

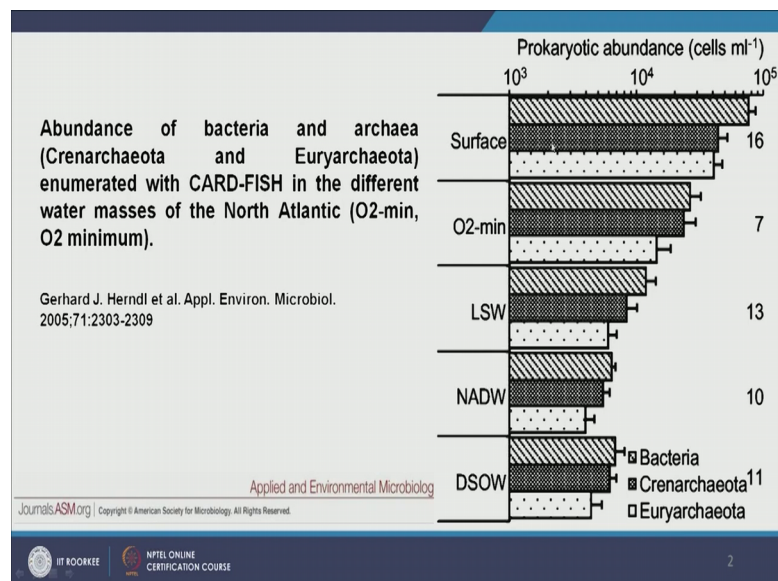
Applied Environmental Microbiology
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Lecture – 20
Microbial Ecosystem V

Dear students, welcome to the last lecture on my microbial ecosystems, in this particular lecture we will wrap up around about the aquatic environmental microbiology, the kinds of microbes that live in aquatic systems and we will focus more on oceans, specially when the pressure and temperature get high and then we move onto our nutrient cycles through the earth as a in collective ecosystem and we will finish today by the human impacts on these nutrient cycles and on the ecosystem in general alright let us begin.

So, as we will start with ocean microbiology and what this is showing you is how the prokaryotic abundance.

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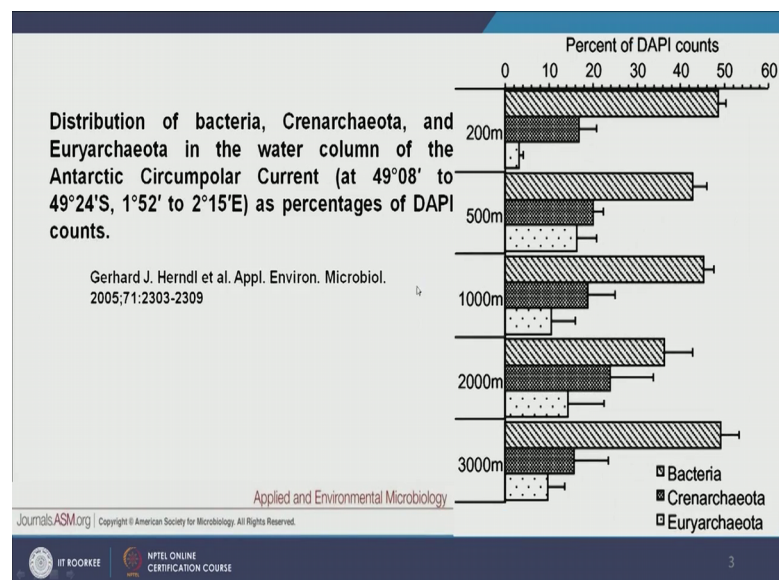


Which is the abundance of Prokaryotic microorganisms changes with depth, Now the ones shaded in diagonal are bacteria, the 1 in the grey in the middle or crenarchaeota kind of archaea and the 1 that are partially shaded with dots euryarchaeota another kind of archaea. So, we are noticing how that the bacteria and archaeal abundance changes as we go below the earth's below the ocean; note that the technology used for this getting

this data is called card fish, which is a fluorescence inflorescent imaging of microorganisms in we will talk about these Mac rebel microbiological techniques later.

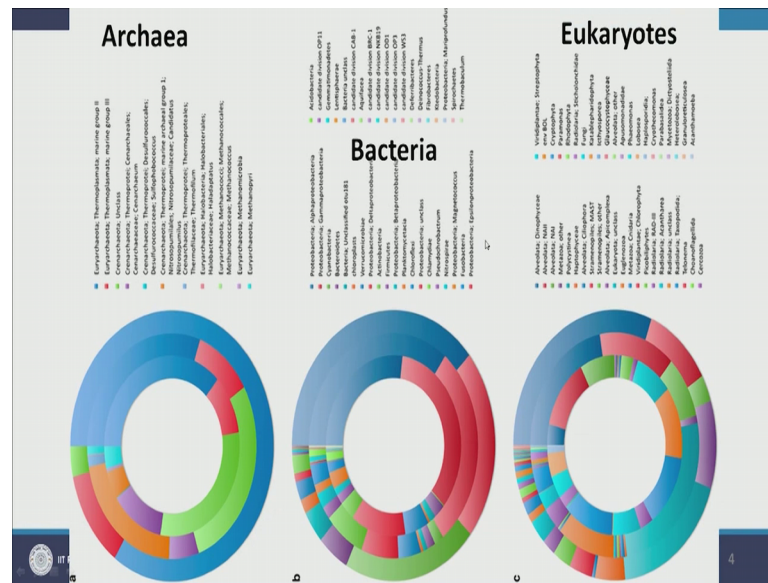
So, this study was done in north Atlantic in 2005, so it has been quite some time and what we are noticing is that as the oxygen drips. So, here is a surface and we have some good amount of dissolved oxygen present maybe even some light and perhaps more nutrients and in this case we notice that there is a very high abundance of bacterial cell, considerable amount of archaeal cell also. In fact, we know the archaeal cell are more than bacterial cell, it is important to note that oceans in general are a good tropics environment they are always lacking a some or other nutrient; whereas the oxygen drops with the abundance drops and with that the abundance keeps continuous to drop.

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The here we have another ocean we have Antarctica, see in Antarctica you notice that it is different from the not Atlantic; here we had more archaeal population than bacterial, but in Antarctica we have up considerably high bacterial population compared to crenarchaeota and euryarchaeota. So, as the depth increases from 200 meter to 3000 meter we notice that the bacteria population first depths and then increases again at high pressure under high depth and the archaeal population remains considerable a constant, it increases in fact between 500 and 2000 in meter. So, at the various surface we have less amount of archaea more euryarchaeota, but with depth archaea also gain some abundance.

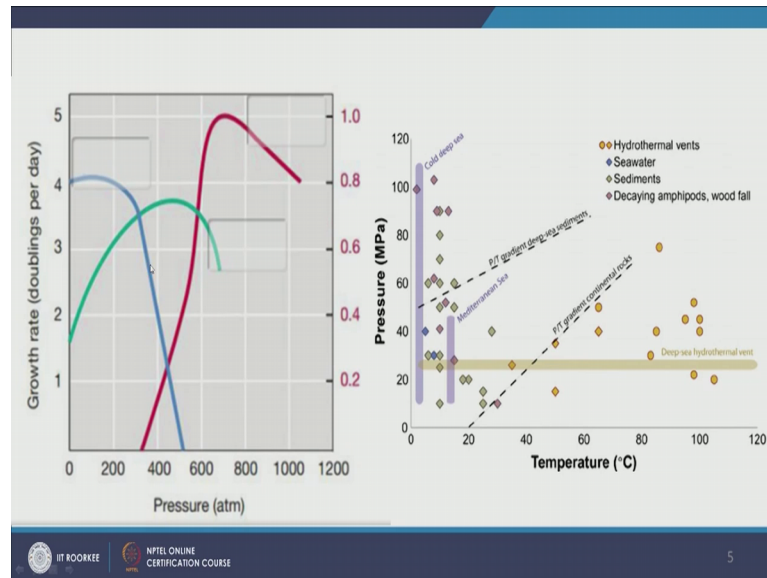
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So, basically what we are noticing here is that with depth and with the kind of ocean whether we are talking about North Atlantic Ocean or we are talking about the Antarctic circumpolar current, which is Antarctic near Antarctica. The microbial population would vary from 1 kingdom 1 domain to another, not only that what we notice is that any given depth this is metagenomic analysis; by the way at any given depth the diversity in archaeal in bacteria and euryarchaeota also vary. So, at the same that we notice that archaea at in final level they had less diversity compared to bacteria and compared to euryarchaeota, which have highest diversity event is visually we can tell that.

Now, note that this is on a final level, so on a class level that is when we go deeper and we go finer in a resolution we might see a different scenario in terms of diversity or richness. Another thing to note is that bacteria and eukaryotes tend to be relatively better characterized than archaea. So, we do not know how many archaea there are that have not been sequenced and thus our databases are not populated enough to tell us what the diversities, but even if we look at them on a basis of operational taxonomic unit, we notice that there is high reversivity of bacteria in eukaryotes. So, not only the number of bacteria, eukaryotes and archaea would vary with that and with oceans but also the diversity.

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Now what happens when we go deep in the ocean, so on x axis you have pressure and on y axis you have the growth rate. So, as we go below in the ocean we the pressure increases because, the weight of the water above the level would be would increase with that and as that increases we have to have bacteria or microorganisms or organisms in general that can tolerate the high pressure and maybe in it. So, on the right plate on the right panel here on x axis you have temperature on y axis, you have pressure in this we are including another factor of temperature.

Many a times the temperature on the ocean surface is in more agreement with the ambient air temperature, but when we go down we might have undersea thermal wends or maybe undersea volcanic wends where we have because, of that we have very high temperatures. So, let us look at the left plate first. So, left plate the y axis is the growth rate doublings per day. So, this is basically the amount of this is the rate at which the microbial population will double. So, we notice when pressure goes from 0 to 200 atmospheres that is lot of pressure, we won't survive this there are microbes that will tolerate this high pressure.

So, the microbes that do not whose the microbial community whose growth rate does not drop a lot. In fact, increases here somewhere in the hundred atmosphere these are pisotolerant microbes. So, pisotolerant microbes are the ones that will survive high pressure, but once we increase the pressure beyond 300 atmospheres their growth rate

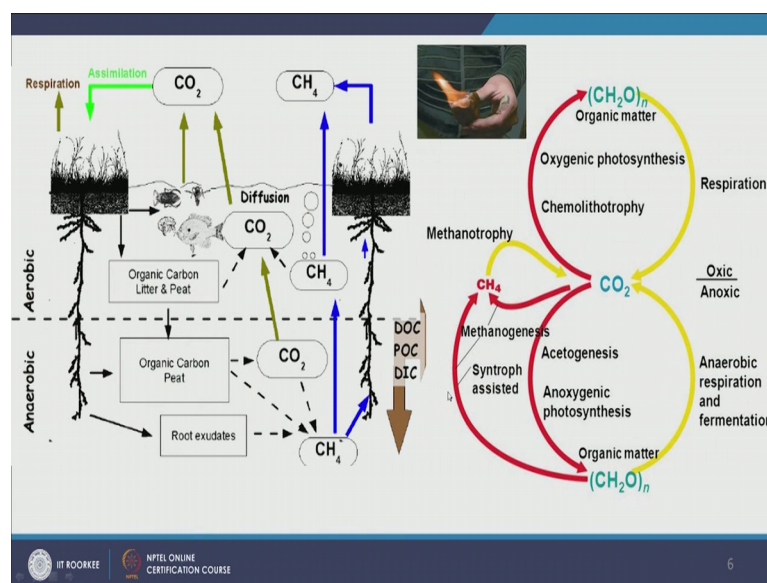
drops very fast, they cannot grow the pressure is too much now. So, we note here that the microbes have a quite broad range of atmospheric pressure that they can tolerate and yet beyond that they won't survive, note that human being on the other hand cannot tolerate such high variation in pressure from 0 to 200 atmospheres.

Now in the green here you piezophilic microbes, so piezophilic microbes love high pressure; when the pressure is not very high their growth rate is lower and then as a pressure increases their growth rate increases. So, per day we will have more doublings up to 3 doublings 4 I mean nearly 4 doublings and then they pick around 500 atmosphere and then pressure increases further they drop they cannot tolerate it in red we have extreme piezophiles. So, these are the microbes that love extremely high pressure, so when the pressure is less than 300 atmosphere; they won't even survive they will give me high pressure and when the pressure starts increasing their growth rate starts increasing it takes around 700 atmosphere and then it drops all the way to 1000 atmosphere.

So, we notice that their as we will go down, the depth of an ocean we will move from moderately high pressure which is 0 to 200 high pressure, from 200 to 600 and then extremely high pressure 600 and beyond atmosphere and with each of these classification we will have different kinds of microbes that we will thrive. Now on the right panel we have put pressure and temperature. So, as when the temperature is high and pressure is high such as deep sea hydrothermal vent, the deep sea is at the high pressure anyway and so we have, but the temperature is very high

So, when the temperature is high we have a very different kind of microorganisms here, versus the microorganisms in cool deep sea as a pressure increases or decreases and meditation and there's not much difference in temperature.

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Now let us look at nutrient cycles so we had doubt. So, far about metabolism we have talked about functions of microbes we have talked about different ecosystems, now very important question arises is how do these microbes drive the nutrients in an ecosystem throughout the earth. Now this is very important question because, this question will lead us to understand how essential microbes are essential it is to protect environmental health; so, that the microbes can do the job of transporting nutrients from 1 end to another in a way that is most suitable for us.

So, let us look here on the left panel you have carbon cycle, so we have plants and there are root system the top layer is aerobic the bottom layer is anaerobic. So, here we have plant respiration and they are assimilating carbon dioxide from atmosphere, now as they do it they die they decay they create organic carbon litter in peat you know the leaves and then they what this is sequestered carbon. So, carbon has been sequestered from atmosphere into form of plant biomass that translates into organic carbon peat c. So, this is very fertile and in anaerobic 0 and we also have root exudates. So, these are the organic materials that are released by roots into the soil.

Now, together they form the carbon in the soil and here because of different microbial processes that there might be anaerobic microbes, that use this organic carbon peat as source of carbon and as electron donor and release carbon dioxide; it is also possible that in highly anaerobic 0 highly reduce 0 we do not have oxygen to make carbon dioxide. In

fact, we have methanogenesis will form methane it is also possible that the carbon dioxide that is made in the less and less reduced 0 will then be converted into methane by methanogens. Now this methanogen can directly be released into the atmosphere directly released into the oceans or it can be taken up by roots which will use it as a nutrient.

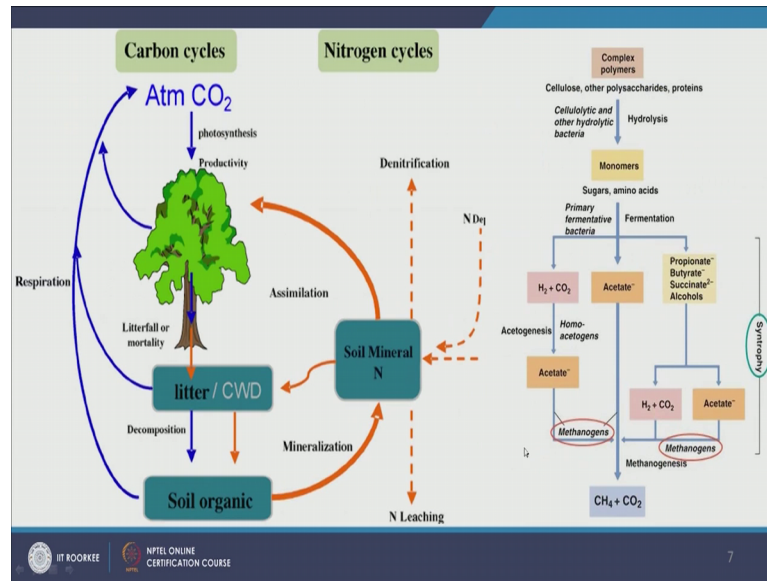
Now, this is a problem when methane directly releases into atmosphere or into water, then we have high methane concentration in the atmosphere and methane is one of the worst gases we going to in our atmospheric high concentration because, it has extremely high a greenhouse gas effect. So, nearly 20 to 25 times higher greenhouse gas effect than carbon dioxide. So, if I increase carbon dioxide by 1 molarity 1 ppm, not molarity 1 ppm in the atmosphere its effect would not be as harmful as increasing 1 ppm of methane.

Now, in many permafrost areas, so permafrost areas are the areas where we have permanent frost. So, there is permanent ice let us say in Siberia and towards the north pole, we have methane that is trapped as ice. So, in the ice here we have methane trapped and as climate is changing this methane is being released out in the atmosphere at a very high rate and people some scientist estimate that in next few years to come most of the permafrost methane trapped in permafrost would release and that will totally overwhelm our climate and we should expect to see a very severe increase in global temperatures and thus we are expecting that things would go really bad very soon in terms of climate.

So, here you have a picture of the methane trapped in permafrost being ignited; now on the right panel again you have a more clearly delineated version of carbon nutrient cycle. So, we have organic matter, organic matter is respired or can be fermented into carbon dioxide and this carbon dioxide can be used by plants for oxygenic photosynthesis on microbes that undergo oxygenic photosynthesis accumulator trophy and they will form organic matter, which can be respired by being like such as our own self heterotrophy and they will make carbon dioxide. Now carbon dioxide can also be converted back into organic matter in by methanogens which will make methane or by acetogenesis and anoxygenic photosynthesis this is make glucose and acid acetate.

Now the methane can be oxidized back to carbon dioxide, so methanotrophy microbes that eat methane or in fact, methane might stay trapped in permafrost as it has stayed for a very long time in Siberia.

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Now these are 2 coupled systems these nutrient cycles do not exist in isolation, when carbon moves in and out of a biomass in and out of a system like roots or soil it interacts with other nutrients; for example, biomass will have nitrogen with live phosphorus. So, when carbon moves nitrogen also moves, when carbon is sequestered nitrogen is also sequestered.

So, let us look at the coupled carbon and nitrogen cycles we have atmospheric carbon dioxide, oxygenic photosynthesis we will convert it into biomass, litter fall for mortality degradation we will composition will increase organic carbon matter in soil and they will be respired by microbes and they we should release carbon dioxide or make methane; which will release appear as methane at the same time when this litter falls to the earth it also in the red is here by the way nitrogen.

So, the nitrogen levels in the soil also increase, this might contribute to the organic matter in the soil which might get mineralized by microbes as they are respiring the organic matter and thus release soil nitrogen which can get denitrified which can reach into groundwater or which can again be assimilated by trees and plants or other microbes.

So, we know they are nitrogen fixing bacteria we know that plants need nitrogen rich soil, so that they can use it to make biomass so this is assimilated process, on the other hand we also have nitrogen deposition. So, the air is very rich in nitrogen nearly 70

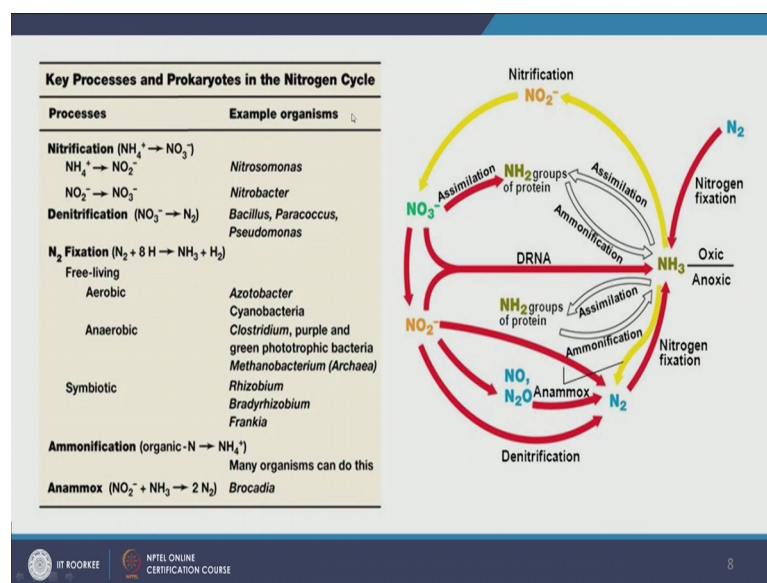
percent of air atmosphere is made of nitrogen. So, nitrogen can get directly deposited here now on the right time we have another kind of degradation. So, since we are talking about biomass one of the most complex not complex one of the most abundant polymers on earth surface is cellulose. So, cellulose is found everywhere and thanks to trees we have plenty of it on the earth, now this cellulose is a very stable form of carbon based polymer and this is very important because the more cellulose we have on earth the more carbon dioxide we have trapped as cellulose.

Because if cellulose degrades it will form carbon dioxide, if you burn trees it is directly goes to carbon dioxide and thus we lose the sequestered carbon and we lose it to atmosphere which makes greenhouse gas effect worse. So, let us look at the cellulose degradation we have complex polymers such as cellulose other polysaccharides proteins maybe lignin which is slow to degrade and then we have cellulitic and hydrolytic bacteria. So, these bacteria are they create they secrete enzyme.

In fact, they are not usually single bacteria, but they are a sweet of hundreds of bacteria, that make different proteins which together attack cellulose resultant cellulose life is that is why they have called cellulitic or hydrolysis. So, they have called hydrolytic and they make monomers of these complex polymers, so we will have cellulose bios we will have glucose.

Now, these sugars or amino acids in case of proteins are either fermented by bacteria into hydrogen and carbon dioxide which can undergo acetogenesis make acetate and then can be used up by methanogens or they are directly converted into acetate which can undergo methanogenesis or they are fermented into proper propionate, butyrate, succinate alcohol etc. We should make S^2 plus CO_2 which can undergo methanogenesis with acetate and the end we will have methane and carbon dioxide. So, here we notice that we are looking at anaerobic this part is mostly anaerobic degradation.

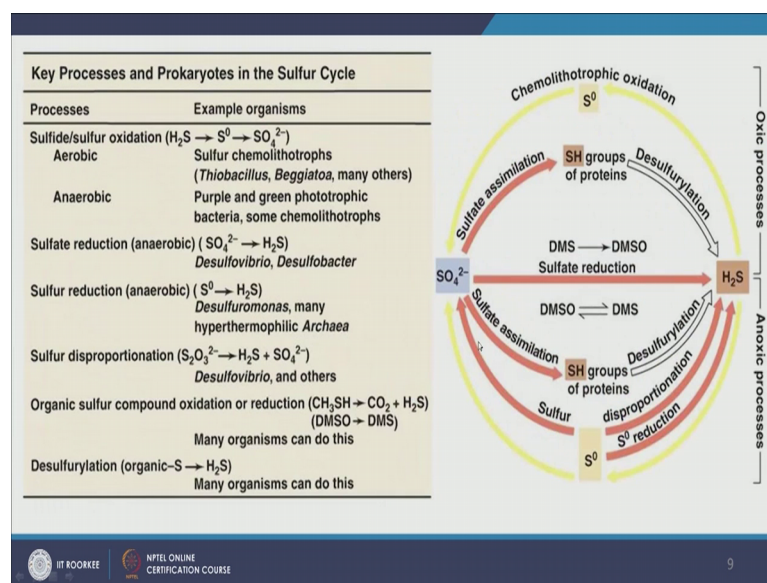
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Now let us go to nitrogen cycle in nitrogen cycle the key processes are nitrification, denitrification and nitrogen fixation ammonification and anammox; we have gone over this before in previous lecture. So, I will be very brief here nitrification is when we are going to higher oxidized states of nitrogen. So, from ammonia to nitrate ammonia to nitrite done by microbes such as, nitrosomonas, nitrobacter, denitrification is when you are reducing the nitrogen, so going from nitrate to nitrogen gas right. So, pseudomonas paracoccus paracilias they do a good job at it.

Nitrogen fixation is when we are fixing the nitrogen in the atmosphere and converting it into a form that can be accelerated by microbes so by plants. So, we have nitrogen with 8 hydrogen from an ammonia and hydrogen gas they can be aerobic they can be anaerobic in here is a list of microbes, the symbiotic nitrogen fixers are in symbiosis with plants the roots of plants. So, rhizobium bradyrhizobium and frankia ammonification is when organic nitrogen converts into ammonia many organisms can do this it is a very important part of multiple treatment plants also and anammox is a recently discovered phenomena and we have talked about it before, when nitrite reacts with ammonia to directly form nitrogen gas and this is your representation of nitrogen cycle.

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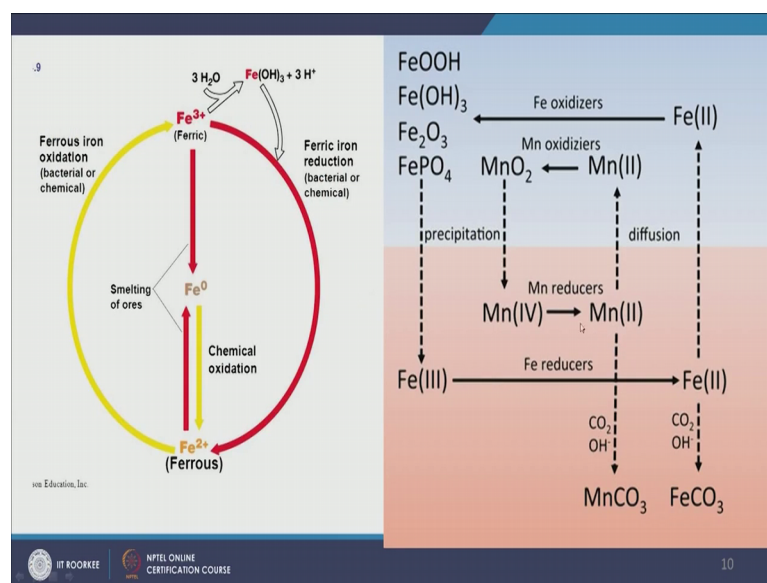


Okay now let us look at the sulfur cycle we have gone through before also, so I will be very quick again. So, in the sulfur cycle the prominent or the dominant activities are oxidation of sulfur. So, which would be converting sulphide to sulfur or elemental sulfur to sulfate, this can be done both aerobically and anaerobically. We have talked about it; purple and green phototrophic bacteria do it. Some chemolithotrophs can do it. Sulfur; chemolithotrophs can do it aerobically. To the next is the reverse direction. So, we have sulfate going to sulfur, elemental sulfur, or going to hydrogen sulfide.

So, delta proteobacteria, *Desulfovibrio*, *Desulfobacter*, they can do this really well, and then we have. So, sulfate to hydrogen sulphide and sulfur to hydrogen sulphide. Many hyperthermophilic archaea can do this too, then we have sulfur disproportionation. So, we have $\text{S}_2\text{O}_3^{2-}$ and then this can convert into this, can disproportionately split into sulfate and hydrogen sulphide. So, one of the sulfurs gets reduced, the other sulfur gets oxidized. *Desulfovibrio* and other microbes can do it. Then we have organic sulfur compound oxidation or reduction.

So, sulfur is not existing; it is an iron, but it is part of an organic compound, and then it is either reduced or oxidized. Many organisms can do it, and then we have desulfurization, when there is sulfur present with an organic material, inherent component of it, and then it is reduced to H_2S .

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Now let us look at the iron cycle and manganese cycle, usually iron and manganese cycles are coupled in the environment, but let us look at iron first. So, this is the first time we are looking at iron so we will spend some time here, the ferric which is highly oxidized from ore of iron can be reduced by bacteria or by chemicals you know the chemistry of ecosystem into ferrous; now ferrous can be further reduced into Fe just elemental iron which very quickly undergoes oxidation, the only way the ferrous will turn into reduced form of our elemental iron is by smelting it and ferric also converts into elemental iron by smelting.

So, smelting is a metallurgical process when you are trying to improve the quality of your iron ore and there are very quick to oxidize. So, ferrous elemental iron very quickly convert into ferrous the ferrous can go very quick undergo quick bacterial or chemical oxidation; which is rusting process and make ferric. Now ferric in presence of water will make ferric hydroxide will solve over in water and it gives water a brown reddish brown color. So, let us see how iron and manganese cycles are coupled together. So, this happens a lot in the water system aquatic systems and this is one of the reason why we have high manganese concentration in some parts of the world like Latin America.

So, here we have iron and then from the ferric form, highly oxidized form and then it goes to this soil gets reduced by iron reducers and it escapes out into ferric cool right cool so far or it can react with carbon dioxide carbonides and form iron carbonate

Now, this can diffuse into the water and then get oxidized, now oxidized version of manganese is not soluble in water, so it precipitates into soil which again can be reduced by manganese reducers. So, what we have here is microbial activity is driving this solution of manganese and because it is driving this solution of manganese we can have a manganese problem.

AIR

$Hg(II)$

Deposition

Cell membranes

CH_3HgCl^{1+}

$HgCl_2$

Hg^0

Food Web

CH_3Hg^+

$Hg(II)$

Hg^0

$Hg-DOM$

WATER

microbes

microbes

Particles

Efflux, Resuspension, Redox

SEDIMENTS: Sedimentation/Sorption/Methylation/Demethylation

- Methyl-mercury – 100 X more toxic
- Mercury resistance:
- Use *organomercury lyase* to convert methyl-Hg to methane and Hg
- *mer* genes

So, we have let it start from the top we have air water and sediment. So, in the air let us say we have mercury exhaust and mercury is being released by industries or initially as it is used to be released by automobiles. So, this mercury is in this oxidation state it is taken by cell membranes, it might convert into methyl mercury which is very toxic enters

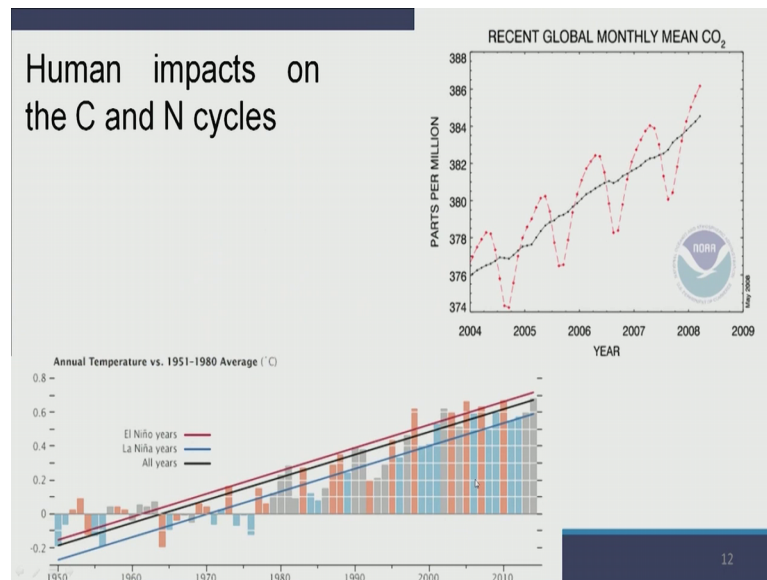
food chain it undergoes bioaccumulation and biomagnifications and it might actually not only stay as CH_3Hg^+ .

But more radicals might join to it and microbes they can convert microbes can convert it back to Hg^{2+} which will form mercury chloride and again intercell membranes or it can find mercuric sulphide and again the microbial activity we can have elemental mercury which will form Hg^0 .DOM and some of these mercury can get deposited into the sediments it can be solved or after methylation it can be or after demethylation when it is removal of methyl radical it can get solved into the sediments.

So, from in the sediment water interface we have a log resuspension in redox reaction and here we have deposition and evasion reaction between air and water. So, some mercury forms will evade, it will they will go into the air form some aerosol or so mercury will come and dissolve in water. Remember methyl mercury is about 100 times more toxic than just mercury, now this is really we need there are some microbes that are resistant to mercury. In fact, I remember when I was doing my doctoral research and I wanted to kill the bacteria I was working with and the reason I wanted to kill them was I wanted an abiotic control so the poison of choice for is mercury chloride.

So, you add mercury chloride and microbes will die, what we notice in many experiments is that there are microbes that do not die when they are present in with mercury chloride. So, now let us look at a more toxic form of mercury which is methyl mercury, interestingly there are microbes that are resistant to methyl mercury also. So, the way they develop resistance to 100 times more toxic methyl mercury is they use this wonderful enzyme organomercuric lyase which converts methyl mercury into methane and mercury they can affect the methane as food source or not and these enzymes are encoded by mer genes. So, if you want to find out the mercury or methyl mercury resistance of microbes look for mer genes and this is a class of genes by the way so there are different kinds of mer genes.

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So this is how I want to end the microbial ecosystem class by talking about, you know about different ecosystems how microbes like to live there what are the different environmental parameters that affect them; you also know about the carbon nitrogen sulfur and heavy metal cycles in our ecosystem. Now the thing is how have human activities impacted the cycles in the ecosystem, this is very important question because perhaps for the first time in history of earth this recent phenomena called humankind or any reasons phenomena has so severely impacted and so severely driven the earth's ecosystem to a mass extinction phase. So, I have said this before in this class and I want to repeat that right now we are under 6th mass extinction stage.

So, the species and the life as we know it is disappearing from earth right. Now at unprecedented rate to such an extent it is with such as high speed that we call it extinction phase and this is worse than any of the other extinction phases ever and most scientists. In fact, all scientists I would say agree on this that human activities are culprit. So, let is try to understand what human impacts have done to carbon nitrogen cycle, most of people have said already familiar with climate change or familiar with greenhouse gas effect so the more we take.

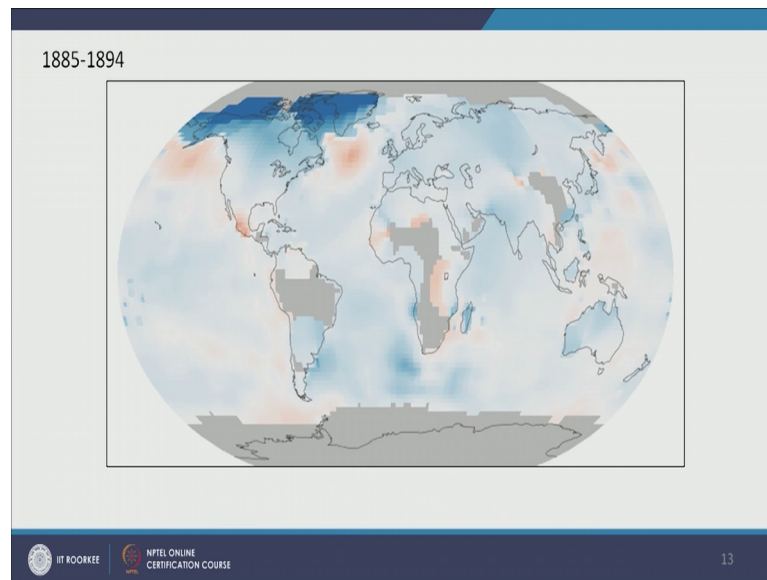
So, think of it this way the petrol is a very stable form of carbon, it stays and under high pressure below the earth's surface for millions of years so it can stay. So, that is the carbon that we have trapped in under the earth's surface, it is sequestered carbonate it has

it is not there in atmosphere we burn it we release it in atmosphere. So, now, we have greenhouse gas effect so as the carbon emissions have increased, our global temperatures have also increased. So, on the right top panel you have how with years the parts per million of global monthly mean on y axis how they have changed.

So, in 2004 we were hovering between 380 and 370 and then by the time it was 2007, we were already talking about 385 and 380 and now in some places we have exceeded 400. So, it is really sad because we are already passed the tipping point now. So, 350 was a number that was given that do not exceed 350 if you want to save the earth we already passed; it way long before this is a decade old data, we are way past it and because we have way past it we know that our climate is changing. We know that our food cycles with change, our weather patterns would change, our agriculture would suffer right, our storm cycles would change and thus the risk of damage we pose to environment; to public health and to health of other species whether it is animal microbial is immense and unprecedented.

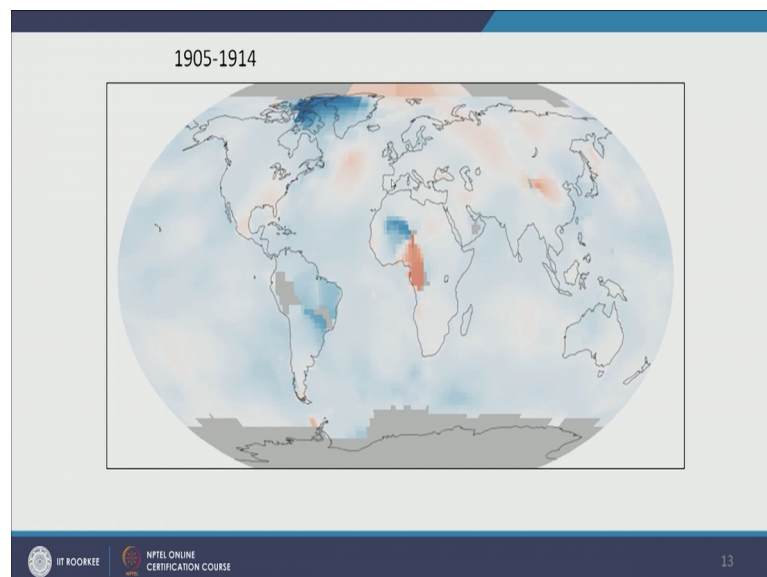
So, on the bottom left panel here we have decades on the x axis and on the y axis you have annual temperature. So, if we take newly 1970 and 1960 as the average and we notice how temperatures have steadily increased over last 6 decades and they keep increasing and they continue increasing. In fact, this rate of increase has never been seen on earth before usually there are annual variations but they are much smaller than what we are staying here.

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So, I want to end this presentation by showing you a slide view this is from NASA website of how global temperatures have changed over years. So, 1885 1894 so the more blue the colder it is the grey we do not have information about and the red is warm; so, we have very cold we have some warm here.

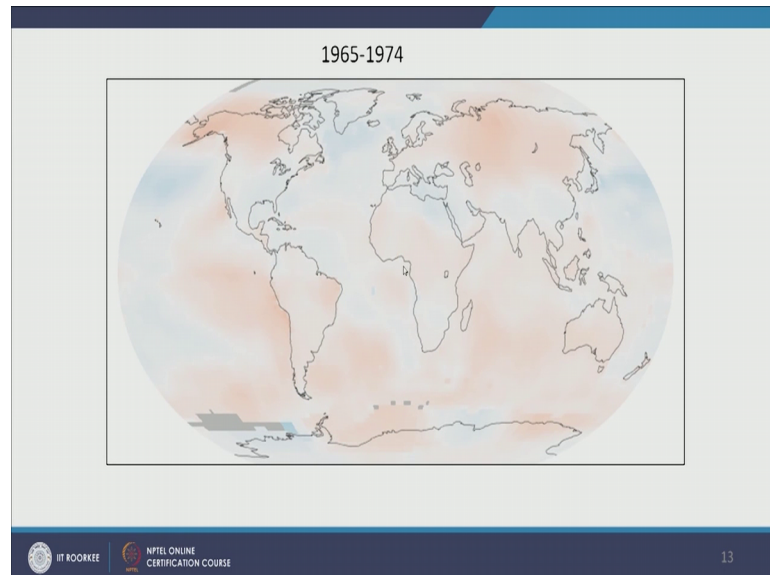
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So this temperate 0 a decade later the coolness look here, the all the blue is gone we have more red the even here even the African continent is getting really red, china is getting

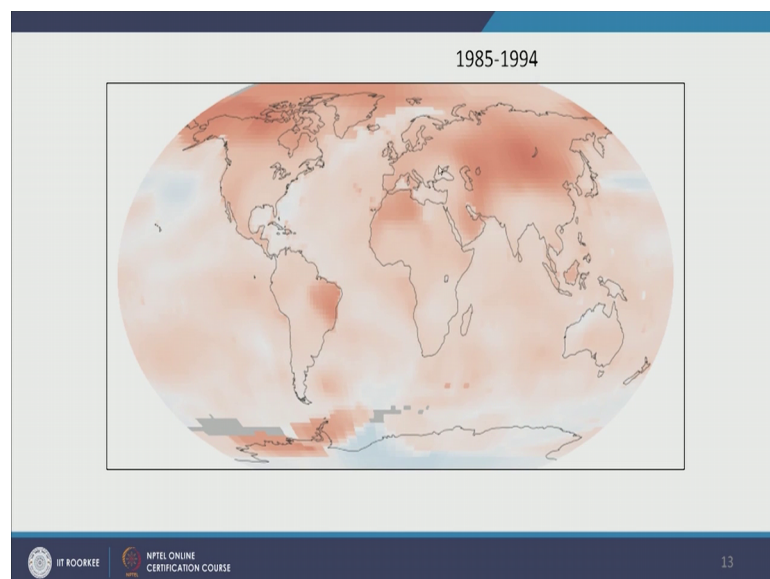
very red here, look at India is relatively cool. So, our ancestor's pre independence as ancestor's pre world war ancestors did not suffer the heat that we do in India now.

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Welcome this 1965 1974 the whole world is much warmer even in south part of India is getting warmer. So, our grandparents must be able to tell us that well south India is to be hot, north India is to still be cool.

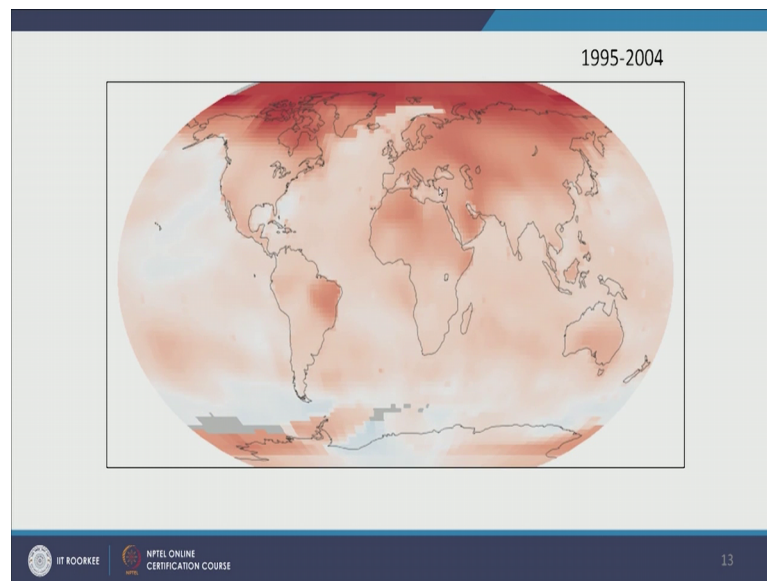
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But it 1985 1994 now the entire world is has heated up really fast very few blue portions left India is now a red zone.

India is a hot country now and you might notice that Russian and North America seem warmer than India, but please note that these are increase. So, they used to be very cold now they are less cold. So, in Siberia for example, they had permafrost but now permafrost is releasing right. So, this is not warmer than India, but the rate of change of temperature is higher here than.

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There this is 1995 and 2014 and this is what I will deal with as we end this the class. So, to recap we started here nearly a century ago and in last hundred years this is where we have reaches.

So, dear students I really hope you understand that in climate change is a very important and very real phenomena, we humans are responsible for it; we are in middle of sixth mass extinction phase not surprisingly it is our duty for to reduce the impact of climate change, to reduce the climate change in at the very first and one of the ways to handle climate changes to understand how the nutrient cycles move in our earth, because carbon climate changes because of higher carbon in air. So, it is the carbon cycle that has been affected.

So, we know there are human emissions, anthropogenic emissions, but we also have to understand the microbial process is how they have changed with this increased release of carbon in atmosphere; how they are wrecking what these changes are going to mean for us and how we can undo them. So, it is very important to understand the microbial

process is that governed the nutrients and thus this course is really important and I really hope that with in coming near future people will appreciate the microbial processes and their role and climate another global problems, so that is all for today.

Thank you so much.