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Sensitivity analysis for the reduction of complex metabolism models

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Abstract

Two different model reduction strategies are studied in order to test their applicability to reduce complex metabolism models. Using a model of one pre-identified model set describing complex metabolic dynamics after glucose pulse stimulation, a model reduction method based on the parameter tuning importance is compared with a pca based approach. Up to 49 of 122 parameters are rejected without significant changes of the simulated trajectories and of the flux distribution.

Applying the reduction procedure to 12 other dynamic models reveals a general model structure inconsistency within the description of the pentose phosphate pathway. That points out the need of additional experiments to reproduce metabolite courses especially of this metabolic pathway.

Thus the sensitivity based model reduction procedure is qualified as a promising tool for the model structure check and can be very useful for the entire model validation process which also includes the critical analysis of the data sets underlying the models.

Keywords: Model reduction; Pca; Parameter sensitivity; Model validation; Metabolism

1. Introduction

The well known, widely-used procaryote *Escherichia coli* K12 possesses approximately 4800 genes encoding 2500 proteins [1]. Several hundreds or even thousands of them are expressed and active at the same time sharing a pool of a comparable number of metabolites, co-factors, nucleotides, etc. Despite this complexity, numerous approaches have been studied up to now to quantitatively describe the cellular metabolism. Stoichiometric models based on flux balance analysis [2], metabolic flux analysis using intensive labelling information [3] or structured metabolism models considering in vitro derived enzyme kinetics [4] are only some of the examples that aim at covering metabolism complexity by modelling.

These approaches have in common that they do not use information about in vivo enzyme kinetics although the knowledge about kinetic and thermodynamic properties of all macromolecules in living cells would offer the possibilities of a *modern* metabolic engineering [5]. In their pioneering work Rizzi et al. [6] aimed at identifying in vivo enzyme kinetic data for *Saccharomyces cerevisiae* by performing glucose pulse experiments. Cells which were cultivated under glucose-limited conditions, were stimulated by a glucose pulse which caused a sudden increase of cellular glucose uptake. As a result, intracellular metabolite pools changed significantly which was monitored during a few minutes time-window by rapid cell sampling combined with immediate metabolism inactivation. Because a series of rapid samples was taken, courses of intracellular metabolite changes were observed which were the basis for the subsequent model identification considering 22 metabolite balances and 99 parameters describing yeast's central metabolism. In the following, similar experiments were performed using *E. coli* [7] and *Zymomonas mobilis* [8].

However, potential pitfalls of the procedure become obvious when the model identifiability and resulting model accuracies are taken into account. The underlying data sets consist of intracellular metabolite concentrations which are only accessible with sophisticated analytical approaches (for instance using LC–MS/MS) considering a significant number of sample preparation steps [9]. As a consequence, dynamically changing metabolite levels of an 'average' cell (neglecting any cell distributions in the population) are given which can

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Nomenclature

Metabolites

AcCoA	acetyl-coenzyme A
ADP	adenosindiphosphate
ala	L-alanine
AMP	adenosinmonophosphate
ATP	adenosintriphosphate
BPG	glycerate-1,3-bisphosphate
chor	chorismate
Cit	citrate
DAHP	7-phospho-2-dehydro-3-deoxy-D-arabinoheptonate
DHAP	glycerone phosphate
E4P	D-erythrose-4-phosphate
FBP	β-D-fructose-1,6-bisphosphate
F6P	β-D-fructose-6-phosphate
GAP	glyceraldehyde-3-phosphate
G3P	glycerol-3-phosphate
G6P	α-D-glucose-6-phosphate
ile	isoleucine
kival	α-ketoisovalerate
leu	leucine
mur	subunit of mureine
NAD	diphosphopyridindinucleotide (oxidized)
NADP	diphosphopyridindinucleotide-phosphate (oxidized)
oaa	oxalocetate
PEP	phophoenolpyruvate
2PG	glycerate-2-phosphate
3PG	glycerate-3-phosphate
6PG	6-phospho-D-gluconate
Pyr	pyruvate
R5P	D-ribose-5-phosphate
Ri5P	D-ribulose-5-phosphate
S7P	D-sedoheptulose-7-phosphate
X5P	D-xylulose-5-phosphate

Enzymes

aldo	aldolase
dahps	dahp synthase
eno	enolase
gapdh	glyceraldehyde-3-phosphate dehydrogenase
g3pdh	glycerol-3-phosphate dehydrogenase
g6pdh	glucose-6-phosphate dehydrogenase
pdh	pyruvate dehydrogenase
pfk	phosphofructokinase
pgdh	6-phosphogluconate dehydrogenase
pgi	glucose-6-phosphate isomerase
pgk	phosphoglyceratekinase
pgm	phosphoglyceratemutase
pk	pyruvate kinase
pts	phosphotransferase system
rpe	ribulose-phosphate epimerase
rpi	ribose-phosphate isomerase
ta	transaldolase
tim	triose phosphate isomerase
tka	Transketolase, reaction A
tkb	Transketolase, reaction B

Mathematical terms, variables and subscripts

DE	differential equation
ODE	ordinary differential equation
pca	principal component analysis
t	time
b	metabolite concentration, measured
c	metabolite concentration, simulated
$\mathbf{c}$	vector of all metabolite concentrations, simulated
$\mathbf{c}^0$	vector of all steady-state metabolite concentrations, simulated
$e(\Delta p)$	objective function with respect to a small parameter change
$\mathbf{F}$	parametric Jacobian matrix
g	index, parameter
$\boldsymbol{\Gamma}$	matrix of eigenvectors
γ	eigenvector, represents a column of $\boldsymbol{\Gamma}$
h	index, parameter
i	index, metabolite/variable
j	index, time interval
$\mathbf{J}$	Jacobian matrix
k	index, parameter (and row in $\boldsymbol{\Gamma}$)
λ	eigenvalue
m	total number of independent metabolites/variables
n	total number of time intervals
$\mathbf{N}$	stoichiometry matrix
v	stoichiometric coefficient
$\mathrm{os}(p)$	overall sensitivity with respect to parameter p
p	total number of parameter
$\mathbf{p}$	vector of parameters
Δp	small parameter change
$\mathbf{R}$	covariance matrix
s	normalized, local sensitivity coefficient
$\mathbf{S}$	sensitivity matrix
v	reaction rate
$\mathbf{v}$	vector of rate equations
x	discarded entry of an eigenvector
ζ	error functional

possess significant measurement errors. To build up the structured metabolism model, well-known enzyme databases like (BRENDA [10], ExPASy [11], etc.) are typically used which have the intrinsic problem that their information is usually derived from in vitro enzyme kinetic experiments. The application of these 'in vitro'