Multi layer network approch to study metabolic switch in *Streptomyces coelicolor* to produce antibiotics

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Abstract

Antimicrobial resistance in human pathogenic bacteria is an increasing threat in modern society, where many infectious diseases no longer respond to treatments with presently available antibiotics. Global health concern as such, expresses the urgent need to produce a higher yield of existing and novel type of antibiotics. In particular, the genus Streptomyces coelicolor is known to produce over 40 % of all known compounds with antibiotic activity, many of which are used to treat serious microbial infections. Antibiotic production by *Streptomyces Coelicolor* (*S.coelicolor*) is a promising technology but to date has not reached competitive rates and titers. In order to find ways to improve the production of antibiotics, one needs to understand the mechanism on genome and metabolome level that leads to antibiotic production.

Metabolic switch (reorganization of metabolome) in the transition from exponential to stationary growth phase of *S.coelicolor* strongly activates secondary metabolism, where antibiotics are being produced. In order to explore such underlying reorganization of the metabolome, epistatic interaction network, together with gene co-expression network are being constructed along the logistic growth curve, analyzed and mapped on our prior reconstructed genome scaled metabolic model of *S.coelicolor* by constructing flux coupling network. Analyses of the constructed networks reveals modules of genes with aggravating and buffering epistatic interactions, genes modules that are highly co-expressed and finally the relevant metabolic reactions and their fully or partially coupling.

Such multilayer network approach enables far better understanding and possible further manipulation of metabolism of *S.coelicolor* in order to produce antibiotics with a better yield and in addition suggests that metabolic switch could be a possible response on existing robustness of the system.

Methods

Constrained-based optimization methods on genome-scale metabolic model, with optimization of objective function (cell growth) under the steady state assumption and under the constraints on upper and lower bound of reaction fluxes has laid the foundation for computational procedures for suggesting system-level epistatic interactions. By computing growth phenotypes of all single and double knockouts of 1318 metabolic genes in *S.coelicolor*, using the framework of flux balance analysis (FBA), we have identified a distinctive trimodal distribution of the epistatic effects, where gene-pairs are classifeid as buffering, aggrevating or non-interacting.

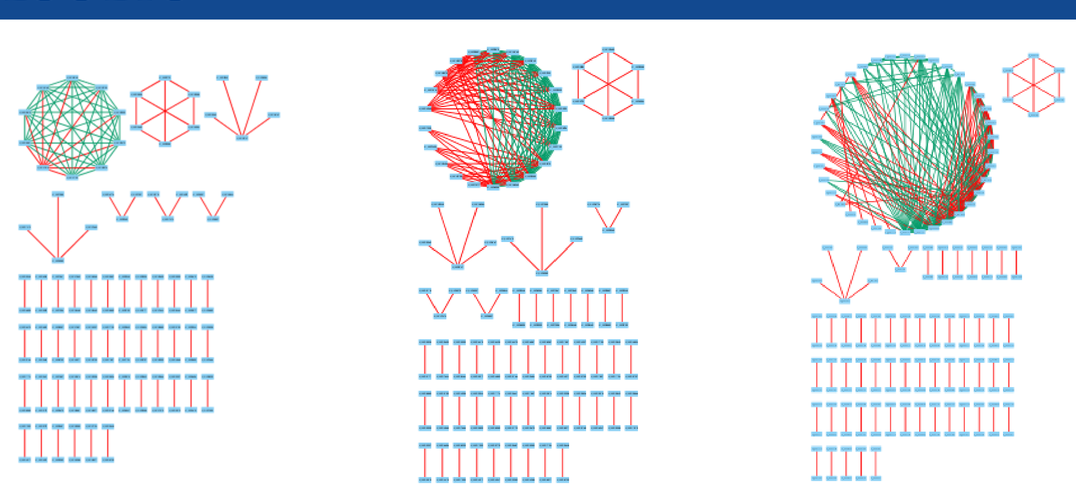
For the purpose of elucidating the toplogical and flux connectivity features of genome scale metabolic reconstruction of *S.coelicolor*, the Flux Coupling Finder framework has been applied to determine whether any two metabolic fluxes are directionally, partially or fully coupled and as such providing additional means to guide genetic manipulations.

Finally weighted gene co-expression network analysis (WGCNA) has been applied to describe correlation patterens among genes across microarray samples and finding clusters (modules) of highly correlated genes.

Conclusion

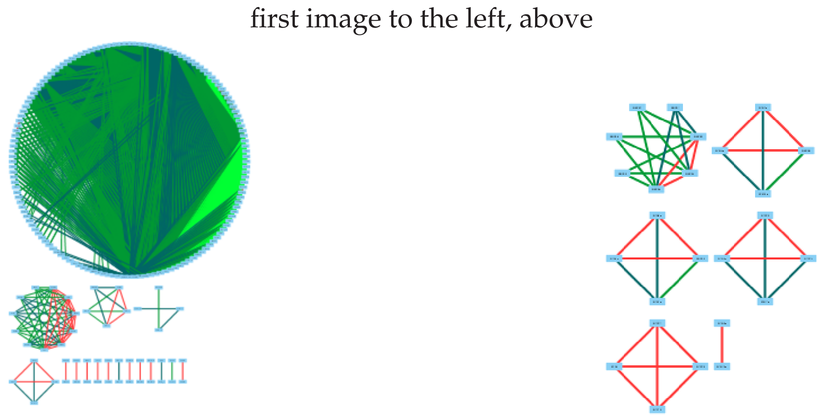
The amount of epistatic interactions as well as the amount of flux coupling between metabolic reactions and gene co-expression network, changes along the growth curve with a higher level of aggrevating/buffering epistasis and a higher level of fully/partially flux coupling upon nutrient depletion, though the ratio between aggrevating vs buffering epistasis and fully vs partally flux coupling stays similar. In addition there is interplay between epistatic, flux coupling and gene co-expression network, where buffering epistasis manly code for fully flux coupled reactions, while aggrevating epistasis catalyze mainly partially and directionally flux coupled metabolic reactions. Such multilayer network approach suggests that metabolic switch could be a possible response on underlaying robustness of the system and enables possible further manipulation of metabolism to produce antibitiotics with a better yield.

Results



Epistatic interaction along logistic growth curve, changing uptake rate of glucose, glutamate and phosphate from 1.0, 0.4 to 0.1 mmol/gDW-hr. Aggrevating and buffering epistasis are marked red and green, respectively. The amount of aggrevating/buffering epistasis increases with nutrient depletion, but the ratio stays similar

Separation of aggrevating(red) and buffering (green) epistatic interactons in connection to the first image to the left, above



Fully flux coupled (red), directionally and partially coupled reactions (versions of green), corresponding to buffering and aggrevating epistatic interactons, respectively. Aggevating epistasis codes mainly for partially flux coupling, while buffering epistasis codes mainly for fixed flux coupling