Wastewater Analysis

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Islamic University-Gaza
Environment and Earth Sciences Department

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Prepared By :Raed M. Al-Khaldi

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ENVT 3123—Wastewater Analysis

Meeting: Mondays 10:30 p.m. – 12:30 p.m. C 302

Instructor:

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Course website:
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Prerequisites:

CHEMA1301: General Chemistry A.
CHEMB1301: General Chemistry B.
CHEM 1202: Practical General Chemistry.
Co-requisite: CHEM 2310: Analytical Chemistry.
CHEM 2110: Analytical Chemistry Laboratory.

Textbook:

Standard Methods for the Examination of Water and Wastewater. 18th Ed. American Public Health EPA.

Other Required Materials:

Students must have a calculator, lab coat, lab report.
Note book that prepared by Instructor.

Course Purpose:

The purpose of this required course is to acquaint students with fundamental laboratory techniques used in wastewater treatment. Students will become familiar with laboratory procedures for the physical, chemical, and biochemical examination of wastewater. In addition, the students will learn to evaluate the data generated by these tests and thereby interpret and draw conclusions about water quality from the results of these tests.

Course Objective:

Upon completion of the course, the student should have knowledge of the following,

1. Know lab safety
2. Use and understand the data train and its importance in reliable data.
3. Know proper units and conversions.
4. Explain the importance of QA/QC
5. Determine sampling preservation procedures.
6. Apply general rules for interpreting results.
7. Basic wastewater quality tests that include methods of measuring these parameters.
8. Evaluate the data generated by these tests.

Class Topics and Schedule:

Laboratory: Students will be divided in to groups to perform laboratory tests. Groups will be assigned by the instructor. Groups work alone; members of each group submit lab reports individually.

Week of:

Lab no. (1)	Syllabus, Schedule, Introduction to lab safety
Lab no. (2)	Data quality, Quality assurance, Accuracy
Lab no. (3)	Introduction to water chemistry, Water/Wastewater Characteristics
Lab no. (4)	pH, Temperature, EC, and DO.
Lab no. (5)	Solids (TS, TDS, TSS)
Lab no. (6)	Solids (FS, VS, Settleable solids)
Lab no. (7)	Dissolved Oxygen, Chemical Oxygen Demand
Lab no. (8)	Biological Oxygen Demand
Lab no. (9)	Mid Term Exams
Lab no. (10)	Ammonia and Cl
Lab no. (11)	Ortho-phosphorous
Lab no. (12)	Total phosphorous
Lab no. (13)	TKN
Lab no. (14)	Final Exam

Evaluation Process

Lab Notebook and Report (5%) Each student will be responsible for keeping a lab notebook for each experiment. Each report should be handed to instructor at the next lab and the notebook will be evaluated at the end of the semester.

Quizzes (5%) Quizzes will be given randomly to assess students understanding of the course material. Quizzes will be administered at the beginning of class to cover the reading assignment for the lesson.

Class Participation (5%) Class participation includes participating in lab and attending class.

Mid-Term Exam (25%) The mid-term exam is comprehensive in nature and will emphasize problem solving, both verbally and mathematically.

Final (60%) The final will combine aspects of the homeworks and tests and include all the labs that taken before the mid term exam.

Contribution of Course to Professional Component:

This course gives students laboratory experience needed for a clear understanding of wastewater analysis. They learn not only, the definitions of physical, chemical and biochemical terms, but also how to quantify and evaluate them experimentally. The course emphasizes the use of modern lab equipment and focuses on the ability to conduct experimental tests and properly evaluate the results.

Attendance

If a student has more than 4 unexcused absences, then his/her overall course grade will drop one letter grade.

Introduction:

Water and wastewater analyses are done by several methods. The most common types of measurements are gravimetric (weighing), electrochemical (using meters with electrodes) and optical (including visual). Instrumental methods are becoming increasingly popular, and instrumentation is getting "smarter" and easier to use with the inclusion of microprocessors.

Measurement Techniques:

- Gravimetric
- Electrochemical
- Colorimetry or spectrophotometry
- Titration
- Chromatography
- Mass Spectrometry

Goals of Wastewater Analysis:

- To design the appropriate wastewater treatment plant (WWTP) for specific area.
- To monitor and evaluate efficiency of treatment processes in WWTP.
- To study the reuse of treated wastewater (in agriculture or recharge).
- To make the Environmental Impact Assessment.

Evaluation of sampling procedures:

To evaluate sampling procedures, assess the following seven areas:

. Sample collection techniques

. Field measurements

. Sample labeling (including location(s)) and documentation

. Sample preservation and holding time

. Transfer of custody and shipment of samples

. Quality control

. Data handling and reporting.

Sampling Procedures and Techniques:

For sound evaluating of sampling program, the lab inspector must be familiar with the procedures and techniques necessary for accurate sampling of wastewaters. The following discussion details the procedures for sample collection, preservation, transfer, quality control, and data handling.

Wastewater Sample Collection Techniques

Sample collection is an important part of the wastewater monitoring program. Without proper sample collection procedures, the results of such monitoring programs are neither useful nor valid, even with the most precise and accurate analytical measurements.

Selection of Representative Sampling Sites

Normally, samples should be collected at the location specified in the plan.

Influent Samples. Document and take these samples at points of high turbulence flow to ensure good mixing. The preferred sampling points for raw wastewater are:

1. Waste flowing from last process in a manufacturing operation
2. Pump wet well (if turbulent)
3. Upstream collection lines, tank, or distribution box following pumping from the wet well or sump
4. Flume throat
5. Aerated grit chamber
6. Upstream siphon following the comminutor (in absence of grit chamber).

If it is not possible to sample at a preferred point, choose an alternative location and document the basis for choosing that location.

Effluent Samples. Collect these samples at the site the permit specifies or, if the permit does not specify a site then the inspector should select the most representative site after final treatment and downstream from all entering waste streams before they enter the receiving waters. Occasionally, municipal plant permits may specify sampling prior to chlorination. For these plants, monitor all parameters at the upstream location except fecal coliforms, pH, and total residual chlorine.

The following general guidelines apply when taking samples:

1. Take samples at a site selected to yield a representative sample.
2. Use a sampling method (grab, composite, continuous) as required in the permit. Some collected are dissolved oxygen, total residual chlorine, oil and grease, coliforms, purgeable organics, sulfides, cyanide, and total phenols.
3. Avoid collecting large nonhomogeneous particles and objects.

4. Collect the sample facing upstream to avoid contamination.

5. Do not rinse sample container with sample when collecting oil and grease and microbiological samples, but fill it directly to within 2.5 to 5 cm from the top.

6. Fill the container completely if the sample is to be analyzed for purgeable organics, oxygen, ammonia, hydrogen sulfide, free chlorine, pH, hardness, sulfite, ammonium, ferrous iron, acidity, or alkalinity.

7. Collect sufficient volume to allow for quality assurance testing. (Table1 provides a guide to numerous sample volumes, but additional volumes may be necessary for quality assurance testing.)

The following general guidelines apply when using automatic samplers:

1. Collect samples where the wastewater is well mixed. Collect the sample near the center of the flow channel at 0.4 to 0.6 depths (mid-depth).

2. Obtain a sufficient volume of sample to perform all required analyses plus any additional amount for quality control. Individual portions of a composite sample should be at least 100 milliliters in order to minimize sampler solids bias.

3. For automatic samplers which use a peristaltic pump, obtain adequate flow rates in the sampler tubing to effectively transport the suspended solids. To avoid solids bias, the velocity of the wastewater in sample tubing should be at least 2 fps and the tubing diameter should be at least 0.25 inch.

4. Time of sample collection begins when the last aliquot is dispensed into the composite sample container.

Sample Volume

The volume of samples collected depends on the type and number of analyses needed, as reflected in the parameters to be measured. Obtain the volume of the sample sufficient for all the required analyses plus an additional amount to provide for any split samples or repeat analyses. Table 1 provides a guide to sample volumes required for determining the constituents in wastewater. Consult the laboratory receiving the sample for any specific volume required. EPA's *Methods for Chemical Analysis of Water and Wastes* (USEPA 1979b) and *Handbook for Sampling and Sample Preservation of Water and Wastewater* (USEPA 1982), and the current Environmental Protection Agency (EPA)-approved edition of *Standard Methods for the Examination of Water and Wastewater* [American Public Health Association (APHA),

Table (1): Volume of Sample Required for Determination of the Various Constituents of Wastewater (Associated Water and Air Resource Engineers, Inc. 1973 Handbook for Monitoring Industrial Wastewater.USEPA Technology Transfer.)

TESTS	Volume of Sample, (1) ml
PHYSICAL	
Color and Odor(2)	100 to 500
Corrosivity(2)	Flowing sample
Electrical conductivity(2)	100
pH, electrometric(2)	100
Radioactivity	100 to 1,000
Specific gravity(2)	100
Temperature(2)	Flowing sample
Toxicity(2)	1,000 to 20,000
Turbidity(2)	100 to 1,000
CHEMICAL	
Dissolved Gases:	
Ammonia,(3) NH(3)	500
Carbon dioxide,(3) free $C0_2$	200
Chlorine,(3) free Cl_2	200
Hydrogen,(3) H_2	1,000
Hydrogen sulfide,(3) H_2S	500
Oxygen,(3) O_2	500 to 1,000
Sulfur dioxide,(3) free SO_2	100
Miscellaneous:	
Acidity and alkalinity	100
Bacteria, iron	500
Bacteria, sulfate-reducing	100
Biochemical oxygen demand (BOD)	100 to 500
Carbon dioxide, total CO_2 (including CO_3^{--}, $H CO_3^-$, and free)	200
Chemical oxygen demand (dichromate)	50 - 100
Chlorine requirement	2,000 to 4,000
Chlorine, total residual Cl_2 (including OCl^-, HOCl, NH_2Cl, $NHCl_2$, and free)	200
Chloroform-extractable matter	1,000
Detergents	100 to 200
Hardness	50 to 100
Hydrazine	50 to 100
Microorganisms	100 to 200
Volatile and filming amines	500 to 1,000
Oily matter	3,000 to 5,000

Organic nitrogen	500 to 1,000
Phenolic compounds	800 to 4,000
pH, colorimetric	10 to 20
Polyphosphates	100 to 200
Silica	50 to 1,000
Solids, dissolved	100 to 20,000
Solids, suspended	50 to 1,000
Tannin and lignin	100 to 20
Cations:	
Aluminum, Al^{+++}	100 to 1,000
Ammonium,(3) NH_4^{+}	500
Antimony, Sb^{+++} to Sb^{+++++}	100 to 1,000
Arsenic, As^{+++} to As^{+++++}	100 to 1,000
Barium, Ba^{++}	100 to 1,000
Cadmium, Cd^{++}	100 to 1,000
Calcium, Ca^{++}	100 to 1,000
Chromium, Cr^{+++} to Cr^{++++++}	100 to 1,000
Copper, Cu^{++}	200 to 4,000
Iron,(3) Fe^{++} and Fe^{+++}	100 to 1,000
Lead, Pb^{++}	100 to 4,000
Magnesium, Mg^{++}	100 to 1,000
Manganese, Mn^{++} to $Mn^{+++++++}$	100 to 1,000
Mercury, Hg^{+} and Hg^{++}	100 to 1,000
Potassium, K^{+}	100 to 1,000
Nickel, Ni^{++}	100 to 1,000
Silver, Ag^{+}	100 to 1,000
Sodium, Na^{+}	100 to 1,000
Strontium, Sr^{++}	100 to 1,000
Tin, Sn^{++} and Sn^{++++}	100 to 1,000
Zinc, Zn^{++}	100 to 1,000
Anions:	
Bicarbonate, HCO_3^{-}	100 to 200
Bromide, Br	100
Carbonate, CO_3^{--}	100 to 200
Chloride, Cl^{-}	25 to 100
Cyanide, CN^{-}	25 to 100
Fluoride, Fl^{-}	200
Hydroxide, OH^{-}	50 to 100

Iodide, I^-	100
Nitrate, NO_3^{--}	10 to 100
Nitrite, NO_2^{--}	50 to 100
Phosphate, ortho, PO_4^{--}, HPO_4^{--}, $H_2PO_4^-$	50 to 100
Sulfate, SO_4^{--}, HSO_4	100 to 1,000
Sulfide, S^{--}, HS^-	100 to 500
Sulfite, SO_3^{--}, HSO_3^-	50 to 100
(1) Consider volumes specified in this table as guides for the approximate quantity of sample necessary for a particular analysis. The exact quantity used should be consistent with the volume prescribed in the standard method of analysis, whenever a volume is specified.	
(2) Use aliquots for other determinations. (3) Obtain samples for unstable constituents in separate containers, preserved as prescribed; containers must be completely filled and sealed against air exposure.	

Sample Containers

The table 2 describes required sample containers, sample preservation, and sample holding time. It is essential that the sample containers be made of chemically resistant material unaffected by the concentrations of the pollutants measured. In addition, sample containers must have a closure that will protect the sample from contamination. Collect wastewater samples for chemical analysis in plastic (polyethylene) containers. Exceptions to this general rule are oil and grease samples, pesticides, phenols, polychlorinated biphenyls (PCBs), and other organic pollutant samples. Collect these in properly cleaned glass jars or bottles and sealed. Collect bacteriological samples in properly sterilized plastic or glass containers. Collect samples that contain constituents that will oxidize when exposed to sunlight (such as iron cyanide complexes) in dark containers.

Ensure sample containers are clean and uncontaminated. Check analytical procedures to determine if they specify container cleaning procedures. Use pre-cleaned and sterilized disposable containers (e.g., polyethylene containers). If these are not used or if the analytical method does not specify procedures, *use the following procedures for cleaning sample containers:*

1. Wash with hot water and detergent.
2. Rinse with acid (e.g., nitric for metals).
3. Rinse with tap water, then rinse three or more times with organic-free water.
4. Rinse glass containers with an interference-free, redistilled solvent (such as acetone or methylene chloride for extractable organics).
5. Dry in contaminant-free area.

Table (2): Required Containers, Preservation Techniques, and Holding Times

Parameter		Container[1]	Preservative [2,3]	Maximum Holding Time[4]
BACTERIAL TESTS				
Coliform, fecal and total		P, G	Cool, 4°C 0.008% $Na_2S_2O_3$ [5]	6 hours
Fecal streptococci		P, G	Cool, 4°C 0.008% $Na_2S_2O_3$ [5]	6 hours
INORGANIC TESTS				
Acidity		P, G	Cool, 4°C	14 day
Alkalinity		P, G	Cool, 4°C	14 day
Ammonia		P, G	Cool, 4°C H_2SO_4 To pH<2	28 day
Biochemical oxygen demand		P, G	Cool, 4°C	48 hours
Biochemical oxygen demand, carbonaceous		P, G	Cool, 4°C	48 hours
Bromide		P, G	None required	28 day
Chemical oxygen demand		P, G	Cool, 4°C H_2SO_4 To pH<2	28 day
Chloride		P, G	None required	28 day
Chlorine, total residual		P, G	None required	Analyze immediately
Color		P, G	Cool, 4°C	48 hours
Cyanide, Total and amenable to chlorination		P, G	Cool, 4 °C NaOH To pH> 12 0.6 g ascorbic acid [5]	14 days [6]
Fluoride		P	None required	28 day
Hardness		P, G	HNO_3 To pH< 2, H_2SO_4 To pH<2	6 months
Hydrogen Ion (pH)		P, G	None required	Analyze immediately
KjeldAhl and organic nitrogen		P, G	Cool, 4°C H_2SO_4 To pH<2	28 day
METALS [7]				
Chromium VI		P, G	Cool, 4°C	24 day
Mercury		P, G	HNO_3 to pH<2	28 day
Metals except above		P, G	HNO_3 to pH<2	6 months
Nitrate		P, G	Cool, 4°C	48 hours
Nitrate-nitrite		P, G	Cool, 4°C H_2SO_4 To pH<2	28 day
Nitrite		P, G	Cool, 4°C	48 hours
Oil and grease		G	Cool,4°C HCl, H_2SO_4 to pH<2	28 day
Organic carbon		P, G	Cool,4°C HCl, H_2SO_4 to pH<2	28 day
Orthophosphate phosphorus		P, G	Filter immediately Cool,4°C	48 hours
Dissolved oxygen	Probe	G bottle & top	None required	Analyze immediately
	Winkler	G bottle & top	Fix onsite and store in the dark	8 hours

Phenols	G	Cool, 4°C H_2SO_4 To pH<2	28 day
Phosphorus (elemental)	G	Cool,4°C	48 hours
Phosphorus, total	P, G	Cool, 4°C H_2SO_4 To pH<2	28 day
Residue, total	P, G	Cool,4°C	7 days
Residue, filterable	P, G	Cool,4°C	7 days
Residue, non filterable (TSS)	P, G	Cool,4°C	7 days
Residue, settleable	P, G	Cool,4°C	48 hours
Residue, volatile	P, G	Cool,4°C	7 days
Silica	P	Cool,4°C	28 day
Specific conductance	P, G	Cool,4°C	28 day
Sulfate	P, G	Cool,4°C	28 day
Sulfide	P, G	Cool,4°C, add zinc acetate plus sodium hydroxide to pH >9	7 days
Sulfite	P, G	None required	Analyze immediately
Surfactants	P, G	Cool,4°C	48 hours
Temperature	P, G	None required	Analyze immediately
Turbidity	P, G	Cool,4°C	48 hours
ORGANIC TESTS[8]			
Purgeable halocarbons	G, teflonlined septum	Cool, 4°C 0.008% $Na_2S_2O_3$ [5]	14 days
Purgeable aromatic hydrocarbons	G, teflonlined septum	Cool,4°C 0.008% $Na_2S_2O_3$ 5 HCl to pH 2[9]	14 days
AcroLein and acrylonitrile	G, teflonlined septum	Cool,4°C 0.008% $Na_2S_2O_3$ [5] Adjust pH to 4-5[10]	14 days
Phenols [11]	G, teflonlined cap	Cool, 4°C 0.008% $Na_2S_2O_3$ [5]	7 days until extraction 40 days after extraction
Benzidenes[11]	G, teflonlined cap	Cool, 4°C 0.008% $Na_2S_2O_3$ [5]	7 days until extraction[13]
Phthalate esters[11]	G, teflonlined cap	Cool,4°C	7 days until extraction; 40 days after extraction
Nitrosamines[11,14]	G, teflonlined cap	Cool, 4°C 0.008% $Na_2S_2O_3$ [5] Store in the dark	7 days until extraction; 40 days after extraction
Polychlorinated biphenyls (PCBs) [11]	G, teflonlined cap	Cool,4°C	7 days until extraction; 40 days after extraction
Nitroaromatics and isophorone[11]	G, teflonlined cap	Cool, 4°C 0.008% $Na_2S_2O_3$ [5] Store in the dark	7 days until extraction; 40 days after extraction
Polynuclear aromatic hydrocarbons[11]	G, teflonlined cap	Cool, 4°C 0.008% $Na_2S_2O_3$ [5] Store in the dark	7 days until extraction; 40 days after extraction
Haloethers[11]	G, teflonlined cap	Cool, 4°C 0.008% $Na_2S_2O_3$ [5] Store in the dark	7 days until extraction; 40 days after extraction
Chlorinated hydrocarbons[11]	G, teflonlined cap	Cool,4°C	7 days until extraction; 40 days after extraction
2,3,7,8-tetrachlorodibenzo-pdioxin[11]	G, teflonlined cap	Cool, 4°C 0.008% $Na_2S_2O_3$ [5]	7 days until extraction; 40 days after extraction

PESTICIDES TEST			
Organochlorine pesticides[11]	G, teflonlined cap	Cool, 4°C pH 5-9[15]	7 days until extraction; 40 days after extraction
RADIOLOGICAL TEST			
Alpha, beta, and radium	P, G	HNO_3 to pH< 2	6 months
WHOLE EFFLUENT TOXICITY TESTS			
Acute and Chronic, for NPDES Compliance	P,G	Cool, 0-6 °C No Additions	36 hours to test initiation

[1] Polyethylene (P) or glass (G).

[2] Perform sample preservation steps immediately upon sample collection. for composite chemical samples, preserve each aliquot at the time of collection. When use of an automatic sampler makes it impossible to preserve each aliquot, then preserve chemical samples by maintaining at 4°C until compositing and sample splitting are completed.

[3] When shipping any sample by common carrier or sent through the United States mail, comply with the Department Of Transportation Hazardous Materials Regulations (49 *CFR* Part172). the person offering such material for transportation is responsible for ensuring such compliance. For the preservation requirements of this Table, 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_3) In water solutions at concentrations of 0.1 5% by weight or less (pH about 1.62 orgreater); sulfuric acid (H_2SO_4) 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 0.08% by weight or less (pH about 12.3 or less).

[5] Used only in the presence of residual chlorine.

[6] Maximum holding time is 24 hours when sulfide is present. Optionally, Test all samples with lead acetate paper before pH adjustments to determine whether sulfide is present. If Sulfide is present, remove by the addition of cadmium nitrate powder until a negative spot test is obtained. Filter The sample then NaOH Is added to pH 12.

[7] Filter samples should be filtered immediately onsite before adding preservative for dissolved metals.

[8] Guidance applies to samples to be analyzed by GC, LC, or GC/MS for specific organic compounds.

[9] Analyze samples receiving no pH adjustment within 7 days of sampling.

[10] pH adjustment is if not needed if not measuring acrolein. Analyze samples for acrolein receiving no pH adjustment within 3 days of sampling.

[11] When the extractable analytes of concern fall within a single chemical category, observe the specified preservation and maximum holding times for optimum safeguarding of sample integrity. When The analytes of concern fall within two or more chemical categories, preserve the sample by cooling to 4 ° C, Reducing residual chlorine with 0.008% sodium thiosulfate, storing in the dark, and adjusting the pH to between 6 and 9; hold samples preserved in this manner for 7 days before extraction and for 40 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 and 13 (re: the analysis of benzidine).

[12] If 1,2-diphenylhydrazine is likely to be present, adjust the pH of the sample to 4.0 ± 0.2 to prevent rearrangement to benzidine.

[13] Store extracts up to 7 days before analysis if storage is conducted under an inert (oxidant-free) atmosphere.

[14] For the analysis of diphenylnitrosamine, add 0.008% $Na_2S_2O_3$ and adjust pH to between 7 and 10 with NaOH within 24 hours of sampling.

[15] Perform the pH Adjustment upon receipt at the laboratory and omit if the samples are extracted within 72 hours of collection. Or the analysis of aldrin, add 0.008% $Na_2S_2O_3$.

Sample labeling:

Identify each sample accurately and completely. Use labels or tags to identify the samples that are moisture-resistant and able to withstand field conditions. Use a waterproof pen to complete the labels or tags. A numbered label or tag associated with a field sample data sheet containing detailed information on the sample is preferable to using only a label or tag for information1.

The information for each sample should include the following:

. Facility name/location

. Sample site location

. Sample number

. Name of sample collector

. Date and time of collection

. Indication of grab or composite sample with appropriate time and volume information

. Identification of parameter to be analyzed

. Preservative used.

Wastewater Sample Preservation and Holding Time:

In most cases, wastewater samples contain one or more unstable pollutants that require immediate (e.g., within 15 minutes) preservation and/or analysis. Provide appropriate chemical preservation before transferring samples to the laboratory. Procedures used to preserve samples include cooling, pH adjustment, and chemical treatment. For some parameters such as cyanide and phenols, add preservatives to sample bottles prior to or immediately following sample collection. For many samples, if preservatives are not appropriately used, bacteria can quickly degrade certain constituents (such as phenols and phosphorus). Other constituents may volatilize (such as volatile organics and sulfides) or may react to form different chemical species (hexavalent chromium, for example). Proper preservation and holding times are essential to ensure sample integrity. Analysis of samples within one day ensures against error from sample deterioration. However, such prompt analysis is not feasible for composite samples in which portions may be stored for as long as 24 hours. Where possible, provide sample preservation during compositing, usually by refrigeration to 4°C (or icing). If using an automatic sampler with ice, replace the ice as necessary to maintain low temperatures. This is a particular limitation of automatic samplers used during the summer when ice must be frequently replaced.

The times listed at the table are the maximum holding times between sample collection and analysis that are allowed for the sample to be considered valid. Typically, the holding time limitations begin upon combination

of the last aliquot in a sample. When use of an automatic sampler makes it impossible to preserve each aliquot, the preservation (chemical) should be done immediately following the composite.

Transfer of Custody and Shipment of Samples:

To ensure the validity of the permit compliance sampling data in court, written records must accurately trace the custody of each sample through all phases of the monitoring program. The primary objective of this chain-of-custody is to create an accurate written record that can be used to trace the possession and handling of the sample from the moment of its collection through its analysis and introduction as evidence.

. Use sample seals to protect the sample's integrity from the time of collection to the time it is opened in the laboratory. The seal should indicate the *collector's name*, the *date* and *time* of *sample collection*, and *sample identification number*.

. Pack samples properly to prevent breakage. Seal or lock the shipping container to readily detect any evidence of tampering can be readily detected. Use of tamper proof evidence tape is recommended.

. Place samples on ice or synthetic ice substitute that will maintain sample temperature at 4°C throughout shipment.

. Accompany every sample with a sample tag and a chain-of-custody record that has been completed, signed, and dated. The chain-of-custody record should include the *names of sample collectors*, *sample identification numbers*, *date* and *time* of *sample collection*, *location of sample collection*, and *names and signatures of all persons handling the sample in the field and in the laboratory*.

. The responsibility for proper packaging, labeling, and transferring of possession of the sample lies with the inspector.

. Accompany all sample shipments with the chain-of-custody record and other pertinent forms. The originator retains a copy of these forms. Also, the originator must retain all receipts associated with the shipment.

. When transferring possession of samples, the transferee must sign and record the date and time on the chain-of-custody record (use the currently approved record). In general, make custody transfers for each sample, although samples may be transferred as a group, if desired. Each person who takes custody must fill in the appropriate section of the chain-of-custody record.

Quality Control:

Conduct control checks during the actual sample collection to determine the performance of sample collection techniques. In general, the most common monitoring errors usually are improper sampling methodology, improper preservation, inadequate mixing during compositing and splitting, and excessive sample holding time. In addition, collect and analyze the following samples to check sample collection techniques:

Blanks:

1. Trip Blank. This is a sample vial(s) filled at the laboratory with deionized water. The blank(s) follows the same handling and transport procedures as the samples collected during the event. The blank(s) functions as a check on sample contamination originating from sample transport, shipping and from site conditions.

Note: Expose the trip blank vial(s), to the same environmental conditions (i.e., light, temperature, etc.) of the sample vial(s) but do not open until it is time for analysis.

2. Field Blank/Field Reagent Blank. These are similar to the trip blanks except they are prepared in the field with deionized water exactly as the sample(s) that are collected. Field blanks are used to check for analytical artifacts and/or background introduced by sampling and analytical procedures.

3. Equipment/Rinsate Blank. Collect a blank when using an automatic sampler or other non-dedicated equipment during the sampling process. The blank is a check of the equipment cleanliness. For automatic samplers, prepare blanks prior to collecting samples, by pumping deionized organic free water through the sampler and collecting the discharge purge water in a sample container for analysis for the constituents of concern.

Field Duplicate. Collect this sample simultaneously from the same source at selected stations on a random time frame by grab samples or from two sets of field equipment installed at the site. Duplicate samples check analytical precision as well as evaluate the "representativeness" of the sample aliquot.

Split Samples. These are samples that have been divided into two containers for analysis by separate laboratories. These samples provide an excellent means of identifying discrepancies in the permittee's analytical techniques and procedures. When filling split samples from a single composite jug, shake the composited sample well and half fill the EPA sample container, then shake the composite again and fill half of the permittee's container. Repeat the procedure for each parameter collected.

The laboratories performing the sample analyses should also use the following control measures:

Prepare/Reagent Blank. A sample consisting of reagent(s), without the target analyte or sample matrix, introduced into the analytical procedure at the appropriate point and carried through all subsequent steps to determine the contribution of the reagents and of the involved analytical steps to error in the observed value.

Quality Control Sample. This is an uncontaminated sample matrix spiked with known amounts of analytes from a source independent from the calibration standards. Use this sample to establish intra laboratory or analyst specific precision and bias or to assess the performance of all or a portion of the measurements' system.

Matrix Spike/Matrix Spike Duplicate (MS/MSD). This sample is three times the normal volume required for a specific chemical analysis to which a known quantity of analyte has been added prior to all sample

preparation. The laboratory utilizes the MS/MSD samples as part of their Quality Assurance/Quality Control Program.

. Use a matrix spike to verify accuracy of the analytical procedures.

. A matrix spike duplicate is a duplicate of a matrix spike sample. It measures the precision of the analysis in terms of relative percent difference.

Data Handling and Reporting:

Verified analytical results are normally entered into a laboratory data management system of some type. The system should contain the sampling data, including:

1. Time and exact location,
2. Analysis dates and times,
3. Names of analysts,
4. Analytical methods/techniques used, and
5. Analytical results.

Data are then reported to the project officer for inclusion into the compliance report.

Sample Types

Two types of sample techniques are used: grab and composite.

Grab Samples: Grab samples are individual samples collected over a period of time not exceeding 15 minutes and are representative of conditions at the time the sample is collected.

The sample volume depends on the type and number of analyses to be performed. The collection of a grab sample is appropriate when a sample is needed to:

1. Sample an effluent that does not discharge on a continuous basis
2. Provide information about instantaneous concentrations of pollutants at a specific time
3. Allow collection of a variable sample volume
4. Corroborate composite samples
5. Monitor parameters not amenable to compositing (e.g., pH, temperature, dissolved oxygen, chlorine, purgeable organics, oil and grease, coliform bacteria. Volatile organics, sulfides, phenols, and phosphorus samples can be composited. If you composite use special handling procedures.

Composite Samples: Collect these samples over time, either by *continuous sampling* or by *mixing discrete samples*, and represent the average characteristics of the waste stream during the compositing period use. Composite samples are used when:

1. Average pollutant concentration during the compositing period is determined
2. Mass per unit time loadings is calculated
3. Wastewater characteristics are highly variable.

Various methods for compositing samples are available, select one based on either time or flow proportioning. Table 3 lists the advantages and disadvantages of various methods. The permit may specify which type of composite sample to use. Collect composite samples either manually or with automatic samplers. Inspectors should consider variability in waste stream flow rate and parameter concentrations carefully when choosing compositing methods, sampling equipment (tubing and containers), and quality assurance procedures.

The compositing methods are as follows:

1. Time Composite Sample: This method requires discrete sample aliquots collected in one container at constant time intervals. This method is appropriate when the flow of the sampled stream is constant (*flow rate does not vary more than 10 percent of the average flow rate*) or when flow monitoring equipment is not available.

2. Flow-Proportional Composite Sample: There are two methods used for this type of sample. One method collects a constant sample volume at varying time intervals proportional to stream flow (e.g., 200 milliliters sample collected for every 5,000 gallons of flow). In the other method, collect the sample by increasing the volume of each aliquot as the flow increases, while maintaining a constant time interval between the aliquots.

3. Sequential Composite Sample: This method requires discrete samples collected in individual containers at constant time intervals or discharge increments. For example, samples collected every 15 minutes, composited into separate containers each hour. The discrete samples can then be manually flow-proportioned to form the composite sample.

Alternatively, take a constant sample volume at constant discharge increments, as measured with a totalizer.

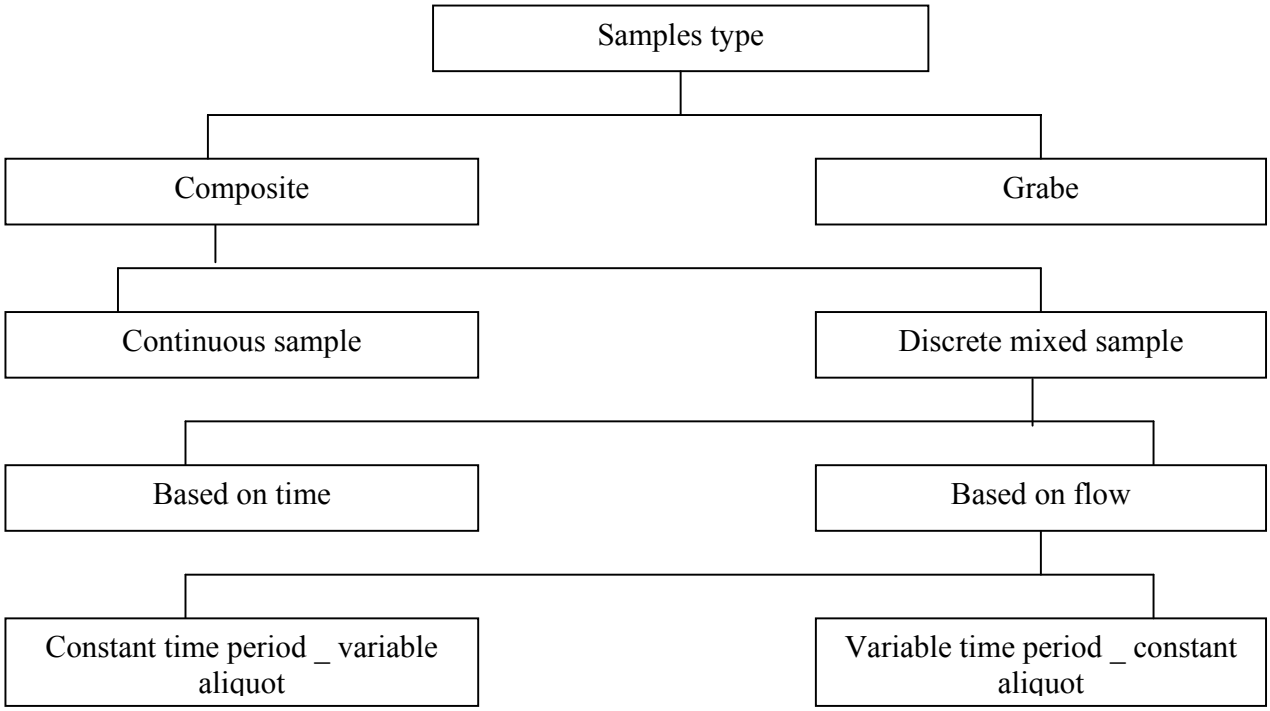
Continuous Composite Sample: Collect this sample continuously from the waste stream.

The sample may be constant volume, or the volume may vary in proportion to the flow rate of the waste stream.

Table (3): The advantages and disadvantages of various compositing methods

Methods	Advantage	Disadvantage	Comments
Time composite			
Constant sample volume, constant time interval between samples	Minimal instrumentation and manual effort; requires no flow measurement	May lack representativeness, especially for highly variable flows	Widely us ed in both automatic samplers and manual handling
Flow proportional composite			
Constant sample volume, time interval between samples proportional to stream flow	Minimal manual effort	Requires accurate flow measurement reading equipment; manual compositing from flowchart	Widely us ed in automatic as well as manual sampling
Constant time interval between samples, sample volume proportional to total stream flow at time of sampling	Minimal instrumentation	Manual compositing from flowchart in absence of prior information on the ratio of minimum to maximum flow; chance of collection g too small or too large individual discrete samples for a given composite volume	Use d in auto ma tic samplers and widely used as manual method
Constant time interval between samples, sample volume proportional to total stream flow since last sample	Minimal instrumentation	Manual compositing from flow chart in absence of prior information on the ratio of minimum to maximum flow; chance of collecting either too small or too large individual discrete samples for a given composite volume	Not widely used in automatic samplers but may be done manually
Sequential composite			
Series of short period composites, constant time intervals between samples	Useful if fluctuations occur and time history is desired	Requires manual compositing of aliquots based on flow	Commonly used; however, manual compositing is labor intensive
Series of short period	Useful if fluctuations occur an	Requires flow totalizer;	Manual com positing is labor

composites, aliquots taken at constant discharge increments	d the time history is desired	requires manual com positing of aliquots based on flow	intensive
Continuous composite			
Constant sample volume	Minim al manual effort, requires no flow measurement highly variable flows	Requires large sample capacity; may lack representativeness for highly variable flows	Practical but not widely used
Sample volume proportional to stream flow	Minim al manual effort, most representative especially for highly variable sample volume, variable pumping capacity and power	Requires accurate flow measurement equipment, large sample volume, variable pumping capacity, and power	Not widely used



Field Measurements

Temperature

Measurement:

Calibrate an immersion thermometer or thermocouple by comparing its readings with those of the reference thermometer at two different temperatures that bracket the temperature range normally measured in the field.

Calibration must be done at least annually and whenever the instrument is suspected of having been misused, damaged, or producing erratic or erroneous readings. The instrument should be within ±1.0 °C compared with a thermometer calibrated to the National Institute of Standards and Technology standards.

Obtain a representative wastewater sample.

Place the temperature probe into the wastewater sample, read the temperature upon stabilization, and record the value in the field notebook.

Some pH and electrical conductivity meters are equipped with a temperature probe, which can be used to simultaneously measure sample temperature.

Dissolved Oxygen

Like solids and liquids, gases can dissolve in water. And, like solids and liquids, different gases vary greatly in their solubilities, *i.e,* how much can dissolve in water. A solution containing the maximum concentration that the water can hold is said to be *saturated*. Oxygen gas, the element which exists in the form of O_2 molecules, is not very water soluble. A saturated solution at room temperature and normal pressure contains only about 9 parts per million of D.O. by weight (9 mg/L). Lower temperatures or higher pressures increase the solubility, and *visa versa*.

Significance: Dissolved oxygen is essential for fish to breathe. Many microbial forms require it, as well. The oxygen bound in the water molecule (H_2O) is not available for this purpose, and is in the wrong "oxidation state", anyway. The low solubility of oxygen in water means that it does not take much oxygen-consuming material to deplete the D.O. As mentioned before, the biodegradation products of bacteria which do not

require oxygen are foul-smelling, toxic, and/or flammable. Sufficient D.O. is essential for the proper operation of many wastewater treatment processes. Activated sludge tanks often have their D.O. monitored continuously. Low D.O.'s may be set to trigger an alarm or activate a control loop which will increase the supply of air to the tank.

Measurement:

D.O. can be measured by a fairly tricky wet chemical procedure known as the Winkler titration. The D.O. is first trapped, or "fixed", as an orange-colored oxide of manganese. This is then dissolved with sulfuric acid in the presence of iodide ion, which is converted to iodine by the oxidized manganese. The iodine is titrated using standard sodium thiosulfate. The original dissolved oxygen concentration is calculated from the volume of thiosulfate solution needed.
Measurements of D.O. can be made more conveniently with electrochemical instrumentation. "D.O. meters" are subject to less interference than the Winkler titration. Most dissolved-oxygen meters may be auto-calibrated on site by simply exposing the probe to saturated air and pressing the calibration button.

Obtain a representative wastewater sample. Place the dissolved-oxygen probe into the wastewater sample and record the final value of dissolved-oxygen concentration to the nearest 0.1 mg/l.

pH

Introduction:

Measurement of pH is one of the most important and frequently used tests in water chemistry. Practically every phase of water supply and wastewater treatment is pH-dependent. At a given temperature the intensity of the acidic or basic character of a solution is indicated by hydrogen ion activity. Modern instruments and the development of a wide variety of electrode designs have made the measurement of pH almost as simple and convenient as the measurement of temperature.

Basic Theory:

In general, pH is a measure of the degree of acidity or alkalinity of a substance. It's related to the active acid concentration of a solution by this equation:

$pH = -\log a_{H^+}$

with a_{H^+} representing the activity of the hydrogen ions in the solution. Neglecting activity effects, the equation above reduces to:

$pH = -\log [H^+]$

with $[H^+]$ representing the concentration of the hydrogen ions in the solution. pH is sometimes referred to as the power of the hydrogen ion in solution. Thus, the pH of the strong acid 0.01 molar HCl is equal to 2, since the hydrogen ion concentration is 10^{-2} molar:

$pH = -\log 10^{-2} = 2$

The pH scale in water usually ranges from 0 to 14. Although concentrations outside this range can and do exist, they are not generally encountered in practice.

The pH scale is based on the dissociation constant of water. In pure water, a very small number of molecules react with one another to form hydronium ions (H_3O^+), which account for acidic properties, and hydroxide ions (OH^-), which account for the basic properties of an aqueous solution.

$$2H_2O \rightleftharpoons H_3O^+ + OH^-$$

or for simplicity:

$$H_2O \rightleftharpoons H^+ + OH^-$$

At 25°C, pure water is very slightly ionized and at equilibrium the acid [H^+] and base [OH^-] concentrations are equal, at 1×10^{-7} molar. The product of both concentrations is the dissociation constant Kw:

$$Kw = [H^+][OH^-] = (1\times10^{-7})(1\times10^{-7}) = 1\times10^{-14}$$

Because the hydrogen ion concentration equals 1×10^{-7} molar, the pH of pure water at 25°C is 7. This is referred to as the neutrality point.

In aqueous solutions at the 25°C, the product of [H^+] and [OH^-] must always remain constant at 1×10^{-14}. Therefore, an increase in either the acid or base concentration will always result in a decrease in the other term. Hence, a solution of the strong base 0.01 M NaOH will have a hydrogen ion concentration of:

$$[H^+] = Kw / [OH^-] = 10^{-14}/10^{-2} = 10^{-12}$$

Thus: $pH = -\log 10^{-12} = 12$

The Standard 0-14 pH Scale*

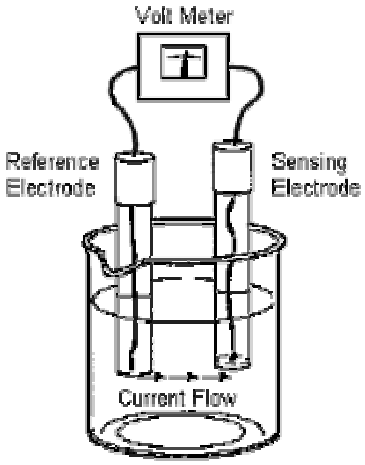
Active Acid Concentration [H^+]	pH	Active Base Concentration [OH^-]
1.0	0	0.00000000000001
0.1	1	0.0000000000001
0.01	2	0.000000000001
0.001	3	0.00000000001
0.0001	4	0.0000000001
0.00001	5	0.000000001
0.000001	6	0.00000001
0.0000001	7	0.0000001
0.00000001	8	0.000001

0.000000001	9	0.00001
0.0000000001	10	0.0001
0.00000000001	11	0.001
0.000000000001	12	0.01
0.0000000000001	13	0.1
0.00000000000001	14	1.0

* For more information, refer to "Determination of pH: Theory and Practice", Roger G. Bates, John Wiley and Sons Inc., New York.

Measurement:

Although the pH value of a sample can be determined using colorimetric indicator solutions or indicator papers, the prefered method is potentionmetric measurement of hydrogen ions activity using a sensing electrod and a reference electrod. When certain electrodes come in contact with a sample, a potential develops across the membrane surface. The magnitude of the potential as a voltage relates to the concentration of ions.



Refer to the operations manual for calibration instructions specific to the pH meter in use. A general calibration procedure is given below.

Commercially prepared buffered calibration solutions are available for pH values of 4.01, 7.0, and 10.01 standard units. At least two standard solutions are required. Check the expiration dates on the buffer solutions to verify that the solutions are still valid for equipment calibration.

Choose two buffer solutions that bracket the expected sample pH. Attempt auto -calibration mode by immersing the pH electrode in the first solution and pressing the calibrate button. After the reading has stabilized, record the value . Rinse electrode with de-ionized water and repeat using the second solution. If manual calibration is required, digitally enter the appropriate pH values after reading each buffer solution.

Always rinse the end of the pH probe and blot dry before putting it into a solution. Gently mix the solution with a magnetic stirrer being careful not to cause a lot of surface disturbance. With the meter in measure mode, re-measure and record the values for each solution. Wait for the pH to stabilize and then record the reading.

The pH probe should be stored wet when not in use. The type of storage solution will depend upon the recommendations of the manufacturer and the type of pH probe. However, in almost all cases a buffer or salt solution is used. Do not use distilled or deionized water as this may dilute the probe's internal solution.

Conductivity

Basic Theory:

Conductance is the inverse of resistance, and conductivity is the inverse of resistivity. Resistivity is the preferred parameter when measuring the electrical properties of a solid, but in liquids the conductivity vanes (approximately) linearly with ionic concentration. The unit for conductivity is the conductance multiplied by a constant that corrects for the geometry of the measuring system, which varies with both the shape and distance between the electrodes and the solution volume. This proportionality constant is called the cell constant K, and is typically expressed in units of cm^{-1}. By convention, conductivity is expressed as microSiemens/cm (equivalent to micromho/cm), and although the display on the instrument shows μS for simplicity, the displayed value is actually μS/cm.

$$\text{conductivity} = K / M\Omega = \mu S/cm$$

The different forms of expressing conductivity can be converted by using the following relationships:

$$\mu\text{Siemen/cm} = \mu\text{mho/cm} = 1000 \times \text{mSiemen/cm}$$
$$\text{mSiemen/cm} = 100 \times \text{mSiemen/m} = 10 \times \mu\text{Siemen/m}$$

The instrument measures the conductivity of the sample solution by measuring the transmission of an AC voltage at 1 kHz, avoiding errors due to polarization of the electrodes.

Tips and Advice:

- Trapped air bubbles can be a source of error. It is easy to see the bubbles if you are measuring water with a beaker cell, but it is more difficult to check when using dip cells. Lightly tap a dip cell to dislodge trapped air.
- If you are measuring below 5 μS/cm, the reading may drift higher as carbon dioxide from the air is absorbed. When measuring conductivity below 1 microSiemen, it is best to use the 0.01 K cell. This special cell is designed as a flow-through cell to avoid carbon dioxide contamination. The stainless steel design also minimizes water velocity and electrochemical errors.
- When measuring very saline solutions (high conductivity: above 50 mS/cm) we recommend the 10K cell.

- In alkaline solutions (i.e. pH> 12) the epoxy body may cause problems. Contact manufacturer for stainless steel or glass cells.

Measurement:

Use a battery operated electrical conductivity meter that is equipped with a temperature compensator and reads directly in mmho/cm at 25°C.

Use certified calibration standards to calibrate the conductivity meter. Commercially available standards come in 100, 500, 1,000, 10,000, 50,000, and 100,000 mmho/cm solutions. Use two standard solutions that are within an order of magnitude of the expected sample value.

Turn on the meter and adjust the conductivity range selector to the appropriate setting. Rinse the conductivity probe with deionized water and then measure the conductivity of the standard solution.

If the meter auto-calibrates, read and record the conductivity value immediately upon stabilization. If the meter requires manual calibration, digitally enter the appropriate solution value. Remeasure the solution in measure mode and record the value.

Obtain a representative wastewater sample.

Rinse the probe with deionized water. With the meter in measure mode, immerse the probe in the wastewater sample and record the reading.

Laboratory Measurements

Solids

These methods quantify solids in wastewater samples using gravimetric analysis following oven drying. Solids refer to matter suspended or dissolved in the water or wastewater and may affect water or effluent quality in adverse ways. Waters with high dissolved solids generally are of inferior palatability and may induce unfavorable physiological reactions in transient consumers. Solids analyses are important in the control of biological and physical wastewater treatment processes and for assessing compliance with regulatory agency limitations.

Total Solids Dried at 103–105°C

1. General Discussion

a. Principle: A well-mixed sample is evaporated in a weighed dish and dried to constant weight in an oven at 103 to 105°C. The increase in weight over that of the empty dish represents the total solids. The results may not represent the weight of actual dissolved and suspended solids in wastewater samples.

b. Interferences*:* Highly mineralized water with a significant concentration of calcium, magnesium, chloride, and/or sulfate may be hygroscopic and require prolonged drying, proper desiccation, and rapid weighing. Exclude large, floating particles or submerged agglomerates of nonhomogeneous materials from the sample if it is determined that their inclusion is not desired in the final result. Disperse visible floating oil and grease with a blender before withdrawing a sample portion for analysis. Because excessive residue in the dish may form a water-trapping crust, limit sample to no more than 200 mg residue.

2. Apparatus

a. Evaporating dishes: Dishes of 100-mL capacity made of one of the following materials:

1) Porcelain, 90-mm diam.

2) Platinum—Generally satisfactory for all purposes.

3) High-silica glass.*

b. Muffle furnace for operation at 550°C.

c. Steam bath.

d. Desiccator, provided with a desiccant containing a color indicator of moisture concentration or an instrumental indicator.

e. Drying oven, for operation at 103 to 105°C.

f. Analytical balance, capable of weighing to 0.1 mg.

g. Magnetic stirrer with TFE stirring bar.

h. Wide-bore pipets.†

i. Graduated cylinder.

j. *Low-form beaker.*‡

3. Procedure

a. Preparation of evaporating dish: If volatile solids are to be measured ignite clean evaporating dish at 550°C for 1 h in a muffle furnace. If only total solids are to be measured, heat clean dish to 103 to 105°C for 1 h. Store and cool dish in desiccator until needed. Weigh immediately before use.

b. Sample analysis: Choose a sample volume that will yield a residue between 2.5 and 200 mg. Pipet a measured volume of well-mixed sample, during mixing, to a preweighed dish. For homogeneous samples, pipet from the approximate midpoint of the container but not in the vortex. Choose a point both middepth and midway between wall and vortex. Evaporate to dryness on a steam bath or in a drying oven. Stir sample with a magnetic stirrer during transfer. If necessary, add successive sample portions to the same dish after evaporation. When evaporating in a drying oven, lower temperature to approximately 2°C below boiling to prevent splattering. Dry evaporated sample for at least 1 h in an oven at 103 to 105°C, cool dish in desiccator to balance temperature, and weigh. Repeat cycle of drying, cooling, desiccating, and weighing until a constant weight is obtained, or until weight change is less than 4% of previous weight or 0.5 mg, whichever is less. When weighing dried sample, be alert to change in weight due to air exposure and/or sample degradation. Analyze at least 10% of all samples in duplicate. Duplicate determinations should agree within 5% of their average weight.

4. Calculation

$$\text{Total solids mg /L} = \frac{(A - B) \text{ X } 1000}{\text{sample volume, mL}}$$

where:

A = weight of dried residue + dish, mg, and

B = weight of dish, mg.

Total Dissolved Solids

Principle: A well-mixed sample is filtered through a standard glass fiber filter, and the filtrate is evaporated to dryness in a weighed dish and dried to constant weight at 180°C. The increase in dish weight represents the total dissolved solids.

Apparatus:

- All apparatus in TS determination is required, and in addition:

- Filtration apparatus.
- Suction flask and suction bump.
- Drying oven, for operation at 180 ± 2°C.

Procedure:

a. *Preparation of glass-filter disk*: Insert disk with wrinkled side up into filtration apparatus. Apply vacuum and wash disk with three successive 20-mL volumes of reagent-grade water.

Continue suction to remove all traces of water. Discard Washings.

b. *Preparation of evaporating dish*: If volatile solids are to be measured, ignite cleaned evaporating dish at 550°C for 1 h in a muffle furnace. If only total dissolved solids are to be measured, heat clean dish to 180 ± 2°C for 1 h in an oven. Store in desiccator until needed. Weigh immediately before use.

c. *Selection of filter and sample sizes*: Choose sample volume to yield between 10 and 200 mg dried residue. If more than 10 min are required to complete filtration, increase filter size or decrease sample volume. When very low total suspended solids are encountered (less than 10 mg/L), less dried residue may be collected; compensate by using a high-sensitivity balance (0.002 mg).

d. *Sample analysis*: stir sample with a magnetic stirrer and pipet a measured volume onto a glass-fiber filter with applied vacuum. Wash with three successive 10-mL volumes of reagent-grade water, allowing complete drainage between washings, and continue suction for about 3 min after filtration is complete. Transfer total filtrate (with washings) to a weighed evaporating dish and evaporate to dryness on a steam bath or in a drying oven. If necessary, add successive portions to the same dish after evaporation. Dry evaporated sample for at

least 1 h in an oven at 180 ± 2°C, cool in a desiccator to balance temperature, and weigh. Repeat drying cycle of drying, cooling, desiccating, and weighing until a constant weight is obtained or until weight change is less than 4% of previous weight or .5 mg, whichever is less. Duplicate determinations should agree within 5% of their average.

Calculation:

Total dissolved solids mg /L = (A-B) × 1000 / sample volume, ml.

Where:

A = weight of dried residue + dish, mg, and

B = weight of dish, mg.

Total Suspended Solids

Principle: A well- mixed sample is filtered through a weighed standard glass-fiber filter and the residue retained on the filter is dried to a constant weight at 103 to 105°C. The increase in weight of the filter represents the total suspended solids. If the suspended material clogs the filter and prolongs filtration. The difference between the total solids and the total dissolved solids may provide an estimate of the total suspended solids.

Apparatus:

- All apparatus in TS determination is required, and in addition:

- Filtration apparatus.
- Suction flask and suction bump.
- Drying oven, for operation at 105 ± 2°C.

Procedure:

a. Preparation of glass-fiber disk: Insert disk with wrinkled side up in a. filtration apparatus. Apply vacuum and wash disk with three successive 20-mL portions of reagents-grade water. Continue suction to remove all traces of water, and discard washings. Remove filter from filtration apparatus and transfer to an inert aluminum-weighing dish. Alternatively weigh dried filter and weighing dish both before and after filtration. Filter material that sticks to the dish must be added to the filter to avoid error. If a Gooch crucible is used, remove crucible and filter combination. Dry in an oven at 103 to 105°C for 1 h. if volatile solids are to be measured, ignite at 550°C for 15 min in a muffle furnace. Cool in desiccator to balance temperature and weigh. Repeat cycle of drying or igniting, cooling, desiccating, and weighing until a constant weight is obtained or until weight change is less than 4% of the previous weighing or 0.5 mg, whichever is less. Store in desiccator until needed.

b. *Sample analysis:* Assemble filtering apparatus and filter and begin suction. Wet filter with a small volume of reagent-grade water to seat it. Stir sample with a magnetic stirrer, and while stirring, pipet a measured volume onto the seated glass-fiber filter. Wash with three successive 10-mL volumes of reagent-grade water, allowing complete drainage between washings, and continue suction for about 3 min after filtration is complete. Sample with high dissolve solids may require additional washings. Carefully remove filter from

filtration apparatus and transfer to an aluminum-weighing dish as a support. Alternatively, remove the crucible and filter combination from the crucible adapter if a Gooch crucible is used. Dry for at least 1 h at 103 to 105°C in an oven, cool in a desiccator to balance temperature, and weigh. Repeat the cycle of drying, cooling, desiccating, and weighing until a constant weight is obtained or until the weight change is less than 4% of the previous weight or 0.5 mg, whichever is less. Duplicate determinations should agree within 5% of their average.

Calculation:

Total suspended solids mg /L= (A-B) × 1000 /sample volume, ml.

Where:

A = weight of filter + dried residue, mg, and

B = weight of the filter, mg.

Fixed and Volatile Solids Ignited At 550°C

Principle: The residue from TS, TDS or TSS is ignited to constant weight at 550°C. The remaining solids represent the fixed total, dissolved, or suspended solids while the weight lost on ignition is the volatile solids.

Apparatus:

- TDS and TSS determination apparatus is required and in addition
- muffle furnace at temperature of 550°C.

Procedure:

Ignite residue produced by TS, TDS or TSS to constant weight in a muffle furnace at a temperature of 550°C. Have furnace up to temperature before inserting sample. Usually, 15 to 20 min ignition is required for 200 mg residue. However, more than one sample and/or heavier residues may overtax the furnace and necessitate longer ignition times. Let dish or filter disk cool partially in air until most of the heat has been dissipated. Transfer to a desiccator for final cooling in a dry atmosphere. Do not over load desiccator. Weight dish or disk as soon as it has cooled to balance temperature. Repeat cycle of igniting, cooling, desiccating, and weighing until a constant weight is obtained or until weight change is less than 4% or 0.5 mg, whichever is less. Duplicate determinations should agree within 5% of their average.

Calculation:

Volatile solids mg /L = (A-B) × 1000 /sample volume, ml.

Fixed solids mg /L = (A-B) × 1000 / sample volume, ml.

Where:

A = weight of residue + dish before ignition, mg.

B = weight of residue + dish or filter after ignition, mg, and

C = weight of dish or filter, mg.

Settleable Solids

1. General Discussion

Settleable solids in surface and saline waters as well as domestic and industrial wastes may be determined and reported on either a volume (mL/L) or a weight (mg/L) basis.

2. Apparatus

The volumetric test requires only an Imhoff cone.

3. Procedure

a. Volumetric: Fill an Imhoff cone to the 1-L mark with a well-mixed sample. Settle for 45 min, gently agitate sample near the sides of the cone with a rod or by spinning, settle 15 min longer, and record volume of settleable solids in the cone as milliliters per liter. If the settled matter contains pockets of liquid between large settled particles, estimate volume of these and subtract from volume of settled solids. The practical lower limit of measurement depends on sample composition and generally is in the range of 0.1 to 1.0 mL/L. Where a separation of settleable and floating materials occurs, do not estimate the floating material as settleable matter. Replicates usually are not required.

Where biological or chemical floc is present, the gravimetric method is preferred.

b. Gravimetric:

1) Determine total suspended solids.

2) Pour a well-mixed sample into a glass vessel of not less than 9 cm diam using not less than 1 L and sufficient sample to give a depth of 20 cm. Alternatively use a glass vessel of greater diameter and a larger volume of sample. Let stand quiescent for 1 h and, without disturbing the settled or floating material, siphon 250 mL from center of container at a point halfway between the surface of the settled material and the liquid surface. Determine total suspended solids (milligrams per liter) of this supernatant liquor. These are the non-settleable solids.

4. Calculation

mg settleable solids/L = mg total suspended solids/L - mg non-settleable solids/L

Tests on Sludge

Settled Sludge Volume

1. General Discussion

The settled sludge volume of a biological suspension is useful in routine monitoring of biological processes. For activated sludge plant control, a 30-min settled sludge volume or the ratio of the 15-min to the 30-min settled sludge volume has been used to determine the returned-sludge flow rate and when to waste sludge. The 30-min settled sludge volume also is used to determine sludge volume index1.

This method is inappropriate for dilute sludges because of the small volume of settled material. In such cases, use the volumetric test for settleable solids using an Imhoff cone. Results from volumetric test for settleable solids are not comparable with those obtained with the procedure herein.

2. Apparatus

a. Settling column: Use 1-L graduated cylinder equipped with a stirring mechanism consisting of one or more thin rods extending the length of the column and positioned within two rod diameters of the cylinder wall. Provide a stirrer able to rotate the stirring rods at no greater than 4 rpm (peripheral tip speed of approximately 1.3 cm/s). See the following figure.

b. Stopwatch.

c. Thermometer.

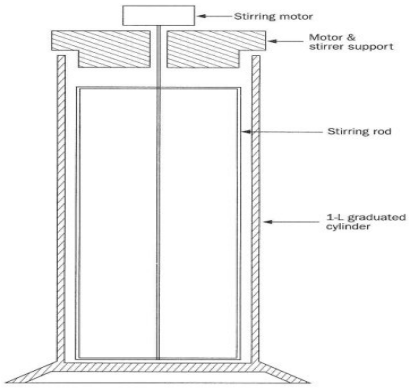


Figure 2710:1. Schematic diagram of settling vessel for settled sludge volume test.

3. Procedure

* Place 1.0 L sample in settling column and distribute solids by covering the top and inverting cylinder three times.
* Insert stirring rods, activate stirring mechanism, start the stop watch, and let suspension settle. Continue stirring throughout test.
* Maintain suspension temperature during test at that in the basin from which the sample was taken.

Determine volume occupied by suspension at measured time intervals, e.g., 5, 10, 15, 20, 30, 45, and 60 min.

* Report settled sludge volume of the suspension in milliliters for an indicated time interval.
* Variations in suspension temperature, sampling and agitation methods, dimensions of settling column, and time between sampling and start of the determination significantly affect results.

4. Precision and Bias

Bias is not applicable. The precision for this test has not been determined.

Sludge Volume Index

1. General Discussion

The sludge volume index (SVI) is the volume in milliliters occupied by 1 g of a suspension after 30 min settling. SVI typically is used to monitor settling characteristics of activated sludge and other biological suspensions. 1 Although SVI is not supported theoretically,2 experience has shown it to be useful in routine process control.

2. Procedure

Determine the suspended solids concentration of a well-mixed sample of the suspension.

Determine the 30 min settled sludge volume.

3. Calculations

$$SVI = \frac{\text{settled sludge volume (mL/L) X 1000}}{\text{suspended solids (mg/L)}}$$

4. Precision and Bias

Precision is determined by the precision achieved in the suspended solids measurement, the settling characteristics of the suspension, and variables associated with the measurement of the settled sludge volume. Bias is not applicable.

Chemical oxygen demand (COD)

Closed Reflux, Titrimetric method

Introduction:

The chemical oxygen demand (COD) is used as a measure of the oxygen equivalent of the organic matter content of a sample that is susceptible to oxidation by a strong chemical oxidant.

For samples from a specific source, COD can be related empirically to BOD. The test is useful for monitoring and control after correlation has been established.

The closed dichromate reflux method (chlorimetric method) is preferred over procedures using other oxidants because of superior oxidizing ability, applicability to a wide Varity of samples, and ease of manipulation.

Oxidation of most organic compounds is 95 to 100 % of the theoretical value.

Principle: Most types of organic matter are oxidized by a boiling mixture of chromic and sulfuric acids. A sample is refluxed in strongly acid solution vessel. after digestion in COD reactor at 160C° for 2hr,oxygen consumed is measured against standard at 620nm with a spectrophotometer.

Interference's and limitations:

* Volatile straight-chain aliphatic compounds are not oxidized to some extent. This failure occurs partly because volatile organics are present in the vapor space and do not come in contact with the oxidizing liquid.

* Chloride can be produce highly interference color that can be eliminated by adding silver sulfate.

Storage and sample preparation:

Homogenize samples containing solids to assure representative sample. The measurement should be done soon after the collection of the sample. Samples treated with sulfuric acid to a pH of less than 2 (at least 2ml per liter) and refrigerated at 4°C can be stored up to 28 days.

Apparatus:

- COD reactor.
- Spectrophotometer.
- Digestion vessel.
- Heating block.

Reagents and solution:

(1) Sulfuric acid reagent:

To 10 gm silver sulfate (Ag_2SO_4) + 35ml distilled water slowly add 965 ml conc. sulfuric acid (H_2SO_4). Let stand for one or two days to dissolve silver sulfate.

(2) Potassium dichromate reagent:

For 0-1500mg/lit COD concentration:

10gm potassium dichromate ($K_2Cr_2O_7$) + 100ml of the sulfuric acid reagent then increase the volume to one liter with conc. sulfuric acid.

(3) COD Standard Solution:

Take 1.7 gm of the dried KHP in distilled water and increase the volume to one liter. Then 1ml equal 2mg COD (or the concentration is 2000 mg COD/lit.). These solutions are stable up to three months, When refrigerated in the absence of visible biological growth.

Method of analysis:

2.8 ml of the dichromate reagent + 36 to 40mg mercuric sulfate ($HgSO_4$) salt, then add 2 ml. Sample, digest in COD reactor for two hours at temperature of 160 C°. Cool then measure at 620 nm for high range (0-1500mg/lit) samples.

Calculation:

COD as mg O_2/L = mg O_2 in final volume × d f.

BIOCHEMICAL OXYGEN DEMAND (BOD)

5-Day BOD Test

1. General Discussion

a. Principle: The method consists of filling with sample, to overflowing, an airtight bottle of the specified size and incubating it at the specified temperature for 5 d. Dissolved oxygen is measured initially and after incubation, and the BOD is computed from the difference between initial and final DO. Because the initial DO is determined shortly after the dilution is made, all oxygen uptake occurring after this measurement is included in the BOD measurement.

b. Sampling and storage: Samples for BOD analysis may degrade significantly during storage between collection and analysis, resulting in low BOD values. Minimize reduction of BOD by analyzing sample promptly or by cooling it to near-freezing temperature during storage. However, even at low temperature, keep holding time to a minimum. Warm chilled samples to 20 ± 3°C before analysis.

1) Grab samples – If analysis is begun within 2 h of collection, cold storage is unnecessary. If analysis is not started within 2 h of sample collection, keep sample at or below 4°C from the time of collection. Begin analysis within 6 h of collection; when this is not possible because the sampling site is distant from the laboratory, store at or below 4°C and report length and temperature of storage with the results. In no case start analysis more than 24 h after grab sample collection. When samples are to be used for regulatory purposes make every effort to deliver samples for analysis within 6 h of collection.

2) Composite samples – Keep samples at or below 4°C during compositing. Limit compositing period to 24 h. Use the same criteria as for storage of grab samples, starting the measurement of holding time from end of compositing period. State storage time and conditions as part of the results.

2. Apparatus

a. Incubation bottles: Use glass bottles having 60 mL or greater capacity (300-mL bottles having a ground-glass stopper and a flared mouth are preferred). Clean bottles with a detergent, rinse thoroughly, and drain before use. As a precaution against drawing air into the dilution bottle during incubation, use a water seal. Obtain satisfactory water seals by inverting bottles in a water bath or by adding water to the flared mouth of special BOD bottles. Place a paper or plastic cup or foil cap over flared mouth of bottle to reduce evaporation of the water seal during incubation.

b. Air incubator or water bath, thermostatically controlled at 20 ± 1°C. Exclude all light to prevent possibility of photosynthetic production of DO.

3. Reagents

Prepare reagents in advance but discard if there is any sign of precipitation or biological growth in the stock bottles. Commercial equivalents of these reagents are acceptable and different stock concentrations may be used if doses are adjusted proportionally.

a. Phosphate buffer solution: Dissolve 8.5 g KH_2PO_4, 21.75 g K_2HPO_4, 33.4 g $Na_2HPO_4 \cdot 7H_2O$, and 1.7 g NH_4Cl in about 500 mL distilled water and dilute to 1 L. The pH should be 7.2 without further adjustment. Alternatively, dissolve 42.5 g KH_2PO_4 or 54.3 g K_2HPO_4 in about 700 mL distilled water. Adjust pH to 7.2 with 30% NaOH and dilute to 1 L.

b. Magnesium sulfate solution: Dissolve 22.5 g $MgSO_4 \cdot 7H_2O$ in distilled water and dilute to 1 L.

c. Calcium chloride solution: Dissolve 27.5 g $CaCl_2$ in distilled water and dilute to 1 L.

d. Ferric chloride solution: Dissolve 0.25 g $FeCl_3 \cdot 6H_2O$ in distilled water and dilute to 1 L.

e. Acid and alkali solutions, 1N, for neutralization of caustic or acid waste samples.

1) Acid – Slowly while stirring, add 28 mL conc sulfuric acid to distilled water. Dilute to 1 L.

2) Alkali – Dissolve 40 g sodium hydroxide in distilled water. Dilute to 1 L.

f. Sodium sulfite solution: Dissolve 1.575 g Na_2SO_3 in 1000 mL distilled water. This solution is not stable; prepare daily.

g. Nitrification inhibitor, 2-chloro-6-(trichloromethyl) pyridine.*

h. Glucose-glutamic acid solution: Dry reagent-grade glucose and reagent-grade glutamic acid at 103°C for 1 h. Add 150 mg glucose and 150 mg glutamic acid to distilled water and dilute to 1 L. Prepare fresh immediately before use.

i. Ammonium chloride solution: Dissolve 1.15 g NH_4Cl in about 500 mL distilled water, adjust pH to 7.2 with NaOH solution, and dilute to 1 L. Solution contains 0.3 mg N/mL.

j. Dilution water: Use demineralized, distilled, tap, or natural water for making sample dilutions.

4. Procedure

a. Preparation of dilution water: Place desired volume of water in a suitable bottle and add 1 mL each of phosphate buffer, $MgSO_4$, $CaCl_2$, and $FeCl_3$ solutions/L of water. Seed dilution water, if desired, as described in 4d. Test dilution water as described in 4h so that water of assured quality always is on hand.

Before use bring dilution water temperature to 20 + 3°C. Saturate with DO by shaking in a partially filled bottle or by aerating with organic-free filtered air. Alternatively, store in cotton-plugged bottles long enough for water to become saturated with DO. Protect water quality by using clean glassware, tubing, and bottles.

b. Dilution water storage: Source water may be stored before use as long as the prepared dilution water meets quality control criteria in the dilution water blank. Such storage may improve the quality of some source waters but may allow biological growth to cause deterioration in others. Preferably do not store prepared dilution water for more than 24 h after adding nutrients, minerals, and buffer unless dilution water blanks consistently meet quality control limits. Discard stored source water if dilution water blank shows more than 0.2 mg/L DO depletion in 5 d.

c. Glucose-glutamic acid check: Because the BOD test is a bioassay its results can be influenced greatly by the presence of toxicants or by use of a poor seeding material. Distilled waters frequently are contaminated with copper; some sewage seeds are relatively inactive. Low results always are obtained with such seeds and waters. Periodically check dilution water quality, seed effectiveness, and analytical technique by making BOD measurements on a mixture of 150 mg glucose/L and 150 mg glutamic acid/L as a "standard" check solution. Glucose has an exceptionally high and variable oxidation rate but when it is used with glutamic acid, the oxidation rate is stabilized and is similar to that obtained with many municipal wastes. Alternatively, if a particular wastewater contains an identifiable major constituent that contributes to the BOD, use this compound in place of the glucose-glutamic acid.

Determine the 5-d 20°C BOD of a 2% dilution of the glucose-glutamic acid standard check solution using the techniques outlined in s 4d-j. Adjust concentrations of commercial mixtures to give 3 mg/L glucose and 3 mg/L glutamic acid in each GGA test bottle. Evaluate data as described in 6, Precision and Bias.

d. Seeding:

1) Seed source – It is necessary to have present a population of microorganisms capable of oxidizing the biodegradable organic matter in the sample. Domestic wastewater, unchlorinated or otherwise-undisinfected effluents from biological waste treatment plants, and surface waters receiving wastewater discharges contain satisfactory microbial populations. Some samples do not contain a sufficient microbial population (for example, some untreated industrial wastes, disinfected wastes, high-temperature wastes, or wastes with extreme pH values). For such wastes seed the dilution water or sample by adding a population of microorganisms. The preferred seed is effluent or mixed liquor from a biological treatment system processing the waste. Where such seed is not available, use supernatant from domestic wastewater after settling at room temperature for at least 1 h but no longer than 36 h. When effluent or mixed liquor from a biological treatment process is used, inhibition of nitrification is recommended.

Some samples may contain materials not degraded at normal rates by the microorganisms in settled domestic wastewater. Seed such samples with an adapted microbial population obtained from the undisinfected effluent or mixed liquor of a biological process treating the waste. In the absence of such a facility, obtain seed from the receiving water below (preferably 3 to 8 km) the point of discharge. When such seed sources also are not available, develop an adapted seed in the laboratory by continuously aerating a sample of settled domestic wastewater and adding small daily increments of waste. Optionally use a soil suspension or activated sludge, or a commercial seed preparation to obtain the initial microbial population. Determine the existence of a satisfactory population by testing the performance of the seed in BOD tests on the sample. BOD values that increase with time of adaptation to a steady high value indicate successful seed adaptation.

2) Seed control – Determine BOD of the seeding material as for any other sample. This is the seed control. From the value of the seed control and a knowledge of the seeding material dilution (in the dilution water) determine seed DO uptake. Ideally, make dilutions of seed such that the largest quantity results in at least 50% DO depletion. A plot of DO depletion, in milligrams per liter, versus milliliters of seed for all bottles having a 2-mg/L depletion and a 1.0-mg/L minimum residual DO should present a straight line for which the slope indicates DO depletion per milliliter of seed. The DO-axis intercept is oxygen depletion caused by the dilution water and should be less than 0.1 mg/L (4h). Alternatively, divide DO depletion by volume of seed in milliliters for each seed control bottle having a 2-mg/L depletion and a 1.0-mg/L residual DO. Average the results for all bottles meeting minimum depletion and residual DO criteria. The DO uptake attributable to the seed added to each bottle should be between 0.6 and 1.0 mg/L, but the amount of seed added should be adjusted from this range to that required to provide glucose-glutamic acid check results in the range of 198 ± 30.5 mg/L. To determine DO uptake for a test bottle, subtract DO uptake attributable to the seed from total DO update.

e. Sample pretreatment: Check pH of all samples before testing unless previous experience indicates that pH is within the acceptable range.

1) Samples containing caustic alkalinity (pH > 8.5) or acidity (pH < 6.0) - Neutralize samples to pH 6.5 to 7.5 with a solution of sulfuric acid (H_2SO_4) or sodium hydroxide (NaOH) of such strength that the quantity of reagent does not dilute the sample by more than 0.5%. The pH of dilution water should not be affected by the lowest sample dilution. Always seed samples that have been pH-adjusted.

2) Samples containing residual chlorine compounds - If possible, avoid samples containing residual chlorine by sampling ahead of chlorination processes. If the sample has been chlorinated but no detectable chlorine residual is present, seed the dilution water. If residual chlorine is present, dechlorinate sample and

seed the dilution water (4f). Do not test chlorinated/dechlorinated samples without seeding the dilution water. In some samples chlorine will dissipate within 1 to 2 h of standing in the light. This often occurs during sample transport and handling. For samples in which chlorine residual does not dissipate in a reasonably short time, destroy chlorine residual by adding Na_2SO_3 solution. Determine required volume of Na_2SO_3 solution on a 100- to 1000-mL portion of neutralized sample by adding 10 mL of 1 + 1 acetic acid or 1 + 50 H_2SO_4, 10 mL potassium iodide (KI) solution (10 g/100 mL) per 1000 mL portion, and titrating with Na_2SO_3 solution to the starch-iodine end point for residual. Add to neutralized sample the relative volume of Na_2SO_3 solution determined by the above test, mix, and after 10 to 20 min check sample for residual chlorine. (Note: Excess Na_2SO_3 exerts an oxygen demand and reacts slowly with certain organic chloramine compounds that may be present in chlorinated samples.)

3) Samples containing other toxic substances - Certain industrial wastes, for example, plating wastes, contain toxic metals. Such samples often require special study and treatment.

4) Samples supersaturated with DO - Samples containing more than 9 mg DO/ L at 20°C may be encountered in cold waters or in water where photosynthesis occurs. To prevent loss of oxygen during incubation of such samples, reduce DO to saturation at 20°C by bringing sample to about 20°C in partially filled bottle while agitating by vigorous shaking or by aerating with clean, filtered compressed air.

5) Sample temperature adjustment - Bring samples to 20 ± 1°C before making dilutions.

6) Nitrification inhibition - If nitrification inhibition is desired add 3 mg 2-chloro-6-(trichloro methyl) pyridine (TCMP) to each 300-mL bottle before capping or add sufficient amounts to the dilution water to make a final concentration of 10 mg/L. (Note: Pure TCMP may dissolve slowly and can float on top of the sample. Some commercial formulations dissolve more readily but are not 100% TCMP; adjust dosage accordingly.) Samples that may require nitrification inhibition include, but are not limited to, biologically treated effluents, samples seeded with biologically treated effluents, and river waters. Note the use of nitrogen inhibition in reporting results.

f. Dilution technique: Make several dilutions of sample that will result in a residual DO of at least 1 mg/L and a DO uptake of at least 2 mg/L after a 5-d incubation. Five dilutions are recommended unless experience with a particular sample shows that use of a smaller number of dilutions produces at least two bottles giving acceptable minimum DO depletion and residual limits. A more rapid analysis, such as COD, may be correlated approximately with BOD and serve as a guide in selecting dilutions. In the absence of prior knowledge, use the following dilutions: 0.0 to 1.0% for strong industrial wastes, 1 to 5% for raw and settled wastewater, 5 to 25% for biologically treated effluent, and 25 to100% for polluted river waters.

Prepare dilutions either in graduated cylinders or volumetric glassware, and then transfer to BOD bottles or prepare directly in BOD bottles. Either dilution method can be combined with any DO measurement technique. The number of bottles to be prepared for each dilution depends on the DO technique and the number of replicates desired.

When using graduated cylinders or volumetric flasks to prepare dilutions, and when seeding is necessary, add seed either directly to dilution water or to individual cylinders or flasks before dilution. Seeding of individual cylinders or flasks avoids a declining ratio of seed to sample as increasing dilutions are made. When dilutions are prepared directly in BOD bottles and when seeding is necessary, add seed directly to dilution water or directly to the BOD bottles. When a bottle contains more than 67% of the sample after dilution, nutrients may be limited in the diluted sample and subsequently reduce biological activity. In such samples, add the nutrient, mineral, and buffer solutions directly to individual BOD bottles at a rate of 1 mL/L (0.33 mL/ 300-mL bottle) or use commercially prepared solutions designed to dose the appropriate bottle size.

1) Dilutions prepared in graduated cylinders or volumetric flasks - If the azide modification of the titrimetric iodometric method is used, carefully siphon dilution water, seeded if necessary, into a 1- to 2-L-capacity flask or cylinder. Fill half full without entraining air. Add desired quantity of carefully mixed sample and dilute to appropriate level with dilution water. Mix well with a plunger-type mixing rod; avoid entraining air. Siphon mixed dilution into two BOD bottles. Determine initial DO on one of these bottles. Stopper the second bottle tightly, water-seal, and incubate for 5 d at 20°C. If the membrane electrode method is used for DO measurement, siphon dilution mixture into one BOD bottle. Determine initial DO on this bottle and replace any displaced contents with sample dilution to fill the bottle. Stopper tightly, water-seal, and incubate for 5 d at 20°C.

2) Dilutions prepared directly in BOD bottles - Using a wide-tip volumetric pipet, add the desired sample volume to individual BOD bottles of known capacity. Add appropriate amounts of seed material either to the individual BOD bottles or to the dilution water. Fill bottles with enough dilution water, seeded if necessary, so that insertion of stopper will displace all air, leaving no bubbles. For dilutions greater than 1:100 make a primary dilution in a graduated cylinder before making final dilution in the bottle. When using titrimetric iodometric methods for DO measurement, prepare two bottles at each dilution. Determine initial DO on one bottle. Stopper second bottle tightly, water-seal, and incubate for 5 d at 20°C. If the membrane electrode method is used for DO measurement, prepare only one BOD bottle for each dilution.

Determine initial DO on this bottle and replace any displaced contents with dilution water to fill the bottle. Stopper tightly, water-seal, and incubate for 5 d at 20°C. Rinse DO electrode between determinations to prevent cross-contamination of samples.

Use the azide modification of the iodometric method or the membrane electrode method to determine initial DO on all sample dilutions, dilution water blanks, and where appropriate, seed controls.

If the membrane electrode method is used, the azide modification of the iodometric method is recommended for calibrating the DO probe.

g. Determination of initial DO: If the sample contains materials that react rapidly with DO, determine initial DO immediately after filling BOD bottle with diluted sample. If rapid initial DO uptake is insignificant, the time period between preparing dilution and measuring initial DO is not critical but should not exceed 30 min.

h. Dilution water blank: Use a dilution water blank as a rough check on quality of unseeded dilution water and cleanliness of incubation bottles. Together with each batch of samples incubate a bottle of unseeded dilution water. Determine initial and final DO as in s 4g and j. The DO uptake should not be more than 0.2 mg/L and preferably not more than 0.1 mg/L Discard all dilution water having a DO uptake greater than 0.2 mg/L and either eliminate source of contamination or select an alternate dilution water source..

i. Incubation: Incubate at 20oC ± 1oC BOD bottles containing desired dilutions, seed controls, dilution water blanks, and glucose-glutamic acid checks. Water-seal bottles as described in 4f.

j. Determination of final DO: After 5 d incubation determine DO in sample dilutions, blanks, and checks as in 4g.

5. Calculation

For each test bottle meeting the 2.0-mg/L minimum DO depletion and the 1.0-mg/L residual DO, calculate BOD5 as follows:

When dilution water is not seeded:

$$\text{BOD5, mg/L} = \frac{D1 - D2}{P}$$

When dilution water is seeded:

$$\text{BOD5, mg/L} = \frac{(D1 - D2) - (B1 - B2)\,f}{P}$$

where:

D1 = DO of diluted sample immediately after preparation, mg/L,

D2 = DO of diluted sample after 5 d incubation at 20oC, mg/L,

P = decimal volumetric fraction of sample used,

B1 = DO of seed control before incubation, mg/L (4d),

B2 = DO of seed control after incubation mg/L (4d), and

f = ratio of seed in diluted sample to seed in seed control = (% seed in diluted sample)/(% seed in seed control).

If seed material is added directly to sample or to seed control bottles:

f = (volume of seed in diluted sample)/(volume of seed in seed control)

BIOCHEMICAL OXYGEN DEMAND (BOD)

(OxiTop Method)

Measuring principle:

(Manometric with pressure sensor) Measurement using OxiTop ® is based on pressure measurement in a closed system: microorganisms in the sample consume the oxygen and form CO_2. This is absorbed by NaOH, creating a vacuum which can be read directly as a measured value in mg/l BOD.

The used sample volume regulates the amount of oxygen available for a complete BOD. Measurement ranges of up to 4000 mg/l can be measured using different volumes.

The OxiTop ® heads green and yellow for differentiation of inflow/ outflow) have an AutoTemp function: if the sample temperature is too cold, the start of measurement is automatically delayed (by at least 1 hour) until a constant temperature has been reached.

Apart from the automatic storage of 5 measured values (1 value per day), further measured values can be read at all times during or after the period of 5 days, which permits the tracking of check values or measurements over longer periods.

Determination of biodegradability

The determination of the biodegradability should be checked before "new" chemicals are used for the first time, not only for environmental reasons but also to minimize disposal charges. The sample and a blank are stirred at a constant temperature for 28 days in closed bottles. The CO_2 Produced is removed from the gas space by means of an absorber so that the resulting negative pressure is a measure of the biodegradability. The continuous recording of the values in the OxiTop ® - C means that the required documentation can be guaranteed in an optimal manner. The Measuring bottles and adapters can be autoclaved at 249.8 ° F (121 ° C).

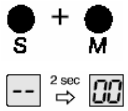
Measurement:

* To measure the sample volume overflow measuring beakers or measuring cylinders are used. Choose the volume according to the measured value expected.
* Put the magnetic stirring rod into the bottle.
* Insert a rubber quiver in the neck of the bottle.

* Put 2 sodium hydroxide tablets into the rubber quiver with a tweezers. (Caution: The Tablets must never come into the sample!)

* Screw OxiTop directly on sample bottle (tightly close).

Start measurement:
Press S and M simutaneously. (2 seconds) until the display shows 00.
Display: Stored Values are deleted.



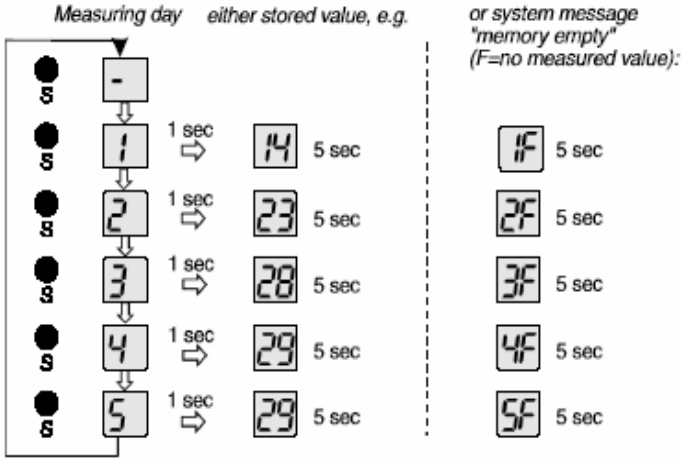
* Keep the measuring bottle with the OxiTop Put on for 5 days at 20°C.

* During the 5 days the sample is continuously stirred. The OxiTop Automatically stores one value every 24 hours for 5 days. To have the current values shown press the M key.

Display current measured value:
Press M until measured value is displayed (1 second).

* Readout Of the stored values after the 5 days have passed.

Recall stored values:
Press S until measured value is displayed (1 second).
Scroll to next day by repressing the S key while the measured value is displayed (5 sec). Fast scrolling by repeatedly pressing the S key.



Convert the displayed measured value (digits) into the BOD Value with the following table (Digits X Factor = BOD_5 in mg/l):

Sample volume (ml)	*Measuring range (mg/l)*	***Factor***
432	*0 - 40*	***1***
365	*0 - 80*	***2***
250	*0 - 200*	***5***
164	*0 - 400*	***10***
97	*0 - 800*	***20***
43.5	*0 - 2000*	***50***

Ammonia Determination

(Nesslerization Method)

Apparatus:

- Spectrophotometer.
- Nessler tubes.
- pH meter.

Reagents:

A - Zinc sulfate solution: Dissolve 100g $ZnSO_4.7H_2O$ and dilute to 1L with water.

B - Stabilizer reagent: Use Rochelle salt to prevent calcium or magnesium precipitation in undistilled samples after addition of alkaline Nessler reagent.

Rochelle salt solution: Dissolve 50g potassium sodium tartrate tetrahydrate, $KNaC_4H_4O_6.4H_2O$, in 100ml water. Remove ammonia usually present in the salt by boiling off 30ml of solution. After cooling , dilute to 100ml.

C - Nessler reagent: Dissolve 100g HgI_2 and 70g KI in a small quantity of water and add this mixture slowly, with stirring, to a cool solution of 160g NaOH dissolved in 500ml water. Dilute to 1L. Store in rubber-stoppered borosilicate glassware and out of sunlight to maintain reagent stability for up a year under normal laboratory conditions.

D - Stock ammonia solution: Dissolve 3.819g anhydrous NH_4Cl, dried at 100C°, in water, and dilute to 1000 ml, 1.00 ml=1.00 mg N= 1.22 mg NH_3.

E - Sodium hydroxide, 6N: Dissolve 240g NaOH in water and dilute to 1L.

Procedure:

a- Treatment of undistilled samples:

Add 0.4 to 0.5 ml 6N NaOH solution to obtain a pH of 10.5, as determined with a pH meter and a high -pH glass electrode, and mix gently. Let treated sample stand for a few minutes, whereupon a heavy flocculent precipitate should fall, leaving a clear and colorless supernate.

Clarify by centrifuging or filtering. Pretest any filter paper used to be sure no ammonia is present as a contaminant. Do this by running water through the filter and testing the filtrate by nesslerization.

b- Color development:

(For undistilled sample) use 50.0ml sample or a portion diluted to 50.0ml with water. If the undistilled portion contain sufficient concentrations of calcium, magnesium, or other ions that produce turbidity or precipitate with nessler reagent, add 1 to 2 drops (0.05 to 0.1 ml) Rochelle salt solution. Mix well. Add 1.0ml nessler reagent.

Mix samples by capping nessler tubes with clean rubber stoppers (washed thoroughly with water) and then inverting tubes at least six times. Keep such conditions as temperature and reaction time the same in blank, samples, and standards. Let reaction proceed for at least 10 min after adding nessler reagent. Measure color in the sample and standards. If NH_3-N is very low, use a 30-min contact time for sample, blank, and standards.

c- Photometric measurement:

Measure absorbance or transmittance with a spectrophotometer or filter photometer. Prepare calibration curve at the same temperature and reaction time used for samples. Measure transmittance readings against a reagent blank and run parallel checks frequently against standards in the nitrogen range of the samples.

Total Kjeldahl Nitrogen

Macro-Kjeldahl Method

1. Principle: In the presence of H_2SO_4, potassium sulfate (K_2SO_4), and cupric sulfate ($CuSO_4$) catalyst, amino nitrogen of many organic materials is converted to ammonium. Free ammonia also is converted to ammonium. After addition of base, the ammonia is distilled from an alkaline medium and absorbed in boric or sulfuric acid. The ammonia can be determined colorimetrically, by titration with a standard mineral acid.

2. Apparatus

a. Digestion apparatus: Kjeldahl flasks with a total capacity of 800 mL yield the best results.

b. Distillation apparatus

c. Apparatus for ammonia determination: Spectrophotometer for use at 640 nm with a light path of 1 cm or greater.

3. Reagents

Prepare all reagents and dilutions in ammonia-free water.

a. Digestion reagent: Dissolve 134 g K_2SO_4 and 7.3 g $CuSO_4$ in about 800 mL water. Carefully add 134 mL conc H_2SO_4. When it has cooled to room temperature, dilute the solution to 1 L with water. Mix well. Keep at a temperature close to 20°C to prevent crystallization.

b. Sodium hydroxide-sodium thiosulfate reagent: Dissolve 500 g NaOH and 25 g $Na_2S_2O_3 \cdot 5H_2O$ in water and dilute to 1 L.

c. Borate buffer solution: *Dissolve 40g boric acid in 600ml distilled water and complete to 900ml.

*Dissolve 0.1g bromocresol green in 100ml methanol.

*Dissolve 0.1g methyle red in 100ml methanol.

*Mix 900ml boric acid solution with 10ml bromocresol and 7ml methyle red solutions and

complete to 1liter by distilled water.

d. Sodium hydroxide, NaOH, 6N.

4. Procedure

a. Selection of sample volume and sample preparation: Place a measured volume of sample in an 800-mL kjeldahl flask. Select sample size from the following tabulation:

Organic Nitrogen in Sample (mg/L)	Sample Size (mL)
0-1	500
1-10	250
10-20	100
20-50	50
50-100	25

b. Ammonia removal: Add 25 mL borate buffer and then 6N NaOH until pH 9.5 is reached. Add a few glass beads or boiling chips and boil off 300 mL. If desired, distill this fraction and determine ammonia nitrogen. Alternately, if ammonia has been determined by the distillation method, use residue in distilling flask for organic nitrogen determination.

c. Digestion: Cool and add carefully 50 mL digestion reagent (or substitute 6.7 mL conc H_2SO_4, 6.7 g K_2SO_4, and 0.365 g $CuSO_4$) to distillation flask. Add a few glass beads and, after mixing, heat under a hood or with suitable ejection equipment to remove acid fumes. Boil briskly until the volume is greatly reduced (to about 25 to 50 mL) and copious white fumes are observed (fumes may be dark for samples high in organic matter). Then continue to digest for an additional 30 min. As digestion continues, colored or turbid samples will become transparent and pale green. After digestion, let cool, dilute to 300 mL with water, and mix. Tilt flask away from personnel and carefully add 50 mL sodium hydroxide-thiosulfate reagent to form an alkaline layer at flask bottom. Connect flask to a steamed-out distillation apparatus and swirl flask to insure complete mixing. The pH of the solution should exceed 11.0.

d. Distillation: Distill and collect 200 mL distillate. Use 50 mL indicating boric acid as absorbent solution when ammonia is to be determined by titration.

e. Final ammonia measurement: Titrate Using HCl (0.1 N) until green color disappear. Record HCl consumed amount and make calculation.

f. Standards: Carry a reagent blank and standards through all steps of the procedure.

5. Calculation

$$\text{TKN mg/l} = \frac{(T - B) \times N \times 14.007 \times 1000}{\text{Volume of sample (ml)}}$$

Total Phosphorus

Phosphorus analyses embody tow general procedural steps:

(a) Conversion of the phosphorus form of interest to dissolved orthophosphate. and (b)colorimetric determination of dissolved orthophosphate.

Storage:

Preserve by freezing at or below -10C°. Add 40mg $HgCL_2$/L to the sample especially when they are table stored for long periods. Do not add either acid or $CHCL_3$ as a preservative when phosphorus forms are to be determined. If total phosphorus alone is to determined add 1ml conc.HCl/L or freeze without any additions.

Sulfuric Acid-Nitric Acid Digestion:

Apparatus:

Digestion rack: An electrically or gas-heated digestion rack with provision for withdrawal of fumes is recommended. Digestion racks typical of those used for micro-Kjeldahl digesitions are suitable.

Micro-kjeldahl flasks.

Reagents:

1) sulfuric acid H_2SO_4 conc.

2) Nitric acid HNO_3 conc.

3) Phenolphthalein indicator aqueous solution .

4) Sodium hydroxide NaOH ,1N.

Procedure:

Into amicro-Kjeldahl flasks. Measure a sample containing the desired amount of phosphorus (this is determined by the colorimetric method used). Add 1ml conc.H_2SO_4 and 5ml conc.HNO_3.

Digest to a volume of 1ml and then continue until solution becomes colorless to remove HNO_3.

- Cool and approximately 20ml distilled water. 0.05ml(1drop) phenolphthalein indicator, and as much 1N NaOH solution as required to produce a faint pink tinge. Transfer neutralized solution filtering if necessary to remove particulate material or turbidity, into a 100-ml volumetric flask. Add filter washings to flask and adjust sample volume to 100ml with distilled water.

Determine phosphorus by ascorbic acid or stannous chloride methods.

Orthophosphate

Ascorbic Acid Method

1. General Discussion

a. Principle: Ammonium molybdate and potassium antimonyl tartrate react in acid medium with orthophosphate to form a heteropoly acid - phosphomolybdic acid - that is reduced to intensely colored molybdenum blue by ascorbic acid.

b. Interference: Arsenates react with the molybdate reagent to produce a blue color similar to that formed with phosphate. Concentrations as low as 0.1 mg As/L interfere with the phosphate determination. Hexavalent chromium and NO_2^- interfere to give results about 3% low at concentrations of 1 mg/L and 10 to 15% low at 10 mg/L. Sulfide (Na_2S) and silicate do not interfere at concentrations of 1.0 and 10 mg/L.

c. Minimum detectable concentration: Approximately 10 µg P/L.

2. Apparatus

a. Colorimetric equipment: One of the following is required:

1) Spectrophotometer, with infrared phototube for use at 880 nm, providing a light path of 2.5 cm or longer.

2) Filter photometer, equipped with a red color filter and a light path of 0.5 cm or longer.

b. Acid-washed glassware.

3. Reagents

a. Sulfuric acid, H_2SO_4, 5N: Dilute 70 mL conc H_2SO_4 to 500 mL with distilled water.

b. Potassium antimonyl tartrate solution: Dissolve 1.3715 g $K(SbO)C_4H_4O_6 \cdot 1/2H_2O$ in 400 mL distilled water in a 500-mL volumetric flask and dilute to volume. Store in a glass-stoppered bottle.

c. Ammonium molybdate solution: Dissolve 20 g $(NH_4)_6Mo_7O_{24} \cdot 4H_2O$ in 500 mL distilled water. Store in a glass-stoppered bottle.

d. Ascorbic acid, 0.1M: Dissolve 1.76 g ascorbic acid in 100 mL distilled water. The solution is stable for about 1 week at 4^oC.

e. Combined reagent: Mix the above reagents in the following proportions for 100 mL of the combined reagent: 50 mL 5N H_2SO_4, 5 mL potassium antimonyl tartrate solution, 15 mL ammonium

molybdate solution, and 30 mL ascorbic acid solution. Mix after addition of each reagent. Let all reagents reach room temperature before they are mixed and mix in the order given. If turbidity forms in the combined reagent, shake and let stand for a few minutes until turbidity disappears before proceeding. The reagent is stable for 4 h.

f. Stock phosphate solution.

g. Standard phosphate solution: Dilute 50.0 mL stock phosphate solution to 1000 mL with distilled water; 1.00 mL = 2.50 µg P.

4. Procedure

a. Treatment of sample: Pipet 50.0 mL sample into a clean, dry test tube or 125-mL erlenmeyer flask. Add 0.05 mL (1 drop) phenolphthalein indicator. If a red color develops add 5N H_2SO_4 solution dropwise to just discharge the color. Add 8.0 mL combined reagent and mix thoroughly. After at least 10 min but no more than 30 min, measure absorbance of each sample at 880 nm, using reagent blank as the reference solution.

b. Correction for turbidity or interfering color: Natural color of water generally does not interfere at the high wavelength used. For highly colored or turbid waters, prepare a blank by adding all reagents except ascorbic acid and potassium antimonyl tartrate to the sample. Subtract blank absorbance from absorbance of each sample.

c. Preparation of calibration curve: Prepare individual calibration curves from a series of six standards within the phosphate ranges indicated in 1c above. Use a distilled water blank with the combined reagent to make photometric readings for the calibration curve. Plot absorbance vs. phosphate concentration to give a straight line passing through the origin. Test at least one phosphate standard with each set of samples.

5. Calculation

$$\text{mg P/L} = \frac{\text{mg P (in approximately final volume) X 1000}}{\text{mL sample}}$$

Chloride

Principle: In a neutral or slightly alkaline solution, potassium chromate can indicate the end point of the silver nitrate titration of chloride. Silver chloride is precipitated quantitatively before red silver chromate is formed.

Apparatus:

- Erlenmeyer flask, 250ml.
- Burt, 50ml.

Reagents:

a- Potassium chromate indicator solution.

10g potassium chromate is dissolved and made up to 100ml with water.

b- Silver standard solution.

4.791g silver nitrate is dissolved and made up to 1L with water. The solution is stored in a brown glass bottle. 1ml corresponds to 1mg chloride.

c- Sodium chloride standard solution.

1.648g sodium chloride (dried for 2 hours at 105C°) is dissolved and made up to 1L with water.1ml of this solution contains 1mg chloride.

Procedure:

-10 ml of sample or a suitable portion diluted to 100ml is placed into an Erlenmeyer flask and 1ml potassium chromate solution added. The mixture is then titrated against a white back ground with silver nitrate solution until the color changes from greenish yellow to reddish brown. Blank sample with distilled water is treated in the same way as the sample.

Calculation of results:

Chloride concentration (mg/L) = (A - B) ×1000mg / C.

A= volume of $AgNO_3$ solution consumed (ml) for sample.

B= volume of $AgNO_3$ solution consumed (ml) for blank.

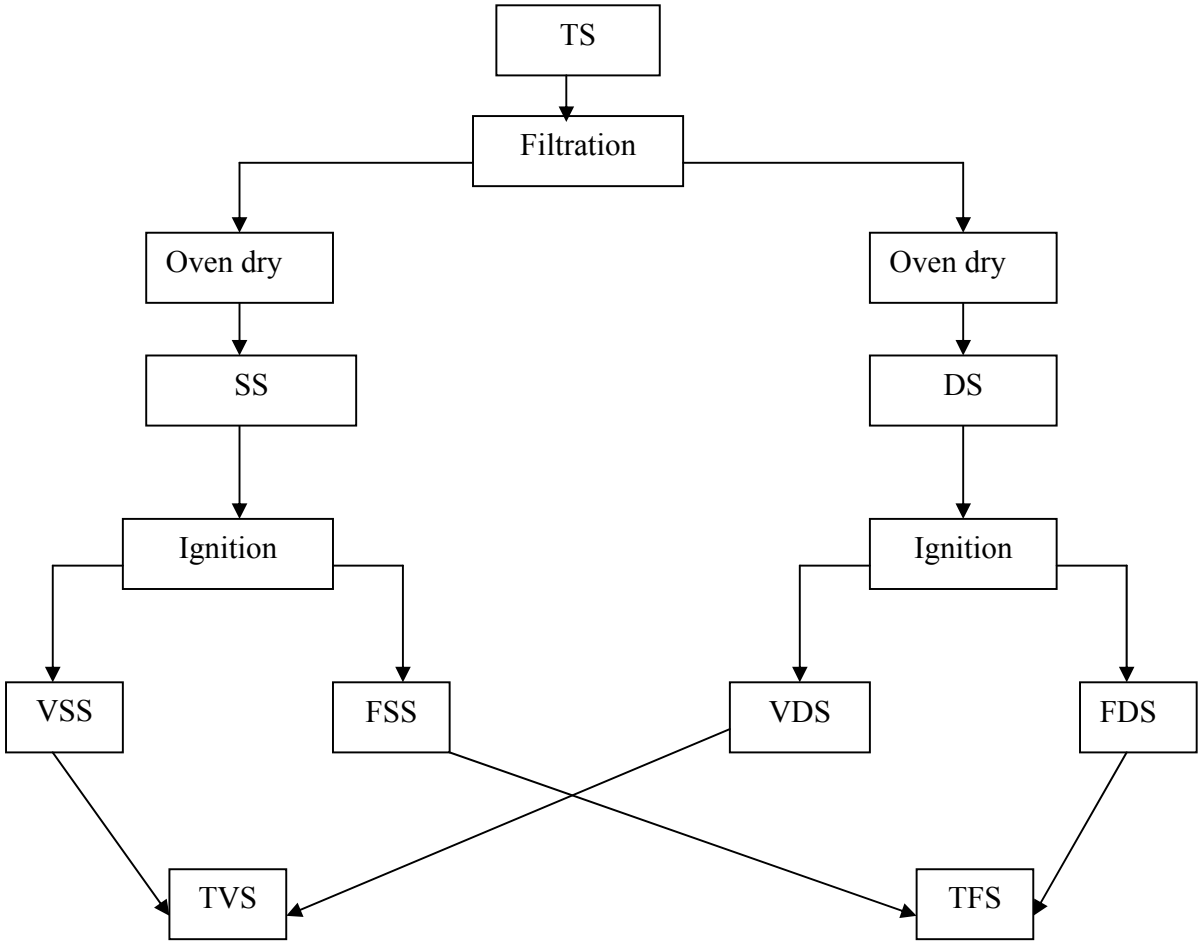
C= sample volume (ml).

Appendixes

Table shows characteristics of wastewater:

pollutant	Unit	Concentration		
		High	Medium	Low
Total Solids (TS)	mg/L	1200	720	350
Total Dissolved Solids(TDS)	mg/L	850	500	250
Fixed Solids (Non volatile)	mg/L	525	300	145
Volatile Solids	mg/L	325	200	105
Total Suspended Solids (TSS)	mg/L	350	220	100
Fixed Suspended Solids (FSS)	mg/L	75	55	20
Volatile Suspended Solids (FSS)	mg/L	275	165	80
Settleable Solids	mg/L	20	10	5
Biochemical Oxygen Demand (BOD_5)	mg/L	400	220	110
Total Organic Carbon (TOC)	mg/L	290	160	80
Chemical Oxygen Demand (COD)	mg/L	1000	500	250
Total Nitrogen (TN)	mg/L	85	40	20
Organic Nitrogen	mg/L	32	15	8
Nitrate (NH3 – N)	mg/L	50	25	12
Nitrite	mg/L	0	0	0
Total Phosphorous	mg/L	15	8	4
Organic Phosphorous	mg/L	5	3	1
Non-Organic Phosphorous	mg/L	10	5	3
Excess Chloride on Drinking water	mg/L	100	50	30
Excess Chloride on Drinking water	mg/L	50	30	20
Hardness (Ca CO3)	mg/L	200	100	50
Oil and Grease	mg/L	150	100	50
Volatile Organic Compounds (VOC)	mg/L	400 <	100-400	100 >
Total Count (MPN)	Count/100ml	10^8-10^9	10^7-10^8	10^6-10^7

Schematic diagram of Solids analysis:



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