Spatio-temporal models of metabolism in microbial communities

Daniel Segrè
Dept. of Biology
Dept. of Biomedical Engineering
Dept. of Physics
Bioinformatics Program
http://www.bu.edu/segrelab/
We depend on microbes and their metabolism
Microbes depend on each other

Kolenbrander et al., Nature Rev. Microbiol., 2010
...often through metabolic exchange

Kolenbrander et al., Nature Rev. Microbiol., 2010
Each microbe faces a complex resource allocation problem
Can the network of interactions between microbes be understood (computationally predicted) based on networks within them?
Can the network of interactions *between* microbes be understood (computationally predicted) based on networks *within* them?

Quantitative predictions of community interactions and dynamics based on genomes

Understanding the metabolic interdependences, unculturability, diversity

Engineering of environments or consortia capable of a given task (e.g. shifting the disease/health balance, producing biofuels)
Can metabolism recapitulate the order of colonization of a biofilm?

Kolenbrander et al, 2010

Mazumdar et al., Metabolic Proximity in the Order of Colonization of a Microbial Community, PLOS1, 2013
Can metabolism recapitulate the order of colonization of a biofilm?

Varun Mazumdar

Pairwise Metabolic Distance

\[ J_{Dist}(A, B) = 1 - \frac{|A \cap B|}{|A \cup B|} \]

Organism A: 1 0 0 1
Organism B: 0 0 1 1

Mazumdar et al., Metabolic Proximity in the Order of Colonization of a Microbial Community, PLOS1, 2013
Can metabolism recapitulate the order of colonization of a biofilm?
Can metabolism recapitulate the order of colonization of a biofilm?

Mazumdar et al., Metabolic Proximity in the Order of Colonization of a Microbial Community, PLOS1, 2013
The real order of colonization has significantly smaller average metabolic distance compared to random.

**ORDER-PRESERVING PATH**

**RANDOM PATH**

- Average Pairwise Metabolic Distance
- Fraction

P-value $\sim 10^{-4}$
(Kolmogorov Smirnov Two Sample Test)
Possible implications and limitations

Metabolism matters in the order of colonization of a biofilm (much weaker signal with non-metabolic genes)

Does this reflect an environmental gradient or inter-species interactions?

Paradox: what prevents a “collapse” into minimal distance boredom? Competition? Also...
Synergy as a function of metabolic overlap predicts an optimum (sweet spot)

\[
\frac{EM(1+2)}{EM(1)+EM(2)}
\]

Relative increase in number of flux modes

Mazumdar et al., PLOS1, 2013
Towards synthetic ecology of microbes

Synthetic cooperation in engineered yeasts

Shou, Ram and Vilar, PNAS 2007

Metabolic synergy in *E. coli* mutants

Wintermute & Silver, MSB 2010
Cellular metabolism as a resource allocation problem (flux balance analysis, FBA) – No kinetics, only fluxes

1. Steady State
   \[ V_1 = V_2 + V_3 \]

2. Capacity constr.
   E.g.: \( V_{\text{GLUC}} < 10 \text{ mmol/gr·h} \)

3. Optimization (LP)
   E.g.: max \( V_{\text{growth}} \)

---

Can we use genome scale flux balance to predict (and perhaps design) microbial consortia based on cross-feeding?

Multi-compartmentalized FBA (Stolyar, Stahl)

± Compartment EFM-based analysis (Carlson)

Multispecies dynamic FBA (dFBA) (Mahadevan)

Multilevel programming (OptCom) (Zomorrodi, Maranas)

...
Inducing metabolic cross-feeding by engineering the environment, rather than the microbes

Klitgord and Segre’, PLoS Computational Biology 2010
Spatio-temporal modeling without a priori assumptions on interactions

Computation of Microbial Ecosystems in Time and Space (COMETS)

Bill Riehl  Will Harcombe  Chris Marx

Ilija Dukovski, Brian Granger, Pankaj Mehta, ...

http://comets.bu.edu  Harcombe et al., Cell Reports, 2014
$B_i =$ Biomass of species $i$
$C_j =$ Amount of metabolite $j$
$U_{ij} =$ Uptake rate of metabolite $j$ in species $i$

$U_{ij}^{\text{MAX}}$

Step-wise dFBA dynamics

Diffusion constants $D_j$

Global spatio-temporal dynamics
\[ B_i = \text{Biomass of species } i \]
\[ C_j = \text{Amount of metabolite } j \]
\[ U_{ij} = \text{Uptake rate of metabolite } j \text{ in species } i \]

FBA

Step-wise dFBA dynamics

Diffusion constants \( D_j \)

Global spatio-temporal dynamics
Recapitulating linear *E. coli* colony growth on different nutrients

### Table 1. COMETS Parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Uptake $V_{\text{max}}$</td>
<td>10 mmol/g/hr</td>
<td>Gosset, 2005</td>
</tr>
<tr>
<td>Uptake $K_m$</td>
<td>10 μM</td>
<td>Gosset, 2005</td>
</tr>
<tr>
<td>Death rate</td>
<td>1%</td>
<td>Saint-Ruf et al., 2004</td>
</tr>
<tr>
<td>Metabolite diffusion</td>
<td>$5 \times 10^{-6}$ cm$^2$/s</td>
<td>Stewart, 2003</td>
</tr>
<tr>
<td>Biomass diffusion</td>
<td>$3 \times 10^{-9}$ cm$^2$/s$^a$</td>
<td>Korolev et al., 2011</td>
</tr>
<tr>
<td>Max. colony height</td>
<td>200 μm</td>
<td>Lewis and Wimpenny, 1981</td>
</tr>
<tr>
<td>Oxygen concentration</td>
<td>250 μmol/cm$^2$</td>
<td>Peters et al., 1987</td>
</tr>
</tbody>
</table>

Data from Lewis and Wimpenny, 1981
Applying COMETS to synthetic microbial consortia

Harcombe, Evolution 2010
Consortium converges to predictable proportions from different initial states

(Methionine secretion coupled to biomass)
Extension to a newly engineered 3-species consortium

Diagram: 
- Lactose → E. coli ΔmetB → S. enterica* → M. extorquens ΔhprA → Acetate → NH₃ → Methylamine → Lactose

Graphs: 
- Initial vs. After 5 transfers for species percentage in community.

Legend: 
- COMETS Experiment

Species percentage in community:
- 0% 25% 50% 75% 100%
Metabolic synergy is distance dependent
Does a competing mutualist help or harm?

- Lactose
- Acetate
- Methionine

- E. coli ΔmetB
- S. enterica*
Does a competing mutualist help or harm?
Does a competing mutualist help or harm?

Eclipse?
Does a competing mutualist help or harm?

Harcombe et al., Cell Reports, 2014
Using COMETS to explore putative interaction mechanisms

Growth rate

Ace (conc)

Ace (flux)

Lactose

Acetate

Methionine

E. coli ΔmetB

S. enterica*

Uptake 0 Secretion

30 50 70 90 110

Time (h)
Towards more accurate microbial growth models in COMETS: Spatial structure and metabolic heterogeneity

Improved COMETS biomass expansion algorithm

Growth above critical density

Pressure

Gradient of pressure $\rightarrow$ Velocity

Convection-diffusion equations

Fluctuations-induced instabilities
Mapping metabolite exchange in small ecosystems (COMETS+VisANT)

Granger et al., submitted
... and large ones
Challenges and opportunities

COMETS is freely available, open source. Easy to implement mutants, complex media, many species (scales linearly). Species uploadable from SBML, COBRA, DOE KBase. New features being added.

Ongoing applications: unculturability and interactions in the oral microbiome, dynamics in health and disease; synthetic ecology for biofuel production.

Big open questions:
* Gene function ignorance is a major limiting step – Community initiatives needed (E.g. http://combrex.bu.edu)
* Theory of metabolism + regulation
* Bridging the gap with top-down approaches (metagenomic data)
Group:
Brian Granger
Ilija Dukovski
Summer Zhuang
Chris Jacobs
Ali Zomorrodi
Amrita Kar
Arion Stettner
Meghan Thommes
David Bernstein
Josh Goldford
Demetrius Dimucci
Ramakrishna Sinadri

Former members
Bill Riehl
Niels Klitgord
Varun Mazumdar
Hsuan-Chao Chiu
David Byrne
Chris Jacobs
Evan Snitkin
Ed Reznik
Sara Baldwin
Qasim Beg
Lina Faller
Emma Briar

Collaborators: Trent Northen (LBNL), Pankaj Mehta (BU), Aimee Dudley (Inst. For Systems Biology),
Chris Marx (U. Idaho), Will Harcombe (U. Minnesota), Nathaniel Cady (Albany), Salomon Amar (BU),
Charles DeLisi (BU), Simon Kasif (BU), Martin Steffen (BU), Rich Roberts (BU), John Finnerty (BU),
Brandon Xia (BU), Claudio Altafini (SISSA), Mattia Zampieri (SISSA), Sid Redner (BU), Paul Krapiwsky
(BU), Oliver Ebenhoeh (U. Aberdeen), Moritz Schuette (Max Planck), Lars Angenent (Cornell), Jenny
Talbot (BU), Mo Khalil (BU)

Thank you
Metabolic Resource Allocation in Individual Microbes Determines Ecosystem Interactions and Spatial Dynamics

William R. Harcombe,1,7,8 William J. Riehl,2,7,9 Ilija Dukovski,2 Brian R. Granger,2 Alex Betts,1,10 Alex H. Lang,3 Gracia Bonilla,2 Amrita Kar,2 Nicholas Leiby,1,4 Pankaj Mehta,2,3 Christopher J. Marx,1,5,11,* and Daniel Segrè2,6,*

Cell Reports 7, 1104–1115, May 22, 2014
\[ UB_m^\alpha = \frac{V_\text{max}[C_m]^n}{[C_m]^n + K_M^{\alpha,m}} , \]

Maximize \[ Z^T v^\alpha \]
Subject to \[ S^\alpha v^\alpha = 0 \]
\[ LB_j^\alpha \leq v_j^\alpha \leq UB_j^\alpha \quad j = 1, \ldots, n \]

\[ B_{(x,y)}^\alpha(t + \Delta t) = B_{(x,y)}^\alpha(t) + B_{(x,y)}^\alpha(t) \cdot v_{\text{growth}}^\alpha \cdot \Delta t \]

\[ Q_{m(x,y)}(t + \Delta t) = Q_{m(x,y)}(t) + u_m^\alpha \cdot B_{(x,y)}^\alpha(t) \cdot \Delta t, \]
A note on FBA, metabolite secretion and transcriptional regulation

In current consortium, we have to force Salmonella to secrete methionine.

General Problem: FBA often fails to predict correctly secreted byproducts

Shewanella oneidensis

Qasim Beg, Mattia Zampieri et al., NAR 2012
Region 4: Bacteroides (phylum)

24%  Pentose and glucuronate interconversions
13%  Starch and sucrose metabolism
13%  Amino sugar and nucleotide sugar metabolism
12%  Other glycan degradation
  7%  Lysosome
  6%  Tryptophan metabolism
  6%  Sphingolipid metabolism
Extension to a newly engineered 3-species consortium
Table 2. A summary of various categories of community modeling approaches using genome-scale metabolic models.

<table>
<thead>
<tr>
<th>Modeling formalism</th>
<th>Modeling condition</th>
<th>Type of optimization problem</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Compartimentalized community-level metabolic models based on MOMA</td>
<td>Steady-state</td>
<td>Quadratic programming</td>
<td>Wintermute and Silver 20</td>
</tr>
<tr>
<td>[De-]Compartimentalized community-level metabolic models based on elementary mode analysis</td>
<td>Steady-state</td>
<td>NA</td>
<td>Taffs et al 208</td>
</tr>
<tr>
<td>Analysis of metabolic model-derived metrics quantifying the degree of cooperation and/or competition</td>
<td>Steady-state</td>
<td>NA</td>
<td>Zeleznia et al 209, Kreimer et al 210, Levy et al 211, Borenstein and Feldman 212, Christian et al 213</td>
</tr>
<tr>
<td>cFBA: Community FBA based on the balanced growth of microorganisms</td>
<td>Steady-state</td>
<td>Linear/Nonlinear programming</td>
<td>Khandelwal et al 297</td>
</tr>
<tr>
<td>OptCom: Multi-level and multi-objective modeling</td>
<td>Steady-state</td>
<td>Nonlinear programming</td>
<td>Zomorrodi and Maranas 198, El-Samman et al 215</td>
</tr>
<tr>
<td>DMNM (Dynamic Multi-species Metabolic Modeling) based on the extension of dynamic FBA for single species</td>
<td>Dynamic</td>
<td>Linear programming</td>
<td>Zuang et al 191, Salimi et al 216, Hanly and Henson 218, 219, 221, Tzamali et al 220, Chiu et al 223</td>
</tr>
<tr>
<td>d-OptCom: Multi-level and multi-objective dynamic metabolic modeling</td>
<td>Dynamic</td>
<td>Nonlinear programming</td>
<td>Zomorrodi et al 193</td>
</tr>
<tr>
<td>COMETS (COmputational modeling of Microbial Ecosystems in Time and Space): Direct integration of dynamic FBA for communities and diffusion models</td>
<td>Spatiotemporal</td>
<td>Linear programming</td>
<td>Harcombe et al 194</td>
</tr>
</tbody>
</table>

Compartmentalized FBA (Stolya, Stahl)

± Compartment EFM-based analysis (Carlsson)

Multispecies dFBA (Mahadevan)

Multilevel programming (OptCom) (Zomorrodi, Maranas)
Cellular metabolism as a resource allocation problem (flux balance analysis, FBA)

1. Steady State
   \[ V_1 = V_2 + V_3 \]

2. Capacity constr.
   E.g.: \[ V_{GLUC} < 10 \text{ mmol/gr·h} \]

3. Optimization (LP)
   E.g.: \[ \text{max } V_{growth} \]