

**Supplementary Table 1 - List of free parameters**

	Nominal Value	Sensitivity test	Comments and References
Flux Balance Analysis			
Stoichiometric matrix ( $S_{ij}$ )	See: <a href="http://www.genome.org/cgi/data/13/2/244/DC1/1">http://www.genome.org/cgi/data/13/2/244/DC1/1</a>	Randomization: [ $\delta S_1$ ] If $S_{ij} \neq 0$ , $S_{ij} \rightarrow S_{ij}(1 \pm 0.5)$ with Prob.=0.01 [ $\delta S_2$ ] If $S_{ij} = 0$ , $S_{ij} \rightarrow \pm 1$ with Prob.= $10^{-3}$	S used in this work: <sup>15</sup> . Other useful resources: <sup>42,43</sup>
Biomass composition	See: <a href="http://www.genome.org/cgi/data/13/2/244/DC1/3">http://www.genome.org/cgi/data/13/2/244/DC1/3</a>	[ $\delta S_1, \delta S_2$ ] Randomized together with $S_{ij}$	Composition used: <sup>15</sup> . See also sensitivity analysis in <sup>44</sup>
Objective function	$Z = V_{\text{growth}}$	-	For testing of objective function, see: <sup>45</sup> . Sensitivity around optima: <sup>13,46</sup>
Nutrient uptake bounds	Glucose: 20 mmol/(grDM*h) Oxygen: 5 mmol/(grDM*h)	[C-] Glucose=10 [C+] Glucose=25 [C++] Glucose=40 [O-] Oxygen=2.5 [O+] Oxygen=10 [cyt] cytr unbound [Thr] Threonine=0 [AC] Acetate=20	Sensitivity also explored in: <sup>47,48</sup>
Thermodynamic and maintenance constraints	Irreversibility constraints from: <a href="http://www.genome.org/cgi/data/13/2/244/DC1/1">http://www.genome.org/cgi/data/13/2/244/DC1/1</a> Maintenance: $V_M=1$	-	-
Deletion constraints	See Supplementary methods for model details	Single and double deletions performed for all metabolic genes	Sensitivity to full and partial deletions: <sup>13,47</sup> . Gene deletions/additions: <sup>49</sup>
Epistatic network analysis			
Direct vs. associative weight ( $\alpha$ )	0.3	[ $\alpha$ ] $\alpha=1$	Prism algorithm; See Methods
Similarity enhancement ( $\gamma$ )	100	[ $\gamma$ ] $\gamma=1$	Prism algorithm; See Methods
Cutoff for aggravating and buffering ( $\theta_-, \theta_+$ )	(-0.25, 0.9)	[ $\theta$ ] (-0.5, 0.5)	Double mutants and epistatic network, Methods
Noise cutoff ( $\epsilon_{\text{noise}}$ )	0.01	[ $\epsilon_{\text{noise}}$ ] $\epsilon_{\text{noise}}=0.1$	Discard pairs of mutants with noisy $\tilde{\epsilon}$ : $\left  \frac{1 - \min(W_X, W_Y)}{1 - W_X W_Y} - 1 \right  < \epsilon_{\text{noise}}$

(For references, see **Supplementary References** online)