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SCHOOL OF BIO AND CHEMICAL ENGINEERING

DEPARTMENT OF BIOTECHNOLOGY

UNIT – I– Environmental & Agricultural Biotechnology – SBB2201

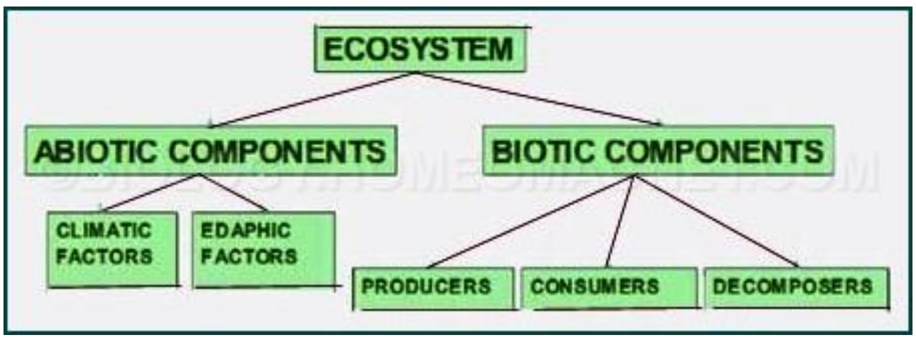
BASIC CONCEPTS OF ECOSYSTEM

The term Ecosystem was first proposed by A.G. Tansley in 1935. he defined it as "the system resulting from the interaction of all the living and non living factors of the Environment.

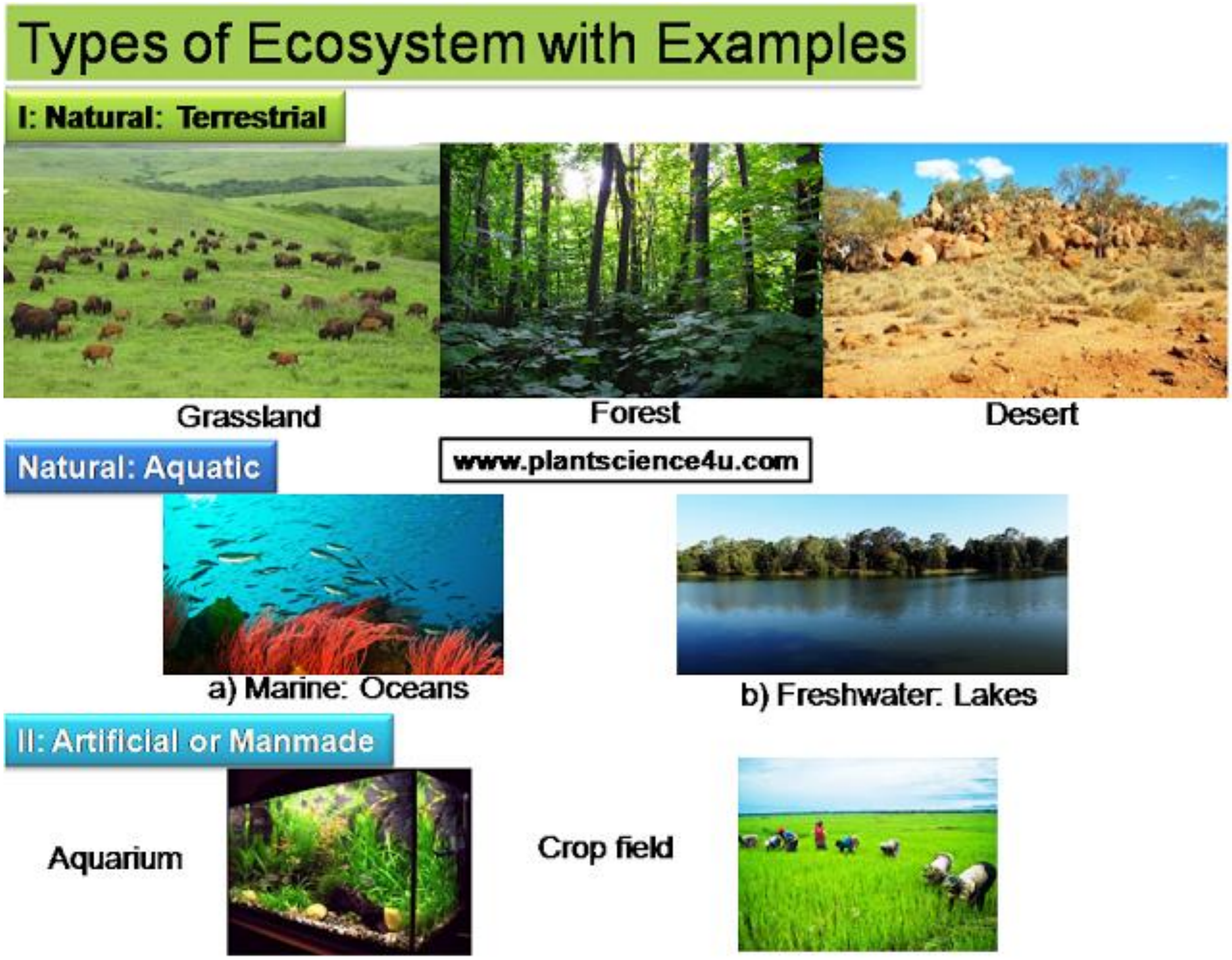
• An ecosystem consists of the biological community that occurs in some locale, and the physical and chemical factors that make up its non-living or abiotic environment. There are many examples of ecosystems

- a pond, a forest, an estuary, a grassland.

STRUCTURE OF ECOSYSTEM



TYPES OF ECOSYSTEM

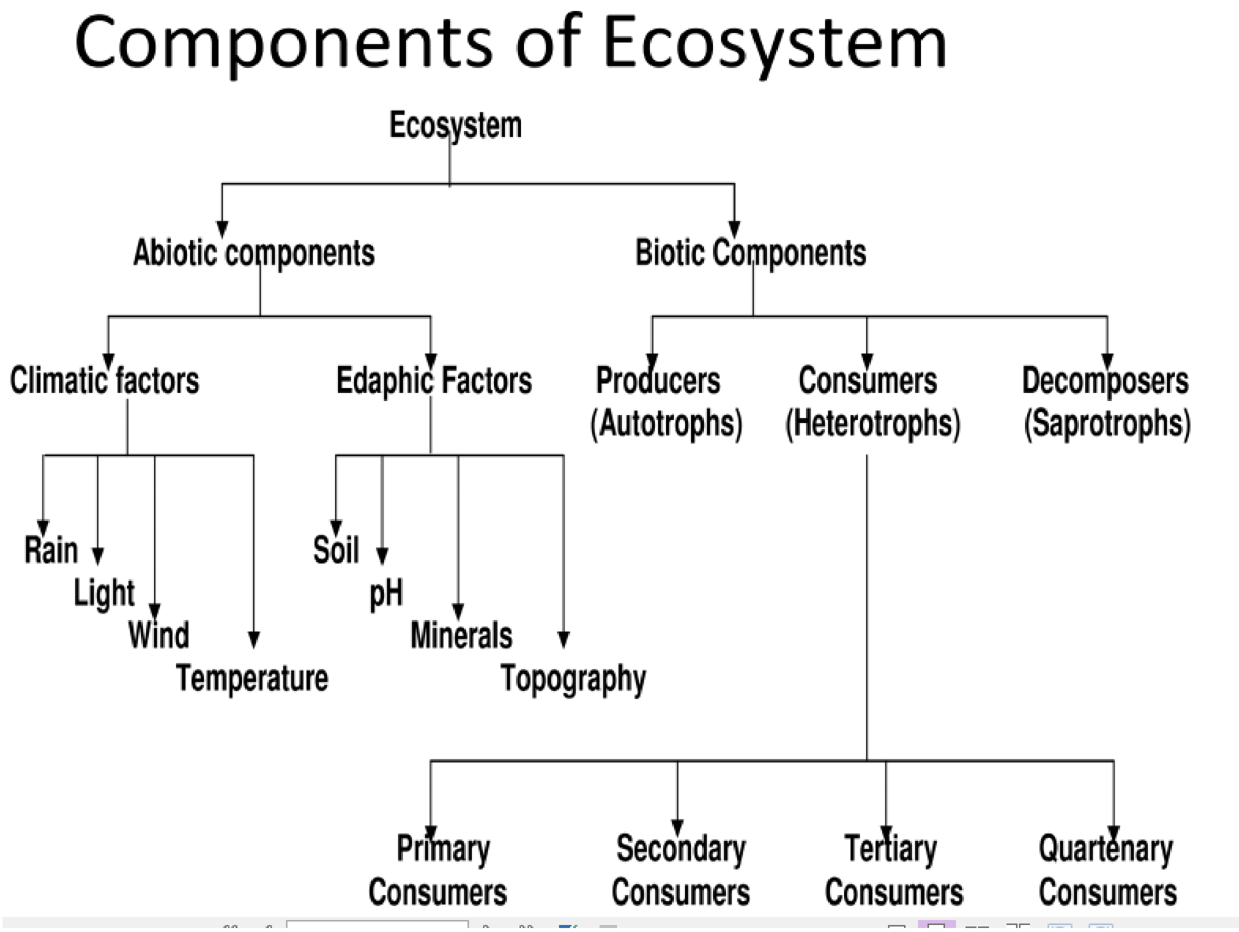


1. Natural Ecosystems : These operate under natural conditions without any major interference by man.

i. Terrestrial Ecosystem : Forest, grassland, desert, etc.

ii. Aquatic Ecosystem : a. Fresh water : Lotic (running water like spring, stream, or rivers) or Lentic (standing water as lake, pond, pools, etc.)

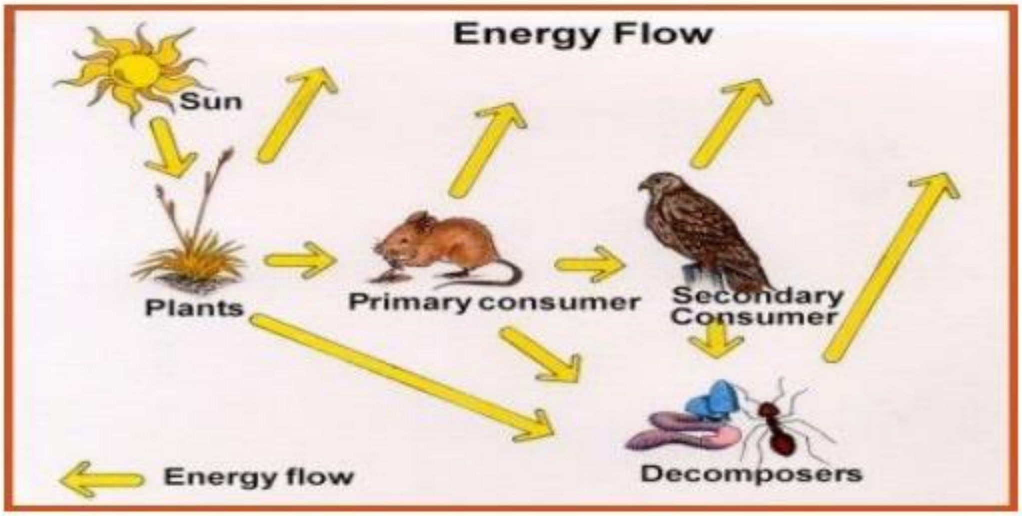
b. Marine water : Such as deep bodies as ocean or shallow ones as Sea or an estuary.



Cybernectics: Cybernetics is a transdisciplinary approach for exploring regulatory systems—their structures, constraints, and possibilities. Norbert Wiener defined cybernetics in 1948 as "the scientific study of control and communication in the animal and the machine"

Homeostatis: homeostasis is the state of steady internal, physical, and chemical conditions maintained by living systems. This is the condition of optimal functioning for the organism and includes many variables, such as body temperature and fluid balance, being kept within certain pre-set limits (homeostatic range). Other variables include the pH of extracellular fluid, the concentrations of sodium, potassium and calcium ions, as well as that of the blood sugar level, and these need to be regulated despite changes in the environment, diet, or level of activity. Each of these variables is controlled by one or more regulators or homeostatic mechanisms, which together maintain life.

Energy transfer in a ecosystem

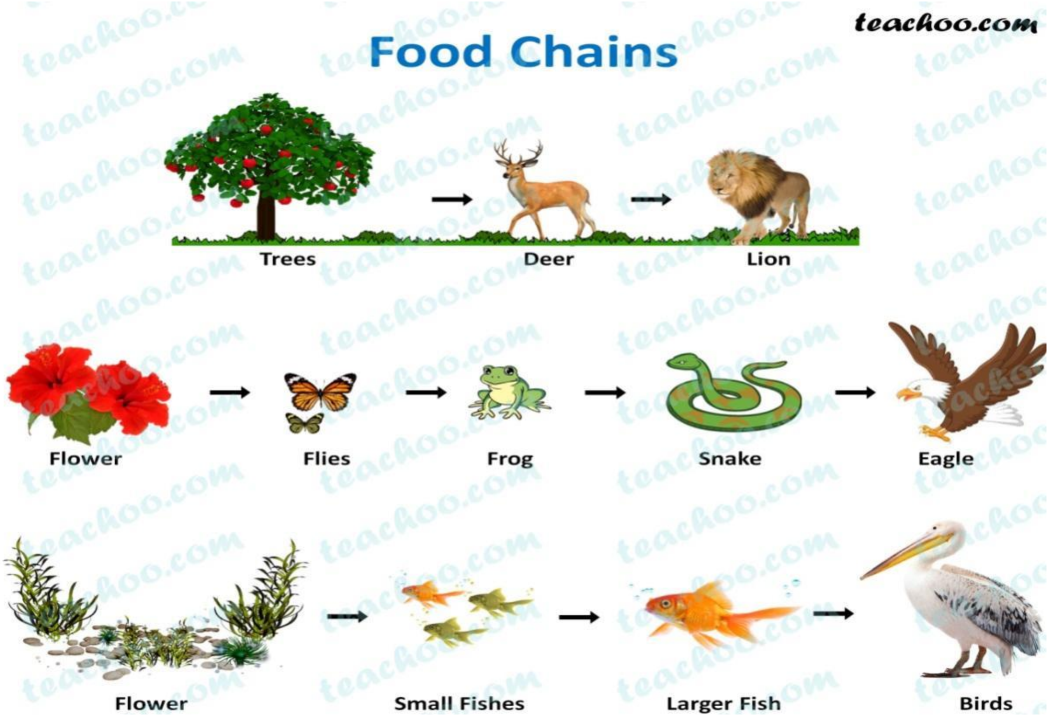


Energy transfer is a one-way process. Once potential energy has been released in some form from its storage in biomass, it cannot all be reused, recycled, or converted to waste heat. This means that if the sun, the ultimate energy source of ecosystems, were to stop shining, life as humans know it would soon end. Fortunately, every day, the sun provides new energy in the form of protons to sustain the food webs of Earth. n 1927, British ecologist Charles Sutherland Elton (1900–1991) wrote that most food webs have a similar pyramidal shape. At the bottom, there are many photosynthetic organisms that collectively have a large biomass and productivity. On each of the following trophic levels, or feeding levels, there are successively fewer heterotrophic organisms, with a smaller productivity. The pyramid of biomass and productivity is now known as the Eltonian pyramid.

Food chain and food web

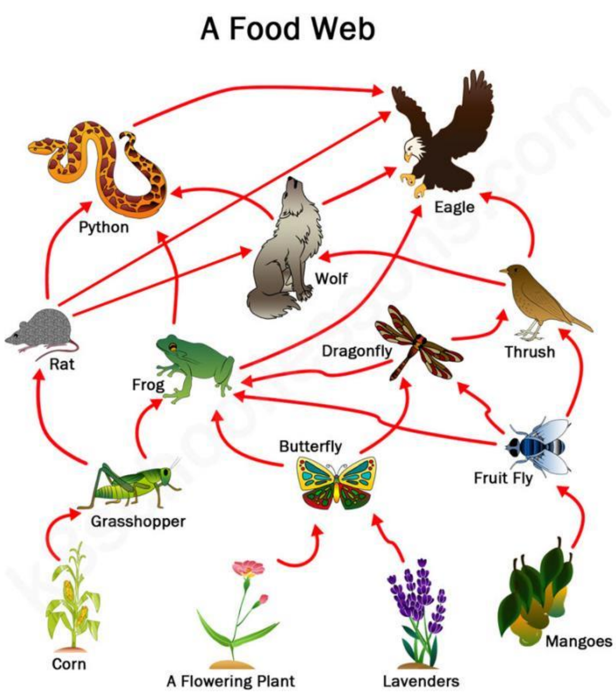
Every organism needs to obtain energy in order to live. For example, plants get energy from the sun, some animals eat plants, and some animals eat other animals.

Food chain



A food chain is the sequence of who eats whom in a biological community (an ecosystem) to obtain nutrition

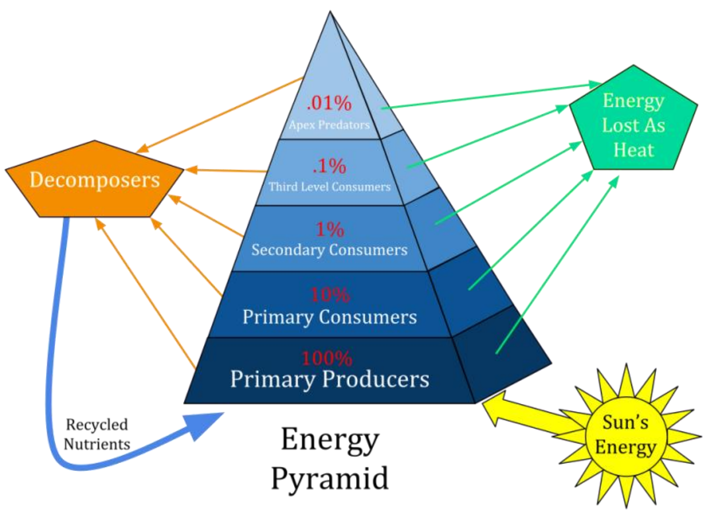
Food Web



A food web(or food cycle) depicts feeding connections (what-eats-what)
in an ecological community and hence is also referred to as a consumerresource system.The food web is a simplified illustration of the various methods of feeding that links an ecosystem into a

unified system of exchange.Various food chains are often interlinked at different tropic levels to form a complex interaction between different species from the point of view of food. Food Web provides more than one alternatives of food to most of the organisms in an ecosystem and thus increases their chances of survival.

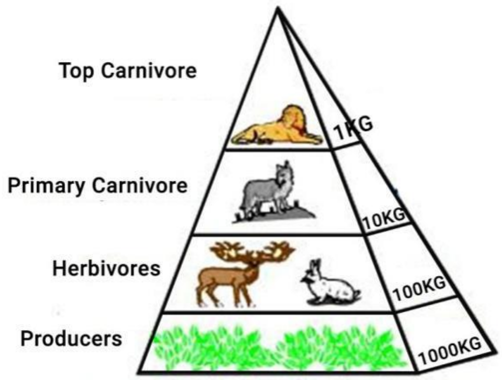
Energy pyramid



Graphic representation of trophic structure & functioning of ecosystem starting with producers at the base & successive trophic levels forming the apex.

All organisms in an ecosystem can be placed in trophic levels depending on what energy source they rely upon and how they provide energy for other organisms in the food web. With the exception of life near hydrothermal vents in the deepbocean, life is always dependent directly or indirectly on the energy from the sun. In everyecosystem, there is an organism at the lowest level that converts energy from the sun into useable energy for other organisms.

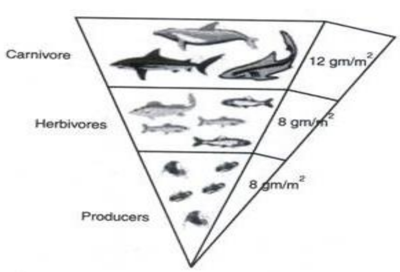
Pyramid Of Numbers



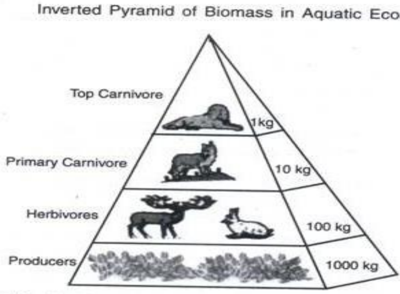
Upright Pyamid of Biomass in aTerrestrial Ecosystem

A pyramid of numbers is a graphical representation of the numbers of individuals in each population in a food chain. Often it is drawn from the autotrophic level up. A pyramid of numbers can be used to examine how the population of a certain species affects another. Often, the autotrophic level in a pyramid of numbers is much larger than any of the higher trophic levels, and the numbers decreases upon ascending the pyramid. There are exceptions, however. For example, in a tree community, a single tree could support many different populations of larger numbers.

Pyramid of Biomass



Inverted Pyramid of Biomass in Aquatic Ecosystem



Upright Pyramid of Biomass for Grassland Ecosystem

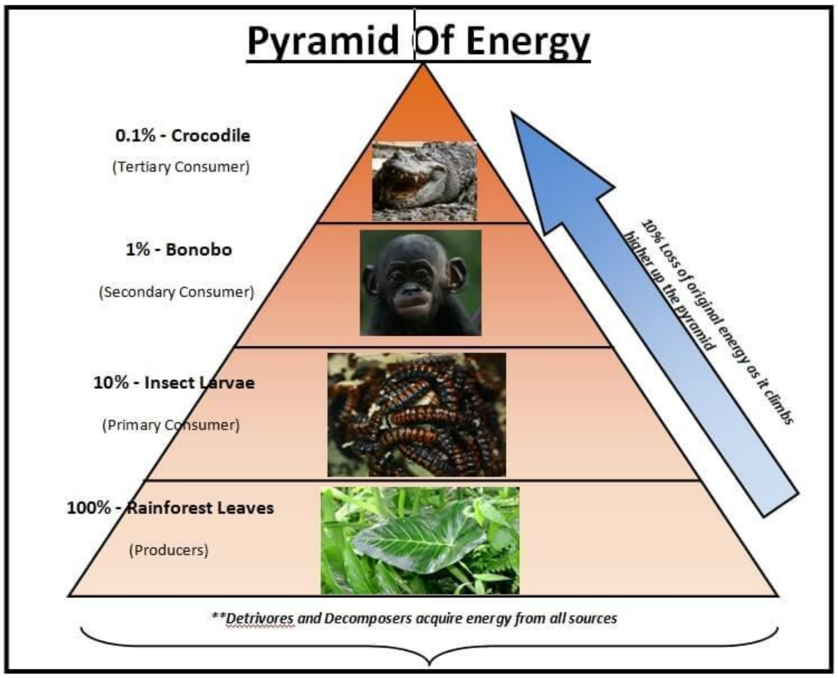
Illustrates the amount of biomass in each trophic level

– Biomass weight is determined after dehydration

• Shows the amount of matterlost between trophic levels.

• Measured in Kg, grams or pounds

Pyramid of Energy



Shows the energy availableat each trophic level.

– The sizeof the blocks represents the proportion of productivity

– Measured in Joules or Calories

Industrial waste water treatment-primary,secondary and tertiary

Wastewater treatment consists of applying known technology to improve or upgrade the quality of a wastewater. Usually wastewater treatment will involve collecting the wastewater in a central, segregated location (the Wastewater Treatment Plant) and subjecting the wastewater to various treatment processes. Most often, since large volumes of wastewater are involved, treatment processes are carried out on continuously flowing wastewaters (continuous flow or "open" systems) rather than as "batch" or a series of periodic treatment processes in which treatment is carried out on parcels or "batches" of wastewaters. While most wastewater treatment processes are continuous flow, certain operations, such as vacuum filtration, involving as it does, storage of sludge, the addition of chemicals, filtration and removal or disposal of the treated sludge, are routinely handled as periodic batch operations.

Wastewater treatment, however, can also be organized or categorized by the nature of the treatment process operation being used; for example, physical, chemical or biological. Examples of these treatment steps are shown below. A complete treatment system may consist of the application of a number of physical, chemical and biological processes to the wastewater.

Some Physical, Chemical and Biological Wastewater Treatment Methods

Physical

- Sedimentation (Clarification)
- Screening

- Aeration
- Filtration
- Flotation and Skimming
- Degassification
- Equalization

Chemical

- Chlorination
- Ozonation
- Neutralization
- Coagulation
- Adsorption
- Ion Exchange

Biological

- **Aerobic**

  Activated Sludge Treatment Methods

  Trickling Filtration

  Oxidation Ponds

  Lagoons

  Aerobic Digestion
- *Anaerobic*

  Anaerobic Digestion

  Septic Tanks

  Lagoons

Physical methods

Include processes where no gross chemical or biological changes are carried out and strictly physical phenomena are used to improve or treat the wastewater.

sedimentation (or clarification): In the process of sedimentation, physical phenomena relating to the settling of solids by gravity are allowed to operate. Usually this consists of simply holding a wastewater for a short period of time in a tank under quiescent conditions, allowing the heavier solids to settle, and removing the "clarified" effluent. Sedimentation for solids separation is a very common process operation and is routinely employed at the beginning and end of wastewater treatment operations.

Aeration: Physically adding air, usually to provide oxygen to the wastewater.

Filtration: Here wastewater is passed through a filter medium to separate solids. An example would be the use of sand filters to further remove entrained solids from a treated wastewater. Certain phenomena will occur during the sedimentation process and can be advantageously used to further improve water quality. Permitting greases or oils, for example, to float to the surface and skimming or physically removing them from the wastewaters is often carried out as part of the overall treatment process.

In certain industrial wastewater treatment processes strong or undesirable wastes are sometimes produced over short periods of time. Since such "slugs" or periodic inputs of such wastes would damage a biological treatment process, these wastes are sometimes held, mixed with other wastewaters, and gradually released, thus eliminating "shocks" to the treatment plant. This is call equalization. Another type of "equalization" can be used to even out wide variations in flow rates. For example, the wet well of a pump station can receive widely varying amounts of wastewater and, in turn, pump the wastes onward at more uniform rates.

Chemical treatment

Consists of using some chemical reaction or reactions to improve the water quality. Probably the most commonly used chemical process is

Chlorination: Chlorine, a strong oxidizing chemical, is used to kill bacteria and to slow down the rate of decomposition of the wastewater. Bacterial kill is achieved when vital biological processes are affected by the chlorine. Another strong oxidizing agent that has also been used as an oxidizing disinfectant is ozone.

Neutralization: Neutralization consists of the addition of acid or base to adjust pH levels back to neutrality. Since lime is a base it is sometimes used in the neutralization of acid wastes.

Coagulation consists of the addition of a chemical that, through a chemical reaction, forms an insoluble end product that serves to remove substances from the wastewater. Polyvalent metals are commonly used as coagulating chemicals in wastewater treatment and typical coagulants would include lime (that can also be used in neutralization), certain iron containing compounds (such as ferric chloride or ferric sulfate) and alum (aluminum sulfate).

Certain processes may actually be physical and chemical in nature. The use of activated carbon to "adsorb" or remove organics, for example, involves both chemical and physical processes. Processes such as ion exchange, which involves exchanging certain ions for others, are not used to any great extent in wastewater treatment.

Biological treatment methods

Use microorganisms, mostly bacteria, in the biochemical decomposition of wastewaters to stable end products. More microorganisms, or sludges, are formed and a portion of the waste is converted to carbon dioxide, water and other end products. Generally, biological treatment methods can be divided into aerobic and anaerobic methods, based on availability of dissolved oxygen.

The purpose of wastewater treatment is generally to remove from the wastewater enough solids to permit the remainder to be discharged to a receiving water without interfering with its best or proper use. The solids which are removed are primarily organic but may also include inorganic solids. Treatment must also be provided for the solids and liquids which are removed as sludge. Finally, treatment to control odors, to retard biological activity, or destroy pathogenic organisms may also be needed.

While the devices used in wastewater treatment are numerous and will probably combine physical, chemical and biological methods, they may all be generally grouped under six methods:

- Preliminary Treatment
- Primary Treatment
- Secondary Treatment
- Disinfection
- Sludge Treatment
- Tertiary Treatment

Degrees of treatment are sometimes indicated by use of the terms primary, secondary and tertiary treatment. Tertiary treatment, properly, would be any treatment added onto or following secondary treatment.

Preliminary Treatment

At most plants preliminary treatment is used to protect pumping equipment and facilitate subsequent treatment processes. Preliminary devices are designed to remove or cut up the larger suspended and floating solids, to remove the heavy inorganic solids, and to remove excessive amounts of oils or greases.

To effect the objectives of preliminary treatment, the following devices are commonly used:

1. Screens -- rack, bar or fine
2. Comminuting devices -- grinders, cutters, shredders
3. Grit chambers
4. Pre-aeration tanks

In addition to the above, chlorination may be used in preliminary treatment. Since chlorination may be used at all stages in treatment, it is considered to be a method by itself. Preliminary treatment devices require careful design and operation.

Primary Treatment

In this treatment, most of the settleable solids are separated or removed from the wastewater by the physical process of sedimentation. When certain chemicals are used with primary sedimentation tanks, some of the colloidal solids are also removed. Biological activity of the wastewater in primary treatment is of negligible importance.

The purpose of primary treatment is to reduce the velocity of the wastewater sufficiently to permit solids to settle and floatable material to surface. Therefore, primary devices may consist of settling tanks, clarifiers or sedimentation tanks. Because of variations in design, operation, and application, settling tanks can be divided into four general groups:

- Septic tanks

- Two story tanks -- Imhoff and several proprietary or patented units
- Plain sedimentation tank with mechanical sludge removal

Upward flow clarifiers with mechanical sludge removal When chemicals are used, other auxiliary units are employed. These are:

1. Chemical feed units
2. Mixing devices
3. Flocculators

The results obtained by primary treatment, together with anaerobic sludge digestion as described later, are such that they can be compared with the zone of degradation in stream self-purification. The use of chlorine with primary treatment is discussed under the section on Preliminary Treatment.

Secondary Treatment

Secondary treatment depends primarily upon aerobic organisms which biochemically decompose the organic solids to inorganic or stable organic solids. It is comparable to the zone of recovery in the self-purification of a stream.

The devices used in secondary treatment may be divided into four groups:

1. Trickling filters with secondary settling tanks
2. Activated sludge and modifications with final settling tanks
3. Intermittent sand filters
4. Stabilization ponds

The use of chlorine with secondary treatment is discussed under the section on Secondary Treatment

Chlorination

This is a method of treatment which has been employed for many purposes in all stages in wastewater treatment, and even prior to preliminary treatment. It involves the application of chlorine to the wastewater for the following purposes:

1. Disinfection or destruction of pathogenic organisms
2. Prevention of wastewater decomposition -- Odor Control, And Protection Of Plant Structures

While chlorination has been commonly used over the years, especially for disinfection, other methods to achieve disinfection as well as to achieve similar treatment ends are also used. Among the most common is the use of ozone. In view of the toxicity of chlorine and chlorinated compounds for fish as well as other living forms, ozonation may be more commonly used in the future. This process will be more fully discussed in the section on disinfection.

Sludge Treatment

The solids removed from wastewater in both primary and secondary treatment units, together with the water removed with them, constitute wastewater sludge. It is generally necessary to subject sludge to some treatment to prepare or condition it for ultimate disposal. Such treatment has two objectives -- the removal of part or all of the water in the sludge to reduce its volume, and the decomposition of the putrescible organic solids to mineral solids or to relatively stable organic solids. This is accomplished by a combination of two or more of the following methods:

1. Thickening
2. Digestion with or without heat
3. Drying on sand bed -- open or covered
4. Conditioning with chemicals
5. Elutriation
6. Vacuum filtration
7. Heat drying
8. Incineration
9. Wet oxidation
10. Centrifuging

Package Units

The term "package units" is used in the field to describe equipment which has been put on the market by a number of manufacturers that is intended to provide wastewater treatment by the use of prefabricated or modular units. Package units can also refer to a complete installation, including both mechanisms and prefabricated containers. This term is also applied to installations where only the mechanisms are purchased and the containers constructed by the purchaser in accordance with plans and specifications prepared by the manufacturer.

Though specific limitations have not been established, individual package units have, in general, been small installations serving a limited population.

Package units have been adapted to practically all the treatment devices, either singly or in various combinations that have been mentioned.

Tertiary and Advanced Wastewater Treatment

The terms "primary" and "secondary" treatment have been used to generally describe a degree of treatment; for example, settling and biological wastewater treatment. Since the early 1970's "tertiary" treatment has come into use to describe additional treatment following secondary treatment. Quite often this merely indicates the use of intermittent sand filters for increased removal of suspended solids from the wastewater. In other cases, tertiary treatment has been used to describe processes which remove plant nutrients, primarily nitrogen and phosphorous, from wastewater.

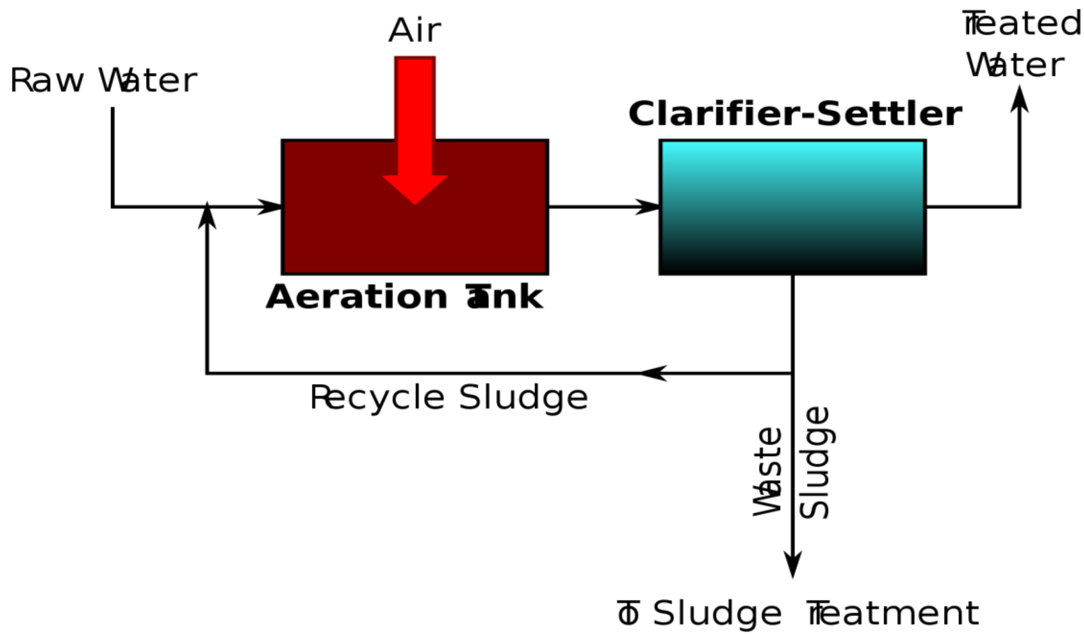
Improvement and upgrading of wastewater treatment units as well as the need to minimize environmental effects has led to the increased use of tertiary treatment.

A term that is also sometimes used to indicate treatment of a wastewater by methods other than primary or biological (secondary) treatment is advanced treatment. This degree of treatment is usually achieved by chemical (for example coagulation) methods as well as physical methods (flocculation, settling and activated carbon adsorption) to produce a high quality effluent water.

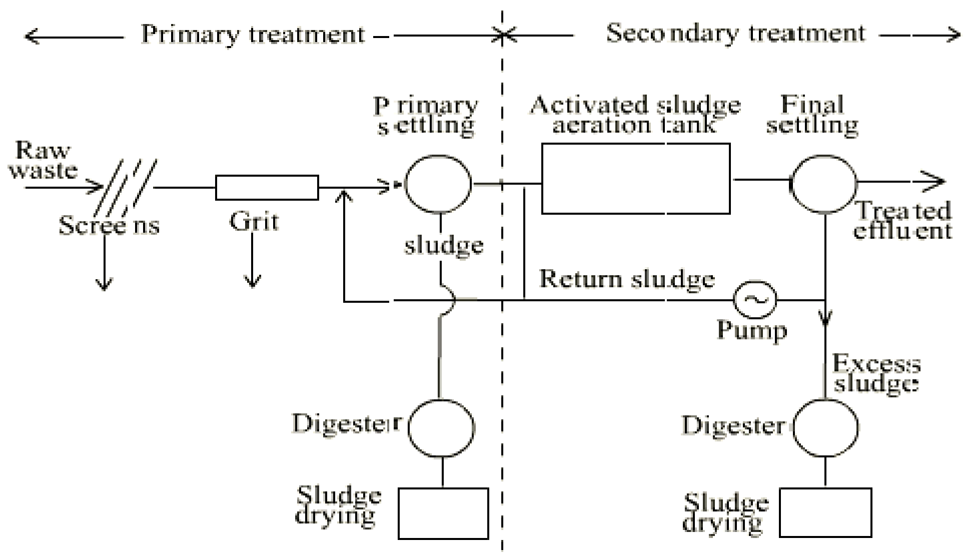
AEROBIC TREATMENT SYSTEMS

1. ACTIVATED SLUDGE PROCESS

The most common suspended growth process used for municipal wastewater treatment is the activated sludge process as shown in figure:



Flow sheet of an activated sludge system



Activated sludge plant involves:

1. wastewater aeration in the presence of a microbial suspension,
2. solid-liquid separation following aeration,
3. discharge of clarified effluent,
4. wasting of excess biomass, and
5. return of remaining biomass to the aeration tank.

In activated sludge process wastewater containing organic matter is aerated in an aeration basin in which micro-organisms metabolize the suspended and soluble organic matter. Part of organic matter is synthesized into new cells and part is oxidized to CO_2 and water to derive energy. In activated sludge systems the new cells formed in the reaction are removed from the liquid stream in the form of a flocculent sludge in settling tanks. A part of this settled biomass, described as activated sludge is returned to the aeration tank and the remaining forms waste or excess sludge. **Activated Sludge Process Variables**

The main variables of activated sludge process are the mixing regime, loading rate, and the flow scheme.

Mixing Regime

Generally two types of mixing regimes are of major interest in activated sludge process: *plug flow* and *complete mixing*. In the first one, the regime is characterized by orderly flow of mixed liquor through the aeration tank with no element of mixed liquor overtaking or mixing with any other element. There may be lateral mixing of mixed liquor but there must be no mixing along the path of flow.

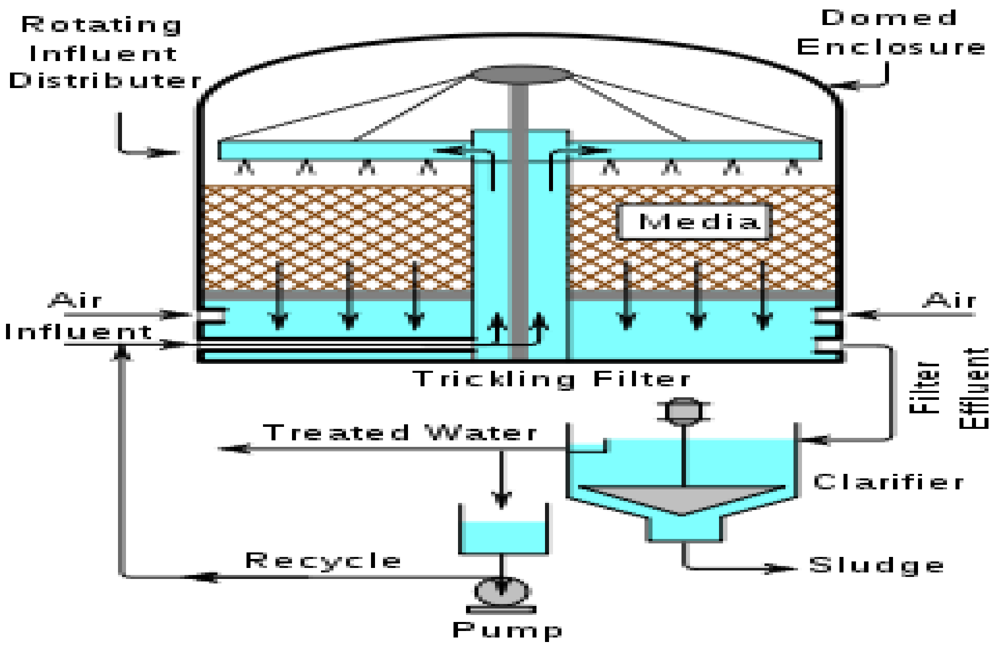
In complete mixing, the contents of aeration tank are well stirred and uniform throughout. Thus, at steady state, the effluent from the aeration tank has the same composition as the aeration tank contents.

The type of mixing regime is very important as it affects (1) oxygen transfer requirements in the aeration tank, (2) susceptibility of biomass to shock loads, (3) local environmental conditions in the aeration tank, and (4) the kinetics governing the treatment process.

2. TRICKLING FILTER

Trickling filter is a type of wastewater treatment system. It consists of a fixed bed of rocks, lava, coke, gravel, slag, polyurethane foam, sphagnum peat moss, ceramic, or plastic media over which sewage or other wastewater flows downward and causes a layer of microbial slime (biofilm) to grow, covering the bed of media. Aerobic conditions are maintained by splashing, diffusion, and either by forced air flowing through the bed or natural convection of air if the filter medium is porous.

The terms trickle filter, trickling biofilter, biofilter, biological filter and biological trickling filter are often used to refer to a trickling filter



Operation

The removal of pollutants from the wastewater stream involves both absorption and adsorption of organic compounds by the layer of microbial biofilm. The filter media is typically chosen to provide a very high surface area to volume. Typical materials are often porous and have considerable internal surface area in addition to the external surface of the medium. Passage of the wastewater over the media furnishes dissolved air, the oxygen which the slime layer requires for the biochemical oxidation of the organic compounds and releases carbon dioxide gas, water and other oxidized end products. As the biofilm layer thickens, it eventually sloughs off into the treated effluent and subsequently forms part of the secondary sludge. Typically, a trickling filter is followed by a clarifier or sedimentation tank for the separation and removal of the sloughing. Other filters utilizing higher-density media such as sand, foam and peat moss do not produce a sludge that must be removed, but require forced air blowers and backwashing or an enclosed anaerobic environment.

The treatment of sewage or other wastewater with trickling filters is among the oldest and most well characterized treatment technologies.

Industrial wastewater treatment trickle filters

Wastewaters from a variety of industrial processes have been treated in trickling filters. Such industrial wastewater trickling filters consist of two types:

- Large tanks or concrete enclosures filled with plastic packing or other media.

- Vertical towers filled with plastic packing or other media.

  The availability of inexpensive plastic tower packings has led to their use as trickling filter beds in tall towers, some as high as 20 meters.[4] As early as the 1960s, such towers were in use at: the Great Northern Oil's Pine Bend Refinery in Minnesota; the Cities Service Oil Company Trafalgar Refinery in Oakville, Ontario and at a kraft paper mill.

  The treated water effluent from industrial wastewater trickling filters is very often subsequently processed in a clarifier-settler to remove the sludge that sloughs off the microbial slime layer attached to the trickling filter media (see Image 1 above).

  Currently, some of the latest trickle filter technology involves aerated biofilters which are essentially trickle filters consisting of plastic media in vessels using blowers to inject air at the bottom of the vessels, with either downflow or upflow of the wastewater.

3. PACKED BED/COLUMN REACTOR

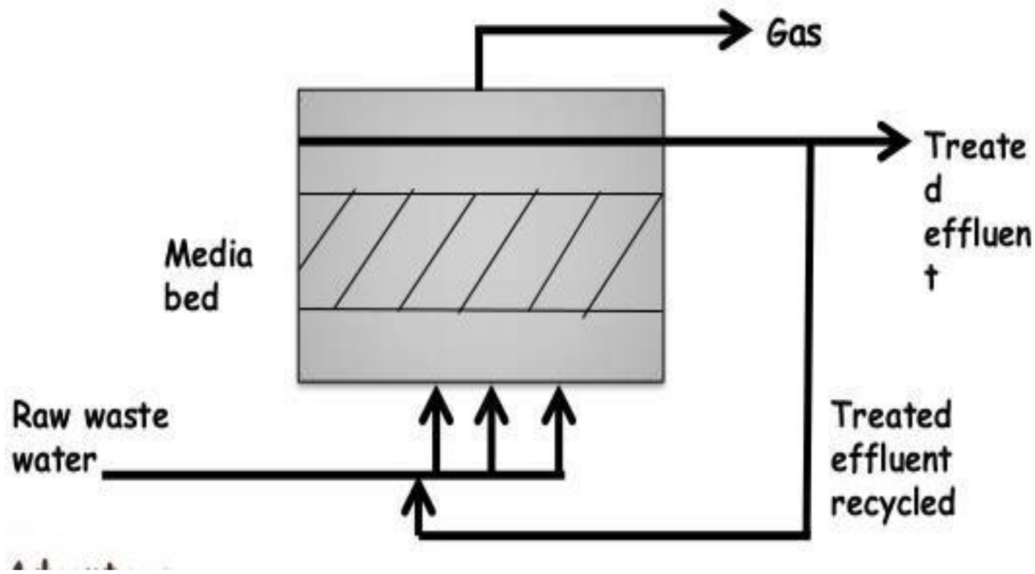
Packed bed ractors recently have been suggested for a number of wastewater treatment applications. Young and Mc-Carty in 1967 reported successful anaerobic treatment of concentrated soluble waste in an upflow submerged anaerobic filter. Tamblyn and Sword and Seidel and Crites reported that satisfactory anaerobic denitrification could be accomplished in similar upflow reactors. Johnson and Baumann described an aerobic sand-medium “pulsed adsorption bed” process for providing additional treatment of low quality effluents from secondary wastewater treatment plants.

The basic designs of the packed bed reactors (PBR’S) used for these applications are essentially the same. Each system consists of a unit containing a porous medium supported over an inlet chamber through which the influent wastewater is distributed (Figure 1).

In aerobic units, air (in some cases, oxygen-enriched air) is passed upward through the unit from an air distribution grid located immediately below or slightly above the bottom of the media bed. The selection of a medium depends on the particular application of the system. The PBR unit may be followed by granular-medium filtration or other treatment units to provide additional effluent polishing through suspended solids (ss) removal.

Johnson and Baumann and Wall reported and removal of carbonaceous biochemical oxygen demand (BODc) in laboratory and pilot scale aerobic PBR units using sand as a medium, but no nitrification forms was observed. Haug and Mccarty obtained satisfactory nitrification in laboratory scale upflow PBR'S using pure oxygen to maintain a high dissolved oxygen (DO) concentration throughout the reactor.

Research by the authors has demonstrated that considerable BODc removal and biological nitrification may be accomplished at low wastewater temperatures in PBR'S using air to maintain an aerobic environment if proper control is maintained over the influent conditions.



The column packed bed reactor used for the removal of BOD and nitrification. Microorganisms gets attached and form the bottom of the reactor.

ADVANTAGE

Easy construction

East operation and

Management Organisms do not

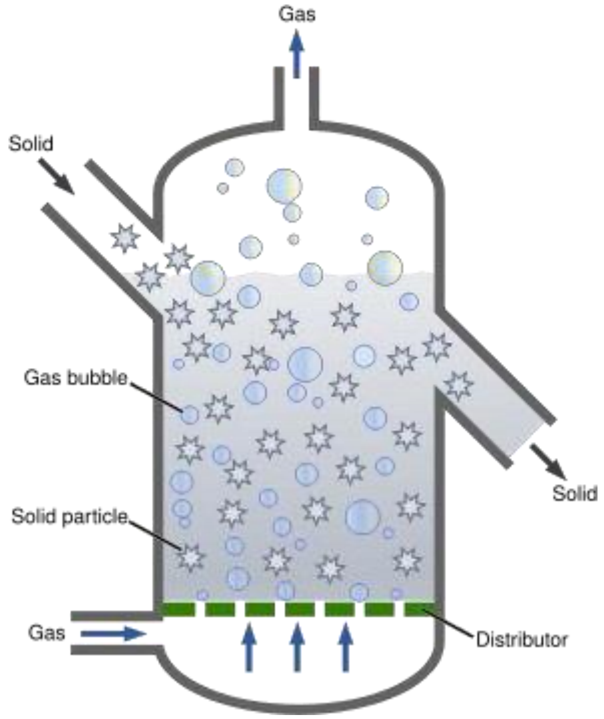
form slime on packing

APPLICATION

Column packed with anthracite, coal particle are used for denitrification of uranium recovery waste water.

4. FLUIDIZED BED REACTOR

Combination of attached growth and suspended growth. Reactor is cylindrical with perforated distribution plates. Biological slime film is developed and maintained on a solid support medium. Support media maintained in suspension by upward flow of influent. Fabricated media developed to support biomass growth within porous internal structure.



Types: 1. Captor: biomass is grown inside polyester foam pads. It can be removed periodically sequeezed. Simple sparger is used.

2. Oxitron: sand particles overflow/clean recycled. High purity oxygen is used.

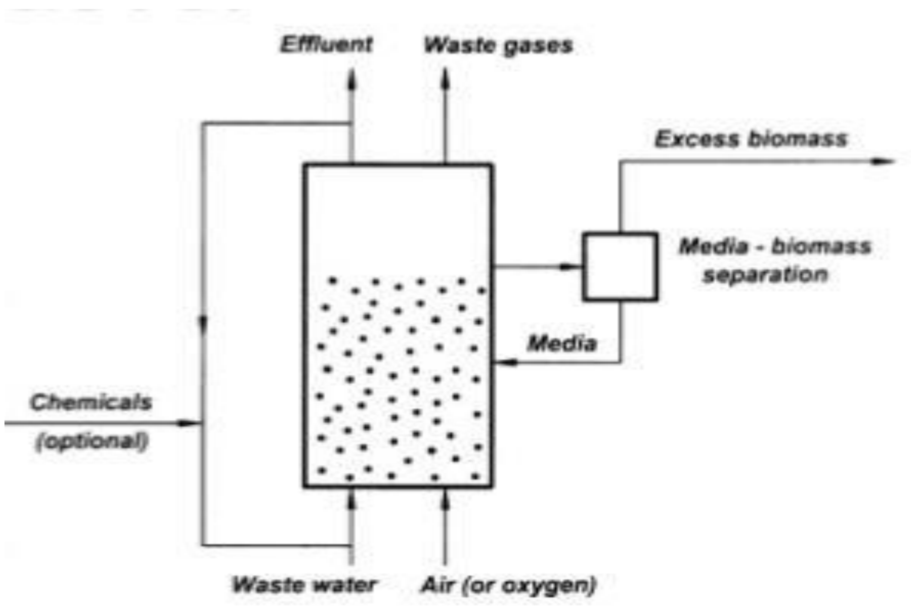
Advantage: low biomass sludge production.

High biomass concentration, high strength waste water treatment, low sludge transport and disposal, high exposed liquid/biofilm surface area, low operation cost.

Application: Aerobic FBR used for nitrate removal in petroleum industry, coke plant, explosive factory nuclear plant. Anoxic FBR used for de-nitrification.

5. INVERSE FLUIDIZED BED BIOFILM

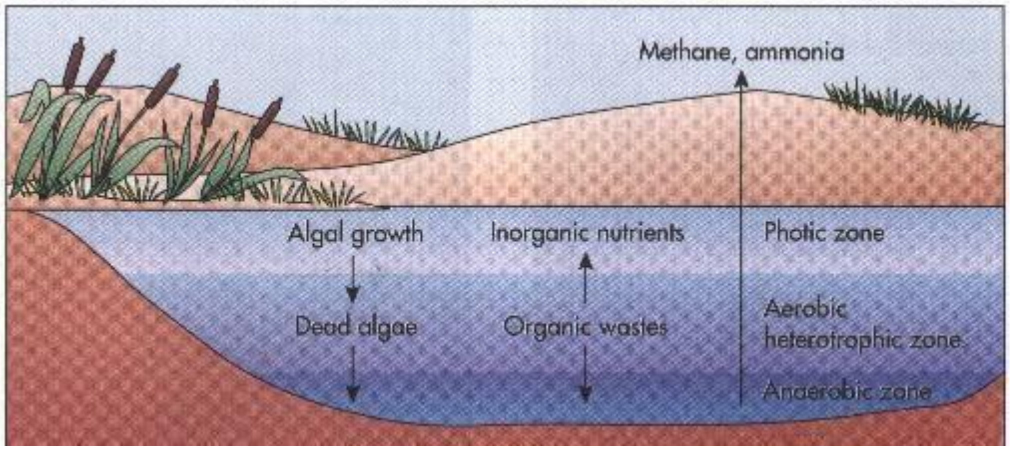
Reactor bed is consisting of biofilm attached to low density polypropylene particles. It expands downwards during fluidization. Allows the control of biomass loading and provides the high oxygen concentration in the reacting liquid media. Fluidization can be conducted by an upward co-current flow of gas and liquid through bed.



ADVANTAGES: Large specific support surface area. 2. Fast biofilm formation, 3. Effective and simple control of biofilm thickness.

6. OXIDATION PONDS

Oxidation Ponds are also known as stabilization ponds or lagoons. They are used for simple secondary treatment of sewage effluents. Within an oxidation pond heterotrophic bacteria degrade organic matter in the sewage which results in production of cellular material and minerals. The production of these supports the growth of algae in the oxidation pond. Growth of algal populations allows furthur decomposition of the organic matter by producing oxygen. The production of this oxygen replenishes the oxygen used by the heterotrophic bacteria. Typically oxidation ponds need to be less than 10 feet deep in order to support the algal growth. In addition, the use of oxidation ponds is largely restricted to warmer climate regions because they are strongly influenced by seasonal temperature changes. Oxidation ponds also tend to fill, due to the settling of the bacterial and algal cells formed during the decomposition of the sewage. Overall, oxidation ponds tend to be inefficient and require large holding capacities and long retention times. The degradation is relatively slow and the effluents containing the oxidized products need to be periodically removed from the ponds. An oxidation pond can be seen in the figure below.



7. Lagoons

An **aerated lagoon** or **aerated basin** is a holding and/or treatment pond provided with artificial aeration to promote the biological oxidation of wastewaters.

Types of aerated lagoons or basins

Suspension mixed lagoons, where there is less energy provided by the aeration equipment to keep the sludge in suspension.

Facultative lagoons, where there is insufficient energy provided by the aeration equipment to keep the sludge in suspension and solids settle to the lagoon floor. The biodegradable solids in the settled sludge then degrade as in an anaerobic lagoon.

Suspension mixed lagoons

Suspension mixed lagoons flow through activated sludge systems where the effluent has the same composition as the mixed liquor in the lagoon. Typically the sludge will have a residence time or sludge age of 1 to 5 days. This means that the chemical oxygen demand (COD) removed is relatively little and the effluent is therefore unacceptable for discharge into receiving waters. The objective of the lagoon is therefore to act as a biologically assisted flocculator which converts the soluble biodegradable organics in the influent to a biomass which is able to settle as sludge. Usually the effluent is then put in a second pond where the sludge can settle. The effluent can then be removed from the top with a low COD, while the sludge accumulates on the floor and undergoes anaerobic stabilization.

Methods of aerating lagoons or basins

There are many methods for aerating a lagoon or basin: Motor-driven floating surface aerators

Motor-driven submerged or

floating jet aerators Motor-driven

fixed-in-place surface aerators

Injection of compressed air through submerged diffusers

Floating surface aerators

A Typical Surface-Aerated Basin (using motor-driven floating aerators)

Ponds or basins using floating surface aerators achieve 80 to 90% removal of BOD with retention times of 1 to 10 days.[5] The ponds or basins may range in depth from 1.5 to 5.0 metres. **In a surface-aerated system**, the aerators provide two functions: they transfer air into the basins required by the biological oxidation reactions, and they provide the mixing required for dispersing the air and for contacting the reactants (that is, oxygen, wastewater and microbes). Typically, the floating surface aerators are rated to deliver the amount of air equivalent to 1.8 to

2.7 kg O_2/kWh. However, they do not provide as good mixing as is normally achieved in activated sludge systems and therefore aerated basins do not achieve the same performance level as activated sludge units.

Biological oxidation processes are sensitive to temperature and, between 0 °C and 40 °C, the rate of biological reactions increase with temperature. Most surface aerated vessels operate at between 4 °C and 32 °C.

Submerged diffused aeration

Submerged diffused air is essentially a form of a diffuser grid inside a lagoon. There are two main types of submerged diffused aeration systems for lagoon applications: floating lateral and submerged lateral. Both these systems utilize fine or medium bubble diffusers to provide aeration and mixing to the process water. The diffusers can be suspended slightly above the lagoon floor or may rest on the bottom. Flexible airline or weighted air hose supplies air to the diffuser unit from the air lateral (either floating or submerged).

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ANAEROBIC BIOLOGICAL TREATMENT

Anaerobic digestion is a sequence of processes by which microorganisms break down biodegradable material in the absence of oxygen.The process is used for industrial or domestic purposes to manage waste or to produce fuels. The fermentation used industrially to produce food and drink products, as well as home fermentation, uses anaerobic digestion.Anaerobic digestion is used as part of the process to treat biodegradable waste and sewage sludge.

Process stages

The four key stages of anaerobic digestion involve **hydrolysis, acidogenesis, acetogenesis** and **methanogenesis.**The overall process can be described by the chemical reaction, where organic material such as glucose is biochemically digested into carbon dioxide (CO_2) and methane (CH_4) by the anaerobic microorganisms.

$C_6H_{12}O_6 \rightarrow 3CO_2 + 3CH_4$

Hydrolysis

In most cases, biomass is made up of large organic polymers. For the bacteria in anaerobic digesters to access the energy potential of the material, these chains must first be broken down into their smaller constituent parts. These constituent parts, or monomers, such as sugars, are readily available to other bacteria. The process of breaking these chains and dissolving the smaller molecules into solution is called hydrolysis. Therefore, hydrolysis of these high-molecular-weight polymeric components is the necessary first step in anaerobic digestion. Through hydrolysis the complex organic molecules are broken down into simple sugars, amino acids, and fatty acids.

Acetate and hydrogen produced in the first stages can be used directly by methanogens. Other molecules, such as volatile fatty acids (VFAs) with a chain length greater than that of acetate must first be catabolised into compounds that can be directly used by methanogens.

Acidogenesis

The biological process of acidogenesis results in further breakdown of the remaining components by acidogenic (fermentative) bacteria. Here, VFAs are created, along with ammonia, carbon dioxide, and hydrogen sulfide, as well as other byproducts.

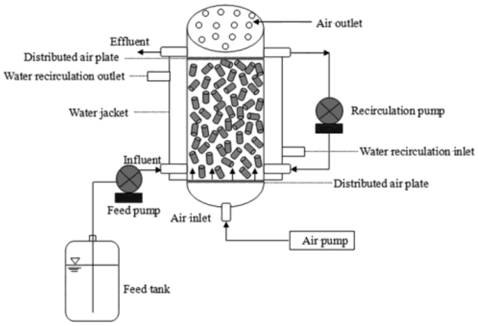
Acetogenesis

The third stage of anaerobic digestion is acetogenesis. Here, simple molecules created through the acidogenesis phase are further digested by acetogens to produce largely acetic acid, as well as carbon dioxide and hydrogen.

Methanogenesis

The terminal stage of anaerobic digestion is the biological process of methanogenesis. Here, methanogens use the intermediate products of the preceding stages and convert them into methane, carbon dioxide, and water. These components make up the majority of the biogas emitted from the system. Methanogenesis is sensitive to both high and low pHs and occurs between pH 6.5 and pH 8.The remaining, indigestible material the microbes cannot use and any dead bacterial remains constitute the digestate.

PACKED COLUMN REACTOR



. A packed bed is a hollow tube, pipe, or other vessel that is filled with a packing material. Packed columns are used for distillation,gas absorption, and liquid-liquid extraction. The gas liquidcontact in packed bed column is continuous, as in a plate column. The liquidflows down the column over the packing surface and the gas or vapor, counter-currently ,up the column , some gas-absorption column are co-current . The packing can be randomly filled with small objects like Raschig rings or else it can be a specificallydesigned structured packing. Packed beds may also contain catalyst particles or adsorbents such as zeolite pellets, granular activated carbon. Packed column utilize packing to contact between the phases (liquid-vapor) on the surface. Packed column performs well at low pressure, low liquidand vapor loadingthat make packed column have the most efficient in these terms. At high flow parameters the capacity and efficiency can significantly reduce, also in heavy fouling applications and corrosive condition. Packed column has less pressure drop than tray column and it reduce

. foaming since generates thin films instead of fine droplets for mass and heat transfer. Packed column is divided by Random, Structured and Grid Packed Columns which is generate a mass transfer area by providing a large surface area over (50%) which the liquidcan transfer heat and mass to the vapor. The performance of a packed column is very dependet on the maintenanceof a good liquidand gas distribution throughout the packed bed Packed Column Description In the Figure illustrates a tower with structured packing.In additionto the packing itself, packed columnsrequire other internals to assure the performance ofthe packing. These internals are:Liquid feed pipes to deliver theSuid to the liquiddistributors,as seen at the top of the tower and at the intermediate distributor.Liquid collectionand mixing as shown below the top bed. Liquiddraw-off sump and pipe as shown below the top bed. Liquid redistributors, as presented between the two beds .Vapor feed pipes as shown

. at the vapor inlet nozzle, at the bottom of the tower. Packing support plates resting on beams and level-led rings welded to the vessel. Hold-down plates. Incorrect design or incorrect installationof any of these elements can lead to tower failure. One of the most critical element, and often the culprit of tower failures, is the liquid distributor. Component of a packed column:

Packing materials: 1-Ceramic: superior wettability ,corrosion resistance at elevated temperature ,bad strength. 2-metal: superior strength and good wettability. 3-plastic:inexpensive, good strength but may have poor wettability at low liquid rate.

Type of packed column:

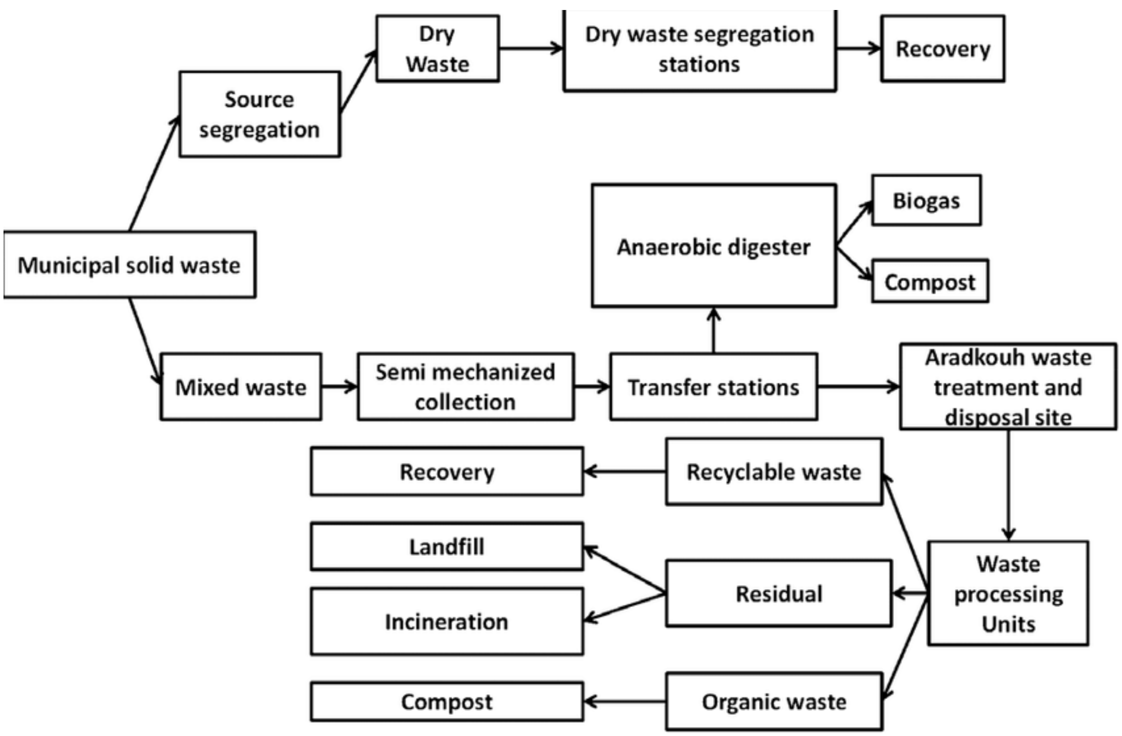
1. Random Packed Column: Random packing is packing of specific geometrical shapes which are dumped into the tower and orient themselves randomly. Random packing has more risk than structured packing and less ability to handle mal distributed liquid.

2. Structured Packed Column : is crimped layers or corrugated sheets which is stacked in the column. Each layer is oriented at 70° to 90° to the layer below. Structured packed offers 30% capacities higher than random packed for equal efficiency up to 50% higher at the same capacity.

3. Grid Packed Column: Is systematically arranged packing use an open- lattice structure. This device is composed of panels that promote mass transfer and enhance entrainment removal. They have high open area, resulting in high capacity, low pressure drop, and high tolerance to fouling.

SOLID WASTE MANAGEMENT

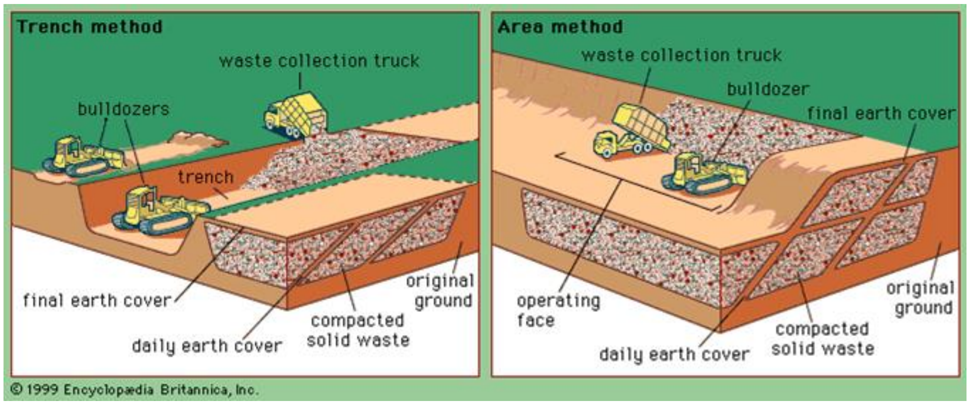
Solid Waste in general can be defined as 'As a material which has negligible value to the producer and there is no direct consumption of the generated waste'. It is generated due to various activities that can be residual and commercial, agricultural, etc. Whatever the origin, content or hazard potential is, solid waste must be managed systematically to ensure environmental best practices.The processing methods available for management of solid waste includes : Segregation , Reduction, Reuse and Recycling, Chemical, Biological And Thermal Conversion, etc.



LANDFILLING

A landfill site is a site for the disposal of waste materials by burial and is the oldest form of waste treatment. E.g Waste is directly dumped into mining voids or borrow pits. Disposed waste is compacted and covered with soil. Gases generated by the decomposing waste materials are

often burnt to generate power.



Landfilling can be done by three methods :

1) Trench method

2) Area method

3) Ramp method

1. TRENCH METHOD

The trench method consists of an excavated trench into which the solid wastes are spread, compacted and covered. The trench method is best suited for nearly level land where the water table is not near the surface.

2. AREA METHOD

The area method is best suited for flat or gently sloping areas where some land depressions may exist. The wastes are spread, compacted and then covered.

3. RAMP METHOD

It is also know as depression method. The slope or ramp is sometimes used in combination with the other two methods. The wastes are spread on an existing slope, compacted and covered.

Advantages :

- Landfill site is a cheap waste disposal option.
- The gases given off by the landfill site could be collected and used for generating power.
- Lots of different types of waste can be disposed of by landfill in comparison to other waste disposal method.

Disadvantages :

- Landfills can pollute air, water and also the soil.
- Landfill can contribute to the global warming

Advantages of a sanitary landfill

Sanitary landfills, as a final disposal method for urban solid wastes, are the best alternative for the countries of our Region. However, it is essential to provide the financial and technical resources for planning, design, construction, operation, and maintenance.

 The initial capital investment is lower than that required to implement either incineration or composting methods.

 Low costs of operation and maintenance.

 It is a complete and final method that can receive all types of municipal solid wastes, eliminating the problem of ashes from incineration and materials that do not degrade during composting.

 It generates employment for unskilled laborers, who are readily available in developing countries.

COMPOSTING

The process of biological degradation of solid organic waste material to stable end products. This can be done in aerobic and anaerobic, aerobic method is more frequently used. In composting solid organic waste converted by aerobic microbiological process in to stable sanitary, nuisance free, humus like material that can be safely disposed into the environments. Composting is cost effective, eco friendly for stable ultimate disposal of sludge. The composting product can be used for soil improvement and used for production of mushroom, helpful for reuse and recycling of organic waste materials from domestic, agriculture and industry.

Organisms involved in composting

Bacteria 80-90% found in the compost possesses a broad range of enzymes to degrade a wide range of organic compounds. Other organisms are actinomycetes, fungi, protozoa, mites, insects, ants and earth worms.

Mechanism of composting

Proteins and lipids decomposed by bacteria, cellulose and other complex organic compounds degraded by actinomycetes and fungi.

Compositing done in 3 stages due to changes in temperature

1. Mesophilic stage: fungi and acid produceing bacteria are active in this stage and the temperature increases from ambient to about 40ºC.
2. Thermophilic stage: as the composting proceeds the temperature raises from 40ºC-70ºC. thermophilic bacteria, thermophilic fungi and actinomycetes are active in this stage.

thermophilic stage is associated with high rate and maximum degrading organic materials.

3. Cooling stage: degradative activity slows down and the thermophilic organisms are replaced by mesophilic bacterial and fungi. Cooling stage is associated with the formationof water, pH stabilization and completion of humic acid formation.

Methods of composting:

Operation of composting involves the following steps.

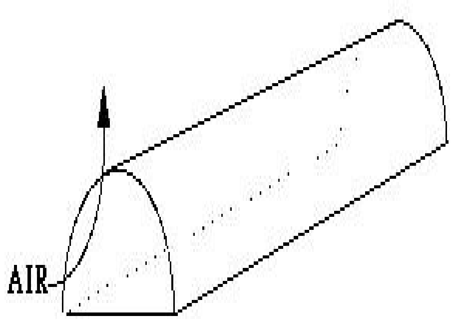
1. Mixing of dewatered sludge with a bulking agent (saw dust, rice hulls, straw or recycle compost). Improve the porosity of the mixture for good aeration.
2. Creating aerobic conditions (aeration) by mechanical or other means. This is needed for the supply of oxygen to control temperature for the removal of water.
3. Removal of bulking agent, if possible.
4. Storage and disposal of the composite.

Composting systems

1. Non-reactor: 2 types i. Field composting, ii. Windrow composting
2. Reactor system:i. Horizontal flow reactor, ii. Vertical flow reactors, iii. Rotating drum reactors

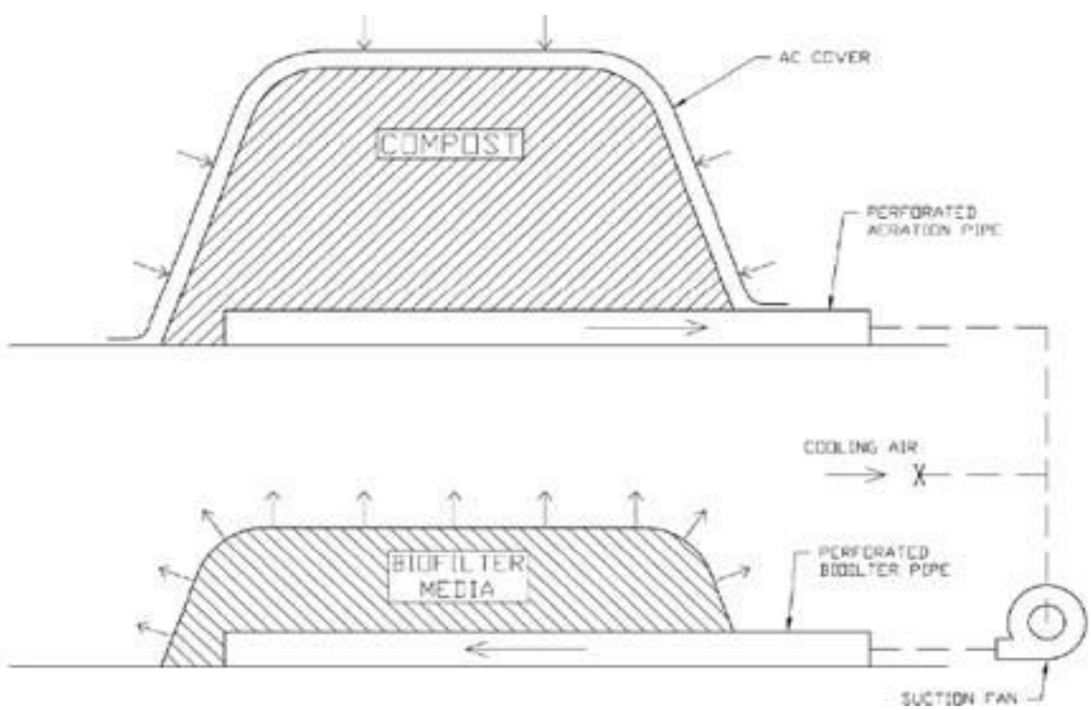
i. FIELD COMPOSTING

It is a natural organic decomposting process, best for hygienic waste. All microbial activity takes place in a thin layer on the soil surface or within a few centimeter of the soil surface. This system is useful for treating both sludge and green wastes, grass, straw, bushwood. To enhance the reaction the waste need to be chopped so the surface of waste exposed to the atmosphere in large, thermal heating does not occur i.e thermal disinfection or killing of weed seeds does not occur.



ii. WINDROW COMPOSTING

This composting is carried out due to the direct contact between the waste material and the atmosphere. The composting process influences the atmosphere by emitting odour, green house gases, spores, germs and dust. The atmosphere carrying oxygen influences the composting by supplying rain water, changes in the temperature, changes in the air humidity and proper aeration. The extent of contact between waste material and the atmosphere can be limited by covering the pile with mature compost material, straw or special textile that allow gas exchange, but reduce the infiltration of rain water. To get high quality compost windrow are disturbed from time –time by turning. This mixing of the material for homogenization dry or wet zones at the surface, wet zones at the bottom. For killing of pathogenic of microorganisms and weed seeds, renewing the structure and free air space and increase evaporation to dry the waste material or the mature compost. The turning frequency depends on the kind and structure of waste and quality requirement of finished compost.



REACTOR COMPOSTING

Composting in an enclosed space (eg. Container, box, bin, tunnel, shed with forced exchange of respiration gases is a type of reactor composting. According to the material flow reactor composting is divided in to

1. Horizontal flow reactor
2. Vertical flow reactor
3. Rotating drum reactor

The reactor have controlled forced aeration and enable the waste air and drainage water to be collected and treated. Addition of water or other additive is possible.

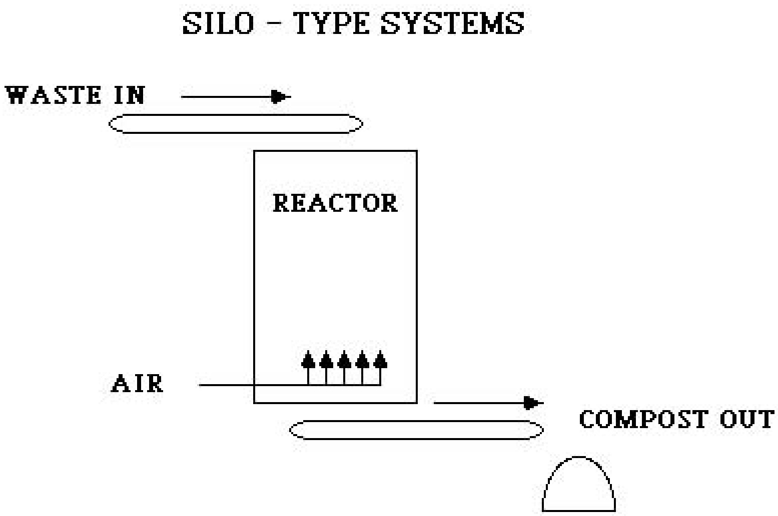
1. Horizontal flow reactor: Static reactor is a batch system where the waste material is loaded with a wheel loader or transport devices in to a horizontal reactor or is covered by a foil or textile material. The waste is placed over a grid and forced aeration was given. It can also pass air through pipes at the bottom of the material or from holes in the floor. The waste air and its odour components and waste water can be treated in a biofilter. In some systems waste air is recycled. Air flow rate is controlled base on oxygen, carbon dioxide and temperature of the material. The retention time in the reactor is between several days to several weeks.

Disadvantages: No turning/mixing, no water addition, forced aeration only from one direction. The end product may be heterogenous (partially too dry) and biologically not stabilized.

2. **Vertical composting reactors** : are generally over 4 meters (yards) high, and can be housed in silos or other large structures. Organic material is typically fed into the reactor at the top through a distribution mechanism, and flows by gravity to an unloading mechanism at the bottom. Process control is usually by pressure-induced aeration, where the airflow is opposite to the downward materials flow. The height of these reactors makes process control difficult due to the high rates of airflow required per unit of distribution surface area. Neither temperature nor oxygen can be maintained at optimal levels throughout the reactors, leading to zones of non-optimal activity. Some manufacturers have minimized these difficulties by enhanced air distribution and collection systems, including changing the airflow direction from vertical to horizontal between alternating sets of inflow and exhaust pipes. As with static pile composting, a stable porous structure is important in vertical reactors which usually lack internal mixing. Tall vertical reactors have been successfully used in the sludge

composting industry where uniform feedstocks and porous amendments can minimize these difficulties in process control, but are rarely used for heterogeneous materials like MSW.

Vertical Reactor



VERMICOMPOST

Vermicomposting

Vermicomposting is a type of composting in which certain species of earthworms are used to enhance the process of organic waste conversion and produce a better end-product. It is a mesophilic process utilizing microorganisms and earthworms. Vermicomposting is an ecofriendly, low cost effective and a effective way to recycle agricultural and kitchen waste. The application of vermicompost not only adds plant nutrients (macro and micro) and growth regulators but also increases soil water retention, microbial population, nutrient content and carbon content of the soil. Vermicompost refers to an organic manure produced by earthworms.

It is mixture of worm castings, organic material including humus, live earth worms and their cocoons and other organisms. Vermicomposting is a appropriate technique for the disposal of non-toxic solid and liquid organic wastes. It helps in cost effective and efficient recycling of animal wastes agricultural residual and industrial wastes using low energy.

Earthworms

Earthworms feed on any organic waste, consume two to five times their body weight and after using 5-10 per cent of the feed stock for their growth, excrete the mucus coated undigested matter as wormcasts worm cast consists of organic matter that has undergone physical and chemical breakdown through the activity of the muscular gizzard which grinds the material to a particle size of 1-2 micron. The nutrients presents in wormicast are readily soluble in water for the uptake of plants. Worm casts are rich source of macro and micro nutrients, vitamins, enzymes, antibiotics, growth harmones and immobilized microfloras.

Earthworms are cosmopolitan in distribution. They are important invertebrate macrofauna. Macroinvertebrates, in general, play a definite and prominent role in regulating soil process; among them, earthworms play a vital role in maintaining soil fertility and efficient nutrient cycling.

Earthworms also act as biological indicators of the soil type and its properties.They also serve to detect toxicity of industrial chemicals and act as indicators of soil fertility because the presence of earthworms support healthy populations of bacteria, actinomycetes, fungi, protozoans, insects, spiders, millipedes and a host of other organisms that are essential for sustaining healthy soil.

Earthworms species for vermicompost

1. *Lampito mauriti* -Sandy loam soil
2. *Octochaetona serrata*- Clay soil
3. *Lumbricus terrestris* -Deep boarings
4. *Allolobophora caliginosa* -shallow boarings
5. *Eudrilus eugeniae*
6. *Eisenia foetida*
7. *Perionyx excavatus*
8. *Pheretima elongate*

The *Eisenia foetida* an epigeic species of earthworms is used for vermicomposting. The general characteristics of *Eisenia foetida* are as under:

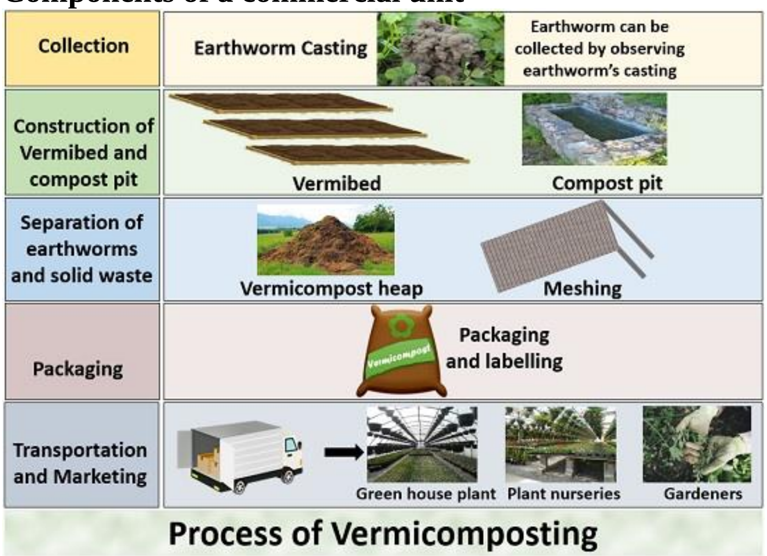
Habit: Epigeic
Size: 8-10 cm
Diameter: 0.8-1.0 mm
Duration of life: 70 days
Temperature range: 15-40 0C
Approximate weight of single earthworm: 1.0-3.0 g
Moisture tolerance range: 30-70%
Habit and Habitats: Living on surface and organic matter rich areas.

Process of vermicomposting

A wide range of organic residues, such as straw, husk, leaves, stalks, weeds etc can be converted into vermicompost. Other potential feedstock for vermicompost production are livestock wastes, poultry litter, dairy wastes, food processing wastes, organic fraction of MSW, bagasse, digestate from biogas plants etc. Earthworms consume organic wastes and reduce the volume by 40–60 percent. Each earthworm weighs about 0.5 to 0.6 gram, eats waste equivalent to its body weight and produces cast equivalent to about 50 percent of the waste it consumes in a day. The moisture content of castings ranges between 32 and 66 percent and the pH is around 7. The level of nutrients in compost depends upon the source of the raw material and the species of earthworm.

There are nearly 3600 types of earthworms which are divided into burrowing and non-burrowing types. **Red earthworm species, like *Eisenia foetid*a**, and are most efficient in compost making. The non-burrowing earthworms eat 10 percent soil and 90 percent organic waste materials; these convert the organic waste into vermicompost faster than the burrowing earthworms. They can tolerate temperatures ranging from 0 to 40°C but the regeneration capacity is more at 25 to 30°C and 40–45 percent moisture level in the pile. The burrowing types of earthworms come onto the soil surface only at night. These make holes in the soil up to a depth of 3.5 m and produce 5.6 kg casts by ingesting 90 percent soil and 10 percent organic waste.

Components of a commercial unit



Location

Suburbs of cities and villages around urban centres can be ideal locations for practice of vermicomposting on a large scale, from the view point of availability of raw material and marketing of the produce. As use of the compost is said to have ameliorative effect on product from fruit, flower and vegetable crops, vermicomposting units may be located in areas with concentration of fruit and vegetable growers and floriculture units.

Sheds

For a vermi-composting unit, whether small or big, this is an essential item and is required for having the vermi beds. They could be of thatched roof supported by bamboo rafters and purlins, wooden trusses and stone pillars. If the size is so chosen as to prevent wetting of beds due to rain on a windy day, they could be open sheds. While designing the sheds adequate room has to be left around the beds for easy movement of the labour attending to the filling and harvesting the beds.

Vermi-beds

Normally the beds are 75 cm - 90 cm thick depending on the provision of filter for drainage of excess water. The entire bed area could be above the ground. Care should be taken to make the bed with uniform height over the entire width to the extent possible to avoid low production owing to low bed volumes. The bed width should not be more that 1.5 m to allow easy access to the centre of the bed.

Land

About 0.5-1 acre of land will be needed to set up a vermiculture production cum extension centre. The centre will have at least 8-10 sheds each of about 180-200 sq.ft. It should also have a bore well, and pump set or watering arrangement and other equipments as described in the scheme economics. The land can be taken on lease of at least 10-15 years. Even sub marginal land also will serve the purpose.

Seed Stock

This is an important item requiring considerable investment. Though the worms multiply fast to give the required numbers over a period of 6 months to a year, it may not be wise to wait till such a time having invested on the infrastructure heavily. Thus, worms @ 350 worms per m3 of bed space should be adequate to start with and to build up the required population in about two cycles or three without unduly affecting the estimated production.

Fencing and Roads/Paths

The site area needs development for construction of structures and development of roads and pathways for easy movement of hand-drawn trolleys/wheel barrows for conveying the raw material and the finished products to and from the vermi-sheds. The entire area has to be fenced to prevent trespass by animals and other unwanted elements. These could be estimated based on the length of the periphery of the farm and the length and type of roads/paths required. The costs on fencing and formation of roads should be kept low as these investments are essential for a production unit, yet would not lead to increase in production.

Water Supply System

As the beds have always to be kept moist with about 50% moisture content, there is need to plan for a water source, lifting mechanism and a system of conveying and applying the water to the vermi-beds. Drippers with round the clock flow arrangement would be quite handy for continuous supply and saving on water. Such a water supply/application system requiring considerable initial investment, however, reduces the operational costs on hand watering and prove economical in the long run. The cost of these items depend on the capacity of the unit and the type of water supply chosen.

Machinery

Farm machinery and implements are required for cutting (shredding) the raw material in small pieces, conveying shredded raw material to the vermi-sheds, loading, unloading, collection of compost, loosening of beds for aeration, shifting of the compost before packing and for air drying of the compost, automatic packing and stitching for efficient running of the unit. Costs of providing necessary implements and the machinery have to be included in the project cost. ;;'

Transport

For any vermi-composting unit transport arrangement is a must. When the source of raw material is away from the production unit, an off-site transport becomes major item of investment. A large sized unit with about 1000 tonnes per annum capacity may require a 3-tonne capacity mini-truck. With small units particularly with the availability of raw material near the site, expending on transport facility may become infructuous. On-site transport facilities like manually drawn trolleys to convey raw material and finished products between the storage point and the vermi-compost sheds could also be included in the project cost.

Furniture

A reasonable amount could also be considered for furnishing the office-cum-stores including the storage racks and other office equipment. These enhance the efficiency of operations.

Operational Costs

In order to operate the unit, expenditure on some items have to be incurred on a recurring basis. These items include salaries of the staff, wages to the labourers, cost of raw material, fuel cost on transport of raw materials and finished goods, packing material cost, repairs and maintenance, power, insurance, etc. The number of office personnel and labourers have to be decided breaking each activity into a number of sub-activities and for each sub-activity estimating the work involved and the capacity of the labour to finish the work in a given time. The number of persons should be so chosen to keep them engaged throughout by providing enough persons at various work points like stores, vermi-beds and equipping them with adequate number of implements to avoid undue waiting.

Applications of Vermicompost

The worm castings contain higher percentage of both macro and micronutrients than the garden compost. Apart from other nutrients, a fine worm cast is rich in NPK which are in readily available form and are released within a month of application. Vermicompost enhances plant growth, suppresses disease in plants, increases porosity and microbial activity in soil, and improves water retention and aeration.

Vermicompost also benefits the environment by reducing the need for chemical fertilizers and decreasing the amount of waste going to landfills. Vermicompost production is trending up worldwide and it is finding increasing use especially in Western countries, Asia-Pacific and Southeast Asia.

A relatively new product from vermicomposting is vermicompost tea which is a liquid produced by extracting organic matter, microorganisms, and nutrients from vermicompost. Unlike vermicompost and compost, this tea may be applied directly to plant foliage, reportedly to enhance disease suppression. Vermicompost tea also may be applied to the soil as a supplement between compost applications to increase biological activity.

Uses

The wastes are pulverized as they pass through the worm,

Surface area of the material increases which in turn helps as base for nutrients

Supply nutrients and growth enhancing hormones to plants

Improves the soil structure leading to increase in water and nutrient holding capacities of soil. Chemical fertilizer in moderate doses can go along with vermicomposting.

Wasteland reclamation using microbes

Wasteland reclamation of wasteland means re-claiming it or to use it for productive purpose. Wasteland reclamation is the process of turning barren, sterile wasteland into something that is fertile and suitable for habitation and cultivation.

The green revolution is the result of intensive agriculture with extensive use of chemical fertilizers. In general, the production of chemical fertilizers has led to depletion of fossil fuel environmental pollution and occupational hazards. In particular, the application of chemical fertilizers in agricultural fields reduces the fertility of the soil.

Due the removal process of desired mineral materials, soil textures have been destroyed, various nutrient cycles have been disturbed, and microbial communities have been altered, affecting vegetation and leading to the destruction of wide areas of land in many countries. Therefore, soil restoration of abandoned mining lands became a very important part of sustainable development strategies.

Contiuous agricultural activities, remove the top soil layer transforming these lands less fertile and more susceptible for soil hazards. Growing cover crops is one of the best practices to improve the organic matter content of the soil and hence soil health and quality. Vegetation layers—besides adding plant material to the soil contributing to its organic matter replenishment—can also fix N in the soil, moderate soil temperature protecting hence soil organisms and can provide habitat for beneficial insects and other organisms. The fertility of soil can be enhanced by Stimulate existing soil microbes, improve the nutrient and water efficiencies, introduce stress tolerance plants, add biofertilizer to increase the nutrient, with agricultural crop rotation and high fertilization doses depending on the location.

ADVANTAGES OF VERMICOMPOSTING :

1. Organic wastes are broken down and fragmented rapidly through earthworms resulting in stable, non-toxic material of potentially high economic value, which is used as a soil conditioner / fertilizer in agricultural and horticultural practices.
2. 2. As with the composting process, vermicomposting provides a reduction in waste bulk density.
3. As an aerobic process composting leads to Nitrogen mineralization. In vermicomposting, earthworms increase and accelerate Nitrogen mineralization rate. The humification process is greater and faster during vermicomposting.
4. A decrease in fulvic acid carbon and increase in the percentage of humic acid carbon are observed in vermicompost. The humic substances show an increase of 40-60%, which was higher than the value obtained through composting process.
5. Vermicomposting also brings down the availability of heavy metals compared to composting.
6. Vermicomposting has hormone like compounds (auxins), which accelerate plant growth.
7. Higher percentages of germination have been recorded with vermicompost than with compost.
8. Chemical analysis of vermicomposts and composts have shown slightly lower pH value for vermicompost and slightly higher nutrient concentrations, particularly of Nitrogen. It is also quite common for vermicomposts to have very low concentration of Ammonia-nitrogen and very high concentration of Nitrate-nitrogen, whereas the opposite is true for many types of compost. The nutritional value of vermin-compost depends upon the raw materials used for feeding.

SCHOOL OF BIO AND CHEMICAL ENGINEERING

DEPARTMENT OF BIOTECHNOLOGY

UNIT – III– Environmental & Agricultural Biotechnology – SBB2201

BIODEGRADATION OF XENOBIOTICS

Biodegradation or biological degradation is the phenomenon of biological transformation of organic compounds by living organisms, particularly the microorganisms.

Biodegradation basically involves the conversion of complex organic molecules to simpler (and mostly non-toxic) ones. The term biotransformation is used for incomplete biodegradation of organic compounds involving one or a few reactions. Biotransformation is employed for the synthesis of commercially important products by microorganisms.

Bioremediation refers to the process of using microorganisms to remove the environmental pollutants i.e. the toxic wastes found in soil, water, air etc. The microbes serve as scavengers in bioremediation. The removal of organic wastes by microbes for environmental clean-up is the essence of bioremediation. The other names used (by some authors) for bioremediation are bio-treatment, bio-reclamation and bio-restoration.

It is rather difficult to show any distinction between biodegradation and bioremediation. Further, in biotechnology, most of the reactions of biodegradation/bioremediation involve xenobiotic.

Xenobiotic:

Xenobiotic (xenos-foregin) broadly refer to the unnatural, foreign and synthetic chemicals such as pesticides, herbicides, refrigerants, solvents and other organic compounds. Microbial degradation of xenobiotic assumes significance, since it provides an effective and economic means of disposing of toxic chemicals, particularly the environmental pollutants.

Pseudomonas — The Predominant Microorganism For Bioremediation:

Members of the genus Pseudomonas (a soil microorganism) are the most predominant microorganisms that degrade xenobiotic. Different strains of Pseudomonas, that are capable of detoxifying more than 100 organic compounds, have been identified. The examples of organic compounds are several hydrocarbons, phenols, organophosphates, polychlorinated biphenyls (PCBs) and polycylic aromatics and naphthalene.

About 40-50 microbial strains of micro-organisms, capable of degrading xenobiotics have been isolated. Besides Pseudomonas, other good examples are Mycobacterium, Alcaligenes, and Nocardia. A selected list of microorganisms and the xenobiotics degraded is given in Table

List of Microorganisms and the Pollutants

A selected list of microorganisms and the pollutants (xenobiotics) that are degraded by bioremediation

Microorganism	*Pollutant chemicals*
Pseudomonas sp	Aliphatic and aromatic hydrocarbons—alkylaminoxides, alkylammonium benzene, naphthalene, anthracene xylene, toluene, polychlorinated biphenyls (PCBs), malathion, parathion, organophosphates.
Mycobacterium sp	Benzene, branched hydrocarbons, cycloparaffins
Alcaligenes sp	Polychlorinated biphenyls, alkyl benzene, halogenated hydrocarbons.
Nocardia sp	Naphthalene, alkylbenzenes, phenoxyacetate.
Arthrobacter sp	Benzene, polycyclic aromatics, phenoxyacetate, pentachlorophenol.
Corynebacterium sp	Halogenated hydrocarbons, phenoxyacetate.
Bacillus sp	Long chain alkanes, phenylurea.
Candida sp	Polychlorinated biphenyls
Aspergillus sp	Phenols
Xanthomonas sp	Polycyclic hydrocarbons
Streptomyces sp	Halogenated hydrocarbons, phenoxyacetate.

Consortia of microorganisms for biodegradation:

A particular strain of microorganism may degrade one or more compounds. Sometimes, for the degradation of a single compound, the synergetic action of a few microorganisms (i.e. a consortium or cocktail of microbes) may be more efficient. For instance, the insecticide parathion is more efficiently degraded by the combined action of Pseudomonas aeruginosa and Psudomonas stulzeri.

Co-metabolism in biodegradation:

In general, the metabolism (breakdown) of xenobiotics is not associated with any advantage to the microorganism. That is the pollutant chemical cannot serve as a source of carbon or energy for the organism. The term co-metabolism is often used to indicate the non-beneficial (to the microorganism) biochemical pathways concerned with the biodegradation of xenobiotics. However, co- metabolism depends on the presence of a suitable substrate for the microorganism. Such compounds are referred to co-substrates.

Factors Affecting Biodegradation:
Several factors influence biodegradation. These include the chemical nature of the xenobiotic, the capability of the individual microorganism, nutrient and O2 supply, temperature, pH and redox potential. Among these, the chemical nature of the substrate that has to be degraded is very important.

Some of the relevant features are given hereunder:

i. In general, aliphatic compounds are more easily degraded than aromatic ones.

ii. Presence of cyclic ring structures and length chains or branches decrease the efficiency of biodegradation.

iii. Water soluble compounds are more easily degraded.

iv. Molecular orientation of aromatic compounds influences biodegradation i.e. ortho > para > meta.

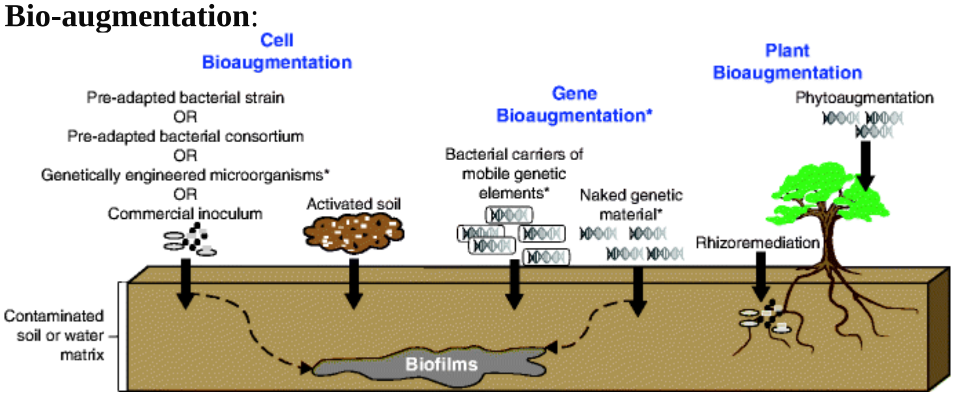
v. The presence of halogens (in aromatic compounds) inhibits biodegradation.

Besides the factors listed above, there are two recent developments to enhance the biodegradation by microorganisms.

Bio-stimulation:

This is a process by which the microbial activity can be enhanced by increased supply of nutrients or by addition of certain stimulating agents (electron acceptors, surfactants).

Bio-augmentation:



It is possible to increase biodegradation through manipulation of genes. More details on this genetic manipulation i.e. genetically engineered microorganisms (GEMs), are described later. Bio-augmentation can also be achieved by employing a consortium of micro-organisms.

Enzyme Systems for Biodegradation:
Several enzyme systems (with independent enzymes that work together) are in existence in the microorganisms for the degradation of xenobiotics. The genes coding for the enzymes of bio-degradative pathways may be present in the chromosomal DNA or more frequently on the plasmids. In certain microorganisms, the genes of both chromosome and plasmid contribute for the enzymes of biodegradation. The microorganism Pseudomonas occupies a special place in biodegradation.

List of Xenobiotics and Plasmids

Xenobiotic	*Name of plasmid in Pseudomonas*
Naphthalene	NAH
Xylene	XYL
Xylene and toluene	TOL, pWWO, XYL-K
Salicylate	SAL
Camphor	CAM
3-Chlorobenzene	pAC25

Recalcitrant Xenobiotics:
There are certain compounds that do not easily undergo biodegradation and therefore persist in the environment for a long period (sometimes in years). They are labeled as recalcitrant.

There may be several reasons for the resistance of xenobiotics to microbial degradation:

i. They may chemically and biologically inert (highly stable).

ii. Lack of enzyme system in the microorganisms for biodegradation.

iii. They cannot enter the microorganisms being large molecules or lack of transport systems.

iv. The compounds may be highly toxic or result in the formation highly toxic products that kill microorganisms.

There are a large number of racalcitrant xenobiotic compounds e.g. chloroform, freons, insecticides (DDT, lindane), herbicides (dalapon) and synthetic polymers (plastics e.g. polystyrene, polyethylene, polyvinyl chlorine).

It takes about 4-5 years for the degradation of DDT (75-100%) in the soil. A group of microorganisms (Aspergillus flavus, Mucor aternans, Fusarium oxysporum and Trichoderma viride) are associated with the slow biodegradation of DDT.

BIO-MAGNIFICATION:

The phenomenon of progressive increase in the concentration of a xenobiotic compound, as the substance is passed through the food chain is referred to as bio-magnification or bioaccumulation. For instance, the insecticide DDT is absorbed repeatedly by plants and microorganism.

When they are eaten by fish and birds, this pesticide being recalcitrant, accumulates, and enters the food chain. Thus, DDT may find its entry into various animals, including man. DDT affects the nervous systems, and it has been banned in some countries.

Biodegradation of Hydrocarbons:
Hydrocarbon are mainly the pollutants from oil refineries and oil spills. These pollutants can be degraded by a consortium or cocktail of microorganisms e.g. Pseudomonas, Corynebacterium, Arthrobacter, Mycobacterium and Nocardia.

Biodegradation of Aliphatic Hydrocarbons:
The uptake of aliphatic hydrocarbons is a slow process due to their low solubility in aqueous medium. Both aerobic and anaerobic processes are operative for the degradation of aliphatic hydrocarbons. For instance, unsaturated hydrocarbons are degraded in both anaerobic and aerobic environments, while saturated ones are degraded by aerobic process. Some aliphatic hydrocarbons which are reclacitrant to aerobic process are effectively degraded in anaerobic environment e.g. chlorinated aliphatic compounds (carbon tetrachloride, methyl chloride, vinyl chloride).

Biodegradation of Aromatic Hydrocarbons:
Microbial degradation of aromatic hydrocarbons occurs through aerobic and anaerobic processes. The most important microorganism that participates in these processes is Pseudomonas.

The biodegradation of aromatic compounds basically involves the following sequence of reactions:

1. Removal of the side chains.

2. Opening of the benzene ring.

Most of the non-halogenated aromatic compounds undergo a series of reactions to produce catechol or protocatechuate. The bioremediation of toluene, L-mandelate, benzoate, benzene, phenol, anthracene, naphthalene, phenanthrene and salicylate to produce catechol is shown in Fig. 59.1. Likewise, Fig. 59.2, depicts the bioremediation of quinate, p-hydroxymandelate, p-hydroxybenzoyl formate, p-toluate, benzoate and vanillate to produce protocatechuate.

Catechol and protocatechuate can undergo oxidative cleavage pathways. In ortho-cleavage pathway, catechol and protocatechuate form acetyl CoA (Fig. 59.3), while in meta-cleavage pathway (Fig. 59.4), they are converted to pyruvate and acetaldehyde. The degraded products of catechol and protocatechuate are readily metabolised by almost all the organisms.

Biodegradation of Pesticides and Herbicides:
Pesticides and herbicides are regularly used to contain various plant diseases and improve the crop yield. In fact, they are a part of the modern agriculture, and have significantly contributed to green revolution. The common herbicides and pesticides are propanil (anilide), propham (carbamate), atrazine (triazine), picloram (pyridine), dichlorodiphenyl trichloroethane (DDT) monochloroacetate (MCA), monochloropropionate (MCPA) and glyphosate (organophosphate). Most of the pesticides and herbicides are toxic and are recalcitrant (resistant to biodegradation). Some of them are surfactants (active on the surface) and retained on the surface of leaves.

Biodegradation of Halogenated Aromatic Compounds:
Most commonly used herbicides and pesticides are aromatic halogenated (predominantly chlorinated) compounds. The bio-degradative path-ways of halogenated compounds are comparable with that described for the degradation of non-halogenated aromatic compounds (Figs. 59.1, 59.2, 59.3 and 59.4). The rate of degradation of halogenated compounds is inversely related to the number of halogen atoms that are originally present on the target molecule i.e. compounds with higher number of halogens are less readily degraded.

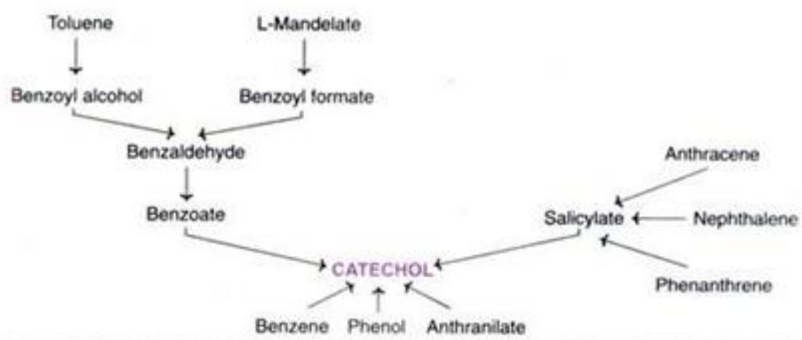


Fig. 59.1 : Bioremediation of certain aromatic compounds by bacteria to produce catechol.

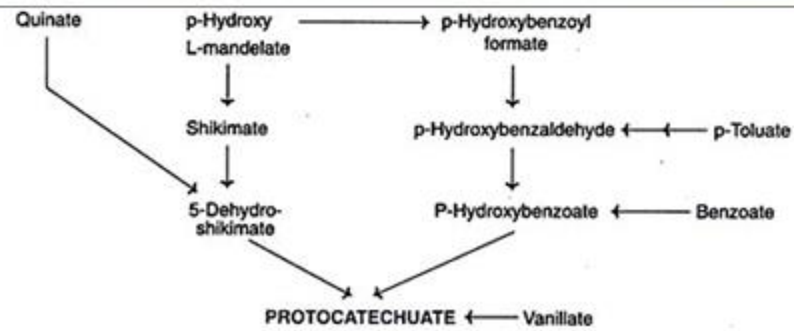
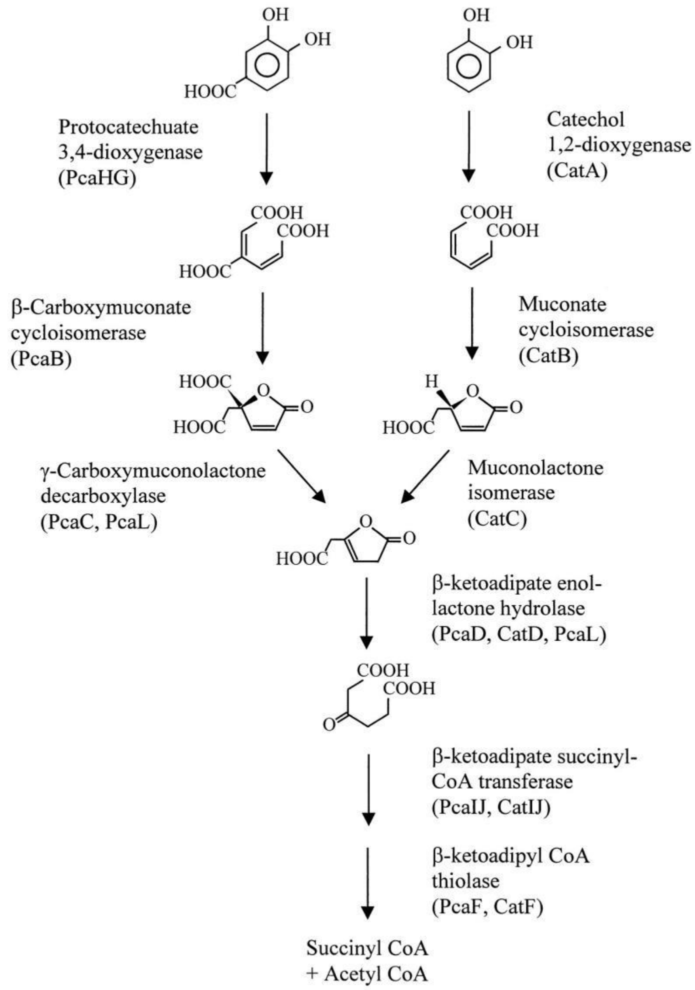


Fig. 59.2 : Bioremediation of certain organic compounds by bacteria to produce protocatechuate.

Bioremediation of Certain Aromatic Compounds Bioremediation of Certain Organic Compounds

Conversion of Catechol and Protocatechuateto Acetyl CoA and Succinate



Conversion of Catechol and Protocatechuate

Dehalogenation (i.e. removal of a halogen substituent from an organic compound) of halogenated compounds is an essential step for their detoxification. Dehalogenation is frequently catalysed by the enzyme di-oxygenase. In this reaction, there is a replacement of halogen on benzene with a hydroxyl group.

Most of the halogenated compounds are also converted to catechol and protocatechuate which can be metabolise. Besides Pseudomonas, other microorganisms such as Azotobacter, Bacillus and E. coli are also involved in the microbial degradation of halogenated aromatic compounds.

Biodegradation of Polychlorinated Biphenyls (PCBs):
The aromatic chlorinated compounds possessing biphenyl ring (substituted with chlorine) are the PCBs e.g. pentachlorobiphenyl. PCBs are commercially synthesized, as they are useful for various purposes — as pesticides, in electrical conductivity (in transformers), in paints and adhesives. They are inert, very stable and resistant to corrosion.

However, PCBs have been implicated in cancer, damage to various organs and impaired reproductive function. Their commercial use has been restricted in recent years, and are now used mostly in electrical transformers.

PCBs accumulate in soil sediments due to hydrophobic nature and high bioaccumulation potential. Although they are resistant to biodegradation, some methods have been recently developed for anaerobic and aerobic oxidation by employing a consortium of microorganisms. Pseudomonas, Alkali genes, Corynebacterium and Acinetobacter. For more efficient degradation of PCBs, the microorganisms are grown on biphenyls, so that the enzymes of biodegradation of PCBs are induced.

Biodegradation of Some Other Important Compounds:
Organo-nitro Compounds:
Some of the toxic organo-nitro compounds can be degraded by microorganisms for their detoxification.2, 4, 6-Trinitrotoluene (TNT):
Certain bacterial and fungal species belonging to Pseudomonas and Clostrium can detoxify TNT.

Synthetic detergents:
They contain some surfactants (surface active agents) which are not readily biodegradable. Certain bacterial plasmid can degrade surfactants.

Genetic Engineering for More Efficient Bioremediation:
Although several microorganisms that can degrade a large number of xenobiotics have been identified, there are many limitations in bio-remediation:

i. Microbial degradation of organic compounds is a very slow process.

ii. No single microorganism can degrade all the xenobiotics present in the environmental pollution.

iii. The growth of the microorganisms may be inhibited by the xenobiotics.

iv. Certain xenobiotics get adsorbed on to the particulate matter of soil and become unavailable for microbial degradation.

It is never possible to address all the above limitations and carry out an ideal process of bioremediation. Some attempts have been made in recent years to create genetically engineered microorganisms (CEMs) to enhance bio-remediation, besides degrading xenobiotics which are highly resistant (recalcitrant) for breakdown. Some of these aspects are briefly described.

Genetic Manipulation by Transfer of Plasmids:
The majority of the genes responsible for the synthesis of bio-degradative enzymes are located on the plasmids. It is therefore logical to think of genetic manipulations of plasmids. New strains of bacteria can be created by transfer of plasmids (by conjugation) carrying genes for different degradative pathways.

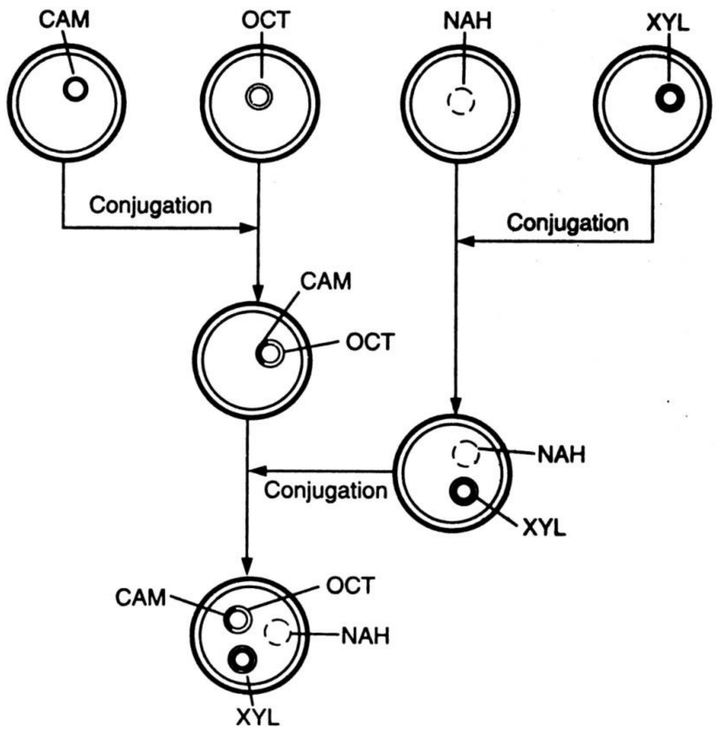
If the two plasmids contain homologous regions of DNA, recombination occurs between them, resulting in the formation of a larger fused plasmid (with the combined functions of both plasmids). In case of plasmids which do not possess homologous regions of DNA, they can coexist in the bacterium (to which plasmid transfer was done).

The first successful development of a new strain of bacterium (Pseudomonas) by manipulations of plasmid transfer was done by Chakrabarty and his co-workers in 1970s. They used different plasmids and constructed a new bacterium called as superbug that can degrade a number of hydrocarbons of petroleum simultaneously.

United States granted patent to this superbug in 1981 (as per the directive of American Supreme Court). Thus, superbug became the first genetically engineered microorganism to be patented. Superbug has played a significant role in the development of biotechnology industry, although it has not been used for large scale degradation of oil spills.

Creation of Superbug by Transfer of Plasmids:
Superbug is a bacterial strain of Pseudomonas that can degrade camphor, octane, xylene and naphthalene.

Creation of the Superbug



The bacterium containing CAM (camphor- degrading) plasmid was conjugated with another bacterium with OCT (octane-degrading) plasmid. These plasmids are not compatible and therefore, cannot coexist in the same bacterium. However, due to the presence of homologous regions of DNA, recombination occurs between these two plasmids resulting in a single CAM-OCT plasmid. This new bacterium possesses the degradative genes for both camphor and octane.

Another bacterium with XYL (xylene-degrading) plasmid is conjugated with NAH (naphthalene-degrading) plasmid containing bacterium. XYL and NAH plasmids are compatible and therefore can coexist in the same bacterium. This newly, produced bacterium contains genes for the degradation of xylene and naphthalene.

The next and final step is the conjugation of bacterium containing CAM-OCT plasmid with the other bacterium containing XYL and NAH plasmids. The newly created strain is the superbug that carries CAM-OCT plasmid (to degrade camphor and octane), XYL (xylene-degrading) plasmid and NAH (naphthalene-degrading) plasmid.

Development of Salicylate—Toluene Degrading Bacteria by Plasmid Transfer:
Some attempts have been made for the creation of a new strain of the bacterium Pseudomonas putida to simultaneously degrade toluene and salicylate. Toluene-degrading (TOL) plasmid was transferred by conjugation to another bacterium that is capable of degrading salicylate (due to the presence of SAL plasmid).

The newly developed strain of Pseudomonas can simultaneously degrade both toluene and salicylate. And this occurs even at a low temperature (0-5°C). However, the new bacterium is not in regular use, as more research is being conducted on its merits and demerits.

Genetic Manipulation by Gene Alteration:
Work is in progress to manipulate the genes for more efficient biodegradation. The plasmid pWWO of Pseudomonas codes for 12 different enzymes responsible for the meta-cleavage pathway (for the conversion of catechol and protocatechuate to pyruvate and acetaldehyde, for degradation of certain aromatic compounds. Some success has been reported to alter the genes of plasmid pWWO for more efficient degradation of toluene and xylene.

Genetically Engineered Microorganisms (GEMs) in Bioremediation:
Superbug is the first genetically engineered microorganism. Several workers world over have been working for the creation of GEMs, specifically designed for the detoxification of xenobiotics. Almost all these CFMs have been created by transferring plasmids.

Bio-surfactant Producing GEM:
A genetically engineered Pseudomonas aeruginosa has been created (by Chakarabarty and his group). This new strain can produce a glycolipid emulsifier (a bio-surfactant) which can reduce the surface tension of an oil water interface. The reduced interfacial tension promotes biodegradation of oils.

SCHOOL OF BIO AND CHEMICAL ENGINEERING

DEPARTMENT OF BIOTECHNOLOGY

UNIT – IV– Environmental & Agricultural Biotechnology – SBB2201

BIOMINING AND BIOLEACHING

Biomining is the broad term that describes the extraction of specific metals from their ores through biological means, usually microorganisms.

• It is an alternative to more traditional physicalchemical methods of mineral processing.

• The application of biomining processes predates by centuries the understanding of the role of

Microorganisms.

Biomining encompasses two processes:

1. **Bioleaching**: Bioleaching is the process by which metals are dissolved from ore

bearing rocks using microorganisms. Bioleaching is also called microbial leaching

2. **Biooxidation**: Biooxidation is a process by which the recovery of a metal is

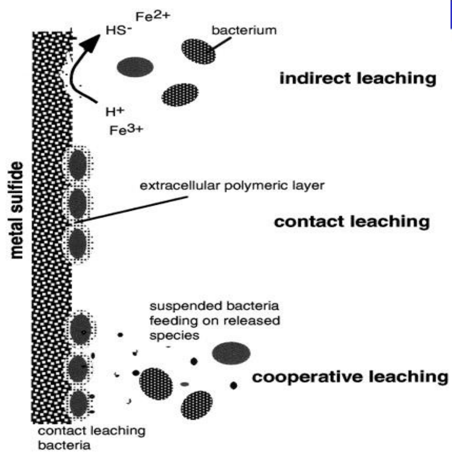
enhanced by microbial decomposition of the mineral, but the target metal is not

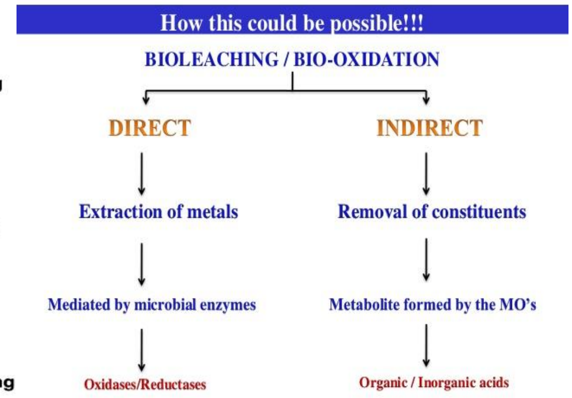
necessarily solubilized.

• In bioleaching the desired metal is leached from the ore. In biooxidation, the

undesired metals and other compounds are leached away from the ore.

Microorganisms Used in Bioleaching
The most commonly used microorganisms in bioleaching are:
• *Thiobacillus thiooxidans*
• *Thiobacillus ferrooxidans*
Other microorganism:
• *Sulfolobus* – Ca, As
• *Saccharomyces cerevisiae* – Cu, Pb, Sn
• *Penicillium simplicissium* - Cr
The bioleaching reaction mechanisms are of two types
• Direct bacterial leaching (Contact leaching)
• Indirect bacterial leaching





Indirect bacterial leaching

• In this process the microbes are not in direct contact with minerals, but bacteria produces strong oxidising agents such as ferric ion and sulfuric acid on oxidation of soluble iron or soluble sulfur respectively. • For indirect bioleaching, acidic environment is absolutely essential in order to keep ferric iron and other metals in solution.

• Acidic environment maintained by oxidation of iron, sulfur, metal sulphides or by dissolution of carbonate ions.

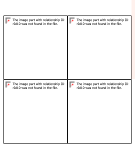
• Example: Bioleaching of uranium

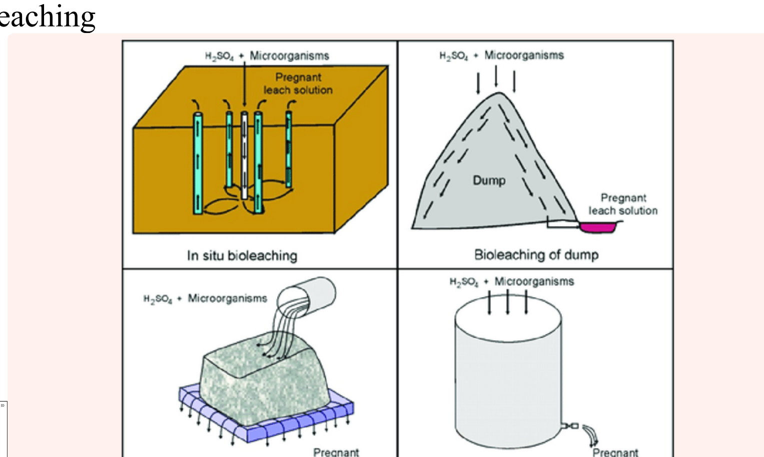
$U_2O + Fe_2(SO_4)_3 \rightarrow UO_2SO_4 + 2FeSO_4$

Types of Bioleaching

There are three commercial process used in bioleaching:

• Slope leaching
• Heap leaching
• In situ leaching





Slope Leaching

In this process the ores are first ground to get fine pieces and then dumped into large leaching dump. Water containing inoculum of thiobacillus is continuously sprinkled over the ore. Water is collected from the bottom and used to extract metals and generate bacteria in an oxidation pond.

Heap Leaching

In this process the ore is dumped into large heaps called leach heaps. Water containing inoculum of thiobacillus is continuously sprinkled over the ore. Water is collected from the bottom and used to extract metal and generate bacteria
in an oxidation pond

In-situ Bioleaching

- In this process the ore remains in its original position in the earth.
- Surface blasting of earth is done to increase the permeability of water.
- Water containing thiobacillus is pumped through drilled passage to the ores.
- Acidic water seeps through the rock and collects at the bottom.
- Again, water is pumped from bottom.
- Mineral is extracted and water is reused after generation of bacteria.

Benefits of Bioleaching

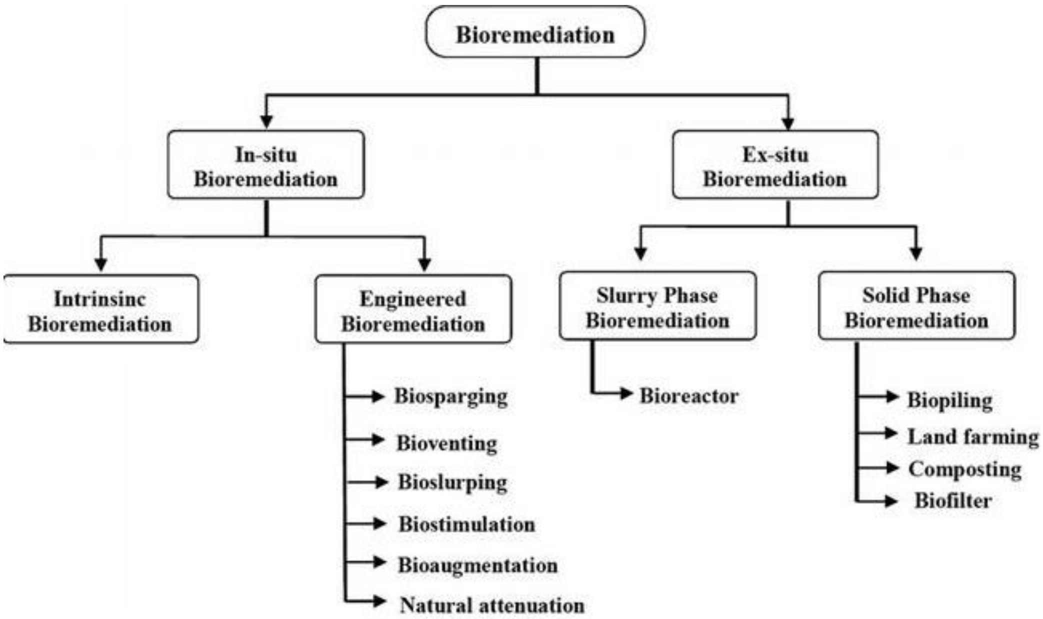
- Simple process.
- Inexpensive technique.
- No poisonous sulfur dioxide emission as in smelter.
- No need of high pressure and temperature.
- Ideal for low grade sulphide ores.
- Environment friendly process

BIOREMEDIATION

This is the process of using microorganisms to remove the environmental pollutants i.e. the toxic wastes found in soil, water, air etc. The microbes serve as scavengers in bioremediation. The removal of organic wastes by microbes for environmental clean-up is the essence of bioremediation. The other names used (by some authors) for bioremediation are bio-treatment, bio-reclamation and bio-restoration

TYPES OF BIOREMEDIATION:

The most important aspect of environmental biotechnology is the effective management of hazardous and toxic pollutants (xenobiotics) by bioremediation. The environmental clean-up process through bioremediation can be achieved in two ways—in situ and ex situ bioremediation.



IN SITU BIOREMEDIATION:

In situ bioremediation involves a direct approach for the microbial degradation of xenobiotics at the sites of pollution (soil, ground water). Addition of adequate quantities of nutrients at the sites promotes microbial growth. When these microorganisms are exposed to xenobiotics (pollutants), they develop metabolic ability to degrade them.

The growth of the microorganisms and their ability to bring out biodegradation are dependent on the supply of essential nutrients (nitrogen, phosphorus etc.). In situ bioremediation has been successfully applied for clean-up of oil spillages, beaches etc. There are two types of in situ bioremediation-intrinsic and engineered.

Intrinsic bioremediation:

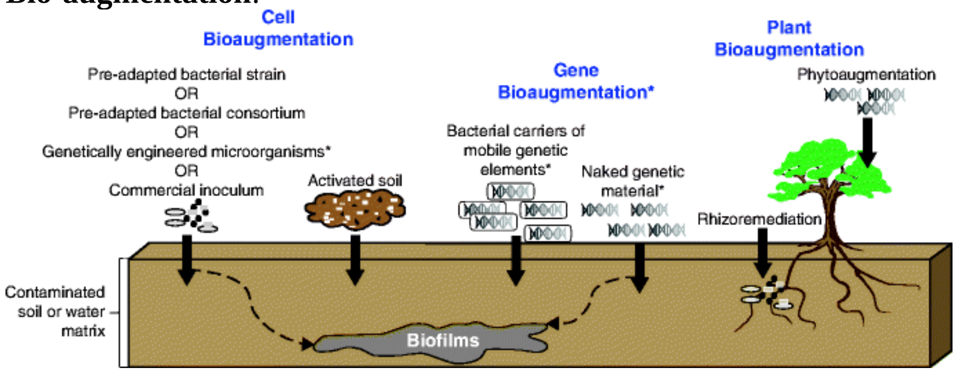
The inherent metabolic ability of the microorganisms to degrade certain pollutants is the intrinsic bioremediation. In fact, the microorganisms can be tested in the laboratory for their natural capability of biodegradation and appropriately utilized.

Engineered in situ bioremediation:

The inherent ability of the microorganisms for bioremediation is generally slow and limited. However, by using suitable physicochemical means (good nutrient and O2 supply, addition of electron acceptors, optimal temperature), the bioremediation process can be engineered for more efficient degradation of pollutants.

Bio-stimulation:
This is a process by which the microbial activity can be enhanced by increased supply of nutrients or by addition of certain stimulating agents (electron acceptors, surfactants).

Bio-augmentation:



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BIO-MAGNIFICATION:
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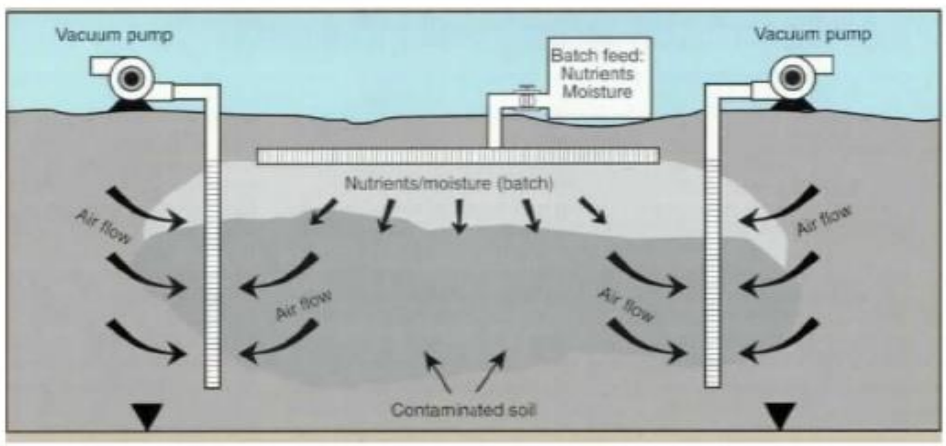
When they are eaten by fish and birds, this pesticide being recalcitrant, accumulates, and enters the food chain. Thus, DDT may find its entry into various animals, including man. DDT affects the nervous systems, and it has been banned in some countries

BIOVENTING

Bioventing



It is a natural biological process in which aerobically degradable compounds bio-degrade by providing oxygen to existing soil microorganisms. Air is slowly pumped into the contaminated area (in the unsaturated zone) through (vertical) injection wells.

The number, location, and depth of the wells depend on many geological factors and engineering considerations. It is a medium-long-term technology (few months-several years).

Enhanced by adding heat, water, nutrients and oxygen to increase the growth rate of MOs.

- An air blower may be used to push or pull air into the soil through the injection wells.
- Nutrients (e.g. Nitrogen and phosphorous) may be pumped into the soil through the injection wells.

Bio-venting requires:

Sufficient concentrations of native (pre-existing) MOs.

Air to be passed through the soil at the apt rate: Quickly enough to maintain aerobic conditions (for microbial activity) BUT slowly enough to minimise VOCs rising to the surface.

Soil pH ~ 6-8 and warm temperatures.

Uses:

- Treats VOCs, petroleum hydrocarbons (adsorbed residuals from LNAPLs non-chlorinated solvents and some pesticides & wood preservatives.
- Contaminants must be in the unsaturated zone of biologically active soil.
- Does not degrade inorganic contaminants (but can change their valence state causing adsorption, uptake, accumulation or stabilisation).

Limitations:

Not effective if water-table is very close to ground surface.

Not effective if extremely high moisture content (lowers the air permeability of the soil decreasing oxygen flow) or extremely low moisture content (too little inhibits microbial activity).

Not when very low temperatures.

Cost:

- highly variable dependent on soil surface area and soil type e.g.

A greater surface area requires more injection/extraction wells - increased cost.

Sand/gravelly strata (better air flow rate) requires less injection/extraction wells - reduced cost

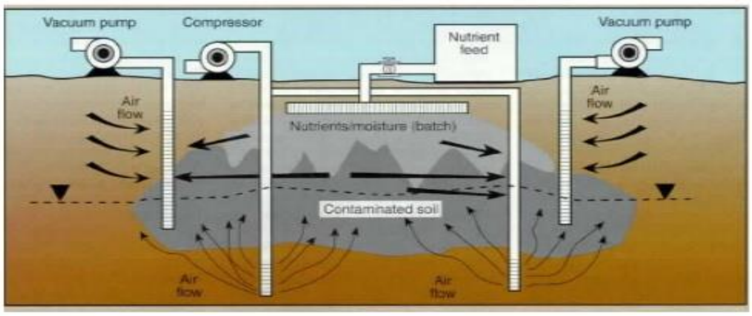
BIOSPARGING

Air is injected (horizontally and vertically) in channels through the contaminated soil, creating an 'underground stripper' that volatises contaminants for their removal.

The process applies to contaminated saturated areas (below the water table) and is commonly used to volatilise NAPLs.The injected air helps to flush (bubble) the contaminants up into the unsaturated zone to be removed by SVE.High air flow rates are used to maintain increased contact between ground water and soil - increasing quantity treated.

The process is enhanced by adding water, nutrients & heat (hot air injection wells).

Biosparging



<u>Uses:</u>

Target contaminant groups:VOCs and fuels

Methane can be added to the sparged air to enhance cometabolism of chlorinated organics.

<u>Limitations:</u>

May get uneven air flow through the saturated zone - could be uncontrolled movement of polluted vapours.

Soil heterogeneity may cause uneven treatment.

Air injection wells designed for site-specific conditions (geology/depth of contaminants)

Highly variable cost - dependent on the surface area & depth of contamination & no. of wells required.

EX SITU BIOREMEDIATION:

Ex-situ bioremediation techniques involve digging pollutants from polluted sites and successively transporting them to another site for treatment. Ex-situ bioremediation techniques are regularly considered based on the depth of pollution, type of pollutant, degree of pollution, cost of treatment and geographical location of the polluted site.

Solid-phase treatment

Solid-phase bioremediation is an ex-situ technology in which the contaminated soil is excavated and placed into piles. It is also includes organic waste like leaves, animal manures and agriculture wastes, domestic, industrial wastes and municipal wastes. Bacterial growth is moved through pipes that are distributed throughout the piles. Air pulling through the pipes is necessary for ventilation and microbial respiration. Solid-phase system required huge amount of space and cleanups require more time to complete as compared to slurry-phase processes. Solid-phase treatment processes include biopiles, windrows, land farming, composting

Slurry-phase bioremediation

Slurry-phase bioremediation is a relative more rapid process compared to the other treatment processes. Contaminated soil is combined with water, nutrient and oxygen in the bioreactor to create the optimum environment for the microorganisms to degrade the contaminants which are present in soil. This processing involves the separation of stones and rubbles from the contaminated soil. The added water concentration depends on the concentration of pollutants, the biodegradation process rate and the physicochemical properties of the soil. After completion of this process the soil is removed and dried up by using vacuum filters, pressure filters and centrifuges. The subsequent procedure is soil disposition and advance treatment of the resultant fluids.

Types of Ex-situ Bioremediation

Biopile

Bioremediation includes above-ground piling of dug polluted soil, followed by aeration and nutrient amendment to improve bioremediation by microbial metabolic activities. This technique comprises aeration, irrigation, nutrients, leachate collection and treatment bed systems. This specific ex-situ technique is progressively being measured due to its useful features with cost effectiveness, which allows operative biodegradation conditions includes pH, nutrient, temperature and aeration are effectively controlled. The biopile use to treat volatile low molecular weight pollutants; it can also be used effectively to remediate polluted very cold extreme environments. The flexibility of biopile allows remediation time to be shortened as heating system can be integrated into biopile design to increase microbial activities and contaminant availability thus increasing the rate of biodegradation . Additionally, heated air can be injected into biopile design to deliver air and heat in tandem, in order to facilitate enhanced bioremediation. Bulking agents such as straw saw dust, bark or wood chips and other organic materials have been added to enhance remediation process in a biopile construct. Although biopile systems connected to additional field ex-situ bioremediation techniques, such as land farming, bioventing, biosparging, robust engineering, maintenance and operation cost, lack of power supply at remote sites, which would facilitate constant air circulation in contaminated piled soil through air pump. Additional, extreme heating of air can lead to soil drying undertaking bioremediation, which will inhibit microbial activities and which stimulate volatilization than biodegradation

Land farming

Land farming is the simplest, outstanding bioremediation techniques due to its low cost and less equipment requirement for operation. It is mostly observed in ex-situ bioremediation, while in some cases of in-situ bioremediation technique. This consideration is due to the site of treatment. Pollutant depth is important in land farming which can be carried out ex-situ or in-situ. In land farming, polluted soils are regularly excavated and tilled and site of treatment speciously regulates the type of bioremediation. When excavated polluted soil is treated on-site, it is ex-situ as it has more in common than other ex-situ bioremediation techniques. Generally, excavated polluted soils are carefully applied on a fixed layer support above the ground surface to allow aerobic biodegradation of pollutant by autochthonous microorganisms [19]. Over all, land farming bioremediation technique is very simple to design and implement, requires low capital input and can be used to treat large volume of polluted soil with minimal environmental impact and energy requirement

Bioreactor

Bioreactor is a vessel in which raw materials are converted to specific product(s) following series of biological reactions. There are different operational modes of bioreactors, which include: batch, fed-batch, sequencing batch, continuous and multistage. Bioreactor provides optimal growth conditions for bioremediation. Bioreactor filled with polluted samples for remediation process. The bioreactor based treatment of polluted soil has several advantages as compared to ex-situ bioremediation procedures. Bioreactor-based bioremediation process having excellent control of pH, temperature, agitation and aeration, substrate and inoculum concentrations efficiently reduces bioremediation time. The ability to control and manipulate process parameters in a bioreactor implies that biological reactions. The flexible nature of bioreactor designs allows maximum biological degradation while minimizing abiotic losses .

Advantages of ex-situ bioremediation

- Suitable for a wide range of contaminants
- Suitability relatively simple to assess from site investigation data
- Biodegradation are greater in a bioreactor system than or in solid-phase systems because the contaminated environment is more manageable and more controllable and predictable.

Disadvantages

- Not applicable to heavy metal contamination or chlorinated hydrocarbons such as trichloroethylene.
- Non-permeable soil requires additional processing.
- The contaminant can be stripped from soil via soil washing or physical extraction before being placed in bioreactor.

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SCHOOL OF BIO AND CHEMICAL ENGINEERING

DEPARTMENT OF BIOTECHNOLOGY

UNIT – V–Environmental & Agricultural Biotechnology– SBB2201

BIOINOCULANT TECHNOLOGY
BIOFERTILIZER

Biofertilizers or microbial inoculants are carrier based ready to use live bacterial or fungal formulations,which on application to plants, soil or composting pits, help in mobilization of various nutrients by their biological activity.

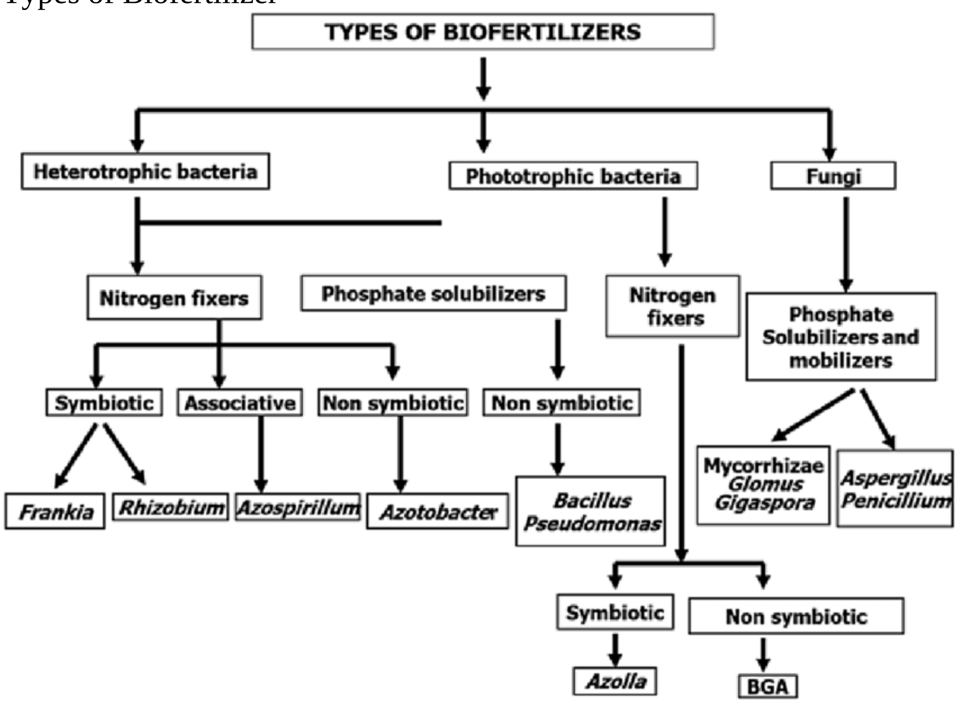
Biofertilizer are classified based on

1) For nitrogen -Rhizobium for legumes crops -Azotobacter/ Azospirilium for non legume crops -Acetobacter for sugarcane only. -Blue –Green Algae (BGA) and Azolla for low land paddy.

2) For phosphorus -Phosphate solubilizing Bacteria (PSB) for all crops to be applied with Rhizobium, Azotobacter, Azospirilium and Acetobacter

3) For enriched compost -Phosphate solubilizing Bacteria (PSB) and Azatobacter culture -Cellulolytic fungal culture

Types of Biofertilizer



Rhizobium

- Inoculum-vary in number, effectiveness and nitrogen fixation
- Inoculation improve nodulation, nitrogen fixation, crop growth and yield
- Inoculation increase yield is well documented

Essentials for inoculation in agriculture

- Population density of species specific rhizobia is low
- The same or symbiotically related legume is not grown in immediate past
- Wastelands have to be reclaimed
- Legumes follows a non-leguminous crop in a rotation
- Soil is poor in mineral N,
- Soils are acidic, alkaline and saline

Factors influence the rhizobium-legume symbiosis

Physico-chemical parameters,Soil temperature, moisture and reaction

Temperature tolerant strain of Rhizobia(up to 40o C)

Moisture stress affect the rhizobium survival

High salt concentration is detrimental, tolerance limit is 0.5 to 5% Na Cl

Fast growers are more tolerant (up to 0.34 M), legumes are more sensitive as compared to Rhizobium.

Acidity-Legumes are more sensitive as compared to Rhizobium.

May limit persistence in soil and reduce nodulation

High soil nitrogen levels, decrease nodulation

Other nutrients like P, K, Mo, Zn, Fe, Mg, S, Co, Ca, Cd, Mn, Cu

organic matter-Favorable effect on nodulation and N_2 fixation.

Indigenous heterotrophic microbes and predators reduces the activity of Rhizobia

Bdellovibrio, an intracellular bacterial parasite.

For increasing the crop yield certain parameters to be studied such as the quality of the inoculants,effective inoculants delivery system and formulation of the policy to exploit symbiotic nitrogen fixation successfully

AZOTOBACTER

These are the freeliving bacteria which grow well on a nitrogen free medium.These bacteria utilize
atmospheric nitrogen gas for their cell protein synthesis. Old population bacteria are encapsulated forms and increase resistance to heat, desiccation and adverse condition.

Factors affecting the growth of Azotobacter

Azotobacter sp are sensitive to acidic pH, high salts and temperature above 35^{o}C. There are six species of Azotobacter, A. chroococcumis are commonly found in Indian soils. Nitrogen fixation. The species of Azotobacterare known to fix from 10-30 mg of N/g of sugar in pure culture. This is a poor competitor for nutrients in soil.

Functions of Azotobacter

It fixes atmospheric nitrogen in the rhizosphere.The most important function is the production of indol acetic acid and gibberellins. Also produce thiamin, riboflavin, nicotine óIt improves seed germination and control plant diseases

Uses as biofertilizer

It is beneficial to cereals, vegetables and certain fruit crops.

It play an importance role in vegetable crops where farmer add sufficient quantities of manures as it is highly respiratory micro-organism and require about 1000 kg of organic carbon to fix 30 kg of N/ha.

Its inoculation increase the crop yield 15-20% over uninoculated treatment

AZOSPIRRILLUM

This is important biofertilizer for maize, paddy and wheat crops.

Increase the nitrogen content of soil after the crop harvest.

It substitute the nitrogen up to 25-30 percentand increase the crop yield by 20-25 percent over uninoculated control.

BLUE GREEN ALGAE AND AZOLLA

Increase the availability of nitrogen and phosphorus and certain micronutrients.
Moderate the soil pH conditions.
Azolla being green manure can substitute 40 -50 kg N per hectare.
Azolla is source of nutrients to poultry, fish and water animals**.**

PHOSPHATE SOLUBILIZER

Phosphate solubilizing bacteria (PSB) are beneficial bacteria capable of solubilizing inorganic phosphorus from insoluble compounds. They have the capability of dissolving soil phosphorus which have been adsorbed and can mineralize organic P to become inorganic P, hence increasing the avalibility of P in the soil

Phosphate solubilizing microbes are
Bacteria-*Bacillus* spp
Fungi- *Penicillium, Aspergillus*
Efficiency of bacteria is more than fungal strains.
Substitute 15-20 percent dose of phosphorus
Mechanism is by phosphatase production

PHOSPHATE MOBILIZER

Gigaspora, Acullosporaare the important sp.
Phosphorus is being mobilized.
Increase the availability of P, Mg, Ca and Zn.
VAM do not produce phosphatase

FORMULATION TRAITS OF BIOFERTILIZER

The product must be carrier based or liquid formulation, capable of holding very high population of specific microorganisms for size able period of time**.**
Incase of carrier based formulations the product should have30-50% of moisture throughout the shelf life period to sustain microbial population.
For carrier based formulations the microbial population should be in the range of107 to 109 cells/g of moist product.
In case of liquid formulations the cell load should be in the range of 1x108
To 1x1010 during the entire period of shelflife.
It should be free from other contaminating microorganisms.
The microbial strain present in the product should be able to produce adequate nodulation in case of Rhizobium, be able to fix atleast 10-15mg of N/g ofcarbon source used in case of free living N_2 fixers and be capable of solubilizing significant quanity of fixed soil P
It should have sufficient shelf life minimum -6 months for carrier based and 12 months for liquid

PRODUCTION TECHNOLOGY

It involves 3 steps:
a) Isolation and identification of appropriate strains of targeted microorganisms.
b) Up-scaling of microbial biomass.
c) Impregnation of carrier with fully grown microbial broth or immobilization of grown cells to obtain liquid formulations

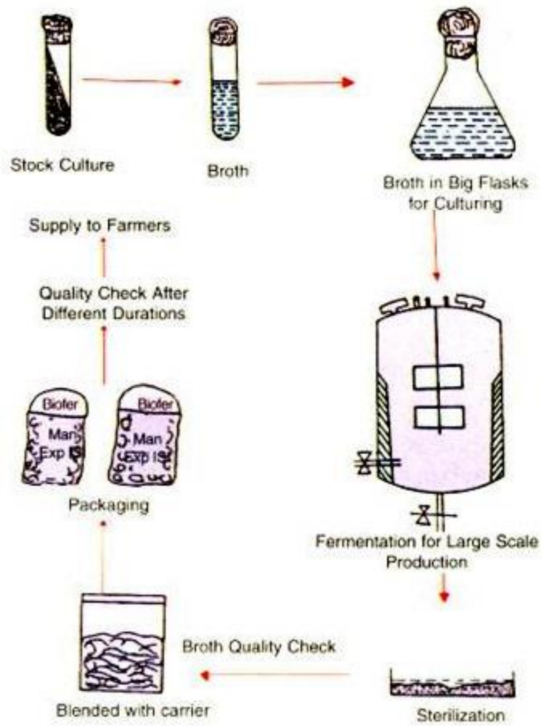


Fig. 34.1 : Commercial production of biofertilizer.

Isolation and identification of strains:

The success of any biofertilizer in the field primarily dependsuponthestrainofthe micro-organisms used in theproduct. The strain,besides possessing specific attributes (such as host specificity, nodulation potential and N_2fixation potential) should also have the ability tocolonize the soil and rhizosphere, be able to successfully compete with the native soil microorganisms and should have enough capacity to survive in the soil for long time in association with other soil microorganisms.

Up-scaling of biomass

To deliver a very high population of microorganismsin biofertilizers, it is very much essential to cultivate these microorganisms under appropriate conditions to achieve very high population Per unit of growing medium. Usually a final cell count of >109 cells per ml of broth should be achieved. This is being done in laboratories under controlled conditions in small glass containers (for small scale production) or large scale fermenters (for large scale production units).

Preparation of carrier based formulations:

Once the optimum growth of microbial cells is achieved in the multiplication process,it has to be mixed with the suitable carrier material, which can provide ideal home for these micro-organisms for about 6 months to 12 months time. The first step in this process involves the selection of suitable carrier materials.

The carrier materials are as follows:

Peat, charcoal, lignite, charcoal-soil mixture, press mud, vermiculite and **coco peat** have been identified as good carrier materials.

Ideal carrier should be:

Cheap in cost
Locally available
High organic content
No toxic chemical to inoculant
Water holding capacity of more than 50%
Easy process

For preparation of finished goods the pure bacterial liquid containing very high population is required. Microorganisms is mixed with the carrier material to obtain moist powdered formulation, which is packed in polythene bags and supplied to the farmers. Depending upon the facilities, mixing of bacterial liquid with carrier.

Material is being done either manually or under complete sterile conditions. Packets prepared by manual mixing method have shorter shelf life (2-3 months), while packets prepared under complete sterile conditions have longer shelf life (6-12months).

Preparation of liquid formulations:

In this process, in some cases the fermentation is taken up with specialized formulations with cell immobilizers added at different stages of growth. Finally prepared brothwith immobilized cell shaving A cell count of $>1x10^{10}$ is harvested and packed in bottles. In somecases,the vegetative cells after being cultivated to desired level are converted into cystsorspores. These spores are further treated to keep the minimal dormant position. Final preparation is packed in bottles. If sterile conditions can be arranged then instead of bottles plastic pouches can also be prepared.

APPLICATION

There are three major ways in applying the biofertilizer to the field namely

- **Seed treatment**
- **Seedling root dip treatment**
- **Soil treatment**

Seed treatment- Suspend200g each of Nfixing and PSB in 300-400ml of water and mix thoroughly. Pour this slurry on10to12kg of seed and mix by hands, till all the seeds are uniformly coated. Dry the treated seeds in shade and sow immediately. For acidic and alkaline soils it is always it is advisable to use1 kg of **slacked lime or gypsum** powder respectively for coating the wet biofertilizer treated seeds.

Seedling root dip treatment

Suspend 1 to 2 kg each of nitrogen fixing (Azotobacter/Azospirillum) and PSB into just Sufficient quantity of water (5-10lit depending upon the quantity of seedlings required to be planted in one acre). Dip the roots of seedlings in this suspension for 20-30 min before transplanting. In case of paddy make a sufficient sizebed (2mtx1.5mtx0.15mt)in the field, fill it with 5cm of water and suspend 2kg each ofAzospirillum and PSB, mix thoroughly. Then dip the roots of seedlings in this bed for 8-12 hours(overnight) and then transplant.

Soil treatment

For soil treatment depending upon the total number of plants per acre 2-4 kg of Azotobacter/Azospirillum and2-4kg of PSB are requiredfor oneacre. Mix two types of biofertilizer in 2-4liters of water separately and sprinkle this suspension on two separate heaps of 50-100 kg of compost. Mix the two heaps separately and leave for incubation overnight. After12 hours,mix the two heaps together. For acidic soils mix 25 kg limes with this mixture.

VERMICOMPOST

Vermicomposting

Vermicomposting is a type of composting in which certain species of earthworms are used to enhance the process of organic waste conversion and produce a better end-product. It is a mesophilic process utilizing microorganisms and earthworms. Vermicomposting is an ecofriendly, low cost effective and a effective way to recycle agricultural and kitchen waste. The application of vermicompost not only adds plant nutrients (macro and micro) and growth regulators but also increases soil water retention, microbial population, nutrient content and carbon content of the soil. Vermicompost refers to an organic manure produced by earthworms.

It is mixture of worm castings, organic material including humus, live earth worms and their cocoons and other organisms. Vermicomposting is a appropriate technique for the disposal of non-toxic solid and liquid organic wastes. It helps in cost effective and efficient recycling of animal wastes agricultural residual and industrial wastes using low energy.

Earthworms

Earthworms feed on any organic waste, consume two to five times their body weight and after using 5-10 per cent of the feed stock for their growth, excrete the mucus coated undigested matter as wormcasts worm cast consists of organic matter that has undergone physical and chemical breakdown through the activity of the muscular gizzard which grinds the material to a particle size of 1-2 micron. The nutrients presents in wormicast are readily soluble in water for the uptake of plants. Worm casts are rich source of macro and micro nutrients, vitamins, enzymes, antibiotics, growth harmones and immobilized microfloras.

Earthworms are cosmopolitan in distribution. They are important invertebrate macrofauna. Macroinvertebrates, in general, play a definite and prominent role in regulating soil process; among them, earthworms play a vital role in maintaining soil fertility and efficient nutrient cycling.

Earthworms also act as biological indicators of the soil type and its properties.They also serve to detect toxicity of industrial chemicals and act as indicators of soil fertility because the presence of earthworms support healthy populations of bacteria, actinomycetes, fungi, protozoans, insects, spiders, millipedes and a host of other organisms that are essential for sustaining healthy soil.

Earthworms species for vermicompost

1. *Lampito mauriti* -Sandy loam soil
2. *Octochaetona serrata*- Clay soil
3. *Lumbricus terrestris* -Deep boarings
4. *Allolobophora caliginosa* -shallow boarings
5. *Eudrilus eugeniae*
6. *Eisenia foetida*
7. *Perionyx excavatus*
8. *Pheretima elongate*

The *Eisenia foetida* an epigeic species of earthworms is used for vermicomposting. The general characteristics of *Eisenia foetida* are as under:

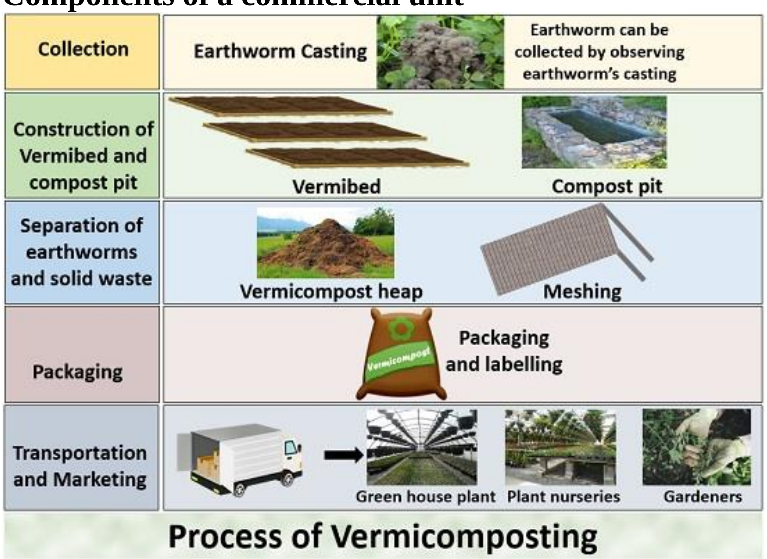
Habit: Epigeic
Size: 8-10 cm
Diameter: 0.8-1.0 mm
Duration of life: 70 days
Temperature range: 15-40 0C
Approximate weight of single earthworm: 1.0-3.0 g
Moisture tolerance range: 30-70%
Habit and Habitats: Living on surface and organic matter rich areas.

Process of vermicomposting

A wide range of organic residues, such as straw, husk, leaves, stalks, weeds etc can be converted into vermicompost. Other potential feedstock for vermicompost production are livestock wastes, poultry litter, dairy wastes, food processing wastes, organic fraction of MSW, bagasse, digestate from biogas plants etc. Earthworms consume organic wastes and reduce the volume by 40–60 percent. Each earthworm weighs about 0.5 to 0.6 gram, eats waste equivalent to its body weight and produces cast equivalent to about 50 percent of the waste it consumes in a day. The moisture content of castings ranges between 32 and 66 percent and the pH is around 7. The level of nutrients in compost depends upon the source of the raw material and the species of earthworm.

There are nearly 3600 types of earthworms which are divided into burrowing and non-burrowing types. **Red earthworm species, like *Eisenia foetid***a, and are most efficient in compost making. The non-burrowing earthworms eat 10 percent soil and 90 percent organic waste materials; these convert the organic waste into vermicompost faster than the burrowing earthworms. They can tolerate temperatures ranging from 0 to 40°C but the regeneration capacity is more at 25 to 30°C and 40–45 percent moisture level in the pile. The burrowing types of earthworms come onto the soil surface only at night. These make holes in the soil up to a depth of 3.5 m and produce 5.6 kg casts by ingesting 90 percent soil and 10 percent organic waste.

Components of a commercial unit



Location

Suburbs of cities and villages around urban centres can be ideal locations for practice of vermicomposting on a large scale, from the view point of availability of raw material and marketing of the produce. As use of the compost is said to have ameliorative effect on product from fruit, flower and vegetable crops, vermicomposting units may be located in areas with concentration of fruit and vegetable growers and floriculture units.

Sheds

For a vermi-composting unit, whether small or big, this is an essential item and is required for having the vermi beds. They could be of thatched roof supported by bamboo rafters and purlins, wooden trusses and stone pillars. If the size is so chosen as to prevent wetting of beds due to rain on a windy day, they could be open sheds. While designing the sheds adequate room has to be left around the beds for easy movement of the labour attending to the filling and harvesting the beds.

Vermi-beds

Normally the beds are 75 cm - 90 cm thick depending on the provision of filter for drainage of excess water. The entire bed area could be above the ground. Care should be taken to make the bed with uniform height over the entire width to the extent possible to avoid low production owing to low bed volumes. The bed width should not be more that 1.5 m to allow easy access to the centre of the bed.

Land

About 0.5-1 acre of land will be needed to set up a vermiculture production cum extension centre. The centre will have at least 8-10 sheds each of about 180-200 sq.ft. It should also have a bore well, and pump set or watering arrangement and other equipments as described in the scheme economics. The land can be taken on lease of at least 10-15 years. Even sub marginal land also will serve the purpose.

Seed Stock

This is an important item requiring considerable investment. Though the worms multiply fast to give the required numbers over a period of 6 months to a year, it may not be wise to wait till such a time having invested on the infrastructure heavily. Thus, worms @ 350 worms per m3 of bed space should be adequate to start with and to build up the required population in about two cycles or three without unduly affecting the estimated production.

Fencing and Roads/Paths

The site area needs development for construction of structures and development of roads and pathways for easy movement of hand-drawn trolleys/wheel barrows for conveying the raw material and the finished products to and from the vermi-sheds. The entire area has to be fenced to prevent trespass by animals and other unwanted elements. These could be estimated based on the length of the periphery of the farm and the length and type of roads/paths required. The costs on fencing and formation of roads should be kept low as these investments are essential for a production unit, yet would not lead to increase in production.

Water Supply System

As the beds have always to be kept moist with about 50% moisture content, there is need to plan for a water source, lifting mechanism and a system of conveying and applying the water to the vermi-beds. Drippers with round the clock flow arrangement would be quite handy for continuous supply and saving on water. Such a water supply/application system requiring considerable initial investment, however, reduces the operational costs on hand watering and prove economical in the long run. The cost of these items depend on the capacity of the unit and the type of water supply chosen.

Machinery

Farm machinery and implements are required for cutting (shredding) the raw material in small pieces, conveying shredded raw material to the vermi-sheds, loading, unloading, collection of compost, loosening of beds for aeration, shifting of the compost before packing and for air drying of the compost, automatic packing and stitching for efficient running of the unit. Costs of providing necessary implements and the machinery have to be included in the project cost. ;;’

Transport

For any vermi-composting unit transport arrangement is a must. When the source of raw material is away from the production unit, an off-site transport becomes major item of investment. A large sized unit with about 1000 tonnes per annum capacity may require a 3-tonne capacity mini-truck. With small units particularly with the availability of raw material near the site, expending on transport facility may become infructuous. On-site transport facilities like manually drawn trolleys to convey raw material and finished products between the storage point and the vermi-compost sheds could also be included in the project cost.

Furniture

A reasonable amount could also be considered for furnishing the office-cum-stores including the storage racks and other office equipment. These enhance the efficiency of operations.

Operational Costs

In order to operate the unit, expenditure on some items have to be incurred on a recurring basis. These items include salaries of the staff, wages to the labourers, cost of raw material, fuel cost on transport of raw materials and finished goods, packing material cost, repairs and maintenance, power, insurance, etc. The number of office personnel and labourers have to be decided breaking each activity into a number of sub-activities and for each sub-activity estimating the work involved and the capacity of the labour to finish the work in a given time. The number of persons should be so chosen to keep them engaged throughout by providing enough persons at various work points like stores, vermi-beds and equipping them with adequate number of implements to avoid undue waiting.

Applications of Vermicompost

The worm castings contain higher percentage of both macro and micronutrients than the garden compost. Apart from other nutrients, a fine worm cast is rich in NPK which are in readily available form and are released within a month of application. Vermicompost enhances plant growth, suppresses disease in plants, increases porosity and microbial activity in soil, and improves water retention and aeration.

Vermicompost also benefits the environment by reducing the need for chemical fertilizers and decreasing the amount of waste going to landfills. Vermicompost production is trending up worldwide and it is finding increasing use especially in Western countries, Asia-Pacific and Southeast Asia.

A relatively new product from vermicomposting is vermicompost tea which is a liquid produced by extracting organic matter, microorganisms, and nutrients from vermicompost. Unlike vermicompost and compost, this tea may be applied directly to plant foliage, reportedly to enhance disease suppression. Vermicompost tea also may be applied to the soil as a supplement between compost applications to increase biological activity.

Uses

The wastes are pulverized as they pass through the worm,
Surface area of the material increases which in turn helps as base for nutrients
Supply nutrients and growth enhancing hormones to plants
Improves the soil structure leading to increase in water and nutrient holding capacities of soil. Chemical fertilizer in moderate doses can go along with vermicomposting.

Wasteland reclamation using microbes

Wasteland reclamation of wasteland means re-claiming it or to use it for productive purpose. Wasteland reclamation is the process of turning barren, sterile wasteland into something that is fertile and suitable for habitation and cultivation.

The green revolution is the result of intensive agriculture with extensive use of chemical fertilizers. In general, the production of chemical fertilizers has led to depletion of fossil fuel environmental pollution and occupational hazards. In particular, the application of chemical fertilizers in agricultural fields reduces the fertility of the soil.

Due the removal process of desired mineral materials, soil textures have been destroyed, various nutrient cycles have been disturbed, and microbial communities have been altered, affecting vegetation and leading to the destruction of wide areas of land in many countries. Therefore, soil restoration of abandoned mining lands became a very important part of sustainable development strategies.

Contiuous agricultural activities, remove the top soil layer transforming these lands less fertile and more susceptible for soil hazards. Growing cover crops is one of the best practices to improve the organic matter content of the soil and hence soil health and quality. Vegetation layers—besides adding plant material to the soil contributing to its organic matter replenishment—can also fix N in the soil, moderate soil temperature protecting hence soil organisms and can provide habitat for beneficial insects and other organisms. The fertility of soil can be enhanced by Stimulate existing soil microbes, improve the nutrient and water efficiencies, introduce stress tolerance plants, add biofertilizer to increase the nutrient, with agricultural crop rotation and high fertilization doses depending on the location.

BIOPESTICIDE

Biopesticides are certain types of pesticides derived from such natural materials as animals, plants, bacteria, and certain minerals. For example, canola oil and baking soda have pesticidal applications and are considered biopesticides. The micro-organisms used as bio-pesticides are viruses, bacteria, protozoa, fungi and mites. Some of the bio-pesticides are being used on a commercial scale.

Most important example is the soil bacterium, Bacillus thuringiensis (Bt). Spores of this bacterium possess the insecticidal Cry protein.Therefore, spores of this bacterium kill larvae of certain insects. The commercial preparations of B. thuringiensis contain a mixture of spores, Cry protein and an inert carrier.This bacterium was the first bio-pesticide to be used on a commercial scale in the world, and is the first bio-pesticide being produced on a commercial scale in India.

Bio-pesticides are of two types: bio-herbicides and bio-insecticides.

(i) **Bio-herbicides:**

Herbicides are chemicals that are used for inhibiting the growth of plants in unwanted places. Herbicides used for controlling weeds in the cultivated areas are called weedicides. A number of risks are involved in the use of chemical herbicides. This can be avoided if herbicide resistance can be introduced in the crop plants. It is possible through genetic engineering or recombinant DNA technology. Transgenic Tomato and Tobacco plants have been developed which show tolerance to specific herbicides.

Certain crop plants do not allow the weeds to grow nearby. They are called smoother crops, e.g., Barley, Rye, Sorghum, Millet, Sweet clover, Alfalfa, Soybean, Sunflower. Smoother crops eliminate weeds through chemicals. Crop rotation with these crops will naturally reduce the incidence of weeds.

Another way of weed control is the introduction of specific insects which feed on the weeds. Extensive growth of Opuntia in India and Australia was checked through the intro-duction of its natural herbivore, cochineal insect (*Cactoblastis cactorum*). Similarly, growth of Hypericum perforatum or Klamath weed was checked by U.S.A. through the introduction of Chrysolina beetles.

An organism which controls or destroys unwanted plant growth without harming the useful plant is called bioherbicide. The first bioherbicide happened to be mycoherbicide. It was put to use in 1981. The herbicide is Phytophthora palmivora. The fungus does not allow the Milkweed Vine to grow in Citrus orchards. Growth of Eichhornia crassipes (Water Hyacinth) is being controlled by Cercospora rodmanii in USA and Alternaria eichhorniae in India.

Puccinia chondrilla has controlled the growth of skeleton weed, Chondrilla juncea in Australia. Fungal spores are now available to be sprayed over weeds for their elimination. Two of them are ‘Devine’ and ‘Collego’. The spores are ideal for marketing because they can tolerate adverse conditions and can remain viable for long periods.

(ii) **Bio-insecticides:**

Bio-insecticides are those biological agents that are used to control harmful insects. They include the following.

(a) **Predators:**

Destructive insects or plant pests can be brought under control through introduction of their natural predators. The predators should be specific and unable to harm the useful insects. Introduction of ladybugs (Lady Bird Beetles) and Praying Mantis has been successful in combating scale insects or aphids which feed on plant sap.

(b) **Parasites and Pathogens**:

This is alternate biological control of plant pests through the search of their natural parasites and pathogens. They include viruses, bacteria, fungi and insect parasitoids. Parasitoids are organisms that live as parasites for some time (as early or larval stage) and free living at other times, e.g., Trichogramma. Nucleopolyhedrovirus (NPV) are species specific.

For example, Baculovirus heliothis (a virus) can control Cotton bollworm (Heliothis Zea). Similarly, Bacillus thuringenesis (a bacterium) is effective against the cabbage looper (Trichoplausiani) and Entomophthora ignobilis (a fungus) the green peach aphid of Potato

(Myzus persicae). In U.S.S.R. the fungus Beauveria bassiana has been successfully employed in controlling Potato beetle and Codling moth.

(c) **Natural Insecticides:**

They are insecticides and related pesticides which are ob-tained from microbes and plants. A number of natural insecticides are available. The common ones include (i) Azadirachtin from Margosa or Neem (Azadirachta indica). It occurs in Margosa extract. Spray of the same keeps away the Japanese beetles and other leaf eating pests because of the antifeedant property of azadirachtin. (ii) Rotenones. They are powerful insecticides which are harmless to warm blooded animals. Chinese are believed to be first to discover their insecticidal properties. Rotenones are obtained from the roots of Derris elliptica and Lonchocarpus nicou. (iii) Squill. The red variety of Sea Onion (Red Squill, Ureginea maritima) produces a radicide which does not have any harmful effect on other animals, (iv) Nicotine. It is obtained from Nicotiana species. The purified chemical is highly poisonous. Nicotine sulphate is one of the most toxic insecticides, (v) Pyrethrum.

It is an insecticide which is obtained from the inflorescence of Chrysanthemum cinerarifolium (Dalmation Pyrethrum), C. coccineum and C. marshallii. The active compounds are pyrethrin and cinerin. Pyrethrin is also used in fly sprays, aerosols, mosquito coils, etc. (vi) Thurioside. It is a toxin produced by bacterium Bacillus thuringenesis. The toxin is highly effective against different groups of insects like moths, flies, mosquitoes and beetles. It does not cause any adverse environmental pollution or disturbance.

Thurioside occurs as crystals in the bacterium. It kills the susceptible insects through inhibiting ion transport in the midgut, formation of pores in gut epithelium, swelling and bursting of cells, (vii) Transgenic Plants. They are crop plants which are modified through genetic engineering to develop natural resistance to insects by inserting cry genes of Bacillus thuringenesis into them, e.g., Bt Cotton. Similarly, transgenic Tomato has been developed which is resistant to homworm larvae.

ORGANIC MANURE

The organic materials most commonly used to improve soil conditions and fertility include farm yard manure (FYM), animal wastes, crop residues, urban organic wastes (either as such or composted), green manures, bio-gas spent slurry, microbial preparations, vermicompost and biodynamic preparations. Sewage sludge and some of the industrial wastes also find application in agriculture.

List of Organic Manures

Manure	Percentage content		
	Nitrogen (N)	Phosphoric acid (P_2O5)	Potash (K_2O)
Coir pith	1.20	1.20	1.20
Blood meal	10-12	1.2	1.0
Press mud	1-1.5	4-5	2-7
Bone meal			
1)Raw bone meal	3-4	20-25	-
2)Steamed bone meal	1-2	25-30	-
Fish meal	4-10	3.9	0.3-1.5
Animal refuse	0.3-0.4	0.1-0.2	0.1-0.3
Cattle dung, fresh	0.4-0.5	0.3-0.4	0.3-0.4
Horse dung ,fresh	0.5 -0.5	0.4-0.6	0.3-1.0
Poultry manure, fresh	1.0-1.8	1.4-1.8	0.8-0.9
Sewage sludge, dry	2.0-3.5	1.0-5.0	0.2-0.5
Sewage sludge, activate dry	4.0-7.0	2.1-4.2	0.5-0.7
Cattle urine	0.9-1.2	trace	0.5-1.0
Horse urine	1.2-1.5	trace	1.3-1.5
Human urine	0.6-1.0	0.1-0.2	0.2-0.3
Sheep urine	1.5-1.7	trace	1.8-2.0
Ash, coal	0.73	0.45	0.53
Ash, household	0.5-1.9	1.6-4.2	2.3-12.0
Ash, wood	0.1-0.2	0.8-5.9	1.5-36.0
Rural compost ,dry	0.5-1.0	0.4-0.8	0.8-1.2
Urban compost ,dry	0.7-2.0	0.9-3.0	1.0-2.0
Farmyard manure ,dry	0.4-1.5	0.3-0.9	0.3-1.9
Filter-press cake	1.0-1.5	4.0-5.0	2.0-7.0
Rice hulls	0.3-0.5	0.2-0.5	0.3-0.5
Groundnut husks	1.6-1.8	0.3-0.5	1.1-1.7
Banana, dry	0.61	0.12	1.00

Rice hulls	0.3-0.5	0.2-0.5	0.3-0.5
Groundnut husks	1.6-1.8	0.3-0.5	1.1-1.7
Banana, dry	0.61	0.12	1.00
Cotton	0.44	0.10	0.66
Maize	0.42	1.57	1.65
Paddy	0.36	0.08	0.71
Tobacco	1.12	0.84	0.80
Pigeon pea	1.10	0.58	1.28
Wheat	0.53	0.10	1.10
Sugarcane trash	0.35	0.10	0.60
Tobacco dust	1.10	0.31	0.93
Tree leaves, dry			
Calotropis gigantea	0.35	0.12	0.36
Careya arborea	1.67	0.40	2.20
Cassia ariculata	0.98	0.12	0.67
Dillenia pentagyna	1.34	O.50	3.20

ADVANTAGES OF ORGANIC MANURES

1. Organic manure provides all the nutrients that are required by plants but in limited quantities.

2. It helps in maintaining C:N ratio in the soil and also increases the fertility and productivity of the soil.

3. It improves the physical, chemical and biological properties of the soil.

4. It improves both the structure and texture of the soils.

5. It increases the water holding capacity of the soil.

6. Due to increase in the biological activity, the nutrients that are in the lower depths are made available to the plants.

7. It acts as much, thereby minimizing the evaporation losses of moisture from the soil.