**Randomized Sampling  
  
Expectations**

**Learning Objectives**

Each student should be able to:

* Explain the Method of Minimization of Metabolic Adjustment (MOMA)
* Explain adaptive laboratory evolution
* Explain extreme pathways
* Explain randomized sampling

**Prerequisites**

* Course Introduction
* Matlab Tutorial
* Flux Balance Analysis Overview
* E.coli Core Model
* Cobra Toolbox
* Robustness Analysis & Phenotype Phase Plane Analysis
* Flux Variability Analysis & Parsimonious Analysis
* Gene/Reaction Knockout

**Resources**

**Required Readings**

1. [Schellenberger, J. and B. O. Palsson (2009). "Use of randomized sampling for analysis of metabolic networks." The Journal of biological chemistry 284(9): 5457-5461.](http://www.ncbi.nlm.nih.gov/pubmed/18940807)
2. [Ibarra, R. U., J. S. Edwards, et al. (2002). "Escherichia coli K-12 undergoes adaptive evolution to achieve in silico predicted optimal growth." Nature 420(6912): 186-189.](http://www.ncbi.nlm.nih.gov/pubmed/12432395)
3. [Segre, D., D. Vitkup, et al. (2002). "Analysis of optimality in natural and perturbed metabolic networks." Proceedings of the National Academy of Sciences of the United States of America 99(23): 15112-15117. - **MOMA**](http://www.ncbi.nlm.nih.gov/pubmed/12415116)
4. Systems Biology: Constraint-based Reconstruction and Analysis, Bernhard O. Palsson, Cambridge University Press, 2015, Chapter 14.

**Recommended Readings**

1. [Papin, J. A., N. D. Price, et al. (2003). "Metabolic pathways in the post-genome era." Trends in biochemical sciences 28(5): 250-258.](http://roger.ucsd.edu/record=b6928588~S9)
2. [Jan Schellenberger, PhD Dissertation, University of California, San Diego, 2010](http://roger.ucsd.edu/record=b6928588~S9)
3. [Price, N. D., J. Schellenberger, et al. (2004). "Uniform sampling of steady-state flux spaces: means to design experiments and to interpret enzymopathies." Biophysical journal 87(4): 2172-2186.](http://www.ncbi.nlm.nih.gov/pubmed/15454420)
4. [Almaas, E., B. Kovacs, et al. (2004). "Global organization of metabolic fluxes in the bacterium Escherichia coli." Nature 427(6977): 839-843.](http://www.ncbi.nlm.nih.gov/pubmed/14985762)

**Classroom Activities**

**Presentations**

* Lecture Presentation *(“Sampling – 2021.pdf”)*
* Supporting Matlab Files *(“Sampling Matlab Files – 2021.zip”)*

**Laboratory**

1. Lab #6 *(“Lab-6.docx”)*

**Reinforcement Activities**

**Examples**

* Adaptive Laboratory Evolution
  + [Ibarra, R. U., J. S. Edwards, et al. (2002). "Escherichia coli K-12 undergoes adaptive evolution to achieve in silico predicted optimal growth." Nature 420(6912): 186-189.](http://www.ncbi.nlm.nih.gov/pubmed/12432395)
* MOMA
  + [Segre, D., D. Vitkup, et al. (2002). "Analysis of optimality in natural and perturbed metabolic networks." Proceedings of the National Academy of Sciences of the United States of America 99(23): 15112-15117](http://www.ncbi.nlm.nih.gov/pubmed/12415116)
  + 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): pp. 1297.
* Extreme Pathways
  + [Papin, J. A., N. D. Price, et al. (2003). "Metabolic pathways in the post-genome era." Trends in biochemical sciences 28(5): 250-258.](http://roger.ucsd.edu/record=b6928588~S9)
* Randomized Sampling
  + [Schellenberger, J. and B. O. Palsson (2009). "Use of randomized sampling for analysis of metabolic networks." The Journal of biological chemistry 284(9): 5457-5461.](http://www.ncbi.nlm.nih.gov/pubmed/18940807)
  + [Price, N. D., J. Schellenberger, et al. (2004). "Uniform sampling of steady-state flux spaces: means to design experiments and to interpret enzymopathies." Biophysical journal 87(4): 2172-2186.](http://www.ncbi.nlm.nih.gov/pubmed/15454420)
  + 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): pp. 1298, 1302, 1304.

**Videos**

* [B. O. Palsson video on Randomized Sampling](https://www.youtube.com/watch?v=FSnE82REBq0&feature=youtu.be)
  + (<http://systemsbiology.ucsd.edu/Publications/Books/SB1-2LectureSlides>)

**Assessment**

**Formative Assessment**

**Reflective Questions**

1. After a gene has been knockout or a new gene has been added to a host cell does the maximum theoretical performance typically match the laboratory results?
2. What Cobra function can be used to approximate the intermediate suboptimal state of the modified host cell?
3. What is a wild-type cell/model? How does it differ from the mutant cell/model?
4. Are the optimized flux values similar to the MOMA flux results?
5. Why don’t the first generation of transformed cells normally achieve the optimal performance predicted in the FBA models?
6. How can cells evolve after 100’s of generations to operate on the line of optimality?
7. What is the relationship between MOMA and adaptive laboratory evolution?
8. What are the number of generations required for adaptive laboratory evolution?
9. What are correlated reaction sets?
10. What is the solution space?
11. What determines the solution space that is used in randomized sampling?
12. Is randomized sampling classified as biased or unbiased assessment?
13. What is hit-and-run sampling?
14. What is the mixed fraction parameter?
15. Under what name are the sample points listed in the sampleStruct?
16. What role does the objective function play in randomized sampling?
17. What Cobra function allows the graphical comparison of different sampled solutions?
18. What Cobra function provides a graphical representation of the correlations between reactions?
19. What Cobra function can be used for randomized sampling?
20. What role do reaction constraints play in the accuracy of the data generated by randomized sampling?

**References**

**Sampling Methods**

1. [David E. Kaufman, Robert L. Smith, (1998) Direction Choice for Accelerated Convergence in Hit-and-Run Sampling. Operations Research 46(1):84-95.](http://pubsonline.informs.org/doi/abs/10.1287/opre.46.1.84)
2. [Lovasz, (1999).”Hit-and-run mixes fast,” Mathematical Programming, 86:443-461.](http://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.28.1700&rep=rep1&type=pdf)

**Sampling Applications**

1. [Thiele, I., N. D. Price, et al. (2005). "Candidate metabolic network states in human mitochondria. Impact of diabetes, ischemia, and diet." The Journal of biological chemistry 280(12): 11683-11695.](http://www.ncbi.nlm.nih.gov/pubmed/15572364)
2. [Price, N. D., J. Schellenberger, et al. (2004). "Uniform sampling of steady-state flux spaces: means to design experiments and to interpret enzymopathies." Biophysical journal 87(4): 2172-2186.](http://www.ncbi.nlm.nih.gov/pubmed/15454420)

**Adaptive Evolution**

1. [Dragosits, M. and D. Mattanovich (2013). "Adaptive laboratory evolution -- principles and applications for biotechnology." Microbial cell factories 12: 64.](http://www.ncbi.nlm.nih.gov/pubmed/23815749)
2. [B. Palsson (2010). “Adaptive Laboratory Evolution.” Microbe,  6(2):69-74](https://www.microbemagazine.org/images/stories/images/feb2011/znw00211000069.pdf)
3. [Conrad, T. M., N. E. Lewis, et al. (2011). "Microbial laboratory evolution in the era of genome-scale science." Molecular Systems Biology 7: 509](http://www.ncbi.nlm.nih.gov/pubmed/21734648)
4. [Fong, S. S., A. P. Burgard, et al. (2005). "In silico design and adaptive evolution of Escherichia coli for production of lactic acid." Biotechnology and bioengineering 91(5): 643-648.](http://www.ncbi.nlm.nih.gov/pubmed/15962337)
5. [Fong, S. S., J. Y. Marciniak, et al. (2003). "Description and interpretation of adaptive evolution of Escherichia coli K-12 MG1655 by using a genome-scale in silico metabolic model." Journal of Bacteriology 185(21): 6400-6408.](http://www.ncbi.nlm.nih.gov/pubmed/14563875)
6. [Ibarra, R. U., J. S. Edwards, et al. (2002). "Escherichia coli K-12 undergoes adaptive evolution to achieve in silico predicted optimal growth." Nature 420(6912): 186-189.](http://www.ncbi.nlm.nih.gov/pubmed/12432395)

**Extreme Pathways**

1. [Price, N. D., J. L. Reed, et al. (2003). "Analysis of metabolic capabilities using singular value decomposition of extreme pathway matrices." Biophysical journal 84(2 Pt 1): 794-804.](http://www.ncbi.nlm.nih.gov/pubmed/12547764)
2. [Papin, J. A., N. D. Price, et al. (2003). "Metabolic pathways in the post-genome era." Trends in biochemical sciences 28(5): 250-258.](http://apps.webofknowledge.com/InboundService.do?product=WOS&SID=1CKBPBG9jKnkdCquSWr&UT=WOS%3A000183468800008&SrcApp=EndNote&DestFail=http%3A%2F%2Fwww.webofknowledge.com&action=retrieve&Init=Yes&SrcAuth=ResearchSoft&customersID=ResearchSoft&Func=Frame&IsProductCode=Yes&mode=FullRecord)
3. [Papin, J. A., N. D. Price, et al. (2002). "The genome-scale metabolic extreme pathway structure in Haemophilus influenzae shows significant network redundancy." J Theor Biol 215(1): 67-82.](http://www.ncbi.nlm.nih.gov/pubmed/12051985)
4. [Papin, J. A., N. D. Price, et al. (2002). "Extreme pathway lengths and reaction participation in genome-scale metabolic networks." Genome research 12(12): 1889-1900.](http://www.ncbi.nlm.nih.gov/pubmed/12466293)
5. [Price, N. D., J. A. Papin, et al. (2002). "Determination of redundancy and systems properties of the metabolic network of Helicobacter pylori using genome-scale extreme pathway analysis." Genome research 12(5): 760-769.](http://www.ncbi.nlm.nih.gov/pubmed/11997342)
6. [Wiback, S. J. and B. O. Palsson (2002). "Extreme pathway analysis of human red blood cell metabolism." Biophysical journal 83(2): 808-818.](http://www.ncbi.nlm.nih.gov/pubmed/12124266)

**Sampling Low-dimensional Solution Space**

1. [Price, N. D., J. Schellenberger, et al. (2004). "Uniform sampling of steady-state flux spaces: means to design experiments and to interpret enzymopathies." Biophysical journal 87(4): 2172-2186.](http://www.ncbi.nlm.nih.gov/pubmed/15454420)
2. [Wiback, S. J., I. Famili, et al. (2004). "Monte Carlo sampling can be used to determine the size and shape of the steady-state flux space." J Theor Biol 228(4): 437-447.](http://www.ncbi.nlm.nih.gov/pubmed/15178193)

**Sampling High-dimensional Solution Space**

1. [Almaas, E., B. Kovacs, et al. (2004). "Global organization of metabolic fluxes in the bacterium Escherichia coli." Nature 427(6977): 839-843.](http://www.ncbi.nlm.nih.gov/pubmed/14985762)
2. [Schellenberger, J. and B. O. Palsson (2009). "Use of randomized sampling for analysis of metabolic networks." The Journal of biological chemistry 284(9): 5457-5461.](http://www.ncbi.nlm.nih.gov/pubmed/18940807)