Evolution and regulation of microbial secondary metabolism

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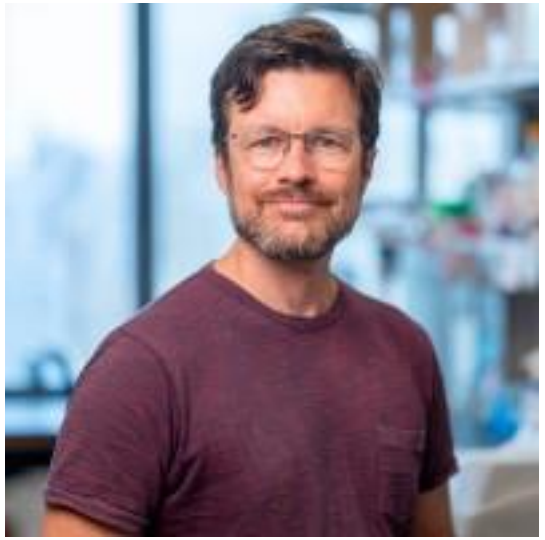

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eLife

RESEARCH ARTICLE

Evolution and regulation of microbial secondary metabolism

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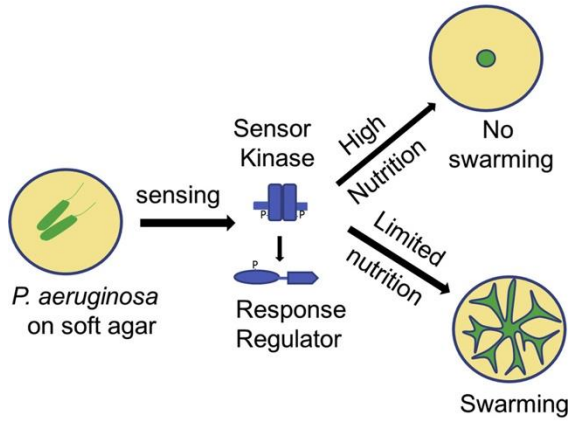
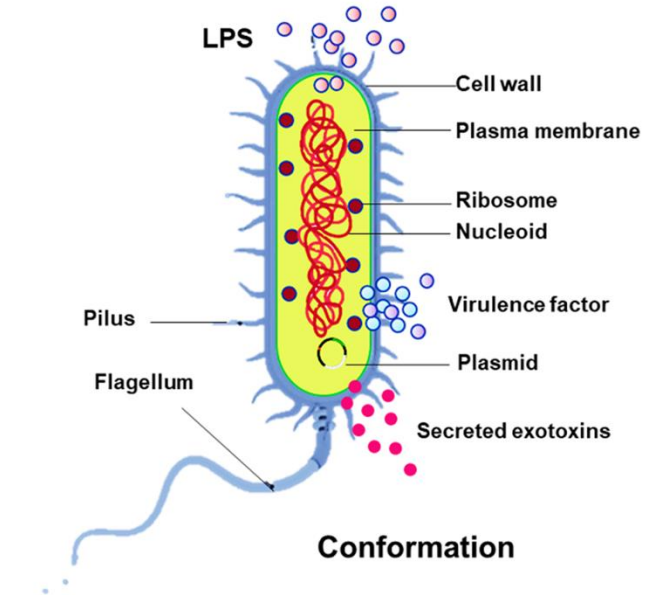
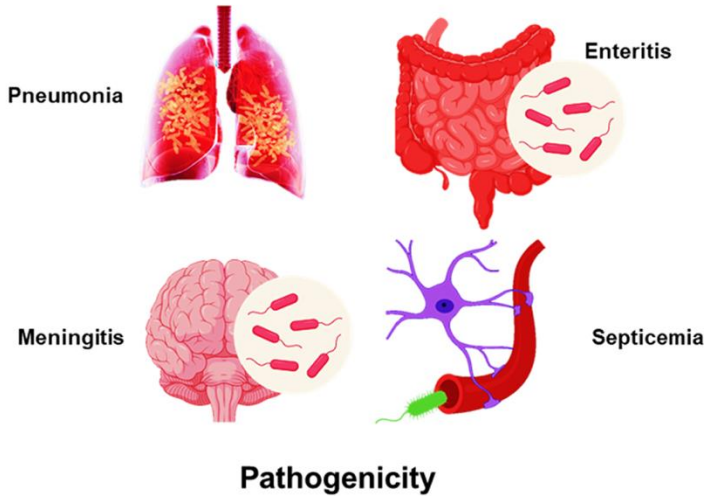
Pseudomonas aeruginosa

- Opportunistic pathogen
- Common infections in cancer patients, cystic fibrosis …
- Biofilm / Swarming behaviour
- Surfactant secretion
- Rhamnolipids

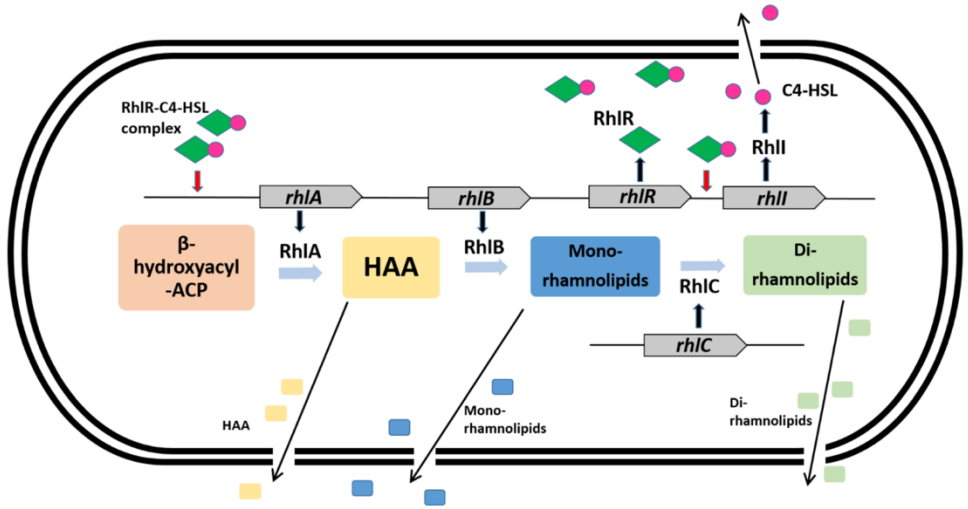


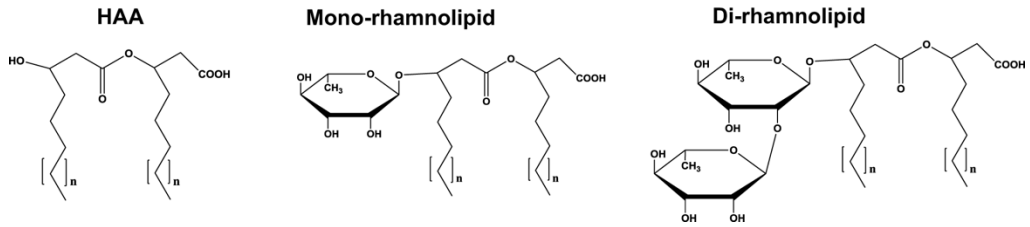


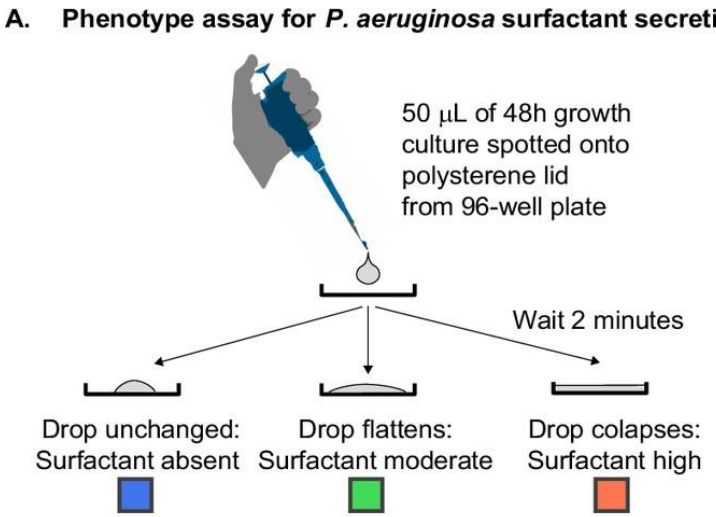


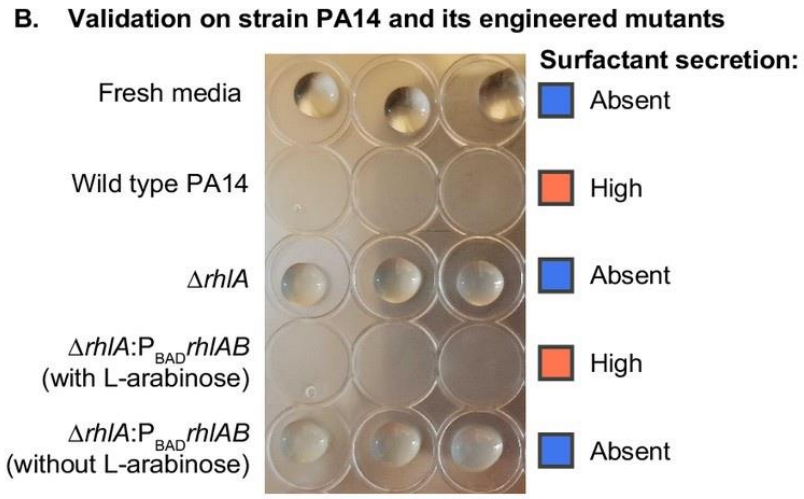
Rhamnolipids

- Secondary metabolites
- (not essential for survival, growth and reproduction)
- Natural surfactants
- Decrease surface tension







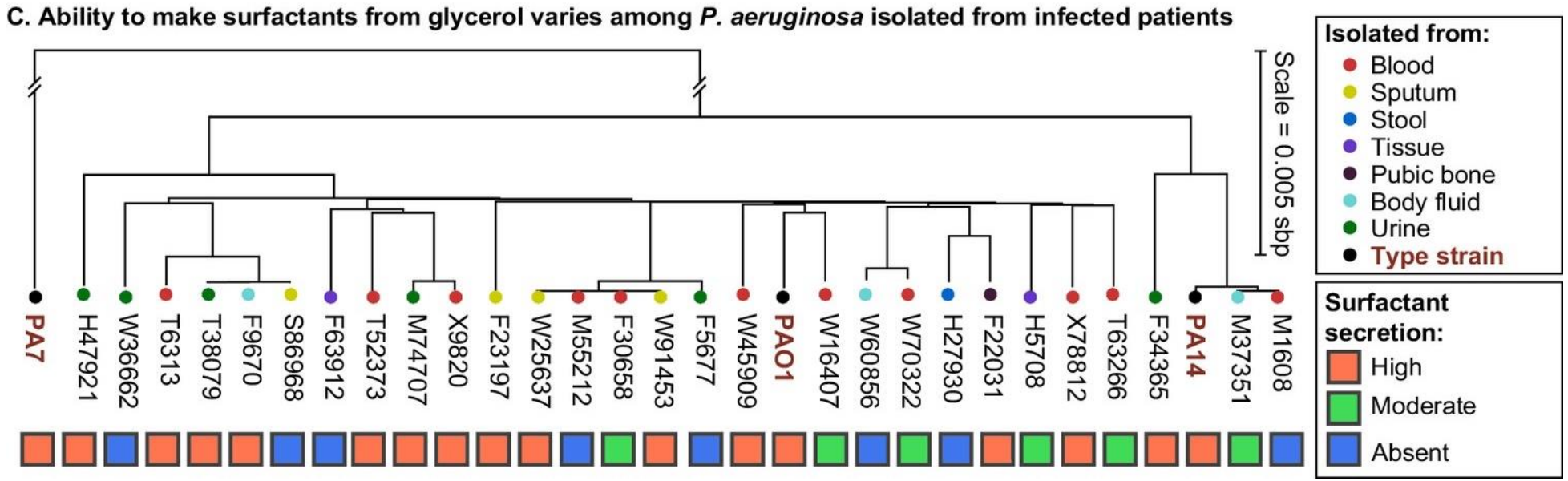


Secondary metabolism

- Relevant for social and ecological interactions
- Surfactant secretion benefits:
  - Motility / Swarming
  - Killing competitors
  - Breaching the epithelial barriers of a host
- Benefit ➔ population, Cost ➔ individual (rB>C)
- Synthesis is highly regulated (Metabolic Prudence strategy):
  - Quorum sensing
  - Nutrients available (high C/N ratio)

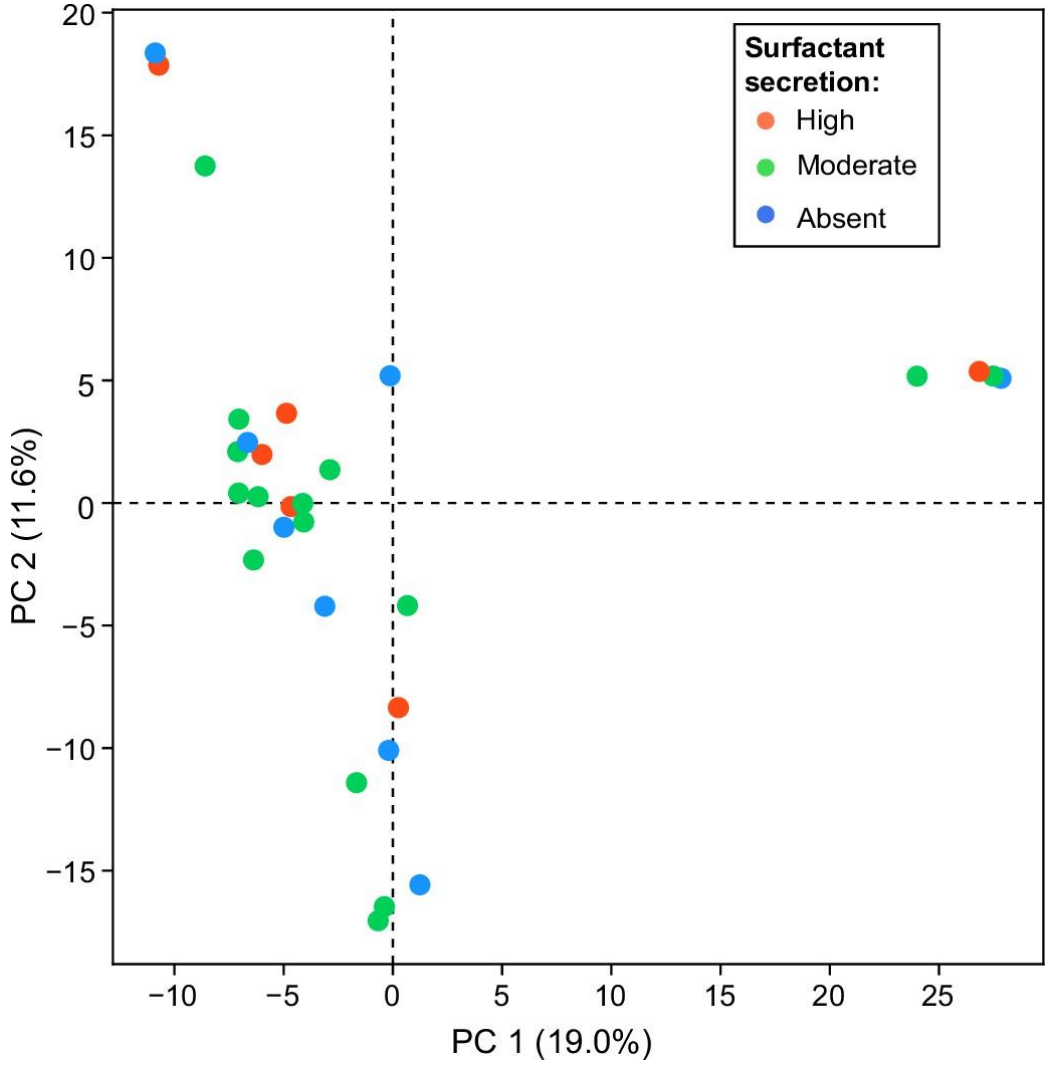
Surfactant production varies inconsistently across the phylogenetic tree

- 28 strains isolated from infected cancer patients (and 3 reference strains)
- Phylogenetic tree from sequence variations in core genome
- Rhamnolipid synthesis genes conserved across all strains



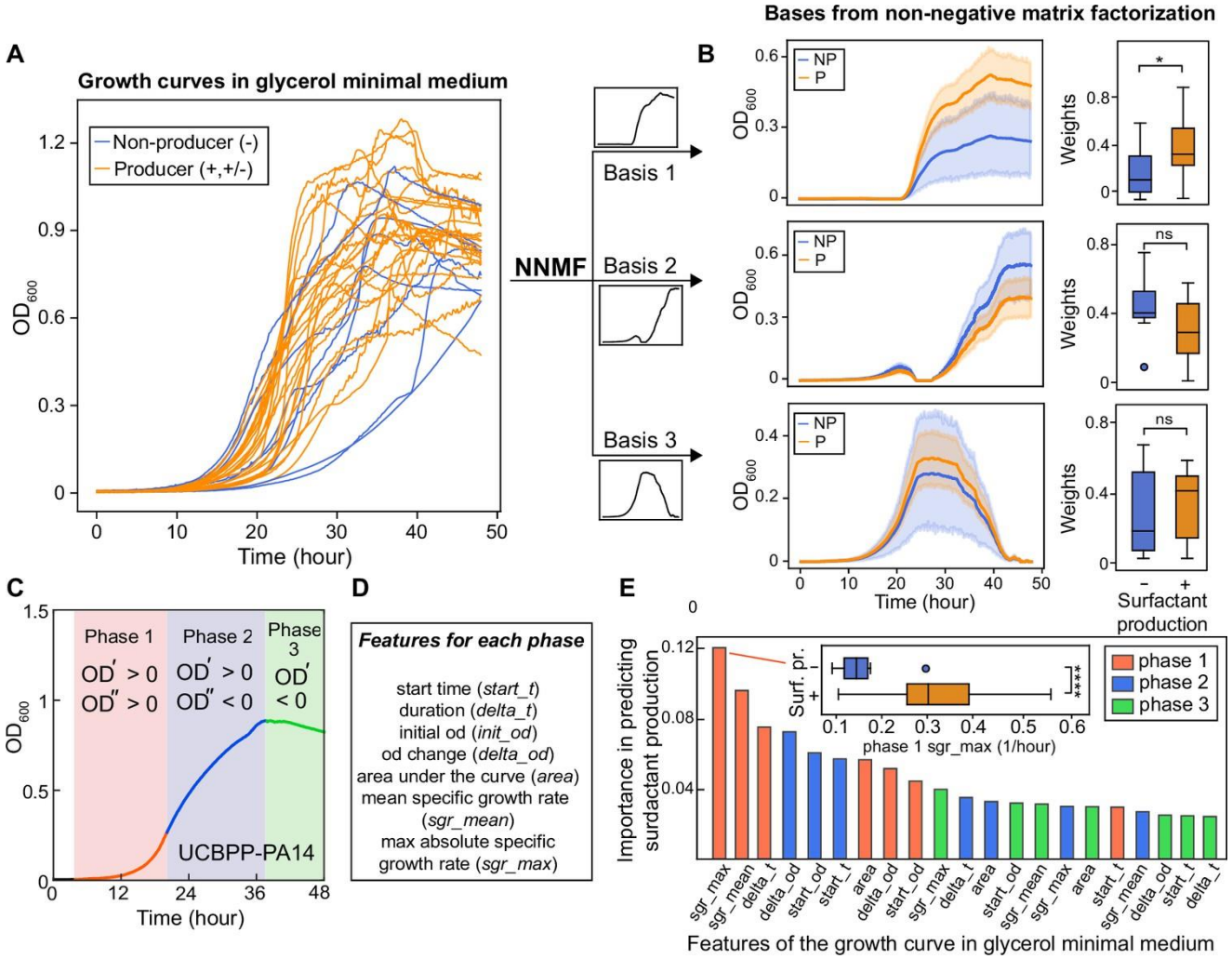
Accessory genome does not explain surfactant secretion phenotype

- Accessory genome gene presence/absence profiles
- 8290 accessory genes, 354 only absent in non-producers
- But no common pattern among non-producers
- No significant differences between surfactant secretion groups



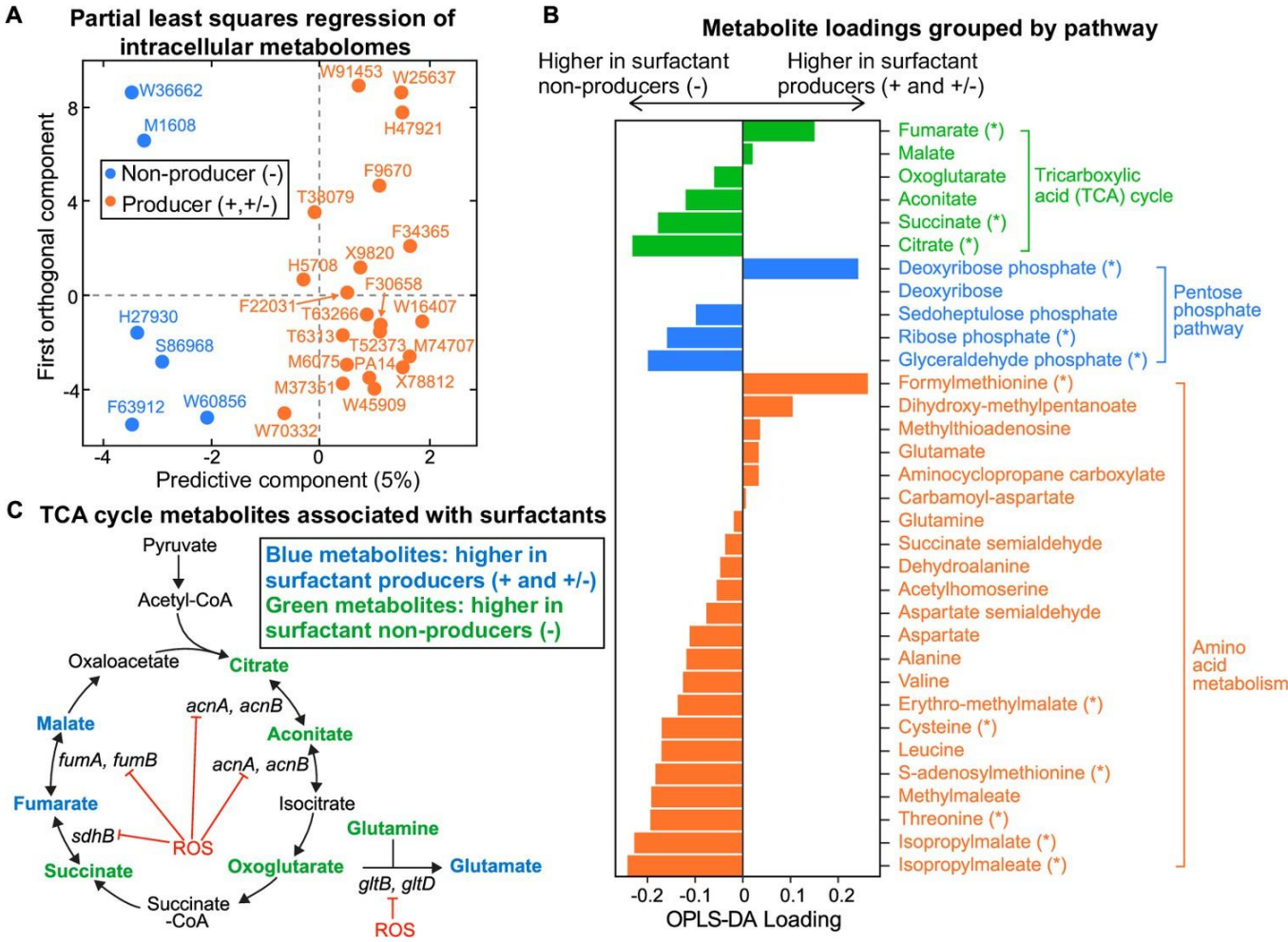
Surfactant secretion lost in strains with slower exponential growth

- Growth on glycerol minimal media
- Growth curves reached similar maximum densities
- Main difference is growth rate in the beginning of exponential phase
- Surfactants are usually produced and secreted only at the end of exponential phase



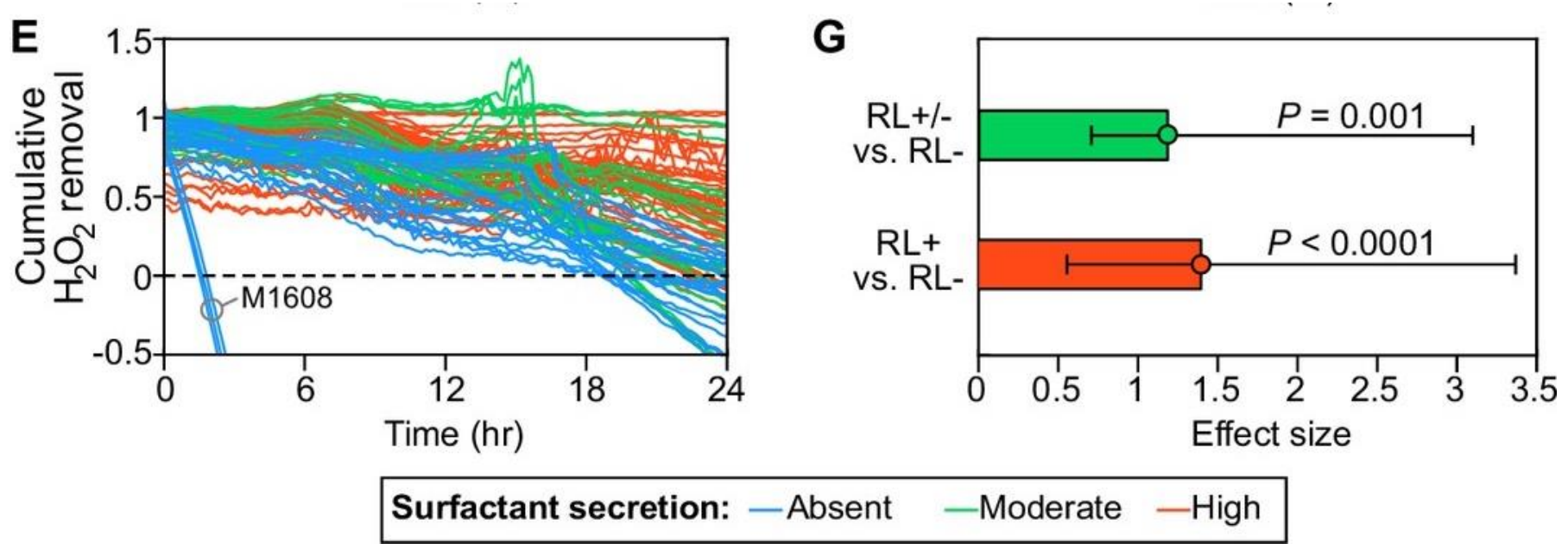
Metabolomics shows differences associated with loss of surfactant production

- OPLS-DA was able to separate producers from non-producers using metabolite profiles
- 15 metabolites had significant differences between groups
- associated with enzymes containing ROS sensitive Fe-S clusters



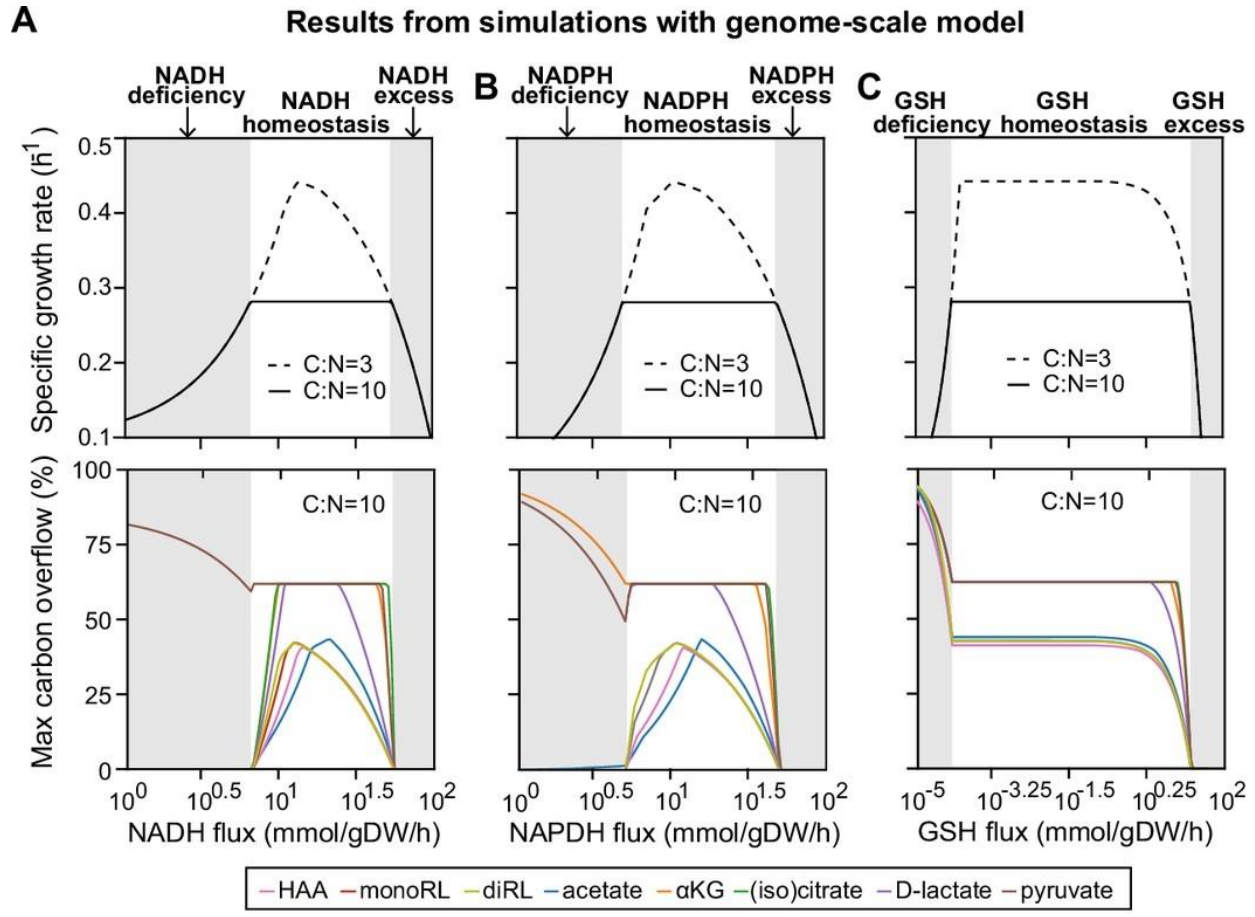
Non-producers are worse at reducing oxidative stress

- Strains were exposed to H_2O_2
- H_2O_2 was measured through fluorescent sensor
- Both moderate and high surfactant producers were significantly more efficient at removing H_2O_2



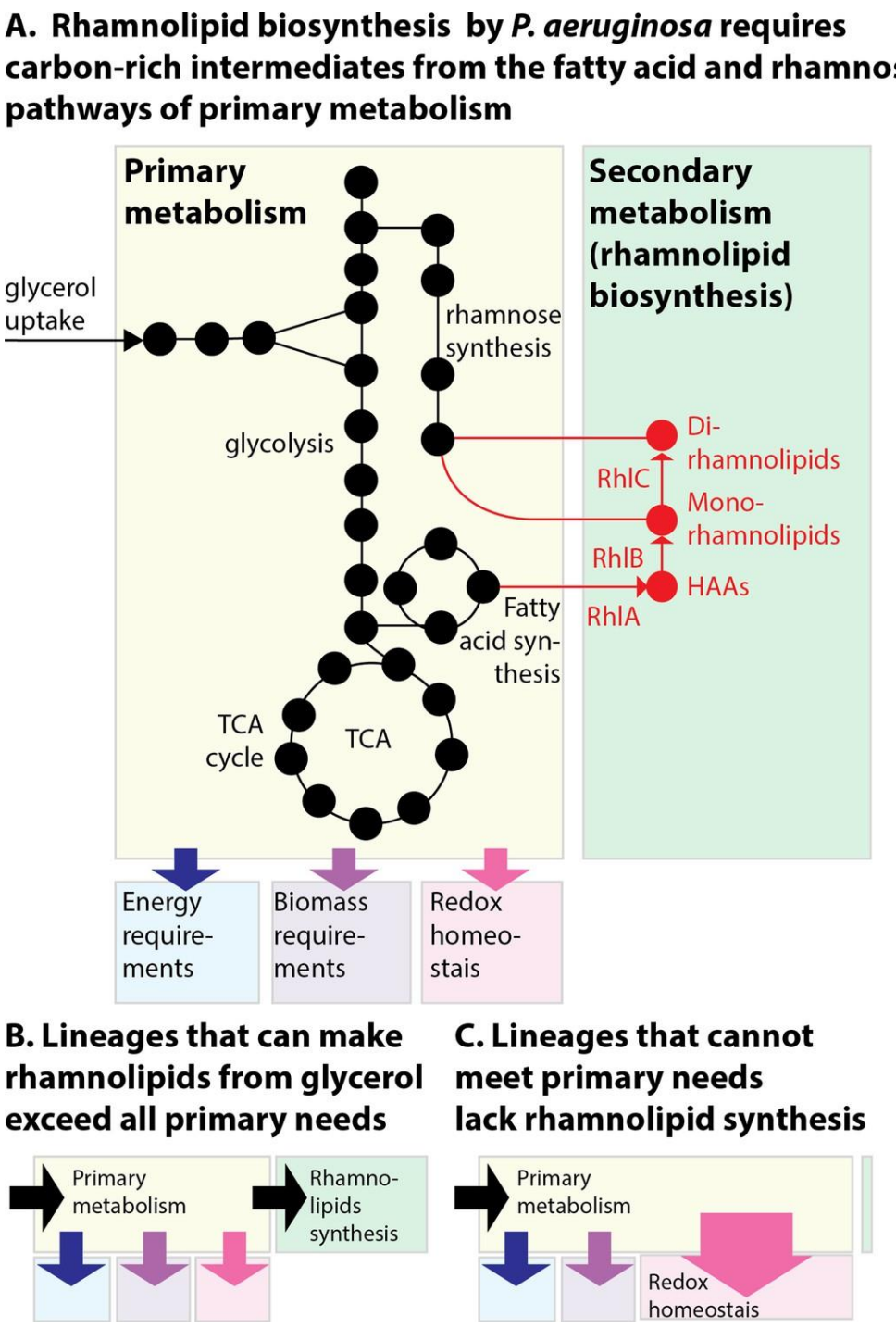
Genome scale metabolic model links oxidative stress, slower growth and lack of surfactants

- Model adapted from *P. putida* model
- C/N=10 – requisite for surfactant production
- Limits growth in normal redox conditions
- Fixed fluxes of NADH, NADPH and GSH synthesis
- High synthesis mimics redox stress
- Rhamnolipid overflow only with redox homeostasis



Explaining Model

- Some strains are not as efficient at maintaining redox homeostasis (why?)
- They require more resources from primary metabolism
- Preventing the activation of rhamnolipid synthesis



Final thoughts

- To fully understand the connection between genotypes and phenotypes we need to understand how they evolved
- Different cellular processes and pathways are highly interdependent
- Some phenotypes can be influenced by genes that we associate to apparently unrelated cellular processes