Computational Systems Biology
Karthik Raman
Department of Biotechnology
Indian Institute of Technology-Madras

Lecture – 81
Constraint-based Modelling of Metabolic Networks: Recap

So, this last recap video for constraint based modeling we will study FSEOF which is flux scanning based on enforced object of flux and we would also look at we will also study the concept of synthetic lethals. And how we can you know simulate or predict synthetic lethals using constraint based approaches and algorithm such as fast SL.

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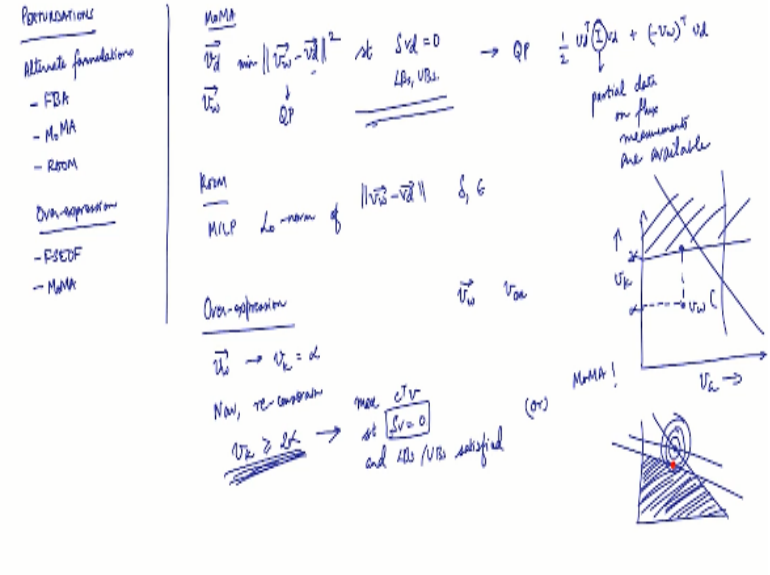

What is the other important concept in over expression we studied FSEOF flux scanning using and enforced objective flux or function?

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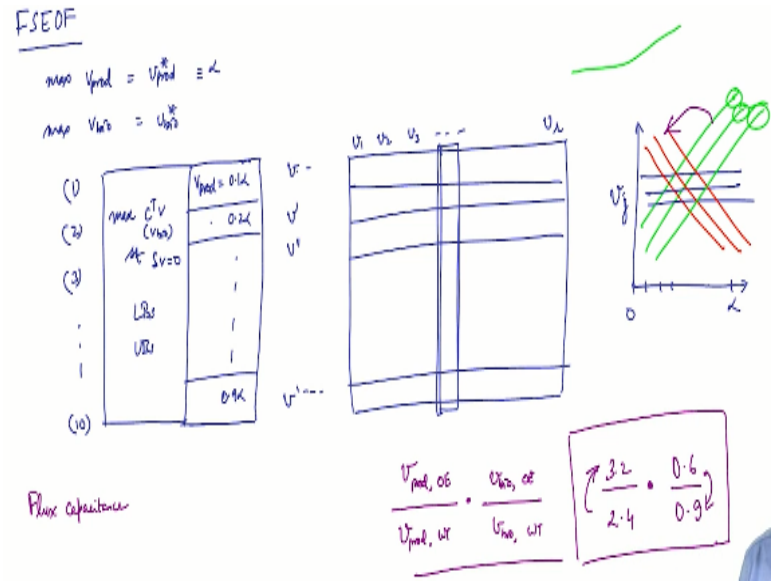

So, what was FSEOF I have not shown you the recent work from our lab we used FSEOF twice with great success in two different system L Lactose and Sunflower and how does FSEOF work Let us say your first maximize v prod you get some v prod star which is alpha here for the moment. Right and then you maximize bio mass usual v bio star now you run a serious of simulations. 1 2 3 v10 right where in you set up right part of the problem is the same.

This is nothing but maximize c transpose v or which is basically v bio such that sv=0 LBS UBS and additionally we v prod=0.1 alpha 0.2 alpha 0.9 alpha. I do not know where you can pull alpha may be bio mass is 0. Right there are cases where you know of product as maximum biomass might basically vanish. So, you perform the simulation so you have these additional constraints and now you will set some v v dash v double dash v several dashes.

Right all these flux distributions you now analyze. Right so how do they look or so this is these are basically going to vectors right with values for v1 v2 v3 vr z. Right and you are going to have 10 or 9 values for these. Now take every column that is here and you plot it. Alpha or let us call it some this will be .1 .2 alpha .3 alpha 0 to alpha right and you plot the flux of v1 or vj what all you like to observe.

You can observe many things you might observe reactions do not care about do not agnostic about any changes in alpha. Any changes in an enforced product flux. You keep increasing

product flux the reaction does not bend does not change there are other reactions that might always decrease. There are other reactions that might always increase right so which of these reactions are now interesting for metabolic engineering.

“Professor - student conversation starts” so, the ones that increase right “**Professor - student conversation ends.”** The reactions that increase that too monotonically right some might you know go like this. There are reactions that can also go like this potentially unlikely but it can happen. Right so the reactions that increase are basically the roads that are adverting more traffic.

So, that the final traffic has improved final traffic has increased. Right so by reversing that argument if I widen these roads. If I over express these reactions these genes, I would be able to support more flux in my product. So, that is the underlying assumption of FSEOF and that is how it works reasonably well in practice. And how would you quantify the improvement you can compute a fraction.

Which says v product over expressed/v product wild type*v bio over expressed/v bio wild type. So, you might get something like let us say 3.2/2.4*0.6/0.9. Right so this might be what you get for example right there is some decrease in biomass and some increase in product flux. So, you can rank the different genes based on this fraction **“Professor - student conversation starts”** biomass flux will decrease in product will increase.

Yes, so even there is an interest we are using that only you know how to increase yeah fair enough but you want to find you want to get the maximum. So, what does the say if I want to have this quantity as high as possible or this quantity it says that I want the maximum gain in product for minimum loss in bio mass. Which is fair explaining the graph you told us that any increase decrease in lines.

So, decrease in lines are not helpful so I am not going to overexpress them and so on. So, they are interesting for example one way is can you delete those out right “**Professor - student conversation ends.”** Or can you in fact one interesting application you should go and read about this it is called flux capacitance the idea is very cool you have a metabolic network. Right can

you build bridges or bypasses in that network to improve product flux.

This may involve heterologe expression of new reactions or you know you need to copy paste reactions or genes from some other organism. But if you build a bypass which happens all the time right in the city. So, you do try to build a bypass or a subway or a fly over which will help us ease traffic right the same day can you improve traffic by building these bypasses so one interesting they way to build a bypass.

That we are actually looking at this can take these reactions and plug them here. There is a reaction that is sucking away lycopene and there are reactions that are supporting lycopene. So, can you take the reaction which is taking away lycopene it is not taking away lycopene it is probably taking a pre-cursive to lycopene by pyruvate or G3p or something. Can you reroute that towards the lycopene production?

It should right because the reaction is actually taking away some pre cursive if you somehow already connected back towards you know producing some other pre cursive **"Professor - student conversation starts."** Yeah you cannot basically take the same reaction and put it right "**Professor - student conversation ends** "so so that is basically a giving b and b is taking of away something from lycopene.

You now convert b to d or something which is a reaction which goes towards like lycopene production d likes lycopene but b does not like lycopene in some sense. So, you now connect b and d. **"Professor - student conversation starts"** (()) (09:02) it depends so but you so you have to simulate all of those and figured it out. You obviously do not want any lethal or any of those things.

So, very well you will find some product forming pathway I mean some biomass central metabolism pathway and so on there right because if it is an on growth associated metabolism that is very likely to happen "**Professor - student conversation ends."**

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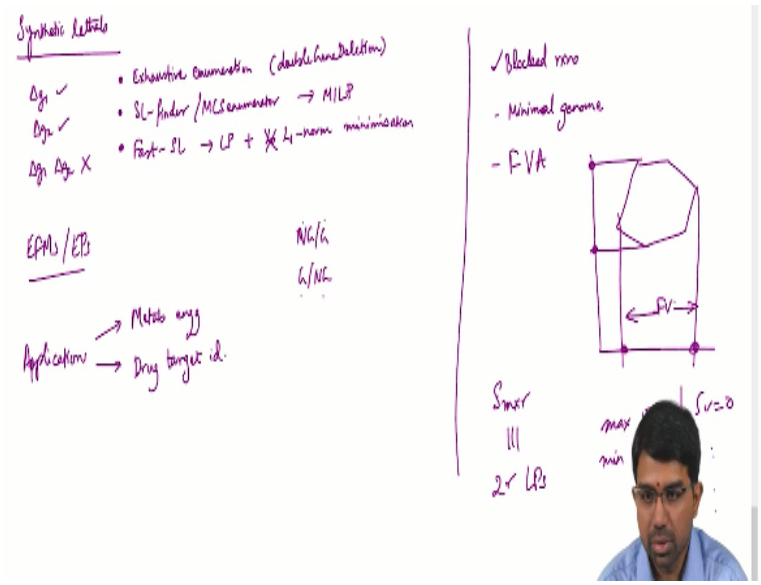

And then we look at the concept of synthetic lethals basically these reactions are if you remove 1 nothing happens if you remove second nothing happens you do 2 genes you have a lethal phenotype. Same thing you can apply to reactions to higher orders and so on and we looked at different methods to do this. The obvious easy way was exhaustive enumeration double gene deletion and cobra.

Better ways are SL finder or MCS enumerator but both use MILP the last thing we saw was fast SL which is basically LP+L0 or actually L1 norm minimization you can use L0 but it is slower. So, we said that any reaction which carries no flux in the beginning is not going to be a lethal reaction we will eliminate those and so on and so forth. Lastly we saw several applications of flux balance analysis and we also looked at elementary nodes.

So, the other thing to remember is EFMs and Eps applications mostly are in metabolic engineering drug target identification. So, these were the major concepts that we looked at in constraint based modeling. A bunch of additional important concepts we quickly looked at were concept of blocked reactions to some extent the minimal genome. We will try to do that again in the lab in one of the later classes.

Then we looked at flux variability analysis so flux variability analysis essentially if this is your flux cone this is your flux cone this tells you the minimum value for and the maximum value for

the fluxes. This is flux variability so flux variability analysis basically assists so if you have a stoichiometry matrix m cross r how many LPs do you need to solve to compute flux variability how would you compute this point max v1 mean v1.

All the time such that sv=0 etc etc so you will have 2r LPs that we need to solve right this will also tell you all the block reactions and so on and remember of course very importantly flux balance analysis gives you an insight into the metabolic capabilities. It does not consider regulation although we saw methods like RFBA or reflux to include regulatory and expression data and there are limitations.

Right so there are cases where you may be predicting false positives false negatives and so on and we also so you remember we studied no growth growth no growth examples and so on. Right exponent predicts as no growth but in model you see growth or exponential there is growth and in model you see there is no growth. And there are a bunch of reasons why this could have happened.

This is essentially the whole set of things that we saw under constrained based modelling. So, let us wrap up constrained based modeling with a lab in the evening.

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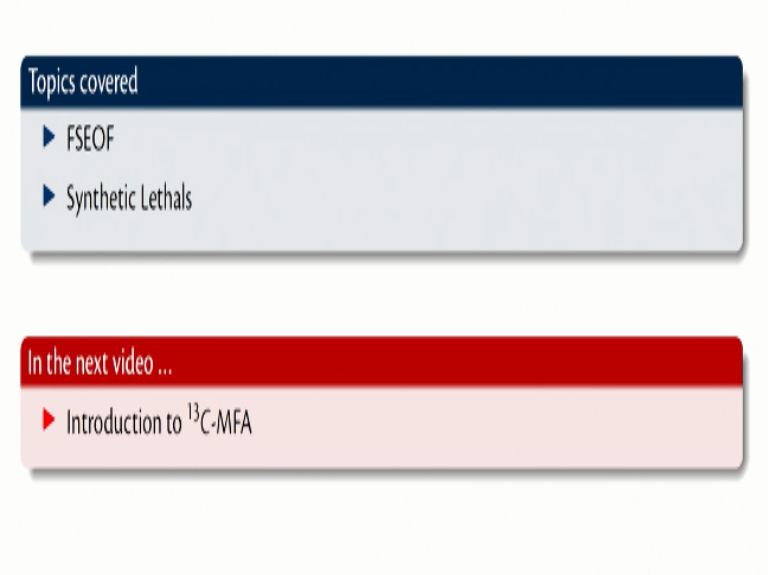

With this video we have nearly come to the end of constrained based modeling. So, I have

recapped all the important concepts and to cap of today have a looked at FSEOF and synthetic lethals. In the next few videos I have another guest lecturer Arthi Ravikrishnan who is my PHD student who will talk to you about how one performs C13 metabolic flux analysis on how one integrated with experiments it GCMS and so on.

To predict and quantify internal fluxes in a metabolic network right so there are many internal fluxes in a metabolic network that cannot be uniquely assigned or you know easily measured without using labelled substrates and that is what we will be discussing in the next few lectures.