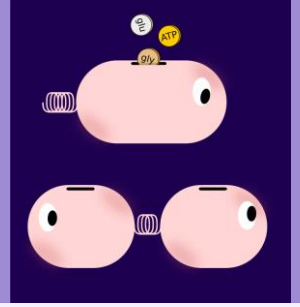
Economic Principles in Cell Biology

Paris, July 8-11, 2024



Return on investment (ROI) in microbial metabolism and interactions

Presented by

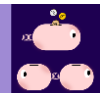
Hyun-Seob Song, University of Nebraska-Lincoln

Doraiswami Ramkrishna, Purdue University

Outline

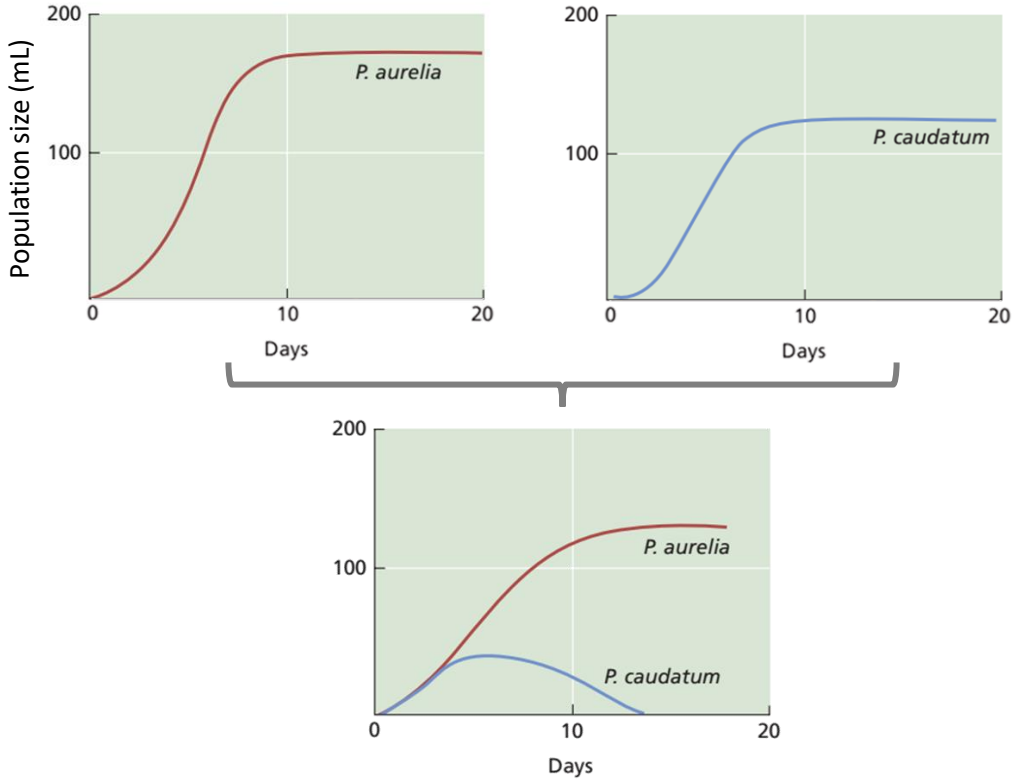
- Survival strategies of microorganisms
- Return-on-investment (ROI) in metabolism
- Cybernetic modeling
- Modeling of microbial interactions

- Modeling of eukaryotic systems
- Difficult to model the whole organism. Suitably define “subsystems”.
- How does one view ROI in eukaryotes?
- How does find meaningful objectives for cybernetic modeling?
- Concluding remarks



Competition drives microorganisms to evolve towards increasing their survival chance

- Microorganisms in natural environments face a constant battle for resources
- Competitive exclusion principle: a cornerstone of community ecology
- Microorganisms have evolved to develop survival strategies into multiple directions
  - Innovation in metabolism
  - Building partnerships

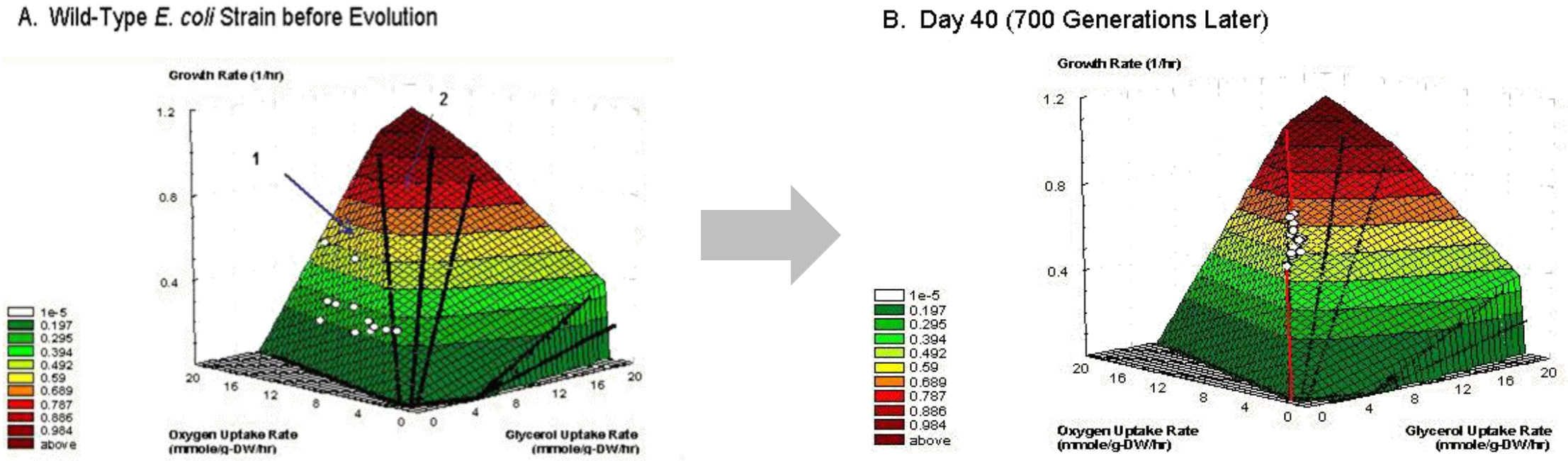


Gause experiment (1934)

(Images taken from Mittelbach and McGill, Community Ecology, 2nd Ed., 2019)

Innovation in metabolism: optimal growth

Evolution of *E. coli* towards optimal growth predicted by flux balance analysis

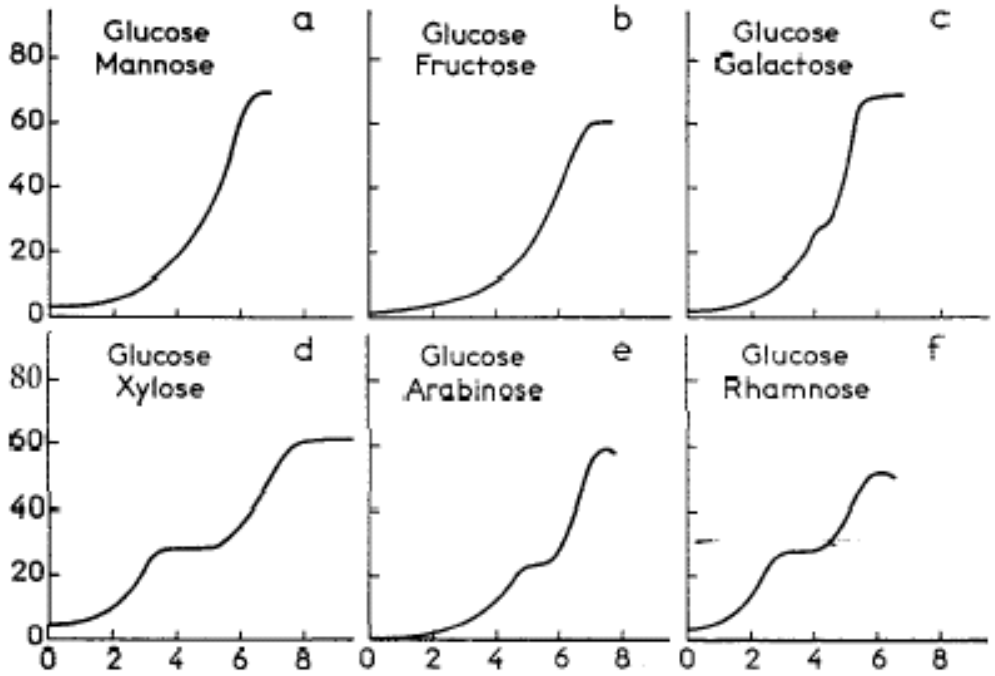


Ibarra et al. (Nature, 2002)

https://www.nsf.gov/od/lpa/news/02/pr0292.htm

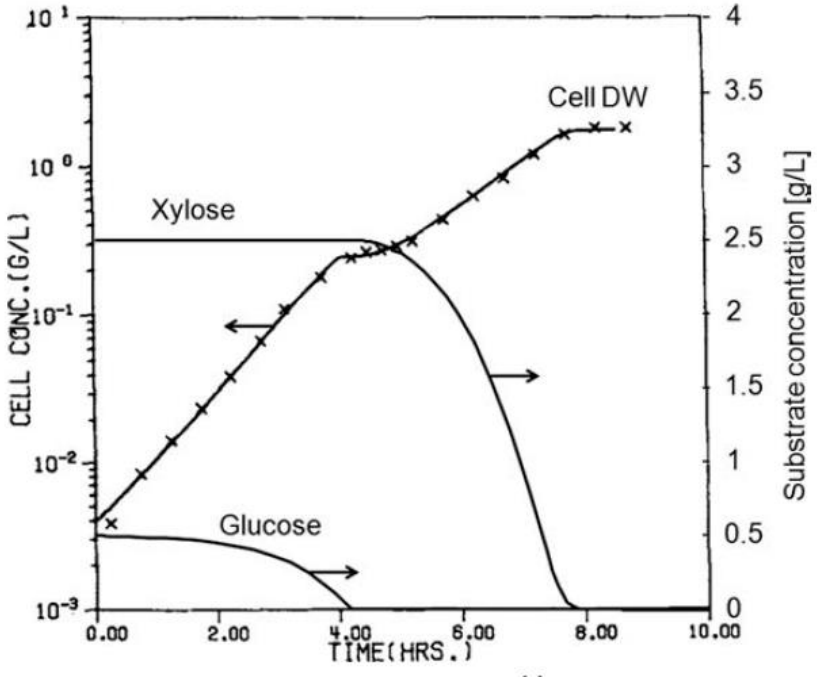
Innovation in metabolism: optimal metabolic switching

Growth of *E. coli* on different carbohydrate pairs (Monod's experiments in 1940s)



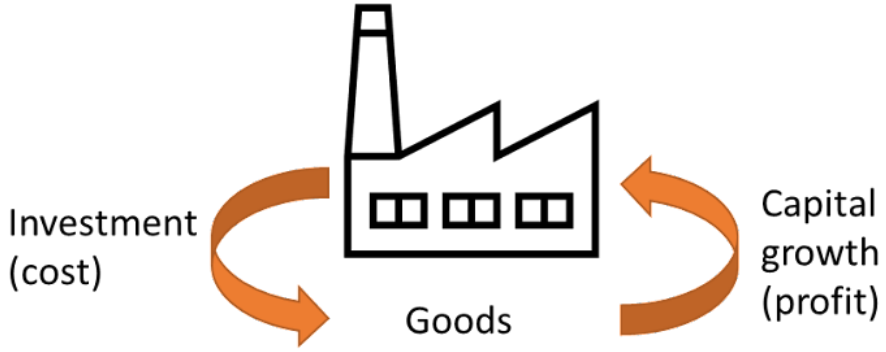
http://science.sciencemag.org/content/154/3748/475

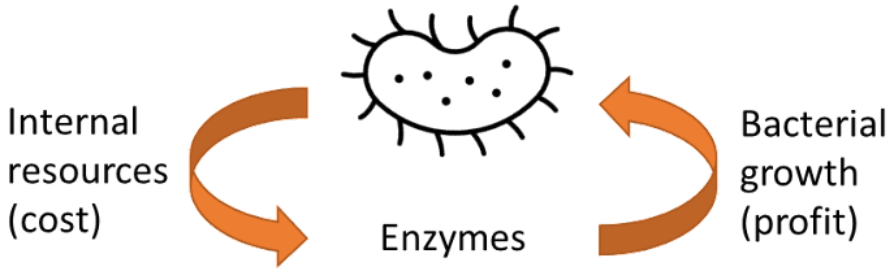
Diauxic growth of *K. oxytoca* on glucose and xylose predicted by cybernetic modeling



Kompala et al. (Biotech Bioeng, 1986)

Return-on-investment (ROI) is an important concept to understand optimal microbial growth





$$ROI = \frac{Net\ Profit}{Cost\ of\ Investment}$$

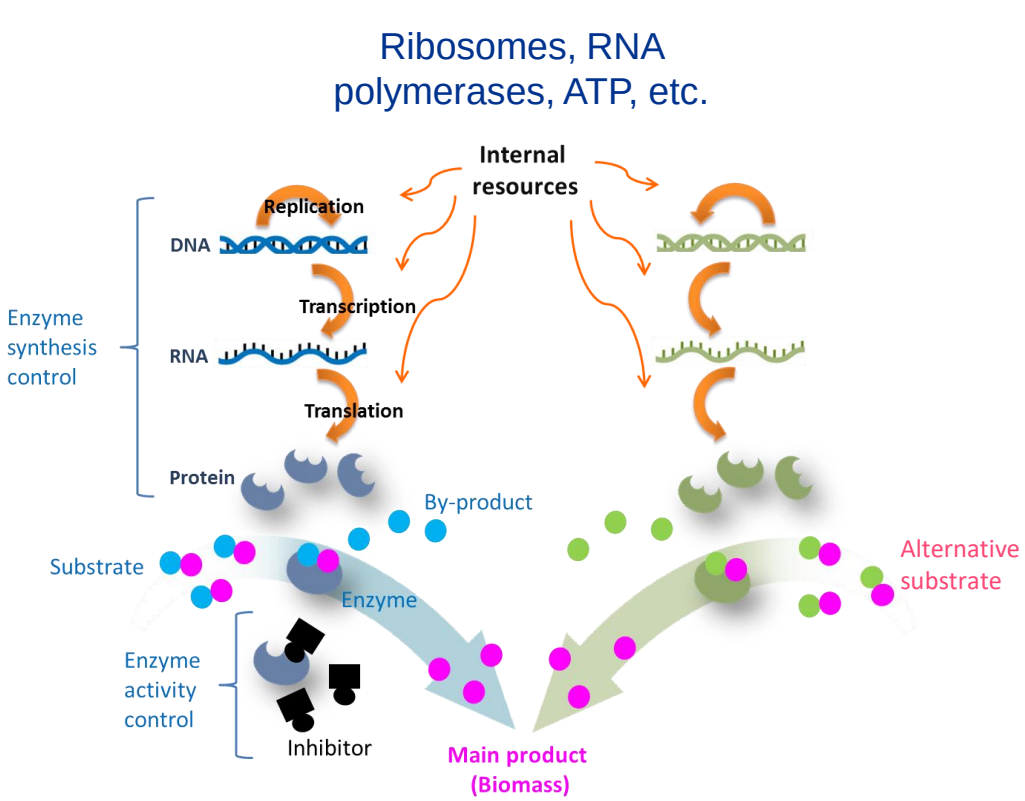
$$ROI = \frac{Gain\ from\ Investment\ - Cost}{Cost}$$

$$mROI = \frac{Net\ Growth}{Cost\ for\ Enzyme\ Production}$$

$$mROI = \frac{\Delta Growth - Cost}{Cost}$$

(mROI = Metabolic ROI)

‘Return’ and ‘cost of investment’ in metabolism

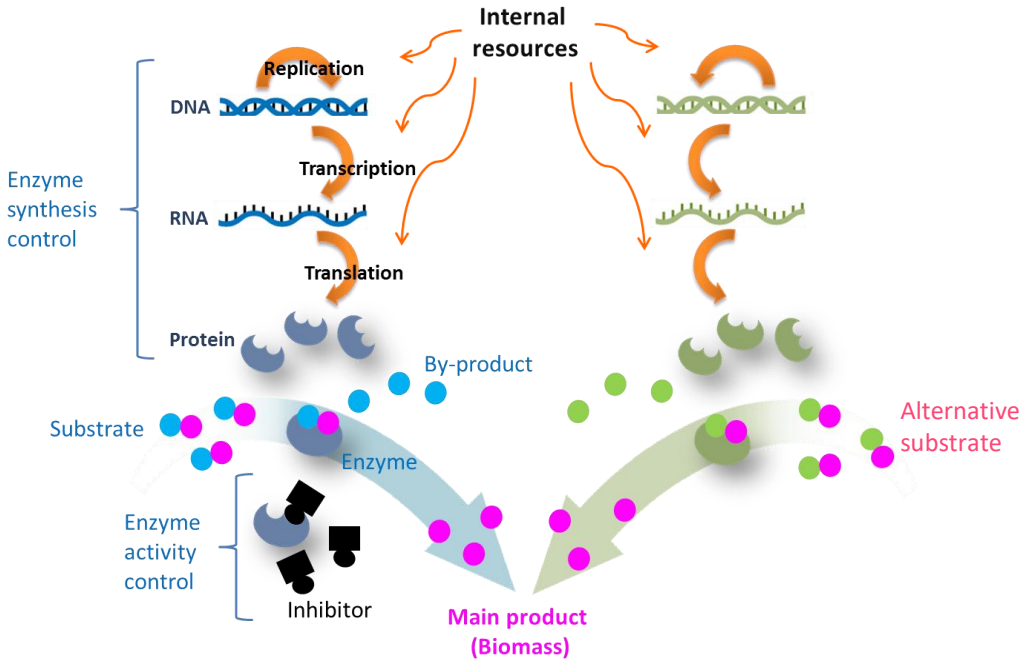


$$mROI = \frac{Gain\ from\ Investment\ - Cost}{Cost}$$

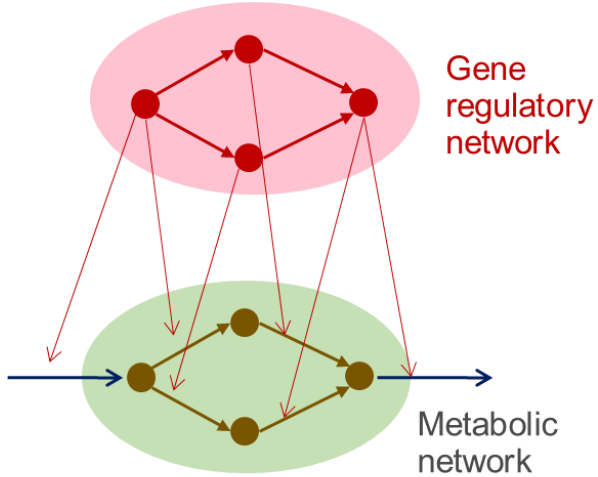
- Net profit or return
  - Cellular growth rate (commonly used)
  - Maintenance (ATP production)
  - Substrate uptake rate
  - Others
- Cost of investment or resources
  - Material and bioenergetic costs required for producing the defined net profit
  - Internal resources: ribosomes, RNA polymerases, ATP, etc.

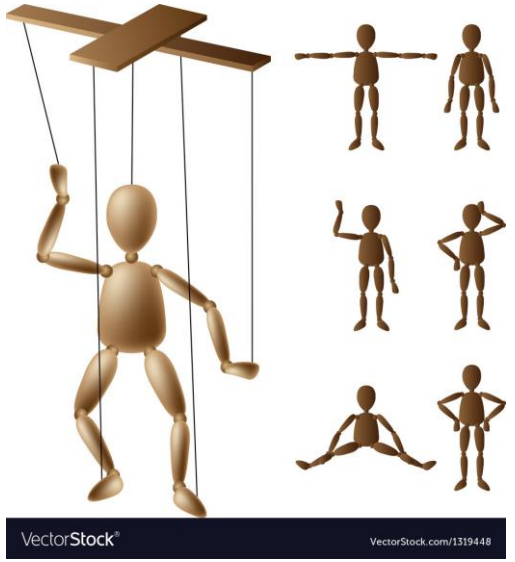
Regulation through optimal allocation of resources is key for maximizing ROI

Ribosomes, RNA polymerases, ATP, etc.

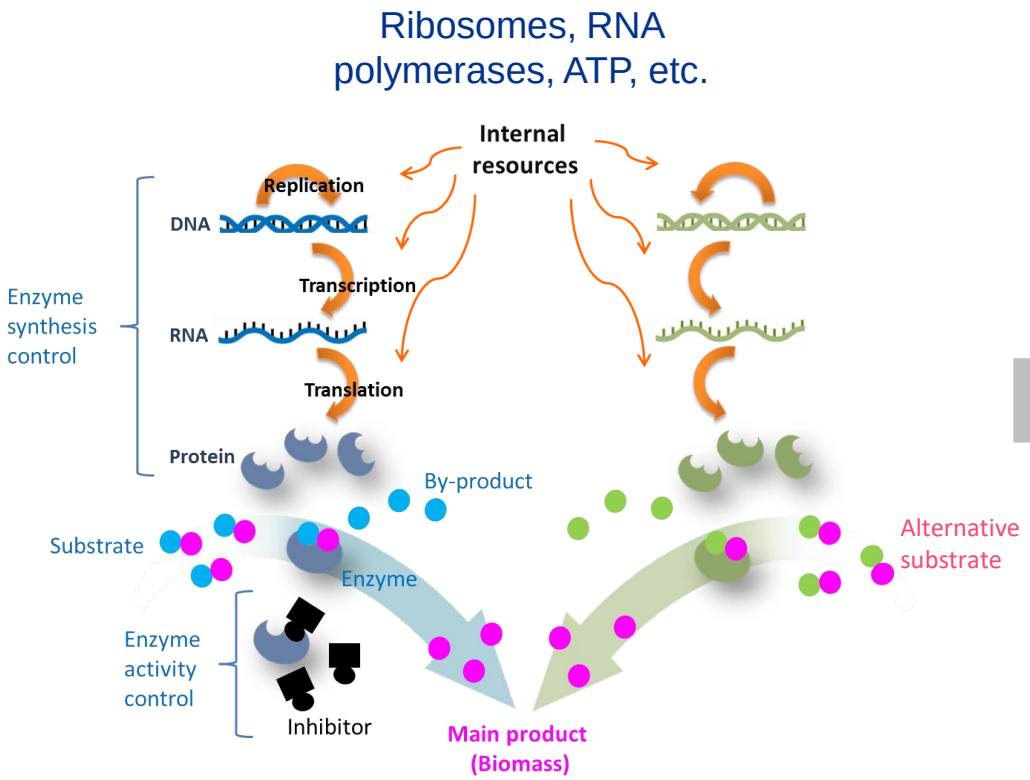


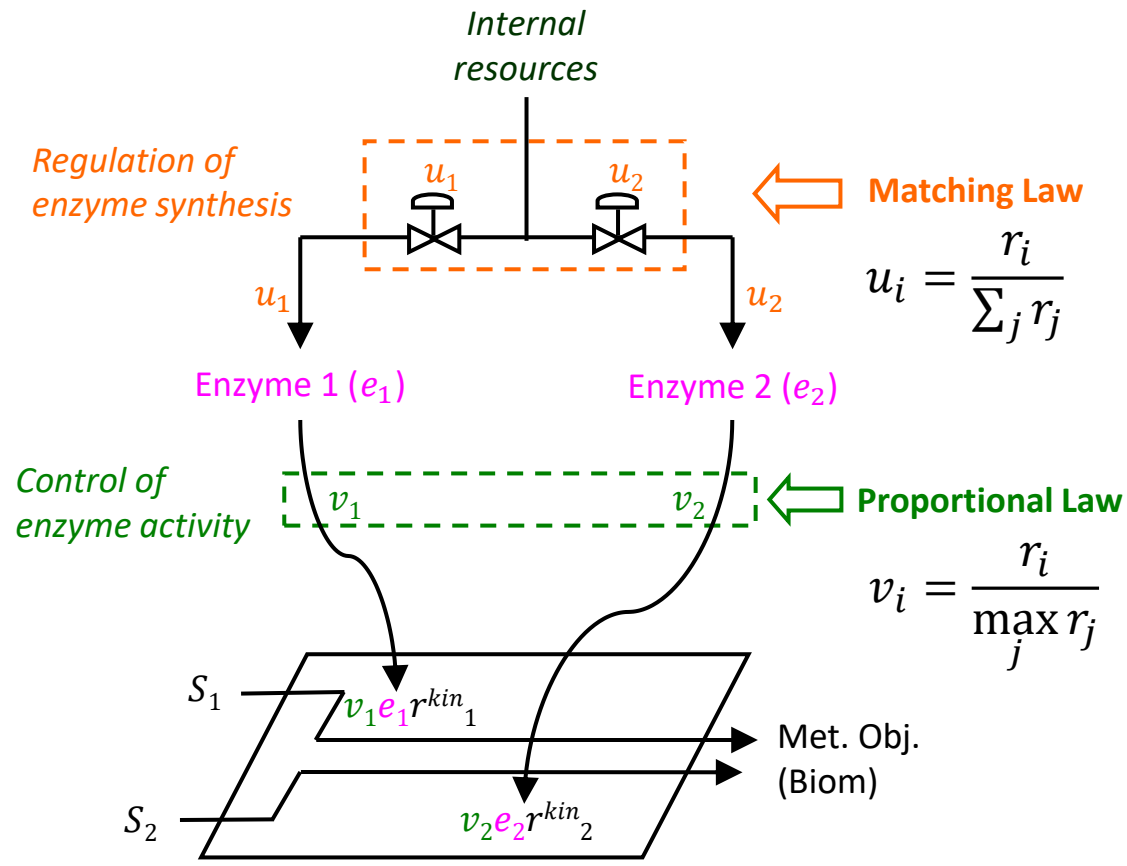
Gene regulatory circuit: control tower





Accounting for ROI and optimal resource allocation in cybernetic modeling





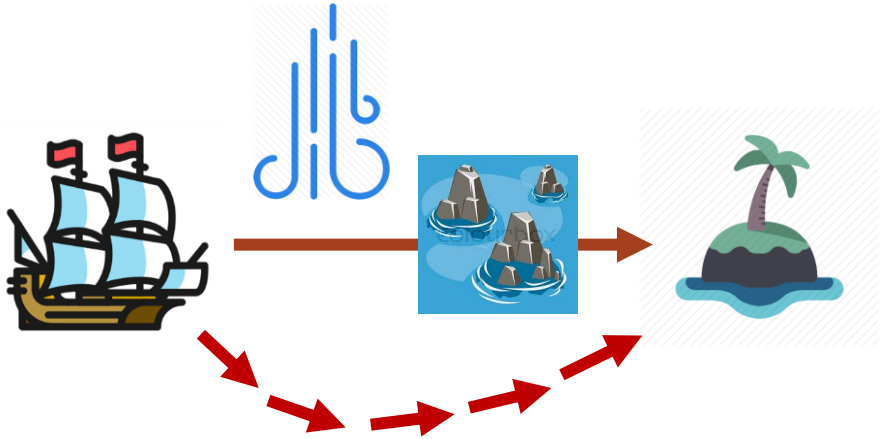
- Mechanistic details of regulation are replaced with the direct description on enzyme synthesis and activity control

The cybernetic approach views microbes as AI systems that optimally regulate metabolic actions towards maximizing ROI

Doraiswami Ramkrishna, the Harry Creighton Peffer Distinguished Professor of Chemical Engineering

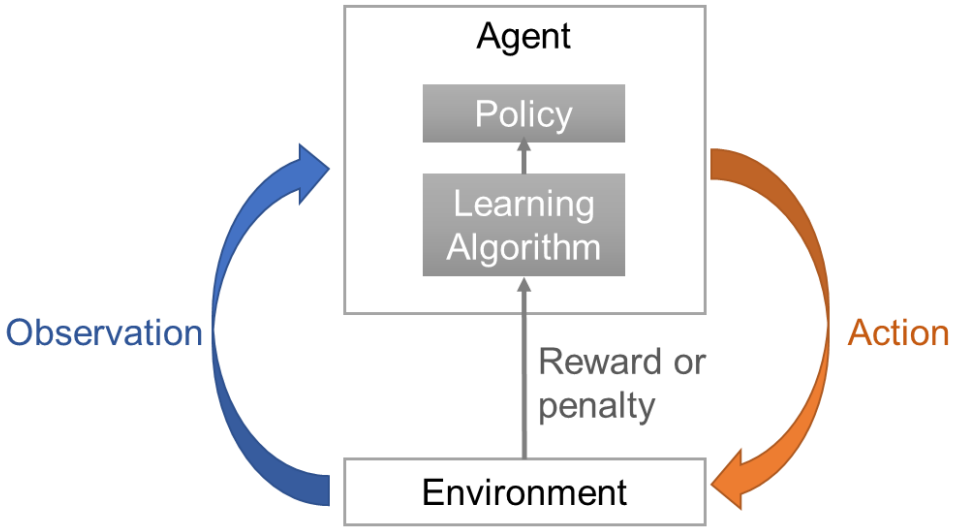
Pioneer of the cybernetic modeling

- “Cybernetics” comes from a Greek word meaning “the art of steering”
- Cybernetics sets a goal and takes action to achieve that goal
- The cybernetic model solves an optimal control problem to simulate cellular behavior

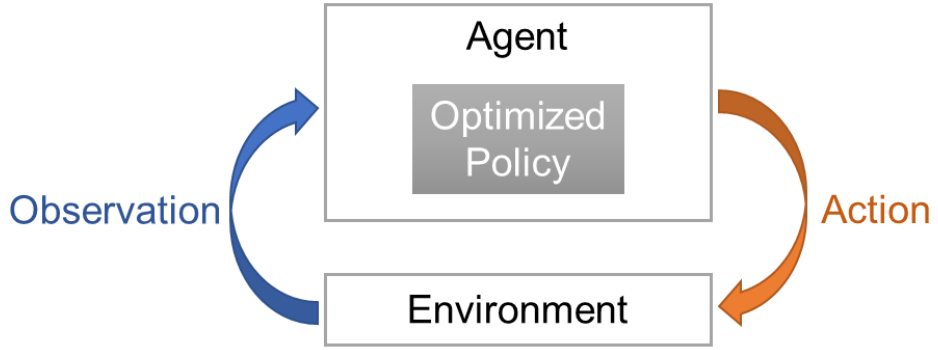


The cybernetic approach views microbes as AI systems that optimally regulate metabolic actions towards maximizing ROI

Reinforcement Learning (RL)



Cybernetic Modeling (CM)



- Both RL and CM implement intelligence through dynamic feedback loops
- RL evaluates the outcomes of taken actions as rewards or penalties to update the policy to get the most reward over time
- CM performs optimal control based on the already optimized policy

In cybernetic modeling, resources are optimally allocated such that metabolic ROI is maximized

Derivation of cybernetic control laws by solving a linear quadratic regulator problem

$$\max J \left(= \underbrace{\mathbf{q}^T \Delta\mathbf{y}(t+\Delta t)}_{= \Delta\text{Growth}} - \underbrace{\frac{\sigma}{2}\int_t^{t+\Delta t} \mathbf{u}^T\mathbf{u}\, d\tau}_{= \text{Cost for enzyme production}} \right)$$

$$= \frac{\mathbf{q}^T \Delta\mathbf{y}(t+\Delta t) - \frac{\sigma}{2}\int_t^{t+\Delta t} \mathbf{u}^T\mathbf{u}\, d\tau}{\underbrace{\sum_i u_i}_{=1}} = mROI$$

The total amount of resources (100%) to be allocated

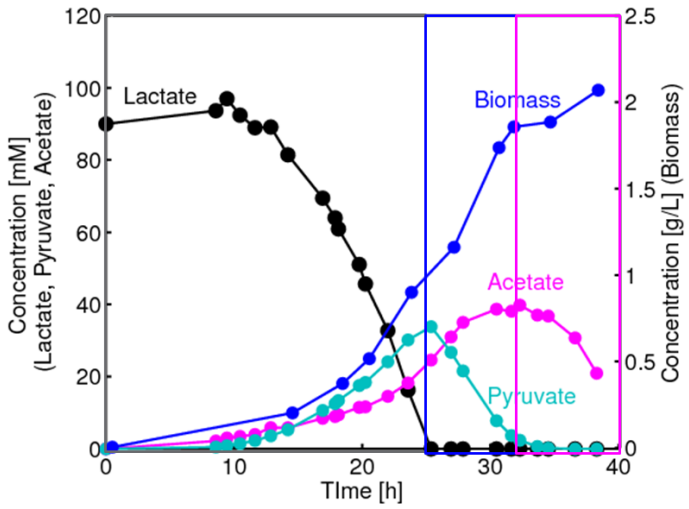
Generalized form of cybernetic control laws

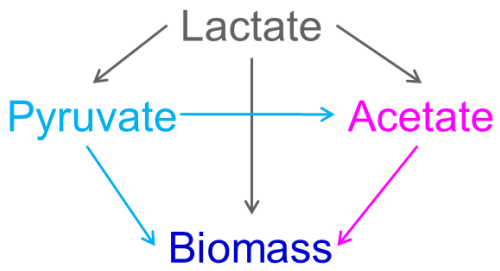
$$\mathbf{u}(t) = \frac{1}{\sigma}\mathbf{B}^T \mathbf{e}^{\mathbf{A}^T \Delta t}\mathbf{q}$$

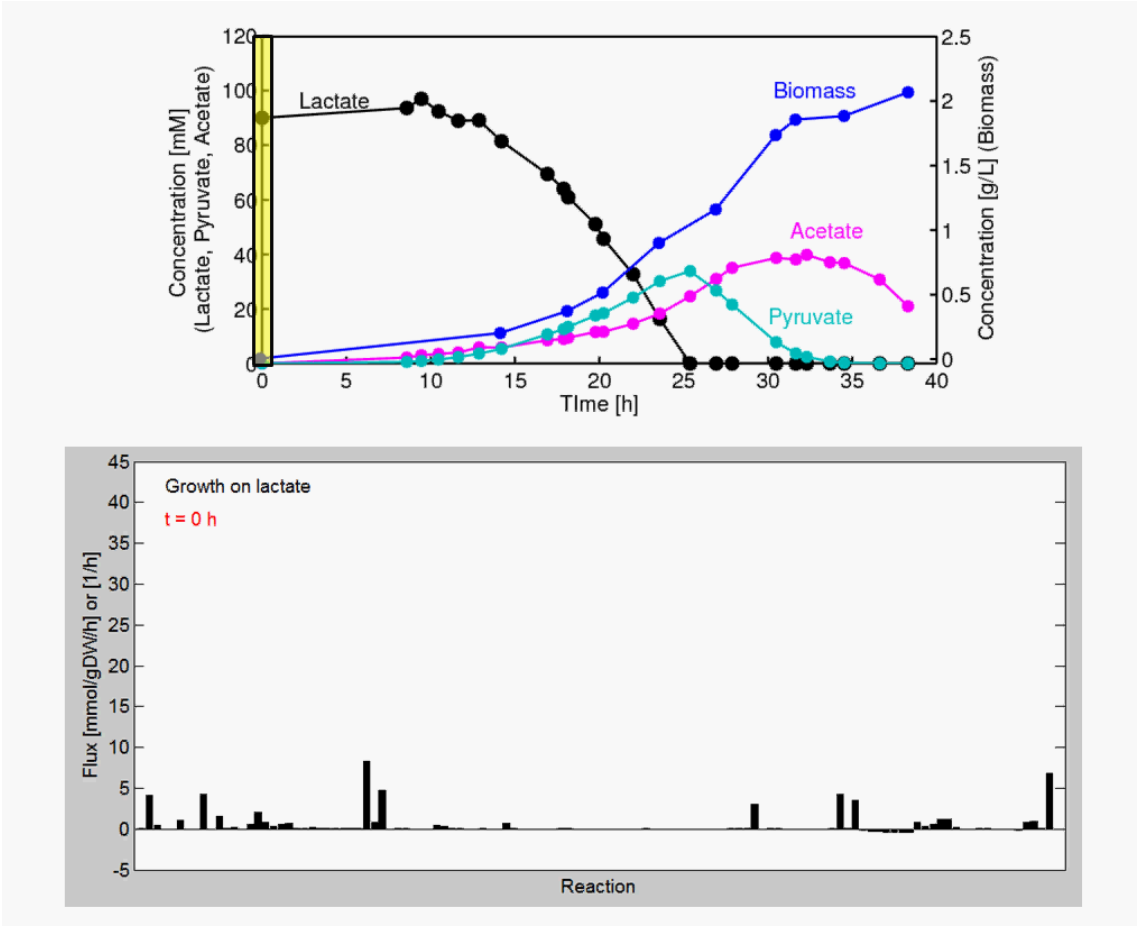
$$\Delta t = 0 \quad \Rightarrow \quad u_i = \frac{r_i}{\sum_j r_j}$$

Young and Ramkrishna (Biotechnol. Prog., 2007)

Simulation of dynamic metabolic switching in *Shewanella oneidensis* MR-1



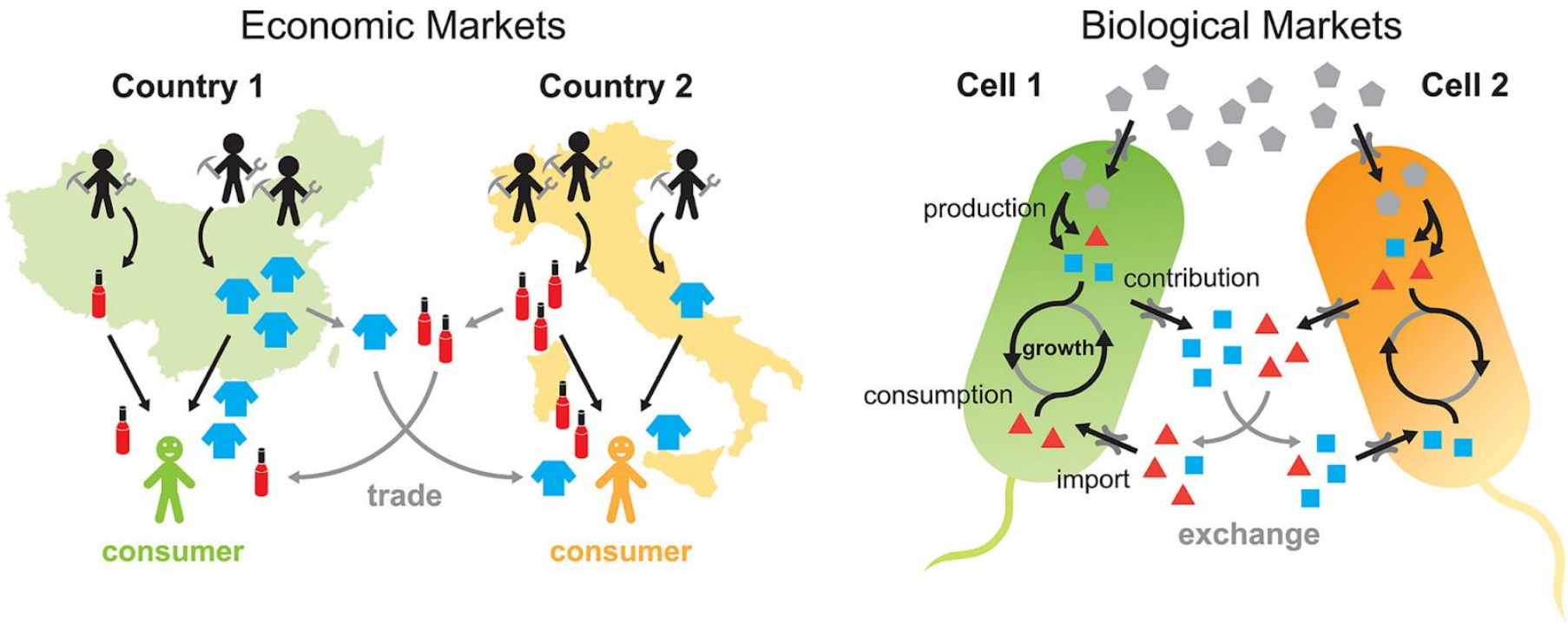




Song et al. (Metab Eng, 2013)

Economics in microbial interactions

- Innovation in metabolism: maximization of **direct** ROI
- Building partnerships: maximization of **indirect** ROI



Tasoff et al. (PLOS ONE, 2015)
https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0132907

What would be a relevant choice of metabolic objective to maximize? Individual vs. community growth

- Maximization of the growth of individual species fails to predict interspecies social behaviors of microorganisms such as division of labor or cross-feeding.

Maximization of individual growth

- The use of maximization of the total (or community) growth is criticized by ecologists favoring individualistic perspectives of microbial communities – it is difficult to justify cell's altruism.

Maximization of community growth

Cybernetic modeling resolves these issues by using generalized cybernetic control laws

- Choice of metabolic objective to maximize
- Context dependency in microbial interactions

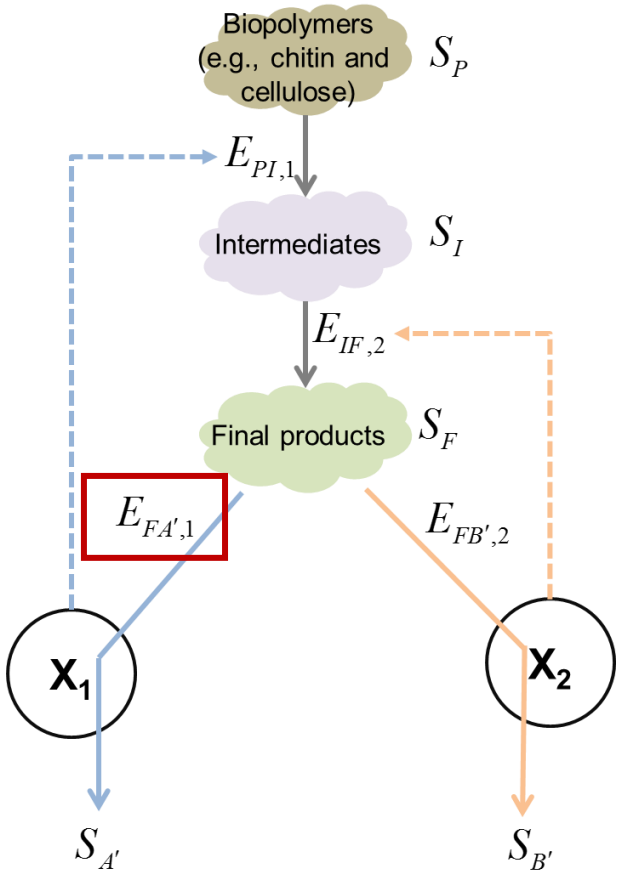
Maximizing **individual ROI** ***over a finite time horizon***

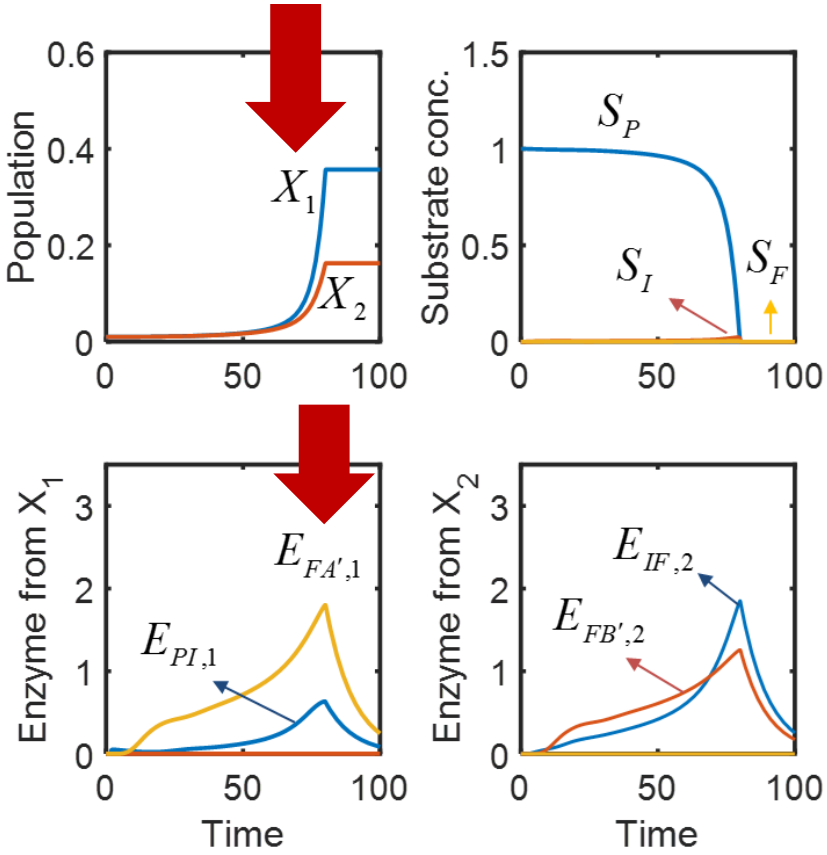
$$\mathbf{u}(t) = \frac{1}{\sigma}\mathbf{B}^T\mathbf{e}^{\mathbf{A}^T\Delta t}\mathbf{q}$$

The resulting community model enables predicting:

- Social behaviors of microorganisms such as division of labor and cross-feeding
- Context-dependent changes in interactions

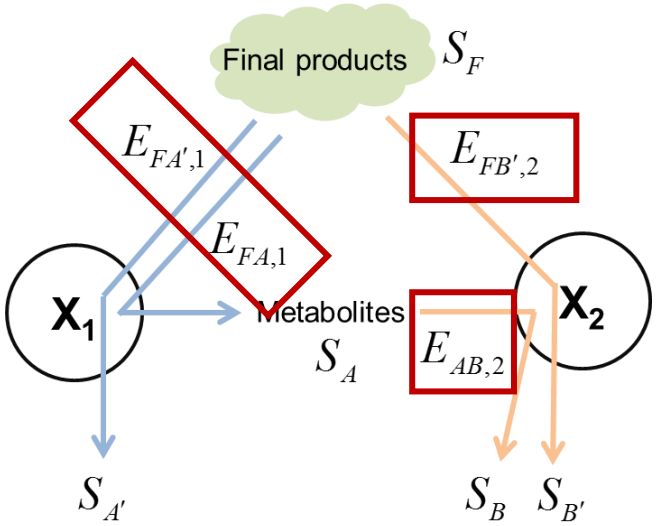
Division of labor in a binary consortium

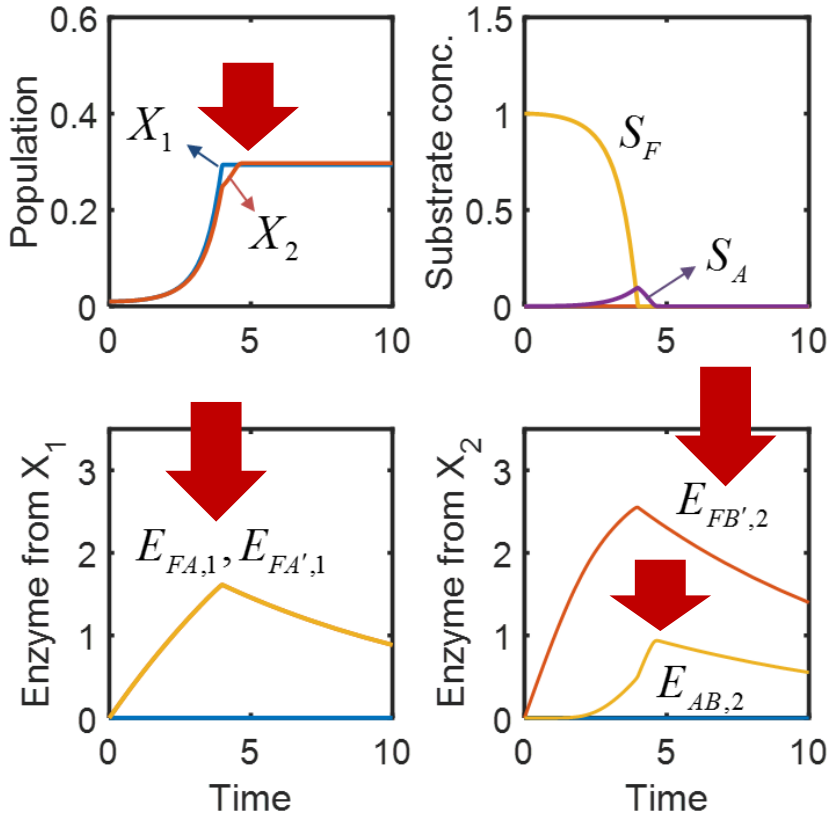




- The primary degrader (X_1) wins the competition
- X_1 proactively prepares for the consumption of hydrolysis products

Cross-feeding in a binary consortium

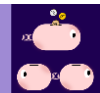




- X_1 constantly synthesizes equal amount of the two enzymes
- Initially, X_2 chooses to compete with X_1 for the hydrolysis product (S_F)
- As S_F becomes less available, X_2 avoids competition by taking S_A
- X_2 temporarily grows less during this transition, but catches up later

Concluding remarks of Part 1

- Microbe's direct and indirect survival strategies are well explained by economic behaviors of microorganisms maximizing return-on-investment (ROI)
- Cybernetic modeling uniquely accounts for metabolic ROI and optimal resource allocation to predict complex microbial dynamics
- Cybernetic modeling enables predicting microorganisms' social behaviors such as division of labor or cross-feeding from individualistic perspectives



Key references

April 2012 Vol. 58, No. 4

JOURNAL REVIEW

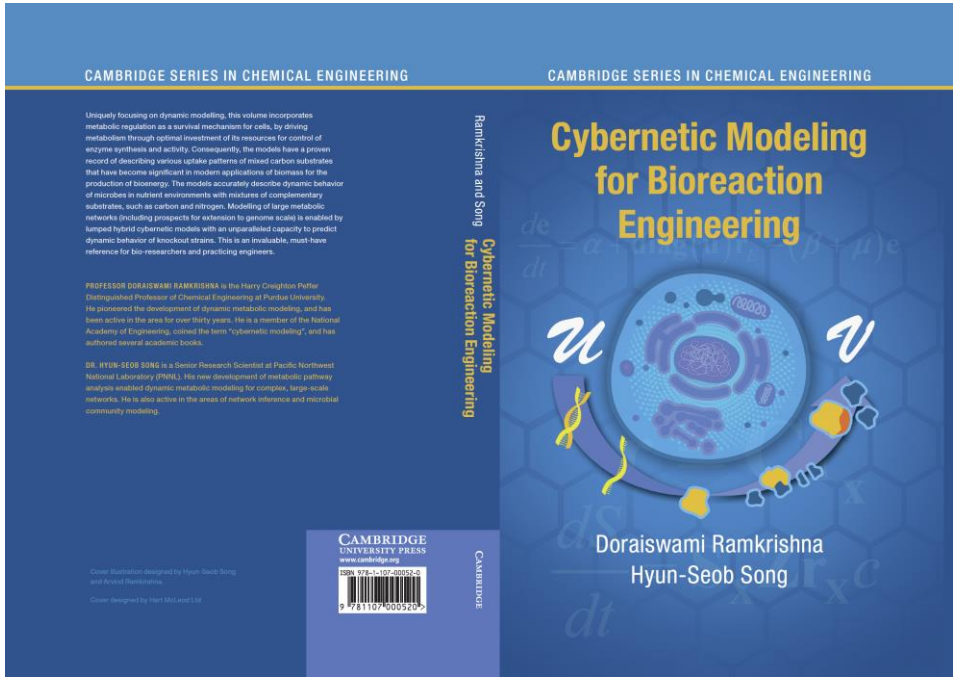
Dynamic Models of Metabolism: Review of the Cybernetic Approach

Doraiswami Ramkrishna and Hyun-Seob Song
School of Chemical Engineering, Purdue University, West Lafayette, IN 47907

Biotechnol. Prog. **2007**, *23*, 83–99

On the Matching and Proportional Laws of Cybernetic Models

Jamey D. Young† and Doraiswami Ramkrishna*

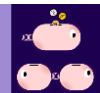


Cambridge University Press (2018)

Outline: Part 2

- Survival strategies of microorganisms
- Return-on-investment (ROI) in metabolism
- Cybernetic modeling
- Modeling of microbial interactions

- Modeling of eukaryotic systems
- Difficult to model the whole organism. Suitably define “subsystems”.
- How does one view ROI in eukaryotes?
- How does find meaningful objectives for cybernetic modeling?
- Concluding remarks



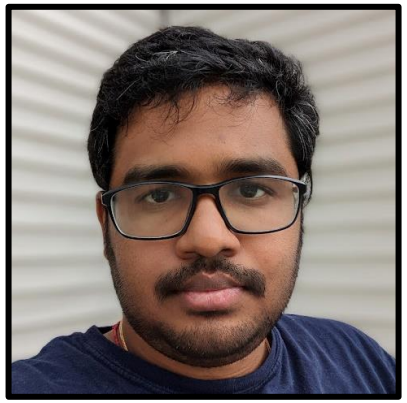
Cybernetic group members

Doraiswami Ramkrishna
Harry Creighton Peffer Distinguished Professor

PURDUE CHEMICAL ENGINEERING

Shankar Subramaniam
Distinguished Professor of Bioengineering, Bioinformatics and Systems Biology, Computer Science & Engineering, Cellular & Molecular Medicine and Nano Engineering

UC San Diego
JACOBS SCHOOL OF ENGINEERING



Rubesh Raja

Sana Khanum

Lina Aboulmouna

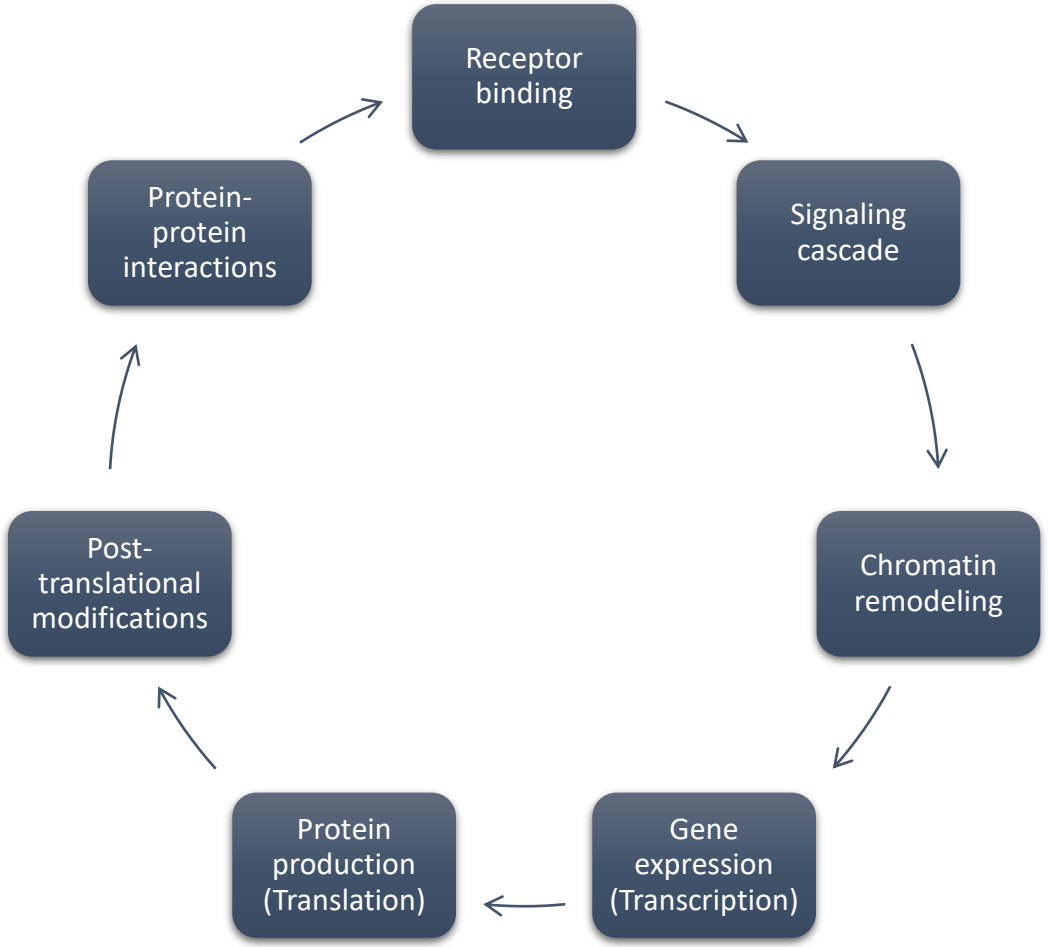
Shakti Gupta

Mano Maurya

Cybernetic modeling of eukaryotic systems

- Eukaryotic cells of complex organisms require **elaborate regulatory mechanisms** involving other **cell types** to maintain their cellular functions.
- Regulation in eukaryotic cells is structured and multifaceted, involving numerous molecules, pathways, and interactions.

- While survival is an important aspect for unicellular systems, such as bacteria, eukaryotic cellular systems can be thought to **optimize themselves towards their cellular functions** and coordinate with other cellular systems to improve the **organism's survival**.
- For example, by using an objective of maximizing certain inflammatory cytokines like TNF-α, our cybernetic models described arachidonic acid metabolism during early inflammatory response.



Cybernetic Regulation (General Considerations)

- **Investment of functional resources to realize a biological objective.**

- **Objective: Maximize the rate of some chosen biological function of system variables associated with the process under study.**

- **Identify all causal relationships between process variables.**
  - **This can be a vexing issue because the system dynamics is experimentally followed by time series measurements with no clear indication as to their causal relationship! This is particularly true of eukaryotic systems.**

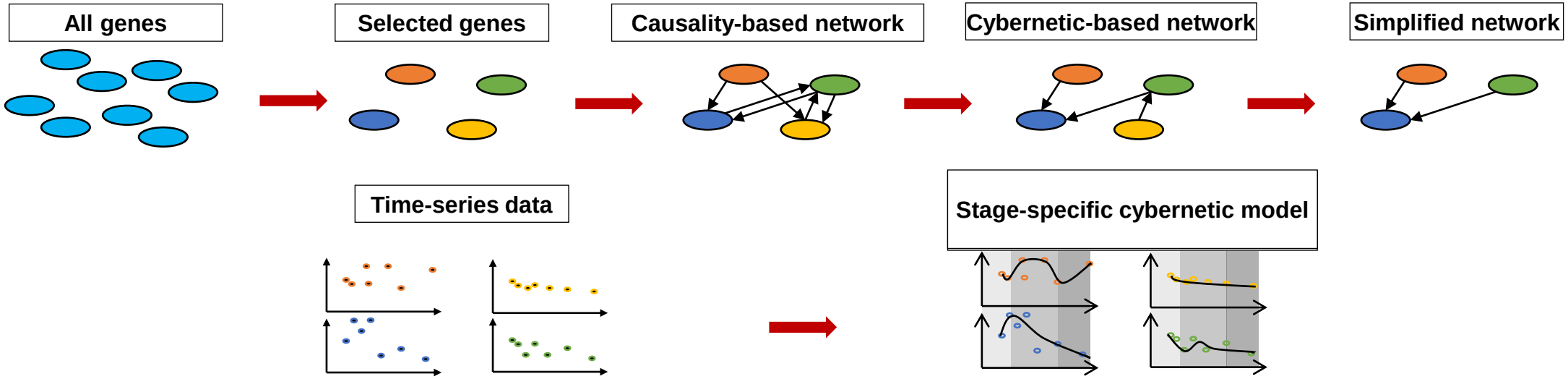
  - **Information-theoretic methods have become extremely important in resolving this issue.**

- **Once all causal relationships are known, we can formulate the**
  - **Kinetic model which is judged to be complete (without regulatory control) when the rate of some biological function, chosen as the objective can be calculated from the system variables.**

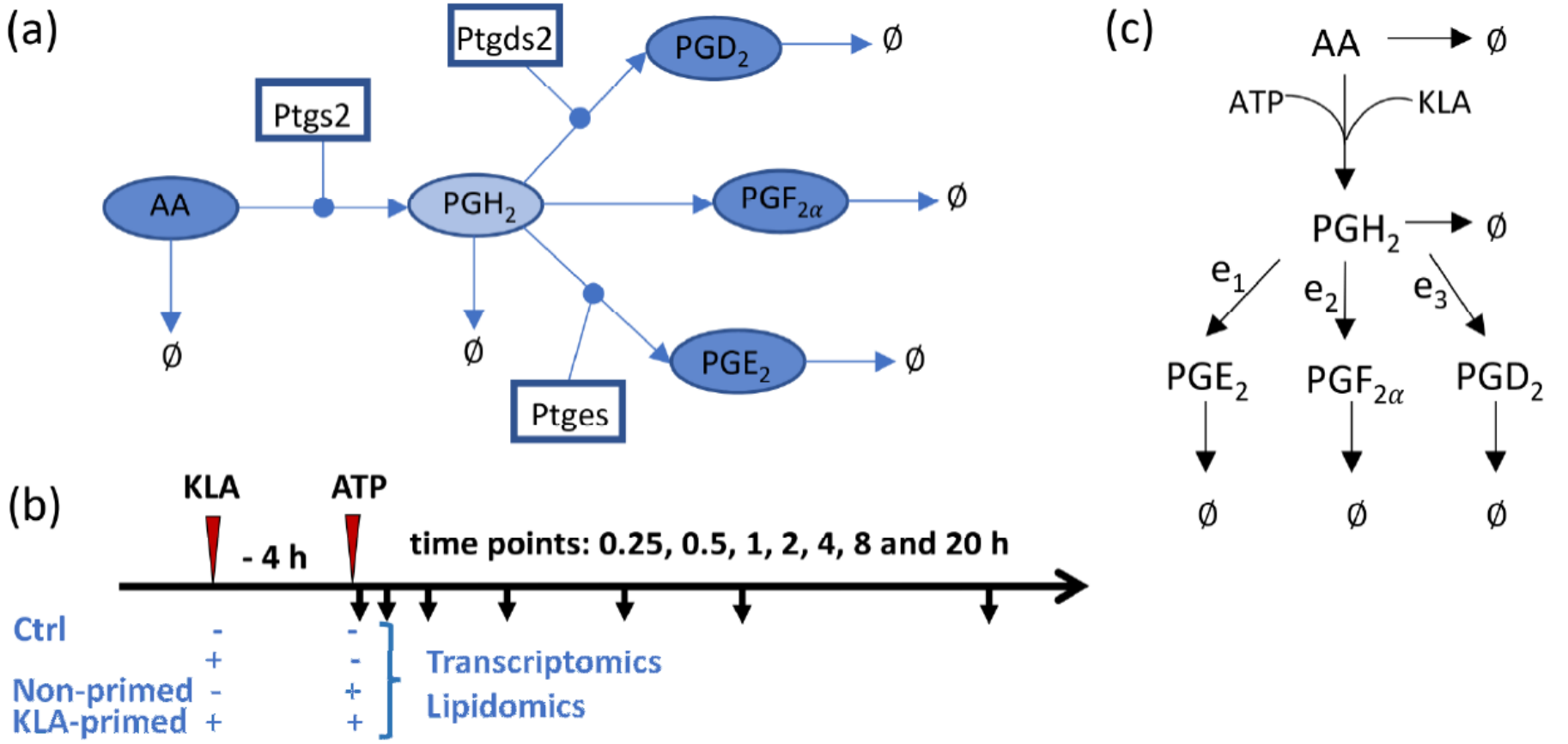
  - **the cybernetic regulation, based on the objective, can now be formulated to include regulatory control of the biological process.**

Model framework for eukaryotic cellular systems

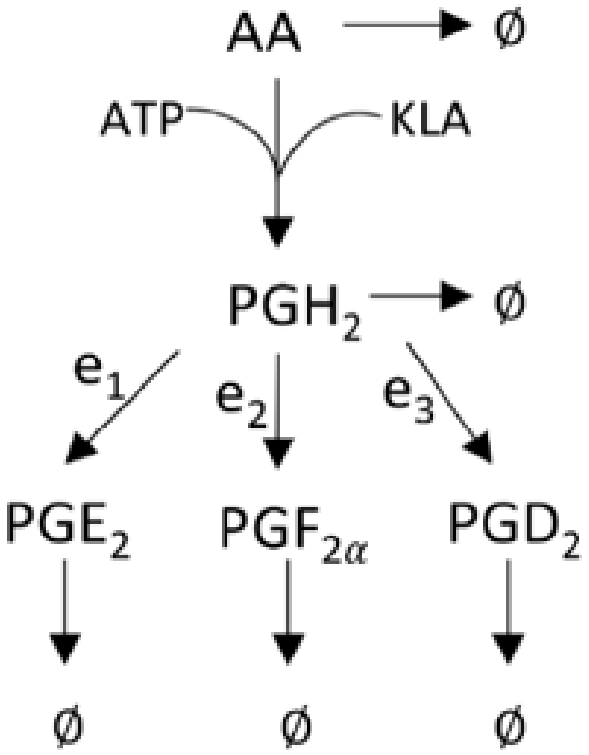
- We have developed a robust modeling framework to help us **dynamically build regulatory processes** in the form of a **network.**
- This is a multi-step framework and currently uses **transcriptomics time-series measurements.**
- Using our framework, we infer key molecules that can be systematically regulated for manipulating the system.
- To model the cellular processes like the cell cycle:
  - The model should incorporate the effects of **multi-level factors** such as signaling pathways, gene expression, protein-protein interactions, and post-translational modifications. These all play crucial roles in regulating cellular functions.
  - Additionally, the model should account for temporal changes, such as chromatin remodeling leading to **distinct cellular stages**.



Prostaglandin Metabolism in Bone Marrow-Derived Mouse Macrophage Cells Stimulated by KLA and ATP



Model Development



$$r_{AA\to PGH_2} = k_{PGH_2}\left[AA\right]\left\{1+k_{ATP}\left[ATP\right]+k_{KLA}\left[KLA\right]\right\}$$

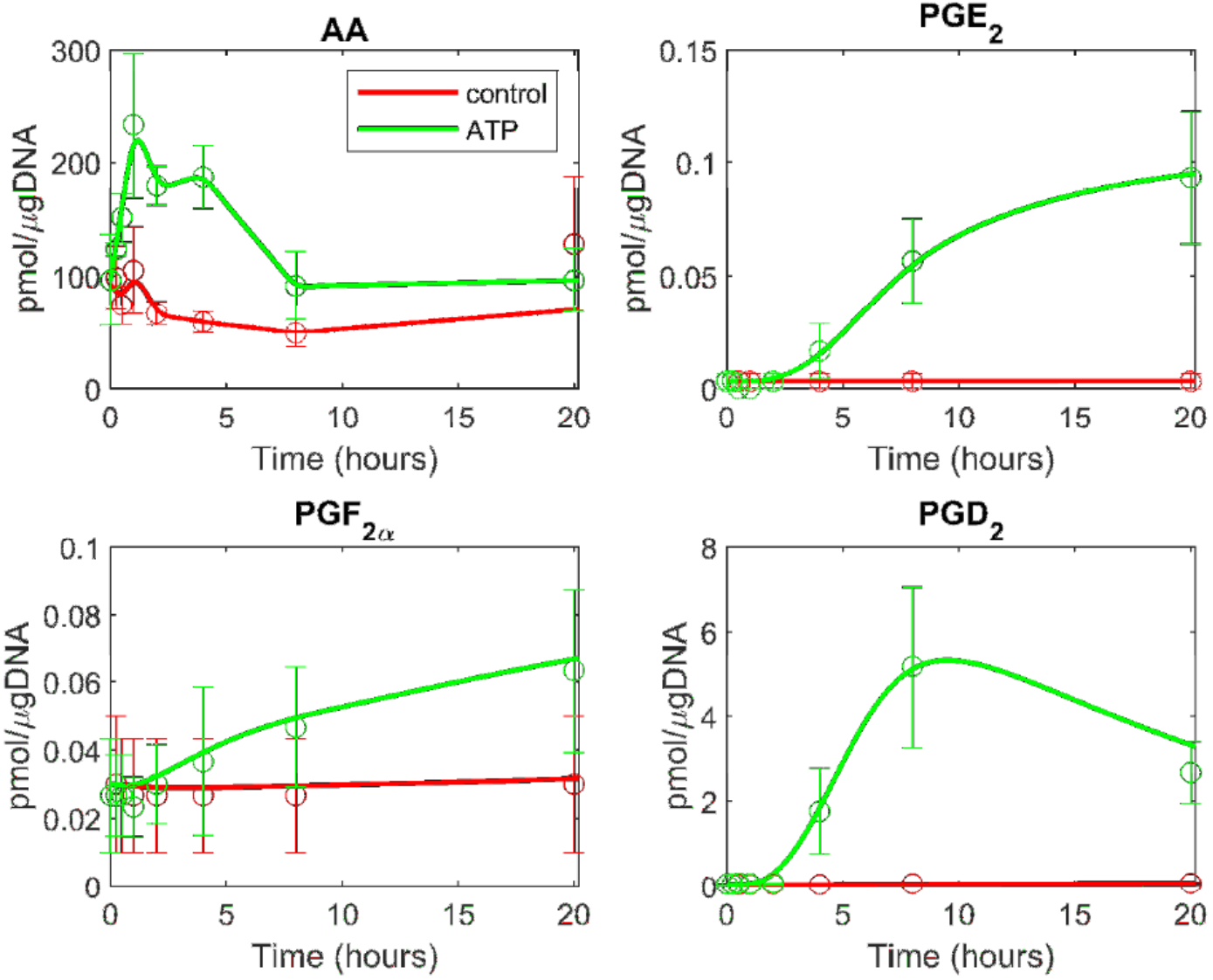
$$r^{kin}_{PGH_2\to PG_i} = e_i k_{PG_i}\left[PGH_2\right] \Rightarrow r^{reg}_{PGH_2\to PG_i} = v_i r^{kin}_{PGH_2\to PG_i}$$

$$\frac{d}{dt}\left[PGH_2\right] = r_{AA\to PGH_2} - r^{reg}_{PGH_2\to PG_i} - \gamma_{PG_i}\left[PGH_2\right]$$

$$r^{kin}_{e_i} = k_{e_i}\left[PGH_2\right] \Rightarrow r^{kin}_{e_i} = u_i r^{reg}_{e_i}$$

$$\frac{de_i}{dt} = \alpha + r^{reg}_{e_i} - \beta e_i \qquad u_i = \frac{\rho_i}{\sum_{k=1}^{3}\rho_k}; \qquad v_i = \frac{\rho_i}{\max_k \rho_k}$$

Fitted dynamic behavior of prostaglandin (PG) formation as an inflammatory response



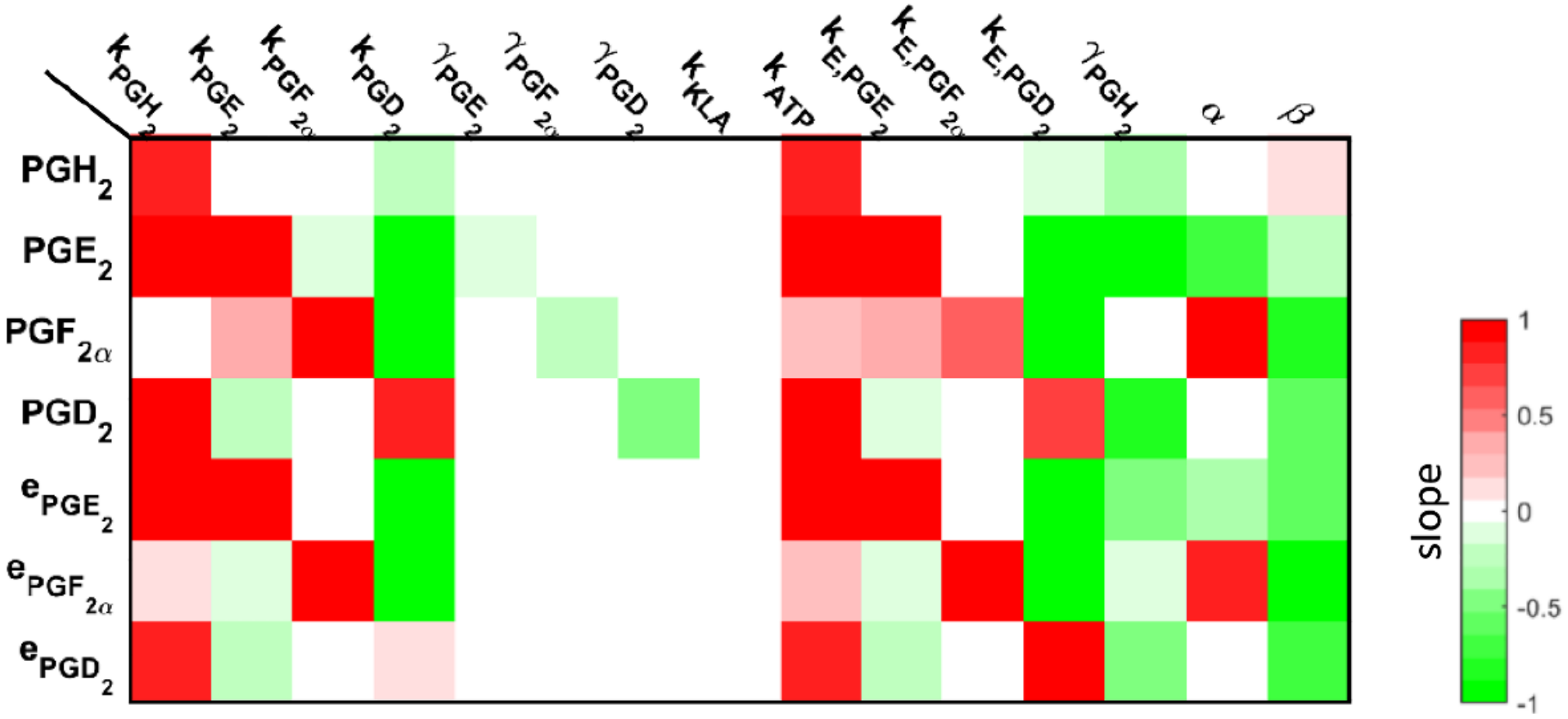
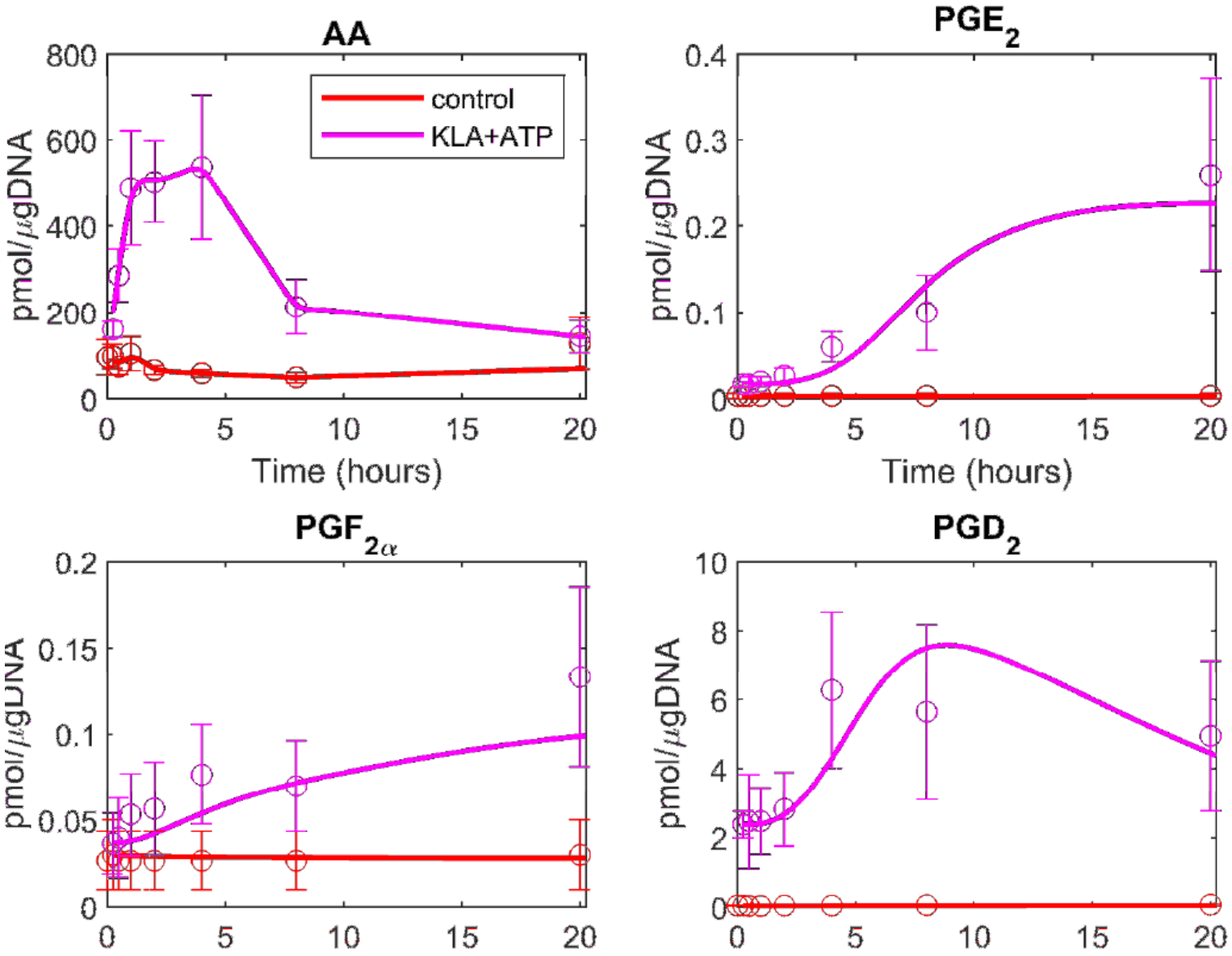
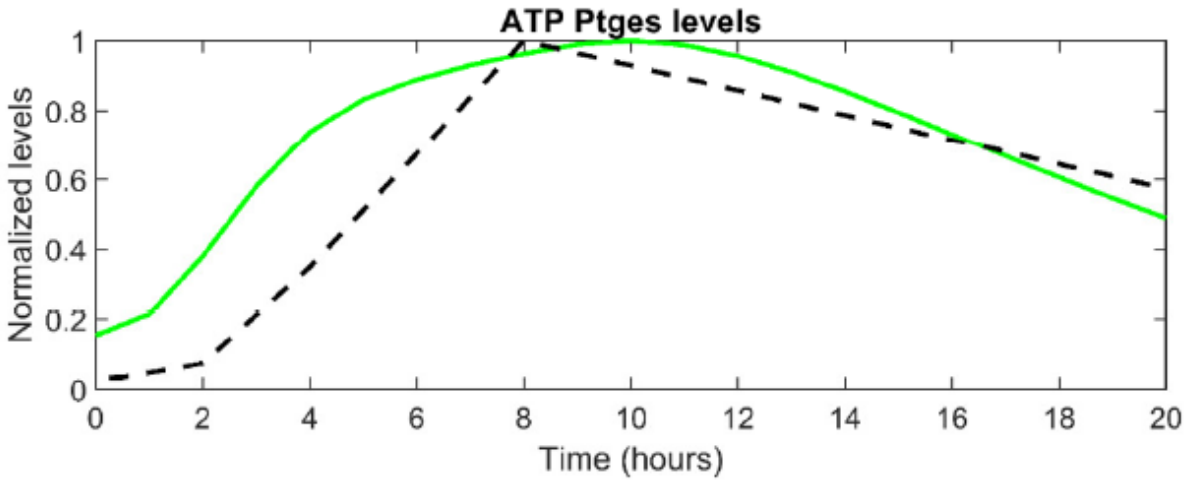


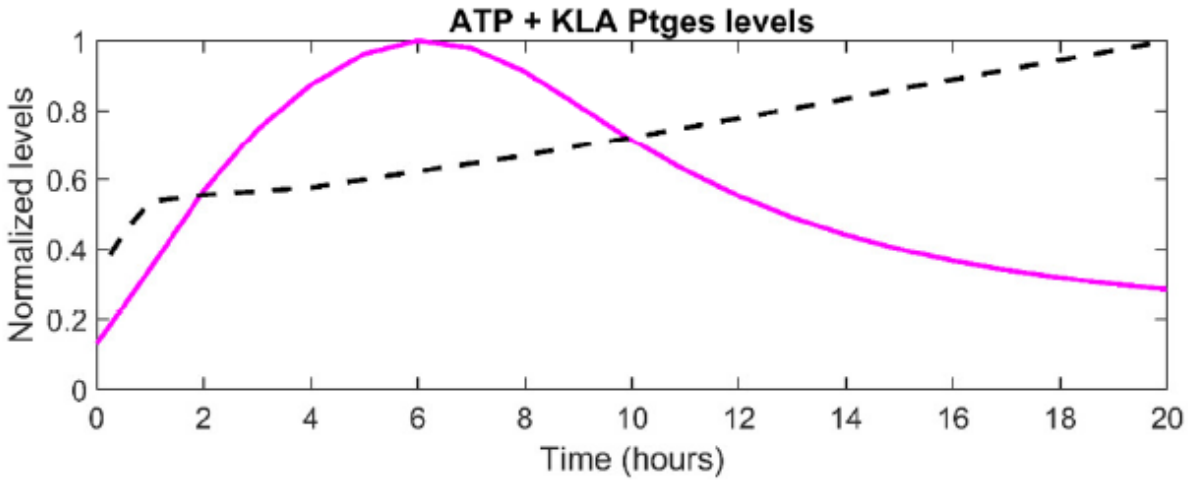
Figure 3. The slope of the sensitivity curves of the arachidonic acid (AA) metabolism are shown as a heat map. For example, the changes in the parameter associated with a conversion of AA into prostaglandin H2 (PGH_2) resulted in an increase in all of the metabolites; whereas, changes in the degradation of PGH_2 resulted in a decrease in all of the metabolites. This is expected, given that PGH_2 is in the upper part of the network, so the changes associated with these parameters will result in an impact on all of the corresponding downstream metabolites.

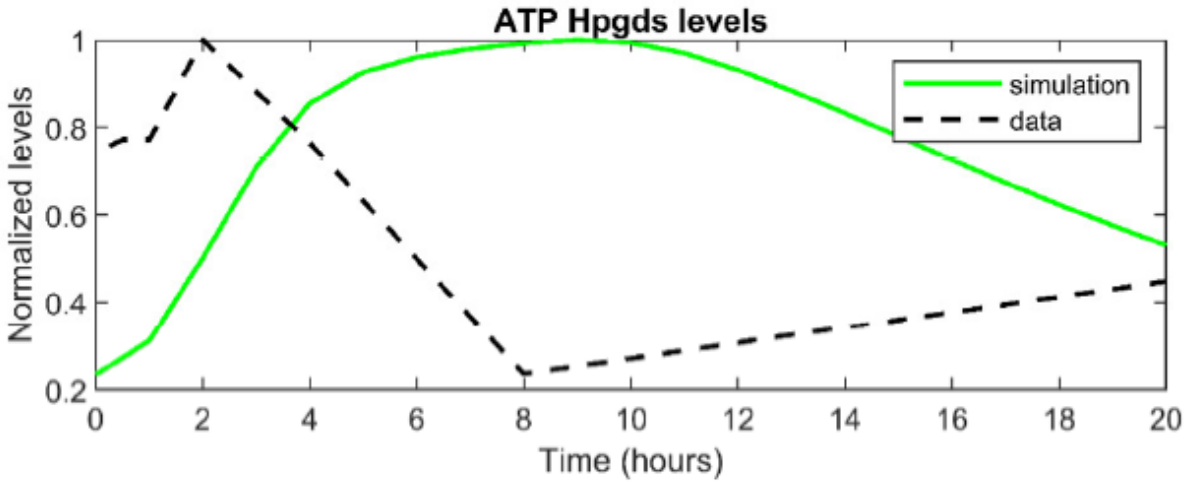
Model Predictions for Prostaglandin

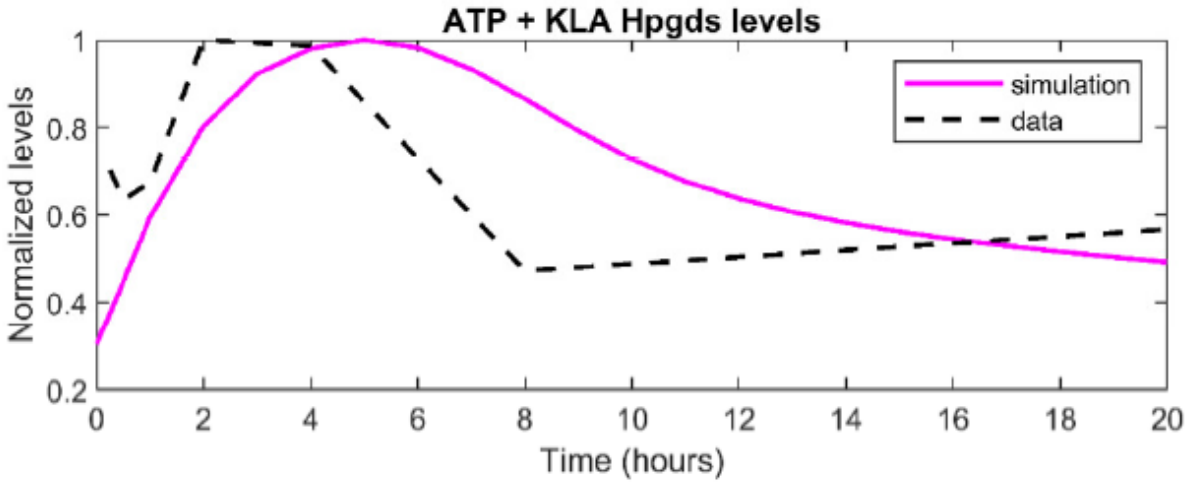


Comparison of scaled enzyme levels cybernetic model with experimental measurements.



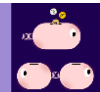






Conclusions

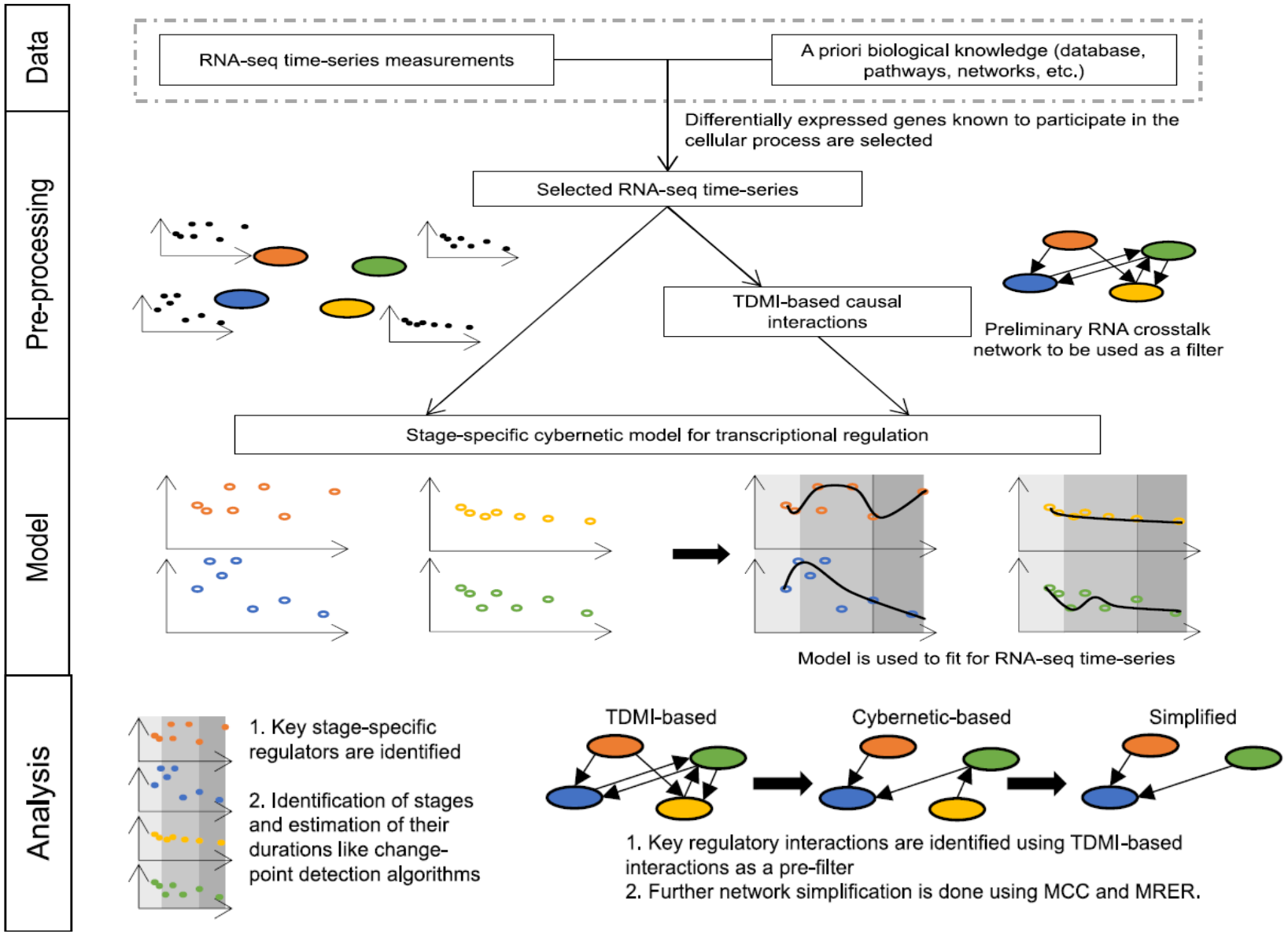
- The cybernetic model provides a robust description of metabolite formation and can be used to predict perturbations to eicosanoid metabolism.
- Our computational model assists in understanding eicosanoid metabolism's complexity and examining complex regulatory phenomena.



More Complex Issues in Modeling of Regulation of Eukaryotic Systems

- Many eukaryotic systems have phase-specific behavior with varying gene regulatory features.

Cybernetics Inspired Model (CIM) Framework for Transcriptional Regulation



Raja et al., Biophys.J, 2024
https://doi.org/10.1016/j.bpj.2023.12.010

Modeling Transcriptional Regulation

Expression rate constant

$$\frac{dRNA_j}{dt} = v_j k_j^r g_j - \gamma_j RNA_j, \qquad \frac{dg_j}{dt} = \alpha + u_j \max\left(k_{i,j}^g RNA_j, 0\right) - \beta g_j$$

Interaction parameter

Cybernetic Objective:

$$\text{Maximize: } \sum_{j=1}^{n} w_j k_j^r g_j,\ w_j \geq 0,\ \sum_{j=1}^{n} w_j = 1 \quad \Rightarrow \quad u_i = \frac{w_i k_i^r}{\sum_{j=1}^{n} w_j k_j^r},\ v_i = \frac{w_i k_i^r}{\max\left\{w_j k_j^r\right\}}$$

Concluding Remarks

- We have shown preliminary extensions of the cybernetic framework for eukaryotic systems.
- Multiple objective functions are essential to handle phase-specific changes in cellular behavior.
- The cybernetic formulation can decipher the most important gene interactions from data.
- Cell cycle systems can be modeled showing phase-specific gene interactions from transcriptomic data.
- A cybernetic regulatory oversight framework is under development to handle more complex regulatory patterns.