

Bioreactor Design and Analysis
Prof. Dr. Smita Srivastava
Department of Biotechnology
Indian Institute of Technology - Madras

Lecture – 23
Mass Transfer in Bioreactors – Practice Problems

(Refer Slide Time: 00:25)

Problem 1

A 20-l stirred fermenter containing a *Bacillus thuringiensis* culture at 30°C is used for production of microbial insecticide, $k_L a$ is determined using the dynamic method. Air flow is shut off for a few minutes and the dissolved-oxygen level drops; the air supply is then re-connected. When steady state is established, the dissolved-oxygen tension is 78% air saturation. The following results are obtained.

(a) Estimate $k_L a$.

(b) An error is made determining the steady-state oxygen level which, instead of 78%, is taken as 70%. What is the percentage error in resulting $k_L a$?

Time (s)	5 (t_1)	15 (t_2)
Oxygen tension (% air saturation)	50 (C_1)	66 (C_2)

Handwritten notes on slide:
 $\frac{dC}{dt} = OTR - OUR$
 $k_L a (C^* - C) = OTR = OUR$
 $k_L a (C^* - 78) = OTR = OUR$
 $\frac{dC}{dt} = k_L a (C^* - C)$
 $\ln \frac{C^* - C_1}{C^* - C_2} = k_L a (t_2 - t_1)$
 $\ln \frac{C^* - 50}{C^* - 66} = k_L a (15 - 5)$
 $\ln \frac{C^* - 50}{C^* - 66} = k_L a (10)$
 $k_L a = \frac{1}{10} \ln \frac{C^* - 50}{C^* - 66}$
 $C^* = 78$
 $k_L a = \frac{1}{10} \ln \frac{78 - 50}{78 - 66} = \frac{1}{10} \ln \frac{28}{12} = \frac{1}{10} \ln 2.33 = \frac{0.85}{10} = 0.085 \text{ s}^{-1}$
 Graph showing oxygen concentration vs time with a step change and exponential recovery curve.

Welcome back students. So today we are going to do some practice problems on the mass transfer module. So, let us begin with the first problem. What you can see on the slide let us read the problem and try to understand. So, the first problem says that there are 20 litres stirred fermenter which contains the bacterium *Bacillus thuringiensis* culture at 30 degree C and it is being used for the production of microbial insecticide.

So, the volumetric mass transfer coefficient which is given here as $k_L a$ is determined using the dynamic method which means if you remember the culture is present in the reactor and they are trying to find the volumetric mass transfer coefficient. Air flow is shut off for a few minutes and the dissolved oxygen level drops okay. The air supply is then reconnected. So, if you remember the profile for the dynamic method was that at certain time point the air flow is shut off it falls down.

And then before it reaches the below the critical concentration of oxygen the air flow is again resumed and the concentration of oxygen in the bulk is measured with respect to time. So they say that the steady state established and the dissolved oxygen tension at this point is

78%, The following results are obtained. So, they have given their data in the form of table where at 5 seconds the oxygen tension calculated is found to be 50%.

And then at 15 seconds, so this was 5 second, it is found to be 66%. Now with this data we need to find the value of the volumetric mass transfer coefficient okay. So, if you remember the mass balance for oxygen the rate of accumulation of oxygen in the medium where C_L is the concentration of dissolved oxygen in the medium was given by the difference between the OTR and the OUR.

Now OTR where it stands for oxygen transport rate and OUR stands for oxygen uptake rate. Now oxygen uptake rate in turn could be defined as the club of the specific oxygen demand of the culture multiplied by the total biomass present at that time. And if you remember the assumptions in the dynamic method while calculating $k_L a$ it was that we will assume that during this process of data collection and finding the value of $k_L a$ the biomass is not changing.

So, we can assume the consumption term to be the same and OTR is given as we all know volumetric mass transfer coefficient multiplied by the concentration gradient, C^* is your saturation concentration of oxygen concentration and C_L is the bulk oxygen concentration at any time point. So how do we calculate the value of $k_L a$ here? Now with this equation for mass transfer of oxygen, the accumulation rate of oxygen in the bulk we know that at steady state 78% is the bulk oxygen concentration which we can measure.

Now at steady state which means dC_L/dt is 0. So OTR will be equal to OUR. So, because we do not know about the q_{O_2} and X given in the problem but we can find the entire OUR which is not changing by finding now the OTR at this time of steady state. So what is OTR? The OTR will be $k_L a$ multiplied by $C^* - 78$. So, they are saying that the dissolved oxygen concentration at steady state is 78.

So, we know that this is equal to our OUR, which we can give as $q_{O_2} X$ specific oxygen demand of the culture multiplied by the biomass concentration. We know now that this is what is OUR. Now at any time points the dC_L/dt can therefore be written as $k_L a$ multiplied by $C^* - C_L$ bulk oxygen concentration at that time $- q_{O_2} X$ which is OUR. Now OUR can be substituted by $k_L a$ multiplied by $C^* - 78$ is not it?

Now if we expand this in turn the dC_L by dt can be written as, so this term will get cancelled if you open up the brackets and it will become $k_L a$ times $78 - C_L$. This will become this. Now if we integrate this if you need to calculate now k_L given the data C_L and t then let us integrate this. So integrating this will become $\ln 78 - C_{L1}$, now let us assume this is C_{L1} C_{L2} limit and this is t_1 and t_2 by $78 - C_{L2} = t_2 - t_1$, is not it?

So, this upon integration can be converted like this. Now let us say this is t_1 , this is t_2 , this is C_{L1} and this is C_{L2} . So if you need to calculate the value of this should be multiplied by $k_L a$. Now if we need to find the value of $k_L a$ and we know t_2 , t_1 one, C_{L1} , C_{L2} $k_L a$ can be obtained.

(Refer Slide Time: 07:34)

Solution

(a)

$$\left(\frac{dC}{dt}\right) = k_L a (C^* - C)$$

$$k_L a = \frac{\ln\left(\frac{C^* - C_1}{C^* - C_2}\right)}{t_2 - t_1}$$

$$k_L a = \frac{\ln\left(\frac{78 - 50}{78 - 66}\right)}{15 - 5}$$

$$k_L a = 0.085 \text{ s}^{-1}$$

(b)

$$k_L a = \frac{\ln\left(\frac{70 - 50}{70 - 66}\right)}{15 - 5}$$

$$k_L a = 0.1609 \text{ s}^{-1}$$

$$\text{Error} = -90\%$$

So, this is what has been done. So $k_L a$ will in turn become $\ln 78 - 50$ divided by $78 - 66$ divided by $15 - 5$, $t_2 - t_1$. So this is what is given here. So our $k_L a$ value can be obtained as 0.085 second inverse. Now let us see the second part. An error is made, I will just erase this so that the question is visible properly. The error is made determining the steady state oxygen levels instead of 78 , they have taken it as 70% by mistake.

Now we need to find what will be the intern percentage error in the resulting $k_L a$ value that should be doable. Now we know that how $k_L a$ is to be determined given the value of C_{L1} , one C_{L2} , t_2 and t_1 . Now the only change here which is being made is that in place of 78 , the steady state concentration measured incorrectly is 70 . So here it becomes $70 - 50$ divided by $70 - 66$ which comes out to be 0.1609 second inverse.

So, if the difference is calculated in terms of percentage it comes out to be 89 point something, so it has been shown as nearly 90%.

(Refer Slide Time: 09:33)

Problem 2

Serratia marcescens bacteria are used for production of threonine. The maximum specific oxygen uptake rate of *S. marcescens* in batch culture is $5 \text{ mmol O}_2 \text{ g}^{-1} \text{ h}^{-1}$. The bacteria are grown in a stirred fermenter to a cell density of 40 g/L ; $k_L a$ under these circumstances is 0.15 s^{-1} . At the fermenter operating temperature and pressure, the solubility of oxygen in the culture liquid is $8 \times 10^{-3} \text{ kg m}^{-3}$. Is the rate of cell metabolism limited by mass-transfer, or dependent solely on metabolic kinetics?

Handwritten notes in red ink:

$$q_{O_2} = 5 \text{ mmol O}_2 \text{ g}^{-1} \text{ h}^{-1}$$

$$X = 40 \text{ g/L}$$

$$k_L a = 0.15 \text{ s}^{-1}$$

$$C^* = 8 \times 10^{-3} \text{ kg/m}^3$$

Comparison: $OTR < OUR$ (with arrow pointing left)

Comparison: $OTR > OUR$ (with arrow pointing right)

Let us see the problem 2. Now *Serratia marcescens* bacteria are used for production of threonine okay. The maximum specific oxygen uptake rate of the bacteria in batch culture is given to us. So means this is nothing but our q_{O_2} the specific oxygen demand of the culture. The bacteria are grown in a stirred fermenter to a cell density of 40 grams per litre. So, we know that the biomass concentration to be achieved is 40 grams per litre.

Now the volumetric mass transfer coefficient in this system is being reported as 15 seconds inverse. So, this is our volumetric mass transfer coefficient. At the operating temperature and pressure, the maximum solubility of oxygen in the liquid is 8 mg per litres. So, this is nothing but our, correct, the saturation concentration of oxygen in the bulk. Now we need to determine if the rate of cell metabolism is limited by mass transfer or not?

Now in order to find that we need to first find or the rate of oxygen consumption due to the growth of the bacteria whether it matches up with the rate of oxygen transferred inside the reactor. So which means that only when $OTR = OUR$, then it is meeting the demand of the culture and if OTR becomes minimum, this is the minimum condition. If OTR becomes less than OUR which means that it will start becoming mass transfer limited. So, ideally it should be greater than OUR , then it is not mass transfer limited but the reaction limited.

(Refer Slide Time: 12:14)

rotational speed and with air being sparged at 0.5 l gas/l reactor volume-minute. E. coli with a q_{O_2} of 10mmol O_2 /g-dry wt-h are to be cultured. The critical dissolved oxygen concentration is

Solution

$$\begin{aligned} \text{Oxygen uptake rate (OUR)} &= 5 \text{ mmol } O_2 \text{ g}^{-1} \text{ h}^{-1} \\ &= \frac{5 \times 10^{-3} \times 32}{3600} \text{ s}^{-1} \\ &= 4.44 \times 10^{-5} \text{ s}^{-1} \\ \text{Cell density} &= 40 \text{ g/L} \end{aligned}$$

$q_{O_2} \times X = \text{OUR}$

Therefore, maximum oxygen requirement = $4.44 \times 10^{-5} \text{ s}^{-1} \times 40 \text{ g/L}$

$$\text{OUR} = 1.77 \times 10^{-3} \text{ kg m}^{-3} \text{ s}^{-1}$$

$$\text{Oxygen transfer rate (OTR)} = k_L a (C^*) = 0.15 \text{ s}^{-1} \times 8 \times 10^{-3} \text{ kg m}^{-3}$$

$$\text{OTR} = 1.2 \times 10^{-3} \text{ kg m}^{-3} \text{ s}^{-1}$$

$k_L a (C^* - 0)$
 $k_L a C^*$

As the oxygen demand is greater than the oxygen transfer rate, the system is limited by mass transfer.

So, let us find out the OUR first. So, q_{O_2} given to us and the cell density is also given. So, OUR will be nothing but q_{O_2} times X which is the biomass. Now we will make the units consistent. So here specific oxygen demand has been converted to millimole per gram per hour. So, this can be converted to hour inverse, so hour further has been converted into seconds, so this becomes second inverse.

So millimole of oxygen has been converted to grams, so into 10 by 3 is moles and 32 is for the oxygen, weight of oxygen per mole and your hour has been converted to seconds by multiplying it with 3600. So ultimately it comes out to be in terms of seconds inverse 4.44 into 10 to the power of -5 . Now cell density is grams per litre. So maximum oxygen uptake rate would be in terms of again they are converting it into kg per meter cube.

So this becomes 1.77 into 10 to the power of -3 kg per meter cube per second okay. Oxygen transfer rate now we need to find, so this was OUR. Now we need to find OTR. Maximum oxygen transfer rate can be what? So, in a reactor the maximum rate at which oxygen can be transferred will be dependent on the $k_L a$ and the concentration gradient which means the difference between the saturation, equilibrium concentration and the bulk concentration.

So maximum gradient can be when the bulk becomes 0, so your maximum OTR possible is $k_L a$ times C^* . Now C^* is given to us as 8 ppm and your $k_L a$ is 0.15 second inverse. So, this again gives us 1.2 into 10 to the power of -3 kg per meter cube per second. Now if you compare the OTR with the OUR value, so this is OUR and this is OTR, so OUR is 1.77

and this is 1.2. So, OUR is much higher than the OTR, which means that this system is going to be mass transfer limited, to be specific oxygen mass transfer limited system.

(Refer Slide Time: 15:15)

Problem 3

A value of $k_L a = 30 \text{ h}^{-1}$ has been determined for a fermentor at its maximum practical agitator rotational speed and with air being sparged at $0.5 \text{ l gas/l reactor volume-minute}$. *E. coli* with a q_{O_2} of $10 \text{ mmol O}_2/\text{g-dry wt-h}$ are to be cultured. The critical dissolved oxygen concentration is 0.2 mg/L . The solubility of oxygen from air in the fermentation broth is 7.3 mg/L at 30°C .

- What is the maximum concentration of *E. coli* accumulated in the system?
- What concentration could be maintained if pure oxygen was used to sparge the reactor?

Handwritten calculations:

$$OUR = q_{O_2} \cdot X$$

$$OTR = k_L a (C^* - C)$$

Given: $C^* = 7.3 \text{ mg/L}$, $C = 0.2 \text{ mg/L}$

Let us see the problem third now. A value of $k_L a$ is given as 30 hours inverse. So volumetric mass transfer coefficient of the system is known for a fermenter at its maximum practical agitator rotational speed with air being sparged at 0.5 vvm okay. So volumetric mass transfer coefficient under the operating conditions of maximum possible agitator 0.5 vvm of air sparging rate is 30 hours inverse. Now *E. coli* is grown in this fermenter.

It has a specific oxygen demand of 10 millimole O_2 per gram dry weight per hour okay. The critical dissolved oxygen concentration which means that oxygen concentration level below which fermentation cannot be done. So, the critical dissolved oxygen concentration is the minimum oxygen concentration bulk possible is 0.2 mg per litre okay. The solubility of oxygen from air in the fermentation broth is 7.3 mg per litre.

So, this is nothing but our saturation oxygen concentration value at the given temperature and pressure. We need to find out what is the maximum concentration of *E. coli* which can be obtained in this fermenter. So maximum biomass possible. So, we know the specific oxygen demand and let us assume X is that maximum biomass possible. So, this is OUR, oxygen uptake rate. So, this will become now the maximum oxygen uptake rate.

Now under any circumstances because the $k_L a$ is fixed, the sparging rate is fixed, so the maximum possible OTR we can find out which will be $k_L a$ times C^* where C^* will

become 0 then only this will become the maximum possible OTR given the $k_L a$ value and the C^* value. C^* is 7.3, $k_L a$ is given to us as 30 hours inverse. So now the maximum possible biomass which can be supported by this fermenter can be calculated by equating OUR and OTR and finding the value of biomass.

So, let us equate $q_{O_2} X = k_L a C^*$. So, your X maximum possible which can be supported can be calculated when you know $k_L a$, C^* and the specific oxygen demand.

(Refer Slide Time: 18:40)

(a)

$$\left(\frac{dC}{dt}\right) = k_L a (C^* - C) - q_{O_2} X$$

At steady state,

$$X = \frac{k_L a (C^* - C)}{q_{O_2}}$$

$$X = 30 \text{ hr}^{-1} (7.3 - 0.2) \text{ mg/L} \frac{1}{10 \text{ mmol g}^{-1} \text{ h}^{-1} \frac{\text{mmol}}{32 \text{ mg}}} = 0.67 \text{ g/L}$$

(b) Solubility of oxygen from air is 7.3 mg/L

$$\text{Using pure oxygen, } C^* = \frac{1 \text{ atm}}{0.21 \text{ atm}} \times 7.3 \text{ mg/L} = 34.8 \text{ mg/L}$$

$$X = 30 \text{ hr}^{-1} (34.8 - 0.2) \text{ mg/L} \frac{1}{10 \text{ mmol g}^{-1} \text{ h}^{-1} \frac{\text{mmol}}{32 \text{ mg}}} = 3.24 \text{ g/L}$$

So, this is what has been done possibly here see at steady state. So, you are making the units consistent. The biomass possible is found to be 0.67 grams per litre. Let us see the second part. What concentration could be maintained if pure oxygen was used to sparge the system? Okay, so we need to find that if pure oxygen is being sparged inside the system under the given operating conditions, then how much amount of biomass can be supported in that case?

So, the maximum amount of biomass which can be supported still remains the same $k_L a$ times C^* by q_{O_2} , but here what has changed is the C^* value. So, we need to first find the C^* value. So now we know by Henry's law we will use that equality. So, for 1 atmosphere where the mole fraction is now completely oxygen, otherwise it is 0.21 is equal to so this is 7.3 mg per litre, what will be the X value?

So, this is your saturation. So, your saturation concentration of pure oxygen can be calculated based on Henry's law as 34.8 mg per litre. So once the saturation concentration of oxygen in

the bulk, then the X by the same equality can be calculated as 3.24 grams per litre. So, as we increase the mole fraction of oxygen in the incoming air the biomass which can be supported by the same fermenter system under the given operating conditions increases.