

Supporting Information

Model Development

As mentioned in the main text, our model will be restricted to the signaling process and gene regulation in the nucleus, with active MAP kinases Fus3* (denoted by x) and Kss1* (y) as the input. A detailed representation of our full model is illustrated in Fig. S1, and can be described by a set of ordinary differential equations (ODEs):

$$\frac{dS_0}{dt} = -g_S(x, y)S_0 + d_S(x, y)S_1 - \lambda_{S_0}S_0 + \nu_p m_S \quad (1)$$

$$\frac{dS_1}{dt} = g_S(x, y)S_0 - d_S(x, y)S_1 - \lambda_{S_1}S_1 - 2a_S S_1^2 + 2b_S S_2 - a_T S_1 T_1 + b_T T_2 + \Delta\lambda_{T_2}(x)T_2 \quad (2)$$

$$\frac{dS_2}{dt} = a_S S_1^2 - b_S S_2 - \lambda_{S_2}S_2 \quad (3)$$

$$\frac{dT_0}{dt} = -g_T(x, y)T_0 + d_T(x, y)T_1 - \lambda_{T_0}(x)T_0 + \nu_p m_T \quad (4)$$

$$\frac{dT_1}{dt} = g_T(x, y)T_0 - d_T(x, y)T_1 - a_T S_1 T_1 + b_T T_2 - \lambda_{T_1}(x)T_1 \quad (5)$$

$$\frac{dT_2}{dt} = a_T S_1 T_1 - b_T T_2 - \lambda_{T_2}(x)T_2 \quad (6)$$

$$\frac{dm_S}{dt} = \mu_S P_S(S_2) - \lambda_m m_S \quad (7)$$

$$\frac{dm_T}{dt} = \mu_T P_T(T_2) - \lambda_m m_T. \quad (8)$$

A description of the symbols appearing in these equations can be found in Table 1. As argued in the main text, we have included Fus3*-mediated degradation of the Tec1*-Ste12* heterodimer that selectively targets Tec1*. This is accomplished via the term $\Delta\lambda_{T_2}(x)$ which is the Fus3*-dependent contribution to the degradation rate: $\Delta\lambda_{T_2}(x) = \lambda_{T_2}(x) - \lambda_{T_2}(0)$, where $\lambda_{T_2}(0)$ is the constitutive degradation rate in the absence of Fus3*. We further assume constant deactivation rates $d_S(x, y) = d_S, d_T(x, y) = d_T$, and assume that the degradation rates are partially homogeneous: $\lambda_{S_0} = \lambda_{S_1} = \lambda_{S_2}/w \equiv \lambda_S$, $\lambda_{T_0}(x) = \lambda_{T_1}(x) = \lambda_{T_2}(x)/w \equiv \lambda_T(x)$. Here, the parameter w controls the relative stability of the active dimers.

This model can be simplified using a quasi-equilibrium approximation for Eqs. 3, 6, 7, and 8:

$$\frac{dS_0}{dt} = -g_S(x, y)S_0 + d_S S_1 - \lambda_S S_0 + \lambda_S S_b P_S(S_2) \quad (9)$$

$$\frac{dS_1}{dt} = g_S(x, y)S_0 - d_S S_1 - \lambda_S S_1 - 2w\lambda_S S_2 - w\lambda_T(0)T_2 \quad (10)$$

$$\frac{dT_0}{dt} = -g_T(x, y)T_0 + d_T T_1 - \lambda_T(x)T_0 + \lambda_T(0)T_b P_T(T_2) \quad (11)$$

$$\frac{dT_1}{dt} = g_T(x, y)T_0 - d_T T_1 - \lambda_T(x)T_1 - w\lambda_T(x)T_2, \quad (12)$$

where

$$S_2 = \frac{S_1^2}{K_S}, \quad T_2 = \frac{S_1 T_1}{K_T(x)}, \quad K_S = \frac{b_S + \lambda_S}{a_S}, \quad \text{and} \quad K_T(x) = \frac{b_T + \lambda_T(x)}{a_T}. \quad (13)$$

By assumption, most Ste12 and Tec1 are locked in inactive forms by abundant Dig1/2 in resting cells. Therefore, the initial conditions of the above ODEs are: $S_0(t=0) = S_b$, $S_1(t=0) = 0$, $T_0(t=0) = T_b$, and $T_1(t=0) = 0$. S_b and T_b denote the initial abundances of Ste12 and Tec1, respectively, and are determined as follows:

$$S_b = \frac{\nu_p \mu_S}{\lambda_m \lambda_S} \quad \text{and} \quad T_b = \frac{\nu_p \mu_T}{\lambda_m \lambda_T(0)}. \quad (14)$$

For the promoter activities we use simple Hill equations

$$P_S(S_2) = \frac{1 + F_S(S_2/S_{th})^{n_1}}{1 + (S_2/S_{th})^{n_1}} \quad \text{and} \quad P_T(T_2) = \frac{1 + F_T(T_2/T_{th})^{n_2}}{1 + (T_2/T_{th})^{n_2}}, \quad (15)$$

where n_1 and n_2 are Hill coefficients describing the cooperativity, and F_S and F_T represent the maximal fold changes of gene expression of *STE12* and *TEC1*, respectively. To capture the saturation at high Fus3* levels, we will take for the degradation rates of Tec1 species

$$\lambda_T(x) = \lambda_T(0) \frac{1 + F_\lambda(x/L_{th})^{n_3}}{1 + (x/L_{th})^{n_3}}. \quad (16)$$

Finally, Fus3* and Kss1*, as mentioned in the main text, are equally competent to activate Ste12 and Tec1. Hence, the activation rates may be expressed in additive forms:

$$g_S(x, y) = g_{Sm} \frac{x + y}{k_S + (x + y)} \quad \text{and} \quad g_T(x, y) = g_{Tm} \frac{x + y}{k_T + (x + y)}. \quad (17)$$

Table 2 lists all the parameters involved in the reduced ODE system. Their values are based on, whenever possible, on experimental data.

Steady State Solution

To analyze the steady state solutions for our 4 variable ODEs, we can first eliminate S_0 and T_0 , resulting in:

$$\frac{dS_1}{dt} = \frac{d_S S_1 + \lambda_S S_b P_S(S_2)}{1 + \lambda_S/g_S(x, y)} - d_S S_1 - \lambda_S S_1 - 2w\lambda_S S_2 - w\lambda_T(0)T_2 \quad (18)$$

$$\frac{dT_1}{dt} = \frac{d_T T_1 + \lambda_T(0)T_b P_T(T_2)}{1 + \lambda_T(x)/g_T(x, y)} - d_T T_1 - \lambda_T(x)T_1 - w\lambda_T(x)T_2. \quad (19)$$

To investigate the dependence of the steady state solution on w we examine two separate cases.

Case I: $w \rightarrow 0$

In the limit of extremely strong cooperative stability for S_2 and T_2 , i.e. $\lambda_{S_2} \rightarrow 0$, we obtain

$$S_1 = \frac{S_b P_S(S_2)}{1 + \frac{\lambda_S + d_S}{g_S(x,y)}} \quad \text{and} \quad T_1 = \frac{T_b P_T(T_2) \frac{\lambda_T(0)}{\lambda_T(x)}}{1 + \frac{\lambda_T(x) + d_T}{g_T(x,y)}}. \quad (20)$$

Now, consider the following input schemes:

(i) $x \rightarrow \infty, y = 0$. For these input values, the mating program is expected to be initiated, leading to high levels of S_2 . Thus,

$$S_1 \approx \frac{S_b P_S(S_2)}{1 + (\lambda_S + d_S)/g_{Sm}} \approx \frac{F_S S_b}{1 + (\lambda_S + d_S)/g_{Sm}} \sim F_S S_b, \quad (21)$$

where the last step is valid for the parameters values we have considered.

Since $T_2 = S_1 T_1 / K_T$ and $K_T = b_T / a_T$ in the case $w = 0$, two counteracting effects must be considered in the solution of T_1 . First, increasing S_1 is likely to raise the level of T_2 in equilibrium. Second, T_1 undergoes accelerating degradation in presence of Fus3*, which suppresses T_2 formation. For small values of the threshold T_{th} appearing in Eq. 15, the Tec1 promoter activity can saturate $P_T(T_2) \approx F_T$. Thus, we have

$$T_1 \approx \frac{T_b P_T(T_2) / F_\lambda}{1 + (F_\lambda \lambda_T(0) + d_T) / g_{Tm}} \approx \frac{F_T T_b / F_\lambda}{1 + (F_\lambda \lambda_T(0) + d_T) / g_{Tm}} \sim F_T T_b / F_\lambda, \quad (22)$$

resulting in an output ratio

$$\frac{S_2}{T_2} = \frac{K_T}{K_S} \frac{S_1}{T_1} \sim \frac{F_\lambda F_S}{q F_T} \frac{S_b}{T_b}, \quad (23)$$

where q controls the ratio K_T / K_S (see Table 2). Notice the importance of the four key parameters of our model.

When T_{th} is very large such that the Tec1 promotor activity is near the basal level, we find

$$T_1 \approx \frac{T_b P_T(T_2) / F_\lambda}{1 + (F_\lambda \lambda_T(0) + d_T) / g_{Tm}} \approx \frac{T_b / F_\lambda}{1 + (F_\lambda \lambda_T(0) + d_T) / g_{Tm}} \sim T_b / F_\lambda, \quad (24)$$

and an output ratio

$$\frac{S_2}{T_2} = \frac{K_T}{K_S} \frac{S_1}{T_1} \sim \frac{F_\lambda F_S}{q} \frac{S_b}{T_b}. \quad (25)$$

Since F_λ , F_S and F_T are all > 1 we see that the ratio can become larger for large values of T_{th} .

(ii) $y \rightarrow \infty, x = 0$. The filamentous growth is expected under this condition. However, since the S_1 solution is independent of T_1 and $g_S(x, y)$ saturates when either x or y becomes large, we obtain the same result for steady-state S_1 as before:

$$S_1 \approx \frac{S_b P_S(S_2)}{1 + (\lambda_S + d_S) / g_{Sm}} \approx \frac{F_S S_b}{1 + (\lambda_S + d_S) / g_{Sm}} \sim F_S S_b. \quad (26)$$

In the absence of Fus3*, T_1 levels are high and T_2 should be much larger than the threshold value T_{th}

$$T_1 \approx \frac{T_b P_T(T_2)}{1 + (\lambda_T(0) + d_T) / g_{Tm}} \approx \frac{F_T T_b}{1 + (\lambda_T(0) + d_T) / g_{Tm}} \sim F_T T_b. \quad (27)$$

Thus,

$$\frac{S_2}{T_2} = \frac{K_T}{K_S} \frac{S_1}{T_1} \sim \frac{F_S}{qF_T} \frac{S_b}{T_b}. \quad (28)$$

Even though it is possible to find parameters such that this ratio is small, the case $w = 0$ gives almost the same value for S_1 and S_2 in both cases ($x \rightarrow \infty$ and $y \rightarrow \infty$). Hence, the signal fidelity V_X is close to zero and a nonvanishing w is required (see also main text).

Case II: $w > 0$

For nonvanishing decay of dimers $w > 0$, we can graphically examine the nullcline equations for the steady state of S_2 and T_2 (see main text). Fig. S2 gives an example of these nullclines in the S_2 vs T_2 space for large x and large y stimulation. For both cases, we always find only one intersection, demonstrating the monostability of our model.

For the case $w \ll K_T(x)/\sqrt{K_S S_2}$ we can approximate Eq. 10 in the main text as follows:

$$T_2 \approx T_b P_T(T_2) \frac{\sqrt{K_S S_2}}{K_T(x)} \left[\frac{\lambda_T(x)}{\lambda_T(0)} \left(1 + \frac{\lambda_T(x)}{g_T} \right) \left(1 + \frac{d_T}{g_T + \lambda_T(x)} \right) \right]^{-1} \propto \frac{\lambda_T(0) T_b \sqrt{K_S S_2}}{K_T(x) \lambda_T(x)} \sim q/F_\lambda, \quad (29)$$

where we have used that under pheromone stimulation $P_T(T_2) \sim 1$. This demonstrates the trade-off between the relative hetero-dimerization rate (q) and the maximal degradation rate of the Tec1*-Ste12* heterodimers (F_λ), as discussed in the main text.

Model Variants

We have tested several model variants and found that the main results and conclusions in the text are robust to the details of the model. For example, in the original model, we have assumed constant deactivation rates $d_S(x, y) = d_S$ and $d_T(x, y) = d_T$ for simplicity. Experiments, however, indicate dual functions of MAPKs: inactive Fus3 and Kss1 are found to inhibit Ste12 and Tec1, while active MAPKs phosphorylate Ste12 and Tec1 (1). Thus, the deactivation rates are actually functions of the MAPKs. Using the simple functional forms $d_S(x, y) = d_{Sm}/(K_{ds} + x + y)$ and $d_T(x, y) = d_{Tm}/(K_{dt} + x + y)$ did not qualitatively alter the previous results. We also assumed originally that the degradation rates are uniform: $\lambda_{S_0} = \lambda_{S_1} \equiv \lambda_S$ and $\lambda_{T_0}(x) = \lambda_{T_1}(x) \equiv \lambda_T(x)$. This assumption can be relaxed as well. For example, we can take $\lambda_{S_0} = \lambda_{T_0}(x) = 0$ and $\lambda_{S_2} = w_S \lambda_{S_1}$, $\lambda_{T_2}(x) = w_T \lambda_{T_1}(x)$. Again, the results did not change appreciably. Also, we have examined a model variant in which $\Delta \lambda_{T_2}(x) = 0$, i.e., no Tec1 selective degradation. For this variant, the coupling term in Eq. 5 becomes $w \lambda_T(x) T_2 = w \lambda_T(x) S_1 T_1 / K_T(x)$. We found that as long as the constitutive degradation of Tec1*-Ste12* (represented by $\lambda_T(0)$) is large enough, this model variant is able to achieve signaling specificity. Finally, we have examined a more micro-oriented model in which S_0 and T_0 do not contain the protein complex Tec1-Ste12-Dig1. Instead, this complex is treated as an additional variable, adding one more equation to the original ODEs. The quasi-equilibrium approximation applied to this new set of equations leads to reduced equations that exhibit an identical model structures. Furthermore, direct simulations demonstrate that the underlying mechanisms remain the same.

Table 1: Symbol Description

x	Fus3*, input for the downstream pathway of the mating pathway
y	Kss1*, input for the downstream network of the filamentous pathway
S_0	Population of protein complexes that contain inactive Ste12, including Ste12-Dig1-Dig2, Tec1-Ste12-Dig1, etc.
T_0	Population of protein complexes that contain inactive Tec1, including Tec1-Ste12-Dig1, etc.
S_1	Active monomer Ste12*
T_1	Active monomer Tec1*
S_2	Active homodimer Ste12*-Ste12*, the model output
T_2	Active heterodimer Tec1*-Ste12*, the model output
PRE	Pheromone Response Elements, a set of mating genes including <i>STE12</i>
FRE	Filamentation Response Elements, a set of filamentation genes including <i>TEC1</i>
m_S	mRNA of gene <i>STE12</i>
m_T	mRNA of gene <i>TEC1</i>
a_S, a_T	On rate of dimerization
b_S, b_T	Off rate of dimerization
λ_i	Degradation rate of i
$g_i(x, y)$	Activation rate of i, as a function of the input x and y
$d_i(x, y)$	Deactivation rate of i, as a function of the input
μ_S, μ_T	Transcriptional rate of gene <i>STE12</i> and <i>TEC1</i>
ν_p	Translational rate of <i>STE12</i> mRNA and <i>TEC1</i> mRNA
$P_S(S_2)$	PRE promoter activity as a function of the homodimer Ste12*-Ste12*
$P_T(T_2)$	FRE promoter activity as a function of the heterodimer Tec1*-Ste12*

Table 2: Parameter values

S_b	$120nM$	initial concentration of Ste12 (2)
T_b	$30nM$	initial concentration of Tec1 (2)
g_{Sm}	$0.1 \min^{-1}$	maximal activation rate of Ste12 (3)
g_{Tm}	$0.1 \min^{-1}$	maximal activation rate of Ste12
k_S	$10nM$	threshold for Ste12 activation (3)
k_T	$10nM$	threshold for Tec1 activation
d_S	$0.05 \min^{-1}$	deactivation rate of Ste12* (3)
d_T	$0.05 \min^{-1}$	deactivation rate of Tec1*
a_S	$0.2 \text{ nM}^{-1} \min^{-1}$	on rate of Ste12*-Ste12*
b_S	$2.0 \min^{-1}$	off rate of Ste12*-Ste12*
a_T	$0.2 \cdot \mathbf{q} \text{ nM}^{-1} \min^{-1}$	on rate of Tec1*-Ste12*
b_T	$2.0 \min^{-1}$	off rate of Tec1*-Ste12*
q	1	control of the ratio K_T/K_S
F_S	2	fold change of <i>STE12</i> gene expression (4)
F_T	8	fold change of <i>TEC1</i> gene expression (4)
F_λ	4	maximum fold of Tec1 degradation rates by Fus3* (5–7)
λ_S	$0.02 \min^{-1}$	constitutive degradation rate of Ste12 (8)
$\lambda_T(0)$	$0.046 \min^{-1}$	constitutive degradation rate of Tec1 (7)
S_{th}	10 nM	threshold for $P_S(S_2)$
T_{th}	10 nM	threshold for $P_T(T_2)$
L_{th}	10 nM	threshold for $\lambda_T(x)$
n_1	1	Hill coefficient of $P_S(S_2)$
n_2	1	Hill coefficient of $P_T(T_2)$
n_3	1	Hill coefficient of $\lambda_T(x)$

References

1. Cook, J. G., L. Bardwell, and J. Thorner., 1997. Inhibitory and activating functions for MAPK Kss1 in the *S. cerevisiae* filamentous-growth signalling pathway. *Nature*. 390:85–88.
2. Ghaemmaghami, S., W. K. Huh, K. Bower, R. W. Howson, A. Belle, N. Dephoure, E. K. O’Shea, and J. S. Weissman., 2003. Global analysis of protein expression in yeast. *Nature*. 425:737–741.
3. Shao, D., W. Zheng, W. Qiu, Q. Ouyang, and C. Tang., 2006. Dynamic Studies of Scaffold-Dependent Mating Pathway in Yeast. *Biophys. J.* 91:3986–4001.
4. Ren, B., F. Robert, J. J. Wyrick, O. Aparicio, E. G. Jennings, I. Simon, J. Zeitlinger, J. Schreiber, N. Hannett, E. Kanin, T. L. Volkert, C. J. Wilson, S. P. Bell, and R. A. Young., 2000. Genome-wide location and function of DNA binding proteins. *Science*. 290:2306–2309.
5. Bao, M. Z., M. A. Schwartz, G. T. Cantin, J. R. Y. III, and H. D. Madhani., 2004. Pheromone-dependent destruction of the Tec1 transcription factor is required for MAP kinase signaling specificity in yeast. *Cell*. 119:991–1000.
6. Chou, S., L. Huang, and H. Liu., 2004. Fus3-regulated Tec1 degradation through SCFCdc4 determines MAPK signaling specificity during mating in yeast. *Cell*. 119:981–990.
7. Wang, Y., and H. G. Dohlman, 2006. Pheromone-regulated sumoylation of transcription factors that mediate the invasive to mating developmental switch in yeast. *J. Biol. Chem.* 281:1964–1969.
8. Esch, R. K., Y. Wang, and B. Errede., 2006. Pheromone-induced degradation of Ste12 contributes to signal attenuation and the specificity of developmental fate. *Eukaryot. Cell*. 5:2147–2160.

Figure Legends

Figure 1.

A schematic representation of our model system, with symbols interpreted in Table 1. Red arrows denote positive or forward reactions; green arrows represent the backward processes in reversible reactions; and blue ones stand for degradation. We use thicker lines to label stronger processes, for example, the degradation of Tec1 species is accelerated in presence of Fus3*.

Figure 2.

The fixed points in the S_2 vs T_2 space obtained from the reduced 2 variable ODEs.

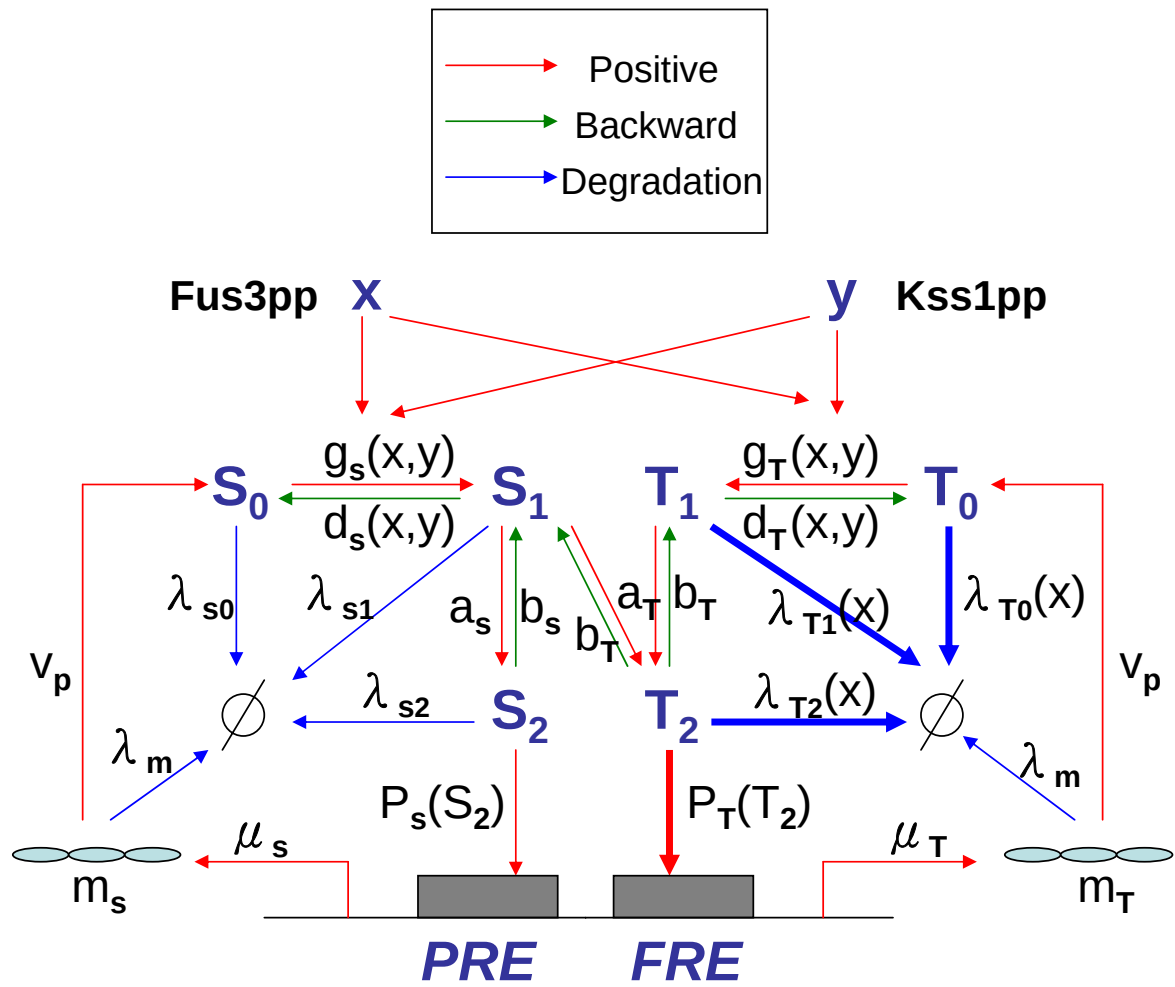


Figure 1:

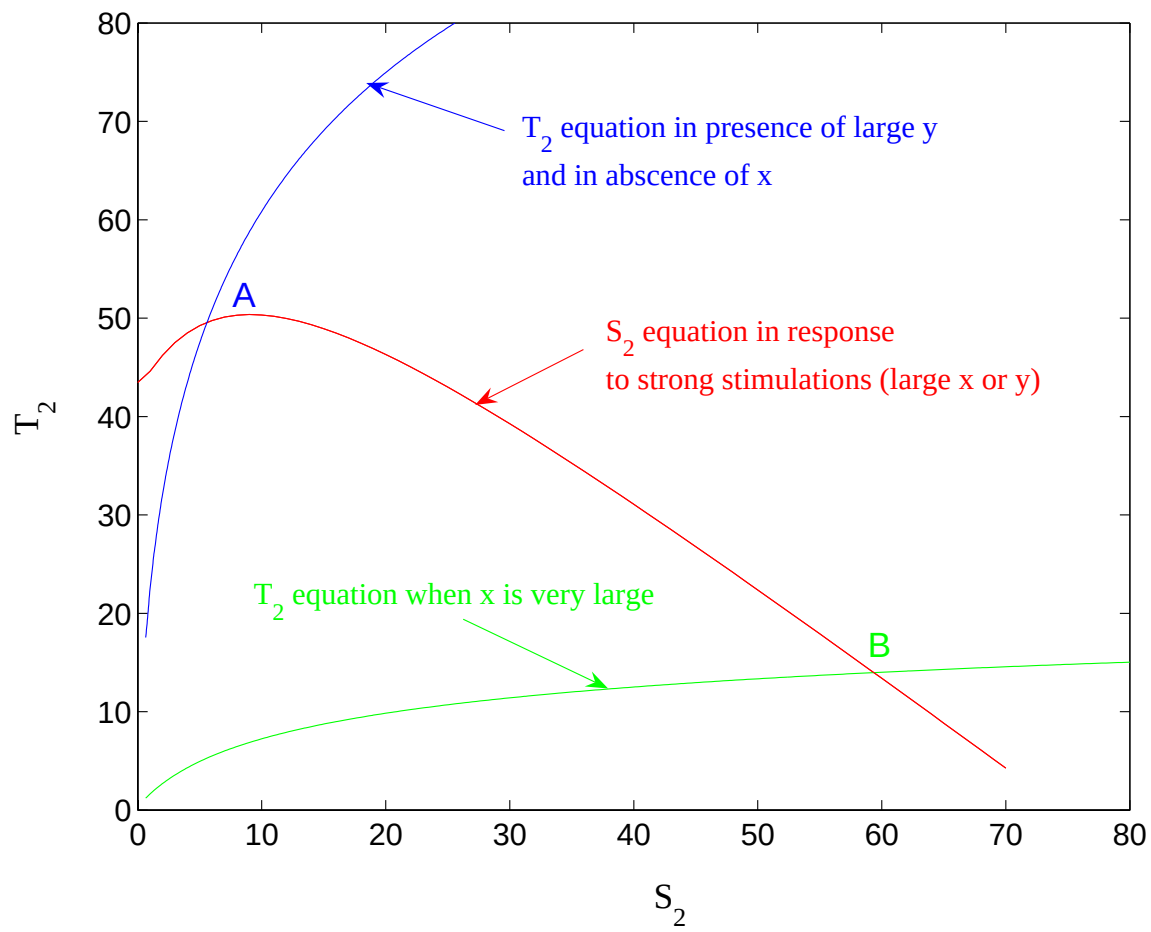


Figure 2: