A modelling approach to quantify dynamic crosstalk between the pheromone and the starvation pathway in baker's yeast

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Supplementary Material

The mathematical model

Model equations

State variables of the system and their respective derivatives are denoted as $c_i(t)$ [nM] and $c_i'(t)$, i=1, ..., 24, reaction rates as v_j, j=1, …, 31 and kinetic constants as k_l, l=1, …, 34. For the respective biological counterparts refer to the mathematical model description in the main test and Figure 2. The functions $\alpha(t)$ and $\beta(t)$ denote the input function, where $\alpha(t)$ was modelled as a unit step function of 10 minutes duration and $\beta(t)$ was modelled as a unit step function of 6 hours duration.

$v_1 = k_1 \cdot c_1(t) \cdot c_2(t)$

$v_2 = k_2 \cdot c_3(t) \cdot c_4(t) \cdot \alpha(t)$

$v_3 = k_3 \cdot c_5(t) \cdot c_6(t)$

$v_4 = k_4 \cdot c_7(t)$

$v_5 = k_5 \cdot c_8(t)$

$v_6 = k_6 \cdot c_6(t) \cdot c_{10}(t)$

$v_7 = k_7 \cdot c_{10}(t)$

$v_8 = k_8 \cdot c_{11}(t)$

$v_9 = k_9 \cdot c_5(t) \cdot c_{12}(t)$

$v_{10} = k_{10} \cdot c_{13}(t)$

$v_{11} = k_{11} \cdot c_{14}(t)$

$v_{12} = k_{12} \cdot c_{10}(t) \cdot c_{12}(t)$

$v_{13} = k_{13} \cdot c_2(t) \cdot \beta(t)$

$v_{14} = k_{14} \cdot c_{16}(t)$

$v_{15} = k_{15} \cdot c_{12}(t)\ c_{16}(t) + k_{30} \cdot c_{12}(t) \cdot c_{11}(t)$

$v_{16} = k_{16} \cdot c_{15}(t) + k_{28} \cdot c_{15}(t) \cdot c_9(t)$

$v_{17} = k_{17} \cdot c_{17}(t)$

$v_{18} = k_{18} \cdot c_{12}(t) \cdot c_{18}(t)$

$v_{19} = k_{19} \cdot c_{18}(t) \cdot c_9(t) + k_{29} \cdot c_{18}(t) \cdot c_{15}(t)$

$v_{20} = k_{20} \cdot c_{19}(t)$

$v_{21} = k_{21} \cdot c_{21}(t)$

$v_{22} = k_{22} \cdot c_{12}(t) \cdot c_{22}(t)$

$v_{23} = k_{23} \cdot c_{22}(t) \cdot c_{15}(t)$

$v_{24} = k_{24} \cdot c_{22}(t) \cdot c_{9}(t)$

$v_{25} = k_{25} \cdot c_{23}(t)$

$v_{26} = k_{26} \cdot c_{9}(t)$

$v_{27} = k_{27} \cdot c_{3}(t)$

$v_{28} = k_{31} \cdot c_{19}(t)$

$v_{29} = k_{32} \cdot c_{20}(t)$

$v_{30} = k_{33} \cdot c_{23}(t)$

$v_{31} = k_{34} \cdot c_{24}(t)$

$c_1'(t) = -v_1 + v_7 + v_{27}$

$c_2'(t) = -v_1 - v_{13} + v_{14} + v_{27}$

$c_3'(t) = v_1 - v_2 - v_{27}$

$c_4'(t) = -v_2 + v_7$

$c_5'(t) = v_2 - v_3 - v_9$

$c_6'(t) = -v_3 - v_6 + v_{26}$

$c_7'(t) = v_3 - v_4$

$c_8'(t) = v_4 - v_5 + v_6$

$c_9'(t) = v_5 - v_{26}$

$c_{10}'(t) = v_5 - v_6 - v_7 + v_{11} - v_{12}$

$c_{11}'(t) = v_7 - v_8$

$c_{12}'(t) = -v_9 - v_{12} - v_{15} + v_{16} + 2\,v_{17} - 2\,v_{18} + v_{21} - v_{22}$

$c_{13}'(t) = v_9 - v_{10}$

$c_{14}'(t) = v_{10} - v_{11} + v_{12}$

$c_{15}'(t) = v_{11} + v_{15} - v_{16}$

$c_{16}'(t) = v_{13} - v_{14}$

$c_{17}'(t) = -v_{17} + v_{18}$

$c_{18}'(t) = v_{17} - v_{18} - v_{19} + v_{28}$

$c_{19}'(t) = v_{19} - v_{28}$

$c_{20}'(t) = v_{20} - v_{29}$

$c_{21}'(t) = -v_{21} + v_{22}$

$c_{22}'(t) = v_{21} - v_{22} - v_{23} - v_{24} + v_{30}$

$c_{23}'(t) = v_{23} - v_{30}$

$c_{24}'(t) = v_{25} - v_{31}$

Modelparameters

The parameters have the units [min^{-1}] or [min^{-1} nM^{-1}] depending on whether they are involved in monomolecular or bimolecular reactions, respectively. Standard parameter value is one for most parameters. This largely concerns binding, dissociation and transformation processes.

$k_2 = k_3 = k_4 = k_5 = k_6 = k_9 = k_{10} = k_{11} = k_{12} = k_{17} = k_{19} = k_{20} = k_{21} = k_{22} = k_{23} = k_{25} = k_{27} = k_{31} = k_{32} = k_{33} = k_{34} = 1$.

Some processes are assumed to occur at a lower rate, like degradation, lumped processes and diffusion limited processes. E.g. the phosphorylations of the MAPK cascade of the filamentous growth pathway are supposed to occur slower than the scaffold protein dependent phosphorylations.

$k_8 = k_{13} = k_{14} = k_{15} = k_{16} = k_{26} = k_{30} = 0.1$

Two processes are assumed to occur fast, i.e. the dissociation of the scaffold complex,

$k_7 = 10$, and we assume a stronger association of Kss1 to Ste12/Ste12 than to Ste12/Tec1, $k_{18} = 10$.

Finally, four processes are assumed to be slow. Three of them describe several processes that are lumped into one, i.e. scaffold complex formation ($k_1 = 0.01$) degradation of Tec1 induced by Fus3PP ($k_{24} = 0.01$) and dephosphorylation of Kss1PP regulated by Fus3PP ($k_{28} = 0.01$). Finally, Kss1 activation of Ste12Ste12 is considered to the activation caused by Fus3PP ($k_{29} = 0.01$).

Initial values

We took the molecule numbers from Yeast GFP Fusion Localization Database (YGFPDB) (http://yeastgfp.ucsf.edu, [1]) and transformed them to nM by assuming an average cell volume of $6.5 \cdot 10^{-17}$ m^3. This resulted in an approximate transformation factor for the molecule numbers of 39^{-1}. Initial concentrations of complexes were derived from their least abundant component. The steady state concentrations according to no input signal were used as initial concentrations for the simulation runs (Supplementary Table S1).

Sensitivity analysis

In the sensitivity analysis all parameters were changed by a factor of 0.1 and 10 and a standard sensitivity measure S was calculated as

$$S = \frac{\Delta M / M}{\Delta p / p},$$

where Δp is the parameter change, p the standard value of the parameter, ΔM is the response of some measure M compared to the standard situation. As response variable we analysed the time integral I, the first local maximum M and the time of the first local maximum t_M of simulated time courses of Fus3PP, Kss1PP, PREPs and FREPs, in a situation of the two stimuli being present, alone and together, respectively. Including the four parameters for length and strength of stimulus and initial conditions, we analysed 47 parameters.

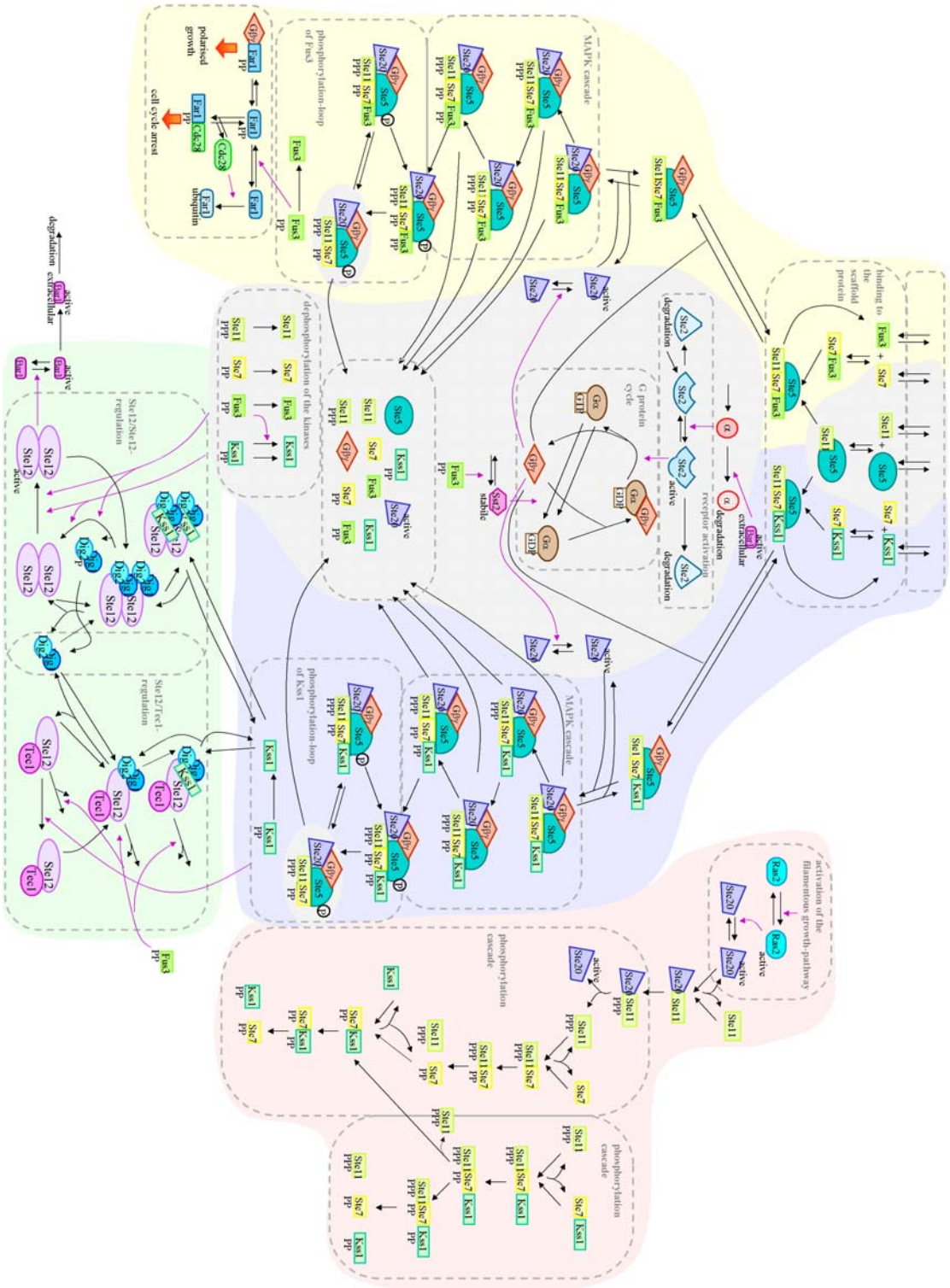
Only few parameter changes prompted a sensitivity greater than five, which was arbitrarily set as a threshold where parameters were considered sensitive. The sensitive parameters upon a pheromone stimulus, i.e. those affecting Fus3PP and PREPs, were those affecting the breakdown rates of Fus3PP, PREPs and the scaffold complex c_{10} as well as the synthesis rate of the inactive transcription complex c_{17} and c_{18}. Regarding the β-stimulus, only the FREPs breakdown rate was sensitive.

References

1. Ghaemmaghami, S., Huh, W. K., Bower, K., Howson, R. W., Belle, A., Dephoure, N., O'Shea, E. K. & Weissman, J. S. (2003) Global analysis of protein expression in yeast, *Nature.* **425**, 737-41.

Supplementary Figures

Supplementary Figure S1: Extended model scheme of the pheromone and the filamentous growth pathway. Yellow background: the pheromone response pathway using the MAPK Fus3. Blue background: the pheromone response pathway using the MAPK Kss1. Yellow-blue background: reactions/components of the pheromone pathway which are independent of the choice of the MAPK. Pink background: the filamentous growth pathway. Green background: regulation of the transcription factors. Pink arrows: regulatory influences. Black arrows: reactions. For sake of clarity, some modules (e.g. receptor activation) are delimited with grey dashed lines.



Supplementary Tables

Supplementary Table S1: Nonzero steady state concentrations used as initial concentrations for the simulations.

Component	**Steady state concentration [nM]**
Ste5 (c_1)	42.3
Ste11 (c_2)	13.3
Ste5/ste11 (c_3)	5.6
Gβγ (c_4)	53.0
Fus3 (c_6)	217.0
Kss1 (c_{12})	54.4
Ste12/Kss1 (c_{17})	35.9
Ste12/Ste12 (c_{18})	0.07
Ste12/tec1/Kss1 (c_{21})	13.7
Ste12/Tec1 (c_{22})	0.25

Supplementary Table S2: Mean Spearman rank correlation coefficients r_S and their respective standard deviations between activation measures for Monte Carlo simulations of 500 runs. The mean was taken over the crosstalk measures C, S_i, S_e, F and S separately for PREPs and FREPs that are depicted in the upper triangle and the lower triangle of the table, respectively.

		PREPs			
		Integral	***Maximum***	t_M	τ
FREPs	***Integral***		0.9 ±0.1	0.2 ± 0.3	0.1 ± 0.4
	Maximum	0.9 ± 0.0		0.2 ± 0.2	0.3 ± 0.3
	t_M	0.3 ± 0.4	0.2 ± 0.3		0.4 ± 0.2
	τ	0.3 ± 0.4	0.1 ± 0.3	0.5 ± 0.1	