Understanding metabolism as an electrical process

Orkun S Soyer

BEE Workshop
Warwick, 29 March 2019

OSS LAB

Bio Electrical Engineering
Innovation Hub @ Warwick

WARWICK
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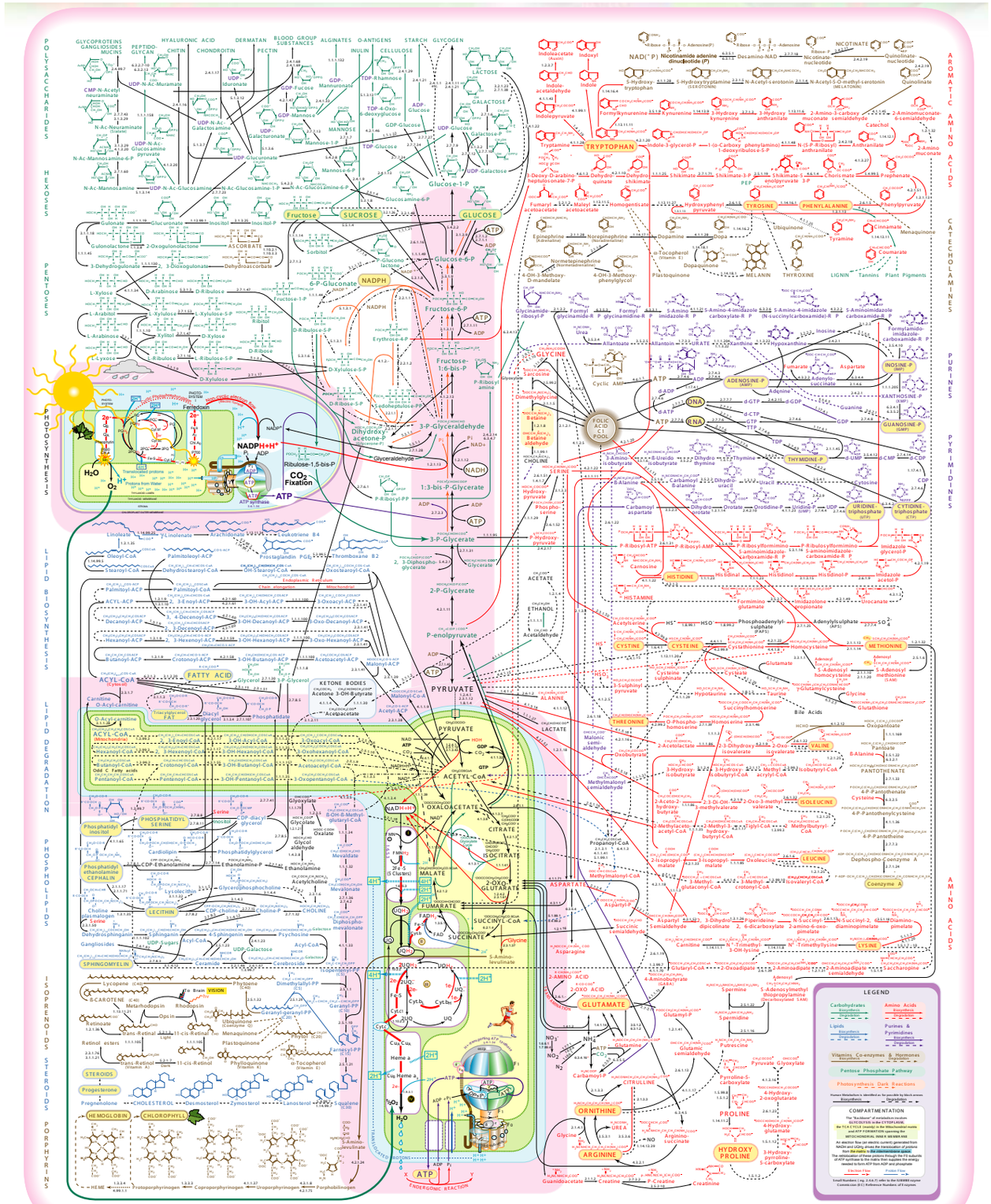
I'm interested in understanding cell metabolism / physiology



Metabolism is the process through which cells acquire energy **to make biomass**

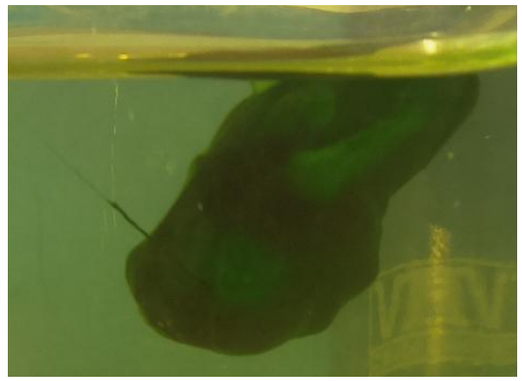
This is the dominant view driving the analysis of metabolism.

e.g; Flux Balance Analysis (FBA) implements biomass optimality, metabolic models consider pathways optimised separately for biomass, etc.

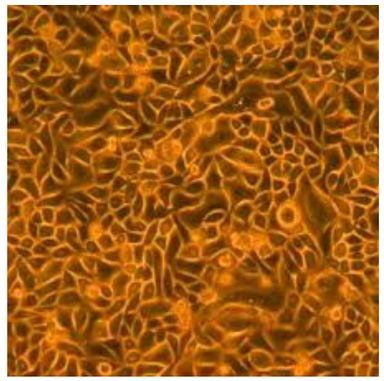
Metabolism before a cell

Selection? Selection on what?

Metabolism without growth



Environmental microbes



Healthy tissue

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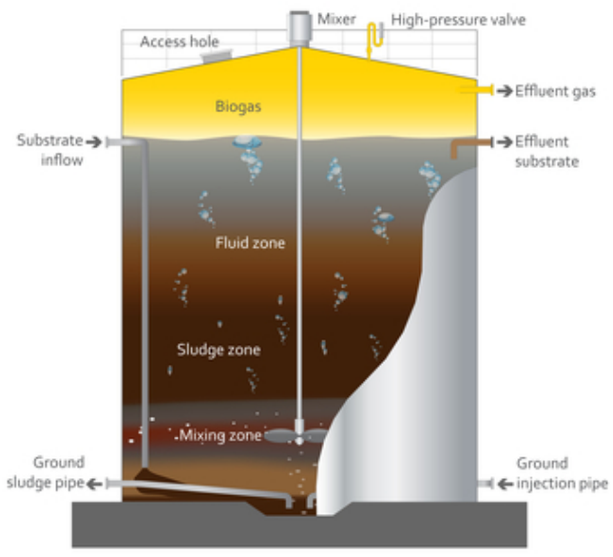
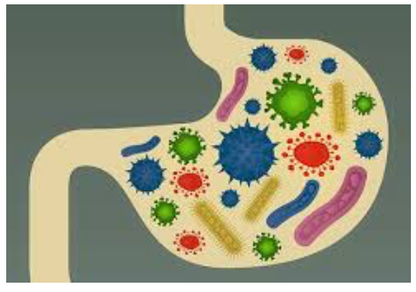
Metabolism across cells



Anaerobic digestion



Animal gut

….

Maybe we need an alternative <u>conceptual</u> view on metabolism?

Adaptive evolution

The dream of every cell is to become two cells

François Monod

Metabolism optimised for biomass production

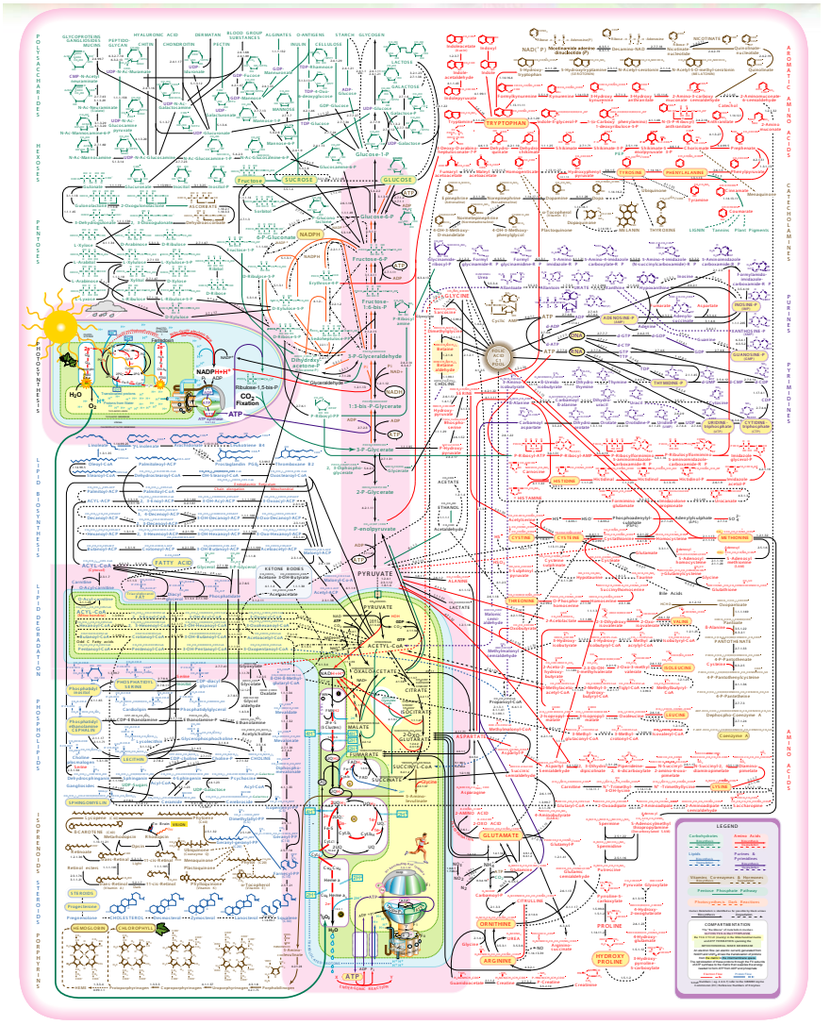
Metabolism invented biomass to stabilise a state of nonequilibrium thermodynamics

Neutral evolution

Life is an electron looking for a place to rest

Albert Szent-Györgyi

Metabolism as an electron flow system shaped by thermodynamic bottlenecks and biophysical tradeoffs

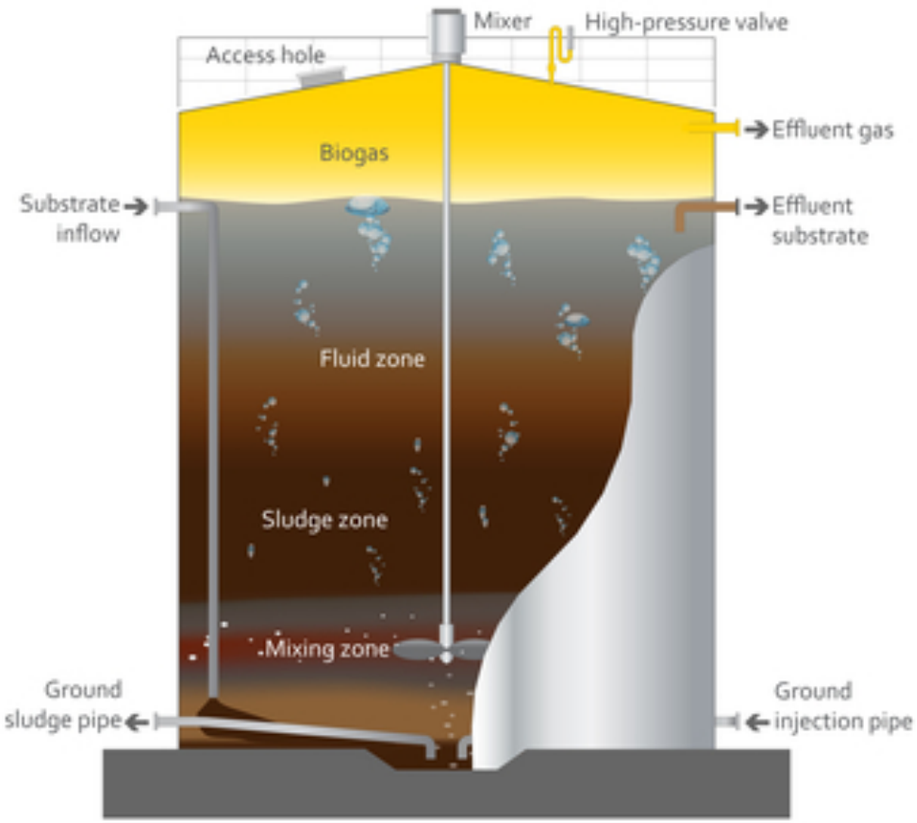


Interrogating metabolism as an electron flow system.

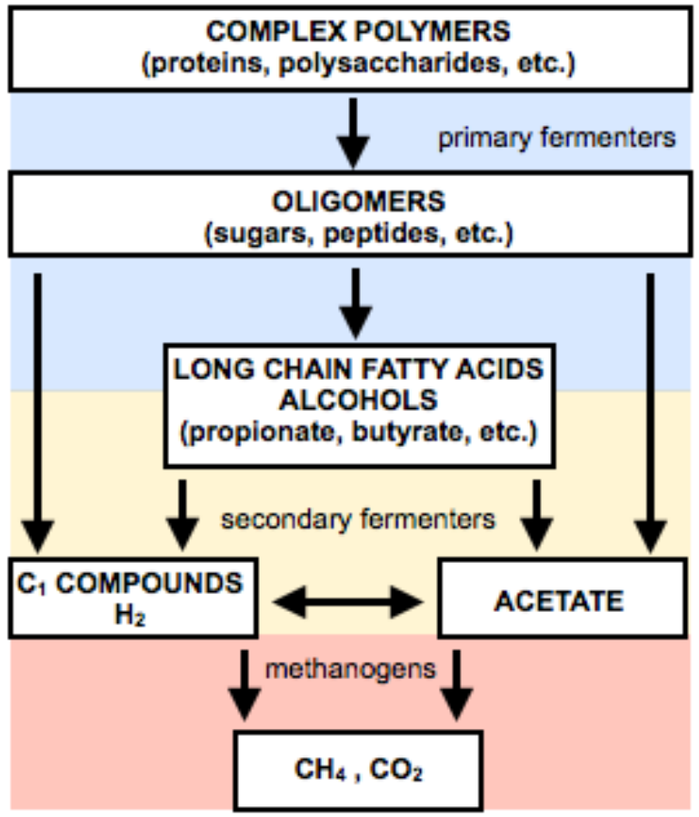
Christian Zerfass, Munehiro Asally, Orkun S Soyer

Current Opinion in Systems Biology *13: 59-67 (2019).*

Vignette 1: Thermodynamic bottlenecks



Anaerobic digestion

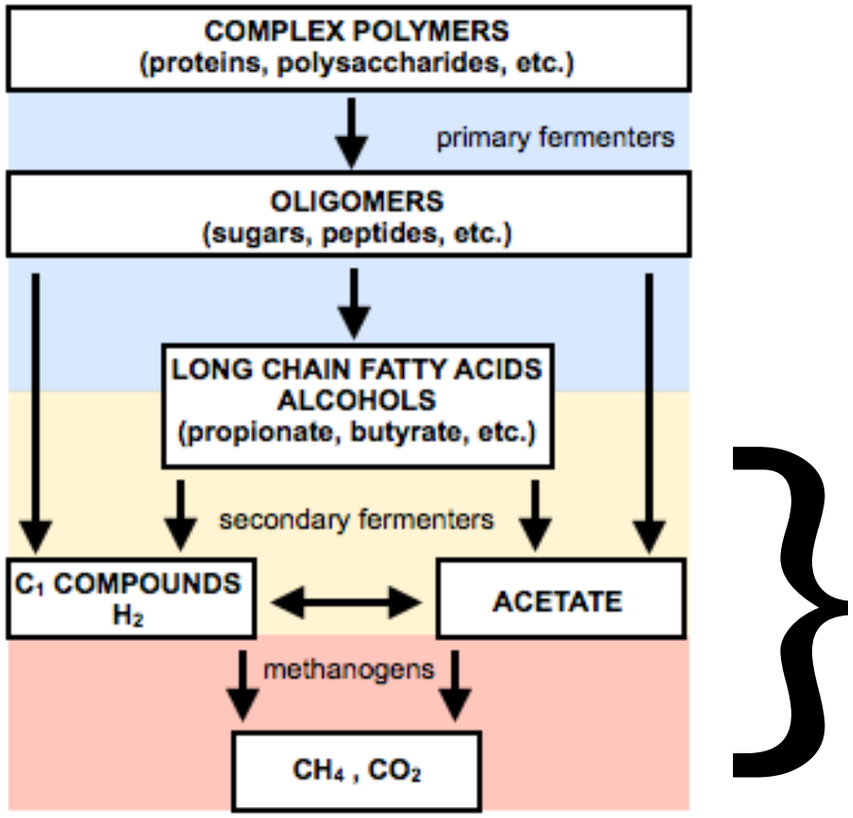


Schink B *Microbiol Mol Biol Rev* 61:2 (1997)

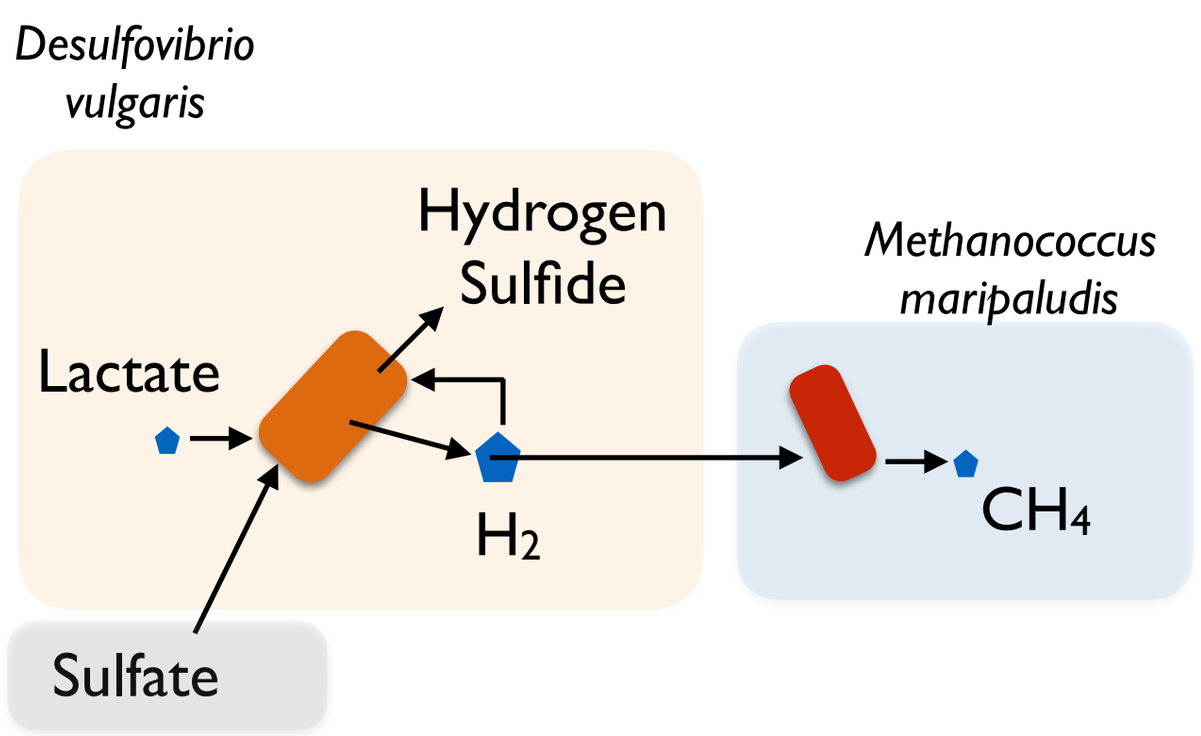
Engineer synthetic communities to learn about biochemical basis of communities

Grosskopf & Soyer, ***Curr. Op. Biotech*** (2014)

Syntrophy: Crucial in all AD systems

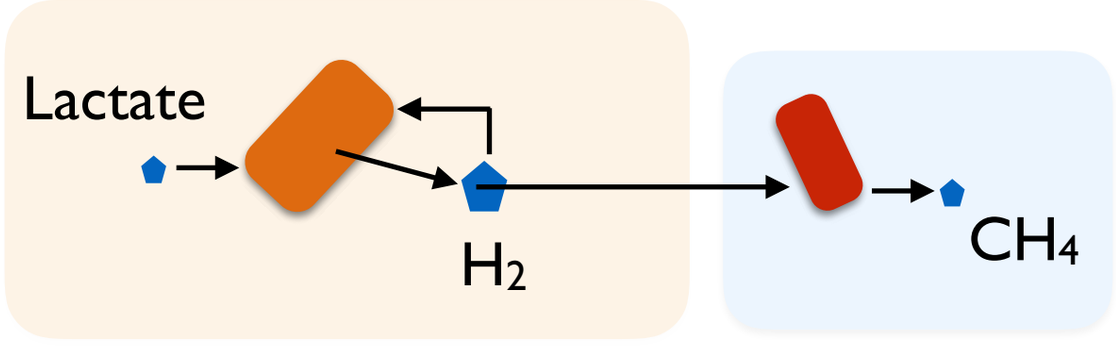


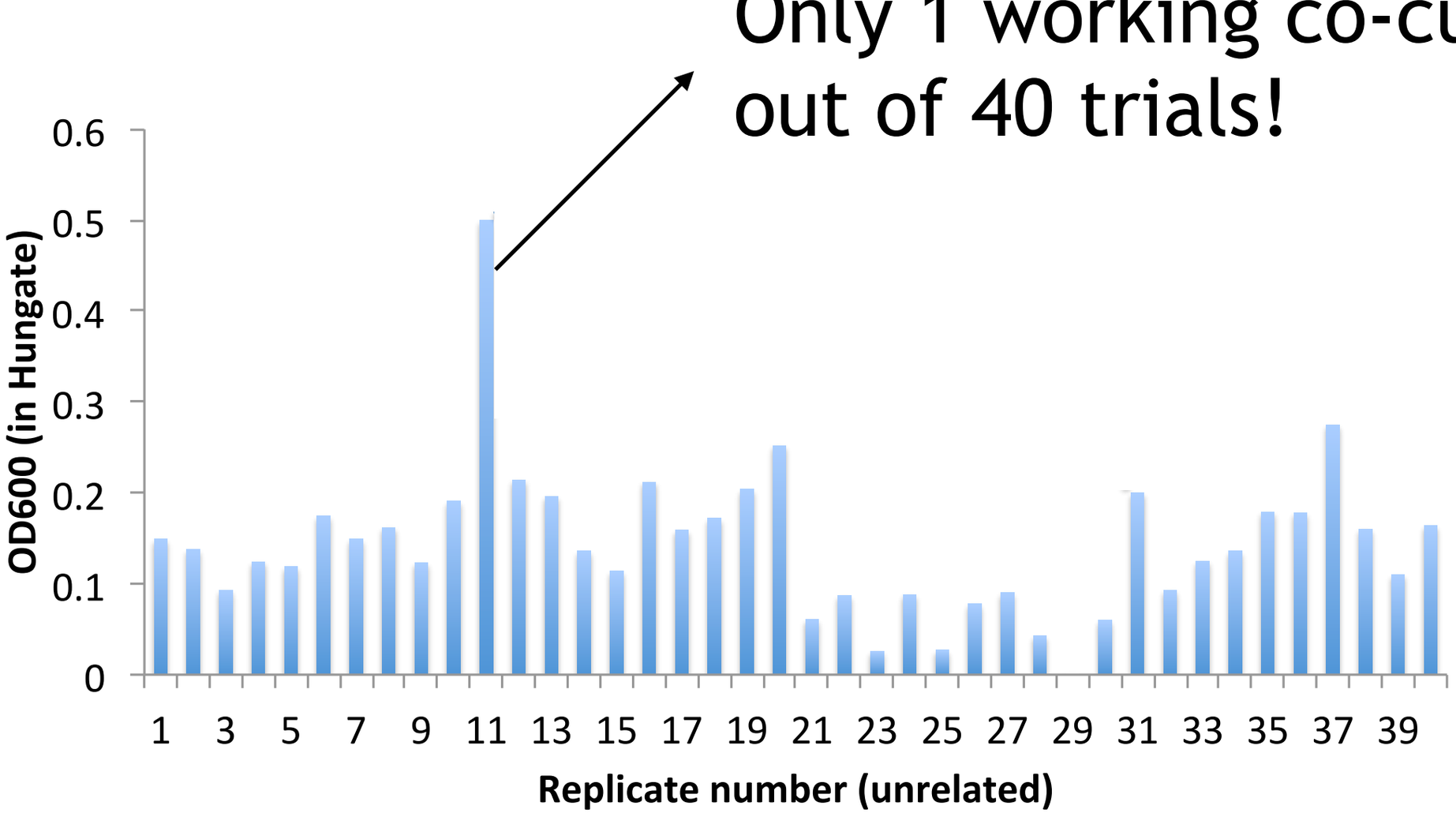
Schink B *Microbiol Mol Biol Rev* 61:2 (1997)



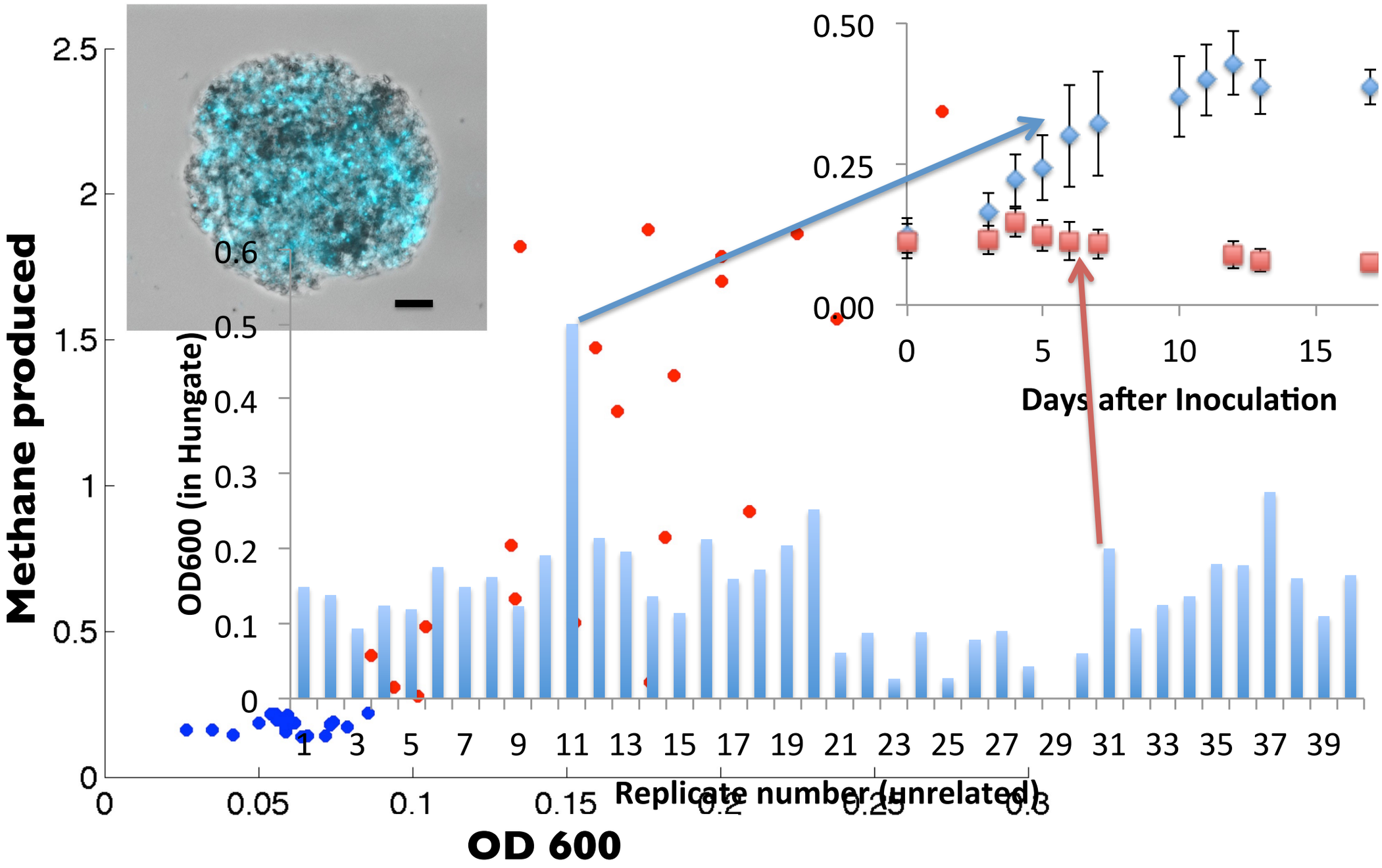
$$\text{Lactate} + 2\text{Sulfate} + 3H^{+} \longrightarrow 3CO_2 + 2HS^{-} + 3H_2O \qquad \Delta G_0 = -259.09 \text{ kJ/mol}$$

Model system to understand the basis of syntrophy:





Isolates from co-culture are consistently “syntrophic”, while those from wild type are not



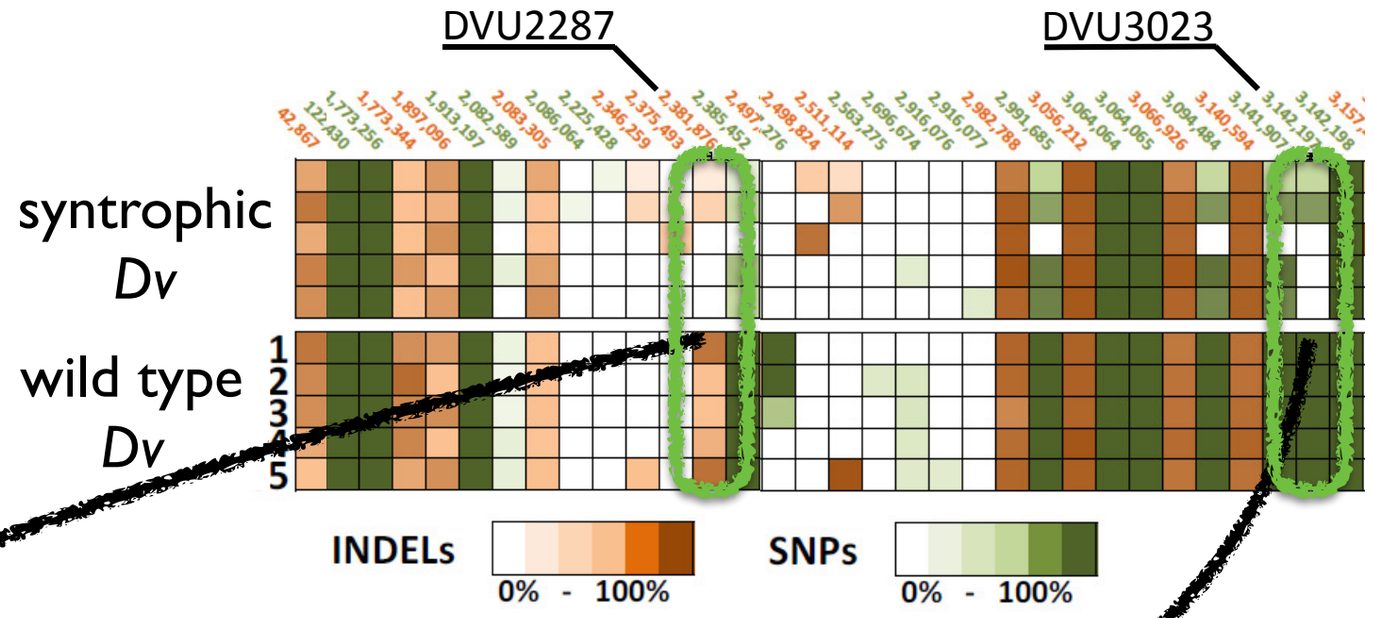
Syntrophy made possible by energy investment to overcome thermodynamics hurdle

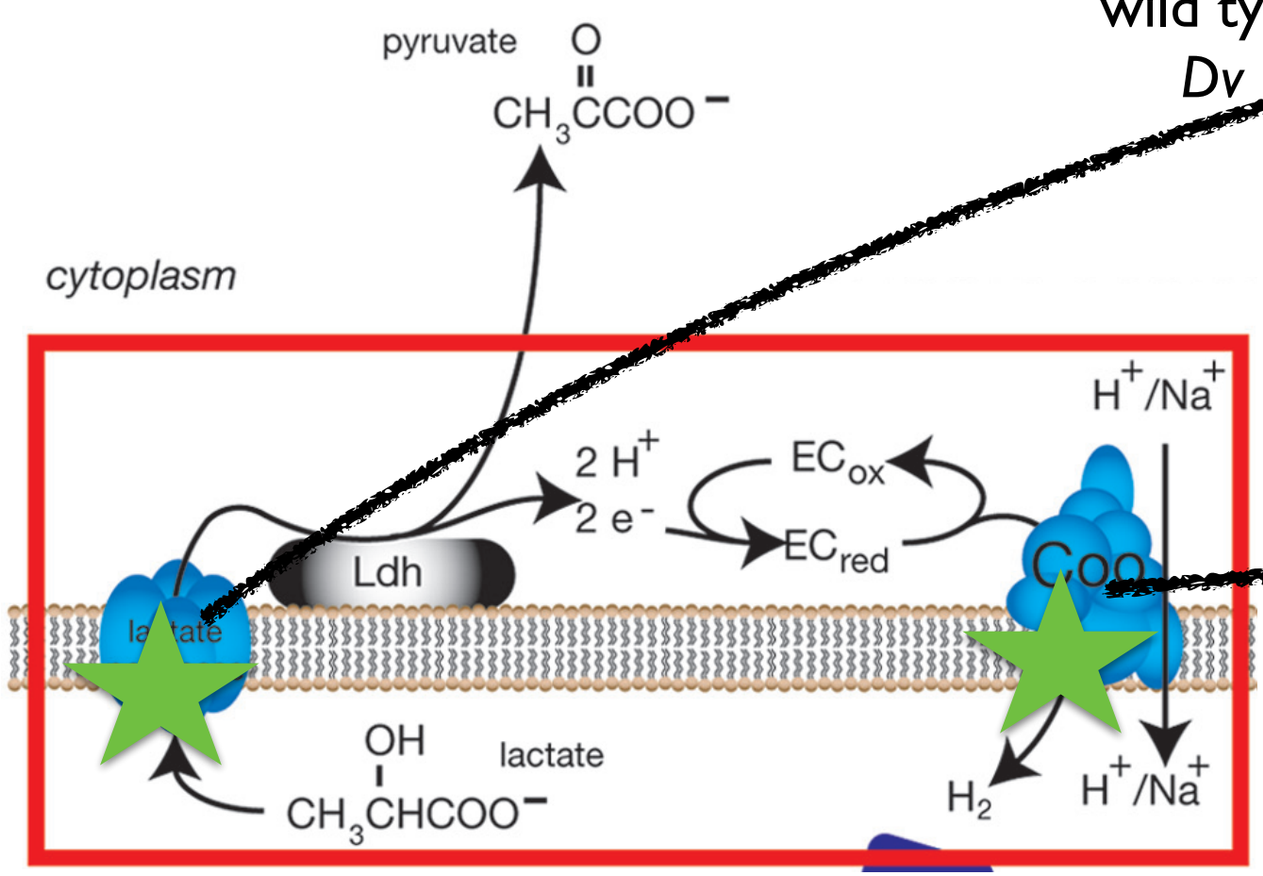
$$\text{Lactate} + H_2O \longrightarrow \text{Acetate} + CO_2 + 2H_2 \qquad \Delta G_0 = -8.79 \text{ kJ/mol}$$

?

$$\text{Lactate} \longrightarrow \text{Pyruvate} + H_2 \qquad \Delta G_0 = 43 \text{ kJ/mol}$$

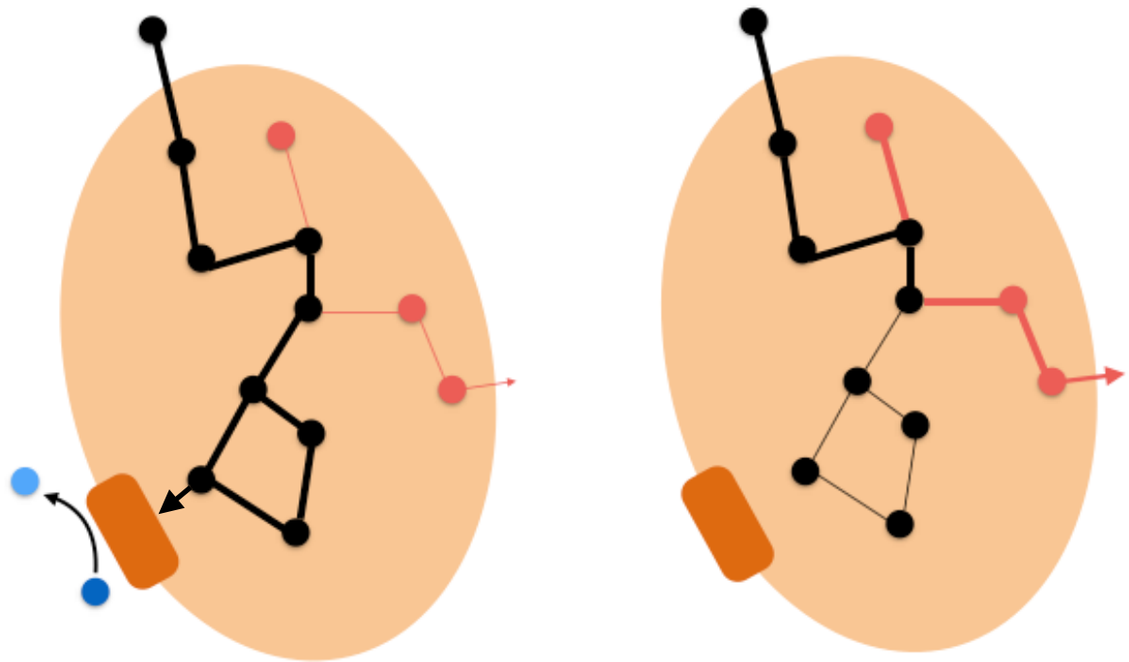
$$Na^+ \text{ (out)} \longrightarrow Na^+ \text{(in)}$$





Grosskopf T. et al. ***ISME J.*** (2016).

Vignette 1: Thermodynamic inhibition due to terminal electron acceptor availability can be a key driver of evolution of metabolic systems (intra- and inter-cellular)



Ethanol, Acetate, Lactate, Butyrate, H_2…

High energy respiratory pathways

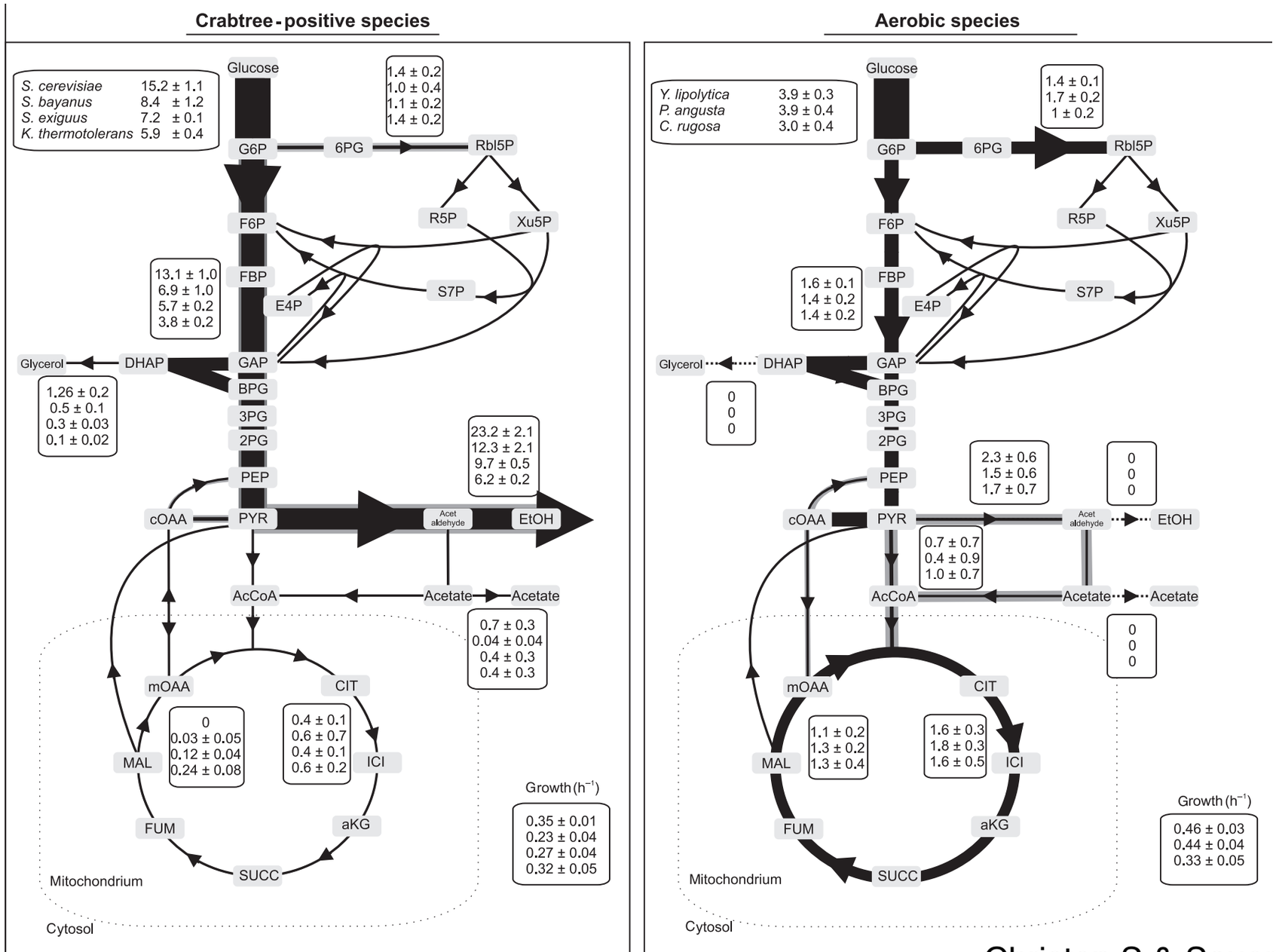
$$\Delta G_0 < -500 kJ / mol$$

Low energy fermentative pathways

$$\Delta G_0 > -300 kJ / mol$$

Test 1: Use electrodes as terminal electron acceptors to control metabolism via electrode potential

Vignette 2: Cellular trade-offs



Christen S & Sauer U, ***FEMS*** (2011)

Fermentation can still happen in the presence of terminal electron acceptor like oxygen

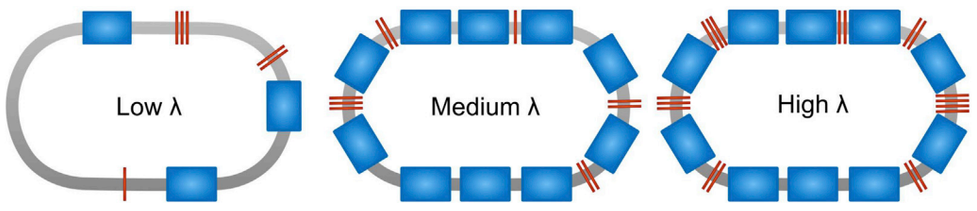
Trade-offs in cellular metabolism can <u>explain</u> respiro-fermentation (overflow metabolism):

Trade-offs in space/enzyme allocation

Szenk M, Dill KA, de Graff AMR, *Cell Systems 5* (2017)

Basan M, et al., *Nature 528* (2015)

….

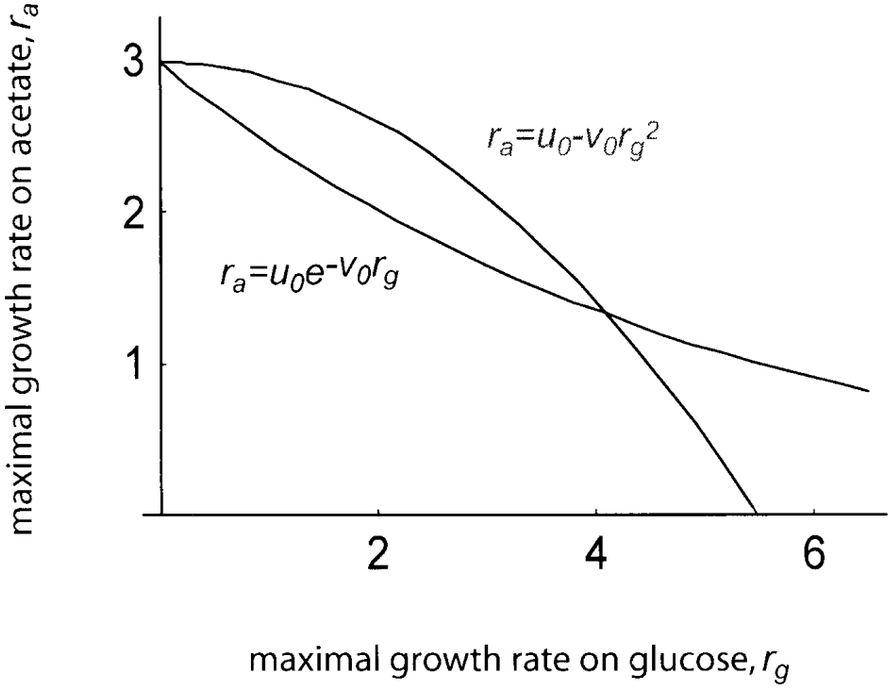


Trade-offs in pathway rate and yield

Pfeiffer T, Schuster S, Bonhoeffer S, *Science* 292 (2001)

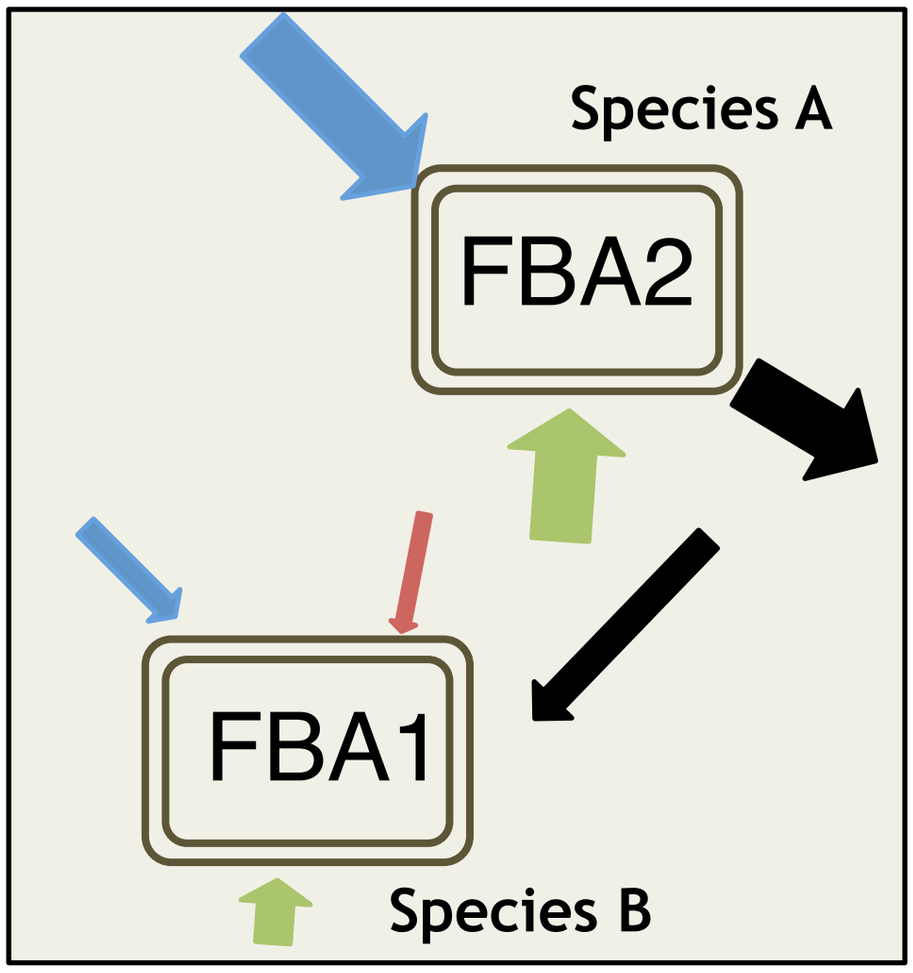
Trade-offs in substrate-based growth rates

Doebeli M, *Pop. Ecology* 44:2 (2002)



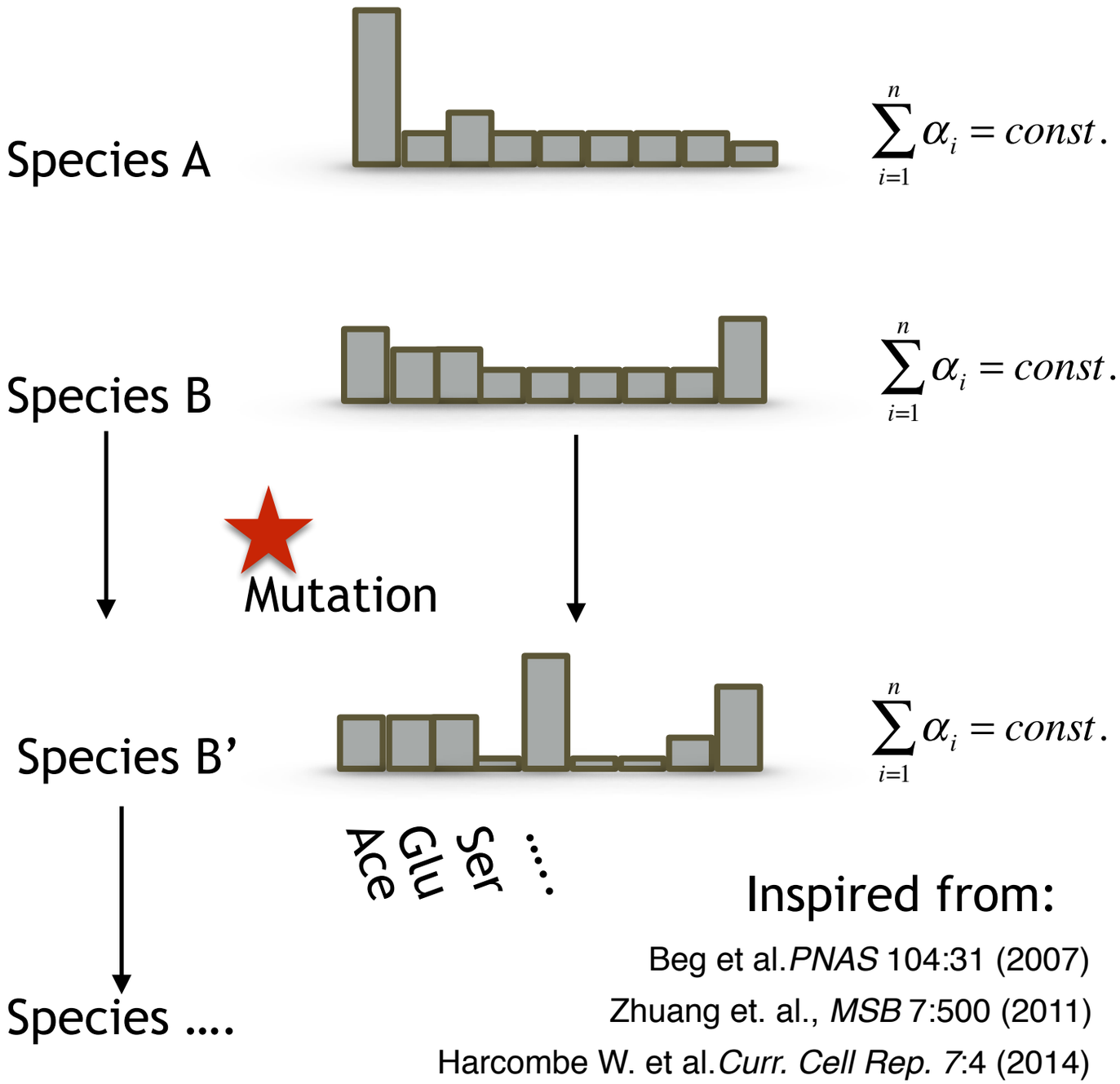
Can trade-offs lead to <u>evolution</u> of overflow metabolism under selection for biomass?

EvoFBA available to download



Continue *ad infinitum*

Uptake Reaction Bounds



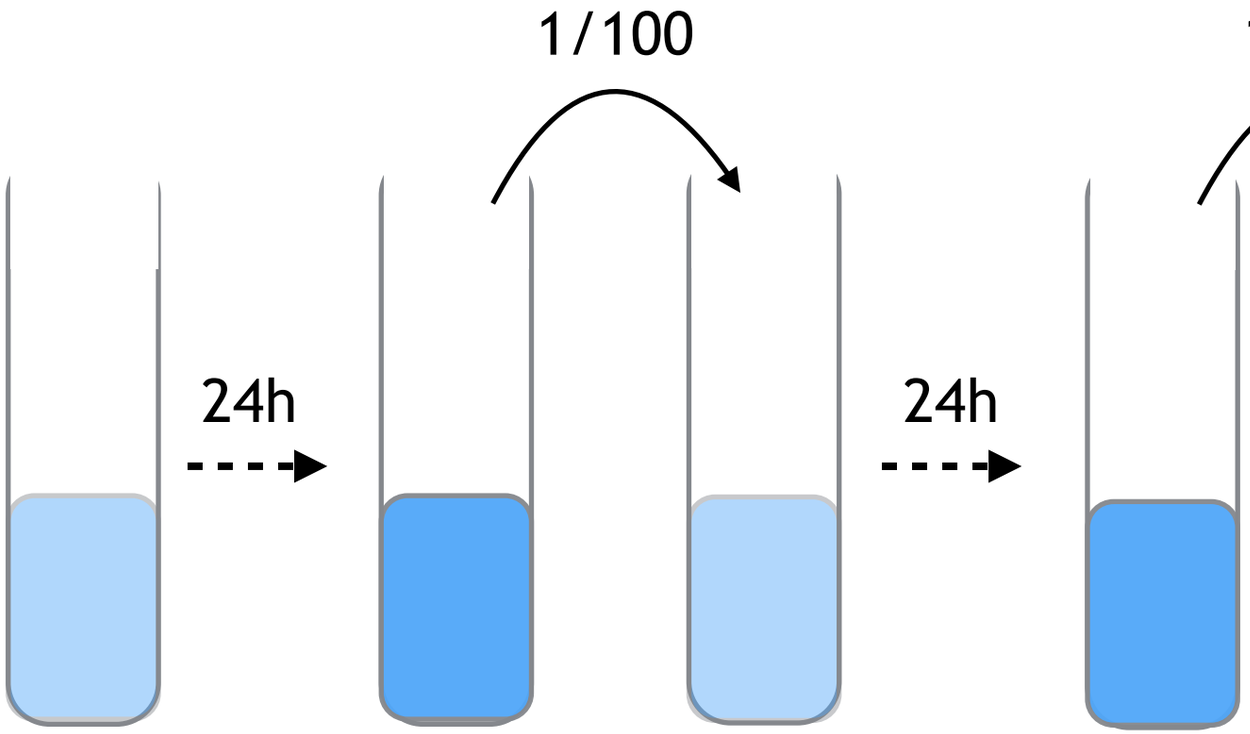
Inspired from:

Beg et al. *PNAS* 104:31 (2007)

Zhuang et. al., *MSB* 7:500 (2011)

Harcombe W. et al. *Curr. Cell Rep. 7*:4 (2014)

Directed evolution of *E.coli* using EvoFBA

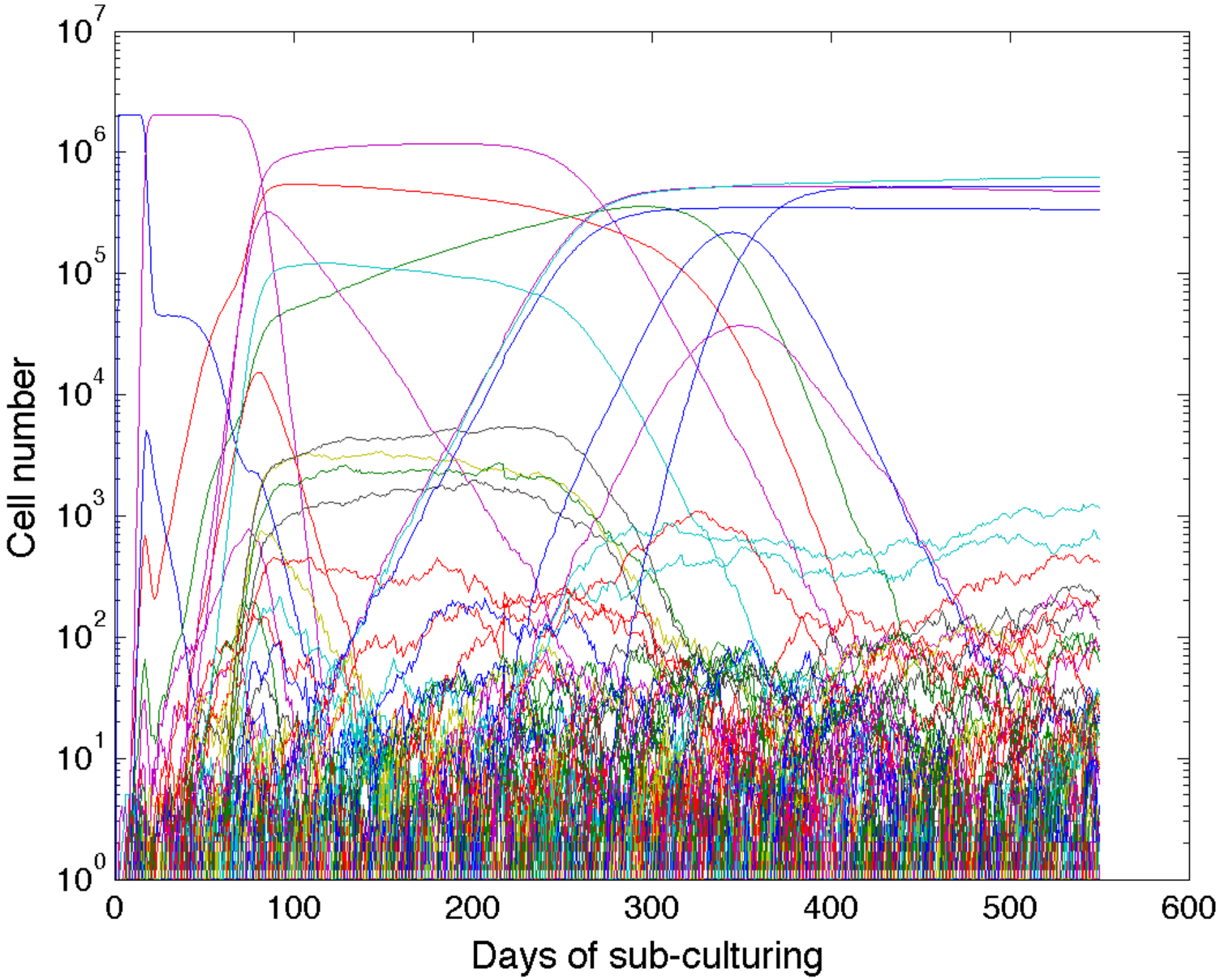


Continue *ad infinitum*

in silico version of

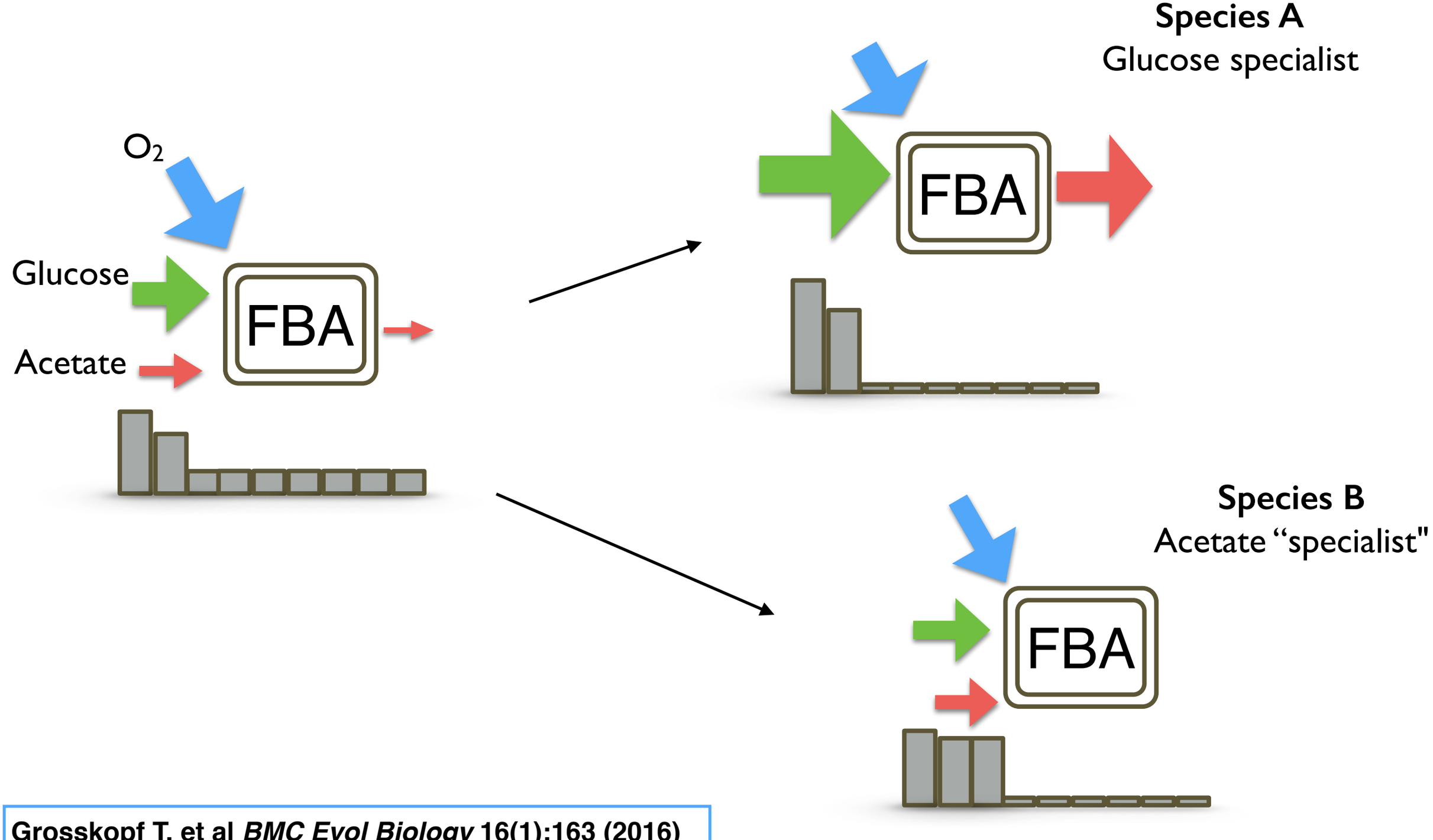
L**enski, R. et al., 1991**. Long-term experimental evolution in Escherichia coli. I. Adaptation and divergence during 2,000 generations. American Naturalist, 138(6), pp.1315–1341.

Many clones emerge over evolution



- 98678 clones generated
- 3978 (~4%) clones survived a sub-culturing event
- 235 +/- 30 clones present each day

Constraints in cellular resources lead to evolution of overflow metabolism and ‘cross-feeding’



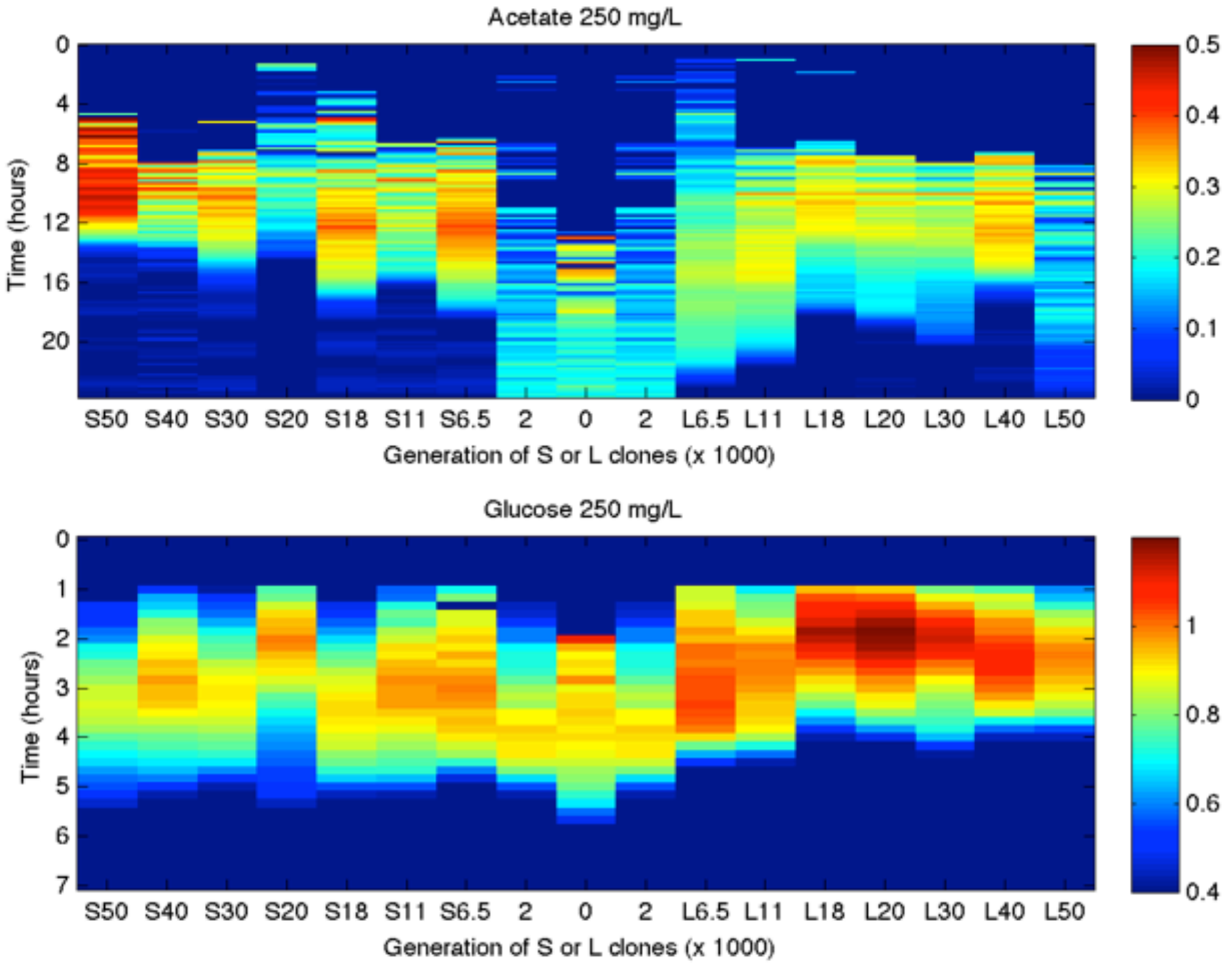
Grosskopf T, et al *BMC Evol Biology* 16(1):163 (2016)

Constraints in cellular resources lead to evolution of ‘cross-feeding’

Ecological
lineages d
Escherichi

Mickaël Le Gac[a,b], Jes

divergence. This pr
distinct niches or,
evolution. Here we
and S, that coexist
verging from a con
tained phenotypic
resource utilization
on the catabolic prc



Vignette 2: Cellular trade-offs can be a key driver of evolution of metabolic systems (intra- and inter-cellular)

Trade-offs in space/enzyme allocation

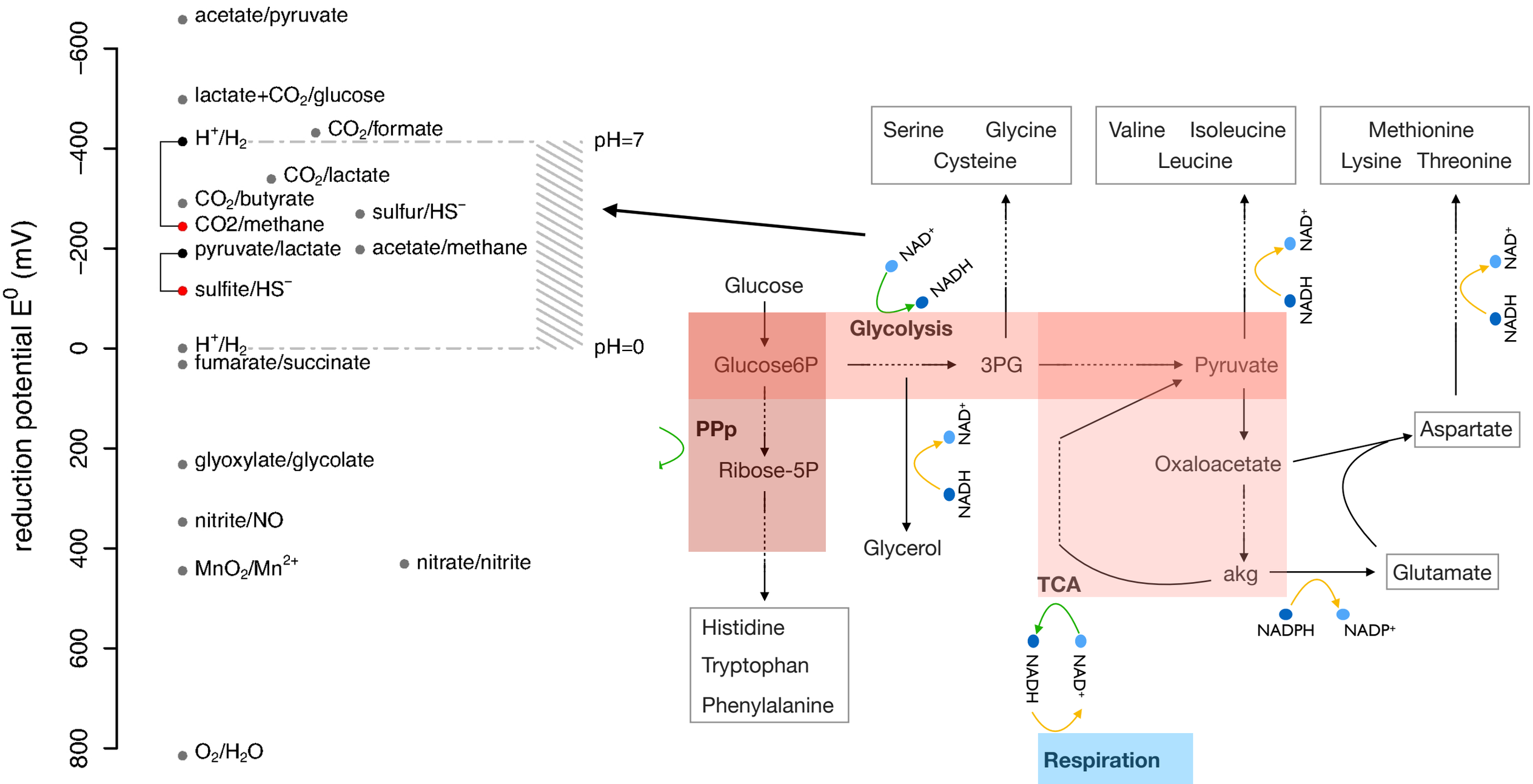
Trade-offs in pathway rate and yield

Trade-offs in substrate-based growth rates

…

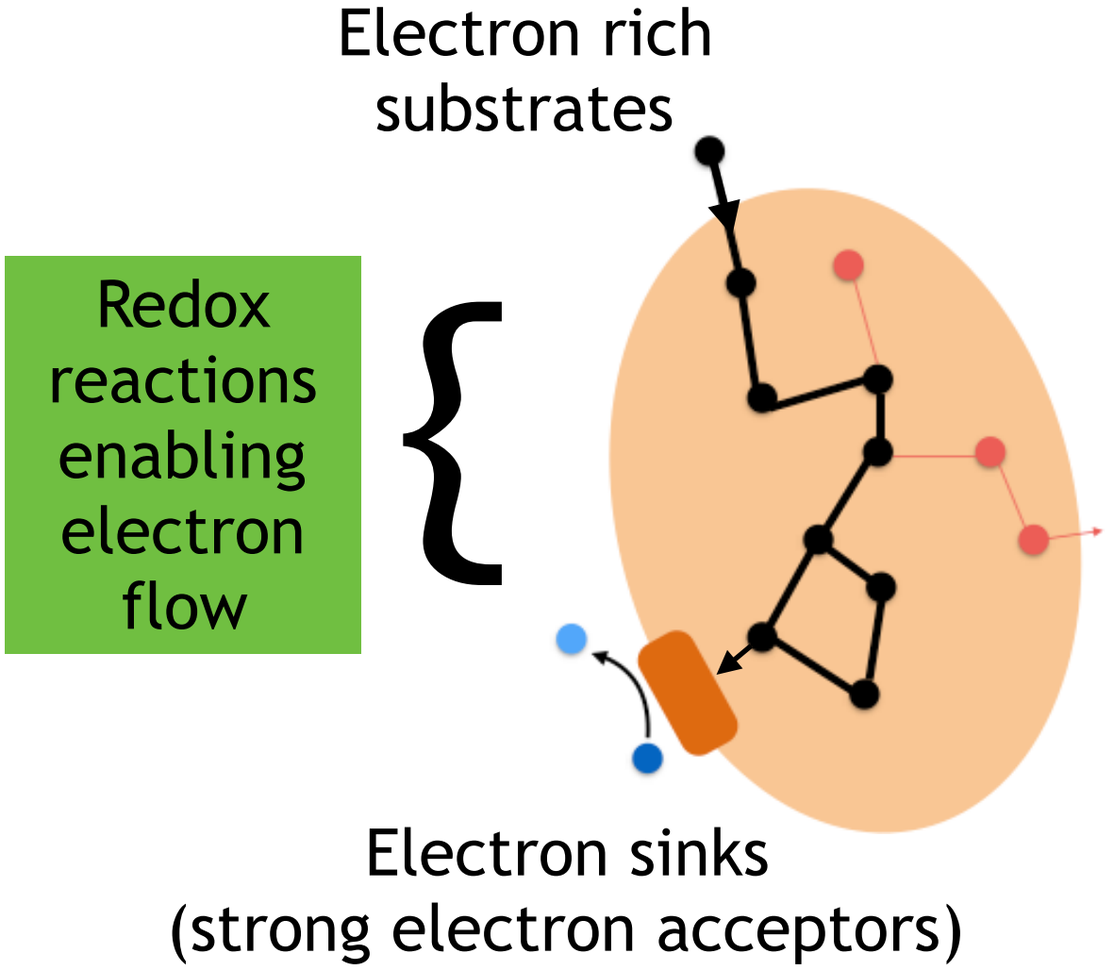
Trade-offs arising from conserved moieties and pH???

Vignette 2: Trade-offs arising from conserved moieties and pH leading to thermodynamic inhibition in different paths



Test 2: Do conserved moieties act as trade-off points?

Metabolism as an electron flow system shaped by thermodynamic bottlenecks and biophysical tradeoffs



Thermodynamic feasibility

Lack of electron acceptors?

Internal redox / charge limitations influencing metabolic fluxes

Charge and redox properties of membranes

Toxic redox effects of respiration?

Ecological drivers

Interrogating metabolism as an electron flow system.

Christian Zerfass, Munehiro Asally, Orkun S Soyer
Current Opinion in Systems Biology *13: 59-67 (2019).*

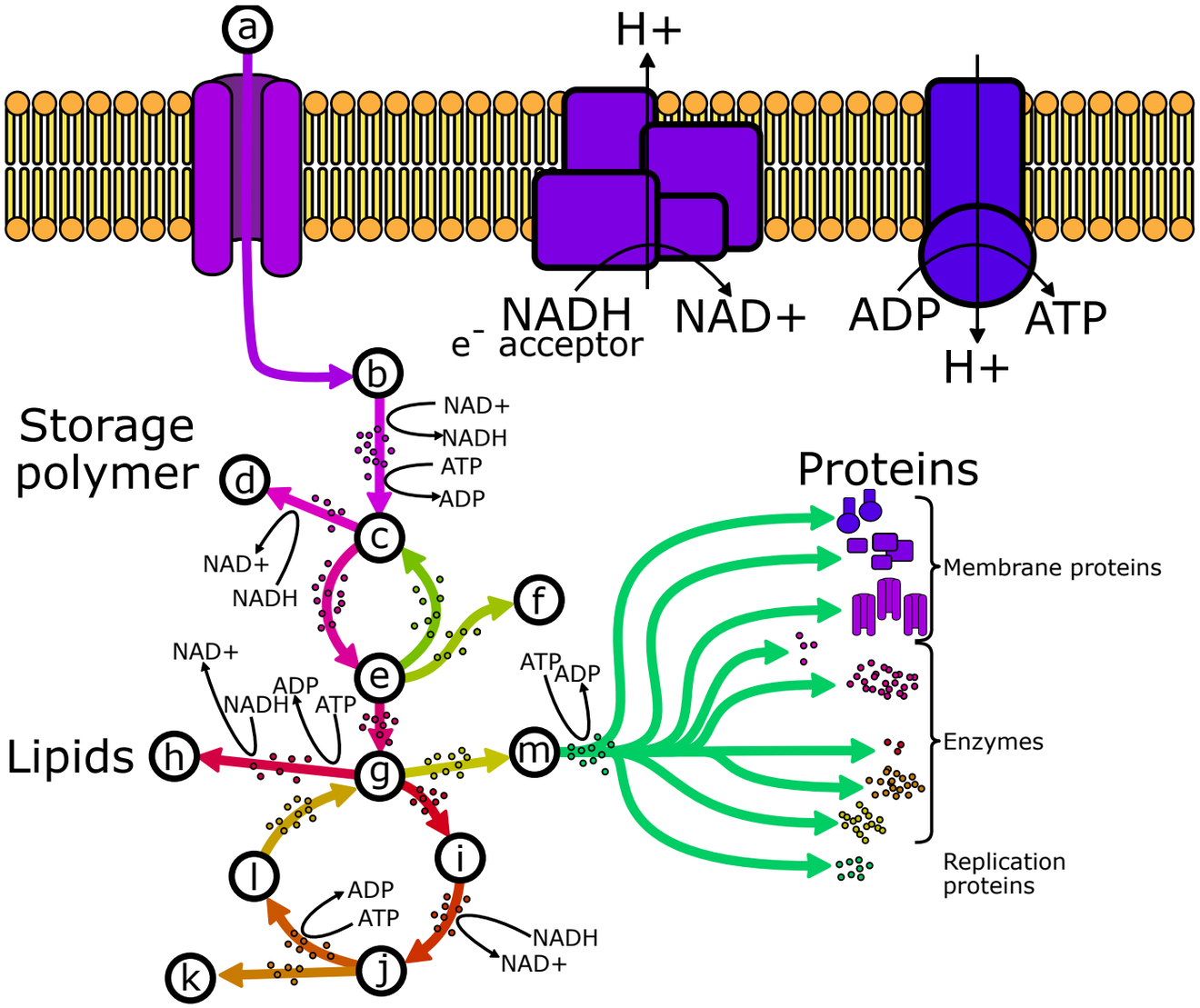
Engineering microbial communities using thermodynamic principles and electrical interfaces.

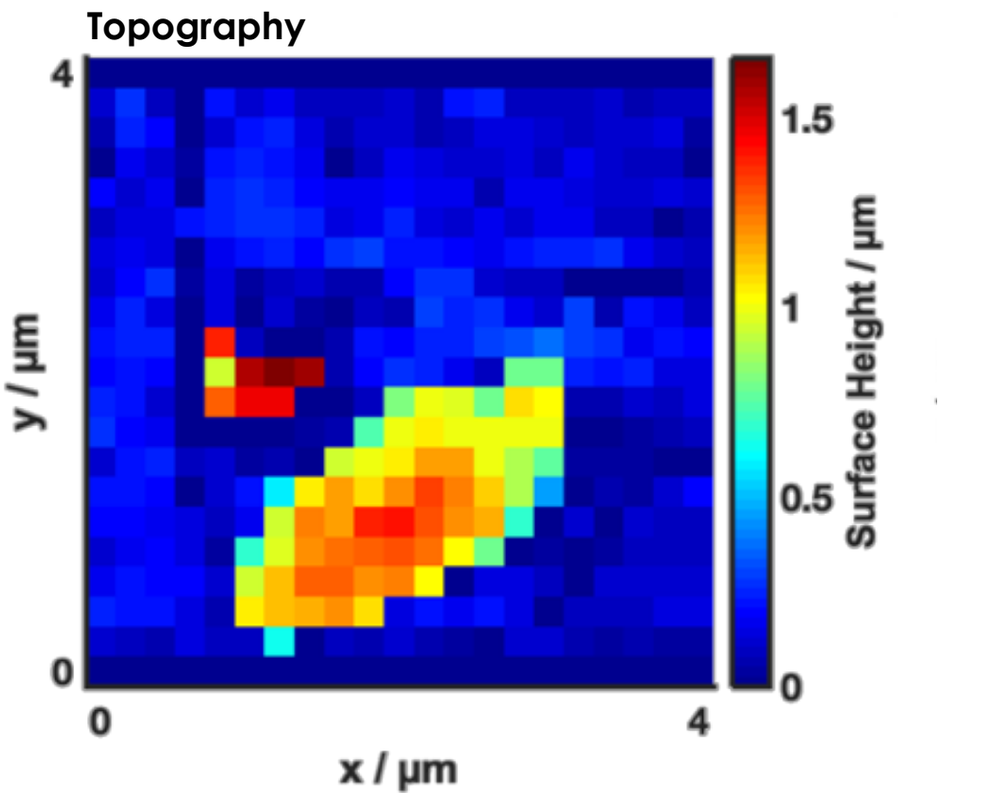
Christian Zerfass, Jing Chen, Orkun S Soyer,
Current Opinion in Biotechnology *50:121-127 (2017).*

Is a view based on electron flows and biophysical limits a useful one to understand and manipulate metabolism?

Red Pill or Blue Pill? The Matrix, 1999, Warner Brothers

Basics: Develop and parameterise a holistic model of metabolism and physiology



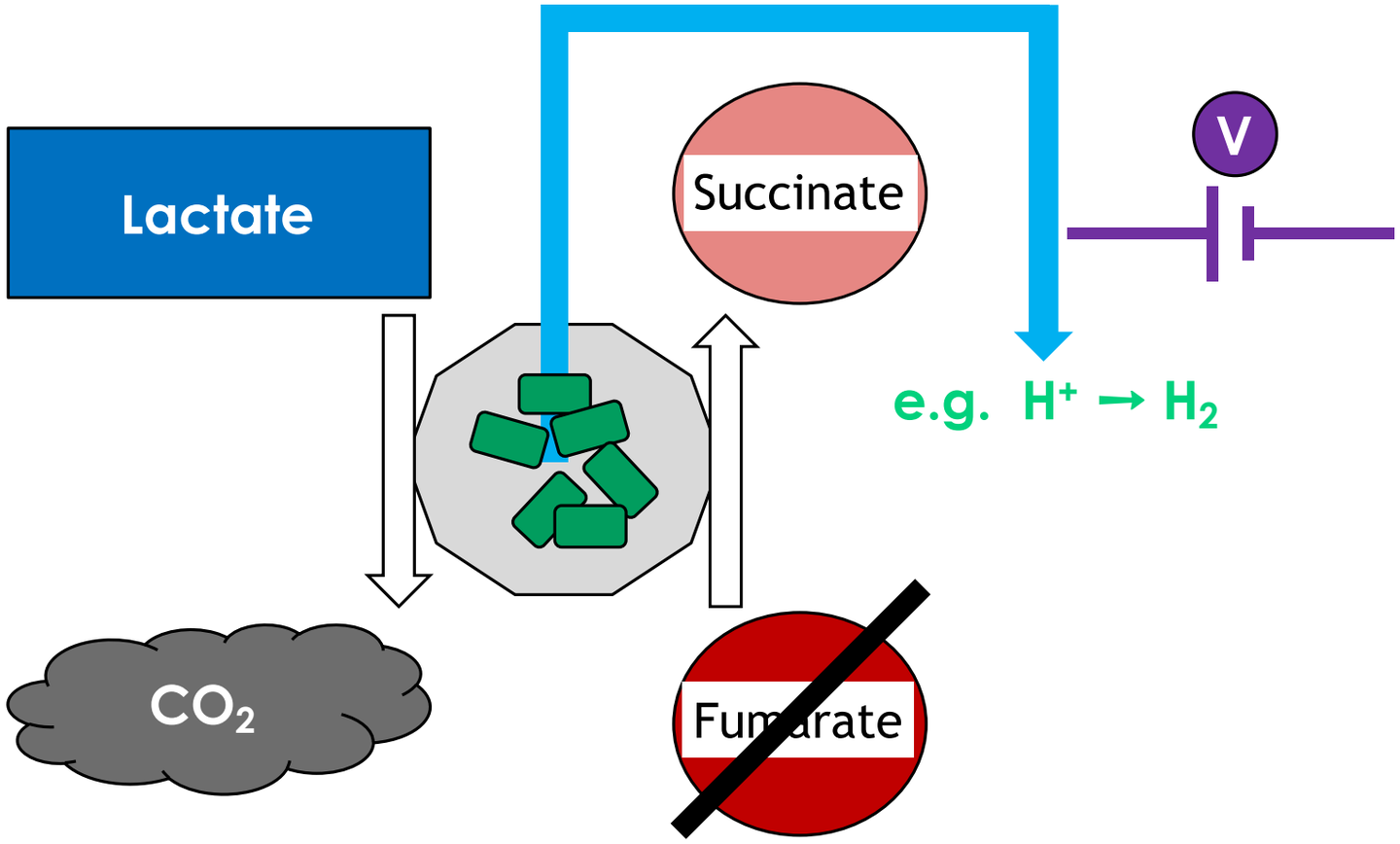


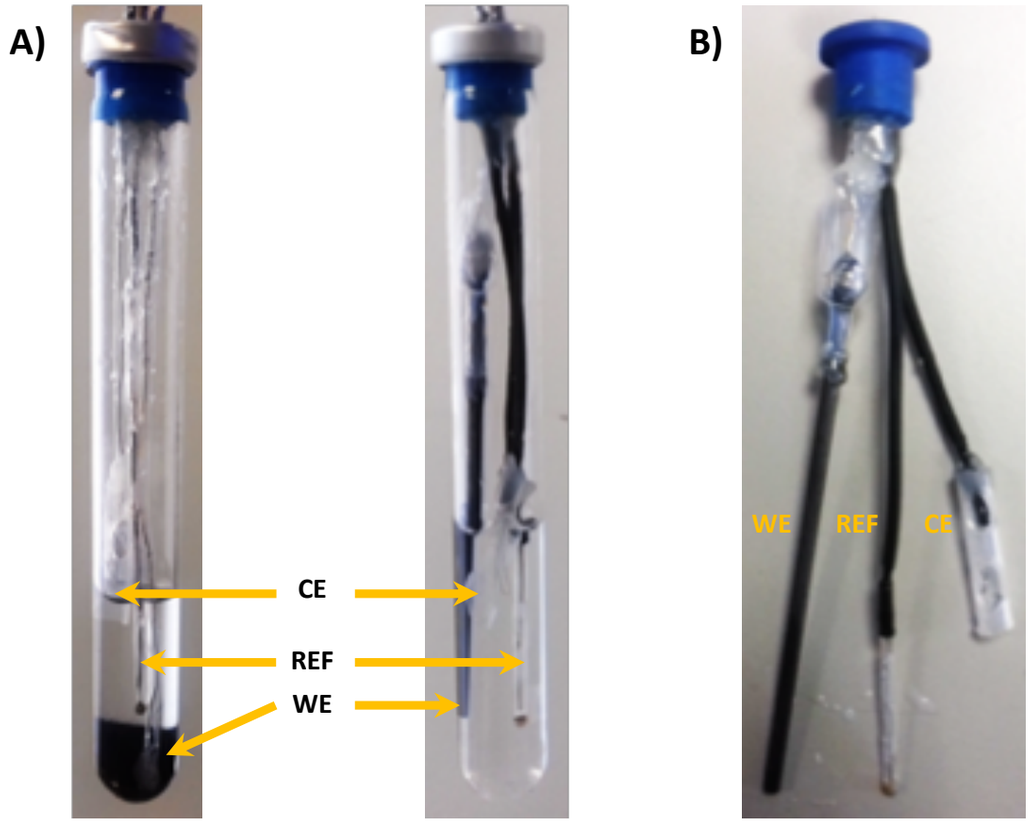
Can we develop a toy model capturing the dynamics of metabolism, conserved moieties, pH, and membrane potential?

Kelsey Cremin

Hadrien Delattre

Test 1: Use electrodes as terminal electron acceptors to shape respiro-fermentation

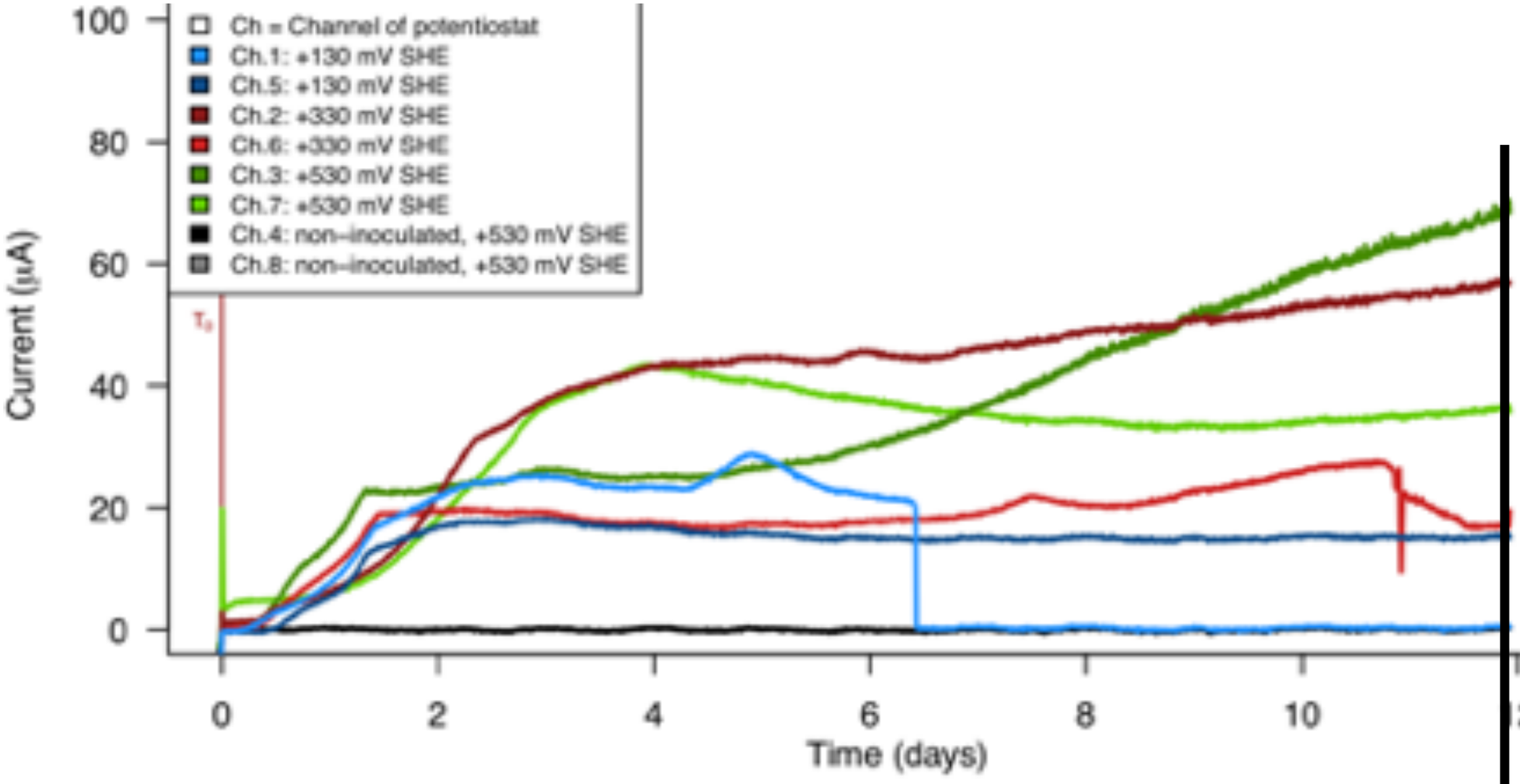




Control of respiration vs. fermentation in *Shewenalla oneidensis* using electrodes poised at specific potentials

Christian Zerfass

Cells utilise both electrode and oxygen



530mV

330mV

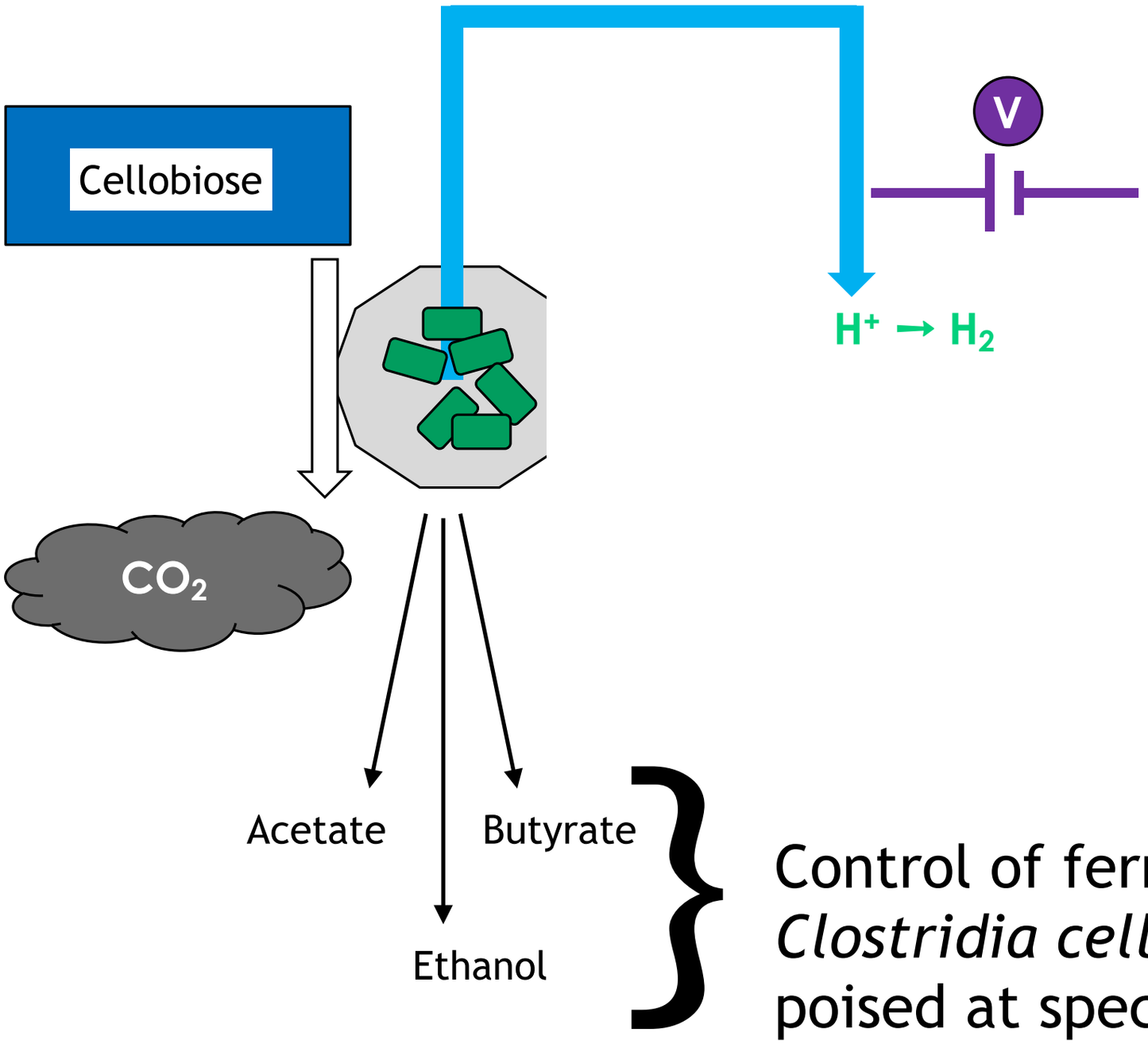
~200mV fum/succ

130mV

Lactate cons.	Acetate prod.	Fum. cons.	Succ. prod.	e⁻ estimate to O_2	e⁻ total calculated	e⁻ measured
20	14	28	28	68	140	140
14	13	-	-	68	132	64
13	12	-	-	68	128	64
12	8	-	-	68	148	80

Assuming 12 (4) e⁻ per lactate resp. (ferm.)

Try with cellulose degrading *Clostridia*



Christian Zerfass

Jing Chen

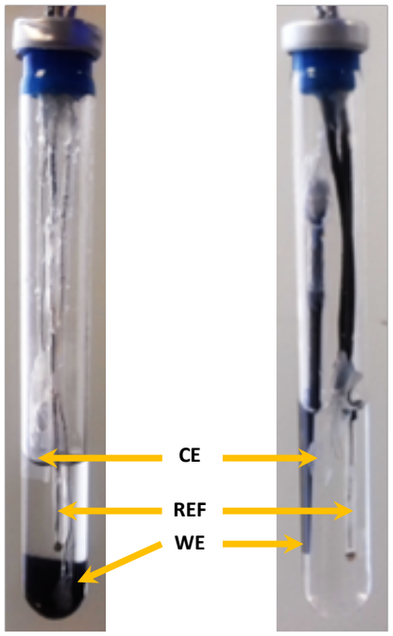
GreenBiologics

Holly Smith, senior scientist, fermentations

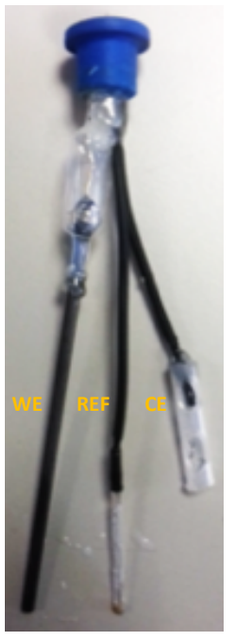
Control of fermentation outputs in *Clostridia celluloticum* using electrodes poised at specific potentials

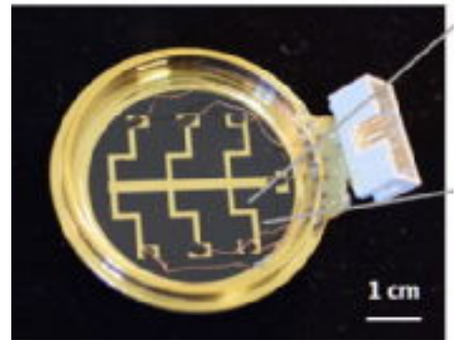
Link to internal metabolism; Upcoming!

A)



B)

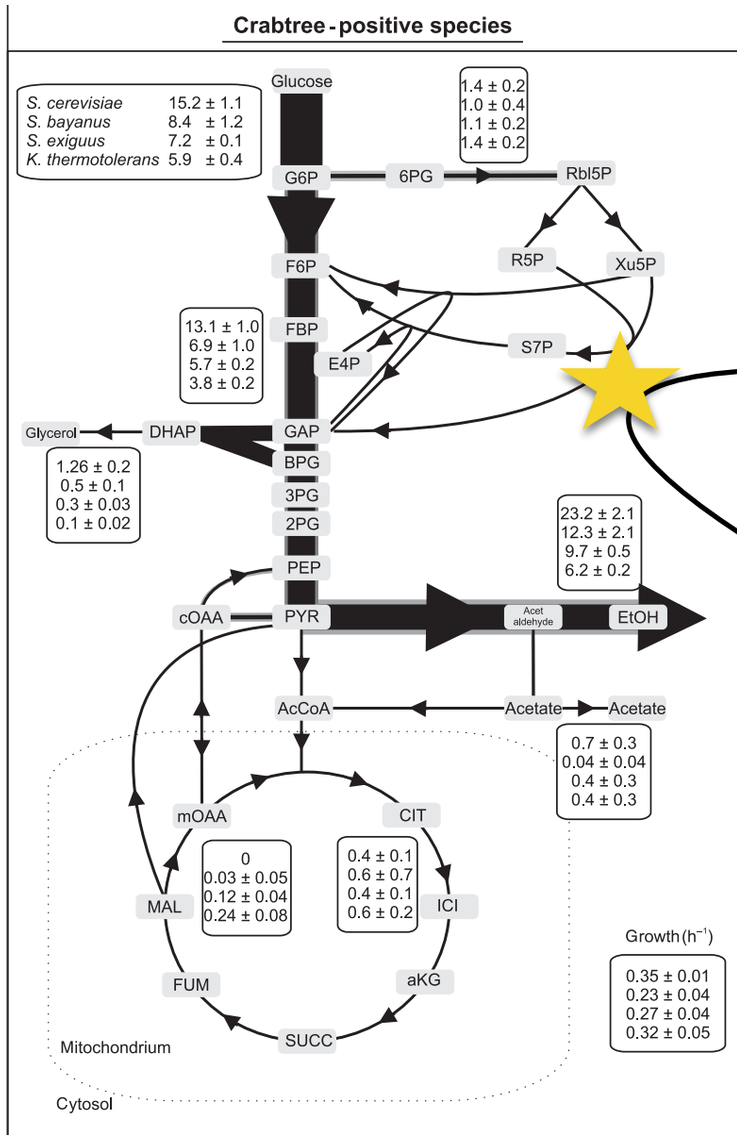


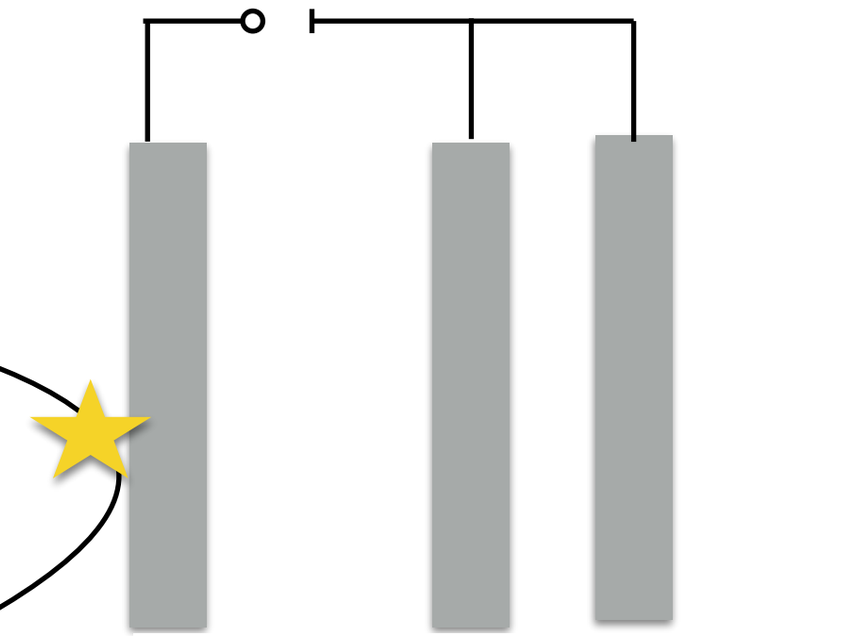


Open PDRA positions

Bio Electrical Engineering Innovation Hub @ Warwick

Test 2: Use electrodes and redox mediators to shape metabolic trade-offs





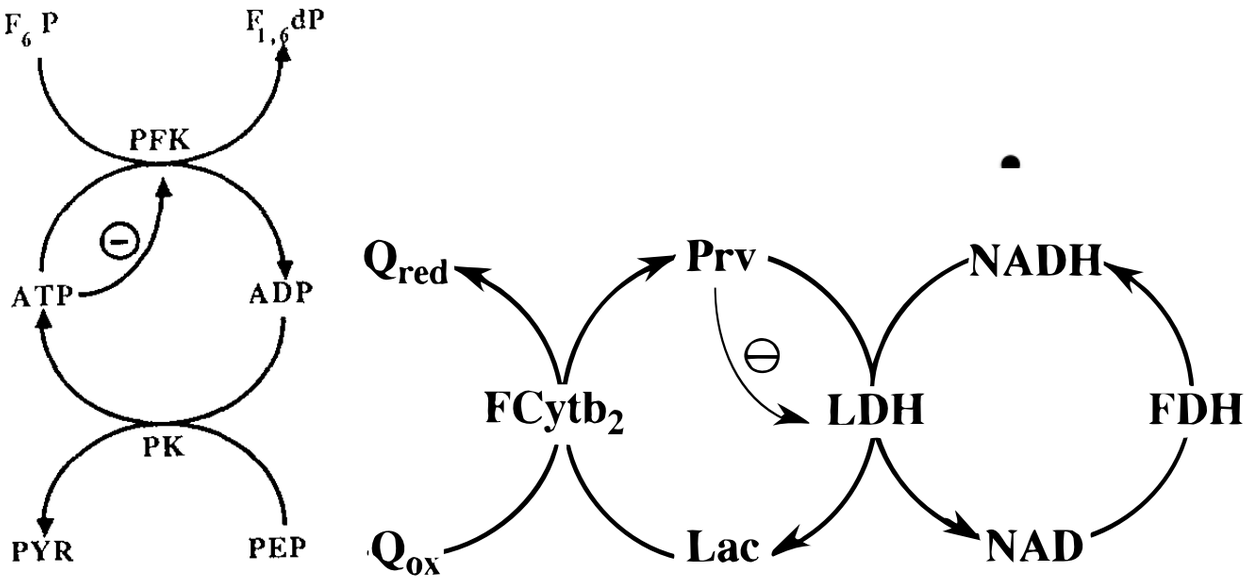
CANCER RESEARCH UK PIONEER AWARD

Control of metabolic pathways in cancer cells with redox mediators

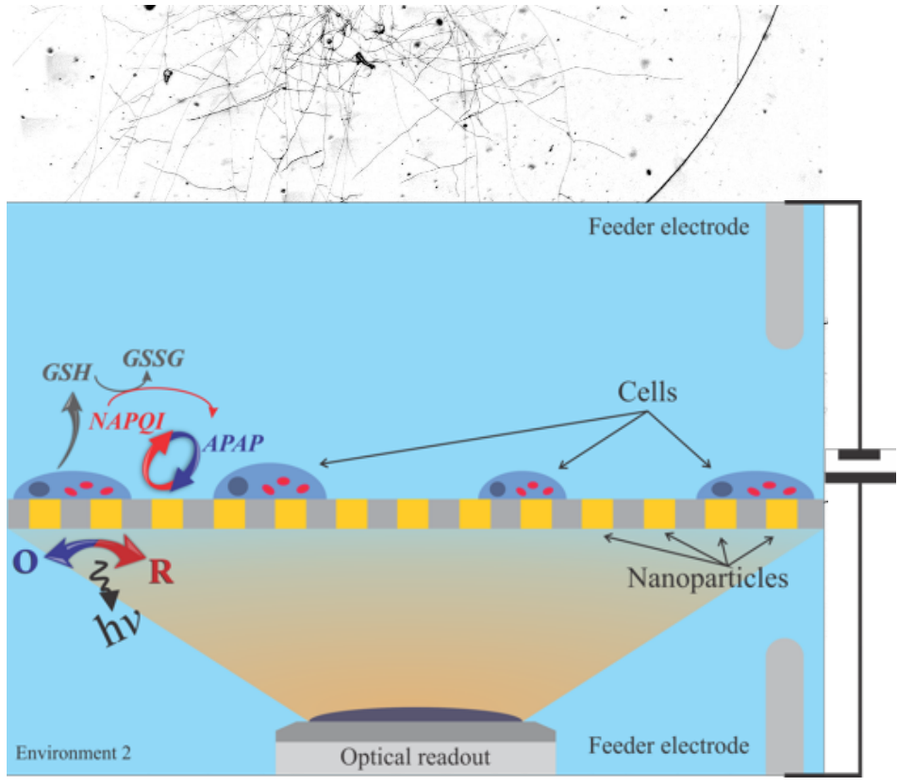
Zoe Schofield **Craig McBeth**

More tests: Metabolism as an electron flow system

Controlling bistability and oscillation in enzymatic reaction motifs



Electric nature of metabolic, spatial oscillations



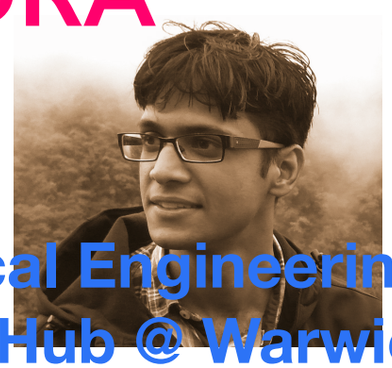
Enzyme-level

Cell-level

Clare Hayes

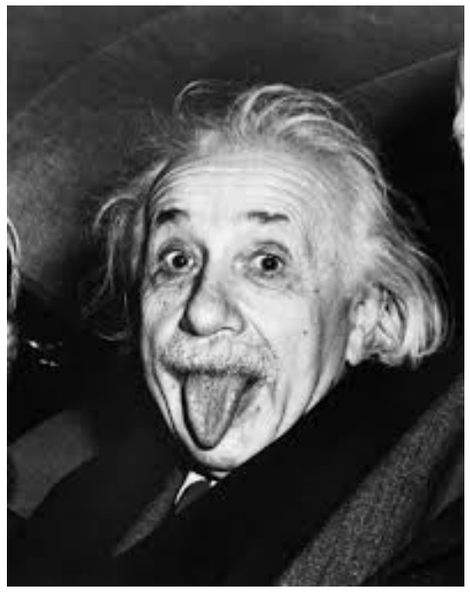
Open PDRA positions

Bio Electrical Engineering Innovation Hub @ Warwick



Praneet Prakash

Metabolism as an electron flow system



Thermodynamics is the only physical theory of universal content, which I am convinced, that within the framework of applicability of its basic concepts will never be overthrown!

THANK YOU

OSS LAB

http://osslab.lifesci.warwick.ac.uk

Craig McBeth
Christian Zerfass
Jing Chen
Zoe Schofield
Connah Johnson
Clare Hayes
Kelsey Cremin
Praneet Prakash
Hadrien Delattre

Funders

BBSRC bioscience for the future
EPSRC Pioneering research and skills
WARWICK THE UNIVERSITY OF WARWICK
CANCER RESEARCH UK PIONEER AWARD

Collaborators

Munehiro Asally, Marco Polin, Pat Unwin, Chris Quince, Joseph Christie-Oleza, Patrick Schaefer (University of Warwick)
Angus Buckling (University of Exeter)

Two PDRA positions

WARWICK CENTRE FOR INTEGRATIVE SYNTHETIC BIOLOGY

Bio Electrical Engineering Innovation Hub @ Warwick