Construction of an enzyme constrained model of the metabolism of *Pseudomonas putida*

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Type of thesis: Computational work

Required competences: MSc Bioinformatics and knowledge of python. Basic knowledge of models of metabolism is a plus (for instance BPE-34306 or SSB-50806).

Acquired competences:

- Construction of Genome scale models (GEM)
- Incorporation of proteomic data to GEM
- High-thruput database mining
- Microbial metabolism

Starting Date: 2021

Description:

Genome-scale, constraint-based models (GEM) and their derivatives are commonly used to model and gain insights into microbial metabolism. They are mathematical representations of cell metabolism based on annotated genomes and able to establish genotype-phenotype relationships linking genes and enzymes with metabolic reactions. In these models the stoichiometry matrix contains stoichiometric relations between metabolites in metabolic reactions, reactions to account for biomass syntheses, ATP hydrolysis due to maintenance, transport of metabolites between the intracellular and extracellular space and among intracellular compartments. This matrix also contains exchange reactions to represent secretion and uptake of metabolites from the medium. In these models the reversibility of the flux through a reaction is included through flux constrains, but information regarding enzyme kinetics and genetic regulation is absent (Gu et al, 2019).

Sanchez et al (2017) introduced the GECKO framework which generates enzyme constrained models (ecModels) adding an additional constraint to GEM linked to the limited enzyme production capacity of the cell. In these models each metabolic reaction includes an extra entity that represents enzyme usage, which is limited by protein abundance, determined by proteomic measurements or a general constrain in the total protein content of the cells. The molecular weight and turnover (k_{cat}) values of the enzymes influence how enzyme availability limits the flux of specific reactions.

Pseudomonas putida is the microbial cell factory of choice for many biotechnological applications. It has a considerable metabolic versatility, remarkable tolerance to various stress conditions and shows rapid growth with simple nutritional requirements.

This thesis project starts applying the GECKO framework to an existing *P. putida* GEM. High-thruput methods for database mining will be used to extract the relevant information from databases such as BRENDA. Finally, predictions with the original model and the enzyme constrained GEM will be compared to experimental data to study the effect of incorporating a protein availability constraint on flux predictions.

Bilbiography:

Gu, C., Bae Kim, G., Jun Kim, W., Uk Kim, H., & Yup Lee, S. (2019). Current status and applications of genome-scale metabolic models. *Genome Biology*. https://doi.org/10.1186/s13059-019-1730-3

Sánchez, B. J., Zhang, C., Nilsson, A., Lahtvee, P.-J., Kerkhoven, E. J., & Nielsen, J. (2017). Improving the phenotype predictions of a yeast genome-scale metabolic model by incorporating enzymatic constraints. *Mol Syst Biol*, *13*, 935. https://doi.org/10.15252/msb.20167411