Supplementary Information for "A multiscale biophysical model for the recruitment of actin nucleating proteins at the membrane interface"

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S1. Numerical solutions for reactions

The proposed two step reaction mechanism for mDia2, NWASP and gelsolin is given as:

(i) $F + M \rightleftharpoons FM$, $N + M \rightleftharpoons AM$ and $G + M \rightleftharpoons GM$
$K_{fm} = \frac{[FM]}{[F][M]}$, $K_{nm} = \frac{[NM]}{[N][M]}$ and $K_{gm} = \frac{[GM]}{[G][M]}$.

(ii) $FM + n_1P \rightleftharpoons FMP_{n_1}$, $NM + n_2P \rightleftharpoons NMP_{n_2}$ and $GM + n_3P \rightleftharpoons GMP_{n_3}$
$K_{fp} = \frac{[FMP_{n_1}]}{[FM][P]^{n_1}}$, $K_{np} = \frac{[NMP_{n_2}]}{[NM][P]^{n2}}$ and $K_{gp} = \frac{[GMP_{n_3}]}{[GM][P]^{n3}}$.

The rates of reactions for mDia2 can be written as:

$$\begin{aligned}\frac{d[FM]}{dt} &= K_{on}^{fm}[F][M] - K_{off}^{fm}[FM] - \frac{d[FMP_{n_1}]}{dt} \\ &= K_{on}^{fm}\left([F]_{tot} - [FM] - [FMP_{n_1}]\right) \\ &\quad \left([M]_{tot} - [FM] - [FMP_{n_1}] - [NM] - [NMP_{n_2}] - [GM] - [GMP_{n_3}]\right) \\ &\quad -K_{off}^{fm}[FM] - \frac{d[FMP_{n_1}]}{dt} \end{aligned} \tag{S1}$$

and

$$\begin{aligned}\frac{d[FMP_{n_1}]}{dt} &= K_{on}^{fp}[FM][P]^{n_1} - K_{off}^{fp}[FMP_{n_1}] \\ &= K_{on}^{fp}[FM]\left([P]_{tot} - n_1[FMP_{n_1}] - n_2[NMP_{n_2}] - n_3[GMP_{n_3}]\right)^{n_1} - K_{off}^{fp}[FMP_{n_1}], \end{aligned} \tag{S2}$$

where K_{on} and K_{off} represent the forward and reverse reaction rate constants. At equilibrium $K_{\alpha\beta} = K_{on}^{\alpha\beta}/K_{off}^{\alpha\beta}$ with $\alpha = f, n, g$ and $\beta = m, p$.

At equilibrium,

$$\frac{d[FM]}{dt} = \frac{d[FMP_n]}{dt} = 0 \tag{S3}$$

Substituting this in equations S1 and S2:

$$\begin{aligned} &K_{on}^{fm}\left([F]_{tot} - [FM] - [FMP_{n_1}]\right)\left([M]_{tot} - [FM] - [FMP_{n_1}] - [NM] - [NMP_{n_2}] - [GM] - [GMP_{n_3}]\right) \\ &\quad -K_{off}^{fm}[FM] = 0 \end{aligned} \tag{S4}$$

and

$$K_{on}^{fp}[FM]\left([P]_{tot} - n_1[FMP_{n_1}] - n_2[NMP_{n_2}] - n_3[GMP_{n_3}]\right)^{n_1} - K_{off}^{fp}[FMP_{n_1}] = 0. \tag{S5}$$

Substituting for K_{on}^{fm}/K_{off}^{fm} and K_{on}^{fp}/K_{off}^{fp}:

$$K_{fm}\left([F]_{tot}-[FM]-[FMP_{n_1}]\right)\left([M]_{tot}-[FM]-[FMP_{n_1}]-[NM]-[NMP_{n_2}]-[GM]-[GMP_{n_3}]\right)$$
$$-[FM]=0 \tag{S6}$$

and

$$K_{fp}[FM]\left([P]_{tot}-n_1[FMP_{n_1}]-n_2[NMP_{n_2}]-n_3[GMP_{n_3}]\right)^{n_1}-[FMP_{n_1}]=0. \tag{S7}$$

Similarly we can write the equations for NWASP as:

$$K_{nm}\left([N]_{tot}-[NM]-[NMP_{n_2}]\right)\left([M]_{tot}-[FM]-[FMP_{n_1}]-[NM]-[NMP_{n_2}]-[GM]-[GMP_{n_3}]\right)$$
$$-[NM]=0 \tag{S8}$$

and

$$K_{np}[NM]\left([P]_{tot}-n_1[FMP_{n_1}]-n_2[NMP_{n_2}]-n_3[GMP_{n_3}]\right)^{n_2}-[NMP_{n_2}]=0 \tag{S9}$$

and the equations for gelsolin as:

$$K_{gm}\left([G]_{tot}-[GM]-[GMP_{n_3}]\right)\left([M]_{tot}-[FM]-[FMP_{n_1}]-[NM]-[NMP_{n_2}]-[GM]-[GMP_{n_3}]\right)(S$$
$$-[GM]=0$$

and

$$K_{gp}[GM]\left([P]_{tot}-n_1[FMP_{n_1}]-n_2[NMP_{n_2}]-n_3[GMP_{n_3}]\right)^{n_3}-[GMP_{n_2}]=0 \tag{S11}$$

Solving equations S6 to S11 numerically we obtain $[FMP_{n_1}]$, $[NMP_{n_2}]$ and $[GMP_{n_3}]$ as a function of $[P]_{tot}$.

The Hill coefficients $n_1=4$, $n_2=5$ and $n_3=4$ are obtained from molecular simulations. For mDia2 we take association constants $K_{fm}=60/\mu M$ and $K_{fp}=0.0001/\mu M^{n1}$ (i.e., dissociation constant $K_{fp}^{d}=10\mu M$) [1]. For NWASP, we take $K_{np}=0.0037/\mu M^{n2}$ (i.e., $K_{np}^{d}=3.07\mu M$) [2] and $K_{nm}=K_{fm}$. For gelsolin we take $K_{gm}=0.01/\mu M$ and $K_{gp}=0.0001/\mu M^{n3}$ (i.e., $K_{gp}^{d}=5\mu M$). The reported dissociation constants for gelsolin varies from $2-20\mu M$ [3], hence in the section S2 we describe the effect of varying K_{gm}^{d}. The membrane binding site concentration is taken to be $[M_{tot}]=10\mu M$.

S2. Bound and free gelsolin for different gelsolin dissociation constants

Here we vary gelsolin dissociation constants K_{gp}^{d} as $2\mu M$, $5\mu M$ and $10\mu M$ when $K_{gm}=K_{fm}=60/\mu M$ and $K_{gm}=0.01/\mu M$.

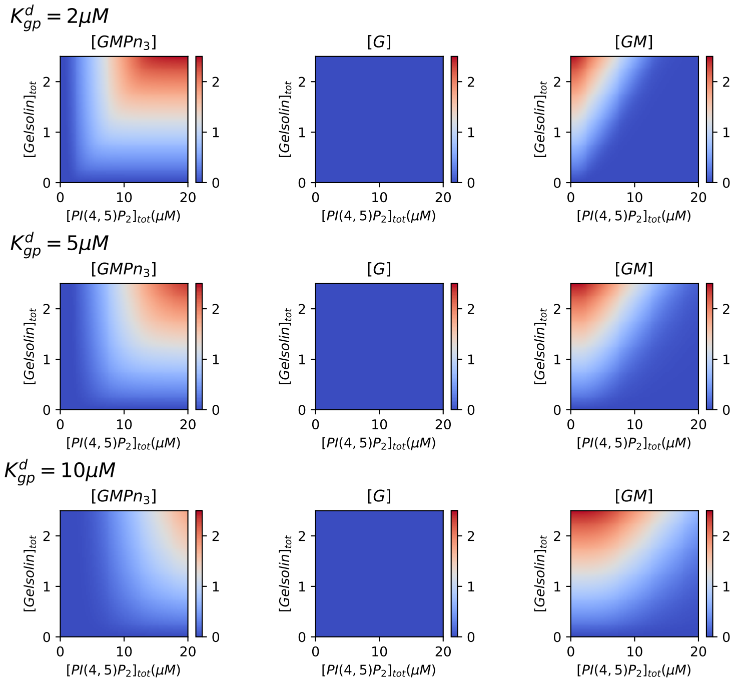

Figure S1. Effect of dissociation constant in gelsolin binding when $K_{gm} = 60/\mu M$.

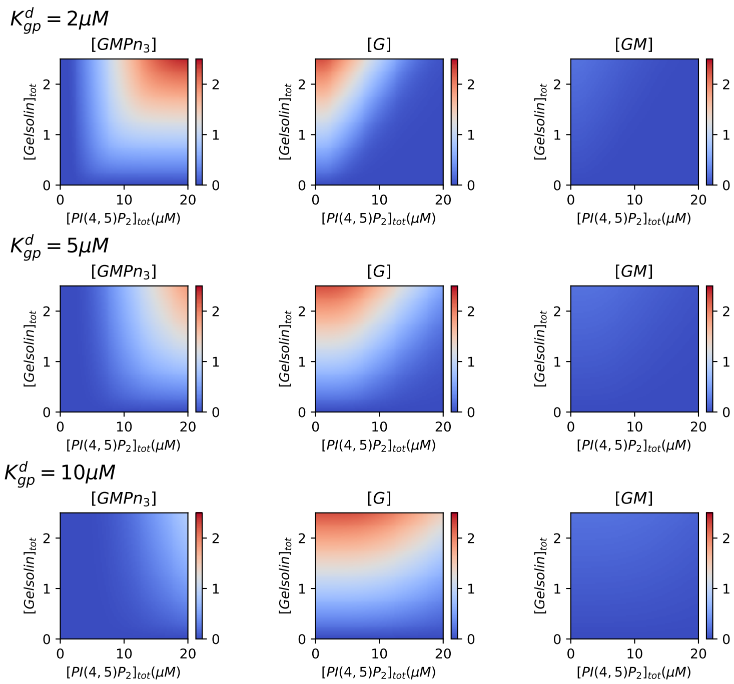

Figure S2. Effect of dissociation constant in gelsolin binding when $K_{gm} = 0.01/\mu M$.

References

[1] Bucki R, Wang Y H, Yang C, Kandy S K, Fatunmbi O, Bradley R, Pogoda K, Svitkina T, Radhakrishnan R and Janmey P A 2019 *Journal of Biological Chemistry* **294** 4704–4722

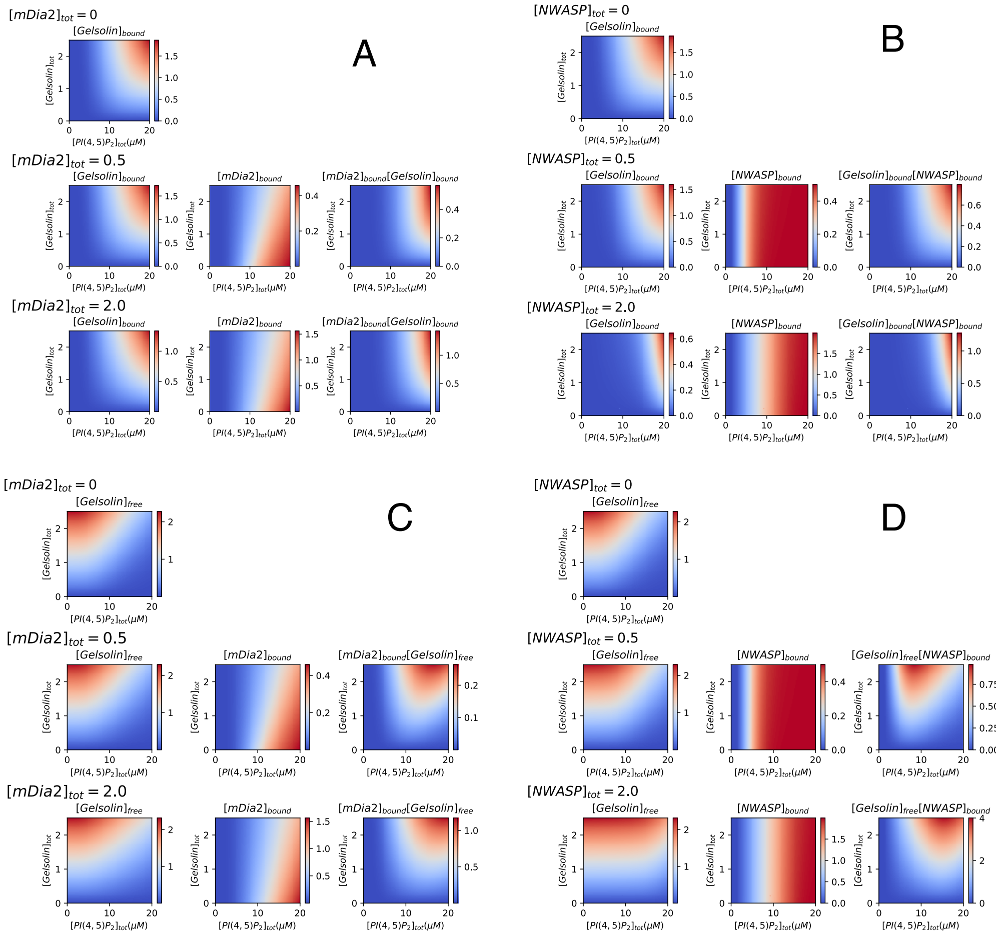

Figure S3. A. Concentration of bound gelsolin and mDia2 as a function of PI(4,5)P_2 and gelsolin at different mDia2 concentrations in the absence of NWASP when $K^d_{gm} = 100\mu M$, $K^d_{gp} = 5\mu M$, $K^d_{fm} = 0.017\mu M$, $K^d_{fp} = 10\mu M$ and $M_{tot} = 10$. B. Concentration of bound gelsolin and NWASP as a function of PI(4,5)P_2 and gelsolin at different NWASP concentrations in the absence of mDia2 when $K^d_{gm} = 100\mu M$, $K^d_{gp} = 5\mu M$, $K^d_{nm} = 0.017\mu M$, $K^d_{np} = 3.07\mu M$ and $M_{tot} = 10$. C. Concentration of free gelsolin and bound mDia2 as a function of PI(4,5)P2 and gelsolin at different mDia2 concentrations in the absence of NWASP when $K^d_{gm} = 100\mu M$, $K^d_{gp} = 5\mu M$, $K^d_{fm} = 0.017\mu M$, $K^d_{fp} = 10\mu M$ and $M_{tot} = 10$. D. Concentration of free gelsolin and bound NWASP as a function of PI(4,5)P2 and gelsolin at different NWASP concentrations in the absence of mDia2 when $K^d_{gm} = 100\mu M$, $K^d_{gp} = 5\mu M$, $K^d_{nm} = 0.017\mu M$, $K^d_{np} = 3.07\mu M$ and $M_{tot} = 10$.

(*Preprint* http://www.jbc.org/content/294/12/4704.full.pdf+html) URL http://www.jbc.org/content/294/12/4704.abstract

[2] Papayannopoulos V, Co C, Prehoda K E, Snapper S, Taunton J and Lim W A 2005 *Molecular Cell* **17** 181 – 191 ISSN 1097-2765 URL http://www.sciencedirect.com/science/article/pii/S1097276504007993

[3] Szatmri D, Xue B, Kannan B, Burtnick L D, Bugyi B, Nyitrai M and Robinson R C 2018 *PLOS ONE* **13** 1–17 URL https://doi.org/10.1371/journal.pone.0201826