Bioreactor Design and Analysis
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Lecture 14
Design of Continuous Bioreactors - Part 2

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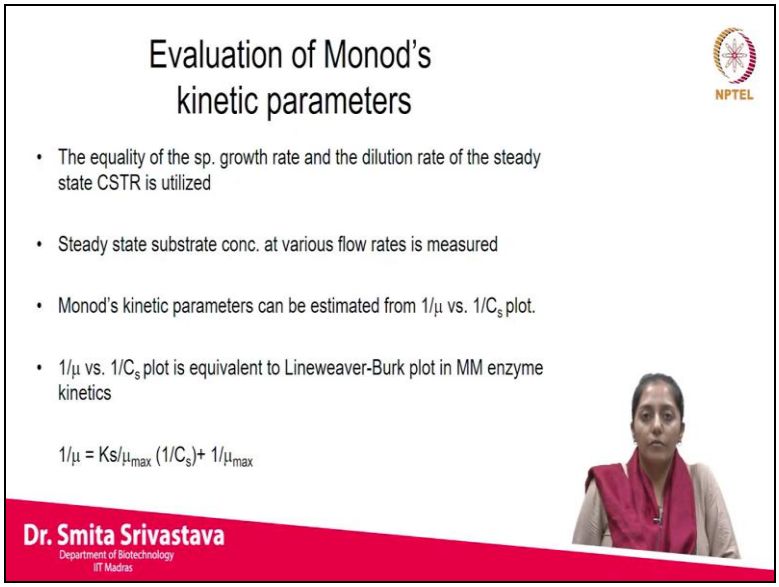

Welcome back students in the last class we were discussing about design of continuous bioreactors. So we will continue the same subject. So let us see how continuous cultivation can be used to determine the constants of Monod's law. So we have been hearing now for quite some time that microbial fermentations mostly follow Monod's kinetics, so now if we want to determine the constants of the Monod's model.

So if you remember the Monod's model Monod model was mu is equals to mu max S upon S + K S. So now if we want to determine the value of mu max and K S for a given fermentation system then we can make use of continuous cultivations how because now we know that in a continuous mode of cultivation one can manipulate the growth rate of the culture by changing the dilution rates. And running a chemostat would mean that whatever is the dilution rate will be equal to the specific growth rate of the culture as we learnt for a chemostat D becomes equal to mu.

So now the equality of the specific growth rate and the dilution rate of the steady state CSTR can be utilized to determine the Monod's constants. Now steady state substrate

concentrations at various flow rates are measured Monod's kinetic parameters can then be estimated by making a plot of 1 by mu versus 1 by S or here this is called as on the slide C with the subscript S which is your steady state substrate concentration.

So 1 by mu versus 1 by C S plot this is equivalent to the Lineweaver-Burk plot in the Michaelis-Menten enzyme kinetics. So if you see 1 by mu versus 1 by S by the rearrangement of the Monod's model then Y stands for 1 by mu and 1 by C S stands for X in a Y by X plot. So this becomes a linear function a line which is having a positive slope of K S by mu max and a Y intercept of 1 by mu max. So here the Y was 1 by mu and the X axis we have 1 by S or 1 by C S here.

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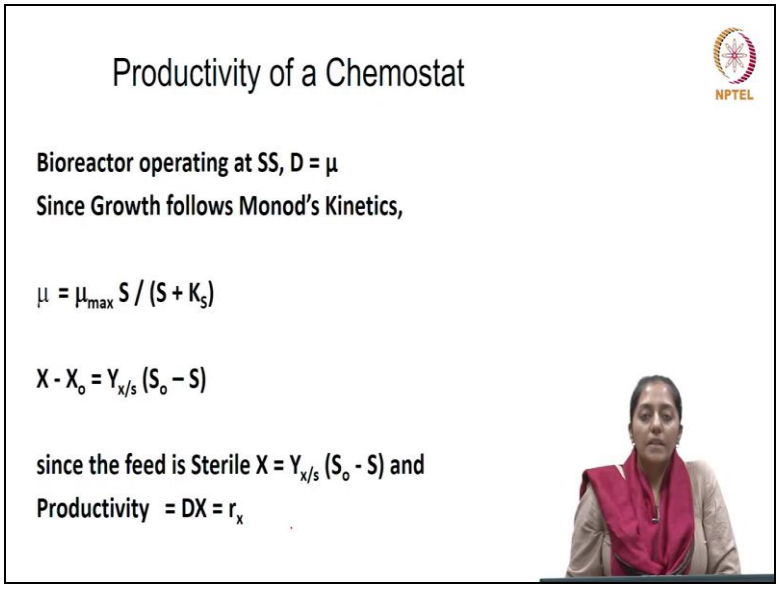

Now let us see how we can calculate the productivity of a chemostat. Now bioreactor operating at a steady state we know gives D is equal to mu. Since the growth we are assuming is following Monod's kinetics mu can be expanded in the form of Monod's model as given here in this equation let us call it as 1. Now equation 2 can further relate the steady state biomass and the steady state substrate concentrations in terms of the yield coefficient and the feed substrate concentration and the biomass concentration.

As the feed is sterile so your X naught goes to zero and therefore your steady state biomass in a chemostat can be given as a function of the feed substrate concentration steady state substrate concentration and the yield coefficient. So then the productivity which is volumetric productivity can be given as D times X which is nothing but your dx by dt or your r x.

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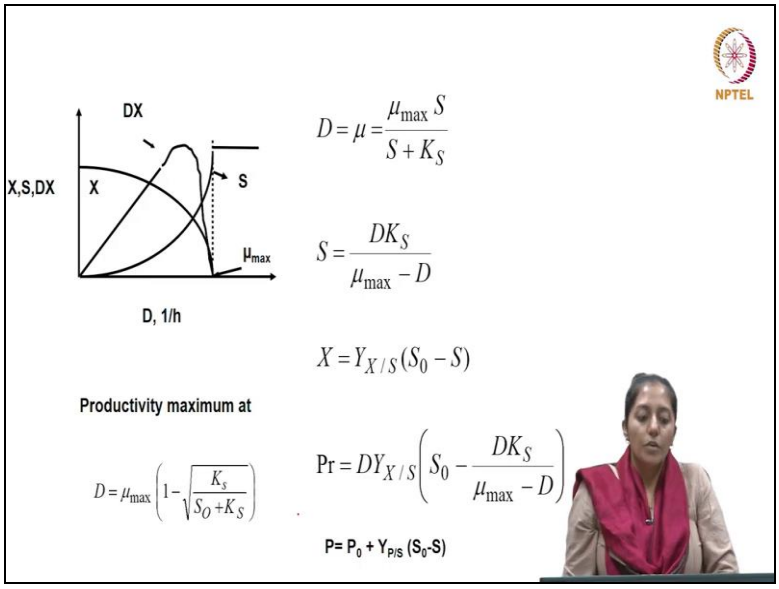

So if you make a plot of dx versus D or X or S versus t how will it look like? It look like as shown on the slide your dx will keep on increasing linearly as the D will increase but beyond a point it will sharply decrease and your productivity will become nearly zero at a D value which is equal to mu max. This is the point which we discussed in the last class as the washout point.

So at the washout if you see your substrate profile here it shows that it reaches a value and it becomes constant. So can you guess what would be the value where it is showing to become now constant after that? So this would be nothing but your feed substrate concentration after the wash out because there is no substrate getting consumed further similarly you can see how the X will change as the dilution rate keeps on increasing the X is initially more or less the same but then as you keep on increasing the dilution rate your biomass concentration in the outlet stream will come down till it reaches 0 at the washout point.

Because this also demonstrates that as the dilution rate is increasing the rate at which the biomass is leaving the reactor is now becoming greater than the rate at which the biomass is growing inside the reactor. So suddenly you will see near the washout there will be a dip in the biomass sharply going down to zero and otherwise it remains nearly same. So if you want to find that dilution rate at which the productivity is maximum, we are here referring to productivity as biomass productivity.

So if we need to determine the dilution rate at which the biomass productivity would be maximum then let us see how we can calculate we know the culture is following Monod

kinetics. We will do the rearrangement and we also know from the last few lectures that for a chemo start at steady state the D state substrate concentration can be related to the dilution rate by this function given let us name it as equation one.

And for a chemostat at steady state the steady state biomass can be related to the steady state substrate concentration as shown in equation 2. So now we know productivity is D times X. So if you want to find the dilution rate then we need to make the entire function as a function of dilution rate right now D is variable and X is also a variable in productivity. So if we substitute for X in terms of D then we first substitute X in terms of S and then S is further substitute substituted in terms of D.

So then you end up in equation 3. Now if you see in equation 3 it is only a function of D variable rest all are known or constants. So then if this productivity; now is a function of D. So if we want to find the maxima of this mathematical function we will set its derivative the first derivative with respect to D as 0 and we will determine that value of D. So that value of D turns out to be this.

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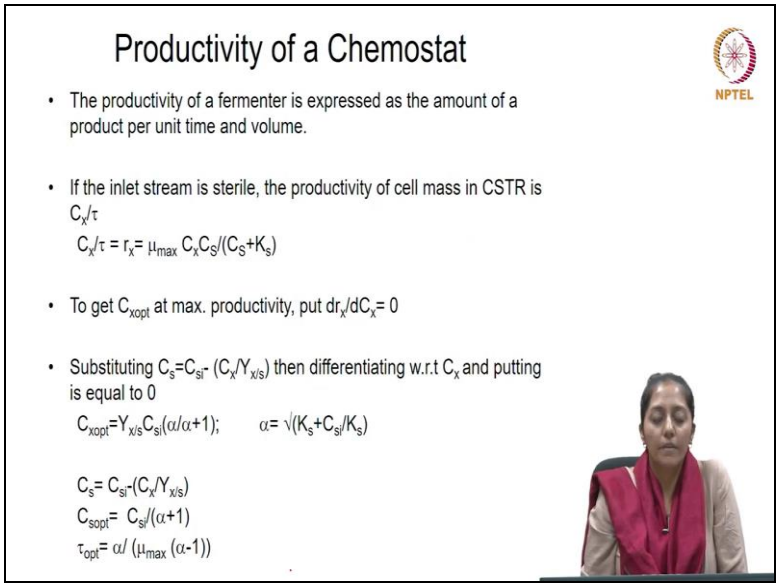

So in terms of residence time the productivity of the fermenter is expressed as the amount of a product per unit time and volume. So this is volumetric productivity if the inlet stream is sterile as I said earlier the productivity of cell mass in a CSTR can be given as C x by tau, tau is the residence time here. So now C x by tau is nothing but your r x which was equal to dx D is an inverse of 1 by tower so therefore this C x by tau.

Then again r x has been expanded in the form of Monod's model. So r x is mu X and mu has been substituted in terms of the Monod's model terms and X here is being represented as C x. Now; to get that if suppose we want to find what will be that optimum biomass concentration to achieve maximum productivity. So then again as we did earlier to find out the optimum dilution rate. So now here r x was productivity so we will set dr x by dC x as 0.

If our r x becomes only a function of C x variable here. So this C s has to be removed in terms of C x so how do we remove that making use of the yield coefficient so where C s can be written as C si which is your feed substrate concentration in the inlet feed minus C x is your steady state biomass Y by X by S is your yield coefficient. So now if this C s is substituted here in equation 1 and then differential of that equation with respect to C x is done and it is set to 0 to find the value of C x.

Then we find that the C x can be written as given here where alpha stands for K s and C s0. So then once you know the C x optimum you can find the value of C s using the yield coefficient and this is your substrate concentration at which your productivity is maximum and your dilution rate as we did earlier also turns out to be the same function as the earlier one. So where alpha so there can be more than one base for reaching to the solution.

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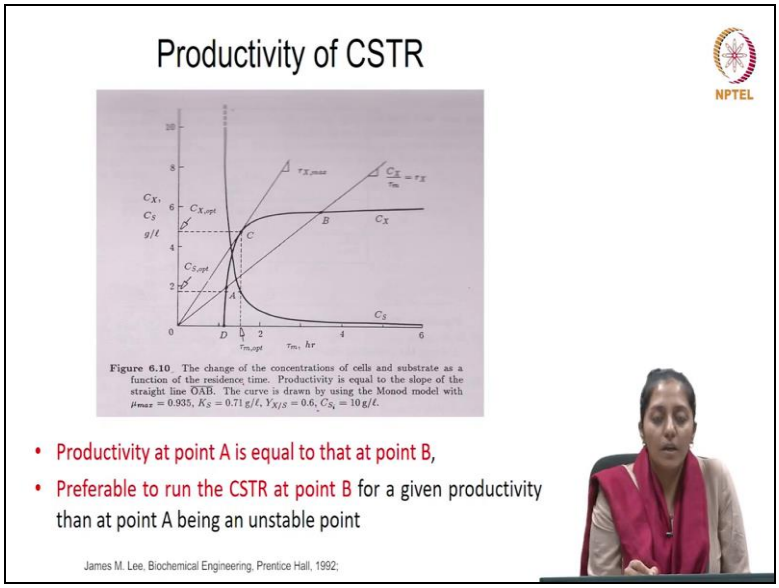

So if you see on the plot it is a plot between the steady state biomass the steady state substrate concentration in the outlet stream versus the residence time. So as the residence time decreases which means the dilution rate is increasing the biomass starts decreasing beyond a point and becomes nearly equal to 0 at point D. Now this D point is nothing but the washout

point. If you make a line crossing this curve wherever it cuts the maximum slope is what is your maximum productivity which is your r x.

So their maximum productivity your C x optima would be the point at which it is a tangent on this curve and on X axis it will be that resonance time or the D optimum. If you see the line OAB it is cutting the curve at 2 points A and B this would mean what that the productivity. Which is nothing but the slope of this line is the same at B and at point A which means we have a choice we can run a chemostat if the desired productivity is the slope of the OAB line either at point B or at point A to reach to that productivity.

So then how do we choose whether one should run this chemostat at point B or at point A can you make a guess? Right so one would like to run the chemostat to achieve this productivity at point B rather than being at point a because point A is very close to D which is the washout point which means the dilution rate is very high at point A even though the biomass is lower. So if you see your productivity which is r x is dependent on the C x and the tau m.

Higher the value of C x higher will be the productivity lower the value of residence time higher will be the productivity. So if you see at point A and B at point b the residence time is high and the biomass is also high. And at point A the residence time is low which means the dilution rate is high while the biomass is less. We do not prefer to run the chemostat at point A because it is close to the washout point which is the point D.

So productivity at point A in the plot is equal to that at point B it is preferable to run the CSTR at point B for a given productivity than at point A because it is an unstable point.

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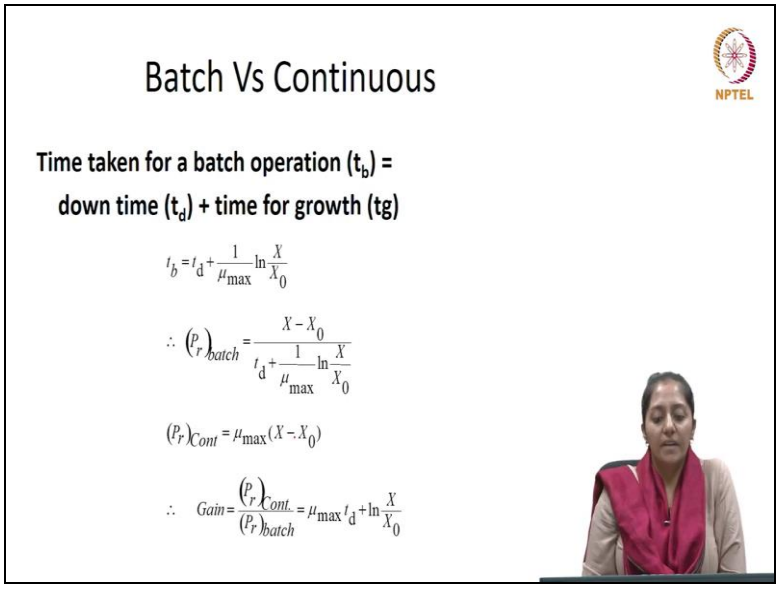

So now let us do a comparison of productivities which we achieve in a batch or and continuous. So let us assume that the time taken for the batch operation is being represented as t b as shown on the slide. The down time as t d and the time the batch time is being given as or for growth to be more precise is being given as t g. So then the total batch time here which is the time spent for the till we run the next batch will be the downtime plus the time spent for the growth which we generally call as the batch time.

So the for the productivity in batch the actual productivity in batch should take into account the downtime as well as the growth time. So therefore your productivity for a batch is written as the final biomass minus the initial biomass which means the total amount of biomass produced by the time taken to produce that. So this is your total time which includes the downtime. Now let us see what is the productivity in a continuous reactor?

So in a continuous reactor we do not consider any downtime here. So it is continuously can be run for a long time that is an advantage over the batch reactors. And your productivities can be given as mu max times X - X naught. Now X naught here is the biomass concentration in the feed which is generally sterile but still let us keep it as it is. So then if you want to know the gain in productivity while running a continuous reactor versus a batch reactor we do the productivity obtained and continuous by the productivity obtained in batch.

And this amount is the gain which we have in the productivity when we run a continuous reactor. So this in a way proves that the productivity in a continuous reactor is higher than in batch by this much fold.

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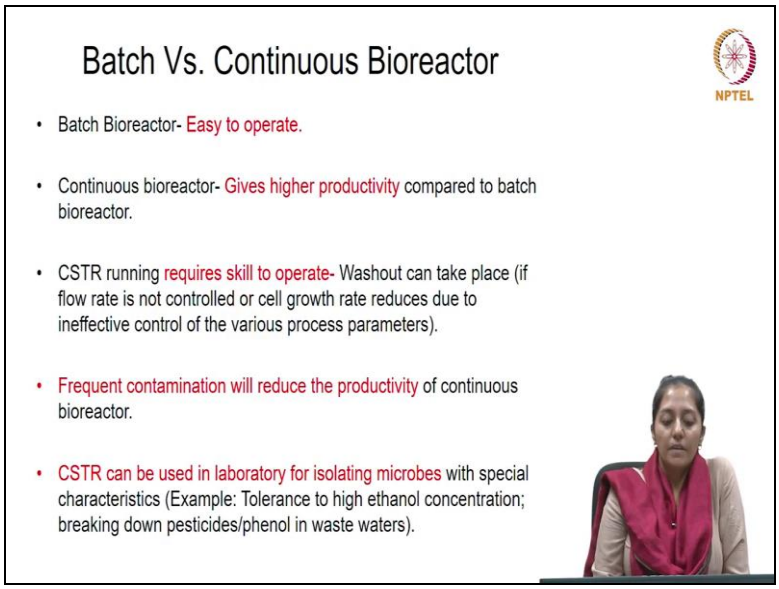

Comparing batch versus continuous reactors batch reactors they are easy to operate continuous reactors they give higher productivity as compared to batch reactors containers reactors to run they require skill to operate washout can take place if the flow rate is not controlled well or the cell growth rate reduces due to the ineffective control of the various process parameters. So which means that it needs continuous monitoring which requires killed personnel, frequent contaminations can reduce the productivity of a continuous reactor.

So that is also a limitation because these are long drawn fermentation processes. Continuous stir tank reactor can be used in laboratory for isolating microbes. So they give us an opportunity of screening under selection pressures. Like for example if one has to find a microbial strain which has higher tolerance to high ethanol concentration or if you want to select a microbial strain which can break down a pesticide or phenol in waste waters and therefore can survive the toxic levels.

Then these selection pressures can be applied to this group of organisms coming inside the continuous reactor and with time there will be due to the selection pressure what remains viable would be the cell line which is able to survive or they are also used for selecting mutants. Again if we want to compare batch versus continuous reactors and we not need to decide the optimum fermenter system given the maximum productivity for the desired conversion or production.

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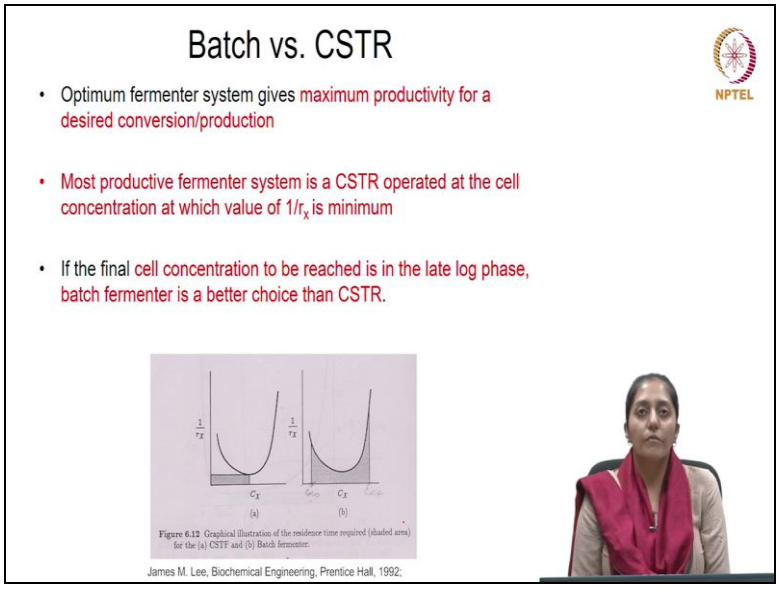

So if we suppose want to choose an optimum fermented fermenter system which can give maximum productivity for a desired conversion or production conversion here means the substrate conversion. So then how do we determine? We know that the most productive system is the CSTR which can be operated at the cell concentration at which your 1 by r x is minimum. If you see the plot of 1 by r x versus C x, so we know that the productivity which is r x is given as C x by tau x is not it.

Now maximum productivity for the desired biomass concentration if your residence time or the time taken to achieve that biomass concentration is minimum. So now if you see the plot how 1 by r x versus C x looks like it looks like a U-shaped curve, why? Yes for the reason you can visualize let us see the extremes when the C x is very less it is because the growth rate is very less.

So as the growth rate improves 1 by r x value is coming down and your C x will keep on improving till all the substrate is consumed and again the growth rate then becomes sharply goes to infinity it is shown here. So that it goes to very low value nearly 0 and that is your maximum biomass which can be achieved in that system. So if you need to find the most optimum system productive fermenter then it will be a CSTR which is running at the cell concentration at which this 1 by r x is minimum.

So that r x is maximum so when is the r x maximum? Your productivity is maximum that tip of the point the turning point. So CSTR would be C x by tau x, so your residence time minimum is this at which this is your C x optimum. Now if the final cell concentration which

is to be achieved is in the late log phase which is here somewhere here in the late log phase when the substrate is getting depleted then what can be a better fermenter?

Then a batch fermenter is a better choice than a CSTR how? Let us suppose this one. So if this is the final biomass concentration which is coming in the late log phase. Then if we run a CSTR at this point the residence time would be the shaded area. Now this shaded area and otherwise if you run a batch let us assume the starter culture concentration is C x naught here. So then; your residence time would be the area under the curve which is nothing but only this much.

So this area is much less than the area for the CSTR to achieve the same final biomass concentration. So your productivity when the biomass concentration desired is towards the end of log phase is higher when you run a batch fermenter than in a continuous reactor otherwise continuous fermenters would be the optimum fermenters to run when the productivity to be achieved is maximum and your biomass there is your C x optimum.

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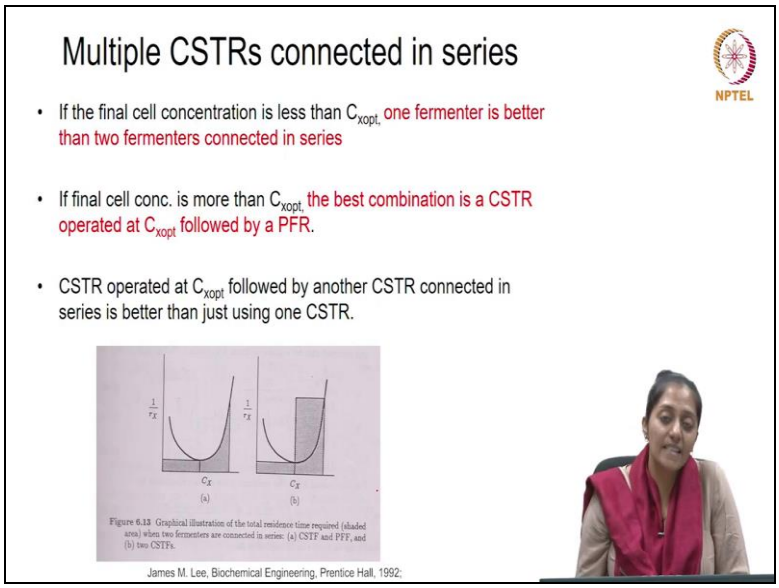

Now if the final cell concentration is less than C x optimum then what can be done then whether one fermenter is better or 2 fermenters connected in series would be better. Let us visualize so if the final cell concentration is less than C x optimum one fermenter is found to be better than the 2 fermenters connected in series because then a batch fermenter is better. If the final cell concentration is more than the C x optimum then the best combination would be a CSTR running at the C x optimum.

Where 1 by r x is minimum followed by a batch reactor which is a plug flow reactor which is similar follows the similar batch kinetics followed by a RFRr which is your plug flow reactor. So if you see on the figure which is shown on the slide and see in terms of the residence time you can clearly make out. Now here the C x of desired is beyond the C x optima point. So this is C x optima.

So C x desired is beyond the C x optima so then in that case there are 2 scenarios in the first figure they have connected the CSTR running at C x optima with a PFR. And in the second figure again there is a C x CSTR running at C x optima but connected with another CSTR. So I hope you can easily make out that the residence time when 2 CSTRs are connected is much more than the residence time when a CSTR is connected with a PFR.

And when this total time increases your productivity would reduce. So therefore the best is to use the first combination where we run one CSTR running at C x optima followed by a PFR to get the final biomass. However running one CSTR only to reach to that final biomass concentration still is not preferable even with the combination of one 2 CSTRs is connected in series because there is again a more time resonance time required if only single CSTR is running.

So therefore CSTR operated at C x optimum followed by another CSTR connected in series is better than just using one CSTR.