# ENVIRONMENTAL MICROBIOLOGY.

Dr. Ariyah Terasawat

### Introduction:

### Definition

Environmental microbiology is the study of microbial interactions microbial processes and microbial communities in the environment.

### **Environmental microbiology includes:**

Study of

Structure and activities of microbial communities.
Microbial interaction and interaction with macroorganisims.
Population biology of microorganisms.
Microbial communities genetic and evolutionary processes.
Element cycles and biogeochemical processes.
Microbial life in extreme and unusual environment.



### **Diversity:**

 $\succ$ The ability of microbes tiny organisms that do big job in our environment play a key role in promoting biodiversity.

> These extremely high levels of biodiversity may help ensure the stability of ecosystem processes in the face of environmental change.

>Dormancy is a critical factor that maintains microbial diversity.

>Microbes are the most abundant and diverse organisms on earth they carry out essential ecosystem services.

>Diversity and abundance are determined by the

biogeographically habitat they occupy



# MICROBIAL HABITATS



#### **Microbial Habitats**

- > These are found in just about every kind of habitat.
- $\succ$  Microbes are incredibly diverse thriving in environments from the very cold to the extremely hot.
- $\succ$  They are also tolerant of many other conditions such as limited water availability high salt content and low oxygen levels.
- $\succ$  Not every microbe can survive in all habitats.





# DISTRIBUTION IN NATURE

Grow where they get food moisture and temperature suitable for growth:

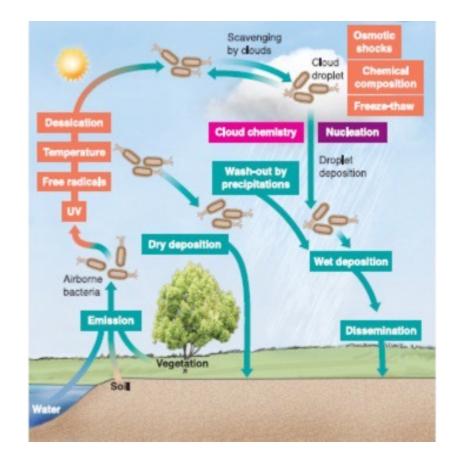
Air

Soil (Terrestrial)

Lake and Oceans (Aquatic)

Another organism

Extreme environment



### TERRESTRIAL HABITATS

>Only one percent of microbes that live in soil have been identified.

> These organisms take part in the formation of soil and are essential components of their ecosystems.

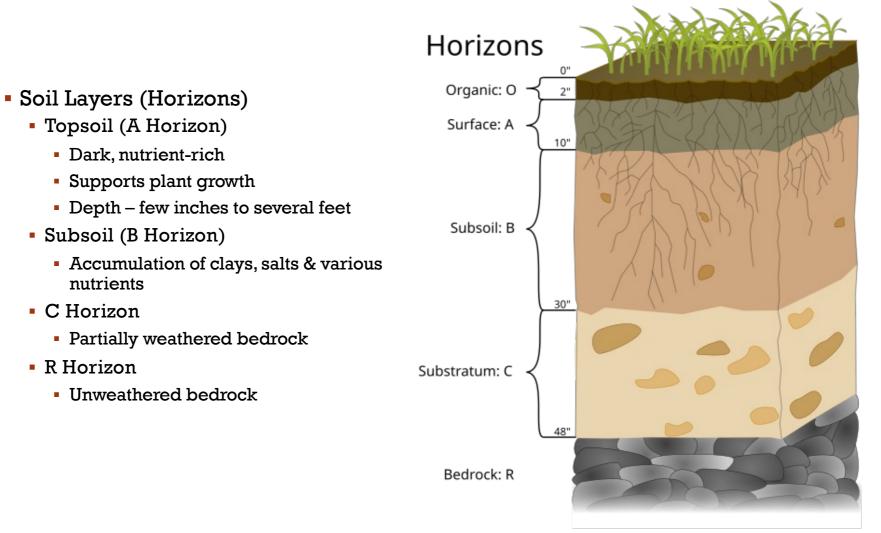
>Bacteria and fungi that live in soil feed mostly on organic matter such as other plants and animals.

>These microbes are very sensitive to their local environment.

>Factors such as the levels of carbon dioxide and oxygen the pH moisture and temperature all affect the growth of microbes in the soil.



# TERRESTRIAL HABITATS





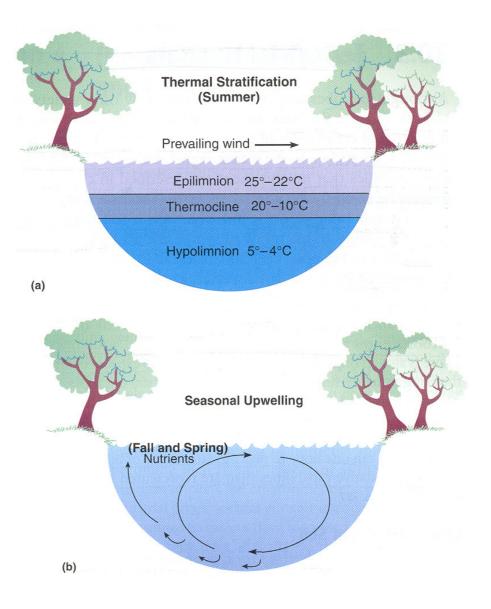
# AQUATIC HABITATS

Microbes live in both fresh and salt water.
These organisms include microscopic plants and animals as well as bacteria fungi and viruses.
As with other microbes the ones that live in water are adapted to the specific conditions of their environment.
Habitats range from ocean water with an extremely high salt content to freshwater lakes or rivers.



# AQUATIC HABITATS

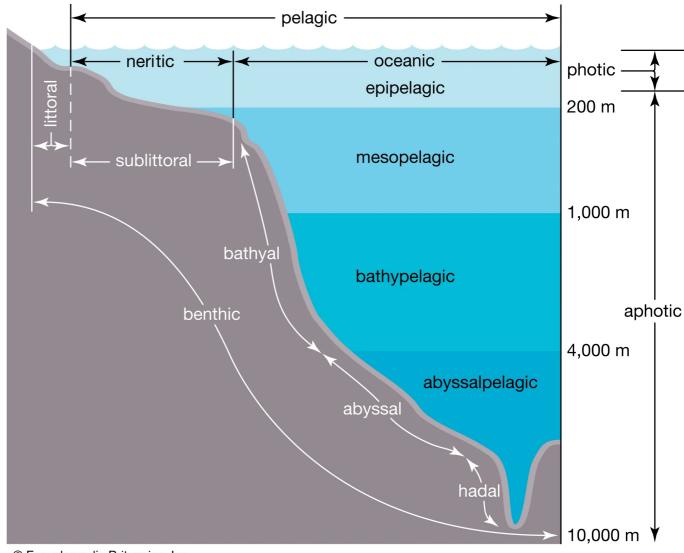
- Freshwater environments
  - Oligotrophic lakes may have anaerobic layers due to thermal stratification
    - Epilimmion
      - Warm upper layer (25°-22°C)
      - Generally oxygen rich due to photosynthetic organisms
      - Generally aerobic
    - Hypolimmion
      - Colder deeper layers (~5°-4°C)
      - May be anaerobic due consumption of O<sub>2</sub> by heterotrophs
      - Water most dense at 4°C (39°F)
    - Thermocline (~20°-10°C)
      - Zone (layer) of rapid temperature change
    - As weather cools, water mixes oxygenating deep water



# AQUATIC HABITATS

- Marine Environment
  - Factors affecting presence of miroorganisms
    - Same Factors as Fresh Water plus
    - Barometric pressure (hydrostatic pressure)
      - 1 atm / 33 feet of seawater
      - ocean 35,750 feet (11,000 meters) deep, hydrostatic pressure - 1,083 atm
      - Organisms are barophilic (barophiles)
    - Salinity
      - Marine averages 3.5% (fresh averages ~0.5%)
      - Organisms are halophilic (halophiles) or halotolerant

#### Zonation of the ocean



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# **MICROORGANISMS IN AIR**

#### **AirPollution:-**

The presence in or introduction into the air of a substance which has harmful or poisonous effects.

### **AIR BORNE POLLUTION:-**

Air borne particles are major causes of respiratory element of human, causing asthma and pathogen infection of the respiratory tract. Airborne fungal spores are also important agents of plan diseases, and the means for dissemination of many common saprotrophic fungi.



#### **Common Example :-**

During a sneeze, millions of tiny droplets of water and mucus are expelled at about 200 miles per hours (100 meters per second). The droplets initially are about 10 - 100micrometers diameter, but they dry rapidly . droplet nuclei of 1 - 4 micrometer containing virus particles or bacteria. This is the major mean of transmission of several diseases of human.

Important diseases of human transmitted from person to person by inhaled air borne particles.

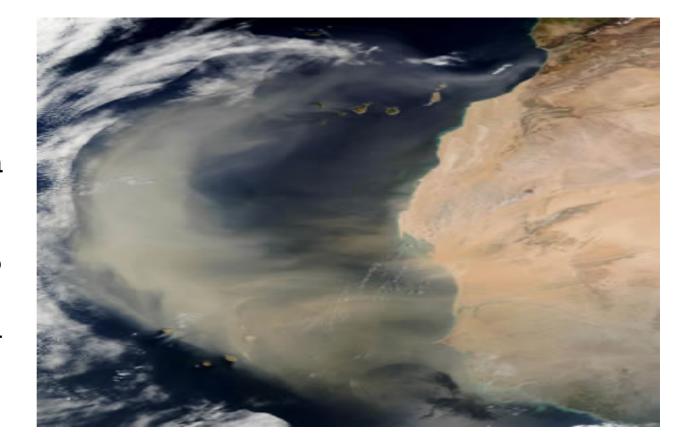
Diseases from Virus:-Chickenpox, smallpox and flu. Diseases from Bacteria :-Tuberculosis (Mycobacterium tuberculosis ), pneumonia





#### **Particulate matter :-**

Air pollutants can be in the form of particulate matter which can be very harmful to our health. The level of effect usually depends on the length of time of exposure, as well as the kind and concentration of chemical particles exposed to short term effects includes irritation to the eyes, nose and so on... Long term heath effect can includes lung cancer, heart diseases and even damage to the brain, liver.

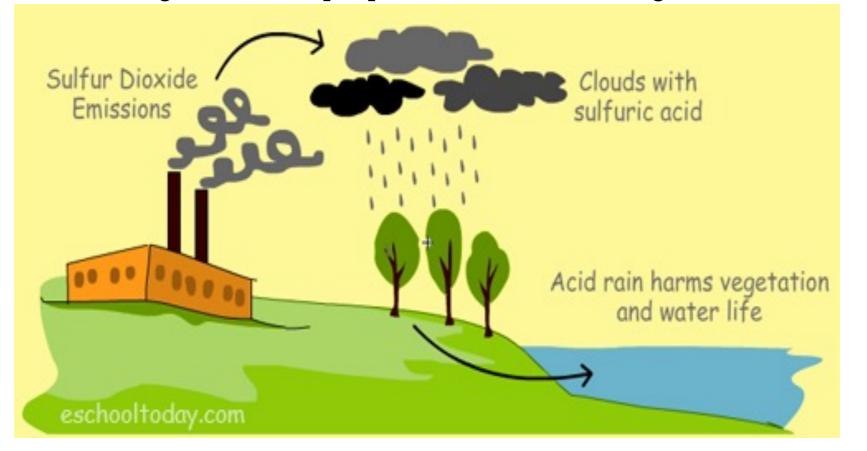




#### **Ground level Ozone :-**

Chemicals reactions involving air pollutants create a toxic gas ozone.

Ozone gas can effect people health and can damage



#### **Extreme Microbial Environments**

>The microbes living in extreme conditions are called extremophiles.

>This literally means that they love the extreme conditions of their habitat.

>The extremophiles are so well adapted to their own environment.

➤Some like the ones in hot springs need extreme temperatures to grow.



#### **Microbial Habitats in Other Organisms**

 $\succ$  Microbes also live on other organisms.

> As with the ones found on people these microbes can be harmful or beneficial to the host.

Example:

> Bacteria grow in nodules on the roots of pea and bean plants. These microbes convert nitrogen from the air into a form that the plants can use.

>In many ways animals and plants have evolved as habitats for the millions of microbes that call them home.



# SYMBIOSIS RELATED TO ENVIRONMENTAL MICROBIOLOGY

#### **SYMBIOSIS**

The relationship in which one organism may depend on another for its survival. Sometimes they need each other. This is called symbiosis.

### FOR EXAMPLE:-

Photosynthetic plants and microbes provide oxygen that humans need to live.

#### **Another Example:-**

Trees offer shelter to other plants and animals.

And many rely on other creatures as sources of food or nutrients



#### **SYMBIOTIC MICROBES**

Often, especially with microbes, one organism lives inside another — the host.

Microbial symbiosis occurs between two microbes. Microbes, however, form associations with other types of organisms, including plants and animals.

Bacteria have a long history of symbiotic relationships, and have evolved in conjunction with their hosts. Other microbes, such as fungi and protists, also form symbiotic relationships with other organisms.

**HOST**- usually the LARGER member **SYMBIONT-** usually the SMALLER member



#### TYPES

**MUTUALISM: (+,+)** In this type of relationship, both partners benefit **Example;** 

E. coli synthesizes vitamin K in the intestine in exchange the large intestine provides nutrients necessary for survival of the microorganisms.

**COMMENSALISM: (+,0)** one organism is benefited and the other is unaffected by this type of relationship. They bring no benefit to the host and yet the microorganisms benefit greatly from the environment they inhabit. **Example;** Staphylococcus on skin

**PARASITISM: (+,-)** one organism benefits at the expense of the other all pathogens are parasites.

**EXAMPLE:-** Its benefits by extracting blood from human host.

#### **BACTERIAL SYMBIOSIS**

Bacteria form symbiotic relationships with many organisms, including humans.

One example is the bacteria that live inside the human digestive system.

These microbes break down food and produce vitamins that humans need. In return, the bacteria benefit from the stable environment inside the intestines.

Bacteria also colonize human skin. The bacteria obtain nutrients from the surface of the skin, while providing people with protection against more dangerous microbes.



### **FUNGI AND PLANT**

Fungi and plants form mutuallybeneficial relationships called mycorrhizal associations.

The fungi increase the absorption of water and nutrients by the plants, and benefit from the compounds produced by the plants during photosynthesis.

The fungus also protects the roots from diseases. Some fungi form extensive networks beneath the ground, and have been known to transport nutrients between plants and trees in different locations.





#### LICHENS

In this mutually-beneficial relationship, the fungus forms the body of the lichen the thallus. This structure attaches to the surface of a rock or tree.

The fungal cells absorb water and nutrients from the surrounding environment. Algal cells grow inside the cells of the fungus. The algal cells convert sunlight to chemical energy through photosynthesis.

This process benefits the fungus. In return, the algal cells are protected from the environment.



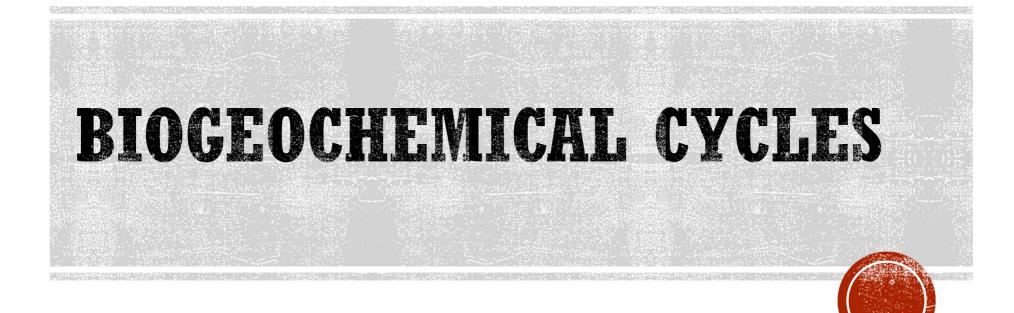


### PROTISTS

An even better-known example of symbiotic protists are the ones that live in the guts of termites. These microbes break down cellulose in the wood particles that termites eat. This enables the termite to obtain nutrition from the wood. Without the help of the protists, the termite would not be able to digest the wood. In this case, the protist is called an **endosymbiont**, which means it lives inside its host, the termite.

#### **Examples of animal-microbe symbiosis**:

Ruminants (such as sheeps and cows) and the bacteria in the rumen.



# What is Biogeochemical Cycles?

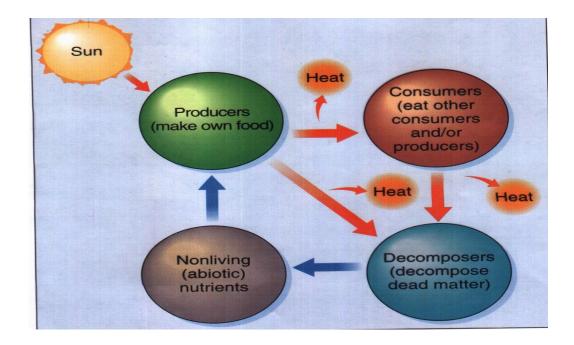
# **Biogeochemical Cycles or Nutrient cycles:**

Is how elements, chemical compounds, and other forms of matter are passed from one organism to another and from one part of the biosphere to another.

### **Types of Biogeochemical Cycle:**

Atmospheric- carbon cycle and nitrogen cycle

Sedimentary - phosphorus cycle and Sulphur cycle

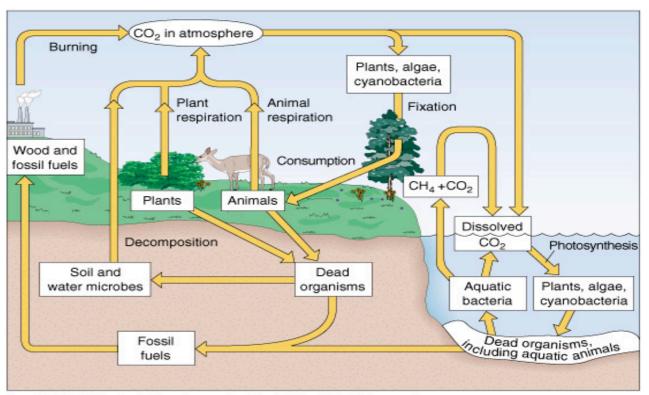


#### The carbon cycle

Carbon is a key ingredient of living tissue. In the atmosphere, carbon is present as carbon dioxide gas, CO2. Carbon dioxide is released into the atmosphere by

- volcanic activity
- respiration
- Human activities

the Plants take in carbon dioxide and use the carbon to build carbohydrates during photosynthesis. The carbohydrates are passed along food webs to animals and other consumers. In the ocean, carbon is also found, along with calcium and oxygen, in calcium carbonate, which is formed by many marine organism decomposition of organic matter.



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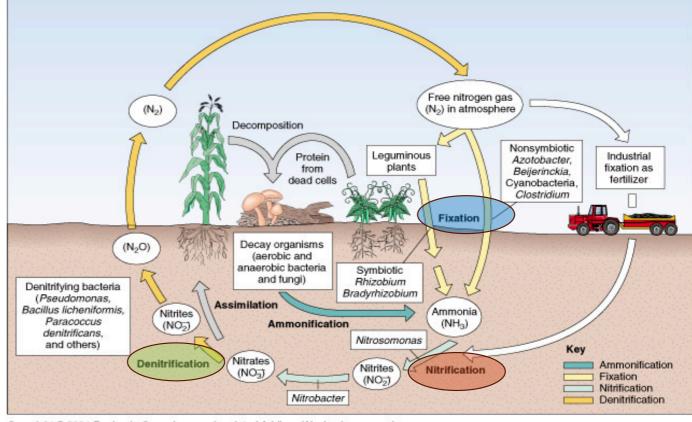


#### The nitrogen cycle

Microbes decompose proteins form dead cells and release amino acids. Ammonia is liberated by microbial ammonification of amino acids.

- Ammonia is oxidized to produce nitrates for energy by nitrifying bacteria.
- Denitrifying bacteria reduce nitrogen in nitrates to molecular nitrogen.
- N<sub>2</sub> is converted into ammonia by nitrogen fixing bacteria

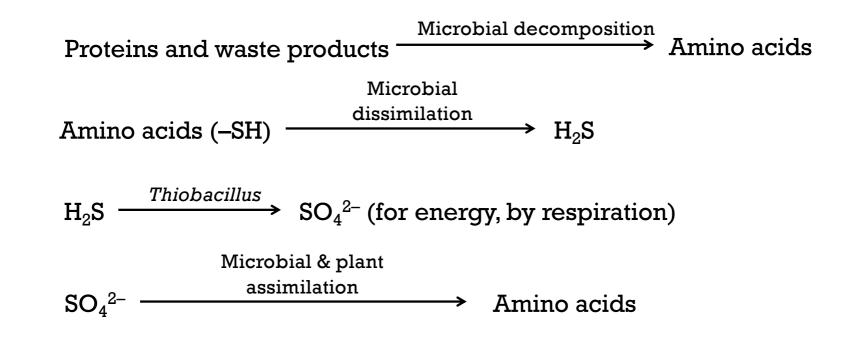
- Ammonium and nitrate are used by bacteria and plants to synthesize amino acids.



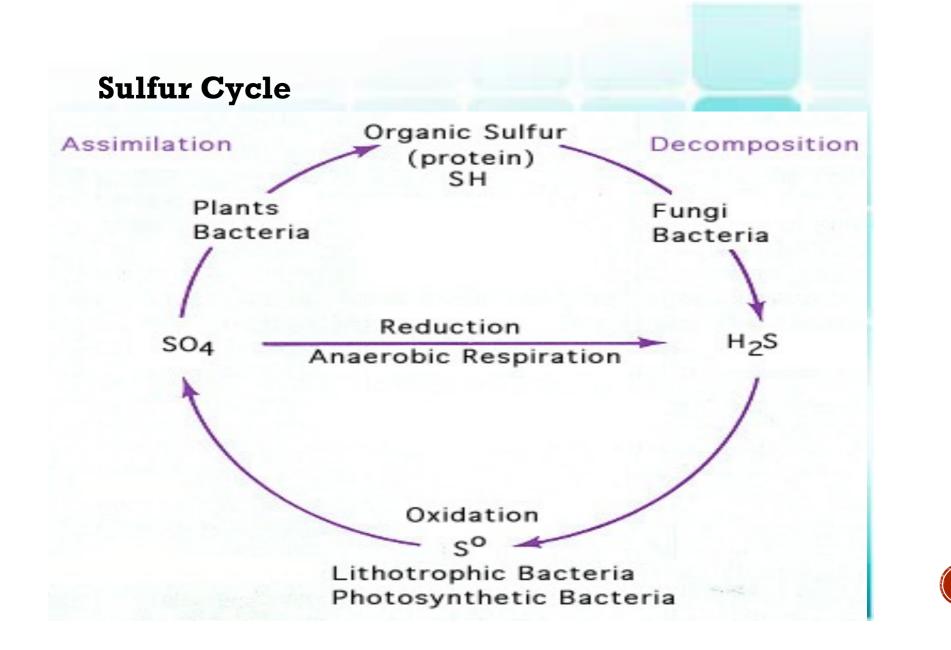
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### SULFUR CYCLE

Plants and certain microbes can use  $SO_4^{2-}$  to make amino acids.  $H_2S$  is oxidized to form  $SO_4^{2-}$ 

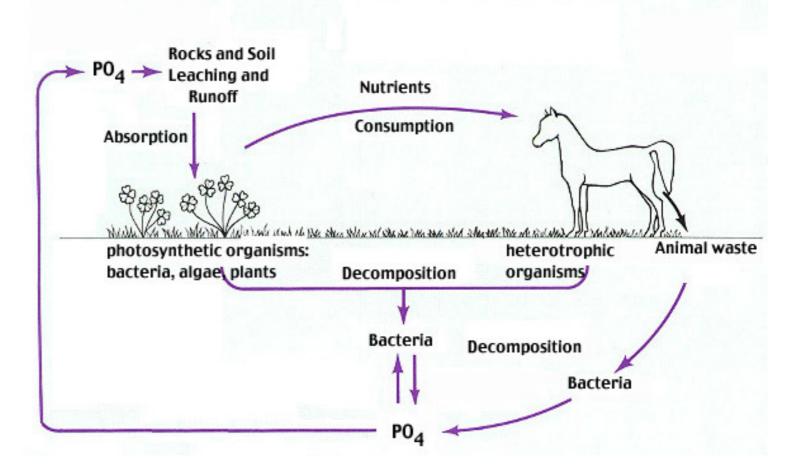






### The Phosphorus Cycle

Inorganic phosphorus is solubilized by microbial acids. Made available to plants and other microbes Is soluble in water Combines with calcium in calcium phosphate .





### LIFE WITHOUT SUNSHINE

- Primary producers in most ecosystems are photoautotroph's.
- Primary producers in deep ocean and endolithic communities are chemoautotrophic bacteria.

$$H_2S \longrightarrow SO_4^{2-}$$

Provides energy for bacteria which may be used to fix  $CO_2$ 

 $CO_2 \xrightarrow{\text{Calvin Cycle}} Sugars$ 

Provides carbon for cell growth



### **Oxygenic photosynthesis**

Algae, cyanobacteria, aerobic heterotrophs

 $\begin{array}{l} \bullet \operatorname{CO}_2 + \operatorname{H}_2\operatorname{O} \to \operatorname{CH}_2\operatorname{O} + \operatorname{O}_2 \\ \\ \operatorname{H}_2\operatorname{O} \text{ is a source of electrons} \\ \bullet \operatorname{CH}_2\operatorname{O} + \operatorname{O}_2 \to \operatorname{CO}_2 + \operatorname{H}_2\operatorname{O} \end{array}$ 

Aerobic respiration

### **AN OXYGENIC PHOTOSYNTHESIS**

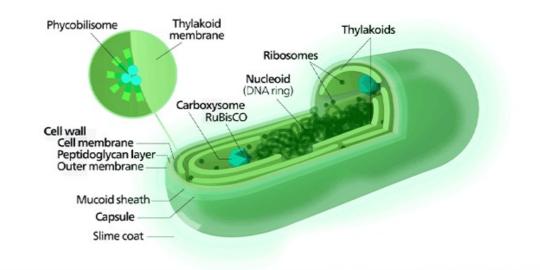
 $\begin{array}{l} {}^{H_2S \text{ oxidizers}} \\ CO_2 + H_2S \rightarrow CH_2O + S + H_2O \\ \\ H_2S \text{ is a source of electrons} \end{array}$ 



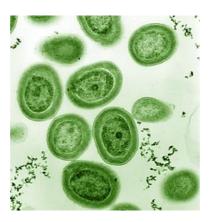
#### Cyanobacteria

- Tremendous ecological importance in the C, O and N cycles.
- Evolutionary relationship to plants.
- Cyanobacteria have chlorophyll *a*, carotenoids and phycobilins.
- Cyanobacteria are very similar to the chloroplasts of red algae (*Rhodophyta*).
- Several species of cyanobacteria are symbionts of liverworts, ferns, cycads, flagellated protozoa, and algae.
- Example: *Prochlorococcus marinus*

### Cyanobacteria



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https://commons.wikimedia.org/wiki/File:Cyanobacterium.svg



# **DEFINITIONS OF BIOREMEDIATION**

 Use of living organisms (e.g., bacteria) to clean up oil spills or remove other pollutants from soil, water, and wastewater."
 Source: United States Environmental Protection Agency, Office of Compliance and Assurance



"clean-up of pollution from soil, groundwater, surface water and air, using biological, usually microbiological processes" Source: Philip et al., 2001

## Introduction

Intensification of agriculture and manufacturing industries has resulted in increased release of a wide range of xenobiotic compounds to the environment.

•Excess loading of hazardous waste has led to scarcity of clean water and disturbances of soil thus limiting crop production.

•Environment is formed by the combination of air , soil, water in the oceans, lakes etc.

Here we discus bioremediation, which clean the environment from pollution.Trees play an important role in the environment.

They may pollute or clean the environment.

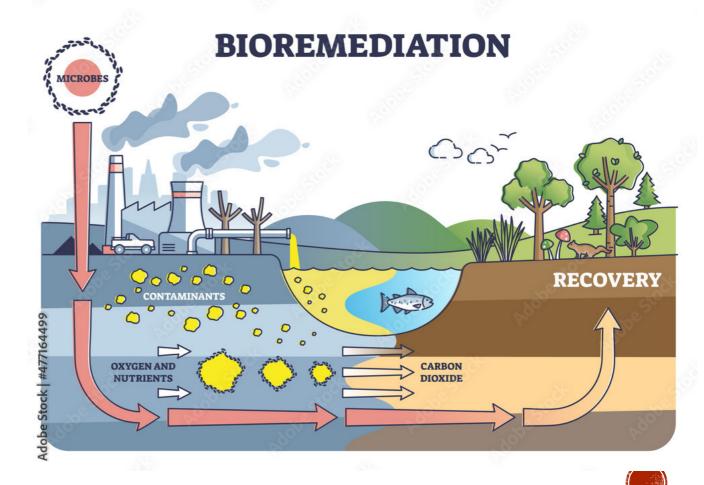


# PRINCIPLES OF BIOREMEDIATION

 Microorganisms use their enzymatic activity for the destruction of pollutants or their transformation to less harmful forms.

Their enzymatic action on the contaminants of environment, break them into digestible form and there microbes get energy by performing normal metabolism.

Here augmentation also involves.



# GENERALLY

Bioremediation can be defined as any process that uses microorganisms or their enzymes to return the environment altered by contaminants to its original condition.

Microbes play its role by the activities of their enzymes, which helps in the destruction of pollutants or their transformation to less harmful forms.
Microorganisms are very important in bioremediation because they have extraordinary metabolic diversity.

## **AGENTS IN BIOREMEDIATION**

In bioremediation, microorganisms are used to destroy or immobilize waste materials.

- Bacteria
- Fungi
- Algae
- Actinomycetes
- (filamentous bacteria)



# **Fundamentals of cleanup reaction**

•Microbes can convert many chemicals into harmless compounds using cleanup reaction.

Aerobic or anaerobically.

Both involve oxidation and reduction reactions.

•Oxidation involves the removal of one or more electrons.

Reduction involves the addition of one or more electrons.

•Oxidizing agents gain electrons and reducing agents lose electron.

•When both reaction occurs at a time, it will be redox reaction.



# What Types of Compounds Can Be Treated Biologically?

#### **Petroleum Hydrocarbons**

Gasoline
Diesel Fuel
Gasoline Additives such as MTBE
Polyaromatic Hydrocarbons
Creosote Chlorinated
Hydrocarbons
Chlorinated Aliphatics: trichloroethylene
Chlorinated Aromatics : PCB's, Pentachlorophenol

#### **Explosives**

RDX, TNT

Inorganic via Reduction to a Lower Valence Causing **Precipitation** 

Uranium, Technicium
Sulfur and Sulfuric Acid

Ammonia or Nitrate/Nitrite





## **How Microbes Use the Contaminant**

#### Primary substrate

enough available to be the sole energy source

#### Secondary substrate

provides energy, not available in high enough concentration

#### Co metabolic substrate

fortuitous transformation of a compound by a microbe relying on some other primary substrate



# **Methods of bioremediation**

On the basis of removal and transportation of wastes for treatment there are basically two methods

## Ex-situ (off-site) strategies

- Excavation/pumping, followed by...
- Landfill
- Incineration
- Chemical removal

## In-situ (on-site) strategies

Immobilization (chemicals, wells, membranes)



# **Advantages of Bioremediation**

Can be highly specific

Less expensive than excavation or incineration processes

If mineralization occurs get complete degradation and clean up

Does not transfer contaminants from one environment to another

- Uses a natural process
- Good public acceptance
- Process is simple

If using ISB you will treat the groundwater and soil at the same time



#### **Disadvantages to Bioremediation**

- Not instantaneous.
- •Often need to develop a system.
- Always need to test and optimize conditions empirically not with computer models.
- May have inhibitors present.
- Compounds may not be in a biodegradable form polymers, plastics.

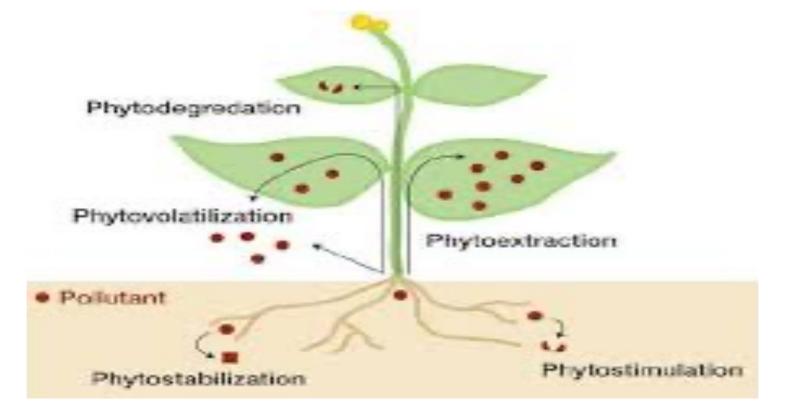
#### Conclusion

- •We should clean the environment including land ,soil, water etc by using biological methods in which bioremediation is very suitable.
- •In bioremediation, microbes play important role by their enzymatic activity.
- They do normal metabolism and destroy the pollutants.
- •We can purify the wastewater, polluted soil, do better crop production, better variety of fruits.
- In this way it is economically important method.



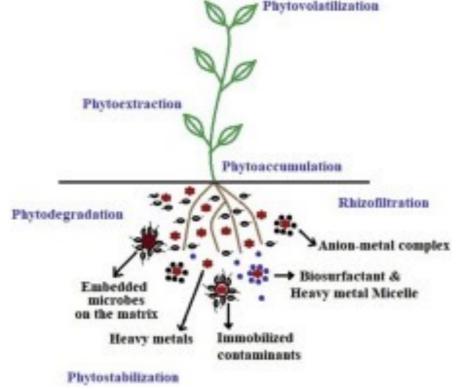
## **TYPES OF BIOREMEDIATION**

**Phytoremediation** describes the treatment of environmental problems through the use of plants that mitigate the environmental problem without the need to excavate the contaminant material and dispose of it elsewhere.



# RHIZOREMEDIATION

**Rhizoremediation,**which is the most evolved process of bioremediation, involves the removal of specific contaminants from contaminated sites by mutual interaction of plant roots and suitable microbial flora.

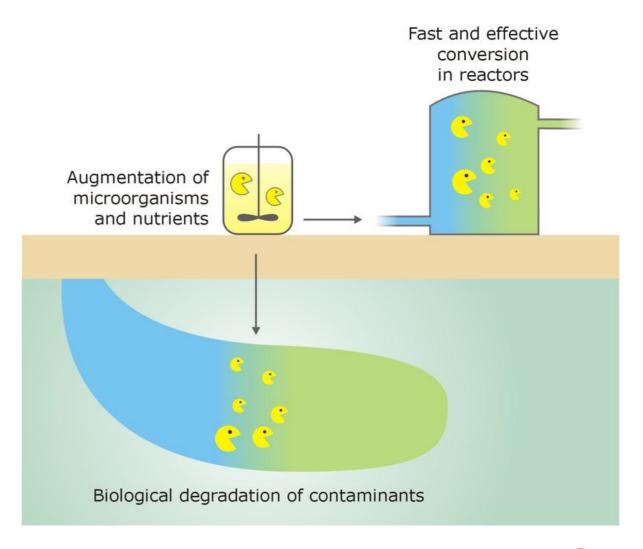




# BIOAUGMENTATION

 Contaminant compounds are transformed by living organisms through reactions that take place as a part of their metabolic processes.

Biodegradation of а compound is often a result of the actions of multiple organisms. When microorganisms are imported to a contaminated site to enhance degradation we have known process a as bioaugmentation.





# Sewage

"Waste matter from domestic or industrial establishments that is carried away in sewers or drains for dumping or conversion into a form that is not toxic".

Sewage is mostly water and contains little particulate matter, perhaps only 0.03%.

Even so in large cities, the solid portion of sewage can total more than 1000 tons of solid material per day.

#### **Dissolved oxygen (DO)** Oxygen concentration in water **Chemical oxygen demand (COD)**

Oxygen is removed from water by Chemical.

#### **Biochemical oxygen demand (BOD)**

Oxygen is removed from water when organic matter is consumed by bacteria.

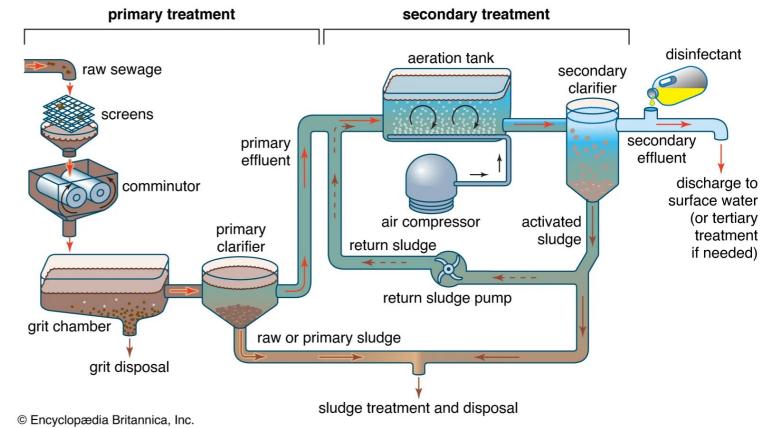
Low oxygen conditions may kill fish and other organisms.



# **Types of Treatment:**

Comprises of:

Primary Treatment.
Secondary Treatment.
Tertiary Treatment.



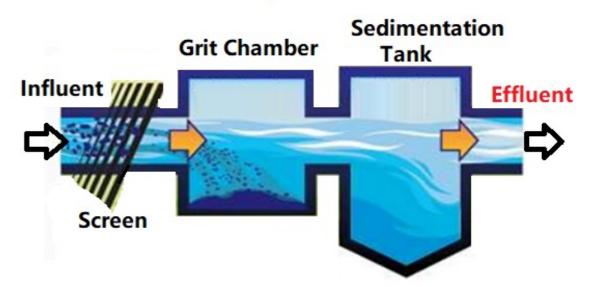


#### **Primary Treatment:**

"Physical process designed to remove materials that settle out of liquid".

- Treatment removes nearly 50% of solids and 25% of BOD
- Sewage passes through series of screens to remove large objects.
- **Skimmers** remove scum and floating debris.
- Sewage allowed to settle in sedimentation tank which facilitates
- the removal of solids.
- Sludge is removed after settling is complete.Primary sewage that remains is sent for secondary treatment.

#### **Primary Treatment**



### Secondary Treatment(chifely biological process)

Designed to convert suspended solids to inorganic compounds and cell mass for removal. This treatment eliminates atleast 95% of BOD.

Microbial growth is encouraged during secondary treatment.

Aerobic organisms degrade organic material to carbon dioxide and water.

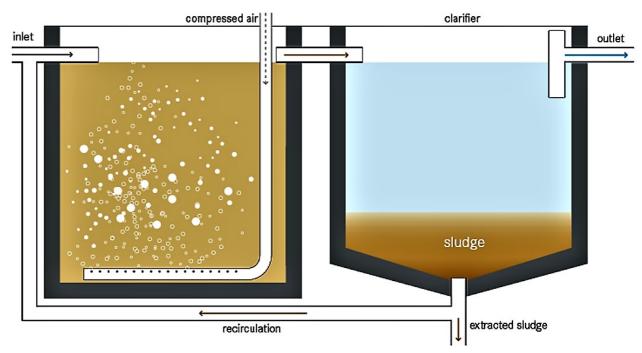
## Methods used in secondary treatment include

Activated sludge
 Anaerobic Digestion of Sludge
 Trickling filter
 Lagoons
 Artificial wetlands



#### **1. Activated Sludge:**

- System employs mixed aerobic microbes, (organisms grow in biofilms called flocs).
- Large number of organisms inoculated into sewage via addition of small amount of leftover sludge.
- Oxygen is supplied and organic matter converted to biomass or waste product
- Sewage is sent to sedimentation where,Flocs settle out and resulting sludge is removed (Portion of sludge is used for next "batch")
- Complication of activated sludge is bulking
- "Interference of filamentous bacteria in sedimentation process."



## **Anaerobic Digestion of Sludge**

Sludges from the primary and secondary treatment settling tanks are pumped into an anerobic digester.

Sludges contain cellulose, proteins, lipid and other insoluble polymers.

Anaerobic bacteria digest the sludge to methane and carbon dioxide.

#### **Types of treatment systems include:**

Septic Tanks or Wastewater Treatment Plants (WWTPs).

**1.Septic Tanks** typically treat small volumes of waste (e.g., from a single household, small commercial/industral).

2.WWTPs typically treat larger volumes of municipal or industrial waste.



#### Pathogen Removal by Activated Sludge

More than 90% of *E.coli*. and Salmonella are destroyed.

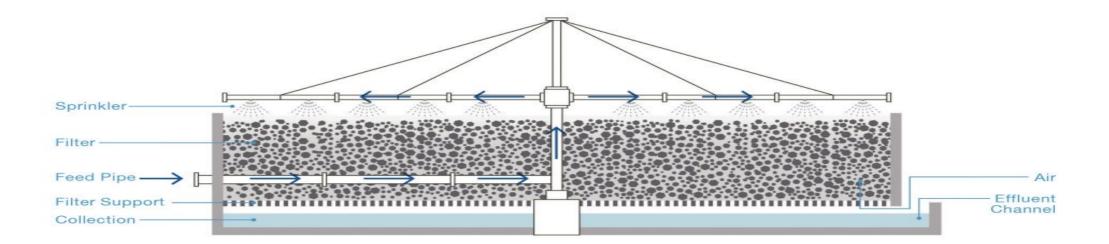
Bacteria are removed by inactivation, grazing by ciliated protozoa, and adsorption to sludge solids.

Viruses are removed mainly by adsorption process.



### **2. Trickling filters:**

- Frequently used in smaller treatment plants.
- Rotating arm sprays sewage over bed of course gravel and rocks.
- Rocks and gravel become coated with biofilms(Consists of heterogeneous mix of bacteria, fungi, algae, protozoa and nematodes).
- Organisms aerobically degrade organic material.

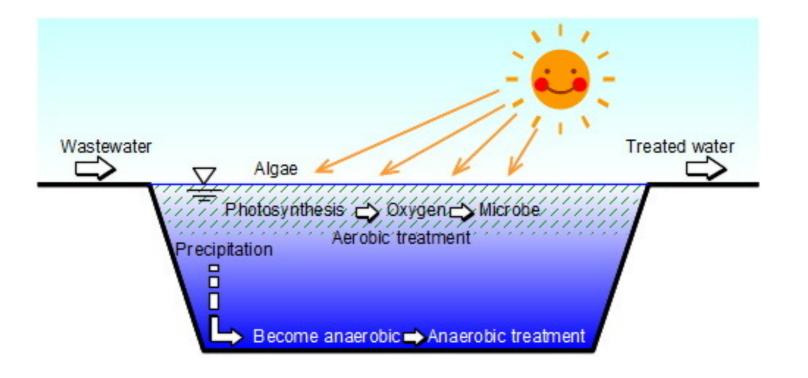


#### 3. Lagoons:

Sewage channeled into shallow ponds or lagoons •Remains there for a period.

Algae and cyanobacteria grow on surface to provide oxygen

•They enable aerobic organisms to degrade sewage.

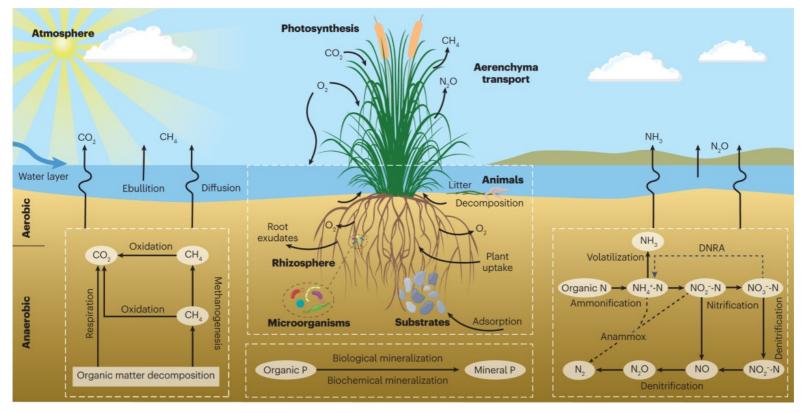


## 4. Artificial wetlands:

Employ same principles as lagoons,has a more advanced design.

Offers means to treat wastewater.

Also provides habitat for birds and other wildlife.





# **Tertiary Treatment** (Advanced Treatment)

Expensive treatment.

Designed to remove specific substance **Ammonia** is removed through ammonia stripping, liberates gaseous ammonia from water.

Nitrates removed using denitrifying bacteria, reduces nitrates to nitrogen gas. Phosphates are removed through chemical precipitation. Pharmaceutical residues

Phenol residues Pesticide





#### **Uses of treated water**

1.Treated water is discharged to waterways
 2.Used for irrigation
 3.Recycled into drinking water

#### Disinfection

Before discharge effluent is disinfected with one or a combination of the following:

- •Chlorine.
- •Ozone.
- •Ultra violet light.

These treatments decrease the numbers of microorganisms and viruses.





## What are biofilms?

- Biofilms are thin usually resistant layer of microorganism(As bacteria, algae, protozoa) that form on and coat various surface.
- $\hfill\square$  These are visible .
- □ It can be in form of single bacerial species.

# **Bio films consist of**

- Bacteria , Fungi, Algae.
- □ Extracellular polymers.
- □ Trapped organic molecule.
- □ Inorganic molecules.



# Where do biofilms form?

Biofilms will form any where there is moisture .
Biofilms happily colonize many household in the bath , kitchen , including toilets , sinks , cutting boards..
Bio films can grow on metals, plastic, tissue, teeth and on soil particles.



# **BIO FILM**



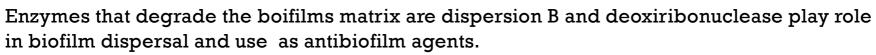


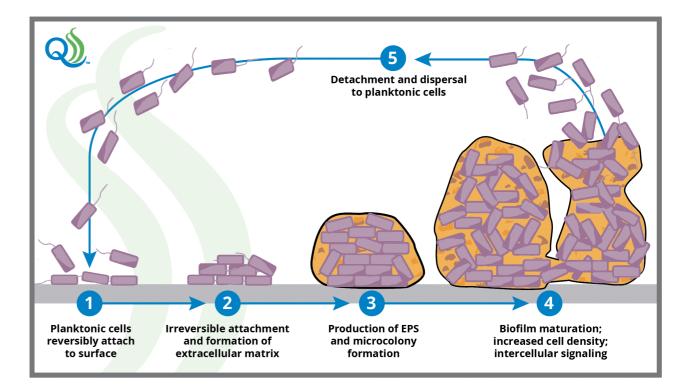




#### How biofilms form?

- 1. Free swimming bacterial cells land on a surface , arrange themselves in cluster , attach.
- 2. The cells begin producing a matrix.
- 3. The cells signal one another to multiply and form a micro colony by EPS.
- 4. The microbial colony promote the coexistence of diverse bacterial species.
- Last step is dispersal.
   Dispersal enable biofilms to spread and colonize new surface.





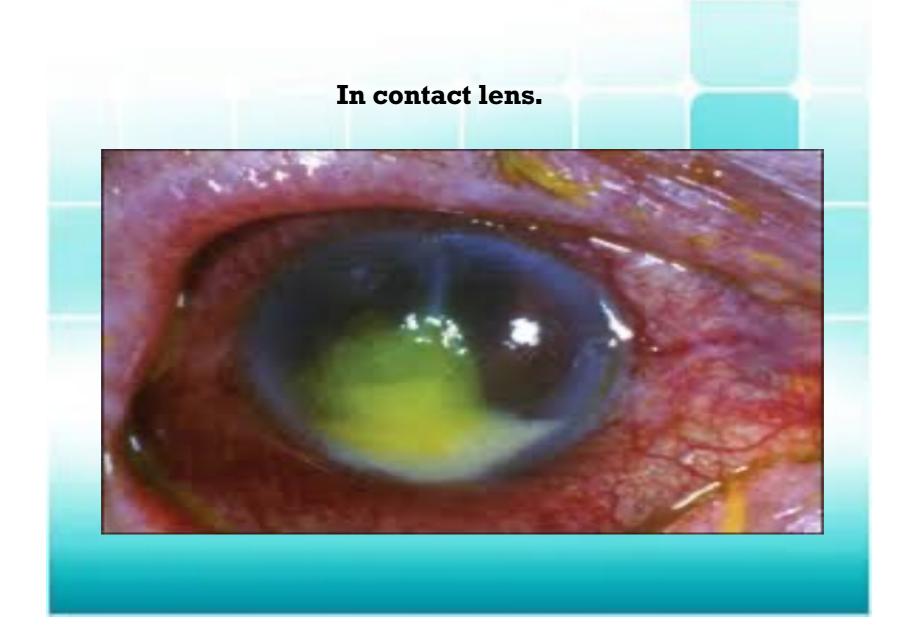
#### **Bio film Infections..**

- Biofilms can cause infection in dental cavity, gum disease, as well as ear, heart and lungs infection.
- Biofilm can also form on contact lenses.
- Biofilms on inner surface of medical equipment cause hospital infections.

#### **Biofilm in teeth**







Who to prevent biofilm?

#### Furanose

can be use to prevent biofilms in agriculture and in marine.
Bound to polymer coating.
In medical devices such as catheter.

Use herbal treatment.

# **Prevention**

Use antibodies , biocides , vibrations that inhibit attachment.

 chose that chemicals in industry like chlorine , hydrogen peroxide, that inhibit attachment .
 Alter the growth conditions of growing biofilms.
 Use mechanical cleaning(high pressure flushing).

