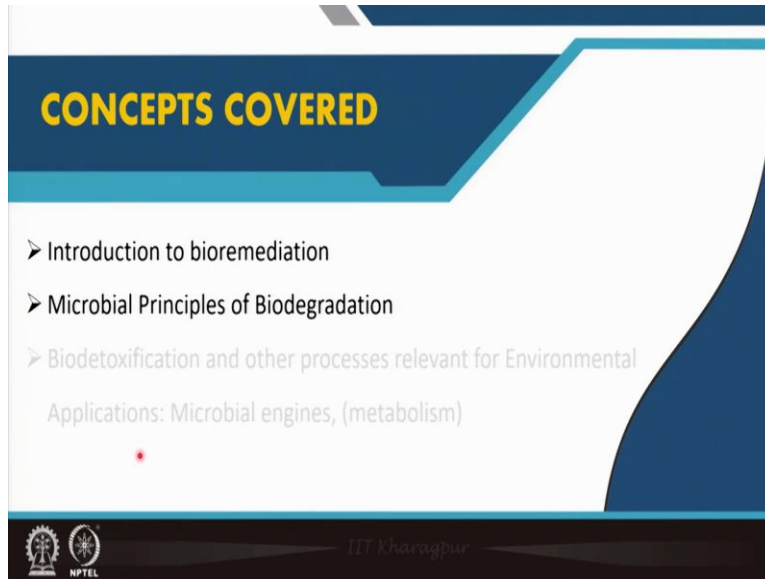


Environmental Biotechnology
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Lecture – 35
Bioremediation

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Welcome to the next lecture of this course environmental biotechnology in this course we are going to start a new topic that is bioremediation. So, this particular lecture will have the following concepts to be discussed. So, we will have the introduction to bioremediation and then followed by the microbial principles of biotic radiation.

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Advancement of microbial ecology paved the road for Environmental Biotechnology : Bioremediation

Pollution treatment technology that uses microbial metabolic processes to reduce, eliminate, contain, or transform various contaminants present in soils, sediments, water, or air to benign products

1900: Municipal wastewater treatment; 1950: Industrial wastewater treatment process; "Microbial infallibility hypothesis" 1970: Bioremediation of gasoline contaminated aquifer; 1980: Importance of biogeochemical processes in bioremediation
 1990: Hybrid approaches – Biological-Chemical
 2000- In situ bioremediation and Monitored Natural Attenuation are widely accepted as cost effective cleanup alternatives



So, what is bioremediation? Bioremediation is considered to be a managed or spontaneous process in which biological catalysis acts on pollutants thereby remedying or eliminating the environmental contamination present in water, wastewater, sludge, soil, aquifer material or gas streams. It is necessary that we understand couple of points within this basic introductory statement on bioremediation.

The first one is the concept of managed or spontaneous process that means I will talk firstly about the spontaneous process spontaneous process refers to those processes which are natural they will they will occur naturally and the human intervention or the technological interventions will be minimum perhaps only to monitor the progress of the pollutant degradation or the state of the environment.

And of course in case of managed it is having some kind of engineered system or rather to say simply it is more like a human controlled system it may be an open system or it may be a closed system but it is controlled by human through different kind of engineering technological and other kind of processes. So, both natural as well as human managed processes could lead to the degradation or remediation of the environmental contamination.

So, the first point was this that it can be a natural or alternatively it can be human controlled or managed. The second point is this that the bioremediation is basically a biological process to say

more specifically it is often a microbiological process although there are examples of plants being used as an important tool. So, I will say biological process. So, biologically it is catalyzed by enzymes. So, there must be some specific enzyme most of the time or certain non specific activities of the cells they are responsible for these biological activities.

And number three third point that it is kind of an action of this biological catalysis on the pollutants. So, if we have the pollutants on this the biological catalyst are going to work and eventually resulting into the remediation that is the removal of the pollutant or reducing its hazard or harm. So, we will talk about the scope of remediation that up to what extent the remediation could be possible.

So, number four would be this action of the biological catalyst or biological processes on the pollutants would result into either remedying or eliminating the environmental contamination. So, elimination is the most desirable process. So, you want the pollutants to be eliminated but it is also a true fact that all pollutants cannot be eliminated for example the heavy metals. So, we cannot eliminate the heavy metals altogether.

So, we can only change their oxidation state their solubility their ability to damage the cells etc. So, possibly for such contaminants we can develop some process or we can rely on the biological catalysis that will offer some kind of remedy. So, that their hazard is reduced and where it works. So, this was the point number four and point number five is very important that it is for everywhere like in water in soil in gas till the minimum requirement that is the physical and chemical constraints.

Mostly the physical constraints like the temperature for example pH for example these are satisfied because biological catalysis must be provided with certain temperature regime we cannot expose the enzymes or the living cells or even the other cellular materials to a very high temperature because then that will totally decompose. So, that is only one constraint. So, except that so wherever the biological catalyst or the enzymes or the cells can maintain their integrity and the structure of the macromolecules are retained.

This kind of catalysts which are the critical component for the bioremediation can be applied and it can be applied to anywhere. So, they can be done using water soil aquifer that is underground water and the gas stream and everything.

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Advancement of microbial ecology paved the road for Environmental Biotechnology : Bioremediation

Pollution treatment technology that uses microbial metabolic processes to reduce, eliminate, contain, or transform various contaminants present in soils, sediments, water, or air to benign products

1900: Municipal wastewater treatment; 1950: Industrial wastewater treatment process; "Microbial infallibility hypothesis" 1970: Bioremediation of gasoline contaminated aquifer; 1980: Importance of biogeochemical processes in bioremediation
1990: Hybrid approaches – Biological-Chemical
2000- In situ bioremediation and Monitored Natural Attenuation are widely accepted as cost effective cleanup alternatives

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Now during the past years what we have learnt that the advancement of microbial ecology basically paved the road for environmental biotechnology and bio remediation. That is the pollution treatment technology that uses microbial metabolic processes to reduce eliminate or contain or transform various contaminants which are present in soil sediment water or air to different benign products.

And the role of microbial ecology that we have already discussed that what is microbial ecology in developing this bio remediation technology it is immense. So, if we look at the background of this bioremediation process we will be able to understand and perhaps appreciate that how actually from a single organism or maybe a handful of organism based bioremediation to a microbial ecology centric bioremediation was possible.

We have started using bio remediation technology through the municipal wastewater treatment during the year 1900. By 1950 we started using bioremediation technology where we deploy the activities of the different microbial cultures microorganisms into wastewater treatment process. And in 1952 the microbial infallibility hypothesis was proposed which basically describes that

for any chemical compound organic compound there would be or there will be or there is one microorganisms.

So, it is just a matter of the time or the ability of the scientist to find out that microorganism. Who can that microorganism can degrade or be used to as a biological catalyst to achieve the remediation. In 1970 and during that time we see that the bio remediation of gasoline contaminated aquifers that is the underground the groundwater systems where gasoline contamination was reported is applied.

And subsequent period helped us to understand that it is not only that the enzyme or the microbe which is of concern but it is of the in the bio geochemical and also the geochemical hydro geological processes which are actually important because they indirectly or directly play a role in controlling the bioremediation process that means the microbes are subjected to their environmental conditions.

So, there may be a pollutant there may be a very capable microorganism but the degradation may not be of appreciable rate or appreciable level only because the biogeochemical conditions or the geochemical or the hydrogeological conditions are not favorable. In 1990 the hybrid approaches biological and chemical coupled approaches were developed and implemented and in next 10 years time in situ bioremediation processes were developed very thoroughly.

We see that across the United States department of energy contaminate different contaminated sites this these approaches of in situ bioremediation during this 1990-2000 period a large number of sites were having bioremediation on a field level by remediation, very successful bioremediation and also the monitored natural attenuation which I referred earlier during my initial deliberation that it can be an absolutely a kind of spontaneous process.

So, monitored natural attenuation relies on basically the natural microbial abilities or biocatalyst which are present naturally and it is only the human intervention is very modest it is the only the monitoring the sites that how the natural processes are actually allowing the contaminants to be eliminated from the system. And that those type of processes like the in situ bioremediation

where these are managed or they may be almost spontaneous process are widely accepted as cost effective cleanup alternatives.

In next two decades like 2000 to 2020 if we consider. So, bioremediation has undergone another remarkable change owing to the development of the microbial ecology tools and microbial ecology approaches that we have seen earlier in our earlier lectures that helped us to understand the role and the significance what can be possible by the microbes who are living with the contaminants. So, we will come back to those discussion may be in our subsequent lectures.

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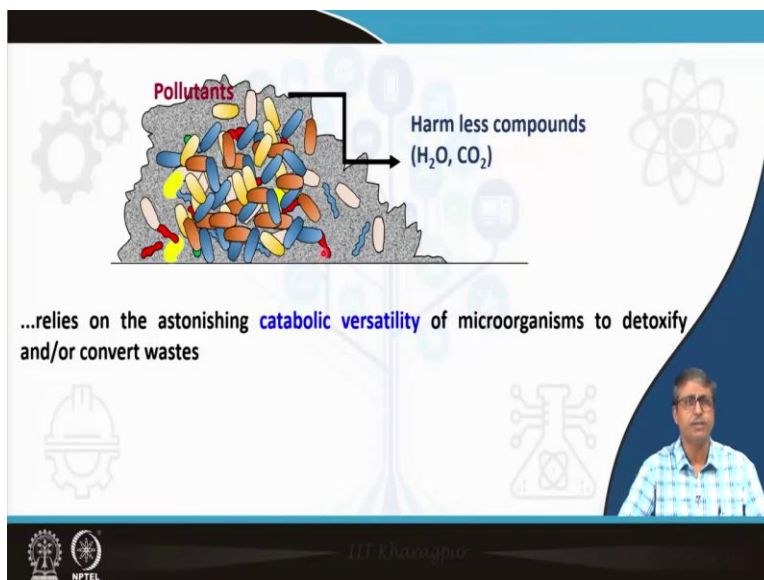
The slide is titled "Bioremediation:" and features a central diagram. On the left, it says "A common pollutant" above "Benzene +", accompanied by a ball-and-stick model of a benzene molecule. An arrow points from this to a yellow box labeled "Microbial cells with desirable enzymes". From the box, an arrow points to the chemical equation $= \text{CO}_2 + \text{H}_2\text{O} +$, which is followed by a cluster of colorful pill-like shapes representing the breakdown products. The background includes faint icons of a gear, a tree, a hard hat, and a flask. In the bottom right corner, there is a small video inset of a man in a blue checkered shirt. At the bottom left, there are logos for IIT Kharagpur and NPTEL.

So, right now we will continue with our basic introductory discussions that in during the bioremediation process it is a very simple otherwise a very simple concept. That you have a contaminant a pollutant like in this case I have taken benzene is organic pollutant and you have to have some microbial cells with desirable enzymes. Why desirable enzymes? Because unless you have these cells with desirable enzymes the pollutant may not be converted to a benign product that means a harmless product or may not be eliminated at all.

So, if we assume that the microbial cells are there or they are applied on the pollutant then they will possibly act on the pollutant molecule and will produce carbon dioxide and hydro water. And also using the energy that is going to be generated by this oxidation process or the breakdown of the pollutant in this case the organic pollutant benzene the microbial cells will

grow. So, I will come again to this particular aspect that it is not only that the pollutant is converted to something which is less toxic but also a large number of cells are produced.

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Now it basically relies on the astonishing catabolic versatility of microorganism to detoxify and to convert waste. So, it is absolutely most of the cases we see it is a natural process. We have a dump site or a dump of waste there most of the cases the microorganisms will naturally colonize and start growing there and these microorganisms are often endowed with the catabolic versatility. Catabolic versatility in this case refers to their ability to withstand the different adverse conditions or the prevailing conditions.

While expressing the required genes or the producing the enzymes which are capable of converting the pollutant molecules into harmless compounds.

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Bioremediation is a technology that can be used to reduce, eliminate, or contain hazardous waste

Transformation Precipitation
Elimination Immobilization

Contaminants
Biotransformation

NPTEL

Dr. Khanna

Now how it works and why to adopt this. So, in a simple term bioremediation is a technology that can be used to reduce eliminate or contain the hazardous waste or the pollutants or the contaminants. So, it can have multiple application like it can be used to reduce or eliminate altogether or contain I will discuss about the contentment in little more detail for radioactive waste for example the containment is a very important topic because from a very large volume of hazardous radioactive waste when we try to minimize or reduce the volume.

So, we try to contain them in smaller volume because we can store few 1000 liters rather than few billions or millions of liters waste because many a times this radio radioactive waste which are often produced in the nuclear power plant or other activities the volume of the waste is of great concern. So, we can actually concentrate those wastes in terms of using microbial process that is the bioremediation.

So, your hazard which is maybe a toxic uranium or toxic technician toxic americium or certain other radioactive elements those are sequesters or accumulated by bacteria and when we capture those bacteria when we concentrate those bacterial cells. So, out of may be few few thousand liters volume we can have only a few liters of bactic bacterial culture or bacterial cell mass that bacterial cell mass which is loaded with the radioactive waste for example or the contaminant can be stored with a minimum volume required.

Certain contaminants like benzene I was referring earlier can be eliminated because these are organic pollutants which can be converted completely to benign product like carbon dioxide or water certain other compounds cannot be converted to complete benign product or eliminated but it can be reduced. So, we will talk about all these different type of processes in due course. So, basically microorganisms and to LSR extent plants as I referred earlier can transform and degrade many types of contaminants.

So, naturally these organisms are microorganisms bacteria archaea and fungi and micro algae as well as well as some plants they have the abilities in fact all living organisms they have some stress management process or they are they are capable of actually converting some of these material into harmless materials. But microbes are specialist. So, they naturally have the exceptional ability to they convert the pollutants or use these pollutants as their nutrients possibly.

This transformation and degradation process which actually is carried out by the microbes these are these vary depending on the physical chemical conditions of the environment and the type of microbial communities present there and the nature of the contaminant. So, it is again is a kind of a quite complex process because we will talk in detail later that it is not as simple as I mentioned earlier that we have a bacterial culture or a bacterial cell or a strain which is capable of degrading a particular contaminant.

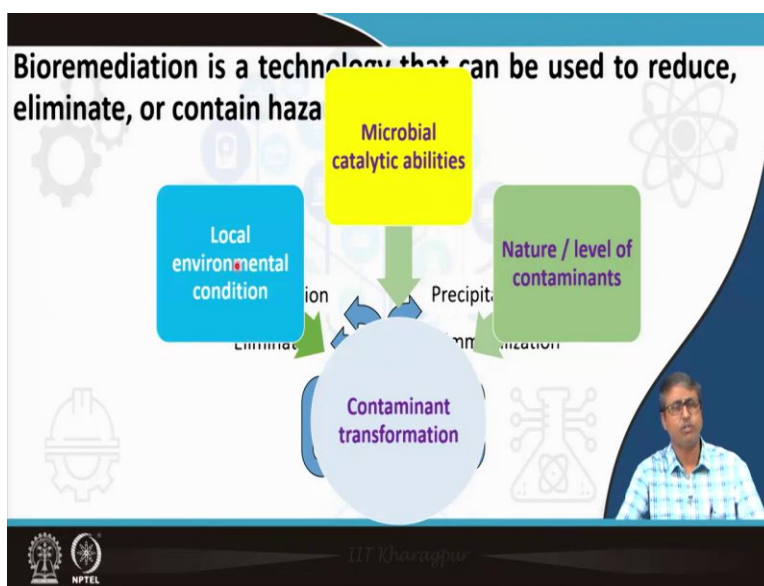
These bacterial strains are subject to the physico chemical conditions because they have their own liking and disliking in terms of the environmental conditions like pH, temperature oxygen requirement redox potential etcetera. At the same time the pollutants are also subjected to this chemical environment and the physical conditions because the solubility of the chemicals the pollutant chemicals their ability to remain absorbed on the solid surface their ability to react with other compounds all these things they change and kind of the nature of the contaminant exactly where is it methylated or it is a normal.

So, these are the type of the contaminants are also there what type of contaminants is the inorganic heavy metal or its organic contaminant. So, there are many minute issues are there but

essentially these are all based on the bio remediation processes are based on biotransformation reactions that means biologically they are transformed into some products which are often less toxic or sometimes they are absolutely non toxic or sometimes it leads to precipitation.

So, they change the solubility product and they leads to or helps to produce certain molecules which are not soluble at all or they allow immobilization of the product molecule or the contaminant molecule. So, that they are their movement of their migration within the environment is limited. So, I am going to discuss this thing in particular.

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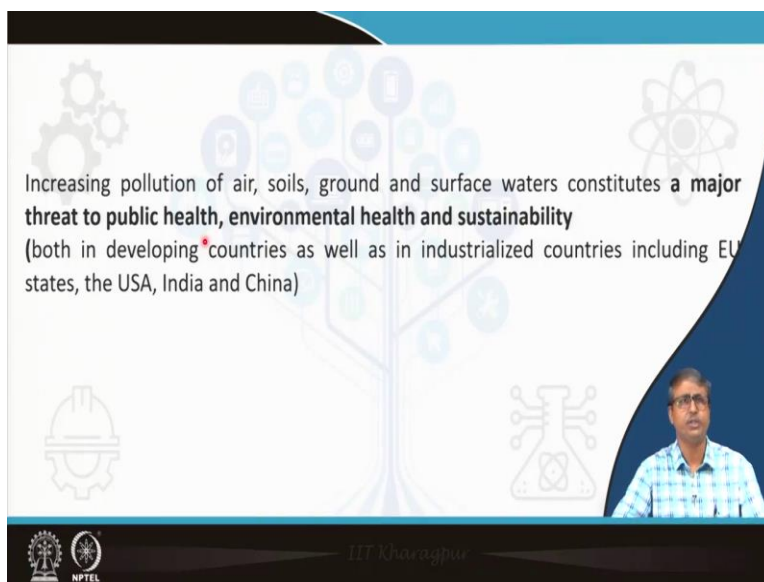


So, now these contaminant transformations which are bio transformation reaction; which might lead to a number of outcomes; including the elimination to immobilization to precipitation etcetera. They are controlled by the local environmental conditions microbial catalytic abilities and also the nature and level of the contaminants. Sometimes the level of the contaminants is so high or the concentration of the contaminants is so high.

Even very capable organism might die or might succumb to the death because of the high concentration of the contaminants. So, it is a huge toxicity because of the toxicity the organism dies. Otherwise if the contaminant is in low concentration or low level this particular contaminant might have been or could possibly be degraded happily by the by the microorganisms present there.

And the catalytic ability of course that is the kind of the backbone of this entire process the microbial enzymes which are responsible for this degradation process or the most of the time the biotransformation reactions and as I mentioned earlier the local environmental conditions which control both the pollutants behaviour or the hazardous contaminants chemical and physical behaviour and also the microbial functions.

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Increasing pollution of air, soils, ground and surface waters constitutes a **major threat to public health, environmental health and sustainability** (both in developing countries as well as in industrialized countries including EU states, the USA, India and China)

Now increasing pollution of air soil ground and surface water constitute a major threat to public health. Now why so, much of stress. So, much of importance was given to bioremediation if we look into the literature if we look into the R and D engagement. So, why so, much of importance emphasis was given to by remediation. Because if we look at the history in last two centuries I will show you certain some data that in developing countries as well as industrialized countries.

We have including European union states and the USA, India, China we have a massive increase in the pollution load all around us.

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Environmental contamination by hazardous substances

Throughout human history, many societies have developed and thrived at the expense of inefficient and unsustainable exploitation of the environment

In the last century, human activities have resulted in to the deterioration of our natural resources :

- ~39–50% of the land surface was modified
- Atmospheric CO₂ concentration increased by 40% over the past 140 years (mainly due to hydrocarbon combustion and deforestation)- global warming

Biodiversity been significantly impacted, many natural resources are impacted, including the widespread contamination of groundwater aquifers by hazardous wastes, soil and our rivers and oceans

 NPTEL

And these environmental contamination by hazardous substances some summary I am going to present here. So, we see that throughout the human history many societies have developed and thrived at the expense of inefficient and unsustainable exploitation of the environment. So, it is a part of our humans development. So, ever since human tried to develop its societal and technological and other parameters other standards. So, we have actually dealt with our environment and most of the most of the time this dealing with the environment was not a very efficient way and was perhaps not very sustainable way also.

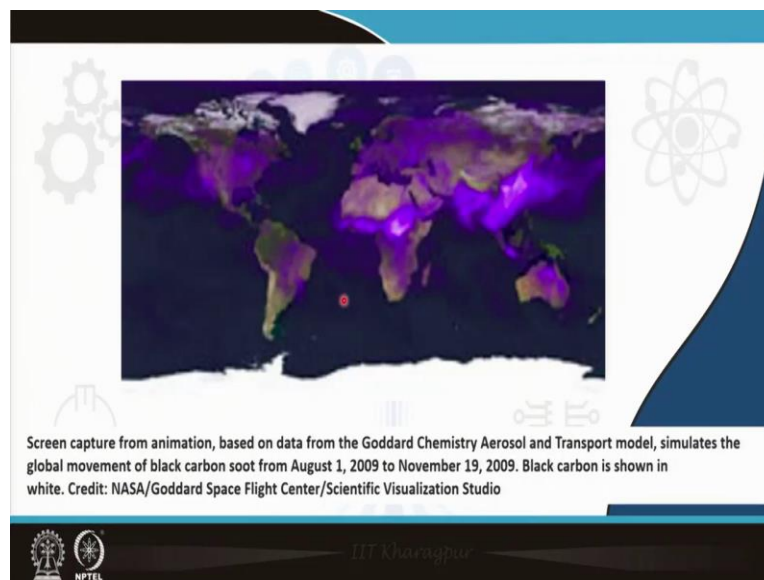
So, in particular if you look at the last what happened in the last century human activities have resulted in the deterioration of the environment of the natural resources very severely like up to 50% of the land surface of our planet earth is modified either because of the urbanization industries our other things and also because of the agricultural hybrid practices atmospheric carbon dioxide concentration increased by 40% over the past 140 years or so mainly due to hydrocarbon combustion and deforestation and leading to global warming.

These all indirectly or directly impacted the biodiversity and also the natural resources are impact very severely. So, direct there are some direct effects like the pollutants and the hazardous contaminants are released and they released into the soil into the water. On the other hand the earth itself has its own bearing capacity managing the pollutants or the managing the chemicals which are present on the earth the earth microorganisms.

They have their inbuilt capacity but sometimes this that capacity was not enough or is not enough because the biodiversity is significantly impacted. So, we lost many of the important species not only for the eukaryotic species which are very well document like for birds for the butterfly to the different animals and different plants but we perhaps lost many microorganisms also.

It is not yet possible for us to cataloging of all the micro organism because impossible task almost. But we can assume that we have lost a significant biodiversity altogether and we because of this the biotransformation ability of the all the biosphere rather I will say is reduced and on the other hand the input of the contaminant contamination has increased. So, I will show you some data.

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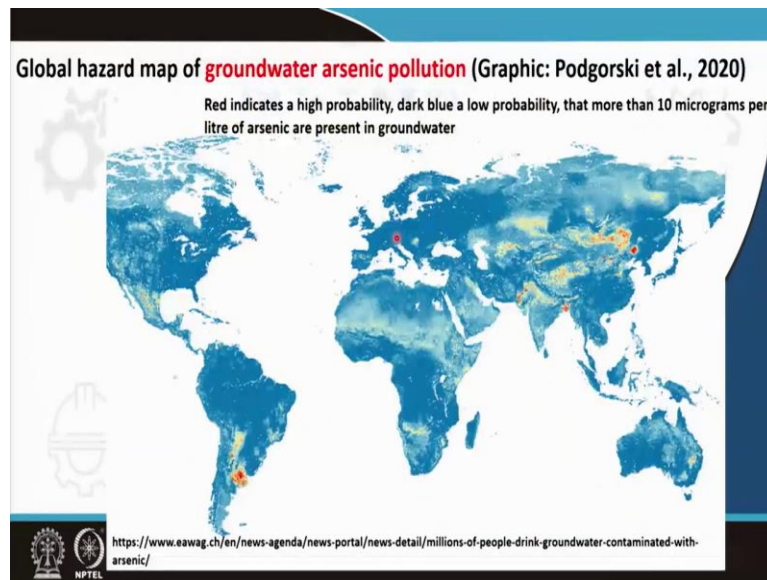


This is one data available from the NASA site that the screen capture from the that it is basically animation and the screen capture is showing the data from the the aerosol and transported model that the soot carbon shoots the black carbon soot from during 2009 to august to 2009 November the data was recorded. And we can see the huge amount of black carbon suits are being released into the atmosphere.

If we see the status of the soil and here there are certain markings the red is very high severity

contamination the human induced soil degradation and the pink one is the high severity. So, if you look at only the red and pink we can understand that red and pink dominants compared to the green and the grey. So, these are human induced soil degradation. So, soils are getting degraded only because of human activity this is only human activity.

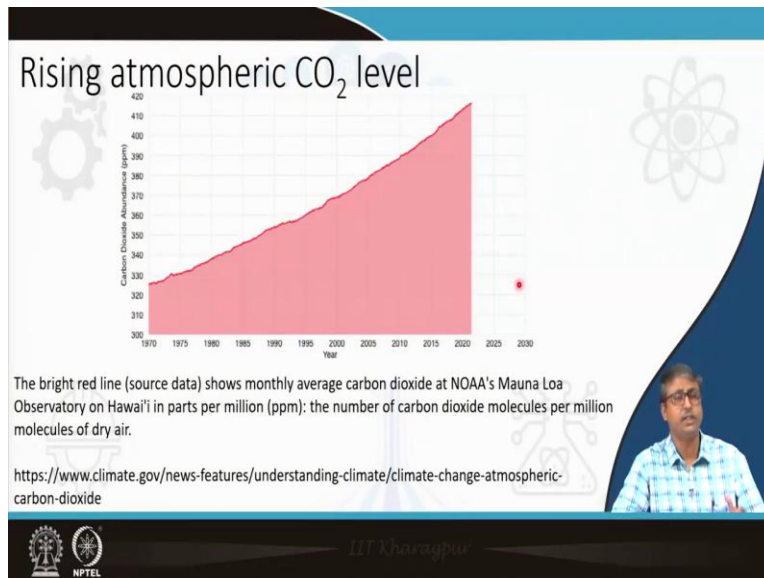
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Natural causes are not included into that if we look at the groundwater arsenic pollution. We can see that these red dots are basically the highest level of arsenic pollution and many times although the arsenic pollutions are often recorded or reported to be geogenic. But still geogenic means it is arsenic is actually coming from the geological settings from the geological sediments minerals which are there in the underground.

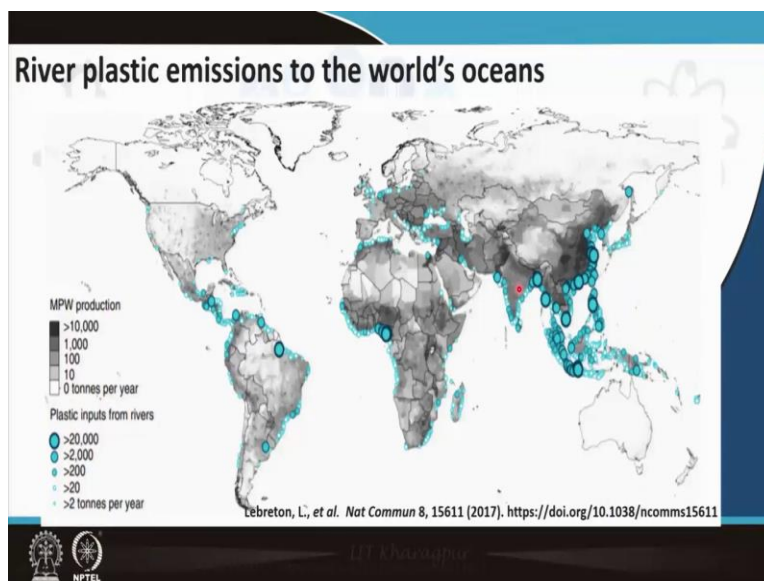
But nevertheless why so, much arsenic is coming into the water? If we look into that we understand that groundwater abstraction how much groundwater we are withdrawing and how much we should there is a big gap between that. And also different type of other release of other organic and other pollutants might be altering the groundwater chemistry groundwater geochemistry or rather biogeochemistry.

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Leading to the; release of pollutants like arsenic which were otherwise there with the minerals for a very long period of time. This data again shows from the climate side that how carbon dioxide level is increasing from 1970 to 2020 data is only plotted. And if we go to this particular site we can actually see the entire history of carbon dioxide rise and the rise in global temperature.

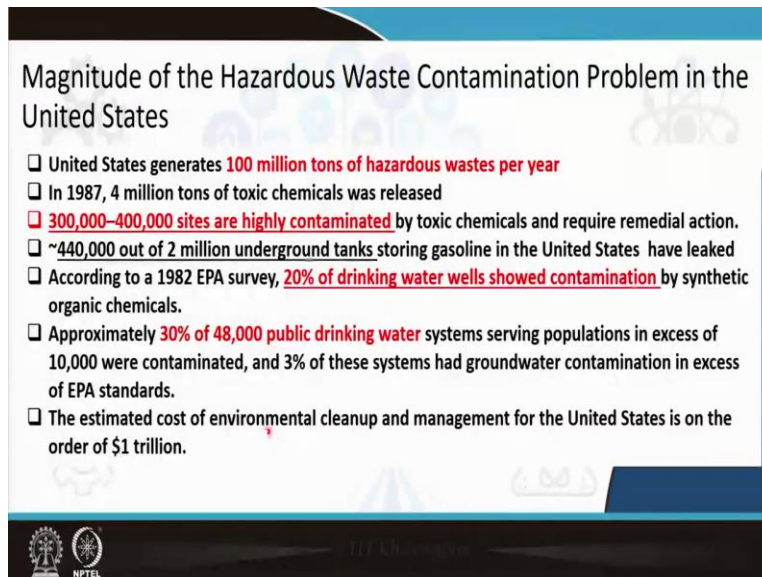
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This particular data is representing the plastic pollution. So, everybody is aware about the plastic pollution but if you look at the plastics released by the rivers into the world ocean. So, almost all the coastal areas if you look at the India and the other parts all the parts are actually having the plastic inputs from different rivers there is a torn spark. So, it is more than like 20000 to 2000


tons of plastic per year it is being deposited into the ocean by different rivers.

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Magnitude of the Hazardous Waste Contamination Problem in the United States

- ❑ United States generates **100 million tons of hazardous wastes per year**
- ❑ In 1987, 4 million tons of toxic chemicals was released
- ❑ **300,000–400,000 sites are highly contaminated** by toxic chemicals and require remedial action.
- ❑ ~**440,000 out of 2 million underground tanks** storing gasoline in the United States have leaked
- ❑ According to a 1982 EPA survey, **20% of drinking water wells showed contamination** by synthetic organic chemicals.
- ❑ Approximately **30% of 48,000 public drinking water** systems serving populations in excess of 10,000 were contaminated, and 3% of these systems had groundwater contamination in excess of EPA standards.
- ❑ The estimated cost of environmental cleanup and management for the United States is on the order of \$1 trillion.



So, what are we doing in 2006 book published by professor Pedro Alvarez mentioned about the magnitude of hazardous waste contamination problem only in United States. It was very explicit in stating from different literature in that particular book that United States generate 100 million tons of hazardous waste per year as per that record published in 2006 book. You can assume that in 2021 or current time what would be the status only in 1987, 4 million tons of toxic chemicals was released into different regions.

So, 3 to 4 lakh sites are highly contaminated by toxic chemicals we only in the U.S. And we had actually four lakh forty thousand out of two million underground tanks storing gasoline in the United States have leaked. And according to 1982 EPA survey 20% of drinking water wells sold contamination and 30% of the 48 000 public drinking water system serving population in excess of 10000 were contaminated.

And the estimated cost during that time of environmental cleanup and management for the United States alone was in the order of trillion dollar.

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Pollutants of concern

The majority of contaminants that affect soils and waters are :

- Heavy metals
- Organic compounds
 - [mineral oil hydrocarbons, polyaromatic hydrocarbons, benzene derivatives, and halogenated hydrocarbons]

Many of organic polluting compounds are xenobiotics (of anthropogenic origin)

- Agricultural (pesticides: dichlorodiphenyltrichloroethane, atrazine, and pentachlorophenol)
- Industrial (solvents: dichloroethane or dielectric fluids such as polychlorinated biphenyls)
- Military use (explosives: 2,4,6-trinitrotoluene)

The slide features a blue header, a white background with faint chemical structures, and a video inset of a man in a blue checkered shirt. Logos for IIT Madras and NPTEL are visible at the bottom left.

So, this is just kind of a tip of the iceberg so if you look at the other countries including India China etc and in current time I think the situation will be devastating. Now what are these pollutants of concern that possibly will be subjected to bioremediation or being already subjected to biomedication or being treated. So, majority of the contaminants that affect the soil and water are found to be heavy metals and organic compounds.

So, these are two classical categories of pollutants and during our lectures also. So, we will discuss separately these heavy metals and organic pollutants. And along with this there are many organic pollutants which are called xenobiotic pollutants of anthropogenic origin they may include different agricultural agriculturally used compounds some industrial solvents and military use including this different explosives.

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Emerging contaminants

[substances long present in the environment whose presence and negative effects have only recently been recognized (Petrie et al., 2015)].

Group of products	Class of chemicals	Examples
Human and veterinary pharmaceuticals	Antibiotics, anti-parasitic agents, ionophores	Amoxicillin, erythromycin, metronidazole tetracycline
	Stimulants and drugs anti-inflammatory, anti-diabetic, anti-epileptic, anti-hypertensive, anti-cancer drugs, anti-coagulants, analgesics	amphetamine, cocaine, caffeine, nicotine, ibuprofen, metformin, morphine, diazepam, tamoxifen
	Hormones including natural and synthetic estrogens, androgens	Estrone, estriol, testosterone, progesterone
Industrial and household wastewater products	Insecticides, plasticizers, detergents, flame retardants, polycyclic aromatic hydrocarbons, antioxidants, fumigants, preservatives	Carbyl, chloropyrifos, diethylphthalate, p-nonylphenol, 1,2,3-trichloropropane, phenol, naphthalene, anthracene, 1,4-dichlorobenzene, acetophenone etc
Personal care products	Insect repellents, polycyclic musks, sunscreen agents, fragrances, antiseptics	Bisphenol A, 1-benzophenone, methylparaben, N,N-diethyltoluamide, triclosan
Nanomaterials	Miscellaneous	Nanosilver, alumina nanoparticles, titanium dioxides, fullerenes, carbon black

Dvorak et al 2017, *Biotechnology Advances*, 35: 845-866

The list can be further broadened with petroleum-derived plastics and some chemicals originally considered to be green, including certain types of bioplastics or ionic liquids

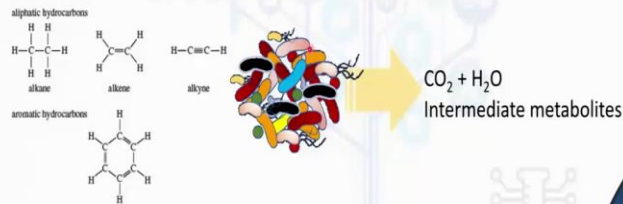
We do have emerging contaminants which are basically substances with long present in the environment whose presence and negative effects have only recently been recognized. So, it is only 2015 we started recognizing that there are something called emerging contaminants. And that possibly is enlisted very nicely that there are the human and veterinary pharmaceuticals industrial and household wastewater products personal care products and different type of nano materials which are being used in different activities human activities.

And some of the class of the chemicals and the examples are also included and that includes obviously the antibiotics to different drugs to different chemical compounds to insecticide plasticizer detergents insect repellents polycyclic mask and different type of nano material cells also.

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Microorganisms (bacteria) are the major entities of transformation of pollutants

The major entity that causes large-scale transformations in the biosphere are microorganisms and their metabolic pathways.



Microbes degrade toxic chemicals via complete mineralization or co-metabolism, in aerobic or anaerobic conditions



Now during this entire process of understanding the pollution around the world and around the corners of the ground groundwater to soil to other water bodies we have realized that there is a tremendous scope for offering remediation or remedial measures through engagement of the microorganisms in doing the proper work in terms of bio transforming these pollutants into harmless products.

So, microbes particularly the bacteria are found to be most favourable candidates for these bio remediation technologies. Now microorganisms that is the bacteria they are the are the major entities of transformation of pollutants the major entity that causes the large scale transformation in the biosphere are microbes and their metabolic pathways because of their long-term evolutionary history that they have been staying in this planet for more than anyone else.

So, they have dealt with all kind of situations. So, naturally what we have seen and we are actually learning from them that they have actually endowed with all kind of abilities to that is the microbial infallibility hypothesis is almost like a true hypothesis that for any kind of organic pollutant there is perhaps some microorganisms who can actually convert these pollutants into some product or certain intermediate metabolites.

And these microbes that degrade the toxic chemicals via complete mineralization that is converting these waste compounds or chemical compounds into carbon dioxide or water

sometimes. The intermediate metabolites are also produced and sometimes the co metabolism or other metabolic of steps are involved during aerobic or anaerobic processes.

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The major entity that causes large-scale transformations in the biosphere are microorganisms and their metabolic pathways

Why microbes? Microbes are:

1. First life form to evolve (~3.8 billion year)
2. Ubiquitous
3. Most Abundant
4. Metabolically most versatile
5. Highest genomic diversity
6. Make up 2/3rd of entire domain of life

- small genome size
- relative simplicity of the cell
- short replication times
- rapid evolution and adaptation to the new environmental conditions

Now the major entity that causes the large scale transformation is the microorganism and their metabolic pathways. Now why microbes? Microbes are important in this regard naturally they have been selected because these are the first life form to evolve close to the 4 billion years ago ubiquitous everywhere microbes are there is no need that we need to always actually search for a microorganism and then apply that MAG we can do that but what we are seeing that in a large number of contaminated environment naturally microorganisms are growing there.

And they are using those pollutants they are living with the pollutants in their own terms. Now when we want the degradation or the transformation process to be faster at more higher levels then perhaps we need to do our engineering and other things otherwise respond the spontaneity of the process is always there. So, it is ubiquitous everywhere we see the microbes are there where it is a petroleum oil spill or it or any other kind of waste we see microorganisms are already living and they are actually dealing with these contaminants.

They are the most abundant living form metabolically they are most versatile because if we see the types of electron donor use electron acceptor use the type of carbon sources they use its is a very long array of processes that they are able to manage. Highest genomic diversity, so highest

genomic diversity is basically connected to the types of different genes they have and their ability to produce different enzymes.

And what we have seen that they make up two third of the entire domain of life there are some specific advantages that is the small genome size relative simplicity of the cell short replication time. So, from a technology development point of view we have we have found that the their growth rate is really something which is manageable and we can have a large amount of biomass which can actually work better compared to the other slow growing cells.

Rapid evolution and adaptation to the new environmental conditions for emerging pollutants and for other different harsh conditions we see microbes are able to learn and adapt very quickly.

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Why bioremediation ?

- 1.Ability and versatility of microbial processes to withstand most inhospitable conditions while tolerating and detoxifying multiple contaminants simultaneously
- 2.No secondary waste generation
- 3.Useful for the high volume dilute wastes where the trace contaminants ultimately limit the acceptability of final wastes
- 4.Economic
- 5.Applicable for otherwise non/slowly degradable organic and inorganic contaminants

The slide features a blue and white color scheme with a background of faint molecular and biological icons. A small inset video of a man in a blue checkered shirt is visible in the bottom right corner. The NPTEL logo is at the bottom left.

Now why bioremediation it is because the ability and versatility of the microbial processes to withstand most inhospitable conditions while tolerating and detoxifying multiple contaminants simultaneously. So, we need to have a remediation process we cannot say move on with the pollution in our water in our soil in our air everywhere we need to develop technologies. So, scientists are all the time working on that and when it when it comes to the micro organism of the biological catalysts we found that among the diverse type of biological entities that we have around us.

The microbial processes are found to be more efficient superior. Superior because they have the ability and the versatility with regard to particularly withstanding the most inhospitable conditions because the wastes are most of the time having very harsh conditions for other living form to survive and grow. The microorganisms they not only transform and decompose or degrade the pollutants but also they tolerate many times up to most of the cases a significant level of concentration threshold.

Unless and until the concentration of the pollutants is very very high the microbes are able to withstand that and they continuously detoxify or degrade and multiple contaminants. It is not always one contaminant being converted to an less toxic form. Often in a realistic situation where you see multiple contaminants are there microbes are capable of handling that very nicely. No secondary waste useful for high volume dilute waste where the contaminant level is actually very low but still we cannot release it.

Because the acceptability of that effluent is not there because of the may be the radioactive contaminated waste for example it is economic. Because the cost of only is the managing the microbial activities and it is applicable for otherwise non or slowly degradable organic and inorganic contaminants. There are actually chemical process which will not possibly work on this kind of compounds but microbial activities will happily work on that.

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The slide illustrates three remediation methods for contaminated soil:

- Bioremediation:** A syringe injects liquid into the soil. A mole character says, "Welcome neighbor!".
- Chemical decomposition:** A syringe injects a different liquid. A mole character says, "You're hurting the community!".
- Excavation:** An excavator digs up the soil. A mole character says, "Help! I'm being taken away!".

A text box states: "Microorganisms adjust to the environment".

URL: <https://www.bri.co.jp/english/bioremediation/index.html>

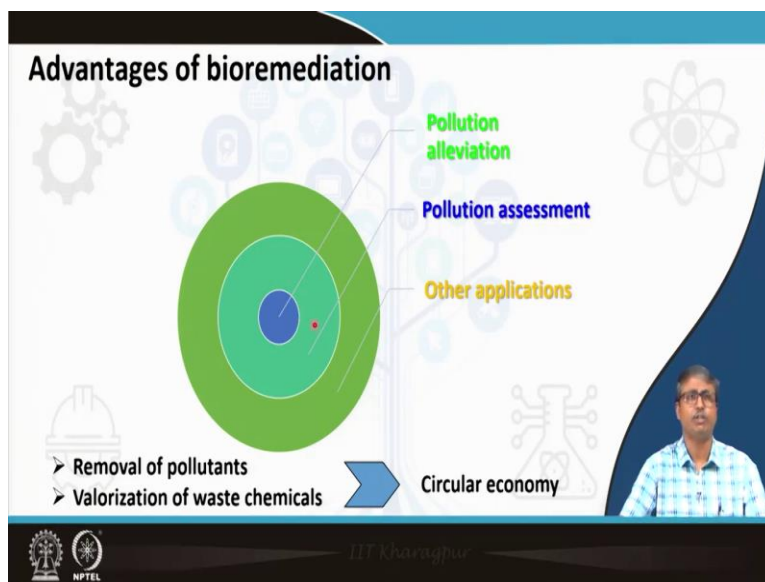
Logos for IIT Kharagpur and NPTEL are visible at the bottom. A small video inset shows a man in a blue checkered shirt.

So, here is the cartoon that describes the process that instead of chemical composition where we add we add basically chemical from outside. So, we interfere with the natural environment natural ecosystem. The moment we add certain chemicals or certain use certain physical method to detoxify or remove the pollutants we actually perturb the ecosystem of the environment. Like this animal is saying you are hurting the community but when we add the biological catalyst doing something in the part of the bioremediation we actually have a shiner we maintain this energy with respect to the overall ecosystem of functioning and its composition.

So, most of the time when we adopt bioremediation strategies they are welcome by the other components of the ecosystem like in this case the animal is saying you are welcome. But on the other way we have the physical remediation that is also not acceptable because many a time it disturbs the entire ecosystem like in this case the excavation process where which are which may be excited treatment or something.

So, we take out the contaminants material may be soil or something like that and take it to some treatment site. So, we destabilize the ecosystem entire ecosystem is destabilized. So, after the thorough survey of the all the options using this chemical decomposition chemical processes the physical interventions and the microbiological or bioremediation interventions in the past 30 years or so we have learnt that the bioremediation is one of the most advantageous.

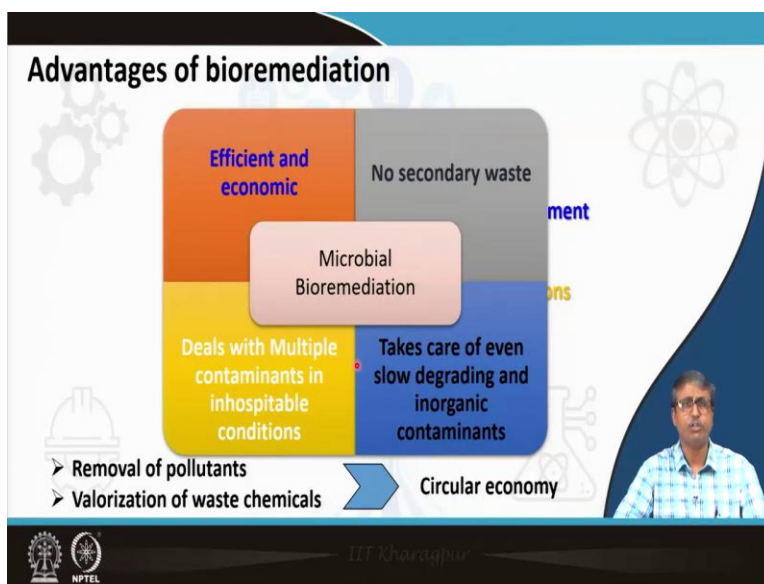
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What are the advantages to be very particular the pollution alleviation is the primary goal followed by associated advantages which are pollution assessment it is not only the pollution elevation which is the central goal. But also we are able to assess how much pollution is there because based on certain microbial genes and microbial processes we can easily actually identify the pollution assessment and how the natural attenuations are going on and different other applications.

Because many of these microorganisms are also capable of producing different products or by-products rather I will say which add to a kind of help us in circular economy. That is if they help in removing the pollutants at the same time they generate certain resources. So, essentially it helps us to gain something out of the resources out of the waste. So, waste is not considered as waste when you use bioremediation this it is conceived as a resource material.

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And add it to the further. So, by microbial bioremediation is found to be efficient and economic deals with multiple contaminants in hospitable condition. So, simultaneously organic inorganic heavy metal containing pollutants which are the ideal scenario if you go to a landfill site you will you will see that all kind of not only plastic there are dyes there are heavy metals there are organic pollutants well lot of compounds are together.

So, for a for any particular type of bacteria it will be very difficult but there are many microbes

who can handle the multiple pollutants at the same time and in inhospitable conditions. Because sometimes the pH or sometimes the presence of other toxic compounds are such that other living forms other living cells might not able to perform there it takes care of even slow degrading and inorganic contaminants and also no secondary waste generation.

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Microorganisms (bacteria and archaea) provide the catabolic machinery for bioremediation

The slide features a background with a stylized tree of icons representing various scientific and technological fields. In the bottom right corner, there is a small video inset of a man in a blue and white checkered shirt. The bottom of the slide contains logos for IIT Kharagpur and NPTEL.

So, microorganisms basically have been through throughout the evolution they have evolved the their metabolic capacity which actually provide with the catabolic machinery we call catabolic machine for bioremediation.

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Developments in bioremediation is closely linked to our understanding on how microorganisms work !

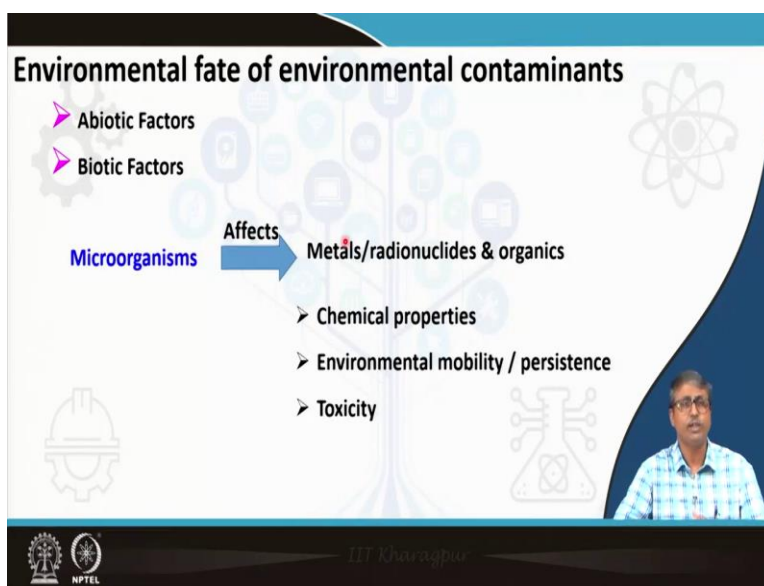
The slide features a background with a stylized tree of icons representing various scientific and technological fields. In the bottom right corner, there is a small video inset of a man in a blue and white checkered shirt. The bottom of the slide contains logos for IIT Kharagpur and NPTEL.

And the development in the bioremediation is basically closely linked to our understanding on

how microorganisms work. So, in order to in the as I mentioned in the past 30 years or so, as we recognize the importance of microorganisms we started learning that how can we use these microorganisms in the best possible way. So, one aspect was the microbial implementation of the knowledge of microbial ecosystems or ecology that the microbes are not singular they always act in association with other species other cells.

They continuously interact with different physical and chemical components or factors in the ecosystem and then they function. So, we have tried to understand how microorganism works.

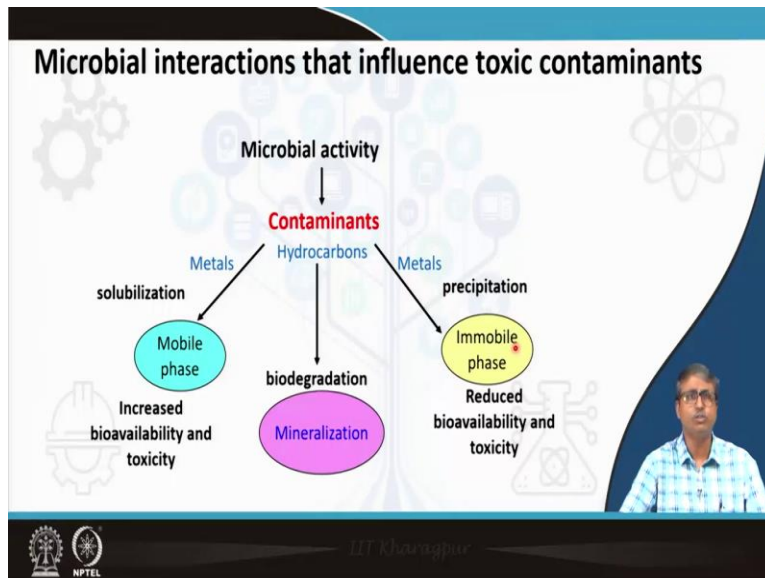
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So, what we have understood and we will discuss about this process in details in our subsequent lecture. So, we today we are going to have our lecture completed by a couple of notes the one aspect is the environmental fate of the contaminant like the among the different factors which might control the environmental fate of the contaminants the abiotic factors and biotic factors are very important.

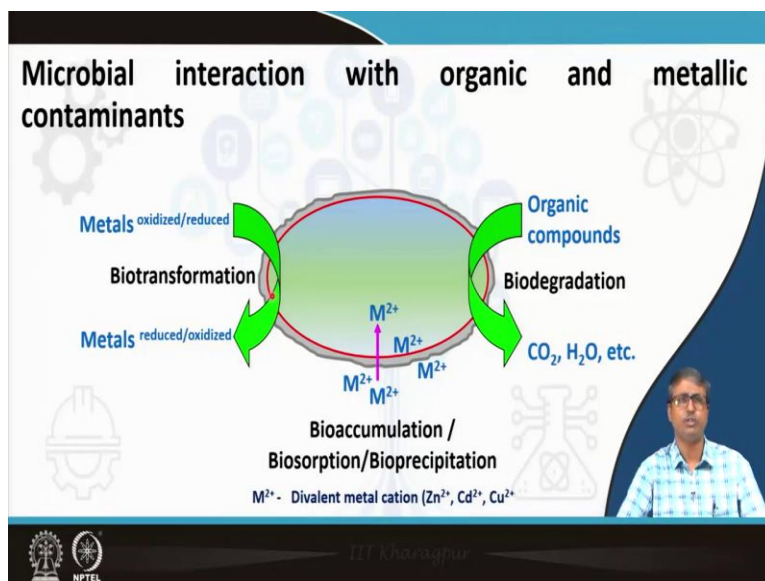
And as a biotic factor the microorganisms the affects the metals radionuclides and the organics in terms of their chemical properties in terms of their environmental mobility and persistence and also in terms of their toxicity.

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So, what they do these microbes they basically interact with these contaminants. So, they can interact with the hydrocarbon contaminants and lead to the mineralization that means convert them to most of the time into carbon dioxide and water or with the metals they can solubilize the metals and they can act in convert the metals into some immobile form. So, which will have reduced availability and reduce toxicity.

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And these processes are very well characterized. So, in a very summarized form if we if we look at the microbial interaction with the toxic or the otherwise organic and metallic contaminants. We see that the biodegradation is the process in which the organic compounds are used by the microorganisms through their microbial different enzymes to convert into CO₂ and H₂O

whereas there are other bio transformation reactions are there which facilitates the oxidation reduction reactions of the different redox active metals inorganic pollutants.

And also there are process processes which involve the bio accumulation bio absorption and bio precipitation particularly for the different type of heavy metals. So, you will discuss individually all these topics like biodegradation biotransformation and bioaccumulation bioabsorption process in our subsequent lecture.

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Basic concept : pollutant as resource

Metabolic strategies

- Organic compounds : C and e donor
- Heavy metals : e donor, acceptor, nutrient
- Other inorganic (N, P, S) : nutrient, e donor, acceptor
- Detoxification strategies
- Co metabolism

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Now why microbes are interested to interact with these pollutants it is it is understood that microbes often they do not consider the pollutant molecules as polluted the chemical molecules with which they interact are pollutant for us pollutant for the overall environment but often up to the levels where the microbes are not feeling very, very stressed they may be acting with them or interacting with them or trying to act with them.

So, that they can utilize them as resource what kind of resource they want to apply a number of metabolic strategies. So, those different organic compounds are used as carbon and electron donor. Heavy metals are used as electron donor or electron acceptor or sometimes as nutrients other inorganic pollutants are inorganic molecules which are actually nutrients again for them like nitrogen phosphorous and sulfur.

They are mostly considered as new general nutrient or electron donor and electron acceptor sometimes as I mentioned if the concentration threshold are really higher and the microbes they feel that we they should do something because it is getting them some toxic toxic signals though they engage different types of detoxification strategies. So, there have been many many evidences that special detoxification strategies for mercury for chromium for the for cadmium etcetera which are not at all any way nutrients.


So, microbes they do not gain anything out of those like from the mercury or from the cadmium they are not going to gain any benefit as a resource. So, they are not going to use them as resource rather they will deploy different detoxification strategies which are very targeted enzymatic reactions those enzymatic reactions will allow the microbes to detoxify some way or the other these toxic contaminants it that is that can be true for different hydrocarbons residues as well.

And also there are co-metabolism co-metabolism meaning that microbes are already having some metabolic reactions and that is actually for other substrate not exactly for the pollutant molecule but somehow because of the chemical structural similarity between the pollutant molecule and the real substrate molecule the there is a kind of overlap and the enzyme is often unable to our enzyme some enzymes are unable to recognize that this is actually a different compound. So, they convert like oxidize or they reduce the pollutant molecule also.

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So, as a reference this literature will be very useful particularly the first one that is the bioremediation and natural attenuation, The Process Fundamentals And Mathematical Models by Pedro Alvarez in 2006 this book was published and also a couple of literature like in the form of journal papers and reviews are mentioned over here.

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CONCLUSION

- The basic concept of bioremediation is introduced
- Role of microorganisms in bioremediation is discussed
- Basic metabolic logic of bioremediation is discussed



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In conclusion in today's lecture we have discussed the basic concept of bioremediation which is introduced the role of microorganisms in bioremediation is discussed and the basic metabolic logic like the microbes they use most of the time are for the many pollutants the pollutants are utilized as metabolic resource for them. So, basic metabolic logic of bioremediation is discussed, thank you.