Water and Wastewater Engineering Dr. Ligy Philip Department of Civil Engineering Indian Institute of Technology, Madras Lecture-19 Microbiology

In the last class we were discussing about the secondary wastewater treatment systems. The secondary wastewater treatment systems are very very important because they try to remove the organic matter present in colloidal form as well as in the dissolved form. These organic matters which are removed in the secondary treatment system are biodegradable and hence the microorganisms are one of the reactions for removing the organic matter. So the other reactant that is required in this system is the dissolved oxygen.

It is essential for us to know about some aspects of microbiology connected with the wastewater treatment system that is secondary treatment of wastewater. So in this particular instance we were discussing about the bacterial growth curve and we also discussed about kinetics of bacterial growth to some extent. These are two important things we should study to understand the secondary wastewater treatment systems which employ the microorganisms.

So, in the kinetics of bacterial growth we did derive a fundamental equation for the growth of microorganisms dx by dt is equal to mu into X this equation we developed so X is the weight of biomass in milligrams per liter and mu is the specific growth rate of microorganisms. The specific growth rate is dependent upon type of food available for the microorganisms and also the type of microorganisms themselves. So it is a function of food as well as the microorganisms.

(Refer Slide Time: 3:33)

Now, this specific growth rate is not a constant but it is a function of the substrate concentration, S is the substrate or the food for the microorganisms. The food for the microorganisms should provide carbon source and energy source that is what we discussed in the last class. This also could be the bio chemical oxygen demand BOD of the wastewater is the substrate. So this is a function of, there is a mu specific growth rate which is the function of the substrate concentration given by the equation mu is equal to mu max multiplied by S over k_s plus S.

(Refer Slide Time: 5:23)

In the last class we had defined all the terms, still I would try to redefine these terms. This equation is Monod's equation developed from mi clase mendense equation. In this mu max is the maximum specific growth rate constant, this is the constant this is the maximum growth rate constant and S is the substrate concentration and k_s is the saturation substrate concentration. So, if want to plot this particular graph (Refer Slide Time: 5:25) the graph looks like this.

(Refer Slide Time: 6:15)



That is if I make a plot between S that is the substrate in milligrams per liter again the substrate is measured in milligrams per liter as well as microorganisms are measured in milligrams per liter versus mu that is specific growth rate of the microorganisms then as the substrate concentration is zero the mu is also zero there is no growth rate and as we increase the substrate concentration mu also increases. This particular thing reaches the maximum value and this is called mu max.

So mu will reach a maximum value for a specific concentration of the substrate and even if we increase the concentration of the substrate the mu will be only mu max. That is, when mu reaches mu max it is independent of substrate concentration, till such time the mu that is the specific growth rate is a function of the concentration of the substrate. When it reaches mu max it is no longer a function of concentration substrate, this particular equation. This particular curve is nothing but a rectangular hyperbola, equation of rectangular hyperbola and now we define the ks here (Refer Slide Time: 7:10) saturation constant is nothing but concentration of the substrate this is ks then mu is equal to half of mu max that is what we discussed in the last class.

Now we move forward from this equation. this is the what is the equation what we have; the dx by dt now can be written as mu max S over k_s plus S multiplied by X so this is the equation substituting for the mu so this is the value of the mu that is the specific growth rate. In terms of maximum growth rate constant, saturation concentration of the substrate and the substrate concentration itself that is what the equation is. Now here this is our fundamental equation may be you call it number one.

(Refer Slide Time: 8:20)



Now we consider two cases; case number one. In case number one, what I consider is I have unlimited concentration of substrate so S is unlimited that is I have very high concentration of substrate available to me available to the microorganisms. That means plenty of food is available. So when plenty of food is available this particular condition is possible only during the initial period of the reaction when we have plenty of food.

During that particular time we can assume substrate is unlimited as a result of which k_s is very very less compared to S that is in the denominator k_s is very less compared to S and hence we can say k_s plus S is approximately equal to S itself.

So substituting this in this equation dx by dt I have now equals mu max over mu max into X. This is one equation I have, this is the second equation (Refer Slide Time: 9:45) I can call it substituting this k_s plus S is equal to S and then that S and this S cancels out and multiplied by X this is the first order equation for the growth of microorganisms.

(Refer Slide Time: 10:00)

And this equation can be solved now. I can solve this particular equation because mu max is constant and dx by dt depends upon the concentration of the microorganisms. That means if you try to solve that equation now, it is very simple; dx by dt dx by X equal to mu max into dt then integrating this particular thing at time t = 0 we assume that the microbial concentration is X_0 and at time t = t the concentration microorganisms is X. Then solving this particular equation I have ln X over X_0 with the boundary conditions mu max into t or $X = X_0$ mu to the power of mu max into t. So this particular equation is the first order equation. It represents exponential growth phase of microbes in microbial growth curve.

(Refer Slide Time: 11:30)

We considered microbial growth curve last time which is nothing but the exponential growth curve. Again may be I can plot this exponential or growth curve of microorganisms. Let us redraw the growth curve and identify where exactly this equation holds good. That is, t is the time and this is the microorganisms concentration, I have a lag phase, this is X_0 , this is the initial organism, lag phase and lag phase is lagging behind and this is log growth phase (Refer Slide Time: 12:30) that is growing exponentially and we have what is called a stationary phase and decline phase. These are the four phases which we discussed. Just I wanted to reiterate that particular thing. So this is the equation whatever I have written, it is for this particular phase log growth phase.

(Refer Slide Time: 13:00)



This represents a log growth phase (Refer Slide Time: 13:06). In log growth phase I have a substrate unlimited. That means this particular curve can be represented by the equation dx by dt equals mu max into X this is the log growth phase. What is happening afterwards in log growth phase is the microorganisms are growing in log exponential order and the food that is available is getting depleted as the result of which with time the food gets depleted. So, as the food is getting reduced, the quantum of food is getting reduced we can consider this as the second case, case number two.

Phase number two is when the food is limited S is limited. The first case is unlimited food and second case is limited food. What would happen in this particular case is the concentration of substrate is less than k_s . That means the concentration of food is less than k_s , k_s is the saturation concentration of substrate and the food available is less than that particular thing.

In most cases, in wastewater treatment plants particularly in domestic wastewater treatment we encounter this particular scenario, this scenario is there while in the industrial wastewater where the BOD of the wastewater is very very high we may get the first exponential growth phase. Here food is limited, food is getting exhausted and it is

limited. So when this is the scenario what we do is k_s plus S is approximately equal to k_s itself. Substituting this particular thing substitute in equation one and substitute in this particular equation what would happen is dx by dt now would be equal to mu max over k_s multiplied by S into X.

(Refer Slide Time: 15:40)

In the denominator k_s plus is made equal k_s only because S is very small and in the numerator I cannot neglect that S so I am writing S into X is equal to, dx by dt is equal to mu max over k_s multiplied by S multiplied by X. So you can see here there are two variables; one is the S and another is X and I cannot definitely solve this particular equation like I could solve the other equation. So mu max is constant, k_s is constant these two things are constants, this particular value is a constant (refer Slide time: 16:30). However, dx by dt varies is a function of concentration substrate and concentration microorganisms because of the simple fact that the food is limited.

When the food is limited the growth rate is the function of the concentration of the substrate. When the food was not limited then it was not the function of the concentration of the substrate that is what it tells here very clearly. So when lot of food is there then the growth rate is not dependent on the concentration of the food but when it is limited it is a function of the concentration of the food.

Now, S and X if you try to look at them as what is this S and what is this X they are dependent variables. What we mean by that? That means, as the substrate concentration is increasing, when the microorganisms utilize the substrate and hence substrate concentration is decreasing then the microbial concentration increases.

In other words as S is decreasing as shown with an arrow mark then X will increase. There is increase in the concentration of X because of decrease in the concentration of S that is why they are dependent variables. Hence I can derive an expression between S and X. So that expression is nothing but what is called yield coefficient. Yield coefficient gives the relationship between S and X, yield coefficient is given by weight of biomass produced. biomass is X here I will put it as X produced per unit time in a given time divided by the weight of substrate that is the food utilized by the microorganisms in unit time. This is what is called yield coefficient.

(Refer Slide Time: 19:05)

So mathematically I can write this y; y is the yield coefficient that is nothing but how many kilograms of microorganisms are produced in a unit time per kilogram utilization of substrate by the microorganisms. If microorganisms utilize one kilogram of substrate how many kilograms biomass is produced that is nothing but yield coefficient. It is just like the yield of the crop.

This can be mathematically written as biomass increase dx and decrease in the substrate concentration minus dx by dt and for a unit time dx by dt I put it here and ds by dt I will put it here (Refer Slide Time: 20:00). Therefore, in other words y is nothing but dx by dt over minus ds by dt. Here I am using the minus sign because substrate is being utilized and hence there is a decrease in the substrate concentration. Now this particular equation can also be written as, in the finite terms of time. Suppose I take a finite time suppose X is the concentration of microorganisms at any time minus X_0 that is initial time X_0 divided by the concentration, initial substrate concentration is S_0 at time t equals to zero minus S. This is the expression we have.

(Refer Slide Time: 20:55)

Hence, in this particular thing X_0 is the concentration of microorganisms at time t = 0, X is the concentration of microorganisms at any time, similarly X_0 is the concentration of substrate at time t = 0 and S is the concentration of substrate at any given time. So this X_0 minus S is the substrate that is being utilized by the microorganisms. When they utilize the microorganisms this much amount of biomass is produced. So this is the equation what we have. Let us see little more about this yield coefficient. When we consider the yield coefficient this is what we have to follow.

Suppose I give the substrate, substrate again I will reiterate, substrate is the food, substrate is the BOD biochemical oxygen demand that is present in the wastewater. So when the substrate is given to the microorganisms that means I will give the substrate to the aerobic microorganisms. Aerobic microorganisms are the organisms which utilize the organic matter in the presence of oxygen that is why they are called aerobic. They will use in this fashion, they will use around 50% of the substrate for production of energy and energy production is manifested by the production of carbon dioxide and water.

In any biological reactions if carbon dioxide and water are produced it means that the energy is produced, the cell is deriving the energy from the substrate, remaining 50% of the substrate is going for the biosynthesis, this is biosynthesis (Refer Slide Time: 23:00). Biosynthesis means, this results in increase in biomass that is nothing but X minus X_0 increase in biomass concentration X minus X_0 that is what I have written there in a unit time that is what it is going to happen.

(Refer Slide Time: 23: 30)



In an aerobic system 50% goes for energy, 50% goes for biosynthesis. If that is the case can you tell me what would be the value of y in this particular case? The value of y in this particular case is 0.5 that is the maximum value. That means as you can see here the denominator is the weight of biomass produced over unit weight of substrate utilized, unit weight of substrate utilized is this, 50% goes for the energy, 50% goes for the synthesis and hence whatever is in synthesis the y where y has the maximum of 0.5, in fact in actual case it will be less than point five which can be the maximum value. That is the importance of the yield coefficient in the aerobic systems.

Suppose the same substrate if I give to an anaerobic systems, anaerobic microorganisms the anaerobic microorganisms do not utilize the oxygen, oxygen is not supply to them, no oxygen. In the absence of oxygen they work. When these works in the absence of oxygen what would happen is they would tend to use 70 to 80% of the substrate for production of energy. For energy production they use about 70 to 80% of the substrate. The end product of this particular thing could be carbon dioxide, water and also another gas called methane. Some energy is locked up in the methane that is why substrate is used for the energy production. Anaerobic organisms are not very efficient in oxidizing the organic matter for the production of energy while remaining part that is 20 to 30% of the substrate goes for the biosynthesis in the anaerobic system, only 20 to 30%.

Now you can see what is the yield coefficient for anaerobic system. The yield coefficient for anaerobic system is approximately 0.2 to 0.3 or even less than that. This is the maximum possible value but in actual practice this will be less than 0.2 also. So you can see here the yield of microorganisms in anaerobic system for the same amount of substrate is less, for aerobic system it is more. That means if you are employing aerobic systems for the treatment of wastewater then you will produce more microorganisms in the anaerobic system.

This is one of the differences we can right away say between the aerobic systems and anaerobic system.

(Refer Slide Time: 26:46)

Continuing our discussion with respect to the yield coefficient you can see here from that equation; dx by dt is equal to minus y times ds by

dt where y is less than one that is why dx by dt is not equal to ds by dt but it is less than ds by dt because y is less than one. Now instead of this negative sign what I will do is the negative sign I can take it inside and write it as ds by dt utilized, ds by dt u is the rate of consumption of substrate or utilization of substrate, rate of consumption of substrate.

(Refer Slide Time: 28:10)

So when I write u utilize, then I take of the minus sign that is all. Now these two equations are; dx by dt is equal to y times ds by dt utilized. These equations are mirror images. Suppose if I want to plot this equation I can plot it like the following.



(Refer Slide Time: 29:40)

I have what is called this is the time, this is X and this is S, also the axis can represent both S microbial concentration and substrate concentration so both are expressed in terms of milligrams per liter, this also is expressed and this also is expressed in terms of milligrams per liter, then at time t = 0 what would be the concentration of S, concentration of S would be S_o let us take the maximum concentration so S_o is the concentration of substrate. As a function of time with respect to this equation dx by dt, again I will write it here as dx by dt is equal to minus y time ds by dt utilized, this dx by dt is equal to mu max S over k_s plus S into X (Refer Slide Time: 29:48). So this is the equation; dx by dt is equal to this or minus y into ds by dt is equal to this. That means at time t = 0 the substrate concentration is S.

(Refer Slide Time: 30:20)



I try to solve this equation and the solution of this equation results in this particular graph like this (Refer Slide Time: 30:10) so this is the variation of concentration of substrate with respect to time, how exactly the concentration of substrate is varying with time, so this curve is for S.

(Refer Slide Time: 31:45)



Now I can also use this equation to find out the variation of X with respect to time. So at time t = 0 the concentration of X equal to X_0 so this is X_0 to start with and as a function of time what is happening is the microorganisms are utilized in the food and they increase in number so that particular number increase can be written like this (Refer Slide Time: 30:56) so this is the equation for X and this is equation for S. You can see the difference

between these two things; difference is not attaining the same level, X is not attaining the same level of S_o the reason for that is because of the yield coefficient yield constant. So the entire substrate is not going into the formation of biomass. This is because of the yield coefficient difference. The difference in yield coefficient is this, taking place here. This is the curve I can represent in this particular equation. They are mirror images. That is dx by dt growth rate and then substrate utilization are mirror images.

Now let us consider one more factor. We have said that y is equal to X minus X_0 over S_0 minus S this is what we have said. Yield coefficient is the weight of biomass produced per weight of substrate utilized in a given amount of time; the same time is what is going to happen. Now this particular thing is y into S_0 minus S is equal to X minus X_0 .

Now let me locate a condition when the entire substrate is utilized entire S is used by microorganisms. That means the microorganisms have consumed the entire food, the food has been completely consumed so in that case what would be the value of S is it will be equal to zero. S is completely consumed that means S is zero. When S is zero that is when the entire food has been consumed then what is the concentration of microorganisms produced at the time that X will take a value of Xmax.

(Refer Slide Time: 33:55)

That means maximum concentration of microorganisms is produced when the entire food is utilized by the microorganisms. So in this particular equation if you substitute Xmax is equal to, and this (Refer Slide Time: 33:28) I will take it this side X_0 plus S is zero y times S_0 . So the maximum concentration of microorganisms is produced as per this equation. The entire substrate has been used. So this is the equation what we can suggest for this particular thing for the maximum growth of microorganisms.

I have been considering till now the bacterial growth rate that is dx by dt. Now I would like to consider another aspect that is substrate utilization rate. According to the equation

the substrate utilization rate ds by dt utilized is equal to 1 by y into mu max S over k_s plus S into X this equation I can write it here. So this is substrate utilization rate is equal to this mu max S over k_s plus S X so this is one equation I can write, the substrate utilization from the dx by dt type of equation where dx by dt is the growth rate of microorganisms.

Now what I can do is I can also write down, suppose this is one equation what we have this is the equation (A) for the utilization of substrate. Now I can also write similar thing like this. In a unit time, suppose if delta S is the substrate that is used in a unit time the rate of substrate utilization is proportional to the X delta t. What is utilizing the substrate? The microorganisms are utilizing the substrate and this is the biomass concentration. The biomass is utilizing the substrate and hence there is a change in substrate. Change substrate is because of the utilization of the substrate by biomass so that is what is proportional to X into dt a small change, elemental change.



(Refer Slide Time: 36:46)

The ds by dt if I write it here put limit delta t tending to zero this will be equal to ds by dt proportional to X or this proportionality I change it by ds by dt is equal to the term called q I will put it as q into X. Now I define what q is. Here q can be defined that means ds by dt the food that is utilized is equal to q into X. The q is known as specific substrate utilization rate. The units of this will be S and X are in the same units that is what is milligrams per liter as what we said so both of them are expressed in the same units, when we are expressing in the same unit then q will take the unit in inverse of time so this is inverse of time the time inverse q is nothing but ds by dt utilized over X so this is what is q value. The specific substrate utilization rate is this particular thing.

(Refer Slide Time: 38:00)



Now we have an expression here. if I plot this graph there is similarity between this expression and another expression called mu, mu is the specific growth rate which is nothing but dx by dt the growth of microorganisms may be I can write it as growth or g simply, I can write it simply g or growth I am writing it here divided by X. So this is specific growth rate and this is specific substrate utilization rate. I do not have to write it here it is already there. Now I would like to know the relationship between the specific substrate utilization rate and specific growth rate. Can you tell me what will be the relationship?

The relationship can be easily derived. The specific substrate utilization rate and specific growth can easily be related. How we can relate them? We can relate them using the yield constant or yield coefficient rather. So the yield coefficient is given by y, y is equal to dx by dt over X divided by ds by dt utilized. Divide both sides both the numerator and denominator by X that is what I am trying to do here. So it is dx by dt over X ds by dt utilized over X. When I do so what happens is, y is equal to, this dx by dt over X is nothing but dx by dt here in this particular equation, you can say that is equal to mu and this particular thing dx by dt utilized over X is equal to q so that is q. That means this is the relationship between specific growth rate and specific substrate utilization rate and the yield coefficient.



So in other words yield coefficient is the ratio of specific growth rate divided by specific substrate utilization rate. That is what we can see. This is one relationship which is important, we will be using it quite often.

Now let us take the equation that is specific substrate utilization rate. It is; q is equal to ds by dt utilized over X is the q value. In other words ds by dt utilized is equal to q into X. Now this q specific substrate utilization rate is not constant just like mu, mu is not a constant so q is also not a constant. Again, q is a function of substrate so this functionality is given by q_{max} S over k_s plus S, $q = q_{max}$ multiplied by S over k_s plus S.

What is q_{max}?

The q_{max} is just like mu max. There is a similarity between q_{max} and mu max. The q_{max} is my maximum substrate utilization rate. It is similar to mu max which we have discussed already. So there is a similarity between these two things. Now let us take condition one again. When the substrate is unlimited similar to the condition which we consider previously in which I have got unlimited substrate, so when you have unlimited substrate what is going to happen to this equation again is, S is greater than k_s and S plus k_s can approximated to S as previously done and hence substituting in this equation may be I call this equation as b so substitute in the equation b for this k_s plus S then this equation will be converted as $q = q_{max}$ into S by S into X so this particular thing cancels out. Therefore, $q = q_{max}$ times X. That is what we have that is $q = q_{max}$ into X so this q (Refer Slide Time: 44:28). Here there is a mistake, $q = q_{max}$ S over S that is in this particular equation I am substituting so this k_s plus S is S and $q = q_{max}$.

(Refer Slide Time: 44:40)

The q takes the value of q_{max} . That means the specific growth rate specific substrate utilization is the maximum value so this particular thing I will substitute in the fundamental equation. The fundamental equation is the following that is my fundamental equation is ds by dt utilized is equal to q into X this is the fundamental equation.

 $q = \left(\frac{ds}{dt}\right)_{a}$ $q = \left(\frac{ds}{dt}\right)_{a}$ $\left(\frac{ds}{dt}\right)_{a} = \left(\frac{q}{q}\right) \times$ (s not const) q = F(S) q = F(S) $q = \frac{q \text{ max} \cdot S}{k_{s} + S} \rightarrow (B)$

(Refer Slide Time: 45:20)

In this now I substitute q_{max} into X ds by dt utilized. This particular thing I will substitute in the particular q_{max} into X. So now this equation can be solved. This particular thing again I will take, if take it to the bottom then it is $q = q_{max}$, this is one equation what I am going to get. This is the equation what we have ds by dt utilized is equal to q_{max} multiplied by X. In other words if you say ds by dt if you asy dx by dt utilized over X this is nothing but for me $q = q_{max}$. So in this equation q equal to q_{max} this also I can write S to the power of zero, a zero order equation with respect to substrate. So this is the zero order equation with respect to substrate concentration.

Therefore, if I make a plot of this particular thing I make a particular equation it is something like this. That is, I plot S here substrate concentration and I plot q here q is nothing but ds by dt utilized divided by X the specific substrate utilization rate this is ds by dt utilized over X. So if take this when S is unlimited when the substrate concentration is very high as we have taken a case when the substrate concentration is very high case number one then this is independent of substrate concentration, this is zero order equation, a line like this will come (Refer Slide Time: 47:35) when the substrate concentration is very high. So this is my q = q_{max} and this is the zero order with respect to substrate concentration. That is the curve we get here.

Now this is one condition always that is food is available in plenty, plenty of food is available. Now when the food becomes limited that is case number two I will take it here, case number two is when S is limited, limited food is available and again I would like to reiterate the fact that most of the times domestic wastewater treatment we come across this particular thing when the food is limited. When the food is limited now we can say that the equation whatever we have is ds by dt utilized is equal to q_{max} S over k_s plus S multiplied by X. In this particular equation what I will do is, concentration of S is less than k_s and when it is less than k_s I have I can say k_s plus S is equal to q_{max} over k_s k_s plus S is also k_s so q_{max} ks multiplied by S into X that is the equation we will get.

So q_{max} is the constant as we know, k_s is a constant, this whole thing is a constant q_{max} by k_s is a constant and this particular bracketed thing is a constant (Refer Slide Time: 49:55) so this constant may be called as k, I can call it as k. So now, again taking that equation we have ds by dt utilized divided by X then immediately this becomes q that is the specific substrate utilization that will be equal to now K times that is the q_{max} over k_s is K multiplied by S. So please look at this equation and the previous equation. This is one equation which you have looked at; this equation is the first order equation with respect to substrate concentration.

(Refer Slide Time: 50:51)



So, if I plot a graph this q that is in this plot will go, S versus q if I plot it this is the first order equation that is $q = q_{max}$ over k_s into S so this is S to the power of 1 so this is the first order equation (Refer Slide Time: 51:25). Now when the substrate is limited it depends on the concentration of the substrate that is substrate utilization depends on the concentration of substrate and when the substrate is unlimited the utilization of substrate is independent of the concentration of substrate.

This is the zero order, this is the first order. So in between this first order should become a zero order or zero order a first order so this particular thing is discontinuous that means there is some other order here between these two things or connecting these two things and hence this model is known as the discontinuous model for substrate utilization. Why it is called discontinuous model? It is zero order when the substrate is available in plenty and it is a first order model when the substrate is limited and in between the situation it is a discontinuous model that is what it is said.



That means we have got two equations now. One is the specific growth rate, another is the specific substrate utilization rate and two cases were considered in both cases. One is when the substrate is limited and the other is when the substrate is unlimited and these two things are connected by what is called the yield coefficient. Yield coefficient is very very important and we have seen that particular thing.

One more point I would like to discuss before going further is, suppose if you give a substrate to the microorganism, the microorganisms could be aerobic or anaerobic and depending upon that the value of y will be different. So let us take into account that a part of this is used for energy and another part is used for the biosynthesis. If you see further this amount of energy that is produced is used for two purposes. One is what is called energy required for growth of microorganism. growth of microorganisms increase the weight of the biomass, energy required for the growth of microbes is one thing and the second thing is, energy is also required not only for growing but energy is also required for maintenance so it is the energy required for maintenance. Maintenance means the cell has to do certain amount of work, it has to move from one place to another place, we are using the flagellum it has to do certain other works so this is called energy required for the maintenance. This energy is called maintenance energy and this energy is called growth energy, energy required for growth. (G. E) is the growth energy (M. E) is the maintenance energy.



When I give a substrate to the microorganism the microorganism utilizes a part of the substrate for energy that energy goes for the growth of microorganism as well as growth of maintenance energy. Now, the situation is the following: if I do not give any substrate to the microorganism but if I give the substrate they will utilize in this fashion. Now what would happen is that there is a condition already we have shown that when the substrate is completely exhausted when S is completely exhausted when there is no food available to the microorganisms what would the microorganisms do?

Definitely you can say that the microorganisms do not grow, grow in number, they will not grow in large size, no growth takes place, growth ceases, cessation of growth, growth disappears however, there is necessity for the maintenance energy. Maintenance energy is to be met even if there is no substrate. Because the cell has to do certain amount of work, it is not growing in size because there is no food but it has to be alive, for alive it has to do certain work so that maintenance under this is required. Hence, this maintenance energy is required for what is called a basal metabolism, to maintain basal metabolism. Basal metabolism is the minimum metabolism that is required for the cells. So what I will do in the next class is that we would see how this maintenance energy is met when the substrate is not available for the microorganisms.