

Water Quality Management Practices

Dr. Gourav Dhar Bhowmick

Agricultural and Food Engineering Department

Indian Institute of Technology Kharagpur

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Bacterial Metabolism and Their Use in Wastewater

Hello everyone, welcome to this NPTEL online certification course on Water Quality Management Practices. My name is Gourav, Professor Gourav Dhar Bhowmick from the Department of Agriculture and Food Engineering of Indian Institute of Technology Kharagpur. So, in this particular module the fundamentals and principle of biological wastewater treatment will be discussed with the first lecture about the bacterial metabolism and their use in wastewater. The concept that we will be covering are the bacterial metabolism in wastewater treatment, the catabolism and anabolism process, the nutritional requirement for microbial growth, what are the type of microbial metabolism, the bacterial growth kinetics in bacterial growth kinetics in batch culture, bacterial growth kinetics in continuous feed culture and the principles of biological wastewater treatment. So, to start with we know the bacterial metabolism it is a chemical process that occur simultaneously within each bacterial cell to provide energy for the growth and it is can be convert it can be bifurcated in two types like the catabolism and the anabolism. Catabolism is what? It is the energy yielding metabolism of the cell and the anabolism is the energy consuming biosynthetic mechanism. It is more like when you have suppose in this reaction it is a it is a aerobic catabolism process. That means, here what is happening here the glucose is like we remove some kind of organic contaminants say suppose glucose is a contaminant here just for the sake of understanding. So, we remove it through the wastewater is using this catabolic metabolism process where the microbes catabolize some kind of organic pollutants. So, in this particular reactions the glucose is getting catabolized.

So, this catabolism again can be of two types aerobic and the anaerobic. Aerobic one where oxygen nitrate or the sulfate oxidize the organic pollutant and that degrade and the in case of anaerobic or the fermentative catabolism the degradation via hydrolysis, acetogenesis, acetogenesis and the methanogenesis happen within this four distinct process. So, it is it is sounds much easier, but actually this degradation process it is takes huge very complicated pathway, but in order to make it easier to make it more much more simpler for you. So, we generally we divide it into four different groups hydrolysis, acetogenesis, acetogenesis and methanogenesis good. So, in general when we talk about the hydrolysis it is it is the first step in anaerobic catabolism what is happening here we like much more larger molecules like protein, carbohydrate or lipid or the fat are converted into some monomeric or dimeric compounds by the action of exo enzymes.

So, say the protein it converted into hydrolyzed into amino acid, the polysaccharide or say like carbohydrate it converted into simple sugar solutions, lipid or the fat components it converted into long chain fatty acids. So, this is the very first process what is happening here? Here we are converting this different higher chain large molecules into the simpler like much more simpler compounds. So, this rate of hydrolysis it depends on various factors such as temperature, pH, particle size and the enzyme concentration. So, in the next step it is called

the acidogen and acetogenesis. In this acidogenesis the hydrolysis product is diffused inside the acidogenic bacterial cell and fermented into short chain fatty acids alcohol, carbon dioxide and H₂ as well as other intermediate organic products also like propionic acid and butyric acid.

So, it is like more rapid process this reaction this acidogenesis and at the end product of acidogenesis are majorly the substrate for acidogenesis and the methanogenesis. So, the first is hydrolysis major this complex large molecule convert into some monomeric or dimeric compounds much more simpler ones. Then those simpler hydrolyzed hydrolyzed product now converted into this short chain fatty acid, alcohol, carbon dioxide and the H₂. Now what is happening in the acetogenesis then? In this process is non gaseous product say whatever it is remain say like propionate butyric and all. So, they are the main substrate and then converted into they normally further oxidized to acetate, carbon dioxide and H₂.

So, the final product here after the acidogenesis is what acetate, carbon dioxide and H₂. After the third process step is done we are left with only acetate, carbon dioxide and H₂. Now one of the one of the major step in any anaerobic reaction is the methanogenesis. What is happening in the methanogenesis? In the methanogenesis this acetate, carbon dioxide and H₂ which is the product of the acetogenesis it converts to CH₄ methane. So, this process the the microorganisms which are involved in it they are called the methanogens. This methanogens they this methanogens they normally convert this all this acetate, carbon dioxide and H₂ into CH₄ the methane. So, there are two types of methanogens acidoclastic and the hydrogenotropic. In case of acetoclastic the acetate utilizing bacteria's are normally involved actually they they are different kind of archae also which are involved in general methanogens are archae in the category of archae if we said correctly. So, it converts the acetate into methane and in case of hydrogenotropic methanogens methanogens they utilize the carbon dioxide and hydrogen found in the acidogenic process to methane. So, our final product here is what is the methane. In general 70 percent of the methane that is produced in the acidoclastic methanogenesis process are because of the acetate utilizing bacteria. And the 30 percent of them are mainly hydrogenotropic methanogens and they are because of the hydrogen utilizing bacteria's are known. This growth rate of this acetate utilizing bacteria are very low and because of this reason most of the anaerobic reactor takes a longer start up time. So, that is one of the major drawback of anaerobic system as well we will discuss about it. The aerobic what is the benefit of aerobic and anaerobic treatment system.

So, anaerobic treatment system treatment system that is one of the major problem that it takes a longer start up time. What is start up time means? Suppose you want to start your reactor it needs couple of couple of days of time to just to acclimatize to the system and also to have enough amount of microorganisms to finish the whole procedure to have this all four steps ready then only it started acting perfectly. So, this inoculum preparation and after then this acclimatization takes a lot of time in case of anaerobic digesters anaerobic reaction processes are known. However, it is the higher sludge concentration and like in general it takes longer start up time and higher sludge concentration, but it normally in its quite stable. However, this sludge concentration actually it is much lesser than the aerobic ones aerobic ones generate more sludge in general. We will discuss about this things with time. So, in general the catabolism and anabolism we normally whatever the available organic or inorganic substrate present in the wastewater it is normally oxidized by the biochemical reactions and it catalyzed by the enzymes produced by macroisms to liberate the energy. Under aerobic conditions oxygen acts as a final oxygen acceptor electron acceptor in case of anoxic conditions sulfate, nitrate, nitrites, carbon dioxide organic compounds act as a electron acceptor. The end

product of this is carbon dioxide, water, ammonia and H₂O water it is please correct it it is only carbon dioxide, water and ammonia. The heterotrophic microorganisms which derive their energy required for the cell synthesis from the oxidation of this organic matter while autotrophic ones from where they derive they derive this energy from synthesis of the oxidation of inorganic compounds or from the photosynthesis. Just remember there is a very basic difference between heterotrophic and autotrophic. Heterotrophic they get their energy from oxidation of organic matter oxidation of organic matter. In case of autotrophic they get it from oxidation of inorganic matter or by the process by the process of photosynthesis. This is the basic difference between autotrophic and the heterotrophic. So, in case of adverse situation microorganisms start oxidizing their own protoplasm and it is called the endogenous respiration or endogenous decay process.

So, that only happens when there is less amount of food available for your microorganism to survive. So, in that case what they do they started decaying their own protoplasm. So, this is called the endogenous decay process. In general the yield of energy is in aerobic in aerobic process is much higher than in anaerobic conditions. Thus the aerobic systems produces more new cells than the anaerobic systems, but at the same times it creates a lot of nuisance also because it creates a lot of new cell it creates a lot of sludge.

So, we call it activated sludge and in general they are called activated sludge. So, this sludge is actually causing a huge problem in aerobic treatment systems. So, what we understood about the catabolism and anabolism like when organic matter is used as an energy source it is transformed into a stable end product it is called the catabolism product process. In case of anabolism what is happening the organic matter is transformed and incorporated into the cell mass. This is called the anabolism process.

They are obviously, interdependent and it occur normally simultaneously. So, what are the nutritional requirement if you talk about the nutritional requirement of microbial growth. So, what they need in general for proper functioning? First a source of energy definitely, second thing a carbon for synthesizing new cellular material and third is nutrient like nitrogen, phosphorus, potassium, sulfur, iron, calcium, magnesium etcetera. The energy required for the cell synthesis may be supplied by light or by a chemical oxidation process reaction catalyzed by the bacteria. So, the sources of energy wise if we differ into two types phototrophs or chemotrophs.

Phototrophs means it uses light as an energy source. So, there can be heterotrophic like there can be phototrophs they may be heterotrophic in nature like sulfate a sulfur reducing bacteria or autotrophic also like most of the photosynthetic bacteria and algae. However, chemotrophs are what who derive their energy from the chemical reactions. They also can be heterotroph like normally they derive energy from protozoa, fungi, bacteria etcetera and also they can be autotrophs like nitrophyne bacteria and all. This nitrophyne bacteria are autotroph in nature that means, they are they uses some kind of chemical to like to get their energy derive their energy.

However, they are autotrophic in nature that means, those chemicals are inorganic chemicals and all that is why they are autotrophic in nature. When they use the phototrophs you see in that case they use the light. So, photosynthesis process photosynthetic bacteria and algae. So, this is the difference between phototrophs and chemotrophs and the heterotrophs and autotrophs. It is very important for us to understand this in a basic nomenclatures.

It will be easier for us to discuss further if we know this basic idea about the bacterial metabolism, the type of bacteria, type of microorganisms, what are the sources of energy that they use and all these things. So, second important requirements is the sources of carbon. Why we need sources of carbon? Because it is necessary for supporting the synthesis of new cell. Our body in majorly the all the microbes and all their cell is majorly comprises of the carbon. We are carbon based living beings.

In general, the earth has a carbon based living beings in general. So, this carbon is very important. So, this carbon sources of carbon can be organic matter as used by the heterotrophs and also can be the carbon dioxide as used by some kind of some type of autotrophs and all. The third is the nutrient or the growth factor requirement. So, not only the sources of energy or the carbon, but another important thing that is we have to make sure that is available in our treatment unit so that the bacteria can grow properly is called is the nutrient different nutrient or growth factor requirements.

So, majorly the primary inorganic nutrients like nitrogen, sulfur, potassium, phosphorus, phosphorus, potassium, magnesium, calcium, iron, sodium, chlorine so, etcetera and nutrient required for the trace amount is like are majorly the zinc, manganese, molybdenum, selenium, cobalt, nickel, copper etcetera. So, they are normally certain mechanisms requires some organic nutrients, they are known as the growth factors also. So, first thing is nutrient requirement. Nutrient requirement can be of principle once as discussed in the first line you see the when we discuss about nitrogen, sulfur, phosphorus, potassium, etcetera and some interest amount also is required for their bodily functions and all, cellular function I would say in this case. And there are some growth factors which are also responsible which nothing, but the organic nutrients it can be it is nothing, but the COD can be easily demonstrated by COD nitrogen phosphorus ratio.

In case of aerobic process the ratio can be 100 like 100 is to 10 is to 1 to 5 COD nitrogen to phosphorus. In case of anaerobic it can be the carbon COD requirement is very high it is can be 350 is to 5 is to 1. So, in case of anaerobic process as a standard ratio that we have to that we normally try to maintain in anaerobic process. So, these are the different type of factors that is required for reproduction and the proper functioning of the microorganisms. And this nutrition nutrient requirement is lower for anaerobic processes due to the slower growth rate of microorganisms than for the aerobic processes.

And the industrial wastewater treatment processes where we add the micro macro like sodium, phosphorus, potassium, sulfur and micro trace you metals nutrients may be required based on the raw wastewater characteristics. So, sometimes when you introduce in the in the secondary treatment process when you have suppose the biological treatment units. In this biological treatment units before that you have to check the wastewater characteristics, raw wastewater which is coming into the biological treatment plant. If it does not have this parameters this nutrition requirement then what we need to do we need to artificially supplement it with this macronutrients and the micronutrients. What are the types of microbial metabolism? First is the aerobic one we all know that is where we molecular oxygen is used for the terminal receptor in respiratory metabolism and the organisms that exist only in presence of molecular oxygen are for the obligately aerobic microorganisms.

Then there come the anoxic, anoxic means the ano microorganisms oxidize the inorganic compounds like sulfate, nitrate, nitrite, but which work as a neutron acceptor in the absence of molecular oxygen. In case of obligately anaerobic means this microorganisms generate energy

by fermentation and can exist and metabolize in the absence of oxygen. In case of facultative anaerobic anaerobes, in case of facultative anaerobes they are these organisms can grow in presence or in absence of oxygen. So, in based on the availability of the oxygen and their characteristics also it can be divided into two types. First one is true facultative anaerobes, true facultative anaerobes this can shift from fermentative to aerobic respiratory metabolism depending on the availability of oxygen and another one is called the aero tolerant ones.

Aero tolerant ones this microorganisms actually follow strictly the fermentative metabolism and are insensitive to the oxygen that does not matter whether oxygen is present or not. They will anyway follow the fermentative metabolism that is why it is called the aero tolerant anaerobes. So, we know first the basic four types of microbial metabolism first one is aerobic, anoxic, obligately anaerobic, facultative anaerobic. In case of facultative anaerobic also can be divided into two types true facultative anaerobes and the aero tolerant anaerobes. So, these are the different types of microbial metabolism those are available.

So, it is very important for you to understand this basic differences. So, let us start discussing about the bacterial growth kinetics, because we need to understand this bacterial growth kinetics in order to understand the functionality of the biological treatment processes. So, to start with the we have to understand the relationship between the specific growth rate of bacteria and the substrate concentration consumed by bacteria and their cellular metabolism. It also predicts the substrate conversion rate into biomass as a product. So, in general this bacterial population growth it follows a geometrical or exponential pattern in which one cell splits into two daughter cells into four, four into eight and so on so forth.

So, after n number of division what will be the number of cell? The number of cell will be 2 to the power n. That means, like after say like 3 division. So, the number of cell that is available from a single one it will from one cell it will become 8 number of cell after 3 division 2 to the power 3. So, this is how it works. So, once the number of divisions you can easily find out the tentative amount of microorganisms present cell living cell present in your system.

So, in this binary fission how it happens? This DNA replication happens from a single cells, chromosomal segregation happens, then the cytokinesis happen we all know about it. So, the time required for this bacterial cell to grow and split into two virtually identical sister cell is known as the generation time or the doubling time. So, this all the whole process to finish this binary fission process it this time that it consumes it is called the doubling time or the generation time. So, please remember this terms this doubling time it is it will be very useful to you and in when we if you see this plot here the cell number versus time if we plot this bacterial growth curve see it follows the certain exponential pattern. Even if we do the logarithmic graph it will follow this straight line.

So, bacterial growth kinetics if we if we discussed on the basis of the cell number the relationship between the number of bacteria that is N_t if we denote in a cultural medium at a given time t is the number of bacteria present at the beginning which can be denoted as N_0 and the number of division occur during this period isn't it. So, so what does that mean? That means, that the amount of bacteria at a certain moment of time and in a culture medium obviously, it is equal to the N_0 that the bacteria present in the beginning multiplied by 2 to the power N isn't it. So, those number of bacteria only now multiplied into N number of division during this during this time. So, once we know the constant division rate like the for a constant division rate say like value of k N can be calculated by k into t. So, what is this k? k is nothing, but the it is it can be it is also like nothing, but the inverse of the doubling time

or the generation time .

So, this doubling time or the generation time g inverse of this g is called as constant division rate or the value k . So, now, if you replace this in this first equation

$N_t = N_0 \times 2^{k \times t} = N_0 \times 2^{1/g \times t}$ instead of N we put k into t instead of k you can also put 1 by g . So, now, the equation becomes N_t equal to N_0 into 2 to the power t by g . So, this g is nothing, but the doubling time or the generation time. Now, we can easily find out the value of k from there if it is unknown to you k is nothing, but 1 by g and 1 by g is nothing, but here if you just simply log from the both side suppose to make it easier in this after this therefore, statement N_t equal to N_0 into 2 to the power kt right.

So, put the N_0 in the left side or say like you simply do the logarithm of logarithm of N_t by N_0 in the left side and then the right side logarithm of 2 to the power kt . So, if you do the 2 to the logarithm of N_t by N_0 is nothing, logarithm of N_t minus N_0 N_t minus $\ln N_0$ logarithm of N_t minus logarithm of N_0 in the left side and 2 to the power kt logarithm of 2 to the power kt kt is a constant it will come in the front and it will become kt into logarithm of 2 . So, now, k if you want to find its k is equal to logarithm of N_t minus N_0 divided by t into $\ln 2$ you understood. Let us make it let us do it let us try to do it again try to write it down when I am saying this things N_t is equal to N_0 into 2 to the power kt . Now N_0 keep it on the left side place it in the left side it will become N_t by N_0 equal to 2 to the power kt .

So, this now do the logarithm in the both the side logarithm of N_t by N_0 equal to logarithm of 2 to the power kt . Now logarithm of N_t by N_0 means it can be written as logarithm of N_t minus logarithm of N_0 the basic logarithm function right. So, now, in the right side logarithm of 2 to the power kt kt is constant. So, kt will come in front.

So, it will become kt logarithm of 2 . Now kt this to find the k , $k = 1/g = \frac{\ln N_t - \ln N_0}{t \times \ln 2}$. So, from there you can easily find out the value of constant division rate for any unknown sample. Now based on the biomass if we now discuss about the growth kinetics based on the biomass the rate of increase in bacterial biomass in a culture media it is normally correlated with the specific growth rate represented by μ and the biomass concentration x at a certain point of time. Biomass concentration means it is a it is nothing, but the MLVS or the VSS volatile suspended solid that actually represents the amount of biomass present in your system. If you remember we discussed about it right volatile suspended solid it represents the x or the amount of biomass.

So, this $\frac{dx}{dt} = \mu \times X$, dx by dt is what it is a increase in bacterial biomass it is a growth rate in the specific it is a rate of increase in the bacterial biomass. Now dx by now if you from the right side x you put it in the left side. So, it will become dx by a capital X equal to μ into dt . So, now, dx by capital X if you do the integration now the both the side it will become integration of dx by x equal to integration of μ into dt . So, μ will come in the in the front and it will become integration of dt only.

So, let us say like integration of 0 to t from both the side. So, it will become $\ln x_t$ minus $\ln x_0$ equal to μ into t . So, now, $\ln x_t$ minus $\ln x_0$. So, if you put the x_0 in the right side it will become $\ln x_t$ equal to $\ln x_0$ plus μ into t . Now $\mu = \frac{\ln X_t - \ln X_0}{t}$ or x_t equal to x_0 into e to the power μt that also you can write it easily simply this simply if you anti logarithm like you know just get rid of the logarithm function from both the side.

So, this $X_t = X_0 \times e^{\mu t}$ is the very important functionality that we need to remember and this actually showcases the any at a certain moment of time what will be the bacterial concentration if the initial ones initial bacterial concentration at and the time and you also know the specific growth rate you can easily find out the amount of bacterial biomass concentration at any moment of time t in your chamber in your reactor. Now, in general the bacterial growth kinetics in the batch culture if we discuss it follows this nice curve if you see in the right side to start with it showcases the lag phase. So, what is a lag phase? Lag phase is nothing, but when a bacterial cell is introduced to a new environment it requires some time to acclimatize it. So, what happened this bacterial cell it is try to adapt by increasing a cell mass only and there is no net increase in the cell number in this phase. And multiple lag phases are observed in when among bacterial species when the culture medium adapts contains more than one carbon source this phenomena is called the diauxic growth and all.

So, in case of diauxic growth you will not see much of a increment in the number of microorganisms, but there is a mass of microorganisms will increase a tiny tiny bit. If you see the lag phase there is a tiny bit of increment in the mass of the microorganisms, but not in the number. In case of continuously operated reactor this bacterial cell will get acclimatized to the environment and over time and that is why in case of continuously operated reactors this lag phase does not exist. So, continuously so, we know that it already acclimatized to the system. So, now, it does not require repetitive adaptation period and all.

The next one is the also we need to understand this majorly the bacteria will first consume the easily metabolized carbon source and therefore, when the carbon source is exhausted the metabolic pathway shift to utilize the other carbon sources and all. But anyway so, in general this lag phase we call it also acclimatization phase where we normally wait for some time to start our reactor in case of batch culture. You remember batch culture right it is batch culture, continuous culture, plug for culture we already discussed. So, in case of batch culture the first phase is called the lag phase then there comes the log phase. What is log phase? Where the amount of food is enough your bacteria is already acclimatized already habituated with the environment they will start growing exponentially.

It is called the exponential growth phase that is why. So, in case of exponential growth phase it is the second phase of the bacterial growth and bacterial growth phase and also it is the highest bacterial growth rate with you can see here. What is happening? It is the cell number will increase exponentially with time because of the availability of the food is enough, the substrate is enough. So, that is why they are they are reproducing in a faster rate and it always follows the first order rate and the and the natural log of cell concentration versus time gives the straight line if you check it, but if you do the logarithmic graph plot the logarithmic graph, but in this case you see there is a sharp exponential growth in the log phase. Then there comes a declining growth phase if you see what is happening in case of declining growth phase we started having the substrate availability started reducing a bit. So, because of the reduction in the substrate availability what is happening? There is no ample amount of substrate available for those bacteria to survive they are growing anyway, but it is not enough.

So, they reach to that certain declination in the growth phase they are not reproducing in a rate that it used to in the log growth phase right because of the availability of substrate is reducing. At certain stage it reaches the stationary phase what do you mean by the stationary phase means the growth rate of bacteria is equal to the death rate of bacteria. So, at the end what is happening $\frac{dx}{dt}$ is 0 that means, the growth in this phase can be expressed as 0

right because there is no increment or decrement because of their growth rate and death rate is almost the same. It only happens because of the depletion of essential nutrient in the media and also the due to the accumulation of toxic byproducts in case there is in the media and in though the bacterial net growth is 0 in this stationary phase the bacterial cell remain still active . They are prone to production of secondary metabolites like antibiotics and other value added products in this stationary phase only .

So, this is very important reaction for the researcher who works on antibiotic and the value added product recovery from the microorganisms and all . Then there comes the death phase or the endogenous decay phase or the endogenous growth phase . So, this is the final phase of bacterial growth in a batch culture where the bacterial growth rate becomes greater than the death rate becomes greater than the growth rate. So, because of that there is a decline in the mass of organisms value because of that you see the declination in the right side of the figure. So, after the stationary phase when the nutrition is limited the bacteria started oxidizing their own cellular mass to survive in that environment.

So, that is why it is called endogenous decay phase or endogenous growth phase . So, this what happened the bacterial cell they started dying and they break down to spill their content into the medium environment making this nutrient available for survival of the all the remaining bacteria . So, this can be expressed by this equation $\frac{dx}{dt}$ equal to minus k_d into x with this k_d is a specific decay constant or also known as the endogenous decay constant. And with this equation it can be represented this growth of the in this phase $\frac{dx}{dt}$ equal to minus k_d into x good. So, now let us discuss about what is happening in case of continuous feed culture.

We discuss about the batch feed culture we understand that what batch culture what is happening. Now, let us try to find out what is happening in the continuous feed culture. In case of continuous feed culture is the most applicable mode of operation for higher biomass productivity for of bacteria. The bacterial cell and the substrate concentration and the product concentration remain almost constant in this method and fresh nutrient medium is constantly supplied to this reactor and the products are withdrawn continuously from the reactor that is why it is called the continuous culture.

This continuous culture can be two types chemostat or the turbidostats. Chemostat means in the nutrient supply rate limits the bacterial growth rate and the flow rate of the culture media. In case of turbidostat what is happening the cell concentration in the reactor is kept constant by varying the flow rate. So, this is the two different types of continuous culture that we normally follow. So, in case of continuous feed cultures when the single limiting substrate the substrate growth rate is normally the function of the limiting substrate concentration S and the maximum specific growth rate or μ_m and the substrate specific constant K_s . So, this μ can be represented this is this equation called the Monod equation named after the scientist Monod.

So, it is a very famous equation in water treatment technology it is called the in specially for the bacterial bacteriological treatment processes. So, $\mu = \frac{\mu_m \times S}{K_s + S}$ where μ_m is the maximum specific growth rate S is the substrate concentration limiting substrate concentration and the K_s is the substrate specific constant. So, in general the bacterial growth rate in continuous culture depends on both the volumetric flow rate of the nutrient supply into the reactor and also the dilution rate. What is dilution rate is nothing, but capital F by capital V .

Capital F means the flow rate of the nutrient supply and V is the reactor volume. From there you can easily find out the dilution rate. So, net change in the cell concentration in the reactor over a time period by if you denote in dx by dt x means the cell the cell biomass right. So, dx by dt represents the net change in the cell concentration. It obviously, equal to the rate of growth in the reactor minus whatever the washout due to the dilution. So, this rate of growth of reactor we already know it is a μ into x and in case of steady state condition this μ is equal to d and this washout due to the dilution will be can be represented by capital D into x and in case of steady state condition this dx by dt will become 0.

So, in case of higher dilution rate the microbial biomass concentration reduces to 0 and which is which is known as the cell washout condition and this dilution point occurs in the critical dilution rate d_c . This d_c can be represented as μ_m into S_r . This S_r is a residual substrate concentration under the steady state condition at a free dilution rate. Now, another important nomenclature I would say like you need to understand it is called the food to microorganism ratio. From the name itself you can understand what is food here is a substrate or say like when we talk about in wastewater treatments research we talk majorly with the with BOD or COD.

This is the food that you are representing it is can be indexed by the BOD or COD. So, this amount of organic substances that is come into the picture divided by the amount of microorganism present there that ratio is called the food to microorganisms. So, how we can find out? Suppose you have a 1 litre of reactor. In this 1 litre of reactor you have certain say like 1 litre per hour of flow rate with the flow rate there is a new wastewater the influent is coming. So, that means, that in the flow rate is the flow rate Q value is 1 litre per hour and it has suppose a BOD of 100 milligram per litre.

So, what is the total food coming? 100 milligram per litre multiplied by 1 litre per hour. That is the total amount of food that is come into the picture. So, divided by what is the microorganisms? Amount of microorganisms present there. So, my amount of microorganism present there is nothing, but the VSS the volatile suspended solid in milligram per litre or gram per meter cube multiplied by the volume of the reactor say 1 litre.

This value represents the microorganism number of microorganisms. So, this ratio this Q into BOD or COD divided by volume into X the VSS represents the food to microorganism ratio. So, this food to microorganism ratio is normally when it is low that means, the food availability is less. When the food availability is less obviously, bacteria will exhibit what happen? They will start consuming their own protoplasm. So, it exhibits the endogenous growth of microorganisms.

In case of higher F by M ratio the food available is abundant. So, they start growing the low growth phase that means, they will reproduce in a much faster rate. Now, if you see this graph if this graph represents the continuous fed reactors. The rate of metabolism that happens in the continuous fed reactors at a different F by M ratio. In this particular reactor in this particular reactor if you see in the X axis in the or the abscissa we have food to microorganism ratio F by M. The lower represents like the less F by M the more you go to the right side it is it will increase.

In the Y axis or the coordinate axis we have rate of microbial metabolism so that means, the more you go upwards it means the rate is increasing. Now, if you see this graph this graph

represents at a certain moment of time if your reactor has a less F by M ratio it represents the endogenous growth phase that means, your reactor your microorganisms present in your reactor already started consuming itself. If it is somewhere in between it represents the decline growth phase and if it is further it represents the low growth phase. So, normally our target is to normal range of biological reactor operations lies between this boundary of endogenous growth phase and the decline growth phase. We do not want it to be in a low growth phase why have why because in that case large production will increase in a in a like it will increase like anything.

So, we want to maintain it in between this in this boundary of endogenous growth phase and the decline growth phase. So, in this region your reactor will perform its in its optimum range. So, in case of continuous growth reactor continuous fed reactor so, we follow this issues this this particular phase we normally try to follow. In case of considering the settling capacity the settling capability of the sludge developed is also important. The sludge produced under the lock phase has a very poor settling characteristics due to their ability to hold water surrounding to the cell.

The sludge produced in the endogenous decay phase has a better settling properties and settle well as it is more stable. In case of endogenous growth phase it settles very well in the secondary sedimentation tank and does not create any problem in recycling the sludge process. Therefore, as in sludge developed under the lock growth phase poses a problem like sludge bulking in the secondary sedimentation tank. What is sludge bulking? Sludge bulking is nothing, but it is what happened there is a chances of some filamentous microorganisms still present in the this waste water and what it happens in the secondary sedimentation tank it creates a problem it does not settle well. The secondary sedimentation tank is introduced to settle the this or all the biological cell right cell biomass, but it does not settle well because of that this phenomena is called the sludge bulking.

And sludge bulking is very predominant when your biological treatment process follows the log growth phase, then in the next stage when it goes to the secondary sedimentation tank it started showcasing the sludge bulking phenomena. So, that is another reason why we need to follow we need to follow this endogenous growth phase this in between endogenous growth phase to decline growth phase this is the perfect or optimum range for biological reactor operation if you are following a continuous system, perfect. So, let us do one example as in the beginning we discussed that about the bacterial cell numbers. So, if you see in a laboratory bacterial growth experiment the culture was inoculated with 10 to the power 9 bacterial cells the doubling time of bacterial cell is 3 hour. So, what will be the bacterial cell number after 6, 24 and 36 hour? Given the N_0 is given as 10 to the power 9, TD is given as 3 hour or the doubling time and NT is to be determined for 6, 24 and 26 hour right.

So, NT we know from the equation N_0 into 2 to the power T by TD. So, after 6 hour N_6 will be 10 to the power 9 N_0 is 10 to the power 9 and T is 6 hour and TD is 3 hour. So, it will become if simply do it with your calculator it will become 4 into 10 to the power 9. So, that means, after 6 hour the number of bacterial cell in your experiment will be 4 into 10 to the power 9 that many bacteria will be there. What will happen after 24 hour? 10 to the power 9 into 2 to the power 24 by 3 24 here T is represented by 24 the amount will be 2.

56 into 10 to the power 11. After 36 hour it will be 10 to the power 9 into 2 to the power 36 by 3 4.096 into 10 to the power 12. So, after 6, 12 and 36 24 and 36 hour the number of bacterial cell now in the culture media will be 4 to the power 10 4 into 10 to the power 9

2.56 into 10 to the power 11 and 4.096 into 10 to the power 12 respectively. Can you imagine it is it is the amount of increment like within 36 hour from 10 to the power 9 it becomes 10 to the power 12 4.

096 into 10 to the power 12. So, this is the double based on doubling time this is how the bacterial cell will double like in a they reproduce and because of that their concentration will also increase in a particular biological reactors and all. Perfect. So, we gather a very good amount of information we if you are put very good concentration in this lecture if it is possible please go through it once more to get some very basic information and very important information I would say when we are going to design a biological reactor you need to understand this lecture very precisely . Because this lecture gives you the basics of how it works . The microorganisms majorly utilize the energy derived from the respiration to synthesize new protoplasm through the sets of reaction catalyzed by enzymes through the from portions of the substrate.

The growth of the bacterial culture is related to the synthesis of cellular components as well as the increase in number by multiplying individual bacterial cells to produce offspring or individual organisms. We also understand the under proper environmental conditions the soluble organic substance present in the wastewater destroyed by biological oxidation. And we also understand the biological treatment system usually consists of a biological reactor and a sedimentation tank to remove the produced biomass called the sludge. And we also understand the different types of reactions, the different type of growth kinetics and also how it differs from the batch process to the continuous process and all. So, we had a very thorough discussions about it these are the references you I would suggest you to like if you are interested please go through this paper it will give you much more in depth understanding about this subject.

And believe me if you want to become a if you want to become a water engineer or wastewater engineer to understand this concept you it is better if you go through this paper. So, it will give you some in depth understanding of these subjects. So, thank you so much we will see you in the next video. Thank you.