**Flux Variability Analysis & Parsimonious Flux Balance Analysis**

**Expectations**

**Learning Objectives**

Each student should be able to:

* Explain alternate optimal solutions.
* Explain flux variability analysis.
* Explain parsimonious FBA.

**Prerequisites**

* Course Introduction
* Matlab Tutorial
* Flux Balance Analysis Overview
* *E.coli* Core Model
* Cobra Toolbox
* Robustness Analysis & Phenotype Phase Plane Analysis

**Resources**

**Required Reading**

1. Systems Biology: Constraint-based Reconstruction and Analysis, Bernhard O. Palsson, Cambridge University Press, 2015, Chapter 20.
2. [Lewis, N. E., K. K. Hixson, et al. (2010). "Omic data from evolved *E. coli* are consistent with computed optimal growth from genome-scale models." Molecular Systems Biology 6: 390.](http://www.ncbi.nlm.nih.gov/pubmed/20664636)

**Recommended Reading**

1. [Reed, J. L. & Palsson, B. Ø. Genome-scale in silico models of *E. coli* have multiple equivalent phenotypic states: assessment of correlated reaction subsets that comprise network states. Genome Res. 14, 1797–1805 (2004).](http://www.ncbi.nlm.nih.gov/pubmed/15342562)

**Classroom Activities**

**Presentations**

* Lecture Presentation *(“Flux Variability Analysis & Parsimonious Flux Balance Analysis-2021.pdf”)*
* Supporting Matlab Files *(“FVA-PFBA Matlab Folders 2021.zip”)*

**Laboratory**

1. Lab #4 *(“Lab-4.docx”)*

**Reinforcement Activities**

**Examples**

* Alternate Optimal Solutions
  + [Reed, J. L. & Palsson, B. Ø. Genome-scale in silico models of *E. coli* have multiple equivalent phenotypic states: assessment of correlated reaction subsets that comprise network states. Genome Res. 14, 1797–1805 (2004).](http://www.ncbi.nlm.nih.gov/pubmed/15342562)
* Flux Variability Analysis
  + [Mahadevan, R. and C. H. Schilling (2003). "The effects of alternate optimal solutions in constraint-based genome-scale metabolic models." Metabolic engineering 5(4): 264-276](http://www.ncbi.nlm.nih.gov/pubmed/14642354)
  + [Schellenberger, J., R. Que, et al. (2011). "Quantitative prediction of cellular metabolism with constraint-based models: the COBRA Toolbox v2.0." Nature protocols 6(9): 1297, 1302.](http://www.ncbi.nlm.nih.gov/pubmed/21886097)
* Parsimonious Flux Balance Analysis
  + [Lewis, N. E., K. K. Hixson, et al. (2010). "Omic data from evolved *E. coli* are consistent with computed optimal growth from genome-scale models." Molecular Systems Biology 6: 390.](http://www.ncbi.nlm.nih.gov/pubmed/20664636)
  + [Schellenberger, J., R. Que, et al. (2011). "Quantitative prediction of cellular metabolism with constraint-based models: the COBRA Toolbox v2.0." Nature protocols 6(9): 1296, 1301.](http://www.ncbi.nlm.nih.gov/pubmed/21886097)

**Assessment**

**Formative Assessment**

* Reflective Questions
  1. What are alternate optimal solutions?
  2. What is the relationship between alternate optimal solutions and a cell’s phenotype?
  3. What are silent phenotypes?
  4. How can you find the alternate optimal solutions using the Cobra Toolbox?
  5. How many alternate optimal solutions can there be for a given phenotype?
  6. How many alternate optimal solutions can there be for a carbon source?
  7. Do aerobic/anaerobic conditions impact the number alternate optimal solutions?
  8. Does the choice of objective function impact the number alternate optimal solutions?
  9. What is flux variability analysis?
  10. What is the relationship between the value of the objective function and the flux values calculated through flux variability analysis?
  11. How is flux variability analysis related to alternate optimal flux vectors?
  12. How can you implement flux variability analysis using the Cobra Toolbox?
  13. Does flux variability analysis identify the specific alternate optimal solutions?
  14. What is the value of knowing which reactions carry flux, which reactions carry no flux, and which reactions span a range of flux values?
  15. Explain the different FVA classifications; hard-coupled, partially-coupled, not-coupled, and no-flux reactions?
  16. Why do they call it parsimonious flux balance analysis?
  17. What are essential genes/reactions?
  18. What are pFBA optima genes/reactions?
  19. What are enzymatically less efficient (ELE) genes/reactions?
  20. What are metabolically less efficient genes/reactions?
  21. What are pFBA no-flux genes/reactions?
  22. What are blocked genes/reactions?
  23. What is the difference between pFBA optima genes/reactions, enzymatically less efficient (ELE) genes/reactions and metabolically less efficient (MLE), genes/reactions?
  24. How can you implement parsimonious flux balance analysis using the Cobra Toolbox
  25. How can parsimonious flux balance analysis be used to metabolically engineer a cell?

**References**

**Flux Variability Analysis**

1. [Reed, J. L. & Palsson, B. Ø. Genome-scale in silico models of *E. coli* have multiple equivalent phenotypic states: assessment of correlated reaction subsets that comprise network states. Genome Res. 14, 1797–1805 (2004).](http://www.ncbi.nlm.nih.gov/pubmed/15342562)
2. [Mahadevan, R. and C. H. Schilling (2003). "The effects of alternate optimal solutions in constraint-based genome-scale metabolic models." Metabolic engineering 5(4): 264-276](http://www.ncbi.nlm.nih.gov/pubmed/14642354)
3. [Phalakornkule, C. et al. A MILP-based flux alternative generation and NMR experimental design strategy for metabolic engineering. Metab. Eng. 3, 124–137 (2001).](http://www.ncbi.nlm.nih.gov/pubmed/11289789)
4. [Lee, S., Phalakornkule, C., Domach, M. M. & Grossmann, I. E. Recursive MILP model for finding all the alternate optima in LP models for metabolic networks.Comp. Chem. Eng. 24, 711–716 (2000).](http://apps.webofknowledge.com/InboundService.do?SID=2ErNXHAB3r1KMtBSIun&product=WOS&UT=000088546800082&SrcApp=EndNote&DestFail=http%3A%2F%2Fwww.webofknowledge.com&Init=Yes&action=retrieve&SrcAuth=ResearchSoft&customersID=ResearchSoft&Func=Frame&IsProductCode=Yes&mode=FullRecord)

**Parsimonious Analysis**

1. [Lewis, N. E., K. K. Hixson, et al. (2010). "Omic data from evolved *E. coli* are consistent with computed optimal growth from genome-scale models." Molecular Systems Biology 6: 390.](http://www.ncbi.nlm.nih.gov/pubmed/20664636)