

WATER BACTERIOLOGY

Training Course 2008

A. S. Altomi

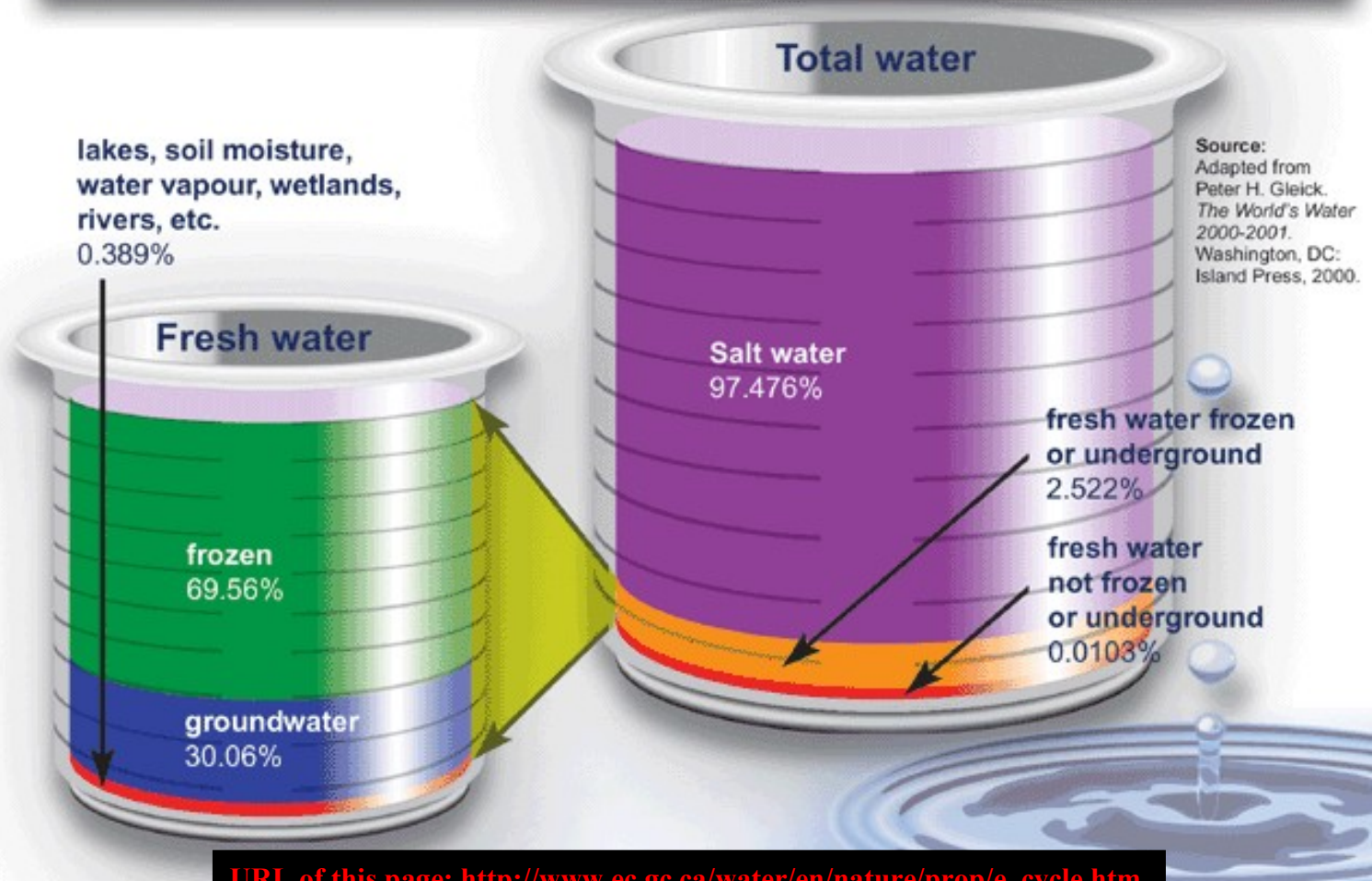


Please shut down your phone or
keep it on silent mode

?How much water is there in the world

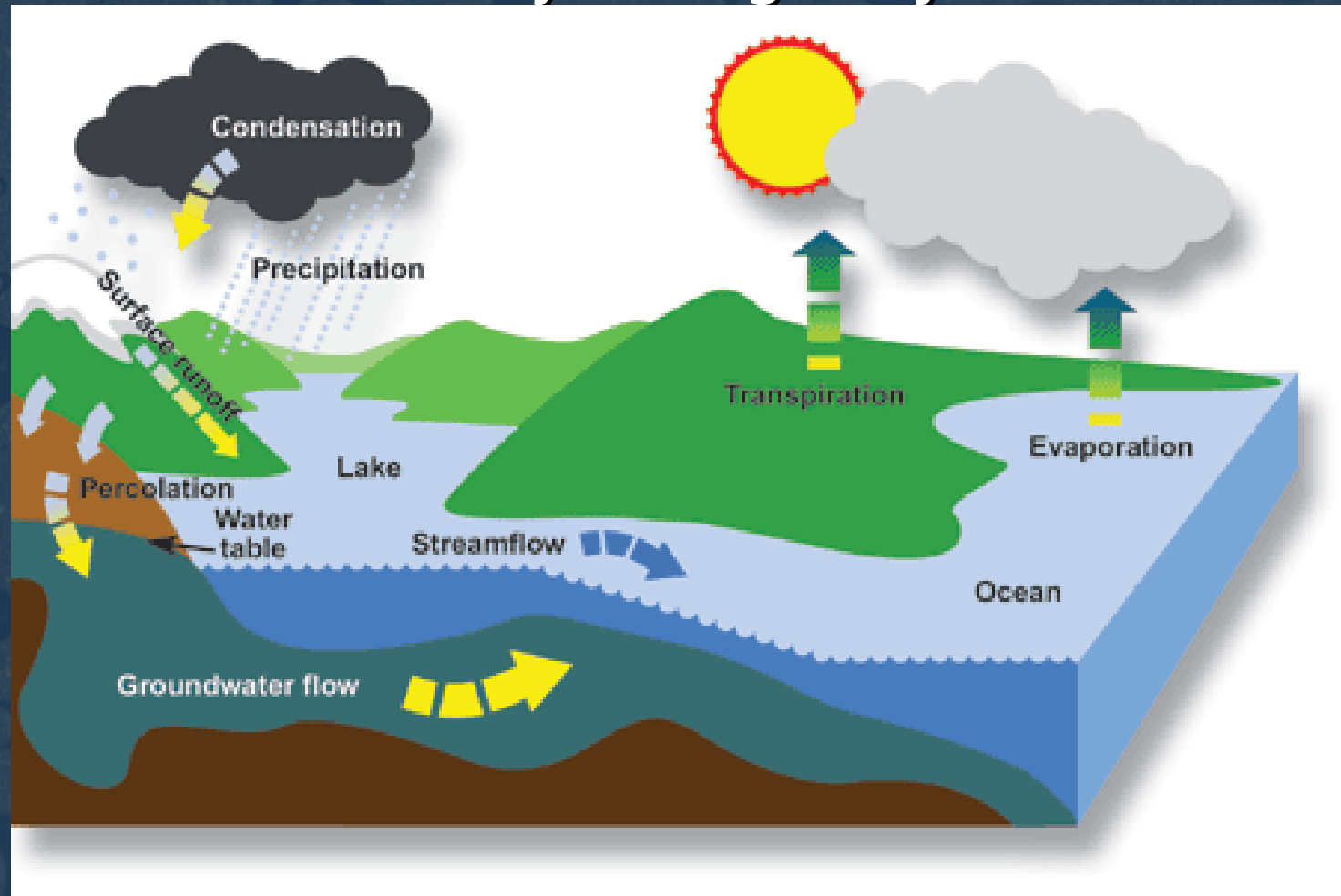
Scientists estimate that the quantity of water was **over one billion cubic kilometers** . And it covers nearly three quarters of the earth's surface in oceans as well as rivers, lakes, snow and glaciers. There is water in the atmosphere and water underground. Water evaporates and returns to the land surface in what is known as the **Hydrologic Cycle**.

The world's water supply



URL of this page: http://www.ec.gc.ca/water/en/nature/prop/e_cycle.htm

The Hydrologic Cycle



URL of this page: http://www.ec.gc.ca/water/en/nature/prop/e_cycle.htm

The Golden Rule

Access to water supply and Sanitation is a fundamental need and a human right. It is vital for dignity and health of all people.

- In September 2000, 189 UN Member States adopted the Millennium Development Goals (MDGs), Achieving these targets will directly affect the lives and future prospects of billions of people around the globe.

Millennium Development Goals ((MDG))

Goal 5: Develop an MDG 5 Partnership for Women's Development.

Goal 7: Ensure Environmental Sustainability.

Adequate treatment and disposal of wastewater contributes to better ecosystem conservation and less pressure on scarce freshwater resources. Careful use of water resources prevents contamination of groundwater and helps minimize the cost of water treatment.

- by 2015, the proportion of people without sustainable access to safe drinking water and basic sanitation



General Introduction

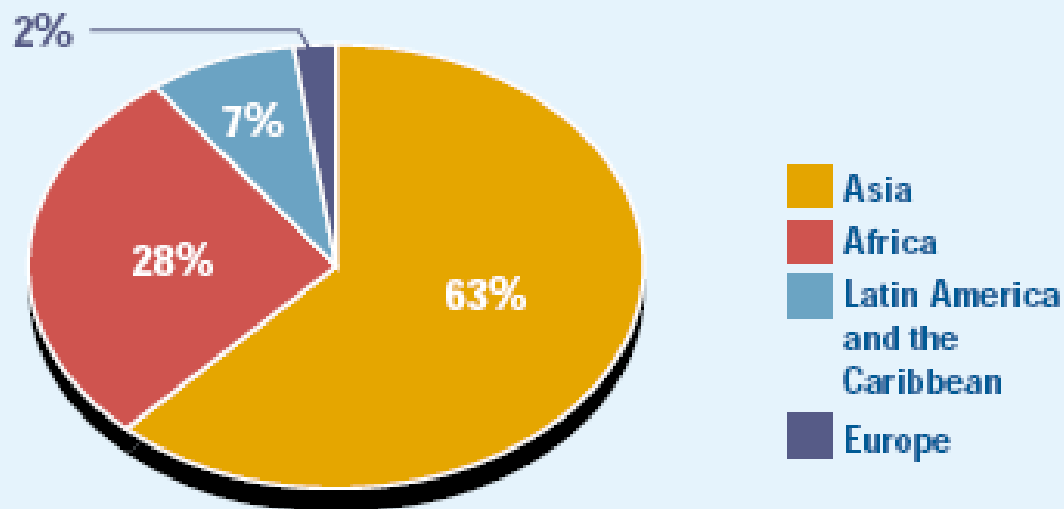
- People served with some form of improved water supply rose from 79% (4.1billion) in 1990 to 825 (4.9billion) in 2000.

- At the same time, the proportion of the world's population with access to excretal disposal facilities increase from 55% (2.9billion) to 60% (3.6 billion people served).

1

- At the beginning of 2000, $\frac{1}{6}$ (1.1 billion people) was without access to improved water supply.

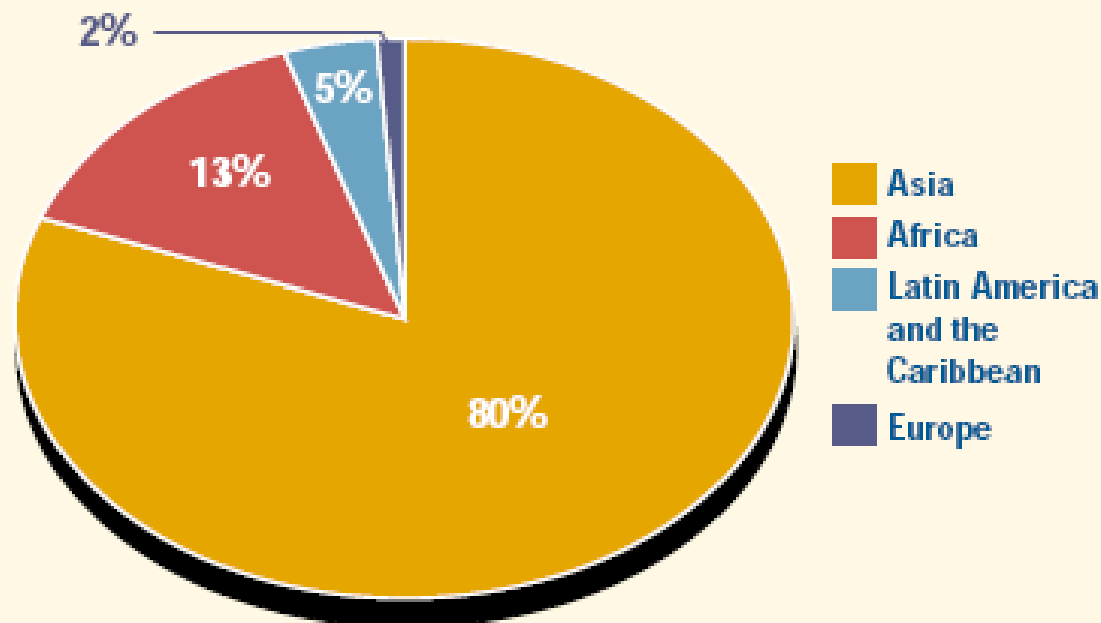
Figure 2.1 Distribution of the global population not served with improved water supply, by region



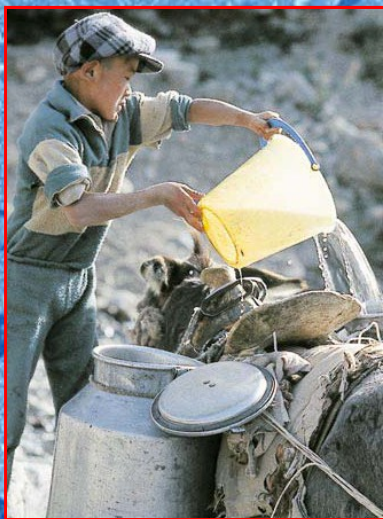
Total unserved: 1.1 billion

- And $\frac{2}{5}$ (2.4 billion people) lacked access to improved sanitation.

Figure 2.2 Distribution of the global population not served with improved sanitation, by region



Total unserved: 2.4 billion



The majority live in Asia and Africa :

- * Fewer than half of all Asians have access to improved Sanitation.

- * 2 out of 5 Africans lack improved water supply.

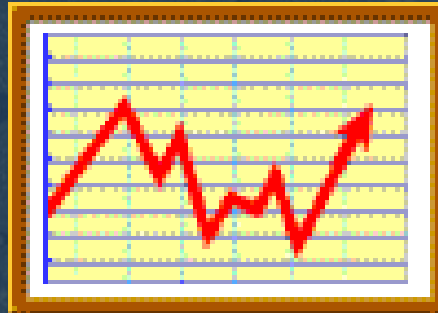
Moreover, Rural services still lag far behind Urban services. 80% of those lacking adequate sanitation (2 billion people) live in Rural areas (1.3 in China and India).

Global Water Supply and Sanitation Assessment 2000 Report

TABLE 6.1 AFRICA: WATER SUPPLY AND SANITATION COVERAGE BY COUNTRY, AREA OR TERRITORY, 1990 AND 2000

		Total population ¹ (thousands)	Urban population (thousands)	Rural population (thousands)	% urban water supply coverage	% rural water supply coverage	% total water supply coverage	% urban sanitation coverage	% rural sanitation coverage	% total sanitation coverage
Libyan Arab Jamahiriya	1990	4 416	3 614	802	72	68	71	97	96	97
	2000	5 604	4 911	693	72	68	72	97	96	97

1990 to 2000 :



- 816 million additional people gaining access to water supply .
- 747 million additional people gaining access to sanitation facilities.

The Water supply and Sanitation sector will face challenges over the coming decades. As the No. of populations in Africa, Asia, Latin America and The Caribbean are expected to increase dramatically :

* The African urban population is expected to more than double over the next 25 years.

* The Asian will almost double.

* Latin Americans and the Caribbeans is expected to Increase by almost 50% over the same period.

So, to achieve the 2015 target in these areas, an additional 2.2 billion people will need access to Sanitation and 1.5 billion will need access to Water supply by that date.

This means providing Water supply services to 280,000 people and Sanitation facilities to 384,000 people every day for the next 15 years.

Health Hazards of Poor Water Supply and Sanitation

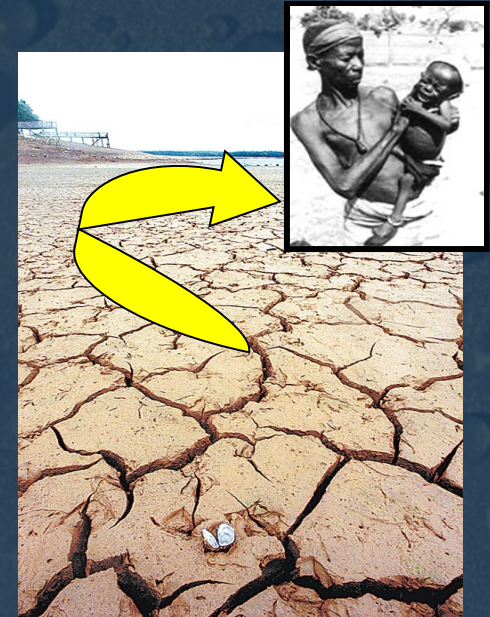
The WHO has estimated that up to 80% of all sickness and disease in the world is caused by inadequate sanitation, polluted water, or unavailability of water.



Cholera



Trachoma



Drought

Diseases that may be associated with contaminated drinking water

	Organism	Disease Caused
1	<i>Bacteria</i>	
	<i>Escherichia coli</i> (some types)	Gastroenteritis
	<i>.Leptospira spp</i>	Leptospirosis
	<i>Salmonella typhi</i>	Typhoid fever
	<i>.Salmonella spp</i>	Salmonellosis
	<i>.Shigella spp</i>	(Shigellosis (bacillary dysentery
	<i>Vibrio cholerae</i>	Cholera
2	<i>Protozoa</i>	
	<i>Balantidium coli</i>	Balantidiasis
	<i>Cryptosporidium parvum</i>	Cryptosporidiosis
	<i>Entamoeba histolytica</i>	(Amebiasis (amoebic dysentery
	<i>Giardia lamblia</i>	Giardiasis

3	Helminths	
	<i>Ascaris lumbricoides</i>	Ascariasis
	<i>T. solium</i>	Taeniasis
	<i>Trichuris trichiura</i>	Trichuriasis
4	Viruses	
	„Enteroviruses (72 types) e.g	Gastroenteritis, hear
	polio echo and coxsackie) (viruses	anomalies, meningitis
	Hepatitis A virus	Infectious hepatitis
	Norwalk agent	Gastroenteritis
	Rotavirus	Gastroenteritis

* Approximately 4 billion cases of Diarrhea each year, mostly among children under the age of Five.

This is equivalent to :

- one child dying every 15 seconds
- 20 jumbo jets crashed every day.

These deaths represent approximately 15% of all child deaths under the age of five in developing countries.

Water, sanitation, and hygiene interventions reduce diarrhoeal disease on average by between one-quarter and one-third

* Intestinal worms infect about 10% of the population of the developing world, Intestinal parasitic infections can lead to malnutrition, anemia and retarded growth, depending upon the severity of the infection.

These can be controlled through better sanitation, hygiene and water supply

* It is estimated that 6 million people are blind from trachoma and the population at risk from this disease is approximately 500 million.

Providing adequate quantities of water reduced the median infection rate by 25%.

* 200 million people in the world are infected with schistosomiasis

a median 77% reduction from well-designed water and sanitation interventions.

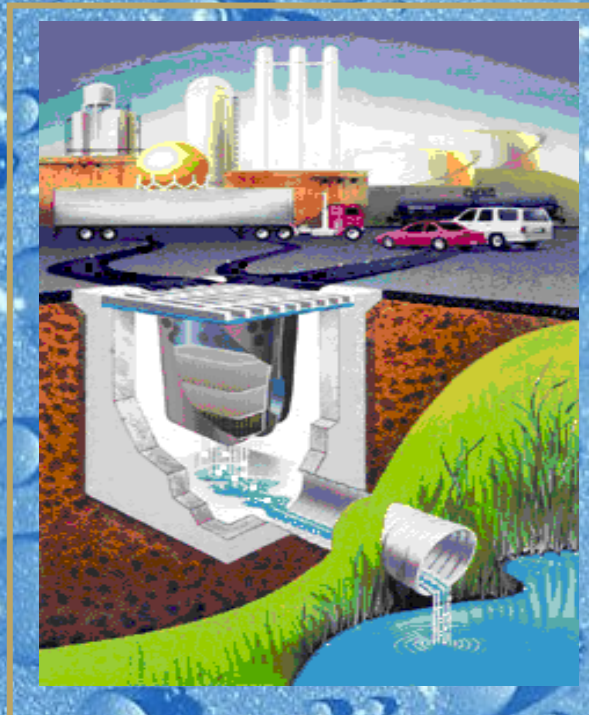
Potable Water

((Should be suitable for human consumption and for all usual domestic purposes including personal hygiene, Washing, Showering and Food preparation)) .



**: Good quality water is
Odorless, Colorless,
Tasteless**

**Free from faecal pollution and chemicals in
harmful amounts.**



General Introduction on Microbiology

Definition – study of living organisms simple in structure and small in size

Include: bacteria, algae, fungi, protozoa, viruses

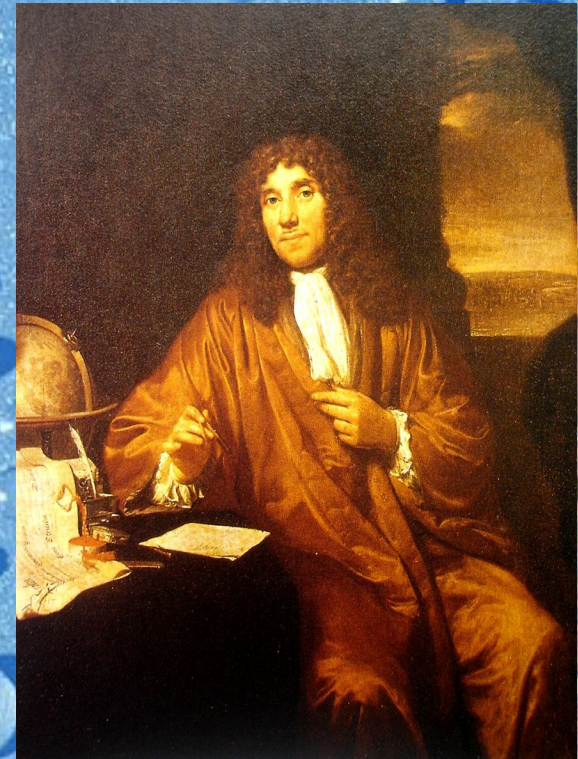
What are microbes?

Microbe is a term for tiny creatures that individually are too small to be seen with the unaided eye. Microbes include bacteria, fungi, parasites, algae and viruses.

Antonie van Leeuwenhoek (1632 – 1723)

Antonie van Leeuwenhoek
(1632 – 1723) simple
microscope (200 x
magnification), „Father of
Microbiology“

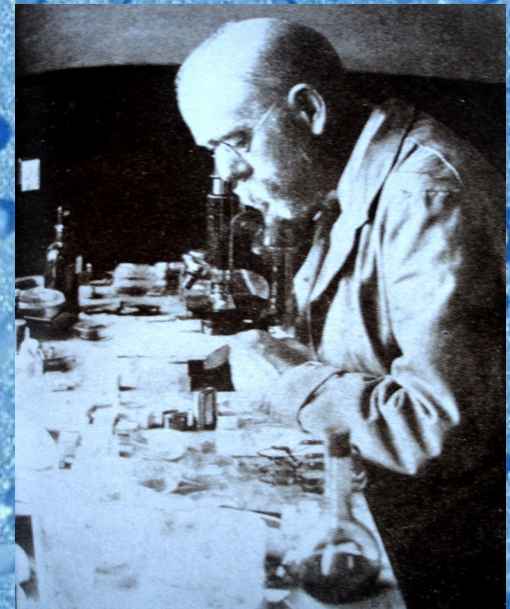
- Descriptions of simple microorganisms („animalcula“)
- bacteria, protozoa, yeasts, erythrocytes, sperms.



Robert Koch (1843-1910)

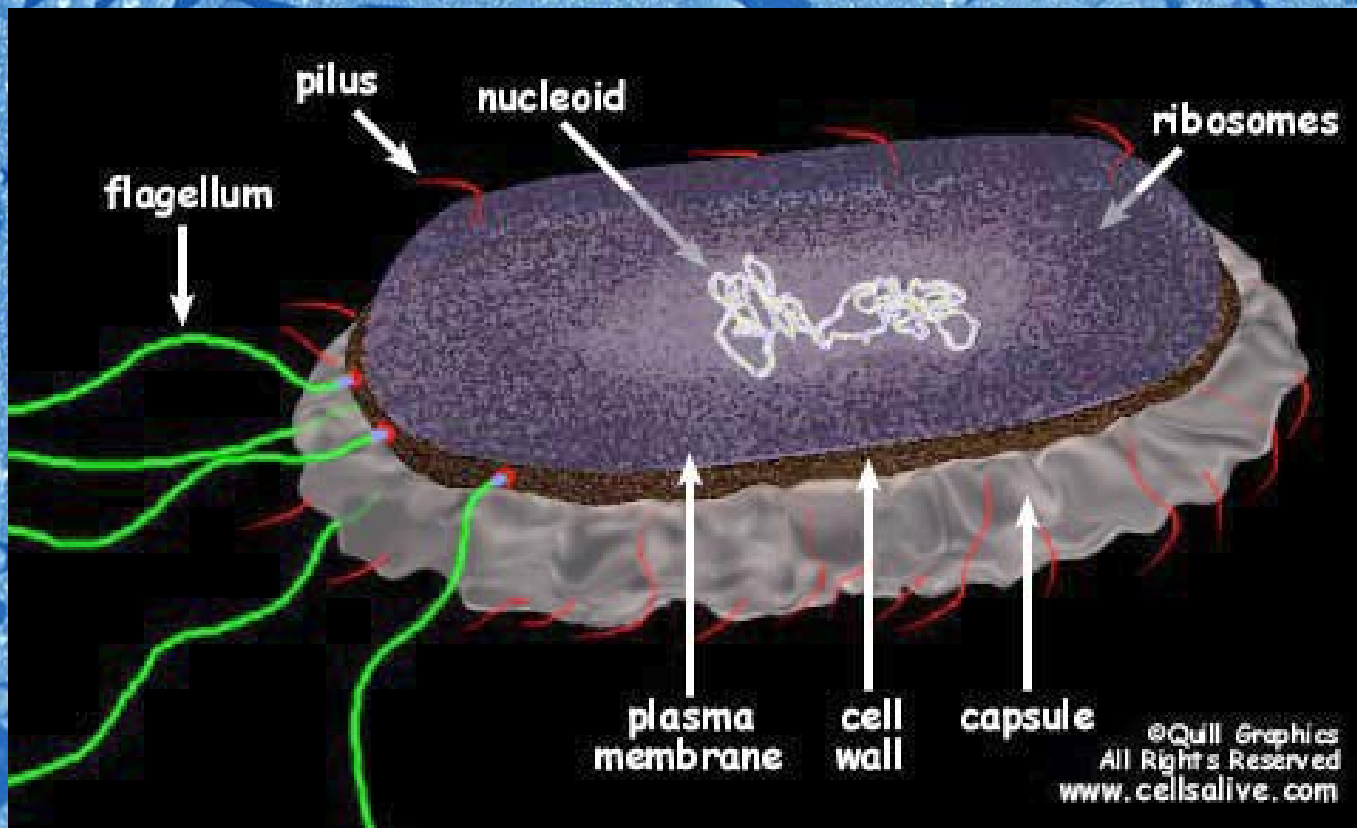
1876 – Koch's postulates identifying the causativity of bacteria and disease:

1. bacteria must be present in all cases of illness
2. must be isolated in a pure culture
3. Application of the pure culture to the experimental animal must induce the illness with characteristic symptoms
4. The same bacteria from infected and ill animal can be again isolated



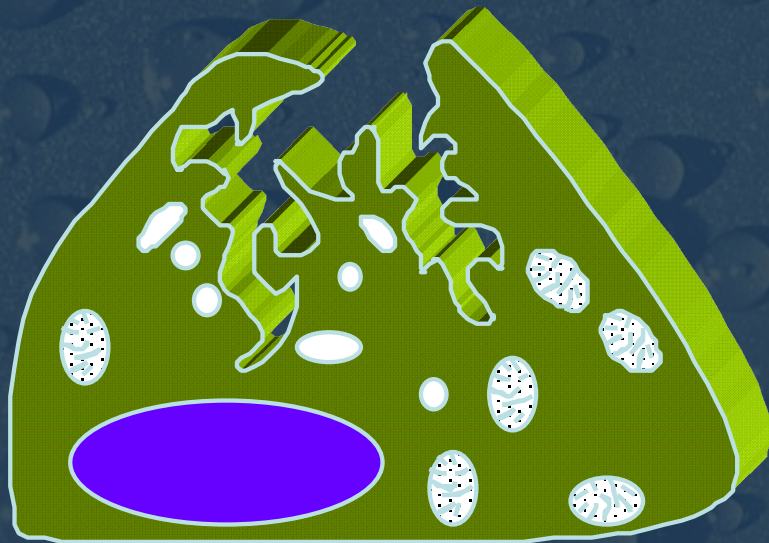
Classification

Structure of Bacterial cell



Size matters

Animal cell



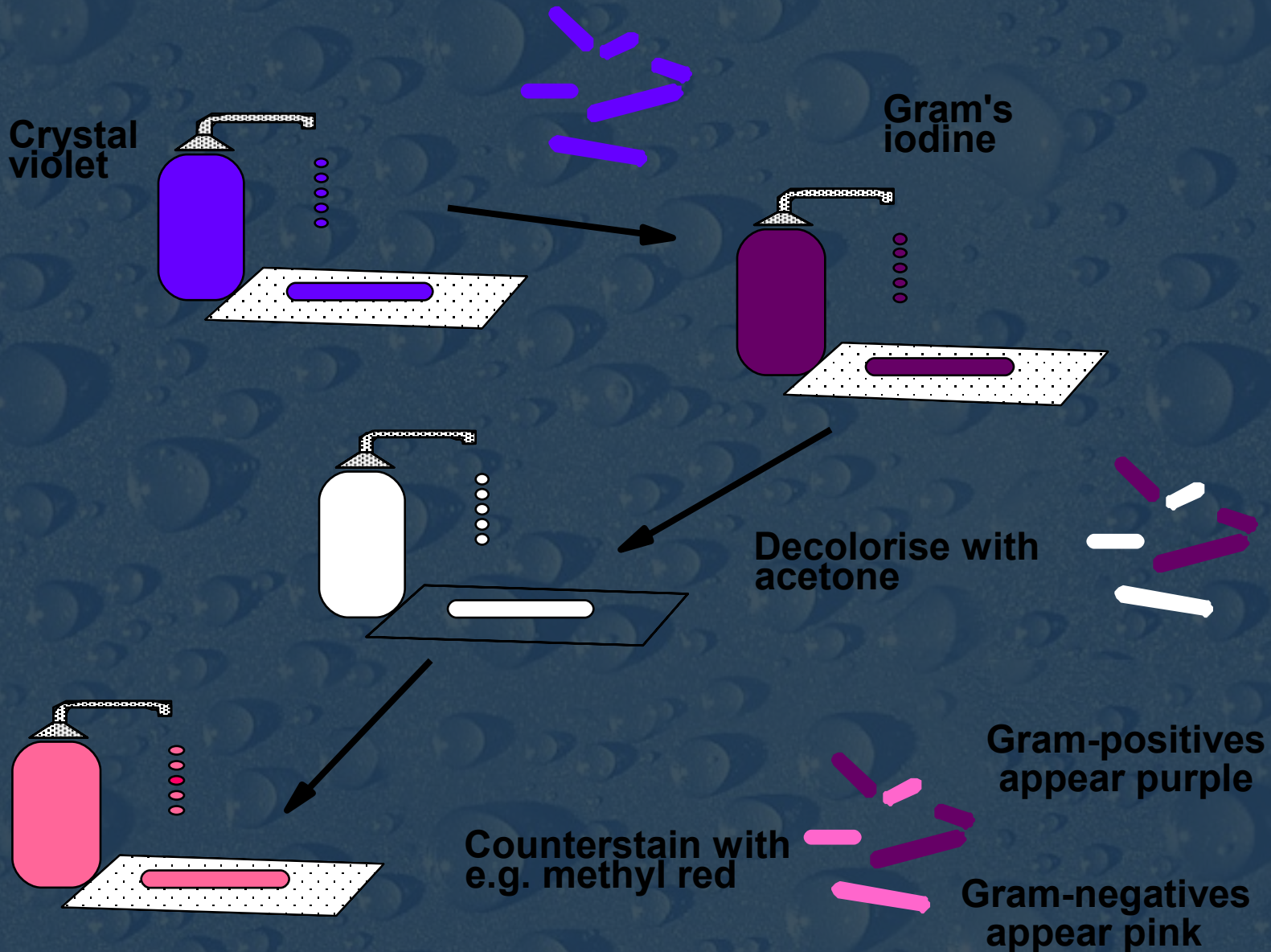
10 microns

Bacterial cells

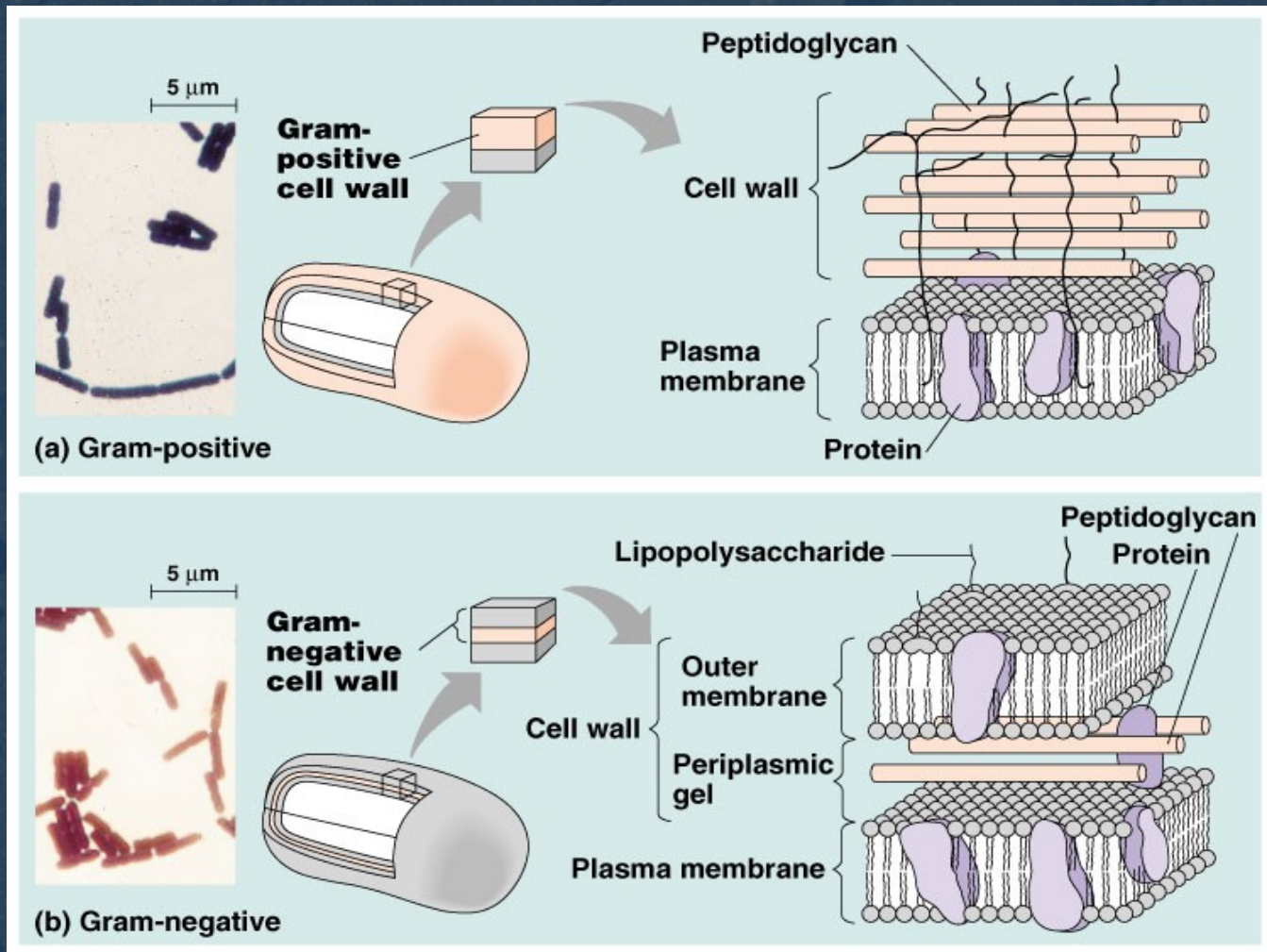


1 micron

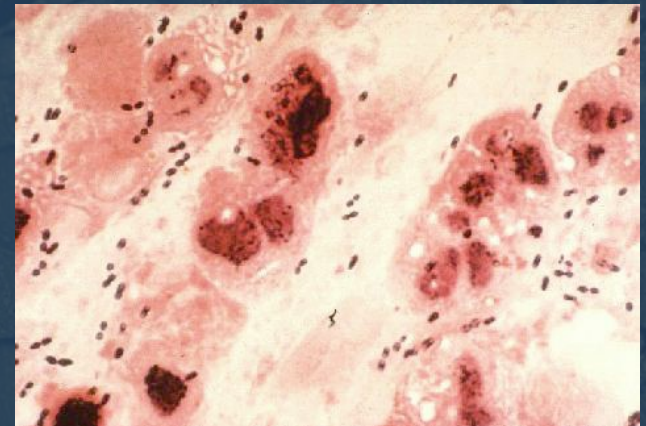
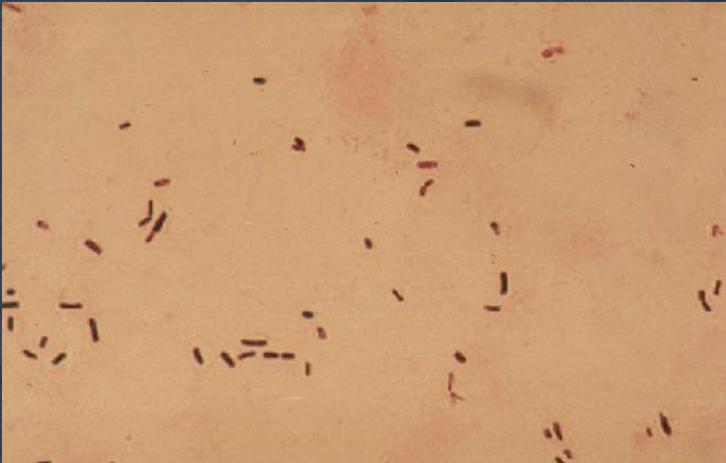
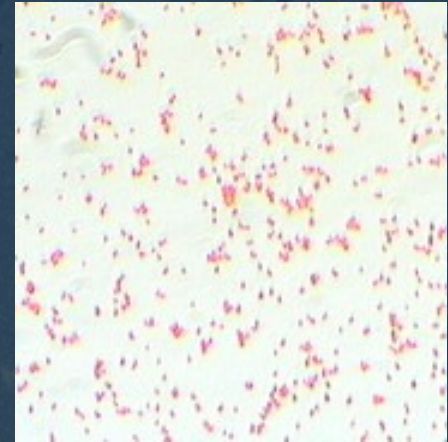
The Gram Stain



Cell Wall Structure

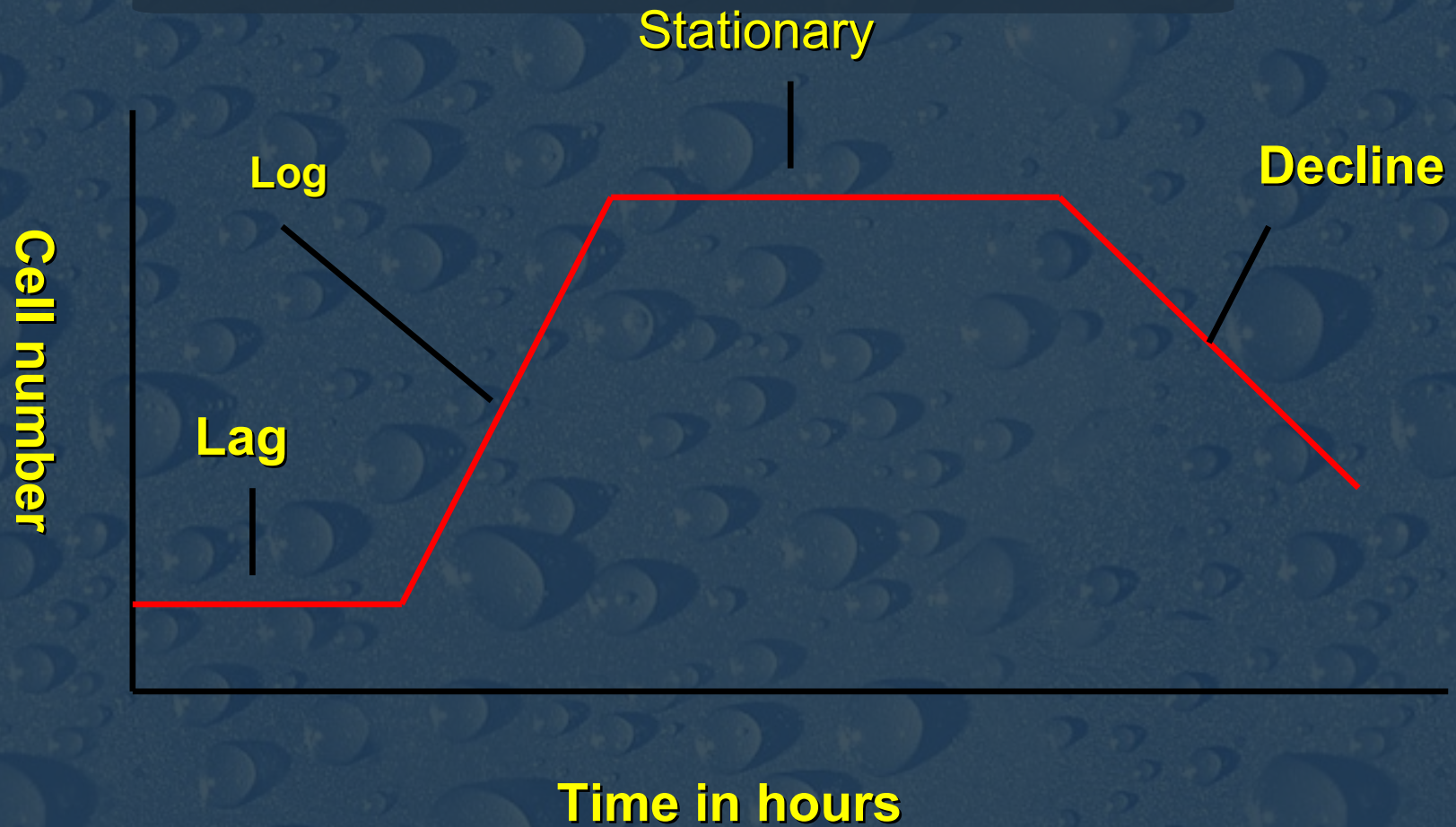


Gram Staining Reaction



Bacterial Growth Curve

A growth curve (divided into 4 stages) :



:Lag phase -1



- . Period from inoculation to beginning multiplication
 - . No or little cell division occurs
 - . Bacteria adapt to the new environment
 - . Clinically corresponds to incubation period of disease

:Logarithmic (Exponential) phase -2

- . Rapid cell division (most active phase).
- . Number of bacteria increase steadily.
- . Clinically corresponds to clinical signs & symptoms of disease.
- . This phase continues until:
 - . Exhaustion of nutrients and/or accumulation of toxic waste products.

Stationary -3

Phase



- . Number of dying cells equals newly formed cells.
- . Number of living bacteria remains constant.
- . Total number of bacteria (living + dead) increases.
- . Slow growth due to Nutrient depletion , waste product accumulation or pH change.
- . Clinically corresponds to recovery stage of disease.

Decline -4

Phase

- . Number of living bacteria decreases steadily.
- . Death rate exceeds multiplication rate.
- . Exhaustion of nutrients and accumulation toxic products.

Clinical Significance of Growth curve

Correlation of 4 stages of growth curve to stages of disease

Phases of growth curve <i>In vitro</i>	Stages of disease <i>In vivo</i>
lag phase	Incubation period of disease
Logarithmic & Stationary phase	Clinical signs & symptoms
Decline phase	Recovery & convalescence

Bacterial Reproduction

- * Bacterial cell division is a asexual
- * Start by duplication of chromosome
 - * Each copy attach to cytoplasmic membrane at mesozome
- * Cytoplasmic Membrane forms a transverse membrane growing inwards
- * A new transverse cell wall grows inwards
 - * A complete transverse septum separate two daughter cells

Growth Requirement Of Bacteria

- * **Growth of bacteria depends on:**

- adequate supply of food

- * **Food is essential for :**

- Build up of protoplasm
- Production of energy

- * **Metabolic activities are brought about:**

- Various

- * **Enzyme activity is conditioned by:**

- Moisture, Temperature, pH

Bacterial Nutrition -1

2- Heterotrophic bacteria:

- Most bacteria of medical importance.
- Require complex preformed organic substance.
- Utilize simple inorganic substances as:
 - Obtained food from plant or animal source.
 - Live in or on animal body (parasitic bacteria).
- Many grow on simple media from:
 - Some require complex organic material (Blood, serum).

Gaseous Requirements -2

Oxygen requirements: 4 groups -1

- b- Facultative anaerobes:
- c- Obligate aerophilic bacteria:
- a- Obligate anaerobes:
- Bacteria that grow in presence or absence of O_2
 - Grow only if complete absence of O_2
 - Only grow in presence of free oxygen
 - Use O_2 to generate energy by aerobic respiration if present
 - In presence of O_2 , toxic molecules are produced (H_2O_2)
 - Energy system depends on O_2 as H acceptor
 - Use anaerobic respiration in absence of O_2
 - Anaerobic bacteria lack enzymes that breakdown toxic molec.

Carbon dioxide (CO₂) requirement

- Bacteria require CO₂ minute quantities as in air
- Some require higher CO₂ concentration (carboxyphilic)
 - e.g. Pathogenic *Neisseria* (5 % CO₂)
 - Brucella abortus* (20 % CO₂)

3- Physical Requirements Of Bacteria

(Hydrogen ion concentration (pH -2

- Pathogenic bacteria grow at a narrow range of pH (7.2 - 7.6)
- Few species require an alkaline pH (*Vibrio cholerae*, pH 8)
- Some prefer an acid pH (*Lactobacilli*, pH 4)

	Temperature range	Optimum temperature
Mesophilic bacteria	C° 42 - 37	C° 37
Psychrophilic bacteria	C° 30 - 5	C° 20 -15
Thermophilic bacteria	C° 80 - 25	C° 60 -50

Microbiology Techniques

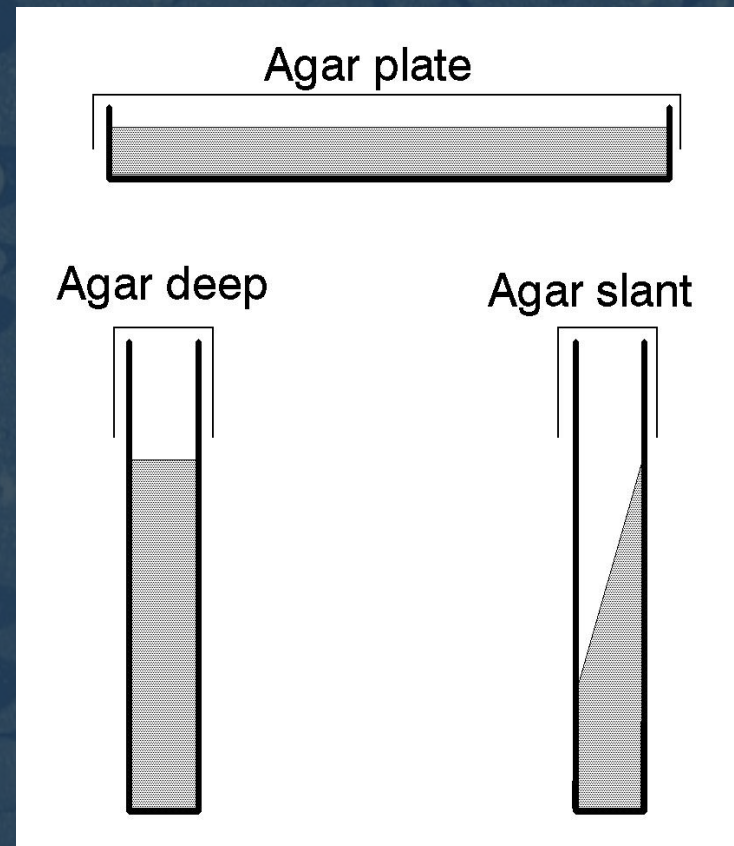
Media Types:

1- Solid Medium

- Agar Plate
- Agar Deep
- Agar Slant

2- Semi-Solid Medium

3- Liquid Medium



How to hold an Inoculating Loop

Inoculating Loop

- The inoculating loop should be held comfortably, much as you would hold a pencil.



Flaming the Loop

Flaming the Loop

- Flaming the loop helps to prevent contamination of the bacteria.
- When flaming the loop, make sure that all of the wire has been heated to **redness**.



Streak Plate



Transfer to tubes

Remove the Caps

- While the wire is cooling, remove the first cap by grasping it with the little finger.



Indicator concept and criteria

- Should be absent in unpolluted water and present when the source of pathogenic microorganisms of concern is present.
- Should be easy to isolate, identify and enumerate. Over time, the health risk to analysts).
- Should not multiply in the environment.
- Should not be a pathogenic microorganism (to minimise the health risk to analysts).
- Should be present in greater numbers than the pathogenic microorganisms.
- The test should be inexpensive thereby permitting numerous samples to be taken.
- Should respond to natural environmental conditions and water treatment processes in a manner similar to the pathogens of concern.

he coliform group :

Total coliforms: Coliform organisms, better referred to as total coliforms to avoid confusion with others in the group, are not an index of faecal pollution or of health risk, but can provide basic information on source water quality. Total coliforms have long been utilised as a microbial measure of drinking water quality, largely because they are easy to detect and enumerate in water.

Capable of fermenting lactose at 35-37°C with the production of acid, gas within 24-48 hours.

Escherichia, Citrobacter, Enterobacter, and Klebsiella.

thermotolerant ('faecal') coliforms:

defined as the group of total coliforms that are able to ferment lactose at 44-45°C. They comprise the genus *Escherichia* and, to a lesser extent, species of *Klebsiella*, *Enterobacter*, and *Citrobacter*.

only *E. coli* is considered to be specifically of faecal origin, being always present in the faeces of humans, other mammals, and birds in large numbers

***Escherichia coli*:**

- *E. coli* is detectable by simple, inexpensive cultural methods that require basic routine bacteriology laboratory facilities, but require well-trained and competent laboratory workers. It grows at 44-45°C on complex media, ferments lactose and mannitol with the production of acid and gas, and produces indole from tryptophan.
- Characterised by possession of the enzymes β -galactosidase and β -glucuronidase. Some strains of this organism are pathogenic.

- **Widely preferred as an index of faecal contamination.**

Enterococci and faecal streptococci :

- All possess the Lancefield group D antigen.
 - Enterococci are detectable by simple, inexpensive culture methods that are easy to perform and can generally be regarded as specific indices of human faecal pollution for most practical purposes.
 - Faecal *streptococci* are more resistant to stress and bacteria are pathogenic.
- chlorination than *E. coli* and the other coliform bacteria.

Sulphite-reducing clostridia **and** ***Clostridium perfringens***

- Obligately anaerobic, spore-forming organisms,
 - *Clostridium perfringens*, is normally present in faeces
 - They are not normally a health risk for laboratory workers but they are pathogenic and if carelessly handled can give rise to food poisoning and wound infections.
- Clostridia are not, however, recommended for the routine monitoring of distribution systems because of their long life span. Because of their long life span they may be detected long after (and far from) the pollution event, leading to possible false alarms

***Pseudomonas aeruginosa* and *Aeromonas* spp.**

- environmentally widespread, with some being opportunistic pathogens.
- ***Ps. aeruginosa*** is commonly found in faeces, soil, water, and sewage but cannot be used as an index of faecal contamination.
- ***Aeromonas*** shows no particular association with faecal pollution.
- Neither *Pseudomonas* nor *Aeromonas* are indices of faecal pollution

Bacteriophages

:

- Divided into two groups, both of which occur in sewage and faecally polluted water.

1- Somatic coliphages :

- frequently detected in human and animal faeces.

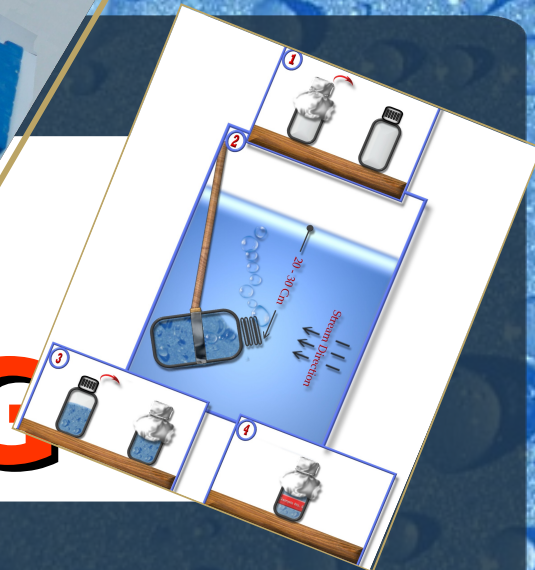
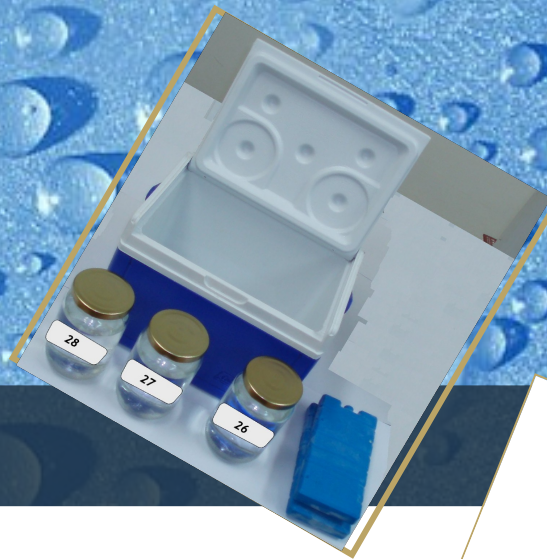
2- F- Specific RNA bacteriophages

:

- Although they are only present in the faeces of a small proportion of people, they are commonly found in high numbers in sewage.

Protozoan parasites :

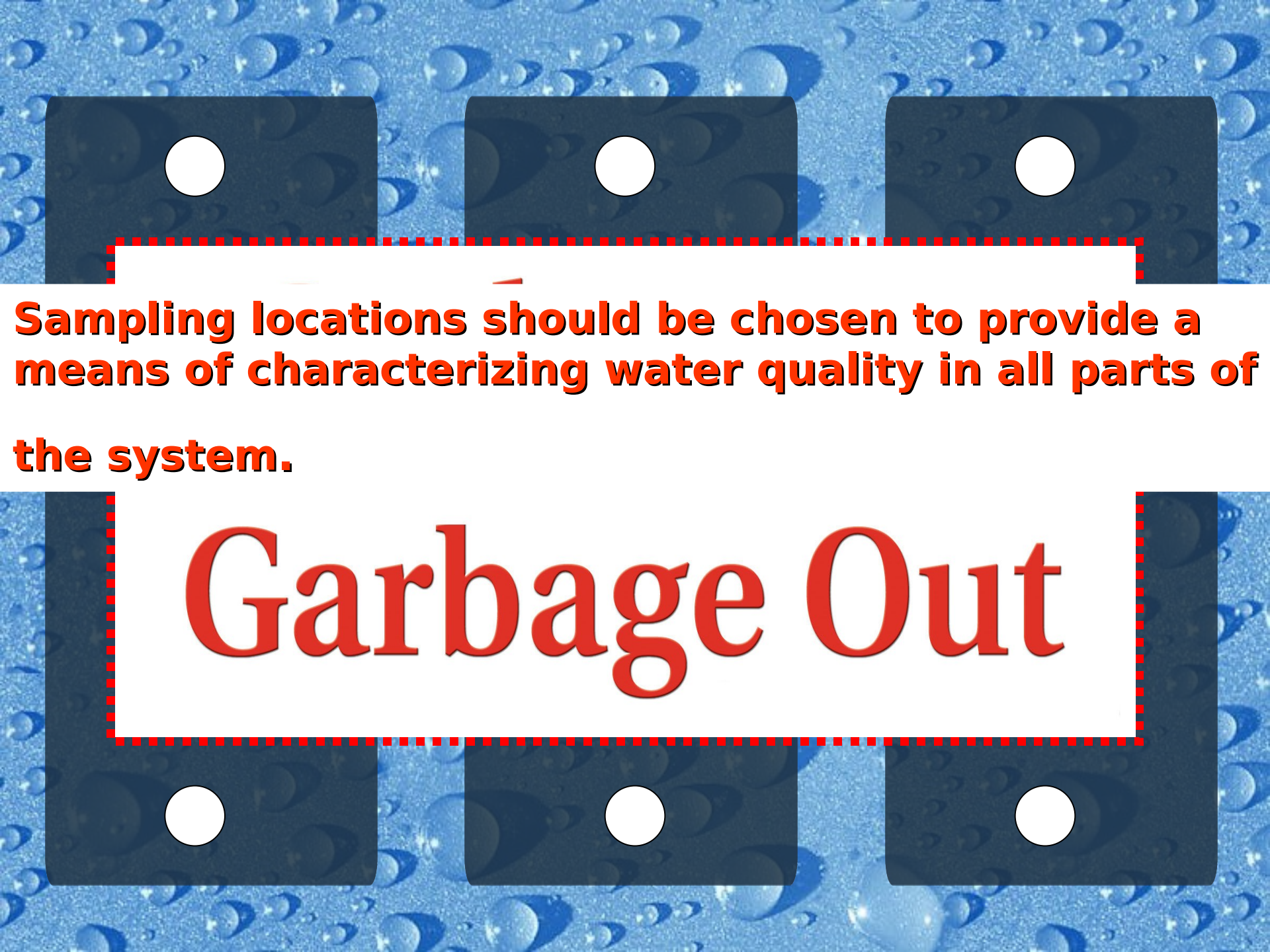
- ***Cryptosporidium*** oocysts and ***Giardia*** cysts are associated with human and animal faecal sources.
- The failure to detect cysts or oocysts does not constitute an indication of the absence of faecal pollution.
- They can survive for very long periods in the environment and are quite resistant to treatment.



SAMPLING



Garbage In
Garbage Out



Sampling locations should be chosen to provide a means of characterizing water quality in all parts of the system.

Garbage Out

Sampling Bottle

- Capacity of at least 200 ml.
- Sterile bottles containing **sodium thiosulphate**.
- When collecting the sample, exercise extreme care to avoid contaminating it with bacteria from the environment.
- Stopper the bottle, label it with full details, and deliver it to the laboratory as quickly as possible.



Storage of samples for microbiological analysis :

Although recommendations vary, the time between sample collection and analysis should, in general,

not exceed 6 hours, and 24 hours is considered the absolute maximum

It is assumed that the samples are immediately placed in a lightproof insulated box containing melting ice or ice-packs with water to ensure rapid cooling.

If ice is not available, the transportation time must not exceed 2 hours. It is imperative that samples are kept in the dark and that cooling is rapid. If these conditions are not met, the samples should be discarded.

Lightproof insulated box containing ice-packs



Information that should be supplied with the samples

Code No

Date of collection

Time of collection

Collected by

Reason of examination

Is there any infected cases

Free residual chlorine

Kind of treatment

.....
Source of sample with exact place ...

Date of Collection	/ / 2005		Code Number: ()		
Time of Collection					
Collected By:					
Reason of Examination	Routine Sample		Otherwise		
Source of Sample	Well		House Tap		other
	Drilled by Hand	Drilled by Driller	Cistern	Direct From the Main	
Depth of the Well			Date of Drilling		
Treatment	Chlorination		Untreated		Other
Possible Source of Pollution	Yes	No			
	Approximate Distance				

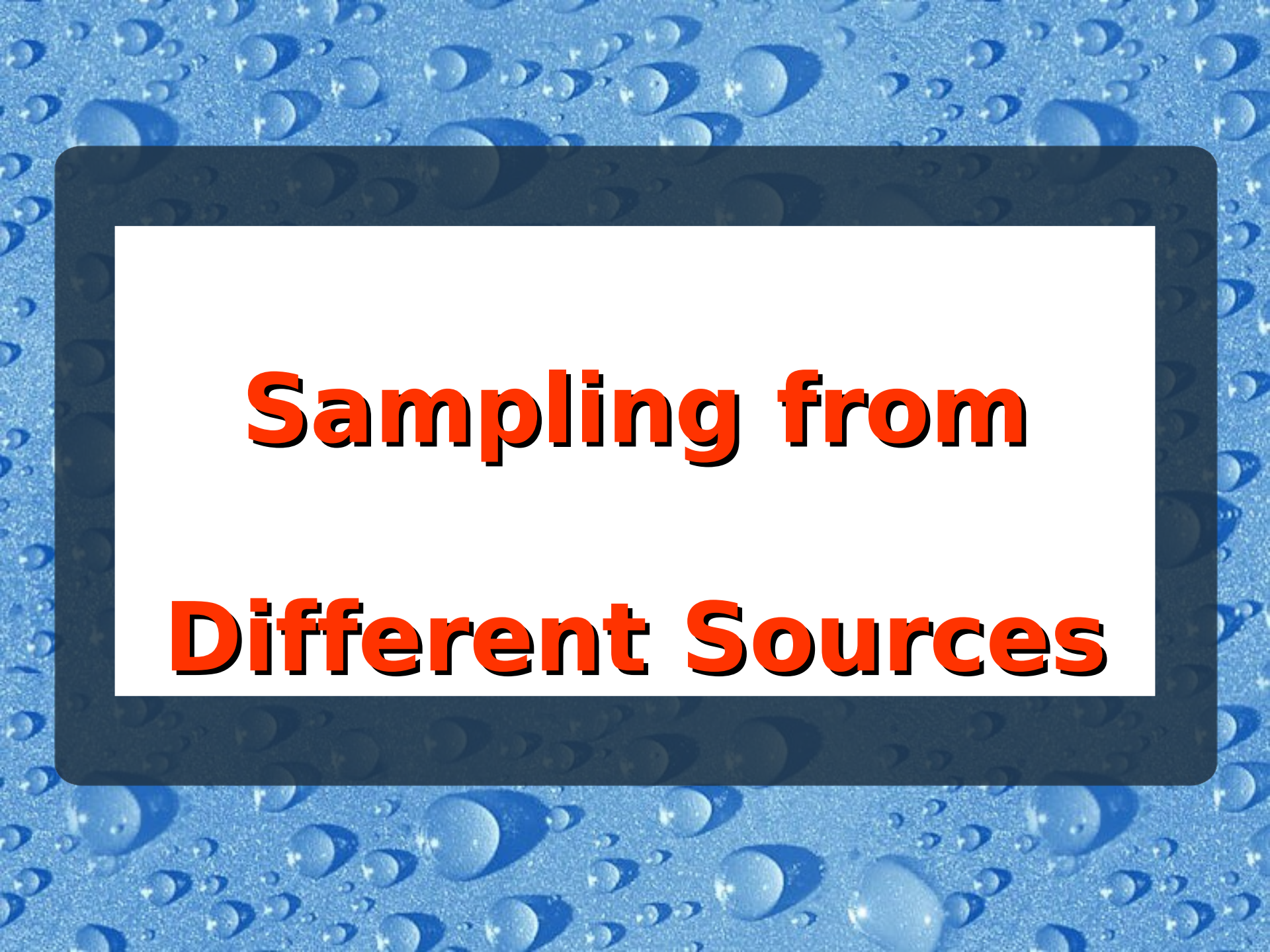
Total Count

Total Coliform

Thermotolerant Coliform

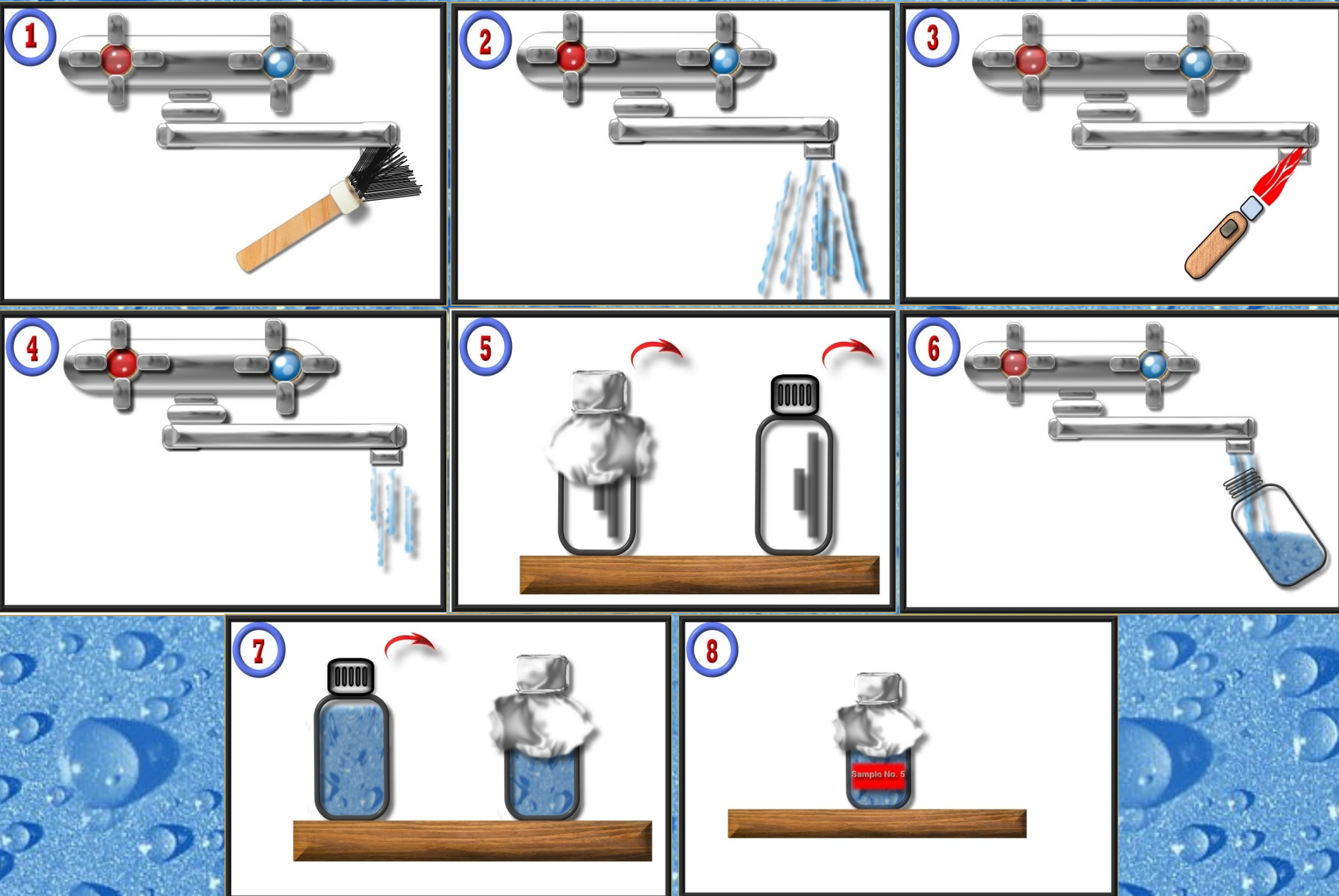
Frequency of Sampling

Population served	Samples to be taken monthly
Less than 5000	sample 1
000 5000-100	sample / 5000 population 1
More than 100 000	sample / 10 000 population, 1 .plus 10 additional samples

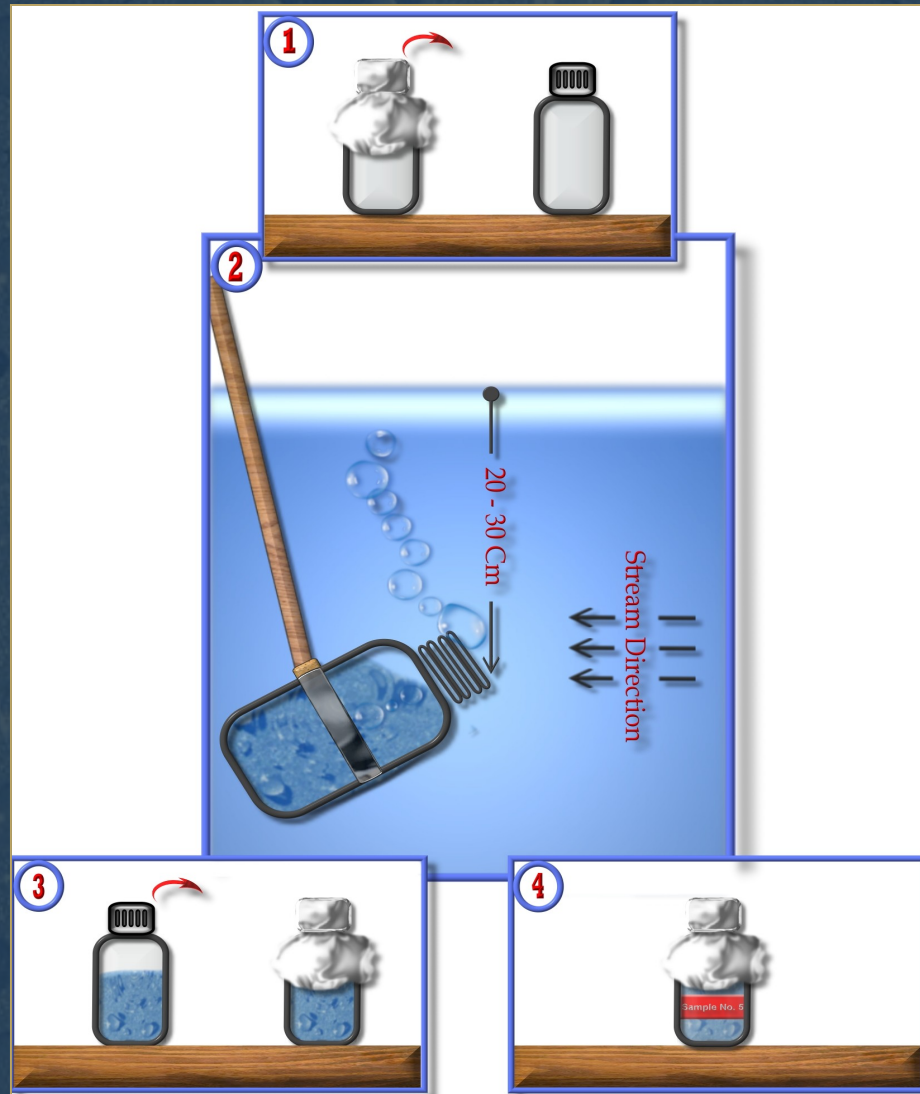


Sampling from Different Sources

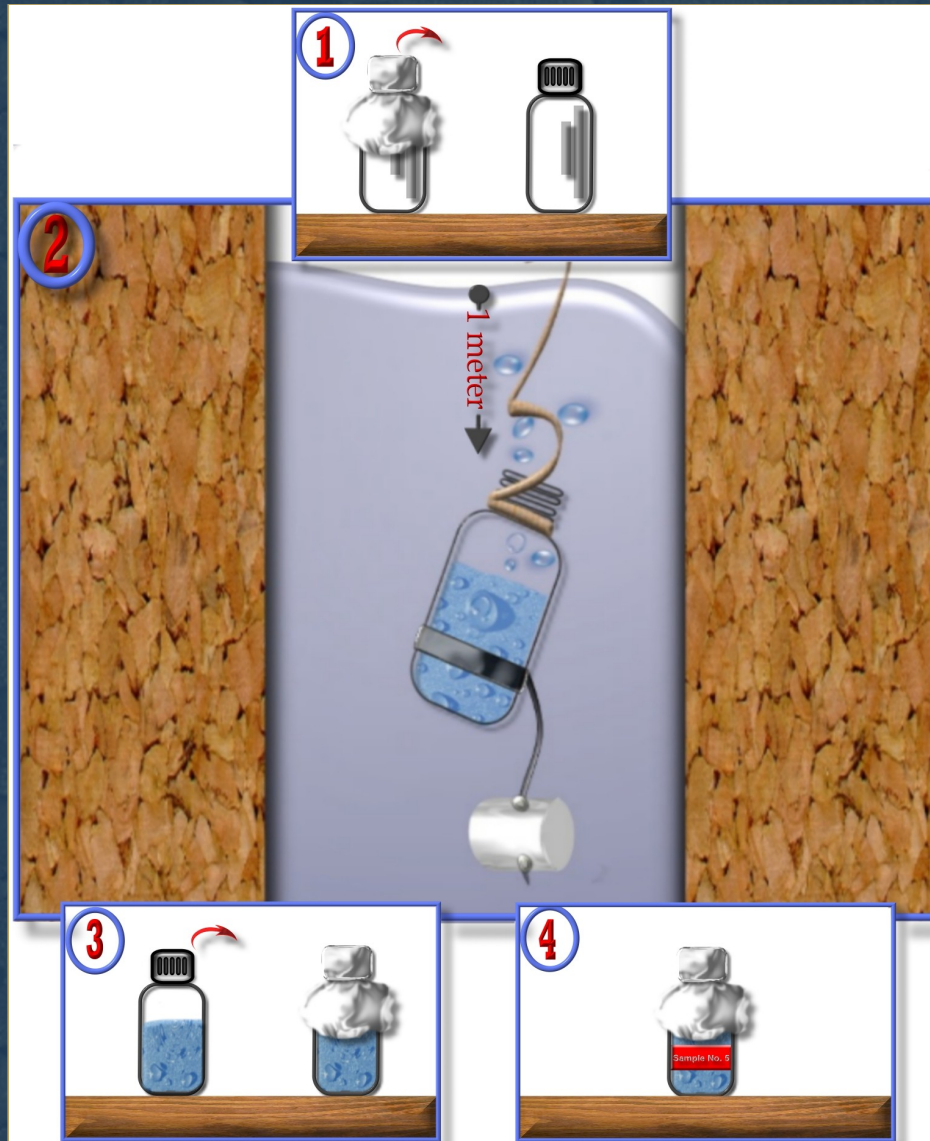
Sampling from Tap

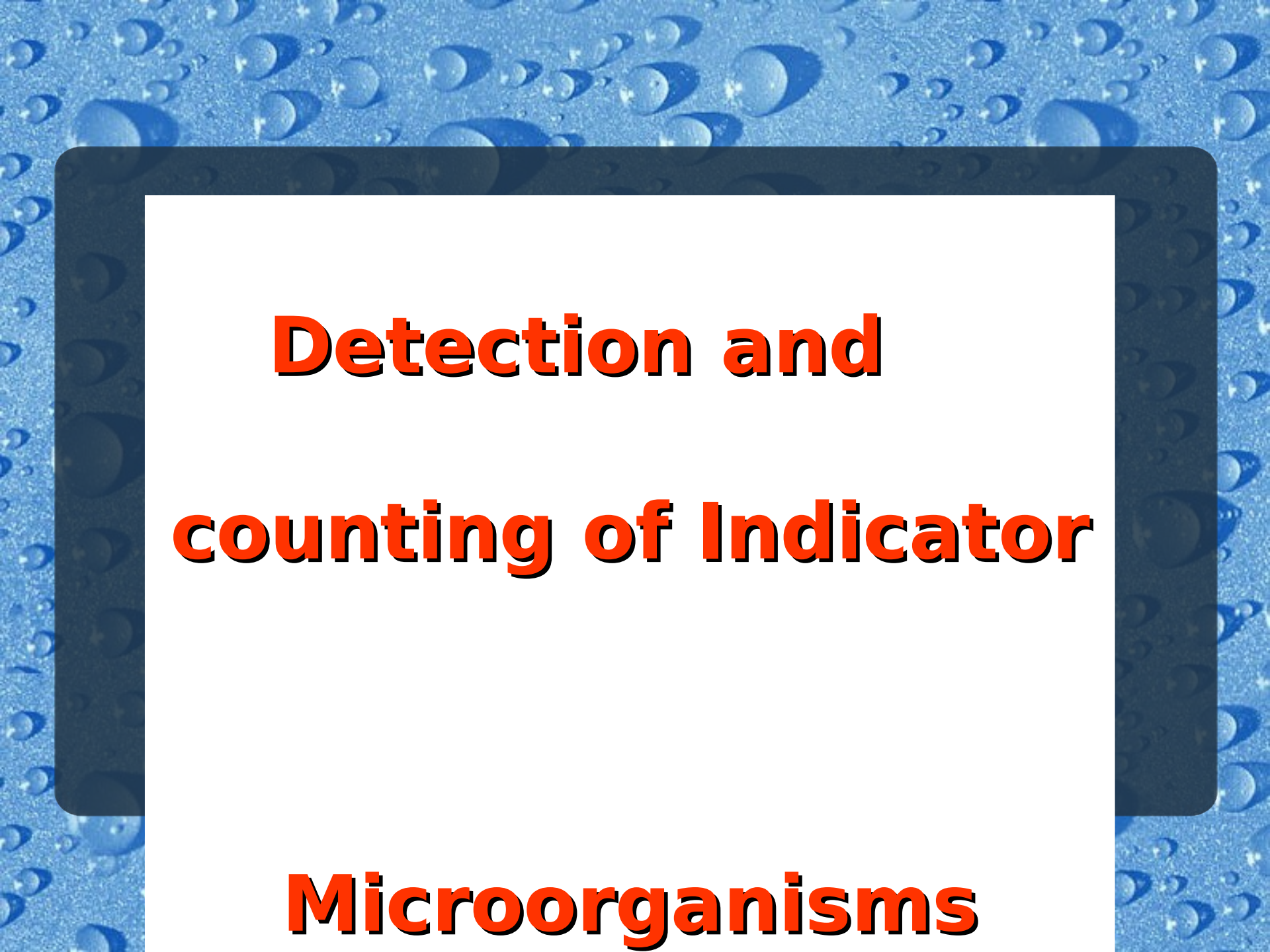


Sampling from Stream or River



Sampling from Well



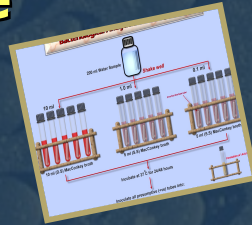


Detection and counting of Indicator

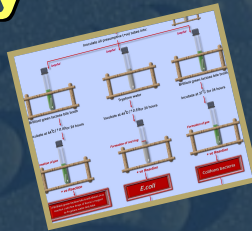
Microorganisms

1- Most Probable Number Technique

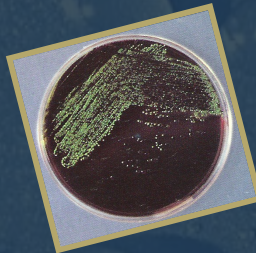
A- Presumptive Test



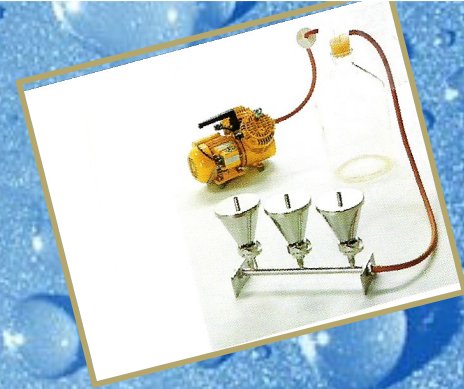
B- Confirmatory Test



C- Completed Test



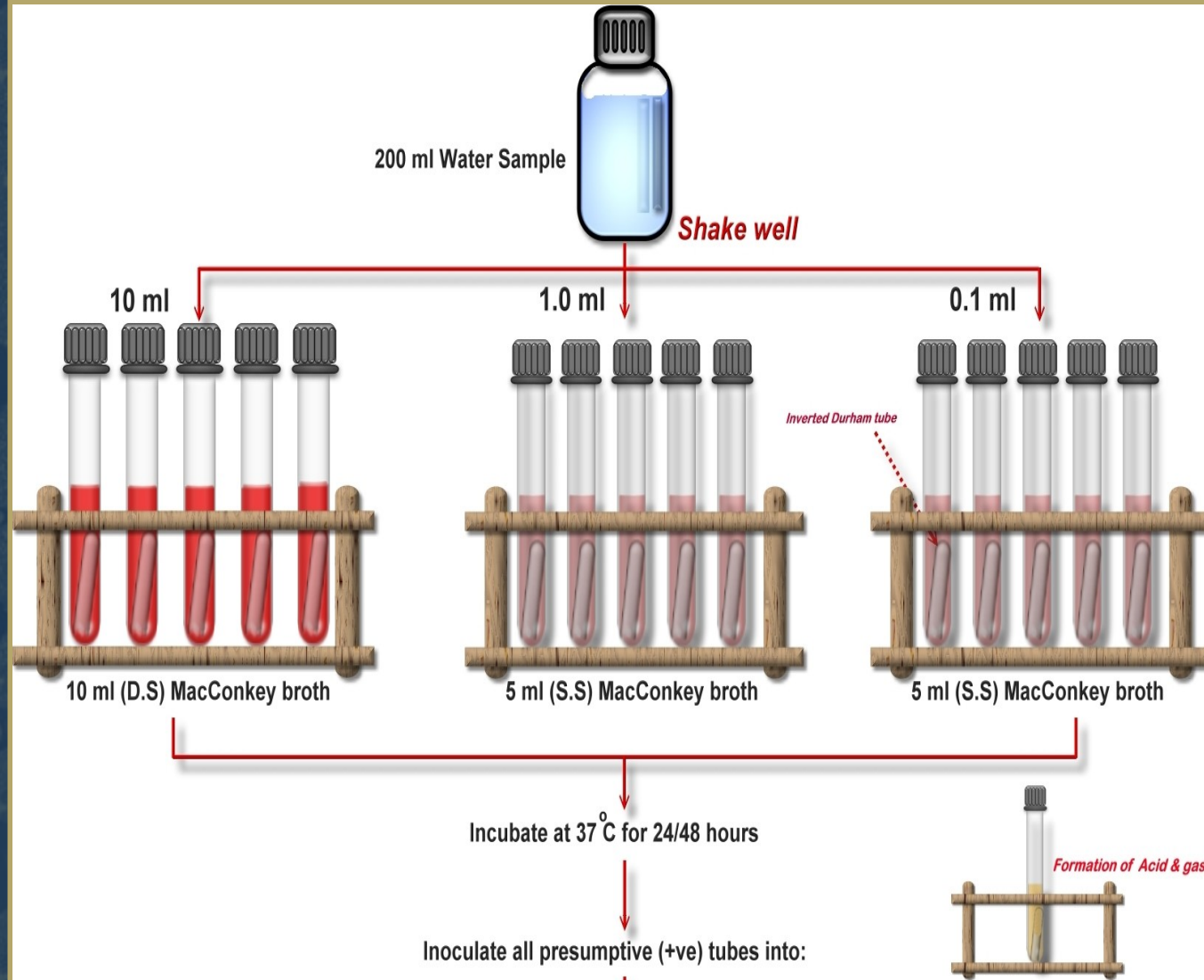
2- Membrane Filtration Technique



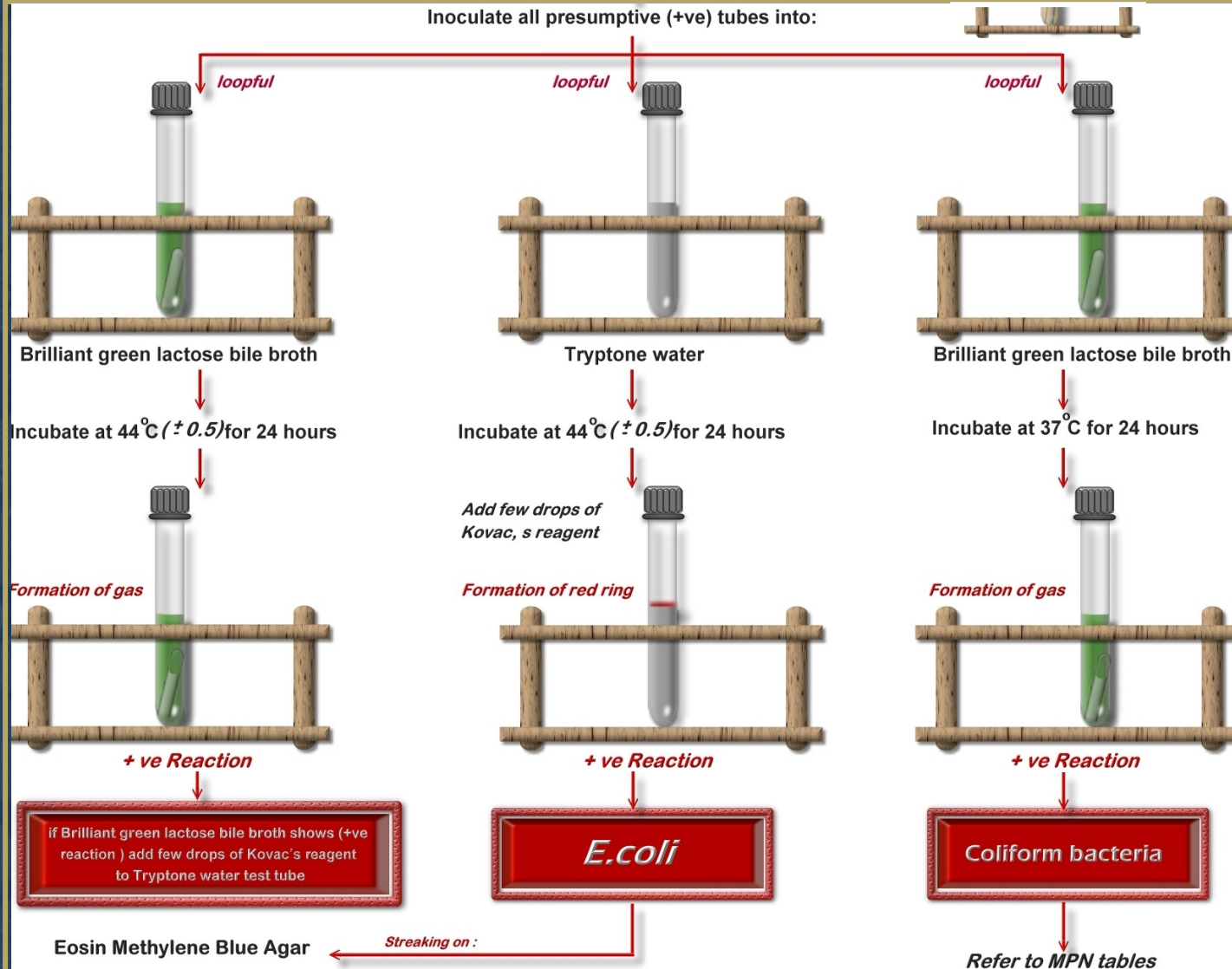
3- Presence-Absence Technique

4-Heterotrophic Plate Count Test

A- Presumptive Test



B- Confirmatory Test



C- Completed Test

Inoculate a loopful of (+ve reaction) test tube of BGB
onto Eosin Methylene Blue Agar

37 °C / 18 – 24
hr.

Green Methallic
Sheen Colonies



E.coli

3 sets of 5 tubes MPN table

Positive Tubes	NdM	Positive Tubes	NdM	Positive Tubes	NdM	Positive Tubes	NdM	Positive Tubes	NdM	Positive Tubes	NdM	Positive Tubes	NdM
10 1.0 0.1		10 1.0 0.1		10 1.0 0.1		10 1.0 0.1		10 1.0 0.1		10 1.0 0.1		10 1.0 0.1	
0 0 0	<1.8	1 0 0	2	2 0 0	4.5	3 0 0	7.8	4 0 0	13	5 0 0	23		
0 0 1	1.8	1 0 1	4	2 0 1	6.8	3 0 1	11	4 0 1	17	5 0 1	31		
0 0 2	3.6	1 0 2	6	2 0 2	9.1	3 0 2	13	4 0 2	21	5 0 2	43		
0 0 3	5.4	1 0 3	8	2 0 3	12	3 0 3	16	4 0 3	25	5 0 3	58		
0 0 4	7.2	1 0 4	10	2 0 4	14	3 0 4	20	4 0 4	30	5 0 4	76		
0 0 5	9	1 0 5	12	2 0 5	16	3 0 5	23	4 0 5	36	5 0 5	95		
0 1 0	1.8	1 1 0	4	2 1 0	6.8	3 1 0	11	4 1 0	17	5 1 0	33		
0 1 1	3.6	1 1 1	6.1	2 1 1	9.2	3 1 1	14	4 1 1	21	5 1 1	46		
0 1 2	5.5	1 1 2	8.1	2 1 2	12	3 1 2	17	4 1 2	26	5 1 2	64		
0 1 3	7.3	1 1 3	10	2 1 3	14	3 1 3	20	4 1 3	31	5 1 3	84		
0 1 4	9.1	1 1 4	12	2 1 4	17	3 1 4	23	4 1 4	36	5 1 4	110		
0 1 5	11	1 1 5	14	2 1 5	19	3 1 5	27	4 1 5	42	5 1 5	130		
0 2 0	3.7	1 2 0	6.1	2 2 0	9.3	3 2 0	14	4 2 0	22	5 2 0	49		
0 2 1	5.5	1 2 1	8.2	2 2 1	12	3 2 1	17	4 2 1	26	5 2 1	70		
0 2 2	7.4	1 2 2	10	2 2 2	14	3 2 2	20	4 2 2	32	5 2 2	95		
0 2 3	9.2	1 2 3	12	2 2 3	17	3 2 3	24	4 2 3	38	5 2 3	120		
0 2 4	11	1 2 4	16	2 2 4	19	3 2 4	27	4 2 4	44	5 2 4	150		
0 2 5	13	1 2 5	17	2 2 5	22	3 2 5	31	4 2 5	50	5 2 5	180		
0 3 0	5.6	1 3 0	8.3	2 3 0	12	3 3 0	17	4 3 0	27	5 3 0	79		
0 3 1	7.4	1 3 1	10	2 3 1	14	3 3 1	21	4 3 1	33	5 3 1	110		
0 3 2	9.3	1 3 2	13	2 3 2	17	3 3 2	24	4 3 2	39	5 3 2	140		
0 3 3	11	1 3 3	16	2 3 3	20	3 3 3	28	4 3 3	45	5 3 3	180		
0 3 4	13	1 3 4	17	2 3 4	22	3 3 4	31	4 3 4	52	5 3 4	210		
0 3 5	15	1 3 5	19	2 3 5	25	3 3 5	35	4 3 5	59	5 3 5	250		
0 4 0	7.5	1 4 0	11	2 4 0	16	3 4 0	21	4 4 0	34	5 4 0	130		
0 4 1	9.4	1 4 1	13	2 4 1	17	3 4 1	24	4 4 1	40	5 4 1	170		
0 4 2	11	1 4 2	15	2 4 2	20	3 4 2	28	4 4 2	47	5 4 2	220		
0 4 3	13	1 4 3	17	2 4 3	23	3 4 3	32	4 4 3	54	5 4 3	280		
0 4 4	16	1 4 4	19	2 4 4	25	3 4 4	36	4 4 4	62	5 4 4	350		
0 4 5	17	1 4 5	22	2 4 5	28	3 4 5	40	4 4 5	69	5 4 5	440		
0 5 0	9.4	1 5 0	13	2 5 0	17	3 5 0	25	4 5 0	41	5 5 0	240		
0 5 1	11	1 5 1	16	2 5 1	20	3 5 1	29	4 5 1	48	5 5 1	350		
0 5 2	13	1 5 2	17	2 5 2	17	3 5 2	32	4 5 2	56	5 5 2	540		
0 5 3	16	1 5 3	19	2 5 3	26	3 5 3	37	4 5 3	64	5 5 3	920		
0 5 4	17	1 5 4	22	2 5 4	29	3 5 4	41	4 5 4	72	5 5 4	1600		
0 5 5	19	1 5 5	24	2 5 5	32	3 5 5	45	4 5 5	81	5 5 5	>1600		

MPN Calculator Software

1. Select MPN type

3 tube, 3 dil 0.1, 0.01, 0.001 Grams

2. Enter MPN data

Grams per test	Tests	Positive tests
0.1	3	3
0.01	3	1
0.001	3	0

Keypad

BS	RTB	TAB
7	8	9
4	5	6
1	2	3
.	0	

3. Calculate MPN

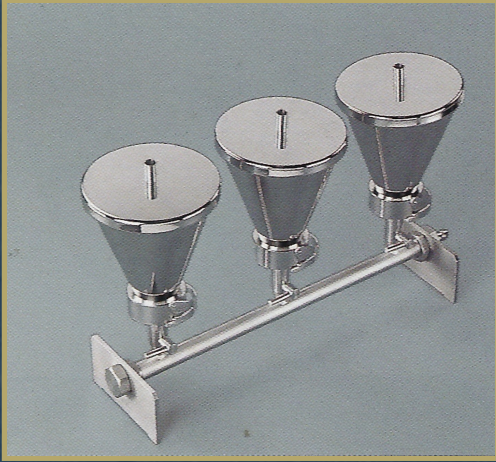
MPN/Gram	42.7
95% CI LL:	10.4
95% CI UL:	175

Calculate MPN

حجم العينة وعدد الأنابيب المستعملة وذلك حسب نوعية المياه

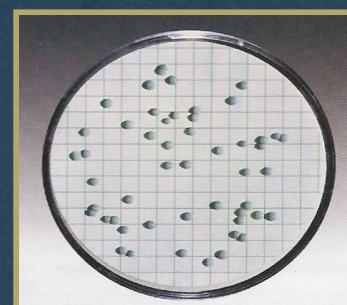
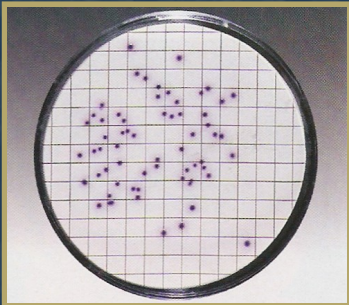
نوع العينة	حجم العينة (مل)				
	50	10	1	0.1	0.01
مياه الشرب المعالجة	1	5			
مياه الشرب المعالجة جزئياً		5	5	5	
مياه الترفيه		5	5	5	
مصادر المياه المحمية		5	5	5	
المياه السطحية			5	5	5

2- Membrane Filtration Method



Recommended for Lab

Analysis



حجم العينة حسب نوعية المياه

نوع العينة	حجم العينة (مل)					
	100	10	1	0.1	0.01	0.001
مياه الشرب المعالجة	X					
مياه الشرب المعالجة جزئياً	X	X				
مياه الترفيه			X	X		
مصادر المياه المحمية	X	X	X			
المياه السطحية			X	X	X	
مياه الصرف الصحي			X	X	X	
مياه الصرف الصحي المعالجة			X	X	X	X
البرك, الأنهار, مياه الفيضان				X	X	X

Small volumes should be added to the filtration apparatus together with a minimum of 9 ml of sterile diluent to ensure adequate dispersal across the surface of the filter membrane.

Advantages & Disadvantages

Most Prob. Number	.Membrane filt. Tech
Slower	Faster
More Labor	Less Labor
More Media Required	LessMedia Required
More Glassware Required	Less Glassware Required
(Less Precise(Statistics	More Precise
More Sensitive	Less Sensitive
Inexpensive	Expensive
Used in the Lab Only	May used in the field
Can be used for all Kinds of Water	Not Recommended for Turbid Water
Enhance Stressed Colonies to Grow	

Heterotrophic Plate Count

Only a small fraction ($\sim 0.01\%$) of waterborne M.Os are thought to belong to the group of culturable HB, and $\sim 1\%$ of the viable bacteria are Not culturable.

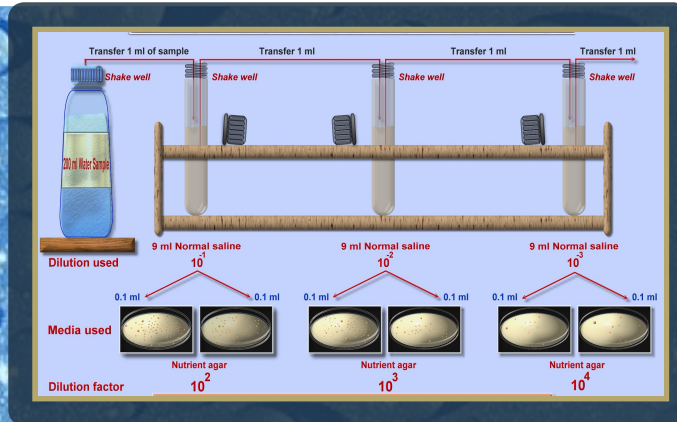
- A longer cultivation time (5-7 days at 27°C).
- There is no clear-cut evidence that HB as such pose a public health risk.



Use of HPC in Water Management

- To indicate the effectiveness of water treatment processes.
- As a measure of No. of growth organisms that may or may not have a significance.
- As a measure of possible interference with coliform Measurements in Lactose-based culture method.

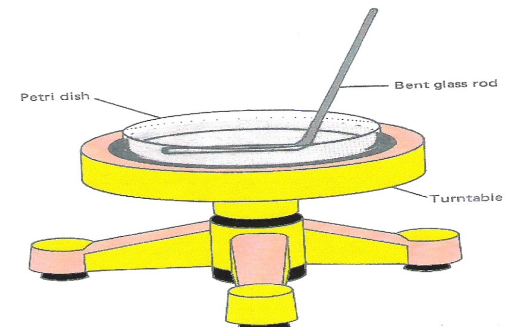
Serial Dilution Method



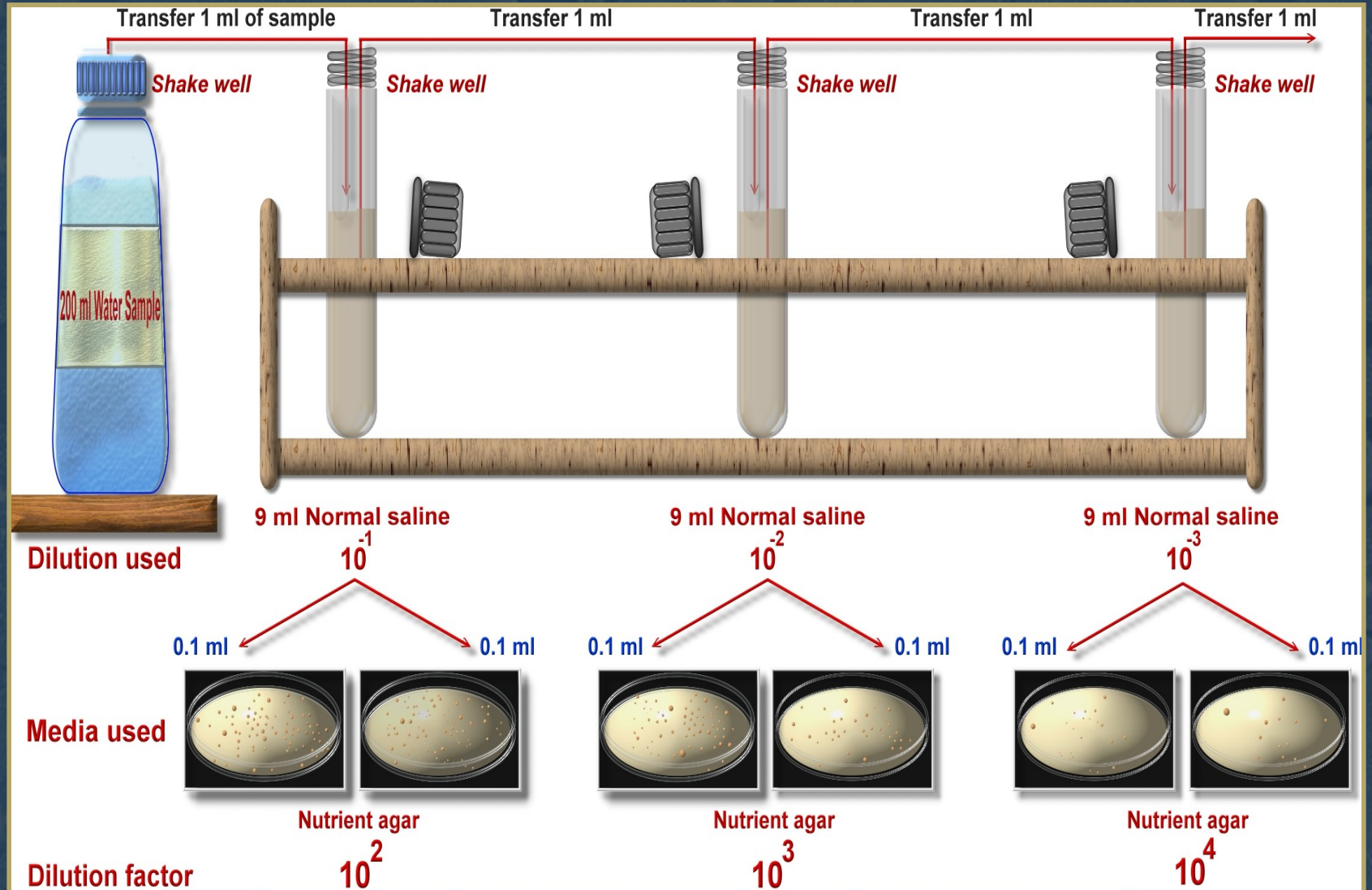
A- Pour Plate Technique



B- Spread Plate Technique



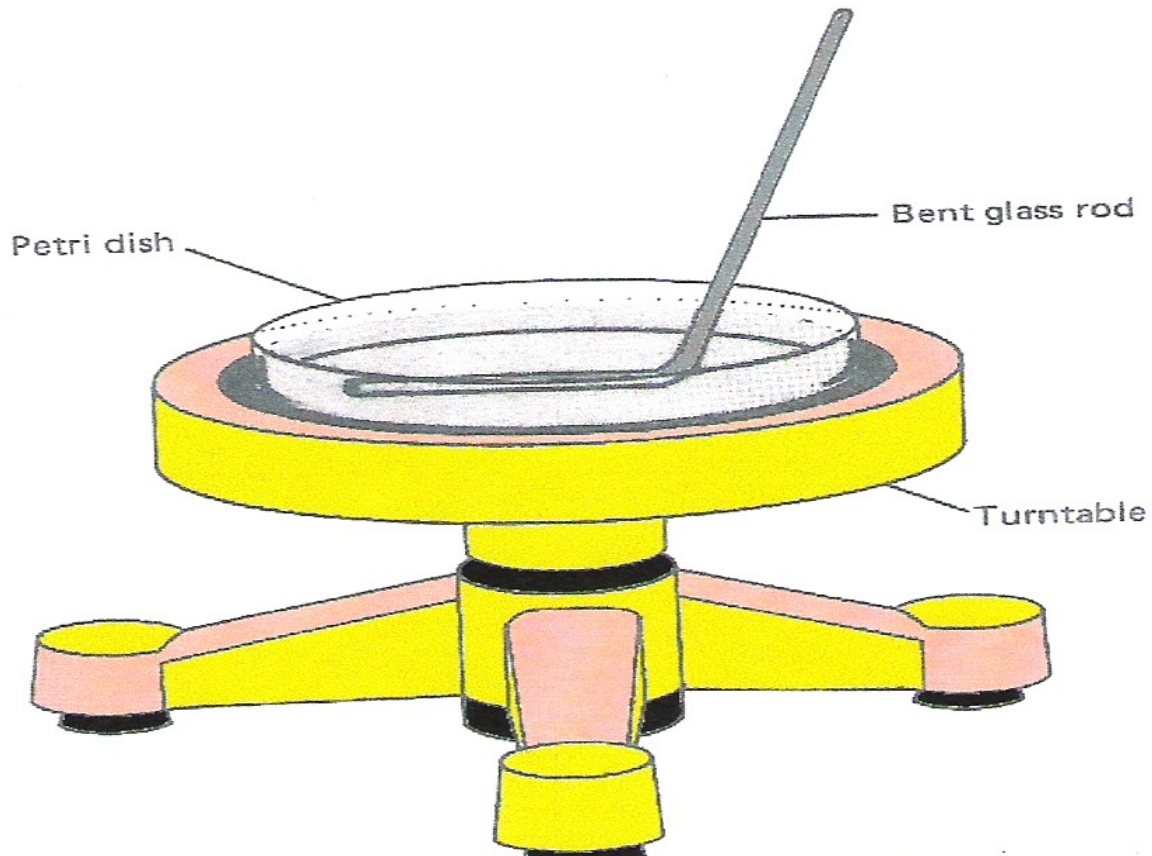
Serial Dilution Method



A- Pour Plate Technique



B- Spread Plate Technique



Quality Control Standard

**A. For Water drawn from the
distribution system**

**B. For Water drawn from
unpiped well**

A. Water drawn from the distribution system:

Quality of supply	Results from routine samples -----		Tolerance
	coliform count/ 100ml	<i>E. coli</i> count/100ml	
Excellent -1	0	0	In all samples
Satisfactory -2	0	3 - 1	Provided that coliform organisms do not occur in consecutive samples or in more than 5% of samples
Intermediate -3	0	9 - 4	
Unsatisfactory -4	coliforms or more, 10 <i>E. coli</i> or any coliform organisms present in consecutive samples. or presence of any coliform organisms in more than 5% of routine samples		In any sample

B. Water drawn from unpiped well:

*Mean count 44°C, 100 ml <i>E. coli</i> count	Category	Comments
0	A	.Excellent
10 – 1	B	Acceptable: But make regular sanitary checks on equipment
50 – 10	C	Unacceptable: Look for and correct structural faults and poor maintenance of pump and plinth. Then disinfect .equipment and source
More than 50	D	Grossly polluted: Look for alternative source, or carryout necessary repairs, .and disinfect well

Result Documentation & Interpretation

Microsoft Excel - Documentation of results 2007

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	A	B	C	D	E	F	G	H	I	J	K
1	MICROBIOLOGY LAB - RESULTS SHEET- 2007										
2	S.N	Date	Sampling Point	Total Count Per 1ml	Total coliforms M.P.N/100 ml		Thermotolerant coliforms M.P.N/100 ml	E.coli Indole test	Residual Chlorine	Collected by	Analysed by
3	65	15-May-2007	23 D 5	112	1:0:0	2	Negative	NO	0.23	Ashraf	Ashraf
4	66	15-May-2007	44 F 12	68	0:1:0	1.8	Negative	NO	0.22	Ashraf	Ashraf
5	67	16-May-2007	NORTH- 221	96	2:0:0	4.5	Negative	NO	0.27	Ashraf	Ashraf
6	68	16-May-2007	DIST. 33	88	0:1:1	3.6	Negative	NO	0.2	Ashraf	Ashraf
7	69	16-May-2007	DIST.26	330	5:2:1	70	Positive	YES	0.25	Nuri	Ashraf
8	70	17-May-2007	BB 131	51	2:0:0	4.5	Negative	NO	0.29	Ashraf	Ashraf
9	71	18-May-2007	DWS 67 M	87	1:0:0	2	Positive	NO	0.28	Ashraf	Ashraf
10	72	18-May-2007	LAB No. 4	NO GROWTH	0:1:1	3.6	Negative	NO	0.22	Ashraf	Ashraf
11	73	18-May-2007	WELL No. 47	112	1:1:1	6.1	Negative	NO	0.22	Ashraf	Ashraf
12	74	18-May-2007	DIST. 39	225	2:0:0	4.5	Negative	NO	0.21	Ashraf	Ashraf
13	75	19-May-2007	BB 131	364	5:2:5	180	Positive	YES	0.23	Nuri	Nuri
14	76	19-May-2007	23 D 5	294	4:2:1	26	Positive	YES	0.25	Nuri	Nuri
15	77	19-May-2007	23 D 5	150	1:0:1	4	Negative	NO	0.25	Ashraf	Ashraf
16	78	19-May-2007	BB 131	NO GROWTH	3:2:5	31	Negative	NO	0.25	Ashraf	Ashraf
17	79	19-May-2007	NORTH- 221	NO GROWTH	2:0:0	4.5	Negative	NO	0.27	Ashraf	Ashraf
18	80	19-May-2007	SS 3G	105	3:1:1	14	Positive	YES	0.01	Ashraf	Ashraf
19	80	20-May-2007	DBT-88	338	1:1:1	6.1	Positive	YES	0.05	Nuri	Nuri
20											

Sheet1 Sheet2 Sheet3

Ready NUM

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Example of classification and color-code scheme for *E. coli* in water supplies

		<i>E.coli</i> count per 100 ml									
		0	1 - 10			10 - 100		100 - 1000		> 1000	
Color code	E										
	D										
	C										
	B										
	A										
		No Action Required	Low Risk: Low Action Priority			Intermediate Risk :Higher Action Priority		High Risk: Urgent Action		very High Risk: Urgent Action	
			Remarks								

W.H.O GUIDELINES

Drinking Water Treatment

- To provide drinking water to consumers that is free of waterborne pathogens.
- No single treatment process can be expected to remove all of the different types of pathogens that can be found in water.

Microbial

treatment :

- 1- The physical removal of the pathogens.
- 2- The inactivation (death) of the pathogen.

- Coagulation and sedimentation

- Filtration

Chemical inactivation

:

Chemicals used include chlorine, chloramine, chlorine dioxide and ozone.

Factors affecting chemical

TTT :

Dose, Contact time, Temperature and sometimes pH.

Chlorination :

chlorine, chloramines, chlorine dioxide and Monochloramine .

- Nearly 100 years of drinking water chlorination has demonstrated its effectiveness in the inactivation of microbial pathogens

Ozonation :

- Ozone has been used for more than a century for water treatment, mostly in Europe.
- The exact mechanism of how ozone inactivates microbes is not well understood.
- *E. coli* is one of the most sensitive to ozone disinfection, while Gram-positive cocci (*Staphylococcus* and *Streptococcus*), the Gram-positive bacilli (*Bacillus*) and the mycobacteria are the most resistant.
- Ozone is effective against *Giardia* and to a lesser extent *Cryptosporidium*.

UV disinfection :

- UV action results from absorption by nucleic acids (DNA and RNA), leading to the dimerisation of pyrimidine bases.
- Usually a dose of 400 J/m^2 (40 mW s/cm^2) is accepted as being sufficient for efficient treatment.

Three types of light source are used for UV disinfection :

- Low-pressure mercury lamp.
- Medium-pressure mercury lamp.
- Pulsed lasers.

Solar water disinfection :

- In low-income countries the sunlight alone can be used to kill or inactivate many, if not all, of the pathogens found in relatively small amount of water at the point of use.

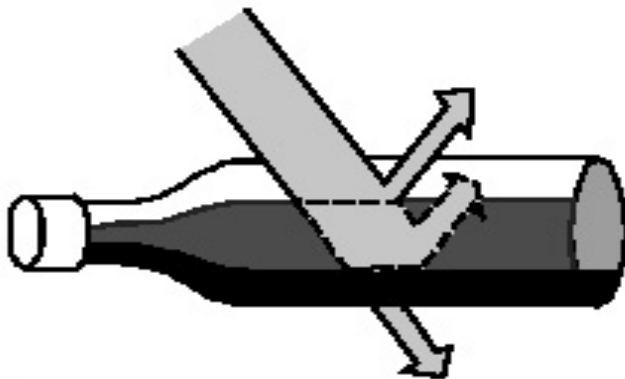
three ways in which solar radiation can be used to eliminate pathogens :

1- Heating.

2- Natural UV radiation.

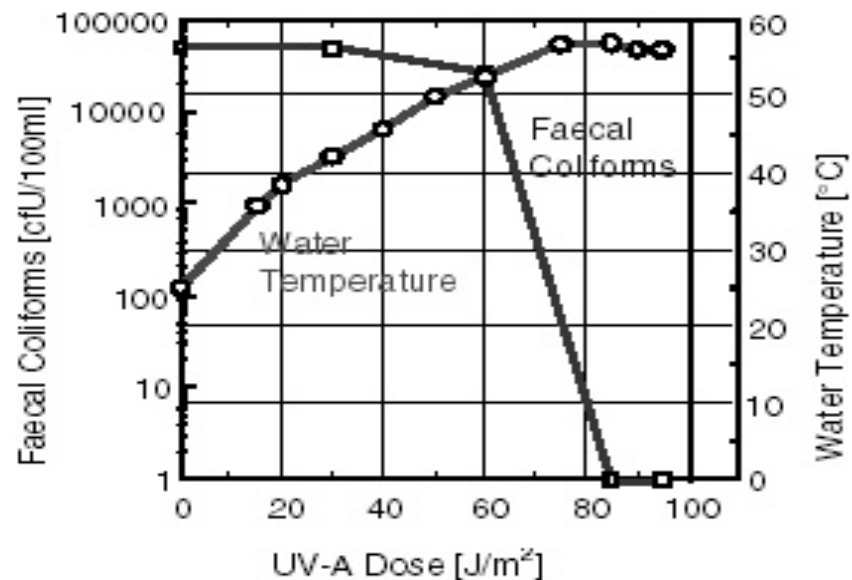
3- Mixture of both thermal and UV effects.

Schematic representation of solar water disinfection and the influence of the water temperature on the UV-inactivation of bacterial cells



SANDEC 8/97

Inactivation of Faecal Coliforms
with Half-Coloured Plastic Bottle



.....Thank you

● Abdurrahman
Tomei



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com



