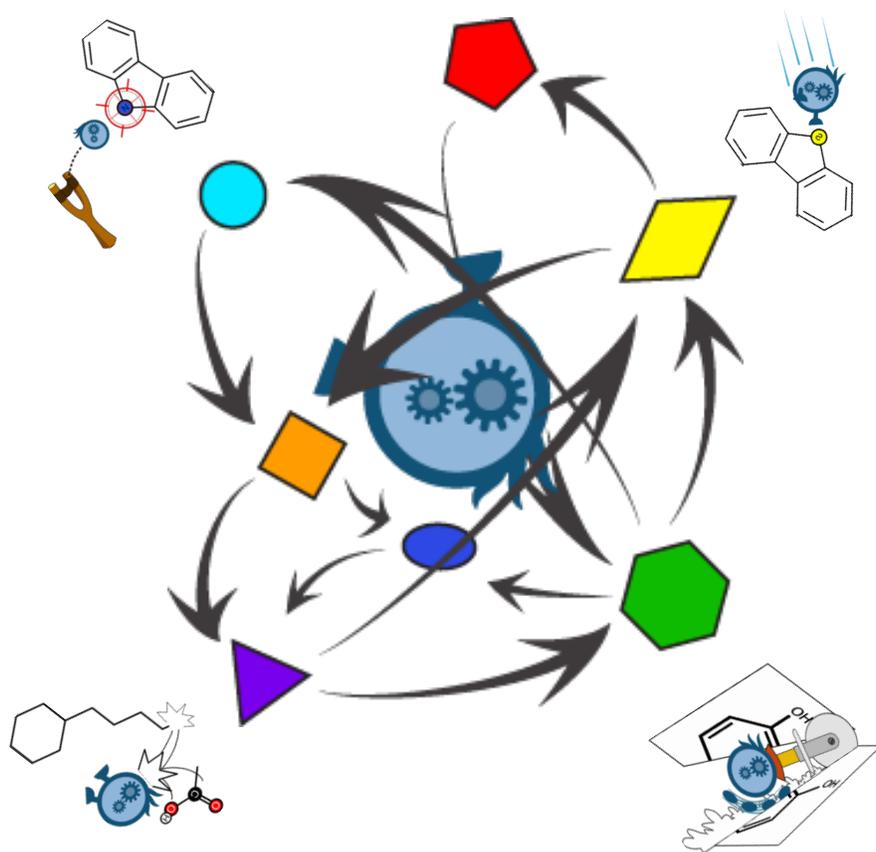


OSCAR OPTIMIZER 1.0

MANUAL



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About

Flux-variability analysis (FVA) was applied to optimize the bioreactor system of OSCAR for the newly incorporated metabolic pathways ([Decarboxylation](#), [Decatecholization](#), [Desulfurization](#), and [Denitrogenation](#)). FVA combines the framework of metabolic pathways with experimental enzymatic data to provide a computational platform for predicting what changes in metabolite levels will result in increased end-product. We developed a model using [MATLAB](#) computer language for predicting what metabolites could be added to your growth media to increase production of hydrocarbons in the *E. coli* chassis. We also created a graphical user interface to make the FVA program user-friendly and to allow current and future iGEM teams/scientists to input their own synthetic pathways into the FVA for analysis. Finally, we validated this model in the wet-lab by optimizing the Petro Brick ([BBa_K590025](#)) system to increase hydrocarbon production, thereby saving time, and resources.

What Are We Trying To Model?

It models the flux rate of metabolic pathways responding to different growth media conditions and generate an optimal set of metabolites that should be added to growth media in order to improve production rate.

How Could Systems Benefit From the Model?

Same as chemical reactions need optimal environmental conditions to achieve maximum production rate, microbes also require optimized growth conditions to accomplish their tasks in maximum speed. In microbiological bioreactor systems the conditions of growth media is much more crucial than in chemical synthesis reactors. If a model can predict an optimal set of metabolites that need to be added into media, this will save time, resources, and funds.

How Does The Program Work?

By running flux variability analysis, the program will give an output correlated to an optimized growth rate. By running the FVA algorithm at optimal biomass flux rates you can get a range of output for your product of interest. This allows you to determine an optimum point where compound production is maximized. Finally, the program will analysis the data with an algorithm to generate a set of media compounds that is expected to accelerate production rate.

For more information, see our [page](#).

Setup

SBML Toolbox setup

1. Download SBML Toolbox from [HERE](#) or <http://sbml.org/Software/SBMLToolbox>
2. Unzip the toolbox file
3. Install:
 - a. Start MATLAB/Octave
 - b. Navigate to the SBMLToolbox/toolbox directory
 - c. Run the install.m script found in that directory
 - i. Type install in MatLab console

Cobra Toolbox setup

1. Download SBML Toolbox from [HERE](#) or <http://opencobra.sourceforge.net/openCOBRA/Welcome.html>
2. Unzip the toolbox file
3. Install:
 - a. Start MATLAB
 - b. Navigate to the Cobra folder Path
 - c. Run initCobraToolbox
 - i. Type initCobraToolbox in MatLab console
 - d. Add the COBRA Toolbox directories to your MATLAB path
 - e. Set the solver
 - i. Type changeCobraSolver in MatLab console
 - f. Type testAll in MatLab console to verify installation

TOMLAB Optimization setup (Option)

1. Download TomLab solver [HERE](#) or <http://tomopt.com/tomlab/download/manuals.php>
2. Install follows the instruction from its manual.

Gurobi Optimizer setup (Option, is chosen to use for iGEM project)

1. Download Gurobi Optimizer [HERE](#) or <http://www.gurobi.com/download/gurobi-optimizer>
2. Install the optimizer
3. Query license (free trail or free academic)
4. Download Gurobi Mex to use Optimizer in MATLAB http://www.convexoptimization.com/wikimization/index.php/Gurobi_Mex:_A_MATLAB_interface_for_Gurobi#Install_Gurobi_Mex_in_MATLAB
5. Configure Gurobi Mex with instruction from its website

OSCAR OPTIMIZER setup

1. Download OSCAR optimizer from [HERE](#) or http://2012.igem.org/wiki/images/5/50/UCalgary2012_OSCAR_optimizer_v1.zip
2. Unzip the optimizer file
3. Add customized Cobra code:
 - a. Copy files in:
OSCAR_optimizer_v1/ Code/ CobraToolboxCustomizedCode
 - b. Paste files to:
<Cobra> folder
4. Install & run:
 - a. Start MATLAB
 - b. Navigate to the OSCAR_optimizer_v1 / Code
 - c. Type FVAapp in MatLab console to run the application

Basic

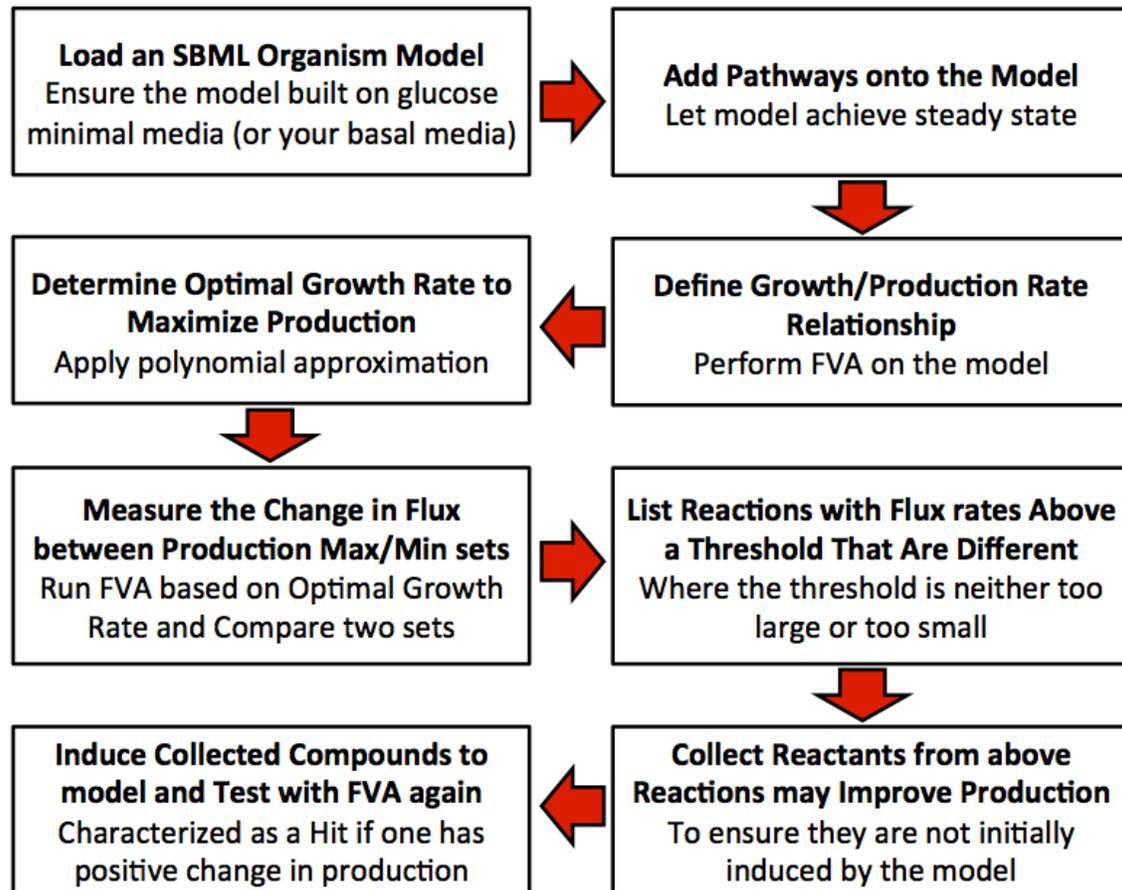
Pathway build tool

The pathway build tool is an interface to help users to reconstruct the original SBML model. Users can create interested pathways and add them into the model via this interface easily.

Pathway analysis tool

The pathway analysis tool is an interface to help users to analysis pathways and get suggested compounds for growth media to improve production. Users are able to select interested pathway for test, to enter parameters, and to view and export results.

Algorithm



Interface

General



1. Tab Switch: Click button to switch Tabs.
2. Model Loading button: Click to load SBML model from machine, the checkbox shows loading status and the text will display the name of current loaded model.

Building Tab

The screenshot shows the 'Building Tab' interface with several components highlighted by numbered callouts:

- 6**: Search input field.
- 7**: Coefficient input field.
- 8**: Compartment dropdown menu.
- 9**: Radio buttons for 'Reactants' and 'Products'.
- 1**: 'Add' button.
- 10**: 'Rxn Name' input field.
- 11**: 'Lower Bound' input field.
- 12**: 'Upper Bound' input field.
- 13**: 'Reversible' input field.
- 2**: 'Create' button.
- 14**: Reactions table.
- 3**: 'Update' button.
- 4**: 'Reset' button.
- 5**: 'Export' button.

Abbreviation	Full Name
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Abbreviation	Coefficient	Comp
--------------	-------------	------

Abbreviation	Coefficient	Comp
--------------	-------------	------

Rxn Name	Formula	Reversible	Lower bound	Upper bound
14				

1. Compound Addition button: Click to add compound with its name, coefficient and compartment information to either Reactants Table or Products Table based on user selection.
2. Reaction Creation button: Click to create a reaction consists of all reactants in Reactants Table and all products in Products Table.
3. Reactions Update button: Click to update changes of reactions that have been already created in Reactions Table.
4. Tab Reset button: Click to clear all the content/data in the Building Tab.
5. Pathway Export button: Click to export pathway that is made up of all the reactions created in Reactions Table. The exported file is written in m script format and able to run FVA test immediately.
6. Compound Name field: Enter compound name here. The name must be non-empty for adding to Reactants Table or Products Table. Once a model is loaded, the Filter button is active. Users can search a compound that is in the model.

7. Coefficient field: Enter Stoichiometric Coefficient here for a compound in a reaction. The value must be numerical and within the range from one to a thousand.
8. Compartment Selection list: Click to select compartment that a compound should locate in a reaction.
9. Compound Role Selection buttons: Click either “Reactants” or “Products” to determine the role of compound in a reaction.
10. Reaction Name field: Enter reaction name here. The name must be non-empty and not duplicate with names of reactions already in Reactions Table.
11. Lower Bound Value field: Enter lower bound value for a reaction here. The value must be numerical.
12. Upper Bound Value field: Enter upper bound value for a reaction here. The value must be numerical.
13. Reversible checkbox: Check the box to indicate a reaction is reversible and uncheck to make the reaction irreversible.
14. Reactions Table: Click any of the cells in the table to change the value entered previously. To update new values in table, click Update button to submit changes.

Analysis Tab

The screenshot displays the Analysis Tab interface. On the left, there are five input fields: Pathway (dropdown menu with '4' selected), Number of Approx. (text box with '5'), Polynomial Degree (text box with '3'), Differences Threshold (text box with '1'), and Zeros Threshold (text box with '1e-05'). To the right is a graph with 'Target compound flux rate (mmol/h/mlDWg)' on the y-axis and 'Biomass flux rate (mmol/h/mlDWg)' on the x-axis, both ranging from 0 to 1. A blue number '9' is plotted on the graph. Below the graph is a table with the following columns: Metabolic Names, Glucose rate, Added met rate, Biomass rate, Target rate, and Value (%). The table is currently empty. At the bottom, there is a 'Button Group' containing three buttons: 'Run' (labeled '1'), 'Reset' (labeled '2'), and 'Export' (labeled '3').

1. Analysis Run button: Click to run analysis on a pathway with all inputted parameters.
2. Analysis Reset button: Click to clear all content/data in Analysis Tab.
3. Analysis Result Export button: Click to export the results showed in table on the interface to a text file.
4. Pathway Selection dropdown menu: Select a pathway in dropdown menu to analyze.
5. Number of Approximation field: Enter a number here to define how many sample points need to exam to determine the relationship between production rate and growth rate.
6. Polynomial Degree field: Enter a number here to define the degree of polynomial will be used to approximate the function between production rate and growth rate.

7. Differences Threshold field: Enter a percentage here to define the threshold of differences of reactions flux in product maximized and product minimized sets.
8. Zeros Threshold field: Enter a value here to define a threshold that makes the program to take any value less than this threshold to consider as zero.
9. Relationship Plot: The plot of a function that represents the relationship between growth rate and production rate will display here.

Credits

All the documents and source code are governed under the BioBrick's Foundation and iGEM 2012 copyright policies.

