Environmental Remediation of Contaminated Sites
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Lecture – 54
Bioremediation: Part - II

Hello everyone, so welcome back to the latest lecture session, so within the context of I believe, remediation of soils and sediments, we were looking at soil vapour extraction, right as in you know, you have relatively volatile compound, we are looking at contamination of the relevant gas let us say in the vadose zone and how do we treat that particular contaminated gaseous phase and so on, right.

And then we started looking at bioremediation now, right that is something we are going to continue with today and may be once we wrap up bioremediation, we are going to look at the relevant case studies for soil vapour extraction in bioremediation let us say, right, so let us look at what we have discussed thus for.

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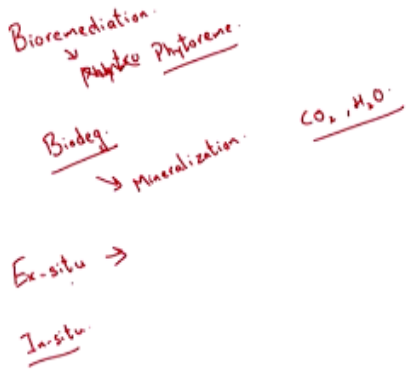

So, we looked at or introduce ourselves to a few terms, one was bioremediation, right, so obviously what do we have out here let us say, you know here let us say, the aspects were that we have living organisms to could be microbes or plants and they bring about let us say either degradation or immobilisation of the relevant contaminant, right, so if it was only living plants

that we are going to talk about or considered that would have been or that would be referred to as phytoremediation let us say, right, phytoremediation.

But when we talk about bioremediation typically, let us say it encompasses both what do we say the action by the microorganisms or the relevant plants let us say, right and again the key aspect is either degradation or even immobilisation let us say is consider bioremediation let us say, right and I believe then we looked at other aspects as in what is biodegradation let us say, right, so I think we more or less I mean it is self-explanatory, right.

You have a transformation of a relevant compound or a conversion of relevant compound from one form to the other and obviously, you have biological process involved, so biologically mediated conversion of relevant compound let us say, that we refer to as biodegradation, right, in that context obviously we have mineralisation, so when you have complete degradation let us say, right towards the relevant end products let us say, right.

You are going to for example to CO_2 and H_2O and so on let us say, right then you say that you know mineralisation has occurred, typically let us say this is what you would want let us say because some of the by-products during degradation could also be toxic let us say, right, so again mineralisation is something that you would typically prefer let us say, right and this is something we you know, these are some of the aspects we looked at.

And then we saw the cases for application of what do we say bioremediation ex situ and in situ let us say, right and ex situ we also looked at land forming or such land piles let us say, right but in that context, we saw that you know you need to have an impermeable or semipermeable layer beneath right because you do not want the relevant contaminant to reach the relevant groundwater or contaminated the soil beneath the trait.

So, there are different aspects and we briefly discuss them and obvious advantages for that let us say you know the costs are going to be less, typically, relatively more acceptable to the relevant communities, right, you know biological process typically more acceptable, typically any way

obviously, right and what does we have and here typically, we actually have degradation or most of the time we have degradation of the relevant compounds, most of the time anyway.

And then obvious disadvantages time, right, they are typically slow and depending up on the varying site conditions or you know site conditions let us say or the heterogeneity, the relevant process can be inhibited let us say right, you can have some toxic compounds being produced as by-products that is certainly an issue right, so these are some of the major disadvantages, right, again we just looked it in over view let us say, right.

So, we are going to obviously look at the relevant aspects or you know the relevant aspects as and what do I need let us say to be able to promote or you know have bioremediation to occur let us say right.

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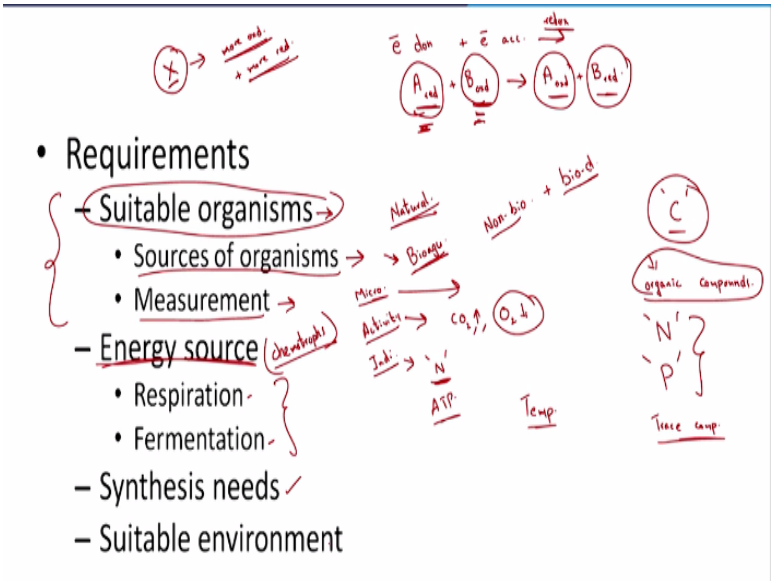

So, obviously the first aspect would be, right to have suitable microorganisms, right, so in this context, right we also looked at you know some aspects or similar aspects in the context of natural attenuation obviously, you want microorganisms, right but what role do these microorganisms typically play now, right and why do they play that role let us say or why do they want to play that role?

So, here you have different compounds and while degrading these compounds, energy will be released let us say or while the redox process takes place let us say typically, energy is released and the microbes let us say for either their growth let us say, right or self-synthesis, they want this energy let us say, so for that particular case, these microbes play a role of let us say catalyst, how do they do that?

They release enzymes which act as catalyst thus shortening the relevant what do we say time required or fastening the kinetics of degradation of the relevant compounds let us say, right, so obviously you need suitable microorganisms now, right. So what are the different sources of organisms let us say, right, so there are 2 aspects obviously, one is natural selection based on natural selection.

As in let us say you know everywhere we are going to have typically microbes present unless the conditions are remarkably toxic to almost all kinds of microbes but that will rarely be or ever be the case let us say, typically you will have some kind of microbes or the other out there, right, so when I refer to natural selection or such, what does that mean now; so based on the relevant conditions at the site let us say, you know certain microbes are thriving, what is that mean let us say?

They are the fittest for thus those conditions now, right, typically we know that you know survey we referred to the term let us say or talk about the term survival of the fittest now, right, so natural selections, so based on those conditions let us say or the stress there or extreme conditions out there in that particular site, certain microbes are thriving and typically those microbes are obviously, relatively better of; at you know taking the process through, right.

So, ideally, you would want to promote growth of those kinds of microbes let us say and thus obviously are from natural selection now, right, so that is one particular case or you can introduce microbes let us say, right and in that context, we refer to that as bio augmentation, right, so you are augmenting the process let us say, right and you are typically going to introduce microbes.

And typically, these particular supplements if I can call them maybe not supplements, these what do we say, non-native species if I can call them in this particular case let us say, right or typically sold but again they are obviously costly, right and also keep in mind that they are not what do we say, very site-specific to those conditions, yes, so obviously you are going to have different variables at play there right.

But typically, when would this particular bio augmentation work; it would typically, work let us say when let us say you have relatively non-biodegradable compound in a mixture of relatively more biodegradable compounds let us say, right, so typically, you knowing in these conditions let us say, you will typically have what do we say greater chances of success for this particular bio augmentation now, right.

Again, 2 sources of microorganisms; one in the natural let us say the one that are thriving or you can introduce non-native species let us say from other locations that you know you have what do we say cultured and grown and so on let us say, right and different aspects out there, right, when we add microbes there, right, so something to keep in mind and how do we you know go about the looking at the relevant measurements now, right.

So, we can look at different aspects; one of them would be directly measuring the microorganisms let us say, right so that is one particular case or measuring the activity that we see or we assume to see, right or indicators of let us say microbial activity let us say, right, so we are going to look at these three aspects, so with respect to measuring microorganisms are just give a very generic overview let us say, right.

So, you extract them from the relevant soil let us say or dilute them let us say right and dilute them, right, you extract the relevant microbes from the soil of your particular matrix and you dilute them let us say and then you are going to grow them let us say, and cultures let us say, you are going to put in conditions that promote the growth of these particular microbes let us say right.

So, you can do that on membrane on plate or in tube let us say right, so once you grow this particular what do we say microorganisms, you are going to have to count them, right or there are different methods now I believe, right and in that manner, you can measure the relevant number of microbes present as in you know the rate at which that they you can typically grow let us say and you can thus estimate the relevant amounts let us say, right.

Again, that is one particular case let us say where how you can measure the microbes, now right or you can obviously measure the activity now, right, so activity in the sense let us say as we looked at in different cases, you need not directly measure the relevant compound but you can look at the relevant by products or so on, now right. So, what are they or how do we go about that let us say?

In this case, they can be carbon dioxide let us say, if typically you know you have your relevant redox process going through, right what is going to happen; you are going to have if there is mineralisation, the carbon let us say is going to be oxidised completely let us say, you know I am assuming the particular example, so what would happen in that case typically, we would estimate or assume that carbon di oxide is going to be produce.

So, you can look for either effect on pH or let us say the carbon dioxide concentrations and so on, right or depending upon the type of process, you can look at the consumption of or decrease in oxygen let us say, right, you can look at the stoichiometric, right how much oxygen is required for particular process to go through and you can look at the rate at which or the level of decrease in oxygen and then estimate the relevant activity let us say right.

So, different ways obviously out there or there are other indicators again, right and for example, let us say, what are we looking at here, for example here you know, microbes have you know the protein content there, so you can look at the nitrogen content, right to be able to indirectly estimate the amount of or the microbial activity let us say right, the nitrogen content or the ATP let us say can be analysed for I believe right.

These are energy carriers here, right, so there are other indicators obviously that you can use to be able to measure the relevant activity at your particular sites let us say right, so these are some of the aspects to keep in mind in the context of measurement now, right. So, let us look at the other aspects so, first obviously you need the organisms; suitable organisms so that is something we looked at.

Sources and measurement of these organisms we looked at that let us say, so now let us say moving on to the relevant aspect, you need an energy source let us say, right, so in the context of energy source obviously, there are 2 aspects to look at; one is obviously that you know of respiration, so you have an electron donor and an electron acceptor, right, so in the context of respiration, you need an electron donor and an electron acceptor.

And then there is going to be the redox process going through, right and then you are going to have the relevant by products, this is the redox process right, reduction oxidation and then you are going to or the microbes are going to facilitate this process by you know releasing the relevant enzymes and thus you know gaining the relevant energy let us say for their own needs now, right.

So, obviously, an example can be let A which was in its more reduced form + B which was in its more oxidised form is now transformed into A in its more oxidised form + B and its more reduced form, so A which was relatively reduce now, transformed into a more oxidised state let us say, right, so, what is happening out there or B which was relatively more oxidised let us say is transforming into relatively more reduced state let us say, right.

So, what is happening out there; oxidised form to reduce right, so oxidised state is decreasing or you can understand that it is taking in electrons, right, so this is taking in or accepting the electrons transforming into a more reduced form and what is happening to this particular compound, obviously, this particular compound is accepting electrons, this particular compound is donating the electrons and transforming it into more oxidised form let us say, right.

So, again one of your particular contaminants will either be A or B right, depending upon on your type of conditions out there right, your compound can; the contaminated compound let us say can either be a relatively more reduced compound or oxidised compound, right and then you can now look promoting which conditions and so on and so forth but obviously, in respiration as we discuss we need to have electron donor or electron acceptor and so on; independent electrons acceptors and donor.

So, how does it differ from fermentation though, right so in fermentation, you have one particular relatively semi-oxidised compound if I may say so, right and I believe we did look at one particular example or a few examples in the context of natural attenuation, right, so this compound let us say well be transformed let us say, you know compound x will be transformed let us say into more oxidised form plus more reduced form let us say, right.

So, obviously this is semi oxidised or such let us say, right, I think we looked at acetic acid or some such example which I am unable to recollect right now, the specific example anyway, so you know in the case of fermentation, this is what you have, you have single compound and its transform into more oxidised form and also more reduced from let us say, right and that it is the case that we looked at that.

So, these are the 2 cases and obviously, you look at chemotrophs, right, they derive energy from this particular chemical process let us say, right or redox process, chemotrophs let us say, right, so what do we have here; we will have microbes and we have the different or major components for deriving the relevant energy, is that enough for the microbes to thrive and degrade the relevant contaminants, not really right.

So, what else is required let us say, right, so what else now, so in this context similar to human beings let us say you just satisfying one particular need is not good enough, you know humans to thrive let us say need have different needs now, similarly, microbes have different needs now, for example they need carbon, right they need carbon now, let us say again for cell synthesis and typically, let us say your organic compounds let us say or let us say heterotrophs let us say if I

may relatively more heterotrophic compounds, pardon me or let me call them organic compounds for now, right.

Organic compounds have your carbon, right and the carbon have the needs of the relevant microbe for carbon let us say for cell synthesis let us say or met by these organic compounds let us say, typically let us say, right and also what else do they need; they need nitrogen and also they need the source of phosphorus let us say, maybe not as high as carbon but this will need you know nitrogen and phosphorous at certain levels.

So, if you do not have some of these key aspects and you just have the electron donor and acceptor, you know the microbes will not be able to thrive, so obviously all these aspects also need to be looked at let us say and then there are other trace compounds that are also required a trace nutrients if I may say so, again you know we are not going to that in that detail in this particular class let us say.

Because that is slightly out of our particular domain here, let us say, right, again what do we need rather than just the microbes and the electron acceptor and donor, we have other major nutrients and minor compounds or trace nutrients that are required let us say, right, so that is something to keep in mind and now, obviously you also need conditions let us say as in similar to human beings, the microbes also have a certain preferable temperature range, right.

So, temperature is a key issue, so you are going to have temperature as in we are done with synthesis needs, we are looking at suitable environment let us say.

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So, what are some of them let us say obviously, temperature is an aspect yes, you know there is an optimal range let us say, typically and obviously pH right, way to acidic or basic typically, you know the microbes cannot withstand these extreme conditions, right, so that is something to keep in mind too, right, so again temperature pH and you have other aspects too obviously, right, so let me look at some of the other aspects we are going to cover now.

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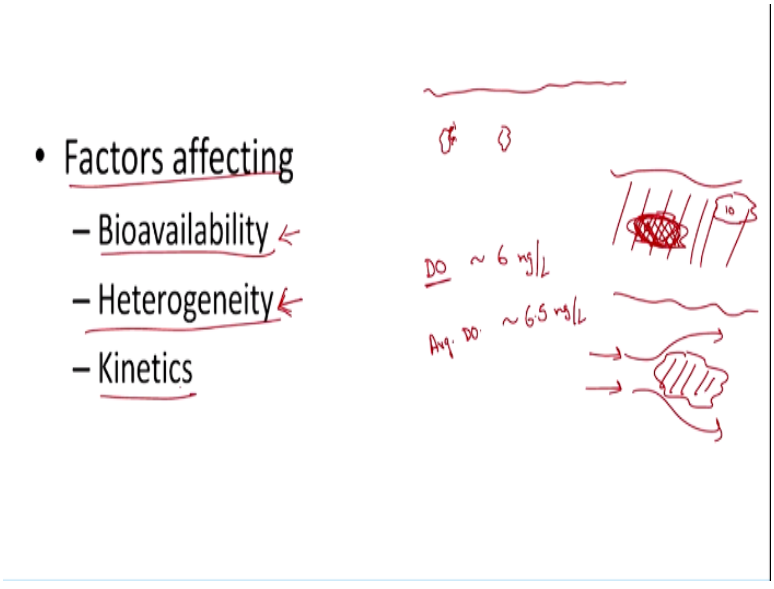

So, here we are going to look at let us say a particular term called bioavailability let us say, right, a in there are bioavailability actually encompasses quite a few aspects but I am going to talk about let us say the access in this particular context let us say, I am only going to talk about or

restrict myself to let us say talking about the availability of a particular what do we say compound let us say for the microbes to utilise let us say, right.

But again as I mention, bioavailability also looks at or encompasses other aspects which are not going to look at let us say, and why is that obviously, an issue let us say right and typically, bioavailability is affected by the extent to which or you know the time to which let us say or after which you are trying to kick start your remediation, we will look at why let us say. So, for example, let us say you have a site contaminated with relatively hydrophobic compounds let us say, right.

So, what do we have out here; we have let us say the hydrophobic compound out here, this is the surface and of you have the soil particles let us say and let us say initially, let us say you have your compound out here let us say, right and then it is adsorbed onto the surface let us say of this soil particle, right, if this was the initial case and you know the microbes are present let us say, they can utilise these particular contaminates.

But what would happen let us say, over time and if there were no microbes present initially let us say, so what would happen now, you would have what do we say this particular compound you know reaching the relative interiors of the relevant soil particle let us say right or it is no more at the relevant surface now, right. So, what does that mean let us say, for the relevant compound is not available for your particular microbes, right.

So, you know the bioavailability is an issue in this particular context, now right, so let us say if considerable time has passed since the initial spill or you know the what do we say contamination of the site, what would happen; the relevant contaminant would travel further into the interior of your relevant soil particle let us say, thus at the surface were the microbes would typically be present or be available let us say, the compound is not available.

As in though within the soil, or if you know if I look at the total concentration, yes the relevant compound is present but it is not available to the relevant microbe, right, so again let us say there are kinetics set play but what kinetics now; the rate at which the compound diffuses out of the

soil particle onto the surface again let us say and these aspects will obviously, play a role, right, so obviously bioavailability is a key issue.

And let us say, any toxicity or such that is affecting the microbes or such are also covered but I am not going into that at this particular stage let us say, so heterogeneity let us say, right, so why is this important or how is this important now right, so heterogeneity let us say you know and there are number of variables out there in the site let us say, so typically let us say if this is my site again side view and if it is a homogeneous mixture let us say, not mixture, matrix.

Obviously, you know it makes the job much easier for me but typically as we know and as we have been witnessing or looking at different case studies, we have never come across the site where you know the conditions are homogeneous, right you have different layers let us say and so on and so forth let us say, so how does it affect your particular case let us say; let us look at one particular example.

As in for example assume, we want dissolved oxygen to be around let us say 6 milligram per metre let us say, right, so let us say somebody comes to you and tells you that the average dissolved oxygen concentration is 6.5 mg per litre, so then they asked would you be able to take a call with about you know is this particular site you know feasible for bioremediation or not let us say, assuming that all the other variables are you know according to order let us say, right.

So, again keep in mind obviously, we are talking about heterogeneity here right, so the key issue is that let us say if you had zones where the dissolved oxygen is 0 and zones let us say, dissolved oxygen is relatively high let us say, right, may be 10 or 9 depending upon the saturation levels let us say and it works out in such a way that the average is around 6 of 6.5 let us say, right, well the rates also the average rate also be you know according to this particular DO values, no, why is that?

If it is 0, let us say for the particular example, I am looking at let us say which requires dissolved oxygen, there will be no removal or degradation within this particular region where the dissolved oxygen is 0 but there will be removal at that particular region where the dissolved oxygen is

available or above the relevant threshold let us say, right but obviously, it would not work out as it, it is an average case, right.

I mean as in you can calculate the average rate and so on, right, obviously heterogeneity is a huge issue now, right, huge issue in the sense that you know you need to have the relevant what do we say nutrients or the electron donor or the electron acceptor or the dissolved oxygen and so on for a comfortable or you know at the relevant levels let us say, right so that is something to keep in mind.

And also another aspect is that let us say you know if your site is heterogeneous and you have relevant contaminant being transported let us say and here you have a relatively impermeable layer here let us say, what is going to happen; your ground water flow patterns might be relatively different, right, so again because of these physical characteristics of the relevant soil or such let us say, what is going to happen now?

You are going to have issues with you know your particular what do we say contaminant concentrations let us say at those particular localised zones let us say, so right, so it is again its you know you need to understand that there are different variables at play and the greater the complexity of the system or the heterogeneity of other system, you know the greater the complexity of the system I guess, right.

Heterogeneity and complexity of the system go hand in hand now, right that is some aspect or one aspect to keep in mind now, right and obviously, the other aspect that we are going to look at is kinetics let us say, right obviously, we talked about one of the disadvantages of the relevant what do we say, microbial process based degradation or bioremediation being kinetics let us say, right, so we obviously need to understand what are some of the factors that affect kinetics here, right.

So, let us briefly talk about that and end that; end this session for today, right, so what do you need; obviously, you need the macronutrients, micronutrients and so on and so forth but

obviously, you typically need your electron acceptor let us say, right or electron donor let us say, if let us assume that is the case here, let us say, right.

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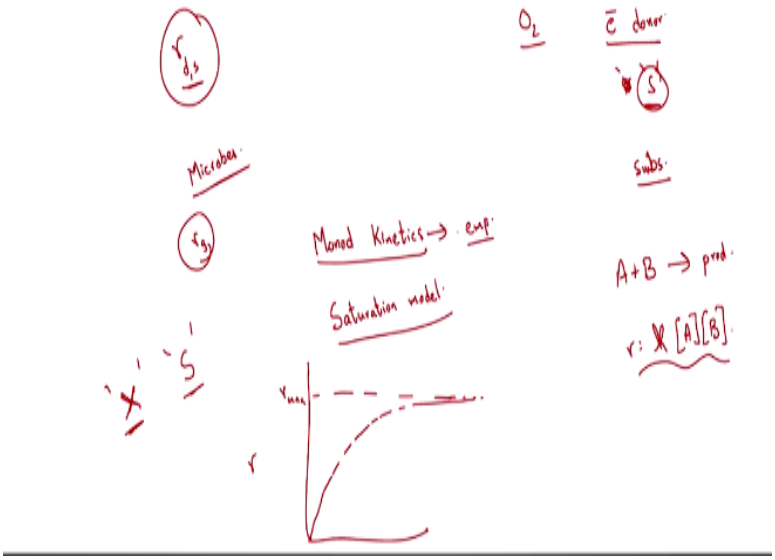

As in I have let us say assume I have oxygen which is going to be my election acceptor let us say, right and I need an electron donor let us say, right and I have some compound x let us say, right or s let us say which is my contaminant which I want to degrade and which can act as an electron donor let us say right, so let us say it some reduce the form of carbon let us say, right, so what will this particular, what do we say rate of or the kinetics of this degradation depend upon now.

So, I am looking at rate of you know, degradation of this particular compound, yes, so in the context of you know, biological process, they look at this or refer to this compound as substrate let us say, right and again this is should be familiar to most people who have had some what do we say knowledge of waste water treatment let us say right typically, they talk about it as substrate let us say, right.

Again, this is the compound that is rate limiting or is required for the relevant microbes let us say to be able to carry out the development degradation let us say, so I am interested in this rate of degradation of these this particular substrate or this particular compound, so what is it depend

upon obviously, it depends upon the microbes and the rate at which they are growing let us say, right.

As in typically let us say if you have compound A + compound B going to products, right the rate would be r = the rate constant times concentration of A, concentration of B and so on, right, yes that is you know, if it is a chemical reaction typically that is what we have out there, right, it depends upon the concentration of the relevant reactants and the rate constant, right but here obviously, there are other aspects it play.

Because you have living organisms present, so the rate of growth of these microbes is obviously very much important for to look at or to understand the rate of decay or not decay, degradation of this particular relevant compound, right, if your rate of growth of the microbes is higher, typically the rate of growth of; not growth pardon me, the decay or the degradation of your relevant contaminant will be higher.

As in let us say, your contaminant is being eaten up by a few microbes let us say, obviously if they are thriving well and there are more number let us say the rate at which this particular contaminant will be eaten up or degrade will be also greater let us say, right, so different aspects of at play, here and typically we referred to the relevant kinetics as mono kinetics, right, so this is what we are going to look at, right.

And again what are the obvious aspect that is going to or they are going to be depend upon, so one case to keep in mind that it is a saturation model, right, it is a saturation model, again we look at these aspects let us say keep in mind that there are certain thresholds above which let us say or the maximum values above which let us say, it cannot increase let us say, right, so that is going to be the case.

As in let us say, if I have this particular case are here and substrate concentration let us say, right, so it is going to be let us say something like this let us say, right and this is my maximum let us say or max let us say, right, so there is going to be a certain maximum, so it does not mean as if I

have lot of what do we say food out there for the microbes, they are going to keep increasing and increasing and increasing, there is some maximum rate of growth, right.

And again, there are other aspects at play, obviously what are they going to be depend upon; they are going to be depend upon the concentration of the microbes, right which I am going to represent by x, concentration of the microbes that are present or the amount of microbes that are present, there it is going to be depend upon the amount of food that is present, right., so again these aspects and there are particular, what do we say variables out there.

And again based on that we are going to come up with in empirical equation which we are going to to refer to as mono kinetics I guess, but we are going to look at this in greater detail in the next class, so for today though, I will end this session for now and thank you.