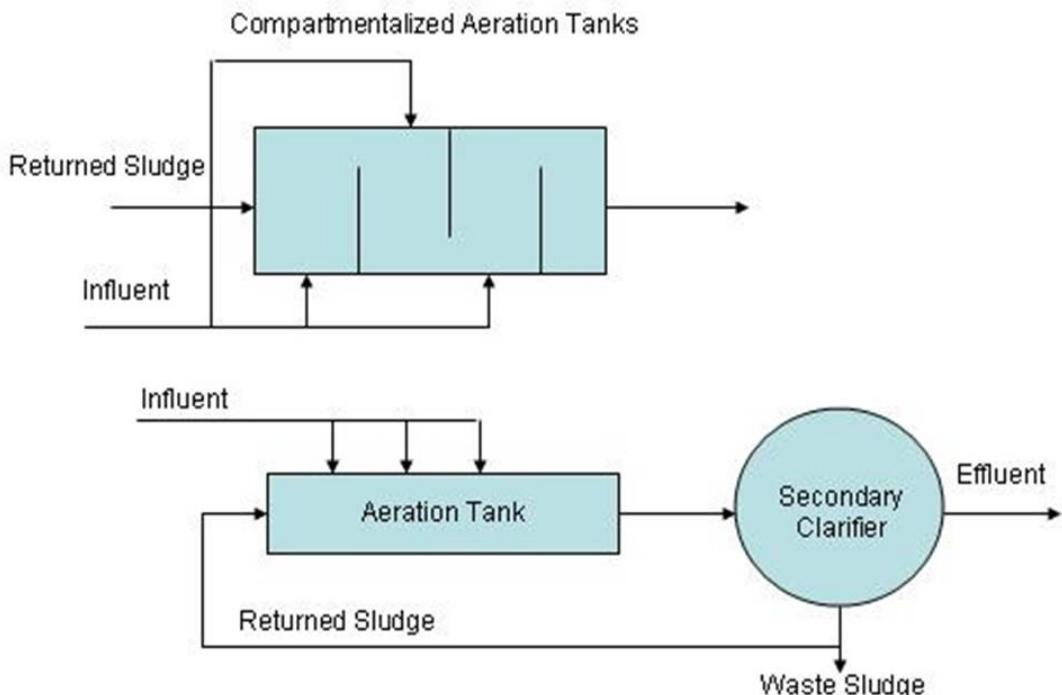
## High Purity Oxygen Activated Sludge



## Oxidation Ditch Activated Sludge Process



## Plug Flow Activated Sludge Process



## Step Feed Activated Sludge Process



Rectangular Clarifiers, notice the weirs are covered and protected from Sun light, the Sun helps the algae to grow on the weirs.



## Key Activated Sludge Word

**Amine:** A functional group consisting of "-NH<sub>2</sub>."

**Amino acid:** A functional group consisting of a carbon with a carboxylic acid, "-COOH" and an amine, "-NH<sub>2</sub>." These compounds are the building blocks for proteins.

**Anabolism:** Biosynthesis, the production of new cellular materials from other organic or inorganic chemicals.

**Anaerobes:** A group of organisms that do not require *molecular* oxygen. These organisms, as well as all known life forms, require oxygen. These organisms obtain their oxygen from inorganic ions such as nitrate or sulfate or from protein.

**Anaerobic process:** A process that only occurs in the absence of molecular oxygen.

**Anoxic process:** A process that occurs only at very low levels of molecular oxygen or in the absence of molecular oxygen.

**Biochemical oxygen demand (BOD):** The amount of oxygen required to oxidize any organic matter present in a water during a specified period of time, usually 5 days. It is an indirect measure of the amount of organic matter present in a water.

**Carbonaceous biochemical oxygen demand (CBOD):** The amount of oxygen required to oxidize any carbon containing matter present in a water.

**Chemical oxygen demand (COD):** The amount of oxygen required to oxidize any organic matter in the water using harsh chemical conditions.

**Decomposers:** Organisms that utilize energy from wastes or dead organisms. Decomposers complete the cycle by returning nutrients to the soil or water and carbon dioxide to the air or water.

**Denitrification:** The anoxic biological conversion of nitrate to nitrogen gas. It occurs naturally in surface waters low in oxygen, and it can be engineered in wastewater treatment systems.

**Deoxygenation:** The consumption of oxygen by the different aquatic organisms as they oxidize materials in the aquatic environment.

**Facultative:** A group of microorganisms which prefer or preferentially use molecular oxygen when available, but are capable of using other pathways for energy and synthesis if molecular oxygen is not available.

**F/M Ratio:** Another method for control wasting is to maintain a constant food-to-microorganism (F:M or F/M) ratio. With this method, the operator will try to increase or decrease the MLVSS to match an increase or decrease in the BOD entering the plant. Most plants will operate best at a specific F/M ratio between 0.05 - 0.1. If the optimum F/M has been determined from experience and can be maintained, a good effluent may be produced with consistent plant operation. The F/M ratio is to be calculated at least weekly and related to the efficiency of treatment plant operation. An F/M ratio between 0.05 - 0.15 BOD/lb MLSS is usually considered acceptable for an extended aeration process.

**Nitrification:** The biological oxidation of ammonia and ammonium sequentially to nitrite and then nitrate. It occurs naturally in surface waters, and can be engineered in wastewater treatment systems. The purpose of nitrification in wastewater treatment systems is a reduction in the oxygen demand resulting from the ammonia.

**Nitrogen fixation:** The conversion of atmospheric (or dissolved) nitrogen gas into nitrate by microorganisms.

**Nitrogenous oxygen demand (NOD):** The amount of oxygen required to oxidize any ammonia present in a water.

**NPDES:** The National Pollutant Discharge Elimination System. The discharge criteria and permitting system established by the U.S. EPA as a result of the Clean Water Act and its subsequent amendments or the permit required by each discharger as a result of the Clean Water Act.

**MCRT Mean Cell Residence Time:** The average time a given unit of cell mass stays in the activated sludge biological reactor. It is typically calculated as the total mixed liquor suspended solids in the biological reactor divided by the combination of solids in the effluent and solids wasted.

**Mixed liquor suspended solids (MLSS):** The total suspended solids concentration in the activated sludge tank.

**Mixed liquor volatile suspended solids (MLVSS):** The volatile suspended solids concentration in the activated sludge tank.

**Organic compound:** Any compound containing carbon except for the carbonates (carbon dioxide, the carbonates and bicarbonates), the cyanides, and cyanates.

**Organic nitrogen:** Nitrogen contained as amines in organic compounds such as amino acids and proteins.

**Oxidative phosphorylation:** The synthesis of the energy storage compound adenosine triphosphate (ATP) from adenosine diphosphate (ADP) using a chemical substrate and molecular oxygen.

**Secondary treatment:** In wastewater treatment, the conversion of the suspended, colloidal and dissolved organics remaining after primary treatment into a microbial mass which is then removed in a second sedimentation process. Secondary treatment includes both the biological process and the associated sedimentation process.

**Sludge:** A mixture of solid waste material and water. Sludges result from the concentration of contaminants in water and wastewater treatment processes. Typical wastewater sludges contain from 0.5 to 10 percent solid matter. Typical water treatment sludges contain 8 to 10 percent solids.

**Thiols:** Organic compounds which contain the "-SH" functional group. Also called mercaptans.

**Total dissolved solids:** (TDS) Is the amount of dissolved matter in a water.

**Total solids:** (TS) Is the amount of organic and inorganic matter that is contained in a water.

**Total suspended solids:** (TSS) Is the amount of suspended (filterable) matter in a water.

**Ultimate biochemical oxygen demand (BOD<sub>u</sub>):** The total amount of oxygen required to oxidize any organic matter present in a water, i.e. after an extended period, such as 20 or 30 days.

**Virus:** A submicroscopic genetic constituent that can alternate between two distinct phases. As a virus particle, or virion, it is DNA or RNA enveloped in an organic capsule. As an intracellular virus, it is viral DNA or RNA inserted into the host organisms DNA or RNA.

**Volatile:** A material that will vaporize easily.

**Volatile solids (VS)** is the amount of matter which volatilizes (or burns) when a water sample is heated to 550°C.





Gravity belt thickeners are often used to remove excess water from sludge.



Dry polymer is being added and used for sludge thickening.

## Bugs or MOs

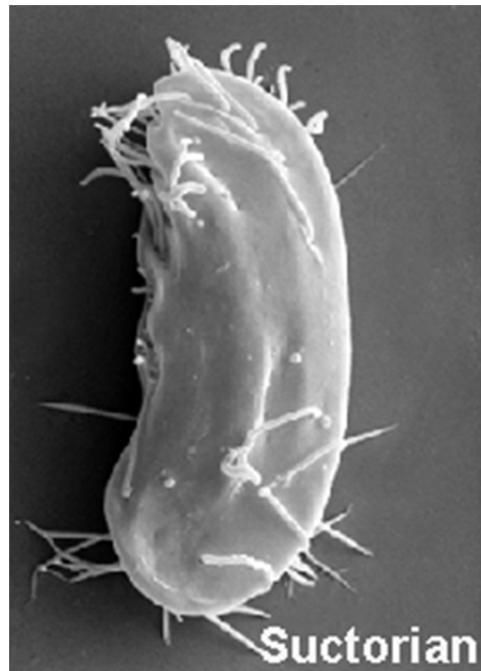
Four groups of bugs do most of the “eating” in the activated sludge process. The first group is the bacteria which eat the dissolved organic compounds. The second and third groups of bugs are microorganisms known as the free-swimming and stalked ciliates. These larger bugs eat the bacteria and are heavy enough to settle by gravity. The fourth group is a microorganism, known as Suctor, which feeds on the larger bugs and assists with settling.

The interesting thing about the bacteria that eat the dissolved organics is they have no mouths. The bacteria have an interesting property, their “fat reserves” are stored on the outside of their bodies. This fat layer is sticky and is what the organics adhere to.

Once the bacteria have “contacted” their food, they start the digestion process. A chemical enzyme is sent out through the cell wall to break up the organic compounds. This enzyme, known as hydrolytic enzyme, breaks the organic molecules into small units which are able to pass through the cell wall of the bacteria.

In wastewater treatment, this process of using bacteria-eating bugs in the presence of oxygen to reduce the organics in water is called activated sludge. The first step in the process, the contact of the bacteria with the organic compounds, takes about 20 minutes. The second step is the breaking up, ingestion and digestion processes, which takes four to 24 hours.

The fat storage property of the bacteria is also an asset in settling. As the bugs “bump” into each other, the fat on each of them sticks together and causes flocculation of the non-organic solids and biomass. From the aeration tank, the wastewater, now called mixed liquor, flows to a secondary clarification basin to allow the flocculated biomass of solids to settle out of the water. The solids biomass, which is the activated sludge, contains millions of bacteria and other microorganisms, is used again by returning it to the influent of the aeration tank for mixing with the primary effluent and ample amounts of air.



**Suctorian**



### **Paramecium sp.**

Paramecium is a medium to large size (100-300  $\mu\text{m}$ ) swimming ciliate, commonly observed in activated sludge, sometimes in abundant numbers. The body is either foot-shaped or cigar-shaped, and somewhat flexible. Paramecium is uniformly ciliated over the entire body surface with longer cilia tufts at the rear of the cell. Paramecium swims with a smooth gliding motion. It may also be seen paired up with another Paramecium which makes a good diagnostic key. The cell has either one or two large water cavities which are also identification tools. This swimmer moves freely in the water column as it engulfs suspended bacteria. It has a large feeding groove used to trap bacteria and form the food cavities that move throughout the body as digestion occurs. Paramecium is described as a filter-feeding ciliate because its cilia move and filter bacteria from the water.



### **Vorticella sp.**

Vorticella is a stalked ciliate. There are at least a dozen species found in activated sludge ranging in length from about 30 to 150  $\mu\text{m}$ . These organisms are oval to round shaped, have a contractile stalk, a domed feeding zone, and a water vacuole located near the terminal end of the feeding cavity. One organism is found on each stalk except during cell division. After reproducing, the offspring develops a band of swimming cilia and goes off to form its own stalk. The evicted organism is called a **"swarmer."**



Vorticella feeds by producing a vortex with its feeding cilia. The vortex draws bacteria into its gullet. Vorticella's principal food source is suspended bacteria.

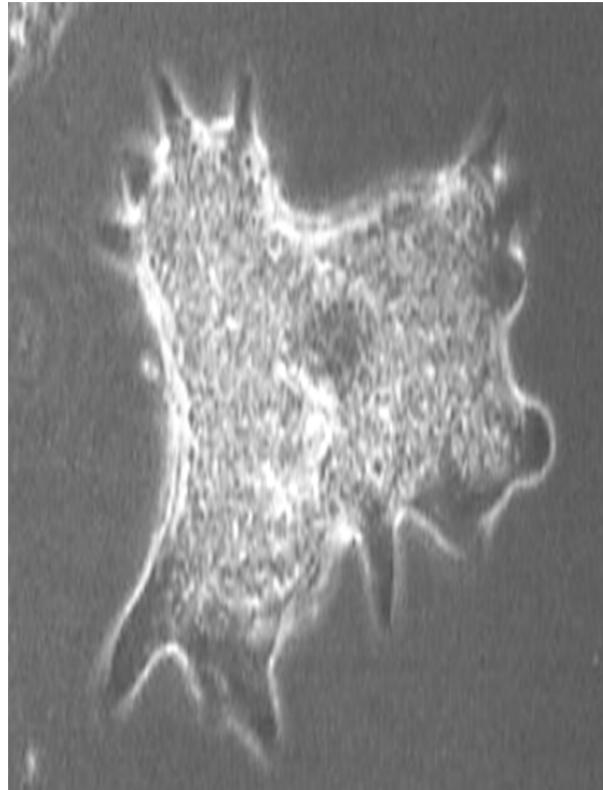
The contracting stalk provides some mobility to help the organism capture bacteria and avoid predators.

The stalk resembles a coiled spring after its rapid contraction. Indicator: If treatment conditions are bad, for example low DO or toxicity, Vorticella will leave their stalks. Therefore, a bunch of empty stalks indicates poor conditions in an activated sludge system. Vorticella sp. are present when the plant effluent quality is high.

**Euglypha sp.**

Euglypha (70-100  $\mu\text{m}$ ) is a shelled (testate) amoeba. Amoebas have jelly-like bodies. Motion occurs by extending a portion of the body (pseudopodia) outward. Shelled amoebas have a rigid covering which is either secreted or built from sand grains or other extraneous materials. The secreted shell of this Euglypha sp. consists of about 150 oval plates. Its spines project backward from the lower half of the shell. Euglypha spines may be single or in groups of two or three. The shell has an opening surrounded by 8-11 plates that resemble shark teeth under very high magnification.

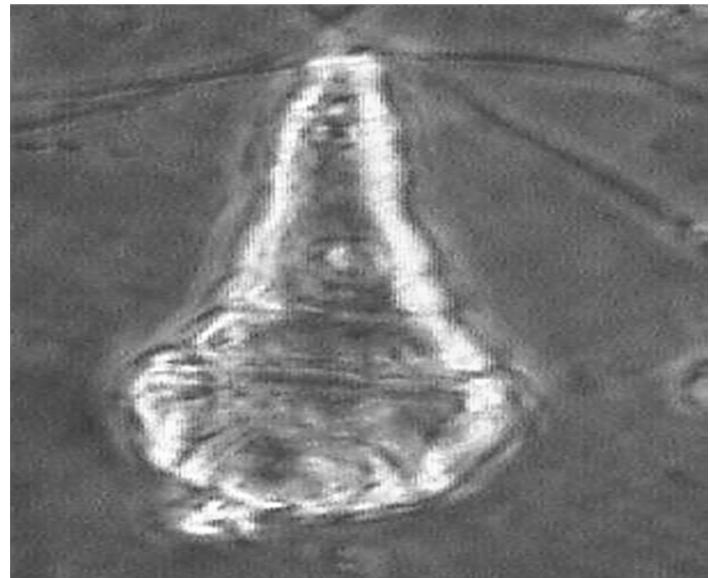
The shell of Euglypha is often transparent, allowing the hyaline (watery) body to be seen inside the shell. The pseudopodia extend outward in long, thin, rays when feeding or moving. Euglypha primarily eats bacteria. Indicator: Shelled amoebas are common in soil, treatment plants, and stream bottoms where decaying organic matter is present. They adapt to a wide range of conditions and therefore are not good indicator organisms.

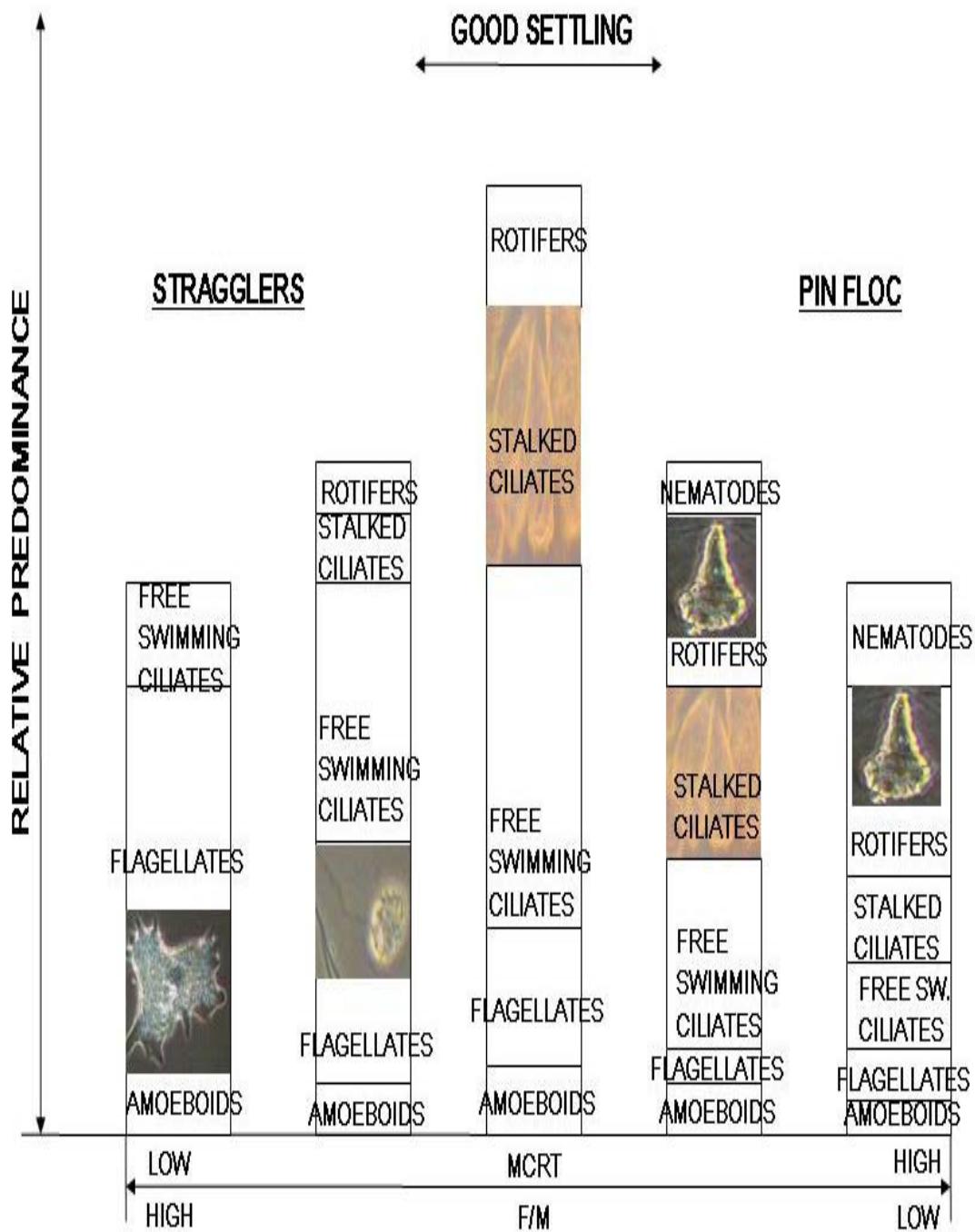
**Euchlanis sp.**

This microscopic animal is a typical rotifer. Euchlanis is a swimmer, using its foot and cilia for locomotion. In common with other rotifers, it has a head rimmed with cilia, a transparent body, and a foot with two strong swimming toes.

The head area, called the "corona," has cilia that beat rhythmically, producing a strong current for feeding or swimming. Euchlanis is an omnivore, meaning that its varied diet includes detritus, bacteria, and small protozoa. Euchlanis has a glassy shell secreted by its outer skin. The transparent body reveals the brain, stomach, intestines, bladder, and reproductive organs.

A characteristic of rotifers is their mastax, which is a jaw-like device that grinds food as it enters the stomach. At times the action of the mastax resembles the pulsing action of a heart. Rotifers, however, have no circulatory system. Indicator: Euchlanis is commonly found in activated sludge when effluent quality is good. It requires a continual supply of dissolved oxygen, evidence that aerobic conditions have been sustained.

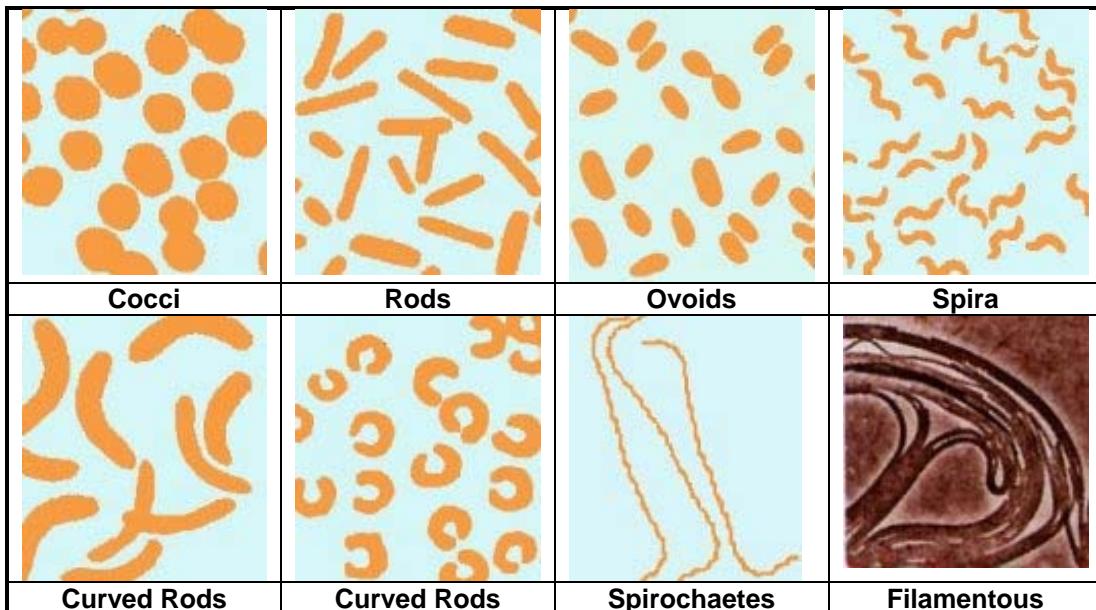




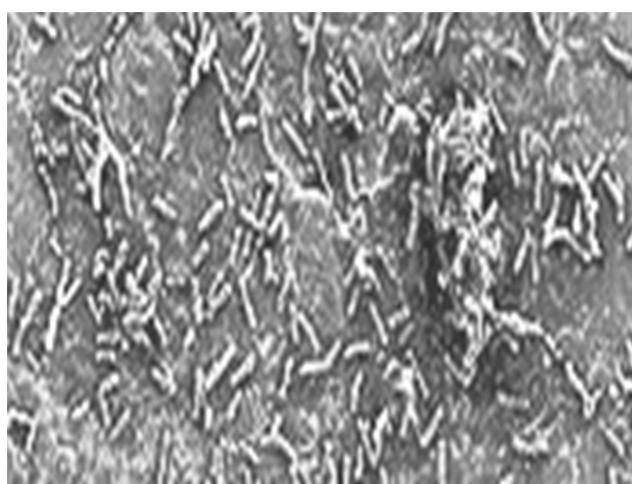
Wastewater Treatment Microlife

## Bacteria Section

Bacteria are one of the most ancient of living things and scientists believe they have been on this planet for nearly 4,000 million years. During this time they have acquired lots of fascinating and different ways of living. They also come in a variety of shapes. The simplest shape is a round sphere or ball. Bacteria formed like this are called cocci (singular coccus). The next simplest shape is cylindrical. Cylindrical bacteria are called rods (singular rod). Some bacteria are basically rods but instead of being straight they are twisted, bent or curved, sometimes in a spiral. These bacteria are called spirilla (singular spirillum). Spirochaetes are tightly coiled up bacteria.



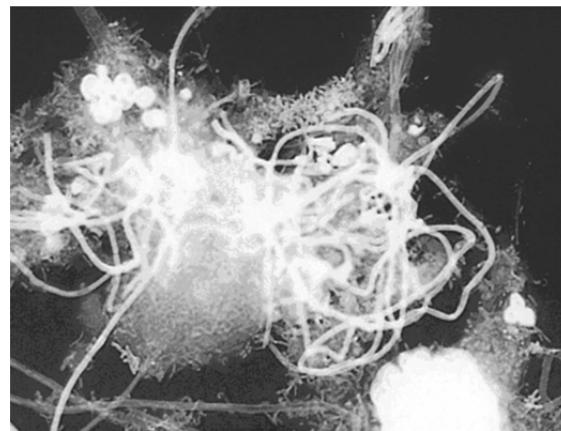
Bacteria are friendly creatures; you never find one bacteria on its own. They tend to live together in clumps, chains or planes. When they live in chains, one after the other, they are called filamentous bacteria - these often have long thin cells. When they tend to collect in a plane or a thin layer over the surface of an object, they are called a biofilm. Many bacteria exist as a biofilm and the study of biofilms is very important. Biofilm bacteria secrete sticky substances that form a sort of gel in which they live. The plaque on your teeth that causes tooth decay is a biofilm.



### **Filamentous Bacteria**

Filamentous Bacteria are a type of bacteria that can be found in a wastewater treatment system.

They function similar to floc forming bacteria since they degrade BOD quite well. In small amounts, they are quite good to a biomass. They can add stability and a backbone to the floc structure that keeps the floc from breaking up or shearing due to turbulence from pumps, aeration or transfer of the water. In large amounts they can cause many problems. Filaments are bacteria and fungi that grow in long thread-like strands or colonies.



### **Site Specific Bacteria**

Aeration and biofilm building are the key operational parameters that contribute to the efficient degradation of organic matter (BOD/COD removal). Over time, the application-specific bacteria become site-specific as the biofilm develops and matures and is even more efficient in treating the site-specific waste stream.

### **Facultative Bacteria**

Most of the bacteria absorbing the organic material in a wastewater treatment system are facultative in nature. This means they are adaptable to survive and multiply in either anaerobic or aerobic conditions. The nature of individual bacteria is dependent upon the environment in which they live. Usually, facultative bacteria will be anaerobic unless there is some type of mechanical or biochemical process used to add oxygen to the wastewater. When bacteria are in the process of being transferred from one environment to another, the metamorphosis from anaerobic to aerobic state (and vice versa) takes place within a couple of hours.

### **Anaerobic Bacteria**

Anaerobic bacteria live and reproduce in the absence of free oxygen. They utilize compounds such as sulfates and nitrates for energy and their metabolism is substantially reduced. In order to remove a given amount of organic material in an anaerobic treatment system, the organic material must be exposed to a significantly higher quantity of bacteria and/or detained for a much longer period of time. A typical use for anaerobic bacteria would be in a septic tank. The slower metabolism of the anaerobic bacteria dictates that the wastewater be held several days in order to achieve even a nominal 50% reduction in organic material. That is why septic tanks are always followed by some type of effluent treatment and disposal process. The advantage of using the anaerobic process is that electromechanical equipment is not required. Anaerobic bacteria release hydrogen sulfide as well as methane gas, both of which can create hazardous conditions. Even as the anaerobic action begins in the collection lines of a sewer system, deadly hydrogen sulfide or explosive methane gas can accumulate and be life threatening.

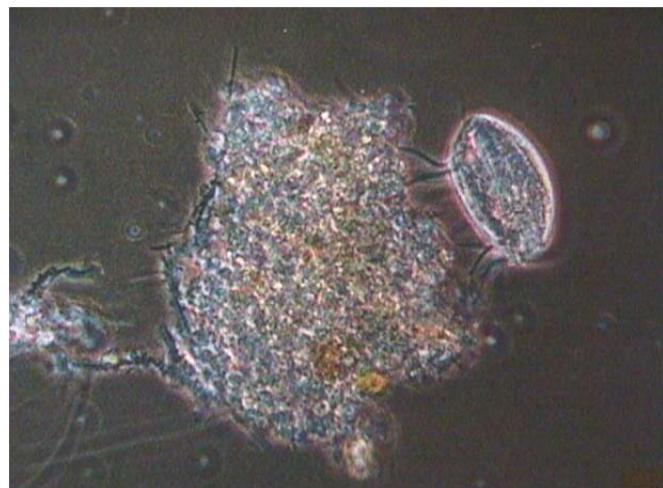
### **Aerobic Bacteria**

Aerobic bacteria live and multiply in the presence of free oxygen. Facultative bacteria always achieve an aerobic state when oxygen is present. While the name "aerobic" implies breathing air, dissolved oxygen is the primary source of energy for aerobic bacteria. The metabolism of aerobes is much higher than for anaerobes. This increase means that 90% fewer organisms are needed compared to the anaerobic process, or that treatment is accomplished in 90% less time. This provides a number of advantages including a higher percentage of organic removal. The by-products of aerobic bacteria are carbon dioxide and water. Aerobic bacteria live in colonial structures called floc and are kept in suspension by the mechanical action used to introduce oxygen into the wastewater. This mechanical action exposes the floc to the organic material while treatment takes place. Following digestion, a gravity clarifier separates and settles out the floc. Because of the mechanical nature of the aerobic digestion process, maintenance and operator oversight are required.

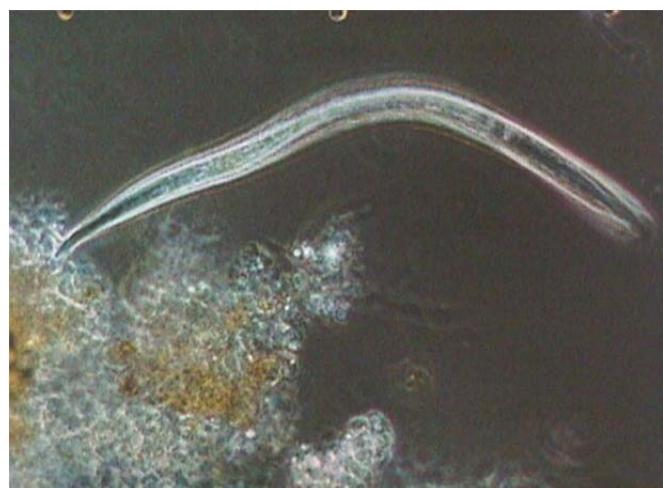
## Protozoans and Metazoans

In a wastewater treatment system, the next higher life form above bacteria is protozoans. These single-celled animals perform three significant roles in the activated sludge process. These include floc formation, cropping of bacteria and the removal of suspended material. Protozoans are also indicators of biomass health and effluent quality. Because protozoans are much larger in size than individual bacteria, identification and characterization is readily performed. Metazoans are very similar to protozoans except that they are usually multi-celled animals. Macroinvertebrates, such as nematodes and rotifers, are typically found only in a well developed biomass.

The presence of protozoans and metazoans and the relative abundance of certain species can be a predictor of operational changes within a treatment plant. In this way, an operator is able to make adjustments and minimize negative operational effects simply by observing changes in the protozoan and metazoan population.



Aspidisca



Nematode

### **Dispersed Growth**

Dispersed growth is material suspended within the activated sludge process that has not been adsorbed into the floc particles. This material consists of very small quantities of colloidal (too small to settle out) bacteria as well as organic and inorganic particulate material. While a small amount of dispersed growth between the floc particles is normal, excessive amounts can be carried through a secondary clarifier. When discharged from the treatment plant, dispersed growth results in higher effluent solids.

### **Taxonomy**

Taxonomy is the science of categorizing life forms according to their characteristics. Eighteen different categories are used to define life forms from the broadest down to the most specific. They are: Kingdom, Phylum, Subphylum, Superclass, Class, Subclass, Cohort, Superorder, Order, Suborder, Superfamily, Family, Subfamily, Tribe, Genus, Subgenus, Species and Subspecies. Identifying the genus is usually specific enough to determine the role of the organisms found in a wastewater treatment system.

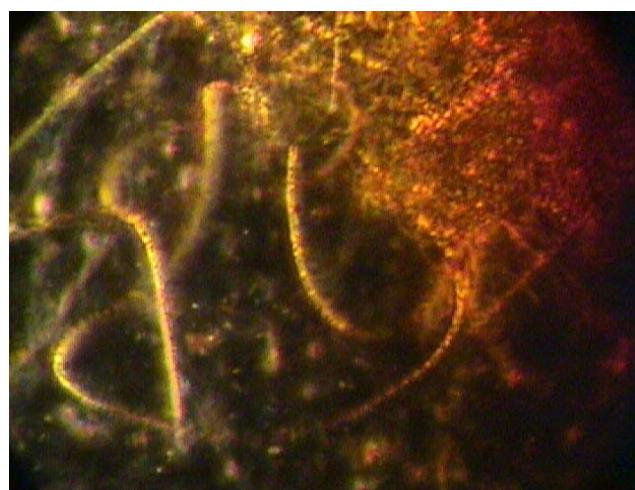
### **Process Indicators**

Following taxonomic identification, enumeration and evaluation of the characteristics of the various organisms and structures present in a wastewater sample, the information can be used to draw conclusions regarding the treatment process.

Numerous industry references, such as **WASTEWATER BIOLOGY: THE MICROLIFE** by the Water Environment Federation, can be used to provide a comprehensive indication of the conditions within a treatment process. As an example, within most activated sludge processes, the shape of the floc particles can indicate certain environmental or operational conditions.

A spherical floc particle indicates immature floc, as would be found during start-up or a process recovery. A mature floc particle of irregular shape indicates the presence of a beneficial quantity of filamentous organisms and good quality effluent. An excess of dispersed growth could indicate a very young sludge, the presence of toxic material, excess mechanical aeration or an extended period of time at low dissolved oxygen levels.

Certain protozoans, such as amoebae and flagellates dominate during a system start-up. Free swimming ciliates are indicative of a sludge of intermediate health and an effluent of acceptable or satisfactory quality. A predominance of crawling ciliates, stalked ciliates and metazoans is an indicator of sludge with excellent health and an effluent of high quality.



**Filamentous Bacteria**

## **Filamentous Bacteria have Positive aspects:**

They are very good BOD removers

They add a backbone or rigid support network to the floc structure

Helps the floc structure filter out fine particulate matter that will improve clarifier efficiency.

They help the floc settle if in small amounts.

They reduce the amount of "pin" floc.

## **Filamentous Bacteria have Negative aspects:**

They can interfere with separation and compaction of activated sludge and cause bulking when predominant.

### **Filamentous Bacteria**

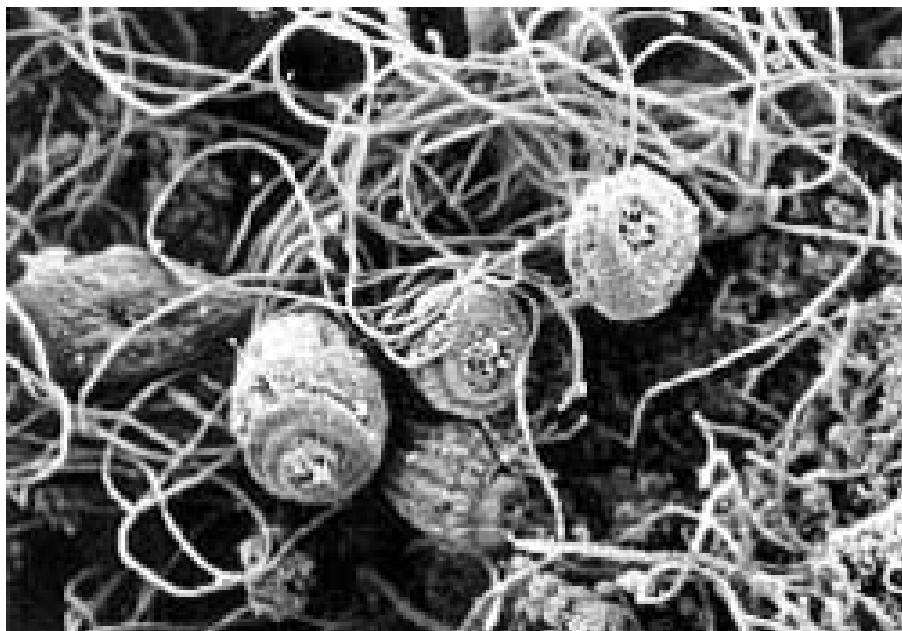
They can affect the sludge volume index (SVI).

They can cause poor settling if dominant.

They can fill up a clarifier and make it hard to settle, causing TSS carryover.

They can increase polymer consumption.

They can increase solids production and cause solids handling costs to increase significantly.



Filamentous bacteria floc (SEM) or Pin Floc.

## Activated Sludge Aerobic Flocs

Aerobic flocs in a healthy state are referred to as activated sludge. While aerobic floc has a metabolic rate approximately 10 times higher than anaerobic sludge, it can be increased even further by exposing the bacteria to an abundance of oxygen. Compared to a septic tank, which takes several days to reduce the organic material, an activated sludge tank can reduce the same amount of organic material in approximately 4-6 hours. This allows a much higher degree of overall process efficiency. In most cases, treatment efficiencies and removal levels are so much improved that additional downstream treatment components are dramatically reduced or totally eliminated.

### Problems may appear during the operation of activated sludge systems, including:

- High solids content in clarified effluent, which may be due to too high or too low solids retention time and to growth of filamentous microorganisms.
- Rising sludge, occurring when sludge that normally settles rises back to the surface after having settled. In most cases, this is caused by the denitrification process, where nitrate present in the effluent is reduced to nitrogen gas, which then becomes trapped in the sludge causing this to float. This problem can be reduced by decreasing the flow from the aeration basin to the settling tank or reducing the sludge resident time in the settler, either by increasing the rate of recycle to the aeration basin, increasing the rate of sludge collection from the bottom, or increasing the sludge wasting rate from the system.
- Bulking sludge, that which settles too slowly and is not compactable, caused by the predominance of filamentous organisms. This problem can be due to several factors of which the most common are nutrient balance, wide fluctuations in organic load, oxygen limitation (too low levels), and an improper sludge recycle rate.
- Insufficient reduction of organic load, probably caused by a low solids retention time, insufficient amount of nutrients such as P or N (rare in fisheries wastewaters), short-circuiting in the settling tank, poor mixing in the reactor and insufficient aeration or presence of toxic substances.
- Odors, caused by anaerobic conditions in the settling tanks or insufficient aeration in the reactor.

### Filamentous Organisms

The majority of filamentous organisms are bacteria, although some of them are classified as algae, fungi or other life forms. There are a number of types of filamentous bacteria which proliferate in the activated sludge process. Filamentous organisms perform several different roles in the process, some of which are beneficial and some of which are detrimental. When filamentous organisms are in low concentrations in the process, they serve to strengthen the floc particles. This effect reduces the amount of shearing in the mechanical action of the aeration tank and allows the floc particles to increase in size.

Larger floc particles are more readily settled in a clarifier. Larger floc particles settling in the clarifier also tend to accumulate smaller particulates (surface adsorption) as they settle producing an even higher quality effluent. Conversely, if the filamentous organisms reach too high a concentration, they can extend dramatically from the floc particles and tie one floc particle to another (interfloc bridging) or even form a filamentous mat of extra-large size. Due to the increased surface area without a corresponding increase in mass, the activated sludge will not settle well. This results in less solids separation and may cause a washout of solid material from the system. In addition, air bubbles can become trapped in the mat and cause it to float, resulting in a floating scum mat.

Due to the high surface area of the filamentous bacteria, once they reach an excess concentration, they can absorb a higher percentage of the organic material and inhibit the growth of more desirable organisms.

## Filamentous Bacteria Identification

Filamentous Identification should be used as a tool to monitor the health of the biomass when a filament problem is suspected. Filamentous Identification is used to determine the type of filaments present so that a cause can be found and corrections can be made to the system to alleviate future problems. All filamentous bacteria usually have a process control variation associated with the type of filament present that can be implemented to change the environment present and select out for floc forming bacteria instead. Killing the filaments with chlorine or peroxide will temporarily remove the filaments, but technically it is a band-aid. A process change must be made or the filaments will return with time eventually. Find out what filaments are present, find out the cause associated with them and make a process change for a lasting fix to the problems.

### Filaments, their causes and suggested controls

Low DO Filaments	Control
Type 1701 <i>S. natans</i>	Adjust the aeration rates or F/M (based on aeration solids)
<i>H. hydrossis</i>	Long RAS lines or sludge held too long in the clarifier can sometimes cause the growth of low DO filaments even if the aeration
Waste with limited Nutrients	Control
Thiothrix I & II 021N and <i>N. Limicola</i> III	Nutrient addition BOD ratio of 100:5:1
Low F/M ratios	Control
0041, <i>Nocardia</i>	Use of selector, increase RAS
Type 1851, 0961, 0803, 0675	Increase WAS

Some filaments have more than one version of the filament species, with slightly different characteristics for identification.

*N. Limicola* I  
*N. Limicola* II  
*N. Limicola* III  
Thiothrix I  
Thiothrix II

### Filamentous Identification

Filaments can be internal or external, and they can be free of the floc structures or found intertwined in the floc. Most labs think that filaments need to be extending from the floc in order to be a problem. This is not true. Internal filaments can cause more problems than external filaments. Think of internal filaments causing a structure like a sponge. It will retain water easily and be harder to dewater, will be hard to compress and will take up more space, thereby increasing solids handling costs.

Filaments present in the system do not always mean there is a problem. Some filaments are good if they form a strong backbone and add a rigid network to the floc. They help give the floc more structure and settle faster. Filaments are good BOD degraders also. They are only a problem when they become dominant. If filament abundance is in the abundant or excessive range, having a Filamentous Identification performed is recommended. When Gram and Neisser stains are performed for filamentous identification, the types of filaments found present will be noted on the Floc Characterization sheet to the right of the filament section and will be noted on the Cover Sheet. A Filament Causes sheet, Filamentous Predominance sheet and corrective actions will be given and included with the report. A Filamentous Worksheet will be included. Individual sheets on the actual filaments present in the sample will be included with more information on that particular filament.

The activated sludge process was invented around 1914 and is today still the most commonly used biological wastewater treatment process. This widespread use is due to the fact that activated sludge can be a rather easy process to implement and one that can attain high treatment efficiency. That is to say, if it works! Activated sludge is susceptible to process disturbances making it a very problematic technology for many of its users. Problems arise most when the wastewater to be treated varies significantly in composition and/or flow.

**Let's do a quick review of the Bugs.... We will go much more into detail later...**

#### **Nocardia amarae**

*Nocardia amarae*, a common cause of disruptive foaming in waste treatment plants, is a slow growing, usually gram-positive, chemoautotrophic, filamentous, strict aerobe that produces the biosurfactant trehalose. Colonies can be brown, pink, orange, red, purple, gray or white, so color alone is not a key to identifying this species. *N. amarae*, member of the Actinomycetes family, is not motile, so it relies on movement of the water to carry it through the system. It produces catalase, urease and nitrate reductase enzymes, but not casease. The foam from *Nocardia amarae* is usually a viscous brown color unless algae are entrapped in it, in which case it appears green and brown.

#### **Nostocoida limicola**

*Nostocoida limicola* is yet another common cause of disruptive foaming in waste treatment plants, motile in its Hormogonia and sometimes Trichome phases. This oxygenic phototrophic species often forms a confluent gel encasing flattened discs or large sheets of cells, forming symbiotic relationships with other species. Staining gram-positive, *Nostocoida* produces round cells within tight coil formations. *Nostocoida* can also be identified by their starburst effect formations using phase contrast microscopy at 400 to 1000x magnification. After chlorination, a few dead cells sticking out identify stress to this species.

#### **Thiothrix**

*Thiothrix* spp., the second most common cause of disruptive foaming in wastewater treatment plants appears as straight to slightly curved cells with rectangular shape form filaments up to 500 microns in length, in multicellular rigid filaments, staining gram-negative, with obligately aerobic respiration. *Thiothrix* are mixotrophic, using several small organic carbons and reduced inorganic sulfur sources for growth and energy. *Thiothrix* I is one of the largest filament found using phase contrast microscopy at 400 to 1000x magnification. *Thiothrix* II produces rectangular filaments up to 200 microns in length and is easily identified by their starburst effect formations using phase contrast microscopy at 400 to 1000x magnification.

#### **Microthrix parvicella**

*Microthrix parvicella* is another common cause of disruptive foaming in waste treatment plants, producing filaments up to 400 microns in length, easily visualized by phase contrast microscopy at 400x magnification. This species is usually found outside floc, tangling with structures in the system, but can also be found hanging out of the floc.

#### **Sphaerotilus natans**

*Sphaerotilus natans* is another filamentous species, and yet it is reputed to increase settleability by branching between flocs, increasing surface area. Cells are straight to slightly curved, up to 1000 microns in length and stain gram-negative. These large cells can be easily visualized by phase contrast microscopy at 100x magnification. Certain conditions favor the proliferation of filamentous species. A low F/M (food to mass) ratio favors filamentous organisms, because their higher ratio of surface area to volume provides them with a selective advantage for securing nutrients in nutrient limited environments. When a plant runs an extremely long sludge age, the slower growing filaments have a better chance to establish a strong colony. As a strict aerobe, high levels of oxygen are necessary to sustain this species. Mesophilic, *Nocardia amarae* thrives in temperatures from 17 to 37 deg. C.

The presence of high levels of fats, oils and greases or hydrocarbons and phenols, can encourage this species, particularly when insufficient levels of nitrogen and phosphorus are present to balance these carbon sources.

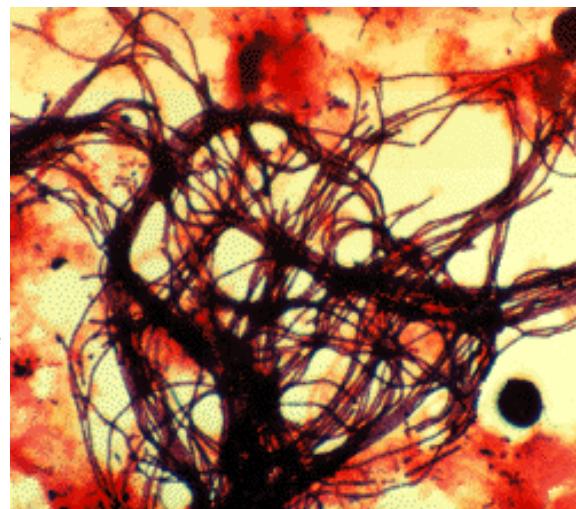
### **Filamentous Bacteria**

A problem that often frustrates the performance of activated sludge is bulking sludge due to the growth of filamentous bacteria. Sludge bulking can often be solved by careful process modifications. However, different filamentous bacteria such as *Microthrix*, *Sphaerotilus*, *Nostocoida*, *Thiothrix* or "Type 021N" and others cause bulking for very different reasons. Many filamentous species have not even been given a scientific name yet. Consequently, in order to make the right kind of process modification, knowledge to identify them and experience with the process ecology are required. The potential for instability with activated sludge is an acute problem when strict demands on treatment performance are in place.

### **PAX - finally, a Fix for Microthrix**

If you ever experienced an overgrowth of *Microthrix parvicella* in your activated sludge plant, you will be aware that it can be very difficult to either eradicate or control. *Microthrix* is the most common cause of bulking and foaming in activated sludge plants (Rosetti et al. 2002), and it appears either essentially alone or in the company of other filaments.

*Microthrix* foams appear in many of the photographs of aeration basins and clarifiers I have collected all over the world, and many of the plant tours on the Internet show the same brown stable scums associated with this organism. Let's face it, *Microthrix* is just about everywhere.



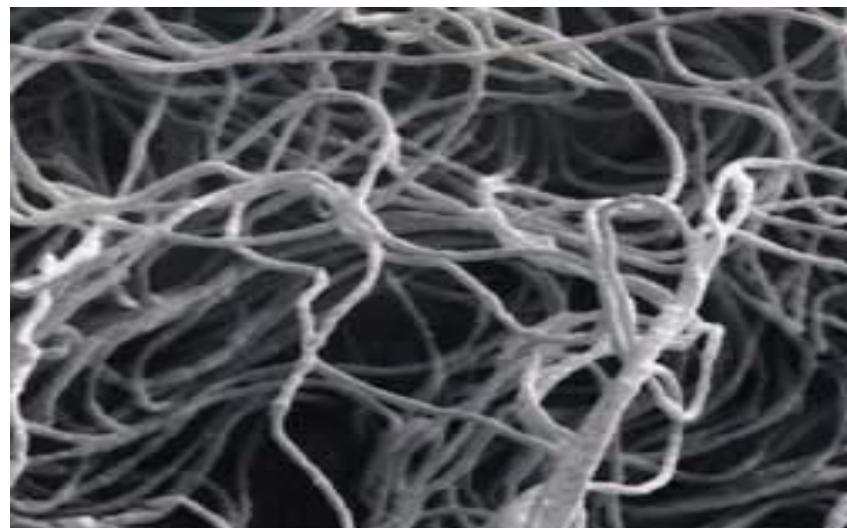
**Figure 1.**  
**A micrograph of *Microthrix parvicella*, gram stain x 1000**

### ***Microthrix* is your enemy - Get to know it!**

*Microthrix* fits into the filamentous bacterial classification of low F/M, which means that it tends to appear in plants with long sludge ages. Lackay et al. (1999) suggested that *M. parvicella* and its low F/M compatriots *Haliscomenobacter hydrossis*, and types 0092, 0041, 1851, 0803 were also encouraged to the point of maximum proliferation by alternating anoxic-aerobic conditions (particularly 30-40% aerobic and 60-70% anoxic) but any alternation of anoxic-aerobic conditions may cause a problem in single reactor, two reactor, or multireactor systems in which nitrate and/or nitrite are present throughout the anoxic period, or in the anoxic reactor just prior to the aerobic reactor. Modern plants incorporating denitrification and/or phosphorus removal are obvious candidates for bulking and foaming due to *Microthrix*.

Figures 1 and 2 show typical views of *Microthrix* by using light microscopy and scanning electron microscopy respectively. It is not difficult to recognize using standard staining and microscopy, giving a positive response to Gram stain and being of fairly easily recognized morphology (Seviour et al. 1999). Of all the filaments creating difficulties in activated sludge plants, it is one of the most easily recognized, but there is a commercial test kit available which uses fluorescent situ hybridization (or "FISH") to permit visual identification should one feel the need.

The design of plants can play a significant part in the proliferation of scums and foams and there are many common mistakes in plant design which assist organisms like *Microthrix* by retaining floating masses in dead areas of the plant which have very high MCRT values and continuously reseed the biomass, (Pitman 1996). These should obviously be avoided (Figs 3, 5 and 6). Similarly poor mixing, poorly designed and inadequate aeration systems, cyclic overloading and low process D.O. levels can contribute to the creation of anoxic and anaerobic zones in what are supposed to be aeration basins.



**Figure 2.** A scanning electron micrograph of *Microthrix parvicella*

#### Current Remedial Techniques

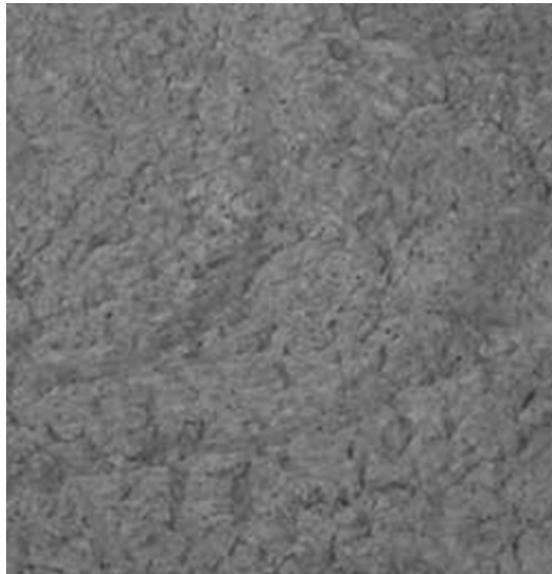
Jenkins *et al.* (1993) presented sludge chlorination as a method of choice in the United States to combat filamentous bulking due to any organism. The success of treatment of *Microthrix* in mixed liquor or foams is poor, due it is believed to resistant filamentous bacteria with hydrophobic cell walls such as *M. parvicella* and *Nostocoida limicola*.

Lakay *et al.* (1988) obtained only a partial elimination of *Microthrix parvicella* bacteria at a high chlorine dose. Hwang and Tanaka found in batch tests that *M. parvicella* remained intact at very high chlorine doses, while the microbial flocs were completely destroyed. Saayman *et al.* (1996) examined the use of non-specific chemical treatment in a BNR plant and assessed the effects of biomass settling characteristics and other operational parameters. While chlorine use was the most effective, it was reported to damage the biomass and cause difficulties in the P removal process when dosed at high levels, while ozone and peroxide were less effective in treating settling problems but less of a problem to the biomass.



**Figure 3.**  
*Dry Microthrix parvicella* foam  
trapped in an anoxic zone of a  
BNR plant. aeration basin.

In recent times the introduction of selectors has been hailed as a major initiative in the control and elimination of filamentous bacteria (bulking and foaming) and the maintenance of moderate biomass SVIs. Evidence on the performance of selectors in controlling low F/M filaments has been described as both controversial and ambiguous and, in the Netherlands, despite incorporating over 80 selectors in full-scale plants, the percentage of plants with bulking associated with *Microthrix parvicella* was unchanged. Other experiences with the aerobic selector showed only little success in controlling the growth of *M. parvicella* in the presence of long chain fatty acids (LCFA), (Lebek and Rosenwinkel, 2002) and a comparison of anoxic selectors at five plants in the US has demonstrated that performance and effectiveness varied significantly (Marten and Daigger, 1997).



**Figure 4. Typical dark brown *Microthrix parvicella* foam on an**

#### **More on *Microthrix***

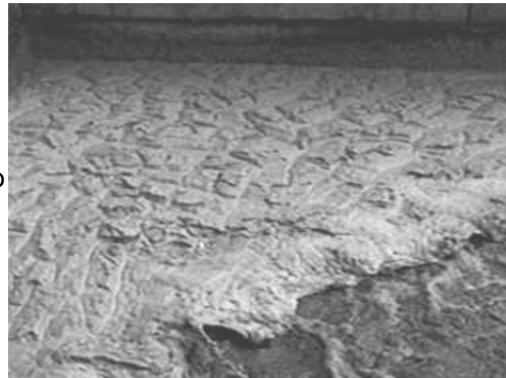
Mamais *et al.* 1998 examined the effect of factors such as temperature, substrate type (easily biodegradable in the form of acetate and slowly biodegradable in the form of oleic acid) on *Microthrix parvicella* growth using complete mix with and without selectors (anoxic and anaerobic) and plug flow reactors. The results indicate that low temperatures and substrates in the form of long chain fatty acids favor the growth of *M. parvicella*. The plug flow configuration was shown to be quite effective in controlling the growth of *M. parvicella* and producing a sludge with good settling characteristics, while the presence of a selector, either anoxic or anaerobic, had no significant effect on the growth of *M. parvicella*. Maintenance of low sludge ages (5) days has also been reported to eliminate *M. parvicella* because it is a slow growing organism, but this is not always operationally possible.

While it is often convenient to group filaments together, it does appear the *Microthrix* has received special attention because of its ability to proliferate. More selective investigation of *Microthrix* has indicated that it has quite well defined requirements. The nature of *Microthrix* is such that it has the capability of using long chain fatty acids (oleic acid) and their esters (triglycerides of palmitic and stearic acid) (fats and oils) as sources of carbon and energy.

Lipids and LCFA are present in all domestic wastewater streams and often constitute a significant part of it. Values of 25-35% of the incoming COD have been reported, and it can support a substantial biomass production in a treatment plant. LCFA are generally easily consumed in activated sludge, and the consumption rate of LCFA under aerobic or anoxic conditions has been found to be rapid.

Studies indicate that *M. parvicella* consumes exclusively long chain fatty acids (LCFA), and that it is able to take up LCFA not only under aerobic, but also under anaerobic and anoxic conditions (Andreasen, K. and Nielsen, P.H. (2000)). It has been reported that *M. parvicella* is able to out-compete other bacteria particularly well in alternating anaerobic-aerobic and anoxic activated sludge systems. This ability is based on a high uptake and storage capacity for LCFA under anaerobic conditions and a subsequent use of the stored substrate for growth with oxygen (or nitrate) as electron acceptor.

Rosetti *et al.* (2002) carried out an extensive examination of *M. parvicella* and found that it was a very versatile organism which could store organic carbon under anaerobic conditions using stored polyphosphate for energy (like the organisms responsible for phosphorus removal). Once exposed to aerobic conditions it would recover rapidly and resume growing. *Microthrix* has a high storage capacity under all operating conditions (anaerobic, anoxic and anaerobic). It has a high "substrate affinity" or low *K<sub>s</sub>*, which means it competes well at low substrate concentration.



**Figure 5.**  
*Microthrix parvicella* foam trapped near a mechanical aerator.

Most interestingly, *M. parvicella* has a maximum growth rate near 22° C, zero growth rate at 30° C and is capable of quite reasonably large growth rates at as low as 7° C which gives it a significant advantage in the competition with floc formers during winter in cold climates.

#### PAX vs. *Microthrix parvicella*

*Microthrix parvicella* is well-equipped to survive, compete and dominate in all kinds of activated sludge systems. With all of the above in mind, it is pleasing to find that *Microthrix* does have a weakness. That weakness is its apparent sensitivity to poly aluminum chloride (PAX) dosing, which seems to attack the ability of *Microthrix parvicella* to use lipids by reducing the activity of extracellular enzymes (lipases) on the surface of the organism rendering the organism relatively uncompetitive (Nielsen *et al.* 2003).

Roels *et al.* (2002) reported a loss of surface scum following PAX-14 dosing which was probably due to a loss of hydrophobicity. Full-scale dosages of PAX-14 range from 1.5 to 4.5 g Al<sup>3+</sup>/kg MLSS/day depending on the sludge retention time (SRT); the lower the SRT, the higher the dosage and certainly lower than 7 g Al<sup>3+</sup>/kg MLSS. Roels *et al.* (2002) offered the following empirical formula to establish the dose:

$$60/\text{SRT} = \#\text{g of Al}^{3+}/\text{kg MLSS}$$

They also recommended the removal of the scum layer before dosing to allow the concentration and time of dosage to be kept at a minimum. Removal of the floating sludge layer from the surface before starting PAX application was necessary to ensure specific and rapid impact of Al-salts on *M. parvicella*.

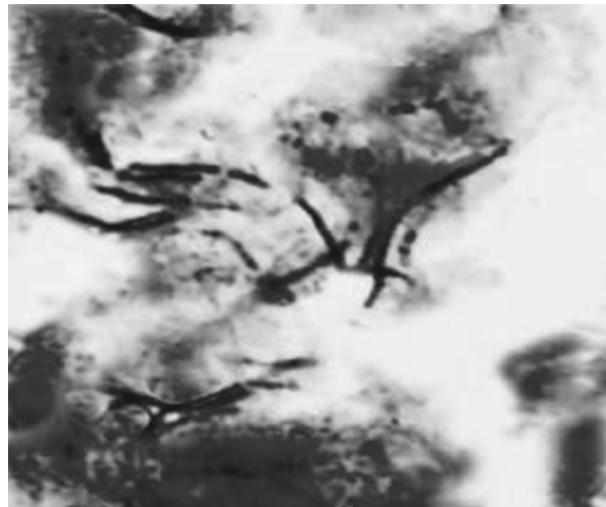
In fact, the stable floating sludge represents an independent microbial system, into which aluminum can penetrate only at a limited extent. Dosage should be combined with high oxygen concentration in the aeration (i.e. above 2.5 mg/L) and the MLSS concentration low (i.e. under 2.5 g/L) since *M. parvicella* competes well at low oxygen levels.



**Figure 6.** A heavy build-up of trapped *Microthrix parvicella* foam during winter.

Of note was that the morphological properties of only *Microthrix parvicella* changed, apparently leaving the other filaments remaining unaffected.

Paris *et al.* (2003) came to a similar conclusion; by dosing  $\text{AlCl}_3$  (3.5 mg  $\text{mgAl}^{3+}$  gMLSS/d), a general improvement of the settling properties of the activated sludge was achieved. As the filamentous population of activated sludge and the occurrence frequency of *M. parvicella* dropped, a decrease of hydrophobicity and floating tendency of activated sludge was observed. With low hydrophobicity the sludge does not tend to float. This has significant relevance for any measure to prevent floating foams.

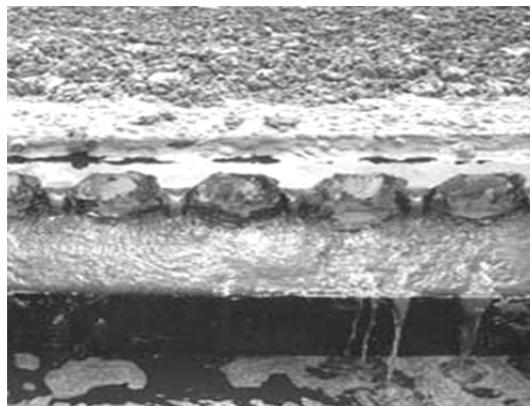


**Figure 7.** An typical view of *Microthrix parvicella* (gram stain x 1000) after extended PAX treatment.

It was observed that by adding PAX a morphological modification of the filamentous bacterium *M. parvicella* occurs. The morphological modification is probably the reason why the hydrophobic property of the filaments decreases. Paris *et al.* (2003) included micrographs which indicated that the *Microthrix parvicella* appeared to shorten in length after dosing (Figure 7) and no longer inhabit the zones between flocs.

## PAX

PAX (or PAX-14 or polyaluminium chloride) used for *Microthrix* control is a flocculant or coagulant commonly used in water and wastewater treatment. The 14 or other number associated with the name refers to the particular grade of the chemical. Nielsen *et al.* (2003) report that PAX-14 is  $\text{Al}_{13}\text{O}_4(\text{OH})_{24}(\text{H}_2\text{O})_{12}^{7+}$  and it is produced from  $\text{Al}(\text{OH})_3$  at high temperature and high pressure. PAX-14 and 18 are being used in several countries with good success for controlling *M. parvicella* - in particular Denmark where PAX-14 has been applied successfully in treatment plants with biological N and/or P removal for 91 out of 500 plants in 2002.



**Figure 8.**  
Foam build-up in a secondary clarifier resulting in solids loss and turbid effluent.

## Proposed Treatment Regime

In the fall, to prevent the normal appearance of *M. parvicella* during the coming winter and to control problems with *M. parvicella* (winter, spring). **Dosage:** 0.5-1.5g Al/kgSS/day usually added to return sludge. PAX should be dosed continuously over the treatment period at the chosen level. Removal of floating sludge before and during dosing is recommended. Microscopic examination of the biomass and regular testing of biomass settling is also a very good idea and the dosing at the chosen remedial rate until a target SVI or preferably DSVI is reached should be the rule. It is not yet fully clear why PAX has the effect that it does, but the research continues. It is known that other Al salts have little effect on surface associated enzymes after 15 min, and no effect on surface hydrophobicity and surface associated enzymes.



Both photographs show the aeration sequence for an SBR.  
Sequence Batch Reactor.



Anoxic zone, denitrification area.



The same area from above photo, but clean and dry, these are porous air diffusers. A rare photo.



This clarifier is used to thicken sludge prior to the digester, this is not a digester but looks like one. Notice the light on the top for Operators to look inside.

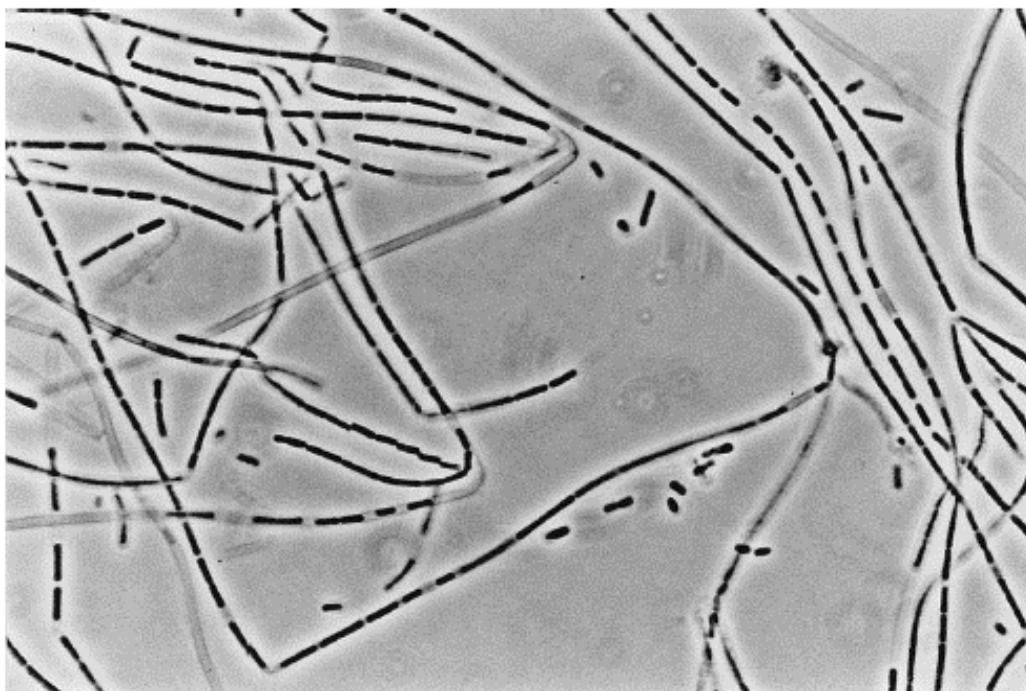


Two massive anaerobic digesters.

## *Sphaerotilus natans*

### Description and Significance

*Sphaerotilus natans* is a filamentous bacterium that is covered in a tubular sheath and can be found in flowing water and in sewage and wastewater treatment plants. While this bacterium sometimes clogs pipes and causes other similar problems, it does not cause major threat to wastewater treatment plants nor is it known to be pathogenic.



Long unbranched and ensheathed filaments produced by *Sphaerotilus natans* IF4.

Relatively long, non-motile filaments (100-1000  $\mu\text{m}$ ). Straight or smoothly curved with tree-like false branching. The cells are round-ended and rod shaped (1.0-1.8 x 1.5-3.0) and are contained in a clear, tightly fitting sheath. **Note:** They can be rectangular when the cells are tightly packed within the sheath. The cell septa are clear and easily observable with indentations. Filaments radiate outward from the floc surface into the bulk solution and can cause sludge settling interference by inter-floc bridging. The filament is usually Gram negative and Neisser negative. There are no sulfur granules. Poly- $\beta$ -hydroxybutyric acid (PHB) is frequently observed as dark intracellular granules. In wastewater that is nutrient deficient, an exocellular slime coat may be present. Attached growth is usually uncommon, but may occur when at low growth rate.

This filament is usually found in environments where there is low DO or low nutrients (Nor P).

### Control

RAS chlorination can be used to get rid of the filaments but process changes should also be made. Cell lysis occurs readily on this type of filament, although the empty sheaths still remain. Sludge wasting is necessary to remove them entirely from the system.



Manipulation of F/M and DO concentration can be used to control the filaments. Nutrient deficient wastes can be checked by effluent values of residual NH<sup>3</sup> and o-PO<sup>4</sup> and should be supplemented if necessary.

#### Rank

*Sphaerotilus natans* ranks 6th in number of predominance. Typically not found in pulp-mills with activated sludge.

#### ***Nostocoida limicola I and II***

*Nostocoida limicola I* is a bent and highly coiled filament. *N. limicola* has cells that are oval (0.6-0.8  $\mu\text{m}$  wide) but are found to be closer to each other and the cell septa are almost indiscernible. The length of the filament can range from 100 to 200  $\mu\text{m}$  and the majority of the time the trichome is found within the floc. *N. limicola* has no sheath and attached growth is rare. It stains Gram positive and Neisser positive.

#### ***Nostocoida limicola II* Identification**

Medium length, non-motile filaments (100-200  $\mu\text{m}$ ). Bent and irregularly coiled filaments with incidental true branching. Knots sometimes seen. Cell septa are clear with indentations. Cells are oval or disc shaped (1.2-1.4  $\mu\text{m}$ ). Filaments are found within the floc structure but may occur in the bulk solution. The filament staining is variable, it is usually Gram negative but sometimes positive and Neisser positive. Usually easy to identify due to its Neisser staining properties. Stains entirely purple and looks like stacked discs (or hockey pucks). In industrial wastes, an organism that is Gram negative and Neisser negative occurs. There is no sheath and there are no sulfur granules. Poly- $\beta$ -hydroxybutyric acid (PHB) granules are frequently observed as dark intracellular granules. Attached growth is usually uncommon. Three subtypes are known. Resembles *M. parvicella* except in its Neisser staining properties.



#### Environment

This filament is usually found in environments where there is low DO or low F/M and the presence of organic wastes. Wastes containing starch seem more selective to this filament. Bulking is more common in industrial wastes. The filament appears to be facultative fermentative, which is unique for most filaments.

#### Control

Manipulation of F/M (usually an increase) and DO concentration can be used to control the filaments. A selector may be used and chlorination. System changes include changing from a complete mix to plug flow aeration basin configuration.

*N. limicola* ranks 12th in number of predominance in industry. Typically not found in kraft mills. Common in municipalities.

## Thiothrix I & II

Thiothrix species consist of two types of Thiothrix and they are Thiothrix I and Thiothrix II. Thiothrix filaments are straight or slightly curved with Thiothrix I having an overall length of 100-500  $\mu\text{m}$  and individual cells having a rectangular shape (1.4-2.5 x 3-5  $\mu\text{m}$ ). Thiothrix II has total length varying from 50-200  $\mu\text{m}$  and its cells are rectangular (0.8-1.4 x 1-2  $\mu\text{m}$ ).

Both types of Thiothrix are found stretching from the floc surface, there is a noticeable septa between cells. Both species are Gram negative and Neisser negative with cells that on occasions have sulfur granules. There are additional structures on Thiothrix trichomes and they include apical gonidia as well as rosettes and a sheath is present, incidental attached growth may be observed. A holdfast may add to the characteristic of radiating out from a common center, the "starburst effect".

Relatively large, non-motile filaments (100-500  $\mu\text{m}$ ). Straight or smoothly curved filaments with no branching. Cells are rectangular (1.4 x 2.5  $\mu\text{m}$ ) and a clear cell septa is present without indentations at the septa. Filaments are found radiating outwards from the floc structure causing inter-floc bridging.

The filament staining is Gram negative or Gram variable when sulfur granules are present and Neisser negative with Neisser positive granules observed frequently.

Exhibits bright sulfur granules in the presence of sulfides under phase contrast (use the S-test). Poly- $\beta$ -hydroxybutyric acid (PHB) is frequently observed as dark intracellular granules. No attached growth when extending into the bulk solution. Can form rosettes and the filaments can have gonidia on the tips. Rosettes are when many filaments radiate outward from a common origin. Prominent heavy sheath. Easy to identify due to its large size.

### Similar Organisms

Type 021N is similar when in the bulk solution and with no attached growth, although Type 021N has no sheath.

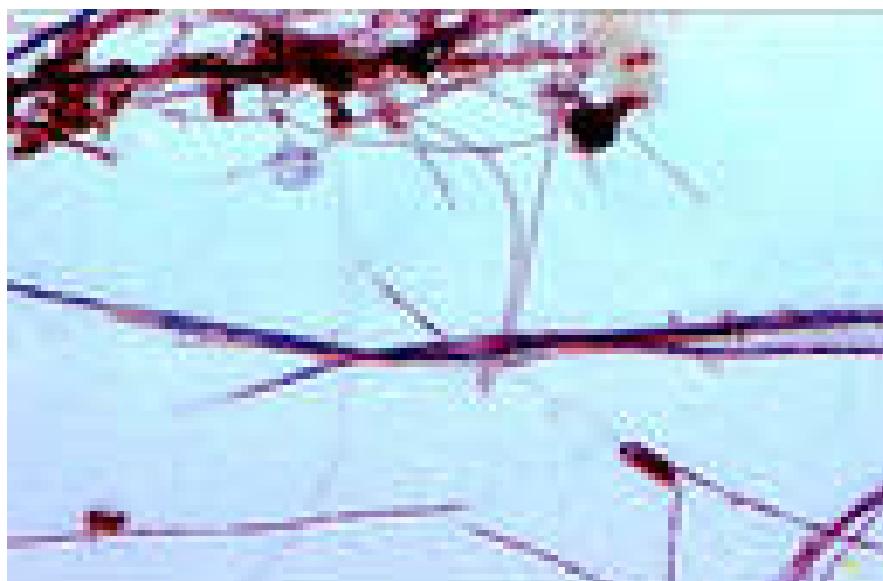
### Environment

This filament is usually found in environments where there are limited nutrients (N or P). It can also be found in wastes containing specific compounds with sulfides and/or organic acids or environments with low DO. Sometimes found in plants with high pH in the aeration system.





Thiothrix II



## Other Wastewater Treatment Components

### Biochemical Oxygen Demand

Biochemical Oxygen Demand (**BOD** or **BOD<sub>5</sub>**) is an indirect measure of biodegradable organic compounds in water, and is determined by measuring the dissolved oxygen decrease in a controlled water sample over a five-day period.

During this five-day period, **aerobic** (oxygen-consuming) bacteria decompose organic matter in the sample and consume dissolved oxygen in proportion to the amount of organic material that is present. In general, a high BOD reflects high concentrations of substances that can be biologically degraded, thereby consuming oxygen and potentially resulting in low dissolved oxygen in the receiving water.

The BOD test was developed for samples dominated by oxygen-demanding pollutants like sewage. While its merit as a pollution parameter continues to be debated, BOD has the advantage of a long period of record.

### Organic Carbon

Most organic carbon in water occurs as partly degraded plant and animal materials, some of which are resistant to microbial degradation. Organic carbon is important in the estuarine food web and is incorporated into the ecosystem by photosynthesis of green plants, then consumed as carbohydrates and other organic compounds by higher animals. In another process, formerly living tissue containing carbon is decomposed as detritus by bacteria and other microbes.

### Total Organic Carbon

(**TOC**) bears a direct relationship with biological and chemical oxygen demand; high levels of TOC can result from human sources, the high oxygen demand being the main concern.



Microscopic identification is essential for any activated sludge process.



Lab tech removing filter for TSS analysis.



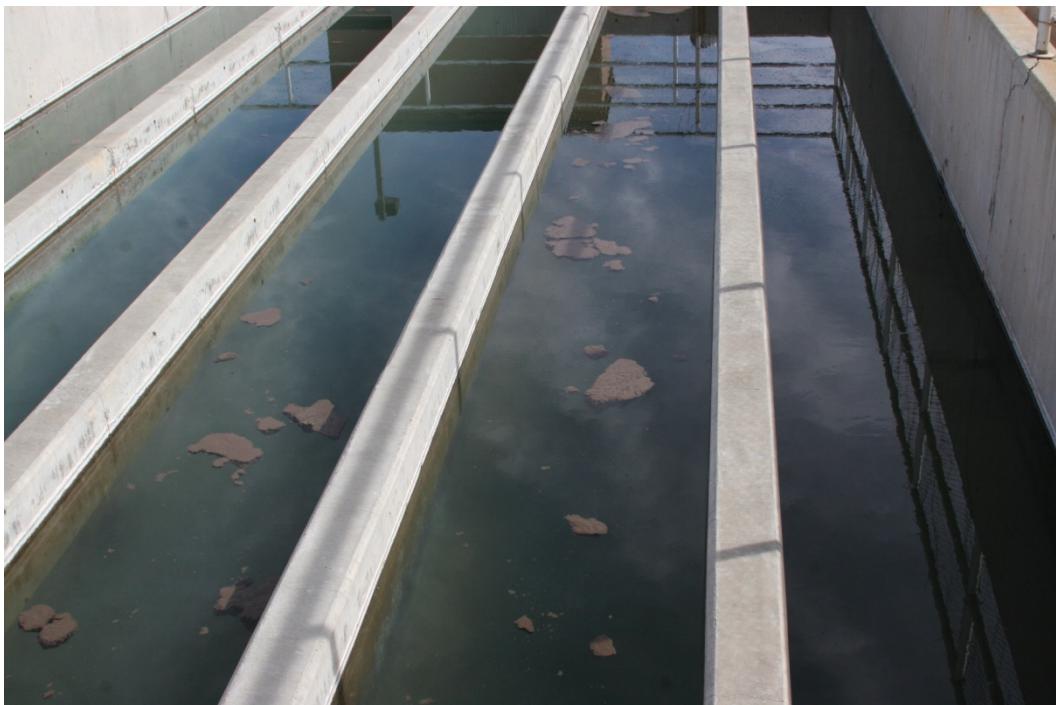
Here is an example of a rectangular clarifier used in the secondary settling process. Operation changes that should be employed if a dark brown foam is developing on the aeration basin is to increase the wasting rate.



Here is Pen floc being carried over the weir do to a process upset. Algae growth in excess can also create several different problems.



These operators are making sure that the backwash pumps are working for the sand filter. Notice the beautiful Arizona background.



During a plant upset, sludge from the filters can be carried over to the chlorine contact channel. In this photograph, it is not too bad, I've seen much worse.

# Need for Nitrogen and Phosphorus Removal at Wastewater Treatment Plants

## Nutrients

Nutrients are chemical elements or compounds essential for plant and animal growth. Nutrient parameters include ammonia, organic nitrogen, Kjeldahl nitrogen, nitrate nitrogen (for water only) and total phosphorus. High amounts of nutrients have been associated with eutrophication, or over-fertilization of a water body, while low levels of nutrients can reduce plant growth and (for example) starve higher level organisms that consume phytoplankton.

The purpose of this section is to provide an overview of the major factors driving decisions to enhance nutrient removal at WWTPs. This section characterizes the industry based on U.S. Environmental Protection Agency (EPA) survey information. This section describes the negative impacts of nutrient enrichment, highlighting the history of water quality changes in key regions of the country. EPA and State initiatives to reduce nutrient pollution from wastewater treatment discharges are summarized in this training course. Lastly, we will highlight several barriers to enhancing nutrient removal at wastewater plants.

## Status of Wastewater Treatment in the U.S.

The 1972 Amendments to the Federal Water Pollution Control Act (FWPCA)(Public Law 92-500), also known as the Clean Water Act (CWA), established the foundation for wastewater discharge control in the U.S. The CWA's primary objective is to "restore and maintain the chemical, physical, and biological integrity of the Nation's waters." The CWA established a program to ensure clean water by requiring permits that limit the amount of pollutants discharged by all municipal and industrial dischargers into receiving waters. Discharges are regulated under the National Pollutant Discharge Elimination System (NPDES) permit program. As of 2004, there were 16,583 municipal wastewater utilities [also known as Publicly Owned Treatment Works (POTWs)] regulated under the CWA, serving approximately 75 percent of the Nation's population (U.S. Public Health Service and USEPA, 2008) with the remaining population served by septic or other onsite systems.

Wastewater treatment has generally been defined as containing one or more of the following four processes: (1) preliminary, (2) primary, (3) secondary, and (4) advanced - also known as tertiary treatment.

Preliminary treatment consists of grit removal, which removes dense inert particles and screening to remove rags and other large debris. Primary treatment involves gravity settling tanks to remove settleable solids, including settleable organic solids. The performance of primary settling tanks can be enhanced by adding chemicals to capture and flocculate smaller solid particles for removal and to precipitate phosphorus. Secondary treatment follows primary treatment in most plants and employs biological processes to remove colloidal and soluble organic matter. Effluent disinfection is usually included in the definition of secondary treatment.

EPA classifies advanced treatment as "a level of treatment that is more stringent than secondary or produces a significant reduction in conventional, non-conventional, or toxic pollutants present in the wastewater" (U.S. Public Health Service and USEPA, 2008). Other technical references subdivide advanced treatment, using the terms "secondary with nutrient removal" when nitrogen, phosphorus, or both are removed and "tertiary removal" to refer to additional reduction in solids by filters or microfilters (Tchobanoglous et al, 2003). Effluent filtration and nutrient removal are the most common advanced treatment processes. The CWA requires that all municipal wastewater treatment plant discharges meet a minimum of secondary treatment. Based on data from the *2004 Clean Watersheds Needs Survey*, 16,543 municipal WWTPs (99.8 percent of plants in the country) meet the minimum secondary waste-water treatment requirements.

Of those that provide at least secondary treatment, approximately 44 percent provide some kind of advanced treatment (U.S. Public Health Service and USEPA, 2008).

### **Nutrient Impairment of U.S. Waterways**

The harmful effects of eutrophication due to excessive nitrogen and phosphorus concentrations in the aquatic environment have been well documented. Algae and phytoplankton growth can be accelerated by higher concentrations of nutrients as they can obtain sufficient carbon for growth from carbon dioxide. In addition to stimulating eutrophication, nitrogen in the form of ammonia can exert a direct demand on dissolved oxygen (DO) and can be toxic to aquatic life. Even if a treatment plant converts ammonia to nitrate by a biological nitrification process, the resultant nitrate can stimulate algae and phytoplankton growth. Phosphorus also contributes to the growth of algae. Either nitrogen or phosphorus can be the limiting nutrient depending on the characteristics of the receiving water.

Nitrogen is typically limiting in estuarine and marine systems and phosphorus in fresh water systems. According to the 2007 report *Effects of Nutrient Enrichment in the Nation's Estuaries: A Decade of Change*, increased nutrient loadings promote a progression of symptoms beginning with excessive growth of phytoplankton and macroalgae to the point where grazers cannot control growth (Bricker et al., 2007). These blooms may be problematic, potentially lasting for months at a time and blocking sunlight to light-dependent submerged aquatic vegetation (SAV). In addition to increased growth, changes in naturally occurring ratios of nutrients may also affect which species dominate, potentially leading to nuisance/toxic algal blooms. These blooms may also lead to other more serious symptoms that affect biota, such as low DO and loss of SAV. Once water column nutrients have been depleted by phytoplankton and macroalgae and these blooms die, the bacteria decomposing the algae then consume oxygen, making it less available to surrounding aerobic aquatic life.

Consequently, fish and invertebrate kills may occur due to hypoxia and anoxia, conditions of low to no DO. Eutrophic conditions may also cause risks to human health, resulting from consumption of shellfish contaminated with algal toxins or direct exposure to waterborne toxins. Eutrophication can also create problems if the water is used as a source of drinking water. Chemicals used to disinfect drinking water will react with organic compounds in source water to form disinfection byproducts, which are potential carcinogens and are regulated by EPA.

Advanced eutrophic conditions can lead to "dead zones" with limited aquatic life, which describes the hypoxia condition that exists in the Northern Gulf of Mexico. A recent U.S. Geological Survey (USGS) report titled *Differences in Phosphorus and Nitrogen Delivery to the Gulf of Mexico from the Mississippi River Basin* documents the contribution of nitrogen and phosphorus from agricultural and non-agricultural sources in the Mississippi River basin (Alexander et al., 2008).

On June 16, 2008 the joint federal-state Mississippi River/Gulf of Mexico Watershed Nutrient Task Force released its *2008 Action Plan for Reducing, Mitigating, and Controlling Hypoxia in the Northern Gulf of Mexico and Improving Water Quality in the Mississippi River Basin*, which builds upon its 2001 plan by incorporating emerging issues, innovative approaches, and the latest science, including findings from EPA's Science Advisory Board.

Improvements include more accountability through an Annual Operating Plan, better tracking of progress, state and federal nutrient reduction strategies, and a plan to increase awareness of the problem and implementation of solutions (USEPA, 2008b). Nutrient pollution has also caused significant problems in the Chesapeake Bay. Elevated levels of both nitrogen and phosphorus are the main cause of poor water quality and loss of aquatic habitats in the Bay. Significant algae blooms on the water surface block the sun's rays from reaching underwater bay grasses. Without sunlight, bay grasses cannot grow and provide critical food and habitat for blue crabs, waterfowl, and juvenile fish.

The Chesapeake Bay Program estimates that 22 percent of the phosphorus loading and 19 percent of the nitrogen loading in the Bay comes from municipal and industrial wastewater facilities (Chesapeake Bay Program, 2008). The first national attention to nutrient contamination occurred in the Great Lakes. In the 1960s Lake Erie was declared “dead” when excessive nutrients in the Lake fostered excessive algae blooms that covered beaches and killed off native aquatic species due to oxygen depletion. At that time, phosphorus was the primary nutrient of concern due to the advent of phosphate detergents and inorganic fertilizers. With the enactment of the CWA and the Great Lakes Water Quality Agreement in 1972, a concerted effort was undertaken to reduce pollutant loadings, including phosphorus in the Lake.

Although the health of the Lake improved dramatically, in recent years, there has been renewed attention to the re-emergence of a “dead” zone in Lake Erie, again due to nutrient loadings. Recent studies by scientists and the National Oceanic and Atmospheric Administration (NOAA) have also hypothesized a relationship between excessive nutrients in the Lake and the presence of two aquatic invasive species – the zebra mussel and the quagga mussel (Vanderploeg et al., 2008). Development and population increases in the Long Island Sound Watershed have resulted in a significant increase in nitrogen loading to the Sound. The increased nitrogen loads have stimulated plant growth, increased the amount of organic matter settling to the benthic zone, lowered DO levels, and changed habitats.

The primary concerns in the Sound include hypoxia, the loss of sea grass, and alterations in the food web. Management efforts are currently underway to reduce nitrogen pollution by more than half with a focus on upgrading WWTPs with new technologies and removing nitrogen by reducing polluted run-off through best management practices on farms and suburban areas (Long Island Sound Study, 2004). The above represent four examples of impaired large water bodies impacted by nutrient loadings. There are more than 80 additional estuaries and bays, and thousands of rivers, streams, and lakes that are also impacted by nutrients in the U.S. In fact, all but one state and two territories have CWA section 303(d) listed<sup>1</sup> water body impairments for nutrient pollution. Collectively, states have listed over 10,000 nutrient and nutrient-related impairments.

Climate change may also be a significant influence on the development of future eutrophic symptoms. According to the report *Effects of Nutrient Enrichment in the Nation’s Estuaries: A Decade of Change*, the factors associated with climate change that are expected to have the greatest impacts on coastal eutrophication are:

- Increased temperatures
- Sea level rise
- Changes in precipitation and freshwater runoff

Increased temperatures will have several effects on coastal eutrophication. Most coastal species are adapted to a specific range of temperatures. Increases in water temperatures may lead to expanded ranges of undesirable species. Higher temperatures may also lead to increased algal growth and longer growing seasons, potentially increasing problems associated with excessive algal growth and nuisance/toxic blooms. Additionally, warmer waters hold less DO, therefore potentially exacerbating hypoxia. Temperature-related stratification of the water column may also worsen, having a further negative effect on DO levels.

Climate change models predict increased melting of polar icecaps and changes in precipitation patterns, leading to sea level rise and changes in water balance and circulation patterns in coastal systems. Sea level rise will gradually inundate coastal lands, causing increased erosion and sediment delivery to water bodies, and potentially flooding wetlands. The increased sediment load and subsequent turbidity increase may cause SAV loss. The positive feedback between increased erosion and algal growth (as erosion increases, sediment associated nutrients also increase, stimulating growth) may also increase turbidity. The loss of wetlands, which act as nutrient sinks, will further increase nutrient delivery to estuaries.

Another report titled *Aquatic Ecosystems and Global Climate Change – Potential Impacts on Inland Freshwater and Coastal Wetland Ecosystems in the United States* notes that climate change of the magnitude projected for the U.S. over the next 100 years will cause significant changes to temperature regimes and precipitation patterns across the U.S. (Poff et al., 2002). Such alterations in climate pose serious risks for inland freshwater ecosystems (lakes, streams, rivers, wetlands) and coastal wetlands, and may adversely affect numerous critical services provided to human populations.

These conclusions indicate climate change is a significant threat to the species composition and function of aquatic ecosystems in the U.S. However, critical uncertainties exist regarding the manner in which specific species and whole ecosystems will respond to climate change. These arise both from uncertainties about how regional climate will change and how complex ecological systems will respond.

Indeed, as climate change alters ecosystem productivity and species composition, many unforeseen ecological changes are expected that may threaten the goods and services that these systems provide to humans. Required by Section 303(d) of the CWA, the 303(d) list is a list of state's water bodies that do not meet or are not expected to meet applicable Water Quality Standards with technology-based controls alone.

### **Federal and State Initiatives to Reduce Nutrient Pollution**

#### **NPDES Permitting**

Established by the FWPCA Amendment of 1972, EPA's NPDES permit program has been the primary mechanism for controlling pollution from point sources. Point sources are discrete conveyances such as pipes or man-made ditches. Individual homes that are connected to a municipal system, use a septic system, or do not have a surface discharge do not need an NPDES permit; however, POTWs and other facilities must obtain permits if they discharge directly to surface waters.

NPDES permits for wastewater discharges contain, among other information, effluent limits for "conventional" pollutants such as biochemical oxygen demand (BOD), total suspended solids (TSS), and pH as well as limits for specific toxicants including various organic and inorganic chemicals. Permits may also include effluent limits for "non-conventional" pollutants such as nitrogen and phosphorus. Effluent limits can be technology-based and/or water-quality based. EPA has established technology-based, secondary treatment effluent limits for BOD as 5-day biochemical oxygen demand (BOD5), TSS, and pH.

Water-quality based effluent limits are set if the technology-based limits are not sufficient to maintain the water quality standards (WQS) of the receiving water. Federal and State regulations related to WQSs and Total Maximum Daily Loads (TMDLs) are expected to drive down NPDES effluent limits for nitrogen and phosphorus. WQS define the goals for a water body by designating its uses, setting criteria to protect those uses, and establishing provisions to protect water bodies from pollutants. Criteria can be narrative or numeric.

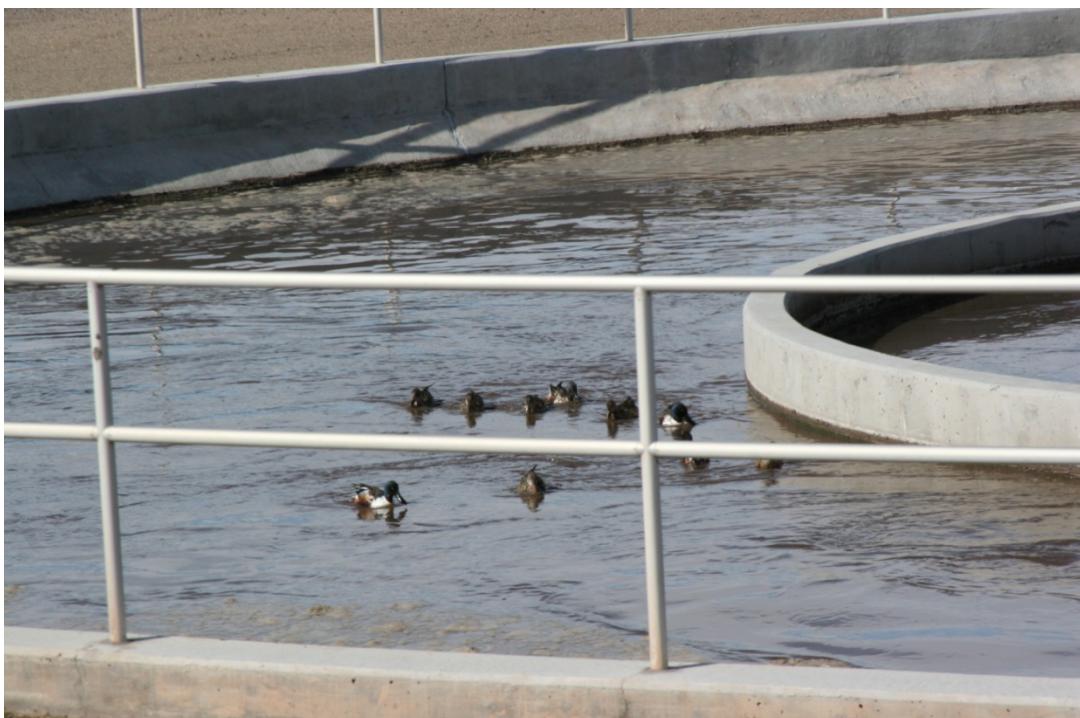
Regulatory agencies can adopt *nutrient criteria* to protect a water body against nutrient over-enrichment and eutrophication caused by nitrogen and phosphorus. In June 1998, EPA issued a *National Strategy for the Development of Regional Nutrient Criteria*. This was followed by publication of recommended nutrient criteria for most streams and lakes in 2001. In a January 9, 2001 *Federal Register* notice, EPA recommended that states and other regulatory agencies develop a nutrient criteria plan to outline their process for adopting such nutrient criteria (*Federal Register*, 2001).

As of May 2007, only a handful of States and Territories had adopted nutrient criteria for nitrogen and phosphorus (USEPA, 2007a), although many have made progress in criteria development.

In a memo dated May 25, 2007, EPA encouraged all regulatory agencies to "...accelerate their efforts and give priority to adopting numeric nutrient standards or numeric translators for narrative standards for all waters in States and Territories that contribute nutrient loadings to our waterways" (USEPA, 2007b).

CWA Section 303(d) requires states to develop TMDLs for water bodies on the 303(d) list of impaired waters. A TMDL is a calculation of the maximum amount of a pollutant a water body can receive and still meet WQS. TMDLs serve as a tool for implementing WQS. The TMDL targets or endpoints represent a number where the applicable WQS and designated uses (e.g., such as public water supply, contact recreation, and the propagation and growth of aquatic life) are achieved and maintained in the water body of concern.

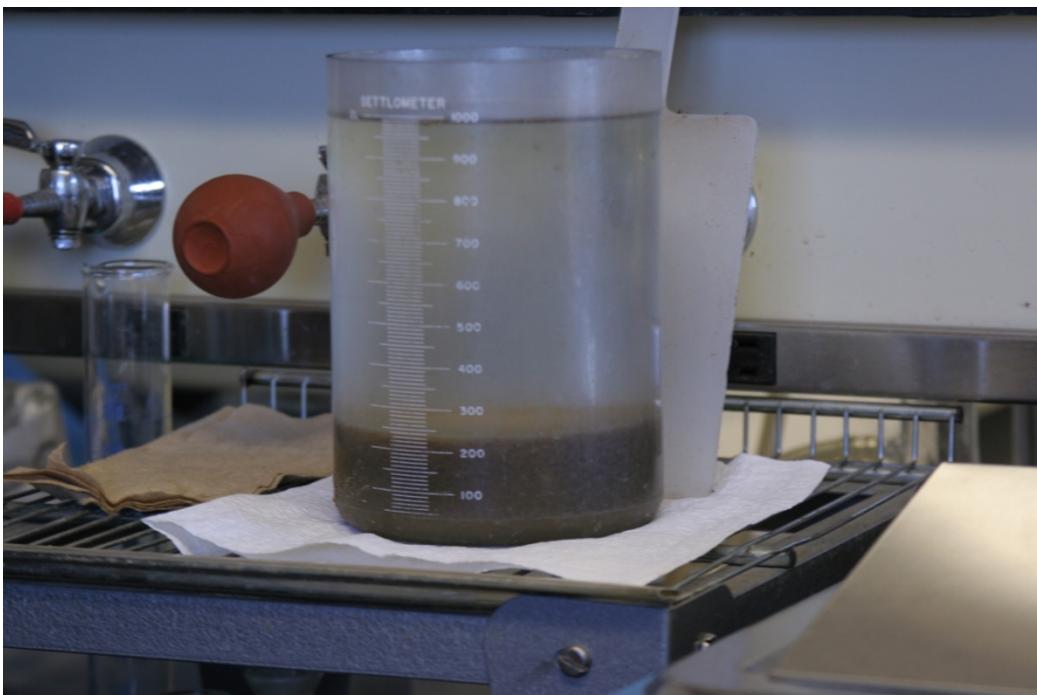
TMDLs identify the level of pollutant control necessary to meet WQS and support the designated uses of a water body. Once a TMDL is set, the total load is allocated among all existing sources. The allocation is divided into two portions - a load allocation representing natural and non-point sources and a waste load allocation representing NPDES permitted point source discharges. In many regions, water bodies have a poor ability to assimilate nutrients or water bodies are already impaired from past pollution and the water body cannot handle large loads of additional nutrients. In these cases, TMDLs may require nutrient permit levels to be even lower than what might be allowed otherwise by nutrient criteria.



No need for operators at the WWT facility, this oxidation ditch is overseen by ducks. Lucky for them the operators do a great job. It is very common to have all types of waterfowl and birds at your facility. I've seen Eagles to Cranes, EVEN BEARS.



The spinning reel for this oxidation ditch is mixing or aerating properly.



This is a 1000 ml settlometer used to determine the Sludge Volume Index (SVI). Increase sludge wasting to decrease MCRT; this may prevent sludge from floating to the surface of a secondary clarifier. Sludge that is rising to the top of the clarifier is a good indication that sludge is not being removed from the primary clarifier often enough.

## Water Quality Trading

Water quality trading is a market-based approach to improve and preserve water quality. Trading can provide greater efficiency in achieving water quality goals by allowing one source to meet its regulatory obligations by using pollutant reductions created by another source that has lower pollution control costs. For example, under a water quality trading program, a POTW could comply with discharge requirements by paying distributed sources to reduce their discharges by a certain amount. The use of geographically-based trading ratios provides an economic incentive, encouraging action toward the most cost effective and environmentally beneficial projects.

EPA issued a Water Quality Trading Policy in 2003 to provide guidance to States and Tribes on how trading can occur under the CWA and its implementing regulations. The policy discusses CWA requirements that are relevant to water quality trading including: requirements to obtain permits, antibacksliding provisions, development of WQSs including an antidegradation policy, NPDES permit regulations, TMDLs and water quality management plans. EPA also developed a number of tools and guidance documents to assist states, permitted facilities, non-point sources, and stakeholders involved in the development of trading programs ([www.epa.gov/owow/watershed/trading.htm](http://www.epa.gov/owow/watershed/trading.htm)). Recently, the U.S. Department of Agriculture (USDA) National Resources Conservation Service released a Nitrogen Trading Tool (NTT) prototype for calculating nitrogen credits based on the Nitrogen Loss and Environmental Assessment Package Model (Gross et al., 2008).

Water quality trading programs have been successfully implemented in several states and individual watersheds across the country. For example, nitrogen pollution from point sources into the Long Island Sound was reduced by nearly 25 percent using an innovative Nitrogen Credit Trading Program. In Connecticut, the program was implemented among 79 sewage treatment plants in the state. Through the Nitrogen Credit Exchange, established in 2002, the Connecticut program has a goal of reducing nitrogen discharges by 58.5 percent by 2014. A recent American Society of Civil Engineers journal article points out, however, that regulatory frameworks for water quality trading programs have yet to be adopted by the majority of States. Barriers to adopting such programs include uncertainty in: (1) the mechanisms for determining appropriate credits and ratios between point sources and distributed sources; and (2) approaches to ensure that promised reductions actually occur (Landers, 2008).



Aeration is often used to refresh the wastewater flow at the influent channel.



The photograph above and below are of an operator taking mixed liquor samples in an oxidation ditch. Always wear latex gloves, many operators quit wearing gloves after a short period of time. Don't become unafraid of the pathogens. Fear the bugs no matter what.



## Nutrient Constituents in Wastewater and Measurement Methods

This section provides an overview of the sources, forms, and measurement methods for nitrogen and phosphorus in wastewater.

### Nitrogen

Nitrogen is an essential nutrient for plants and animals. Approximately 80 percent of the earth's atmosphere is composed of nitrogen and it is a key element of proteins and cells. The major contributors of nitrogen to wastewater are human activities such as food preparation, showering, and waste excretion. The per capita contribution of nitrogen in domestic wastewater is about 1/5th of that for BOD. Total nitrogen in domestic wastewater typically ranges from 20 to 70 mg/L for low to high strength wastewater (Tchobanoglous et al., 2003). Factors affecting concentration include the extent of infiltration and the presence of industries. Influent concentration varies during the day and can vary significantly during rainfall events, as a result of inflow and infiltration to the collection system.

**The most common forms of nitrogen in wastewater are:**

- Ammonia ( $\text{NH}_3$ )
- Ammonium ion ( $\text{NH}_4^+$ )
- Nitrite ( $\text{NO}_2^-$ )
- Nitrate ( $\text{NO}_3^-$ )
- Organic nitrogen

Nitrogen in domestic wastewater consists of approximately 60 to 70 percent ammonia-nitrogen and 30 to 40 percent organic nitrogen (Tchobanoglous et al., 2003; Crites and Tchobanoglous, 1998). Most of the ammonia-nitrogen is derived from urea, which breaks down rapidly to ammonia in wastewater influent. EPA approved methods for measuring ammonia, nitrate, and nitrite concentration use colorimetric techniques. Organic nitrogen is approximated using the standard method for Total Kjeldahl Nitrogen (TKN) (APHA, AWWA, and WEF, 1998).

**The TKN method has three major steps:**

- (1) digestion to convert organic nitrogen to ammonium sulfate;
- (2) conversion of ammonium sulfate into condensed ammonia gas through addition of a strong base and boiling; and
- (3) measurement using colorimetric or titration methods. Because the measured concentration includes ammonia, the ammonia-nitrogen concentration is subtracted from the TKN to determine organic nitrogen.

Nitrogen components in wastewater are typically reported on an "as nitrogen" basis so that the total nitrogen concentration can be accounted for as the influent nitrogen components are converted to other nitrogen compounds in wastewater treatment.

WWTPs designed for nitrification and denitrification can remove 80 to 95 percent of inorganic nitrogen, but the removal of organic nitrogen is typically much less efficient (Pehlivanoglu-Mantash and Sedlak, 2006). Domestic wastewater organic nitrogen may be present in particulate, colloidal or dissolved forms and consist of proteins, amino acids, aliphatic N compounds, refractory natural compounds in drinking water (e.g. Humic substances), or synthetic compounds (e.g. ethylene Diamine tetraacetic acid (EDTA)). Organic nitrogen may be released in secondary treatment by microorganisms either through metabolism or upon death and lysis. Some nitrogen may be contained in recondensation products. Hydrolysis of particulate and colloidal material by microorganisms releases some organic nitrogen as dissolved, biodegradable compounds. Amino acids are readily degraded during secondary biological treatment, with 90 to 98 percent removal in activated sludge systems and 76 to 96 percent removal in trickling filters. However, other forms of organic nitrogen may be more persistent in wastewater treatment processes.

The importance of organic nitrogen has increased as effluent limits on nitrogen have become more stringent. With more impaired waterways from nutrient loads, effluent limits for total nitrogen (TN) concentrations of 3.0 mg/L or less are becoming more common. The dissolved organic nitrogen (DON) concentration in the effluent from biological nutrient removal treatment facilities was found to range from 0.50 to 1.50 mg/L in 80 percent of 188 plants reported by Pagilla (STAC-WERF, 2007) and values as high as 2.5 mg/L were observed. Thus, for systems without effluent filtration or membrane bioreactors (MBRs) that are trying to meet a TN treatment goal of 3.0 mg/L, the effluent DON contribution can easily be 20 to 50 percent of the total effluent nitrogen concentration, compared to only about 10 percent for conventional treatment (Pehlivanoglu-Mantas and Sedlak, 2004).

The chemical composition of DON in wastewater effluents is not completely understood. Sedlak (2007) has suggested that only about 20 percent of the DON has been identified as free and combined amino acids, EDTA, and other trace nitrogen compounds. About 45 percent may be unidentified low molecular weight compounds and the other 35 percent as unidentified high molecular weight compounds containing Humic acids and amides. Similar results were found by Khan (2007). Early work by Parkin and McCarty (1981) suggested that 40 to 60 percent of effluent DON is non-bioavailable. The non-bioavailable portion is also referred to as recalcitrant DON (rDON).



Primary clarifier

## Phosphorus

Total phosphorus (TP) in domestic wastewater typically ranges between 4 and 8 mg/L but can be higher depending on industrial sources, water conservation, or whether a detergent ban is in place. Sources of phosphorus are varied. Some phosphorus is present in all biological material, as it is an essential nutrient and part of a cell's energy cycle. Phosphorus is used in fertilizers, detergents, and cleaning agents and is present in human and animal waste.

### Phosphorus in wastewater is in one of three forms:

- Phosphate (also called Orthophosphate)
- Polyphosphate, or
- Organically bound phosphorus.

The orthophosphate fraction is soluble and can be in one of several forms (e.g., phosphoric acid, phosphate ion) depending on the solution pH. Polyphosphates are high-energy, condensed phosphates such as pyrophosphate and trimetaphosphate. They are also soluble but will not be precipitated out of wastewater by metal salts or lime. They can be converted to phosphate through hydrolysis, which is very slow, or by biological activity.

Organically bound phosphorus can either be in the form of soluble colloids or particulate. It can also be divided into biodegradable and non-biodegradable fractions. Particulate organically bound phosphorus is generally precipitated out and removed with the sludge. Soluble organically bound biodegradable phosphorus can be hydrolyzed into orthophosphate during the treatment process.

Soluble organically bound non-biodegradable phosphorus will pass through a wastewater treatment plant. A typical wastewater contains 3 to 4 mg/L phosphorus as phosphate, 2 to 3 mg/L as polyphosphate, and 1 mg/L as organically bound phosphorus (WEF and ASCE, 2006).

### Phosphorus content in wastewater can be measured as

- Orthophosphate
- Dissolved orthophosphate
- Total phosphorus
- Total dissolved phosphorus (i.e., all forms except particulate organic phosphorus)

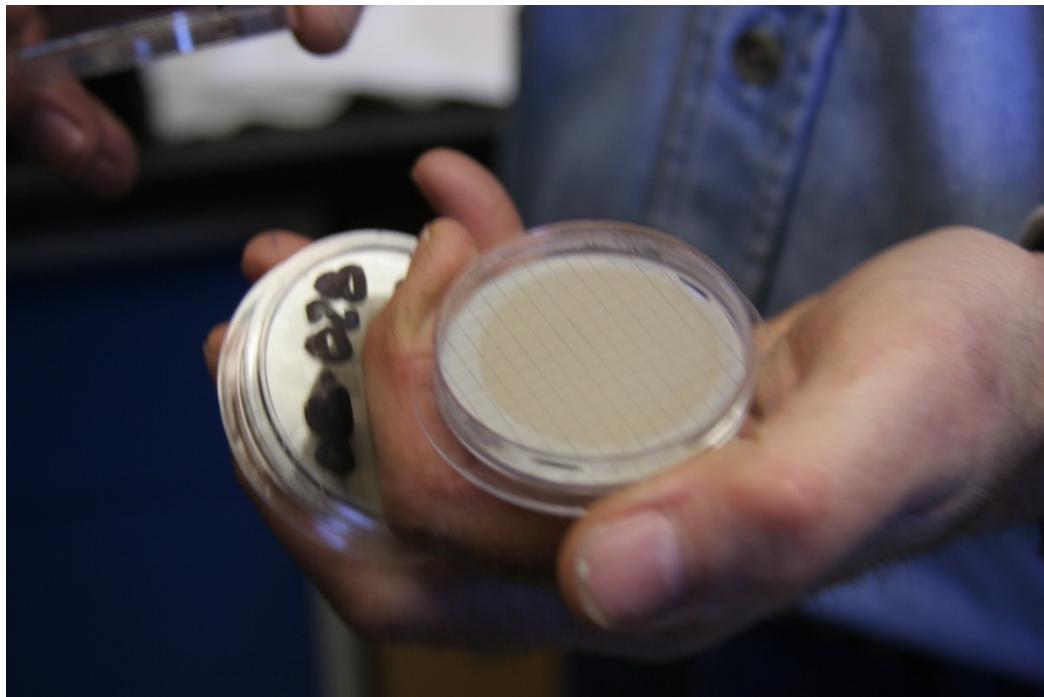
EPA approved laboratory methods rely on colorimetric analysis. Colorimetric analysis measures orthophosphate only, so a digestion step is needed to convert polyphosphate and organic phosphorus to orthophosphate to measure TP. The persulfate method is reported to be the most common and easiest method (WEF and ASCE, 2006). To determine dissolved phosphorus (either total dissolved phosphorus or total dissolved orthophosphate), the sample is first filtered through a 0.45 micron filter.

USEPA approved colorimetric methods are routinely used to measure phosphorus levels as low as 0.01 mg/L. On-line analyzers that use the colorimetric method are available from vendors (e.g., the Hach Phosphax™ SC phosphate analyzer).

Ion chromatography is a second common technique used to measure orthophosphate in wastewater. As with colorimetric methods, digestion is required for TP measurement, with persulfate digestion recommended (WEF and ASCE, 2006).



Microscopes are used to see indicator bugs and other MO's microorganisms. This examination is used so that the operator knows how well the process is working.



This is a filter used for the coliform test.  
**Phosphorus Removal by Chemical Addition**

The purpose of this section is to describe techniques for phosphorus removal by chemical addition. It summarizes issues associated with chemical feed location, mixing, and sludge production. An overview of advanced solids separation processes is also provided.

### **Principles**

Chemical precipitation for phosphorus removal is a reliable, time-tested, wastewater treatment method that has not drastically changed over the years. To achieve removal, various coagulant aids are added to wastewater where they react with soluble phosphates to form precipitates. The precipitates are removed using a solids separation process, most commonly settling (clarification). Chemical precipitation is typically accomplished using either lime or a metal salt such as aluminum sulfate (alum) or ferric chloride. The addition of polymers and other substances can further enhance floc formation and solids settling. Operators can use existing secondary clarifiers or retrofit primary clarifiers for their specific purposes.

### **Aluminum and Iron Salts**

Alum and ferric or ferrous salts are commonly used as coagulant and settling aids in both the water and wastewater industry. They are less corrosive, create less sludge, and are more popular with operators compared to lime. Alum is available in liquid or dry form, can be stored on site in steel or mild concrete, and has a near unlimited shelf life. Ferric chloride is similar although care is needed during handling because of corrosivity. If an industrial source is available such as waste pickle liquor, ferrous chloride or ferrous sulfate have been used for phosphorus removal. Ferrous forms should be added directly to aerobic reactors rather than to anaerobic reactors such as primary settling basins because the ferrous iron needs to oxidize to ferric iron for best results.

The molar ratio of aluminum to phosphorus required for phosphorus removal ranges from about 1.38:1 for 75 percent removal, 1.72:1 for 85 percent removal, and 2.3:1 for 95 percent removal.

For iron compounds, a ratio of about 1:1 is required, with a supplemental amount of iron (10 mg/L) added to satisfy the formation of hydroxide (WEF and ASCE, 1998). For additional removal of phosphorus with aluminum and iron salts, a ratio of between 2 and 6 parts metal salt to 1 part phosphorus may be required for adequate phosphorus removal.

To supplement stoichiometry calculations, designers should consider jar tests and, in some cases, full-scale pilot tests to gauge the effects on the required dose of competing reactions; the influence of pH and alkalinity, adsorption, and co-precipitation reactions; and the interaction with polymers that are added to increase coagulation and flocculation (WEF and ASCE, 1998; Bott et al. 2007).

Aluminum or ferric iron salts can be added to the primary clarifier, secondary clarifier, tertiary clarifier, or directly into the activated sludge aeration tank. Multiple additions can increase phosphorus removal efficiency. Ferrous salts can only be added to the aeration basin since it needs to be oxidized to ferric to precipitate the phosphorus.

The solubility of aluminum and iron salts is a function of pH. The optimum solubility for alum was previously reported to occur at a pH range of 5.5 to 6.5, significantly lower than most influent wastewater. Recent studies (Szabo et al., 2008) showed that the range for both iron and alum is between 3.5 and 7.5 with the highest efficiency between pH 5.5 and 7.

Chemicals such as lime compounds, caustic soda, and soda ash can be used to raise the pH of the waste stream prior to biological treatment processes or discharge. It is important to understand that alkalinity is consumed during the precipitation reactions, and precipitation will be incomplete if insufficient alkalinity is present.

### Lime

Although lime had lost favor due to issues associated with chemical handling, sludge production, and re-carbonation, it has recently been considered more often because of its ability to reduce phosphorus to very low levels when combined with effluent filtration and the microbial control properties associated with its high pH. When lime is added to wastewater, it first reacts with the bicarbonate alkalinity to form calcium carbonate ( $\text{CaCO}_3$ ). As the pH increases to more than 10, excess calcium ions will react with phosphate to precipitate hydroxylapatite [ $\text{Ca}_5(\text{OH})(\text{PO}_4)_3$ ].

Because it reacts first with alkalinity, the lime dose is essentially independent of the influent phosphorus concentration. Tchobanoglous et al. (2003) estimates the lime dose to typically be 1.4 to 1.6 times the total alkalinity expressed as  $\text{CaCO}_3$ .

The typical reaction between calcium compounds and phosphorus is represented below:



The molar ratio required for phosphorus precipitation with lime is approximately 5:3, but can vary from between 1.3 to 2, depending on the composition of the wastewater. As with iron and aluminum salts, jar tests can be used to determine correct doses for a specific wastewater stream (WEF, 1998).

Lime addition can raise the pH to greater than 11. Because activated sludge processes require pH levels below 9, lime cannot be added directly to biological treatment processes or it will cause process upsets. Lime can be added to primary sedimentation tanks and removed with the primary sludge or it can be added as a tertiary treatment process after biological treatment. When added to primary tanks, it will also result in the removal of colloidal material through coagulation and settling, with a concomitant removal of TSS up to 80 percent and chemical oxygen demand (COD) up to 60 percent.

In either case, pH adjustment is needed and typically accomplished by adding  $\text{CO}_2$  or a liquid acid such as sulfuric acid, nitric acid, or hypochlorite (Tchobanoglous et al., 2003; USEPA, 1999a).

Hortskotte et al. (1974) showed that when the primary effluent is discharged directly to a nitrifying activated sludge plant, the hydrogen ions produced may neutralize the high pH. However, when denitrification is practiced and the operator wishes to make use of the soluble COD in the primary effluent, the effluent must be neutralized before discharging it to the anoxic zone.

Lime requires special handling and operations practices that further set it apart from chemical precipitation by metal salts. Although the formation of carbonate scaling on equipment and pipes is a drawback of lime treatment, lime slaking, where quicklime ( $\text{CaO}$ ) is reacted with water to form calcium hydroxide ( $\text{Ca}(\text{OH})_2$ ), is the biggest operational disadvantage.

## Location of Chemical Feed and Mixing

Lime or metal salts can be added at several locations throughout the treatment plant to remove phosphorus.

“Pre-precipitation” is when chemicals are added to raw water to precipitate phosphorus in the primary sedimentation basins.

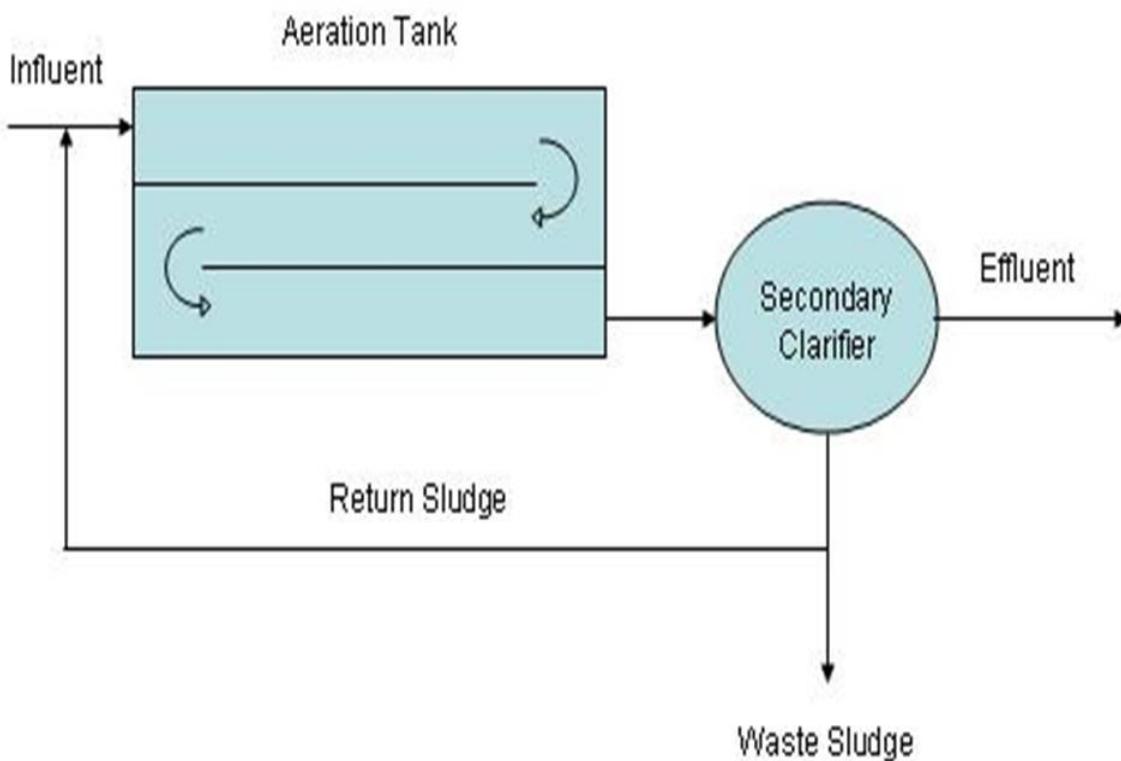
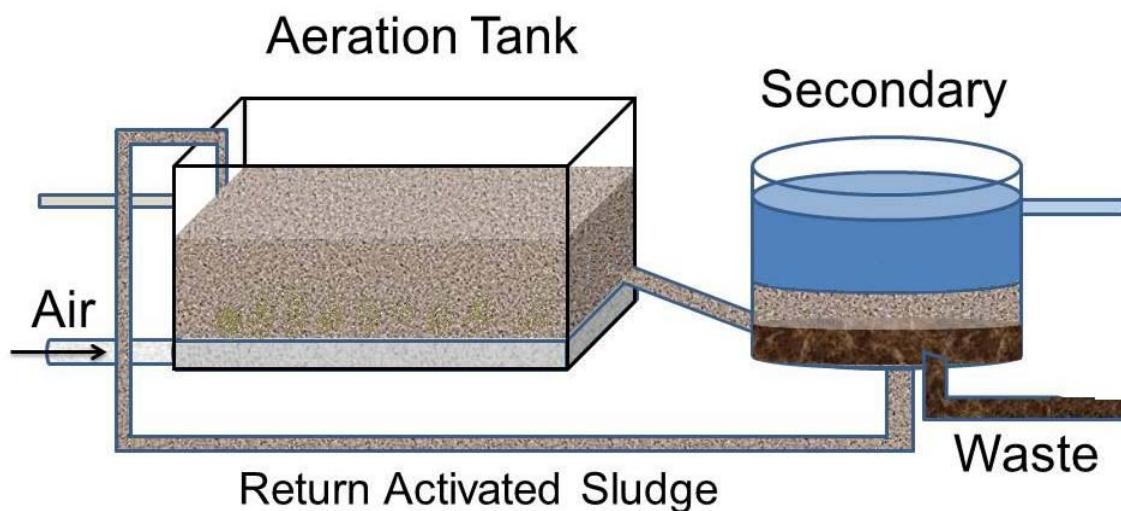
“Co-precipitation” involves adding chemicals to form precipitates that can be removed with biological sludge.

“Post-precipitation” is when chemicals are added after secondary sedimentation and precipitants are removed in a tertiary process such as sedimentation or filtration (Tchobanoglous et al., 2003). Because it requires a high pH to achieve a low phosphorus concentration, lime cannot be added directly to biological reactors or to the secondary clarifiers.

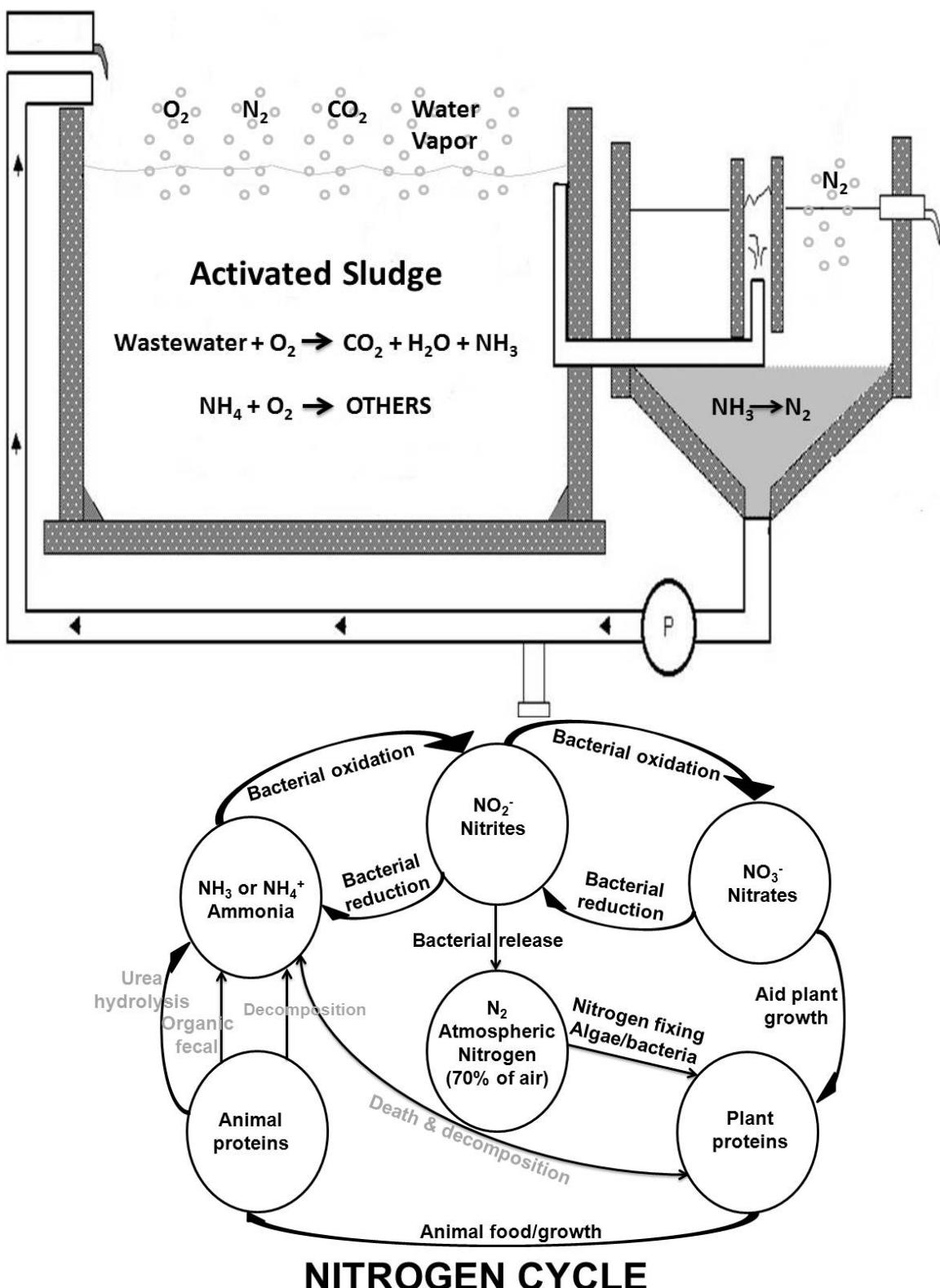
Multipoint additions of iron or aluminum salts have been very effective and can typically remove more phosphorus than single-point applications. There are several advantages to post-precipitating phosphorous using a tertiary treatment technique (after biological processes in a separate reactor):

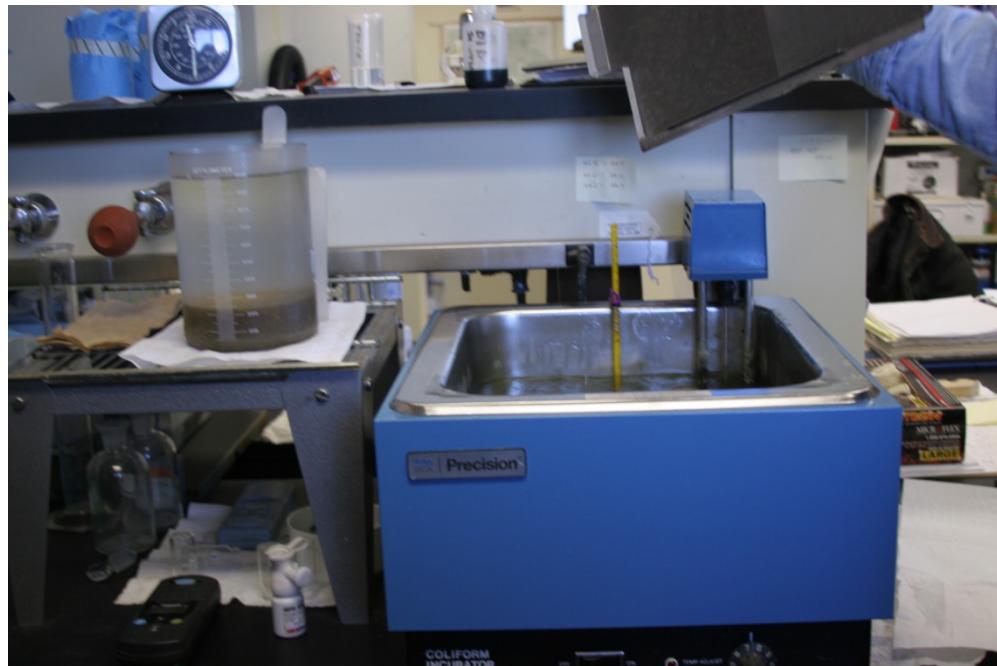
- Microorganisms rely on phosphorus as a food source. If too much phosphorus is removed prior to biological treatment, biological processes may suffer. For activated sludge, the minimum ratio of phosphorus to BOD5 for a rapidly growing (low solids retention time (SRT)) system is typically about 1:100 (WEF and ASCE, 1998).
- Competing chemicals in the primary sedimentation basins can increase the required dose.
- Phosphorus enters the treatment plant as soluble orthophosphate, soluble polyphosphates, and organically bound phosphorus. Most of the polyphosphates and much of the organically bound phosphorus are converted to more simple orthophosphates during biological treatment. If the influent contains significant polyphosphates and/or organically bound phosphorus, locating chemical treatment after biological processes would be more efficient and achieve lower effluent levels.
- The removal of carbonate alkalinity and phosphorus by lime prior to biological treatment can have a negative impact on nitrification processes (WEF and ASCE, 1998). Also, removing phosphorus to very low concentrations upstream of denitrification filters can negatively affect the denitrification process. Previous studies showed that the hydroxide alkalinity can be balanced by the hydrogen ions produced during nitrification.
- Sludge recalcification can be used to achieve high removal efficiencies using lime in tertiary treatment. One potential advantage to adding chemicals during primary treatment instead of tertiary treatment is reduced capital costs and space requirements as a result of removing additional BOD and TSS and reducing the load to downstream processes, thereby reducing the size of the subsequent activated sludge basins and the amount of oxygen transfer needed.

Chemicals should be well mixed with the wastewater to ensure reaction with soluble phosphates and formation of precipitates. Chemicals may either be mixed in separate tanks or can be added at a point in the process where mixing already occurs. Bench-scale and pilot scale tests are often used to determine the correct mixing rate for a given composition of wastewater and chemicals used, including polymer (USEPA, 1999a).



## Plug Flow Activated Sludge Process





Here is an incubator for the coliform test. The operator will place the sample in this device for 24 to 48 hours depending on the desired results. There are several different methods to calculate coliform bacteria. This is an older true and tested method.



This glass bottle is used for quality control (QA/QC) for bacteria samples tubes.

## Advanced Solids Separation Processes

The effectiveness of phosphorus removal by chemical addition is highly dependent on the solids separation process following chemical precipitation. Direct addition of metal salts to activated sludge processes followed by conventional clarification can typically remove TP to effluent levels between 0.5 and 1.0 mg/L (Bott et al., 2007). Tertiary processes (post-secondary treatment) can be used to remove phosphorus to very low (< 0.1 mg/L) concentrations. For example, Reardon (2005) reports that four WWTP with tertiary clarifiers achieved TP levels of between 0.032 and 0.62 mg/L. Two common tertiary processes are clarification and effluent filtration. These approaches can be used separately or in combination. The next section presents a detailed discussion of effluent filtration. Advances in tertiary clarification processes are discussed below.

The types of clarifiers used for tertiary processes include conventional, one or two-stage lime, solids-contact, high-rate, and ballasted high-rate (BHRC). Several patented BHRC using different types of ballast such as recycled sludge, microsand, and magnetic ballast (USEPA, 2008a) have been developed in recent years. The advantages of high-rate clarification are that the clarifiers have a smaller footprint and are able to treat larger quantities of wastewater in a shorter period of time. In addition, as an add-on during wet weather, they can help prevent sanitary sewer overflows (SSOs) and combined sewer overflows (CSOs).

**The following patented processes are examples of high rate clarification including performance estimates:**

- DensaDeg® uses a coagulant in a rapid mix basin to destabilize suspended solids. The water flows into a second tank where polymer (for aiding flocculation) and sludge are added. The sludge acts as the “seed” for formation of high density floc. This floc is removed in settling tubes (USEPA, 2008). The main advantages of this process are a smaller footprint and denser sludge which is easier to dewater. Pilot testing for City of Fort Worth, Texas found a phosphorus removal rate of 88-95% for DensaDeg® (USEPA, 2003).
- Actiflo® uses a coagulant in a rapid mix basin to destabilize suspended solids. The water flows to a second tank where polymer (for aiding flocculation) and microsand are added. Microsand provides a large surface onto which suspended solids attach, creating a dense floc that settles out quickly. Clarification is assisted by lamella settling. Product pilot testing in Fort Worth, Texas showed a phosphorus removal efficiency of 92-96% for Actiflo®(USEPA, 2003).
- The CoMag process uses the addition of magnetic ballast with metal salts to promote floc formation. Settling is followed by high gradient magnetic separation for effluent polishing and recovery of the magnetic ballast (USEPA, 2008a). CoMag is currently in operation at a 4.0 million gallons per day (MGD) wastewater treatment plant in Concord, Massachusetts. The vendor has guaranteed an effluent phosphorus concentration not to exceed 0.05 mg/L (EPA Region 10, 2007).

### Other Design and Operational Issues

Phosphorus removal by chemical addition is limited to the soluble phosphates in the waste stream. Organically bound phosphorus and polyphosphates will not be removed by chemical treatment unless they are coagulated with the chemicals and removed in the sludge. Chemicals can be added after biological treatment to capitalize on the conversion of polyphosphates and organically bound phosphorus to phosphates by microorganisms in activated sludge.

The success of phosphorus removal by chemical addition depends on proper instrumentation and control. Dosage control typically takes the form of manual operation (for small systems), adjustments based on automatic flow measurements, or the more advanced on-line analyzers with computer-assisted dosage control.

Chemical properties of any water used for making solutions should be considered – tap water high in suspended solids could cause sludge to form when mixed with coagulants (WEF and ASCE, 1998) and could lead to clogging of chemical feed lines. Smith et al. (2008) found that factors such as pH, complexation, mixing, and the coagulant used can limit the removal of phosphorus, especially in the range of <0.1 mg/L.

### **Impacts on Sludge Handling and Production**

Sludge handling and production is generally considered to be one of the main downsides of chemical addition. Chemical precipitation methods always produce additional solids due to generation of metal- or calcium- phosphate precipitates and additional suspended solids (WEF and ASCE, 1998). Chemically treated sludge has a higher inorganic content compared to primary and activated sludge and will increase the required size of aerobic and anaerobic digesters. Additional sludge production can be estimated using reaction equations. The use of metal salts can result in increased inorganic salts (salinity) in the sludge and in the effluent.

Salinity can create problems when biosolids are land applied or when the effluent is returned to existing water supply reservoirs. Biological phosphorus removal was developed in South Africa due to the high rate of indirect recycling of wastewater effluent which led to excessive total dissolved solids (TDS) during dry periods. High total salts can reduce germination rates for crops and negatively affect the soil structure.

Lime traditionally produces a higher sludge volume compared to metal salts because of its reaction with natural alkalinity. An advantage of lime sludge is that some stabilization can occur due to the high pH levels required. One disadvantage is that lime can cause scaling in mechanical thickening and dewatering systems. There are also differences in the amount and characteristics of sludge generated by alum versus ferric salts. Although alum tends to produce less sludge than do ferric salts, alum sludge can be more difficult to concentrate and dewater compared to ferric sludge.

### **Biological Nitrogen Removal**

This section provides an overview of the principles behind biological nitrogen removal and describes the common design configurations in use today. It identifies key operational and design issues (including impacts on sludge handling and production), provides general guidelines on process selection, and summarizes ongoing research efforts in this area. Process configurations that are designed to remove both nitrogen and phosphorus are described latter.

### **Principles**

In wastewater treatment, nitrogen removal occurs in two sequential processes: nitrification and denitrification. An overview of each process is provided below.

#### **Nitrification**

Nitrification is an aerobic process in which autotrophic bacteria oxidize ammonia or nitrite for energy production. Nitrification is normally a two-step aerobic biological process for the oxidation of ammonia to nitrate. Ammonia-nitrogen (NH<sub>3</sub>-N) is first converted to nitrite (NO<sub>2</sub><sup>-</sup>) by ammonia oxidizing bacteria (AOB). The nitrite produced is then converted to nitrate (NO<sub>3</sub><sup>-</sup>) by nitrite oxidizing bacteria (NOB). Both reactions usually occur in the same process unit at a wastewater treatment plant (e.g. activated sludge mixed liquor or fixed film biofilm).

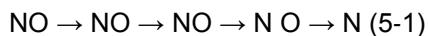
The group of AOB most associated with nitrification is the *Nitrosomonas* genus, although other AOB such as *Nitrosococcus* and *Nitrosospira* can contribute to the process. *Nitrobacter* are the NOB most associated with the second step, although other bacteria including *Nitrospina*, *Nitrococcus*, and *Nitrospira* have been found to also oxidize nitrite (Tchobanoglous et al., 2003; USEPA, 2007c).

AOB and NOB are classified as autotrophic bacteria because they derive energy from the oxidation of reduced inorganic compounds (in this case, nitrogenous compounds) and use inorganic carbon (CO<sub>2</sub>) as a food source. Nitrifying bacteria require a significant amount of oxygen to complete the reactions, produce a small amount of biomass, and cause destruction of alkalinity through the consumption of carbon dioxide and production of hydrogen ions. For each gram (g) of NH<sub>3</sub>-N converted to nitrate, 4.57 g of oxygen are used, 0.16 g of new cells are formed, 7.14 g of alkalinity are removed, and 0.08 g of inorganic carbon are utilized in formation of new cells (Tchobanoglous et al., 2003).

Nitrifying bacteria grow slower and have much lower yields as a function of substrate consumed, compared to the heterotrophic bacteria in biological treatment processes. The maximum specific growth rate of the nitrifying bacteria is 10 to 20 times less than the maximum specific growth rate of heterotrophic bacteria responsible for oxidation of carbonaceous organic compounds in wastewater treatment. Thus, the nitrification process needs a significantly higher SRT to work compared to conventional activated sludge processes. The SRT needed for nitrification in an activated sludge process is a function of the maximum specific growth rate (which is related to temperature), the reactor dissolved oxygen concentration, and pH. Nitrification rates decline as the DO concentration decreases below 3.0 mg/L and the pH decreases below 7.0 mg/L. With sufficient DO and adequate pH, typical nitrification design SRTs range from 10 to 20 days at 10°C and 4 to 7 days at 20°C (Randall et al., 1992).

### **Denitrification**

In municipal and industrial wastewater treatment processes, denitrification is the biological reduction of nitrate or nitrite to nitrogen gas (N<sub>2</sub>) as indicated by equation below.



It is accomplished by a variety of common heterotrophic microorganisms that are normally present in aerobic biological processes. Most are facultative aerobic bacteria with the ability to use elemental oxygen, nitrate, or nitrite as their terminal electron acceptors for the oxidation of organic material.

Heterotrophic bacteria capable of denitrification include the following genera: *Achromobacter*, *Acinetobacter*, *Agrobacterium*, *Alcaligenes*, *Arthrobacter*, *Bacillus*, *Chromobacterium*, *Corynebacterium*, *Flavobacterium*, *Hypomicrobium*, *Moraxella*, *Nesseria*, *Paracoccus*, *Propionibacteria*, *Pseudomonas*, *Rhizobium*, *Rhodopseudomonas*, *Spirillum* and *Vibrio* (Tchobanoglous et al., 2003).

Recent research has shown that nitrite reduction is accomplished by a much more specialized group of heterotrophic bacteria than those performing the conversion of nitrate to nitrite (Katehis, 2007).

Denitrification by heterotrophic nitrifying bacteria and by autotrophic bacteria has also been observed. An example of a heterotrophic nitrifying bacteria that can denitrify is *Paracoccus pantotropha*, which obtains energy by nitrate or nitrite reduction while oxidizing ammonia under aerobic conditions. A readily available carbon source, such as acetate, is needed (Robertson and Kuenen, 1990). The conditions required for this form of denitrification are not practical in biological wastewater treatment.

An autotrophic denitrifying bacteria of practical significance in wastewater treatment is that in the Anammox process used to remove nitrogen in return streams from anaerobic digestion sludge dewatering filtrate or centrate. These bacteria have been identified as a member of bacteria in the order *Planctomycetales* (Strous et al, 1999). Under anaerobic conditions, ammonia is oxidized with the reduction of nitrite with the final product as nitrogen gas. The reaction is best accomplished at temperatures above 25°C and they are slow growing organisms.

Facultative denitrifying bacteria will preferentially use oxygen instead of nitrate. In the absence of oxygen, however, they will carry out nitrite and/or nitrate reduction. Microbiologists generally use the term anaerobic to describe biological reactions in the absence of oxygen. To distinguish anaerobic conditions for which the biological activity occurs mainly with nitrate or nitrite as the electron acceptor, the term “anoxic” has been applied.

Although oxygen is known to inhibit denitrification, denitrification has been observed in activated sludge and fixed film systems in which the bulk liquid DO concentration is positive. This is due to the establishment of an anoxic zone within the floc or biofilm depth. Hence, a single system can carry out simultaneous nitrification and denitrification. The DO concentration that is possible for simultaneous nitrification and denitrification depends on a number of factors including the mixed liquor concentration, temperature, and substrate loading. The DO concentration above which denitrification is inhibited may vary from 0.10 to 0.50 mg/L (WEF and ASCE, 2006; Tchobanoglous et al., 2003; Barker and Dold, 1997).

**The organic carbon source for denitrifying bacteria can be in the form of:**

- Soluble degradable organics in the influent wastewater
- Soluble organic material produced by hydrolysis of influent particulate material
- Organic matter released during biomass endogenous decay

A general rule of thumb is that 4 g of wastewater influent BOD is needed per g of NO<sub>3</sub>-N to be removed through biological treatment (Tchobanoglous et al., 2003). When denitrification occurs after secondary treatment, there is little BOD remaining so a supplemental carbon source is often needed. The most common exogenous carbon source in use is methanol; however, due to issues regarding its safety, cost, and availability, some wastewater systems are using alternative carbon sources such as acetic acid, ethanol, sugar, glycerol, and proprietary solutions depending on the needs of their particular facility (deBarbadillo et al., 2008).

Biological denitrification reactions produce alkalinity and heterotrophic biomass. Based on the stoichiometry of the reactions, denitrification will produce a 3.57 mg/L of alkalinity as CaCO<sub>3</sub> for each mg/L of NO<sub>3</sub><sup>-</sup>-N consumed. Heterotrophic biomass produced can be estimated as 0.4 g volatile suspended solids (VSS) produced for every gram of COD consumed. Growth kinetics for denitrifiers are dependent on a number of factors including carbon substrate type and concentration, DO concentration, alkalinity, pH, and temperature, with carbon source being the most important.

**Current Configurations**

Biological nitrogen removal can be accomplished by a variety of treatment configurations using suspended growth, attached growth, or combined systems. In the past, some WWTPs were required to only remove ammonia-nitrogen in wastewater to reduce toxicity to aquatic organisms with no limits on nitrate or total nitrogen. However, most treatment plants are now required to remove nitrogen because both ammonia-nitrogen and nitrate-nitrogen can stimulate algae and phytoplankton growth and lead to eutrophication of U.S. waterways. For biological nitrogen removal, it is essential that nitrification occur first followed by denitrification.

**Biological Nitrogen Removal Process Configurations**

Biological nitrogen removal systems achieve nitrification and denitrification along with BOD reduction in bioreactors followed by secondary clarification. Processes can be either suspended growth or hybrid systems that use a combination of attached growth (biofilms) and suspended growth technologies. Configurations within each of these classifications are discussed below. Note that biological processes that remove both nitrogen and phosphorus are discussed later in this manual.

### **Suspended Growth Systems Modified Ludzck Ettinger (MLE) process**

The most common nitrogen removal process used at WWTPs is the Modified Ludzck Ettinger (MLE) process, which is considered a pre-denitrification, single sludge system. The process includes an initial anoxic zone, followed by an aerobic zone. In the anoxic zone, nitrate produced in the aerobic zone is reduced to nitrogen gas. This process uses some of the BOD in the incoming waste. Nitrification occurs in the aerobic zone along with the removal of most of the remaining BOD. At the end of the aerobic zone, pumps recycle the nitrate-rich mixed liquor to the anoxic zone for denitrification.

Total nitrogen removal for the MLE process is typically 80 percent, and the process achieves total effluent nitrogen concentrations ranging from approximately 5 to 8 mg/L with internal nitrate recycle ratios of 2 to 4 based on the influent flowrate (2-4Q).

### **Four-Stage Bardenpho Process**

The four-stage Bardenpho process builds on the MLE process, with the first two stages being identical to the MLE system (anoxic zone followed by an aeration zone with a nitrate-rich recycle from the aeration to the anoxic zone). The third stage is a secondary anoxic zone to provide denitrification to the portion of the flow that is not recycled to the primary anoxic zone. Methanol or another carbon source can be added to this zone to enhance denitrification. The fourth and final zone is a re-aeration zone that serves to strip any nitrogen gas and increase the DO concentration before clarification. Some configurations have used an oxidation ditch instead of the first two stages. This process can achieve effluent TN levels of 3 to 5 mg/L.

### **Sequencing Batch Reactors**

Sequencing batch reactors (SBRs) are fill and draw batch systems in which all treatment steps are performed in sequence for a discreet volume of water in a single or set of reactor basins.

#### **SBRs use four basic phases for most systems:**

**Fill:** water is added to the basin and is aerated and mixed

**React:** Biological processes are performed

**Settle:** All aeration and mixing is turned off and the biomass is allowed to settle

**Decant:** Clarified effluent is removed and biomass is wasted as necessary

The SBR control system allows it to mimic most other suspended growth processes such as the MLE or Four-Stage Bardenpho system. It typically completes 4 to 6 cycles per day per tank for domestic wastewater. If properly designed and operated, SBRs can achieve about 90 percent removal of nitrogen (WEF and ASCE, 2006).

### **Oxidation Ditches**

Oxidation ditches are looped channels that provide continuous circulation of wastewater and biomass. A number of operating methods and designs have been developed to achieve nitrogen removal, all of which work by cycling the flow within the ditch between aerobic and anoxic conditions. DO can be added to the aerated zone using horizontal brush aerators, diffused aerators with submersible mixers, or vertical shaft aerators (WEF and ASCE, 2006). Patented designs include the NITROX process, Carrousel, and BioDenitro (WERF, 2000a). Many oxidation ditch configurations can achieve simultaneous nitrogen and phosphorus removal.

### **Step Feed**

The step feed biological nitrogen removal process splits the influent flow and directs a portion of it to each of several anoxic zones, with the highest proportion of influent flow going to the first zone and steadily decreasing until the last anoxic zone prior to clarification. The biomass in the later stages are not just treating influent flow but are also used to reduce nitrate from the upstream zones. The step feed system provides flexibility for systems to handle wet-weather events. It can also be compatible with existing conventional “plug flow” activated sludge processes and it does not require the installation of recycle pumps and piping.

Disadvantages include the need to control the DO concentration of aeration zones preceding the downstream anoxic zones and the need to control the flow splitting to the step feed points.

### **Attached Growth and Hybrid Systems Integrated Fixed-Film Activated Sludge (IFAS)**

Integrated fixed-film activated sludge (IFAS) is any suspended growth system (e.g., MLE, Four-Stage Bardenpho) that incorporates an attached growth media within the suspended growth reactor in order to increase the amount of biomass in the basin. IFAS systems have higher treatment rates than suspended growth systems and generate sludge with better settling characteristics. Many types of fixed and floating media are available, including:

- **Rope:** also called looped-cord or strand media. Consists of a polyvinyl chloride-based material woven into rope with loops along the length to provide surface area for the biomass (WERF, 2000b). Proprietary designs include Ringlace, Bioweb, and Biomatrix (USEPA, 2008a).
- **Moving Bed with Sponge:** proprietary products include Captor and Linpor (USEPA, 2008a).
- **Plastic Media:** several types of free-floating plastic media are available from Kaldness. Other media types include fabric mesh (e.g., AccuWeb) and textile material (Cleartec).

### **Moving-Bed Biofilm Reactor (MBBR)**

The moving-bed biofilm reactor (MBBR) is similar to the IFAS system in that it uses plastic media with a large surface area to increase biomass within the biological reactor. The MBBR media is submerged in a completely mixed anoxic or aerobic zone. The plastic media are typically shaped like small cylinders to maximize surface area for biomass growth. The difference between MBBR and IFAS is that MBBR does not incorporate return sludge (WERF, 2000b).

### **Membrane Bioreactor (MBR)**

MBRs are commonly designed for nitrogen removal, using membranes for liquid-solids separation following the anoxic and aerobic zones instead of conventional clarification. Membranes can be submersed in the biological reactor or located in a separate stage or compartment. Low-pressure membranes (ultrafiltration or microfiltration) are commonly used. Systems can be pressure driven or vacuum. All systems use an air scour technique to reduce buildup on the membranes (USEPA, 2007d; USEPA, 2008a).

Membrane materials are either organic polymers or inorganic materials such as ceramics. They are designed in modular units and are typically configured as either hollow fiber bundles or plate membranes (USEPA, 2007d) For biological nutrient removal applications, the design SRTs and design principals for MBR systems are similar to those used for systems with secondary clarifiers.

One of the main differences is that the MBR systems operate at a higher MLSS concentration which results in smaller tanks and smaller space requirements. In addition, membrane separation provides for greatly reduced TSS in the effluent, typically below 1.0 mg/L, and hence slightly greater removal of nitrogen and phosphorus. Operational issues include potential for membrane biofouling and increase pumping costs (USEPA, 2007d; WEF, 2005).

## Activated Sludge Process Section



### Key Terms

**Aerobic (AIR-O-bick)** a condition in which free or dissolved oxygen is present in the aquatic environment

**Aerobic Bacteria** – bacteria which will live and reproduce only in an environment containing oxygen.

**(aerobes)** Oxygen combined chemically, such as in water molecules ( $H_2O$ ), cannot be used for respiration by aerobes

**Anaerobic (AN-air O-bick)**- a condition in which “**free**” or dissolved oxygen is not present in the aquatic environment.

**Anaerobic Bacteria** – bacteria that thrive without the presence of oxygen.  
**(anaerobes)**

**Saprophytic bacteria** – bacteria that break down complex solids to volatile acids.

**Methane Fermenters** – bacteria that break down the volatile acids to methane ( $CH_4$ ) carbon dioxide ( $CO_2$ ) and water ( $H_2O$ ).

**Oxidation** – the addition of oxygen to an element or compound, or removal of hydrogen or an electron from an element or compound in a chemical reaction. The opposite of reduction.

**Reduction** – the addition of hydrogen, removal of oxygen or addition of electrons to an element or compound. Under anaerobic conditions in wastewater, sulfur compounds or elemental sulfur are reduced to  $H_2S$  or sulfide ions.



#### Water Bear Tardigrade

This creepy "water bear" has forced scientists to reconsider their definition of what's "alive." When unable to find water, this insect like critter (which is the size of a grain of sand) stops moving, breathing, and eating. Even its cells shut down. Dead as a doornail, right? Wrong! Add water and the critter springs back to life. Scientists have exposed dried-up water bears to extreme heat, bitter cold, and even massive doses of radiation -- and the teeny animals still revived. The key to the critters' survival may be a sugar they produce as they dry out. By coating structures inside and between the critters' cells, the sugar keeps the cells from sticking together and breaking. When water is added, the sugar dissolves -- and the creepy crawlies burst back into action.



## Basic System Components of Activated Sludge

In the basic “activated” sludge process, emphasis on *activated*, the wastewater enters an aerated tank (the dome) where previously developed biological floc particles are brought into contact with the organic matter (foot-long hot dogs) of the wastewater.

The organic matter is a carbon and an energy source for the bug's cell growth and is converted into cell tissue. The oxidized end product is mainly carbon dioxide,  $\text{CO}_2$ . The substance in the sports dome is referred to as mixed liquor. The stuff in the mixed liquor is suspended solids and consists mostly of microorganisms, suspended matter, and non-biodegradable suspended matter (**MLVSS**).

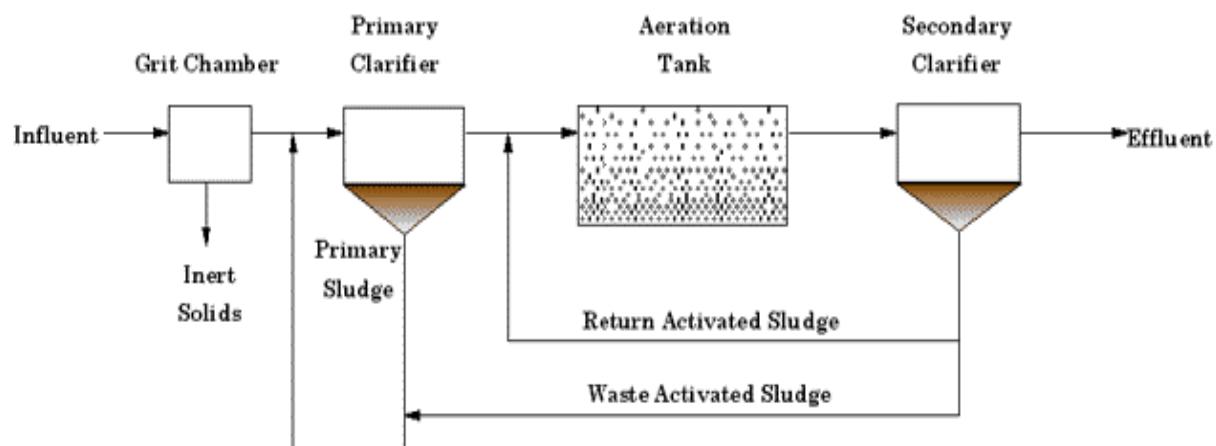
The make-up of the microorganisms are around 70 to 90% organic and 10 to 30% inorganic matter. The makeup of cells varies depending on the chemical composition of the wastewater and the specific characteristics of the organisms in the biological mass. The picture below shows the basic outline of an aeration tank. Just remember that pretreatment is crucial prior to the activated sludge process.

**Before we dive into the tank, in the space provided, list three key components of pretreatment (headworks) and how each benefits the process.**

1.

2.

3.



### Mixed Liquor

Back to the mixed liquor, as it leaves the aeration tank, it usually goes to a clarifier to separate the suspended solids (**SS**) from the treated wastewater. The concentrated biological solids are then recycled back to the aeration tank, as returned activated sludge (**RAS**), to maintain a concentrated population of bugs (the team players) to treat the wastewater.

Before we start the game, we need to make sure we have a stadium and all components are in place and operating properly. In the space provided, define the following terms:  
See Glossary in Rear.

**Anaerobic:**

**Aerobic:**

**DO:**

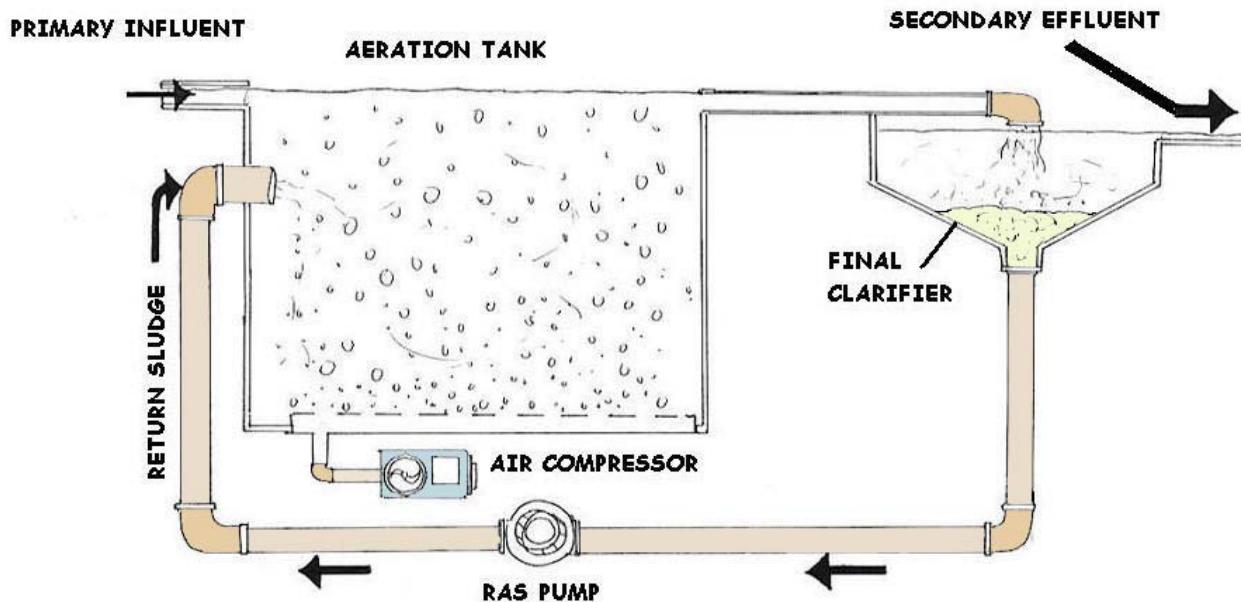
**BOD:**

**COD:**

### Process Design

Let's first look at the different aeration tank designs and how they function. We will focus on the following:

## ACTIVATED SLUDGE PROCESS

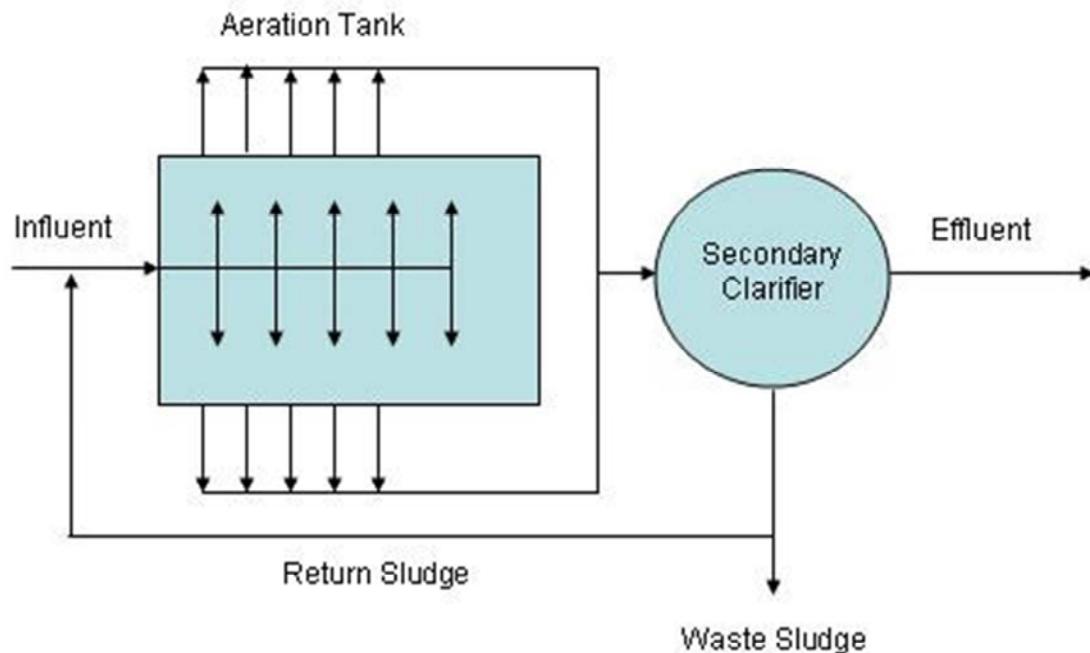


## Complete Mix Activated Sludge Process

In a complete mix activated sludge process, the mixed liquor is similar throughout the aeration tank. The operating characteristics measured in terms of solids, oxygen uptake rate (**OUR**), MLSS, and soluble BOD 5 concentration are identical throughout the tank.

Because the entire tank contents are the same quality as the tank effluent, there is a very low level of food available at any time to a large mass of microorganisms.

This is the major reason why the complete mix modification can handle surges in the organic loading without producing a change in effluent quality. The type of air supply used could be either diffused air or a mechanical aerator. Complete mix process may be resistant to shock loads but is susceptible to filamentous growths.

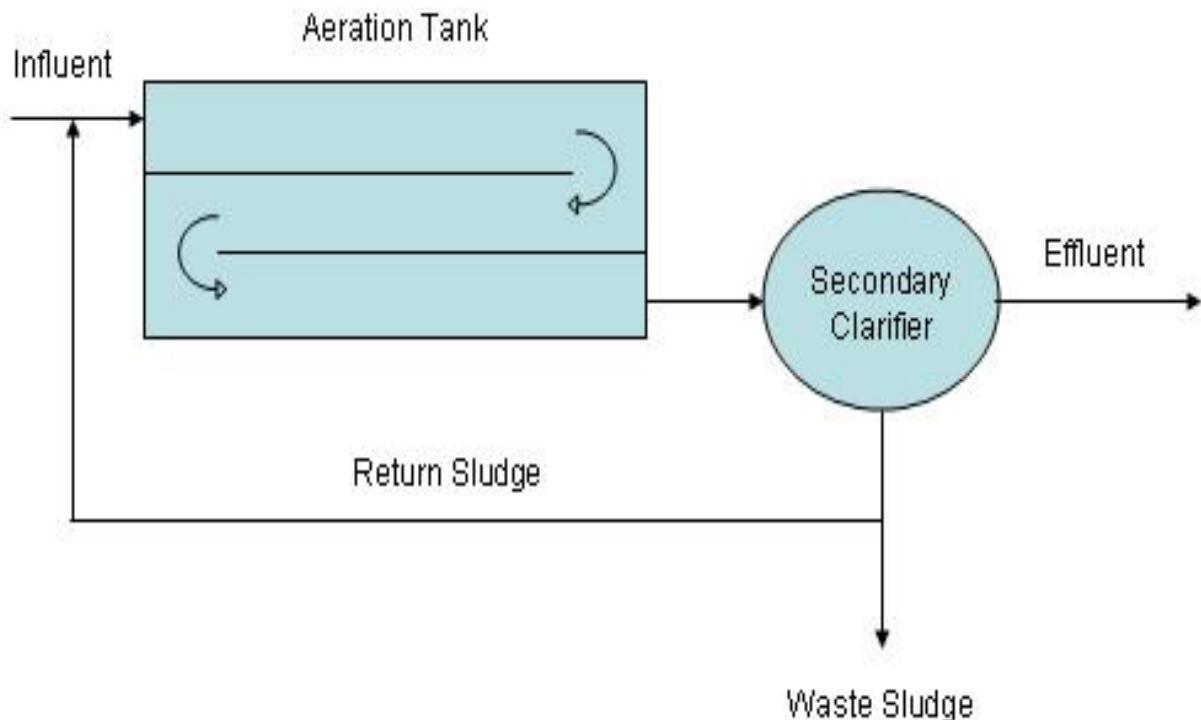


## Plug Flow Activated Sludge Process

Plug flow tanks are the oldest and most common form of aeration tank. They were designed to meet the mixing and gas transfer requirements of diffused aeration systems. One characteristic of the plug flow configuration is a very high organic loading on the MLSS in the initial part of the tank. The loading is then reduced and the organic material in the raw wastewater is oxidized.

At the end of the tank, depending on detention time, the oxygen consumption may primarily be the result of endogenous respiration or nitrification, which we will talk more about later on. The same characteristics are present when the aeration tank is partitioned into a series of compartments.

Each compartment must have the oxygen supply and design to meet the individual compartment needs. Plug flow configurations have the ability to avoid “**bleed through**,” the passage of untreated organics during peak flow. These configurations are often preferred when high effluent DO’s are sought because only a small section of the tank will operate at a high DO. In a complete mix configuration, the entire tank must operate at the elevated DO.



## Contact Stabilization Activated Sludge Process

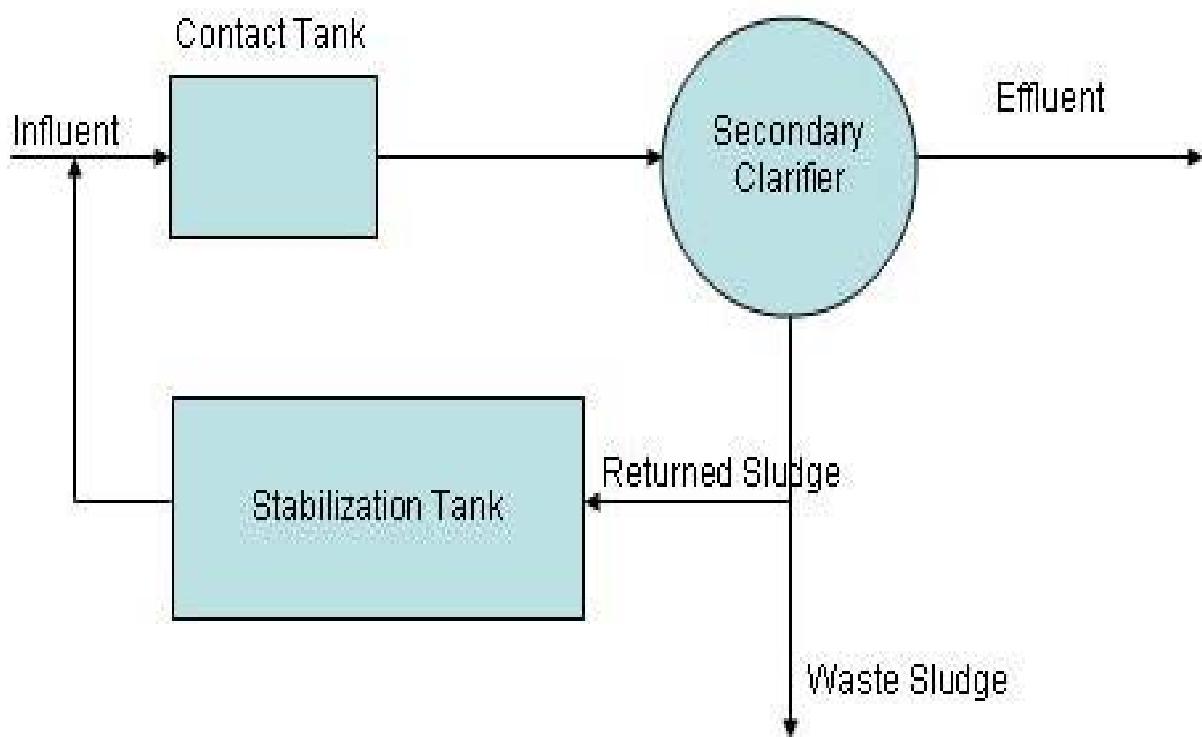
Contact stabilization activated sludge is both a process and a specific tank configuration. The contact stabilization encompasses a short-term contact tank, secondary clarifier, and a sludge stabilization tank with about six times the detention time used in the contact tank.

Contact stabilization is best for smaller flows in which the MCRT desired is quite long.

Therefore, aerating return sludge can reduce tank requirements by as much as 30 to 40 % versus that required in an extended aeration system. The volumes for the contact and stabilization tanks are often equal in size and secondary influent arrangements.

### ***What does this all mean?***

They can be operated either in parallel as an extended aeration facility or as a contact stabilization unit. This flexibility makes them suitable for future expansion to conventional activated sludge, without increasing the aeration tank, by merely adding more clarification capacity.



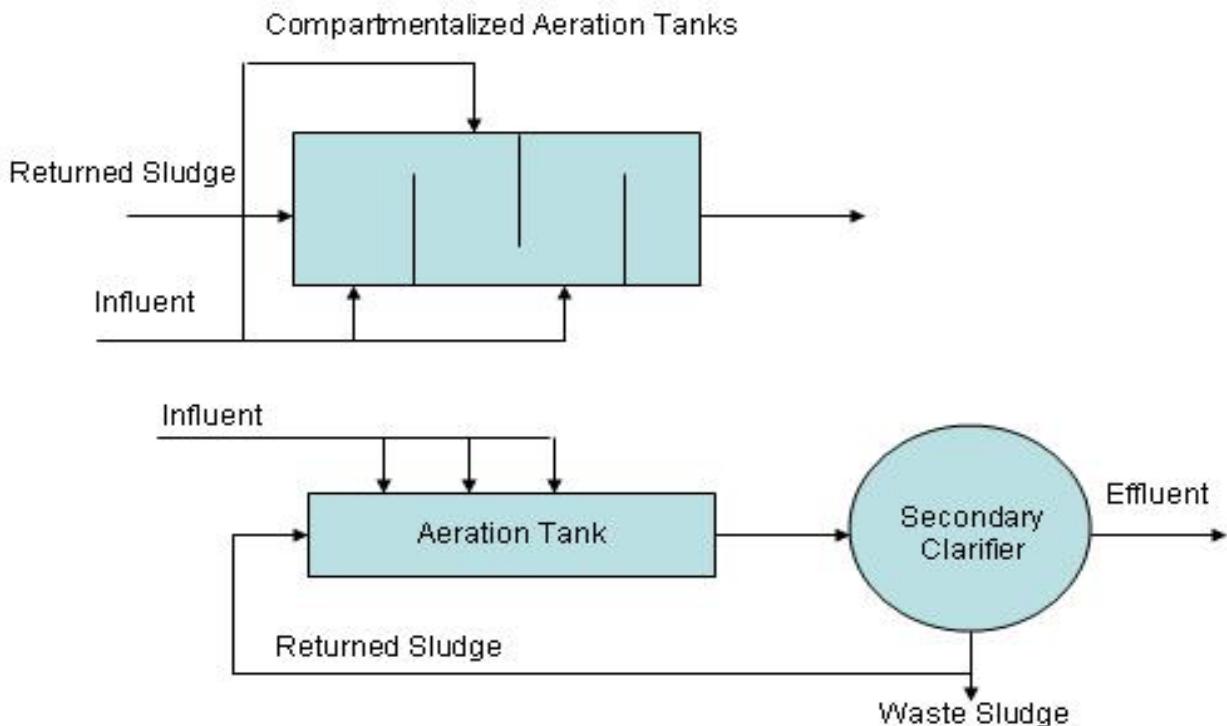
## Step Feed Activated Sludge Process

Step feed is a modification of the plug flow configuration in which the secondary influent is fed at two or more points along the length of the aeration tank.

With this arrangement, oxygen uptake requirements are relatively even and the need for tapered aeration is eliminated.

Step feed configurations generally use diffused aeration equipment. The step feed tank may be either the long rectangular or the folded design. Secondary influent flow is added at two or more points to the aeration tank, usually in the first 50 to 75% of the length.

It is also possible to use the same process approach by compartmentalizing the tank and directing flow lengthwise through the compartments. Usually, the last compartment does not receive any raw waste.



## Extended Aeration Activated Sludge Process

The extended aeration process uses the same flow scheme as the complete mix or plug flow processes but retains the wastewater in the aeration tank for 18 hours or more.

This process operates at a high MCRT (low F/M), resulting in a condition where there is not enough food in the system to support all of the microorganisms present. The microorganisms therefore compete very actively for the remaining food and even use their own cell structure for food.

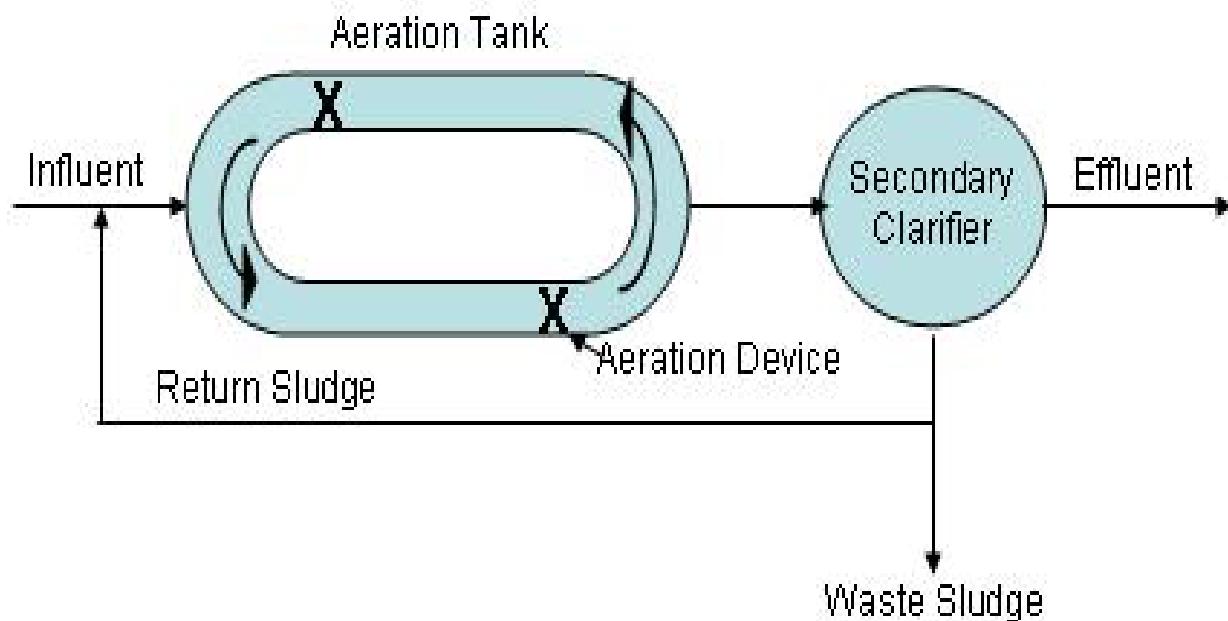
This highly competitive situation results in a highly treated effluent with low sludge production. (Many extended aeration systems do not have primary clarifiers and they are package plants used by small communities.)

The main disadvantages of this system are the large oxygen requirements per unit of waste entering the plant and the large tank volume needed to hold the wastes for the extended period.

### Oxidation Ditch Activated Sludge Process

The oxidation ditch is a variation of the extended aeration process. The wastewater is pumped around a circular or oval pathway by a mechanical aerator/pumping device at one or more points along the flow pathway. In the aeration tank, the mixed liquor velocity is maintained between 0.8 and 1.2 fps in the channel to prevent solids from settling.

Oxidation ditches use mechanical brush disk aerators, surface aerators, and jet aerator devices to aerate and pump the liquid flow. Combination diffused aeration and pumping devices are commonly used in Europe.



## High Purity Oxygen Activated Sludge Process

The most common high purity oxygen activated sludge process uses a covered and staged aeration tank configuration. The wastewater, return sludge, and oxygen feed gas enter the first stage of this system and flow concurrently through the tank.

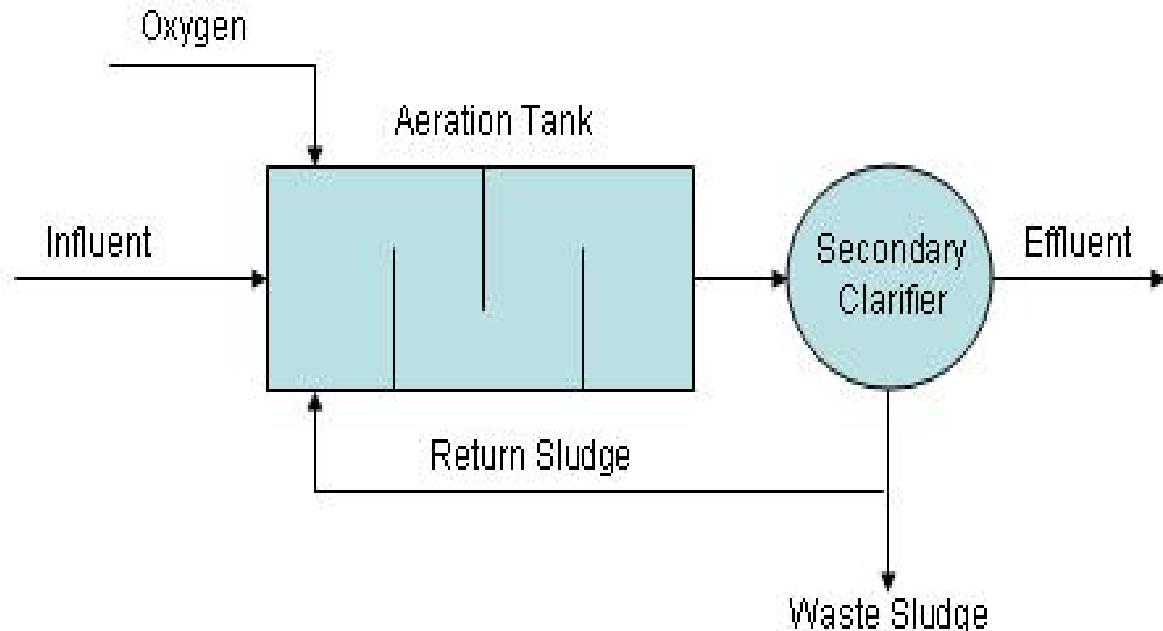
The tanks in this system are covered to retain the oxygen gas and permit a high degree of oxygen use. A prime advantage of the staged reactor configuration of the oxygenation system is the system's ability to match the biological uptake rate with the available oxygen gas purity.

The dissolution of oxygen and the mixing of the biological solids within each stage of the system are accomplished with either surface aeration devices or with submerged turbine-aeration systems. The selection of either of these two types of dissolution systems largely depends on the aeration tank geometry selected.

The particular configuration of oxygenation tank selected for a given system, that is, size of each stage, number of stages per aeration tank, and number of parallel aeration tanks, is determined by several parameters including waste characteristics, plant size, land availability, and treatment requirements.

Aside from the aeration tank, the other key factor in an oxygen activated sludge system is the oxygen gas source. There are three sources of oxygen supply: liquid oxygen storage, cryogenic oxygen generation, and pressure-swing adsorption generation.

The first of these requires no mechanical equipment other than a storage tank that is replenished by trucked-in liquid oxygen. This method is economically feasible for small (less than 4 mgd) or temporary installations.



## Aeration Section

There are several designs and applications for aerators:

- Diffused Aerators
- Mechanical Surface Aerators
- Submerged Turbine Aerators

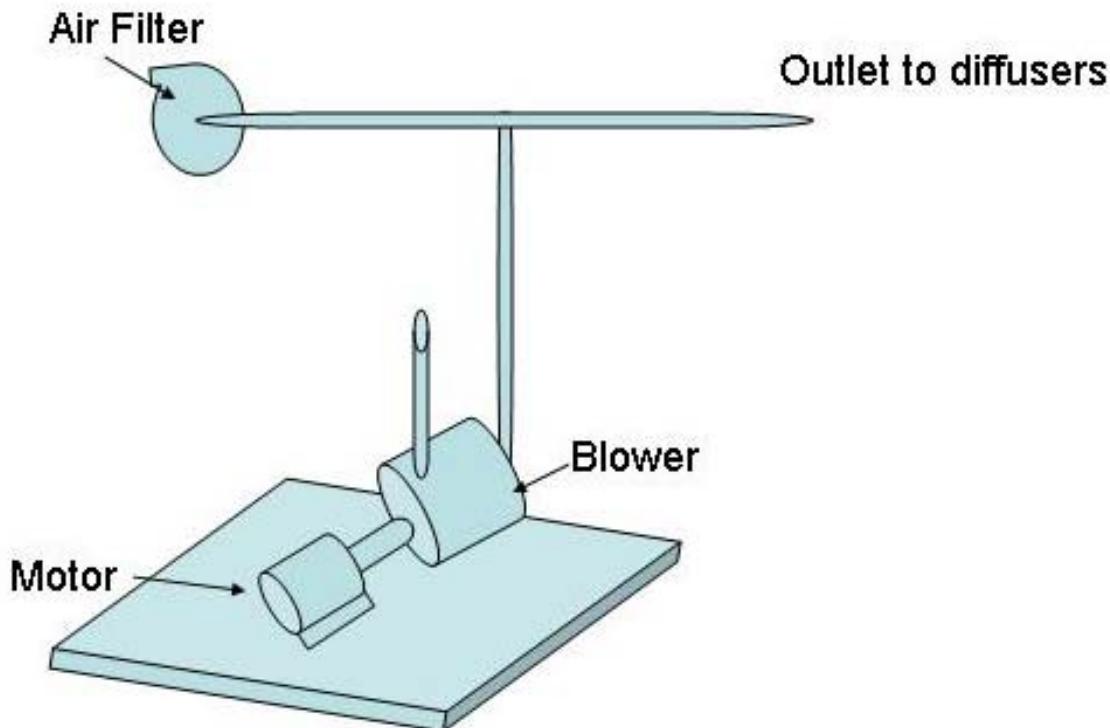
The two most common types of aeration systems are subsurface diffusion and mechanical aeration. Diffused air systems have been around longer than you.

Opened tubes were used or perforated pipes located at the bottom of aeration tanks. But a more efficient process was desired, born to the process, porous plate diffusers. In the diffused air system, compressed air is introduced near the bottom of the tank. Let's look at the definition for diffused aeration:

***"The injection of a gas, air or oxygen, below a liquid surface."***

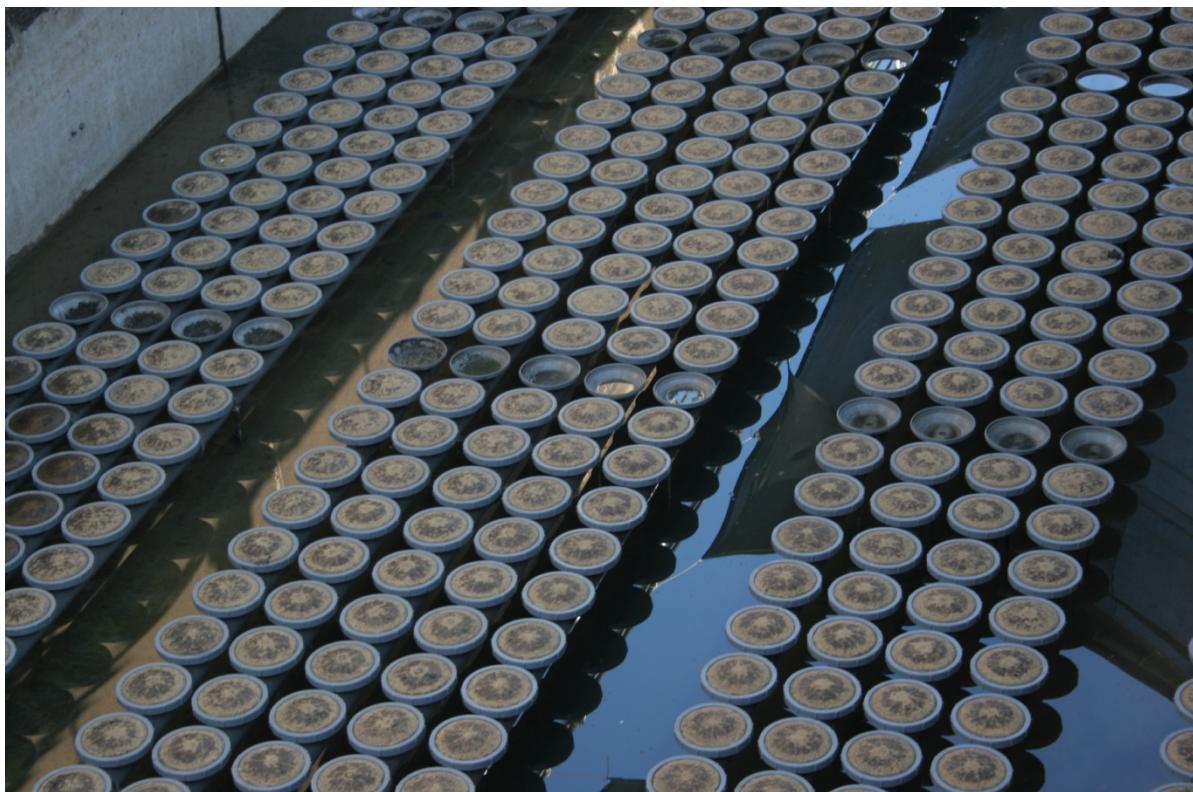
There are a variety of hybrid air diffusion systems used in the process; we will focus on the basic components.

The following diagram highlights the main parts of the diffused aeration system.





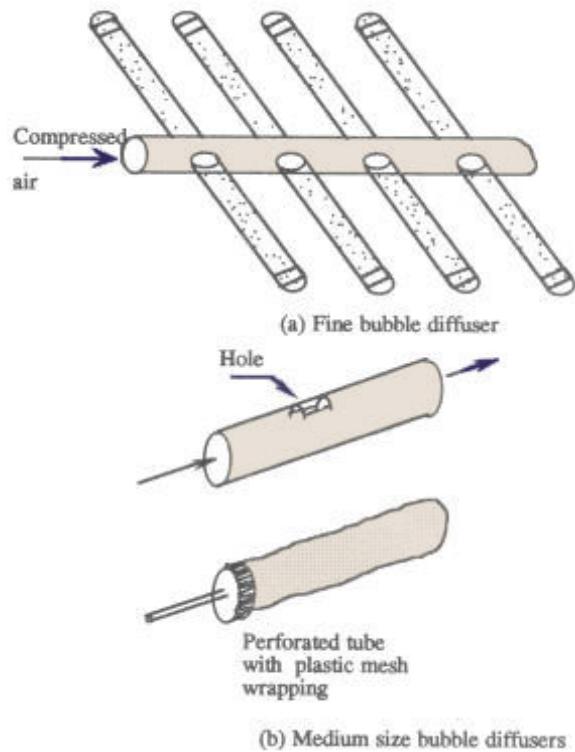
Here is a rare and up-close view of non-porous diffuser heads. Notice the heads that are missing in the bottom photograph.



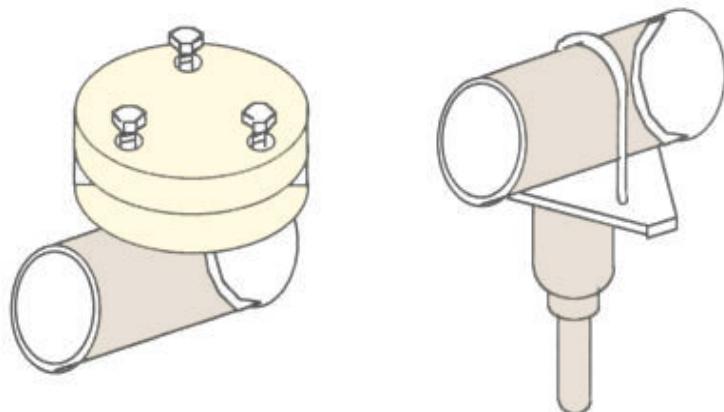
## Aeration

The aerated systems described above need an oxygen supply. Depending on the characteristics of the process, different designs may be used. The oxygen can be supplied to the activated sludge by either diffused aeration, by turbine agitation, by static aerators, or by surface coarse or large bubble diffusers. The last two are also used in lagoon systems.

The diffused aeration systems are also divided into fine bubble, medium and coarse or large bubble diffusers. The fine bubble diffusers are built of porous materials (grains of pure silica or aluminum oxide are bonded ceramicly or by resins) which provide very small bubbles of high surface area that favor the oxygen transfer from the air to the wastewater. The medium bubble diffusers are perforated pipes or tubes wrapped with plastic or woven fabric. The coarse or large bubble diffusers can be orifice devices of various types, some of which are designed to be non-clogging.



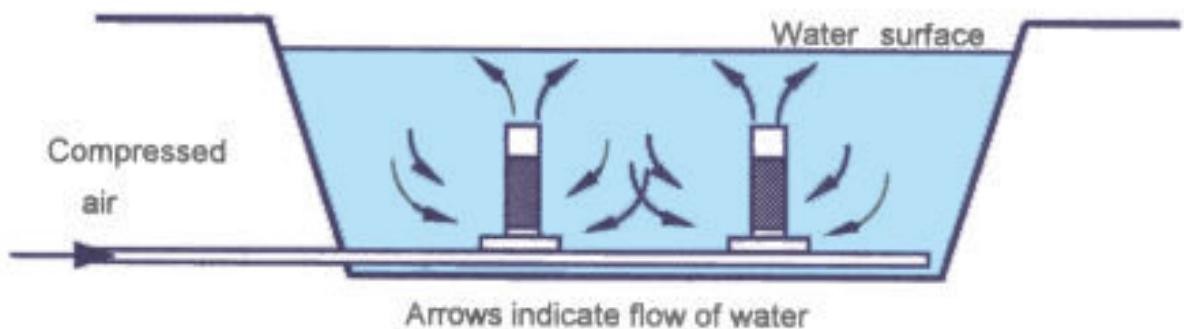
Fine and medium bubble diffusers



### Coarse or Large Bubble Diffusers

With the small or fine bubble diffusers, it is important to use air free of particles that would otherwise clog them. Although somewhat less efficient for oxygen transfer, the coarse bubble diffusers are sometimes preferred because the presence of particles in the air is not a critical problem, and also for their lower cost and maintenance requirements. The diffusers are placed along air manifolds, close to the bottom of the aeration tanks.

The static aerators are vertical tubes placed at the bottom of the aeration tank, with packing material along its length. The compressed air is supplied from the bottom of the tubes, forcing a mixture of air and water through the packing, where most of the oxygen transfer to the wastewater takes place. They have been used mainly in aerated lagoons.



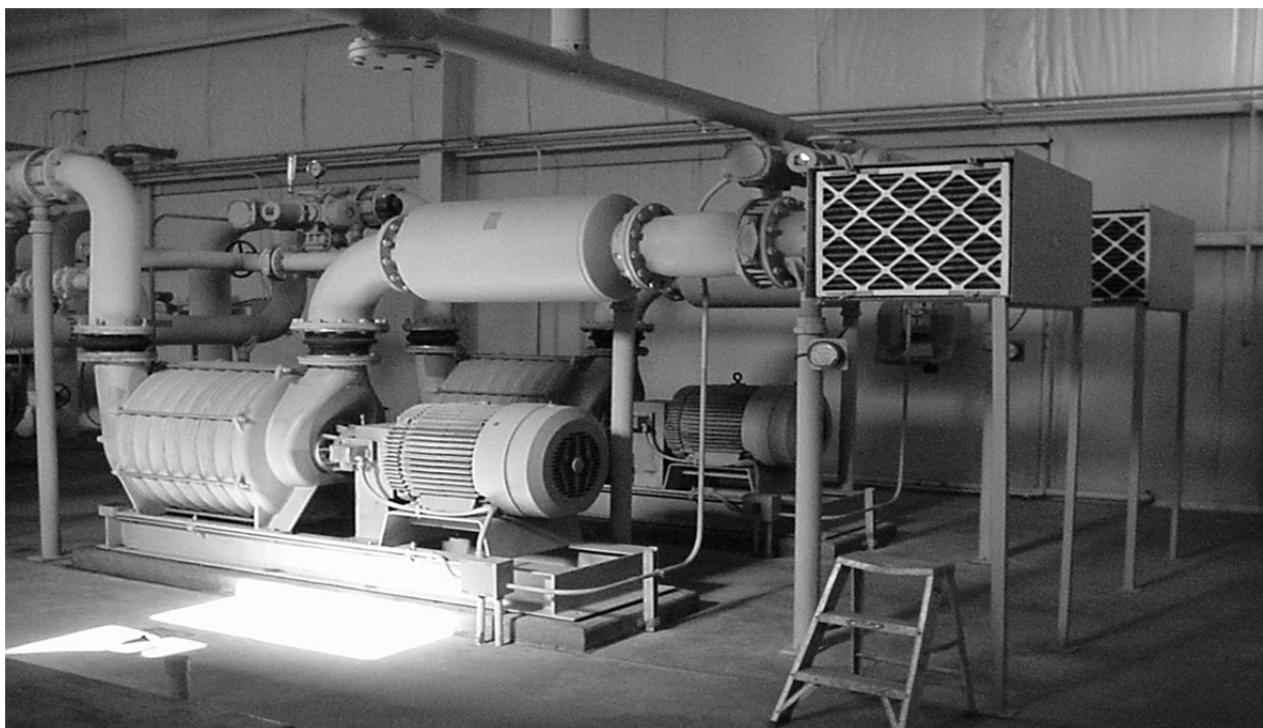
Oxidation ditches are used for nutrient removal.

## Blowers

In the diffused aeration system, blowers are used to circulate the tank's contents by the air-lift effect. The air filter on the blower removes dirt from the air. Therefore, helps prevent diffuser clogging. Before all this begins, we need a power source to drive the blower. Usually, electric motors are used but in remote locations, gas or diesel engines can be used as well. In some states, solar energy is available to provide the power.

As illustrated in the photograph below, the rotation of the motor shaft is transferred to the blower shaft by means of a flexible coupling or through drive belts. The blowers that we will refer to are centrifugal blowers.

The centrifugal blower works like a centrifugal pump or a fan. Rotating impellers or fans cause movement of the air through the blowers. You have an intake side that takes in the air and the discharge side the forces the air out. The number of impellers you have will determine if it is a multi-stage or single-stage blower. The photograph below illustrates the major components of a centrifugal blower.



A lobe blower utilizes positive displacement; it also has an intake and a discharge side. The lobes turn in opposite directions in the casing. As they turn, the air is drawn in through the blower inlet and is trapped. The lobes keep turning, opening the blower discharge, and forcing the trapped air through the outlet. Usually, an electric motor drives the blower with belt pulleys or flexible couplings.



**Before we continue let's review what you just read about the blowers and motors.**

1. What are two ways that the motor and the blowers can be attached?
2. When using flexible couplings, what are some maintenance concerns to consider?

Blowers may be provided with additional equipment. For example, safeguards can be installed to protect equipment and operators. Temperature sensors can be used for bearing housing, vibration sensors protect the unit by shutting it down if limits are exceeded. Condensation drains should be provided on the bottom of blowers to drain off any accumulated moisture.

The compressed air from the blowers moves into a system of pipes and valves. The amount of air supplied from the blower is controlled by regulating valves mounted on the intake and/or discharge side of the blower. Usually butterfly valves are used and depending on your budget, you could have manually operated or use automation.

Blowers usually discharge to a common manifold, so check valves are installed at the discharge of each blower. The intake and discharge pipes are called the air mains. They are connected by a flexible connection to allow for vibration and heat expansion in the piping. In the winter months, the best place to be is in the blower room. There is a pressure relief valve on the discharge manifold to protect the blower from excessive back pressure overload. When this occurs the operator will be awakened on the midnight shift. Pressure gauges are used in several areas on the discharge side of the blowers. In some cases you may see them on the intake side for use in calculations of pump efficiency.

On the intake side, where air is supplied, you would have some type of filtering to remove dirt particles that could clog the diffusers. It also protects the blowers from excessive wear. Replaceable filter units are the simplest for operations. Bag house dust collectors are bulky and expensive, though maintenance may be less. In some cases, electrostatic precipitators may be an advantage, shocking if operators are not careful, in areas of poor air quality. Most systems have utilized pressure drop measuring to indicate when it is time to replace or clean the units.



The above photograph shows air being unevenly distributed.

## Diffusers

There are many different design layouts and patterns of diffuser placement. Systems that allow longer and more complete contact between the air and the liquid are preferred. We will focus on fine bubble (porous) diffusers and coarse bubble (nonporous).

Coarse bubble diffusion devices, or large-hole diffusers, produce larger bubbles than porous plates, porous tubes, or synthetic socks. The larger bubbles provide less surface area for air-liquid contact and will result in less oxygen transfer efficiency than that obtained with fine bubble diffusers.

**Answer this question:**

An air stone like those used in aquariums is a good example of a?

- A. Porous material
- B. Nonporous material

### Mechanical Aeration

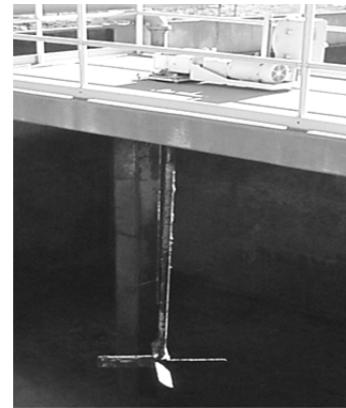
There are several main types of mechanical aeration devices. The floating and fixed bridge aerators are quite common. Some use a blade to agitate the tank's surface and disperse air bubbles into the aeration liquor. Others circulate the mixed liquor by an updraft or downdraft pump or turbine. This action produces surface and subsurface turbulence, while diffusing air through the mixed liquor.



The motor speeds are usually in the 1800 rpm range. This speed is reduced to the 30 to 70 rpm range with gear reducers.



Most vertical motors are mounted on a gear reduction unit as seen in the photograph on the right. The impeller drive shaft can be enclosed in a housing connected directly to the gear box. There is a bearing at the bottom of the shaft that steadies and aligns this shaft. This bearing needs lubrication, always check your manufacturer's recommendations.



Some plants use an oxidation ditch in which rotating brushes, blades, or disks are rotated partially submerged in the mixed liquor. The turbulence produced traps the air bubbles and keeps the mixed liquor in motion.

Other systems use both compressed air and a mechanical device to trap the bubbles. In one such system, submerged turbine aeration, air is injected below a rotating turbine blade that shears and disperses the air.

Submerged turbine applications have also used a draft tube operating in a downdraft-pumping mode.

#### **Jet and Aspirator**

Aerators provide oxygen transfer by mixing pressurized air and water within a nozzle and then discharging the mixture into the aeration tank. The velocity of the discharged liquid and the rising air plume provide the necessary mixing action.





## Secondary Clarifiers

Because microorganisms are continually produced, a way must be provided for wasting some of the generated biological solids produced. This is generally done from the round or rectangular shaped clarifiers.

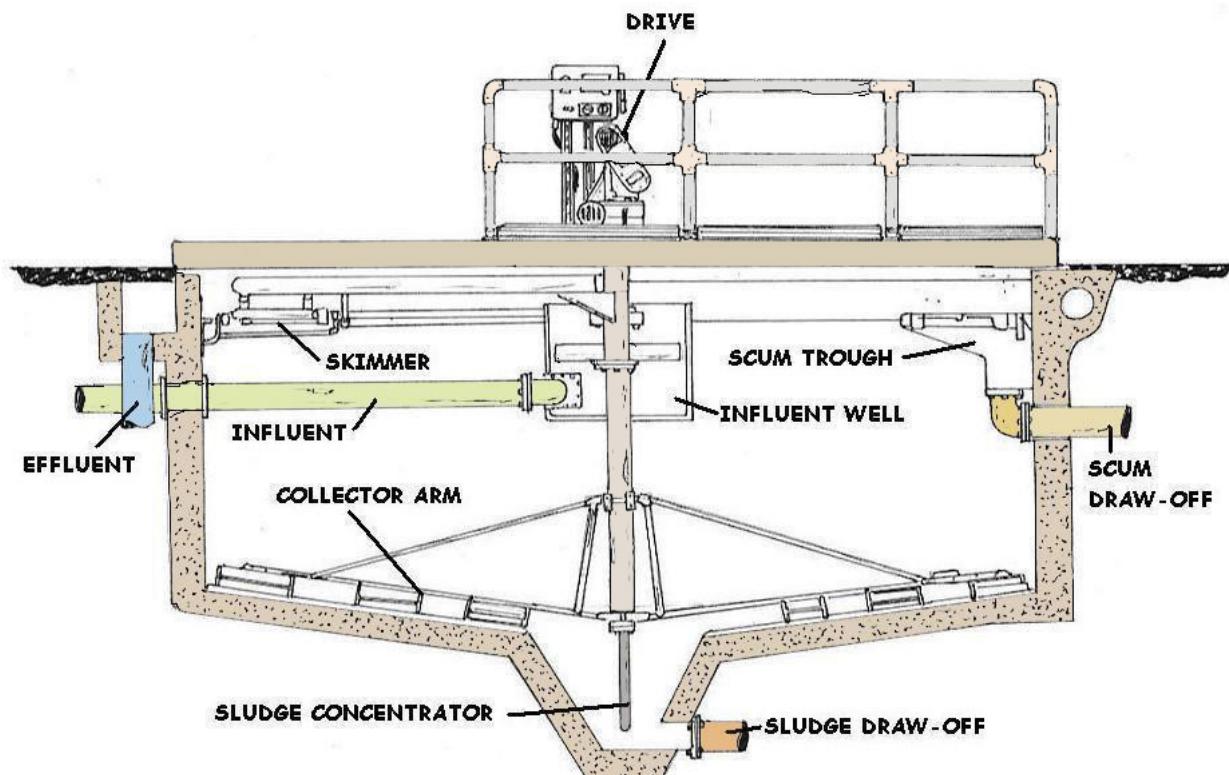
Let's first look at the components of a rectangular clarifier. Most are designed with scrapers on the bottom to move the settled activated sludge to one or more hoppers at the influent end of the tank. It could have a screw conveyor or a traveling bridge used to collect the sludge. The most common is a chain and flight collector. Most designs will have baffles to prevent short-circuiting and scum from entering the effluent.

The activated sludge is removed from the hopper(s) and returned by a sludge pump to the aeration tank or wasted. Since we mentioned return and waste what do the following terms represent?

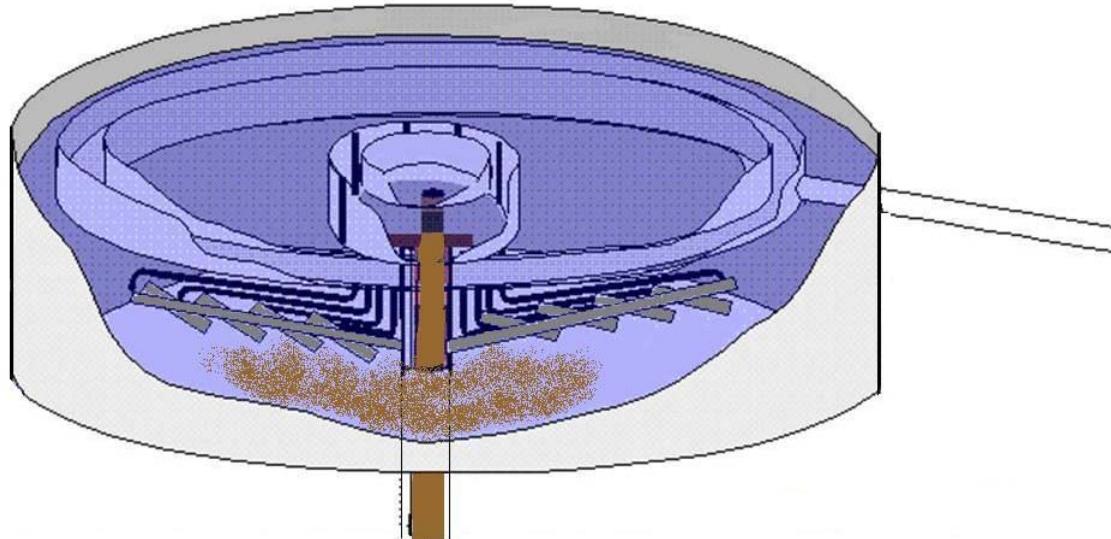
**RAS:**

**WAS:**

## CLARIFIER



**Clarifier** - A settling tank used to remove suspended solids by gravity settling. Commonly referred to as sedimentation or settling basins, they are usually equipped with a motor driven chain and flight or rake mechanism to collect settled sludge and move it to a final removal point.

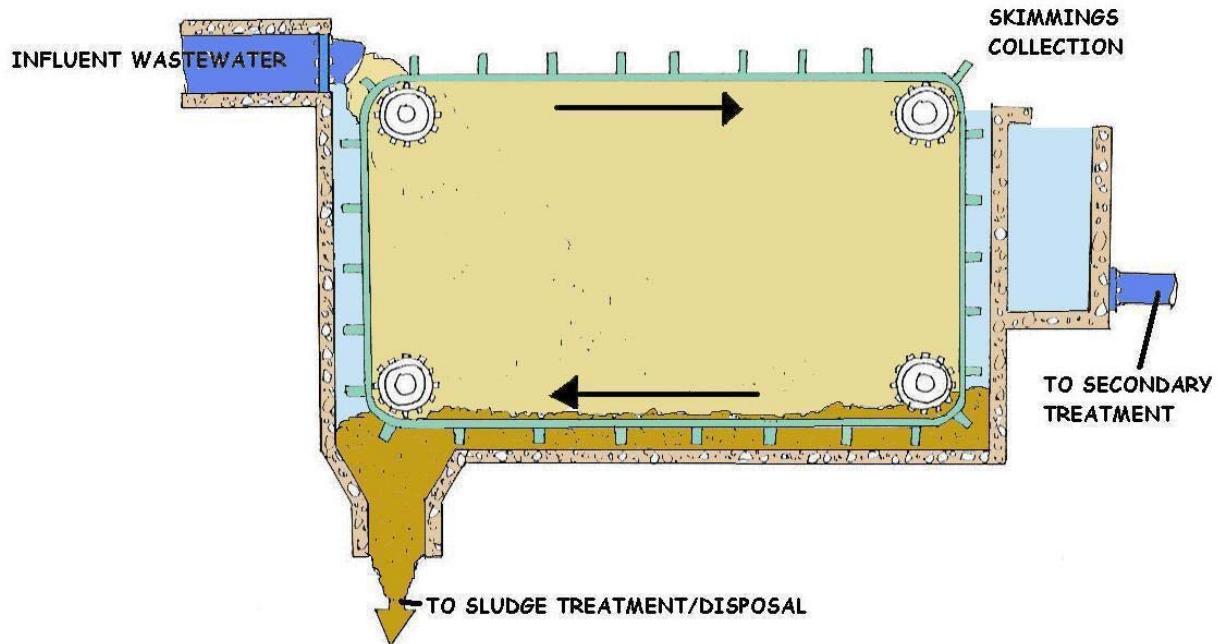
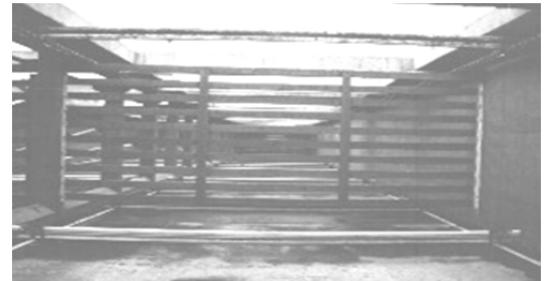


## Scum Removal Equipment

Scum removal equipment is desirable on secondary clarifiers. Skimmers are either of the type that rotates automatically or manually. The most important thing to consider is the sludge and scum collection mechanism. We will talk about "flights and chains". They move the settled sludge to the hopper in the clarifier for return and they also remove the scum from the surface of the clarifier. The flights are usually wood or nonmetallic flights mounted on parallel chains. The motor shaft is connected through a gear reducer to a shaft which turns the drive chain. The drive chain turns the drive sprockets and the head shafts. The shafts can be located overhead or below.

Some clarifiers may not have scum removal equipment so the configuration of the shaft may vary. As the flights travel across the bottom of the clarifier, wearing shoes are used to protect the flights. The shoes are usually metal and travel across a metal track.

To prevent damage due to overloads, a shear pin is used. The shear pin holds the gear solidly on the shaft so that no slippage occurs. Remember, the gear moves the drive chain. If a heavy load is put on the sludge collector system, the shear pin should break. This means the gear would simply slide around the shaft and movement of the drive chain would stop.





Top photograph, a clarifier's raking mechanism. Bottom, scum armature equipment.



## Scum Removal Equipment

In some circular or square tanks rotating scrapers are used. The diagram below shows an typical scum removal equipment.

The most common type has a center pier or column. The major mechanical parts of the clarifier are the drive unit; the sludge collector mechanism; and the scum removal system. There is also some related equipment that we will consider briefly. Let's look at the drive unit first. There are three main parts to the drive unit; the motor (or gear motor); the gear reducer; and the turntable.

The motor is connected to a gear reduction unit which is commonly connected to additional gearing. The drive cage is rotated around a center column by the motor and gear reduction unit. Although the drive motor runs at about 1800 rpm, the gear reducer lowers the output speed so that the sludge collector mechanism goes through one revolution every 20 to 30 minutes. Usually, the motors used on clarifier mechanisms are totally enclosed, fan cooled motors, suitable for outside operation.



The horsepower of the motor is dependent on the size of the clarifier. The motor drives the chain and sprocket which drives the worm gear. The worm gear drives the gear that is mounted on a shaft that drives the turntable. The motor shaft speed is reduced by a series of gear reducers.

We looked at the main parts of the drive unit. Now let's take a look at the sludge collector and the scum removal system mechanism. The main parts of the unit are: the rake arm; the scraper blades; the adjustable squeegees; the surface skimmer; the scum baffles; and the scum box.



The surface skimmer rotates at the same speed as the collector mechanism and is usually supported by the collector rake arm. The scum baffle prevents scum from flowing over the effluent weir. The surface skimmer collects the scum and deposits it in the scum box. The stilling well or influent baffle projects above the liquid and directs the influent downwards to assist in settling suspected solids and reduce short circuiting. Another important part of the secondary clarifier is the effluent weir, launder and pipe. An effluent weir goes around the circumference of the tank and allows clarified liquid to flow evenly from the tank. The effluent launder collects the tank overflow and takes it to a low point in the launder where a pipe is used to take the effluent to the chlorine contact basin or other means of treatment.

Some clarifiers may have a scum trough heater. The scum removal system rotates around the clarifier at a very slow rate. In subfreezing temperatures, the scum box and pipe could freeze. This problem can be overcome by using immersion heaters, or putting infrared lamps over the scum box. Some clarifiers are covered.



**As you have previously read, depending on the design and operation of the process, activated sludge has several interrelated components:**

1. Single aeration tank or multiple aeration tanks designed for completely mixed or plug flow.
2. An aeration source to provide adequate oxygen and mixing: sources can be compressed air, mechanical aeration, or pure oxygen.
3. A clarifier to separate the biological solids (activated sludge) from the treated wastewater.
4. A means of collecting the biological solids in the clarifier and recycling most of them (return activated sludge, RAS) to the aeration tank.
5. A means of removing or wasting excess biological solids (waste activated sludge, WAS) from the system.

## Review Process Goals

As previously noted, the activated sludge process can be used to remove carbonaceous BOD and also ammonia (nitrification). We can take the wastewater oxygen demand separated into two categories: carbonaceous and nitrogenous.

### Carbonaceous BOD Removal

The carbonaceous demand should be expressed as a function of the number of days that the demand will be measured; 3-day, 5-day (most common), 7-day, and 20-day time periods are commonly used. To obtain only carbonaceous oxygen demand, it may be necessary to inhibit nitrification by adding chemicals.

The rate and extent of BOD<sub>5</sub> (5-day BOD) removal in primary treated (settled) or untreated wastewater depends on the relative quantities of soluble, colloidal, and suspended BOD<sub>5</sub>, and a soluble BOD<sub>5</sub> content of approximately 20 to 40% of the total. These proportions may vary, particularly in warmer climates where long collection system residence times and the higher wastewater temperatures may result in a higher proportion of soluble BOD<sub>5</sub>. This is caused by the bacterial degradation of a portion of the colloidal and settleable fractions.

With typical municipal wastewater, a well-designed activated sludge process should achieve a carbonaceous, soluble BOD<sub>5</sub> effluent quality of 5mg/L or less. Similarly, with clarifiers designed to maximize solids removal at peak flows and adequate process control, the average SS in the effluent should not exceed 15 mg/L. On a practical basis, an effluent with 20/20 mg/L BOD<sub>5</sub> and SS should be attained, assuming proper operation. Potential capabilities of the process are 10/15 mg/L Bod<sub>5</sub> and SS. To consistently achieve values lower than 10/15 mg/L, some type of tertiary treatment is required.

### Nitrification

Of the total oxygen demand exerted by the wastewater, there is often a sizeable fraction associated with the oxidation of ammonia to nitrate. The autotrophic bacteria *Nitrosomonas* and *Nitrobacter* are responsible for this two-state conversion. Being autotrophic, these nitrifying organisms must reduce oxidized carbon compounds in the wastewater, such as CO<sub>2</sub> and its related ionic species, for cell growth. As a result, this characteristic markedly affects the ability of the nitrifying organisms to compete in a mixed culture.

The nitrifying bacteria obtain their energy by oxidizing ammonia nitrogen to nitrite nitrogen and then to nitrate nitrogen. Because very little energy is obtained from these oxidation reactions, and because energy is needed to change CO<sub>2</sub> to cellular carbon, the population of nitrifiers in activated sludge is relatively small. When compared to the normal bacteria in activated sludge, the nitrifying bacteria have a slower reproduction rate.

**Nitrifying organisms** are present to some extent in all domestic wastewaters. However, some wastewaters are not nitrified in existing plants because they are designed for the higher growth rate of bacteria responsible for carbonaceous removal. As the MCRT is increased, nitrification generally takes place. The longer MCRT prevents nitrifying organisms from being lost from the system when carbonaceous wasting occurs or, more accurately, the longer MCRT permits the build-up of an adequate population of nitrifiers.

Because of the longer MCRT required for nitrification, some systems are designed to achieve nitrification in the second stage of a two-stage activated sludge system.

The oxygen demand for complete nitrification is high. For most domestic wastewaters, it will increase the oxygen supply and power requirements by 30 to 40% because complete nitrification requires from 4.3 to 4.6 lb. of oxygen for each lb. of ammonia nitrogen (4.3 to 4.6 mg/mg) converted into nitrate, and wastewaters generally contain 10 to 30 mg/L of reduced nitrogen. Nitrification systems generally are not operated at intermediate (40 to 80%) removals; stable operation is achieved when essentially complete nitrification (greater than 90%) occurs.

Minimum acceptable dissolved oxygen (**DO**) concentrations of 2 to 3 mg/L have been reported, but nitrification appears to be inhibited when the oxygen concentration is lower than 1 mg/L.

Optimum growth of nitrifying bacteria has been observed in the pH range of 8 to 9 although other ranges have been reported. A substantial reduction in nitrification activity usually occurs at pH levels below 7, although nitrification can occur at low pH.

While nitrification occurs over a wide temperature range, temperature reduction results in a slower reaction rate.

The temperature effect is made less severe by increasing the MCRT. During the conversion of ammonia to nitrate, mineral acidity is produced. If insufficient alkalinity is present, the system's pH will drop and nitrification may be inhibited.

### **Bacteria Highlights**

A change in the numbers or predominance of microorganisms in activated sludge is usually gradual. The time required for a complete shift from one species to another will normally be seen in: 2 to 3 MCRT's.

A large amount of long filamentous bacteria will prevent good settling. The liquid above this mass is called the supernatant.

Endogenous respiration of microorganisms in an extended aeration plant will complete the oxidation process of an organic material.

The bug Nocardia causes frothing.

Saprophytic type bacteria produces the most acid in an anaerobic digester.

The best location for microscopic examination of activated sludge in a conventional system is at the effluent end of the aeration system. The examination can reveal a predominant number of rotifers and nematodes, this condition may indicate that the F/M ratio is too low and this would be normal in an extended aeration process.

**Food to microorganism ratio.** A measure of food provided to bacteria in an aeration tank.

$$\frac{\text{Food}}{\text{Microorganism}} = \frac{\text{BOD, lbs/Day}}{\text{MLVSS, lbs}}$$
$$= \frac{\text{Flow, MGD} \times \text{BOD, mg/L} \times 8.34 \text{ lbs/gal}}{\text{Volume, MG} \times \text{MLVSS, mg/L} \times 8.34 \text{ lbs/gal}}$$

$$\text{or} \quad = \frac{\text{BOD, kg/day}}{\text{MLVSS, kg}}$$

## Separate Stage Nitrification and Denitrification Systems

### Suspended Growth Nitrification

Single-sludge systems for BOD removal and nitrification require that the biomass inventory be retained long enough to establish a stable population of nitrifiers and that the HRT be such that the biomass can react with the ammonia-nitrogen entering the system. The overall approach for designing such systems is to determine the target SRT for the system based on influent characteristics (i.e., BOD, ammonia-nitrogen, organic nitrogen), environmental conditions such as temperature and flow characteristics (i.e., average daily, maximum monthly, diurnal peak).

Most activated sludge treatment plants will readily nitrify if they have sufficient aerobic SRT and can deliver sufficient oxygen maintaining 2 mg/L DO or greater. For plants having difficulty in nitrifying due to insufficient tank volume, there are some emerging technologies which can improve the process.

One of these is bioaugmentation. Bioaugmentation is accomplished by seeding the activated sludge process with an external source of nitrifying bacteria (also known as external bioaugmentation) or making process improvements to increase the activity of or enrich the nitrifier population (also known as *in situ* bioaugmentation).

External bioaugmentation uses either commercial sources of nitrifiers or sidestream processes to grow nitrifiers onsite. Early experiences with commercial sources were not consistent, so most work to date has been with sidestream production onsite (USEPA, 2008a). Two patented sidestream configurations for external bioaugmentation are the Single reactor High-activity Ammonia Removal Over Nitrite (SHARON) process and the In-Nitri® process. Both provide high temperature sidestream nitrification using ammonia from the anaerobically digested sludge dewatering liquid or digested supernatant. The nitrifiers grown in the sidestream reactor are fed to the main liquid treatment stream.

Both use flow through reactors with hydraulic retention times (HRT) in the 2 to 3 day range. In the SHARON process, nitrification is stopped mainly at nitrite by such process control methods as low DO concentration, low pH and/or low SRT. Full-scale operating systems for the SHARON process include installations at Utrecht, Rotterdam, Zwolle, Beverwijk, Groningen, The Hague in the Netherlands, and a system in New York City. Seeding from a diffused air biological nutrient removal process to stimulate nitrification in a parallel oxygen process has proved successful at a number of locations (Bott et al., 2007). Emerging *in situ* bioaugmentation technologies used to enhance nitrifier growth and shown to be successful in bench, pilot, and/or full-scale trials are described briefly below (USEPA, 2008a):

- The Bio-Augmentation Regeneration/Reaeration (BAR) process was developed in the U.S. and is identical to the Regeneration-DeNitrification (R-DN) process developed independently in the Czech Republic. It works by recycling ammonia-laden filtrate or centrate from dewatering of aerobically digested sludge to the head of the aeration tank. The sidestream is fully nitrified, seeding the aeration tank with additional nitrifying bacteria which allows for reduced SRT.
- Aeration Tank 3 (AT3) is similar to the BAR process except that it sends a smaller fraction of the return activated sludge (RAS) to the aeration tank in order to stop the nitrification process at the nitrite stage.
- Bio-Augmentation Batch Enhanced (BABE) process uses a SBR to grow nitrifiers by feeding it RAS and reject water from the sludge dewatering process. After treatment, concentrated nitrifiers are recycled to the head of the aeration tank.

- The Mainstream Autotrophic Recycle Enhanced N-removal (MAUREEN) Process was developed for the two-sludge treatment configuration at the Blue Plains Advanced Wastewater Treatment Plant in Washington, DC. The process involves sidestream treatment of WAS from the second stage to preferentially select AOB for bioaugmentation to the first sludge stage.

### **Attached Growth Nitrification**

Attached growth processes will also nitrify. Trickling filters and rotating biological contactors (RBCs) have historically been used for biological treatment of wastewater and can achieve nitrification with a low organic loading and a relatively high media volume. Typically, nitrification is achieved on the media after most of the BOD is removed since the heterotrophic population competes with the nitrifying organisms for oxygen and space on the media.

A major disadvantage of these technologies compared to suspended growth systems is that denitrification is fully dependent on addition of a supplemental carbon source. Suspended growth processes, on the other hand, can be designed to denitrify 80 percent or more of nitrate using the incoming BOD as the carbon source, which is a lower cost solution.

Consequently, trickling filters and RBCs have fallen out of favor for nutrient removal applications. In recent years, manufacturers have developed new technologies called biological aerated filters (BAF) to achieve BOD removal and nitrification. USEPA (2008a) identifies two existing BAF designs as established technologies: the Biofor® system and the Biostyr® system. The Biofor® filtration system is a fixed bed, upflow system with a dense granular media that is designed to expand during filtration. Air is sprayed into the filter to maintain an aerobic environment. The Biostyr® system is similar but uses a media that is less dense than water and held in place during operation by a screen at the top of the cell.

BAF can be configured in series to remove BOD in one unit and ammonia-nitrogen in the next or it can be designed for BOD removal and nitrification in a single unit depending on process goals. Advantages of BAF include its smaller footprint, higher hydraulic loading rates, and less susceptibility to washout than suspended sludge systems (Verma et al., 2006).

Another fixed film process that has gained popularity lately is moving bed biofilm reactors (MBBR). These reactors involve biofilm attached to a plastic media in a series of fluidized bed reactors. The plastic media help promote specialization of the biofilm within each reactor for either nitrification or denitrification (WEF and ASCE, 2006). Mixers or medium bubble diffuse aeration are used to keep the media suspended, depending on whether the system is anaerobic or aerobic. MBBR has a shorter SRT and smaller footprint than activated sludge processes. It has also proven to be effective in cold temperatures (Bott et al., 2007).

### **Separate-Stage Denitrification**

A separate-stage denitrification system may be appropriate for plants that are regularly achieving nitrification and need to add denitrification capabilities. Attached growth systems (denitrifying filters) are more common than suspended growth systems, although suspended growth systems have been used for some treatment plants. Suspended growth reactors typically have short SRTs (2 to 3 hrs.) and a small aerated zone following the denitrification zone to oxidize excess methanol and release contained nitrogen gas bubbles (WEF and ASCE, 2006).

Denitrification filters typically have a small footprint compared to suspended growth systems and have the added advantage of achieving denitrification and solids removal simultaneously. They were first installed in the 1970s and have evolved into two main process configurations (USEPA, 2007c):

- Downflow denitrification filters are deep bed filters consisting of media, support gravel, and a block underdrain system. Wastewater flow is directed over weirs onto the top of the filter where a supplemental carbon source, typically methanol, is added. Backwashing (typically air scouring and backwashing with air and water) is conducted at regular intervals to remove entrapped solids from the filter.

During operation, nitrate is converted to nitrogen gas and becomes entrained in the filter media, increasing head loss through the filter. To release entrained nitrogen, most denitrification systems have a nitrogen-release cycle operation that essentially “bumps” the filter by turning on the backwash pump(s) for a short period of time.

- Upflow continuous backflow filters do not have to be taken off-line for backwashing, as it is an integral part of the filtering process. Wastewater enters the bottom of the filter where a carbon source, typically methanol, is added. Water flows up through an influent pipe and is dispersed into the filter media through distributors. Filtered water discharges at the top of the filter. Filter media continuously travels downward, is drawn into an airlift pipe at the center of the filter, and is scoured before being returned to the filter bed.

**Performance of denitrifying filters depends on many factors including:**

- Influent weir configuration
- Filter media
- Underdrain system
- Backwash system
- Flow and methanol feed control

One wastewater system in Connecticut reported that key design issues for them were influent piping design to minimize aeration, maintaining a consistent flow to the filters, and control of methanol feed based on influent COD (Pearson et al., 2008).



The tubes held in this photograph should be placed in an autoclave. One tube is a standard or QA and the other would indicate contamination.



The refrigerated automatic WWT sampler will have a Data programmer that will allow you to set the time to collect the sample or samples. This machine can also measure the amount of the sample. These can also be used for the collection of composite samples. Sometimes you will see a pH probe with real-time readings sent to the Operator's Command Center. These are a common sight at most wastewater plants and SIUs.



## Key Design and Operational Issues

### Temperature

In general, as temperature of the wastewater increases, the rate of nitrification and denitrification increases. For the typical range of liquid temperatures between 10 and 25° C, the nitrification rate will approximately double for every 8 to 10° C increase in temperature (WEF and ASCE, 2006). Rapid decreases in temperature without acclimation time will, however, cause even slower nitrification rates than predicted, strictly by the temperature change. Denitrification rates will also increase with increasing temperature, although not at the same magnitude as nitrification rates.

### Dissolved Oxygen

Nitrifying bacteria are also more sensitive to DO levels as compared to aerobic heterotrophic bacteria, with growth rates starting to decline below 3 to 4 mg/L with significant reduction below 2 mg/L. The nitrification rate at a DO concentration of 0.50 mg/L is only about 60 percent of that at a 2.0 mg/L DO concentration. Studies have shown that the amount of oxygen available to nitrifying bacteria can be limited by floc size, requiring higher bulk DO concentrations under higher organic loading conditions (Stenstrom and Song, 1991). At DO concentrations less than 0.5 mg/L, the effect is greater for *Nitrobacter* than for *Nitrosomonas*. This can result in higher NO<sub>2</sub>-N in the effluent and have a negative impact on chlorine disinfection as 1 g of NO<sub>2</sub>-N consumes 5 g chlorine for oxidation. DO must normally be less than 0.2 to 0.5 mg/L, otherwise there will be inhibition of the denitrification process.

### pH and Alkalinity

Nitrification generally operates well within a pH range of 6.8 to 8.0 (WEF and ASCE, 2006). At lower pH values the nitrification rate is much slower and at pH values near 6.0 the nitrification rate may only be about 20 percent of that with a pH of 7.0 (Tchobanoglous et al., 2003). Alkalinity is consumed during the nitrification process but partially replenished (up to 62.5 percent) during the denitrification process. Depending on the influent wastewater alkalinity, there may be a sufficient alkalinity reduction due to nitrification to decrease to unacceptable levels. Addition of chemicals such as lime, sodium hydroxide, or soda ash can be used to replace the alkalinity consumed by nitrification to maintain acceptable pH levels.

### Carbon Sources for Denitrification

Denitrifying bacteria need a readily available carbon food source, such as soluble BOD, to ultimately convert nitrate to nitrogen gas. WWTPs that meet very low total nitrogen limits typically use a secondary anoxic zone in which supplemental carbon is added. Supplemental sources can be "internal" such as fermented wastewater or sludge, or "external" sources such as purchased chemicals.

#### **Methanol is currently the most common external carbon source used in denitrification because of its low cost. It has several drawbacks, however, namely:**

- It is highly flammable and implicated in some storage tank explosions and fires (Dolan, 2007); however with proper design and operation problems can be minimized.
- It is not the most efficient source for most treatment configurations.
- Costs have begun to fluctuate widely (deBarbadillo et al., 2008).
- Availability is a problem in some areas (Neethling et al. 2008).
- Reported low growth rates under cold temperatures (Dold et al. 2008).

Other sources of carbon include ethanol, acetic acid, corn syrup, molasses, glucose, glycerol, and industrial waste products. The WEF Nutrient Challenge Research Plan (2007) identified research on alternative carbon sources as priority for operators, owners, and engineers of wastewater systems. In December of 2007, the 2nd External Carbon Workshop was held in Washington, DC to discuss the state of the technology and research needs. WERF is also currently formulating a standard protocol for evaluation of external carbon alternatives.

### **Nitrification Inhibition from Toxic Chemicals**

Nitrifying bacteria are very sensitive to heavy metals and other inorganic compounds in waste-water. The Local Limits Development Guidance Manual (USEPA 2004) has been the main source of information on inhibitory effects for a variety of wastewater treatment processes including nitrification. Appendix G of the 2004 version provides a summary table with the reported range of nitrification inhibition threshold levels for a number of metals, non-metal inorganics, and organic compounds. Actual inhibitory effects are site-specific and depend on many factors including the nature of biodegradable organic material, microorganism speciation, acclimation effects, temperature, and water quality conditions.

### **Wet Weather Events**

Wet weather events can increase inflow and infiltration into the collection system and subsequently increase the hydraulic load to the wastewater treatment plant. This can in turn reduce the SRT leading to reduced performance of nitrification process units. In addition, wet weather flows have different characteristics than typical wastewater influent flow and can be less favorable for nitrification and denitrification. Conditions that are less favorable for nitrification include decreased alkalinity and sudden temperature drops. Lower biodegradable COD concentrations and increased DO make wet weather flows less amenable to denitrification.

Flow equalization basins can be used to handle wet weather events; however, this requires available space and capital investment. USEPA (2008a) identifies a number of innovative storage and treatment technologies used to manage influent flows during wet weather events.

### **Guidance for Selecting Process Modifications**

Nitrogen removal requires first that a biological nitrification process be present or that the facility be modified to accomplish nitrification. Considerably more volume is needed for activated sludge nitrification compared to designs for BOD removal only. If there is insufficient space to accommodate the increased volume, suspended growth or hybrid process options that require less space such as the MBR process or IFAS systems with suspended media in the activated sludge process should be considered. Another option is to use a fixed film nitrification process after the suspended growth process clarification step. This could be a BAF or a plastic media trickling filter. However, if nitrogen removal is required, an exogenous carbon source is needed, which has higher operating costs than using the influent BOD for denitrification.

Nitrification systems need sufficient oxygen transfer for ammonia oxidation in addition to BOD removal. Such systems should consider the impact to diurnal loadings and ammonia addition in recycle streams. The influent TN concentration may have daily peak values that are 1.5 to 2.0 times the daily average loading. Higher peak loadings require longer SRTs to assure that sufficient nitrifying bacteria are present to remove ammonia at a greater rate, while maintaining a low effluent ammonia concentration. Often anaerobic digester sludge dewatering operations occur during the day and produce return recycle streams high in ammonia concentration (500-1000 mg/L) at times that coincide with the high influent diurnal ammonia loads. Recycle equalization or treatment helps to provide a more stable nitrification system and lower effluent NH<sub>3</sub>-N concentrations.

**In many cases, it is advantageous to incorporate a denitrification pre-anoxic step with nitrification (MLE process) due to the many benefits and improved operational stability. The advantages include**

- 1) less aeration energy as the nitrate produced can be used for BOD removal,**
- 2) the production of alkalinity to offset the alkalinity used by nitrification, which in some cases eliminates the need to purchase alkalinity, and**
- 3) a more stable, better settling activated sludge process as the anoxic-aerobic processes favor good settling floc-forming bacteria over filamentous growth.**

The effluent nitrogen goals greatly affect the process design choices and system operation. For an effluent goal of 10 mg/L TN, an MLE process is often sufficient for activated sludge treatment with secondary clarifiers or membrane separation. However, with water conservation leading to more concentrated wastewater, these processes alone may not be sufficient due to the fact that they are limited to 80-85% removal of the influent TN.

For TN effluent goals of 3 to 5 mg/L or lower, some form of post anoxic treatment is generally needed. One option is to convert an MLE process to a Bardenpho process by adding another anoxic aerobic set of tanks. Although the endogenous respiration rate of the bacteria can be used to consume nitrate in the post anoxic tanks, it is often necessary to add an exogenous carbon source. Other alternatives to using exogenous carbon sources include denitrification filters instead of adding more activated sludge tank volume, step feed with carbon addition in the last pass, and IFAS processes.

Denitrification processes require sufficient carbon to drive the nitrate/nitrite reduction reactions. Characterization of the influent wastewater with regard to its organic strength and soluble fraction and the TN and ammonia concentrations is needed to fully understand a system's carbon needs. In addition, design and operating methods that eliminate or minimize DO feeding to anoxic zones can reduce the amount of exogenous carbon needed and provide a more stable operation. Low DO zones prior to downstream anoxic tanks or for withdrawal of recycle to preanoxic zones should be considered.

### **Impacts on Sludge Production and Handling**

It has been documented by both research and full scale experiments that BOD removal by activated sludge using nitrate as the electron acceptor instead of DO will result in a 20% or more reduction in waste activated sludge (WAS) production for the same operating conditions. Full-scale investigations near Melbourne, Australia achieved as high as a 40% reduction in WAS, and implementation of nitrogen removal at the York River, VA, plant resulted in a reduction of more than 50% in WAS production. The impact this will have on total sludge production by a treatment plant will depend upon how much waste sludge is produced by other treatment units such as primary clarifiers and chemical treatment with precipitating chemicals.

Additionally, implementation of nitrogen removal at conventional activated sludge plants can improve the thickening characteristics due to decreasing the amounts of filamentous bacteria in the activated sludge. If an external carbon source is added to improve the rate of denitrification, there will be an increase in WAS production compared to when no external carbon source is added. If an external carbon source is used to supplement denitrification, it is likely that the small increase in solids production will be offset by endogenous respiration due to longer SRTs. Solids produced from nitrogen removal processes generally thicken and dewater well and show no negative impact on any solids processing system.

## What is in Municipal Wastewater?

**Municipal wastewater** consists primarily of domestic wastes from households and industrial wastewater from manufacturing and commercial activities. Both types of wastewater are collected in sanitary sewers, and are usually treated at a municipal wastewater treatment plant. After treatment, the wastewater is discharged to its *receiving water* (i.e., a river, an estuary, or an ocean).

Wastewater entering a treatment plant may contain organic pollutants (including raw sewage), metals, nutrients, sediment, bacteria, and viruses.

**Toxic substances** used in the home – motor oil, paint, household cleaners, and pesticides - or substances released by industries, also make their way into sanitary sewers.



**Industrial processes**, such as steel or chemical manufacturing, produce billions of gallons of wastewater daily. Some industrial pollutants are similar to those in municipal sewage, but often are more concentrated. Other industrial pollutants are more exotic and include a variety of heavy metals and synthetic organic compounds. In sufficient dosages, they may present serious hazards to human health and aquatic organisms. Unlike municipal or industrial sources of pollution, which come from a single discrete facility, other sources are usually more diffuse. For example, rainwater or snowmelt washing over farmlands may carry topsoil and fertilizer residues into nearby streams.

### Stormwater

This type of runoff, called *stormwater*, may carry oil and gasoline, agricultural chemicals, nutrients, heavy metals, and other toxic substances, as well as bacteria, viruses, and oxygen-demanding compounds. A recent EPA study indicated roughly one-third of identified cases of water quality impairment nationwide are attributable to stormwater, whether from farmland, streets, parking lots, construction sites, or other sources.

### Animal Feeding Operations (AFOS)

Animal Feeding Operations are livestock-raising operations, such as hog, cattle and poultry farms, that confine and concentrate animal populations and their waste. Animal waste, if not managed properly, can run off to nearby water bodies and cause serious water pollution and public health risks. There are approximately 450,000 AFOs in the United States.

### Acid Mine Drainage

Acid Mine Drainage is one of the most significant environmental impacts resulting from past and current mining activities. It has been cited as a major cause of stream pollution in northern Appalachia (PA, W. VA, VA, MD); more than 50 percent of stream miles in PA and WV do not meet water quality standards because of acid mine drainage impacts. In addition, there are an estimated 200,000 abandoned hardrock mines nationwide and somewhere between 2,000 and 10,000 active ones. Some of these mining operations produce waste material and other conditions that result in acid mine drainage as well as discharges of heavy metals which affect aquatic life and drinking water sources.

## Biological Phosphorus Removal and Combination Processes

This section provides an overview of the principles behind biological phosphorus removal (BPR). It describes existing configurations that can achieve phosphorus removal and in many cases, simultaneous nitrogen removal. Key operational issues, impacts on sludge handling, and a summary of ongoing research related to BPR removal are also provided.

### Principles

Biological phosphorus removal is achieved by contacting phosphorus accumulating organisms (PAOs) in the RAS with feed, containing volatile fatty acids (VFA), in a zone free of nitrates and DO (anaerobic zone). Phosphorus is released in this zone providing energy for uptake of VFAs that are polymerized and stored inside the PAO cells. The anaerobic zone is followed by an aeration zone where the polymerized VFAs are metabolized and phosphorus is taken up again to store excess energy from the metabolism.

The phosphorus content of the mixed liquor suspended solids (MLSS) would be similar to that of the waste activated sludge (WAS). When nitrification occurs in the aeration basin, nitrates will be present in the RAS, resulting in some metabolism of the VFA before storage, thereby reducing subsequent phosphorus uptake. Some form of denitrification (anoxic zones) must be used to reduce/remove the nitrates from the RAS. The process flow sheets now known as Pho-redox (A/O) and 3 Stage Pho-redox (A2/O) as well as the modified Bardenpho process were first published by Barnard (1975) as the Pho-redox flow sheets for the removal of phosphorus. The theory for the functioning of the PAO was first suggested by Fuhs & Chen (1975).

### Fuhs & Chen Theory

PAOs have the ability to store a large mass of phosphorus in their cells in the form of polyphosphates. Polyphosphates are formed by a series of high-energy bonds. The organisms can subsequently get energy from breaking these bonds. The polyphosphate globules within the cells function just like energy storage batteries. The storage of polyphosphates (energy), takes place in the aeration zone. In the anaerobic zone, these obligate aerobic bacteria can take up short chain VFA such as acetate and propionate and store them in the form of intermediate products such as poly- $\beta$ -hydroxybutyrate (PHB). The energy for transferring the food across the cell membranes in the anaerobic zone is derived from breaking phosphorus bonds. Excess phosphates are released to the liquid in the anaerobic zone.

Some magnesium and potassium ions are co-transported across the cell walls with phosphates. PAOs can only get energy from the food they have taken up in the anaerobic zone when they pass to the aerobic zone where oxygen is available. They use oxygen to metabolize the stored products, deriving enough energy to take up all the released phosphates as well as those in the influent, and store them in the cells. The WAS will contain sufficient phosphate-enriched PAOs to remove most of the phosphorus from the waste steam once enhanced BPR is established.

The right carbon source, in this case a combination of acetates and propionates, is essential for BPR. The wastewater characteristics are thus important. In general, it can be said that you need at least 40:1 COD:TP or about 18:1 BOD<sub>5</sub>:TP in the process influent wastewater to reduce effluent phosphorus to less than 1.0 mg/L. In addition, some of the COD should consist of short chain VFAs. More COD may be required if nitrates must also be denitrified.

Biological phosphorus removal can work in with or without nitrification. When nitrification occurs, denitrification within the process is important to reduce the nitrates that may be returned with the RAS. While the anaerobic zone serves mostly as a contact zone for VFAs and PAOs, some fermentation of easily biodegradable carbon compounds (rbCOD) to acetate and propionate may take place. In most plants the readily biodegradable material is in short supply and must be reserved for the PAOs.

**When nitrate or oxygen is discharged to the anaerobic zone, two things may happen, both undesirable:**

- They will prevent fermentation of rbCOD to acetic and propionic acid.
- Nitrates or DO could serve as electron acceptors for PAOs and other organisms that will metabolize the VFA and so deprive the PAOs of the substance that they need to store for growth and phosphorus removal.

In the absence of electron acceptors such as DO and nitrates in the anaerobic zone, PAOs are favored to grow since they can take up and store the VFA under anaerobic conditions, thereby making it unavailable for other aerobic and facultative heterotrophs in the aerobic zone.

Biological removal of both nitrogen and phosphorus at the same WWTP is common. Both functions require a carbon source. While denitrification organisms can feed on quite a number of easily degradable materials such as methanol, sugar, glucose, acetate and propionate, PAOs are restricted to the latter two for polymerization and storage (e.g. adding methanol to the anaerobic zone will reduce nitrates but not assist in the removal of phosphorus).

### **Current Configurations**

The basic design of anaerobic, anoxic, and aerobic zones, in that order, has been achieved in many different configurations. The configurations vary in the number of stages, the nature and location of recycles, and the operation of the process. Each process was modified from the standard biological activated sludge design in order to accomplish various design goals (e.g., protection of the anaerobic zone from excess nitrate recycle). The primary processes are listed below.

**Of these, all will also biologically remove nitrogen except for the Pho-redox process.**

- Pho-redox (A/O)
- 3 Stage Pho-redox (A2/O)
- Modified Bardenpho
- University of Capetown (UCT) and Modified UCT (MUCT)
- Johannesburg (JHB), Modified Johannesburg, and Westbank
- Orange Water and Sewer Authority (OWASA)
- Oxidation ditches with anaerobic zones or phases added
- SBR operated with an anaerobic phase
- Hybrid chemical/biological processes

The performance of these technologies depends on many site specific factors, including but not limited to temperature, hydraulic and organic loading, recycle rates, and return streams. The technologies described in this section are generally capable of phosphorus removal to effluent levels between 0.5 and 1.0 mg/L. Operating strategies that can be used to enhance biological treatment and achieve these and, in some cases, even lower effluent levels.

Biological phosphorus removal can be combined with other technologies to achieve very low effluent concentrations (< 0.2 mg/L). Chemical addition combined with biological removal of phosphorus has been used to consistently achieve low levels. WEF and ASCE (1998) recommend that WWTPs have chemical addition capabilities even for well operating BPR plants to provide backup phosphorus removal in the event of power outages, pipe breaks, or other unforeseen events.

Solids removal can also be a limiting factor in achieving phosphorus removal below 0.2 mg/L. Very low phosphorus levels generally require a TSS level of less than 5 mg/L. Tertiary filtration (see membrane bioreactors), and advanced clarification processes can achieve TSS levels less than 5 mg/L.

## **Pho-redox (A/O) and 3 Stage Pho-redox (A2/O)**

The Pho-redox (A/O) process is a conventional activated sludge system with an anaerobic zone at the head of the aeration basin. The RAS is pumped from the clarifier to the anaerobic zone. It is a low SRT process, operated to avoid nitrification. With no nitrates in the RAS the process is reliable and easy to operate except at temperatures in excess of 25°C when nitrification is difficult to avoid. The 3 Stage Pho-redox (A2/O) process adds an anoxic zone after the anaerobic zone to achieve de-nitrification.

In addition, a nitrate rich liquor is recycled from the end of the aerobic zone to the head of the anoxic zone to enhance de-nitrification. A shortcoming of the 3 Stage Pho-redox process is that there will be nitrates present in the RAS, potentially making the process unreliable.

### **Modified Bardenpho**

The Bardenpho process removes nitrogen to low concentrations. The addition of an anaerobic zone at the head of the process enables phosphorus removal as well. The process consists of 5 stages: an anaerobic stage followed by alternating anoxic and aerobic stages. A nitrate-rich liquor is recycled from the first aerobic stage, designed for complete nitrification, to the first anoxic stage. The RAS is recycled from the clarifier to the beginning of the anaerobic zone. Since the nitrates in the RAS ranges from 1 to 3 mg/L, it does not seriously interfere with the mechanism for phosphorus removal as can happen in the 3 Stage Pho-redox process.

### **University of Cape Town (UCT) and Modified UCT (MUCT)**

The UCT process was designed to reduce nitrates to the anaerobic zone when high removal of nitrates in the effluent is not required. It consists of three stages: an anaerobic stage, an anoxic stage, and an aerobic stage. The RAS is returned from the clarifier to the anoxic zone instead of the anaerobic zone to allow for denitrification and to avoid interference from nitrate with the activation of the PAOs in the anaerobic stage. A nitrate rich stream is recycled from the aerobic zone to the anoxic zone. Denitrified mixed liquor is recycled from the anoxic zone to the anaerobic zone. Several modifications of the process exist. Sometimes it can be difficult to achieve the level of denitrification in the anoxic zone required to protect the anaerobic zone from nitrates when the zone is receiving both RAS and high internal nitrate recycle flows. This problem led to the development of the modified UCT process, which splits the anoxic zone into two stages. The nitrate rich recycle from the aerobic zone is recycled to the head of the second anoxic stage. The nitrate containing RAS is recycled to the first anoxic stage where it is denitrified. Next, the denitrified RAS is recycled from the end of the first anoxic stage back to the head of the anaerobic stage and mixed with the incoming wastewater.

### **Johannesburg (JHB), Modified Johannesburg and Westbank**

The JHB process is similar to the 3 Stage Pho-redox process, but has a pre-anoxic tank ahead of the anaerobic zone to protect the zone from nitrates when low effluent nitrates are not required. The low COD of the wastewater limited the de-nitrification capacity in the original plant (Northern Works), resulting in nitrates in the RAS. This reduced BPR so much that a pre-anoxic tank was included on the RAS line to remove the nitrates from the RAS flow using endogenous respiration, before the flow entered the anaerobic zone. The modified JHB process adds a recycle from the end of the anaerobic zone to the head of the pre-anoxic zone to provide residual, readily biodegradable compounds for denitrification.

The Westbank process is similar to the JHB process but adds some primary effluent to the anaerobic zone to assist in denitrification with the remainder of the primary effluent being discharged to the anaerobic zone. During storm flows, excess flow is passed directly to the main anoxic zone. VFA obtained from acid fermentation of the primary sludge is passed to the anaerobic zone.

### **Orange Water and Sewer Authority (OWASA)**

The OWASA process was developed by adding activated sludge from a biological nitrogen removal process to a trickling filter plant. Then, nitrified effluent from the trickling filter is fed to the aerobic zone of the activated sludge system. Because the VFAs have been destroyed by the trickling filter, it is necessary to ferment the settled organic solids from the primary clarifier to produce sufficient VFAs for BPR.

Next, the fermented supernatant is passed to an anaerobic (nutrition) zone and mixed with the RAS to initiate BPR. Mixed liquor then flows from the nutrition zone to an anoxic zone and then to an aerobic zone. Alternatively, simultaneous nitrification and denitrification takes place in the aeration zone.

### **Oxidation Ditches**

There are several oxidation ditch designs that can remove phosphorus. They normally consist of an anaerobic zone ahead of the oxidation ditch whereas simultaneous nitrification and denitrification takes place within the ditches. Oxidation ditches typically operate as racetrack configurations around a central barrier, with forward mixed liquor flows of approximately 1 foot per second or more. It is possible, by manipulating the DO transferred to the mixed liquor, to establish both anoxic, aerobic and near anaerobic zones within the racetrack configuration, even though the high flow velocities accomplish complete mixing of the wastewater with the RAS.

There are many forms of oxidation ditches, such as the Carousel, the Pasveer Ditch and the Orbital process. The Orbital process creates anaerobic and anoxic zones in the outer of three concentric oval shaped ditches with the RAS recycled from the clarifier to the anoxic zone. It is also possible to introduce an anaerobic tank before the ditch to accomplish BPR in the combined system. The Pasveer Ditch and the Carousel system also can be used in conjunction with an anaerobic zone to accomplish BPR, in addition to simultaneous nitrification and denitrification within the ditches. Because of the very high internal recycle within the ditches, very low nitrate concentrations can be achieved in the mixed liquor before settling, and anaerobic conditions are easy to maintain in the anaerobic zone, thereby resulting in efficient BPR. The layout would resemble a Pho-redox process with simultaneous nitrification-denitrification (SND) in the aeration basin. Alternatively the Carousel or Pasveer Ditch could be used as the aeration stage in either the 3 Stage Pho-redox or the Modified Bardenpho process.

The VT2 process at Bowie, MD, operates two Pasveer ditches in series with dedicated anoxic, near anaerobic and aerobic zones. It also has a side stream anaerobic zone that receives only 30 percent of the influent flow to enhance BPR. Denitrified MLSS for the anaerobic zone are obtained from the end of the near anaerobic zone of the adjacent ditch. Operated without primary sedimentation, the system consistently obtains very low (<0.25 mg/L) effluent TP without chemicals or effluent filtration. The ditches are operated in series because the plant has limited clarification capacity, and series operation results in lower MLSS concentrations to the clarifiers. The biodenipho process also uses pairs of ditches.

The ditches in the biodenipho process operate in alternating anoxic-aerobic modes. An anaerobic tank is placed before the ditches for BPR and the ditches are alternated between nitrification and denitrification.

### **Sequencing Batch Reactors (SBR)**

SBRs are fill-and-draw reactors that operate sequentially through the various phases by means of adjusting the mixing and aeration. The reactor phases can be set and automated to allow the mixed liquor to go through an anaerobic/anoxic/aerobic progression as is necessary for removal of phosphorus and nitrates. Because of the fill-and-draw nature of SBRs, it actually is necessary to remove the nitrates remaining from the previous cycle before anaerobic conditions can be established, thus the typical treatment progression becomes anoxic/anaerobic/aerobic.

However, SBRs are almost always operated without primary sedimentation, so they still usually have a favorable BOD<sub>5</sub>:TP ratio for effluent TP of somewhat less than 1.0 mg/L during the settling phase.

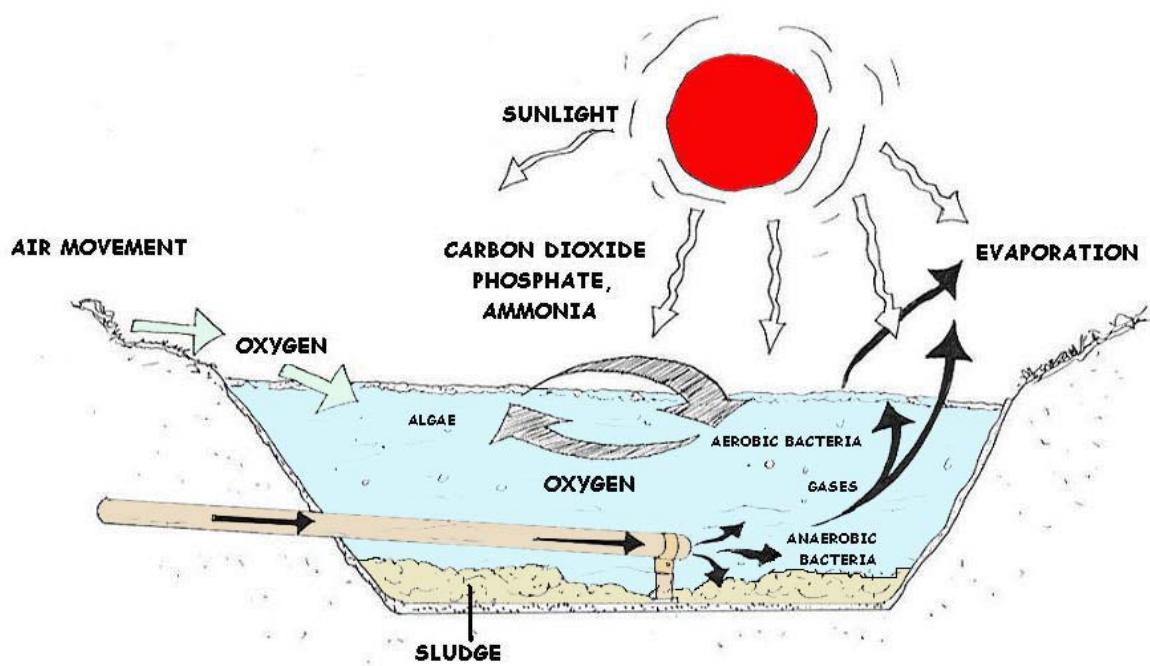
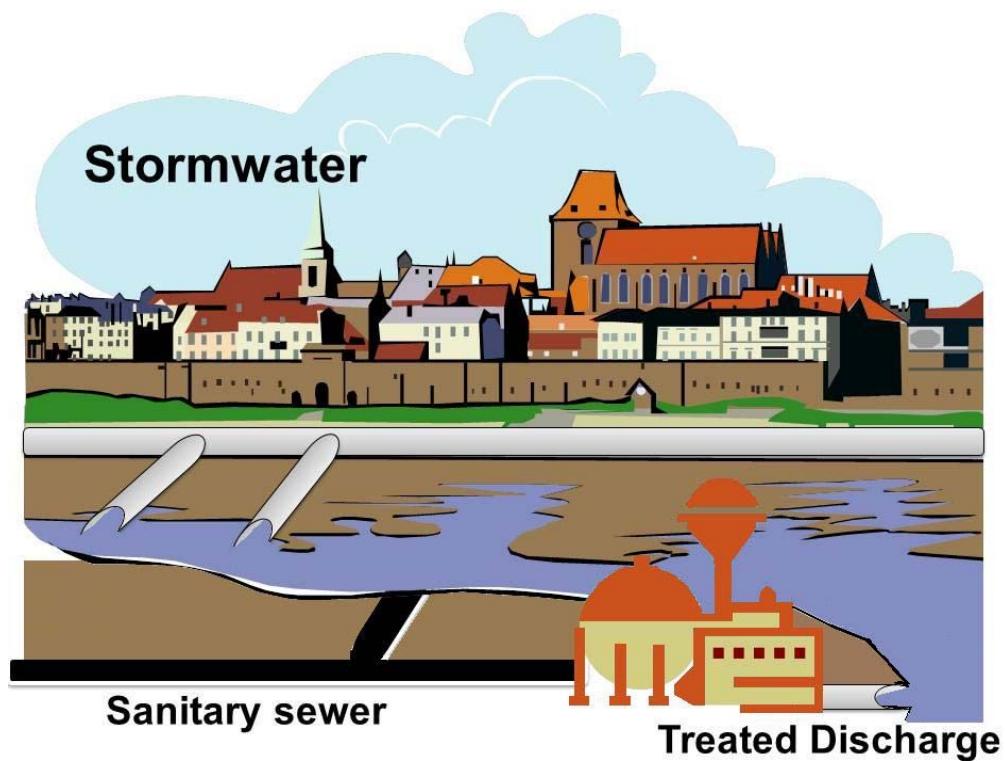
#### **Hybrid Chemical / Biological Processes**

The PhoStrip configuration, used mainly in non-nitrifying plants, pulls a side stream off the RAS in a conventional activated sludge plant. The side stream is concentrated and retained for a day or more in a thickening tank where the solids blanket is deep enough to produce anaerobic conditions and fermentation, resulting in the release of phosphates by the microorganisms. Lime is then added to the supernatant stream to precipitate and remove phosphate. The thickened, fermented sludge is passed back to the main aeration basin. Existing plants include Seneca Falls, NY; Lansdale, PA; Adrian, MI; Savage, MD; Southtowns, NY; Amherst, NY; and Reno-Sparks, NV.

The Biological Chemical Phosphorus and Nitrogen Removal (BCFS) configuration is similar to the modified UCT process. In this process, a sludge stream is removed from the anaerobic zone. Ferric chloride is added to the sludge thickener to remove phosphate. This provides an advantage over chemical addition to the secondary clarifier because it does not require the chemical sludge to be recycled. There is an existing plant at Holten in the Netherlands (WEF and ASCE, 2006), but no performance data are available.



Microscope being utilized to view activated sludge MO's. Thiothrix is a type of filament that can grow in the aeration basin of an activated sludge plant. Low DO levels are a possible cause to the growth of this long filament.



**Stabilization pond** - A large shallow basin used for wastewater treatment by natural processes involving the use of algae and bacteria to accomplish biological oxidation of organic matter.

## Return and Waste Activated Sludge Systems

The RAS system pumps the settled sludge from the secondary clarifier back to the aeration tank. It is important that this system returns the RAS to the aeration tank before the microorganisms deplete the entire DO. The RAS must also be as concentrated as possible and the flow must be accurately measured and controlled.

To accomplish this, the RAS pumping system must have a positive variable flow control device and the RAS flow must be adjustable between the minimum and maximum range for proper process control. The desired return flow to the aeration tank could also be automatically paced to secondary influent flow.

All activated sludge processes must have a WAS system to remove excess microorganisms. This is necessary to control the F/M and MCRT. If the process is to reliably meet discharge requirements, this system must provide a positive, flexible, and reliable means of removing excess microorganisms.

It is essential for the system to have flow-metering and pumping equipment that function completely independent of other activated sludge control devices. The most positive and flexible system will include an independent pumping system with flow adjustability (for example, variable speed drive) and a flow meter that provides feedback into a flow-control device.



Such a system can be set for a given wasting rate with complete assurance that variable system head or concentration conditions will not affect its ability to remove the microorganisms required. WAS systems must have sufficient capacity to deal with both the hydraulic and/or organic load changes and process changes.

### Aeration and DO Control

The purpose of aeration is two-fold: oxygen must be dissolved in the liquid in sufficient quantities to maintain the organisms and the contents of the tank must be sufficiently mixed to keep the sludge in suspension.

Mixing energy and oxygen transfer are provided through mechanical or diffused aeration. The amount of oxygen that has to be transferred by the aeration system is theoretically equal to the amount of oxygen required by the organisms in the system to oxidize the organic material.

The DO concentration in the aeration tank must be sufficient to sustain at all times the desirable microorganisms in the aeration tank, clarifier, and return sludge line back to the aeration tank. When oxygen limits the growth of microorganisms, filamentous organisms may predominate and the settleability and quality of the activated sludge may be poor.

On the other hand, over aeration can create excess turbulence and may result in the breakup of the biological floc and waste energy. Poor settling and high effluent solids will result. For these reasons, it is very important to periodically monitor and adjust the aeration tank DO levels and, for diffused air systems, the air flow rates.

In practice, the DO concentration in the aeration tank should normally be maintained at about 1.5 to 4 mg/L in all areas of the aeration tank at all times for adequate microorganism activity. Poor sludge settling as a result of filamentous organisms has been associated with mixed liquor DO concentrations below 0.5 mg/L. Above 4 mg/L, treatment usually does not significantly improve but power usage increases aeration costs considerably.

#### **RAS Control**

To properly operate the activated sludge process, a good settling mixed liquor must be achieved and maintained. The MLSS are settled in a clarifier and then returned to the aeration tank as the RAS. This keeps a sufficient concentration of activated sludge in the aeration tanks so that the required degree of treatment can be obtained in the allotted time period. The return of activated sludge from the secondary clarifier to the aeration tank is a key control parameter of the process.

**The secondary clarifiers have two basic functions:**

- ◆ to clarify the secondary effluent through solids/liquid separation; and
- ◆ to rapidly collect and thicken the settled solids for return to the aeration tanks or wasting to the sludge processing facilities.



Example of a Sludge Press.

## Constant Rate Versus Constant Percentage Return

There are two basic ways for returning sludge to the aeration tank:

- ◆ at a constant rate, independent of the secondary influent flow rate, and
- ◆ at a constant percentage of the varying secondary influent flow.

Clarifier size and hydraulics may limit the range of practical return adjustments. Regardless of calculated values, return rates should not be reduced to the level where slowly moving, thick clarifier sludge will plug the sludge withdrawal pipes.

Also, low return rates during the night should be increased to approach the anticipated higher return rates during the day before, rather than after, the increased wastewater flows actually reach the plant. Increasing the return sludge flow after the flow increase may cause a hydraulic overload condition resulting in a carryover of solids into the clarifiers (washout).

### Constant Rate Control

Returning activated sludge at a constant flow rate that is independent of the secondary influent wastewater flow rate results in a continuously varying MLSS concentration that will be at a minimum during peak secondary influent flows and a maximum during minimum secondary influent flows.

The aeration tank and the secondary clarifier must be looked at as a system where the MLSS are stored in the aeration tank during minimum wastewater flow and then transferred to the clarifier as the wastewater flow and then transferred to the clarifier as the wastewater flows initially increase.

The clarifier acts as a storage reservoir for the MLSS during periods of high flow. The clarifier has a constantly changing depth of sludge blanket as the MLSS moves from the aeration tank to the clarifier and vice versa.

### Constant Percentage Control

The second approach is to pace the return flow at a fixed percentage of the influent wastewater flow rate (Q), at a constant rate (R)  $R/Q$ . This may be done automatically with instruments, or manually with frequent adjustments.

This approach keeps the MLSS and sludge blanket depths more constant throughout high and low flow periods and also tends to maintain a more constant F/M and MCRT.

### Settleability

The settleability test can be used to estimate the desirable sludge return rate. This method uses the sludge volume in a 2-L settleometer at the end of a 30-minute settling period to represent the underflow and the supernatant volume to represent the overflow.



Above photograph, Flagella.



Lagoons in series.

## Rotating Biological Contactors RBC

Rotating Biological Contactors is a remediation technology used in the secondary treatment of wastewater. This technology involves allowing wastewater to come in contact with a biological medium in order to facilitate the removal of contaminants.

In its simplest form, a rotating biological contactor consists of a series of discs or media blocks mounted on a shaft which is driven, so the media rotates at right angles to the flow of sewage. The discs or media blocks are normally made of plastic (polythene, PVC, expanded polystyrene) and are contained in a trough or tank so that about 40% of their area is immersed.

The biological growth that becomes attached to the media assimilates the organic materials in the wastewater. Aeration is provided by the rotating action, which exposes the media to the air after contacting them with the wastewater. The degree of wastewater treatment is related to the amount of media surface area and the quality and volume of the inflowing wastewater.



Rotating Biological Contactors can be supplied as part of an integral package plant to treat sewage from various communities. Integral units are provided in sizes of up to a 500-population equivalent. A smaller version is also available for small private installations.

Modular systems can also be adapted to cater to populations of any number.

Multiple units have been used for populations in excess of 5000.

Each plant is designed to meet the specific requirements of the site and the effluent quality required.

### Key Advantages

- Short contact periods are required because of the large active surface.
- Capable of handling a wide range of flows.
- Sloughed biomass generally has good settling characteristics and can easily be separated from the waste stream.
- Operating costs are low, as little skill is required in plant operation.
- Retention times are short.
- Low power requirements.
- Low sludge production and excellent process control.

### Problems

White biomass over most of a RBC disc can be resolved by increasing the age of the sludge.

### RBC Principles

The principles of the rotating biological contactor originated in the early 1900's but its application to sewage treatment did not occur until the 1960's when the present system was developed. The process employed relies on the well-established principle of biological oxidation using naturally occurring organisms to ensure even the most stringent effluent standards can be achieved.

## Primary Settlement Zone



## Rotating Biological Contactors

Incoming flows of crude sewage enter the RBC primary settlement zone, which is designed to have a buffering capacity of balancing flows up to 6 mgd (million gallons a day). Settlement solids are retained in the tank's lower region while the partially clarified liquor passes forward to the biozone where it makes contact with the slowly rotating disks.

### Contactors

#### Installation of Rotating Biological Contactors

Rotating Biological Contactors are available in sizes from 1100mm diameter up to 3800mm in diameter. The media packs that form the rotors are manufactured from vacuum formed black polyethylene sheets supported on the central shaft with a galvanized steel framework. The central shaft is manufactured from mild steel tube, protected internally against corrosion and fitted with end stub shafts, which are supported on split bearings.



#### Gearbox and Drive mechanism

Rotation is provided by a shaft mounted gearbox and motor fitted at one end.

### **Biozone**

The rotor assembly is suspended within the biozone with 40% of the diameter submerged in the liquor at any one time. The disks slowly rotate and the continuous alternate exposure to air and sewage results in a growth of organisms known as biomass which adheres to the disks. These organisms occur naturally in the sewage and carry out the purification process by feeding off the impurities present in the sewage. As they have a short life cycle, these organisms are continually shearing off the rotating disks and pass from the biozone to the final zone.

The biozone is fitted with a series of baffles between each bank of media to prevent short circuiting and to ensure maximum performance.

### **Final Settlement Zone**



*The recently completed installation at Culbokie, for Scotland Water.*

The biomass passes from the biozone into the final settlement zone where it settles to form humus sludge. This is then regularly pumped out using either an air lift system or submersible pumps and returned to the primary zone.

The clarified liquid decants from the top of the tank as effluent that can be discharged to a reed bed for further clarification or direct to a watercourse.



Top left, filters being baked at 105°C. Right photograph, filters in desiccant.  
Bottom photograph, preparation for the fecal test.



## Emerging Technologies

Many plants that are not specifically configured for BPR nevertheless achieve phosphorus removal to less than 1 mg/L. The first such observation in a nitrifying plant was in a four-stage Bardenpho plant where mixed liquor was recycled from the second anoxic zone to an unstirred fermenter, then returned to the anoxic zone. The CATABOL™ and Cannibal Processes claim to reduce excess secondary sludge production by passing mixed liquor or RAS through an anaerobic (fermenting) stage and then back to the main stream aeration system. In addition, both processes pass the mixed liquor through a process for removal of some of the inert solids. Both processes claim to get similar phosphorus removal to that for the Bardenpho plant described above.

All of these processes rely on the fermentation of some of the mixed liquor for producing VFA that assists in the biological removal of phosphorus. The Town of Cary, NC, has been using a system by which some of the sludge in the return streams of a biological nitrogen removal plant is subjected to anaerobic conditions similar to that of the other processes described above resulting in an effluent phosphorus concentration of less than 0.5mg/L.

There is a similarity between these processes and *ad hoc* processes for switching off aeration in plug-flow plants for promoting phosphorus removal. These *ad hoc* processes take various forms. The Piney Water, CO, plant is a 5-stage Bardenpho plant with no primary sedimentation and little VFA in the influent, which resulted in little phosphorus removal. By switching off a mixer in one of the anaerobic zones, sludge settled to the bottom and fermented, which supplied the VFAs for reducing the orthophosphorus to less than 0.2 mg/L.

A similar operation at the Henderson, NV, plant in a JHB type process had the same effect. Some plug-flow aeration plants succeeded in reducing phosphorus to below 1 mg/L by turning off aeration at the feed end of the plant, such as the Blue Lakes and Seneca plants operated by the Metropolitan Council Environmental Service in Minnesota and the St. Cloud, MN, plant.

The Joppatowne plant operated by Harford County, MD, consists of an MLE plant with some sludge accumulation in the anoxic zone while reducing the phosphorus from 7 mg/L in the influent to around 1 mg/L in the effluent. All of these plants use the same principle of fermenting some of the mixed liquor sludge or underflow from the final clarifiers, either inside the main stream tanks or in a side stream basin. There are many instances where enterprising operators can achieve 80 percent or more phosphorus removal by turning off air or mixers in conventional treatment plants. There is a Catabol plant in Cartersville, GA (USEPA, 2008a); however, there are no published data for this plant.

### Operational and Design Considerations

Important factors that affect BPR include:

- Bioavailable COD:P ratio in the anaerobic zone influent, including adjustments by VFA addition and sludge fermentation
- SRT and HRT
- Presence of oxygen or nitrate in the anaerobic zone
- Backmixing of oxygen
- Temperature
- pH
- Secondary release under anaerobic conditions
- Sufficient oxygen in the aerobic zone
- Inhibition
- Flow and load balancing

## COD:P Ratio

The PAOs need VFAs in the form of acetic and propionic acid. These acids may be in the feed or can be produced through fermentation of soluble rbCOD such as sugar, ethanol, etc., in the anaerobic zone. As a rough estimate of the propensity for phosphorus removal to an effluent concentration less than 1.0 mg/L, the COD:P ratio typically should be about 40 or more. VFA is produced through fermentation of municipal wastewater or it can be added as a commercial or waste product. Some wastewater collection systems that are relatively flat and have long collection times may have sufficient fermentation in the collection system to provide the necessary VFAs, but it will vary monthly depending upon the temperature and flow conditions in the collection system. Force mains are excellent fermenters for the production of VFA. Systems that do not have a COD/P ratio of at least 40 will most likely need to supplement VFAs to achieve effluent phosphorus concentrations below 1.0 mg/L. However, they will still achieve substantial BPR with lower ratios if appropriately operated. See below for a more detailed discussion of VFAs.

Recent studies suggest that the instantaneous COD:P ratio is more important than the overall average (Neethling et al., 2005). Short term drops in the BOD:P ratio in the primary effluent to below that required for sustainable phosphorus removal correlated well with rises in effluent phosphorus. Intermittent recycles of phosphorus rich return streams may cause short term variability in the BOD:P ratio. Controlling or eliminating these recycles can improve plant performance. Weekend changes in the BOD:P ratio also can affect performance.

Another group of organisms, glycogen accumulating organisms (GAOs), also has the ability to take up acetate in the anaerobic zone, not by using energy in phosphate bonds but by using stored glycogen as the energy source. Under certain conditions, such as high temperatures or low phosphorus concentrations relative to the influent bioavailable COD, they may out-compete PAOs for the VFAs, which would result in less or no release of phosphorus in the anaerobic zone. This in turn will result in less or no overall phosphorus removal. GAOs use the stored energy in the form of glycogen to take up VFAs and store them as a complex carbohydrate containing poly-hydroxy valerate (PHV), instead of PHB formed with poly-phosphorus as the energy source. When this begins to happen, there is a slow decline of phosphorus removal by the biological system.

There is still a debate amongst researchers about the conditions likely to favor GAOs over PAOs. Summarizing a number of publications, it would appear that the following conditions favor the growth of GAOs over that of PAOs:

- High SRT
- High temperature over 28 °C
- Longer non-aerated zones
- Stronger wastes with low TKN content
- Periods of intermittent low BOD loads
- If the VFA consists mostly of either acetate or propionate
- Polysaccharides such as glucose are fed to the anaerobic zone.
- Low pH in the aerobic zone

Further confirmation is needed for some of these factors.

### Volatile Fatty Acid Addition

Only VFAs such as acetic and propionic are taken up by PAOs. Reported doses of VFA for successful phosphorus removal range from 3 to 20 mg/L VFA per gram of phosphorus removed. These numbers, however, do not take into account the rbCOD that is fermented in the anaerobic zone. It is more accurate to look at the rbCOD/P ratio for good phosphorus removal, which ranges from 10 to 16. (Barnard, 2006). Surveys show that it is rare for a WWTP treating municipal sewage to achieve more than 95 percent removal of phosphorus by biological processes without adding VFAs (Neethling et al., 2005).

An Australian study shows that while both PAOs and GAOs could use acetate, PAOs will have a competitive advantage when the VFAs consist of roughly equal parts of acetic and propionic acid as a growth medium. PAOs that are fed on acetate are able to switch to propionate much more quickly and effectively than GAOs (Oehmen et al., 2005). This finding led to a strategy to feed equal amounts of acetic acid and propionic acid as the optimal for stimulating PAO growth (Oehmen et al., 2006, Bott et al., 2007). One study shows that isovaleric acid drives BPR even better than acetic acid (Bott et al., 2007).

Isovaleric acid, however, is much more expensive than acetic acid and is more odorous. It also is not significantly generated in the primary sludge fermentation process. Addition of rbCOD such as sugars and alcohols containing two carbons or more can increase phosphorus uptake by PAOs when added to the anaerobic zone but may cause sludge bulking if dosed in excess (Jenkins and Harper, 2003).

### **Sludge Fermentation**

Anaerobic fermentation produces VFA consisting mainly of acetic and propionic acid. Some configurations, such as the Westbank and OWASA configurations, make use of anaerobic fermentation of the primary sludge to provide VFAs to the nutrient removal process. A fermentation process, however, can be added to any configuration to provide VFAs, especially in areas where little fermentation takes place in the collection system. Fermentation of the primary sludge or the RAS will produce VFA. Primary sludge fermentation is used more frequently.

There are several primary sludge fermenter designs that can accomplish this. The simplest configuration involves allowing the formation of a thicker sludge blanket in the primary clarifier itself and returning some of the thickened sludge to either the primary clarifier or to a mixing tank ahead of the primary clarifier to allow elutriation of the VFA to the primary effluent. This is referred to as an activated primary sedimentation tank (Barnard, 1984). Another variation is to pump some sludge to a complete-mix tank ahead of the primary clarifier, to accomplish fermentation. The sludge is then passed to the primary clarifier for elutriation of the VFA. Both of these processes lead to an increased load on the primary clarifier and some VFA may be lost due to aeration between the primary clarifier and the anaerobic zone. Sludge age should also be controlled to prevent methanogenic bacteria from growing and converting the VFA to methane. Usually, a SRT less than 4 days is sufficient for this.

Alternative methods accomplish fermentation in a gravity sludge thickener by holding the sludge under anaerobic conditions for 4 to 8 days. The supernatant can then be fed directly to the anaerobic zone and a high load on the primary clarifier can be avoided. Thickening can either be accomplished with a single thickener or in two stages. The two-stage process can either be a complete mix tank, followed by a thickener or two thickeners in series. It has been shown that adding molasses or other sources of readily biodegradable COD can improve the performance of fermenters (Bott et al., 2007).

RAS can also be fermented in a side stream process. The fermentation zone is similar to the anaerobic or anoxic zone of many biological processes. RAS fermentation could be used in any BPR process, but is most common in processes without primary clarifiers. Research and experience have revealed some key design considerations for primary fermenters (WEF and ASCE, 2006). These processes can have high solids content and may need a positive displacement pump to operate properly. Because fermentation can lower the pH and produce carbon dioxide and hydrogen sulfide, corrosion resistant materials should be used. Odor control may also be necessary if hydrogen sulfide is produced. Monitoring of pH and oxidation reduction potential (ORP) may be desirable to control the process.

## Retention Time

The concentration of phosphorus in the sludge typically increases as the SRT increases, although the impact is very small over the SRT range of 4 to 30 days. Efficient phosphorus uptake typically requires a minimum SRT of 3 to 4 days depending on temperature. Higher SRTs will not increase phosphorus uptake, given there is sufficient VFAs available. If SRT becomes too great, however, effluent quality can degrade. This can be due to release of phosphorus as biomass degrades (WEF and ASCE, 2006). Both anaerobic and aerobic HRT can affect the amount of phosphorus stored by PAOs. Sufficient time should be allowed for the formation of VFAs and storage of the Polyhydroxyalkanoates (PHAs) in the anaerobic zone, although the reactions are relatively fast. If the time is too short, phosphorus uptake in the aerobic zone will be lower than achievable because insufficient PHAs were stored in the anaerobic zone. It has been reported that the ratio of HRT in the anaerobic zone to the HRT in the aerobic zone is important. One study found that a ratio of between 3 and 4 for aerobic HRT to anaerobic HRT led to optimal plant operation (Neethling et al., 2005).

### Temperature

High temperatures can have an adverse effect on phosphorus removal. At temperatures greater than 28° C, phosphorus removal will generally be impaired, apparently by the predominance of the GAOs (Bott et al., 2007). At the low end of the temperature scale, Erdal et al. (2002) found that PAOs outcompeted GAOs at 5° C even though the PAO metabolism was slower at 5° C than at 20° C. The GAOs virtually disappeared in the 5° C reactor. Modeling studies have shown that GAOs can predominate at higher temperatures because of their increased ability to uptake acetate at those temperatures compared to PAOs (Whang et al., 2007). Low temperatures can also lower phosphorus uptake but have been shown to not be an issue in well operated and properly acclimatized plants (WEF and ASCE, 2006).

### Presence of Oxygen or Nitrate in the Aerobic Zone

If oxygen or nitrate is present in the anaerobic zone, organisms that use oxygen or nitrates as electron acceptors will preferentially grow by fully oxidizing the organics to CO<sub>2</sub> and H<sub>2</sub>O, thereby reducing the VFAs available for polymerization and storage by the PAOs. Nitrate can also inhibit fermentation of rbCOD because most of the fermenters are facultative and can use the nitrate as an electron acceptor to fully oxidize the rbCOD instead of producing VFAs as an end product of fermentation, thus depriving the PAOs of organics they can polymerize and store. Therefore, recycle of streams containing high DO and nitrate concentrations to the anaerobic zone should be avoided. Introduction of oxygen through pumps and other devices should also be avoided.

### Avoiding Backmixing of Oxygen

Another potential source of oxygen and nitrates to the anaerobic zone is backmixing from downstream zones. In configurations where the anaerobic zone is followed immediately by an anoxic or aerobic zone, backmixing can cause elevated concentrations of nitrates and/or DO in the anaerobic zone leading to favoring of organisms other than PAOs. The problem can be avoided by increased baffling or changing the mixing rates. This problem is more likely to occur when the downstream zone is aerated, because aeration of mixed liquor increases the liquid depth, making the liquid level in the aerobic zone higher than in the non-aerated zone.

### pH

Low pH can reduce and even prevent BPR. Below pH 6.9 the process has been shown to decline in efficiency (WEF and ASCE, 2006). This is possibly due to competition with GAOs. Filipe, et al. (2001), found that GAOs grow faster than PAOs at a pH of less than 7.25. Because many wastewater processes such as chemical addition and nitrification can lower pH, this should be monitored and adjusted if necessary. It also has been shown that it is not possible to establish enhanced biological phosphorus removal (EBPR) when the pH is less than 5.5, even though an abundant amount of acetic acid is present in the anaerobic zone (Tracy and Flammino, 1987; Randall and Chapin, 1997).

## Anaerobic Release

Secondary release of phosphorus occurs when the PAOs are under anaerobic conditions in the absence of a source of VFA. The energy stored as polyphosphate is used for cell maintenance and phosphorus is released to the liquid phase (Barnard, 1984). There will then be no stored food to supply energy for the uptake of phosphorus upon subsequent aeration.

### **This may occur in the following process stages:**

- In the anaerobic zone if the retention time is too high and the VFA is depleted well within the required retention time.
- In the main anoxic zone when that runs out of nitrates.
- In the second anoxic zone there are no nitrates to be removed.
- In the sludge blankets of final clarifiers when the RAS rate is too low and sludge is not removed fast enough.

Additionally, release may happen in aerobic zones that are too large, resulting in stored substrate depletion and destruction of PAO cells by endogenous metabolism. Since there was no food storage associated with the phosphorus release, additional carbon is then required to take up the phosphorus released, but the amount in the influent may be insufficient.

Therefore, chemicals must be added to remove the excess phosphorus. Over-design of biological nutrient removal systems could thus lead to a higher demand for an external source of VFA. Phosphorus will be released in sludge treatment processes that are anaerobic. Gravity thickening of BPR sludge can lead to phosphorus release if long retention times are used. Using mechanical dewatering instead of gravity dewatering allows less retention time and less phosphorus release (Bott et al., 2007). It is usually recommended that dissolved air flotation (DAF) be used to thicken BPR sludge to reduce the amount of phosphorus release. DAF thickening can be quite successful for the reduction of release, but if the thickened sludge is left on the DAF beach too long before removal, excessive release will occur, just as it will when the sludge is left too long in a gravity thickener.

Anaerobic digestion will also lead to phosphorus release although some phosphorus will be precipitated as either a metal salt (e.g. calcium phosphate) or as struvite (magnesium ammonium phosphate,  $MgNH_4PO_4$ ). BPR sludge takes up and releases magnesium along with phosphates, and these two ions combine with ammonium, also present in abundance in anaerobic digesters, to form struvite.

Struvite formation is very fast, and will continue until one of the three ions is reduced to that ion's solubility level. Magnesium is usually present in the lowest concentration, and its depletion typically limits struvite formation within the anaerobic digester. Calcium phosphate precipitates also tend to form in anaerobic digesters, but they form much more slowly than struvite and the formation tends to be non-stoichiometric. If substantial amounts of phosphates are precipitated by calcium along with the struvite formation, there will be little if any propensity for struvite to form when the sludge exits the anaerobic digesters. Also, if the digested sludge is composted after dewatering, the resulting Class A sludge will be enriched in magnesium, phosphorus, nitrogen, and, to a lesser extent, potassium, which also is taken up and released with phosphorus by PAOs. Thirty percent of the phosphorus entering the anaerobic digesters at the York River plant during BPR experimentation was recycled back to the headworks from belt filter press dewatering (Randall et al., 1992).

Alternatives to anaerobic digestion such as composting, drying, or alkaline treatment can be used to reduce phosphorus release. There have been several studies which have examined using struvite precipitation as a way of recovering phosphorus from supernatant from digesters.

When anaerobic release of phosphorus occurs, recycling these streams can overload phosphorus removal processes. The effect can be worsened when the waste handling process is only operated intermittently. In some instances there is a high degree of phosphorus precipitation in the anaerobic digesters and with sufficient VFA in the influent the returned phosphorus may be removed. However, in most circumstances, some chemicals need to be added to the return streams or to the anaerobic digester itself so that the metal precipitate will be removed with the dewatered sludge.

### **Sufficient Oxygen in the Aerobic Zone**

It is necessary for oxygen to be present in the aerobic zone for phosphorus to be taken up and retained in the activated sludge. Maintaining a sufficiently high DO transfer in the aerobic zone enhances process stability and has been found to be a key factor in phosphorus removal. (Bott et al., 2007)

### **Inhibition**

EBPR, like any biological process, can be inhibited by chemicals toxic to the organisms. Although not as sensitive to inhibition as nitrification and rare in practice, the BPR process can be inhibited by toxic chemicals, including high concentrations of acetate (Randall and Chapin, 1997).

### **Flow and Load Balancing**

Flows and loads to wastewater treatment plants can vary widely because of regular diurnal use patterns and because of larger, more irregular disturbances such as storm events. Peaks in either flow, or nutrient load can stress the system and cause poor performance. Peaks can be evened out using equalization tanks to balance the flow. Equalization tanks in combination with nutrient sensors can also be used to balance nutrient loads. In this case, recycle streams high in nutrient concentrations such as digester supernatant can be stored during peak nutrient loads and recycled during times when concentrations are lower.

### **Impacts on Sludge Handling and Removal**

Stored phosphorus adds dry weight to the sludge; however, the actual PAO VSS production will be less because the reaction is less efficient than heterotrophic metabolism using DO as the electron acceptor. Sludge from BPR will be similar to sludge from conventional activated sludge plants, although it will have a higher phosphorus content. Varying results have been found with some plants reporting little or no change in settling and dewatering (Knocke et al., 1992) and others reporting enhanced settling and dewatering properties (Bott et al., 2007). The sludge produced from the process will also have higher magnesium and potassium concentrations due to co-uptake of these elements with phosphorus.

Struvite can precipitate in anaerobic processes. With abundant phosphorus and ammonia it is usually only the magnesium that is in short supply. Some magnesium is released from the digested cells with the phosphorus and may increase struvite precipitation. Some processes have proposed precipitating out struvite or other phosphate solids to avoid phosphorus return in recycle streams (Bott et al., 2007). The struvite crystals, however, depending upon where they form, can plug centrifuge ports, and pumps and pipes used to convey the sludge, if not controlled. Plugged lines are very difficult to clean.

### **Guidance for Selecting Process Modifications**

If an existing activated sludge WWTP needs to lower phosphorus levels in its effluent, a number of options are available. Some key considerations are summarized below. For systems that do not have BPR, an anaerobic zone can be added at the head of the plant. This may be achieved by switching off aerators at the head of the reactor or by adding a separate reactor. Mixing in the anaerobic zone should be sufficient to retain biological solids in suspension without introducing oxygen. If baffling is not already present, it could be added to achieve separation of the anaerobic and aerobic zones.

Note that baffling is essential to prevent backmixing because the liquid level in the aerated zone will always be higher than that in the non-aerated zone. Therefore, an overflow baffle should be used between zones. Considerations should also be made for additional pumping needed for any recycle streams. Proper sizing of the anaerobic zone is important to ensure sufficient VFA is formed and taken up in the aeration basin. If an aerobic zone is converted to an anaerobic zone, care should be taken to ensure that the remaining aerobic zone is sufficiently sized to achieve treatment objectives. This usually is not a problem because the anaerobic zone seldom needs to be more than 15 percent of the total volume, and can be considerably less if fermentation is practiced or VFA are added. Note that much of the BOD in typical municipal sewage will be removed from solution in the anaerobic zone, and this reduces the required size of the aerobic zone, even though most of the stored BOD will be stabilized in either the anoxic or aerobic zone, or both.

For plants that already have BPR but need additional phosphorus removal, the designers should start by identifying areas that may be limiting the current process. For example, if recycle streams are intermittent, overloading of the process may occur during recycle and the process performance may suffer. Flow equalization to enable constant recycle flows may be an option in these cases. RAS when returned to the anaerobic zone may introduce nitrates or oxygen that will interfere with PAO performance. The phosphorus content of the return streams could be reduced by adding some chemicals to precipitate some of the phosphorus. Reducing oxygen introduction to the anaerobic zone from upstream processes may be needed to optimize phosphorus removal.

Plants looking to improve phosphorus removal performance should also closely examine the plant for secondary release of phosphorus. If sludge blankets in clarifiers are too deep, anaerobic conditions can develop and cause secondary phosphorus release. This can be minimized by using deeper clarifiers, maintaining low sludge blankets, and increasing the RAS rate, so that the released phosphorus is pumped from the bottom of the clarifier rather than flowing over the effluent weir. Sludge handling can also cause excessive phosphorus release such as in gravity thickeners, DAFs and anaerobic digesters. If supernatant from these processes when poorly managed is recycled, it can overload the process. Options in this case would be to eliminate the recycle, improve operation of the process, change the process, or treat the recycle stream to remove phosphorus before it is returned to the plant.

Another area to examine in seeking improved phosphorus removal is the COD:P ratio. If the ratio is low, supplementing the current process with VFAs may provide additional removal. VFAs can either be added as a chemical addition process or produced through fermentation of primary or secondary sludge.

Other ways of improving TP removal include filtration and chemical addition. Phosphorus is often attached to colloidal particles and very low phosphorus levels usually require removal of TSS. Membrane bioreactors (MBR) in combination with biological and/or chemical phosphorus removal can result in very low effluent levels due to enhanced solids removal. Chemical addition with or without filtration can also achieve low phosphorus levels.

### **Effluent Filtration**

Effluent filtration in combination with chemical precipitation can be used to remove phosphorous down to very low levels (< 0.1 mg/L). USEPA Region 10 (2007) found that 2-stage filtration through use of a first and second stage filter or by providing tertiary clarification prior to filtration, resulted in the lowest effluent phosphorus concentrations of 23 WWTPs evaluated. Effluent filtration can also be used to remove soluble organic nitrogen that is not removed through biological treatment or settling.

**A wide variety of filter types have been used for wastewater treatment, including:**

- Conventional down-flow filters
- Deep-bed down-flow filters
- Continuous backwashing upflow sand filters
- Pulsed bed filters
- Traveling bridge filters
- Fuzzy filters
- Discfilter
- Cloth media disk filters
- Membranes
- Blue PROTm process
- Pressure filters



NO<sup>2</sup>/NO<sup>3</sup>, Fluoride, Sulfide, Metals, BOD-TDS-TSS  
Wide-mouth Sludge/Metals bottle.

## Types of Filters

This section describes the various filters listed on above page, presents key design and operating principles, and summarizes ongoing research and emerging technologies in this area.

### Conventional Down-flow Filters

These filters consist of fixed-media beds typically up to 3 feet in depth and are similar to filters used to treat drinking water. Media can be single media, dual media, or multi media. Single media is typically sand or anthracite. Dual media combines anthracite and sand. Multi-media filters include a layer of garnet or limonite. Flow in these filters is by gravity from the top down. Most of the removal occurs in the top few inches of the media. The filter must be taken off-line periodically to backwash the filter to prevent clogging and too high of a pressure loss.

### Deep-bed Down-flow Filters

These filters are similar to conventional down-flow filters but have deeper beds and larger media size. This gives the advantage of longer run times between backwashes. The size of the media is limited by the ability to backwash the filter. Because these filters are more difficult to backwash, air scour is necessary to fully clean the filter bed.

### Continuous Backwashing Upflow Sand Filters

During operation of the continuous backwashing upflow filter, water is introduced through risers at the bottom of a deep sand bed. Water flows upward through the sand bed and over an overflow weir. Sand and trapped solids flow downward through the filter and are drawn into the suction of an airlift pipe in the center of the filter. As the sand travels up the airlift pipe, energy from the air scours the particles and separates the sand from filtered solids. At the top of the airlift pipe, the clean sand settles back onto the top of the filter and the solids are carried away into a reject line. These filters have the advantage of having no moving parts other than the air compressor and requiring less energy and maintenance than traditionally backwashed filters. They are sometimes referred to by the trade name Dynasand.

### Pulsed Bed Filters

Pulsed bed filters are shallow filters with an unstratified fine sand media. An air pulse disturbs the media and allows penetration of solids into media bed, allowing the entire filter bed to be used for removal of solids. The pulse is designed to expand the filter operation and reduce the number of backwash cycles, although the filter must still be periodically backwashed to remove the solids.

### Traveling-Bridge Filters

Traveling-bridge filters consist of long shallow beds of granular media. Wastewater is applied to the top of the media and flows downward. Each cell is individually backwashed by a traveling-bridge while the other cells continue to operate. The bridge uses filtered water to backwash the filters and includes surface wash to breakup matted solids or clumps of solids.

### Fuzzy Filters

The fuzzy filter uses a proprietary synthetic filter media that is highly porous. Water flows not only around the media but also through it, allowing much higher filtration rates. The media is held in place by a metal plate and flow is from the bottom of the bed upwards. The filter is backwashed by raising the plate and introducing a horizontal air stream from alternating sides causing the media to roll back and forth. The effluent is returned to the plant.

### Discfilters

Discfilters are a series of parallel mounted disks used to support a cloth filter media. Water enters a central tube and flows out between the two layers of cloth in each disk. The disks rotate and are normally 60 to 70 percent submerged. The portion above the water is backwashed using spray nozzles.

### **Cloth Media Disk Filters**

The cloth media disk filter is similar to the discfilter listed above. In this case the water flows from the outside of the partially submerged cloth disks and into a center pipe. Disks continue to rotate during backwash and water is sucked into the disc using suction heads.

### **Membranes**

Membrane systems use a pressure head to drive water through a permeable membrane. Membrane filters are typically classified by their pore size which in turn determines the size of the particles they exclude. Microfiltration, ultrafiltration, nanofiltration, and reverse osmosis (RO) remove increasingly smaller particles. Microfiltration and ultrafiltration remove 3 to 6 logs of bacteria, 95 percent or more BOD, along with most particles (WEF, 2006). Nanofiltration removes nearly all particles including some viruses. RO removes all particles as well as most large dissolved constituents. The energy cost for applying the pressure head and the need to replace membranes make membrane filtration a more expensive technology. It can achieve very low concentrations of nutrients and other contaminants, however, and is common in water re-use projects.

Membranes can be configured a number of ways including hollow fiber, spiral wound, plate and frame, cartridge, or in pressure vessels. Membranes can foul from organics, biological activity, or metals in the wastewater. Typically the water must be pre-treated before using these membranes. Pretreatment could be conventional filters, cartridge filters, or larger membrane filters. Disinfection may also be required to prevent biological fouling.

### **Blue PROTM Process**

The Blue PROTM process uses a continuous backwashing filter that is designed remove phosphorus. Filters can be run in series for even greater removal. The filter media (sand) is coated with a hydrous ferric oxide coating, which enhances phosphorus removal through adsorption. A ferric salt is added prior to the filter to aid in coagulation and to replace the ferric coating which is abraded from the sand. Water flows up through the filter while the sand travels down. An airlift tube at the bottom of the filter carries the sand upward. Turbulence from the compressed air knocks accumulated iron and phosphorus along with any solids off the particle as it travels upward. The iron, phosphorus, and particles are wasted, while the clean sand is deposited on the top of the bed. The filters can be run biologically active to achieve denitrification.

The Blu-CAT process combines the Blu-Pro process with addition of advanced oxidants. Early pilot tests show that this process is capable of removing other emerging contaminants along with phosphorus and microorganisms (USEPA, 2008a).

### **Pressure Filters**

Pressure filters are similar to conventional media filters except they are contained in closed containers and are filtered under pressure. The increased pressure creates a greater head loss and allows longer times between backwashes.

### **Design and Operating Principles**

Filtration is mainly affected by the concentration and size distribution of particles entering the filter. Turbidity is often used as a surrogate for particle concentration. The concentration of particles will affect run-time in filters and will also affect the required surface area to achieve the desired filtration. The size distribution of the particles and its relevance to pore size of the granular or membrane filters will affect the removal mechanisms. Filtration rate is also an important design parameter. Too fast of a filtration rate can cause floc to break up and pass through the filter. The optimal filtration rate depends on floc strength, which in turn depends on the biological treatment processes prior to filtration (e.g., Higher SRTs lead to weaker flocs). The filtration rate, along with the loading rate will determine the area of the filter required. The higher the loading rate, the more frequent backwashes will be required and the greater the head loss across the filters. Typical filtration rates are 5 to 15 meters per hour for gravity filters and up to 20 meters per hour for pressure filters (WEF and ASCE, 1998).

Addition of polymers or other coagulant aids can greatly aid filtration. Typical doses for filter influent are 0.05 to 0.15 mg/L of organic polyelectrolyte (WEF and ASCE, 1998), although jar tests are conducted to determine the proper dose. Too low a dose can allow uncoagulated particles through the filter and too high a dose can lead to mudballs and filter clogging.

There are several ways the flow rate can be controlled in filters. Constant-rate fixed head filtration maintains a constant flow through the filter. This will lead to an increased head above the filter as the filter run progresses. In constant-rate variable head filtration the rate is kept the same and the filter is backwashed when the head reaches a certain value. In variable-rate filtration, the rate of filtration decreases throughout the filter run until it reaches a minimum value and is backwashed. Variable-rate filtration is less common than constant-rate filtration.

Proper backwashing is also important to filter operation. Without proper backwashing there can be breakthrough of particles and turbidity. Lack of a proper backwash can also lead to accumulation of materials on the surface of the filter that can form mudballs and cracks, which can allow solids to pass through the filter. A surface wash or air scour may also be helpful to prevent accumulation of mudballs or grease. Surface wash or air scour is also helpful for traveling bridge filters. Without surface wash traveling bridge filters are limited to an influent TSS concentration of 40 to 50 mg/L (WEF and ASCE, 1998).

If membrane filters are used, fouling can be an important consideration. Cellulose acetate membranes can be damaged by biological activity. Disinfection is often used to prevent biological fouling of the membranes. Some membrane materials such as polyacramides, however, can be damaged by chlorine. This can be avoided by using an alternative disinfectant, a different membrane material, or by de-chlorination. Lowering the pH can help to prevent mineral fouling of nanofiltration or reverse osmosis membranes. Besides pre-treatment, chemical cleaning of the membranes may also be required periodically. Monitoring of effluent quality and pressure differential can be important to help identify membrane fouling or failure.

### **Ongoing Research and Emerging Technologies**

The use of membranes as tertiary filtration is an area that has recently expanded. Research continues on various membrane configurations along with topics such as pre-treatment, membrane cleaning, and removal of emerging contaminants. Fuzzy filters are also an innovative technology that is beginning to be established in the wastewater community with several full scale projects.

Other research has focused on enhancements to existing technology. For example, the Blue-Pro system combines continuous backwashing filters, a well-known technology, with a hydrous ferric oxide coating and ferric salt addition to remove phosphorus by adsorption as well as filtration.

### **Mathematical Modeling**

#### **8.1 The Need for Models**

WWTPs are complex systems that depend on numerous biological, chemical, and physical processes to achieve effluent goals. Because of the complex behavior of the processes and the variability in wastewater characteristics, biological populations, and plant design, it is not always possible to predict how changing any one variable will affect the effluent quality.

Plant designs that work for one influent wastewater and climate may not perform well in different conditions. Pilot scale or full scale trials can help to determine the effect of various parameters, but costs and time to cover all possibilities may be prohibitive. Therefore, models fill an important need by enabling simulation of a process and estimating the impact that changing parameters will have on the treatment effectiveness.

Models can be used for a number of purposes including the design of new WWTPs, the design of retrofits or upgrades to existing plants, determining how changes in operations may affect effluent concentrations of permitted contaminants, determining how plants will respond to changes in influent quality or flow, and for training operators. Not all models can achieve all of these purposes, so models should be selected with the desired use in mind. There is some disagreement in the literature in the use of the term model.

Some references use the term to refer to sets of mathematical equations that characterize a process, other references use model to refer to the computer program used to solve these equations. This section will use the former and will use the term “simulator” to describe the computer program.

### **Overview of Available Models**

Models are sets of equations, generally based on theory and grounded in empirical data that represent a wastewater treatment process. Each unit process is represented by its own model.

Model equations for processes such as clarification and settling are well known and fairly simple. Modeling biological wastewater processes such as activated sludge, however, is much more complicated. The primary set of models for activated sludge processes has been compiled by the International Water Association (IWA). The first model was developed in 1986 and was called the activated sludge model (ASM). Later known as ASM1, this model was able to model the biological oxidation of carbon, nitrification, and de-nitrification.

Although the ASM model gained widespread use among both academia and industry, it had limitations. For example the model assumed constant temperature and pH, did not include EBPR, and the biological reactions did not depend on the carbon source.

In order to improve the model, IWA developed four other ASM models; ASM2, ASM2d, ASM3, and ASM3 with BioP. ASM2 and ASM2d were intended to add EBPR. The ASM3 models were intended to deal with limitations such as the independence of the ASM1 model of temperature and carbon source. In addition, other models were developed to seek to improve upon the ASM model.

The metabolic biological phosphorus model of the Delft University of Technology (TUDP) was developed to fully account for the metabolism occurring in PAOs during EBPR. Barker and Dold (1997) developed a model (B&D) to include different rates of growth depending on the carbon source.

When selecting models, the processes required and the range of normal operating parameters for the plant should be considered and compared to the available models. For example, if chemical phosphorus removal is to be used in a plant, the plant is limited to using either the ASM2 or ASM2d models. Each model also has a range of temperatures and pH over which it is valid.

## Wastewater Sampling Information

### Required Containers, Preservation Techniques, and Holding Times

40 CFR 136.3

Parameter No./name	Container	Preservation	Maximum holding time
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#### Table IA--Bacteria Tests:

1-4 Coliform, fecal and total. P, G..... Cool, 4C, 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$ ..... 6 hours.  
5 Fecal streptococci..... P, G..... Cool, 4C, 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$  ..... 6 hours.

#### Table IA--Aquatic Toxicity Tests:

6-10 Toxicity, acute and chronic. P, G..... Cool, 4C ..... 36 hours.

#### Table IB--Inorganic Tests:

1. Acidity..... P, G..... Cool, 4C..... 14 days.
2. Alkalinity..... P, G..... Cool, 4C..... 14 days.
4. Ammonia..... P, G..... Cool, 4C,  $\text{H}_2\text{SO}_4$  to  $\text{pH} < 2$ ..... 28 days.
9. Biochemical oxygen demand.. P, G..... Cool, 4C..... 48 hours.
10. Boron..... P, PFTE, or  $\text{HNO}_3$  TO pH2..... 6 months. Quartz.
11. Bromide..... P, G..... None required..... 28 days.
14. Biochemical oxygen demand, P, G..... Cool, 4C..... 48 hours. carbonaceous.
15. Chemical oxygen demand.... P, G..... Cool, 4C,  $\text{H}_2\text{SO}_4$  to  $\text{pH} < 2$ ..... 28 days.
16. Chloride..... P, G..... None required..... 28 days.
17. Chlorine, total residual.. P, G..... None required ..... Analyze immediately.
21. Color..... P, G..... Cool, 4C..... 48 hours.
- 23-24. Cyanide, total and amenable to chlorination. P, G..... Cool, 4 deg.C,  $\text{NaOH}$  to  $\text{pH} > 12$ , 0.6g Ascorbic acid 14 days.
25. Fluoride..... P..... None required..... 28 days.
27. Hardness..... P, G.....  $\text{HNO}_3$  to  $\text{pH} < 2$ ,  $\text{H}_2\text{SO}_4$  to  $\text{pH} < 2$ ..... 6 months.
28. Hydrogen ion (pH)..... P, G..... None required..... Analyze immediately.
- 31, 43. Kjeldahl and organic nitrogen. P, G..... Cool, 4 deg.C,  $\text{H}_2\text{SO}_4$  to  $\text{pH} < 2$ ..... 28 days.

#### Metals:

18. Chromium VI..... P, G..... Cool, 4C..... 24 hours.
35. Mercury..... P, G.....  $\text{HNO}_3$  to  $\text{pH} < 2$ ..... 28 days.
- 3, 5-8, 12, 13, 19, 20, 22, 3, 5-8, 12, 13, 19, 20, 22, P, G..... do..... 6 months.
- 26, 29, 30, 32-34, 36, 37, 45, 47, 51, 52, 58-60, 62,63, 70-72, 74, 75. Metals, except boron, chromium VI and mercury.
38. Nitrate..... P, G..... Cool, 4 deg.C..... 48 hours.
39. Nitrate-nitrite..... P, G..... Cool, 4 deg.C,  $\text{H}_2\text{SO}_4$  to  $\text{pH} < 2$ ..... 28 days.
40. Nitrite..... P, G..... Cool, 4 deg.C..... 48 hours.
41. Oil and grease..... G..... Cool to 4 deg.C,  $\text{HCl}$  or  $\text{H}_2\text{SO}_4$  to  $\text{pH} < 2$  to 28 days.
42. Organic Carbon..... P, G..... Cool to 4 deg.C  $\text{HCl}$  or  $\text{H}_2\text{SO}_4$  to  $\text{pH} < 2$  or 28 days.
44. Orthophosphate..... P, G..... Filter immediately, Cool, 4 deg.C. ..... 48 hours.
46. Oxygen, Dissolved Probe... G Bottle and top. None required..... Analyze immediately.
47. Winkler..... G Bottle and top. Fix on site and store in dark... 8 hours.
48. Phenols..... G only..... Cool, 4 deg.C,....  $\text{H}_2\text{SO}_4$  to  $\text{pH} < 2$  28 days.
49. Phosphorus (elemental).... G..... Cool, 4 deg.C..... 48 hours.
50. Phosphorus, total..... P, G..... Cool, 4 deg.C,  $\text{H}_2\text{SO}_4$  to  $\text{pH} < 2$ ..... 28 days.
53. Residue, total..... P, G..... Cool, 4 deg.C..... 7 days.
54. Residue, Filterable..... P, G..... Cool, 4 deg.C..... 7 days.
55. Residue, Nonfilterable P, G..... Cool, 4 deg.C..... 7 days.

56. Residue, Settleable(TSS)..... P, G..... Cool, 4 deg. C..... 48 hours.  
 57. Residue, volatile..... P, G..... Cool, 4 deg. C..... 7 days.  
 61. Silica..... P, PFTE, or Quartz..... Cool, 4 deg. C..... 28 days.  
 64. Specific conductance..... P, G..... Cool, 4 deg. C..... 28 days.  
 65. Sulfate..... P, G..... Cool, 4 deg. C..... 28 days.  
 66. Sulfide.... P, G..... Cool, 4 deg. C add zinc acetate plus sodium hydroxide to pH>9. 7 days.  
 67. Sulfite..... P, G..... None required..... Analyze immediately.  
 68. Surfactants..... P, G..... Cool, 4 deg. C..... 48 hours.  
 69. Temperature..... P, G..... None required..... Analyze.  
 73. Turbidity..... P, G..... Cool, 4 deg. C..... 48 hours.

**Table IC--Organic Tests**

13, 18-20, 22, 24-28, 34-37, G, Teflon-lined septum Cool, 4 deg. C, 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$  14 days.  
 39-43, 45-47, 56, 76, 104, 105, 108-111, 113.  
 Purgeable Halocarbons. 6, 57, 106.  
 Purgeable aromatic hydrocarbons G, Teflon-lined septum Cool, 4 deg.C, 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$  14 days.  
 3, 4. Acrolein and acrylonitrile G, Teflon-lined septum Cool, 4 deg.C, 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$  pH 4-5 14 days.  
 23, 30, 44, 49, 53, 77, 80, 81, 98, 100, 112. G, Teflon-lined Cool, 4 deg.C, 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$  14 days.  
 Phenols G, Teflon-lined septum Cool, 4 deg.C, 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$  pH 4-5 7 days until extraction; 40 days after extraction.  
 7, 38. Benzidines G, Teflon-lined septum..... Cool, 4 deg.C, 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$  7 days until extraction.  
 14, 17, 48, 50-52. Phthalate G, Teflon-lined septum Cool, 4 deg.C..... 7 days until extraction; esters 40 days after extraction.  
 82-84. Nitrosamines G, Teflon-lined septum .....Cool, 4 deg. C, 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$ ...Store in dark  
 88-94. PCBs G, Teflon-lined septum Cool, 4 deg.C ..... 7 days until extraction; 40 days after extraction.  
 54, 55, 75, 79. Nitroaromatics ..... G, Teflon-lined septum.....Cool, 4 deg.C, 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$  and isophorone  
 1, 2, 5, 8-12, 32, 33, 58, 59, 74, 78, 99, 101. Polynuclear aromatic hydrocarbons. Cool, 4 deg.C, 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$ ....Store in dark  
 15, 16, 21, 31, 87. Haloethers ..... G, Teflon-lined septum..... Cool, 4 deg.C, 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$  7 days until extraction; 40 days after extraction.  
 29, 35-37, 63-65, 73, 107. Chlorinated hydrocarbons G, Teflon-lined septum.....Cool, 4 deg.C, 7 days until extraction; 40 days after extraction.  
 60-62, 66-72, 85, 86, 95-97, 102, 103. CDDs/CDFs aqueous: field and lab G..... Cool, 0-4 deg.C, pH9, 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$  1 year preservation.  
 Solids, mixed phase, and .....do..... Cool, 4 deg.C..... 7 days. Tissue: field preservation.  
 Solids, mixed phase, and .....do..... Freeze, -10 deg.C..... 1 year. Tissue: lab preservation.

**Table ID--Pesticides Tests:**

1-70. Pesticides \11\..... .....do..... Cool, 4 deg.C, pH 5-9 ..... Do.

**Table IE--Radiological Tests:**

1-5. Alpha, beta and radium... P, G.....  $\text{HNO}_3$  to pH2..... 6 months.

Polyethylene (P) or glass (G). For microbiology, plastic sample containers must be made of sterilizable materials (polypropylene or other autoclavable plastic).

Sample preservation should be performed immediately upon sample collection. For composite chemical samples each aliquot should be preserved at the time of collection. When use of an automated sampler makes it impossible to preserve each aliquot, then chemical samples may be preserved by maintaining at 4 degrees C until compositing and sample splitting is completed.

When any sample is to be shipped by common carrier or sent through the United States Mails, it must comply with the Department of Transportation Hazardous Materials Regulations (49 CFR part 172). The person offering such material for transportation is responsible for ensuring such compliance. For the preservation requirements of Table II, the Office of Hazardous Materials, Materials Transportation Bureau, Department of Transportation has determined that the Hazardous Materials Regulations do not apply to the following materials: Hydrochloric acid (HCl) in water solutions at concentrations of 0.04% by weight or less (pH about 1.96 or greater); Nitric acid (HNO<sub>3</sub>) in water solutions at concentrations of 0.15% by weight or less (pH about 1.62 or greater); Sulfuric acid (H<sub>2</sub>SO<sub>4</sub>) in water solutions at concentrations of 0.35% by weight or less (pH about 1.15 or greater); and Sodium hydroxide (NaOH) in water solutions at concentrations of 0.080% by weight or less (pH about 12.30 or less).

Samples should be analyzed as soon as possible after collection. The times listed are the maximum times that samples may be held before analysis and still be considered valid. Samples may be held for longer periods only if the permittee, or monitoring laboratory, has data on file to show that for the specific types of samples under study, the analytes are stable for the longer time, and has received a variance from the Regional Administrator under Sec. 136.3(e). Some samples may not be stable for the maximum time period given in the table. A permittee, or monitoring laboratory, is obligated to hold the sample for a shorter time if knowledge exists to show that this is necessary to maintain sample stability. See Sec. 136.3(e) for details. The term "analyze immediately" usually means within 15 minutes or less of sample collection. Should only be used in the presence of residual chlorine.

Maximum holding time is 24 hours when sulfide is present. Optionally all samples may be tested with lead acetate paper before pH adjustments in order to determine if sulfide is present. If sulfide is present, it can be removed by the addition of cadmium nitrate powder until a negative spot test is obtained. The sample is filtered and then NaOH is added to pH 12.

Samples should be filtered immediately on-site before adding preservative for dissolved metals.

Guidance applies to samples to be analyzed by GC, LC, or GC/MS for specific compounds.

Sample receiving no pH adjustment must be analyzed within seven days of sampling.

The pH adjustment is not required if acrolein will not be measured. Samples for acrolein receiving no pH adjustment must be analyzed within 3 days of sampling.

When the extractable analytes of concern fall within a single chemical category, the specified preservative and maximum holding times should be observed for optimum safeguard of sample integrity. When the analytes of concern fall within two or more chemical categories, the sample may be preserved by cooling to 4 deg. C, reducing residual chlorine with 0.008% sodium thiosulfate, storing in the dark, and adjusting the pH to 6-9; Samples preserved in this manner may be held for seven days before extraction and for forty days after extraction. Exceptions to this optional preservation and holding time procedure are noted in footnote 5 (re the requirement for thiosulfate reduction of residual chlorine), and footnotes 12, 13 (re the analysis of benzidine).

If 1,2-diphenylhydrazine is likely to be present, adjust the pH of the sample to 4.0<plus-minus>0.2 to prevent rearrangement to benzidine.

Extracts may be stored up to 7 days before analysis if storage is conducted under an inert (oxidant-free) atmosphere.

For the analysis of diphenylnitrosamine, add 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$  and adjust pH to 7-10 with NaOH within 24 hours of sampling.

The pH adjustment may be performed upon receipt at the laboratory and may be omitted if the samples are extracted within 72 hours of collection. For the analysis of aldrin, add 0.008%  $\text{Na}_2\text{S}_2\text{O}_3$ .

Sufficient ice should be placed with the samples in the shipping container to ensure that ice is still present when the samples arrive at the laboratory. However, even if ice is present when the samples arrive, it is necessary to immediately measure the temperature of the samples and confirm that the 4°C temperature maximum has not been exceeded.

In the isolated cases where it can be documented that this holding temperature cannot be met, the permittee can be given the option of on-site testing or can request a variance. The request for a variance should include supportive data which show that the toxicity of the effluent samples is not reduced because of the increased holding temperature.

Common Wastewater Sample Collection Bottles



Above, 625/608, 1657, TTO/Organics, TPH/Oil/Grease  
Smaller bottles-TOCs, VOCs, 601/602 and 502.2.

## Wastewater/Pretreatment Sampling General Information

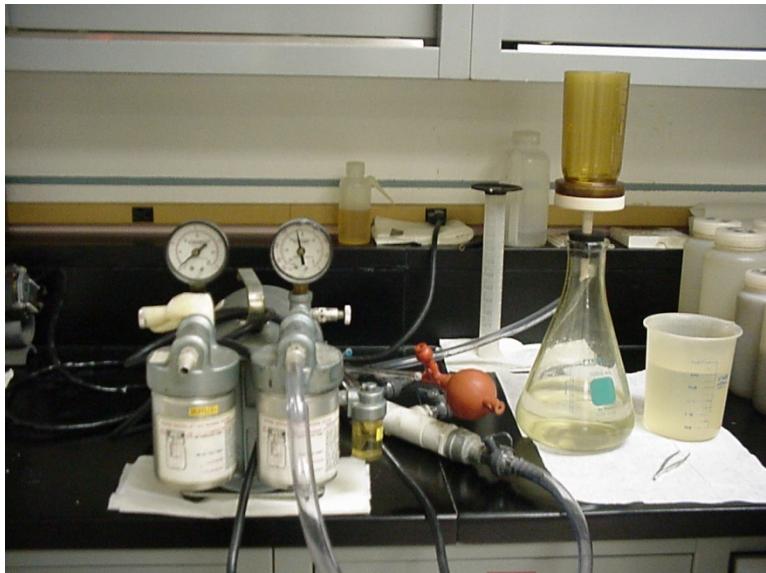
In accordance with the Clean Water Act and General Pretreatment Program Regulations, the POTW conducts a variety of sampling activities which must be closely coordinated.

Each of these activities is briefly described below.

### Permit Application Policy Example

All industrial users that require a permit must be sampled to determine the characteristics of the wastes to be discharged into the POTW's sewer system. Prior to the issuance of a permit for existing industrial users, the POTW samples the user's effluent, and performs the analyses required by the applicable discharge standards (i.e., Categorical standards or local limits).

For new industrial users, estimates of the wastes to be discharged into the POTW's sewer system must be submitted along with the permit application. No sampling would be performed at these new facilities, since they do not presently discharge wastes into the sewer system. A four-day sampling program is usually conducted at each site to collect both composite and grab (for pollutants not amenable to composite sampling) samples as needed.



### Sewer System Evaluation Policy Example

On a regular basis, selected locations in the sewer system are sampled to develop background data for purposes of updating the local limits, and to screen areas for higher than "**background**" pollutant levels. In addition, problem areas are sampled on an as needed basis to determine potential sources of POTW Code violations that either occur on a frequent basis, or are the result of a slug load to the sewer system. To monitor sewers for background information, the sampling program would typically be conducted over a four-day period. In instances where the intent is to determine sources of pollutants and/or slug loads, the length of the program would vary.

### Multi-City Users (Metering Stations) Policy Example

All wastewater, which is transported to the POTW Treatment Plant from the Multi-City users, is analyzed for pollutants of concern to the Industrial Pretreatment Program. The sampling program is conducted over a five-day period to obtain four days of sampling data at each sewer location (i.e., a metering station) on a quarterly basis. Once the sampling dates have been determined, the Water Quality Inspector will notify, in writing, the Sub-regional Organizational Group (**SROG**) or equivalent representative for that City of the dates when the sampling will be conducted.

### **Sampling Safety Policy Example**

Upon arrival at the site, safety is the priority. A visual inspection must be completed prior to any entry. The site must be free of any obstructions or hazards which may cause injury when entering the sampling area. If there are any problems detected, the SROG or equivalent representative and the Water Quality Inspector should be notified, and no entry should be attempted until the problem has been corrected.

### **Metering and Sampling Stations Qualify As Confined Spaces**

If all safety criteria have been met, prepare equipment for the site. Check the assignment sheet to determine what parameters are required to be sampled, which in turn determines the type of tubing to be used, (i.e. Tygon or Teflon).

The sampler must be completely assembled before performing QA/QC procedures. After QA/QC is complete, a sufficient amount of weight must be attached to the tubing to keep the strainer submerged in the effluent for proper siphoning of the sample, without allowing the strainer to hit the bottom of the flume. Make sure the intake tubing does not kink.

If the metering station has a flow meter, you may connect either their cable or a POTW cable to the sampler from the flow meter. Occasionally, you will set up a flow meter to have a comparison reading. Determine the pulse rate and proper setting from the flow, and program the sampler. After entering the data into the sampler, wait to make sure the equipment is pulling samples. After the initial set-up of the sampling equipment, samples will be collected during the remainder of the sampling period. Split samples may be requested by the SROG or equivalent representative. If the volume of the sample is adequate, these may be given, provided the representative supplies the containers and allows the POTW Inspector to pour off the samples.



Upon exiting the confined space, continue to follow the confined space entry procedures as outlined by OSHA Standards. When you return to the sampling vehicle, you must immediately perform field tests and preserve the samples according to the techniques set forth by Standard Methods or the State/Federal Rule.

All paper work must be filled out completely before the sampling crew's departure. This paperwork includes the chain of custody which is turned in to the laboratory with the samples, **"Metering Station Field Observation Form"** or equivalent form that remains with the sampling site file, and the Multi-City Metering Station Sample Record of which the original is given to the Water Quality Inspector or representative and the copy is given to the SROG or equivalent representative. If there is not a representative at the site, these copies will be turned over to the Water Quality Inspector with the originals at the end of the week.

Remember, all paperwork should be completed prior to leaving site or sampling location.

## Wastewater and Pretreatment Compliance Monitoring

There are two types of sampling activities that are performed as part of compliance monitoring for permitted industries: unscheduled and demand.

**Unscheduled sampling** is used to determine the compliance status of the user. Instances of noncompliance are often identified during unannounced monitoring visits. No notice is given for this type of sampling. This type of sampling is performed two to four times a year, at each industrial user site, over a two to five-day period to obtain sampling data.

**Demand sampling** is usually initiated in response to a known or suspected violation, discovered as a result of a self-monitoring report, routine sampling visit, public complaint, unusual influent condition at the wastewater treatment plant, or emergency situations (e.g., plant upsets, sewer line blockages, fires, explosions, etc.).

Most often, this type of sampling is conducted to support enforcement actions against an industrial user. This type of sampling activity is performed on an as needed basis. The length of the sampling program depends on the flow, nature of the wastes, and type of samples (i.e., grab or composite) to be collected. Typically, composite and grab samples are collected at each user site.



### Nonpermitted Industrial Users (User Rate Charge Program) Policy Example

On a periodic basis (i.e., once every two to three years), commercial and minor industrial users are sampled to determine discharge concentrations of various pollutants. Typical types of users which may be sampled include: restaurants, photo processing laboratories, laundries, car washes, and printing shops.

A three- to four-day sampling program is usually conducted at each assigned site. Commercial establishments are sampled to establish BOD and SS levels for various groups of users for the POTW's Finance or Utilities Division.

This activity is also helpful in identifying industrial or commercial users which may discharge pollutants of concern.

### Wastewater Treatment Plant Sampling

POTW samples are collected in accordance with the National Pollutant Discharge Elimination System (**NPDES**) permit which sets discharge limits for certain pollutants and specifies sampling frequencies and sample types.

The POTW is responsible for coordinating the plant sampling activity with laboratory personnel who prepare any special sampling bottles and laboratory appurtenances necessary (i.e. trip blanks, etc.) to complete the sampling objectives.

**Pre-Treatment Monitoring Locations Should:**

- be appropriate for waste stream conditions;
- be representative of the discharge;
- have no bypass capabilities; and
- allow for unrestricted access at all times.

Control Authorities should measure flow to allow for collection of flow-proportioned composite samples, which are required, unless flow-proportional sampling is not feasible. Flow-proportional composite samples are preferred over time composite samples particularly where the monitored discharge is intermittent or variable.

Desired analyses dictate the preparation protocols, equipment, and collection bottles to use to avoid contamination of samples or loss of pollutants through improper collection. Sampling for such pollutants as pH, cyanide, oil and grease, flashpoint, and volatile organic compounds require manual collection of grab samples. Similar to composite samples, grab samples must be representative of the monitored discharge and are to be collected from actively flowing wastestreams. Fluctuations in flow or the nature of the discharge may require collection of and hand-compositing of more than one grab sample to accurately assess compliance.

To ensure defensibility of data, Control Authorities should develop and implement standard operating procedures and policies detailing sample collection and handling protocols in accordance with 40 CFR Part 136. Adherence to proper sample collection and handling protocols, 40 CFR Part 136 approved analytical methodologies, and record keeping requirements [40 CFR §403.12(o)(1)] can be verified through review of field measurement records, chain of custodies, and lab reports. Field measurement records may require information regarding sample location, condition of and programmed settings for sampling equipment, wastewater meter readings, and information for such parameters as pH and temperature which require analysis in the field.



Chain of custody forms serve as a link between field personnel and the laboratory and contain information regarding the sample matrix, type, and handling. Lab reports should contain the minimum information specified in 40 CFR §403.12(o)(1)(ii-iv) as well as any additional information necessary to demonstrate compliance with 40 CFR Part 136 requirements (e.g., analytical methodology, sample preparation date and time, time of analysis).

Use of standardized forms which prompt recording of information necessary for demonstrating compliance with applicable requirements, will aid in ensuring it can be used as admissible evidence in enforcement proceedings or in judicial actions.

## ◆ ***Wastewater Plant Sampling Procedure Example***

Set up two samplers or equivalent at the plant influent channel and two samplers at the plant effluent channel. Two samplers are used to provide sufficient sample quantity and to minimize sampler failure. All sampling equipment must be prepared and cleaned as established in your POTW's procedures. Teflon hose or equivalent is required. Sampling sites are specified in each plants NPDES permit.

**Collect the following composite samples at all sites.**

- (1) Metals Sample - (one 2-liter plastic bottle)

Preserve with 1:1 nitric acid to a pH < 2. Store sample on ice to four degrees Centigrade.

- (2) Cyanide Sample - one (2-liter plastic bottle)

Collect the cyanide sample as a composite in accordance with NPDES permit. Check the sample for chlorine. If  $\text{Cl}_2$  is present, use ascorbic acid to eliminate chlorine. Add NaOH to a pH > 12. Store samples on ice to four degrees Centigrade.

- (3) EPA Test Method 608 and 625<sup>i</sup> samples are informational samples only. These results are used for local limits data.

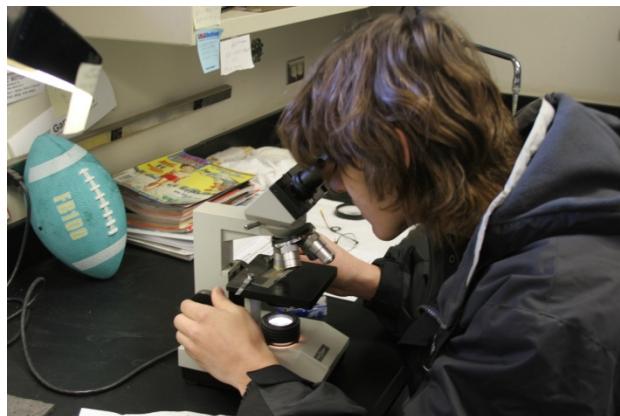
608 and 625 samples are collected as composite samples. At the influent channel: Collect one 1-liter amber glass bottle of each sample (608, 625). Check samples for chlorine. At the effluent channel: Collect one 4-liter amber glass bottle of each sample (608, 625). Check samples for chlorine. If  $\text{Cl}_2$  is present in the samples, use sodium thiosulfate ( $\text{Na}_2\text{S}_2\text{O}_3$ ) to eliminate chlorine. Store samples on ice to four degrees Centigrade.

- (4) **625/Phenols** are collected as a grab sample. Collect one 4-liter amber glass bottle at the effluent channel only. Check the sample for chlorine. If  $\text{Cl}_2$  is present, use sodium thiosulfate ( $\text{Na}_2\text{S}_2\text{O}_3$ ) to eliminate chlorine. Store sample on ice to four degrees Centigrade.

### **Bio-Solids Sampling Example**

Bio-solids (dried sludge) samples are collected at POTWs. Normally, bio-solid samples will be collected from the final storage area for dry sludge. The location of the dried bio-solids may vary based on the individual plants. Sampling frequency will be determined on an as needed basis and to comply with the EPA requirements.

All samples collected are grabs. All samples are collected using a sterile plastic scoop or equivalent in order to avoid any contamination.



**The following is a list of samples that are normally collected:**

PARAMETER	CONTAINER
Helminth Ova & Enteric Virus	1 Qt Plastic Bag (Ziploc)
Metals +	500 ml Plastic Bottle
Nitrogen (total)	4 oz Glass Bottle
TOC (Total Organic Carbon)	4 oz Glass Bottle
Fecal Coliform	(autoclaved from lab)
6 hr hold time	500 ml Plastic Bottle

**Sample Scheduling Example**

An active file is maintained on each sampling location which contains historical data, including past process discharge flow readings, water meter readings, sampling dates, and conditions of sampling site.

**Treated Wastewater Effluent River Sampling Activities Example**

When developing a sampling plan for river sampling, the following considerations must be observed:

- (1) Sampling sites must meet the objectives of the program or study.
- (2) At the sampling sites the river must be flowing freely and the sample must be as representative as possible of river flow at that site. Consideration of all safety factors must be observed.
- (3) Samples must be collected at midstream of the main channel at approximately two-thirds of the depth unless specific depths have been requested.
- (4) All safety precautions must be observed during sampling which includes the use of harnesses, waterproof boots and other equipment.

**Samples from Sewers Example**

Sewer system and user rate sampling are conducted in manholes. General guidelines for selection of sampling locations include the following:

- (1) Samples should be taken at points of high turbulent flow to ensure good mixing and prevent the deposition of solids.
- (2) The sample location should be easily accessible and free of any major safety hazards.
- (3) Sample lines should not be located where there is surface scum.
- (4) If a flow study or a flow/proportional sampling event is required, make sure that the sewer pipe does not have a curve, a drop in the line or any obstructions. These would cause false readings.



## Co-Removal of Emerging Contaminants

This section provides a brief background on emerging contaminants and key findings from studies on the co-removal of emerging contaminants by nutrient removal technologies.

### Background on Emerging Contaminants

The term “emerging contaminants” refers broadly to those synthetic or naturally occurring chemicals, or to any microbiological organisms, that have not been commonly monitored in the environment but which are of increasing concern because of their known or suspected adverse ecological or human health effects. Emerging contaminants can fall into a wide range of groups defined by their effects, uses, or by their key chemical or microbiological characteristics. Two groups of emerging contaminants that are of particular interest and concern at present are endocrine disrupting chemicals (EDCs) and pharmaceutical and personal care products (PPCPs). These compounds are found in the environment, often as a result of human activities.

EDCs may interfere with the endocrine systems by damaging hormone-producing tissues, changing the processes by which hormones are made or metabolized, or mimicking hormones. In addition to natural and synthetic forms of human hormones that are released into the environment, there are a multitude of synthetic organic compounds that are able to disrupt the endocrine system. Public concern about EDCs in the environment has been rapidly increasing since the 1990s when researchers reported unusual sexual characteristics in wildlife. A report by the USGS, found that fish in many streams had atypical ratios of male and female sex hormones (Goodbred et al., 1997). In England, researchers found that male trout kept in cages near WWTP outfalls were developing eggs on their testes and had increased levels of the protein that is responsible for egg production (vitellogenin) (Sumpter, 1995; Kaiser, 1996). Follow-up laboratory studies showed that synthetic forms of estrogen (17 $\alpha$ -ethynodiol (EE2)) could increase vitellogenin production in fish at levels as low as 1-10 ng/L, with positive responses seen down to the 0.1-0.5 ng/L level (Purdom et al., 1994).

Human estrogens have the ability to alter sexual characteristics of aquatic species at trace concentrations as low as 1 ng/L (Purdom et al., 1994). WWTP effluents have been identified as a primary source for EDCs in the environment, with the bulk of their endocrine disrupting activity resulting from human estrogen compounds (Desbrow et al., 1998, Snyder et al., 2001). The synthetic estrogen, EE2, and the natural estrogens, estrone (E1) and 17 $\beta$ -estradiol (E2), are the greatest contributors to endocrine disrupting activity in WWTP effluent (Johnson et al., 2001) with EE2 showing the greatest recalcitrance in WWTPs (Joss et al., 2004). Influent concentrations range from below detection to 70 ng/L for EE2, 670 ng/L for E1 and 150 ng/L for E2 (Vethaak et al., 2005, Clara et al., 2005b). Other EDCs include tributyl tin, which was previously used in paints to prevent marine organisms from sticking to ships, nonylphenol (a surfactant), and bisphenol A (plasticizer and preservative).

PPCPs encompass a wide variety of products that are used by individuals for personal health or cosmetic reasons, and also include certain agricultural and veterinary medicine products. PPCPs comprise a diverse collection of thousands of chemical substances, including prescription and over-the counter therapeutic drugs, veterinary drugs, fragrances, sun-screen products, vitamins, and cosmetics. Many of these products, notably the pharmaceuticals for human or animal use, are specifically designed to be biologically active, and some PPCPs may also fall into the category of EDCs described previously.

### Estrogens of Concern

#### *Name Chemical Structure Name Chemical Structure*

E1	Estrone	C18H22O2
E2	17 $\beta$ -estradiol	C18H24O2
E3	Estriol	C18H24O3
EE2	17 $\alpha$ -ethynodiol	C20H24O2

Currently, municipal sewage treatment plants are engineered to remove conventional pollutants such as solids and biodegradable organic material but are not specifically designed for PPCP removal or for other unregulated contaminants. Wastewater treatment commonly consists of primary settling followed by biological treatment, secondary settling, and disinfection. This treatment can remove more than 90 percent of many of the most commonly known or suspected EDCs found in wastewater influent; however, low concentrations of some suspected EDCs may remain in the wastewater treatment sludge or effluent (WERF, 2005). As discussed in the next section, studies have shown enhanced nutrient removal technologies to be effective in removing low concentrations of some emerging contaminants.

### **Removal of Emerging Contaminants by Nutrient Removal Technologies**

Several studies have examined the effectiveness of current wastewater treatment technologies in the removal of emerging contaminants. Some of these studies are discussed below and their major findings are organized under three subsections: role of activated sludge SRT in removal efficiency, role of nitrifying bacteria in biodegradation, and use of RO to improve removal efficiencies. Details regarding the study design, such as evaluated treatments and contaminants, and a summary of major study findings are provided at the end of this section.

#### **The significant findings are also presented as follows:**

- Removal efficiencies were enhanced for several investigated contaminants at longer SRTs, with critical SRTs for some beyond which removal rates did not improve.
- Longer SRTs allow for the establishment of slower growing bacteria (e.g., nitrifying bacteria in activated sludge), which in turn provide a more diverse community of microorganisms with broader physiological capabilities.
- Nitrifying bacteria may play a key role in biodegradation but the role of heterotrophic bacteria may also play a significant role.
- Reverse osmosis has been found to effectively remove PPCPs below detection limits including those that were not consistently removed at longer SRTs.

One caveat regarding studies on emerging contaminants is that their concentrations in wastewater influent are often quite low (e.g., concentrations of ng/L to  $\mu$ g/L range) and may be close to method detection limits. Therefore, small variations between measured influent and effluent concentrations may show large variations in apparent removal efficiencies, possibly even producing negative calculated removals.

### **Role of Solids Retention Time in Removal Efficiency**

The focus of several studies has been the relationship of the SRT to the removal of emerging contaminants. In particular, many investigated whether longer SRTs would result in increased removal efficiencies for estrogens and other categories of PPCPs. Longer activated sludge SRTs allow for the establishment of slower growing bacteria (e.g., nitrifying bacteria in activated sludge), which in turn provide a more diverse community of microorganisms with broader physiological capabilities.

Clara et al. (2005a), Kreuzinger et al. (2004), and Oppenheimer et al. (2007) observed enhanced removal with increasing SRTs for most of the EDCs and pharmaceuticals tested and found no significant differences in removal performances between conventional activated sludge systems and MBR when operated at similar SRT $_{10}^{\circ}$  C. This is likely due to the molecular weight of the study compounds, which was smaller than the molecular weight cut-off of the ultrafiltration membranes in the MBR.

Researchers have observed similar findings for natural estrogens with higher removal percentages at longer SRTs. Effluent concentrations for three natural estrogens were measured near their detection limits at SRT $_{10}^{\circ}$  C higher than 10 days, with their critical SRT $_{10}^{\circ}$  C estimated between 5 and 10 days (Clara et al., 2005a).

High removal rates of > 90 percent were also observed by Joss et al. (2004) in a study in which they evaluated the removal of E1, E2, and EE2 under aerobic and anaerobic conditions in WWTPs designed for nutrient removal. Joss et al. (2004) also reported that the maximum efficiency is dependent on redox conditions, with the highest removal rate occurring during the reduction of E1 to E2 under aerobic conditions. Clara et al. (2005a) cited examples where conflicting results were obtained for EE2.

Ternes et al. (1999) found no significant elimination of this compound during batch experiments; however, Baronti et al. (2000) and Joss et al. (2004) report greater than 85 percent removal in full-scale WWTPs. For the pharmaceuticals ibuprofen and bezafibrate, Clara et al. (2005a) reported more than 95 percent removal during treatment and calculated the critical value for SRT $10^{\circ}$  C at 5 days for ibuprofen and about 10 days for bezafibrate. Analogous removal results were obtained in several other studies (Stumpf et al., 1998; Buser et al., 1999; Zwiener et al., 2001, as cited in Clara et al., 2005a; Oppenheimer et al., 2007). Clara et al. (2005b) noted no or slight removal of these two pharmaceuticals and two musk fragrances (tonalide and galaxolide) at a WWTP with a low SRT of 1 to 2 days.

Clara et al. (2005a, 2005b) also found that the pharmaceutical carbamazepine was not removed during wastewater treatment. In addition, these studies found contradictory results for diclofenac (e.g., removal rates ranged from no removal to > 70 percent at SRTs of > 10 days (Clara et al., 2005b)). Clara et al. (2005a) also cited several examples where conflicting results were obtained for diclofenac. No significant removal was reported by Buser et al. (1999) and Heberer (2002a); whereas, Ternes et al. (1998) observed elimination rates of up to 70 percent.

Clara et al. (2005a, 2005b) concluded that the removal potential for conventional WWTPs and MBRs depends on the SRT. They further concluded that high removal rates can be achieved at SRTs $10^{\circ}$  C of more than 10 days. These parameters correspond to the design criteria for nitrogen removal in the German Association for Water, Wastewater and Waste (ATV-DVWK, 2000) and the urban wastewater directive of the European Community (91/271/EEC) for WWTPs in sensitive areas. In its 2005, technical brief, "Endocrine Disrupting Compounds and Implications for Wastewater Treatment," WERF summarized information from several studies that examined the effectiveness of current wastewater treatment technologies in the removal of EDCs.

#### **The classes of EDCs included:**

steroids/sterols (naturally occurring, synthetic, and phytoestrogens), organohalides, metals/organometals, alkyl phenols, polycyclic aromatic hydrocarbons (PAHs)/crude oil, and plasticizers.

Although the WERF 2005 technical brief states that in general, EDC treatment effectiveness is improved with increased SRT, it does not provide the specific SRTs that are associated with the cited removal rates.

Oppenheimer et al. (2007) examined the relationship of SRT to treatment removal efficiencies for 20 PPCPs that are commonly found in the influent of U.S. treatment facilities. Many of the studies already discussed here have been conducted primarily in Europe, were conducted at small-scale WWTPs and bench/pilot plants under controlled conditions, and focused on estrogens and prescription pharmaceuticals rather than PPCPs. The Oppenheimer et al. (2007) study also noted trends regarding the effect of HRT and pure oxygen systems compared to conventional aeration systems on PPCP removal.

Oppenheimer et al. (2007) defined a minimum critical SRT as the minimum time needed to consistently demonstrate greater than 80 percent removal. The results of the study showed that this critical SRT was compound dependent but that the majority of the 20 PPCPs were consistently removed in those treatment plants operating at SRTs of 5 to 15 days. Specifically, 9 of 12 frequently occurring PPCPs were effectively removed through secondary treatment (e.g., ibuprofen).

Conversely, six compounds that are routinely detected in influent (i.e., detected in at least 20 percent of the influent samples) were not well removed by secondary treatment (BHA, DEET, musk ketone, triclosan, benzophenone, galaxolide).

The results for galaxolide conflicted with those reported by Clara et al. (2005b) who generally found high removal rates with SRTs > 10 days and Kreuzinger et al. (2004) who reported removal at SRT between 25 to 40 days. Oppenheimer et al. (2007) found that some compounds such as octylphenol, tri-(chloroethyl) phosphate, and triphenylphosphate were not well removed by secondary treatment; however, these were seldom detected in the influent samples. Based on these results, Oppenheimer et al. (2007) concluded that secondary treatment provides an “effective first barrier” for the 20 PPCPs in the study.

Oppenheimer et al. (2007) also noted trends regarding the effect of HRT and pure oxygen systems compared to conventional aeration systems on PPCP removal but determined that insufficient data existed to make any definitive conclusions. When the PPCP removal performance of a high-purity oxygenated activated sludge plant was compared to a conventional aeration system, the pure oxygen system showed higher removal rates although its SRT was shorter than the conventional aeration plant (i.e., 1 day versus 3 days). In addition, different HRTs operating at similar SRTs had similar removal rates, and therefore suggested that HRT does not significantly affect removal effectiveness in the investigated PPCPs.

### **Role of Nitrifying Bacteria in Biodegradation**

As discussed above, longer SRTs allow for the establishment of slow-growing nitrifying bacteria (i.e., ammonia oxidizing bacteria and nitrite-oxidizing bacteria). Several studies evaluated whether nitrifying bacteria improve the biodegradation of certain emerging contaminants. Major findings from some of these studies are discussed in this section.

The WERF (2005) technical brief indicated that secondary biological treatment that includes nitrification, nutrient removal, and disinfection may remove more than 90 percent of certain steroids, and >95 percent of alkyl phenols; whereas, secondary biological treatment without nitrification and disinfection may decrease removal of these by more than 15 percent. Batt et al. (2006) investigated the role of nitrifying bacteria in activated sludge in the biodegradation of two pharmaceuticals, iopromide and trimethoprim.

The biodegradation of these compounds was conducted in two lab-scale bioreactors using biomass from a stage-2 activated sludge WWTP (operated at an SRT of 49 days). In one of the bioreactors, nitrification was not inhibited (Batch-1 reactor); in the other, nitrification was inhibited with allylthiourea (Batch-2 reactor). Monitoring was also conducted in the WWTP and compared to results obtained from the batch reactors. Both reactors exhibited high removal rates for iopromide; however for trimethoprim, Batch-1 showed a high removal rate of 70 percent, contrasted to the Batch-2 reactor removal rate of approximately 25 percent with nitrification inhibited. Removal rates within the treatment plant, however, were consistent for both pharmaceuticals, showing significantly higher removal rate after nitrification (approx. 60 percent for iopromide and 50 percent for trimethoprim) compared to activated sludge treatment only (<1 percent for both).

Based on these results, Batt et al. (2006) concluded that nitrifying bacteria have a key role in the biodegradation of pharmaceuticals in WWTP that are operated at higher SRTs. This conclusion is supported by Marttinen et al. (2003), who investigated the fate of phthalates in a WWTP with nitrogen removal and observed that about one third of the removal occurred in the nitrification/denitrification treatment phase.

Studies by Yi and Harper (2007), Khunjar et al. (2007), and others have focused on the mechanisms of estrogen removal during nitrification. Possible mechanisms include sorption of estrogens to solids and biotransformation within the treatment facility, especially in the presence of nitrifying activated sludges (Khunjar et al., 2007).

Ammonia oxidizing bacteria have monooxygenase enzymes for ammonia oxidation and these enzymes have been shown previously to be nonspecific and able to accomplish cometabolic degradation of recalcitrant organics.

Cometabolic degradation is a reasonable hypothesis for estrogen degradation because this compound is present at low ng/L concentrations that are below those expected to support microbial growth on that compound alone. One goal of the Yi and Harper (2007) study was to establish whether biotransformation of EE2 is due to cometabolic activity. They conducted batch experiments using enriched cultures of autotrophic ammonia oxidizers. Their study and others (Vader et al., 2000, Shi et al., 2004, as reported in Yi and Harper, 2007) showed a strong relationship between nitrification and EE2 removal in enriched nitrifying cultures. Based on batch tests with and without a nitrifying bacteria inhibitor, they concluded that EE2 biotransformation can be cometabolically mediated in bioreactors that are enriched for autotrophic nitrifiers. However, Yi and Harper (2007) noted that the heterotrophic microorganisms, if present in activated sludge processes, may also be responsible for some micropollutant biotransformations. Further work is needed in this area as these tests did not identify the EE2 degradation product to confirm cometabolic degradation and the role of heterotrophs was not accounted for in some tests.

The focus of a Khunjar et al. (2007) study was to identify the role of ammonia oxidizing bacteria compared to heterotrophic bacteria in the biotransformation of EE2. They used pure cultures of ammonia oxidizing *Nitrosomonas europaea* and heterotrophic cultures that were enriched with monooxygenase and dioxygenase enzyme systems. Nitrifying activated sludge mixed liquors were taken from two WWTPs to seed the cultures. EE2 concentrations were 10-15 µg/L. The results of their study showed significant sorption of EE2 to the predominantly heterotrophic culture but none to the *N. europaea* culture.

In addition, biotransformation of EE2 was significant in the *N. europaea* culture. They observed three major EE2 metabolites at different phases of *N. europaea* culture growth that suggest differential action on each byproduct by the nitrifying bacteria; however, additional work is needed to identify these byproducts. The authors also noted that additional research is needed with continuous flow cultivated *N. europaea* to determine whether these metabolites are likely to be present in nitrifying activated sludge. Also, *N. europaea* was not significantly inhibited at EE2 concentrations at or below 10 µg EE2/L, suggesting that ammonia oxidation may not be significantly impacted by concentrations of EE2 that may be typical of those found in the environment.

#### **Use of Reverse Osmosis to Improve Removal Efficiencies**

Several studies describe the effectiveness of RO in the removal of PPCP and EDCs from secondary wastewater effluent. Braghetta et al. (2002) calculated the removal rates that could be achieved with a RO step following tertiary treatment for 17 PPCPs. They estimated removals to be > 90 percent for most of the selected compounds. Lower removal rates were estimated for diclofenac (55.2 to 62 percent), ketoprofen (64.3 percent), and paraxanthine (73.7 percent).

As previously discussed, the WERF (2005) technical brief evaluated RO removal rates for several compounds. Specifically, the WERF brief cites numerous studies in which RO achieved removal rates of 90 percent or better for naturally occurring and synthetic steroids, organohalides, metals /organometals, and alkyl phenols.

In addition, Oppenheimer et al. (2007) found that RO was effective in removing all 20 investigated PPCPs below the detection limit including those that were not consistently removed at SRTs of 30 days (i.e., galaxolide) using conventional activated sludge treatment or media filtration.

## Hand Compositing

Hand compositing is a series of time proportional grab samples which are collected and composited by hand. Provided the sample volumes are equal and are collected at even intervals, the results should be the same as if done by an automatic sampler (i.e., flow proportional composite sampling). A specific instance where this sampling method may be used is in metal plating shops which have batch discharges from the treatment tank. Provided the tank contains a homogeneous mixture, a minimum of four grab samples are taken of equal amounts and at evenly spaced intervals of time during discharge, to accurately represent the entire tank.

This should represent the waste characteristics of the entire batch discharge to the sewer. One hand composite per batch discharge would be equivalent to a 24-hour composite sample taken at other types of facilities. The sampling data would be compared with the average daily categorical standards or local limits where applicable.



Parshall Flume and Ultrasonic Flow Meter. Here is a great trick if you do not have a stand for your ultrasonic probe, simply use a reflective street cone to hold your probe. Notice the debris and most POTW's will write a NOV for not maintaining the flume and/or uncleanness.



## POTW's Wastewater Samples

### General

There are four types of samples that are collected by the POTW's Sampling Section: grab, time proportional composites, flow proportional composites, and hand composites. The sampling method used depends largely on the types of analyses to be run, and the nature of the wastestream being sampled. Each sampling method is described in this section.

Most POTW's will define the sampling methods which must be used by industrial users (**IUs**) to obtain representative samples to show compliance with their permits: **Example**

- (1) A grab sample is an individual sample collected in less than 15 minutes without regard for flow or time of day. pH, cyanide, oil and grease, sulfide, and volatile organics must be collected as grab samples.
- (2) 24-hour flow proportional composite samples where feasible. The POTW may waive this requirement if the IU demonstrates that this method is not feasible. Samples would then be taken by means of time proportional composite sampling methods or by hand composite where the IU can demonstrate that this will provide a representative sample of the effluent being discharged.

The volume of sample to be collected by any of these methods is dependent on the number and types of analyses that must be performed.

### Wastewater Grab Samples

Grab samples are individual samples collected in less than 15 minutes without regard to flow or time of day. Grab samples are normally taken manually, but can be pumped. Oil and grease samples and purgeable organics are exceptions and must be taken manually.

**A grab sample is usually taken when a sample is needed to:**



- (1) Provide information about an instantaneous concentration of pollutants at a specific time.
- (2) Quantify the pollutants in a non-continuous discharge (e.g., batch discharge).
- (3) Corroborate composite samples if the waste is not highly variable.
- (4) Monitor parameters not amenable to compositing such as pH, temperature, dissolved oxygen, chlorine, purgeable organics and sulfides, oil and grease, coliform bacteria, and sulfites.

## **Collecting Procedure for Water/Wastewater Grab Samples**

### **Policy Example**

Lower dipper or mouth of the bottle into water just below surface. In some cases, you will need to rinse the bottle or dipper three times in the sample before obtaining the sample.

Retrieve collected sample to clean processing area.

Rinse the outside of the bottle 3 times to remove contamination.

Pour the sample into the required laboratory bottle.

#### **Filtering (for ortho-P and NOx samples)**

You may need to filter the sample; this is true with some water and wastewater samples. Some surface water virus samples need to be filtered.

- Secure caps tightly.
- Bottle preservation is performed in the truck or lab before sampling.
- Secure sample container caps tightly.
- Label the sample containers and place them in an iced cooler before storage.

#### **Timed Composites**

Timed samples are usually taken in instances where the intention is to characterize the wastes over a period of time without regard to flow, or where the flow is fairly constant. Timed composite samples consist of a series of equal volume grab samples taken at regular intervals. Usually the interval is 15 minutes with a maximum sampling duration of 24 hours.

However, other intervals can be used and may be more appropriate under some circumstances. Samplers are available which can take up to 10 discreet samples per bottle, for a total of 240 discreet samples. The sampler may be programmed to take any number of samples into one composite bottle which has a 2.5-gallon capacity.

#### **Flow Proportional Composites**

Flow proportional composite samples consist of: a series of grab samples whose volumes are equal in size and proportion to the flow at the time of sampling. Samples are taken at varying time intervals, or continuous samples taken over a period of time based on the flow. Wherever possible, flow proportional sampling is recommended because it most accurately reflects the nature of the wastestream.

Equal volume samples taken at varying time intervals are most often collected by the sampling inspectors. A flow measuring device should be used in conjunction with the automatic sampler.

This sampling method is used for all sampling activities except for instances where grab samples are required or time proportional sampling is more expedient and can provide the same accuracy as flow proportional sampling (i.e., constant flow levels).

## **Pre-Sampling Procedures Example**

To ensure acceptable analytical results, numerous steps must be followed before a sampling program can be initiated:

- (1) Sampling equipment must be clean and in good working order.
- (2) Sampling site must be selected.
- (3) Types of analyses must be determined.
- (4) Proper sample containers must be selected and prepared.

### **Wastewater Sampling Equipment**

The POTW may use one or more of the following portable samplers, ISCO Ultra-Sonic flow meters, SIGMA Depth Sensor samplers, and SIGMA pH Probe samplers. Safety equipment and other necessary equipment are also used.

The equipment that is kept in the sampling vehicle is dependent on the types of sampling activities planned each week, while the equipment stored in the storeroom is for back-up needs and future sampling demands.

Each sampling vehicle should be equipped with at least one sampler and one flow meter more than is needed for the particular sampling period. For example, three scheduled flow proportionate sampling sites would require a vehicle to be equipped with four samplers and four flow meters. At least one spare battery for each type of equipment taken into the field should also be placed in the sampling vehicle.

Ancillary equipment, such as supports, harnesses, blowers, etc., that must be carried in each vehicle will depend on the nature of the sampling location.

In order to keep the equipment in good working order, it should be maintained and cleaned on a regular basis. Routine maintenance and cleaning procedures should be written into the procedures.

### **Sampling Equipment Maintenance Policy Example**

Basic maintenance for samplers includes: periodic calibration, general equipment checking, and replacement of the internal desiccant and fuses. Routine cleaning should be done as covered in SOP or equivalence.

Basic maintenance of the flow meters includes: periodic replacement of the internal desiccant, plotter paper, ribbon, fuses, and any broken re-roll spool assemblies. **Note:** on this assembly there are two tabs on the sides of this piece which are extremely thin and easily broken.

The NiCad and Gel Cell batteries need to be recharged on a regular basis.

Any battery that reads less than 12.50 when checked should not be installed or left on any of the sampling equipment. At the battery charging station, areas are set aside for batteries that need to be charged and batteries already charged.

To prolong battery life, batteries should be charged for a maximum of 24 hours, in accordance with the procedures described in the manufacturer's operations and maintenance manuals.

It is important to note that charged NiCad batteries, if left unused for a long time, are nevertheless slowly discharging.

Gel cell batteries are generally more stable. Voltage readings should be taken **before** the charged batteries are taken into the field to be sure that they still have a full charge.

When a sampler, flow meter, or ancillary equipment needs more specific repairs, the manufacturer representative should be contacted and arrangements made for repair or replacement of the equipment.

#### **Cleaning Automatic Samplers Policy Example**

Samplers, sample jars, grab beakers, and all other equipment used in collecting samples must be cleaned between their uses at each site, to avoid the possibility of cross contamination. Latex or nitrile gloves or equivalent should be worn to protect against infections and acid burns. The following steps should be taken to ensure the proper cleaning of the sampling equipment.

- (1) Break down the sampler and lay the three components in a row.
- (2) Place the strainers and weights in a plastic bucket.
- (3) Set the glass composite jars and Teflon caps off to the side, to be cleaned separately from the samplers.
- (4) Pour a small amount of diluted (1:128) O-Syl disinfectant and MICRO soap into each sampler component, the bucket containing the strainers and weights, and the composite jars.
- (5) To clean the sampler components:
  - (a) Partially fill the sampler bases and cover with water.
  - (b) Use a brush to scrub the inside and outside of each sampling component. Using a small bottle brush, thoroughly scrub the inside of the intake tube and the float housing of the sampler head (these are critical areas since they come in contact with the sample).
  - (c) Rinse off the soap with fresh water.
  - (d) Stack each component so that it will dry quickly and thoroughly.
  - (e) Reassemble the sampler after the components are dry, and store it in the proper compartment of the sampling van. Leave the sampler lid loose so moisture won't be trapped.
  - (f) Clean the strainers and weights in the bucket. Empty the contents of the bucket and rinse the bucket, strainers, and weights. After they have dried, place them in the proper storage areas of the sampling van.
  - (g) Drain the wastewater tank of the sampling van into the sewer drain.
  - (h) Refill the fresh-water tank on the sampling van with potable water.

## Sampler Bottle Cleaning and Preparation Policy Example

- (1) Fill each jar with O-Syl (same dilution as used in the sampler disinfection), MICRO soap, and fresh water.
- (2) Thoroughly scrub the inside and outside of the jars until they are sparkling clean. Make sure that all oil and grease are removed.
- (3) Rinse the jars with fresh water.
- (4) Pour a small amount of 1:1 nitric acid into one jar, and securely place the proper Teflon cap on the jar. Swirl the nitric acid throughout the jar, remove the lid, and pour the nitric acid into the next jar. Repeat this procedure until all the bottles have been treated. Rinse bottles with water after the acid wash. **NOTE:** Wear safety glasses or a full-face shield to protect your eyes.
- (5) Place jars in the drying oven. If jars are to air dry use Acetone to clean the bottles the same way as stated in (4) above. Let the jars and caps dry completely.
- (6) Place the jars, with their caps on loosely, in their respective places on the sampling van.

### Selection of Pretreatment Sampling Site

In order to ensure the collection of valid samples, a representative sampling site must be selected. For industrial sampling, the sites are designated in the permit.

### Industrial Users - Permitted/Non-permitted Example

The sampling points within an industry vary with each industry depending on the nature of the process and location of pretreatment facilities. Therefore, exact locations must be identified on a case by case basis. However, the following general principles apply in all cases:

- (1) A permanent sampling location(s) must be identified for use by the POTW and the IU.

All permitted industries are required to install a sampling vault. The location of the vault is designated by the enforcement inspector. The enforcement inspector responsible for an individual company or site is responsible for providing directions (**maps**) of the specific sampling points, as well as current copies of permits and the name of the contact person and phone number. This information needs to be kept current in the sampling file. Locations of sampling points need to be compared to what is listed on the current permit. If sampling points that the POTW is using do not agree with permit location, do not sample - refer to Chief Inspector or Supervisor.

- (2) The sampling location should be easily accessible and relatively free of safety hazards.
- (3) For categorical industries, there should be, if possible, no discharge present other than that from the regulated process.

If other wastestreams are combined with the regulated wastestream prior to the sampling location, the combined wastestream formula will need to be utilized. The sampling crew must be aware of lower limits to correctly show analysis on chain of custody.

- (4) If the rate of industrial process discharge flow is needed (i.e., where mass limitations are applied), the sampling location will need to be located where the flow of the wastestream is known or can be measured or estimated and flow rates for the other wastestreams obtained.
- (5) In instances where sampling must be performed in the sewer outside of the building, the IU must install a sampling vault in accordance with Code.

### **Sample Type and Analyses**

Different sample volumes are required for various analyses. In addition, the laboratory has developed standard volumes for routine analyses performed on industrial waste samples as follows:

- (1) BOD/COD/TSS (1000-2000 ml, plastic) or equivalent
- (2) Heavy metals (500-2000 ml, plastic) or equivalent
- (3) Cyanide (2000 ml, plastic) or equivalent
- (4) Oil and grease (1000 ml, level-one glass) or equivalent

### **Selection and Preparation of Sample Containers**

The selection of a sample container is based on the parameter to be measured. The inspector should be familiar with the type of sampling containers and preservatives that are needed.

It is essential that the sample containers be made of chemically resistant material, and do not affect the concentrations of the pollutants to be measured.

In addition, sample containers should have a closure (i.e., leak proof/resistant, Teflon lined) that protects the sample from contamination and be properly labeled before leaving the sampling site.

### **Wastewater Sample Preservation**

Wastewater usually contains one or more unstable pollutants that require immediate analysis or preservation until an analysis can be made.

Sample preservation is needed for composite samples, for example, which may be stored for as long as 24 hours prior to transferring them to the laboratory.

Recommended preservatives and holding times that should be used for specific pollutants are presented in the front of this section.

## **Quality Assurance/Quality Control Policy Example**

Quality Assurance/Quality Control (**QA/QC**) measures taken by the sampling crew include equipment blanks, trip blanks, split samples and duplicate samples. Equipment blanks and trip blanks are routine QA/QC measures.

Split samples are taken for Local Limits (pretreatment) sampling and when requested by an industry or laboratory. Split samples requested by an industry are analyzed by their lab at their expense.

Duplicate samples should be run when requested by a Supervisor or Project Leader.

The laboratory prepares all trip blanks/travel blanks used by the sampling crews. This is performed in the laboratory rather than in the field in order to assure that there is no field contamination in the blanks.

Any contamination detected in the blanks would result from field exposure which could in turn affect collected samples.

### **Chain-of-Custody**

Documentation of all pertinent data concerning the collection, preservation and transportation of samples is critical to the overall success of the Wastewater Sampling Program. If sampling is performed for the Pretreatment program, any sampling data may be used as evidence in court proceedings against a noncompliant industrial user. In this case documentation becomes critical. This form is a legal document and is of major importance in a court hearing. Specific procedures with regard to chain of custody are outlined below:

- (1) The sampling crew takes a sufficient supply of pre-numbered Industrial Waste Lab Reports, (custody forms) and sample containers into the field.
- (2) The sampling crew fills in the sampling form at the time of sample collection, and returns the form to the lab along with the collected sample. Specific information to be completed on the form includes:
  - (a) CODE: The company ID number assigned by supervisor.
  - (b) SITE No.: The sampling point ID number assigned by supervisor.
  - (c) DATE SAMPLED: From - Date sampling began To - Date sample is pulled. If it is a grab sample, only the date the sample was taken will be entered with the other line crossed out.
  - (d) SUBMITTED BY: This will have a preprinted truck number. The sampling crew will write in their initials on the blank line which follows.
  - (e) LABEL: A letter is checked and the type of analysis to be performed.
  - (f) PRESERVATIVE: The method of preservation used. See preservation section to see which preservatives to use.
  - (g) TYPE OF SAMPLE: Check off whether proportional, timed composite, hand composite, flow or grab sample.

- (h) TIME: The time frame needed for collection of the sample. A starting time for sample collection, an ending time, and a total time in hours and quarter hours is recorded, such as 23.25 hours. On a grab sample only, the end time, which is the time the sample was taken, will be entered and the other two lines will be crossed out.
- (i) RELINQUISHED BY: This is the signature of person that relinquishes sample to lab personnel, or to any other person taking custody of the sample.
- (j) DATE: Date sample is submitted to the laboratory or relinquished to another person.
- (k) NOTES TO LAB: Includes any special notes to the lab, such as special analysis required of the sample, a letter code which is assigned to the entity being tested the amount of flow if sample is flow proportional, grab sample pH and temperature, and/or actual sample temperature.
- (l) FIELD TEST: Results of any field tests including sample pH, hexavalent chromium, dissolved sulfides, copper, and residual chlorine.
- (m) RESULTS: The appropriate box(es) need to be checked to correspond to the label designation chosen above.

(3) When the sampling is completed at a site, the sampling crew labels the bottles with the label letter designation. The samples are sealed with chain of custody seals and placed in an ice chest for transportation to the lab.

(4) The sampling crew submits the samples and the chain of custody form to the laboratory.

(5) The laboratory logs the samples and assigns a Lab Reference Number to the sample. The sample is tracked by means of this number.

(6) Laboratory personnel sign and date the form, and return it to the sampling crew who makes two copies of the form. One copy is for the sampling crew files and the other is for data entry. The original form is returned to the laboratory. It is also important to note that the sampling vehicle should be kept locked at all times when the sampling crew is not in the vehicle, or in full view of the vehicle.

## Equipment Maintenance Cleaning Techniques *Example*

It is important to keep all equipment used for sampling clean to reduce the risk of cross contamination.

Is your automatic sampler and equipment clean?

All components of an automatic sampler need to be clean prior to setup, since contamination can occur.

Automatic sampler - Each section should be clean, especially the inlet tube.

Intake tubing - Cleaned or new Tygon or Teflon should be used according to the samples you are collecting.

- Intake line strainer - Clean or stainless steel or Teflon.
- Pump tubing - Clean or new medical grade silicone tubing.

Composite bottle - Generally glass is used for this and it's extremely important that it is clean.

Automatic sampling equipment - all components of an automatic sampler should be cleaned using phosphate free soap and rinsed thoroughly prior to setup.

### **Follow safety procedures (goggles, gloves, etc...) When cleaning equipment!**

Composite bottles used in the automatic sampler need to be cleaned prior to setup.

1. Rinse bottle and cap with tap water to remove any residual contaminants.
2. Wash bottle and cap with a phosphate free soap and tap water.
3. Rinse bottle and cap with tap water to flush off any soap.
4. Add 1:1 nitric acid to bottle, cap and shake to cover entire inside of bottle and cap.  
Pour out 1:1 nitric acid.
5. Triple rinse bottle and cap with analyte free water.
6. Allow bottle and cap to dry.
7. Store Bottle with cap loosely screwed onto bottle to keep contaminants out.

### **Tubing - clean or new**

Using clean tubing (intake and pump) for each sampling event will eliminate the need to clean the tubing. If tubing is not going to be changed before a sampling event, then the tubing will need to be washed.

1. Pump clean tap water through tubing, using the automatic sampler pump to transfer water from one container to another.
2. Place tubing into another container that has a phosphate free soap in it and pump the soapy water through the tubing 3 or 4 times while catching the discharge in a separate container.
3. Pump clean tap water through tubing to rinse out soap.
4. Add 1:1 nitric acid to clean tap water and pump this solution through tubing 3 or 4 times.
5. Pump analyte-free water through tubing until all residual acid has been removed.
6. Keep tubing in a clean place until next sampling event.

## Regular Maintenance Includes the Following

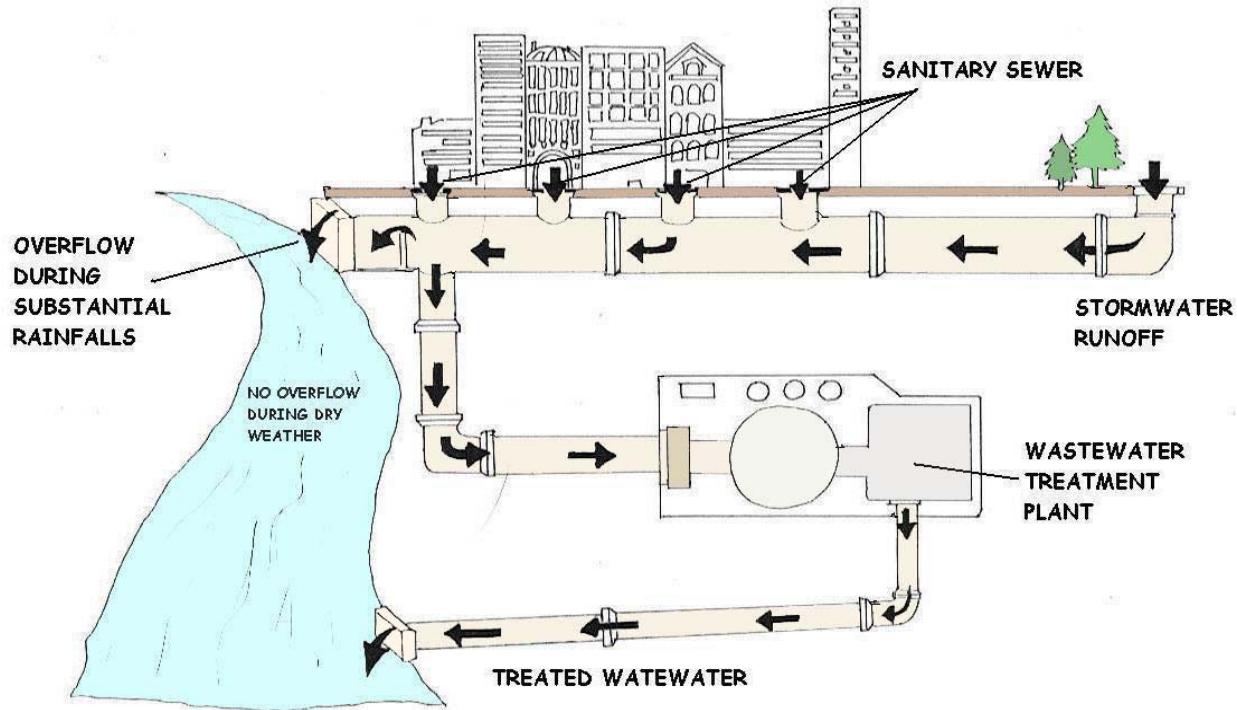
### Automatic Sampler

- Washing
- Drying
- Change desiccant
- Check full bottle shut-off float
- Recharge battery

### Flowmeter

- Washing
- Drying
- Change plotter paper
- Change printer ribbon
- Change desiccant
- Recharge battery

**Record keeping - keep a record of cleaning dates. What was cleaned, and when.**



A modern wastewater treatment facility may have up to 10 different sampling sites some are necessary for compliance purposes, some sites are to please certain agendas or for political purposes. These sampling sites may include Local Limits, QA/QC, Influent, Outfall, Chlorine residual, and many more sites to maintain compliance and to ensure that the plant is running efficiently.

## Proper Sample Handling

The proper handling of water quality samples also includes wearing gloves. Gloves not only protect field personnel, but also prevent potential contamination to the water sample. Always wear powderless, disposable gloves. When sampling for inorganics, wear latex gloves. Nitrile gloves are appropriate for organics.

The following sections provide a field reference for chain of custody procedures, sampling surface water and ground water, and further provides procedures for measuring field parameters and handling water-quality samples.

Use chain-of-custody procedures when coolers and containers are prepared, sealed and shipped. They will remain sealed until used in the field. When making arrangements with the laboratory, make sure you request enough containers, including those for blank and duplicate samples. Order extra sample bottles to allow for breakage or contamination in the field.

Some samples require low-temperature storage and/or preservation with chemicals to maintain their integrity during shipment and before analysis in the laboratory. The most common preservatives are hydrochloric, nitric, sulfuric and ascorbic acids, sodium hydroxide, sodium thiosulfate, and biocides. Many laboratories provide pre-preserved bottles filled with measured amounts of preservatives. Although most federal and state agencies allow the use of pre-preserved sample containers, some may require either cool temperatures or added preservatives in the field.

When the containers and preservatives are received from the laboratory, check to see that none have leaked. Be aware that many preservatives can burn eyes and skin, and must be handled carefully. Sampling bottles should be labeled with type of preservative used, type of analysis to be done and be accompanied by a Material Safety Data Sheet (**MSDS**).

Make sure you can tell which containers are pre-preserved, because extra care must be taken not to overfill them when collecting samples in the field. Check with the laboratory about quality control procedures when using pre-preserved bottles. Coolers used for sample shipment must be large enough to store containers, packing materials and ice. Obtain extra coolers, if necessary. Never store coolers and containers near solvents, fuels or other sources of contamination or combustion. In warm weather, keep coolers and samples in the shade.



### Field Parameters

Measure and record the field parameters of temperature, electrical conductivity, pH and dissolved oxygen in an undisturbed section of stream flow. Other parameters may be measured, if desired.



Water Sample Station commonly found at most water or wastewater treatment plants. This tap will allow the operator to obtain Grab Samples for pH, Temperature, COD, Bacterial, ORP, OUP, Organics and Inorganic field parameters.



pH Meter pH scale is between 0-14 0 being Acid and 14 being Base. For fun, we measured Orange Juice and that is pH 3.5 or so - acid! We also measured Welch's Grape Juice and the meter showed 3.4 - also an acid (and slightly stronger). Here is a close-up of the pH41 meter we use and its pH 7.0 buffer solution (used for calibration and storage - there are a few drops of the buffer solution in the cap of the pH41 to keep it properly moist when not in use).

## QA/QC Field Procedures for Plant Sampling *Example*

### ◆ Duplicate Sampling Procedure

The purpose of Duplicate Samples is to check the laboratory's ability to reproduce analytical results. Duplicate Samples are to be collected using these steps:

1. Determine amount of sample needed. If a flow proportion sample is required, then base the amount of sample needed on the current flow reading. If a flow-proportion sample is not required, then use the predetermined amount for the sampling site.
2. Collect sample using a grab type sampler or a sampling head.
3. Measure the amount determined in Step 1 using a graduated cylinder or other accurate measuring device.
4. Pour measured sample into sample container that is not marked as the Duplicate Sample.
5. Measure same amount as in Step 1.
6. Pour second measured quantity into sample container marked for Duplicate Sample.
7. Process both samples using standard procedures and submit both samples to laboratory.

### ◆ Split Sampling Procedure

The purpose of Split Samples is to check analytical procedures by having the samples analyzed by two different laboratories. Split Samples are to be collected using these steps:

1. Determine amount of sample needed. If a flow proportion sample is required, then base the amount of sample needed on the current flow reading. If a flow-proportion sample is not required, then use the predetermined amount for the sampling site.
2. Collect sample using a grab type sampler or a sampling head.
3. Measure the amount determined in Step 1 using a graduated cylinder or other accurate measuring device.
4. Pour measured sample into sample container that is not marked as the Split Sample.
5. Measure same amount as in Step 1
6. Pour second measured quantity into sample container marked for Split Sample.
7. Process both samples using standard procedures and submit both samples to the laboratory. The laboratory will be responsible for submitting the samples to the outside laboratory that will be analyzing the Split Sample.

### ◆ Trip Blank Procedure

The purpose of Trip Blanks is to determine if the sample bottles have been adequately cleaned, and if sample contamination occurs between the time sample bottles leave the laboratory to the time that samples are returned to the lab.

Trip blanks are prepared by the laboratory using bottles supplied by the sampler. They are picked up by the person who begins the sampling day. Trip blanks are placed in the cooler which contains the other samples and remain there until the samples are turned into the laboratory.



♦ A normal day for a WWT sampler. Here she is looking up a flow rate to determine the volume of wastewater that is flowing. Many samplers do not know simple math formulas to determine flow and rely solely upon the electric measuring devices. These measuring devices can fail or be programmed incorrectly, or batteries can die. You better be prepared for the worst case scenario. Pickle bottle is shown in the middle photograph.

## Field Equipment Blank Procedure

The purpose of Field Equipment Blanks are to test the procedure for cleaning the sample measuring container to determine if cross contamination between sample sites has occurred. These Blanks are needed only at sites where flow-proportion samples are taken. Follow these steps when collecting a Field Equipment Blank:

1. Collect Field Equipment Blank **AFTER** collecting a sample and **BEFORE** moving to the next sampling location.
2. After collecting sample, triple rinse sample measuring container, usually a graduated cylinder, using High Purity water.
3. Open a sealed bottle of High Purity Water.
4. Pour the High Purity Water into the sample measuring container that was just rinsed.
5. Pour the High Purity water from sample measuring device into sample bottles labeled for the Field Equipment Blanks.
6. Repeat Steps 3 through 5 until all Field Equipment Blank sample bottles have been filled.
7. Process samples using standard procedures and submit to laboratory.

An equipment blank is high purity water which has been collected in a composite sample bottle or a series of discrete bottles from an automatic sampler. Equipment blanks are used to evaluate the reliability of composite samples collected in the field.

The data produced from the equipment blank indicates the performance of the sample collection system, which involves the cleaning of sampling equipment, and accessories, preservation techniques, and handling of samples.

The objective is to demonstrate that the samples are not contaminated by inadequate cleaning of equipment, contaminated preservation additives or sample collection techniques, and to provide documented records on Quality Assurance Practices.

Procedures to be followed in collecting the equipment blanks are outlined below.  
(Also see QA/QC check list, example).

- (1) The sampler is to be assembled completely in the manner determined by the parameters the crew will be sampling (i.e. if sampling for organics, Teflon suction tubing must be used at that site). The composite jar inside the sampler must always be rinsed out thoroughly with high purity water.
- (2) Program the sampler to collect the proper amount of high purity water that is representative of the sample parameters that will be collected at that site. Grab samples are excluded. Pump high purity water through the strainer and intake tubing prior to filling the sampler bottle. Then, place the strainer into as many fresh, uncontaminated bottles of high purity water as needed to collect the necessary volume of sample.
- (3) If the sampler is set up in the discrete mode, the crew must then transfer the collected samples into the field composite bottle and shake to mix thoroughly.
- (4) Transfer the sample from the field composite bottle into its respective lab sample bottles. Test and preserve the samples as appropriate for the parameters being analyzed.

- (5) Follow the chain of custody procedures outlined in SOP for turning the samples in to the laboratory. All paperwork must be completed at this time, and all bottles must be marked accordingly. Custody seals must be used. The crew must note the sampling activity in a logbook that is kept specifically for documenting preparation of equipment blanks and/or any other QA activities.

## **Wastewater Sampling Procedures/Techniques**

### **General Guidelines**

In general, the following guidelines should be observed in conducting sampling activities:

- (1) Samples being collected must be representative of the wastestream being tested.
- (2) Samples shall be collected in uncontaminated containers and preserved properly.
- (3) Samples should be of sufficient volume for the required analyses.
- (4) Samples should be stored in a manner which does not alter the properties of the sample prior to chain of custody transfer.
- (5) Samples should be properly and completely identified by marking them with the proper information.
- (6) Sample lines should be as short as possible and the smallest practical diameter to facilitate purging, reduce lag time, and give adequate consideration to maximum transport velocity. Also, they should have sufficient strength to prevent structural failure.
- (7) Sample lines should be pitched downward at least 10 percent to prevent settling or separation of solids contained by the sample.
- (8) Samples should be delivered as quickly as possible to the laboratory.



### **Specific Techniques**

Sampling techniques in addition to the above general guidelines must also recognize differences in sampling methodology, preservation, and analytical methods.

The following sections specify techniques that differ by pollutant group and discuss such factors as sampling methodology (e.g., composite, grab, etc.), type of container, preservation and holding time.

## Sampling Techniques for Volatile Organics

Volatile organics are analyzed in accordance with EPA methods 601, 602, and 603.

Due to the volatility of these compounds, only grab samples can be taken. If a composite sample is needed, individual grab samples must be collected and composited in the laboratory prior to analysis.

The procedures that must be followed in taking these samples are outlined below.

**NOTE:** Gloves, clothing, face, and eye protection must be worn when handling volatile organics.

In addition, the sampling crew must thoroughly clean those parts of the body that have been exposed to these materials.

- (1) For each sampling date, the lab will also provide two additional bottles to be used as a backup in case of breakage. These sampling vials are only good for one week. If any are unused, they must be returned to the lab for disposal.
- (2) The lab will provide one sample trip blank per sampling date. This bottle is to be kept on ice until the samples are submitted to the lab. At least one day prior to sampling, go to the lab and request the sample bottles (40 ml vials) for the specific sampling site, as indicated by the sampling plan. The laboratory will arrange to have the appropriate number of sample bottles prepared, based on the number of analyses to be performed. The sampling crew should make sure that all bottles are provided for these samples by the lab technicians.
- (3) Collect the sample in a clean glass beaker. Test for chlorine with the Hach test kit. If there is any chlorine residual, neutralize the chlorine with sodium thiosulfate ( $\text{Na}_2\text{S}_2\text{O}_3$ ) and retest for chlorine. Repeat until there is no chlorine residual. Make notes on chain of custody sheet if extra amounts of sodium thiosulfate are required for neutralization.
- (4) Remove the vials from the ice. There will be two empty vials for the 601 sample and two vials with HCl for the 602. The HCl will already have been measured into the vials by the lab personnel.
- (5) Fill the vial to just overflowing in such a manner that no air bubbles pass through the sample as the vial is being filled. This is accomplished by pouring the sample from the beaker into the vial along the side of the vial to minimize the possibility of entrapping air in the sample. Do not rinse out or overfill the vials, this will wash out the preservative in the vial.
- (6) Seal the vial so that no air bubbles are entrapped in it. Remember to put the Teflon side of the cap facing down onto the vial.
- (7) To be sure there are no air bubbles, turn the vial upside down and tap it against the palm of the hand. Check to see if there are air bubbles along the sides or bottom of the vial. If there are bubbles,

unseal the vial, top off the vial, and reseal. Check the vial again for the presence of bubbles.

- (8) All samples must be maintained at four degrees centigrade from the time of collection until the time of extraction. Custody seals must be placed on all samples, and all paper work must be filled out properly.
- (9) Return the sample bottles and QA/QC bottles to the laboratory the same day the sample is collected.

#### **Acid/Base/Neutral Extractable Organics and Pesticides Example**

Acid extractable organics are analyzed in accordance with EPA methods 604 and 625. Base/neutral extractable organics are analyzed in accordance with EPA method 625, or individual methods for various groups of compounds including EPA methods 605, 606, 607, 609, 611, and 612. Pesticides are analyzed in accordance with EPA method 608.

The procedures that must be followed in taking these samples are outlined below.

- (1) Samples must be collected in certified clean one-gallon amber glass bottles with Teflon lids.
- (2) Travel blanks or QA/QC bottles may not be required with the samples.
- (3) Grab samples must be collected in amber glass bottles. They do not have to be completely filled, but must be a minimum of 1/3 to 1/2 full. Bottles should not be pre-washed with samples prior to filling.
- (4) For composite sampling, glass composite bottles must be used and pre-cleaned. Teflon tubing must be used for the suction piping. The pump tubing must be medium grade silicone rubber.
- (5) The composite bottle in the sampler must be kept refrigerated (putting ice in the sampler) at 4 degrees Celsius. If amber glass is not used, (i.e. 2 1/2-gallon clear composite sampler bottle,) the sample must be protected from the light during collection and compositing. The compositing must be done in the field, (i.e. when discrete sampling has been used).
- (6) All samples must be iced at four degrees Celsius from the time of collection until extraction.
- (7) The sample should be checked for the presence of chlorine using field test kits that provide results in accordance with EPA methods 330.4 and 330.5. If chlorine is determined to be present, 80 mg of sodium thiosulfate should be added to each bottle. The sample must be retested for chlorine. This procedure must be repeated until there is no residual of chlorine shown. The amount of sodium thiosulfate added must be noted on the chain of custody if in excess of 80 mg.
- (8) All necessary paperwork must be completed at sampling site. All bottles must be properly labeled, and have custody seal.

## Sampling Techniques for Heavy Metals

- (1) Generally, all metal samples collected are to be composite samples, i.e., flow/composite, time/composite, or hand composite.
- (2) For composite sampling, place the lid on the bottle and agitate the bottle to completely mix the composite sample.
- (3) Transfer the required amount from the composite container to either a 500 ml or 2000 ml clean plastic bottle. Check the pH of the sample as described in Section 8.7.2.5.

**Note:** For inductively coupled plasma (**ICP**) metal analysis, a 500 ml clean plastic bottle is required. For extra metals or metals by furnace, a 2000 ml clean plastic bottle is required.
- (4) Add nitric acid (1:1 solution) to the sample to reduce the pH to below 2.0. Usually, 2 ml/500 ml is sufficient. Recheck the pH to be sure it is below 2.0. Make a note on the lab sheet if more than two ml of acid is required to bring the pH below 2.0.
- (5) Label the sample bottle with the corresponding IW number and proper analysis code letter. Attach the custody seal to the sample, then store in the ice chest until transferred to the laboratory. Fill out the IW lab sheet with all the pertinent information, being careful to include all required parameters and the type of analysis required, e.g., ICP/furnace.
- (6) When a grab sample is necessary, rinse out the receiving sample bottle with an aliquot of the sample stream at least three times. Then fill the sample bottle and proceed with steps two through four described above.
- (7) When a split sample is requested (i.e., one for the samplers and one for the user), the composite sample is prepared as described in item one. Providing there is sufficient sample, a portion is transferred into the bottle provided by the user.
- (8) If more than one site is sampled per day, a clean composite container (i.e., two and one half-gallon glass jar), must be used at each site.
- (9) If a discreet sampler is being used, at the time of collection combine all the samples that have been collected into a single clean composite bottle. Then follow the preceding steps one through four, and refer to step six if a split is requested.



## Cyanide

To assure that the sample can be analyzed for cyanide, no chlorine can be present in the sample. Procedures for taking cyanide samples are as follows:

- (1) This sample is normally a grab sample. The cyanide sample is a composite sample when collected as part of Priority Pollutants or Plant Sampling at the waste treatment plants.
  - (a) In the sampling file, check the industries' wastewater discharge permit and locate all cyanide (CN) sampling sites. If the sampling sites are located in a confined space, follow Confined Space procedures before collecting the sample or samples.
  - (b) Collect 2000 ml (maximum), 1000 ml (minimum), of CN sample into a type C plastic bottle.

**NOTE:** 2000 ml is the standard, but for batch dischargers 1000 ml is adequate.

- (c) Test the cyanide sample for pH and temperature with the pH meter. Record the results on the custody sheet (Industrial Waste (IW) lab sheet).
- (d) Test for chlorine with **Hach Total Chlorine Test Kit** (the instructions are located in the kit)
- (e) If chlorine is present in the CN sample, neutralize it with Ascorbic Acid ( $C_6H_8O_6$ ). For ascorbic acid neutralization, add  $C_6H_8O_6$ , a few crystals at a time, until five mls of sample in the test tube produces no color. Then add an additional 0.06 g of  $C_6H_8O_6$  for each liter of sample volume.
- (f) Once all  $Cl_2$  has been neutralized, preserve the sample with Sodium Hydroxide (NaOH) and raise the pH to  $>12$ . Verify the  $>12$  pH with a pH meter or pH test strips.
- (g) Mark on the side of the CN sample bottle the Lab sheet number (using a water proof marker), and place a corresponding custody seal across the sample bottle tightened cap. Place a Cyanide label on the bottle if cyanide is suspected of being present in the sample.
- (h) Store the CN sample in the ice at four degrees Celsius and transport it to the laboratory.

## Total Sulfides

- (1) The Total Sulfide sample is collected as a grab sample only. Use a clean 500 ml plastic bottle to collect the sample. This sample may be pumped into the sample container or collected directly from the discharge side of the sampling device.
- (2) Preserve the sample with 1 ml of 2N Zinc Acetate ( $C_4H_6O_4Zn$ ) and then add Sodium Hydroxide (NaOH) to raise the pH  $> 9$ .
- (3) Label and seal the sample with a custody seal. Cool to  $4^{\circ}C$ .

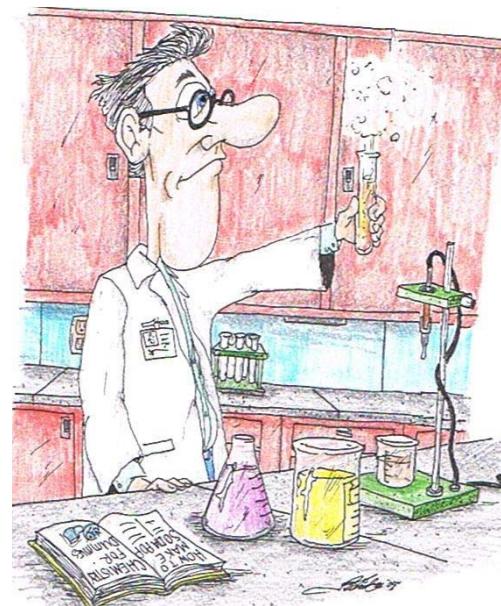
## Oil and Grease/TPH

Oil and grease samples are collected as two separate samples:

**METHOD 413.1** (Oil and Grease). Non-volatile hydrocarbons: vegetable oils, animal fats, waxes, soaps, and related matters.

**METHOD 418.1** (TPH). Extractable petroleum hydrocarbons: light fuels and mineral oils.

- (1) This is a grab sample only. The bottle used to take the sample must be the same bottle given to the laboratory for analysis. Do not pump or transfer the wastewater sample into the bottle. Obtain a level one clean 1000 ml glass bottle, do not use a pre-preserved bottle because you will lose the preservative when collecting the sample.
- (2) Collect the sample by placing the bottle neck down (up-side down) into the effluent stream below the surface. This should be as close to the discharge pipe or point as physically possible. Turn the bottle, allowing the bottle to fill, while keeping the bottle below the surface. Remove the filled bottle and cap it. Never skim the surface of the effluent stream.
- (3) Preserve the sample using five ml of sulfuric acid ( $H_2SO_4$ ) for method 413.1 or hydrochloric acid (HCL) for method 418.1 (6:1 Ratio) to a pH of less than two. Reference 42 of methods 418.1 and 41 of methods 413.1. When more than five ml of HCL is used to lower the pH to less than two, make note of how much additional acid is used, and record this on the lab sheet. Also indicate required analyses method on lab sheet.
- (4) After making sure the sample is well mixed and preserved, seal and attach the proper identification (custody) label to the bottle. Then attach a custody seal across the lid. Store all samples at four degrees centigrade.
- (5) Under no circumstances are Inspectors to collect an oil and grease sample or any other grab sample for IUs.
- (6) All samples must be taken from a good representative flow. If there is any question as to whether there is sufficient flow for a representative sample, do not collect any sample. Make the necessary notes in the file report as to why no sample was obtained.



## BOD/COD/SS

- (1) 24-hour composite sampling is always used for this test. Agitate the bottle to completely mix the composite sample. Do not allow the solids to settle out before you pour off the sample.
- (2) When more than one sample is being taken from a composite bottle, the BOD/COD/SS is taken first. The lab needs 1000 ml if the sample is cloudy or has solids. If the sample is clear, you must collect 2000 ml. Transfer the appropriate volume to the sample bottle.
- (3) Take the pH/temperature of the sample with either pH paper and a thermometer, or the pH meter carried on the sampling trucks.
- (4) Label the sample bottle and place a custody seal over the lid. Store on ice at four degrees centigrade.
- (5) Should split samples be requested, they are given when it is sure there is enough sample for POTW's requirements. Users must provide their own sample containers and allow POTW's staff to pour off samples.



### Rotating Bar Screens

The wastewater headworks is a key sampling location both for compliance and for process control.

## **Virus Sampling Example**

Viruses are microbiological organisms which can cause infectious diseases. Wastewater recharge and sewage disposal into the environment may contribute to the occurrence of viruses in surface water and groundwater. Viruses are the most mobile and infectious of the waterborne pathogens. Large volumes of water must be filtered to detect viruses. This involves passing the water samples through a cartridge filter by use of a gasoline driven pump.

### **(1) Equipment Needed**

Most of the equipment required for virus sampling should be available on the sampling trucks. However, some equipment is virus sampling specific. The needed equipment is as follows:

- (a) Gasoline/oil powered water pump or equivalent
- (b) Hoses - intake (supplied with pump) and discharge (garden type, with female connectors at both ends)
- (c) Two 55-gallon plastic containers or equivalent
- (d) Filter apparatus
- (e) Cartridge filters
- (f) Sodium thiosulfate (two 500 gram bottles/site)
- (g) Gasoline can with gas/oil mixture
- (h) Hach total chlorine test kit
- (i) Large plastic Zip-lock bags (supplied with cartridges)
- (j) Chain of custody sheets
- (k) Thermometer
- (l) Water-proof marker
- (m) Latex gloves
- (n) Liquid bleach
- (o) Cooler with blue ice
- (p) pH meter

### **(2) Sampling Procedure**

Check the pump for gas/oil prior to starting (**Note:** do not fill while it is running). Make sure the gas/oil mixture is correct by checking the mixing instructions on the side of the two-cycle pump oil can. Latex gloves should be worn for protection, and to prevent contamination of the filters.

Connect the hoses and filter housing (with no filter) to the pump, and run the effluent through it for one to two minutes to flush the system. Next, pump effluent into the two 55-gallon drums and rinse them out. (**Note:** If disinfection was not possible after the last sampling, then 50-100 gallons of effluent should be pumped through the entire equipment set up prior to placing the filter in the housing.)

Pump effluent almost to the top (just above the handles) of both containers. While the drums are filling, check the water in the drums for chlorine using the Hach test kit and record the results and the temperature on the custody sheet.

If chlorine is present and needs to be eliminated, add 500 grams of sodium thiosulfate to each container to eliminate it.

After visual observation has determined that all the sodium thiosulfate has dissolved, retest to make sure there is a <0.1 ppm chlorine residual. If chlorine

was removed, take the hose from the channel, allow it to drain, and re-prime the pump with the de-chlorinated water.

Pump this water through the system to flush it, and adjust the flow to fill a one-gallon jug in about 15-20 seconds. Don't waste too much water, as the flow can be adjusted after the filter is inserted. Install the filter into the blue holder, being very careful not to touch it with your hands (wear clean latex gloves). There are two black washers that go with the filter, one on the bottom and the other on the top. Make sure these are aligned with the filter housing to prevent leaking. Screw the holder and filter onto the apparatus.

Refuel the pump, restart it, and adjust the water flow so that it is close to 15-20 seconds per gallon. Make sure the housing doesn't leak. Try to keep this amount of flow, since too great a flow will cause pass-through in the filter. Pump the water from both containers until they are empty.

Stop the pump, remove the filter (wear clean latex gloves), and place it in its original zip-lock bag. The washers do not need to go with the filter, but if they fall into the bag it is better to leave them than take the chance of contaminating the filter trying to remove them.

Fill in the information area on the zip-lock bag with a marker, indicating the plant being sampled and the date, and put it in the cooler with the blue ice provided. The blue ice keeps the temperature at 4 degrees Celsius to prevent significant die-off of the viruses.

While at the site, or later at the plant, mix a half-gallon of bleach to 10 gallons of clean water. Pump it through the flow system and the containers. Rinse everything with fresh water and drain it so it is ready for the next time. Let the pump cool before storing it. Store the gas/oil mixture in the warehouse flammable storage cabinet.



### **Parasitological Sampling**

Parasitological sampling utilizes the same equipment and techniques as in the virus sampling described above. However, a different type of filter, which is provided by the Lab, is used.

## Field Tests *Example*

### Sampling Procedures for Hexavalent Chromium (Hach Kit)

- (1) Rinse out the two color viewing tubes with a portion of the sample to be tested.
- (2) Refill one of the color viewing tubes to the 5 ml mark with a sample (this is the test sample). Using the clippers provided in the test kit, open one ChromaVer three chromium reagent powder pillow. Add the contents of the pillow to the sample. Stopper and shake to mix and put the tube in the color comparator.
- (3) Fill the other viewing tube with a sample and put it in the left side of the color comparator (this is the blank).
- (4) Let the viewing tubes sit in the color comparator for approximately 5 minutes. The samples should not be exposed to direct sunlight.
- (5) Hold the color comparator up to a light source and view the two samples through the two openings in the front. Rotate the dial on the holder until the color appears the same in both samples. Record the results from the dial (which is read in mg/l Cr +6) onto the chain of custody form.

### Sampling Techniques for Dissolved Sulfides (Chemetrics, Inc. Kit)

- (1) Collect a 25 ml grab sample in the container provided.
- (2) Add three drops of activator (amber colored liquid) and mix well.
- (3) Break a sulfide chemet Type S glass ampule and add the contents to the 25 ml container.
- (4) Let stand five minutes.
- (5) Take a reading and record the results on the chain of custody form. If the reading is 0.0 then show the results less than 0.1 mg/l.

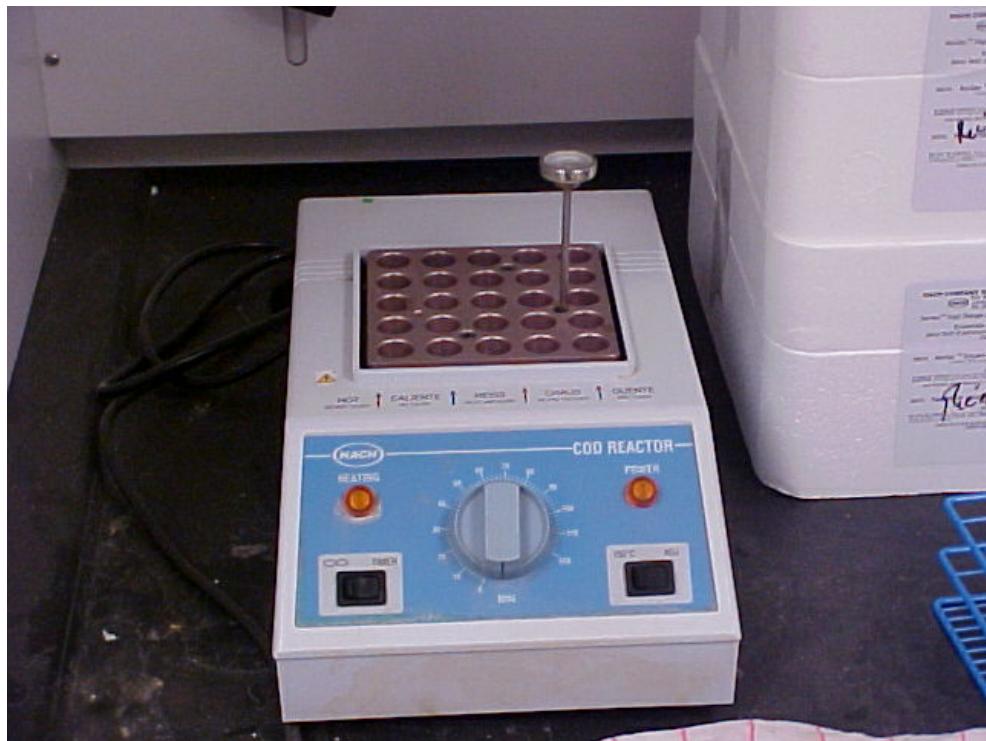
### Sampling Techniques for Free and Total Chlorine (older colorwheel Hach Kit)

Procedures for determining free chlorine are as follows.

- (1) Rinse out the two color viewing tubes with a portion of the sample to be tested.
- (2) Refill one of the color viewing tubes to the 5 ml mark with a sample (this is the test sample). Using the clippers provided in the test kit, open one DPD free chlorine reagent powder pillow. Add the contents of the pillow to the sample. Stopper and shake to mix and put the tube in the color comparator. All of the powder does not have to dissolve to obtain correct readings.
- (3) Fill the other viewing tube with the original sample and put it in the left side of the color comparator (this is the blank).
- (4) Let the viewing tubes sit in the color comparator for approximately 1 minute. The samples should not be exposed to direct sunlight.
- (5) Hold the color comparator up to a light source and view the two samples through the two openings in the front. Rotate the dial on the holder until the color appears the same in both samples. Record the results from the dial (which is read in mg/l free chlorine) onto the chain of custody form.

**Procedures for determining total chlorine are as follows.**

- (1) Rinse out the two color viewing tubes with a portion of the sample to be tested.
- (2) Refill one of the color viewing tubes to the 5 ml mark with a sample (this is the test sample). Using the clippers provided in the test kit, open one DPD total chlorine reagent powder pillow. Add the contents of the pillow to the sample. Stopper and shake to mix and put the tube in the color comparator. All of the powder does not have to dissolve to obtain correct readings.
- (3) Fill the other viewing tube with a sample and put it in the left side of the color comparator (this is the blank).
- (4) Let the viewing tubes sit in the color comparator for approximately 3 minutes. The samples should not be exposed to direct sunlight.
- (5) Hold the color comparator up to a light source and view the two samples through the two openings in the front. Rotate the dial on the holder until the color appears the same in both samples. Record the results from the dial (which is read in mg/l total chlorine) onto the chain of custody form.



COD Reactor

# Dissolved Oxygen

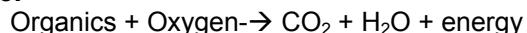
Dissolved oxygen (DO) in water is not considered a contaminant. However, the (DO) level is important because too much or not enough dissolved oxygen can create unfavorable conditions. Generally, a lack of (DO) in natural waters creates anaerobic conditions. Anaerobic means without air. Certain bacteria thrive under these conditions and utilize the nutrients and chemicals available to exist. *Under anaerobic conditions the reaction is:*

## **Anaerobic:**



Where the intermediates are butyric acid, mercaptans and hydrogen sulfide gas. At least two general forms of bacteria act in balance in a wastewater digester: Saprophytic organisms and Methane Fermenters. The saprophytes exist on dead or decaying materials. The methane fermenters live on the volatile acids produced by these saprophytes. The methane fermenting bacteria require a pH range of 6.6 to 7.6 to be able to live and reproduce. Aerobic conditions indicate that dissolved oxygen is present. Aerobic bacteria require oxygen to live and thrive. When aerobes decompose organics in the water, the result is carbon dioxide and water.

## **Aerobic:**



Dissolved Oxygen in a water sample can be detrimental to metal pipes in high concentrations because oxygen helps accelerate corrosion. Oxygen is an important component in water plant operations. Its primary value is to oxidize iron and manganese into forms that will precipitate out of the water. It also removes excess carbon dioxide. The amount of dissolved oxygen in a water sample will affect the taste of drinking water also.

## **Methods of Determination**

There are two methods that we will be using in the lab. The membrane electrode method procedure is based on the rate of diffusion of molecular oxygen across a membrane. The other is a titrimetric procedure (Winkler Method) based on the oxidizing property of the (DO). Many factors determine the solubility of oxygen in a water sample. Temperature, atmospheric pressure, salinity, biological activity and pH all have an effect on the (DO) content.



## **Iodometric Test**

The iodometric (titration) test is very precise and reliable for (DO) analysis of samples free from particulate matter, color and chemical interferences. Reactions take place with the addition of certain chemicals that liberate iodine equivalent to the original (DO) content. The iodine is then measured to the starch iodine endpoint. We then calculate the dissolved oxygen from how much titrate we use. Certain oxidizing agents can liberate iodine from iodides (positive interference), and some reducing agents reduce iodine to iodide (negative interferences). The alkaline Iodide-Azide reagent effectively removes interference caused by nitrates in the water sample, so a more accurate determination of (DO) can be made.

Methods of analysis are highly dependent on the source and characteristics of the sample. The membrane electrode method involves an oxygen permeable plastic membrane that serves as a diffusion barrier against impurities. Only molecular oxygen passes through the membrane and is measured by the meter. This method is excellent for field testing and continuous monitoring. Membrane electrodes provide an excellent method for (DO) analysis in polluted, highly colored turbid waters and strong waste effluents.

These interferences could cause serious errors in other procedures. Prolonged usage in waters containing such gases as  $\text{H}_2\text{S}$  tends to lower cell sensitivity. Frequent changing and calibrating of the electrode will eliminate this interference.

Samples are taken in BOD bottles where agitation or contact with air is at a minimum. Either condition can cause a change in the gaseous content. Samples must be determined immediately for accurate results. The dissolved oxygen test is the one of the most important analyses in determining the quality of natural waters. The effect of oxidation wastes on streams, the suitability of water for fish and other organisms and the progress of self-purification can all be measured or estimated from the dissolved oxygen content. In aerobic sewage treatment units, the minimum objectionable odor potential, maximum treatment efficiency and stabilization of wastewater are dependent on maintenance of adequate dissolved oxygen. Frequent dissolved oxygen measurement is essential for adequate process control.

## Terms

**Aerobic (AIR-O-bick)** a condition in which free or dissolved oxygen is present in the aquatic environment.

**Aerobic Bacteria (aerobes)**— bacteria which will live and reproduce only in an environment containing oxygen. Oxygen combined chemically, such as in water molecules ( $\text{H}_2\text{O}$ ), cannot be used for respiration by aerobes.

**Anaerobic (AN-air O-bick)**- a condition in which “**free**” or dissolved oxygen is not present in the aquatic environment.

**Anaerobic Bacteria (anaerobes)** – bacteria that thrive without the presence of oxygen.

**Saprophytic bacteria** – bacteria that break down complex solids to volatile acids.

**Methane Fermenters** – bacteria that break down the volatile acids to methane ( $\text{CH}_4$ ) carbon dioxide ( $\text{CO}_2$ ) and water ( $\text{H}_2\text{O}$ ).

**Oxidation** – the addition of oxygen to an element or compound, or removal of hydrogen or an electron from an element or compound in a chemical reaction. The opposite of reduction.

**Reduction** – the addition of hydrogen, removal of oxygen or addition of electrons to an element or compound. Under anaerobic conditions in wastewater, sulfur or compounds elemental sulfur are reduced to  $\text{H}_2\text{S}$  or sulfide ions.

## Procedure for Dissolved Oxygen Determination

### METER-PROBE METHOD

1. Collect a water sample in the clean 300-ml glass stoppered BOD bottle for two or three minutes to make sure there are no air bubbles trapped in the bottle. Do one Tap water sample and one DI water sample. Mark the BOD bottles.
2. Insert the DO probe from the meter into your BOD bottles. Record the DO for Tap and DI water. Now continue with the Winkler Burette method.

### PROCEDURES FOR WINKLER BURET METHOD

1. Add the contents of one **MANGANESE SULFATE** powder pillow and one **ALKALINE IODIDE-AZIDE** reagent powder pillow to each of your BOD bottles (TAP and DI)
2. Immediately insert the stoppers so that no air is trapped in the bottles and invert several times to mix. A flocculent precipitate will form. It will be brownish-orange if dissolved oxygen is present or white if oxygen is absent.
3. Allow the samples to stand until the floc has settled and leaves the solution clear (about 10 minutes). Again invert the bottles several times to mix and let stand until the solution is clear.
4. Remove the stoppers and add the contents of one **SULFAMIC ACID** powder pillow to each bottle. Replace the stoppers, being careful not to trap any air bubbles in the bottles, and invert several times to mix. The floc will dissolve and leave a yellow color if dissolved oxygen is present.
5. Measure 200 ml of the prepared solution by filling a clean 250-ml graduated cylinder to the 200-ml mark. Pour the solutions into clean 250-ml Erlenmeyer flasks. Save the last 100 mls for a duplicate.
6. Titrate the prepared solutions with PAO Titrant, 0.025N, to a pale yellow color. Use a white paper under the flask.
7. Add two droppers full of Starch Indicator Solution and swirl to mix. A dark blue color will develop.
8. Continue the titration until the solution changes from dark blue to colorless (end point). Go Slow- drop by drop. Record the burette reading to the nearest 0.01mls.
9. The total number of ml of PAO Titrant used is equal to the mg/L dissolved oxygen.

## Dissolved Oxygen

### Meter Results

1. De-ionized water \_\_\_\_\_ mg/L
2. Tap water \_\_\_\_\_ mg/L
3. What is the meter procedure measuring?
4. What factors would determine which the best method to use is?
5. What are two forms of bacteria present in a wastewater digester?

### Winkler Method Results

#### 6. De-ionized Water

200ml final Burette reading-  
Sample initial Burette reading- - \_\_\_\_\_ = \_\_\_\_\_ mg/L

100ml final Burette reading-  
duplicate initial Burette reading- - \_\_\_\_\_ dup= \_\_\_\_\_ mg/L  
mls x 2

#### 7. Tap water

200ml final Burette reading-  
Sample initial Burette reading- - \_\_\_\_\_ = \_\_\_\_\_ mg/L  
mls

100ml final Burette reading  
Sample initial Burette reading- - \_\_\_\_\_ = \_\_\_\_\_ mg/L  
mls x 2

8. What are some factors that can alter the (DO) content prior to testing?
9. Were your samples anaerobic or aerobic?
10. Why is it important to monitor the (DO) content of water and wastewater?

Be specific and give a detailed explanation.

## Sludge Volume Index (SVI)

### Sludge Volume Index Lab

The Sludge Volume Index (**SVI**) of activated sludge is defined as the volume in milliliters occupied by 1g of activated sludge after settling for 30 minutes. The lower the (**SVI**), the better is the settling quality of the aerated mixed liquor. Likewise, high (**SVI**) of 100 or less is considered a good settling sludge.

### Calculation:

The results obtained from the suspended matter test and settleability test on aerated mixed liquor are used to obtain the SVI.

### Calculation:

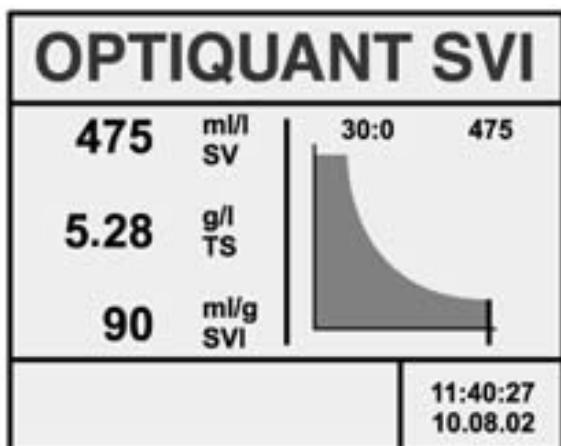
$$\text{SVI} = \frac{\text{ml/L of sludge in settled mixed liquor in 30 min} \times 1000 \text{ mg/g}}{\text{mg/L of suspended matter in mixed liquor}}$$



### At last! Automated sludge volume index monitoring

Your wastewater treatment facility relies on timely monitoring of pH, flow, phosphate, ammonia, nitrate, or DO. Now, real-time assessment of sludge conditions with the new OptiQuant SVI™ Sludge Volume and Sludge Volume Index Analyzer complements these key control parameters. Gone are manual samplings and hasty trips to the lab for analysis – it lets operators operate! No more re-mixing, dilutions, or questionable results. The SVI Analyzer's in-situ sampling yields an accurate, representative sample. It automatically detects bulking that signals upset conditions, gives operators better indication of upset root cause and corrective action, and provides on-the-spot response to chemical dosing adjustments. And the SVI Analyzer doesn't make more work for operators, because its unique sampling vessel construction discourages fouling. For complete information contact Hach at [WWW.Hach.Com](http://WWW.Hach.Com).

Operators select a graphical or numeric SVI controller display. The controller and sampling vessel provide sludge volume monitoring, while an optional OptiQuant™ TS-line suspended solids probe allows automatic calculation of sludge volume index.



## Suspended Matter for Mixed Liquor and Return Sludge

Suspended matter in mixed liquor and return sludge can be used to determine process status, estimate the quantity of biomass, and evaluate the results of process adjustments.

### Apparatus

- Buchner funnel and adaptor
- Filter flask
- Filter paper 110 mm diam., Whatman 1-4
- 103<sup>0</sup> drying oven
- Desiccator
- Balance
- Graduated Cylinder

### Procedure

1. Dry the filter papers in oven at 103<sup>0</sup> c to remove all traces of moisture.
2. Remove papers from oven and desiccate to cool for approximately 5 minutes.
3. Weigh to the nearest 0.01g and record the mass (W<sub>1</sub>)
4. Place the paper in the bottom of the Buchner funnel and carefully arrange so that the outer edges lay snugly along the side. Careful not to touch it with your finger. Use a glass rod. Wet the paper, turn on the vacuum and make a good seal, make a pocket covering the bottom of the funnel.
5. Add 20 to 100 mls of sample at a sufficient rate to keep the bottom of the funnel covered, but not fast enough to overflow the pocket made by the filter paper. Record the Volume used.
6. Remove the filter paper with tweezers. Dry in a 103<sup>0</sup> c oven for 30 minutes. Remove and desiccate. Reweigh the filter paper (W<sub>2</sub>) to the nearest 0.01g.

### Calculation:

#### mg/L Suspended Matter

$$\frac{(W_2) - (W_1) \times 1000}{ML \text{ Sample}} \text{ ML/L}$$

**Where:** (W<sub>1</sub>) and (W<sub>2</sub>) are expressed in mg.

(W<sub>1</sub>) = mass of the prepared filter.

(W<sub>2</sub>) = mass of the filter and sample after the filtration step.

## Settleability Lab

The settled sludge volume of a biological suspension is useful for routine activated sludge plant control. Variations in temperature, sampling and agitation methods, diameter of the settling column, and time between sampling and start of the test can significantly affect results. The same procedure and apparatus should be used each time the test is performed.

### Apparatus

- Two settling columns with a minimum volume of 1000 ml.
- A 1000 ml or larger graduated cylinder or Mallory settlometer may be used as a settling column.

### Procedure

The settleability test on activated sludge should be run immediately after the sample is taken. The mixed liquor sample should be taken at the effluent end of the aeration tanks, while the return sludge sample should be taken at some point between the final settling tank and the point at which the sludge is mixed with primary effluent.

1. Determine the settle ability of mixed liquor and return sludge by allowing 1000 mls of well mixed samples of each to settle in 1000 ml grad. Cylinder or Mallory settleometer. Care should be taken to minimize floc break up during the transfer of the sample to the cylinder.
2. After 30 minutes, record the volume occupied by the sludge to the nearest 5 ml.
3. The reading at the end of 30 minutes is generally used for plant control. Although the settleability test on return sludge is not used in any of the calculations for activated sludge, the result is helpful in determining whether too much or too little sludge is being returned from the final settling tank.

### Calculation: % Settled Sludge

$$\frac{\text{ml of sludge in settled mixed liquor or return sludge} \times 100}{1000}$$

## Sludge Volume Index Lab Report Worksheet

### Suspended Mater Calculations:

$(W_1)$  = \_\_\_\_\_ mg      Duplicate  $(W_1)$  = \_\_\_\_\_ mg

$(W_2)$  = \_\_\_\_\_ mg       $(W_2)$  = \_\_\_\_\_ mg

mls Sample = \_\_\_\_\_ mls Sample = \_\_\_\_\_

mg/L suspended matter = \_\_\_\_\_ dup. \_\_\_\_\_

### Settleability Calculations:

% settled sludge = \_\_\_\_\_

$\frac{(\text{ml of sludge in settled mixed liquor or returned sludge} \times 100)}{1000}$

### Sludge Volume Index Calculations:

$\frac{(\text{ml of sludge in settled mixed liquor in 30 minutes} \times 1000 \text{ mg/g})}{\text{mg/L of suspended matter in mixed liquor}}$

## Glossary

**ABIOGENESIS:** The concept of spontaneous generation (that life can come from non-life). This idea was refuted by Pasteur.

**ABIOTIC:** The non-living components of an organism's environment. The term abiotic is also used to denote a process which is not facilitated by living organisms.

**ABORAL:** Pertaining to the region of the body opposite that of the mouth. Normally used to describe radially symmetrical animals.

**ABSCISIC ACID (ABA):** A plant hormone that generally acts to inhibit growth, promote dormancy, and help the plant withstand stressful conditions.

**ABSENCE OF OXYGEN:** The complete absence of oxygen in water described as Anaerobic.

**ABSORPTION SPECTRUM:** The range of a material's ability to absorb various wavelengths of light. The absorption spectrum is studied to evaluate the function of photosynthetic pigments.

**ACCURACY:** How closely an instrument measures the true or actual value.

**ACCESSORY PIGMENT:** A photosynthetic pigment which absorbs light and transfers energy to chlorophylls during photosynthesis. Because accessory pigments have different absorption optima than chlorophylls, presence of accessory pigments allows photosynthetic systems to absorb light more efficiently than would be possible otherwise.

**ACELLULAR:** Not within cells. Sometimes used as a synonym for unicellular (but multinucleate). Unicellular also pertains to single-celled organisms.

**ACETYL COA:** Acetyl CoenzymeA is the entry compound for the Krebs cycle in cellular respiration; formed from a fragment of pyruvic acid attached to a coenzyme.

**ACETYLCHOLINE:** A neurotransmitter substance that carries information across vertebrate neuromuscular junctions and some other synapses.

**ACID:** Slowly add the acid to water while stirring. An operator should not mix acid and water or acid to a strong base.

**ACID AND BASE ARE MIXED:** When an acid and a base are mixed, an explosive reaction occurs and decomposition products are created under certain conditions.

**ACID RAIN:** Rain that is excessively acidic due to the presence of acid: causing pollutants in the atmosphere. Pollutants include nitrogen and sulfur oxides due to burning of coal and oil.

**ACIDOSIS:** A condition whereby the hydrogen ion concentration of the tissues is increased (and pH decreased). Respiratory acidosis is due to the retention of CO<sub>2</sub>; metabolic acidosis by retention of acids due either to kidney failure or diarrhea.

**ACOELOMATE:** Lacking a coelom.

**ACQUIRED IMMUNITY:** Results from exposure to foreign substances or microbes (also called natural immunity).

**ACROSOME:** An organelle at the tip of a sperm cell that helps the sperm penetrate the egg.

**ACTH (adrenocorticotropic hormone):** A proteinaceous hormone from the anterior pituitary that stimulates the adrenal cortex. Used to stimulate the production of cortisol.

**ACTIN:** A globular protein that links into chains, two of which twist helically about each other, forming microfilaments in muscle and other contractile elements in cells.

**ACTION POTENTIAL:** The stimulus-triggered change in the membrane potential of an excitable cell, caused by selective opening and closing of ion channels.

**ACTION SPECTRUM:** A graph which illustrates the relationship between some biological activity and wavelength of light.

**ACTIVATING ENZYME:** An enzyme that couples a low-energy compound with ATP to yield a high-energy derivative.

**ACTIVATION ENERGY:** In a chemical reaction, the initial investment required to energize the bonds of the reactants to an unstable transition state that precedes the formation of the products.

**ACTIVE SITE:** That specific portion of an enzyme that attaches to the substrate by means of weak chemical bonds.

**ACTIVE TRANSPORT:** The movement of a substance across a biological membrane against its concentration or electrochemical gradient with the help of energy input and specific transport proteins.

**ACTIVATED SLUDGE:** The biologically active solids in an activated sludge process wastewater treatment plant.

**ACTIVATED SLUDGE PROCESS:** A biological wastewater treatment process in which a mixture of wastewater and biologically enriched sludge is mixed and aerated to facilitate aerobic decomposition by microbes.

**ADAPTATION:** Any genetically controlled characteristic that increases an organism's fitness, usually by helping the organism to survive and reproduce in the environment it inhabits.

**ADAPTIVE RADIATION:** This refers to the rapid evolution of one or a few forms into many different species that occupy different habitats within a new geographical area.

**ADHESION:** In chemistry, the phenomenon whereby one substance tends to cling to another substance. Water molecules exhibit adhesion, especially toward charged surfaces.

**ADP (Adenosine diphosphate):** A doubly phosphorylated organic compound that can be further phosphorylated to form ATP.

**ADRENAL GLAND:** An endocrine gland located adjacent to the kidney in mammals. It is composed of an outer cortex, and a central medulla, each involved in different hormone-mediated phenomena.

**ADRENALIN:** A hormone produced by the pituitary that stimulates the adrenal cortex.

**ADSORB:** Hold on a surface.

**ADSORPTION:** *Not to be confused with absorption.* Adsorption is a process that occurs when a gas or liquid solute accumulates on the surface of a solid or a liquid (adsorbent), forming a film of molecules or atoms (the adsorbate). It is different from absorption, in which a substance diffuses into a liquid or solid to form a solution. The term sorption encompasses both processes, while desorption is the reverse process. Adsorption is present in many natural physical, biological, and chemical systems, and is widely used in industrial applications such as activated charcoal, synthetic resins, and water purification. Adsorption, ion exchange, and chromatography are sorption processes in which certain adsorbates are selectively transferred from the fluid phase to the surface of insoluble, rigid particles suspended in a vessel or packed in a column. Similar to surface tension, adsorption is a consequence of surface energy. In a bulk material, all the bonding requirements (be they ionic, covalent, or metallic) of the constituent atoms of the material are filled by other atoms in the material. However, atoms on the surface of the adsorbent are not wholly surrounded by other adsorbent atoms, and therefore can attract adsorbates. The exact nature of the bonding depends on the details of the species involved, but the adsorption process is generally classified as physisorption (characteristic of weak van der Waals forces) or chemisorption (characteristic of covalent bonding).

**ADVANCED:** New, unlike the ancestral condition.

**AERATION:** The addition of air or oxygen to water or wastewater, usually by mechanical means, to increase dissolved oxygen levels and maintains aerobic conditions.

**AEROBIC:** The condition of requiring oxygen; an aerobe is an organism which can live and grow only in the presence of oxygen.

**AEROBIC DIGESTION:** Sludge stabilization process involving direct oxidation of biodegradable matter and oxidation of microbial cellular material.

**AGE STRUCTURE:** The relative numbers of individuals of each age in a population.

**AGONISTIC BEHAVIOR:** A type of behavior involving a contest of some kind that determines which competitor gains access to some resource, such as food or mates.

**AIDS (acquired immune deficiency syndrome):** A condition in which the body's helper T lymphocytes are destroyed, leaving the victim subject to opportunistic diseases.

**AIR GAP SEPARATION:** A physical separation space that is present between the discharge vessel and the receiving vessel; for an example, a kitchen faucet.

**AIR ENTRAINMENT:** The dissolution or inclusion of air bubbles into water.

**ALCOHOL:** Any of a class of organic compounds in which one or more - OH groups are attached to a carbon compound.

**ALDEHYDE:** An organic molecule with a carbonyl group located at the end of the carbon skeleton.

**ALGAE:** Microscopic plants that are free-living and usually live in water. They occur as single cells floating in water, or as multicellular plants like seaweed or strands of algae that attach to rocks.

**ALPHA AND BETA RADIOACTIVITY:** Represent two common forms of radioactive decay. Radioactive elements have atomic nuclei so heavy that the nucleus will break apart, or disintegrate spontaneously. When decay occurs, high-energy particles are released. These high-energy particles are called radioactivity. Although radioactivity from refined radioactive elements can be dangerous, it is rare to find dangerous levels of radioactivity in natural waters. An alpha particle is a doubly-charged helium nucleus comprised of two protons, two neutrons, and no electrons. A beta particle is a high-speed electron. Alpha particles do not penetrate matter easily, and are stopped by a piece of paper. Beta particles are much more penetrating and can pass through a millimeter of lead.

**ALKALINE:** Having a pH of more than 7. Alkaline solutions are also said to be basic.

**ALKALINITY:** Alkalinity or AT is a measure of the ability of a solution to neutralize acids to the equivalence point of carbonate or bicarbonate. Alkalinity is closely related to the acid neutralizing capacity (ANC) of a solution and ANC is often incorrectly used to refer to alkalinity. However, the acid neutralizing capacity refers to the combination of the solution and solids present (e.g., suspended matter, or aquifer solids), and the contribution of solids can dominate the ANC (see carbonate minerals below). The alkalinity is equal to the stoichiometric sum of the bases in solution. In the natural environment carbonate alkalinity tends to make up most of the total alkalinity due to the common occurrence and dissolution of carbonate rocks and presence of carbon dioxide in the atmosphere. Other common natural components that can contribute to alkalinity include borate, hydroxide, phosphate, silicate, nitrate, dissolved ammonia, the conjugate bases of some organic acids and sulfide. Solutions produced in a laboratory may contain a virtually limitless number of bases that contribute to alkalinity. Alkalinity is usually given in the unit mEq/L (milliequivalent per liter). Commercially, as in the pool industry, alkalinity might also be given in the unit ppm or parts per million. Alkalinity is sometimes incorrectly used interchangeably with basicity. For example, the pH of a solution can be lowered by the addition of CO<sub>2</sub>. This will reduce the basicity; however, the alkalinity will remain unchanged.

**ALLANTOIS:** One of the four extraembryonic membranes found associated with developing vertebrates; it serves in gas exchange and as a repository for the embryo's nitrogenous waste. In humans, the allantois is involved in early blood formation and development of the urinary bladder.

**ALLEL:** Alternate forms of a gene which may be found at a given location (locus) on members of a homologous set of chromosomes. Structural variations between alleles may lead to different phenotypes for a given trait.

**ALLOMETRIC:** The variation in the relative rates of growth of various parts of the body, which helps shape the organism.

**ALLOPATRIC SPECIATION:** A type of speciation which occurs when a population becomes segregated into two populations by some sort of geographic barrier (also called geographic speciation). This phenomenon is presumed to have been the mechanism whereby many species of organisms evolved.

**ALLOPOLYPLOID:** A common type of polyploid species resulting from two different species interbreeding and combining their chromosomes.

**ALL-OR-NONE:** (event) An action that occurs either completely or not at all, such as the generation of an action potential by a neuron.

**ALLOSTERIC ENZYME:** An enzyme that can exist in two or more conformations.

**ALLOSTERIC SITE:** A receptor on an enzyme molecule which is remote from the active site. Binding of the appropriate molecule to the allosteric site changes the conformation of the active site, making it either more or less receptive to the substrate.

**ALPHA HELIX:** A spiral shape constituting one form of the secondary structure of proteins, arising from a specific hydrogen bonding structure.

**ALTERNATION OF GENERATIONS:** Occurrences of a multicellular diploid form, the sporophyte, with a multicellular haploid form, the gametophyte.

**ALTERNATIVE DISINFECTANTS:** Disinfectants - other than chlorination (halogens) - used to treat water, e.g. ozone, ultraviolet radiation, chlorine dioxide, and chloramine. There is limited experience and scientific knowledge about the by-products and risks associated with the use of alternatives.

**ALTRUISM:** The willingness of an individual to sacrifice its fitness for the benefit of another.

**ALUMINUM SULFATE:** The chemical name for Alum. The molecular formula of Alum is Al<sub>2</sub>(SO<sub>4</sub>)<sub>3</sub>·14H<sub>2</sub>O. It is a cationic polymer.

**ALVEOLUS:** One of the dead-end, multilobed air sacs that constitute the gas exchange surface of the lungs.

**AMINO ACID:** An organic molecule possessing a carboxyl (COOH) and amino group. Amino acids serve as the monomers of polypeptides and proteins.

**AMINO GROUP:** A functional group consisting of a nitrogen atom bonded to two hydrogens; can act as a base in solution, accepting a hydrogen ion and acquiring a charge of +1.

**AMINOACYL:** tRNA synthetases- A family of enzymes, at least one for each amino acid, that catalyze the attachment of an amino acid to its specific tRNA molecule.

**AMOEBA:** Amoeba (sometimes amœba or ameba, plural amoebae) is a genus of protozoa that moves by means of pseudopods, and is well-known as a representative unicellular organism. The word amoeba or ameba is variously used to refer to it and its close relatives, now grouped as the Amoebozoa, or to all protozoa that move using pseudopods, otherwise termed amoeboids.

**AMOEBOID:** (cell) A cell which has the tendency to change shape by protoplasmic flow. (movement) A streaming locomotion characteristic of Amoeba and other protists, as well as some individual cells, such as white blood cells, in animals.

**AMMONIA:** A chemical made with Nitrogen and Hydrogen and used with chlorine to disinfect water. Most ammonia in water is present as the ammonium ion rather than as ammonia.

**AMP (Adenosine monophosphate):** A singly phosphorylated organic compound that can be further phosphorylated to form ADP.

**AMYLASE:** A starch-digesting enzyme.

**ANABOLISM:** A metabolic pathway of biosynthesis that consumes energy to build a large molecule from simpler ones.

**ANAEROBIC:** Without oxygen. An organism which lives in the absence of oxygen is called an anaerobe. An abnormal condition in which color and odor problems are most likely to occur.

**ANAEROBIC CONDITIONS:** When anaerobic conditions exist in either the metalimnion or hypolimnion of a stratified lake or reservoir, water quality problems may make the water unappealing for domestic use without costly water treatment procedures. Most of these problems are associated with Reduction in the stratified waters.

**ANAEROBIC DIGESTION:** Sludge stabilization process where the organic material in biological sludges are converted to methane and carbon dioxide in an airtight reactor.

**ANAGENESIS:** A pattern of evolutionary change involving the transformation of an entire population, sometimes to a state different enough from the ancestral population to justify renaming it as a separate species; also called phyletic.

**ANALOGOUS:** Characteristics of organisms which are similar in function (and often in structure) but different in embryological and/or evolutionary origins.

**ANALYST:** The analyst must have at least 2 years of college lecture and laboratory course work in microbiology or a closely related field. The analyst also must have at least 6 months of continuous bench experience with environmental protozoa detection techniques and IFA microscopy, and must have successfully analyzed at least 50 water and/or wastewater samples for *Cryptosporidium* and *Giardia*. Six months of additional experience in the above areas may be substituted for two years of college.

**ANCESTRAL TRAIT:** Trait shared by a group of organisms as a result of descent from a common ancestor.

**ANEUPLOIDY:** A chromosomal aberration in which certain chromosomes are present in extra copies or are deficient in number.

**ANION:** A negatively charged ion.

**ANISOGAMOUS:** Reproducing by the fusion of gametes that differ only in size, as opposed to gametes that are produced by oogamous species. Gametes of oogamous species, such as egg cells and sperm, are highly differentiated.

**ANNUAL:** A plant that completes its entire life cycle in a single year or growing season.

**ANOXIC:** A biological environment that is deficient in molecular oxygen, but may contain chemically bound oxygen, such as nitrates and nitrites.

**ANTERIOR:** Referring to the head end of a bilaterally symmetrical animal.

**ANTHROPOMORPHISM:** Attributing a human characteristic to an inanimate object or a species other than a human.

**ANTIBIOTIC:** A chemical that kills or inhibits the growth of bacteria, often via transcriptional or translational regulation.

**ANTIBODY:** A protein, produced by the B lymphocytes of the immune system that binds to a particular antigen.

**ANTICODON:** The specialized base triplet on one end of a tRNA molecule that associates with a particular complementary codon on an mRNA molecule during protein synthesis.

**ANTIDIURETIC HORMONE:** A hormone important in osmoregulation (it acts to reduce the elimination of water from the body).

**ANTIGEN:** A foreign macromolecule that does not belong to the host organism and that elicits an immune response.

**APOMORPHIC CHARACTER:** A derived phenotypic character, or homology, that evolved after a branch diverged from a phylogenetic tree.

**APOSEMATIC COLORATION:** Serving as a warning, with reference particularly to colors and structures that signal possession of defensive device.

**AQUEOUS SOLUTION:** A solution in which water is the solvent.

**ARCHAEBACTERIA:** A lineage of prokaryotes, represented today by a few groups of bacteria inhabiting extreme environments. Some taxonomists place archaeabacteria in their own kingdom, separate from the other bacteria.

**ARCHENTERON:** The endoderm-lined cavity formed during the gastrulation process that develops into the digestive tract of the animal.

**ARISTOTLE:** A Greek philosopher often credited as the first to use empirical and deductive methods in logic.

**ARTIFICIAL SELECTION:** The selective breeding of domesticated plants and animals to encourage the occurrence of desirable traits.

**AS:** The chemical symbol of Arsenic.

**AS NITROGEN:** An expression that tells how the concentration of a chemical is expressed mathematically. The chemical formula for the nitrate ion is  $\text{NO}_3^-$ , with a mass of 62. The concentration of nitrate can be expressed either in terms of the nitrate ion or in terms of the principal element, nitrogen. The mass of the nitrogen atom is 14. The ratio of the nitrate ion mass to the nitrogen atom mass is 4.43. Thus a concentration of 10 mg/L nitrate expressed as nitrogen would be equivalent to a concentration of 44.3 mg/L nitrate expressed as nitrate ion. When dealing with nitrate numbers it is very important to know how numeric values are expressed.

**ASCUS:** The elongate spore sac of a fungus of the Ascomycota group.

**ASEXUAL:** A type of reproduction involving only one parent that produces genetically identical offspring by budding or division of a single cell or the entire organism into two or more parts.

**ASSORTATIVE MATING:** A type of nonrandom mating in which mating partners resemble each other in certain phenotypic characters.

**ASYMMETRIC CARBON:** A carbon atom covalently bonded to four different atoms or groups of atoms.

**ATOM:** The general definition of an ion is an atom with a positive or negative charge. Electron is the name of a negatively charged atomic particle.

**ATOMIC NUMBER:** The number of protons in the nucleus of an atom, unique for each element.

**ATOMIC THEORY:** The physical theory of the structure, properties and behavior of the atom.

**ATOMIC WEIGHT:** The total atomic mass, which is the mass in grams of one mole of the atom (relative to that of  $^{12}\text{C}$ , which is designated as 12).

**ATP (Adenosine triphosphate):** A triply phosphorylated organic compound that functions as "energy currency" for organisms, thus allowing life forms to do work; it can be hydrolyzed in two steps (first to ADP and then to AMP) to liberate 7.3 Kcal of energy per mole during each hydrolysis.

**ATPASE:** An enzyme that functions in producing or using ATP.

**AUTOGENOUS MODEL:** A hypothesis which suggests that the first eukaryotic cells evolved by the specialization of internal membranes originally derived from prokaryotic plasma membranes.

**AUTOIMMUNE DISEASE:** An immunological disorder in which the immune system goes awry and turns against itself.

**AUTONOMIC NERVOUS SYSTEM:** A subdivision of the motor nervous system of vertebrates that regulates the internal environment; consists of the sympathetic and parasympathetic subdivisions.

**AUTOPOLYPLOID:** A type of polyploid species resulting from one species doubling its chromosome number to become tetraploids, which may self-fertilize or mate with other tetraploids.

**AUTOSOME:** Chromosomes that are not directly involved in determining sex.

**AUTOTROPH:** An organism which is able to make organic molecules from inorganic ones either by using energy from the sun or by oxidizing inorganic substances.

**AUXIN:** One of several hormone compounds in plants that have a variety of effects, such as phototropic response through stimulation of cell elongation, stimulation of secondary growth, and development of leaf traces and fruit.

**AUXOTROPH:** A nutritional mutant that is unable to synthesize and that cannot grow on media lacking certain essential molecules normally synthesized by wild-type strains of the same species.

**AXON:** A typically long outgrowth, or process, from a neuron that carries nerve impulses away from the cell body toward target cells.

**AXONEME:** An internal flagellar structure that occurs in some protozoa, such as *Giardia*, *Spironucleous*, and *Trichomonas*.

**B-CELL LYMPHOCYTE:** A type of lymphocyte that develops in the bone marrow and later produces antibodies, which mediate humoral immunity.

**BACKFLOW:** To reverse the natural and normal directional flow of a liquid, gases, or solid substances back in to the public potable (drinking) water supply. This is normally an undesirable effect.

**BACKFLOW PREVENTION:** To stop or prevent the occurrence of, the unnatural act of reversing the normal direction of the flow of liquid, gases, or solid substances back in to the public potable (drinking) water supply. See Cross-connection control.

**BACKSIPHONAGE:** A liquid substance that is carried over a higher point. It is the method by which the liquid substance may be forced by excess pressure over or into a higher point.

**BACTERIA:** Small, one-celled animals too small to be seen by the naked eye. Bacteria are found everywhere, including on and in the human body. Humans would be unable to live without the bacteria that inhabit the intestines and assist in digesting food. Only a small percentage of bacteria cause disease in normal, healthy humans. Other bacteria can cause infections if they get into a cut or wound. Bacteria are the principal concern in evaluating the microbiological quality of drinking water, because some of the bacteria-caused diseases that can be transmitted by drinking water are potentially life-threatening.

**BACTERIOPHAGE:** A bacteriophage (from 'bacteria' and Greek phagein, 'to eat') is any one of a number of viruses that infect bacteria. The term is commonly used in its shortened form, phage. Typically, bacteriophages consist of an outer protein hull enclosing genetic material. The genetic material can be ssRNA (single stranded RNA), dsRNA, ssDNA, or dsDNA between 5 and 500 kilo base pairs long with either circular or linear arrangement. Bacteriophages are much smaller than the bacteria they destroy - usually between 20 and 200 nm in size.

**BACTERIUM:** A unicellular microorganism of the Kingdom Monera. Bacteria are prokaryotes; their cells have no true nucleus. Bacteria are classified into two groups based on a difference in cell walls, as determined by Gram staining.

**BALANCED POLYMORPHISM:** A type of polymorphism in which the frequencies of the coexisting forms do not change noticeably over many generations.

**BARR BODY:** The dense object that lies along the inside of the nuclear envelope in cells of female mammals, representing the one inactivated X chromosome.

**BASAL BODY:** A cell structure identical to a centriole that organizes and anchors the microtubule assembly of a cilium or flagellum.

**BASE PAIRING:** Complementary base pairing refers to the chemical affinities between specific base pairs in a nucleic acid: adenine always pairs with thymine, and guanine always pairs with cytosine. In pairing between DNA and RNA, the uracil of RNA always pairs with adenine. Complementary base pairing is not only responsible for the DNA double helix, but it is also essential for various *in vitro* techniques such as PCR (polymerase chain reaction). Complementary base pairing is also known as Watson-Crick pairing.

**BASE:** A substance that reduces the hydrogen ion concentration in a solution.

**BASEMENT MEMBRANE:** The floor of an epithelial membrane on which the basal cells rest.

**BASIDIUM:** The spore-bearing structure of Basidiomycota.

**BATESIAN MIMICRY:** A type of mimicry in which a harmless species looks like a different species that is poisonous or otherwise harmful to predators.

**BEHAVIORAL ECOLOGY:** A heuristic approach based on the expectation that Darwinian fitness (reproductive success) is improved by optimal behavior.

**BELT PRESS:** A dewatering device utilizing two opposing synthetic fabric belts, revolving over a series of rollers to "squeeze" water from the sludge.

**BENIGN TUMOR:** A noncancerous abnormal growth composed of cells that multiply excessively but remain at their place of origin in the body.

**BENTHIC:** Pertaining to the bottom region of an aquatic environment.

**BENCH TEST:** A small-scale test or study used to determine whether a technology is suitable for a particular application.

**BEST AVAILABLE TECHNOLOGY ECONOMICALLY ACHIEVABLE (BAT):** A level of technology based on the best existing control and treatment measures that are economically achievable within the given industrial category or subcategory.

**BEST MANAGEMENT PRACTICES (BMPs):** Schedules of activities, prohibitions of practices, maintenance procedures, and other management practices to prevent or reduce the pollution of waters of the U.S. BMPs also include treatment requirements, operating procedures and practices to control plant site runoff, spillage or leaks, sludge or waste disposal, or drainage from raw material storage.

**BEST PRACTICABLE CONTROL TECHNOLOGY CURRENTLY AVAILABLE (BPT):**

A level of technology represented by the average of the best existing wastewater treatment performance levels within an industrial category or subcategory.

**BEST PROFESSIONAL JUDGMENT (BPJ):** The method used by a permit writer to develop technology-based limitations on a case-by-case basis using all reasonably available and relevant data.

**BETA PLEATED SHEET:** A zigzag shape, constituting one form of the secondary structure of proteins formed of hydrogen bonds between polypeptide segments running in opposite directions.

**BILATERAL SYMMETRY:** The property of having two similar sides, with definite upper and lower surfaces and anterior and posterior ends. The Bilateria are members of the branch of Eumetazoa (Kingdom Animalia) which possess bilateral symmetry.

**BILE:** A mixture of substances containing bile salts, which emulsify fats and aid in their digestion and absorption.

**BINARY FISSION:** The kind of cell division found in prokaryotes, in which dividing daughter cells each receive a copy of the single parental chromosome.

**BINOMIAL NOMENCLATURE:** Consisting of two names. In biology, each organism is given a *genus* name and a species name (i.e., the human is *Homo sapiens*).

**BIOCHEMICAL OXYGEN DEMAND (BOD):** The BOD test is used to measure the strength of wastewater. The BOD of wastewater determines the milligrams per liter of oxygen required during stabilization of decomposable organic matter by aerobic bacteria action. Also, the total milligrams of oxygen required over a five-day test period to biologically assimilate the organic contaminants in one liter of wastewater maintained at 20 degrees Centigrade.

**BIOGENESIS:** A central concept of biology, that living organisms are derived from other living organisms (contrasts to the concept of abiogenesis, or spontaneous generation, which held that life could be derived from inanimate material).

**BIOGEOCHEMICAL CYCLE:** A circuit whereby a nutrient moves between both biotic and abiotic components of ecosystems.

**BIOGEOGRAPHY:** The study of the past and present distribution of species.

**BIOLOGICAL MAGNIFICATION:** Increasing concentration of relatively stable chemicals as they are passed up a food chain from initial consumers to top predators.

**BIOLOGICAL SPECIES:** A population or group of populations whose members have the potential to interbreed. This concept was introduced by Ernst Mayr.

**BIOMASS:** The total weight of all the organisms, or of a designated group of organisms, in a given area

**BIOME:** A large climatic region with characteristic sorts of plants and animals.

**BIOSOLIDS:** Solid organic matter recovered from municipal wastewater treatment that can be beneficially used, especially as a fertilizer. "Biosolids" are solids that have been stabilized within the treatment process, whereas "sludge" has not.

**BIOSPHERE:** The region on and surrounding the earth which is capable of supporting life. Theoretically, the concept may be ultimately expanded to include other regions of the universe.

**BMR:** The basal metabolic rate is the minimal energy (in kcal) required by a homeotherm to fuel itself for a given time. Measured within the thermoneutral zone for a postabsorptive animal at rest.

**BODY FEED:** Coating or bulking material added to the influent of material to be treated. This adds "body" to the material during filtration cycle.

**BREAK POINT CHLORINATION:** The process of chlorinating the water with significant quantities of chlorine to oxidize all contaminants and organic wastes and leave all remaining chlorine as free chlorine.

**BROMINE:** Chemical disinfectant (HALOGEN) that kills bacteria and algae. This chemical disinfectant has been used only on a very limited scale for water treatment because of its handling difficulties. This chemical causes skin burns on contact, and a residual is difficult to obtain.

**BUFFER:** Chemical that resists pH change, e.g. sodium bicarbonate

**BULKING SLUDGE:** A poor or slow settling activated sludge that results from the prevalence of filamentous organisms. A phenomenon that occurs in activated sludge plants whereby the sludge occupies excessive volumes and will not concentrate readily. This condition refers to a decrease in the ability of the sludge to settle and consequent loss over the settling tank weir. Bulking in activated sludge aeration tanks is caused mainly by excess suspended solids (SS) content. Sludge bulking in the final settling tank of an activated sludge plant may be caused by improper balance of the BOD load, SS concentration in the mixed liquor, or the amount of air used in aeration.

**Ca:** The chemical symbol for calcium.

**CADMIUM:** A contaminant that is usually not found naturally in water or in very small amounts.

**CAKE:** Dewatered sludge material with a satisfactory solids concentration to allow handling as a solid material.

**CALCIUM HARDNESS:** A measure of the calcium salts dissolved in water.

**CALCIUM ION:** Is divalent because it has a valence of +2.

**CALCIUM, MAGNESIUM AND IRON:** The three elements that cause hardness in water.

**CaOCl<sub>2</sub>.4H<sub>2</sub>O:** The molecular formula of Calcium hypochlorite.

**CARBON DIOXIDE GAS:** The pH will decrease and alkalinity will change as measured by the Langelier index after pumping carbon dioxide gas into water.

**CARBONATE HARDNESS:** Carbonate hardness is the measure of Calcium and Magnesium and other hard ions associated with carbonate (CO<sub>3</sub><sup>2-</sup>) and bicarbonate (HCO<sub>3</sub><sup>-</sup>) ions contained in a solution, usually water. It is usually expressed either as parts per million (ppm or mg/L), or in degrees (KH - from the German "Karbonathärte"). One German degree of carbonate hardness is equivalent to about 17.8575 mg/L.

Both measurements (mg/L or KH) are usually expressed "as CaCO<sub>3</sub>" – meaning the amount of hardness expressed as if calcium carbonate was the sole source of hardness. Every bicarbonate ion only counts for half as much carbonate hardness as a carbonate ion does. If a solution contained 1 liter of water and 50 mg NaHCO<sub>3</sub> (baking soda), it would have a carbonate hardness of about 18 mg/L as CaCO<sub>3</sub>. If you had a liter of water containing 50 mg of Na<sub>2</sub>CO<sub>3</sub>, it would have a carbonate hardness of about 29 mg/L as CaCO<sub>3</sub>. Carbonate hardness supplements non-carbonate (a.k.a. "permanent") hardness where hard ions are associated with anions such as Chloride that do not precipitate out of solution when heated. Carbonate hardness is removed from water through the process of softening. Softening can be achieved by adding lime in the form of Ca(OH)<sub>2</sub>, which reacts first with CO<sub>2</sub> to form calcium carbonate precipitate, reacts next with multi-valent cations to remove carbonate hardness, then reacts with anions to replace the non-carbonate hardness due to multi-valent cations with non-carbonate hardness due to calcium. The process requires recarbonation through the addition of carbon-dioxide to lower the pH which is raised during the initial softening process.

**CARBONATE, BICARBONATE AND HYDROXIDE:** Chemicals that are responsible for the alkalinity of water.

**CAROLUS LINNAEUS:** Swedish botanist and originator of the binomial nomenclature system of taxonomic classification

**CATHODIC PROTECTION:** An operator should protect against corrosion of the anode and/or the cathode by painting the copper cathode. Cathodic protection interrupts corrosion by supplying an electrical current to overcome the corrosion-producing mechanism. Guards against stray current corrosion.

**CAUSTIC:** NaOH (also called Sodium Hydroxide) is a strong chemical used in the treatment process to neutralize acidity, increase alkalinity or raise the pH value.

**CAUSTIC SODA:** Also known as sodium hydroxide and is used to raise pH.

**CENTRATE:** The liquid remaining after solids have been removed in a centrifuge.

**CENTRIFUGE:** A dewatering device relying on centrifugal force to separate particles of varying density such as water and solids.

**CENTRIFUGAL FORCE:** That force when a ball is whirled on a string that pulls the ball outward. On a centrifugal pump, it is that force which throws water from a spinning impeller.

**CENTRIFUGAL PUMP:** A pump consisting of an impeller fixed on a rotating shaft and enclosed in a casing, having an inlet and a discharge connection. The rotating impeller creates pressure in the liquid by the velocity derived from centrifugal force.

**CHAIN OF CUSTODY (COC):** A record of each person involved in the possession of a sample from the person who collects the sample to the person who analyzes the sample in the laboratory.

**CHECK VALVE:** Allows water to flow in only one direction.

**CHELATION:** A chemical process used to control scale formation in which a chelating agent "captures" scale-causing ions and holds them in solution.

**CHEMICAL FEED RATE:** Chemicals are added to the water in order to improve the subsequent treatment processes. These may include pH adjusters and coagulants. Coagulants are chemicals, such as alum, that neutralize positive or negative charges on small particles, allowing them to stick together and form larger particles that are more easily removed by sedimentation (settling) or filtration. A variety of devices, such as baffles, static mixers, impellers and in-line sprays, can be used to mix the water and distribute the chemicals evenly.

**CHEMICAL OXIDIZER:** KMnO<sub>4</sub> is used for taste and odor control because it is a strong oxidizer which eliminates many organic compounds.

**CHEMICAL REACTION RATE:** In general, when the temperature decreases, the chemical reaction rate also decreases. The opposite is true for when the temperature increases.

**CHEMICAL SLUDGE:** Sludge resulting from chemical treatment processes of inorganic wastes that are not biologically active.

**CHEMICAL OXYGEN DEMAND (COD):** The milligrams of oxygen required to chemically oxidize the organic contaminants in one liter of wastewater.

**CHLORAMINATION:** Treating drinking water by applying chlorine before or after ammonia. This creates a persistent disinfectant residual called chloramines.

**CHLORAMINES:** A group of chlorine ammonia compounds formed when chlorine combines with organic wastes in the water. Chloramines are not effective as disinfectants and are responsible for eye and skin irritation as well as strong chlorine odors (also known as Combined Chlorine).

**CHLORINATION:** The process in water treatment of adding chlorine (gas or solid hypochlorite) for purposes of disinfection.

**CHLORINE:** A chemical used to disinfect water. Chlorine is extremely reactive, and when it comes in contact with microorganisms in water it kills them. Chlorine is added to swimming pools to keep the water safe for swimming. Chlorine is available as solid tablets for swimming pools. Some public water system's drinking water treatment plants use chlorine in a gas form because of the large volumes required. Chlorine is very effective against algae, bacteria and viruses. Protozoa are resistant to chlorine because they have thick coats; protozoa are removed from drinking water by filtration.

**CHLORINE DEMAND:** Amount of chlorine required to react on various water impurities before a residual is obtained. Also, means the amount of chlorine required to produce a free chlorine residual of 0.1 mg/l after a contact time of fifteen minutes as measured by iodometric method of a sample at a temperature of twenty degrees in conformance with Standard methods.

**CHLORINE FEED:** Chlorine may be delivered by vacuum-controlled solution feed chlorinators. The chlorine gas is controlled, metered, introduced into a stream of injector water and then conducted as a solution to the point of application.

**CHLORINE, FREE:** Chlorine available to kill bacteria or algae. The amount of chlorine available for sanitization after the chlorine demand has been met. Also known as chlorine residual.

**CIRCULATION:** The continual flow of drilling fluid from injection to recovery and recirculation at the surface.

**ClO<sub>2</sub>:** The molecular formula of Chlorine dioxide.

**CLARIFIER:** A settling tank used to remove suspended solids by gravity settling. Commonly referred to as sedimentation or settling basins, they are usually equipped with a motor driven chain and flight or rake mechanism to collect settled sludge and move it to a final removal point.

**COMPOSITE SAMPLE:** To have significant meaning, samples for laboratory tests on wastewater should be representative of the wastewater. The best method of sampling is proportional composite sampling over several hours during the day. Composite samples are collected because the flow and characteristics of the wastewater are continually changing. A composite sample will give a representative analysis of the wastewater conditions.

**COAGULATION:** The best pH range for coagulation is between 5 and 7. Mixing is an important part of the coagulation process you want to complete the coagulation process as quickly as possible. A chemical added to initially destabilize, aggregate, and bind together colloids and emulsions to improve settleability, filterability, or drainability.

**COLIFORM:** Bacteria normally found in the intestines of warm-blooded animals. Coliform bacteria are present in high numbers in animal feces. They are an indicator of potential contamination of water. Adequate and appropriate disinfection effectively destroys coliform bacteria. Public water systems are required to deliver safe and reliable drinking water to their customers 24 hours a day, 365 days a year. If the water supply becomes contaminated, consumers can become seriously ill. Fortunately, public water systems take many steps to ensure that the public has safe, reliable drinking water. One of the most important steps is to regularly test the water for coliform bacteria. Coliform bacteria are organisms that are present in the environment and in the feces of all warm-blooded animals and humans. Coliform bacteria will not likely cause illness. However, their presence in drinking water indicates that disease-causing organisms (pathogens) could be in the water system. Most pathogens that can contaminate water supplies come from the feces of humans or animals. Testing drinking water for all possible pathogens is complex, time-consuming, and expensive. It is relatively easy and inexpensive to test for coliform bacteria. If coliform bacteria are found in a water sample, water system operators work to find the source of contamination and restore safe drinking water. There are three different groups of coliform bacteria; each has a different level of risk.

**COLIFORM TESTING:** The effectiveness of disinfection is usually determined by Coliform bacteria testing. A positive sample is a bad thing and indicates that you have bacteria contamination.

**COLLOIDAL SUSPENSIONS:** Because both iron and manganese react with dissolved oxygen to form insoluble compounds, they are not found in high concentrations in waters containing dissolved oxygen except as colloidal suspensions of the oxide.

**COLORIMETRIC MEASUREMENT:** A means of measuring an unknown chemical concentration in water by measuring a sample's color intensity.

**CHRONIC:** A stimulus that lingers or continues for a relatively long period of time, often one-tenth of the life span or more. Chronic should be considered a relative term depending on the life span of an organism. The measurement of chronic effect can be reduced growth, reduced reproduction, etc., in addition to lethality.

**COMBINED CHLORINE:** The reaction product of chlorine with ammonia or other pollutants, also known as chloramines.

**COMPOSITE SAMPLE:** A water sample that is a combination of a group of samples collected at various intervals during the day. A combination of individual samples of water or wastewater taken at predetermined intervals to minimize the effect of variability of individual samples.

**COMPOSTING:** Stabilization process relying on the aerobic decomposition of organic matter in sludge by bacteria and fungi.

**CONDENSATION:** The process that changes water vapor to tiny droplets or ice crystals.

**CONTACT STABILIZATION PROCESS:** Modification of the activated sludge process where raw wastewater is aerated with activated sludge for a short time prior to solids removal and continued aeration in a stabilization tank.

**CONTACT TIME:** If the water temperature decreases from 70°F (21°C) to 40°F (4°C). The operator needs to increase the detention time to maintain good disinfection of the water.

**CONTAINS THE ELEMENT CARBON:** A simple definition of an organic compound.

**CONTAMINANT:** Any natural or man-made physical, chemical, biological, or radiological substance or matter in water, which is at a level that may have an adverse effect on public health, and which is known or anticipated to occur in public water systems.

**CONTAMINATE:**

1. To make impure or unclean by contact or mixture.
2. To expose to or permeate with radioactivity.

**CONTAMINATION:** A degradation in the quality of groundwater in result of it's becoming polluted with unnatural or previously non-existent constituents.

**COPPER:** The chemical name for the symbol Cu.

**CORROSION:** The removal of metal from copper, other metal surfaces and concrete surfaces in a destructive manner. Corrosion is caused by improperly balanced water or excessive water velocity through piping or heat exchangers.

**CORROSIVITY:** The Langelier Index measures corrosivity.

**CROSS-CONNECTION:** A physical connection between a public water system and any source of water or other substance that may lead to contamination of the water provided by the public water system through backflow. Might be the source of an organic substance causing taste and odor problems in a water distribution system.

**CROSS-CONTAMINATION:** The mixing of two unlike qualities of water. For example, the mixing of good water with a polluting substance like a chemical.

**CRYPTOSPORIDIUM:** A disease-causing parasite, resistant to chlorine disinfection. It may be found in fecal matter or contaminated drinking water. Cryptosporidium is a protozoan pathogen of the Phylum Apicomplexa and causes a diarrheal illness called cryptosporidiosis. Other apicomplexan pathogens include the malaria parasite Plasmodium, and Toxoplasma, the causative agent of toxoplasmosis. Unlike Plasmodium, which transmits via a mosquito vector, Cryptosporidium does not utilize an insect vector and is capable of completing its life cycle within a single host, resulting in cyst stages which are excreted in feces and are capable of transmission to a new host.

**CYANURIC ACID:** Chemical used to prevent the decomposition of chlorine by ultraviolet (UV) light.

**CYANOBACTERIA:** Cyanobacteria, also known as blue-green algae, blue-green bacteria or Cyanophyta, is a phylum of bacteria that obtain their energy through photosynthesis. The name "cyanobacteria" comes from the color of the bacteria (Greek: *kyanós* = blue). They are a significant component of the marine nitrogen cycle and an important primary producer in many areas of the ocean, but are also found on land.

**CYST:** A phase or a form of an organism produced either in response to environmental conditions or as a normal part of the life cycle of the organism. It is characterized by a thick and environmentally resistant cell wall.

**DAILY MAXIMUM LIMITATIONS:** The maximum allowable discharge of pollutants during a 24 hour period. Where daily maximum limitations are expressed in units of mass, the daily discharge is the total mass discharged over the course of the day. Where daily maximum limitations are expressed in terms of a concentration, the daily discharge is the arithmetic average measurement of the pollutant concentration derived from all measurements taken that day.

**DANGEROUS CHEMICALS:** The most suitable protection when working with a chemical that produces dangerous fumes is to work under an air hood.

**DECANT:** Separation of a liquid from settled solids by removing the upper layer of liquid after the solids have settled.

**DECIBELS:** The unit of measurement for sound.

**DECOMPOSE:** To decay or rot.

**DECOMPOSITION OF ORGANIC MATERIAL:** The decomposition of organic material in water produces taste and odors.

**DEMINERALIZATION PROCESS:** Mineral concentration of the feed water is the most important consideration in the selection of a demineralization process. Acid feed is the most common method of scale control in a membrane demineralization treatment system.

**DENITRIFICATION:** A biological process by which nitrate is converted to nitrogen gas.

**DEPOLARIZATION:** The removal of hydrogen from a cathode.

**DESICCANT:** When shutting down equipment which may be damaged by moisture, the unit may be protected by sealing it in a tight container. This container should contain a desiccant.

**DESORPTION:** Desorption is a phenomenon whereby a substance is released from or through a surface. The process is the opposite of sorption (that is, adsorption and absorption). This occurs in a system being in the state of sorption equilibrium between bulk phase (fluid, i.e. gas or liquid solution) and an adsorbing surface (solid or boundary separating two fluids). When the concentration (or pressure) of substance in the bulk phase is lowered, some of the sorbed substance changes to the bulk state. In chemistry, especially chromatography, desorption is the ability for a chemical to move with the mobile phase. The more a chemical desorbs, the less likely it will adsorb, thus instead of sticking to the stationary phase, the chemical moves up with the solvent front. In chemical separation processes, stripping is also referred to as desorption as one component of a liquid stream moves by mass transfer into a vapor phase through the liquid-vapor interface.

**DIATOMACEOUS EARTH:** A fine silica material containing the skeletal remains of algae.

**DIGESTER:** A tank or vessel used for sludge digestion.

**DIGESTION:** The biological decomposition of organic matter in sludge resulting in partial gasification, liquefaction, and mineralization of putrescible and offensive solids.

**DIRECT CURRENT:** A source of direct current (**DC**) may be used for standby lighting in a water treatment facility. The electrical current used in a DC system may come from a battery.

**DISINFECT:** The application of a chemical to kill most, but not all, microorganisms that may be present. Chlorine is added to public water drinking systems drinking water for disinfection. Depending on your state rule, drinking water must contain a minimum of 0.2 mg/L free chlorine. Disinfection makes drinking water safe to consume from the standpoint of killing pathogenic microorganisms including bacteria and viruses. Disinfection does not remove all bacteria from drinking water, but the bacteria that can survive disinfection with chlorine are not pathogenic bacteria that can cause disease in normal healthy humans.

**DISINFECTION:** The treatment of water to inactivate, destroy, and/or remove pathogenic bacteria, viruses, protozoa, and other parasites.

**DISSOLVED OXYGEN:** Can be added to zones within a lake or reservoir that would normally become anaerobic during periods of thermal stratification.

**DISSOLVED SOLIDS:** Solids in solution that cannot be removed by filtration with a 0.45 micron filter.

**DISTILLATION, REVERSE OSMOSIS AND FREEZING:** Processes that can be used to remove minerals from the water.

**DRY ACID:** A granular chemical used to lower pH and or total alkalinity.

**E. COLI, Escherichia coli:** A bacterium commonly found in the human intestine. For water quality analyses purposes, it is considered an indicator organism. These are considered evidence of water contamination. Indicator organisms may be accompanied by pathogens, but do not necessarily cause disease themselves.

**ECDYSONE:** A steroid hormone that triggers molting in arthropods.

**ECOLOGICAL EFFICIENCY:** The ratio of net productivity at one trophic level to net productivity at the next lower level.

**ECOLOGICAL NICHE:** The sum total of an organism's utilization of the biotic and abiotic resources of its environment. The fundamental niche represents the theoretical capabilities and the realized niche represents the actual role.

**ECOLOGY:** The study of how organisms interact with their environments.

**ECOSYSTEM:** The sum of physical features and organisms occurring in a given area.

**ECTODERM:** The outermost tissue layer of an animal embryo. Also, tissue derived from an embryonic ectoderm.

**ECTOTHERM:** An organism that uses environmental heat and behavior to regulate its body temperature.

**EDWARD JENNER:** A pioneer of vaccination; used vaccination with material from cowpox lesions to protect people against smallpox.

**EFFECTIVENESS OF CHLORINE:** The factors which influence the effectiveness of chlorination the most are pH, turbidity and temperature. Effectiveness of Chlorine decreases occurs during disinfection in source water with excessive turbidity.

**EFFECTOR:** The part of an organism that produces a response to a stimulus.

**EFFLUENT:** Partially or completely treated water or wastewater flowing out of a basin or treatment plant.

**ELECTRICAL SYNAPSE:** A junction between two neurons separated only by a gap junction, in which the local currents sparking the action potential pass directly between the cells.

**ELECTROCARDIOGRAM:** A plot of electrical activity of the heart over the cardiac cycle; measured via multiple skin electrodes.

**ELECTROCHEMICAL GRADIENT:** Combined electrostatic and osmotic-concentration gradient, such as the chemiosmotic gradient of mitochondria and chloroplasts.

**ELECTROGENIC PUMP:** An ion transport protein generating voltage across a membrane.

**ELECTROMAGNETIC SPECTRUM:** The entire spectrum of radiation; ranges in wavelength from less than a nanometer to more than a kilometer.

**ELECTRON:** The name of a negatively charged atomic particle. A negatively charged subatomic particle of an atom or ion. In atoms, the number of electrons present is equal to the number of positively charged protons present. Hence, atoms are electrically neutral.

**ELECTRON MICROSCOPE:** A microscope that focuses an electron beam through a specimen, resulting in resolving power a thousandfold greater than that of a light microscope. A transmission EM is used to study the internal structure of thin sections of cells; a scanning EM is used to study the ultrastructure of surfaces.

**ELECTRON TRANSPORT CHAIN:** A series of enzymes found in the inner membranes of mitochondria and chloroplasts. These are involved in transport of protons and electrons either across the membrane during ATP synthesis.

**ELECTRONEGATIVITY:** A property exhibited by some atoms whereby the nucleus has a tendency to pull electrons toward itself.

**ELECTRONIC CHARGE UNIT:** The charge of one electron ( $1.6021 \times 10^{-19}$  coulomb).

**ELECTROSTATIC FORCE:** The attraction between particles with opposite charges.

**ELECTROSTATIC GRADIENT:** The free-energy gradient created by a difference in charge between two points, generally the two sides of a membrane.

**ELEMENT:** Any substance that cannot be broken down into another substance by ordinary chemical means.

**ELIMINATION:** The release of unabsorbed wastes from the digestive tract.

**EMERGENT PROPERTY:** A property exhibited at one level of biological organization but not exhibited at a lower level. For example, a population exhibits a birth rate, an organism does not.

**EMULSION:** A suspension, usually as fine droplets of one liquid in another. A mixture made up of dissimilar elements, usually of two or more mutually insoluble liquids that would normally separate into layers based on the specific gravity of each liquid.

**ENDERGONIC:** A phenomenon which involves uptake of energy.

**ENDOCRINE:** A phenomenon which relates to the presence of ductless glands of the type typically found in vertebrates. The endocrine system involves hormones, the glands which secrete them, the molecular hormone receptors of target cells, and interactions between hormones and the nervous system.

**ENDOCYTOSIS:** A process by which liquids or solid particles are taken up by a cell through invagination of the plasma membrane.

**ENDODERM:** The innermost germ layer of an animal embryo.

**ENDODERMIS:** A plant tissue, especially prominent in roots, that surrounds the vascular cylinder; all endodermal cells have Caspary strips.

**ENDOMEMBRANE SYSTEM:** The system of membranes inside a eukaryotic cell, including the membranous vesicles which associate with membrane sheets and/or tubes.

**ENDOMETRIUM:** The inner lining of the uterus, which is richly supplied with blood vessels that provide the maternal part of the placenta and nourish the developing embryo.

**ENDONUCLEASE:** An enzyme that breaks bonds within nucleic acids. A restriction endonuclease is an enzyme that breaks bonds only within a specific sequence of bases.

**ENDOPLASMIC RETICULUM:** A system of membrane-bounded tubes and flattened sacs, often continuous with the nuclear envelope, found in the cytoplasm of eukaryotes. Exists as rough ER, studded with ribosomes, and smooth ER, lacking ribosomes.

**ENDORPHIN:** A hormone produced in the brain and anterior pituitary that inhibits pain perception.

**ENDOSKELETON:** An internal skeleton.

**ENDOSPERM:** A nutritive material in plant seeds which is triploid (3n) and results from the fusion of three nuclei during double fertilization.

**ENDOSYMBIOTIC:** 1) An association in which the symbiont lives within the host 2) A widely accepted hypothesis concerning the evolution of the eukaryotic cell: the idea that eukaryotes evolved as a result of symbiotic associations between prokaryote cells. Aerobic symbionts ultimately evolved into mitochondria; photosynthetic symbionts became chloroplasts.

**ENDOTHELIUM:** The innermost, simple squamous layer of cells lining the blood vessels; the only constituent structure of capillaries.

**ENDOTHERMIC:** In chemistry, a phenomenon in which energy is absorbed by the reactants. In physiology, this term concerns organisms whose thermal relationship with the environment is dependent substantially on internal production of heat.

**ENDOTOXIN:** A component of the outer membranes of certain gram-negative bacteria responsible for generalized symptoms of fever and ache.

**ENERGY:** The capacity to do work by moving matter against an opposing force.

**ENHANCER:** A DNA sequence that recognizes certain transcription factors that can stimulate transcription of nearby genes.

**ENTAMOEBA HISTOLYTICA:** Entamoeba histolytica, another water-borne pathogen, can cause diarrhea or a more serious invasive liver abscess. When in contact with human cells, these amoebae are cytotoxic. There is a rapid influx of calcium into the contacted cell, it quickly stops all membrane movement save for some surface blebbing. Internal organization is disrupted, organelles lyse, and the cell dies. The ameba may eat the dead cell or just absorb nutrients released from the cell.

**ENTERIC:** Rod-shaped, gram-negative, aerobic but can live in certain anaerobic conditions; produce nitrite from nitrate, acids from glucose; include Escherichia coli, Salmonella (over 1000 types), and Shigella.

**ENTEROVIRUS:** A virus whose presence may indicate contaminated water; a virus that may infect the gastrointestinal tract of humans.

**ENTROPY:** A type of energy which is not biologically useful to do work (in contrast to free energy).

**ENVELOPE:** 1) (nuclear) The surface, consisting of two layers of membrane, that encloses the nucleus of eukaryotic cells. 2) (virus) A structure which is present on the outside of some viruses (exterior to the capsid).

**ENVIRONMENT:** Water, air, and land, and the interrelationship that exists among and between water, air and land and all living things. The total living and nonliving aspects of an organism's internal and external surroundings.

**ENZYME:** A protein, on the surface of which are chemical groups so arranged as to make the enzyme a catalyst for a chemical reaction.

**EPICOTYL:** A portion of the axis of a plant embryo above the point of attachment of the cotyledons; forms most of the shoot.

**EPIDERMIS:** The outermost portion of the skin or body wall of an animal.

**EPINEPHRINE:** A hormone produced as a response to stress; also called adrenaline.

**EPIPHYTE:** A plant that nourishes itself but grows on the surface of another plant for support, usually on the branches or trunks of tropical trees.

**EPISOME:** Genetic element at times free in the cytoplasm, at other times integrated into a chromosome.

**EPISTASIS:** A phenomenon in which one gene alters the expression of another gene that is independently inherited.

**EPITHELIUM:** An animal tissue that forms the covering or lining of all free body surfaces, both external and internal.

**EPITOPE:** A localized region on the surface of an antigen that is chemically recognized by antibodies; also called antigenic determinant.

**EQUATION:** A precise representation of the outcome of a chemical reaction, showing the reactants and products, as well as the proportions of each.

**EQUILIBRIUM:** In a reversible reaction, the point at which the rate of the forward reaction equals that of the reverse reaction. (constant) At equilibrium, the ratio of products to reactants. (potential) The membrane potential for a given ion at which the voltage exactly balances the chemical diffusion gradient for that ion.

**ERNST MAYR:** Formulated the biological species concept.

**ERYTHROCYTE:** A red blood corpuscle.

**ESOPHAGUS:** An anterior part of the digestive tract; in mammals it leads from the pharynx to the stomach.

**ESSENTIAL:** 1) An amino or fatty acid which is required in the diet of an animal because it cannot be synthesized. 2) A chemical element required for a plant to grow from a seed and complete the life cycle.

**ESTIVATION:** A physiological state characterized by slow metabolism and inactivity, which permits survival during long periods of elevated temperature and diminished water supplies.

**ESTRADIOL:** 1,3,5(10)-estratriene- 3,17 beta-diol C<sub>18</sub>H<sub>24</sub>O<sub>2</sub>. This is the natural hormone - present in pure form in the urine of pregnant mares and in the ovaries of pigs.

**ESTROGEN:** Any of a group of vertebrate female sex hormones.

**ESTROUS CYCLE:** In female mammals, the higher primates excepted, a recurrent series of physiological and behavioral changes connected with reproduction.

**ESTRUS:** The limited period of heat or sexual receptivity that occurs around ovulation in female mammals having estrous cycles.

**ESTUARY:** That portion of a river that is close enough to the sea to be influenced by marine tides.

**ETHYLENE:** The only gaseous plant hormone, responsible for fruit ripening, growth inhibition, leaf abscission, and aging.

**EUBACTERIA:** The lineage of prokaryotes that includes the cyanobacteria and all other contemporary bacteria except archaeabacteria.

**EUCHROMATIN:** The more open, unraveled form of eukaryotic chromatin, which is available for transcription.

**EUCOELOMATE:** An animal whose body cavity is completely lined by mesoderm, the layers of which connect dorsally and ventrally to form mesenteries.

**EUGLENA:** Euglena are common protists, of the class Euglenoidea of the phylum Euglenophyta. Currently, over 1000 species of Euglena have been described. Marin et al. (2003) revised the genus so and including several species without chloroplasts, formerly classified as Astasia and Khawinea. Euglena sometimes can be considered to have both plant and animal features. Euglena gracilis has a long hair-like thing that stretches from its body. You need a very powerful microscope to see it. This is called a flagellum, and the euglena uses it to swim. It also has a red eyespot. Euglena gracilis uses its eyespot to locate light. Without light, it cannot use its chloroplasts to make itself food.

**EUKARYOTE:** A life form comprised of one or more cells containing a nucleus and membrane - bound organelles. Included are members of the Kingdoms Protista, Fungi, Plantae and Animalia.

**EUMETAZOA:** Members of the subkingdom that includes all animals except sponges.

**EUTROPHIC:** A highly productive condition in aquatic environments which owes to excessive concentrations of nutrients which support the growth of primary producers.

**EVAGINATED:** Folded or protruding outward.

**EVAPORATIVE COOLING:** The property of a liquid whereby the surface becomes cooler during evaporation, owing to the loss of highly kinetic molecules to the gaseous state.

**EVERSIBLE:** Capable of being turned inside out.

**EVOLUTION:** A theory that all of the changes that have transformed life on earth from its earliest beginnings to the diversity that characterizes it today. As used in biology, the term evolution means descent with change. See Intelligent Design.

**EXCITABLE CELLS:** A cell, such as a neuron or a muscle cell that can use changes in its membrane potential to conduct signals.

**EXCITATORY POSTSYNAPTIC POTENTIAL:** An electrical change (depolarization) in the membrane of a postsynaptic neuron caused by the binding of an excitatory neurotransmitter from a presynaptic cell to a postsynaptic receptor. This phenomenon facilitates generation of an action potential in the PSP.

**EXCRETION:** Release of materials which arise in the body due to metabolism (e.g., CO<sub>2</sub>, NH<sub>3</sub>, H<sub>2</sub>O).

**EXERGONIC:** A phenomenon which involves the release of energy.

**EXOCYTOSIS:** A process by which a vesicle within a cell fuses with the plasma membrane and releases its contents to the outside.

**EXON:** A part of a primary transcript (and the corresponding part of a gene) that is ultimately either translated (in the case of mRNA) or utilized in a final product, such as tRNA.

**EXOSKELETON:** An external skeleton, characteristic of members of the phylum, Arthropoda.

**EXOTHERMIC:** A process or reaction that is accompanied by the creation of heat.

**EXOTOXIN:** A toxic protein secreted by a bacterial cell that produces specific symptoms even in the absence of the bacterium.

**EXPONENTIAL:** (population growth) The geometric increase of a population as it grows in an ideal, unlimited environment.

**EXTRAEMBRYONIC MEMBRANES:** Four membranes (yolk sac, amnion, chorion, allantois) that support the developing embryo in reptiles, birds, and mammals.

**EXTRINSIC:** External to, not a basic part of; as in extrinsic isolating mechanism.

**F:** The chemical symbol of Fluorine.

**F PLASMID:** The fertility factor in bacteria, a plasmid that confers the ability to form pili for conjugation and associated functions required for transfer of DNA from donor to recipient.

**F1 GENERATION:** The first filial or hybrid offspring in a genetic cross-fertilization.

**F2 GENERATION:** Offspring resulting from interbreeding of the hybrid F1 generation.

**FACILITATED DIFFUSION:** Passive movement through a membrane involving a specific carrier protein; does not proceed against a concentration gradient.

**FACULTATIVE:** An organism which exhibits the capability of changing from one habit or metabolic pathway to another, when conditions warrant. (anaerobe) An organism that makes ATP by aerobic respiration if oxygen is present but that switches to fermentation under anaerobic conditions.

**FAT:** A biological compound consisting of three fatty acids linked to one glycerol molecule.

**FATE MAP:** A means of tracing the fates of cells during embryonic development.

**FATTY ACID:** A long carbon chain carboxylic acid. Fatty acids vary in length and in the number and location of double bonds; three fatty acids linked to a glycerol molecule form fat.

**FAUNA:** The animals of a given area or period.

**FEATURE DETECTOR:** A circuit in the nervous system that responds to a specific type of feature, such as a vertically moving spot or a particular auditory time delay.

**FECAL COLIFORM:** A group of bacteria that may indicate the presence of human or animal fecal matter in water. Total coliform, fecal coliform, and E. coli are all indicators of drinking water quality. The total coliform group is a large collection of different kinds of bacteria. Fecal coliforms are types of total coliform that mostly exist in feces. E. coli is a sub-group of fecal coliform. When a water sample is sent to a lab, it is tested for total coliform. If total coliform is present, the sample will also be tested for either fecal coliform or E. coli, depending on the lab testing method.

**FECES:** Indigestible wastes discharged from the digestive tract.

**FEEDBACK:** The process by which a control mechanism is regulated through the very effects it brings about. Positive feedback is when the effect is amplified; negative feedback is when the effect tends toward restoration of the original condition. Feedback inhibition is a method of metabolic control in which the end-product of a metabolic pathway acts as an inhibitor of an enzyme within that pathway.

**FERMENTATION:** Anaerobic production of alcohol, lactic acid or similar compounds from carbohydrate resulting from glycolysis.

**FERRIC CHLORIDE:** An iron salt commonly used as a coagulant. Chemical formula is FeCl<sub>3</sub>.

**FIBRIN:** The activated form of the blood: clotting protein fibrinogen, which aggregates into threads that form the fabric of the clot.

**FIBROBLAST:** A type of cell in loose connective tissue that secretes the protein ingredients of the extracellular fibers.

**FIBRONECTINS:** A family of extracellular glycoproteins that helps embryonic cells adhere to their substrate as they migrate.

**FILTER:** A device utilizing a granular material, woven cloth or other medium to remove pollutants from water, wastewater or air.

**FILTER AID:** A polymer or other material added to improve the effectiveness of the filtration process.

**FILTER CAKE:** The layer of solids that is retained on the surface of a filter.

**FILTER CLOGGING:** An inability to meet demand may occur when filters are clogging.

**FILTER PRESS:** A dewatering device where sludge is pumped onto a filtering medium and water is forced out of the sludge, resulting in a "cake".

**FILTRATE:** Liquid remaining after removal of solids with filtration.

**FILTRATION RATE:** A measurement of the volume of water applied to a filter per unit of surface area in a given period of time.

**FITNESS:** The extent to which an individual passes on its genes to the next generation. Relative fitness is the number of offspring of an individual compared to the mean.

**FIXATION:** 1) Conversion of a substance into a biologically more usable form, for example, CO<sub>2</sub> fixation during photosynthesis and N<sub>2</sub> fixation. 2) Process of treating living tissue for microscopic examination.

**FIXED ACTION PATTERN (FAP):** A highly: stereotyped behavior that is innate and must be carried to completion once initiated.

**FLACCID:** Limp; walled cells are flaccid in isotonic surroundings, where there is no tendency for water to enter.

**FLAGELLIN:** The protein from which prokaryotic flagella are constructed.

**FLAGELLUM:** A long whip-like appendage that propels cells during locomotion in liquid solutions. The prokaryote flagellum is comprised of a protein, flagellin. The eukaryote flagellum is longer than a cilium, but as a similar internal structure of microtubules in a "9 + 2" arrangement.

**FLAME CELL:** A flagellated cell associated with the simplest tubular excretory system, present in flatworms: it acts to directly regulate the contents of the extracellular fluid.

**FLOCCULATION:** The process of bringing together destabilized or coagulated particles to form larger masses that can be settled and/or filtered out of the water being treated. Conventional coagulation-flocculation-sedimentation practices are essential pretreatments for many water purification systems—especially filtration treatments. These processes agglomerate suspended solids together into larger bodies so that physical filtration processes can more easily remove them. Particulate removal by these methods makes later filtering processes far more effective. The process is often followed by gravity separation (sedimentation or flotation) and is always followed by filtration. A chemical coagulant, such as iron salts, aluminum salts, or polymers, is added to source water to facilitate bonding among particulates. Coagulants work by creating a chemical reaction and eliminating the negative charges that cause particles to repel each other. The coagulant-source water mixture is then slowly stirred in a process known as flocculation. This water churning induces particles to collide and clump together into larger and more easily removable clots, or "flocs." The process requires chemical knowledge of source water characteristics to ensure that an effective coagulant mix is employed. Improper coagulants make these treatment methods ineffective. The ultimate effectiveness of coagulation/flocculation is also determined by the efficiency of the filtering process with which it is paired.

**FLOCCULANTS:** Flocculants, or flocculating agents, are chemicals that promote flocculation by causing colloids and other suspended particles in liquids to aggregate, forming a floc. Flocculants are used in water treatment processes to improve the sedimentation or filterability of small particles. For example, a flocculant may be used in swimming pool or drinking water filtration to aid removal of microscopic particles which would otherwise cause the water to be cloudy and which would be difficult or impossible to remove by filtration alone. Many flocculants are multivalent cations such as aluminum, iron, calcium or magnesium. These positively charged molecules interact with negatively charged particles and molecules to reduce the barriers to aggregation. In addition, many of these chemicals, under appropriate pH and other conditions such as temperature and salinity, react with water to form insoluble hydroxides which, upon precipitating, link together to form long chains or meshes, physically trapping small particles into the larger floc. Long-chain polymer flocculants, such as modified polyacrylamides, are manufactured and sold by the flocculant producing business. These can be supplied in dry or liquid form for use in the flocculation process. The most common liquid polyacrylamide is supplied as an emulsion with 10-40 % actives and the rest is a carrier fluid, surfactants and latex. Emulsion polymers require activation to invert the emulsion and allow the electrolyte groups to be exposed.

The following chemicals are used as flocculants:

- \* alum
- \* aluminum chlorohydrate
- \* aluminum sulfate
- \* calcium oxide
- \* iron(III) chloride
- \* iron(II) sulfate
- \* polyacrylamide
- \* sodium aluminate
- \* sodium silicate

**FLOC SHEARING:** Likely to happen to large floc particles when they reach the flocculation process.

**FLOCCULATION BASIN:** A compartmentalized basin with a reduction of speed in each compartment. This set-up or basin will give the best overall results.

**FLOOD RIM:** The point of an object where the water would run over the edge of something and begin to cause a flood.

**FLORA:** The plants of a given area or period.

**FLOW CYTOMETER:** A particle-sorting instrument capable of counting protozoa.

**FLOW MUST BE MEASURED:** A recorder that measures flow is most likely to be located in a central location.

**FLUID FEEDER:** An animal that lives by sucking nutrient-rich fluids from another living organism.

**FLUID MOSAIC MODEL:** The currently accepted model of cell membrane structure, which envisions the membrane as a mosaic of individually inserted protein molecules drifting laterally in a fluid bilayer of phospholipids.

**FLUX:** The term flux describes the rate of water flow through a semipermeable membrane. When the water flux decreases through a semipermeable membrane, it means that the mineral concentration of the water is increasing.

**FLY ASH:** The noncombustible particles in flue gas. Often used as a body feed or solidification chemical.

**FOLLICLE STIMULATING HORMONE (FSH):** A gonadotropic hormone of the anterior pituitary that stimulates growth of follicles in the ovaries of females and function of the seminiferous tubules in males.

**FOLLICLE:** A jacket of cells around an egg cell in an ovary.

**FOOD CHAIN:** Sequence of organisms, including producers, consumers, and decomposers, through which energy and materials may move in a community.

**FOOD WEB:** The elaborate, interconnected feeding relationships in an ecosystem.

**FOOT CANDLE:** Unit of illumination; the illumination of a surface produced by one standard candle at a distance of one foot.

**FORMULA:** A precise representation of the structure of a molecule or ion, showing the proportion of atoms which comprise the material.

**FOUNDER EFFECT:** The difference between the gene pool of a population as a whole and that of a newly isolated population of the same species.

**FRACTIONATION:** An experimental technique which involves separation of parts of living tissue from one another using centrifugation.

**FRAGMENTATION:** A mechanism of asexual reproduction in which the parent plant or animal separates into parts that reform whole organisms.

**FRAMESHIFT MUTATION:** A mutation occurring when the number of nucleotides inserted or deleted is not a multiple of 3, thus resulting in improper grouping into codons.

**FREE CHLORINE:** In disinfection, chlorine is used in the form of free chlorine or as hypochlorite ion.

**FREE CHLORINE RESIDUAL:** Regardless of whether pre-chlorination is practiced or not, a free chlorine residual of at least 10 mg/L should be maintained in the clear well or distribution reservoir immediately downstream from the point of post-chlorination. The reason for chlorinating past the breakpoint is to provide protection in case of backflow.

**FREE ENERGY OF ACTIVATION:** See Activation energy.

**FREE ENERGY:** Usable energy in a chemical system; energy available for producing change.

**FREE OIL:** Non-emulsified oil that separates from water, in a given period of time.

**FREQUENCY DEPENDENT SELECTION:** A decline in the reproductive success of a morph resulting from the morph's phenotype becoming too common in a population; a cause of balanced polymorphism in populations.

**FUNCTIONAL GROUP:** One of several groups of atoms commonly found in organic molecules. A functional group contributes somewhat predictable properties to the molecules which possess them.

**FUNDAMENTAL NICHE:** The total resources an organism is theoretically capable of utilizing.

**G:** (protein) A membrane protein that serves as an intermediary between hormone receptors and the enzyme adenylyl cyclase, which converts ATP to cAMP in the second messenger system in non-steroid hormone action. Depending on the system, G proteins either increase or decrease cAMP production.

**G1 PHASE:** The first growth phase of the cell cycle, consisting of the portion of interphase before DNA synthesis is initiated.

**G2 PHASE:** The second growth phase of the cell cycle, consisting of the portion of interphase after DNA synthesis but before mitosis.

**GAIA HYPOTHESIS:** An idea, first formulated by James E. Lovelock in 1979, which suggests that the biosphere of the earth exists as a "superorganism" which exhibits homeostatic self- regulation of the environment-biota global system.

**GAMETANGIUM:** The reproductive organ of bryophytes, consisting of the male antheridium and female archegonium; a multi-chambered jacket of sterile cells in which gametes are formed.

**GAMETE:** A sexual reproductive cell that must usually fuse with another such cell before development begins; an egg or sperm.

**GAMETOPHYTE:** A haploid plant that can produce gametes.

**GANGLION:** A structure containing a group of cell bodies of neurons.

**GAP JUNCTION:** A narrow gap between plasma membranes of two animal cells, spanned by protein channels. They allow chemical substances or electrical signals to pass from cell to cell.

**GASTRULA:** A two-layered, later three-layered, animal embryonic stage.

**GASTRULATION:** The process by which a blastula develops into a gastrula, usually by an involution of cells.

**GATED ION CHANNEL:** A membrane channel that can open or close in response to a signal, generally a change in the electrostatic gradient or the binding of a hormone, transmitter, or other molecular signal.

**GEL ELECTROPHORESIS:** In general, electrophoresis is a laboratory technique used to separate macromolecules on the basis of electric charge and size; the technique involves application of an electric field to a population of macromolecules which disperse according to their electric mobilities. In gel electrophoresis, the porous medium through which the macromolecules move is a gel.

**GEL:** Colloid in which the suspended particles form a relatively orderly arrangement.

**GENE:** The hereditary determinant of a specified characteristic of an individual; specific sequences of nucleotides in DNA.

**GENE AMPLIFICATION:** Any of the strategies that give rise to multiple copies of certain genes, thus facilitating the rapid synthesis of a product (such as rRNA for ribosomes) for which the demand is great.

**GENE CLONING:** Formation by a bacterium, carrying foreign genes in a recombinant plasmid, of a clone of identical cells containing the replicated foreign genes.

**GENE DELIVERY:** This is a general term for the introduction of new genetic elements into the genomes of living cells. The delivery problem is essentially conditioned by the fact that the new genetic elements are usually large, and by the presence of the outer cell membrane and the nuclear membrane acting as barriers to incorporation of the new DNA into the genome already present in the nucleus. Viruses possess various natural biochemical methods for achieving gene delivery; artificial gene delivery is one of the essential problems of "genetic engineering". The most important barrier is apparently the outer cell membrane, which is essentially a lipid barrier, and introduction of any large complex into the cell requires a fusion of one kind or another with this membrane. Liposomes, which consist of lipid membranes themselves, and which can fuse with outer cell membranes, are thus potential vehicles for delivery of many substances, including DNA.

**GENE FLOW:** The movement of genes from one part of a population to another, or from one population to another, via gametes.

**GENE POOL:** The sum total of all the genes of all the individuals in a population.

**GENE REGULATION:** Any of the strategies by which the rate of expression of a gene can be regulated, as by controlling the rate of transcription.

**GENETIC DRIFT:** Change in the gene pool as a result of chance and not as a result of selection, mutation, or migration.

**GENETIC RECOMBINATION:** The general term for the production of offspring that combine traits of the two parents.

**GENETICS:** The science of heredity; the study of heritable information.

**GENOME:** The cell's total complement of DNA.

**GENOMIC EQUIVALENCE:** The presence of all of an organism's genes in all of its cells.

**GENOMIC IMPRINTING:** The parental effect on gene expression. Identical alleles may have different effects on offspring depending on whether they arrive in the zygote via the ovum or via the sperm.

**GENOMIC LIBRARY:** A set of thousands of DNA segments from a genome, each carried by a plasmid or phage.

**GENOTYPE:** The particular combination of genes present in the cells of an individual.

**GENUS:** A taxonomic category above the species level, designated by the first word of a species' binomial Latin name.

**GIARDIA LAMBLIA:** Giardia lamblia (synonymous with *Lamblia intestinalis* and *Giardia duodenalis*) is a flagellated protozoan parasite that colonizes and reproduces in the small intestine, causing giardiasis. The giardia parasite attaches to the epithelium by a ventral adhesive disc, and reproduces via binary fission. Giardiasis does not spread via the bloodstream, nor does it spread to other parts of the gastro-intestinal tract, but remains confined to the lumen of the small intestine. Giardia trophozoites absorb their nutrients from the lumen of the small intestine, and are anaerobes.

**GIARDIASIS, HEPATITIS OR TYPHOID:** Diseases that may be transmitted through the contamination of a water supply, but not AIDS.

**GIS – GRAPHIC INFORMATION SYSTEM:** Detailed information about the physical locations of structures such as pipes, valves, and manholes within geographic areas with the use of satellites.

**GLIAL CELL:** A non-conducting cell of the nervous system that provides support, insulation, and protection for the neurons.

**GLIDING:** Rod-shaped, gram-negative, mostly aerobic; glide on secreted slimy substances; form colonies, frequently with complex fruiting structures.

**GLOMERULUS:** A capillary bed within Bowman's capsule of the nephron; the site of ultrafiltration.

**GLUCOSE:** A six carbon sugar which plays a central role in cellular metabolism.

**GLYCOCALYX:** The layer of protein and carbohydrates just outside the plasma membrane of an animal cell; in general, the proteins are anchored in the membrane, and the carbohydrates are bound to the proteins.

**GLYCOGEN:** A long, branched polymer of glucose subunits that is stored in the muscles and liver of animals and is metabolized as a source of energy.

**GLYCOLYSIS:** A metabolic pathway which occurs in the cytoplasm of cells and during which glucose is oxidized anaerobically to form pyruvic acid.

**GLYCOPROTEIN:** A protein with covalently linked sugar residues. The sugars may be bound to OH side chains of the polypeptide (O: linked) or the amide nitrogen of asparagine side chains (N: linked).

**GLYCOSIDIC:** A type of bond which links monosaccharide subunits together in di- or polysaccharides.

**GLYOXYSOME:** A type of microbody found in plants, in which stored lipids are converted to carbohydrates.

**GOLGI APPARATUS:** A system of concentrically folded membranes found in the cytoplasm of eukaryotic cells. Plays a role in the production and release of secretory materials such as the digestive enzymes manufactured in the pancreas.

**GONADOTROPIN:** Refers to a member of a group of hormones capable of promoting growth and function of the gonads. Includes hormones such as follicle stimulating hormone (FSH) and luteinizing hormone (LH) which are stimulatory to the gonads.

**GOOD CONTACT TIME, pH and LOW TURBIDITY:** These are factors that are important in providing good disinfection when using chlorine.

**GPM:** Gallons per minute.

**GRAB SAMPLE:** A sample which is taken from a water or wastestream on a one-time basis with no regard to the flow of the water or wastestream and without consideration of time. A single grab sample should be taken over a period of time not to exceed 15 minutes.

**GRAB SAMPLE:** A single water or wastewater sample taken at a time and place representative of total discharge.

**GRADED POTENTIAL:** A local voltage change in a neuron membrane induced by stimulation of a neuron, with strength proportional to the strength of the stimulus and lasting about a millisecond.

**GRANUM:** A stack-like grouping of photosynthetic membranes in a chloroplast

**GRAVITY BELT THICKENER:** A sludge dewatering device utilizing a filter belt to promote gravity drainage of water. Usually precedes additional dewatering treatment.

**GRAVITY FILTER:** A filter that operates at atmospheric pressure.

**GRAVITY THICKENING:** A sedimentation basin designed to operate at high solids loading rates.

**GRAVITROPISM:** A response of a plant or animal in response to gravity.

**GREENHOUS EFFECT:** The warming of the Earth due to atmospheric accumulation of carbon dioxide which absorbs infrared radiation and slows its escape from the irradiated Earth.

**GREGOR MENDEL:** The first to make quantitative observations of the patterns of inheritance and proposing plausible explanations for them.

**GROWTH FACTOR:** A protein that must be present in a cell's environment for its normal growth and development.

**GT:** Represents (Detention time) x (mixing intensity) in flocculation.

**GUARD CELL:** A specialized epidermal cell that regulates the size of stoma of a leaf.

**GYMNOSPERM:** A vascular plant that bears naked seeds not enclosed in any specialized chambers.

**H2SO4:** The molecular formula of Sulfuric acid.

**HABIT:** In biology, the characteristic form or mode of growth of an organism.

**HABITAT:** The kind of place where a given organism normally lives.

**HABITUATION:** The process that results in a long-lasting decline in the receptiveness of interneurons to the input from sensory neurons or other interneurons (sensitization, adaptation).

**HALF:** The average amount of time it takes for one-half of a specified quantity of a substance to decay or disappear.

**HALIDES:** A halide is a binary compound, of which one part is a halogen atom and the other part is an element or radical that is less electronegative than the halogen, to make a fluoride, chloride, bromide, iodide, or astatide compound. Many salts are halides. All Group 1 metals form halides with the halogens and they are white solids. A halide ion is a halogen atom bearing a negative charge. The halide anions are fluoride (F), chloride (Cl), bromide (Br), iodide (I) and astatide (At). Such ions are present in all ionic halide salts.

**HALOACETIC ACIDS:** Haloacetic acids are carboxylic acids in which a halogen atom takes the place of a hydrogen atom in acetic acid. Thus, in a monohaloacetic acid, a single halogen would replace a hydrogen atom. For example, chloroacetic acid would have the structural formula  $\text{CH}_2\text{ClCO}_2\text{H}$ . In the same manner, in dichloroacetic acid two chlorine atoms would take the place of two hydrogen atoms ( $\text{CHCl}_2\text{CO}_2\text{H}$ ).

**HAPLOID:** The condition of having only one kind of a given type of chromosome.

**HARD WATER:** Hard water causes a buildup of scale in household hot water heaters. Hard water is a type of water that has high mineral content (in contrast with soft water). Hard water primarily consists of calcium ( $\text{Ca}^{2+}$ ), and magnesium ( $\text{Mg}^{2+}$ ) metal cations, and sometimes other dissolved compounds such as bicarbonates and sulfates. Calcium usually enters the water as either calcium carbonate ( $\text{CaCO}_3$ ), in the form of limestone and chalk, or calcium sulfate ( $\text{CaSO}_4$ ), in the form of other mineral deposits. The predominant source of magnesium is dolomite ( $\text{CaMg}(\text{CO}_3)_2$ ). Hard water is generally not harmful. The simplest way to determine the hardness of water is the lather/froth test: soap or toothpaste, when agitated, lathers easily in soft water but not in hard water. More exact measurements of hardness can be obtained through a wet titration. The total water 'hardness' (including both  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$  ions) is read as parts per million or weight/volume (mg/L) of calcium carbonate ( $\text{CaCO}_3$ ) in the water. Although water hardness usually only measures the total concentrations of calcium and magnesium (the two most prevalent, divalent metal ions), iron, aluminum, and manganese may also be present at elevated levels in some geographical locations.

**HARDNESS:** A measure of the amount of calcium and magnesium salts in water. More calcium and magnesium lead to greater hardness. The term "hardness" comes from the fact that it is hard to get soap suds from soap or detergents in hard water. This happens because calcium and magnesium react strongly with negatively-charged chemicals like soap to form insoluble compounds.

**HARDY-WEINBERG THEOREM:** An axiom maintaining that the sexual shuffling of genes alone cannot alter the overall genetic makeup of a population.

**HAUSTORIUM:** In parasitic fungi, a nutrient-absorbing hyphal tip that penetrates the tissues of the host but remains outside the host cell membranes.

**HAVERSIAN SYSTEM:** One of many structural units of vertebrate bone, consisting of concentric layers of mineralized bone matrix surrounding lacunae, which contain osteocytes, and a central canal, which contains blood vessels and nerves.

**HAZARDS OF POLYMERS:** Slippery and difficult to clean-up are the most common hazards associated with the use of polymers in a water treatment plant.

**HEAD:** The measure of the pressure of water expressed in feet of height of water. 1 PSI = 2.31 feet of water or 1 foot of head equals about a half a pound of pressure or .433 PSI. There are various types of heads of water depending upon what is being measured. Static (water at rest) and Residual (water at flow conditions).

**HEADWORKS:** The facility at the "head" of the water source where water is first treated and routed into the distribution system.

**HEALTH ADVISORY:** An EPA document that provides guidance and information on contaminants that can affect human health and that may occur in drinking water, but which the EPA does not currently regulate in drinking water.

**HEAT OF VAPORIZATION:** The amount of energy absorbed by a substance when it changes state to a gas. Water absorbs approximately 580 calories per gram when it changes from liquid water to water vapor.

**HEAT:** The total amount of kinetic energy due to molecular motion in a body of matter. Heat is energy in its most random form.

**HELPER T CELL:** A type of T cell that is required by some B cells to help them make antibodies or that helps other T cells respond to antigens or secrete lymphokines or interleukins.

**HEMAGGLUTININ:** A surface antigen on influenza viruses which controls infectivity by associating with receptors on host erythrocytes or other cells.

**HEMATOPOIESIS:** The formation of blood.

**HEMATOPOIETIC STEM CELLS:** Cells found in the bone marrow of adult mammals which give rise to erythroid stem cells, lymphoid stem cells, and myeloid stem cells. Such cells give rise to erythrocytes and a variety of types of lymphocytes and leucocytes.

**HEMOGLOBIN:** An iron-containing respiratory pigment found in many organisms.

**HEMOLYMPH:** In invertebrates with open circulatory systems, the body fluid that bathes tissues.

**HEMOPHILIA:** A genetic disease resulting from an abnormal sex-linked recessive gene, characterized by excessive bleeding following injury.

**HEPATIC:** Pertaining to the liver.

**HEREDITY:** A biological phenomenon whereby characteristics are transmitted from one generation to another by virtue of chemicals (i.e. DNA) transferred during sexual or asexual reproduction.

**HERPESVIRUS:** A double stranded DNA virus with an enveloped, icosahedral capsid.

**HERTZ:** The term used to describe the frequency of cycles in an alternating current (AC) circuit. A unit of frequency equal to one cycle per second.

**HETEROCHROMATIN:** Non-transcribed eukaryotic chromatin that is so highly compacted that it is visible with a light microscope during interphase.

**HETEROCHRONY:** Evolutionary changes in the timing or rate of development.

**HETEROCYST:** A specialized cell that engages in nitrogen fixation on some filamentous cyanobacteria.

**HETEROGAMY:** The condition of producing gametes of two different types (contrast with isogamy).

**HETEROMORPHIC:** A condition in the life cycle of all modern plants in which the sporophyte and gametophyte generations differ in morphology.

**HETEROSPOROUS:** Referring to plants in which the sporophyte produces two kinds of spores that develop into unisexual gametophytes, either male or female.

**HETEROTROPH:** An organism dependent on external sources of organic compounds as a means of obtaining energy and/or materials. Such an organism requires carbon ("food") from its environment in an organic form. (synonym-organotroph).

**HETEROTROPHIC PLATE COUNT:** A test performed on drinking water to determine the total number of all types of bacteria in the water.

**HETEROZYGOTE ADVANTAGE:** A mechanism that preserves variation in eukaryotic gene pools by conferring greater reproductive success on heterozygotes over individuals homozygous for any one of the associated alleles.

**HETEROZYGOUS:** The condition whereby two different alleles of the gene are present within the same cell.

**HF:** The molecular formula of Hydrofluoric acid.

**HIGH TURBIDITY CAUSING INCREASED CHLORINE DEMAND:** May occur or be caused by the inadequate disinfection of water.

**HISTAMINE:** A substance released by injured cells that causes blood vessels to dilate during an inflammatory response.

**HISTOLOGY:** The study of tissues.

**HISTONE:** A type of protein characteristically associated with the chromosomes of eukaryotes.

**HIV-1:** Acute human immunodeficiency virus type 1 is the subtype of HIV (human immune deficiency virus) that causes most cases of AIDS in the Western Hemisphere, Europe, and Central, South, and East Africa. HIV is a retrovirus (subclass lentivirus), and retroviruses are single-stranded RNA viruses that have an enzyme called reverse transcriptase. With this enzyme the viral RNA is used as a template to produce viral DNA from cellular material. This DNA is then incorporated into the host cell's genome, where it codes for the synthesis of viral components. An HIV-1 infection should be distinguished from AIDS. Acquired immunodeficiency syndrome (AIDS) is a secondary immunodeficiency syndrome resulting from HIV infection and characterized by opportunistic infections, malignancies, neurologic dysfunction, and a variety of other syndromes.

**HOLOBLASTIC:** A type of cleavage in which there is complete division of the egg, as in eggs having little yolk (sea urchin) or a moderate amount of yolk (frog).

**HOME RANGE:** An area within which an animal tends to confine all or nearly all its activities for a long period of time.

**HOMEobox:** Specific sequences of DNA that regulate patterns of differentiation during development of an organism.

**HOMEOSTASIS:** A phenomenon whereby a state or process (for example, within an organism) is regulated automatically despite the tendency for fluctuations to occur.

**HOMEOTHEMIC:** Capable of regulation of constancy with respect to temperature.

**HOMEOTIC GENES:** Genes that control the overall body plan of animals by controlling the developmental fate of groups of cells.

**HOMEOTIC:** (mutation) A mutation in genes regulated by positional information that results in the abnormal substitution of one type of body part in place of another.

**HOMOLOGOUS CHROMOSOMES:** Chromosomes bearing genes for the same characters.

**HOMOLOGOUS STRUCTURES:** Characters in different species which were inherited from a common ancestor and thus share a similar ontogenetic pattern.

**HOMOLOGY:** Similarity in characteristics resulting from a shared ancestry.

**HOMOPLASY:** The presence in several species of a trait not present in their most common ancestor. Can result from convergent evolution, reverse evolution, or parallel evolution.

**HOMOSPOROUS:** Referring to plants in which a single type of spore develops into a bisexual gametophyte having both male and female sex organs.

**HOMOZYGOUS:** Having two copies of the same allele of a given gene.

**HORMONE:** A control chemical secreted in one part of the body that affects other parts of the body.

**HOST RANGE:** The limited number of host species, tissues, or cells that a parasite (including viruses and bacteria) can infect.

**HUMORAL IMMUNITY:** The type of immunity that fights bacteria and viruses in body fluids with antibodies that circulate in blood plasma and lymph, fluids formerly called humors.

**HYBIRD VIGOR:** Increased vitality (compared to that of either parent stock) in the hybrid offspring of two different, inbred parents.

**HYBIRD:** In evolutionary biology, a cross between two species. In genetics, a cross between two genetic types.

**HYBIRDIZATION:** The process whereby a hybrid results from interbreeding two species; 2) DNA hybridization is the comparison of whole genomes of two species by estimating the extent of hydrogen bonding that occurs between single-stranded DNA obtained from the two species.

**HYBRIDOMA:** A hybrid cell that produces monoclonal antibodies in culture, formed by the fusion of a myeloma cell with a normal antibody-producing lymphocyte.

**HYDRATED LIME:** The calcium hydroxide product that results from mixing quicklime with water. Chemical formula is  $\text{CaOH}_2$ .

**HYDRATION SHELL:** A "covering" of water molecules which surrounds polar or charged substances in aqueous solutions. The association is due to the charged regions of the polar water molecules themselves.

**HYDRIDES:** Hydride is the name given to the negative ion of hydrogen, H. Although this ion does not exist except in extraordinary conditions, the term hydride is widely applied to describe compounds of hydrogen with other elements, particularly those of groups 1–16. The variety of compounds formed by hydrogen is vast, arguably greater than that of any other element. Various metal hydrides are currently being studied for use as a means of hydrogen storage in fuel cell-powered electric cars and batteries. They also have important uses in organic chemistry as powerful reducing agents, and many promising uses in hydrogen economy.

Every element of the periodic table (except some noble gases) forms one or more hydrides. These compounds may be classified into three main types by the predominant nature of their bonding:

- \* Saline hydrides, which have significant ionic character,
- \* Covalent hydrides, which include the hydrocarbons and many other compounds, and
- \* Interstitial hydrides, which may be described as having metallic bonding.

**HYDROCARBON:** Any compound made of only carbon and hydrogen.

**HYDROCHLORIC AND HYPOCHLOROUS ACIDS:** The compounds that are formed in water when chlorine gas is introduced.

**HYDROFLUOSILIC ACID:** ( $\text{H}_2\text{SiF}_6$ ) a clear, fuming corrosive liquid with a pH ranging from 1 to 1.5. Used in water treatment to fluoridate drinking water.

**HYDROGEN BOND:** A type of bond formed when the partially positive hydrogen atom of a polar covalent bond in one molecule is attracted to the partially negative atom of a polar covalent bond in another.

**HYDROGEN SULFIDE OR CHLORINE GAS:** These chemicals can cause olfactory fatigue.

**HYDROGEN ION:** A single proton with a charge of +1. The dissociation of a water molecule (H<sub>2</sub>O) leads to the generation of a hydroxide ion (OH<sup>-</sup>) and a hydrogen ion (H<sup>+</sup>).

**HYDROGEN SULFIDE:** A toxic gas formed by the anaerobic decomposition of organic matter. Chemical formula is H<sub>2</sub>S.

**HYDROLYSIS:** The chemical reaction that breaks a covalent bond through the addition of hydrogen (from a water molecule) to the atom forming one side of the original bond, and a hydroxyl group to the atom on the other side.

**HYDROPHILIC:** Having an affinity for water.

**HYDROPHOBIC INTERACTION:** A type of weak chemical bond formed when molecules that do not mix with water coalesce to exclude the water.

**HYDROPHOBIC:** The physicochemical property whereby a substance or region of a molecule resists association with water molecules.

**HYDROSTATIC:** Pertaining to the pressure and equilibrium of fluids. A hydrostatic skeleton is a skeletal system composed of fluid held under pressure in a closed body compartment; the main skeleton of most cnidarians, flatworms, nematodes, and annelids.

**HYDROXYL GROUP:** A functional group consisting of a hydrogen atom joined to an oxygen atom by a polar covalent bond. Molecules possessing this group are soluble in water and are called alcohols.

**HYDROXYL ION:** The OH<sup>-</sup> ion.

**HYPEROSMOTIC:** A solution with a greater solute concentration than another, a hypoosmotic solution. If the two solutions are separated from one another by a membrane permeable to water, water would tend to move from the hypo- to the hyperosmotic side.

**HYPERPOLARIZATION:** An electrical state whereby the inside of the cell is made more negative relative to the outside than was the case at resting potential. A neuron membrane is hyperpolarized if the voltage is increased from the resting potential of about -70 mV, reducing the chance that a nerve impulse will be transmitted.

**HYPERTROPHY:** Abnormal enlargement, excessive growth.

**HYPHA:** A fungal filament.

**HYPOCHLORITE AND ORGANIC MATERIALS:** Heat and possibly fire may occur when hypochlorite is brought into contact with an organic material.

**HYPOCOTYL:** The portion of the axis of a plant embryo below the point of attachment of the cotyledons; forms the base of the shoot and the root.

**HYPOOSMOTIC SOLUTION:** A solution with a lesser solute concentration than another, a hyperosmotic solution. If the two solutions are separated from one another by a membrane permeable to water, water would tend to move from the hypo- to the hyperosmotic side.

**HYPOTHESIS:** A formal statement of supposition offered to explain observations. Note that a hypothesis is only useful if it can be tested. Even if correct, it is not scientifically useful if untestable.

**HYPOTHETICO-DEDUCTIVE:** A method used to test hypotheses. If deductions formulated from the hypothesis are tested and proven false, the hypothesis is rejected.

**IMAGINAL DISK:** An island of undifferentiated cells in an insect larva, which are committed (determined) to form a particular organ during metamorphosis to the adult.

**IMBIBITION:** The soaking of water into a porous material that is hydrophilic.

**IMMUNE RESPONSE:** 1) A primary immune response is the initial response to an antigen, which appears after a lag of a few days. 2) A secondary immune response is the response elicited when the animal encounters the same antigen at a later time. The secondary response is normally more rapid, of greater magnitude and of longer duration than the primary response.

**IMMUNOGLOBULINE:** The class of proteins comprising the antibodies.

**IMMUNOLOGICAL:** 1) Immunological distance is the amount of difference between two proteins as measured by the strength of the antigen: antibody reaction between them. 2) Immunological tolerance is a mechanism by which an animal does not mount an immune response to the antigenic determinants of its own macromolecules.

**IMMUNOMAGNETIC SEPARATION (IMS):** A purification procedure that uses microscopic, magnetically responsive particles coated with an antibodies targeted to react with a specific pathogen in a fluid stream. Pathogens are selectively removed from other debris using a magnetic field.

**IMPELLERS:** The semi-open or closed props or blades of a turbine pump that when rotated generate the pumping force.

**IMPERVIOUS:** Not allowing, or allowing only with great difficulty, the movement of water.

**IMPRINTING:** A type of learned behavior with a significant innate component, acquired during a limited critical period.

**IN SERIES:** Several components being connected one to the other without a bypass, requiring each component to work dependent on the one before it.

**IN SITU:** Treatment or disposal methods that do not require movement of contaminated material.

**IN VITRO FERTILIZATION:** Fertilization of ova in laboratory containers followed by artificial implantation of the early embryo in the mother's uterus.

**INCINERATION:** The process of reducing the volume of a material by burning and reducing to ash if possible.

**INCLINED PLATE SEPARATOR:** A series of parallel inclined plates that can be used to increase the efficiency of clarifiers and gravity thickeners.

**INCOMPLETE DOMINANCE:** A type of inheritance in which F1 hybrids have an appearance that is intermediate between the phenotypes of the parental varieties.

**INDETERMINATE:** 1) A type of cleavage exhibited during the embryonic development in deuterostomes, in which each cell produced by early cleavage divisions retains the capacity to develop into a complete embryo; 2) A type of growth exhibited by plants: they continue to grow as long as they live, because they always retain meristematic cells capable of undergoing mitosis.

**INDIRECT REUSE:** The beneficial use of reclaimed water into natural surface waters or groundwater.

**INDUCED FIT:** The change in shape of the active site of an enzyme so that it binds more snugly to the substrate, induced by entry of the substrate.

**INDUCTION:** 1) The ability of one group of embryonic cells to influence the development of another. 2) A method in logic which proceeds from the specific to general and develops a general statement which explains all of the observations. Commonly used to formulate scientific hypotheses.

**INDUSTRIAL MELANISM:** Melanism which has resulted from blackening of environmental surfaces (tree bark, etc.) by industrial pollution. This favors survival of melanic forms such as moths which rest on tree bark and are less likely to be seen by predators.

**INDUSTRIAL WASTEWATER:** Liquid wastes resulting from industrial processes.

**INFECTIOUS:** 1) An infectious disease is a disease caused by an infectious microbial or parasitic agent. 2) Infectious hepatitis is the former name for hepatitis A. 3) Infectious mononucleosis is an acute disease that affects many systems, caused by the Epstein: Barr virus.

**INFECTIOUS PATHOGENS/MICROBES/GERMS:** are considered disease-producing bacteria, viruses and other microorganisms.

**INFLAMMATORY RESPONSE:** A line of defense triggered by penetration of the skin or mucous membranes, in which small blood vessels in the vicinity of an injury dilate and become leakier, enhancing infiltration of leukocytes; may also be widespread in the body.

**INFLUENT:** Water or wastewater flowing into a basin or treatment plant.

**INGESTION:** A heterotrophic mode of nutrition in which other organisms or detritus are eaten whole or in pieces.

**INHIBITORY POSTSYNAPTIC POTENTIAL:** An electrical charge (hyperpolarization) in the membrane of a postsynaptic neuron caused by the binding of an inhibitory neurotransmitter from a presynaptic cell to a postsynaptic receptor.

**INITIAL PRECISION AND RECOVERY (IPR):** Four aliquots of spiking suspension analyzed to establish the ability to generate acceptable precision and accuracy. An IPR is performed prior to the first time this method is used and any time the method or instrumentation is modified.

**INNER CELL MASS:** A cluster of cells in a mammalian blastocyst that protrudes into one end of the cavity and subsequently develops into the embryo proper and some of the extraembryonic membranes.

**INORGANIC COMPOUND:** Compounds that contain no carbon or contain only carbon bound to elements other than hydrogen.

**INORGANIC CONTAMINANTS:** Mineral-based compounds such as metals, nitrates, and asbestos. These contaminants are naturally-occurring in some water, but can also get into water through farming, chemical manufacturing, and other human activities. EPA has set legal limits on 15 inorganic contaminants.

**INORGANIC IONS:** Present in all waters. Inorganic ions are essential for human health in small quantities, but in larger quantities they can cause unpleasant taste and odor or even illness. Most community water systems will commonly test for the concentrations of seven inorganic ions: nitrate, nitrite, fluoride, phosphate, sulfate, chloride, and bromide. Nitrate and nitrite can cause an illness in infants called methemoglobinemia. Fluoride is actually added to the drinking water in some public water systems to promote dental health. Phosphate, sulfate, chloride, and bromide have little direct effect on health, but high concentrations of inorganic ions can give water a salty or briny taste.

**INSOLUBLE COMPOUNDS:** are types of compounds cannot be dissolved. When iron or manganese reacts with dissolved oxygen (DO) insoluble compound are formed.

**INTAKE FACILITIES:** One of the more important considerations in the construction of intake facilities is the ease of operation and maintenance over the expected lifetime of the facility. Every intake structure must be constructed with consideration for operator safety and for cathodic protection.

**INOSITOL TRIPHOSPHATE:** The second messenger, which functions as an intermediate between certain non-steroid hormones and the third messenger, a rise in cytoplasmic  $Ca^{++}$  concentration.

**INSERTION:** A mutation involving the addition of one or more nucleotide pairs to a gene.

**INSIGHT LEARNING:** The ability of an animal to perform a correct or appropriate behavior on the first attempt in a situation with which it has had no prior experience.

**INSULIN:** The vertebrate hormone that lowers blood sugar levels by promoting the uptake of glucose by most body cells and promoting the synthesis and storage of glycogen in the liver; also stimulates protein and fat synthesis; secreted by endocrine cells of the pancreas called islets of Langerhans.

**INTEGRAL PROTEIN:** A protein of biological membranes that penetrates into or spans the membrane.

**INTERBREED:** To breed with another kind or species; hybridize.

**INTERFERON:** A chemical messenger of the immune system, produced by virus: infected cells and capable of helping other cells resist the virus.

**INTERLEUKIN:** 1: A chemical regulator (cytokine) secreted by macrophages that have ingested a pathogen or foreign molecule and have bound with a helper T cell; stimulates T cells to grow and divide and elevates body temperature. Interleukin: 2, secreted by activated T cells, stimulates helper T cells to proliferate more rapidly.

**INTERMEDIATE FILAMENT:** A component of the cytoskeleton that includes all filaments intermediate in size between microtubules and microfilaments.

**INTERNEURON:** An association neuron; a nerve cell within the central nervous system that forms synapses with sensory and motor neurons and integrates sensory input and motor output.

**INTERNODE:** The segment of a plant stem between the points where leaves are attached.

**INTERSTITIAL CELLS:** Cells scattered among the seminiferous tubules of the vertebrate testis that secrete testosterone and other androgens, the male sex hormones.

**INTERSTITIAL FLUID:** The internal environment of vertebrates consisting of the fluid filling the spaces between cells.

**INTERTIDAL ZONE:** The shallow zone of the ocean where land meets water.

**INTRINSIC RATE OF INCREASE:** The difference between number of births and number of deaths, symbolized as  $r_{max}$ ; maximum population growth rate.

**INTROGRESSION:** Transplantation of genes between species resulting from fertile hybrids mating successfully with one of the parent species.

**INTRON:** The noncoding, intervening sequence of coding region (exon) in eukaryotic genes.

**INVAGINATION:** The buckling inward of a cell layer, caused by rearrangements of microfilaments and microtubules; an important phenomenon in embryonic development.

**INVERSION:** 1) An aberration in chromosome structure resulting from an error in meiosis or from mutagens; reattachment in a reverse orientation of a chromosomal fragment to the chromosome from which the fragment originated. 2) A phenomenon which occurs during early development of sponges at which time the external ciliated cells become inward-directed.

**INVERTEBRATE:** An animal without a backbone; invertebrates make up about 95% of animal species.

**ION:** A charged chemical formed when an atom or group of atoms has more or less electrons than protons (rather than an equal number).

**ION EXCHANGE:** An effective treatment process used to remove iron and manganese in a water supply. The hardness of the source water affects the amount of water an ion exchange softener may treat before the bed requires regeneration.

**IONIC BOND:** A chemical bond due to attraction between oppositely charged ions.

**IRON:** The elements iron and manganese are undesirable in water because they cause stains and promote the growth of iron bacteria.

**IRON AND MANGANESE:** In water they can usually be detected by observing the color of the inside walls of filters and the filter media. If the raw water is pre-chlorinated, there will be black stains on the walls below the water level and a black coating over the top portion of the sand filter bed. When significant levels of dissolved oxygen are present, iron and manganese exist in an oxidized state and normally precipitate into the reservoir bottom sediments. The presence of iron and manganese in water promote the growth of Iron bacteria. Only when a water sample has been acidified then you can perform the analysis beyond the 48 hour holding time. Iron and Manganese in water may be detected by observing the color of the filter media. Maintaining a free chlorine residual and regular flushing of water mains may control the growth of iron bacteria in a water distribution system.

**IRRUPTION:** A rapid increase in population density often followed by a mass emigration.

**ISOGAMY:** A condition in which male and female gametes are morphologically indistinguishable.

**ISOMER:** Molecules consisting of the same numbers and kinds of atoms, but differing in the way in which the atoms are combined.

**ISOSMOTIC:** Solutions of equal concentration with respect to osmotic pressure.

**ISOTOPE:** An atomic form of an element, containing a different number of neutrons than another isotope. Isotopes vary from one another with respect to atomic mass.

**JUXTAGLOMERULAR APPARATUS (JGA):** Specialized tissue located near the afferent arteriole that supplies blood to the kidney glomerulus; JGA raises blood pressure by producing renin, which activates angiotensin.

**K- SELECTION:** The concept that life history of the population is centered upon producing relatively few offspring that have a good chance of survival.

**KARYOGAMY:** The fusion of nuclei of two cells, as part of syngamy.

**KARYOTYPE:** A method of classifying the chromosomes of a cell in relation to number, size and type.

**KEYSTONE PREDATOR:** A species that maintains species richness in a community through predation of the best competitors in the community, thereby maintaining populations of less competitive species.

**KILOCALORIE:** A thousand calories; the amount of heat energy required to raise the temperature of 1 kilogram of water by primary C.

**KILL = C X T:** Where other factors are constant, the disinfecting action may be represented by: Kill=C x T.

**KIN SELECTION:** A phenomenon of inclusive fitness, used to explain altruistic behavior between related individuals.

**KINESIS:** A change in activity rate in response to a stimulus.

**KINETIC ENERGY:** The ability of an object to do work by virtue of its motion. The energy terms that are used to describe the operation of a pump are pressure and head. The energy of motion. Moving matter does work by transferring some of its kinetic energy to other matter.

**KINETOCHORE:** A specialized region on the centromere that links each sister chromatid to the mitotic spindle.

**KINGDOM:** A taxonomic category, the second broadest after domain.

**KREBS CYCLE:** A chemical cycle involving eight steps that completes the metabolic breakdown of glucose molecules to carbon dioxide; occurs within the mitochondrion; the second major stage in cellular respiration. Also called citric acid cycle or tricarboxylic acid (TCA) cycle.

**LABORATORY BLANK:** See Method blank

**LABORATORY CONTROL SAMPLE (LCS):** See Ongoing precision and recovery (OPR) standard

**LACTEAL:** A tiny lymph vessel extending into the core of the intestinal villus and serving as the destination for absorbed chylomicrons.

**LACTIC ACID:** Gram-positive, anaerobic; produce lactic acid through fermentation; include *Lactobacillus*, essential in dairy product formation, and *Streptococcus*, common in humans.

**LAGGING STRAND:** A discontinuously synthesized DNA strand that elongates in a direction away from the replication fork.

**LAMARCK:** Proposed, in the early 1800s, that evolutionary change may occur via the inheritance of acquired characteristics. This idea, which has since been discredited, holds that the changes in characteristics which occur during an individual's life can be passed on to its offspring.

**LAND APPLICATION:** The disposal of wastewater or municipal solids onto land under controlled conditions.

**LAND DISPOSAL:** Application of municipal wastewater solids to the soil without production of usable agricultural products.

**LANDFILL:** A land disposal site that employs an engineering method of solid waste disposal to minimize environmental hazards and protect the quality of surface and subsurface waters.

**LANGELIER INDEX:** A measurement of Corrosivity. The water is becoming corrosive in the distribution system causing rusty water if the Langelier index indicates that the pH has decreased from the equilibrium point. Mathematically derived factor obtained from the values of calcium hardness, total alkalinity, and pH at a given temperature. A Langelier index of zero indicates perfect water balance (i.e., neither corroding nor scaling). The Langelier Saturation Index (sometimes Langelier Stability Index) is a calculated number used to predict the calcium carbonate stability of water. It indicates whether the water will precipitate, dissolve, or be in equilibrium with calcium carbonate. Langelier developed a method for predicting the pH at which water is saturated in calcium carbonate (called pHs). The LSI is expressed as the difference between the actual system pH and the saturation pH.

$$\text{LSI} = \text{pH} - \text{pHs}$$

If the actual pH of the water is below the calculated saturation pH, the LSI is negative and the water has a very limited scaling potential. If the actual pH exceeds pHs, the LSI is positive, and being supersaturated with CaCO<sub>3</sub>, the water has a tendency to form scale. At increasing positive index values, the scaling potential increases.

Langelier saturation index is defined as:

$$\text{LSI} = \text{pH (measured)} - \text{pHs}$$

\* For LSI > 0, water is super saturated and tends to precipitate a scale layer of CaCO<sub>3</sub>

\* For LSI = 0, water is saturated (in equilibrium) with CaCO<sub>3</sub>. A scale layer of CaCO<sub>3</sub> is neither precipitated nor dissolved

\* For LSI < 0, water is under saturated and tends to dissolve solid CaCO<sub>3</sub>

In practice, water with an LSI between -0.5 and +0.5 will not display enhanced mineral dissolving or scale forming properties. Water with an LSI below -0.5 tends to exhibit noticeably increased dissolving abilities while water with an LSI above +0.5 tends to exhibit noticeably increased scale forming properties.

It is also worth noting that the LSI is temperature sensitive. The LSI becomes more positive as the water temperature increases. This has particular implications in situations where well water is used. The temperature of the water when it first exits the well is often significantly lower than the temperature inside the building served by the well or at the laboratory where the LSI measurement is made.

**LARVA (pl. larvae):** A free-living, sexually immature form in some animal life cycles that may differ from the adult in morphology, nutrition, and habitat.

**LATERAL LINE SYSTEM:** A mechanoreceptor system consisting of a series of pores and receptor units (neuromasts) along the sides of the body of fishes and aquatic amphibians; detects water movements made by an animal itself and by other moving objects.

**LATERAL MERISTEMS:** The vascular and cork cambia, cylinders of dividing cells that run most of the length of stems and roots and are responsible for secondary growth.

**LAW OF INDEPENDENT ASSORTMENT:** Mendel's second law, stating that each allele pair segregates independently during gamete formation; applies when genes for two traits are located on different pairs of homologous chromosomes.

**LAW OF SEGREGATION:** Mendel's first law, stating that allele pairs separate during gamete formation, and then randomly re-form pairs during the fusion of gametes at fertilization.

**LEACHATE:** Fluid that trickles through solid materials or wastes and contains suspended or dissolved materials or products of the solids.

**LEACHING:** A chemical reaction between water and metals that allows for removal of soluble materials.

**LEADING STRAND:** The new continuously complementary DNA strand synthesized along the template strand in the 5' --> 3' direction.

**LEUKOCYTE:** A white blood cell; typically functions in immunity, such as phagocytosis or antibody production.

**LEVELS OF ORGANIZATION:** A basic concept in biology is that organization is based on a hierarchy of structural levels, with each level building on the levels below it.

**LICHEN:** An organism formed by the symbiotic association between a fungus and a photosynthetic alga.

**LIFE:** A table of data summarizing mortality in a population.

**LIGAMENT:** A type of fibrous connective tissue that joins bones together at joints.

**LIGAND:** A ligand is a molecule that binds specifically to a receptor site of another molecule. A ligase is an enzyme which catalyzes such a reaction. For example, a DNA ligase is an enzyme which catalyzes the covalent bonding of the 3' end of a new DNA fragment to the 5' end of a growing chain.

**LIGASE:** Ligases are enzymes that catalyze the "stitching together" of polymer fragments. DNA ligase, for example, catalyzes phosphodiester bond formation between two DNA fragments, and this enzyme is involved in normal DNA replication, repair of damaged chromosomes, and various in vitro techniques in genetic engineering that involve linking DNA fragments.

**LIGNIN:** A hard material embedded in the cellulose matrix of vascular plant cell walls that functions as an important adaptation for support in terrestrial species.

**LIMBIC SYSTEM:** A group of nuclei (clusters of nerve cell bodies) in the lower part of the mammalian forebrain that interact with the cerebral cortex in determining emotions; includes the hippocampus and the amygdala.

**LIME:** The term generally used to describe ground limestone (calcium carbonate), hydrated lime (calcium hydroxide), or burned lime (calcium oxide).

**LIME SOFTENING:** Lime softening is primarily used to "soften" water—that is to remove calcium and magnesium mineral salts. But it also removes harmful toxins like radon and arsenic. Though there is no consensus, some studies have even suggested that lime softening is effective at removal of Giardia. Hard water is a common condition responsible for numerous problems. Users often recognize hard water because it prevents their soap from lathering properly. However, it can also cause buildup ("scale") in hot water heaters, boilers, and hot water pipes. Because of these inconveniences, many treatment facilities use lime softening to soften hard water for consumer use. Before lime softening can be used, managers must determine the softening chemistry required. This is a relatively easy task for groundwater sources, which remain more constant in their composition. Surface waters, however, fluctuate widely in quality and may require frequent changes to the softening chemical mix. In lime softening, lime and sometimes sodium carbonate are added to the water as it enters a combination solids contact clarifier. This raises the pH (i.e., increases alkalinity) and leads to the precipitation of calcium carbonate. Later, the pH of the effluent from the clarifier is reduced again, and the water is then filtered through a granular media filter. The water chemistry requirements of these systems require knowledgeable operators, which may make lime softening an economic challenge for some very small systems.

**LIME STABILIZATION:** The addition of lime to untreated sludge to raise the pH to 12 for a minimum of 2 hours to chemically inactivate microorganisms.

**LINKED GENES:** Genes that are located on the same chromosomes.

**LIPID:** One of a family of compounds, including fats, phospholipids, and steroids, that are insoluble in water.

**LIPOPROTEIN:** A protein bonded to a lipid; includes the low-density lipoproteins (LDLs) and high-density lipoproteins (HDLs) that transport fats and cholesterol in the blood.

**LIPOSOME:** Liposomes are vesicles (spherules) in which the lipid molecules are spontaneously arranged into bilayers with hydrophilic groups exposed to water molecules both outside the vesicle and in the core.

**LISTED HAZARDOUS WASTE:** The designation for a waste material that appears on an EPA list of specific hazardous wastes or hazardous waste categories.

**LOCUS:** A particular place along the length of a certain chromosome where a specified allele is located.

**LOGISTIC POPULATION GROWTH:** A model describing population growth that levels off as population size approaches carrying capacity.

**L.O.T.O.:** If a piece of equipment is locked out, the key to the lock-out device the key should be held by the person who is working on the equipment. The tag is an identification device and the lock is a physical restraint.

**LYMPHOCYTE:** Lymphocytes (lymph cells, lympho- leukocytes) are a type of leukocyte (white blood cell) responsible for the immune response. There are two classes of lymphocytes: 1) the B- cells, when presented with a foreign chemical entity (antigen), change into antibody producing plasma cells; and, 2) the T- cells interact directly with foreign invaders such as bacteria and viruses. The T- cells express various surface marker macromolecules. For example, CD4+ is the notation for a specific expressed T- cell surface marker that can be identified by assay.

**LYSIS:** The destruction of a cell by rupture of the plasma membrane.

**LYSOGENIC CYCLE:** A type of viral replication cycle in which the viral genome becomes incorporated into the bacterial host chromosome as a prophage.

**LYSOSOME:** A membrane-bounded organelle found in eukaryotic cells (other than plants). Lysosomes contain a mixture of enzymes that can digest most of the macromolecules found in the rest of the cell.

**LYSOZYME:** An enzyme in perspiration, tears, and saliva that attacks bacterial cell walls.

**LYTIC CYCLE:** A type of viral replication cycle resulting in the release of new phages by death or lysis of the host cell.

**M-ENDO BROTH:** The coliform group are used as indicators of fecal pollution in water, for assessing the effectiveness of water treatment and disinfection, and for monitoring water quality. m-Endo Broth is used for selectively isolating coliform bacteria from water and other specimens using the membrane filtration technique. m-Endo Broth is prepared according to the formula of Fifield and Schaufus.<sup>1</sup> It is recommended by the American Public Health Association in standard total coliform membrane filtration procedure for testing water, wastewater, and foods.<sup>2,3</sup> The US EPA specifies using m-Endo Broth in the total coliform methods for testing water using single-step, two-step, and delayed incubation membrane filtration methods.

**M PHASE:** The mitotic phase of the cell cycle, which includes mitosis and cytokinesis.

**MACROEVOLUTION:** Evolutionary change on a grand scale, encompassing the origin of novel designs, evolutionary trends, adaptive radiation, and mass extinction.

**MACROMOLECULE:** A giant molecule of living matter formed by the joining of smaller molecules, usually by condensation synthesis. Polysaccharides, proteins, and nucleic acids are macromolecules.

**MACROPHAGE:** An amoeboid cell that moves through tissue fibers, engulfing bacteria and dead cells by phagocytosis.

**MAGNESIUM HARDNESS:** Measure of the magnesium salts dissolved in water – it is not a factor in water balance.

**MAGNETIC STARTER:** Is a type of motor starter should be used in an integrated circuit to control flow automatically.

**MAJOR HISTOCOMPATIBILITY COMPLEX:** A large set of cell surface antigens encoded by a family of genes. Foreign MHC markers trigger T-cell responses that may lead to rejection of transplanted tissues and organs.

**MAKEUP WATER:** Fluid introduced in a recirculating stream to maintain an equilibrium of temperature, solids concentration or other parameters. Also refers to the quantity of water required to make a solution.

**MALIGNANT TUMOR:** A cancerous growth; an abnormal growth whose cells multiply excessively, have altered surfaces, and may have unusual numbers of chromosomes and/or aberrant metabolic processes.

**MALPIGHIAN TUBULE:** A unique excretory organ of insects that empties into the digestive tract, removes nitrogenous wastes from the blood, and functions in osmoregulation.

**MANTLE:** A heavy fold of tissue in mollusks that drapes over the visceral mass and may secrete a shell.

**MARBLE AND LANGELIER TESTS:** Are used to measure or determine the corrosiveness of a water source.

**MASS NUMBER:** The sum of the number of protons plus the number of neutrons in the nucleus of an atom; unique for each element and designated by a superscript to the left of the elemental symbol.

**MATRIX SPIKE (MS):** A sample prepared by adding a known quantity of organisms to a specified amount of sample matrix for which an independent estimate of target analyte concentration is available. A matrix spike is used to determine the effect of the matrix on a method's recovery efficiency.

**MATRIX:** The nonliving component of connective tissue, consisting of a web of fibers embedded in homogeneous ground substance that may be liquid, jellylike, or solid.

**MATTER:** Anything that takes up space and has mass.

**MAXIMUM CONTAMINANT LEVEL (MCLs):** The maximum allowable level of a contaminant

**MECHANICAL SEAL:** A mechanical device used to control leakage from the stuffing box of a pump. Usually made of two flat surfaces, one of which rotates on the shaft. The two flat surfaces are of such tolerances as to prevent the passage of water between them. Held in place with spring pressure.

**MECHANORECEPTOR:** A sensory receptor that detects physical deformations in the body environment associated with pressure, touch, stretch, motion, and sound.

**MEDIAN BODIES:** Prominent, dark-staining, paired organelles consisting of microtubules and found in the posterior half of *Giardia*. In *G. intestinalis* (from humans), these structures often have a claw-hammer shape, while in *G. muris* (from mice), the median bodies are round.

**MEDIUM WATER SYSTEM:** More than 3,300 persons and 50,000 or fewer persons.

**MEDULLA OBLONGATA:** The lowest part of the vertebrate brain; a swelling of the hindbrain dorsal to the anterior spinal cord that controls autonomic, homeostatic functions, including breathing, heart and blood vessel activity, swallowing, digestion, and vomiting.

**MEDUSA:** The floating, flattened, mouth-down version of the cnidarian body plan. The alternate form is the polyp.

**MEGAPASCAL:** A unit of pressure equivalent to 10 atmospheres of pressure.

**MEGGER:** Used to test the insulation resistance on a motor.

**MEIOSIS:** A two-stage type of cell division in sexually reproducing organisms that results in gametes with half the chromosome number of the original cell.

**MEMBRANE:** A thin barrier that permits passage of particles of a certain size or of particular physical or chemical properties.

**MEMBRANE POTENTIAL:** The charge difference between the cytoplasm and extracellular fluid in all cells, due to the differential distribution of ions. Membrane potential affects the activity of excitable cells and the transmembrane movement of all charged substances.

**MESENTERIES:** Membranes that suspend many of the organs of vertebrates inside fluid-filled body cavities.

**MESODERM:** The middle primary germ layer of an early embryo that develops into the notochord, the lining of the coelom, muscles, skeleton, gonads, kidneys and most of the circulatory system.

**MESOSOME:** A localized infolding of the plasma membrane of a bacterium.

**MESSENGER:** (RNA) A type of RNA synthesized from DNA in the genetic material that attaches to ribosomes in the cytoplasm and specifies the primary structure of a protein.

**METABOLISM:** The sum total of the chemical and physical changes constantly taking place in living substances.

**METALLOID:** Metalloid is a term used in chemistry when classifying the chemical elements. On the basis of their general physical and chemical properties, nearly every element in the periodic table can be termed either a metal or a nonmetal. A few elements with intermediate properties are, however, referred to as metalloids. (In Greek metallon = metal and eidos = sort)

There is no rigorous definition of the term, but the following properties are usually considered characteristic of metalloids:

\* metalloids often form amphoteric oxides.

\* metalloids often behave as semiconductors (B,Si,Ge) to semimetals (e.g. Sb).

The concepts of metalloid and semiconductor should not be confused. Metalloid refers to the properties of certain elements in relation to the periodic table. Semiconductor refers to the physical properties of materials (including alloys, compounds) and there is only partial overlap between the two.

**METAMORPHOSIS:** The resurgence of development in an animal larva that transforms it into a sexually mature adult.

**METANEPHRIDIUM:** A type of excretory tubule in annelid worms that has internal openings called nephrostomes that collect body fluids and external openings called nephridiopores.

**METASTASIS:** The spread of cancer cells beyond their original site.

**METAZOAN:** A multicellular animal. Among important distinguishing characteristics of metazoa are cell differentiation and intercellular communication. For certain multicellular colonial entities such as sponges, some biologists prefer the term "parazoa".

**METHANE:** Methane is a chemical compound with the molecular formula CH<sub>4</sub>. It is the simplest alkane, and the principal component of natural gas. Methane's bond angles are 109.5 degrees. Burning methane in the presence of oxygen produces carbon dioxide and water. The relative abundance of methane and its clean burning process makes it a very attractive fuel. However, because it is a gas at normal temperature and pressure, methane is difficult to transport from its source.

In its natural gas form, it is generally transported in bulk by pipeline or LNG carriers; few countries still transport it by truck. Methane is a relatively potent greenhouse gas with a high global warming potential of 72 (averaged over 20 years) or 25 (averaged over 100 years).<sup>[1]</sup> Methane in the atmosphere is eventually oxidized, producing carbon dioxide and water. As a result, methane in the atmosphere has a half-life of seven years (if no methane was added, then every seven years, the amount of methane would halve). The abundance of methane in the Earth's atmosphere in 1998 was 1745 parts per billion, up from 700 ppb in 1750. In the same time period, CO<sub>2</sub> increased from 278 to 365 parts per million. The radiative forcing effect due to this increase in methane abundance is about one-third of that of the CO<sub>2</sub> increase. In addition, there is a large, but unknown, amount of methane in methane clathrates in the ocean floors. Global warming could release this methane, which could cause a further sharp rise in global temperatures. Such releases of methane may have been a major factor in previous major extinction events. The Earth's crust also contains huge amounts of methane. Large amounts of methane are produced anaerobically by methanogenesis. Other sources include mud volcanoes which are connected with deep geological faults.

**METHOD BLANK:** An aliquot of reagent water that is treated exactly as a sample, including exposure to all glassware, equipment, solvents, and procedures that are used with samples. The method blank is used to determine if analytes or interferences are present in the laboratory environment, the reagents, or the apparatus.

**Mg/L:** Stands for "milligrams per liter." A common unit of chemical concentration. It expresses the mass of a chemical that is present in a given volume of water. A milligram (one one-thousandth of a gram) is equivalent to about 18 grains of table salt. A liter is equivalent to about one quart.

**MICROBE OR MICROBIAL:** Any minute, simple, single-celled form of life, especially one that causes disease.

**MICROBIAL CONTAMINANTS:** Microscopic organisms present in untreated water that can cause waterborne diseases.

**MICROBIOLOGICAL:** Is a type of analysis in which a composite sample unacceptable.

**MICROBODY:** A small organelle, bounded by a single membrane and possessing a granular interior. Peroxisomes and glyoxysomes are types of microbodies.

**MICROEVOLUTION:** A change in the gene pool of a population over a succession of generations.

**MICROFILAMENT:** Minute fibrous structure generally composed of actin found in the cytoplasm of eukaryotic cells. They play a role in motion within cells.

**MICROFILTRATION:** A low pressure membrane filtration process that removes suspended solids and colloids generally larger than 0.1 micron diameter.

**MICROORGANISMS:** Very small animals and plants that are too small to be seen by the naked eye and must be observed using a microscope. Microorganisms in water include algae, bacteria, viruses, and protozoa. Algae growing in surface waters can cause off-taste and odor by producing the chemicals MIB and geosmin. Certain types of bacteria, viruses, and protozoa can cause disease in humans. Bacteria are the most common microorganisms found in treated drinking water. The great majority of bacteria are not harmful. In fact, humans would not be able to live without the bacteria that inhabit the intestines. However, certain types of bacteria called coliform bacteria can signal the presence of possible drinking water contamination.

**MICROSCOPE:** An instrument which magnifies images either by using lenses in an optical system to bend light (light microscope) or electromagnets to direct the movement of electrons (electron microscope).

**MICROTUBULE:** A minute tubular structure found in centrioles, spindle apparatus, cilia, flagella, and other places in the cytoplasm of eukaryotic cells. Microtubules play a role in movement and maintenance of shape.

**MICROVILLUS:** Collectively, fine, fingerlike projections of the epithelial cells in the lumen of the small intestine that increase its surface area.

**MILLIGRAMS PER LITER:** (mg/L) A common unit of measurement of the concentration of a material in solution.

**MILLILITER:** One one-thousandth of a liter; A liter is a little more than a quart. A milliliter is about two drops from an eye dropper.

**MIMICRY:** A phenomenon in which one species benefits by a superficial resemblance to an unrelated species. A predator or species of prey may gain a significant advantage through mimicry.

**MISCIBLE:** Capable of being mixed together.

**MISSENSE:** (mutation) The most common type of mutation involving a base-pair substitution within a gene that changes a codon, but the new codon makes sense, in that it still codes for an amino acid.

**MITOCHONDRIAL MATRIX:** The compartment of the mitochondrion enclosed by the inner membrane and containing enzymes and substrates for the Krebs cycle.

**MITOCHONDRION:** An organelle that occurs in eukaryotic cells and contains the enzymes of the citric acid cycle, the respiratory chain, and oxidative phosphorylation. A mitochondrion is bounded by a double membrane.

**MITOSIS:** A process of cell division in eukaryotic cells conventionally divided into the growth period (interphase) and four stages: prophase, metaphase, anaphase, and telophase. The stages conserve chromosome number by equally allocating replicated chromosomes to each of the daughter cells.

**MIXED LIQUOR SUSPENDED SOLIDS:** Suspended solids in the mixture of wastewater and activated sludge undergoing aeration in the aeration basin.

**MODEM SYNTHESIS:** A comprehensive theory of evolution emphasizing natural selection, gradualism, and populations as the fundamental units of evolutionary change; also called Neo-Darwinism.

**MOISTURE:** If a material is hygroscopic, it must be protected from water.

**MOISTURE AND POTASSIUM PERMANGANATE:** The combination of moisture and potassium permanganate produces heat.

**MOLARITY:** A common measure of solute concentration, referring to the number of moles of solute in 1 L of solution.

**MOLD:** A rapidly growing, asexually reproducing fungus.

**MOLE:** The number of grams of a substance that equals its molecular weight in daltons and contains Avogadro's number of molecules.

**MOLECULAR FORMULA:** A type of molecular notation indicating only the quantity of the constituent atoms.

**MOLECULAR WEIGHT:** The molecular mass (abbreviated Mr) of a substance, formerly also called molecular weight and abbreviated as MW, is the mass of one molecule of that substance, relative to the unified atomic mass unit u (equal to 1/12 the mass of one atom of carbon-12). This is distinct from the relative molecular mass of a molecule, which is the ratio of the mass of that molecule to 1/12 of the mass of carbon 12 and is a dimensionless number. Relative molecular mass is abbreviated to Mr.

**MOLECULE:** Two or more atoms of one or more elements held together by ionic or covalent chemical bonds.

**MOLTING:** A process in arthropods in which the exoskeleton is shed at intervals to allow growth by secretion of a larger exoskeleton.

**MONERA:** The kingdom of life forms that includes all of the bacteria.

**MONOCLONAL ANTIBODY:** A defensive protein produced by cells descended from a single cell; an antibody that is secreted by a clone of cells and, consequently, is specific for a single antigenic determinant.

**MONOECIOUS:** Referring to an organism having the capacity of producing both sperm and eggs.

**MONOHYBRID CROSS:** A breeding experiment that employs parental varieties differing in a single character.

**MONOMER:** A small molecule, two or more of which can be combined to form oligomers (consisting of a few monomers) or polymers (consisting of many monomers).

**MONOPHYLETIC:** A term used to describe any taxon derived from a single ancestral form that gave rise to no species in other taxa.

**MONOSACCHARIDE:** A simple sugar; a monomer.

**MONOZYGOTIC TWINS:** Monozygotic twins are genetically identical, derived from the division and autonomous development of a single zygote (fertilized egg).

**MORPHOGENESIS:** The development of body shape and organization during ontogeny.

**MORPHOSPECIES:** Species defined by their anatomical features.

**MOSAIC:** A pattern of development, such as that of a mollusk, in which the early blastomeres each give rise to a specific part of the embryo. In some animals, the fate of the blastomeres is established in the zygote.

**MOTOR NERVOUS SYSTEM:** In vertebrates, the component of the peripheral nervous system that transmits signals from the central nervous system to effector cells.

**MPF:** M: phase promoting factor: A protein complex required for a cell to progress from late interphase to mitosis; the active form consists of cyclin and cdc2, a protein kinase.

**M.S.D.S.:** A safety document must an employer provide to an operator upon request.

**MUCOSA:** Refers to the mucous tissue lining various tubular structures in the body.

**MUD BALLS IN FILTER MEDIA:** Is a possible result of an ineffective or inadequate filter backwash.

**MULLERIAN MIMICRY:** A mutual mimicry by two unpalatable species.

**MULTIGENE FAMILY:** A collection of genes with similar or identical sequences, presumably of common origin.

**MUNICIPAL WASTE:** The combined solid and liquid waste from residential, commercial and industrial sources.

**MUNICIPAL WASTEWATER TREATMENT PLANT (MWTP):** Treatment works designed to treat municipal wastewater.

**MURIATIC ACID:** An acid used to reduce pH and alkalinity. Also used to remove stain and scale.

**MUST:** This action, activity, or procedural step is required.

**MUTAGEN:** A chemical or physical agent that interacts with DNA and causes a mutation.

**MUTAGENESIS:** The creation of mutations.

**MUTATION:** A spontaneous or induced change in a gene's or chromosome's structure or number. The resulting individual is termed a mutant.

**MUTUALISM:** A symbiotic relationship in which both the host and the symbiont benefit.

**MYCELIUM:** The densely branched network of hyphae in a fungus.

**MYCOBACTERIUM:** Pleomorphic spherical or rod-shaped, frequently branching, no gram stain, aerobic; commonly form yellow pigments; include Mycobacterium tuberculosis, cause of tuberculosis.

**MYCOPLASMA:** Spherical, commonly forming branching chains, no gram stain, aerobic but can live in certain anaerobic conditions; without cell walls yet structurally resistant to lysis; among smallest of bacteria; named for superficial resemblance to fungal hyphae (myco-means "fungus").

**MYELIN SHEATH:** An insulating coat of cell membrane from Schwann cells that is interrupted by nodes of Ranvier where saltatory conduction occurs.

**MYOFIBRILS:** Fibrils arranged in longitudinal bundles in muscle cells (fibers); composed of thin filaments of actin and a regulatory protein and thick filaments of myosin.

**MYOGLOBIN:** An oxygen-storing, pigmented protein in muscle cells.

**MYOSIN:** A type of protein filament that interacts with actin filaments to cause cell movement, such as contraction in muscle cells.

**NAD<sup>+</sup>:** Nicotinamide adenine dinucleotide (oxidized); a coenzyme present in all cells that assists enzymes in transferring electrons during the redox reactions of metabolism.

**NANO-FILTRATION:** A specialty membrane filtration process that rejects solutes larger than approximately one nanometer (10 angstroms) in size.

**NANOMETER:** A unit of measure (length). 1 nm is equal to  $1 \times 10^{-9}$  m, or 1/1,000,000 mm.

**NaOCl:** Is the molecular formula of Sodium hypochlorite.

**NaOH:** Is the molecular formula of Sodium hydroxide.

**NATURAL ORGANIC MATTER:** Organic matter present in natural waters.

**NEGATIVE CONTROL:** See Method blank.

**NEGATIVE FEEDBACK:** A primary mechanism of homeostasis, whereby a change in a physiological variable that is being monitored triggers a response that counteracts the initial fluctuation.

**NEURAMINIDASE:** A surface enzyme possessed by some influenza viruses which help the virus penetrate the mucus layer protecting the respiratory epithelium and also plays a role in budding of new virus particles from infected cells.

**NEUTRALIZATION:** The chemical process that produces a solution that is neither acidic nor alkaline. Usually with a pH between 6 and 8.

**NEURON:** A nerve cell; the fundamental unit of the nervous system, having structure and properties that allow it to conduct signals by taking advantage of the electrical charge across its cell membrane.

**NEUROSECRETORY CELLS:** Cells that receive signals from other nerve cells, but instead of signaling to an adjacent nerve cell or muscle, release hormones into the blood stream.

**NEUROTRANSMITTER:** The chemical messenger released from the synaptic terminals of a neuron at a chemical synapse that diffuses across the synaptic cleft and binds to and stimulates the postsynaptic cell.

**NEUTRAL VARIATION:** Genetic diversity that confers no apparent selective advantage.

**NEUTRALIZATION REACTIONS:** Chemical reactions between acids and bases where water is an end product.

**NEUTRON:** An uncharged subatomic particle of about the same size and mass as a proton.

**NH3:** The molecular formula of Ammonia.

**NH4<sup>+</sup>:** The molecular formula of the Ammonium ion.

**NITRATES:** A dissolved form of nitrogen found in fertilizers and sewage by-products that may leach into groundwater and other water sources. Nitrates may also occur naturally in some waters. Over time, nitrates can accumulate in aquifers and contaminate groundwater.

**NITROGEN:** Nitrogen is a nonmetal, with an electronegativity of 3.0. It has five electrons in its outer shell and is therefore trivalent in most compounds. The triple bond in molecular nitrogen (N<sub>2</sub>) is one of the strongest in nature. The resulting difficulty of converting (N<sub>2</sub>) into other compounds, and the ease (and associated high energy release) of converting nitrogen compounds into elemental N<sub>2</sub>, have dominated the role of nitrogen in both nature and human economic activities. At atmospheric pressure molecular nitrogen condenses (liquefies) at 77 K (-195.8 °C) and freezes at 63 K (-210.0 °C) into the beta hexagonal close-packed crystal allotropic form. Below 35.4 K (-237.6 °C) nitrogen assumes the alpha cubic crystal allotropic form. Liquid nitrogen, a fluid resembling water, but with 80.8% of the density, is a common cryogen. Unstable allotropes of nitrogen consisting of more than two nitrogen atoms have been produced in the laboratory, like N<sub>3</sub> and N<sub>4</sub>.<sup>[1]</sup> Under extremely high pressures (1.1 million atm) and high temperatures (2000 K), as produced under diamond anvil conditions, nitrogen polymerizes into the single bonded diamond crystal structure, an allotrope nicknamed "nitrogen diamond."

**NITROGEN AND PHOSPHORUS:** Pairs of elements and major plant nutrients that cause algae to grow.

**NITROGEN-FIXING:** Rod-shaped, gram-negative, aerobic; convert atmospheric nitrogen gas to ammonium in soil; include Azotobacter, a common genus.

**NO<sub>3</sub><sup>-</sup>:** The molecular formula of the Nitrate ion.

**NOMENCLATURE:** The method of assigning names in the classification of organisms.

**NON-CARBONATE HARDNESS:** The portion of the total hardness in excess of the alkalinity.

**NON-CARBONATE IONS:** Water contains non-carbonate ions if it cannot be softened to a desired level through the use of lime only.

**NON-POINT SOURCE POLLUTION:** Air pollution may leave contaminants on highway surfaces. This non-point source pollution adversely impacts reservoir water and groundwater quality.

**NONCOMPETITIVE INHIBITOR:** A substance that reduces the activity of an enzyme by binding to a location remote from the active site, changing its conformation so that it no longer binds to the substrate.

**NONCYCLIC ELECTRON FLOW:** A route of electron flow during the light reactions of photosynthesis that involves both photosystems and produces ATP, NADPH, and oxygen; the net electron flow is from water to NADP<sup>+</sup>.

**NONCYCLIC PHOTOPHOSPHORYLATION:** The production of ATP by noncyclic electron flow.

**NONDISJUNCTION:** An accident of meiosis or mitosis, in which both members of a pair of homologous chromosomes or both sister chromatids fail to separate normally.

**NONPOLAR:** Electrically symmetrical. For example, in many molecules with covalent bonds, the electrons are shared equally; the poles are electrically neutral.

**NONSENSE MUTATION:** A mutation that changes an amino acid codon to one of the three stop codons, resulting in a shorter and usually nonfunctional protein.

**NORM OF REACTION:** The range of phenotypic possibilities for a single genotype, as influenced by the environment.

**NORMALITY:** It is the number of equivalent weights of solute per liter of solution. Normality highlights the chemical nature of salts: in solution, salts dissociate into distinct reactive species (ions such as H<sup>+</sup>, Fe<sup>3+</sup>, or Cl<sup>-</sup>). Normality accounts for any discrepancy between the concentrations of the various ionic species in a solution. For example, in a salt such as MgCl<sub>2</sub>, there are two moles of Cl<sup>-</sup> for every mole of Mg<sup>2+</sup>, so the concentration of Cl<sup>-</sup> as well as of Mg<sup>2+</sup> is said to be 2 N (read: "two normal"). Further examples are given below. A normal is one gram equivalent of a solute per liter of solution. The definition of a gram equivalent varies depending on the type of chemical reaction that is discussed - it can refer to acids, bases, redox species, and ions that will precipitate. It is critical to note that normality measures a single ion which takes part in an overall solute. For example, one could determine the normality of hydroxide or sodium in an aqueous solution of sodium hydroxide, but the normality of sodium hydroxide itself has no meaning. Nevertheless it is often used to describe solutions of acids or bases, in those cases it is implied that the normality refers to the H<sup>+</sup> or OH<sup>-</sup> ion. For example, 2 Normal sulfuric acid (H<sub>2</sub>SO<sub>4</sub>), means that the normality of H<sup>+</sup> ions is 2, or that the molarity of the sulfuric acid is 1. Similarly for 1 Molar H<sub>3</sub>PO<sub>4</sub> the normality is 3 as it contains three H<sup>+</sup> ions.

**NTU:** (Nephelometric turbidity unit): A measure of the clarity or cloudiness of water.

**NUCLEAR:** 1) (envelope) The surface, consisting of two layers of membrane, that encloses the nucleus of eukaryotic cells. 2) (pore) An opening of the nuclear envelope which allows for the movement of materials between the nucleus and surrounding cytoplasm.

**NUCLEASE:** This term refers to any enzyme that acts on nucleic acids, e.g., Dnase, Rnase, endonuclease, etc.

**NUCLEIC:** (acid) A polymer composed of nucleotides that are joined by covalent bonds (phosphodiester linkages) between the phosphate of one nucleotide and the sugar of the next nucleotide.

**NUCLEOID:** The region that harbors the chromosome of a prokaryotic cell. Unlike the eukaryotic nucleus, it is not bounded by a membrane.

**NUCLEOLUS (pl. nucleoli):** A specialized structure in the nucleus, formed from various chromosomes and active in the synthesis of ribosomes.

**NUCLELUS:** A small, generally spherical body found within the nucleus of eukaryotic cells. The site of ribosomal RNA synthesis.

**NUCLEOSIDE:** An organic molecule consisting of a nitrogenous base joined to a five- carbon sugar.

**NUCLEOSOME:** The basic, beadlike unit of DNA packaging in eukaryotes, consisting of a segment of DNA wound around a protein core composed of two copies of each of four types of histone.

**NUCLEOTIDE:** The basic chemical unit (monomer) of a nucleic acid. A nucleotide in RNA consists of one of four nitrogenous bases linked to ribose, which in turn is linked to phosphate. In DNA, deoxyribose is present instead of ribose.

**NUCLEUS:** A membrane-bound organelle containing genetic material. Nuclei are a prominent internal structure seen both in *Cryptosporidium* oocysts and *Giardia* cysts. In *Cryptosporidium* oocysts, there is one nucleus per sporozoite. One to four nuclei can be seen in *Giardia* cysts.

**NUCLEUS:** The membrane bound organelle of eukaryotic cells that contains the cell's genetic material. Also the central region of an atom composed of protons and neutrons.

**NULL:** In the scientific method, the hypothesis which one attempts to falsify.

**O<sub>3</sub>:** The molecular formula of ozone.

**OLIGOTROPHIC:** A reservoir that is nutrient-poor and contains little plant or animal life. An oligotrophic ecosystem or environment is one that offers little to sustain life. The term is commonly utilized to describe bodies of water or soils with very low nutrient levels. It derives etymologically from the Greek oligo (small, little, few) and trophe (nutrients, food). Oligotrophic environments are of special interest for the alternative energy sources and survival strategies upon which life could rely.

**ONGOING PRECISION AND RECOVERY (OPR) STANDARD:** A method blank spiked with known quantities of analytes. The OPR is analyzed exactly like a sample. Its purpose is to assure that the results produced by the laboratory remain within the limits specified in this method for precision and recovery.

**OOCYST AND CYST SPIKING SUSPENSION:** See Spiking suspension.

**OOCYST AND CYST STOCK SUSPENSION:** See Stock suspension.

**OOCYST:** The encysted zygote of some sporozoa; e.g., *Cryptosporidium*. The oocyst is a phase or form of the organism produced as a normal part of the life cycle of the organism. It is characterized by a thick and environmentally resistant outer wall.

**ORGANIC:** Relating to, or derived from, a living thing. A description of a substance that contains carbon atoms linked together by carbon-carbon bonds.

**ORGANIC MATTER:** Substances containing carbon compounds, usually of animal or vegetable origin.

**ORGANIC PRESURORS:** Natural or man-made compounds with chemical structures based upon carbon that, upon combination with chlorine, leading to trihalomethane formation.

**OSMOSIS:** Osmosis is the process by which water moves across a semi permeable membrane from a low concentration solute to a high concentration solute to satisfy the pressure differences caused by the solute.

**OVER-RANGE PROTECTION DEVICES:** Mechanical dampers, snubbers and an air cushion chamber are examples of surging and overrange protection devices.

**OXIDE:** An oxide is a chemical compound containing at least one oxygen atom as well as at least one other element. Most of the Earth's crust consists of oxides. Oxides result when elements are oxidized by oxygen in air. Combustion of hydrocarbons affords the two principal oxides of carbon, carbon monoxide and carbon dioxide. Even materials that are considered to be pure elements often contain a coating of oxides. For example, aluminum foil has a thin skin of Al<sub>2</sub>O<sub>3</sub> that protects the foil from further corrosion. Virtually all elements burn in an atmosphere of oxygen. In the presence of water and oxygen (or simply air), some elements - lithium, sodium, potassium, rubidium, caesium, strontium and barium - react rapidly, even dangerously to give the hydroxides. In part for this reason, alkali and alkaline earth metals are not found in nature in their metallic, i.e., native, form. Caesium is so reactive with oxygen that it is used as a getter in vacuum tubes, and solutions of potassium and sodium, so called NaK are used to deoxygenate and dehydrate some organic solvents. The surface of most metals consists of oxides and hydroxides in the presence of air. A well-known example is aluminum foil, which is coated with a thin film of aluminum oxide that passivates the metal, slowing further corrosion. The aluminum oxide layer can be built to greater thickness by the process of electrolytic anodizing. Although solid magnesium and aluminum react slowly with oxygen at STP, they, like most metals, will burn in air, generating very high temperatures. As a consequence, finely divided powders of most metals can be dangerously explosive in air.

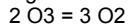
**OXIDIZED:**

1. to convert (an element) into an oxide; combine with oxygen.
2. to cover with a coating of oxide or rust.
3. to take away hydrogen, as by the action of oxygen; add oxygen or any nonmetal.
4. to remove electrons from (an atom or molecule), thereby increasing the valence. Compare REDUCE (def. 12). –verb (used without object)
5. to become oxidized.
6. (esp. of white wine) to lose freshness after prolonged exposure to air and often to darken in color.

**OXIDIZING:** The process of breaking down organic wastes into simpler elemental forms or by products. Also used to separate combined chlorine and convert it into free chlorine.

**OXYGEN DEFICIENT ENVIRONMENT:** One of the most dangerous threats to an operator upon entering a manhole.

**OZONE:** Ozone or trioxygen ( $O_3$ ) is a triatomic molecule, consisting of three oxygen atoms. It is an allotrope of oxygen that is much less stable than the diatomic  $O_2$ . Ground-level ozone is an air pollutant with harmful effects on the respiratory systems of animals. Ozone in the upper atmosphere filters potentially damaging ultraviolet light from reaching the Earth's surface. It is present in low concentrations throughout the Earth's atmosphere. It has many industrial and consumer applications. Ozone, the first allotrope of a chemical element to be recognized by science, was proposed as a distinct chemical compound by Christian Friedrich Schönbein in 1840, who named it after the Greek word for smell (ozein), from the peculiar odor in lightning storms. The formula for ozone,  $O_3$ , was not determined until 1865 by Jacques-Louis Soret and confirmed by Schönbein in 1867. Ozone is a powerful oxidizing agent, far better than dioxygen. It is also unstable at high concentrations, decaying to ordinary diatomic oxygen (in about half an hour in atmospheric conditions):



This reaction proceeds more rapidly with increasing temperature and decreasing pressure. Deflagration of ozone can be triggered by a spark, and can occur in ozone concentrations of 10 wt% or higher.

**PACKING:** Material, usually of woven fiber, placed in rings around the shaft of a pump and used to control the leakage from the stuffing box.

**PAINT FILTER TEST:** Test to determine free water content of sludge or dewatered solids sample. Usually used as the criteria for admission to a landfill.

**PARAMECIUM:** Paramecia are a group of unicellular ciliate protozoa formerly known as slipper animalcules from their slipper shape. They are commonly studied as a representative of the ciliate group. Simple cilia cover the body which allows the cell to move with a synchronous motion (like a caterpillar). There is also a deep oral groove containing inconspicuous compound oral cilia (as found in other peniculids) that is used to draw food inside. They generally feed upon bacteria and other small cells. Osmoregulation is carried out by a pair of contractile vacuoles, which actively expel water absorbed by osmosis from their surroundings. Paramecia are widespread in freshwater environments, and are especially common in scums. Paramecia are attracted by acidic conditions. Certain single-celled eukaryotes, such as Paramecium, are examples for exceptions to the universality of the genetic code (translation systems where a few codons differ from the standard ones).

**PARTS PER MILLION (PPM):** A common unit of measure used to express the number of parts of a substance contained within a million parts of a liquid, solid, or gas.

**PASTEURIZATION:** A process for killing pathogenic organisms by applying heat for a specific period of time.

**PATHOGENS:** Disease-causing pathogens; waterborne pathogens A pathogen may contaminate water and cause waterborne disease.

**Pb:** The chemical symbol of Lead.

**PCE:** abbr. perchloroethylene. Known also as perc or tetrachloroethylene, perchloroethylene is a clear, colorless liquid with a distinctive, somewhat ether-like odor. It is non-flammable, having no measurable flashpoint or flammable limits in air. Effective over a wide range of applications, perchloroethylene is supported by closed loop transfer systems, stabilizers and employee exposure monitoring.

**pCi/L:** Picocuries per liter A curie is the amount of radiation released by a set amount of a certain compound. A picocurie is one quadrillionth of a curie.

**PEAK DEMAND:** The maximum momentary load placed on a water treatment plant, pumping station or distribution system.

**PERKINESIS:** The aggregation resulting from random thermal motion of fluid molecules.

**PERMEATE:** The term for water which has passed through the membrane of a reverse osmosis unit. The liquid that passes through a membrane.

**pH:** A unit of measure which describes the degree of acidity or alkalinity of a solution. The pH scale runs from 0 to 14 with 7 being the mid-point or neutral. A pH of less than 7 is on the acid side of the scale with 0 as the point of greatest acid activity. A pH of more than 7 is on the basic (alkaline) side of the scale with 14 as the point of greatest basic activity. The term pH is derived from "p", the mathematical symbol of the negative logarithm, and "H", the chemical symbol of Hydrogen. The definition of pH is the negative logarithm of the Hydrogen ion activity.  $pH = -\log[H^+]$ .

**pH OF SATURATION:** The ideal pH for perfect water balance in relation to a particular total alkalinity level and a particular calcium hardness level, at a particular temperature. The pH where the Langelier Index equals zero.

**PHENOLPHTHALEIN/TOTAL ALKALINITY:** The relationship between the alkalinity constituent's bicarbonate, carbonate, and hydroxide can be based on the P and T alkalinity measurement.

**PHENOL RED:** Chemical reagent used for testing pH in the range of 6.8 - 8.4.

**PHOSPHATE, NITRATE AND ORGANIC NITROGEN:** Nutrients in a domestic water supply reservoir may cause water quality problems if they occur in moderate or large quantities.

**PHYSICAL CHEMICAL TREATMENT:** Treatment processes that are non-biological in nature.

**PICOCURIE:** A unit of radioactivity. "Pico" is a metric prefix that means one one-millionth of one one-millionth. A picocurie is one one-millionth of one one-millionth of a Curie. A Curie is that quantity of any radioactive substance that undergoes 37 billion nuclear disintegrations per second. Thus a picocurie is that quantity of any radioactive substance that undergoes 0.037 nuclear disintegrations per second.

**PIEZOMETRIC SURFACE:** See potentiometric surface.

**PIN FLOC:** Small flocculated particle size.

**PLATE AND FRAME PRESS:** A batch process dewatering device in which sludge is pumped under high pressure through a series of parallel plates, in which a chamber is created between the plates. Each plate is fitted with filter cloth and the solids are collected in the chambers and the water is filtered from the sludge.

**POINT SOURCE DISCHARGE:** A pipe, ditch, channel or other container from which pollutants may be discharged.

**POLLUTANT:** A substance, organism or energy form present in amounts that impair or threaten an ecosystem to the extent that its current or future uses are prevented.

**POLLUTION:** To make something unclean or impure. See Contaminated.

**POLYMER:** A type of chemical when combined with other types of coagulants aid in binding small suspended particles to larger particles to help in the settling and filtering processes. Chemical used for flocculation in dewatering. Also known as a "polyelectrolyte" which is a substance made of giant molecules formed by the union of simple smaller molecules.

**POLYPHOSPHATES:** Chemicals that may be added to remove low levels of iron and manganese.

**PORE SPACE:** The interstitial space between sediments and fractures that is capable of storing and transmitting water.

**POROSITY:** A factor representing a rock, soil, or formations percentage of open space available for the percolation and storage of groundwater.

**POSITIVE CONTROL:** See Ongoing precision and recovery standard.

**POST-CHLORINE:** Where the water is chlorinated to make sure it holds a residual in the distribution system.

**POST TREATMENT:** Treatment of finished water or wastewater to further enhance its quality.

**POTABLE:** Good water which is safe for drinking or cooking purposes. Non-Potable: A liquid or water that is not approved for drinking.

**POTENTIAL ENERGY:** The energy that a body has by virtue of its position or state enabling it to do work.

**PPM:** Abbreviation for parts per million.

**PRE-CHLORINE:** Where the raw water is dosed with a large concentration of chlorine.

**PRE-CHLORINATION:** The addition of chlorine before the filtration process will help:

- > Control algae and slime growth
- > Control mud ball formation
- > Improve coagulation
- > Precipitate iron

The addition of chlorine to the water prior to any other plant treatment processes.

**PRECIPITATE:** A solid that separates from a solution.

**PRECIPITATION:** The phenomenon that occurs when a substance held in solution passes out of solution into a solid form.

**PRELIMINARY TREATMENT:** Treatment steps including comminution, screening, grit removal, pre-aeration, and/or flow equalization that prepares wastewater influent for further treatment.

**PRESSURE:** Pressure is defined as force per unit area. It is usually more convenient to use pressure rather than force to describe the influences upon fluid behavior. The standard unit for pressure is the Pascal, which is a Newton per square meter. For an object sitting on a surface, the force pressing on the surface is the weight of the object, but in different orientations it might have a different area in contact with the surface and therefore exert a different pressure.

**PRESSURE FILTER:** Filter unit enclosed in a vessel that may be operated under pressure.

**PRESSURE HEAD:** The height of a column of water capable of being maintained by pressure. See also Total Head, Total Dynamic Head.

**PRESSURE MEASUREMENT:** Bourdon tube, Bellows gauge and Diaphragm are commonly used to measure pressure in waterworks systems. A Bellows-type sensor reacts to a change in pressure.

**PREVENTION:** To take action. Stop something before it happens.

**PRIMARY CLARIFIER:** Sedimentation basin that precedes secondary wastewater treatment.

**PRIMARY SLUDGE:** Sludge produced in a primary waste treatment unit.

**PRIMARY TREATMENT:** Treatment steps including sedimentation and/or fine screening to produce an effluent suitable for biological treatment.

**PROCESS WASTEWATER:** Wastewater generated during manufacture or production processes.

**PROCESS WATER:** Water that is used for, or comes in contact with an end product or the materials used in an end product.

**PROPIONIC ACID:** Rod-shaped, pleomorphic, gram-positive, anaerobic; ferment lactic acid; fermentation produces holes in Swiss cheese from the production of carbon dioxide.

**PROTON, NEUTRON AND ELECTRON:** Are the 3 fundamental particles of an atom.

**PROTOZOA:** Microscopic animals that occur as single cells. Some protozoa can cause disease in humans. Protozoa form cysts, which are specialized cells like eggs that are very resistant to chlorine. Cysts can survive the disinfection process, then "hatch" into normal cells that can cause disease. Protozoa must be removed from drinking water by filtration, because they cannot be effectively killed by chlorine.

**PSEUDOMONAD:** Rod-shaped (straight or curved) with polar flagella, gram-negative, aerobic; can use up to 100 different compounds for carbon and energy.

**PUMPING LIFT:** The height to which water must be pumped or lifted to, feet of head.

**PTFE:** Polytetrafluoroethylene.

**QUANTITATIVE TRANSFER:** The process of transferring a solution from one container to another using a pipette in which as much solution as possible is transferred, followed by rinsing of the walls of the source container with a small volume of rinsing solution (e.g., reagent water, buffer, etc.), followed by transfer of the rinsing solution, followed by a second rinse and transfer.

**QUICKLIME:** A calcium oxide material produced by calcining limestone to liberate carbon dioxide, also called "calcined lime" or "pebble lime", commonly used for pH adjustment. Chemical formula is CaO.

**RAW TURBIDITY:** The turbidity of the water coming to the treatment plant from the raw water source.

**REAGENT:** A substance used in a chemical reaction to measure, detect, examine, or produce other substances.

**REDOX POTENTIAL:** Reduction potential (also known as redox potential, oxidation / reduction potential or ORP) is the tendency of a chemical species to acquire electrons and thereby be reduced. Each species has its own intrinsic reduction potential; the more positive the potential, the greater the species' affinity for electrons and tendency to be reduced. In aqueous solutions, the reduction potential is the tendency of the solution to either gain or lose electrons when it is subject to change by introduction of a new species. A solution with a higher (more positive) reduction potential than the new species will have a tendency to gain electrons from the new species (i.e. to be reduced by oxidizing the new species) and a solution with a lower (more negative) reduction potential will have a tendency to lose electrons to the new species (i.e. to be oxidized by reducing the new species). Just as the transfer of hydrogen ions between chemical species determines the pH of an aqueous solution, the transfer of electrons between chemical species determines the reduction potential of an aqueous solution. Like pH, the reduction potential represents an intensity factor. It does not characterize the capacity of the system for oxidation or reduction, in much the same way that pH does not characterize the buffering capacity.

**RELAY LOGIC:** The name of a popular method of automatically controlling a pump, valve, chemical feeder, and other devices.

**RESERVOIR:** An impoundment used to store water.

**RICKETTSIA:** Spherical or rod-shaped, gram-negative, aerobic; cause Rocky Mountain spotted fever and typhus; closely related to Agrobacterium, a common gall-causing plant bacterium.

**ROTIFER:** Rotifers get their name (derived from Greek and meaning "wheel-bearer"; they have also been called wheel animalcules) from the corona, which is composed of several ciliated tufts around the mouth that in motion resemble a wheel. These create a current that sweeps food into the mouth, where it is chewed up by a characteristic pharynx (called the mastax) containing a tiny, calcified, jaw-like structure called the trophi. The cilia also pull the animal, when unattached, through the water. Most free-living forms have pairs of posterior toes to anchor themselves while feeding. Rotifers have bilateral symmetry and a variety of different shapes. There is a well-developed cuticle which may be thick and rigid, giving the animal a box-like shape, or flexible, giving the animal a worm-like shape; such rotifers are respectively called loricate and illoricate.

**RAW SEWAGE:** Untreated wastewater and its contents.

**RAW SLUDGE:** Undigested sludge recently removed from a sedimentation basin.

**RAW WATER:** Untreated surface or groundwater.

**REAGENT WATER BLANK:** see Method blank.

**REAGENT WATER:** Water demonstrated to be free from the analytes of interest and potentially interfering substances at the method detection limit for the analyte.

**RECLAIMED WATER:** Wastewater that has been treated to a level that allows for its reuse for a beneficial purpose.

**RECLAMATION:** The process of improving or restoring the condition of land or other material to a better or more useful state.

**RECYCLING:** The process by which recovered materials are transformed into new products.

**RELATIVE STANDARD DEVIATION (RSD):** The standard deviation divided by the mean times 100.

**RESIDENCE TIME:** The period of time that a volume of liquid remains in a tank or system.

**RESPIRATION:** Intake of oxygen and discharge of carbon dioxide as a result of biological oxidation.

**RETURN ACTIVATED SLUDGE:** Settled activated sludge that is returned to mix with raw or primary settled wastewater.

**ROBERT HOOKE:** Coined the term "cell" to describe the structures he saw while examining a piece of cork using a microscope.

**ROTARY DRUM SCREEN:** Cylindrical screen used to remove floatable and suspended solids.

**RSD:** See Relative standard deviation.

**SANITARY SURVEY:** Persons trained in public health engineering and the epidemiology of waterborne diseases should conduct the sanitary survey. The importance of a detailed sanitary survey of a new water source cannot be overemphasized. An on-site review of the water sources, facilities, equipment, operation, and maintenance of a public water systems for the purpose of evaluating the adequacy of the facilities for producing and distributing safe drinking water. The purpose of a non-regulatory sanitary survey is to identify possible biological and chemical pollutants which might affect a water supply.

**SANITIZER:** A disinfectant or chemical which disinfects (kills bacteria), kills algae and oxidizes organic matter.

**SATURATION INDEX:** See Langmuir's Index.

**SATURATOR:** A device which produces a fluoride solution for the fluoride process. Crystal-grade types of sodium fluoride should be fed with a saturator. Overfeeding must be prevented to protect public health when using a fluoridation system.

**SATURATED ZONE:** Where an unconfined aquifer becomes saturated beneath the capillary fringe.

**SCADA:** A remote method of monitoring pumps and equipment. 130 degrees F is the maximum temperature that transmitting equipment is able to withstand. If the level controller may be set with too close a tolerance 45 could be the cause of a control system that is frequently turning a pump on and off.

**SCALE:** Crust of calcium carbonate, the result of unbalanced water. Hard insoluble minerals deposited (usually calcium bicarbonate) which forms on pool and spa surfaces and clog filters, heaters and pumps. Scale is caused by high calcium hardness and/or high pH. The regular use of stain prevention chemicals can prevent scale.

**SCREENINGS PRESS:** A mechanical press used to compact and/or dewater material removed from mechanical screening equipment.

**SCROLL AND BASKET:** The two basic types of centrifuges used in water treatment.

**SCRUBBER:** A device used to removal particulates or pollutant gases from combustion or chemical process exhaust streams.

**SCUM:** Floatable materials found on the surface of primary and secondary settling tanks consisting of food wastes, grease, fats, paper, foam, and similar matter.

**SECONDARY CLARIFIER:** A clarifier following a secondary treatment process, designed for gravity removal of suspended matter.

**SECONDARY SLUDGE:** The sludge from the secondary clarifier in a wastewater treatment plant.

**SECONDARY TREATMENT:** The treatment of wastewater through biological oxidation after primary treatment.

**SEDIMENTATION:** The removal of settleable suspended solids from water or wastewater by gravity in a quiescent basin or clarifier.

**SEDIMENTATION BASIN:** A quiescent tank used to remove suspended solids by gravity settling. Also called clarifiers or settling tanks, they are usually equipped with a motor driven rake mechanism to collect settled sludge and move it to a central discharge point.

**SEDIMENTATION BASIN:** Where the thickest and greatest concentration of sludge will be found. Twice a year sedimentation tanks should be drained and cleaned if the sludge buildup interferes with the treatment process.

**SEDIMENTATION:** The process of suspended solid particles settling out (going to the bottom of the vessel) in water.

**SEDIMENT:** Grains of soil, sand, gravel, or rock deposited by and generated by water movement.

**SENSOR:** A float and cable system are commonly found instruments that may be used as a sensor to control the level of liquid in a tank or basin.

**SEPTIC:** Condition characterized by bacterial decomposition under anaerobic conditions.

**SETTLEABILITY:** The tendency of suspended solids to settle.

**SETTLEABLE SOLIDS:** That portion of suspended solids which are of a sufficient size and weight to settle to the bottom of an Imhoff cone in one hour.

**SETTLED SLUDGE VOLUME:** Volume of settled sludge measured at predetermined time increments for use in process control calculations.

**SETTLED SOLIDS:** Solids that have been removed from the raw water by the coagulation and settling processes.

**SEWAGE:** Liquid or waterborne wastes polluted or fouled from households, commercial or industrial operations, along with any surface water, storm water or groundwater infiltration.

**SEWER GAS:** A gas mixture produced by anaerobic decomposition of organic matter usually containing high percentages of methane and hydrogen sulfide.

**SHEATHED:** Filamentous, gram-negative, aerobic; "swarmer" (colonizing) cells form and break out of a sheath; sometimes coated with metals from environment.

**SHOCK:** Also known as superchlorination or break point chlorination. Ridding a water of organic waste through oxidization by the addition of significant quantities of a halogen.

**SHOCK LOAD:** A sudden hydraulic or organic load to a treatment plant, also descriptive of a change in the material being treated.

**SHORT-CIRCUITING:** Short Circuiting is a condition that occurs in tanks or basins when some of the water travels faster than the rest of the flowing water. This is usually undesirable since it may result in shorter contact, reaction or settling times in comparison with the presumed detention times.

**SHOULD:** This action, activity, or procedural step is suggested but not required.

**SINGLE PHASE POWER:** The type of power used for lighting systems, small motors, appliances, portable power tools and in homes.

**SLOP OIL:** Separator skimmings and tramp oil generated during refinery startup, shutdown or abnormal operation.

**SLUDGE:** Accumulated and concentrated solids generated within a treatment process that have not undergone a stabilization process.

**SLUDGE BASINS:** After cleaning sludge basins and before returning the tanks into service the tanks should be inspected, repaired if necessary, and disinfected.

**SLUDGE BLANKET:** The accumulated sludge suspended in a clarifier or other enclosed body of water.

**SLUDGE DEWATERING:** The removal of a portion or majority of the water contained in sludge by means of a filter press, centrifuge or other mechanism.

**SLUDGE DRYING BED:** A closed area consisting of sand or other porous material upon which sludge is dewatered by gravity drainage and evaporation.

**SLUDGE REDUCTION:** Organic polymers are used to reduce the quantity of sludge. If a plant produces a large volume of sludge, the sludge could be dewatered, thickened, or conditioned to decrease the volume of sludge. Turbidity of source water, dosage, and type of coagulant used are the most important factors which determine the amount of sludge produced in a treatment of water.

**SLURRY:** A mixture of a solid and a liquid that facilitates the transfer of the solid into a treatment solution.

**SOC:** A common way for a synthetic organic chemical such as dioxin to be introduced to a surface water supply is from an industrial discharge, agricultural drainage, or a spill.

**SODA ASH:** Chemical used to raise pH and total alkalinity (sodium carbonate)

**SODIUM BICARBONATE:** Commonly used to increase alkalinity of water and stabilize pH.

**SODIUM BISULFATE:** Chemical used to lower pH and total alkalinity (dry acid).

**SODIUM HYDROXIDE:** Also known as caustic soda, a by-product chlorine generation and often used to raise pH.

**SOFTENING WATER:** When the water has a low alkalinity it is advantageous to use soda ash instead of caustic soda for softening water.

**SOFTENING:** The process that removes the ions which cause hardness in water.

**SOLID, LIQUID AND VAPOR:** 3 forms of matter.

**SOLID WASTE:** Garbage, refuse, sludge and other discarded material resulting from community activities or commercial or industrial operations.

**SOLUBILITY:** The amount of a substance that can dissolve in a solution under a given set of conditions.

**SPADNS:** The lab reagent called SPADNS solution is used in performing the Fluoride test.

**SPIKING SUSPENSION:** Diluted stock suspension containing the organism(s) of interest at a concentration appropriate for spiking samples.

**SPIRILLUM:** Spiral-shaped, gram-negative, aerobic; include Bdellovibrio, predatory on other bacteria.

**SPIROCHETE:** Spiral-shaped, gram-negative, mostly anaerobic; common in moist environments, from mammalian gums to coastal mudflats; complex internal structures convey rapid movement; include *Treponema pallidum*, cause of syphilis.

**SPOROZOITE:** A motile, infective stage of certain protozoans; e.g., *Cryptosporidium*. There are four sporozoites in each *Cryptosporidium* oocyst, and they are generally banana-shaped.

**SPRAY BOTTLE OF AMMONIA:** An operator should use ammonia to test for a chlorine leak around a valve or pipe. You will see white smoke if there is a leak.

**SPRING PRESSURE:** Is what maintains contact between the two surfaces of a mechanical seal.

**STABILIZATION POND:** A large shallow basin used for wastewater treatment by natural processes involving the use of algae and bacteria to accomplish biological oxidation of organic matter.

**STERILIZED GLASSWARE:** The only type of glassware that should be used in testing for coliform bacteria.

**STOCK SUSPENSION:** A concentrated suspension containing the organism(s) of interest that is obtained from a source that will attest to the host source, purity, authenticity, and viability of the organism(s).

**STUFFING BOX:** That portion of the pump that houses the packing or mechanical seal.

**SUBNATANT:** Liquid remaining beneath the surface of floating solids.

**SUCCESSION:** Transition in the species composition of a biological community, often following ecological disturbance of the community; the establishment of a biological community in an area virtually barren of life.

**SULFIDE:** The term sulfide refers to several types of chemical compounds containing sulfur in its lowest oxidation number of -2. Formally, "sulfide" is the dianion, S<sub>2</sub><sup>-</sup>, which exists in strongly alkaline aqueous solutions formed from H<sub>2</sub>S or alkali metal salts such as Li<sub>2</sub>S, Na<sub>2</sub>S, and K<sub>2</sub>S. Sulfide is exceptionally basic and, with a pKa > 14, it does not exist in appreciable concentrations even in highly alkaline water, being undetectable at pH < ~15 (8 M NaOH). Instead, sulfide combines with electrons in hydrogen to form HS, which is variously called hydrogen sulfide ion, hydrosulfide ion, sulfhydryl ion, or bisulfide ion. At still lower pH's (<7), HS<sup>-</sup> converts to H<sub>2</sub>S, hydrogen sulfide. Thus, the exact sulfur species obtained upon dissolving sulfide salts depends on the pH of the final solution. Aqueous solutions of transition metals cations react with sulfide sources (H<sub>2</sub>S, NaSH, Na<sub>2</sub>S) to precipitate solid sulfides. Such inorganic sulfides typically have very low solubility in water and many are related to minerals. One famous example is the bright yellow species CdS or "cadmium yellow". The black tarnish formed on sterling silver is Ag<sub>2</sub>S. Such species are sometimes referred to as salts. In fact, the bonding in transition metal sulfides is highly covalent, which gives rise to their semiconductor properties, which in turn is related to the practical applications of many sulfide materials.

**SULFATE- AND SULFUR- REDUCING:** Commonly rod-shaped, mostly gram-negative, anaerobic; include *Desulfovibrio*, ecologically important in marshes.

**SULFUR- AND IRON- OXIDIZING:** Commonly rod-shaped, frequently with polar flagella, gram-negative, mostly anaerobic; most live in neutral (nonacidic) environment.

**SUPERNATANT:** The liquid layer which forms above the sludge in a settling basin.

**SURFACE SEAL:** The upper portion of a wells construction where surface contaminants are adequately prevented from entering the well, normally consisting of surface casing and neat cement grout.

**SURFACTANT:** Surfactants reduce the surface tension of water by adsorbing at the liquid-gas interface. They also reduce the interfacial tension between oil and water by adsorbing at the liquid-liquid interface. Many surfactants can also assemble in the bulk solution into aggregates. Examples of such aggregates are vesicles and micelles. The concentration at which surfactants begin to form micelles is known as the critical micelle concentration or CMC. When micelles form in water, their tails form a core that can encapsulate an oil droplet, and their (ionic/polar) heads form an outer shell that maintains favorable contact with water. When surfactants assemble in oil, the aggregate is referred to as a reverse micelle. In a reverse micelle, the heads are in the core and the tails maintain favorable contact with oil. Surfactants are also often classified into four primary groups; anionic, cationic, non-ionic, and zwitterionic (dual charge).

**SUSPENDED SOLIDS:** Solids captured by filtration through a 0.45 micron filter membrane.

**TCE, trichloroethylene:** A solvent and degreaser used for many purposes; for example dry cleaning, it is a common groundwater contaminant. Trichloroethylene is a colorless liquid which is used as a solvent for cleaning metal parts. Drinking or breathing high levels of trichloroethylene may cause nervous system effects, liver and lung damage, abnormal heartbeat, coma, and possibly death. Trichloroethylene has been found in at least 852 of the 1,430 National Priorities List sites identified by the Environmental Protection Agency (EPA).

**TDS-TOTAL DISSOLVED SOLIDS:** An expression for the combined content of all inorganic and organic substances contained in a liquid which are present in a molecular, ionized or micro-granular (colloidal sol) suspended form. Generally, the operational definition is that the solids (often abbreviated TDS) must be small enough to survive filtration through a sieve size of two micrometers. Total dissolved solids are normally only discussed for freshwater systems, since salinity comprises some of the ions constituting the definition of TDS. The principal application of TDS is in the study of water quality for streams, rivers and lakes, although TDS is generally considered not as a primary pollutant (e.g. it is not deemed to be associated with health effects), but it is rather used as an indication of aesthetic characteristics of drinking water and as an aggregate indicator of presence of a broad array of chemical contaminants.

**TDS:** Ion exchange is an effective treatment process used to remove iron and manganese in a water supply. This process is ideal as long as the water does not contain a large amount of TDS. When determining the total dissolved solids, a sample should be filtered before being poured into an evaporating dish and dried. Demineralization may be necessary in a treatment process if the water has a very high value Total Dissolved Solids.

**TELEMETERING:** The use of a transmission line with remote signaling to monitor a pumping station or motors. Can be used to accomplish accurate and reliable remote monitoring and control over a long distribution system.

**TEMPERATURE SAMPLE:** This test should be performed immediately in the field, a grab sample.

**TERTIARY TREATMENT:** The use of physical, chemical, or biological means to improve secondary wastewater effluent quality.

**THE RATE DECREASES:** In general, when the temperature decreases, the chemical reaction rate decreases also.

**THICKENING:** A procedure used to increase the solids content of sludge by removing a portion of the liquid.

**THICKENING, CONDITIONING AND DEWATERING:** Common processes that are utilized to reduce the volume of sludge.

**THOMAS MALTHUS:** Formulated the concept that population growth proceeds at a geometric rate.

**TIME FOR TURBIDITY BREAKTHROUGH AND MAXIMUM HEADLOSS:** Are the two factors which determine whether or not a change in filter media size should be made.

**TITRATION:** A method of testing by adding a reagent of known strength to a water sample until a specific color change indicates the completion of the reaction.

**TOTAL ALKALINITY:** A measure of the acid-neutralizing capacity of water which indicates its buffering ability, i.e. measure of its resistance to a change in pH. Generally, the higher the total alkalinity, the greater the resistance to pH change.

**TOTAL COLIFORM:** Total coliform, fecal coliform, and E. coli are all indicators of drinking water quality. The total coliform group is a large collection of different kinds of bacteria. Fecal coliforms are types of total coliform that mostly exist in feces. E. coli is a sub-group of fecal coliform. When a water sample is sent to a lab, it is tested for total coliform. If total coliform is present, the sample will also be tested for either fecal coliform or E. coli, depending on the lab testing method.

**TOTAL DISSOLVED SOLIDS (TDS):** The accumulated total of all solids that might be dissolved in water. The weight per unit volume of all volatile and non-volatile solids dissolved in a water or wastewater after a sample has been filtered to remove colloidal and suspended solids.

**TOTAL DYNAMIC HEAD:** The pressure (psi) or equivalent feet of water, required for a pump to lift water to its point of storage overcoming elevation head, friction loss, line pressure, drawdown and pumping lift.

**TOTAL SOLIDS:** The sum of dissolved and suspended solids in a water or wastewater.

**TOTAL SUSPENDED SOLIDS:** The measure of particulate matter suspended in a sample of water or wastewater.

**TOXIC:** Capable of causing an adverse effect on biological tissue following physical contact or absorption.

**TREATABILITY STUDY:** A study in which a waste is subjected to a treatment process to determine treatment and/or to determine the treatment efficiency or optimal process conditions for treatment.

**TRIHALOMETHANES (THM):** Four separate compounds including chloroform, dichlorobromomethane, dibromochloromethane, and bromoform. The most common class of disinfection by-products created when chemical disinfectants react with organic matter in water during the disinfection process. See Disinfectant Byproducts.

**TUBE SETTLERS:** This modification of the conventional process contains many metal tubes that are placed in the sedimentation basin, or clarifier. These tubes are approximately 1 inch deep and 36 inches long, split-hexagonal shape and installed at an angle of 60 degrees or less. These tubes provide for a very large surface area upon which particles may settle as the water flows upward. The slope of the tubes facilitates gravity settling of the solids to the bottom of the basin, where they can be collected and removed. The large surface settling area also means that adequate clarification can be obtained with detention times of 15 minutes or less. As with conventional treatment, this sedimentation step is followed by filtration through mixed media.

**TUBERCLES:** The creation of this condition is of the most concern regarding corrosive water effects on a water system. Tubercles are formed due to joining dissimilar metals, causing electro-chemical reactions. Like iron to copper pipe. We have all seen these little rust mounds inside cast iron pipe.

**TURBIDIMETER:** Monitoring the filter effluent turbidity on a continuous basis with an in-line instrument is a recommended practice. Turbidimeter is best suited to perform this measurement.

**TURBIDITY:** A measure of the cloudiness of water caused by suspended particles. A qualitative measurement of water clarity which results from suspended matter that scatters or otherwise interferes with the passage of light through the water.

**ULTRAFILTRATION:** A low pressure membrane filtration process which separates solutes up to 0.1 micron size range.

**UNDER PRESSURE IN STEEL CONTAINERS:** After chlorine gas is manufactured, it is primarily transported in steel containers.

**UP FLOW CLARIFIER:** Clarifier where flocculated water flows upward through a sludge blanket to obtain floc removal by contact with flocculated solids in the blanket.

**U.S. ENVIRONMENTAL PROTECTION AGENCY:** In the United States, this agency responsible for setting drinking water standards and for ensuring their enforcement. This agency sets federal regulations which all state and local agencies must enforce.

**VANE:** That portion of an impeller that throws the water toward the volute.

**VAPOR:** The gaseous phase of a material that is in the solid or liquid state at standard temperature and pressure.

**VARIABLE DISPLACEMENT PUMP:** A pump that will produce different volumes of water dependent on the pressure head against it.

**VELOCITY HEAD:** The vertical distance a liquid must fall to acquire the velocity with which it flows through the piping system. For a given quantity of flow, the velocity head will vary indirectly as the pipe diameter varies.

**VENTURI:** If water flows through a pipeline at a high velocity, the pressure in the pipeline is reduced. Velocities can be increased to a point that a partial vacuum is created.

**VERTICAL TURBINE:** A type of variable displacement pump in which the motor or drive head is mounted on the wellhead and rotates a drive shaft connected to the pump impellers.

**VIBRIO:** Rod- or comma-shaped, gram-negative, aerobic; commonly with a single flagellum; include *Vibrio cholerae*, cause of cholera, and luminescent forms symbiotic with deep-water fishes and squids.

**VIRUSES:** Very small disease-causing microorganisms that are too small to be seen even with microscopes. Viruses cannot multiply or produce disease outside of a living cell.

**VITRIFICATION:** Vitrification is a process of converting a material into a glass-like amorphous solid that is free from any crystalline structure, either by the quick removal or addition of heat, or by mixing with an additive. Solidification of a vitreous solid occurs at the glass transition temperature (which is lower than melting temperature,  $T_m$ , due to supercooling). When the starting material is solid, vitrification usually involves heating the substances to very high temperatures. Many ceramics are produced in such a manner. Vitrification may also occur naturally when lightning strikes sand, where the extreme and immediate heat can create hollow, branching rootlike structures of glass, called fulgurite. When applied to whiteware ceramics, vitreous means the material has an extremely low permeability to liquids, often but not always water, when determined by a specified test regime. The microstructure of whiteware ceramics frequently contain both amorphous and crystalline phases.

**VOLATILE:** A substance that evaporates or vaporizes at a relatively low temperature.

**VOLATILE ORGANIC COMPOUNDS (VOCs):** Solvents used as degreasers or cleaning agents. Improper disposal of VOCs can lead to contamination of natural waters. VOCs tend to evaporate very easily. This characteristic gives VOCs very distinct chemical odors like gasoline, kerosene, lighter fluid, or dry cleaning fluid. Some VOCs are suspected cancer-causing agents. Volatile organic compounds (VOCs) are organic chemical compounds that have high enough vapor pressures under normal conditions to significantly vaporize and enter the atmosphere. A wide range of carbon-based molecules, such as aldehydes, ketones, and other light hydrocarbons are VOCs. The term often is used in a legal or regulatory context and in such cases the precise definition is a matter of law. These definitions can be contradictory and may contain "loopholes"; e.g. exceptions, exemptions, and exclusions. The United States Environmental Protection Agency defines a VOC as any organic compound that participates in a photoreaction; others believe this definition is very broad and vague as organics that are not volatile in the sense that they vaporize under normal conditions can be considered volatile by this EPA definition. The term may refer both to well characterized organic compounds and to mixtures of variable composition.

**VOID:** An opening, gap, or space within rock or sedimentary formations formed at the time of origin or deposition.

**VOLTAGE:** Voltage (sometimes also called electric or electrical tension) is the difference of electrical potential between two points of an electrical or electronic circuit, expressed in volts.[1] It measures the potential energy of an electric field to cause an electric current in an electrical conductor. Depending on the difference of electrical potential it is called extra low voltage, low voltage, high voltage or extra high voltage. Specifically Voltage is equal to energy per unit charge.

**VOLUTE:** The spiral-shaped casing surrounding a pump impeller that collects the liquid discharge by the impeller.

**VORTICELLA:** Vorticella is a genus of protozoa, with over 100 known species. They are stalked inverted bell-shaped ciliates, placed among the peritrichs. Each cell has a separate stalk anchored onto the substrate, which contains a contractile fibril called a myoneme. When stimulated this shortens, causing the stalk to coil like a spring. Reproduction is by budding, where the cell undergoes longitudinal fission and only one daughter keeps the stalk. Vorticella mainly lives in freshwater ponds and streams - generally anywhere protists are plentiful. Other genera such as Carchesium resemble Vorticella but are branched or colonial.

**VORTEX:** The helical swirling of water moving towards a pump.

**VIRUSES:** are very small disease-causing microorganisms that are too small to be seen even with microscopes. Viruses cannot multiply or produce disease outside of a living cell.

**VULNERABILITY ASSESSMENT:** An evaluation of drinking water source quality and its vulnerability to contamination by pathogens and toxic chemicals.

**WAIVERS:** Monitoring waivers for nitrate and nitrite are prohibited.

**WASTE ACTIVATED SLUDGE:** Excess activated sludge that is discharged from an activated sludge treatment process.

**WASTEWATER:** Liquid or waterborne wastes polluted or fouled from households, commercial or industrial operations, along with any surface water, storm water or groundwater infiltration.

**WATER HAMMER:** A surge in a pipeline resulting from the rapid increase or decrease in water flow. Water hammer exerts tremendous force on a system and can be highly destructive.

**WATER QUALITY CRITERIA:** Comprised of both numeric and narrative criteria. Numeric criteria are scientifically derived ambient concentrations developed by EPA or States for various pollutants of concern to protect human health and aquatic life. Narrative criteria are statements that describe the desired water quality goal.

**WATER QUALITY STANDARD:** A statute or regulation that consists of the beneficial designated use or uses of a waterbody, the numeric and narrative water quality criteria that are necessary to protect the use or uses of that particular waterbody, and an antidegradation statement.

**WATERBORNE DISEASE:** A disease, caused by a virus, bacterium, protozoan, or other microorganism, capable of being transmitted by water (e.g., typhoid fever, cholera, amoebic dysentery, gastroenteritis).

**WATER RECLAMATION:** The restoration of wastewater to a state that will allow its beneficial reuse.

**WHOLE EFFLUENT TOXICITY:** The total toxic effect of an effluent measured directly with a toxicity test.

**WPCF:** Water Pollution Control Facility

**WTP:** Water Treatment Plant

**WWTP:** Wastewater Treatment Plant

**ZERO DISCHARGE:** A facility that discharges no liquid effluent to the environment.

## Glossary References

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Foster, Laurence, M.D. 1985. "Waterborne Disease - It's Our Job to Prevent It". PIPELINE newsletter, Oregon Health Division, Drinking Water Program, Portland, Oregon 1(4): 1-3.

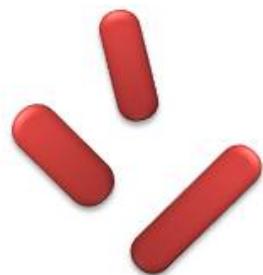
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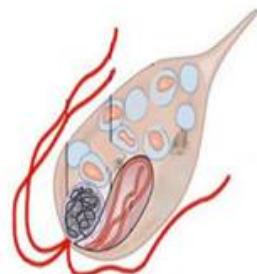
## Microorganism Appendix



**COCCI  
(SPHERICAL)**



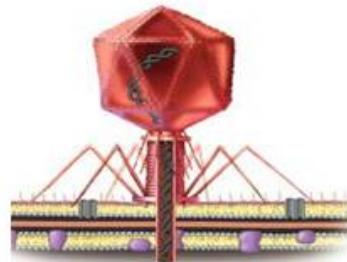
**BACILLI  
(ROD-SHAPED)**



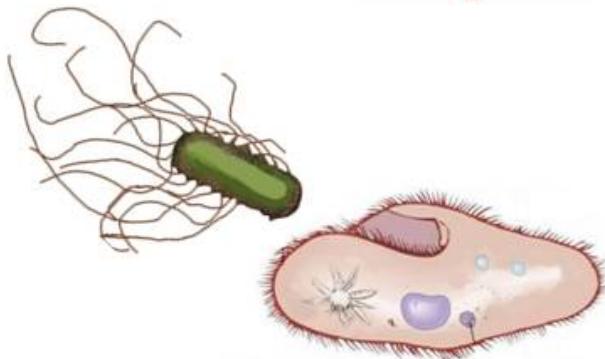
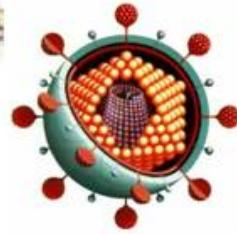
**PROTOZOA**



**SPIRILLI  
(SPIRAL)**



**VIRUSES**

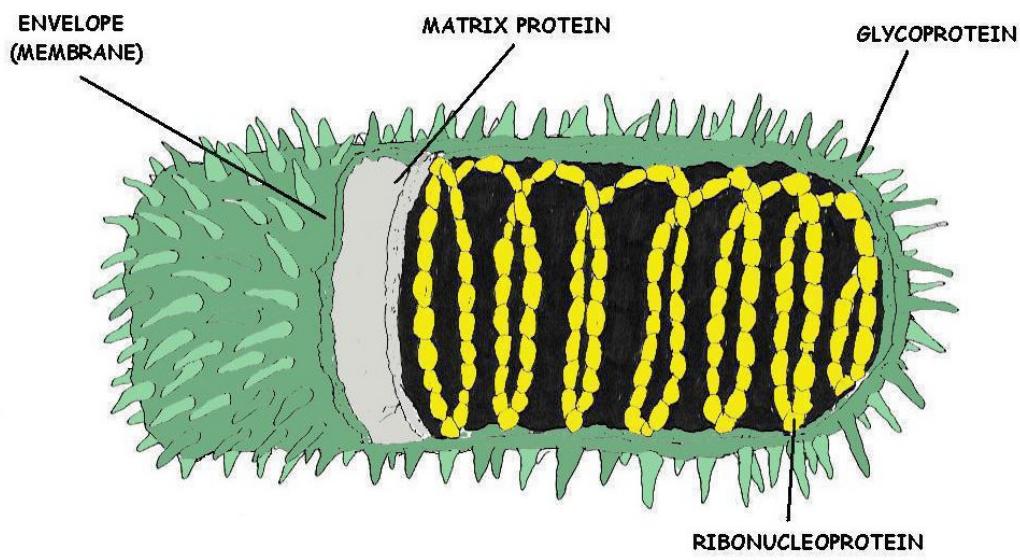


**BACTERIA**

## **BACTERIA TYPES**

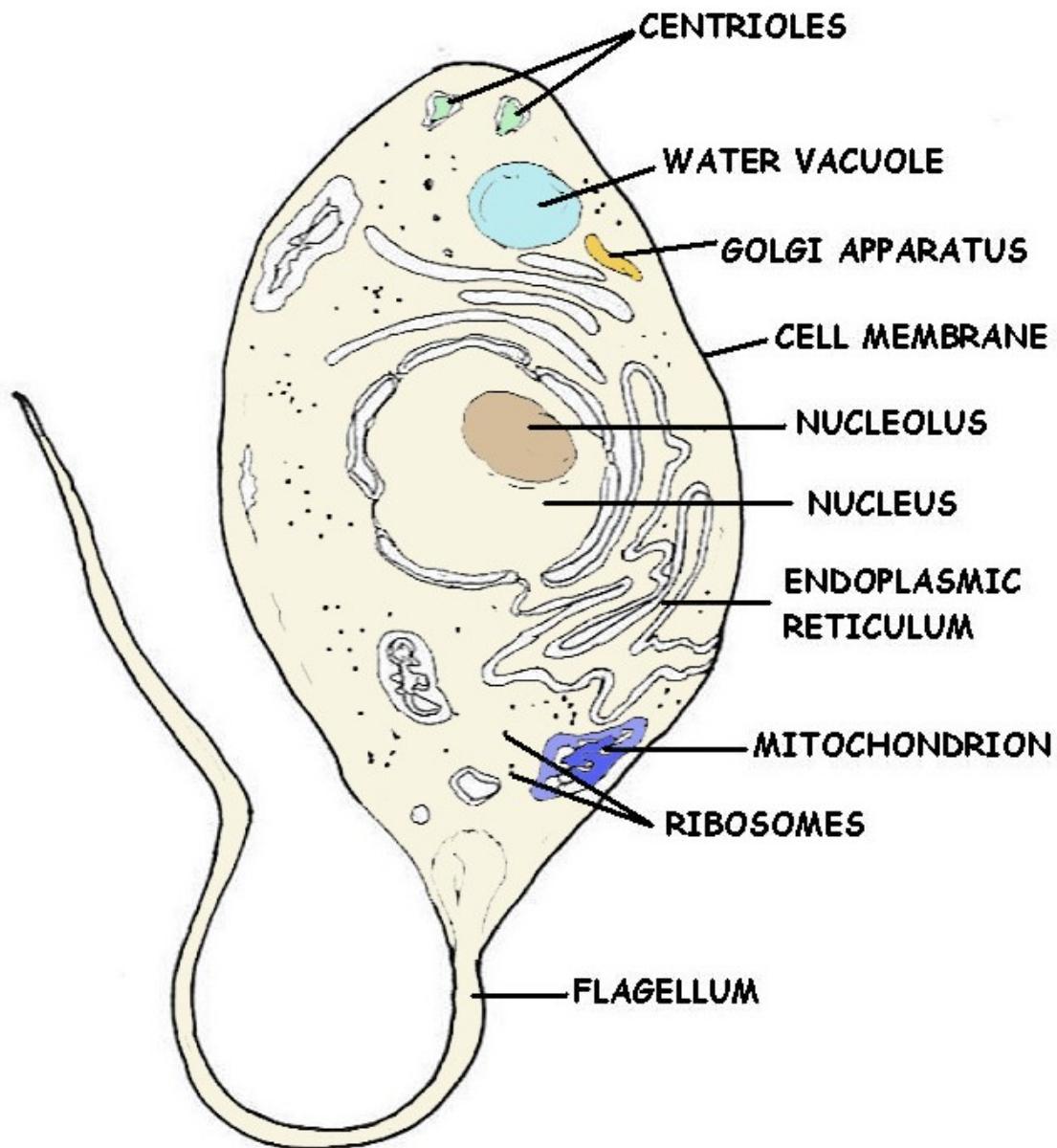


This section will give a close-up and short explanation of the major microorganisms found in water and in wastewater.



VIRUS

## Protozoa



## PROTOZOAN CELL

Protozoa are around 10–50 micrometer, but can grow up to 1 mm and can easily be seen under a microscope. Protozoa exist throughout aqueous environments and soil. Protozoa occupy a range of trophic levels. As predators, they prey upon unicellular or filamentous algae, bacteria, and microfungi.

Protozoa play a role both as herbivores and as consumers in the decomposer link of the food chain. Protozoa also play a vital role in controlling bacteria populations and biomass. As components of the micro- and meiofauna, protozoa are an important food source for microinvertebrates. Thus, the ecological role of protozoa in the transfer of bacterial and algal production to successive trophic levels is important. Protozoa such as the malaria parasites (*Plasmodium* spp.), trypanosomes and leishmania are also important as parasites and symbionts of multicellular animals.

Most protozoa exist in 5 stages of life which are in the form of trophozoites and cysts. As cysts, protozoa can survive harsh conditions, such as exposure to extreme temperatures and harmful chemicals, or long periods without access to nutrients, water, or oxygen for a period of time. Being a cyst enables parasitic species to survive outside of the host, and allows their transmission from one host to another. When protozoa are in the form of trophozoites (Greek, *tropho*=to nourish), they actively feed and grow. The process by which the protozoa takes its cyst form is called encystation, while the process of transforming back into trophozoite is called excystation.

Protozoa can reproduce by binary fission or multiple fission. Some protozoa reproduce sexually, some asexually, and some both (e.g. *Coccidia*). An individual protozoan is hermaphroditic.

### **Classification**

Protozoa were commonly grouped in the kingdom of Protista together with the plant-like algae and fungus-like water molds and slime molds. In the 21st-century systematics, protozoans, along with ciliates, mastigophorans, and apicomplexans, are arranged as animal-like protists. However, protozoans are neither Animalia nor Metazoa (with the possible exception of the enigmatic, moldy Myxozoa).

### **Sub-groups**

Protozoa have traditionally been divided on the basis of their means of locomotion, although this is no longer believed to represent genuine relationships:

- \* Flagellates (e.g. *Giardia lamblia*)
- \* Amoeboids (e.g. *Entamoeba histolytica*)
- \* Sporozoans (e.g. *Plasmodium knowlesi*)
- \* Apicomplexa
- \* Myxozoa
- \* Microsporidia
- \* Ciliates (e.g. *Balantidium coli*)

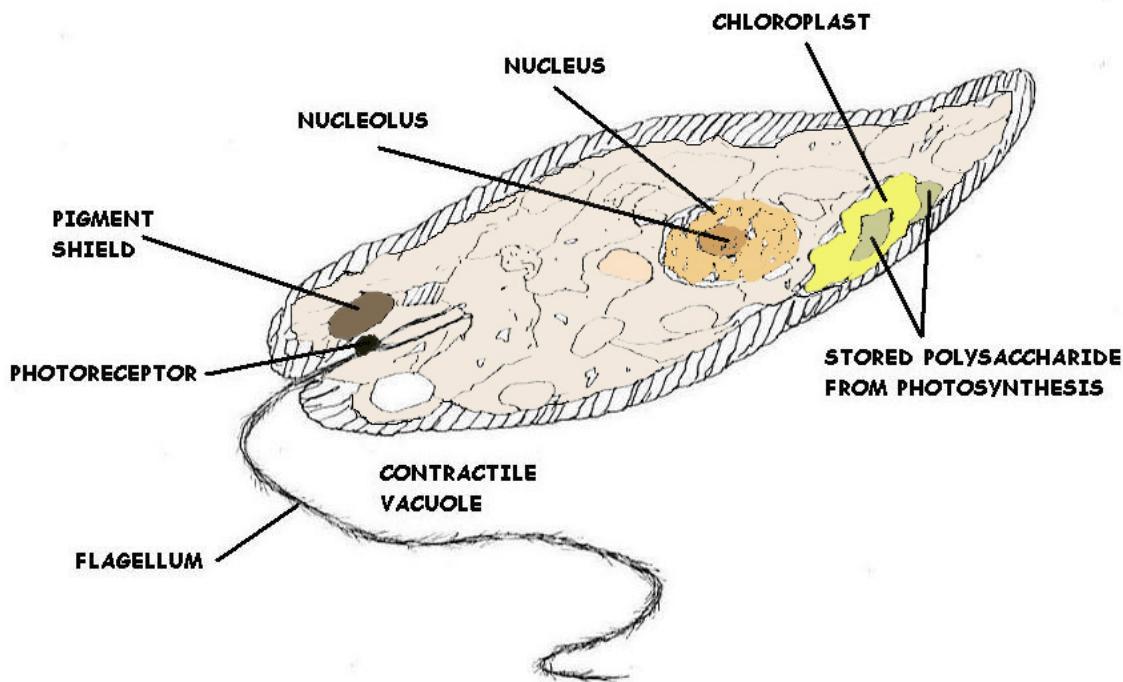
There are many ways that infectious diseases can spread. Pathogens usually have specific routes by which they are transmitted, and these routes may depend on the type of cells and tissue that a particular agent targets. For example, because cold viruses infect the respiratory tract, they are dispersed into the air via coughing and sneezing.

Once in the air, the viruses can infect another person who is unlucky enough to inhale air containing the virus particles.

Agents vary greatly in their stability in the environment. Some viruses may survive for only a few minutes outside of a host, while some spore-forming bacteria are extremely durable and may survive in a dormant state for a decade or more.

## Protozoa Section

### EUGLENA



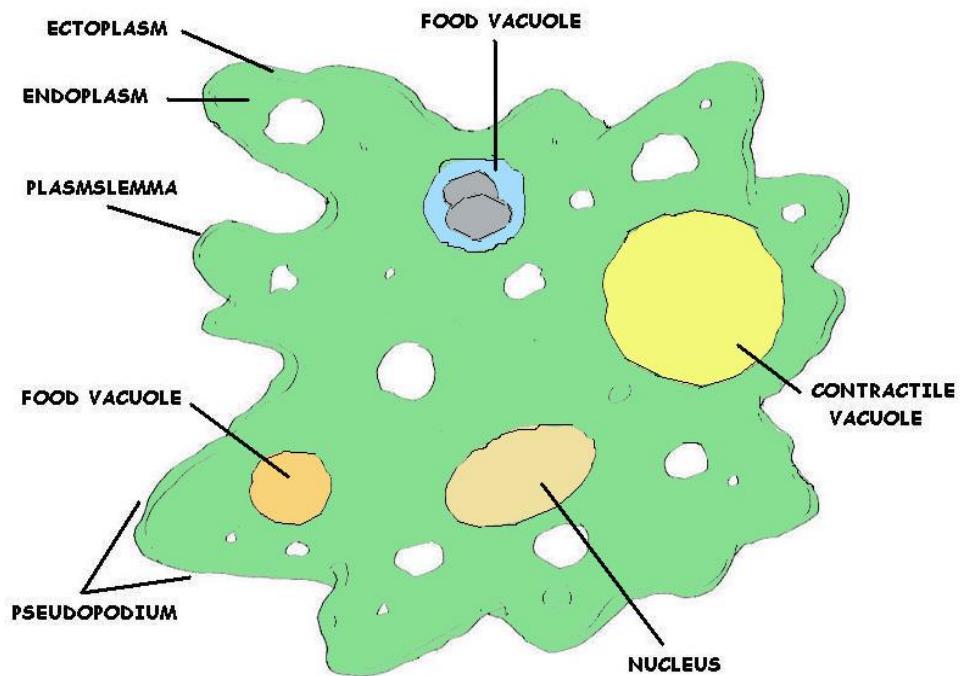
The diverse assemblage of organisms that carry out all of their life functions within the confines of a single, complex eukaryotic cell are called protozoa.

Paramecium, Euglena, and Amoeba are well-known examples of these major groups of organisms. Some protozoa are more closely related to animals, others to plants, and still others are relatively unique. Although it is not appropriate to group them together into a single taxonomic category, the research tools used to study any unicellular organism are usually the same, and the field of protozoology has been created to carry out this research. The unicellular photosynthetic protozoa are sometimes also called algae and are addressed elsewhere. This report considers the status of our knowledge of heterotrophic protozoa (protozoa that cannot produce their own food).

#### Free-living Protozoa

Protozoans are found in all moist habitats within the United States, but we know little about their specific geographic distribution. Because of their small size, production of resistant cysts, and ease of distribution from one place to another, many species appear to be cosmopolitan and may be collected in similar microhabitats worldwide (Cairns and Ruthven 1972). Other species may have relatively narrow limits to their distribution.

Marine ciliates inhabit interstices of sediment and beach sands, surfaces, deep sea and cold Antarctic environments, planktonic habitats, and the algal mats and detritus of estuaries and wetlands.



AMOEBA



*Amoeba proteus*, pseudopods slowly engulf the small desmid *Staurastrum*.

## Amoebas

Amoebas (Phylum Rhizopoda) are unicellular protists that are able to change their shape constantly. Each species has its own distinct repertoire of shapes.

### How does an amoeba locomote?

Amoebas locomote by way of cytoplasmic movement. (cytoplasm is the cell content around the nucleus of the cell) The amoeba forms pseudopods (false feet) with which they 'flow' over a surface. The cytoplasma not only flows, it also changes from a fluid into a solid state.

These pseudopods are also used to capture prey, they simply engulf the food. They can detect the kind of prey and use different 'engulfing tactics'.

The image from the last page shows several cell organelles. Left from the center we can see aspherical water expelling vesicle and just right of it, the single nucleus of this species can be seen. Other species may have many nuclei. The cell is full of brown food vacuoles and also contains small crystals.

### Protozoa Information

Our actual knowledge of salinity, temperature, and oxygen requirements of marine protozoa is poor (although some groups, such as the foraminifera, are better studied than others), and even the broadest outlines of their biogeographic ranges are usually a mystery. In general, freshwater protozoan communities are similar to marine communities except the specialized interstitial fauna of the sand is largely missing. In freshwater habitats, the foraminifera and radiolaria common in marine environments are absent or low in numbers while testate amoebae exist in greater numbers. Relative abundance of species in the marine versus freshwater habitat is unknown.

Soil-dwelling protozoa have been documented from almost every type of soil and in every kind of environment, from the peat-rich soil of bogs to the dry sands of deserts. In general, protozoa are found in greatest abundance near the soil surface, especially in the upper 15 cm (6 in), but occasional isolates can be obtained at depths of a meter (yard) or more.

Protozoa do not constitute a major part of soil biomass, but in some highly productive regions such as forest litter, the protozoa are a significant food source for the microinvertebrates, with a biomass that may reach 20 g/m<sup>2</sup> of soil surface area there.

### Environmental Quality Indicators

Polluted waters often have a rich and characteristic protozoan fauna. The relative abundance and diversity of protozoa are used as indicators of organic and toxic pollution (Cairns et al. 1972; Foissner 1987; Niederlehner et al. 1990; Curds 1992). Bick (1972), for example, provided a guide to ciliates that are useful as indicators of environmental quality of European freshwater systems, along with their ecological distribution with respect to parameters such as amount of organic material and oxygen levels.

Foissner (1988) clarified the taxonomy of European ciliates as part of a system for classifying the state of aquatic habitats according to their faunas.

## **Symbiotic Protozoa**

### **Parasites**

Protozoa are infamous for their role in causing disease, and parasitic species are among the best-known protozoa. Nevertheless, our knowledge has large gaps, especially of normally free-living protozoa that may become pathogenic in immunocompromised individuals. For example, microsporidia comprise a unique group of obligate, intracellular parasitic protozoa. Microsporidia are amazingly diverse organisms with more than 700 species and 80 genera that are capable of infecting a variety of plant, animal, and even other protist hosts.

They are found worldwide and have the ability to thrive in many ecological conditions. Until the past few years, their ubiquity did not cause a threat to human health, and few systematists worked to describe and classify the species. Since 1985, however, physicians have documented an unusual rise in worldwide infections in AIDS patients caused by four different genera of microsporidia (*Encephalitozoon*, *Nosema*, *Pleistophora*, and *Enterocytozoon*). According to the Centers for Disease Control in the United States, difficulties in identifying microsporidian species are impeding diagnosis and effective treatment of AIDS patients.

### **Protozoan Reservoirs of Disease**

The presence of bacteria in the cytoplasm of protozoa is well known, whereas that of viruses is less frequently reported. Most of these reports simply record the presence of bacteria or viruses and assume some sort of symbiotic relationship between them and the protozoa. Recently, however, certain human pathogens were shown to not only survive but also to multiply in the cytoplasm of free-living, nonpathogenic protozoa. Indeed, it is now believed that protozoa are the natural habitat for certain pathogenic bacteria. To date, the main focus of attention has been on the bacterium *Legionella pneumophila*, the causative organism of Legionnaires' disease; these bacteria live and reproduce in the cytoplasm of some free-living amoebae (Curds 1992). More on this subject in the following pages.

### **Symbionts**

Some protozoa are harmless or even beneficial symbionts. A bewildering array of ciliates, for example, inhabit the rumen and reticulum of ruminates and the cecum and colon of equids. Little is known about the relationship of the ciliates to their host, but a few may aid the animal in digesting cellulose.

### **Data on Protozoa**

While our knowledge of recent and fossil foraminifera in the U.S. coastal waterways is systematically growing, other free-living protozoa are poorly known. There are some regional guides and, while some are excellent, many are limited in scope, vague on specifics, or difficult to use. Largely because of these problems, most ecologists who include protozoa in their studies of aquatic habitats do not identify them, even if they do count and measure them for biomass estimates (Taylor and Sanders 1991).

Parasitic protozoa of humans, domestic animals, and wildlife are better known although no attempt has been made to compile this information into a single source. Large gaps in our knowledge exist, especially for haemogregarines, microsporidians, and myxosporidians (see Kreier and Baker 1987).

## **Museum Specimens**

For many plant and animal taxa, museums represent a massive information resource. This is not true for protozoa. In the United States, only the National Natural History Museum (Smithsonian Institution) has a reference collection preserved on microscope slides, but it does not have a protozoologist curator and cannot provide species' identification or verification services. The American Type Culture Collection has some protozoa in culture, but its collection includes relatively few kinds of protozoa.

## **Ecological Role of Protozoa**

Although protozoa are frequently overlooked, they play an important role in many communities where they occupy a range of trophic levels. As predators upon unicellular or filamentous algae, bacteria, and microfungi, protozoa play a role both as herbivores and as consumers in the decomposer link of the food chain. As components of the micro- and meiofauna, protozoa are an important food source for microinvertebrates. Thus, the ecological role of protozoa in the transfer of bacterial and algal production to successive trophic levels is important.

## **Factors Affecting Growth and Distribution**

Most free-living protozoa reproduce by cell division (exchange of genetic material is a separate process and is not involved in reproduction in protozoa). The relative importance for population growth of biotic versus chemical-physical components of the environment is difficult to ascertain from the existing survey data. Protozoa are found living actively in nutrient-poor to organically rich waters and in fresh water varying between 0°C (32°F) and 50°C (122°F). Nonetheless, it appears that rates of population growth increase when food is not constrained and temperature is increased (Lee and Fenchel 1972; Fenchel 1974; Montagnes et al. 1988).

Comparisons of oxygen consumption in various taxonomic groups show wide variation (Laybourn and Finlay 1976), with some aerobic forms able to function at extremely low oxygen tensions and to thereby avoid competition and predation.

Many parasitic and a few free-living species are obligatory anaerobes (grow without atmospheric oxygen). Of the free-living forms, the best known are the plagiopylid ciliates that live in the anaerobic sulfide-rich sediments of marine wetlands (Fenchel et al. 1977). The importance of plagiopylids in recycling nutrients to aerobic zones of wetlands is potentially great.

Because of the small size of protozoa, their short generation time, and (for some species) ease of maintaining them in the laboratory, ecologists have used protozoan populations and communities to investigate competition and predation.

The result has been an extensive literature on a few species studied primarily under laboratory conditions. Few studies have been extended to natural habitats with the result that we know relatively little about most protozoa and their roles in natural communities. Intraspecific competition for common resources often results in cannibalism, sometimes with dramatic changes in morphology of the cannibals (Giese 1973). Field studies of interspecific competition are few and most evidence for such species interactions is indirect (Cairns and Yongue 1977).

### Contractile Vacuoles

Many protozoa have contractile vacuoles, which collect and expel excess water, and extrusomes, which expel material used to deflect predators or capture prey. In multicellular organisms, hormones are often produced in vesicles. In higher plants, most of a cell's volume is taken up by a central vacuole or tonoplast, which maintains its osmotic pressure. Many eukaryotes have slender motile projections, usually called flagella when long and cilia when short. These are variously involved in movement, feeding, and sensation. These are entirely distinct from prokaryotic flagella. They are supported by a bundle of microtubules arising from a basal body, also called a kinetosome or centriole, characteristically arranged as nine doublets surrounding two singlets. Flagella also may have hairs or mastigonemes, scales, connecting membranes, and internal rods. Their interior is continuous with the cell's cytoplasm.

### Centrioles

Centrioles are often present even in cells and groups that do not have flagella. They generally occur in groups of one or two, called kinetids that give rise to various microtubular roots. These form a primary component of the cytoskeletal structure, and are often assembled over the course of several cell divisions, with one flagellum retained from the parent and the other derived from it. Centrioles may also be associated in the formation of a spindle during nuclear division. Some protists have various other microtubule-supported organelles. These include the radiolaria and heliozoa, which produce axopodia used in flotation or to capture prey, and the haptophytes, which have a peculiar flagellum-like organelle called the haptoneema.

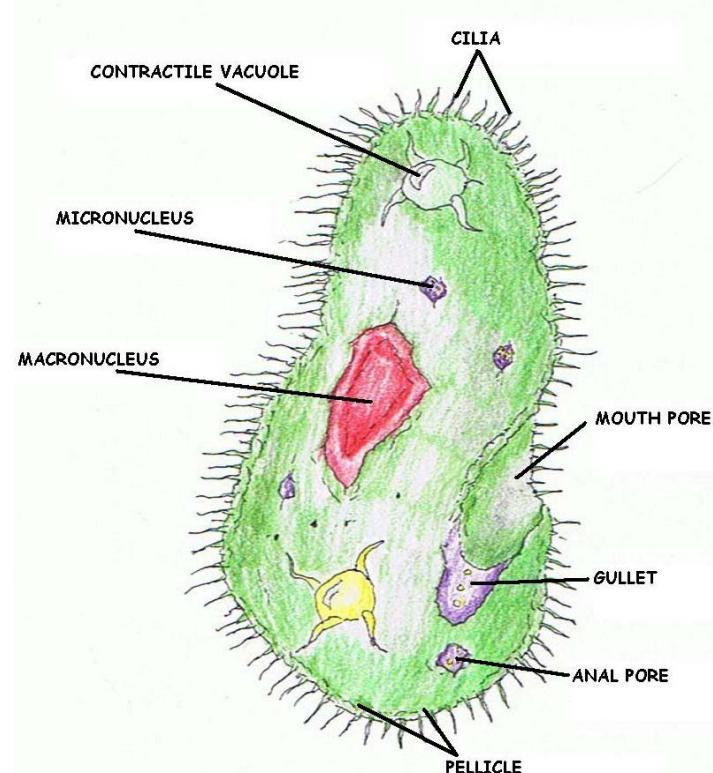
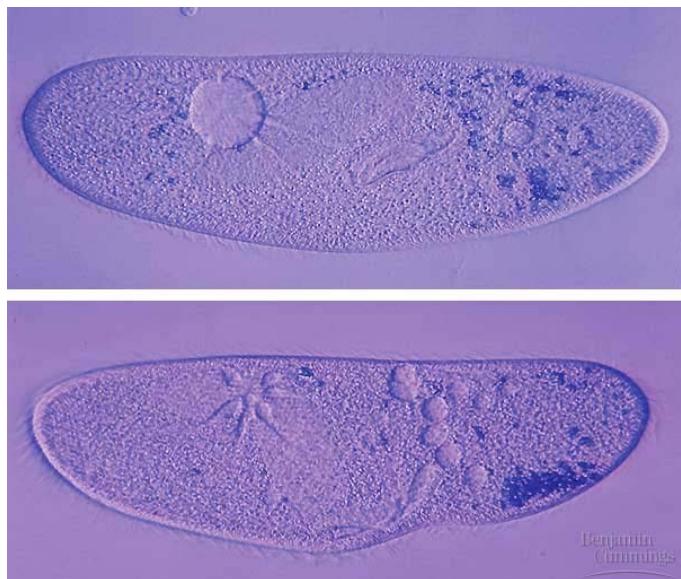


Figure 1. A diagram of *Paramecium* sp. with major organelles indicated.



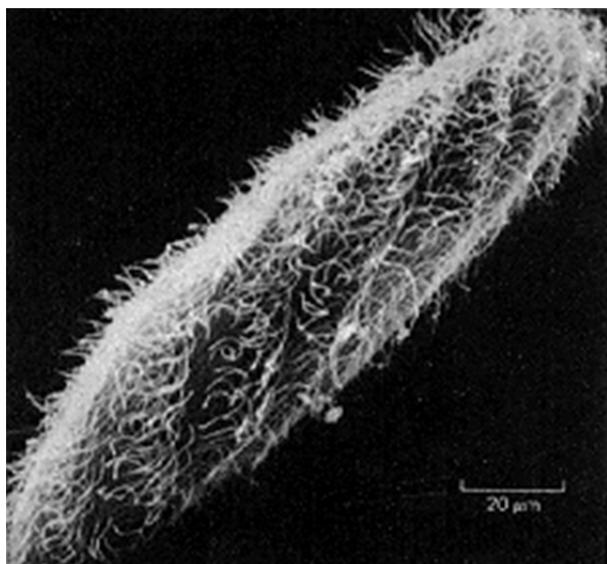
### Contractile Vacuoles

**Figure 2.** The contractile vacuole when full (top) and after contraction (bottom).

#### Paramecium

Members of the genus *Paramecium* are single-celled, freshwater organisms in the kingdom Protista. They exist in an environment in which the osmotic concentration in their external environment is much lower than that in their cytoplasm. More specifically, the habitat in which they live is **hypotonic** to their cytoplasm. As a result of this, *Paramecium* is subjected to a continuous influx of water, as water diffuses inward to a region of higher osmotic concentration.

If *Paramecium* is to maintain homeostasis, water must be continually pumped out of the cell (against the osmotic gradient) at the same rate at which it moves in. This process, known as **osmoregulation**, is carried out by two organelles in *Paramecium* known as **contractile vacuoles** (Figures 1 and 2).





Here is a wastewater Inspector is utilizing the auto sampler's manual to help adjust the time and adjust for the correct flow for a composite sample. You can see that she is pouring off the pickle jar. This inspector is also the course author.

## Protozoan Diseases

Protozoan pathogens are larger than bacteria and viruses, but still microscopic. They invade and inhabit the gastrointestinal tract. Some parasites enter the environment in a dormant form, with a protective cell wall called a “**cyst**.” The cyst can survive in the environment for long periods of time and be extremely resistant to conventional disinfectants such as chlorine. Effective filtration treatment is therefore critical to removing these organisms from water sources.

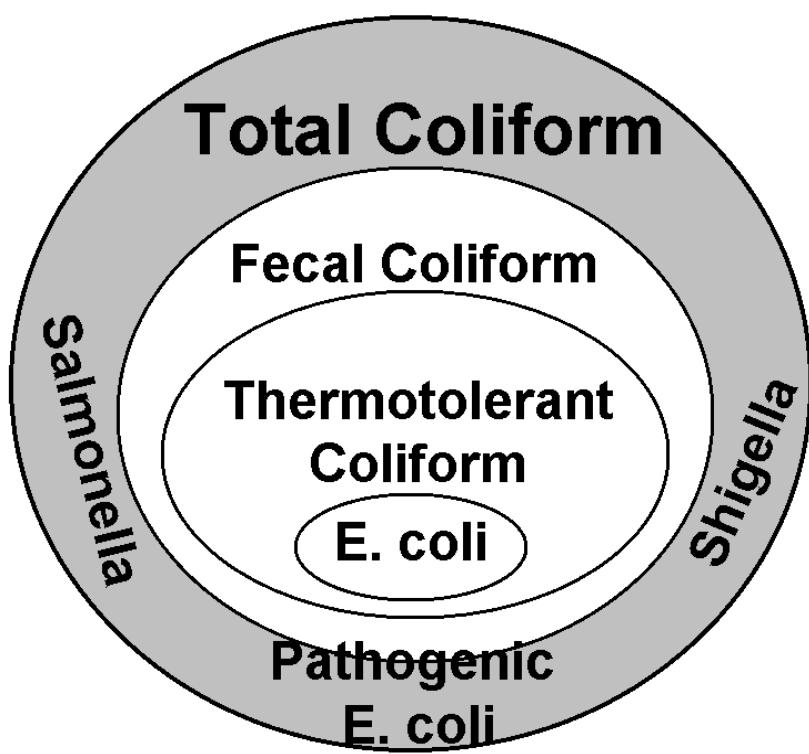
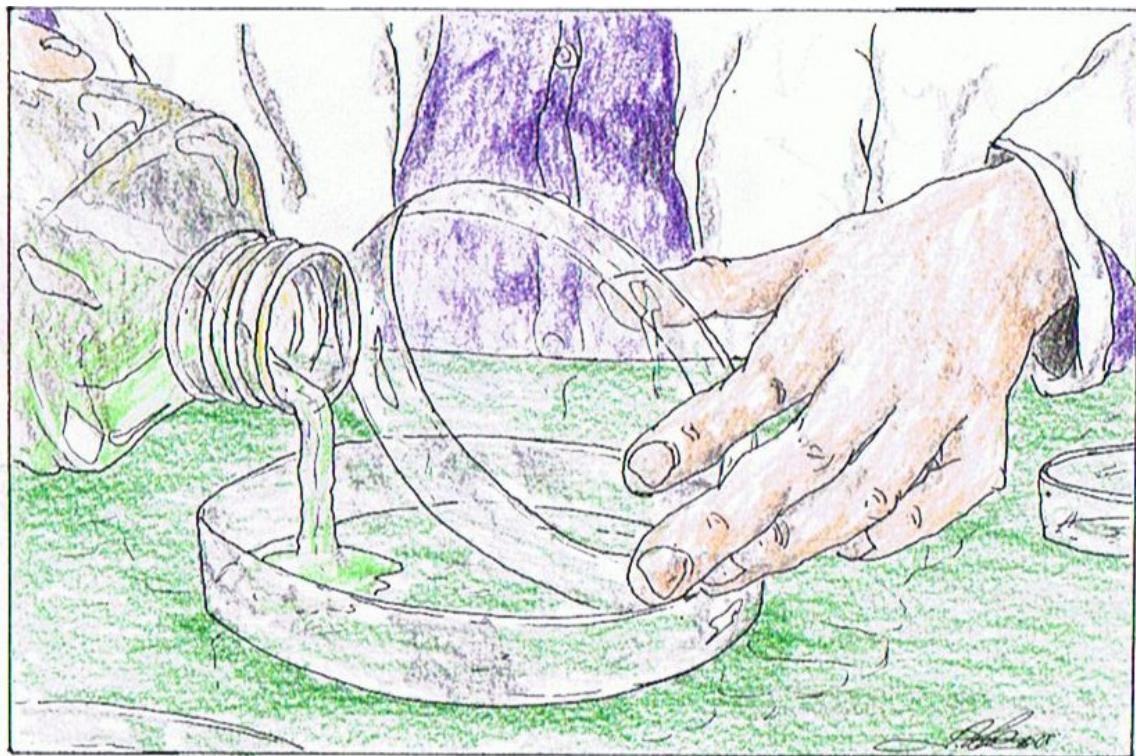
### Giardiasis

Giardiasis is a commonly reported protozoan-caused disease. It has also been referred to as “**backpacker’s disease**” and “**beaver fever**” because of the many cases reported among hikers and others who consume untreated surface water. Symptoms include chronic diarrhea, abdominal cramps, bloating, frequent loose and pale greasy stools, fatigue and weight loss. The incubation period is 5-25 days or longer, with an average of 7-10 days. Many infections are asymptomatic (no symptoms). Giardiasis occurs worldwide. Waterborne outbreaks in the United States occur most often in communities receiving their drinking water from streams or rivers without adequate disinfection or a filtration system. The organism, ***Giardia lamblia***, has been responsible for more community-wide outbreaks of disease in the U.S. than any other pathogen. Drugs are available for treatment but are not 100% effective.

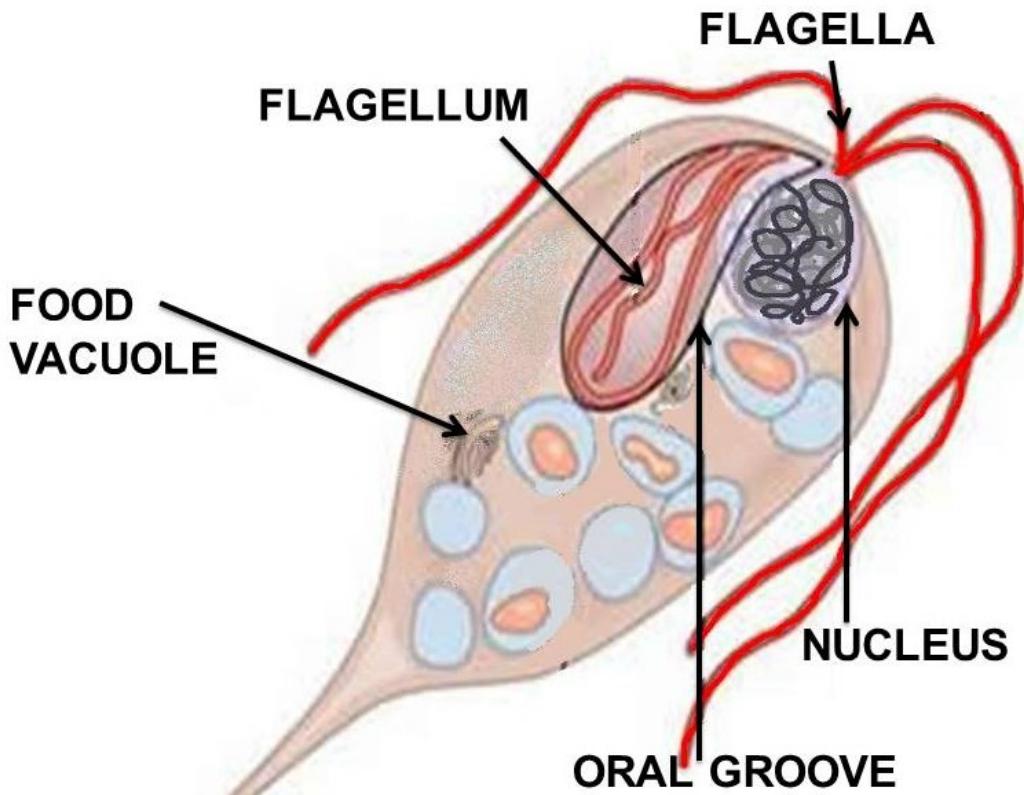
### Cryptosporidiosis

Cryptosporidiosis is an example of a protozoan disease that is common worldwide, but was only recently recognized as causing human disease. The major symptom in humans is diarrhea, which may be profuse and watery. The diarrhea is associated with cramping abdominal pain. General malaise, fever, anorexia, nausea, and vomiting occur less often. Symptoms usually come and go, and end in fewer than 30 days in most cases. The incubation period is 1-12 days, with an average of about seven days. ***Cryptosporidium*** organisms have been identified in human fecal specimens from more than 50 countries on six continents. The mode of transmission is fecal-oral, either by person-to-person or animal-to-person. There is no specific treatment for ***Cryptosporidium*** infections.

All of these diseases, with the exception of hepatitis A, have one symptom in common: diarrhea. They also have the same mode of transmission, fecal-oral, whether through person-to-person or animal-to-person contact, and the same routes of transmission, being either foodborne or waterborne. Although most pathogens cause mild, self-limiting disease, on occasion, they can cause serious, even life threatening illness. Particularly vulnerable are persons with weak immune systems such as those with HIV infections or cancer. By understanding the nature of waterborne diseases, the importance of properly constructed, operated and maintained public water systems becomes obvious. While water treatment cannot achieve sterile water (no microorganisms), the goal of treatment must clearly be to produce drinking water that is as pathogen-free as possible at all times. For those who operate water systems with inadequate source protection or treatment facilities, the potential risk of a waterborne disease outbreak is real. For those operating systems that currently provide adequate source protection and treatment, operating and maintaining the system at a high level on a continuing basis is critical to prevent disease.



## Giardia Lamblia



## GIARDIA LAMBLIA

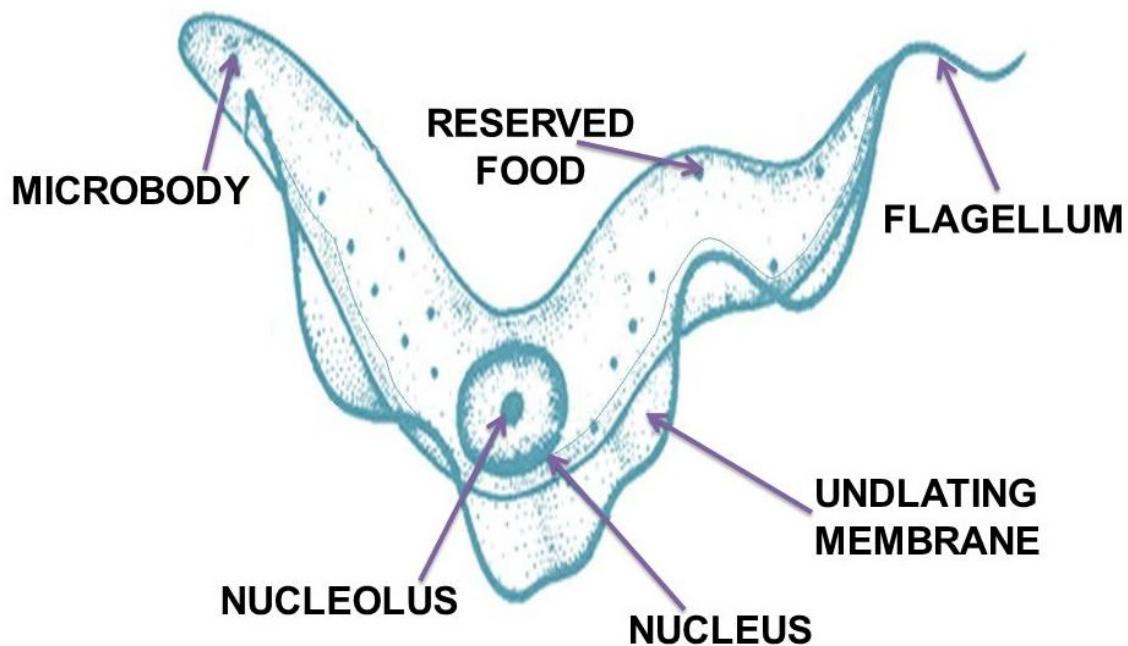
Giardia lamblia (synonymous with *Lamblia intestinalis* and *Giardia duodenalis*) is a flagellated protozoan parasite that colonizes and reproduces in the small intestine, causing giardiasis. The giardia parasite attaches to the epithelium by a ventral adhesive disc, and reproduces via binary fission. Giardiasis does not spread via the bloodstream, nor does it spread to other parts of the gastro-intestinal tract, but remains confined to the lumen of the small intestine. Giardia trophozoites absorb their nutrients from the lumen of the small intestine, and are anaerobes.

Giardia infection can occur through ingestion of dormant cysts in contaminated water, or by the fecal-oral route (through poor hygiene practices). The Giardia cyst can survive for weeks to months in cold water and therefore can be present in contaminated wells and water systems, and even clean-looking mountain streams, as well as city reservoirs, as the Giardia cysts are resistant to conventional water treatment methods, such as chlorination and ozonolysis. Zoonotic transmission is also possible, and therefore Giardia infection is a concern for people camping in the wilderness or swimming in contaminated streams or lakes, especially the artificial lakes formed by beaver dams (hence the popular name for giardiasis, "Beaver Fever"). As well as water-borne sources, fecal-oral transmission can also occur, for example in day care centers, where children may have poorer hygiene practices.

Those who work with children are also at risk of being infected, as are family members of infected individuals. Not all Giardia infections are symptomatic, so some people can unknowingly serve as carriers of the parasite.

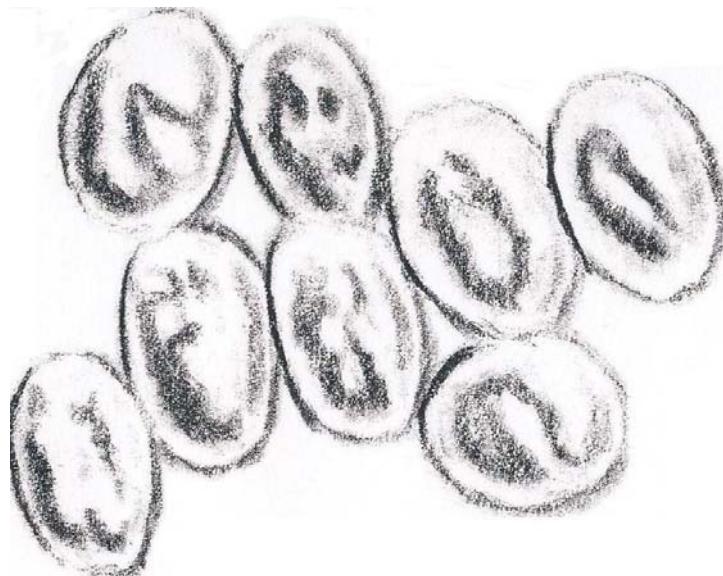
The life cycle begins with a non-infective cyst being excreted with feces of an infected individual. Once out in the environment, the cyst becomes infective. A distinguishing characteristic of the cyst is 4 nuclei and a retracted cytoplasm. Once ingested by a host, the trophozoite emerges to an active state of feeding and motility. After the feeding stage, the trophozoite undergoes asexual replication through longitudinal binary fission. The resulting trophozoites and cysts then pass through the digestive system in the feces. While the trophozoites may be found in the feces, only the cysts are capable of surviving outside of the host.

Distinguishing features of the trophozoites are large karyosomes and lack of peripheral chromatin, giving the two nuclei a halo appearance. Cysts are distinguished by a retracted cytoplasm. This protozoa lacks mitochondria, although the discovery of the presence of mitochondrial remnant organelles in one recent study "indicate that Giardia is not primitively amitochondrial and that it has retained a functional organelle derived from the original mitochondrial endosymbiont"



## PROTOZOAN PARASITE

## Cryptosporidium

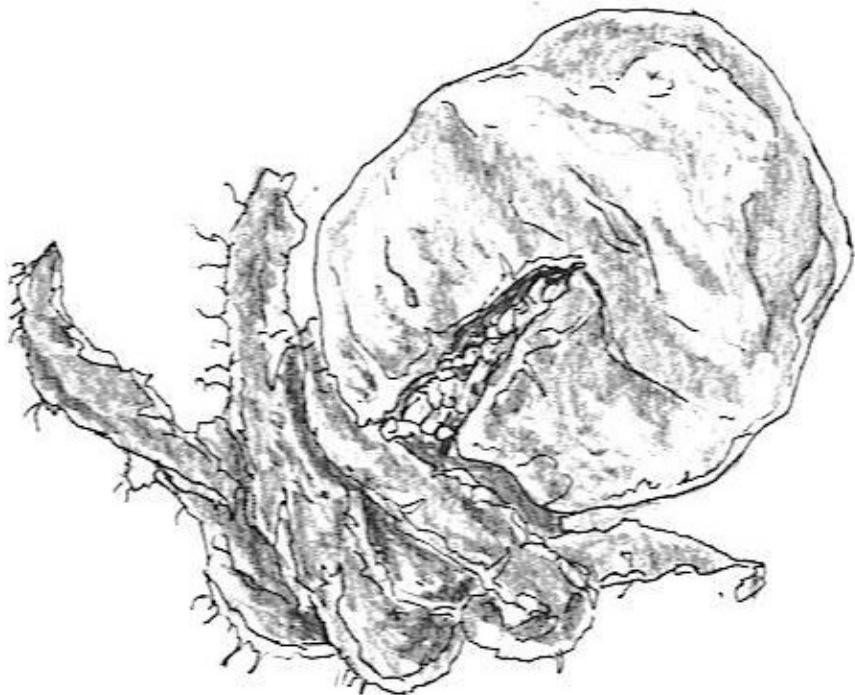


Cryptosporidium is a protozoan pathogen of the Phylum Apicomplexa and causes a diarrheal illness called cryptosporidiosis. Other apicomplexan pathogens include the malaria parasite Plasmodium, and Toxoplasma, the causative agent of toxoplasmosis. Unlike Plasmodium, which transmits via a mosquito vector, Cryptosporidium does not utilize an insect vector and is capable of completing its life cycle within a single host, resulting in cyst stages which are excreted in feces and are capable of transmission to a new host.

A number of species of Cryptosporidium infect mammals. In humans, the main causes of disease are *C. parvum* and *C. hominis* (previously *C. parvum* genotype 1). *C. canis*, *C. felis*, *C. meleagridis*, and *C. muris* can also cause disease in humans. In recent years, cryptosporidiosis has plagued many commercial Leopard gecko breeders. Several species of the Cryptosporidium family (*C. serpentis* and others) are involved, and outside of geckos it has been found in monitor lizards, iguanas, tortoises as well as several snake species.

Cryptosporidiosis is typically an acute short-term infection but can become severe and non-resolving in children and immunocompromised individuals. The parasite is transmitted by environmentally hardy cysts (oocysts) that, once ingested, excyst in the small intestine and result in an infection of intestinal epithelial tissue.

The genome of *Cryptosporidium parvum* was sequenced in 2004 and was found to be unusual amongst Eukaryotes in that the mitochondria seem not to contain DNA. A closely-related species, *C. hominis*, also has its genome sequence available. CryptoDB.org is a NIH-funded database that provides access to the *Cryptosporidium* genomics data sets.



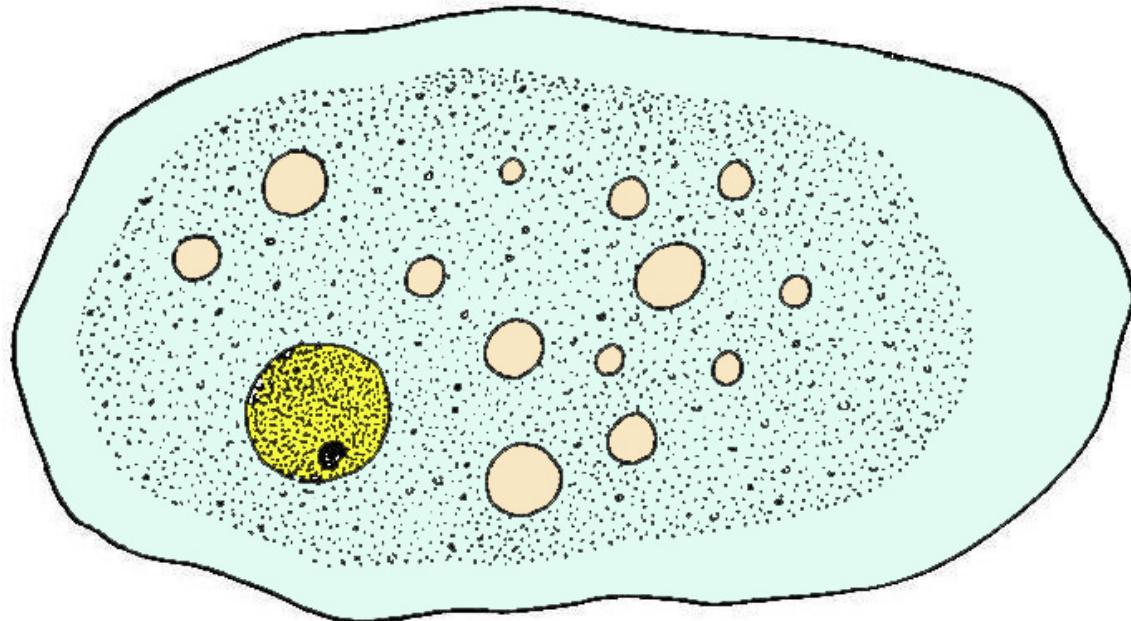
## CRYPTO-PARVUM

When *C. parvum* was first identified as a human pathogen, diagnosis was made by a biopsy of intestinal tissue (Keusch, *et al.*, 1995). However, this method of testing can give false negatives due the "patchy" nature of the intestinal parasitic infection (Flanigan and Soave, 1993). Staining methods were then developed to detect and identify the oocysts directly from stool samples. The modified acid-fast stain is traditionally used to most reliably and specifically detect the presence of cryptosporidial oocysts.

There have been six major outbreaks of cryptosporidiosis in the United States as a result of contamination of drinking water (Juranek, 1995). One major outbreak in Milwaukee in 1993 affected over 400,000 persons. Outbreaks such as these usually result from drinking water taken from surface water sources such as lakes and rivers (Juranek, 1995). Swimming pools and water park wave pools have also been associated with outbreaks of cryptosporidiosis. Also, untreated groundwater or well water public drinking water supplies can be sources of contamination.

The highly environmentally resistant cyst of *C. parvum* allows the pathogen to survive various drinking water filtrations and chemical treatments such as chlorination. Although municipal drinking water utilities may meet federal standards for safety and quality of drinking water, complete protection from cryptosporidial infection is not guaranteed. In fact, *all* waterborne outbreaks of cryptosporidiosis have occurred in communities where the local utilities met all state and federal drinking water standards (Juranek, 1995).

## Entamoeba histolytica

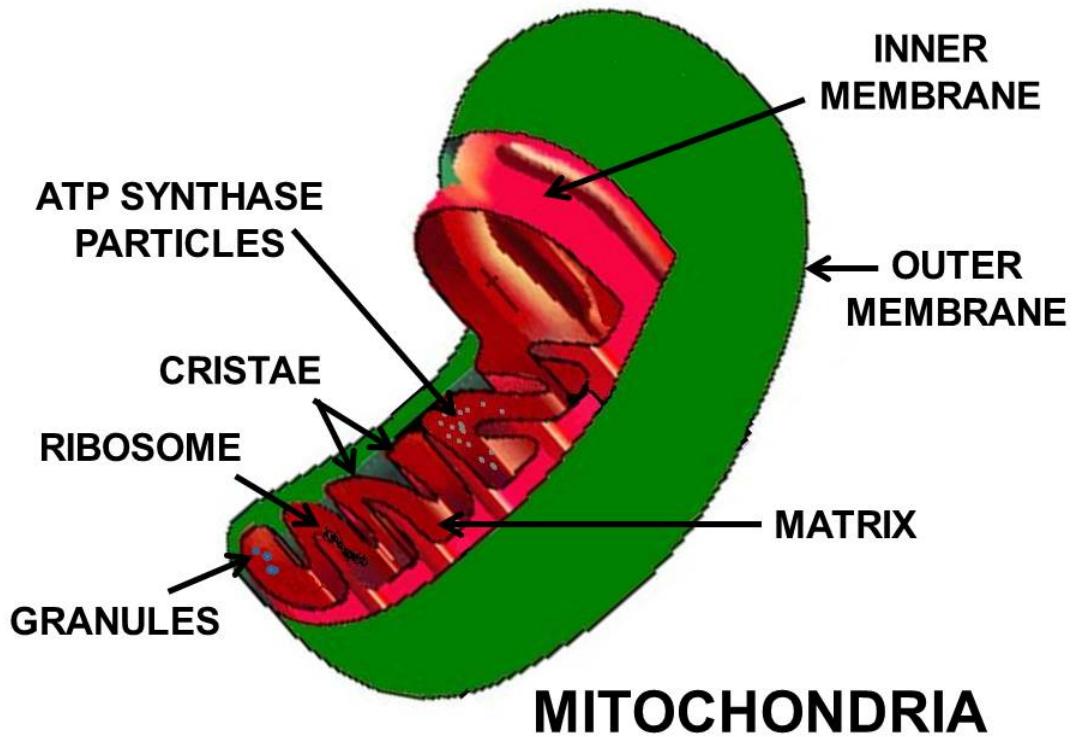


Entamoeba histolytica, another water-borne pathogen, can cause diarrhea or a more serious invasive liver abscess. When in contact with human cells, these amoebae are cytotoxic. There is a rapid influx of calcium into the contacted cell, it quickly stops all membrane movement save for some surface blebbing. Internal organization is disrupted, organelles lyse, and the cell dies. The ameba may eat the dead cell or just absorb nutrients released from the cell.

On average, about one in 10 people who are infected with *E. histolytica* becomes sick from the infection. The symptoms often are quite mild and can include loose stools, stomach pain, and stomach cramping. Amebic dysentery is a severe form of amebiasis associated with stomach pain, bloody stools, and fever. Rarely, *E. histolytica* invades the liver and forms an abscess. Even less commonly, it spreads to other parts of the body, such as the lungs or brain.

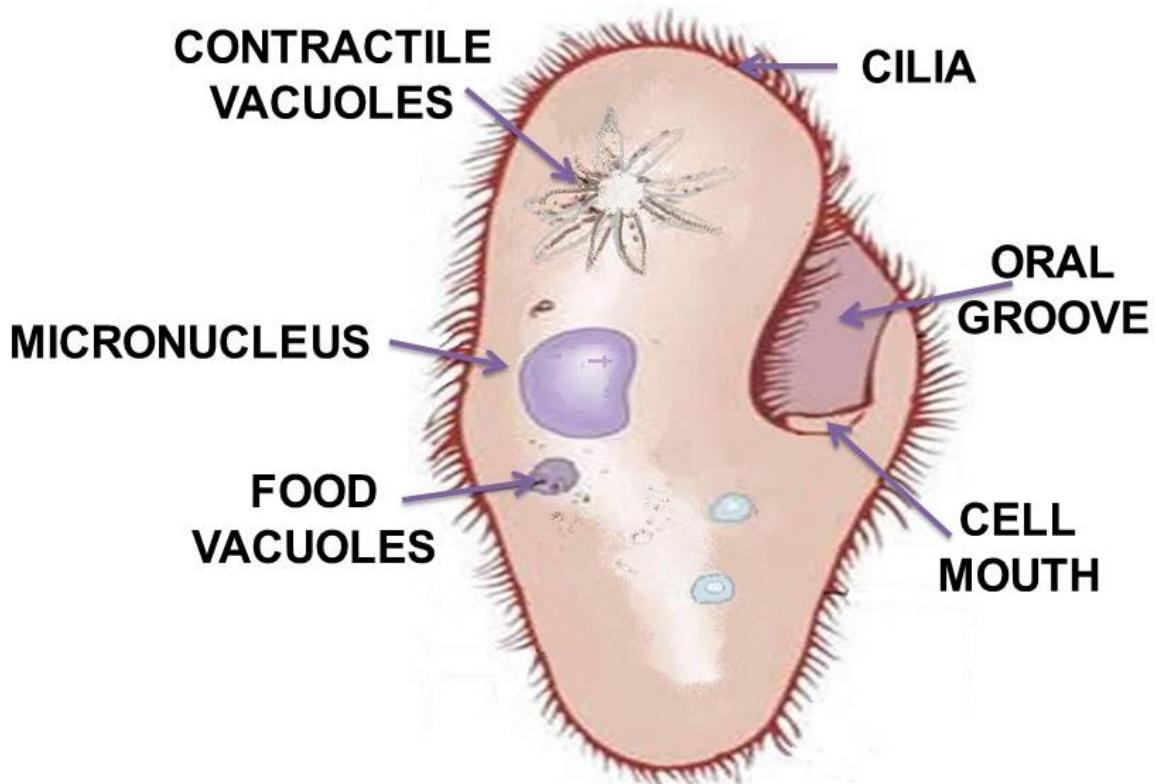
### Scientific classification

Domain: Eukaryota  
Phylum: Amoebozoa  
Class: Archamoebae  
Genus: Entamoeba  
Species: *E. histolytica*



### Mitochondria

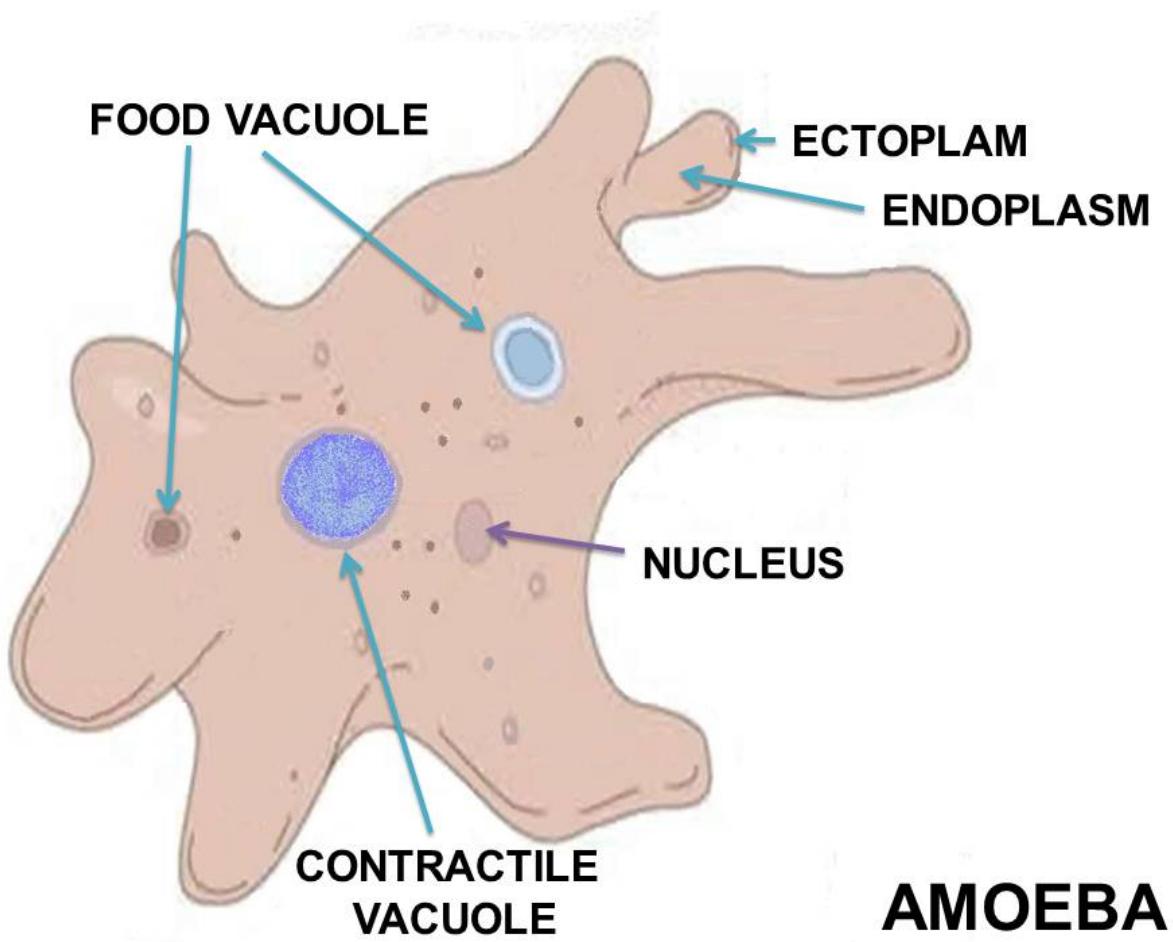
The bacterial cell is surrounded by a lipid membrane, or cell membrane, which encloses the contents of the cell and acts as a barrier to hold nutrients, proteins and other essential components of the cytoplasm within the cell. As they are prokaryotes, bacteria do not tend to have membrane-bound organelles in their cytoplasm and thus contain few large intracellular structures. They consequently lack a nucleus, mitochondria, chloroplasts and the other organelles present in eukaryotic cells, such as the Golgi apparatus and endoplasmic reticulum.



## PARAMECIUM

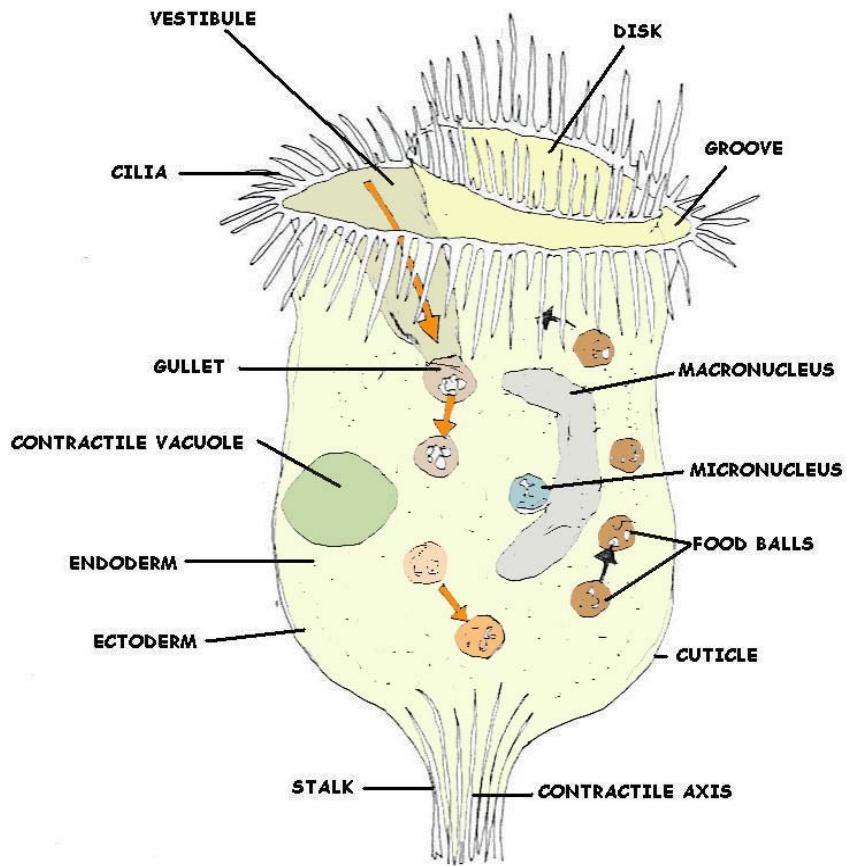
### Paramecia

Paramecia are a group of unicellular ciliate protozoa formerly known as slipper animalcules from their slipper shape. They are commonly studied as a representative of the ciliate group. Simple cilia cover the body which allows the cell to move with a synchronous motion (like a caterpillar). There is also a deep oral groove containing inconspicuous compound oral cilia (as found in other peniculids) that is used to draw food inside. They generally feed upon bacteria and other small cells. Osmoregulation is carried out by a pair of contractile vacuoles, which actively expel water absorbed by osmosis from their surroundings. Paramecia are widespread in freshwater environments, and are especially common in scums. Paramecia are attracted by acidic conditions. Certain single-celled eukaryotes, such as Paramecium, are examples for exceptions to the universality of the genetic code (translation systems where a few codons differ from the standard ones).



## Amoeba

Amoeba (sometimes *amœba* or *ameba*, plural *amoebae*) is a genus of protozoa that moves by means of pseudopods, and is well-known as a representative unicellular organism. The word amoeba or ameba is variously used to refer to it and its close relatives, now grouped as the Amoebozoa, or to all protozoa that move using pseudopods, otherwise termed amoeboids.



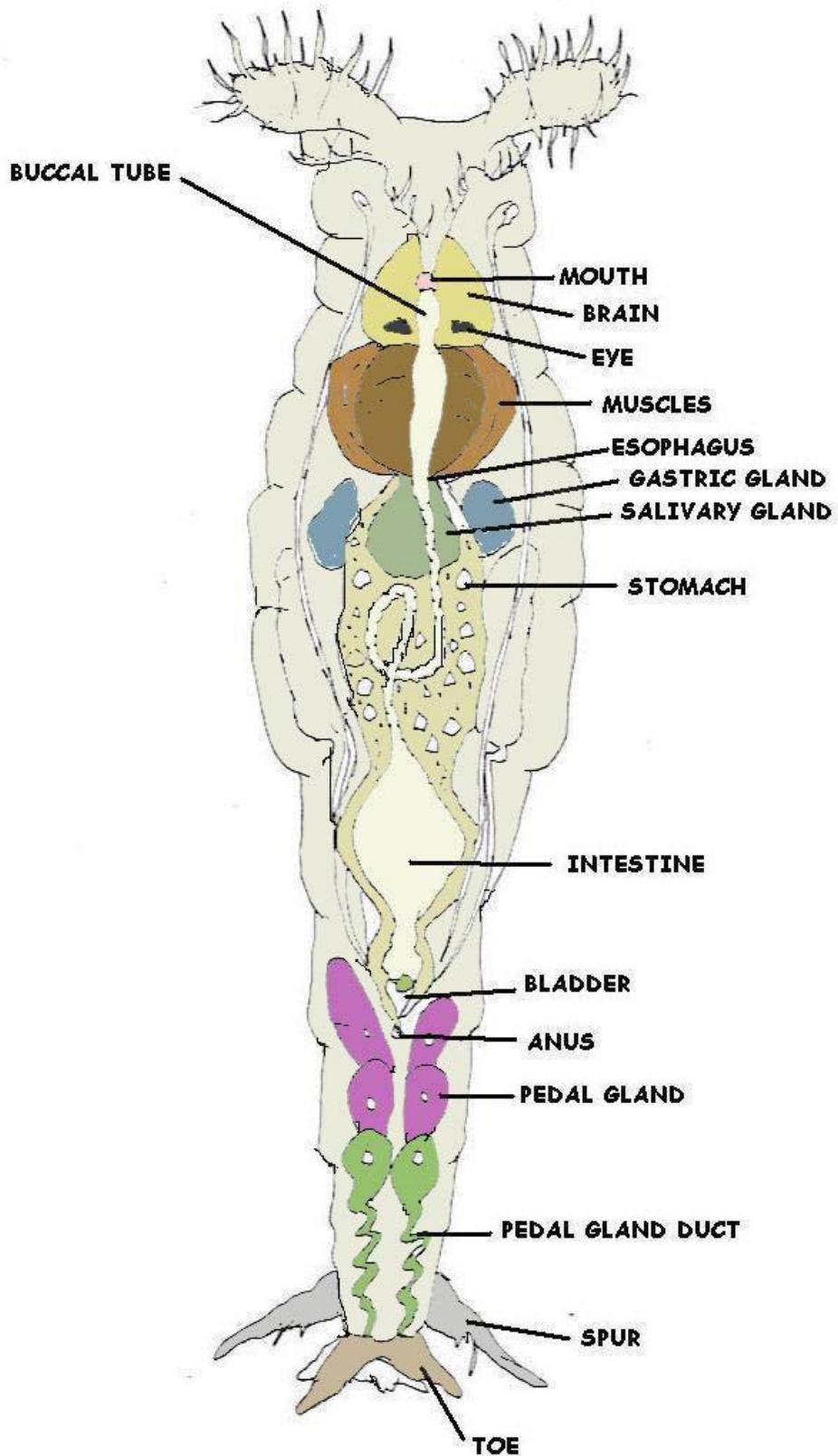
## VORTICELLA

(TYPE OF PROTOZOAN FOUND IN STAGNANT WATER)

### Vorticella

Vorticella is a genus of protozoa, with over 100 known species. They are stalked inverted bell-shaped ciliates, placed among the peritrichs. Each cell has a separate stalk anchored onto the substrate, which contains a contractile fibril called a myoneme. When stimulated this shortens, causing the stalk to coil like a spring. Reproduction is by budding, where the cell undergoes longitudinal fission and only one daughter keeps the stalk. Vorticella mainly lives in freshwater ponds and streams - generally anywhere protists are plentiful. Other genera such as Carchesium resemble Vorticella but are branched or colonial.

**Domain:** Eukaryota  
**Phylum:** Ciliophora  
**Class:** Oligohymenophorea  
**Subclass:** Peritrichia  
**Order:** Sessilida  
**Family:** Vorticellidae  
**Genus:** Vorticella



## ROTIFER

## Rotifer

The rotifers make up a phylum of microscopic and near-microscopic pseudocoelomate animals. They were first described by John Harris in 1696 (Hudson and Gosse, 1886). Leeuwenhoek is mistakenly given credit for being the first to describe rotifers but Harris had produced sketches in 1703. Most rotifers are around 0.1-0.5 mm long, and are common in freshwater throughout the world with a few saltwater species. Rotifers may be free swimming and truly planktonic, others move by inch worming along the substrate, whilst some are sessile, living inside tubes or gelatinous holdfasts. About 25 species are colonial (e.g. *Sinantherina semibullata*), either sessile or planktonic.

Rotifers get their name (derived from Greek and meaning "wheel-bearer"; they have also been called wheel animalcules) from the corona, which is composed of several ciliated tufts around the mouth that in motion resemble a wheel. These create a current that sweeps food into the mouth, where it is chewed up by a characteristic pharynx (called the mastax) containing a tiny, calcified, jaw-like structure called the trophi. The cilia also pull the animal, when unattached, through the water. Most free-living forms have pairs of posterior toes to anchor themselves while feeding. Rotifers have bilateral symmetry and a variety of different shapes. There is a well-developed cuticle which may be thick and rigid, giving the animal a box-like shape, or flexible, giving the animal a worm-like shape; such rotifers are respectively called lorate and illorate.

Like many other microscopic animals, adult rotifers frequently exhibit eutely - they have a fixed number of cells within a species, usually on the order of one thousand. Males in the class Monogononta may be either present or absent depending on the species and environmental conditions. In the absence of males, reproduction is by parthenogenesis and results in clonal offspring that are genetically identical to the parent. Individuals of some species form two distinct types of parthenogenetic eggs; one type develops into a normal parthenogenetic female, while the other occurs in response to a changed environment and develops into a degenerate male that lacks a digestive system, but does have a complete male reproductive system that is used to inseminate females thereby producing fertilized 'resting eggs'. Resting eggs develop into zygotes that are able to survive extreme environmental conditions such as may occur during winter or when the pond dries up. These eggs resume development and produce a new female generation when conditions improve again. The life span of monogonont females varies from a couple of days to about three weeks.

Bdelloid rotifers are unable to produce resting eggs, but many can survive prolonged periods of adverse conditions after desiccation. This facility is termed anhydrobiosis, and organisms with these capabilities are termed anhydrobionts. Under drought conditions, bdelloid rotifers contract into an inert form and lose almost all body water; when rehydrated, however, they resume activity within a few hours. Bdelloids can survive the dry state for prolonged periods, with the longest well-documented dormancy being nine years. While in other anhydrobionts, such as the brine shrimp, this desiccation tolerance is thought to be linked to the production of trehalose, a non-reducing disaccharide (sugar), bdelloids apparently lack the ability to synthesize trehalose. Bdelloid rotifer genomes contain two or more divergent copies of each gene. Four copies of hsp82 are, for example, found. Each is different and found on a different chromosome, excluding the possibility of homozygous sexual reproduction.

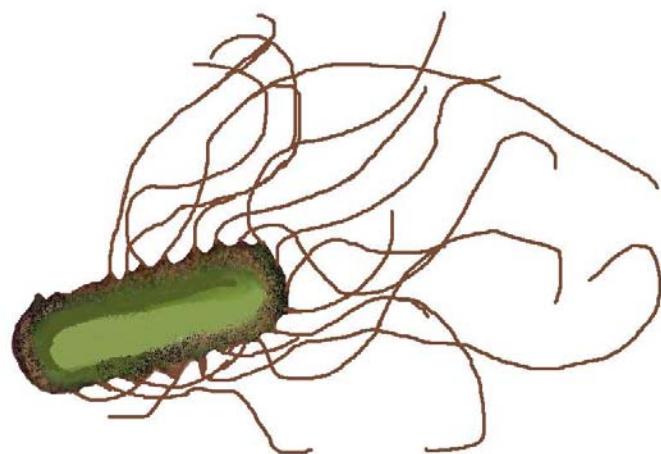
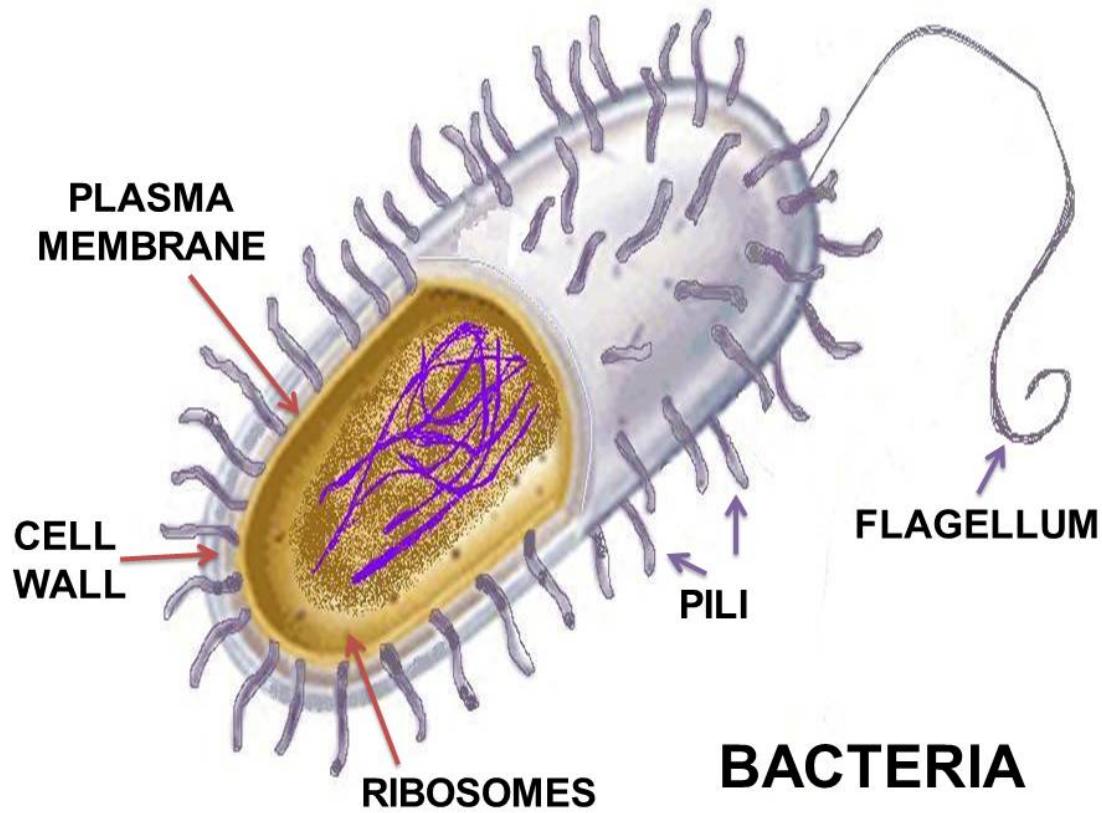
## Waterborne Diseases

Name	Causative organism	Source of organism	Disease
Viral gastroenteritis	<b>Rotavirus</b> (mostly in young children)	Human feces	Diarrhea or vomiting
Norwalk Agent	<b>Noroviruses</b> (genus <i>Norovirus</i> , family <i>Caliciviridae</i> ) <sup>*1</sup>	Human feces; also, shellfish; lives in polluted waters	Diarrhea and vomiting
Salmonellosis	<b>Salmonella</b> (bacterium)	Animal or human feces	Diarrhea or vomiting
Gastroenteritis <i>Escherichia coli</i>	-- <b><i>E. coli O1 57:H7</i></b> (bacterium): Other <i>E. coli</i> organisms:	Human feces	Symptoms vary with type caused
Typhoid	<b>Salmonella typhi</b> (bacterium)	Human feces, urine	Inflamed intestine, enlarged spleen, high temperature-sometimes fatal
Shigellosis	<b>Shigella</b> (bacterium)	Human feces	Diarrhea
Cholera	<b>Vibrio cholera</b> (bacterium)	Human feces; also, shellfish; lives in many coastal waters	Vomiting, severe diarrhea, rapid dehydration, mineral loss-high mortality
Hepatitis A	<b>Hepatitis A</b> virus	Human feces; shellfish grown in polluted waters	Yellowed skin, enlarged liver, fever, vomiting, weight loss, abdominal pain-low mortality, lasts up to four months
Amebiasis	<b>Entamoeba histolytica</b> (protozoan)	Human feces	Mild diarrhea, dysentery, extra intestinal infection
Giardiasis	<b>Giardia lamblia</b> (protozoan)	Animal or human feces	Diarrhea, cramps, nausea, and general weakness — lasts one week to months
Cryptosporidiosis	<b>Cryptosporidium parvum</b>	Animal or human feces	Diarrhea, stomach pain — lasts (protozoan) days to weeks

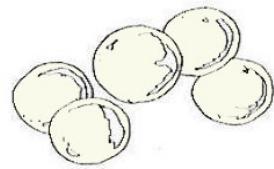
Notes:

\*1 <http://www.cdc.gov/ncidod/dvrd/revb/gastro/norovirus.htm>  
<http://www.cdc.gov/mmwr/preview/mmwrhtml/rr5009a1.htm>

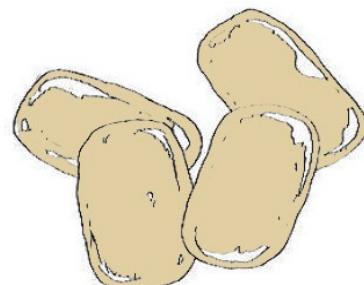
## Bacteria Section



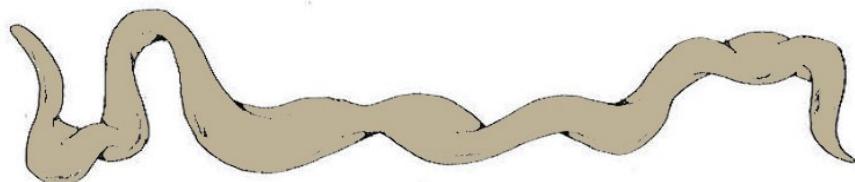
**Peritrichous Bacteria**



**COCCI**  
(SPHERICAL)



**BACILLI**  
(ROD-SHAPED)



**SPIRILLI**  
(SPIRAL)

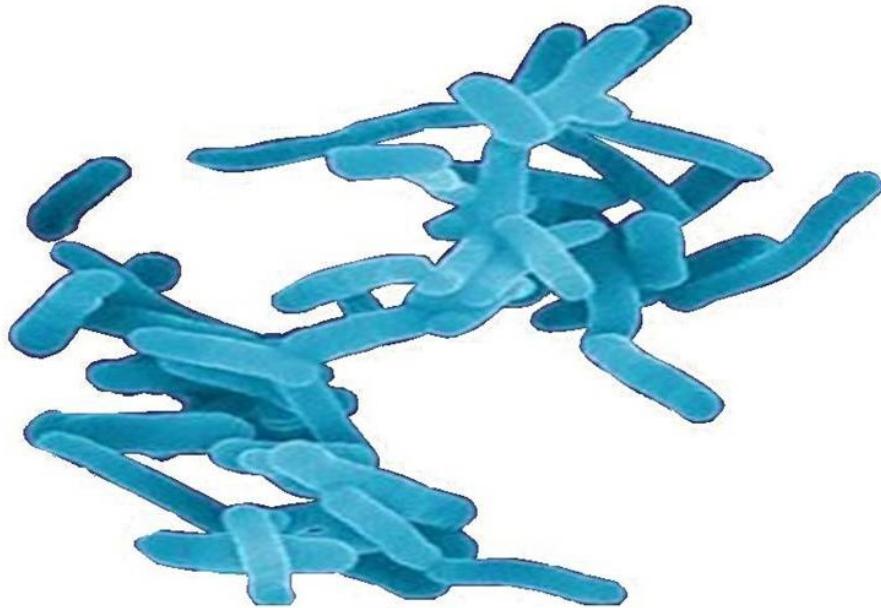
## **BACTERIA TYPES**

Microbiologists broadly classify bacteria according to their shape: spherical, rod-shaped, and spiral-shaped. Pleomorphic bacteria can assume a variety of shapes. Bacteria may be further classified according to whether they require oxygen (aerobic or anaerobic) and how they react to a test with Gram's stain. Bacteria in which alcohol washes away Gram's stain are called gram-negative, while bacteria in which alcohol causes the bacteria's walls to absorb the stain are called gram-positive.



**COLORLESS FILAMENTOUS  
SULFUR BACTERIA**

## **Shigella dysenteriae**



# **SHIGELLA DYSENTERIAE**

Shigella dysenteriae is a species of the rod-shaped bacterial genus Shigella. Shigella can cause shigellosis (bacillary dysentery). Shigellae are Gram-negative, non-spore-forming, facultatively anaerobic, non-motile bacteria.

*S. dysenteriae*, spread by contaminated water and food, causes the most severe dysentery because of its potent and deadly Shiga toxin, but other species may also be dysentery agents. Shigella infection is typically via ingestion (fecal-oral contamination); depending on age and condition of the host as few as ten bacterial cells can be enough to cause an infection. Shigella causes dysentery that result in the destruction of the epithelial cells of the intestinal mucosa in the cecum and rectum. Some strains produce enterotoxin and Shiga toxin, similar to the verotoxin of *E. coli* O157:H7. Both Shiga toxin and verotoxin are associated with causing hemolytic uremic syndrome.

Shigella invades the host through epithelial cells of the large intestine. Using a Type III secretion system acting as a biological syringe, the bacterium injects IpaD protein into cell, triggering bacterial invasion and the subsequent lysis of vacuolar membranes using IpaB and IpaC proteins. It utilizes a mechanism for its motility by which its IcsA protein triggers actin polymerization in the host cell (via N-WASP recruitment of Arp2/3 complexes) in a "rocket" propulsion fashion for cell-to-cell spread.

The most common symptoms are diarrhea, fever, nausea, vomiting, stomach cramps, and straining to have a bowel movement. The stool may contain blood, mucus, or pus (e.g. dysentery). In rare cases, young children may have seizures. Symptoms can take as long as a week to show up, but most often begin two to four days after ingestion. Symptoms usually last for several days, but can last for weeks. Shigella is implicated as one of the pathogenic causes of reactive arthritis worldwide.



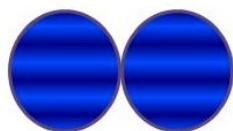
**Coccus**



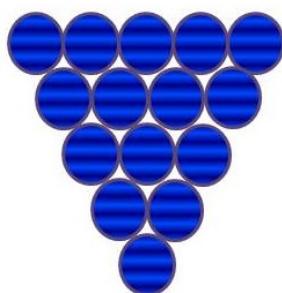
**Bacillus**



**Spirillum**



**Diplo-**



**Staphylo-**



**Strepto-**

Bacteria Types

Type	Characteristics
<b>Acetic acid</b>	Rod-shaped, gram-negative, aerobic; highly tolerant of acidic conditions; generate organic acids
<b>Actinomycete</b>	Rod-shaped or filamentous, gram-positive, aerobic; common in soils; essential to growth of many plants; source of much of original antibiotic production in pharmaceutical industry
<b>Coccoid</b>	Spherical, sometimes in clusters or strings, gram-positive, aerobic and anaerobic; resistant to drying and high-salt conditions; <i>Staphylococcus</i> species common on human skin, certain strains associated with toxic shock syndrome
<b>Coryneform</b>	Rod-shaped, form club or V shapes, gram-positive, aerobic; found in wide variety of habitats, particularly soils; highly resistant to drying; include <i>Arthrobacter</i> , among most common forms of life on earth
<b>Endospore-forming</b>	Usually rod-shaped, can be gram-positive or gram-negative; have highly adaptable, heat-resistant spores that can go dormant for long periods, possibly thousands of years; include <i>Clostridium</i> (anaerobic) and <i>Bacillus</i> (aerobic)
<b>Enteric</b>	Rod-shaped, gram-negative, aerobic but can live in certain anaerobic conditions; produce nitrite from nitrate, acids from glucose; include <i>Escherichia coli</i> , <i>Salmonella</i> (over 1000 types), and <i>Shigella</i>
<b>Gliding</b>	Rod-shaped, gram-negative, mostly aerobic; glide on secreted slimy substances; form colonies, frequently with complex fruiting structures
<b>Lactic acid</b>	Gram-positive, anaerobic; produce lactic acid through fermentation; include <i>Lactobacillus</i> , essential in dairy product formation, and <i>Streptococcus</i> , common in humans
<b>Mycobacterium</b>	Pleomorphic, spherical or rod-shaped, frequently branching, no gram stain, aerobic; commonly form yellow pigments; include <i>Mycobacterium tuberculosis</i> , cause of tuberculosis
<b>Mycoplasma</b>	Spherical, commonly forming branching chains, no gram stain, aerobic but can live in certain anaerobic conditions; without cell walls yet structurally resistant to lysis; among smallest of bacteria; named for superficial resemblance to fungal hyphae ( <i>myco-</i> means 'fungus')
<b>Nitrogen-fixing</b>	Rod-shaped, gram-negative, aerobic; convert atmospheric nitrogen gas to ammonium in soil; include <i>Azotobacter</i> , a common genus
<b>Propionic acid</b>	Rod-shaped, pleomorphic, gram-positive, anaerobic; ferment lactic acid; fermentation produces holes in Swiss cheese from the production of carbon dioxide
<b>Pseudomonad</b>	Rod-shaped (straight or curved) with polar flagella, gram-negative, aerobic; can use up to 100 different compounds for carbon and energy
<b>Rickettsia</b>	Spherical or rod-shaped, gram-negative, aerobic; cause Rocky Mountain spotted fever and typhus; closely related to <i>Agrobacterium</i> , a common gall-causing plant bacterium
<b>Sheathed</b>	Filamentous, gram-negative, aerobic; 'swarmer' (colonizing) cells form and break out of a sheath; sometimes coated with metals from environment

<b>Spirillum</b>	Spiral-shaped, gram-negative, aerobic; include <i>Bdellovibrio</i> , predatory on other bacteria
<b>Spirochete</b>	Spiral-shaped, gram-negative, mostly anaerobic; common in moist environments, from mammalian gums to coastal mudflats; complex internal structures convey rapid movement; include <i>Treponema pallidum</i> , cause of syphilis
<b>Sulfate- and Sulfur-reducing</b>	Commonly rod-shaped, mostly gram-negative, anaerobic; include <i>Desulfovibrio</i> , ecologically important in marshes
<b>Sulfur- and iron-oxidizing</b>	Commonly rod-shaped, frequently with polar flagella, gram-negative, mostly anaerobic; most live in neutral (nonacidic) environment
<b>Vibrio</b>	Rod- or comma-shaped, gram-negative, aerobic; commonly with a single flagellum; include <i>Vibrio cholerae</i> , cause of cholera, and luminescent forms symbiotic with deep-water fishes and squids

## Gram<sup>+</sup>

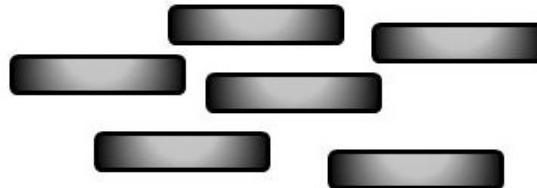


**Lactobacillus acidophilus**



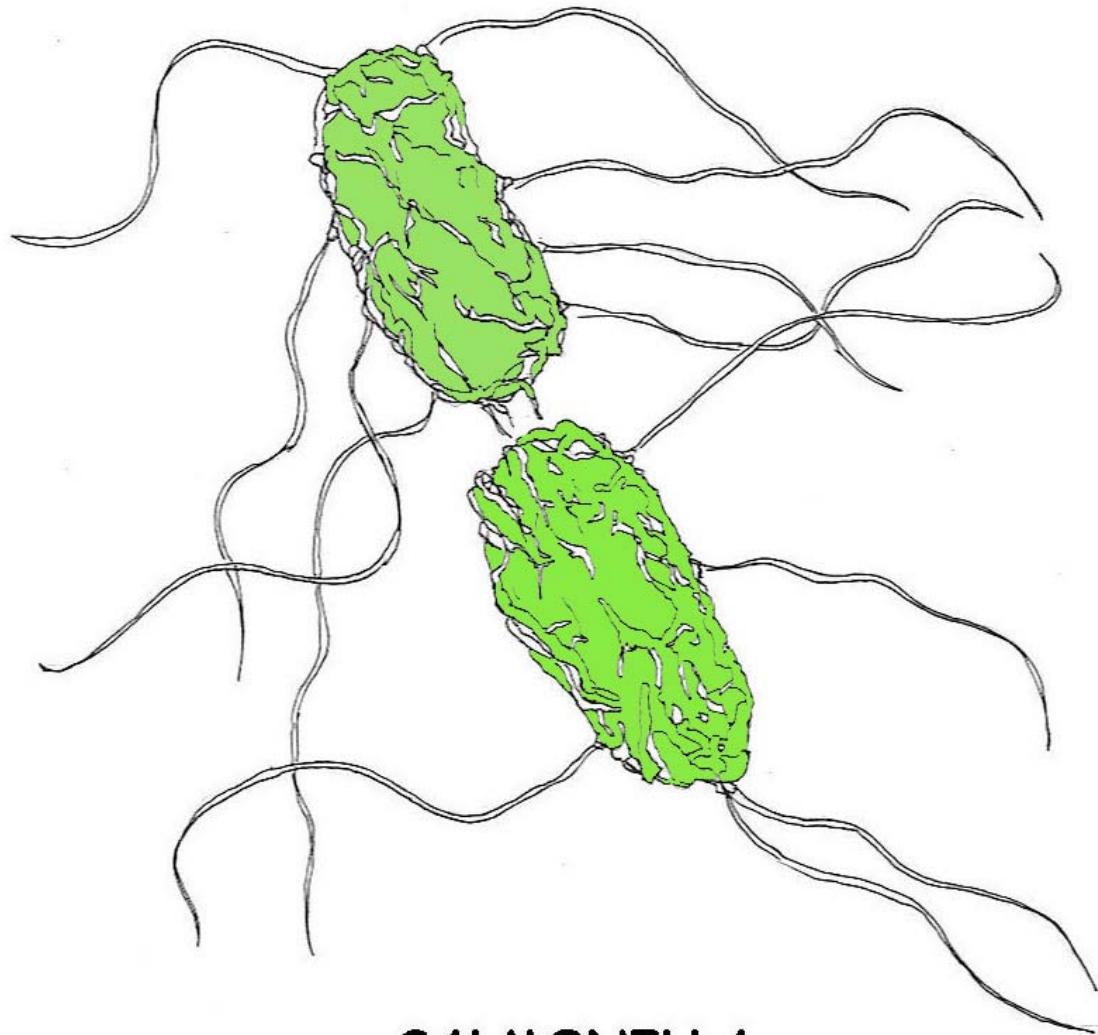
**Streptococcus thermophilus**

## Gram-



**Escherichia coli**

## Salmonella



## SALMONELLA

Salmonella is a Gram-negative bacterium. It is found in many turtles and other reptiles. In clinical laboratories, it is usually isolated on MacConkey agar, XLD agar, XLT agar, DCA agar, or Önöz agar. Because they cause intestinal infections and are greatly outnumbered by the bacteria normally found in the healthy bowel, primary isolation requires the use of a selective medium, so use of a relatively non-selective medium such as CLED agar is not often practiced. Numbers of salmonella may be so low in clinical samples that stools are routinely also subjected to "enrichment culture", where a small volume of stool is incubated in a selective broth medium, such as selenite broth or Rappaport Vassiliadis soya peptone broth, overnight. These media are inhibitory to the growth of the microbes normally found in the healthy human bowel, while allowing salmonellae to become enriched in numbers. Salmonellae may then be recovered by inoculating the enrichment broth on one or more of the primary selective media. On blood agar, they form moist colonies about 2 to 3 mm in diameter.

When the cells are grown for a prolonged time at a range of 25—28°C, some strains produce a biofilm, which is a matrix of complex carbohydrates, cellulose and proteins. The ability to produce biofilm (a.k.a. "rugose", "lacy", or "wrinkled") can be an indicator of dimorphism, which is the ability of a single genome to produce multiple phenotypes in response to environmental conditions. *Salmonellae* usually do not ferment lactose; most of them produce hydrogen sulfide which, in media containing ferric ammonium citrate, reacts to form a black spot in the centre of the creamy colonies.

### Classification

*Salmonella* taxonomy is complicated. As of December 7, 2005, there are two species within the genus: *S. bongori* (previously subspecies V) and *S. enterica* (formerly called *S. choleraesuis*), which is divided into six subspecies:

- \* I—*enterica*
- \* II—*salamae*
- \* IIIa—*arizonae*
- \* IIIb—*diarizonae*
- \* IV—*houtenae*
- \* V—obsolete (now designated *S. bongori*)
- \* VI—*indica*

There are also numerous (over 2500) serovars within both species, which are found in a disparate variety of environments and which are associated with many different diseases. The vast majority of human isolates (>99.5%) are subspecies *S. enterica*. For the sake of simplicity, the CDC recommends that *Salmonella* species be referred to only by their genus and serovar, e.g.

*Salmonella* Typhi instead of the more technically correct designation, *Salmonella enterica* subspecies *enterica* serovar Typhi.



## Escherichia Coli Section

### Fecal Coliform Bacteria

Fecal coliform bacteria are microscopic organisms that live in the intestines of warm-blooded animals. They also live in the waste material, or feces, excreted from the intestinal tract. When fecal coliform bacteria are present in high numbers in a water sample, it means that the water has received fecal matter from one source or another. Although not necessarily agents of disease, fecal coliform bacteria may indicate the presence of disease-carrying organisms, which live in the same environment as the fecal coliform bacteria.

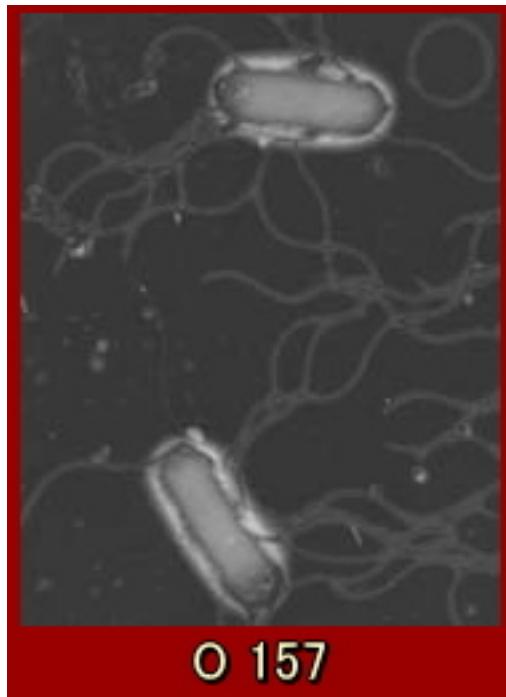
### Reasons for Natural Variation

Unlike the other conventional water quality parameters, fecal coliform bacteria are living organisms. They do not simply mix with the water and float straight downstream. Instead they multiply quickly when conditions are favorable for growth, or die in large numbers when conditions are not. Because bacterial concentrations are dependent on specific conditions for growth, and these conditions change quickly, fecal coliform bacteria counts are not easy to predict. For example, although winter rains may wash more fecal matter from urban areas into a stream, cool water temperatures may cause a major die-off. Exposure to sunlight (with its ultraviolet disinfection properties) may have the same effect, even in the warmer water of summertime.

### Expected Impact of Pollution

The primary sources of fecal coliform bacteria to fresh water are wastewater treatment plant discharges, failing septic systems, and animal waste. Bacteria levels do not necessarily decrease as a watershed develops from rural to urban. Instead, urbanization usually generates new sources of bacteria. Farm animal manure and septic systems are replaced by domestic pets and leaking sanitary sewers. In fact, stormwater runoff in urbanized areas has been found to be surprisingly high in fecal coliform bacteria concentrations.

The presence of old, disintegrating storm and sanitary sewers, misplaced sewer pipes, and good breeding conditions are common explanations for the high levels measured.



### Coliform Standards (in colonies/100ml )

Drinking water.....	1FC
Total body contact (swimming).....	200FC
Partial body contact (boating).....	1000FC
Threatened sewage effluent .....	not to exceed 200 FC

\*Total coliform (TC) includes bacteria from cold-blooded animals and various soil organisms. According to recent literature, total coliform counts are normally about 10 times higher than fecal coliform (FC) counts.

### Indicator Connection Varies

General coliforms, E. Coli, and Enterococcus bacteria are the "indicator" organisms generally measured to assess microbiological quality of water. However, these aren't generally what get people sick. Other bacteria, viruses, and parasites are what we are actually worried about.

Because it is so much more expensive and tedious to do so, actual pathogens are virtually never tested for. Over the course of a professional lifetime pouring over indicator tests, in a context where all standards are based on indicators, water workers tend to forget that the indicators are not the things we actually care about.

### What are these indicators? More information in the Laboratory section.

- **General coliforms** indicate that the water has come in contact with plant or animal life. General coliforms are universally present, including in pristine spring water. They are of little concern at low levels, except to indicate the effectiveness of disinfection. Chlorinated water and water from perfectly sealed tube wells is the only water I've tested which had zero general coliforms. At very high levels they indicate there is what amounts to a lot of compost in the water, which could easily include pathogens (Ten thousand general coliform bacteria will get you a beach closure, compared to two or four hundred fecal coliforms, or fifty enterococcus).
- **Fecal coliforms**, particularly E. coli, indicate that there are mammal or bird feces in the water.
- **Enterococcus bacteria** also indicate that there are feces from warm blooded animals in the water. Enterococcus are a type of fecal streptococci. They are another valuable indicator for determining the amount of fecal contamination of water.

According to studies conducted by the EPA, enterococci have a greater correlation with swimming-associated gastrointestinal illness in both marine and fresh waters than other bacterial indicator organisms, and are less likely to "die off" in saltwater.

The more closely related the animal, the more likely pathogens excreted with their feces can infect us. Human feces are the biggest concern, because anything which infects one human could infect another. There isn't currently a quantitative method for measuring specifically human fecal bacteria (expensive genetic studies can give a presence/absence result). Ingesting a human stranger's feces via contaminated water supply is a classic means for infections to spread rapidly. The more pathogens an individual carries, the more hazardous their feces. Ingesting feces from someone who is not carrying any pathogens may gross you out, but it can't infect you. Infection rates are around 5% in the US, and approach 100% in areas with poor hygiene and contaminated water supplies. Keep in the back of your mind that **the ratio of indicators to actual pathogens is not fixed**. It will always be different, sometimes very different. Whenever you are trying to form a mental map of reality based on water tests, you should include in the application of your water intuition an adjustment factor for your best guess of the ratio between indicators and actual pathogens.

## Membrane Filter Total Coliform Technique

The membrane filter total Coliform technique is used at Medina County for drinking water quality testing. The following is a summary of this test. A sampling procedure sheet is given to all sample takers by Medina County.

The samples are taken in sterile 100 mL containers. These containers, when used for chlorinated water samples, have a sodium thiosulfate pill or solution to dechlorinate the sample.

The sample is placed in cold storage after proper sample taking procedures are followed. (See sample procedures below)

The samples are taken to the laboratory with a chain of custody to assure no tampering of samples can occur.

These samples are logged in at the laboratory.

No longer than 30 hours can lapse between the time of sampling and time of test incubation. (8 hours for heterotrophic, nonpotable 6 hours, others not longer than 24 hours)

All equipment is sterilized by oven and autoclave. Glassware in oven at  $170^{\circ}\text{C} \pm 10^{\circ}\text{C}$  with foil (or other suitable wrap) loosely fitting and secured immediately after sterilization.

Filtration units in autoclave at  $121^{\circ}\text{C}$  for 30 minutes.

Use sterile petri dishes, grid, and pads bought from a reliable company – certified, quality assured - test for satisfactory known positive amounts.

Incubators –  $35^{\circ}\text{C} \pm .5^{\circ}\text{C}$  (60% relative humidity)

M-endo medium is prepared and heated to near boiling removed from heat cooled to  $45^{\circ}\text{C}$  pH adjusted to  $7.2 \pm .2$  and immediately dispensed 8ml to plates. Keep refrigerated and discard after 2 weeks.

Fisher Scientific

**Microbiological Test Data Sheet**

Laboratory	Medina County SD 500 Laboratory	Test Method	Year	2006	Page #	43		
Certificate Number	421	Membrane Filter (MF)	MMO-MUG/Coliform (DL)	MMO-MUG/Coliform (C5)				
Lab #	Collected	Incubated	Sample Location	Total Coliform	E. coli	Controls	Test Method	Chlorine
1161	5-8 9:05	13-14 10:10	Imperial Beach	+	+	+	Flow	Flow
1162	9:20		14th Subdivision Down					
1163	9:40		505 Subdivision					
1164	10:05		580 Liverpool					
1165	10:20		720 Liverpool					
1166	10:35		740 Adams St					
1167	10:45		5321 Center					
1168	10:50		5329 Center					
1169	9:30		Baldwin Park					
			58 Pauls Church					
			Final Blank					
			Final					



Plates can be stored in a dated box with expiration date and discarded if not used. No denatured alcohol should be used. Everclear or 95% proof alcohol or absolute methyl may be used for sterilizing forceps by flame.

**Procedure:**

Counters are alcohol wiped.

Bench sheets are filled out.

Samples are removed from refrigeration.

Sterile wrapped utensils are placed on counters.

Filtration units are placed onto sterile membrane filters by aseptic technique using sterile forceps.

Sterile petri dishes are labeled.

The samples closures are clipped.

The sample is shaken 25 times 1 foot in length within 7 seconds.

100 mL is filtered and rinsed with sterile distilled water 3 times.

The membrane filter is aseptically removed from filter holder.

A sterile padded petri dish is used and the membrane filter is rolled onto the pad making sure no air bubbles form.

The sterile labeled lid is placed on the petri dish.

2 blanks and a known is run with each series of samples.

The samples are placed in the  $35^{\circ}\text{C} \pm .5^{\circ}\text{C}$  incubator stacked no higher than 3 for 22 – 24 hours (Humidity can be maintained by saturated paper towels placed under containers holding petri dishes.)

After 22- 24 hours view the petri dishes under a 10 –15 power magnification with cool white fluorescent light.

Count all colonies that appear pink to dark red with a metallic surface sheen – the sheen may vary in size from a pin head to complete coverage.

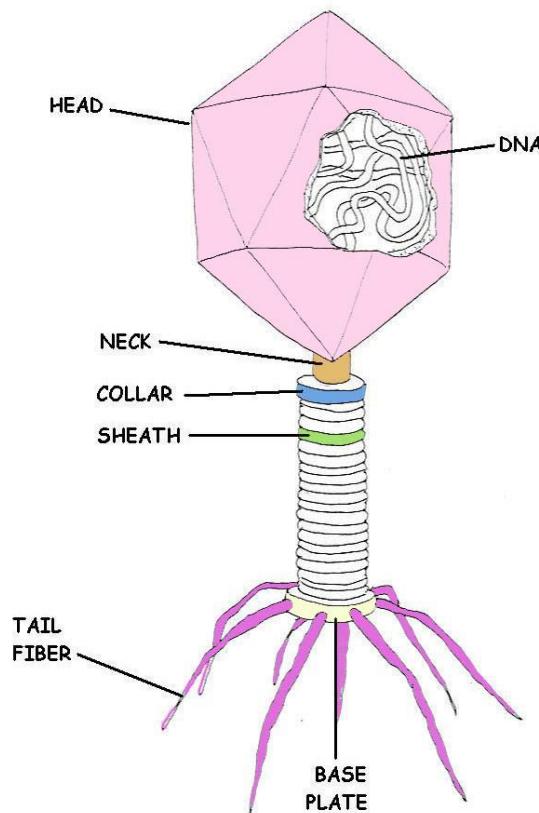
Report as Total Coliform per 100 mL.

If no colonies are present report as <1 coliform/100mL.

Anything greater than 1 is over the limit for drinking water for 2 samples taken 24 hours apart. Further investigation may be necessary – follow Standard Methods accordingly.



Photograph and Credits to Mary McPherson  
Aran™ Aqua Analytical Laboratory Director.



## BACTERIOPHAGE

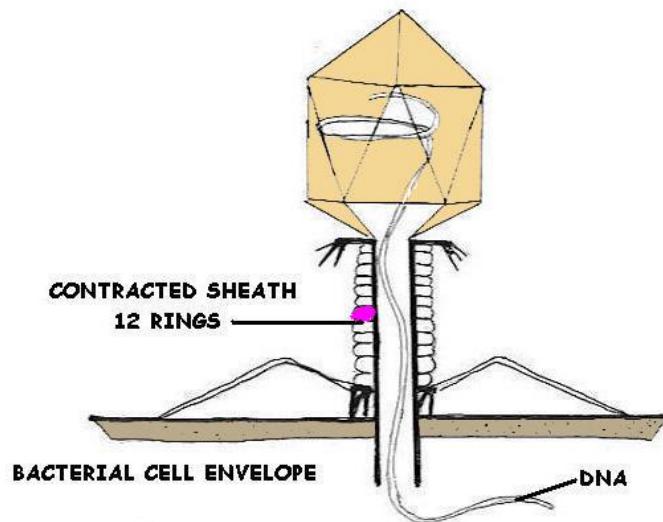
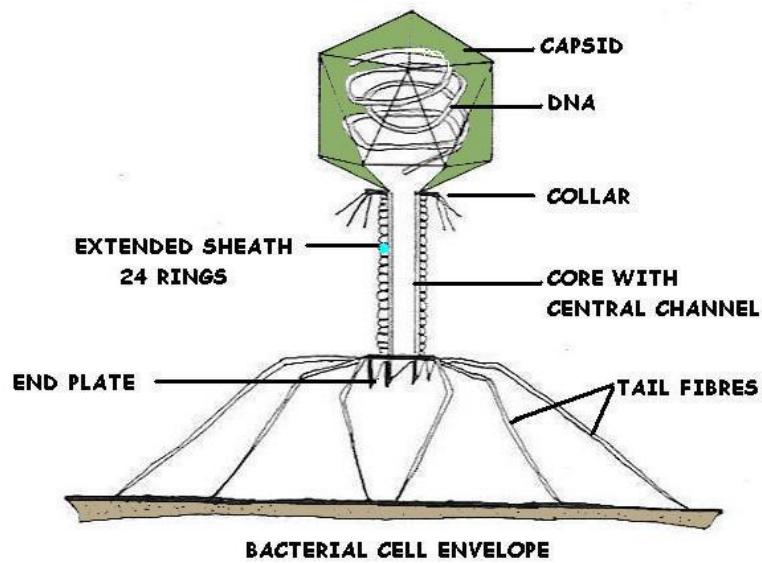
### Bacteriophage

A bacteriophage (from 'bacteria' and Greek *phagein*, 'to eat') is any one of a number of viruses that infect bacteria. The term is commonly used in its shortened form, *phage*.

Typically, bacteriophages consist of an outer protein hull enclosing genetic material. The genetic material can be ssRNA (single stranded RNA), dsRNA, ssDNA, or dsDNA between 5 and 500 kilo base pairs long with either circular or linear arrangement. Bacteriophages are much smaller than the bacteria they destroy - usually between 20 and 200 nm in size.

Phages are estimated to be the most widely distributed and diverse entities in the biosphere. Phages are ubiquitous and can be found in all reservoirs populated by bacterial hosts, such as soil or the intestine of animals. One of the densest natural sources for phages and other viruses is sea water, where up to  $9 \times 10^8$  virions per milliliter have been found in microbial mats at the surface, and up to 70% of marine bacteria may be infected by phages.

## VIRUS CAPSID (BACTERIOPHAGES)

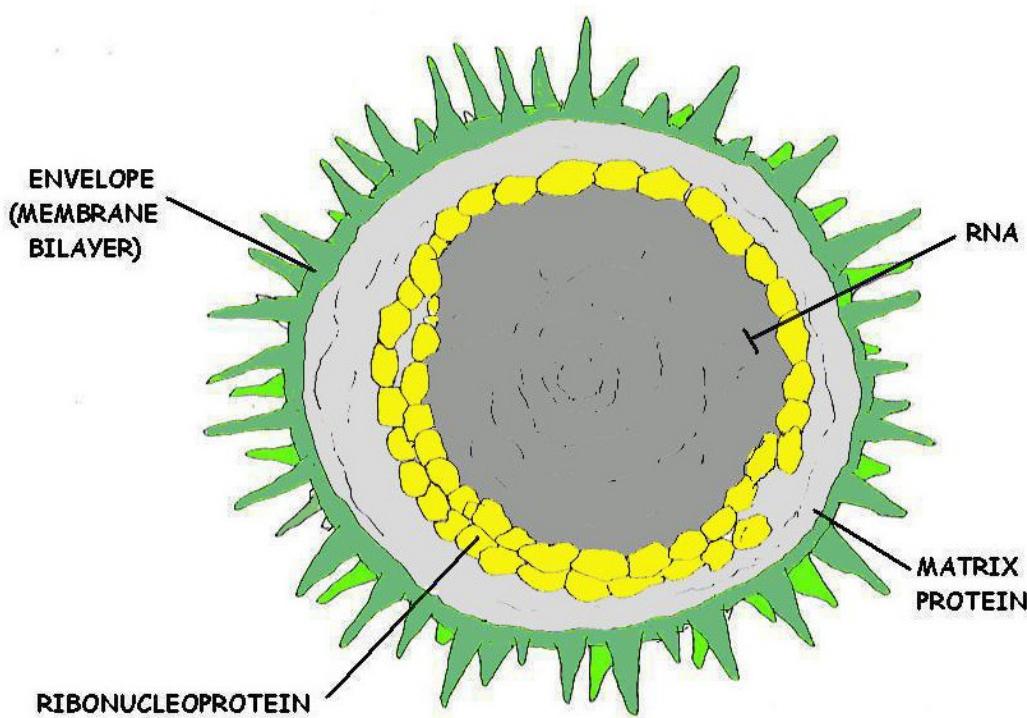


### Release of Virions

Phages may be released via cell lysis or by host cell secretion. In the case of the T4 phage, in just over twenty minutes after injection upwards of three hundred phages will be released via lysis within a certain timescale. This is achieved by an enzyme called endolysin which attacks and breaks down the peptidoglycan. In contrast, "lysogenic" phages do not kill the host but rather become long-term parasites and make the host cell continually secrete more new virus particles. The new virions bud off the plasma membrane, taking a portion of it with them to become enveloped viruses possessing a viral envelope. All released virions are capable of infecting a new bacterium.

## Viruses

Viruses are acellular microorganisms. They are made up of only genetic material and a protein coat. Viruses depend on the energy and metabolic machinery of the host cell to reproduce. A virus is an infectious agent found in virtually all life forms, including humans, animals, plants, fungi, and bacteria. Viruses consist of genetic material—either deoxyribonucleic acid (DNA) or ribonucleic acid (RNA)—surrounded by a protective coating of protein, called a capsid, with or without an outer lipid envelope. Viruses are between 20 and 100 times smaller than bacteria and hence are too small to be seen by light microscopy.



### CROSS SECTIONAL VIEW

Viruses vary in size from the largest poxviruses of about 450 nanometers (about 0.000014 in) in length to the smallest polioviruses of about 30 nanometers (about 0.000001 in). Viruses are not considered free-living, since they cannot reproduce outside of a living cell; they have evolved to transmit their genetic information from one cell to another for the purpose of replication. Viruses often damage or kill the cells that they infect, causing disease in infected organisms. A few viruses stimulate cells to grow uncontrollably and produce cancers. Although many infectious diseases, such as the common cold, are caused by viruses, there are no cures for these illnesses. The difficulty in developing antiviral therapies stems from the large number of variant viruses that can cause the same disease, as well as the inability of drugs to disable a virus without disabling healthy cells. However, the development of antiviral agents is a major focus of current research, and the study of viruses has led to many discoveries important to human health.

## **Virions**

Individual viruses, or virus particles, also called virions, contain genetic material, or genomes, in one of several forms. Unlike cellular organisms, in which the genes always are made up of DNA, viral genes may consist of either DNA or RNA. Like cell DNA, almost all viral DNA is double-stranded, and it can have either a circular or a linear arrangement. Almost all viral RNA is single-stranded; it is usually linear, and it may be either segmented (with different genes on different RNA molecules) or non-segmented (with all genes on a single piece of RNA).

## **Capsids**

The viral protective shell, or capsid, can be either helical (spiral-shaped) or icosahedral (having 20 triangular sides). Capsids are composed of repeating units of one or a few different proteins. These units are called protomers or capsomers. The proteins that make up the virus particle are called structural proteins. Viruses also carry genes for making proteins that are never incorporated into the virus particle and are found only in infected cells. These viral proteins are called nonstructural proteins; they include factors required for the replication of the viral genome and the production of the virus particle.

Capsids and the genetic material (DNA or RNA) they contain are together referred to as nucleocapsids. Some virus particles consist only of nucleocapsids, while others contain additional structures.

Some icosahedral and helical animal viruses are enclosed in a lipid envelope acquired when the virus buds through host-cell membranes. Inserted into this envelope are glycoproteins that the viral genome directs the cell to make; these molecules bind virus particles to susceptible host cells.

## **Bacteriophages**

The most elaborate viruses are the bacteriophages, which use bacteria as their hosts. Some bacteriophages resemble an insect with an icosahedral head attached to a tubular sheath. From the base of the sheath extend several long tail fibers that help the virus attach to the bacterium and inject its DNA to be replicated, direct capsid production, and virus particle assembly inside the cell.

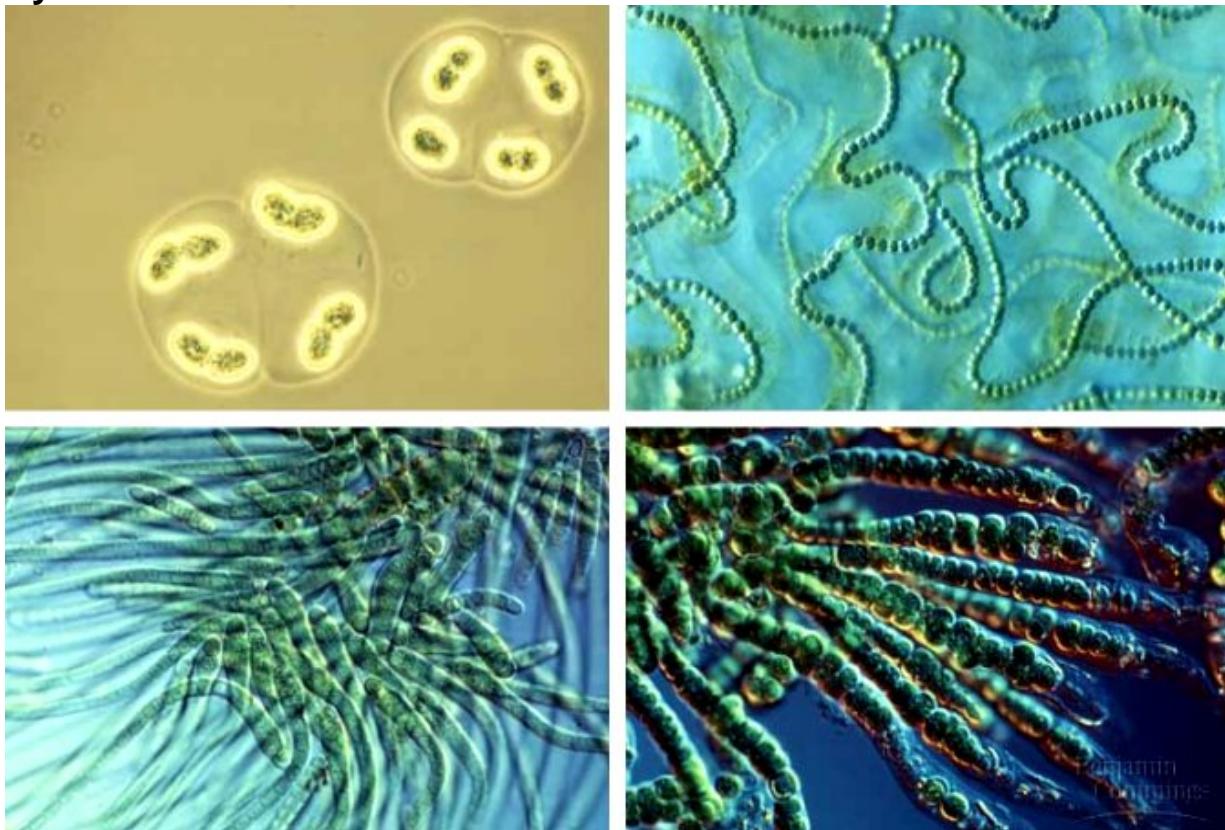
## **Viroids and Prions**

Viroids and prions are smaller than viruses, but they are similarly associated with disease. Viroids are plant pathogens that consist only of a circular, independently replicating RNA molecule. The single-stranded RNA circle collapses on itself to form a rod-like structure. The only known mammalian pathogen that resembles plant viroids is the deltavirus (hepatitis D), which requires hepatitis B virus proteins to package its RNA into virus particles. Co-infection with hepatitis B and D can produce more severe disease than can infection with hepatitis B alone. Prions are mutated forms of a normal protein found on the surface of certain animal cells.

## **Virus Classification**

Viruses are classified according to their type of genetic material, their strategy of replication, and their structure. The International Committee on Nomenclature of Viruses (ICNV), established in 1966, devised a scheme to group viruses into families, subfamilies, genera, and species. The ICNV report published in 1995 assigned more than 4000 viruses into 71 virus families. Hundreds of other viruses remain unclassified because of the lack of sufficient information.

## Cyanobacteria



## Cyanobacteria

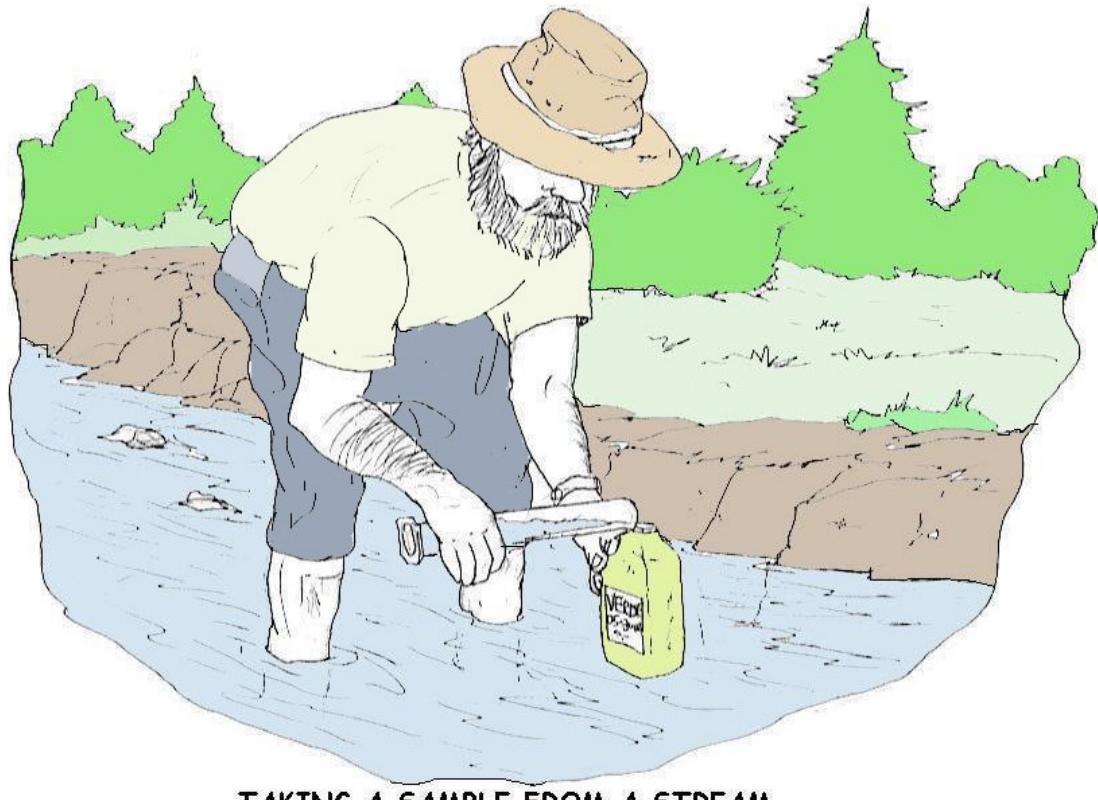
Cyanobacteria, also known as blue-green algae, blue-green bacteria or Cyanophyta, is a phylum of bacteria that obtain their energy through photosynthesis. The name "cyanobacteria" comes from the color of the bacteria (Greek: *kyanós* = blue). They are a significant component of the marine nitrogen cycle and an important primary producer in many areas of the ocean, but are also found on land.

Cyanobacteria include unicellular and colonial species. Colonies may form filaments, sheets or even hollow balls. Some filamentous colonies show the ability to differentiate into several different cell types: vegetative cells, the normal, photosynthetic cells that are formed under favorable growing conditions; akinetes, the climate-resistant spores that may form when environmental conditions become harsh; and thick-walled heterocysts, which contain the enzyme nitrogenase, vital for nitrogen fixation. Heterocysts may also form under the appropriate environmental conditions (anoxic) wherever nitrogen is necessary. Heterocyst-forming species are specialized for nitrogen fixation and are able to fix nitrogen gas, which cannot be used by plants, into ammonia ( $\text{NH}_3$ ), nitrites ( $\text{NO}_2$ ) or nitrates ( $\text{NO}_3$ ), which can be absorbed by plants and converted to protein and nucleic acids.

The rice paddies of Asia, which produce about 75% of the world's rice, could not do so were it not for healthy populations of nitrogen-fixing cyanobacteria in the rice paddy fertilizer too.

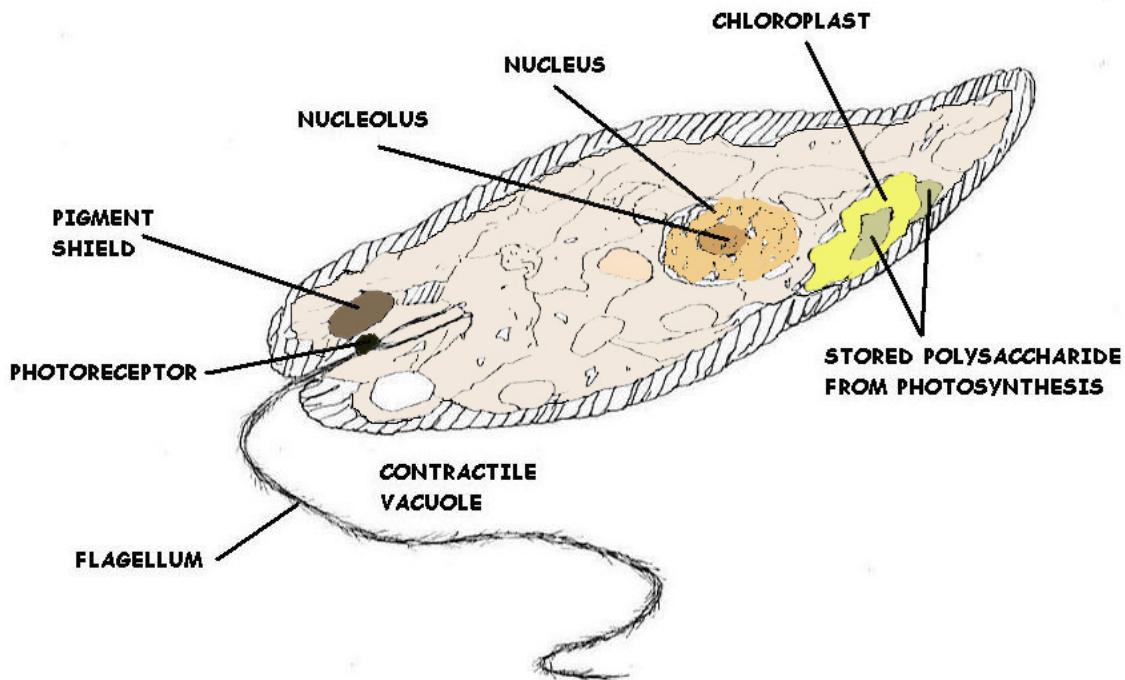
Many cyanobacteria also form motile filaments, called hormogonia, that travel away from the main biomass to bud and form new colonies elsewhere. The cells in a hormogonium are often thinner than in the vegetative state, and the cells on either end of the motile chain may be tapered. In order to break away from the parent colony, a hormogonium often must tear apart a weaker cell in a filament, called a necridium.

Each individual cell of a cyanobacterium typically has a thick, gelatinous cell wall. They differ from other gram-negative bacteria in that the quorum sensing molecules autoinducer-2[4] and acyl-homoserine lactones are absent. They lack flagella, but hormogonia and some unicellular species may move about by gliding along surfaces. In water columns some cyanobacteria float by forming gas vesicles, like in archaea.



TAKING A SAMPLE FROM A STREAM

# EUGLENA



## Euglena

Euglenas are common protists, of the class Euglenoidea of the phylum Euglenophyta. Currently, over 1000 species of Euglena have been described. Marin et al. (2003) revised the genus so, and including several species without chloroplasts, formerly classified as *Astasia* and *Khawinea*. Euglena sometimes can be considered to have both plant and animal features.

Euglena gracilis has a long hair-like thing that stretches from its body. You need a very powerful microscope to see it. This is called a flagellum, and the euglena uses it to swim. It also has a red eyespot. Euglena gracilis uses its eyespot to locate light. Without light, it cannot use its chloroplasts to make itself food. In order for Euglena gracilis to make more Euglena gracilis it will complete a process called mitosis. That means it can split itself in half and become two Euglena gracilis. It can only do this if it is well-fed and if the temperature is right. Euglena gracilis can reproduce better in warm temperatures.

Euglena gracilis, and other euglena, are harmless to people, but they are often signs that water is polluted, since they do well where there is a lot of green algae to eat. Green algae does well where there is a lot of nitrogen (comes from waste) in the water. If you don't clean your swimming pool, leaves and twigs get in the water and turn into waste. Then algae and euglena show up.

KINGDOM: Protist, PHYLUM: Euglenophyta, CLASS: Euglenophyceae, ORDER: Euglenales, FAMILY: Euglenidae, GENUS: Euglena, SPECIES: Euglena gracilis

## Peptidoglycan

**Peptidoglycan**, also known as murein, is a polymer consisting of sugars and amino acids that forms a mesh-like layer outside the plasma membrane of eubacteria. The sugar component consists of alternating residues of  $\beta$ -(1,4) linked N-acetylglucosamine and N-acetylmuramic acid residues. Attached to the N-acetylmuramic acid is a peptide chain of three to five amino acids. The peptide chain can be cross-linked to the peptide chain of another strand forming the 3D mesh-like layer.



## Hepatitis



# HEPATITUS VIRUS

There are five types of hepatitis -- A through E -- all of which cause inflammation of the liver. Type D affects only those who also have hepatitis B, and hepatitis E is extremely rare in the United States.

- Type A hepatitis is contracted through anal-oral contact, by coming in contact with the feces of someone with hepatitis A, or by eating or drinking hepatitis A contaminated food or water.
- Type B hepatitis can be contracted from infected blood, seminal fluid, vaginal secretions, or contaminated drug needles, including tattoo or body-piercing equipment. It can also be spread from a mother to her newborn.
- Type C hepatitis is not easily spread through sex. You're more likely to get it through contact with infected blood, contaminated razors, needles, tattoo and body-piercing equipment, or manicure or pedicure tools that haven't been properly sanitized, and a mother can pass it to her baby during delivery.
- Type D hepatitis can be passed through contact with infected blood, contaminated needles, or by sexual contact with an HIV-infected person.
- Type E hepatitis is most likely to be transmitted in feces, through oral contact, or in water that's been contaminated.



Operators analyze sludge samples to improve wasting.



## MATH CONVERSION FACTORS

1 PSI = 2.31 Feet of Water  
1 Foot of Water = .433 PSI  
1.13 Feet of Water = 1 Inch of Mercury  
454 Grams = 1 Pound  
1 Gallon of Water = 8.34 pounds/gal  
1 mg/L = 1 PPM  
17.1 mg/L = 1 Grain/Gallon  
1% = 10,000 mg/L  
694 Gallons per Minute = MGD  
1.55 Cubic Feet per Second = 1 MGD  
60 Seconds = 1 Minute  
1440 Minutes = 1 Day  
.746 kW = 1 Horsepower  
1 + 1 = 2

### LENGTH

12 Inches = 1 Foot  
3 Feet = 1 Yard  
5280 Feet = 1 Mile

### AREA

144 Square Inches = 1 Square Foot  
43,560 Square Feet = 1 Acre

### VOLUME

1000 Milliliters = 1 Liter  
3.785 Liters = 1 Gallon  
231 Cubic Inches = 1 Gallon  
7.48 Gallons = 1 Cubic Foot  
62.38 Pounds = 1 Cubic Foot

## DIMENSIONS

SQUARE:      Area (sq. ft.) = Length X Width

Volume (cu. ft.) = Length (ft) X Width (ft) X Height (ft)

CIRCLE: Area (sq. ft.) =  $3.14 \times \text{Radius (ft)} \times \text{Radius (ft)}$

CYLINDER: Volume (Cu. ft) =  $3.14 \times \text{Radius (ft)} \times \text{Radius (ft)} \times \text{Depth (ft)}$

SPHERE:  $\frac{(3.14) (\text{Diameter})^3}{(6)}$       Circumference =  $3.14 \times \text{Diameter}$



## GENERAL

POUNDS PER DAY AKA SOLIDS APPLIED =  
Concentration (mg/L) X Flow (MG) X 8.34

PERCENT EFFICIENCY =  $\frac{\text{In} - \text{Out}}{\text{In}} \times 100$

TEMPERATURE:  ${}^{\circ}\text{F} = {}^{\circ}\text{C} \times \frac{9}{5} + 32$   
 ${}^{\circ}\text{C} = ({}^{\circ}\text{F} - 32) \times \frac{5}{9}$

CONCENTRATION: Conc. (A) X Volume (A) = Conc. (B) X Volume (B)

FLOW RATE (Q):  $Q = A \times V$  (Quantity = Area X Velocity)

FLOW RATE (gpm): Flow Rate (gpm) =  $\frac{2.83 \times (\text{Diameter, in})^2 \times (\text{Distance, in})}{\text{Height, in}}$

% SLOPE =  $\frac{\text{Rise (feet)}}{\text{Run (feet)}} \times 100$

ACTUAL LEAKAGE =  $\frac{\text{Leak Rate (GPD)}}{\text{Length (mi.)} \times \text{Diameter (in)}}$

VELOCITY =  $\frac{\text{Distance (ft)}}{\text{Time (Sec)}}$

N = Manning's Coefficient of Roughness

R = Hydraulic Radius (ft.)

S = Slope of Sewer (ft/ft.)

HYDRAULIC RADIUS (ft) =  $\frac{\text{Cross Sectional Area of Flow (ft)}}{\text{Wetted pipe Perimeter (ft)}}$

WATER HORSEPOWER =  $\frac{\text{Flow (gpm)} \times \text{Head (ft)}}{3960}$

BRAKE HORSEPOWER =  $\frac{\text{Flow (gpm)} \times \text{Head (ft)}}{3960} \times \text{Pump Efficiency}$

MOTOR HORSEPOWER =  $\frac{\text{Flow (gpm)} \times \text{Head (ft)}}{3960} \times \text{Pump Eff.} \times \text{Motor Eff.}$

MEAN OR AVERAGE =  $\frac{\text{Sum of the Values}}{\text{Number of Values}}$

TOTAL HEAD (ft) = Suction Lift (ft) X Discharge Head (ft)

SURFACE LOADING RATE =  $\frac{\text{Flow Rate (gpm)}}{(\text{gal/min/sq. ft.}) \times (\text{Surface Area (sq. ft)})}$

$$\text{MIXTURE} = \frac{(\text{Volume 1, gal}) (\text{Strength 1, \%}) + (\text{Volume 2, gal}) (\text{Strength 2, \%})}{(\%) (\text{Volume 1, gal}) + (\text{Volume 2, gal})} \quad \text{STRENGTH}$$

$$\text{INJURY FREQUENCY RATE} = \frac{(\text{Number of Injuries})}{\text{Number of hours worked per year}} \cdot 1,000,000$$

$$\text{DETENTION TIME (hrs)} = \frac{\text{Volume of Basin (gals)} \times 24 \text{ hrs}}{\text{Flow (GPD)}}$$

$$\text{Slope} = \frac{\text{Rise (ft)}}{\text{Run (ft)}} \quad \text{Slope(\%)} = \frac{\text{Rise (ft)}}{\text{Run (ft)}} \times 100$$

POPULATION EQUIVALENT (PE):

1 PE = .17 Pounds of BOD per Day

1 PE = .20 Pounds of Solids per Day

1 PE = 100 Gallons per Day

$$\text{LEAKAGE (GPD/inch)} = \frac{\text{Leakage of Water per Day (GPD)}}{\text{Sewer Diameter (inch)}}$$

CHLORINE DEMAND (mg/L) = Chlorine Dose (mg/L) – Chlorine Residual (mg/L)

$\tau Q$  = Allowable time for decrease in pressure from 3.5 PSU to 2.5 PSI

$\tau q$  = As below

$$\tau Q = (0.022) (d_1^2 L_1) / Q \quad \tau q = \frac{[0.085]}{q} [(d_1^2 L_1) / (d_1 L_1)]$$

Q = 2.0 cfm air loss

$\theta$  = .0030 cfm air loss per square foot of internal pipe surface

$d$  = Pipe diameter (inches)

L = Pipe Length (feet)

$$V = \frac{1.486}{v} R^{2/3} S^{1/2}$$

V = Velocity (ft./sec.)

v = Pipe Roughness

R = Hydraulic Radius (ft)

S = Slope (ft/ft)

$$\text{HYDRAULIC RADIUS (ft)} = \frac{\text{Flow Area (ft. 2)}}{\text{Wetted Perimeter (ft.)}}$$

$$\text{WIDTH OF TRENCH (ft)} = \text{Base (ft)} + (2 \text{ Sides}) \times \frac{\text{Depth (ft 2)}}{\text{Slope}}$$

$$\text{AMPERAGE} = \frac{\text{Voltage}}{\text{Ohms}}$$

$$\text{VOLTAGE IMBALANCE} = \frac{\text{Maximum Voltage Deviation (Volts)} \times 100}{\text{Average Voltage (Volts)}}$$

## LABORATORY

TSS (mg/L) =  $\frac{\text{Paper Wt. And Dried Solids (g)} - \text{Paper Wt. (g)} \times 1,000,000}{\text{Milliliters of Sample}}$

BOD (mg/L) =  $\frac{(\text{Initial DO} - \text{Final DO}) \times 300}{(\text{unseeded}) \text{ Milliliters of Sample}}$

LANGELIER INDEX =  $\text{pH} - \text{pH}_s$

## STABILIZATION PONDS

DETENTION TIME (Days) =  $\frac{\text{Volume of Ponds (gals)}}{\text{Flow Rate (gals/day)}}$

ORGANIC LOADING (Lbs. Of BOD/Acre/Day) =  $\frac{\text{Pounds of BOD Applied per Day}}{\text{Surface Areas (Acres)}}$

## FIXED MEDIA

HYDRAULIC LOADING (gals/1000 cu. ft./day) =  $\frac{\text{Flow Rate (gals./day)}}{1000\text{'S Cubic Feet of Media}}$

ORGANIC LOADING (lbs BOD/day/1000 cu. ft.) =  $\frac{\text{Pounds of BOD applied per Day}}{1000\text{'S OF Cubic Feet of Media}}$

## ACTIVATED SLUDGE

DETENTION TIME (hrs.) =  $\frac{\text{Volume of the tank (gals)}}{\text{Flow Rate (gals/hour)}}$

SVI (mg/L) =  $\frac{\text{Settled Sludge Volume (mls)} \times 1000}{\text{MLSS (mg/L)}}$

SDI (g/ml) =  $\frac{1 \times 100}{\text{SVI}}$

F/M =  $\frac{\text{BOD (applied to aerator)} \times \text{Flow (MGD)} \times 8.34}{\text{Pounds of Solids under Aeration}}$

MCRT (Days) =  $\frac{\text{Pounds of Solids under Aeration} - \text{Pounds of Solids in Clarifier}}{\text{Pounds of Solids Wasted} - \text{B Pounds of Solids over the Weirs}}$

## DIGESTER AND SOLIDS HANDLING

OXYGEN UPTAKE RATE (OUR) =  $\frac{\text{mg of O}_2 \text{ used}}{\text{Minute}}$

ORGANIC LOADING (lbs./day/cu. ft.) =  $\frac{\text{Pounds of Volatile Solids applied per Day}}{\text{Volume of Digester (cu. ft.)}}$

VOLATILE SOLIDS REDUCTION =  $\frac{(\text{In} - \text{Out}) (100\%)}{\text{In} - (\text{In} - \text{Out})}$

DRY POLYMER (Lbs) = (Gal. Of solution) X (8.34 lbs./gal.) X (% polymer solution)

SLUDGE APPLICATION (lbs) = (Gal. Of Sludge) X (8.34 lbs./gal.) X (% Solids in sludge)

1 TON = 2,000 lbs    1 METER = 3.28 Feet





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