

Cellular Economics of Exchanged Metabolites Alter Ratios of Microbial Trading Partners in a Predictable Manner



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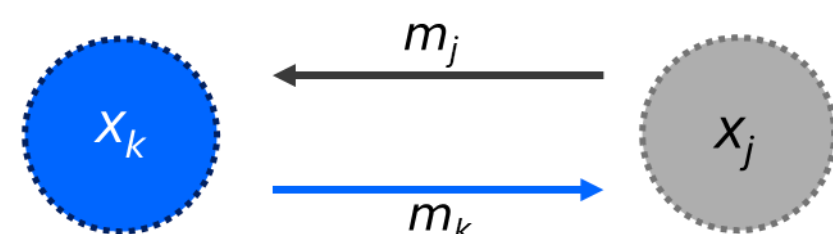
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Abstract

Most microorganisms exist in interacting consortia, yet the principles behind consortia assembly, including trading partner structure, still require deciphering despite being central to understanding, building, and controlling consortia. This study tests a cellular economy-based hypothesis that proposes consortia engaged in obligate metabolite exchange will assemble at ratios where the metabolic burden associated with the exchanges is predictably shared. The metabolic burden was quantified using ATP equivalents, the quantity of high energy ATP phosphoanhydride bonds that could be produced if the exchanged metabolites were instead retained and catabolized for cellular energy. The burden of metabolite exchange was manipulated by changing O_2 availability, which altered the bioavailability of substrate energy, and by deactivating the ATP synthase enzyme, which prevented oxidative phosphorylation. Three synthetic cocultures assembled at predicted strain ratios as a function of the perturbations. The strain ratios represented an equivalent metabolic burden between the trading partners even though the exchanged metabolites varied substantially in flux magnitude (300+ fold), molecular weight (174 vs. 88 g mole⁻¹), enzyme requirements (8 specialized enzymes vs. none), and biological energy density (27.5 vs. 9 ATP molecule⁻¹). The strain ratios could be rationally altered up to 20-fold using O_2 availability and cellular phenotype. The metabolic burden theory was applied to seven additional published cocultures and found to accurately predict trading partner ratios. Quantifying exchanged metabolites on an ATP equivalents basis provides a theory for interpreting natural consortia and a toolbox for controlling bioprocess consortia.

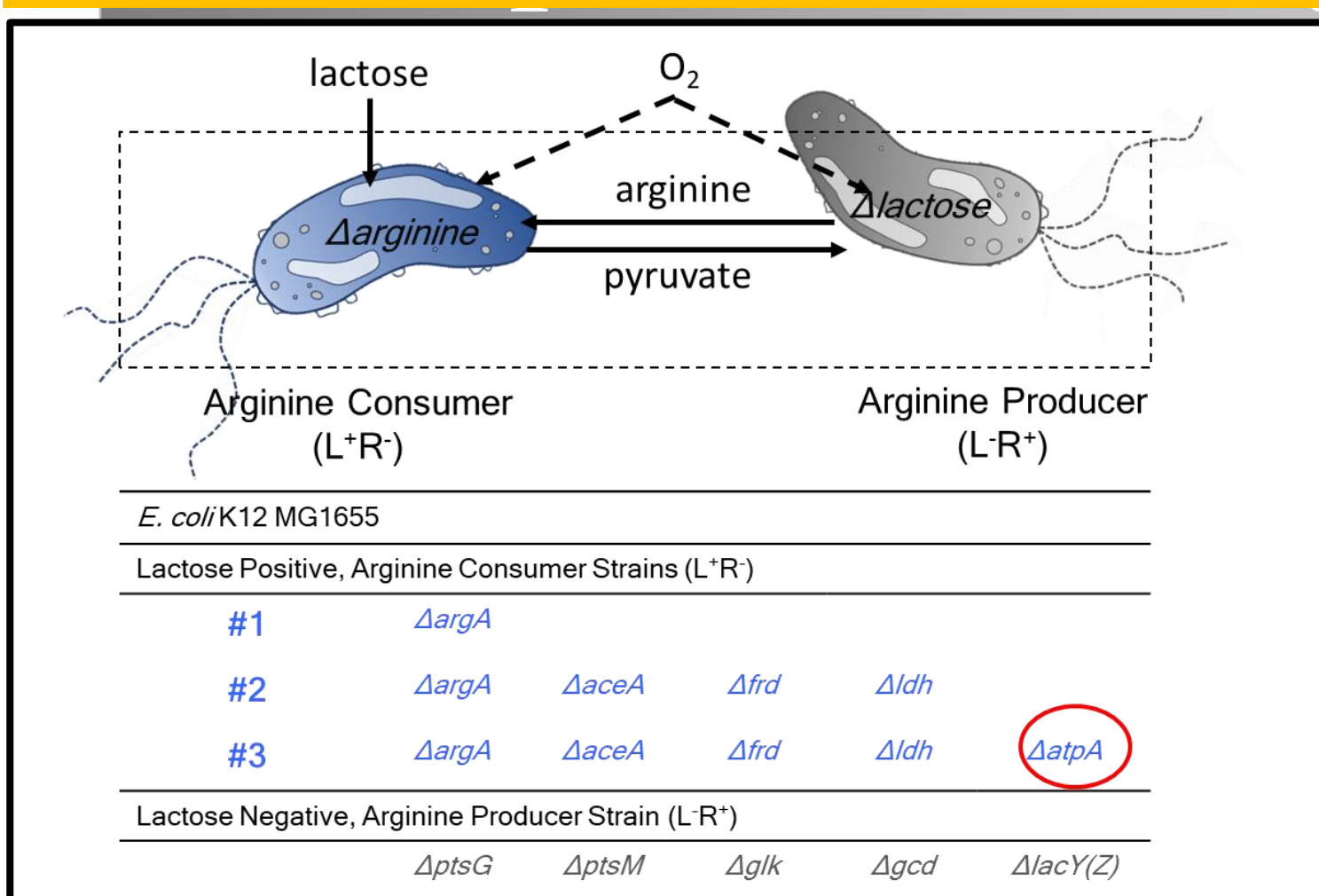
Opportunity Costs and Metabolic Burden



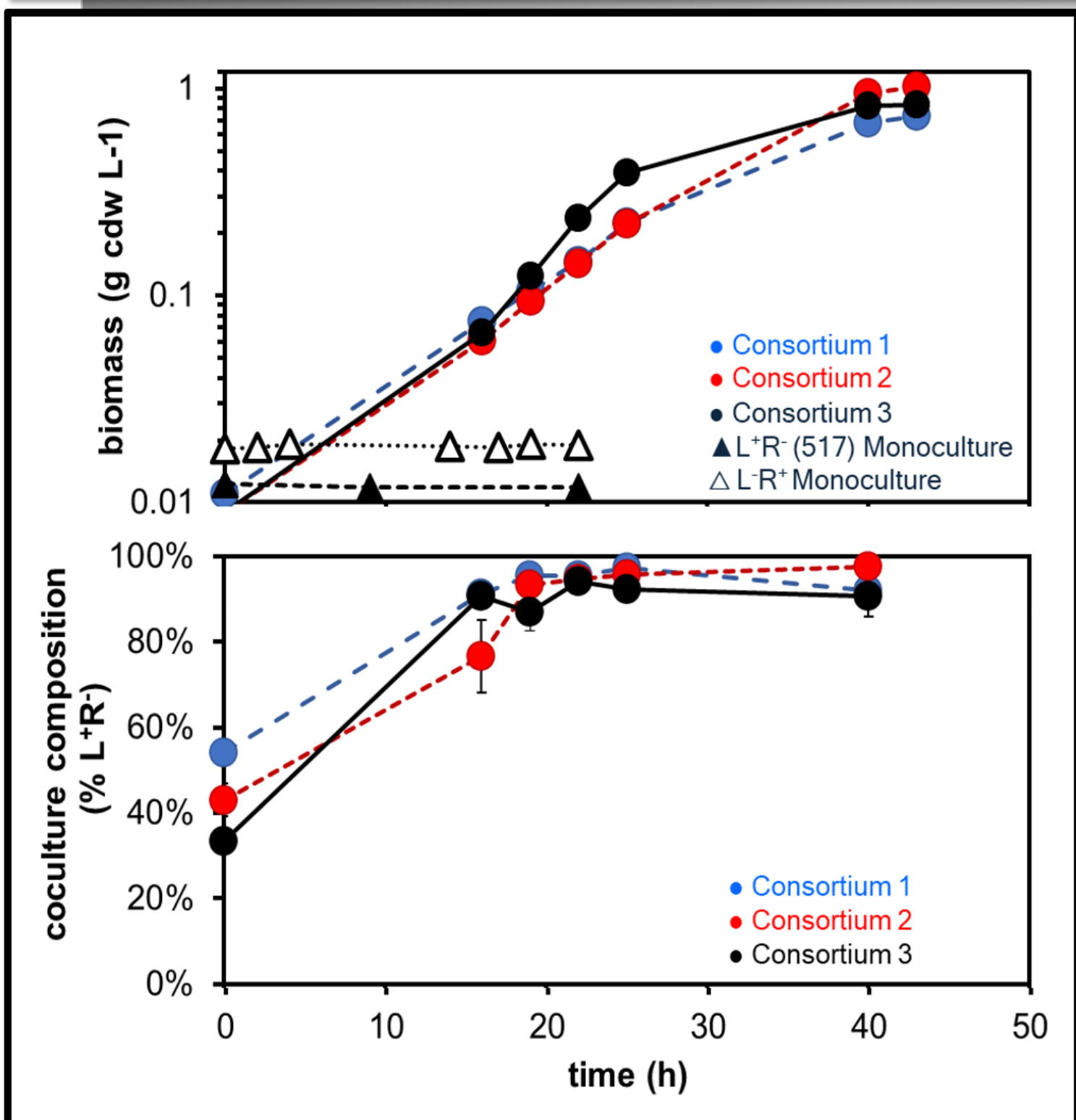
Hypothesis: during obligate metabolite exchange, cells exchange equitable quantities of the resources.

Equitable can be quantified as the number of ATP bonds that could be made if the exchanged metabolite were instead retained and catabolized.

Experimental System



Three synthetic consortia were built for obligate exchange of pyruvate and arginine. Consortium 3, L⁻R⁺ strain had altered cellular economics and was not able to generate ATP using ATP synthase.



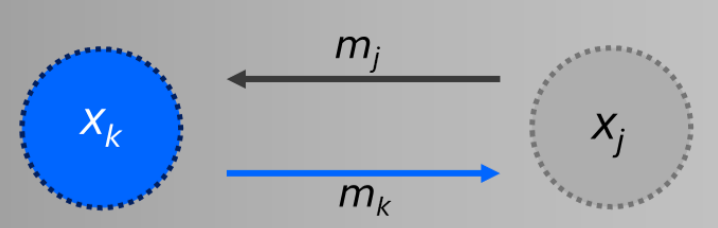
Consortia had similar growth rates, biomass yields, and strain ratios during O_2 limited planktonic, batch growth. Metabolite exchange was obligatory as the strains did not grow as monocultures on lactose salts medium.

Theory

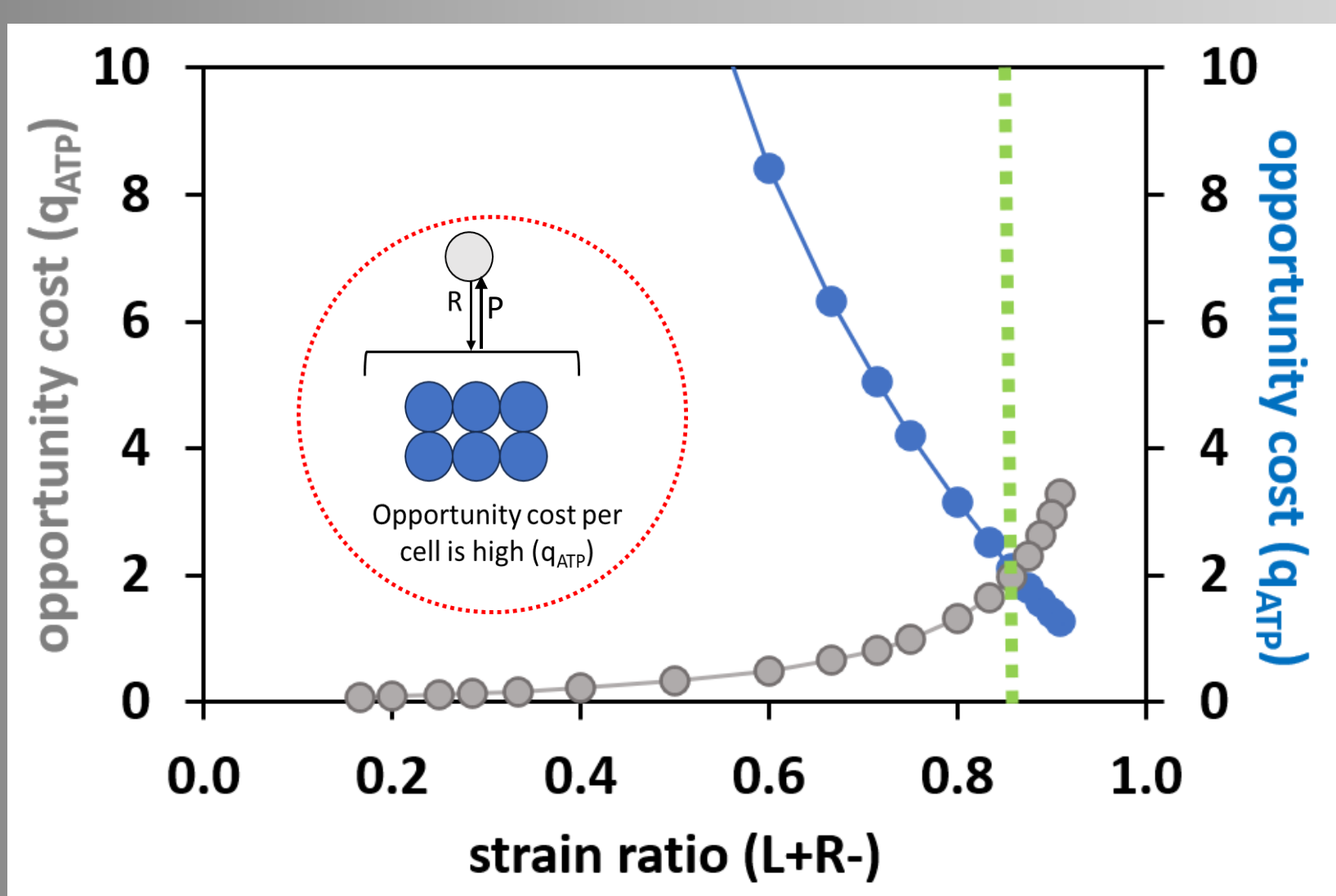
ATP equivalent balance:

$$\frac{dATP_k}{dt} = G_{atp,k} - q_{x,k} - q_{atp,k} - q_{mx,k} + q_{mx,j}$$

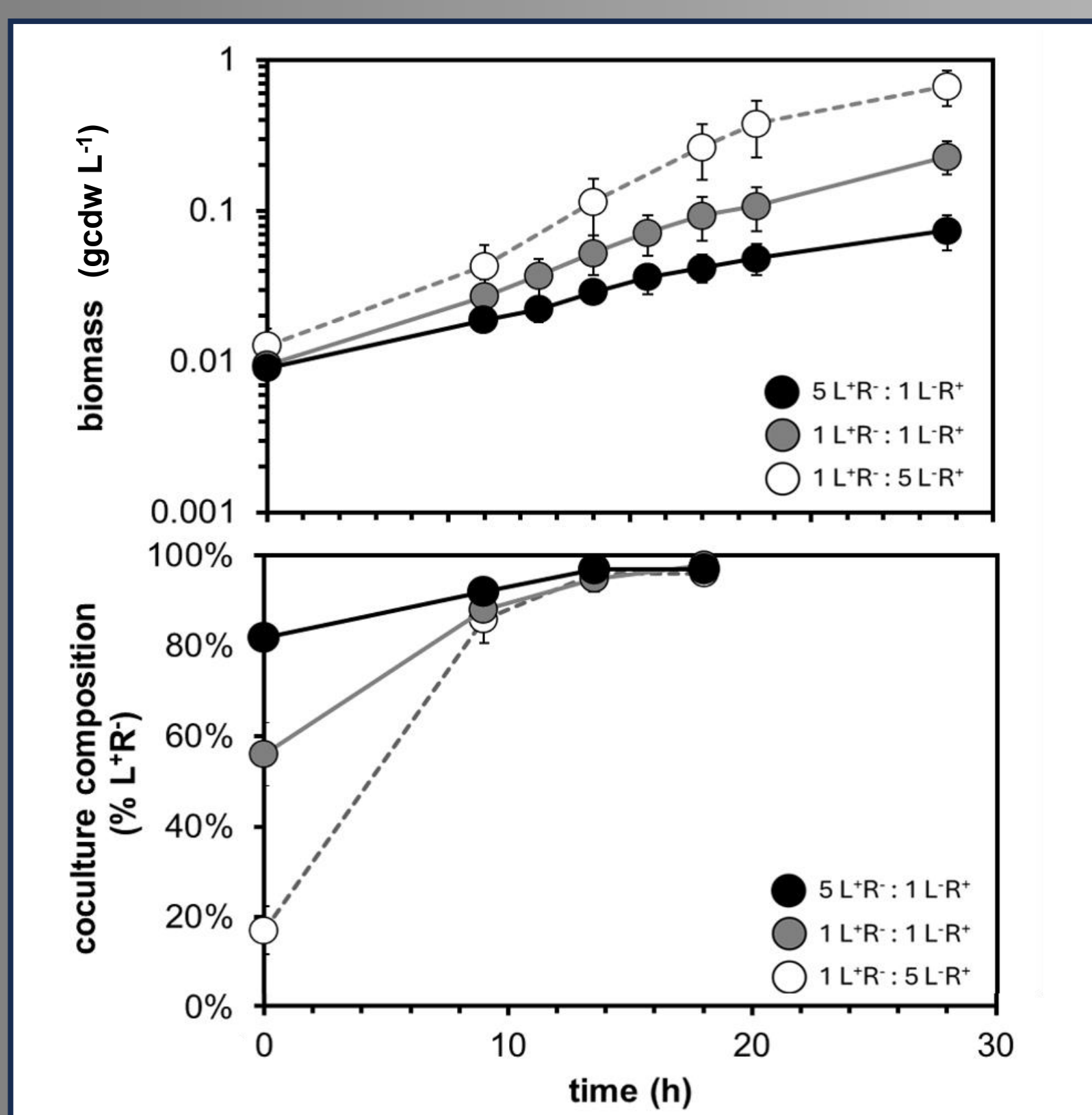
Pyruvate: 9 (~1) ATP
 $q_{pyr} = 2$ (11) mmol (g cdw h)⁻¹



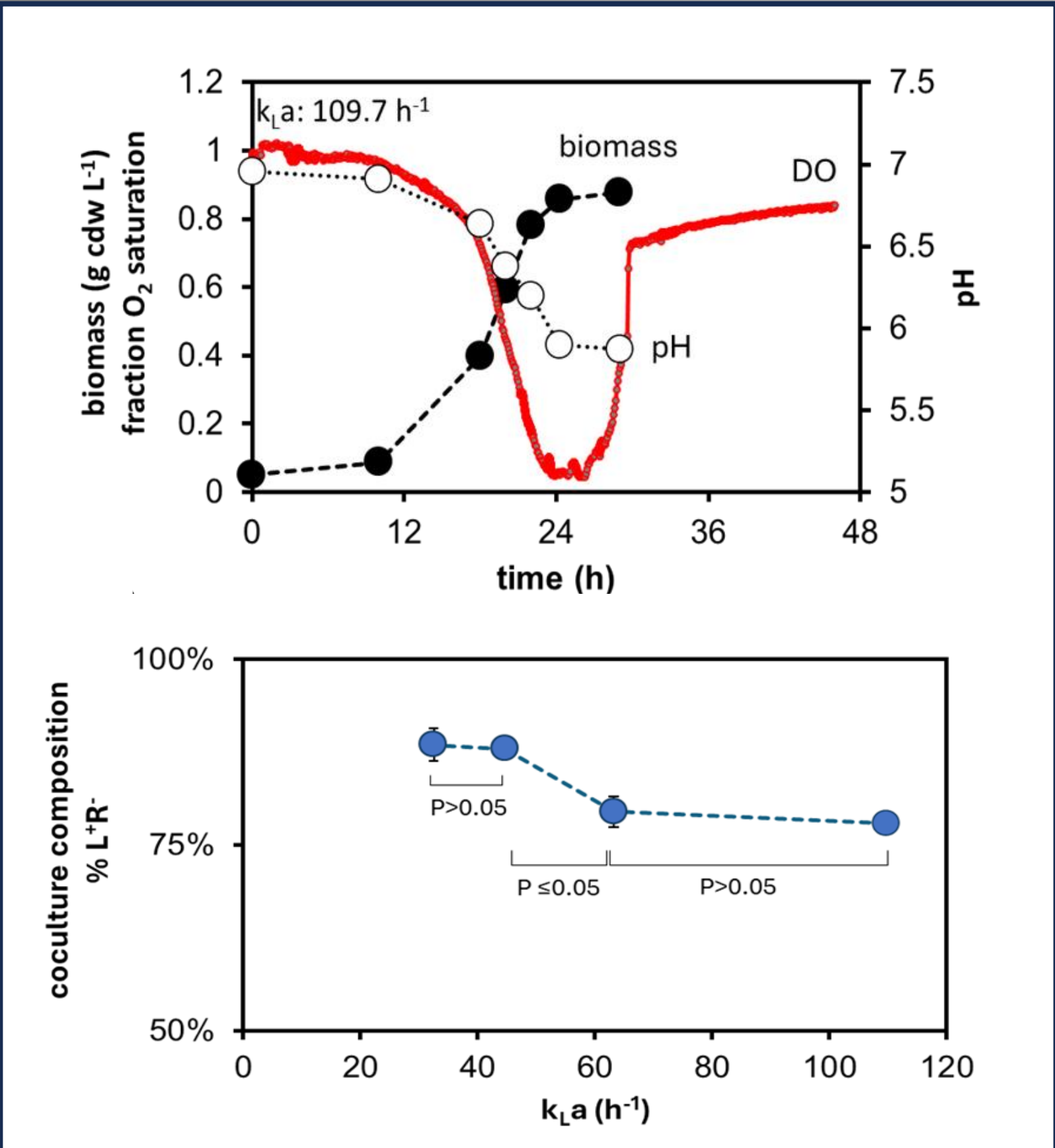
Arginine: 28 (9) ATP
 $q_{arg} = 0.03$ mmol (g cdw h)⁻¹



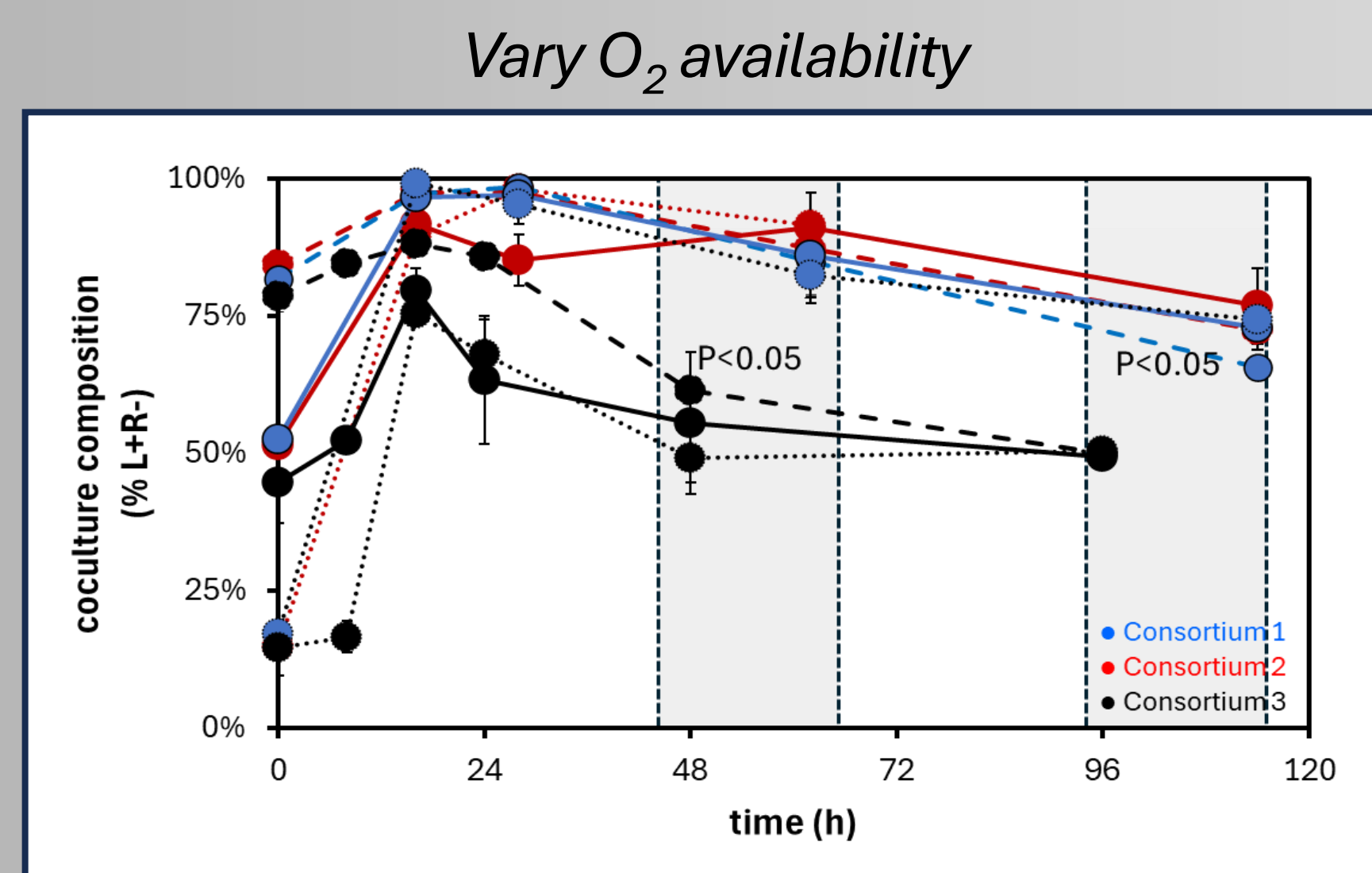
Results



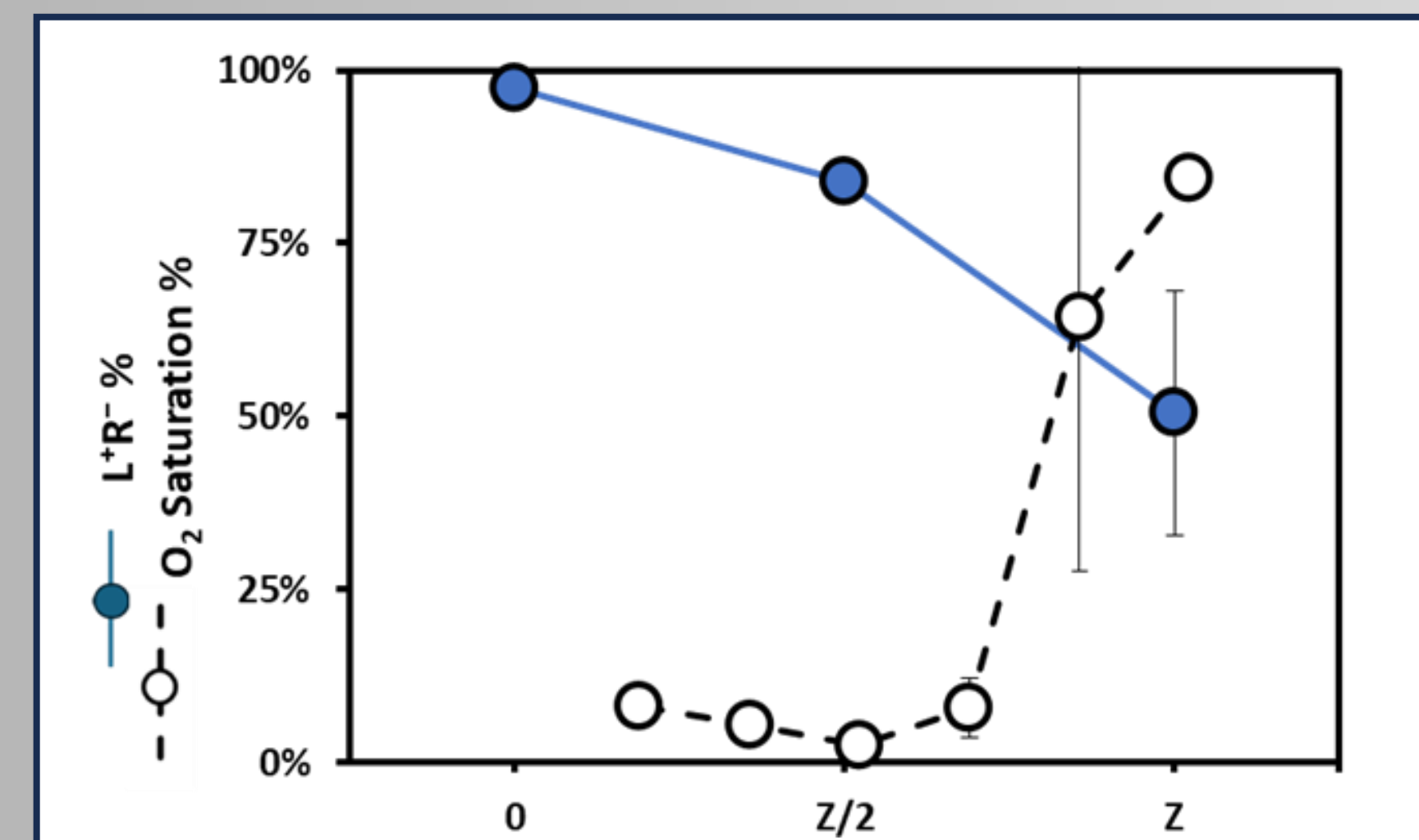
Planktonic consortia were robust to starting strain ratios. Consortia quickly approached stable, repeatable ratios influenced by O_2 availability and ability to use ATP synthase.



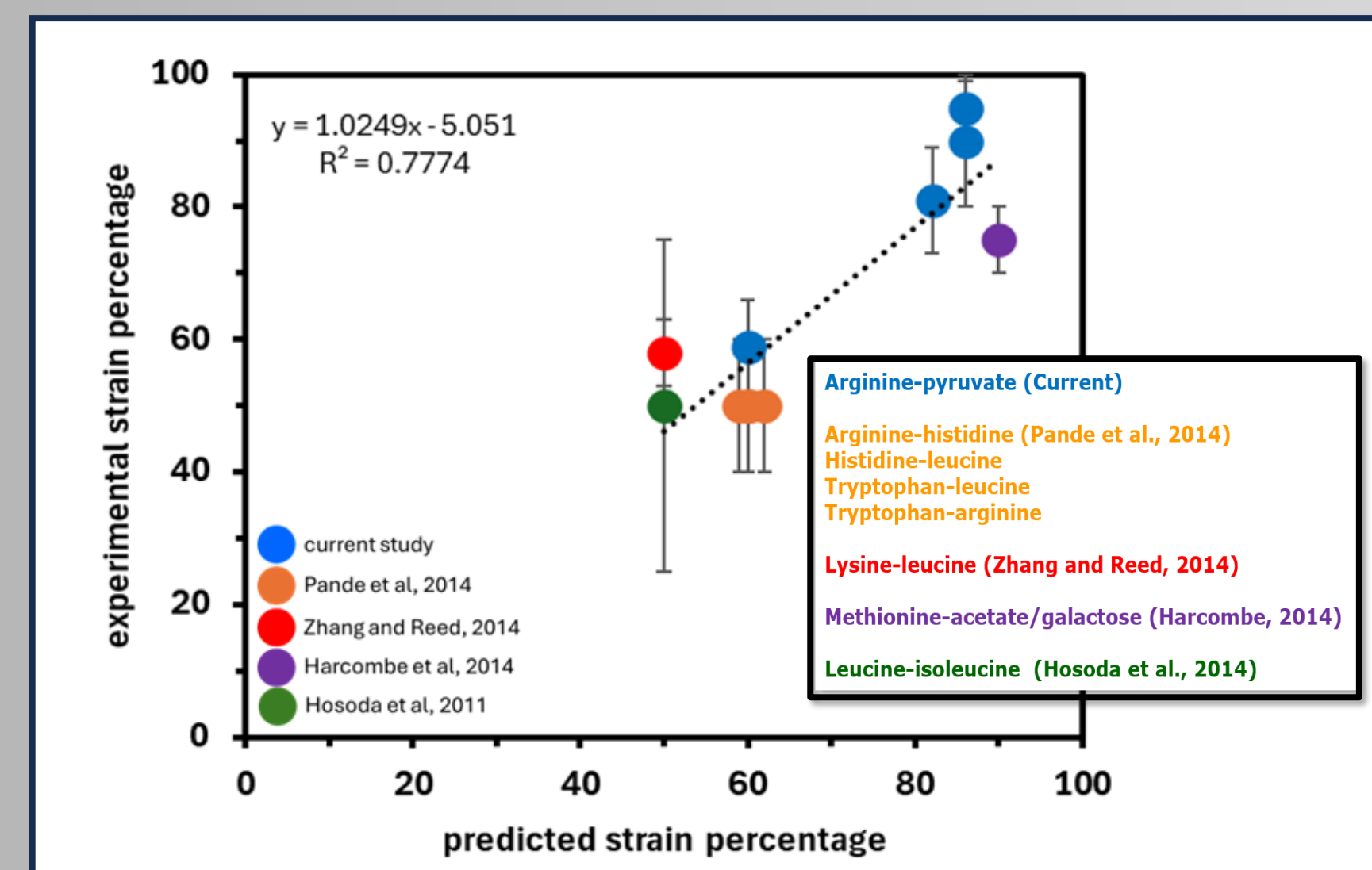
O_2 availability in planktonic consortia controlled with bioreactor $k_L a$ values and via electron donor : electron acceptor ratios in sealed microcalorimetry bioreactors.



Biofilm consortia were robust to starting strain ratios. Consortia quickly approached stable, repeatable ratios influenced by O_2 availability and ability to use ATP synthase.

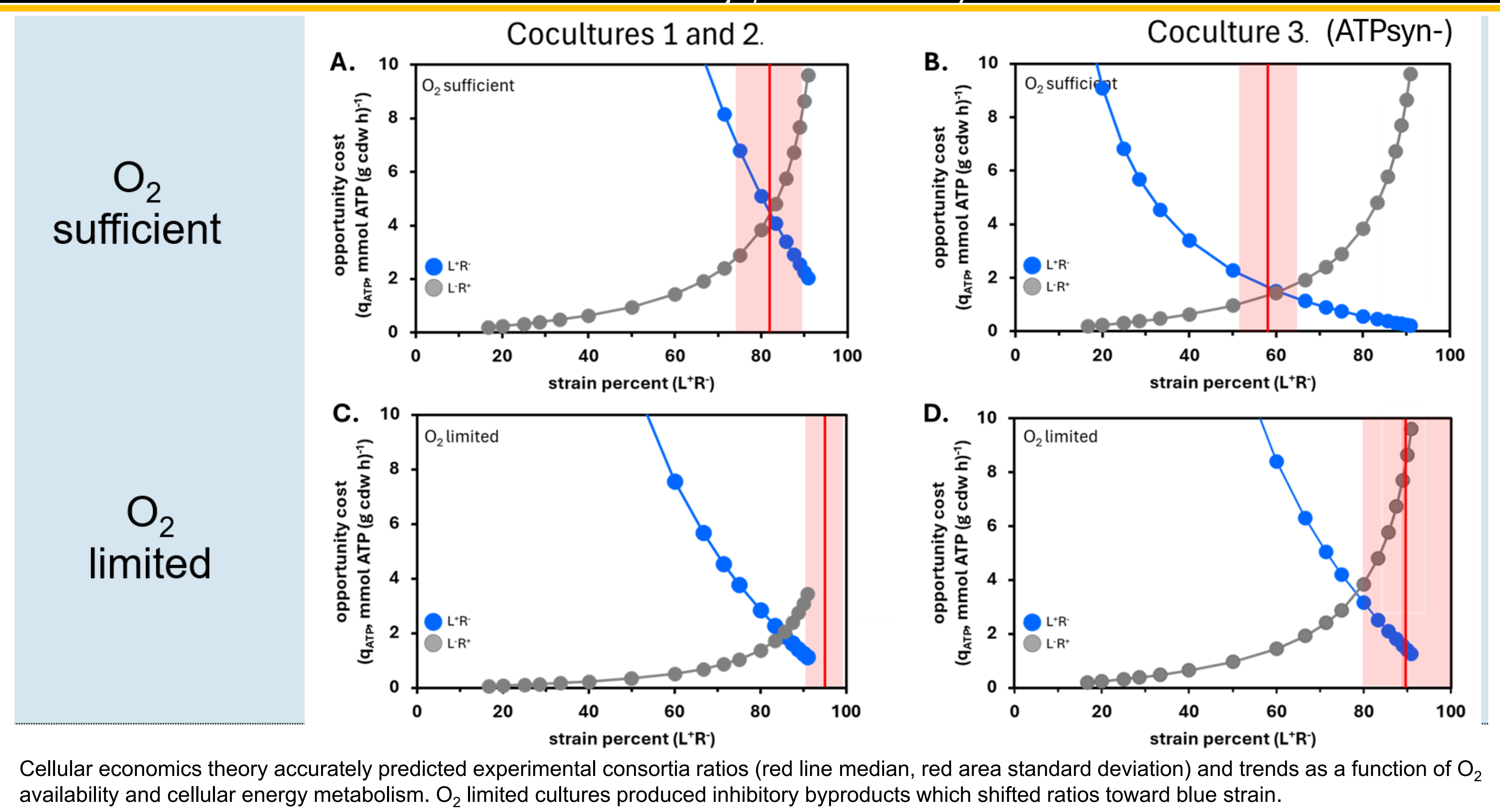


Biofilms: *In situ* strains ratios correlated with *in situ* O_2 levels.

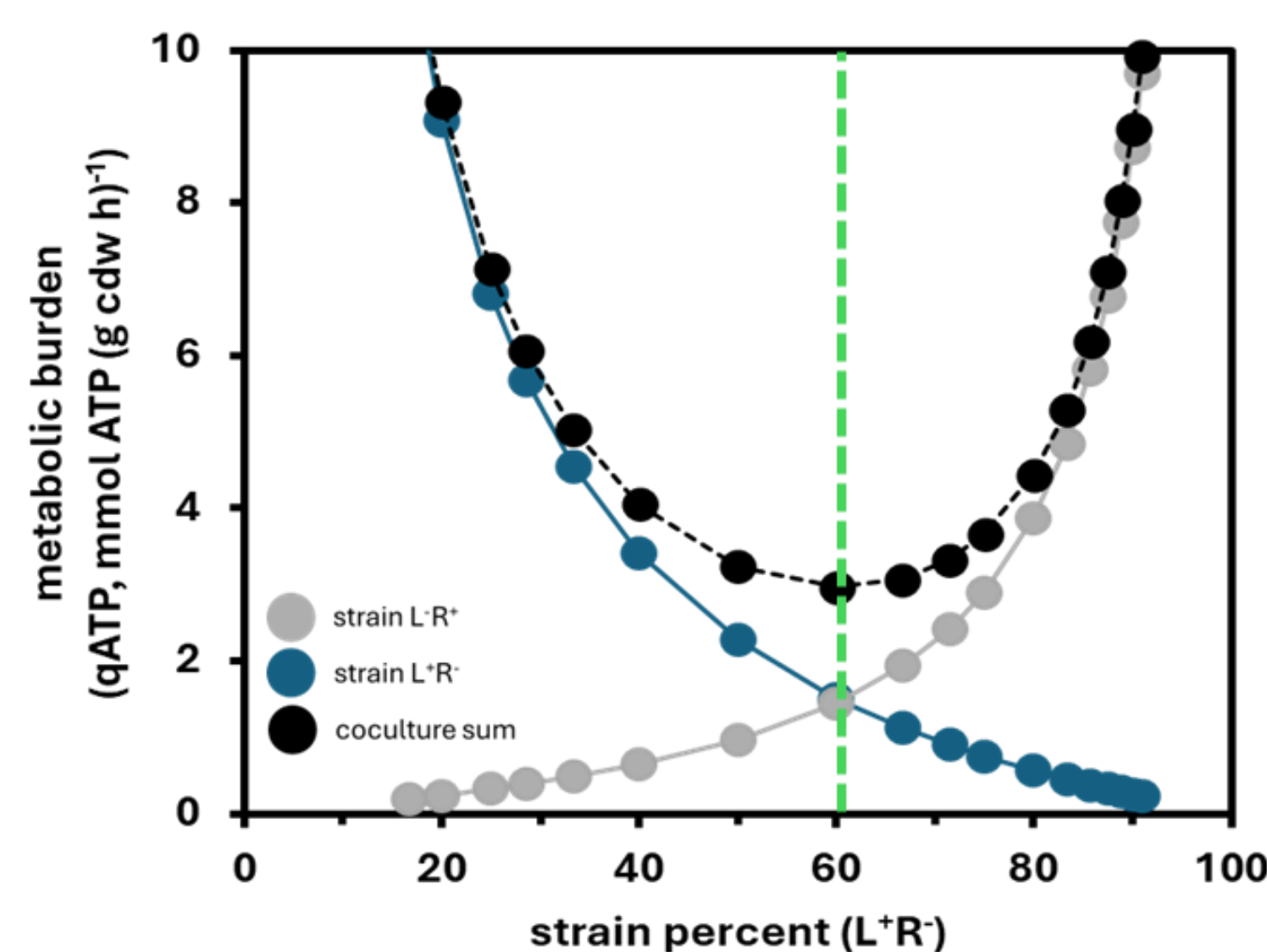


Cellular economics theory worked on seven additional published consortia exchanging amino acids and organic byproducts.

Testing Theory



Cellular economics theory accurately predicted experimental consortia ratios (red line median, red area standard deviation) and trends as a function of O_2 availability and cellular energy metabolism. O_2 limited cultures produced inhibitory byproducts which shifted ratios toward blue strain.



When consortia share the metabolic burden of obligate metabolite exchange, the consortia minimize the overall cost of exchange.

Summary

- Obligate cross feeding consortia assemble at ratios consistent with equal metabolic burden.
- Altering the cellular cost of exchanged metabolites shifted consortia ratios in predictable manner.
- Theory consistent with seven published consortia.
- When obligate cross feeding consortium members have equitable metabolic burden, the consortium as a whole minimizes cost of exchange.

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Reference:

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