Aspects of Biochemical Engineering

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Lecture – 06
Stoichiometry of Biochemical Processes-I

Welcome back to my course, Aspects of Biochemical Engineering. In the last lecture, I tried to give the information on different bio products. As you know bio products of different varieties; one way different classifications we have, low value high volume products, medium value medium volume products and high value and low volume products. Now on the basis of cost this product may be classified. Now today I will be starting one very interesting topic which is very essential for the mass and energy balance of any kind of Biochemical Processes.

The topic that I am going to discuss that is Stoichiometry of Biochemical Processes. Now what do you mean by Stoichiometry? Stoichiometry we mean that interrelationship different component present in the reaction mixture. And also, it gives the information that how much that heat evolved that we have in the biochemical system. As you know that in the Biochemical Process, most of the Biochemical Processes, they are exothermic in nature. So, exothermic means they are heat liberating. So most of the Biochemical Processes are usually operated at ambient temperature and atmospheric pressure. So, it is close to 30-35 degree centigrade. Now particularly, since we are in the tropical country and during summer the temperature rise to as high as 40-45 degree centigrade.

So, (Refer Time: 02:08) since we have heat evolution in this particular bioreactor or Biochemical Processes. So, we should have more cooling arrangement. Question comes how much cooling arrangement is required; that can be determined if you find out how much is evolve in the Biochemical Process? So, Stoichiometry not only gives you the information of interactive molecular relationship of the different component present in the reaction mixture, but also heat evolve in this process. And also, it gives another information which is very important in the Biochemical Process that is the validity of the experimental result. Because we carry out some kind of experiments in the laboratory and naturally the results that we get that how to evaluate that whether the experimental data that I am reporting that is we are reporting that is right or wrong. So, these are the 3 different information we can get from the Stoichiometry of Bioprocess.

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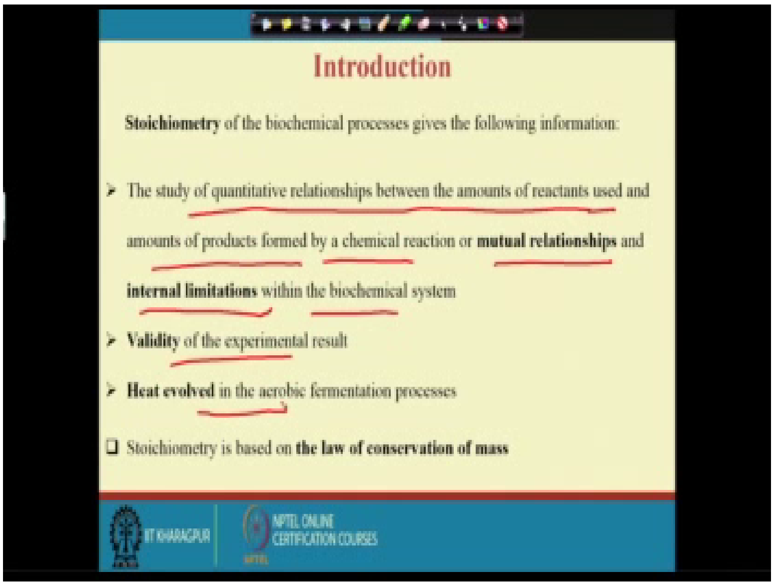

Now, here if you look at, so what I told it is like this, that the Stoichiometry of the Biochemical Process give the following information, study of quantitative relationship between among the reactants used and the product forms of a chemical reaction or mutual relationship and internal limitation within the biochemical system; that means, what I want to point out, how much of the substrate molecule gives how much a product, that theoretically that you can find out on the stoichiometry. Then another thing I told you the validity of experimental results. And third one is the heat evolved particularly in aerobic fermentation process.

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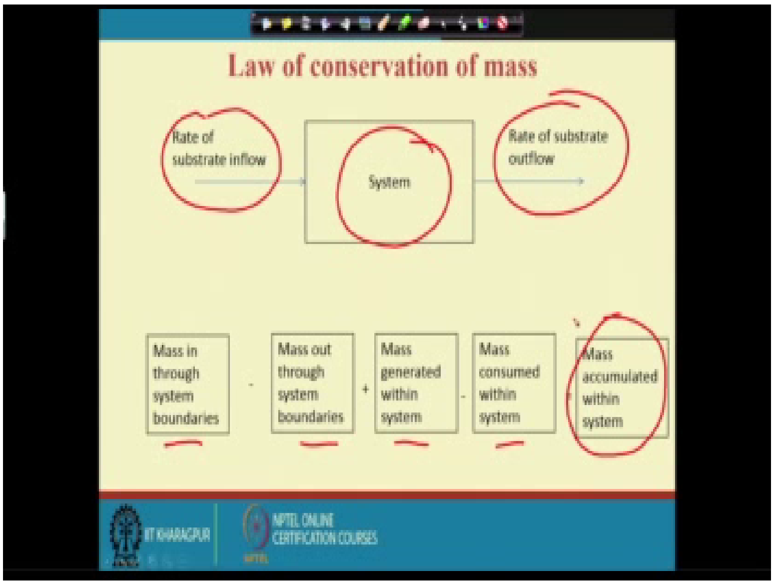

Now, law of; what do you mean by law of conservation of mass. Conservation of mass if you look at here that is rate of substrate inflow and rate of substrate outflow and this is passing through a system like this. Now that means, a law of conservation of mass that nothing can be destroyed nothing can be (Refer Time: 04:33). Overall balance of the process that should be remaining same. Now here if you look at the mass in through the system boundaries and minus mass out through the system boundaries and plus mass generated within the system minus must consumed within the system is equal to mass accumulation within the system. This is called conservation of mass. The ultimately that should remain same.

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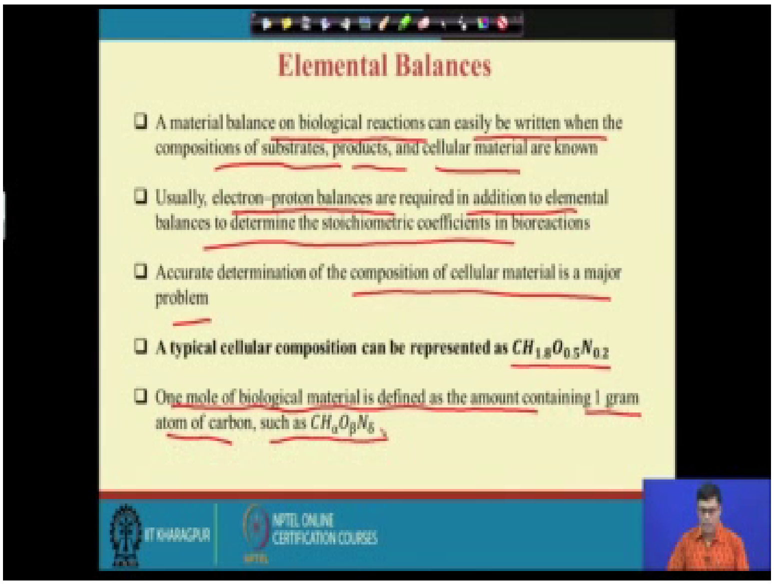

Now, that only the one, when you do the Stoichiometry of The Bioprocess that we shall have to do some elemental analysis and through these elemental analysis we can find out how much of substrate molecule reacts with how much of a, b, c, d. How much of a reacts with how much of b reacts to give how much of c and how much of d. So, this is you can you have some kind of elemental analysis we have. Now if you look at this elemental balance, the material balance; if you look at material balance of the biological reaction can be easily written within the composition of substrate product and similar mass are known. Because you know that in the biological system, because since we are talking about the bio system, what is happening in the bio system, you are putting your substrate then you are getting the product.

Now when you get the products your products may be of different either in the form of cell mass. Or maybe if it is cell mass mostly you get the cell mass, if you have other than cell mass. So, you get cell mass as well as the product. Now the usually the electron proton balance are required in addition to elemental balance to determine the stoichiometric coefficient of the. This either I shall discuss interest details, how you can find out the Stoichiometric Coefficient of The Biochemical Process. This is very interesting. Accurate determination of the composition of the cellular composition is the major problem. So, you what we what you do usually, we assume the ampere that kind of cellular composition of the biomass. This is CH 1.8 O 0.5 and N 0.2.

It is find within 5 percent limitations, it give the empirical that formula like this of the cell mass. So, another very important thing is one mole of biological material is defined as one gram atom of carbon that is CH alpha O beta and N delta. Now let me explain that.

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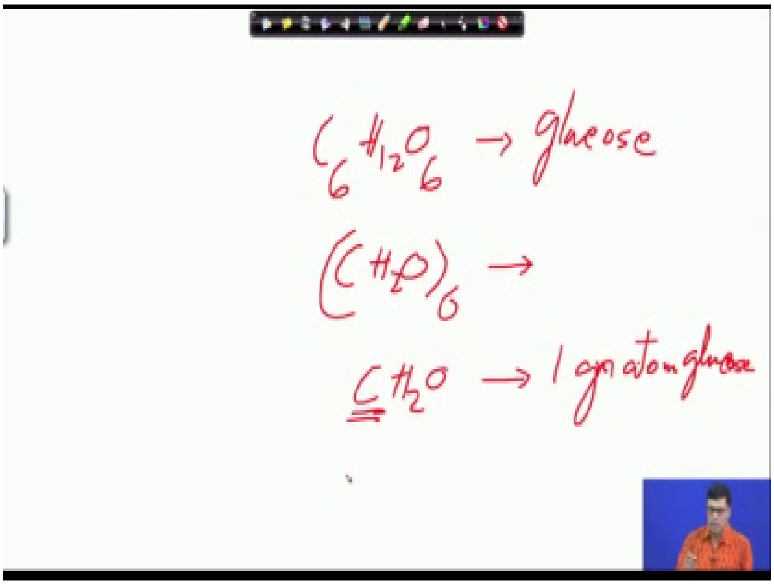

Because you know, suppose we are talking about Glucose C 6 H 12 O 6, this is we call it glucose. Now when you write this glucose in the form of this 1 gram atom of carbon, how you can write? This is CH 2 O 6. So, 1 gram atom carbon is of glucose is like this. This is 1 gram atom glucose, when you use a other substrate. Am I right?

So, that this is the similarly it is the applicable. Suppose any compounds when you determine the empirical formula, we can easily convert in the terms of 1 gram atom of carbon. This we can easily convert in terms of that not very difficult.

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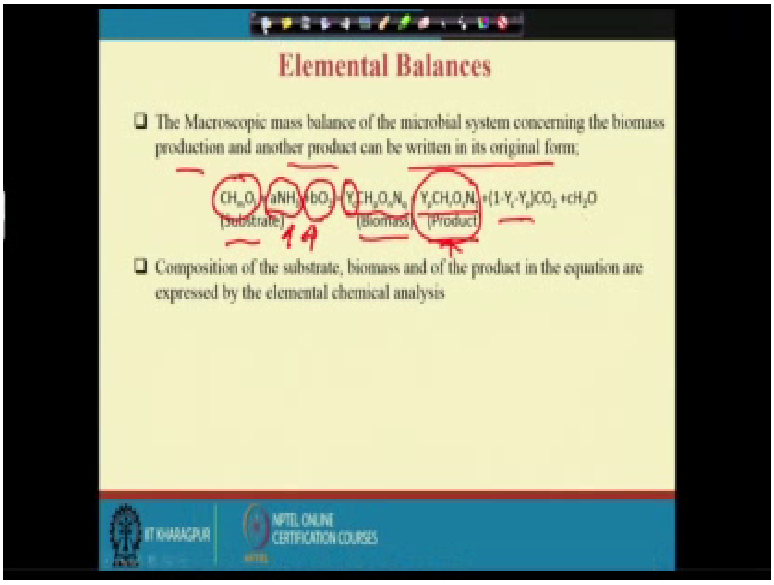

Now, here is the let me show you the macroscopic biomass balance of the microbial system concerning the biomass production and another product can be written in the original form. So, you know that when you write any kind of stoichiometry of the bioprocess it is essential that we should have some preliminary information on the system. As for examples, I want to point out, suppose I want to produce some kind of baker cyst.

Now in the baker cyst fermentation process, we are targeting to produce the biomass only. We are not targeting, may be carbon dioxide is a kind of byproduct of the system. As we know the carbon dioxide that produce in the baker cyst fermentation process or in case of ethanol fermentation processes. We have more days in the purified form and it is used for the production of dry ice. So, commonly that how we can write this equation, if you look at this is the empirical formula of substrate, this is CH m O l, this is the substrate. Now when you talk about biomass, we required some kind of nitrogen source. So, usually we put nitrogen in the form of ammonia. So, we can consider a mole of ammonia that is required. Then if it is the aerobic system, say aerobic system, we required molecular oxygen.

So, we can write b O 2, this is poly; (Refer Time: 10:07). So, in what we get, we get the products like biomass. This is the empirical formula of biomass. This is the empirical formula of product. So, Y c is the Y c moles of gram atomic biomass. And Y p gram atom of product. That is equal to now it is kind of carbon balance. You can see that 1 mole of carbon is converted to Y c moles of biomass and Y p moles of product. So, what will be the carbon dioxide 1 minus Y c minus Y p, this CO 2 and plus cH 2. So, but suppose we want to produce some kind of biomass. So, we can ignore the other than the product formation. Here the product formation that we have that we can ignore. But if we want to produce some kind of product as for example.

Eternal fermentation process or cyclic acid fermentation processes or penicillin fermentation process, then we can ignore this biomass formation. But if it is the anaerobic system, we know anaerobic system, that amount of cell must produce drastically reduced as compared to that of aerobic system. So, if we ignore that your relationship will be simple you know. One thing is that this oxygen will not be required; the nitrogen also mostly will not be required. Because if you don't consider biomass either you need the product formation show your carbon source is mostly converted to your product if product does not contain any nitrogen and also carbon dioxide and water. This is, but in case of hydrophobic fermentation process when you consider the cell mass, then you can ignore the product formation.

This you can ignore and you can write this is the substrate. This is the ammonia, this is carbon dioxide, this is oxygen, molecular oxygen, this is biomass carbon dioxide and water. So these kind of, suppose I can give you another example; suppose in the anaerobic digestion process the organic biomass, organic mass is converted to methane and carbon dioxide. Now when organic mass converted to methane and carbon dioxide, what we considered, since it is the anaerobic process you should not consider any kind of oxygen because it does not (Refer Time: 12:37) oxygen harmful for the system. And also at the anaerobic system we don't produce significant amount of biomass. So, nitrogen sources also we do not consider. That what we considered since this organic residue is like a, is a polymeric material. So, we required water molecule for the hydrolysis of this initial hydrolysis breakdown of the polymeric molecule then, when you get the monomer this is converted to methane and carbon dioxide.

So, what will be the equation you have? You have substrate plus water equal to methane plus carbon plus that carbon dioxide that you will get in the system may be with a hydrogen ion, that excess hydrogen ion might be present there. This is how you can write the balance equation.

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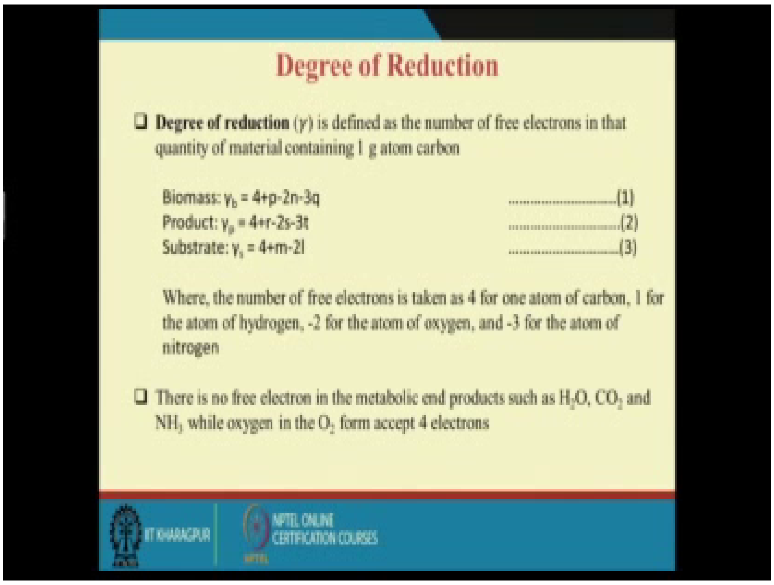

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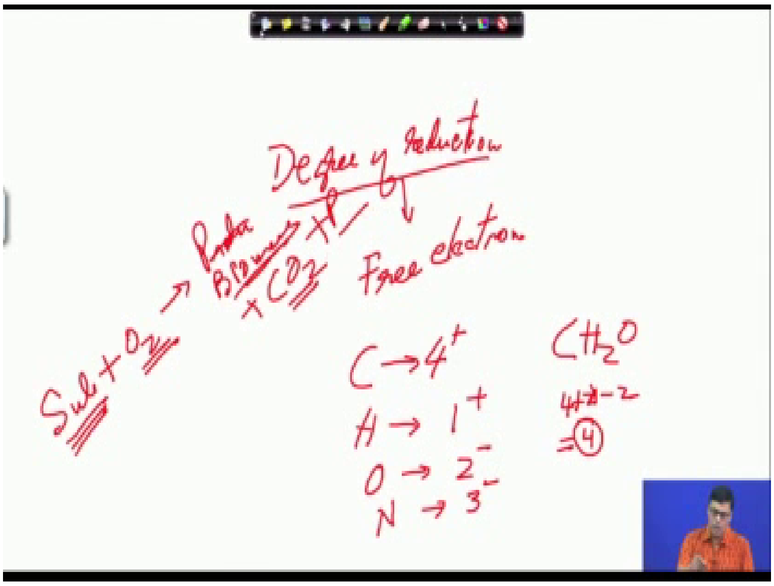

Now let me discuss about, discuss about the Degree of reduction. What do you mean by degree of reduction? Degree of reduction is basically it is the free electron present in a in the in any kind of reactant or product. Now question come how you calculate the free electrons? Because we considered that carbon if you look at carbon, what is carbon it has 4 electrons in the outermost orbit. So, you can take 4 more electrons. We consider we will see 4 plus. Now in case of hydrogen, we can consider it as 1 plus. Am I right? Now in case of this oxygen, it usually take 2 electrons. So, it is minus. The 4 electrons can go out 1 electrons can go out. So, it is plus and this since it can take 2 electrons, we can had nitrogen what we can write that in the outermost orbit it can it can take 3 electrons.

So, it is 3 minus. So, you know that that we can write like this. Now if you this is I told you this is the formula of glucose and 1gram atom the how if you do the electron balance a free electron, but this is 4 plus 2 minus 2 because, 2 into 1, because this is basically 2 into 1. So, it is coming around 4. So, 4 is the free electrons. So, degree of reduction basically it deals with that how much free electron present in the particular substrate molecule. Now suppose for ammonia carbon dioxide and water, we considered that degree of reduction is 0 that is the free electron is 0. So, when you write.

So, we can suppose we write a empirical equation like substrate plus oxygen give the kind of product or you know biomass, you can say biomass plus carbon dioxide. So, you know that we can write the balance equation how much that you know free electron present in the substrate, how much free electron present in the oxygen molecules, how much present in the biomass, how much this is will be 0. So, maybe if some product is there, then the product you find out that how much free electron. So, we can write a balance equation that particular. That I shall show you how you can do that.

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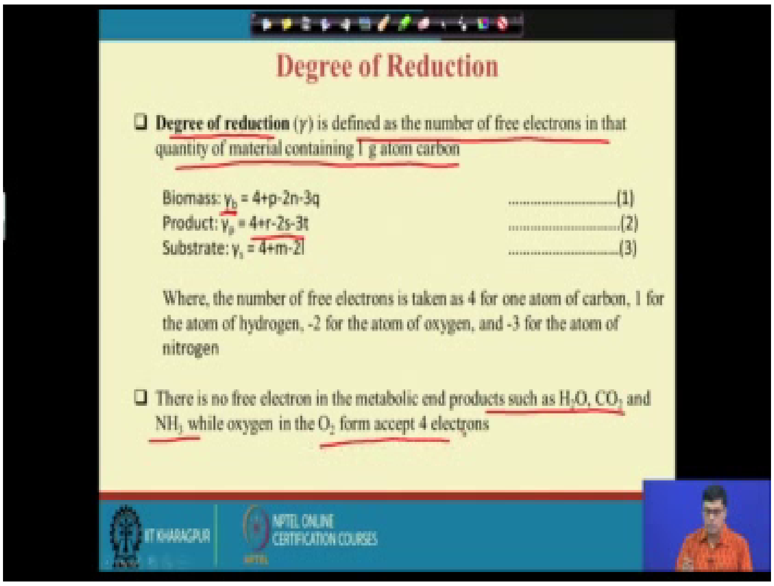

Now, here I have. So, let us go here, this is you see the degree of reduction is defined as the number of free electrons in that quantity of the material contents 1 gram atom carbon.

Now biomass, that you know if you look at the biomass that, this is biomass, for this is the formula of biomass CH p O n N q. Now if you look at this that how you do this balance therefore, 1 p is hydrogen and 2 n that is oxygen and 3 q is the nitrogen. So, this is how you can write this nitrogen and the free electron balance for with respect to biomass. So, it is we express with the gamma b. Now product similarly we can write the balance in case of process product. If you look at the product that, the product also you have this formula that product has.

So, what will be the product that is, this is the product. So, this is 4 plus r minus 2 s minus 2 t. This is how you can we can write that similarly substrate we can find out this. Now where the number of free electron taking 4 for 1 atom of carbon and 1 for hydrogen minus 2 for oxygen and minus 3 for nitrogen, this we shall have to remember. And no free electrons of the metabolic end products, that end product we have like carbon, water, carbon dioxide ammonia while oxygen O 2 except 4 electrons.

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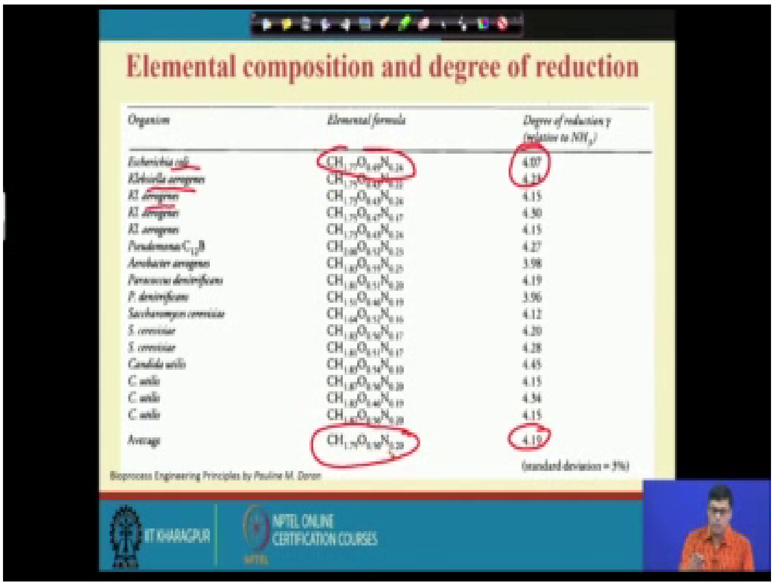

Now, here I have given some typical examples of elemental composition of different biomass. I can again this is the different like Escherichia coli this is the empirical formula we have. Then klebsiella aerogenes like klebsiella aerogene. So, different type of and you know that respective degree of freedom. So, I ask all of you, you can do the exercise and try to find out whether you are getting this value or not when in case of different types. So, this is the different and that is why on the basis of that we select that average composition empirical compound of the biomass is a CH 1.79 O 0.5 and N 0.2.

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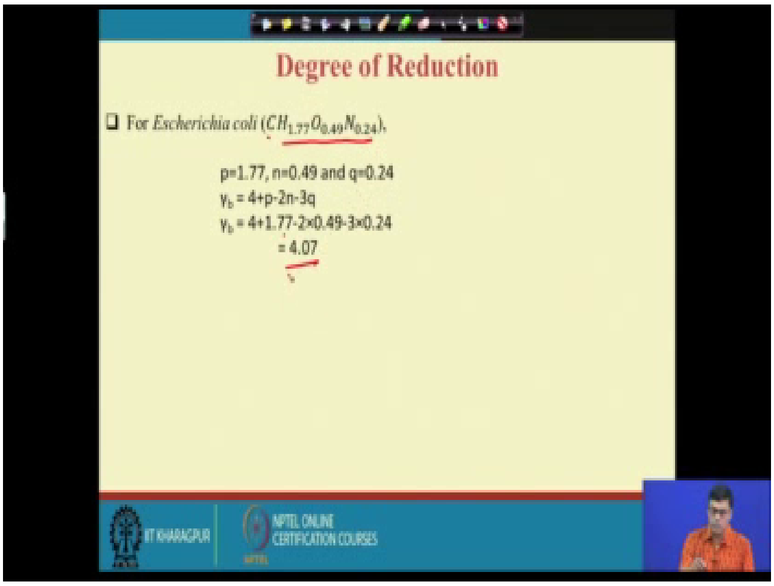

Now, in case of a Escherichia coli, that if you look at this is the formula that we have, just now we have found out, and what will be the free electrons this is 4 then 1.777 into 1, then 2 then 2 into 0.49 and 3 into 2.4. So, it is coming about 4.07. So, I hope it is clear to you.

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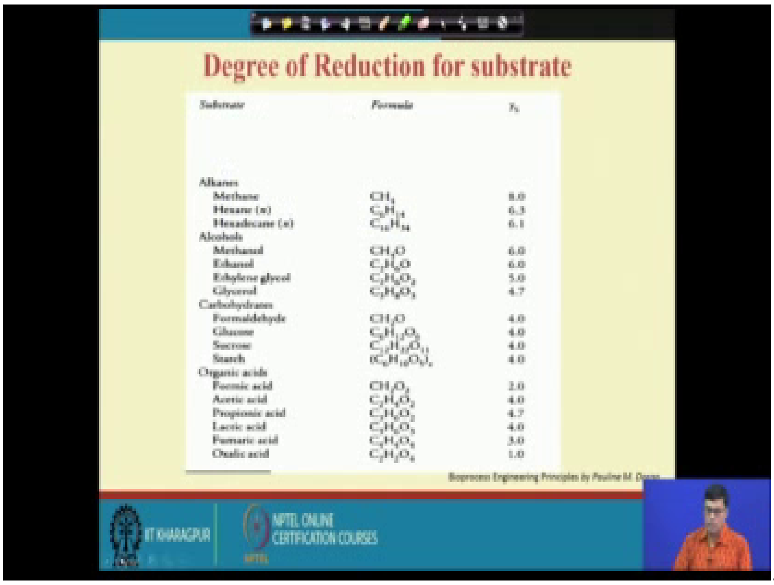

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Now, we similarly we have done some degree of reduction calculation with respect to some substrate molecule like alkanes, alcohol, carbohydrate and organic acid. This has come like that you can practice.

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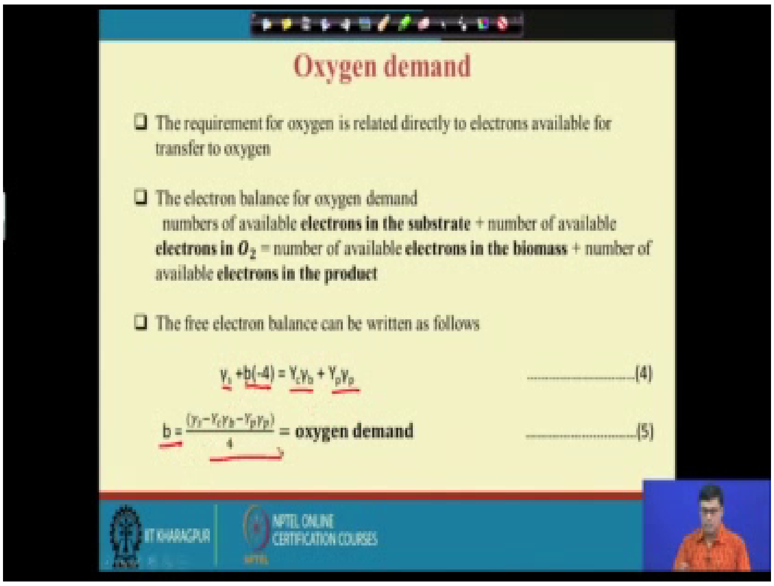

Now, oxygen demand how you can calculate because this is very important. So, I have already shown that you know our that common equation is this. There is a substrate plus ammonia plus oxygen it gives biomass product carbon dioxide and. Now if we do the balance this is with respect to substrate, this is the degree of reduction of substrate, then we know that 1 O 2 can accept 4 electrons.

So, it I can write b moles of O 2 that is b into 0.4 minus 4. And then Y c is the Y c moles of Y c gram atom of biomass. So, this is Y c gamma b and Y p gamma p. So, I can write that oxygen demand this b is the oxygen demand we can calculate like this.

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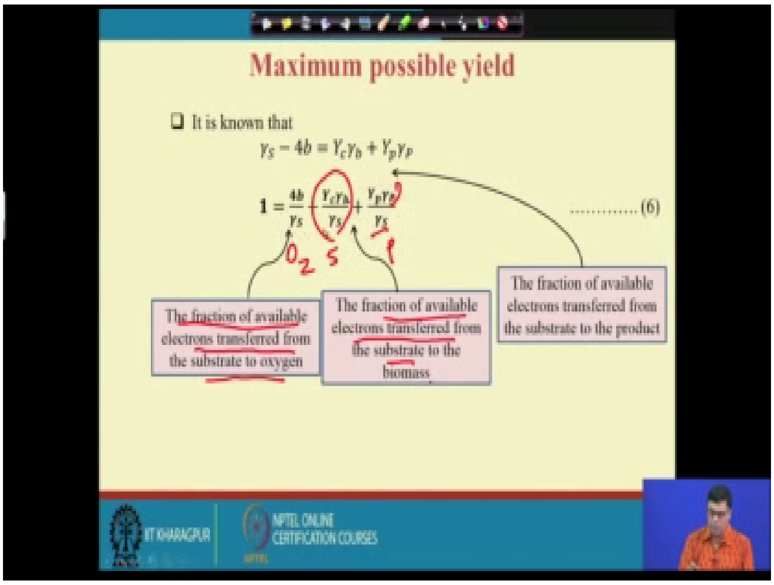

Now, again if we divide both side by gamma s we will get that you know that like this the now 4 b by gamma s is considered as the fraction, this considers the fraction available electron transferred from substrate to oxygen and this is the similarly, this is we consider the fraction available electron transfer to substrate to biomass, this is biomass and this is product. So, again this is fraction of electron that is transfer to substrate to this is biomass. So, you know that. So, this is oxygen, this is substrate, this is product, this is how electrons are transferring. So, we have 3 different portions here.

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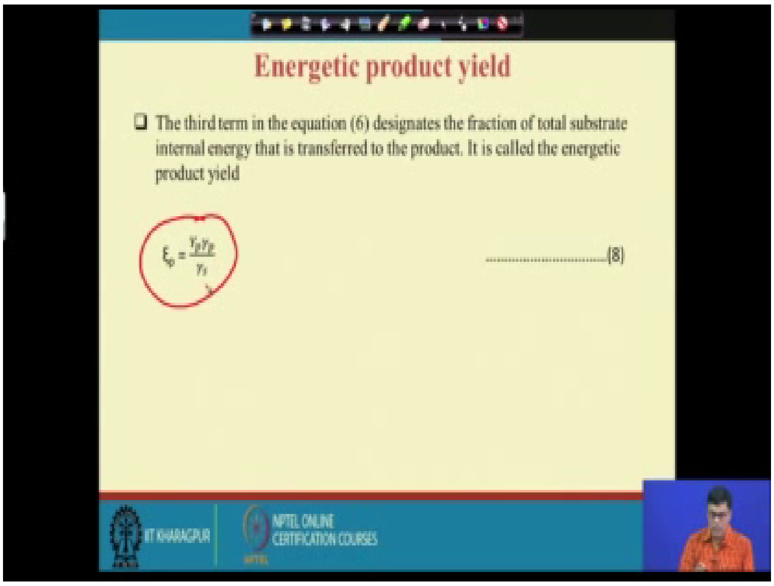

Now, this you know if you look at previous one, that you know this particular this portion if you look at this portion.

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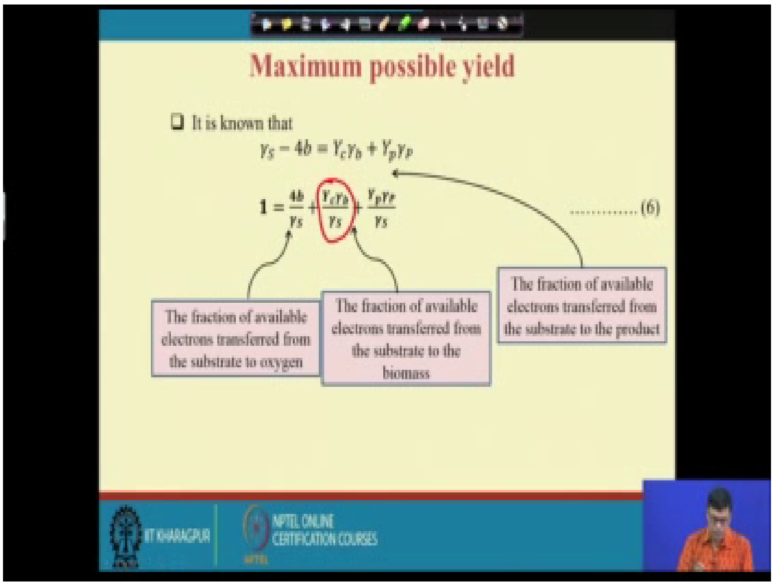

This is called energetic growth yield. And this is like this is. This is eta then Y c gamma p by y gamma s we consider that this is called energetic and growth yield. This is how we can write it. Now if you go through that second portion that here we can find out this portion, this portion we call energetic product yield. If you look at, this is this is called energetic product yield. So, energetic product yield Y p gamma p by gamma s. That we can we can do that like this.

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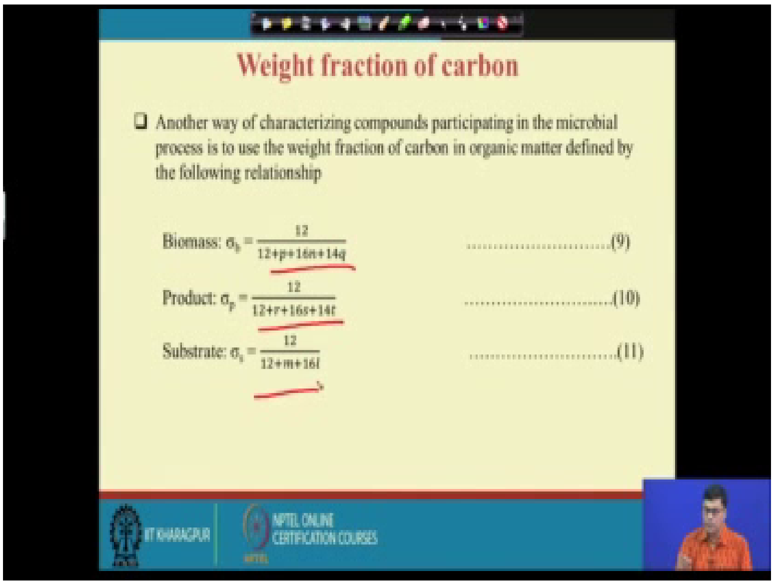

Now, another way we can differentiate this biomass product and substrate that is, on the basis of what weight fraction of carbon because. Let me explain that.

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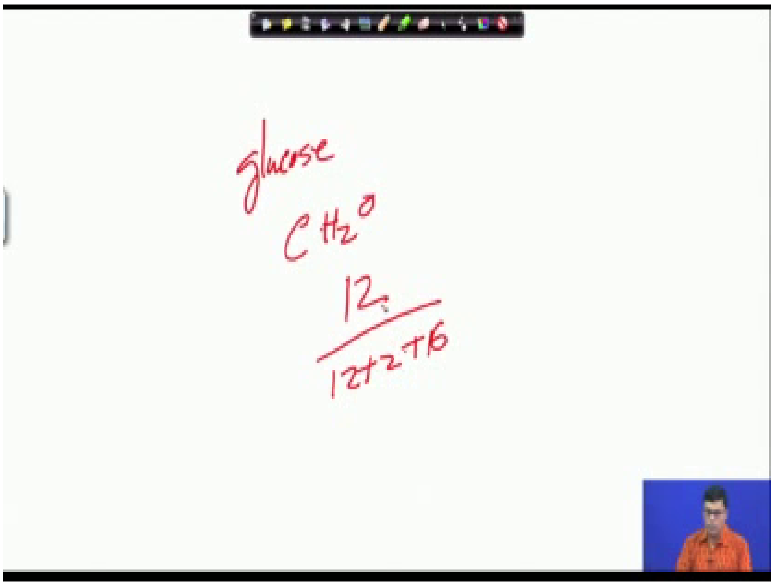

That suppose, that I was talking about glucose is CH 2 O. So, gamma fraction that weight fraction of carbon. So, what is the why to make weight over 12 and what is the molecular weight 12 plus 2 plus 16. So, this ratio is the weight fraction of carbon. So, you know that I can do it very easily.

So, this is similarly now in case of biomass, I can do that in case of product we can do that, we can do in case of substrate. Now biomass yield that another very interesting thing that is, let me explain that.

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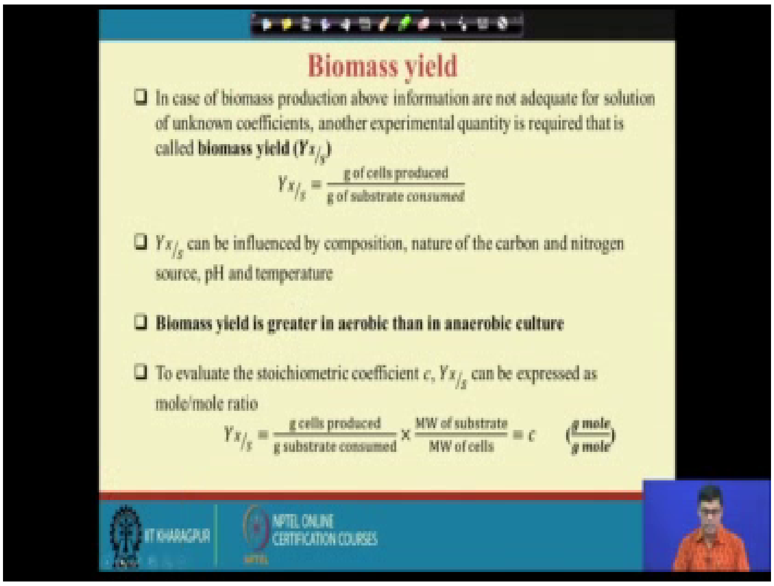

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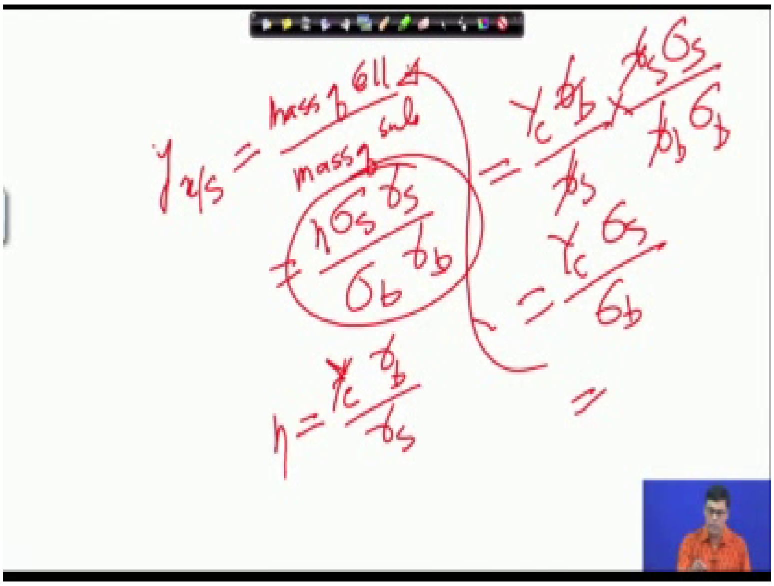

Now biomass yield is very interesting y x by s is a, what is equal to Mass of cells per Mass of substrate consumed. Now this is equal to.

Actually this is equal to eta sigma s gamma s then we have gamma b and sigma b. 'b' stands for biomass. Now question comes, how this equation has come? Now this we can easily find out that like this, that if you look at the eta we have already seen eta equal to Y c into gamma b by gamma s am I right. So, here if you put why c gamma b by gamma s multiplied by you have gamma s sigma s and gamma b and sigma b. So, this the gamma and gamma b and gamma b will cancel and this will cancel.

So, what will basically we have, this is we have equal to Y c into sigma s by sigma b. And if you, what is this, this is the atomic weight of carbon by molecular weight or gamma atomic weight of substrate, this is the atomic weight of carbon by gram atom of biomass. Now when you do that in biomass will go there and you multiply by this, you will find you will across this equation you will come across. So, you can easily find out how it has come. So, this is how it has come. This is.

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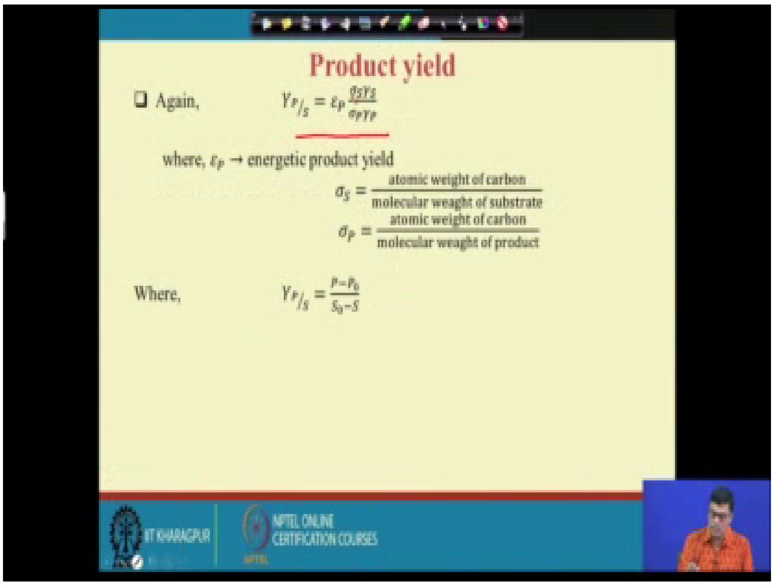

Now, similarly we will, we can calculate that other thing also. Then Y x by s is like this and then Y p by s is like this, epsilon p gamma s gamma means say sigma s gamma s sigma b gamma b. So, p this with respect to product we can find out. So, and gamma Y p by s is equal to P minus P 0 by S 0 minus S. So, we can easily calculate that.

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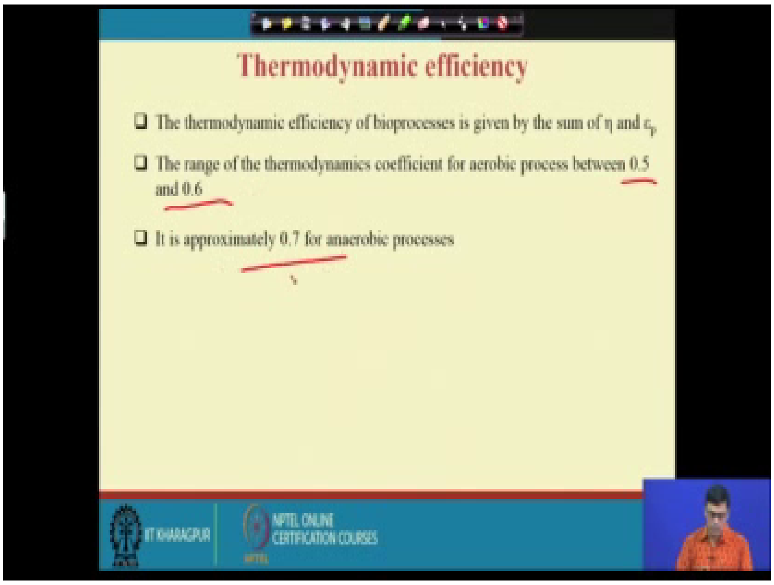

So, we know that, another thing I want to point out in case of biochemical system Thermodynamics ill coefficient that coefficient efficiency of the process plays very important role. Now in Thermodynamic efficiency in case of aerobic system, it varies from 0.5 to 0.6. And in case of an anaerobic system 0.7. Now this will help us to find out the validity, I told you the Stoichiometry help us to find out the validity of the experimental results.

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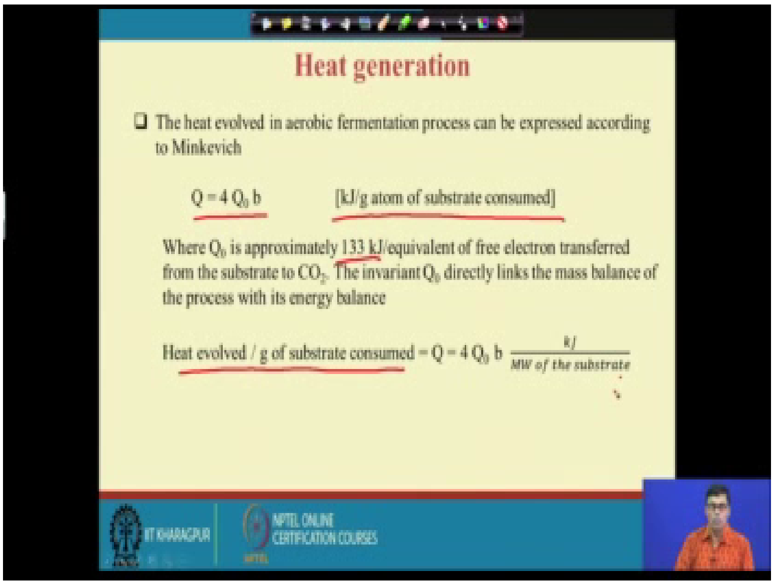

So, this will help us. And another thing I told you that the Stoichiometry also determined how much heat evolved in the biochemical process. The Q equal to 4 Q 0 into b and the Q 0 is equal to 133 kilo joule per equivalent of free electron transferred and to b. So, this unit is kg per gram atom substrate consumed. Now if you divide, this you can convert into if you divide by molecular weight of substrate, then it will be heat evolved per gram of substrate consumed. Now if you divide by yield coefficient, you can easily find out how much heat is devolved per gram of the same mass production. So, this is what I want to tell about the preliminary part of stoichiometry.

And here we get the information that how we can develop the inter molecular relationship with respect to elemental balance, with respect to electron and proton balance and then we find out that validity of the experiment result. Because on the basis of this thermodynamic ill coefficient, I will show you in the next couple of lectures and also we can find out the amount of heat evolved in this process.

Thank you very much.