Introduction to Dynamical Models in Biology
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Lecture 18
Modeling Molecular Processes - 3

Hello. Welcome to module 6 of third week of our course. In this course we will in this module, we will discuss about writing ODE based equation for a very common process in cell biology that is transcription and translation which ultimately controls the concentration of proteins inside a cell. So if I look into the steps, simplified steps involving transcription and translation, I can write a (sche) scheme that's shown here in the slide.

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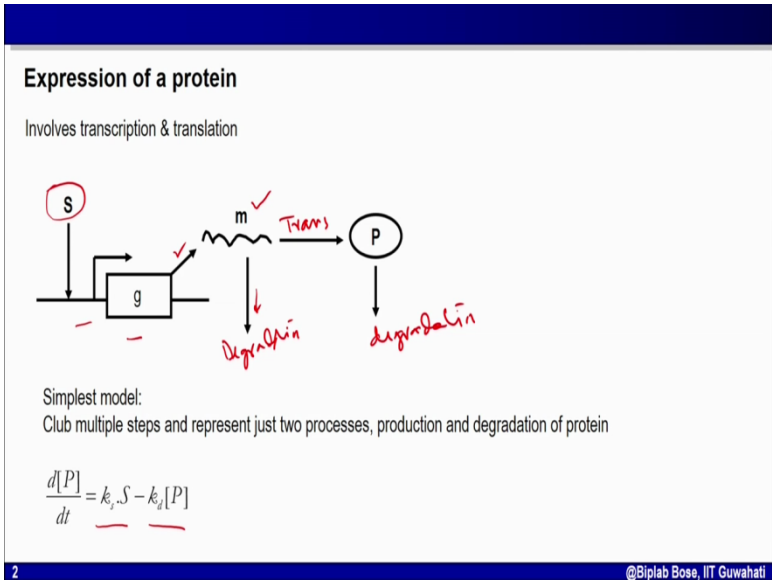

I have a gene G which is under control of the promoter. A transcription factor or external signal comes (a) activates this promoter and mRNA M is formed. Remember mRNA is formed by transcription shown by this arrow. Now as once the mRNA is formed, that can get degraded by this method so I have degradation here and while translation mRNA is getting (conver) is giving rise to protein P and protein P can also get degraded so this is a generalized scheme, very simplified.

You can have much more complications inside the whole thing. For example, you carry out thing, your promoter may be condensed, it may have to open and all these (issu) issues are there and for example, in case of translation, you may assume that ribosome comes, binds, scan the initiation side, all these things, we are not considering. What we have done, we have clubbed them together and we have (sim) created a simplified schematic representation here and we will try to write down the ODEs based on this.

Now if I don't require to separate transcription and translation as a separate entity, in some cases, I don't need. I just I am interested only in the dynamics of the protein and I am not much bothered about how the whole process is broken down in different parts. I can write a simple ODE representing the formation (an) the rate of change of protein concentration and we have seen it earlier that it will be equal to $Ks*S$ that is the rate constant of synthesis of the protein into S that is the signal which is inducing the production of the protein - $Kd*P$, this is the first order degradation of protein.

You have seen it in a earlier module. These are simplest to all, I will always recommend or try to use if I am not bothered about the detailed mechanism multiple steps involved in the production of the protein but in some cases, actually I have to bother about these multiple steps in or transcription and translation separately because maybe I want to explore the dynamics, in terms of the dynamics of external signal so remember this external signal or the transcription factor which goes and bind to the promoter and controls or induces or inhibits the expression of (transcri) transcription of a gene may have a particular dynamics and I want to know the input output relation in this transcription induction or inhibition.

So in that case, I cannot club transcription and translation together, I have to separate them out so I have to consider transcription as a separate process in which the input signal S or the (transcri) controlling transcription factor will have a role to play and translation as a separate process.

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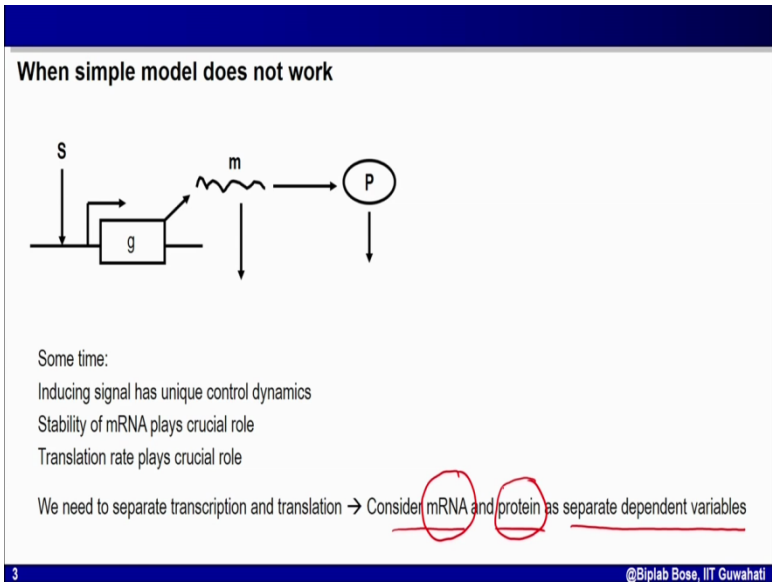

And many a times actually we forget that actually the degradation or stability of mRNA is very crucial in the dynamics of the protein as a whole, so suppose I want to study the effect of stability of mRNA on the dynamics of the protein concentration in the cell then I have to consider transcription and translation separate because I want mRNA as a dependant variable also.

So similarly if I want to look into the rate of translation as a separate feature, feature parameter and I want to study the effect of different rate of translation on the dynamics of the protein then I may separate out transcription and translation separately and I have to model it and write the ODE separately. So if I separate out transcription and translation then I will consider mRNA and protein as separate dependant variable so I will write 1 ODE for mRNA, 1 ODE for the protein so let us try to do that.

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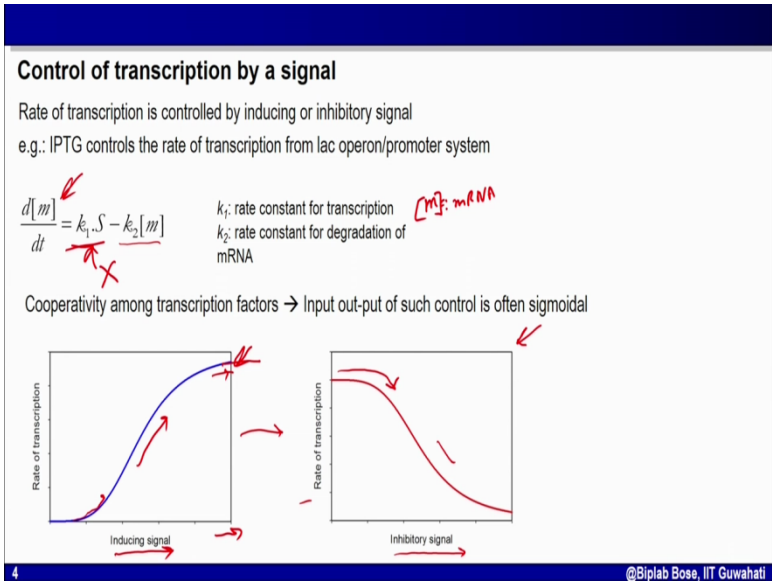

I will start with writing the ODE for the mRNA. Now mRNA concentration is controlled by transcription and degradation of the mRNA. Now transcription is again controlled by the signal which may be itself a transcription factor or its molecule is goes and activates the transcription factor so that transcription factor or inducing signal um will control the rate of synthesis of mRNA. For inhibitory signal, it will also control the rate of synthesis of the mRNA so that means I have to incorporate this signal into the ODE for the mRNA and for example, I have written here, it is a very common thing.

You add IPTG in your bacterial culture where the bacteria is carrying a plasmid having a Lac Operon and and controlling the expression of a gene so you vary the concentration of IPTG and the amount of the gene transcribed changes so I can write a simple ODE, the way I have shown here. $\frac{d[M]}{dt}$ is equal to that is the M is concentration of mRNA, M is a concentration of mRNA so the change in rate of change of concentration of mRNA is equal to $K1*[S]$, K1 is the rate constant for transcription, S is the signal. For example, concentration of ITPG added to the culture minus $K2*[M]$, K2 is the rate constant for degradation, M is the concentration of free (conen) free mRNA so this is the first order degradation so the first part is synthesis by transcription. Second part is degradation.

Now see the first part. The (sa) transcription part. K1* S so that means if I keep on increasing S linearly then the rate will also increase linearly but the fact is, in many experimental system, it has been seen that his relation of rate of transcription with the amount of input signal is actually not linear, rather it is nonlinear and particularly sigmoidal, S shaped, as have shown here in this graphs. In many experiments, for example an experiment involving IPTG as a external signal for inducing the expression, when inducing signal is increased, the rate of transcription does not increase linearly rather follow a sigmoidal path as I have shown and eventually saturates, it does not increase further, if you increase signal further, it will still remain here at the saturation so it is a nonlinear behavior.

Similar has been observed from many transcription factor molecule, factor which goes and inhibit transcription of a particular gene where inhibitory signal if you increase, the rate of transcription does not decrease linearly rather non linearly in S shaped sigmoidal fashion and it is just reverse of the previous one. So that means I have a nonlinear relation between signal S and the rate of transcription and it is now, well understood that in many cases, these nonlinear relation, this sigmoidal behavior arises because transcription factor works with cooperativity.

One transcription factor does not work alone. It binds with multiple copy of the same transcription factor, then make a large complex and that complex is functional. Many a time, not just 1 molecule is involved, 1 transcription factor goes, attach to the promoter and then it will recruit other transcription factor to create a large complex. Unless and until this large complex is formed, the effect of the transcription factor is not manifested. So it is a cooperative phenomena and as I have cooperative phenomena, I have this sigmoidal behavior. Now that means if I have a system where transcription rate is have a (rela) sigmoidal relation with input signal, I cannot use this simple linear relation say of $K1*[S]$. So how can I modify this equation to reflect the sigmoidal behavior that people are observing in experiments.

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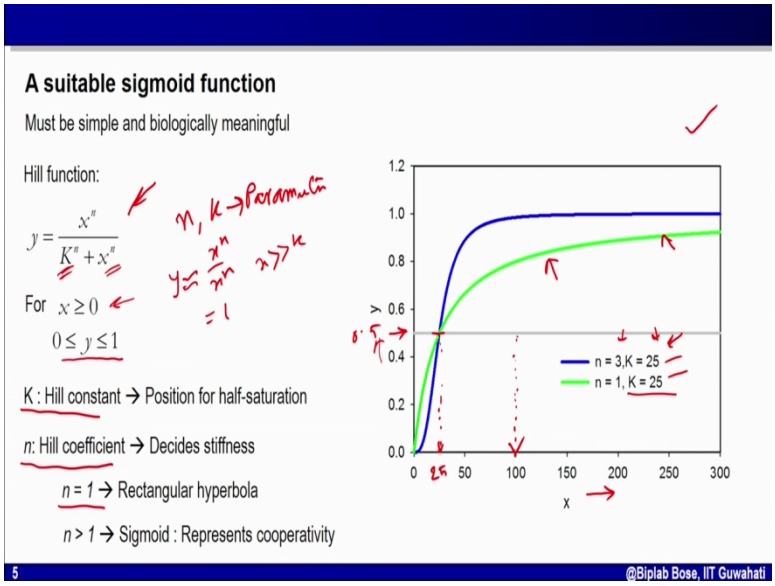

Now remember different sigmoidal function, you can write mathematically, large number of them. You can (im) make a imagination and write a function which will have sigmoidal input output behavior but we have to choose a function which will be sigmoidal in behavior, at the same time, it should be very simple and has some biological meaning and in case of modeling biological system, we very frequently use Hill function to represent a sigmoid function because it has very simple and it has less number of parameters and those parameters can be actually determined from experiments.

So let us look into what it Hill function. Hill function is nothing Y that may be output is X^n/K^n , K is again a parameter and X^n , n and K are parameters. It means these are constants and I have to get those values, either I have to guess them or from experimental data, I have to get them. If you look into this function, you can easily see if X varies from 0 to any higher (val) any positive value, Y will increase initially. When X=0, obviously $Y=0$ but as you keep on increasing X, Y increases but eventually when X becomes very large with respect to K, this Y becomes nothing but equivalent to $\frac{X^n}{X^n}$ because X has now become very large with respect to K so this becomes 1.

So that means this function Y changes from 0 to 1, 1 is the maximum value and that helps because it is within a particular range and if you look into diagram here, shown in your your slide, I have varied X in a positive value from 0 to 300 and Y in the vertical axis. You can easily see this blue curve is a sigmoidal one. Here I have considered N = 3 and K = 25 so now, look into this parameter K and n, what they are. Before I move into, I will (rem) remind you that you may have seen this type of Hill function in biochemistry textbooks where hemoglobin has been discussed. Hemoglobin has multiple subunit and work (cooperati) they have cooperativity and the partial pressure, the oxygen saturation of hemoglobin has a sigmoidal behavior and Hill function is used there to represent that sigmoidal behavior, you can look into that.

Now let us go back to the parameters K and N. K is called Hill constant and it represent the position of half saturation. Let us look into that. I will change the value of K from 25 to something else say suppose 100 then I get this pink curve. This gray line is representing 50%, 0.5 so it is intersecting this blue as this value which is nothing but 25 and this gray line is intersecting the pink line at 100 so that is value of K so 0.5 is half saturation because the maximum value is 1 so half saturation is 0.5 so and K represent where I reach that half saturation value and you can easily imagine that by experiment, I can actually measure this half (satur) half saturation value that means I can measure the amount of the value of X for which the value of Y will be 0.5, the half saturation and that is my K and this K is called Hill constant.

N is interesting parameter. N is called Hill coefficient and it decides how stiff this S type figure would be, how stiff this sigmoid curve would be. If you take N = 1, I don't get much stiffness, I get a rectangular hyperbola type thing. Let us take the value of N. What I have done here, remember K is same in both the curves, that is 25. So half saturation is at 25 but I have changed in (fro) in blue, N is 3 and in the green one, N is 1 so I have reduced the value and taken the value of N, the Hill coefficient to 1 and you can see the curve has lost its sigmoid behavior and it has become a rectangular hyperbola.

And in fact, it will be like that till your you have N equal to 1 and if you have N greater than 1 then I start getting the sigmoidal behavior so Hill coefficient tells me whether the system has sigmoidal nature or not and if it is sigmoidal, how stiff that sigmoid is whereas K tells me that

how sharp this sigmoid rise. Is it rising very fast with respect to the value of X because it determine the half saturation at which we reach the half of the value of Y.

So using these 2 parameters, K and N, I can actually manipulate the Hill function and get different sigmoidal behavior and that helps helps me in my modeling so I have a simple function,

Y = $\frac{X^n}{K^n + X^n}$ and just 2 parameter, I can measure this parameter from experimental data and it gives this function depending upon the value of N and K, give me large number of sigmoidal behavior.

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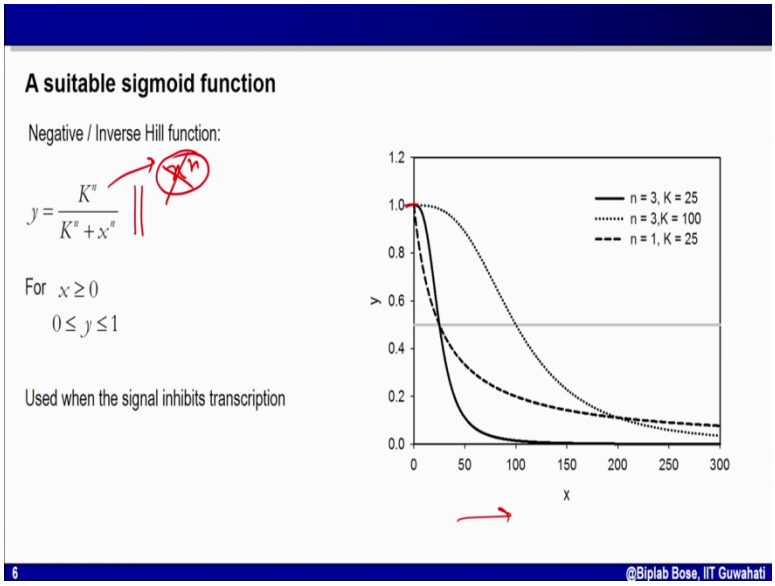

Now remember for induction, it is sigmoidal rise as inducing signal increases, you have sigmoidal rise in the output, the rate of transcription but for inhibitory signal, the rate of transcription drops so I require another function, I can actually tweak the Hill function and get this inverse Hill function where you don't have X^n on the (denomin) numerator, you don't have it so rather you have K^n, that is a Hill constant to the power N divided by the same numerator, $K^n + X^n$. Again here, when X = 0, Y will be 1 because that's what you want when there is no X maximum value and then as you increase X from 0, Y start dropping and it reaches towards 0. So this inverse of negative Hill function when I will have a inhibitory signal. The transcription factor I have in a system which goes and inhibit the process.

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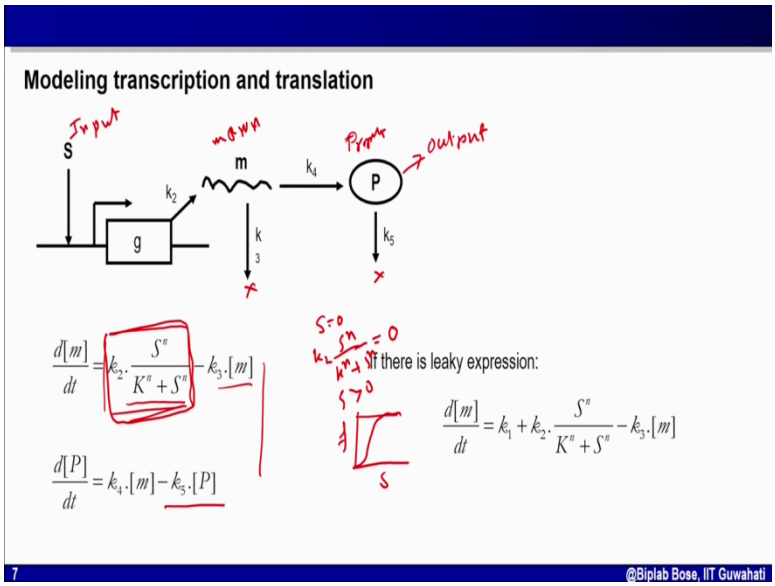

So once I have these Hill functions, inverse and the normal Hill function in hand. Let us try to model a transcription translation unit using this Hill function. So what I have the scheme. S is the signal, this I can consider as the input which gives the positive induction for transcription of the gene G and I get that mRNA in which get degraded and which may get translated by ribosome forming protein P and this is my output so both mRNA and protein can get degraded so they are getting degraded here and I want to write ODE for both M and for P because I am considering them a separate entities, separate dependent variable. So now I will use Hill function.

So $\frac{d[M]}{dt}$, rate of change in concentration of M is equal to K2 into the Hill function here where S is the signal into $K^n + S^n$. Look into this part very carefully. When S=0, $K\frac{2 * S^n}{K^n + S^n}$ will become 0. When S > 0 then this function will have this behavior, sigmoidal behavior depending upon S so this function has the S so that means as S will increase, this whole term will also increase and reach a maximum value K2 because the Hill function has a maximum value 1 so 1 into K2 is K2 so initially when S= 0, K2 * 0 so rate is 0, no transcription.

As you increase S, $\frac{S^n}{K^n+S^n}$, this Hill function will move towards 1 depending (on) on the value of S in a sigmoidal fashion so when I am multiplying that with K2, the overall rate of transcription is also behaving sigmoidally and reaching the maximum value K2 at the maximum value of F, that is the Hill function so this rate of transcription has a sigmoidal behavior now with respect to S and K3 * N is nothing but first order degradation of the mRNA.

Let us look into the protein part so $\frac{d[P]}{dt}$ is the rate of change in concentration of protein will be equal to now K4 *[M], M is the amount or concentration of mRNA present so obviously, translation will depend upon how many copies of the mRNA is present at that time for that particular protein so it is K4*[M], first order reaction - K3 (in) K5 * P, that is a first order degradation of the protein so I have a simple 2 ODEs. The first thing only has this Hill function and the transcription rate control. Now we have to remember 1 critical issue here.

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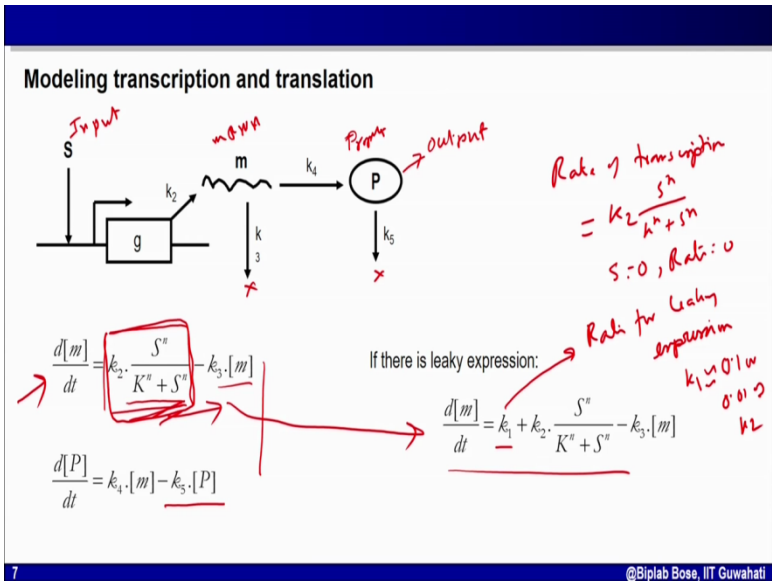

Let us clean this part a bit so that you can see it clearly. If I consider the rate of transcription is equal to nothing but $K\frac{2*S^n}{K^n+S^n}$, the way I have done here then when S = 0, rate is also 0. That means when there is no signal, there is no production of the mRNA for this gene so if there is no

mRNA, there has been no protein production but that may not be true in many cases. In fact for a IPTG based system also, even in absence of IPTG, there is some amount of transcription from Lac Operon and that transcription and translation of the protein is actually required to run the Lac Operon system.

So in some time, even in absence of the inducing signal, there is a low but steady expression of the mRNA. We call it leaky expression and sometimes for the physiology or for the dynamics of the whole system, actually that leaky expression is required so how can I represent it because the ODE that I have written here does not incorporate that because when S = 0, rate of transcription will be 0, there will be no formation of the mRNA so there will be no formation of the (prot) protein also. I can simply change this equation by adding a constant term K1.

K1 is the rate constant, rate for leaky expression so when S will be 0 then this Hill function will become 0 so K2 into Hill function will be 0 but K1 which may have a very low value, it will give me rise of mRNA, it will keep on producing mRNA, usually we take K1 as around 0.1 or 0.01 of K2 so 10% of or 1% of the maximum rate of transcription that can happen by induction. You take a very small value of K2 with respect to K1 with respect to K2. So I can simply change this ODE to this one and incorporate K1 so my rate of change of M that is mRNA, $\frac{d[M]}{dt}$ becomes K1 +K2 into the Hill function involving [S] -K3 *[M].

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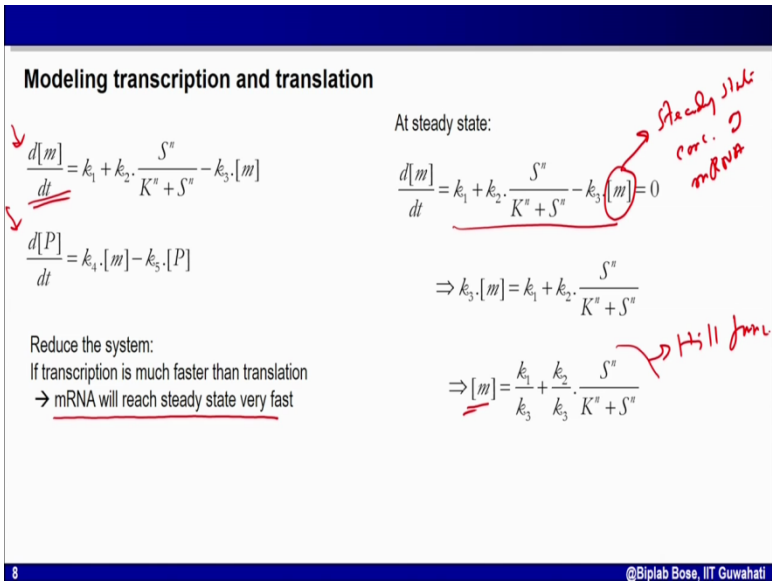

So if I have these 2 now, the first one considering leaky expression and the second ODE is for my protein so if I have this, I have 2 ODEs. Now imagine I have a cascade of transcriptional network, it means 1 protein is (transcrib) produced and then that protein acts as a transcription factor for the next protein and induces the production of that protein. That protein also works as a transcription factor and control expression of another protein so if have multiple transcription translation unit in my model then for each case, I have write these 2 ODE for 1 for mRNA, another for the protein and that may complicate my calculations so I will try to reduce my ODE.

So how can I reduce my number of ODE for a transcription or translation system? That's what I will discuss now. We all know that usually transcription is a faster process than translation particularly in case of eukaryotic because transcription happens inside the nucleus and then the nucleus has to get it transported outside the nucleus into the cytoplasm where the ribosome will buy and many other processing of mRNA will happen before that.

So if I don't consider nucleus and cytoplasm as a separate chamber, separate compartment and as a whole I consider a cell as single unit then I can consider that the production of proteins is bit slower with respect to transcription so I can safely consider that before the protein production reaches the steady state, mRNA production reaches a steady state because mRNA is produced in a faster rate, it also gets degraded so I reach steady state. So what I can do, I can assume that if transcription is much faster than translation, mRNA will reach steady state very fast. That means

I can consider this $\frac{d[M]}{dt}=0$ so let us do that so I can consider steady state for the mRNA so I can put $\frac{d[M]}{dt}=0$.

That means this whole relation K1 + K2 into the Hill function $-K3*M=0$ so this M is the steady state concentration of my mRNA so I want to separate that out from this algebraic relation so I keep K3 and M in left hand side and keep everything else on the right hand side further I can divide both sides by K3. Then I get the steady state concentration of M, this is my steady state concentration M is equal to $K1/K3$ considering that I have leaky expression plus $\frac{K2}{K3}$ into the Hill function, $\frac{S^n}{K^n+S^n}$, , this is my Hill function.

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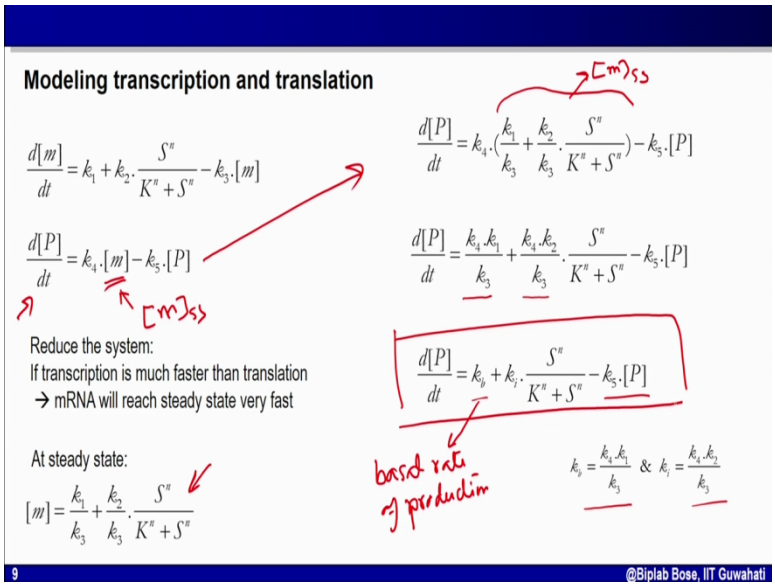

So now I can rewrite the whole thing because I have reduced 1 ODE, I do not require the ODE for (di) for the mRNA because I have considered that it has reached a steady state, I am left with only the ODE of ODE of protein so $\frac{d[P]}{dt}$ and I will place this value of N here, replace this value of N with the steady state value of N, that's what I have calculated just now using this relation. So I will replace that so I can write from this relation, I can write $\frac{d[P]}{dt} = K4$, that's the rate of (translate) rate constant for translation into this is the steady state concentration of steady state that just we have calculated, $\frac{K1}{K3} + \frac{K2}{K3}$ into the Hill function - K5 * P.

Now I will club multiple parameters together, I will take K4, K1 and K3 together. K4, K2, K3 together, I can replace this one by a new parameter, I can replace this one with a new parameter and I get this relation. $\frac{d[P]}{dt} = ¿$ KB that is the basal rate of production, this is my basal rate of production plus KI, that is the rate constant (max) for (indu) inducible production of the protein, KI into the Hill function involving the signal S that is $\frac{S^n}{K^n}$, K is the Hill constant + $S^n - ¿$

K5P that is the first order degradation. I have written down spelled out here $Kb = \frac{K4 * K1}{K3}$

whereas KI is $K\frac{4 * K2}{K3}$ so I have only 1 ODE now.

So this ODE will give me the dynamics of the protein when the input signal is S and the input output relation at a transcriptional level has a sigmoidal behavior.

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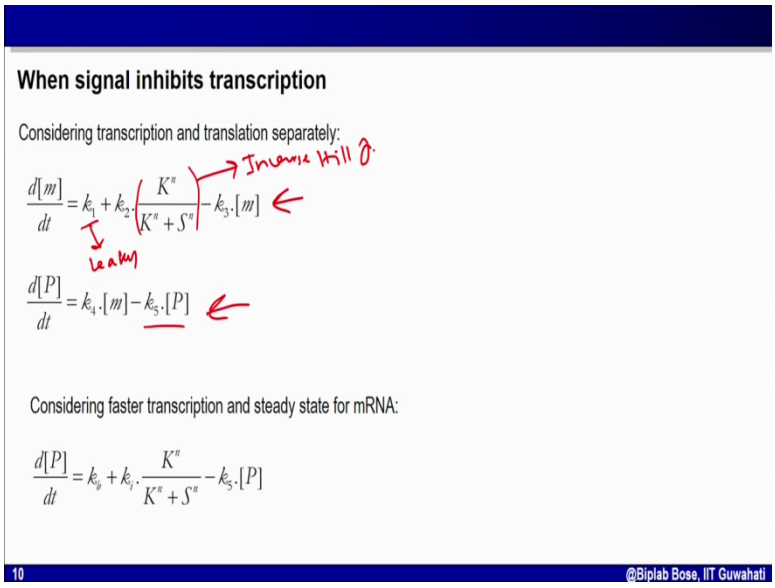

Now suppose the signal inhibits my production, inhibits my transcription then I have to use the inverse or negative Hill function and I can write the ODE the way I have shown here. $\frac{d[M]}{dt} = K1$ that is the leaky expression, rate of leaky expression plus K2 into the inverse Hill function here or negative Hill function here minus K3 * M, K3 * M is nothing but your rate of first order degradation of mRNA and the second ODE that is shown here is K4 * M, that is (bi) the translation of, the rate of translation of the protein and depends upon the concentration of mRNA present minus degradation of the protein K5 * P.

Now just like the previous one, here also I can consider the transcription is much faster than translation and the transcript, the mRNA reaches the steady state much before the protein reaches

the steady state so I can consider this $\frac{d[M]}{dt}=0$, $\frac{d[M]}{dt}=0$ and I can replace M steady state value with some algebraic relation from this first ODE and put that one in the second one and I can get just I have (done) the way I have got got in the last slide, $\frac{d[P]}{dt}$, that is the rate of change in protein concentration equal to KB, this is the basal rate of production plus KI into the Hill function inverse Hill function or negative Hill function in this case minus K5 * P.

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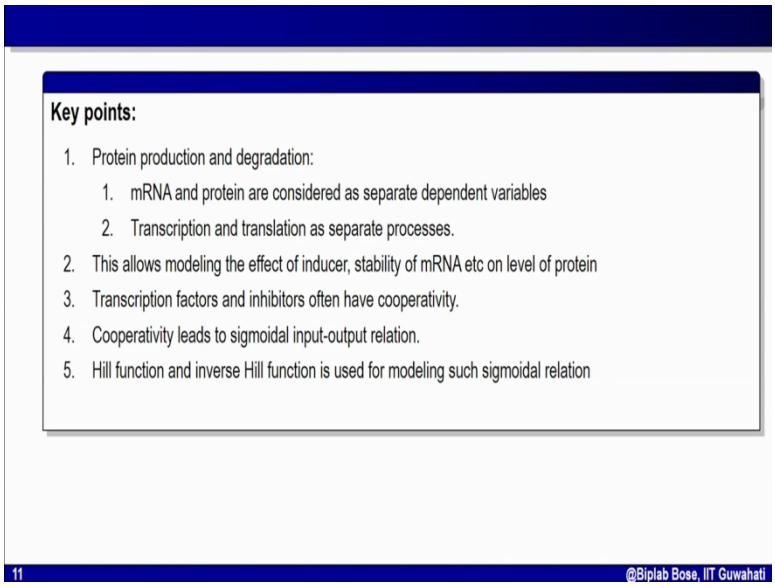

So if I jot down what we have discussed in this module, we have discussed that the protein production and degradation, usually we may consider transcription (tra) we may not consider transcription or translation separately, we may club down together in a single process but some time, we have to separate them, just consider them as a separate process and in those cases, I have to consider mRNA and protein as separate dependent variable that means I will have 2 different ODEs for them and for these, I have considered transcription and translation as separate process.

Now if I do that, it allows me to investigate role of different inducing and inhibitory signal in transcriptional process. It also helps me to understand the effect of stability of mRNA on the level of protein, all these questions can be approached. Now we have to remember in most of the

cases, transcription factors or inhibitors work with cooperativity so if you have cooperativity, this cooperativity sometime leads to a sigmoidal input output relation so input output is not linear here but non linear and a S shaped and S saturate as a higher value.

If you have that type of behavior, if experimental data is telling you that you have a sigmoidal input output relation for the transcription factor involved in this system, you can use Hill function which is very simple or a inverse Hill function to model this type of sigmoidal behavior and remember Hill function is defined by 2 parameters, 1 is called Hill constant which represent the half saturation value where I get the half saturation and the Hill coefficient N which tells me whether I have the sigmoid (S) or the S shape in the curve or the sharpness of rise of the curve.

If N is equal to 1, I get a rectangular hyperbola, I don't have a sigmoidal behavior. That means I don't have any cooperativity in the system. When you have cooperativity in the system, when the transcription factors work with cooperativity, you will have sigmoidal nature and N will be greater than 1. That's all for module, for this module. Thank you for watching it.