APPENDIX S1: Model of Pheromone-Induced Yeast Cell Polarization

We updated a previous model [19] of yeast cell polarization using insights gained from this study. This model was based on the spatial dynamics of the heterotrimeric and Cdc42p G-protein cycles. Receptor (R) binds ligand (L) and becomes activated (RL). Activated receptor converts heterotrimeric G-protein (G) into activated $\alpha$-subunit (Ga) and free G$\beta\gamma$ (Gbg). All of these species are on the membrane. The connection between the two cycles is the fact that free G$\beta\gamma$ recruits cytoplasmic Cdc24p to the membrane. Membrane-bound Cdc24p (C24m) activates Cdc42p. Activated Cdc42p (C42a) recruits the scaffold protein Bem1p (B1) to the membrane. Finally, a positive feedback loop is created because membrane-bound Bem1p can bind and recruit Cdc24p to the membrane.

The connection between the yeast model and the generic model (Model 1) is best seen in Equation 4 of the yeast model, describing the dynamics of membrane-bound, active Cdc24p. There, recruitment of Cdc24p to the membrane depends on a cooperative term that is a function of G$\beta\gamma$, $(k_{24cm0}(Gbg^*_n)(C24c))$, and a positive feedback term, $(k_{24cm1}(B1^*)(C24c))$, that depends on Bem1p which in turn is a function of active Cdc42p and hence active Cdc24p.

We made two important modifications to the previous model. First, we added a negative feedback loop for better regulation. The loop includes the protein kinase Cla4p which is activated by Cdc42p and which phosphorylates and inhibits Cdc24p resulting in negative feedback [29]. Second, there is a feedforward/feedback coincidence detection term in the positive feedback loop for better tracking. We changed the $B1^*$ term from

$$\left(\frac{B1^*}{1+(\gamma[B1m]^{-h})}\right)$$

to

$$\left(\frac{B1^*_{\gamma}}{1+(\gamma[Gbg^*_n][B1m]^{-h})}\right)$$

where now G$\beta\gamma$ (the output of the heterotrimeric G-protein cycle and the input to the Cdc42 cycle) influences the positive feedback via
two terms: the cooperative input Hill-term and the positive feedback term. Biologically, the model hypothesizes that Gβγ directly modulates the positive feedback loop presumably through direct and indirect protein-protein interactions with Bem1p, Cdc24p, and Cdc42p. For example, Gβγ is known to bind to Ste20p which in turn binds Bem1p and Cdc42p [34].

\[
\begin{align*}
\frac{\partial [R]}{\partial t} &= D_s \nabla^2_s[R] - k_{RL}[L][R] + k_{RLm}[RL] - k_{Rd0}[R] + k_{Rs} \\
\frac{\partial [RL]}{\partial t} &= D_s \nabla^2_s[RL] + k_{RL}[L][R] - k_{RLm}[RL] - k_{Rd1}[RL] \\
\frac{\partial [G]}{\partial t} &= D_s \nabla^2_s[G] - k_{Ga}[RL][G] + k_{Ga}[Gd][Gb] \\
\frac{\partial [Ga]}{\partial t} &= D_s \nabla^2_s[Ga] + k_{Ga}[RL][G] - k_{Gd}[Ga] \\
\frac{\partial [C24m]}{\partial t} &= D_s \nabla^2_s[C24m] + k_{24cm0}(Gb^*_n)[C24c] + k_{24cm1}(B1^*)[C24c] - k_{24mc}[C24m] \\
&\quad - k_{24d}[Cla4a][C24m] \\
\frac{\partial [C42]}{\partial t} &= D_s \nabla^2_s[C42] - k_{42a}[C24m][C42] + k_{42d}[C42a] \\
\frac{\partial [C42a]}{\partial t} &= D_s \nabla^2_s[C42a] + k_{42a}[C24m][C42] - k_{42d}[C42a] \\
\frac{\partial [B1m]}{\partial t} &= D_s \nabla^2_s[B1m] + k_{B1cm}[C42a][B1c] - k_{B1mc}[B1m] \\
\frac{\partial [Cla4a]}{\partial t} &= k_{Cla4a}[C42a^*_n] - k_{Cla4d}[Cla4a]
\end{align*}
\]

\[Gb^*_n = \frac{R}{1 + (\delta(Gb^*_n))^{-q}},\]  

where \(\delta = SA/\int_S(Gb^*_n)ds\) and \(q = 100, R = 1.\)

\[B1^* = \frac{B1^*_t}{1 + (\gamma Gb^*_n[B1m])^{-h}},\]  

where \(B1^*_t = \int_S[B1m]ds/SA; \gamma = SA/(2 \int_S[B1m]ds).\) \(SA = \int_S ds\) is the surface area of the cell, and \(h = 8.\)
\[
C42a^*_t = \frac{\int_s [C42a] ds}{SA}
\]

The initial conditions and conservation equations are as follows. We may assume that \([C42], [R],\) and \([G]\) are equally distributed along the surface with a total amount of \(C42_t, R_t,\) and \(G_t,\) respectively.

\[
[R]_0 = R_t / SA, \quad R_t = 10,000 \text{ molecules/cell},
\]
\[
[G]_0 = G_t / SA, \quad G_t = 10,000 \text{ molecules/cell},
\]
\[
[C42]_0 = C42_t / SA, \quad C42_t = 10,000 \text{ molecules/cell},
\]
\[
[RL]_0 = 0, \quad [Ga]_0 = 0, \quad [C24m]_0 = 0, \quad [C42a]_0 = 0, \quad [B1m]_0 = 0.
\]

\[
\begin{align*}
[Gd] &= [G]_0 - [G] - [Ga], \\
[Gbg] &= [G]_0 - [G], \\
Gbg_m &= [Gbg]/G_0, \\
V \cdot [C24c] &= C24_t - \int_s [C24m] ds, \quad C24_t = 2000 \text{ molecules/cell}, \\
V \cdot [B1c] &= B1_t - \int_s [B1m] ds, \quad B1_t = 3000 \text{ molecules/cell}.
\end{align*}
\]

The surface area and volume of the cell (ellipsoid with major axis 2 \(\mu m\) and minor axis 1 \(\mu m\)) were \(SA = 21.5 \mu m^2\) and \(V = 8.4 \mu m^3\).

The rate constants are listed below:

\[
\begin{align*}
k_{RL} &= 2 \times 10^6 \text{ M}^{-1} \text{ s}^{-1}; \quad k_{R_{Lm}} = 1 \times 10^{-2} \text{ s}^{-1}; \quad k_{R_s} = 4 \text{ (molecules) s}^{-1} / SA; \quad k_{Rd0} = 4 \times 10^{-4} \text{ s}^{-1}; \quad k_{G1} = 1 \text{ (molecules) s}^{-1} \times SA; \\
k_{Ga} &= 1 \times 10^{-5} \text{ (molecules)}^{-1} \text{ s}^{-1} \times SA; \quad k_{Gd} = 0.1 \text{ s}^{-1}; \quad k_{24cm0} = 0.04 \text{ s}^{-1} \times V / SA; \quad k_{24cm1} = 3.3 \times 10^{-3} \text{ or } 3.3 \times 10^{-2} \text{ (molecules)}^{-1} \text{ s}^{-1} \times V; \quad k_{24mc} = 1 \text{ s}^{-1}; \quad k_{24d} = SA / 3000 \text{ s}^{-1}; \\
k_{42a} &= 1 \times 10^{-5} \text{ (molecules)}^{-1} \text{ s}^{-1} \times SA; \quad k_{42d} = 0.02 \text{ s}^{-1}; \\
k_{B1cm} &= 1 \times 10^{-5} \text{ (molecules)}^{-1} \text{ s}^{-1} \times V; \quad k_{B1mc} = 0.01 \text{ s}^{-1}; \quad k_{C104a} = 0.006 \text{ s}^{-1}; \quad k_{C104d} = 0.01 \text{ s}^{-1}.
\end{align*}
\]

\(D_s = 0, 0.001, 0.01, \text{ or } 0.1 \mu m^2 / s.\)