Supplementary Information

To accompany Jakobson, Slininger, Tullman-Ercek, and Mangan; A Systems-Level Model Reveals That 1,2-Propanediol Utilization Microcompartments Enhance Pathway Flux Through Intermediate Sequestration.

The following describes the equations used in the numerical and analytical code used to generate the plots and other figures in the text. The codebase will be made available on GitHub following publication under an XXX license.

Governing equations

As described in the Models section, the equations describing the concentrations of P and A in the MCP are as follows:

$$D\nabla^2 P(r) - R_{CDE} = 0 \tag{1}$$

$$D\nabla^2 A(r) + R_{CDE} - R_{PQ} = 0 \tag{2}$$

And the concentrations in the cytosol are described by the following:

$$P(r) = \frac{k_m^P P_{MCP}(r = R_c) - P_{out}(j_c + k_m^P)}{\frac{D}{R_b^2} + k_m^p X}\left(\frac{1}{r} - \frac{D}{k_c^P R_c^2} - \frac{1}{R_c}\right) + P_{MCP}(r = R_c) \tag{3}$$

$$A(r) = \frac{A_{MCP}(r = R_c) - A_{out}}{\frac{D}{k_m^A R_b^2} + X}\left(\frac{1}{r} - \frac{D}{k_c^A R_c^2} - \frac{1}{R_c}\right) + A_{MCP}(r = R_c) \tag{4}$$

Where $X = \left(\frac{D}{R_c^2 k_c} + \frac{1}{R_c} - \frac{1}{R_b}\right)$

The following boundary conditions hold at the cell and MCP membranes, respectively:

$$D\frac{\partial P}{\partial r}|_{r=R_b} = j_c P_{out} + k_m^P \left(P_{out} - P_{cytosol}(r = R_b)\right) \tag{5}$$

$$D\frac{\partial A}{\partial r}|_{r=R_b} = k_m^A \left(A_{out} - A_{cytosol}(r = R_b)\right) \tag{6}$$

$$\frac{\partial P}{\partial r}|_{r=R_c} = -\left(\frac{1}{R_c^2}\right)\frac{k_m^P P_{MCP}(r = R_c) - P_{out}(j_c + k_m^P)}{\frac{D}{R_b^2} + k_m^p X} \tag{7}$$

$$\frac{\partial A}{\partial r}|_{r=R_c} = -\left(\frac{1}{R_c^2}\right)\frac{A_{MCP}(r = R_c) - A_{out}}{\frac{D}{k_m^A R_b^2} + X} \tag{8}$$

Non-dimensional equations

We then recast the system in terms of the following non-dimensional variables:

$$\rho = \frac{r}{R_c} \tag{9}$$

$$a = \frac{A}{K_{PQ}} \tag{10}$$

$$p = \frac{P}{K_{CDE}} \tag{11}$$

Applying the non-dimensionalization and letting $\kappa = \frac{K_{CDE}}{K_{PQ}}$, we obtain the following governing equation for A in the MCP:

$$\frac{K_{PQ}D}{R_c^2}\frac{1}{\rho^2}\frac{\partial}{\partial\rho}\left(\rho^2\frac{\partial a}{\partial\rho}\right) = \frac{2V_{PQ}a}{1+a} - \frac{V_{CDE}p}{1+p} \tag{12}$$

Now let $\gamma = \frac{2V_{PQ}}{V_{CDE}}$ and $\xi = \frac{K_{PQ}D}{V_{CDE}R_c^2}$, yielding:

$$\xi\frac{1}{\rho^2}\frac{\partial}{\partial\rho}\left(\rho^2\frac{\partial a}{\partial\rho}\right) = \xi\nabla^2 a = \gamma\frac{a}{1+a} - \frac{p}{1+p} \tag{13}$$

Similarly for P in the MCP,

$$\kappa\xi\frac{1}{\rho^2}\frac{\partial}{\partial\rho}\left(\rho^2\frac{\partial p}{\partial\rho}\right) = \kappa\xi\nabla^2 p = \frac{p}{1+p} \tag{14}$$

We can then nondimensionalize the boundary conditions as follows:

$$\frac{\partial a}{\partial\rho}|_{\rho=1} = \epsilon_a + \beta_a a \tag{15}$$

$$\epsilon_a = \frac{A_{out}}{R_c K_{PQ}\left(\frac{D}{k_m^A R_b^2} + X\right)} \tag{16}$$

$$\beta_a = \frac{-1}{R_c\left(\frac{D}{k_m^A R_b^2} + X\right)} \tag{17}$$

Similarly for P,

$$\frac{\partial p}{\partial\rho}|_{\rho=1} = \epsilon_p + \beta_p p \tag{18}$$

$$\epsilon_p = \frac{P_{out}\left(j_c + k_m^P\right)}{K_{CDE}R_c\left(\frac{D}{R_b^2} + k_m^P X\right)} \tag{19}$$

$$\beta_p = \frac{-k_m^P}{R_c\left(\frac{D}{R_b^2} + k_m^P X\right)} \tag{20}$$

These nondimensional equations can then be solved numerically by a finite-difference approach to find the steady-state concentrations in the MCP, and the solutions in the cytosol follow directly. We solve the spherical finite-difference equations using the ODE15s solver in MATLAB.

Analytical solution

For ease of computation, we cast the analytical solution (assuming constant concentrations in the MCP) differently than in the Models section.

First, consider the mass balance on A_{MCP} :

$$0 = 4\pi R_c^2 k_c^A (A_{cyt} - A_{MCP}) + \frac{4}{3}\pi R_c^3 (\frac{V_{CDE} P_{MCP}}{K_{CDE} + P_{MCP}} - \frac{2V_{PQ} A_{MCP}}{K_{PQ} + A_{MCP}}) \tag{21}$$

And similarly for P_{MCP}:

$$0 = 4\pi R_c^2 k_c^P (P_{cyt} - P_{MCP}) - \frac{4}{3}\pi R_c^3 \frac{V_{CDE} P_{MCP}}{K_{CDE} + P_{MCP}} \tag{22}$$

First we solve for P_{MCP}, as this does not depend on A_{MCP} due to the irreversibility of PduCDE:

$$k_c^P (P_{cyt} - P_{MCP}) = \frac{1}{3} R_c \frac{V_{CDE} P_{MCP}}{K_{CDE} + P_{MCP}} \tag{23}$$

$$\frac{3DK_{CDE}}{V_{CDE} R_c^3 (\frac{D}{k_m^P R_b^2} + X)} \Big[\frac{P_{out}(1 + \frac{j_c}{k_m^P})}{K_{CDE}} - p \Big] = \frac{p}{1+p} \tag{24}$$

Let $Y = \frac{V_{CDE} R_c^3 (\frac{D}{k_m^P R_b^2} + X)}{3DK_{CDE}}$ and let $Z = \frac{P_{out}(1 + \frac{j_c}{k_m^P})}{K_{CDE}}$.

$$\frac{1}{Y}(Z - p) = \frac{p}{1+p} \tag{25}$$

Let $E = Y - Z + 1$.

$$p = \frac{-E \pm \sqrt{E^2 + 4Z}}{2} \tag{26}$$

Now we can find A_{MCP} similarly, given the solution for P_{MCP}.

$$\frac{3D}{R_c^3 (\frac{D}{k_m^A R_b^2} + X)} (A_{out} - A_{MCP}) = \frac{2V_{PQ} A_{MCP}}{K_{PQ} + A_{MCP}} - \frac{V_{CDE} P_{MCP}}{K_{CDE} + P_{MCP}} \tag{27}$$

$$\frac{3DK_{PQ}}{2V_{PQ} R_c^3 (\frac{D}{k_m^A R_b^2} + X)} (\frac{A_{out}}{K_P Q} - a) + \frac{1}{2}\frac{V_{CDE}}{V_{PQ}} \frac{p}{1+p} = \frac{a}{1+a} \tag{28}$$

Let $U = \frac{2V_{PQ} R_c^3 (\frac{D}{k_m^P R_b^2} + X)}{3DK_{PQ}}$, let $V = \frac{A_{out}}{K_{PQ}}$, and let $W = \frac{1}{2}\frac{V_{CDE}}{V_{PQ}}$.

$$\frac{1}{U}(V - a) + W \frac{p}{1+p} = \frac{a}{1+a} \tag{29}$$

Let $F = 1 + U - V$.

$$a = \frac{-(F - UW\frac{p}{1+p}) \pm \sqrt{(F - UW\frac{p}{1+p})^2 + 4(UW\frac{p}{1+p} + V)}}{2} \tag{30}$$

Again, the solutions in the cytosol follow directly from these MCP solutions.

Equations for no MCP case

In the case when there is no Pdu MCP, we assume that the same number of enzymes are now distributed throughout the cell. The equations in the cell are therefore now as follows:

$$0 = D\nabla^2 A + R_{CDE} - R_{PQ} \tag{31}$$

$$0 = D\nabla^2 P - R_{CDE} \tag{32}$$

These can be non-dimensionalized as follows (*c.f.* with above for MCP case):

$$\xi \frac{1}{\rho^2}\frac{\partial}{\partial \rho}\left(\rho^2 \frac{\partial a}{\partial \rho}\right) = \xi \nabla^2 a = \gamma \frac{a}{1+a} - \frac{p}{1+p} \tag{33}$$

Similarly for P,

$$\kappa\xi \frac{1}{\rho^2}\frac{\partial}{\partial \rho}\left(\rho^2 \frac{\partial p}{\partial \rho}\right) = \kappa\xi \nabla^2 p = \frac{p}{1+p} \tag{34}$$

Now considering the boundary conditions,

$$\frac{\partial A}{\partial r}|_{r=R_b} = k_m^A (A_{out} - A) \tag{35}$$

$$\frac{\partial a}{\partial \rho}|_{\rho=1} = \epsilon_a + \beta_a a \tag{36}$$

$$\epsilon_a = \frac{R_b A_{out} k_m^A}{D K_{PQ}} \tag{37}$$

$$\beta_a = \frac{-R_b k_m^A}{D} \tag{38}$$

Similarly for P,

$$\frac{\partial p}{\partial \rho}|_{\rho=1} = \epsilon_p + \beta_p p \tag{39}$$

$$\epsilon_p = \frac{R_b P_{out}}{D K_{CDE}}(j_c + k_m^P) \tag{40}$$

$$\beta_p = \frac{-R_b k_m^P}{D} \tag{41}$$

These equations can once again be solved numerically by the same finite difference approach described above.

Supplementary Figures

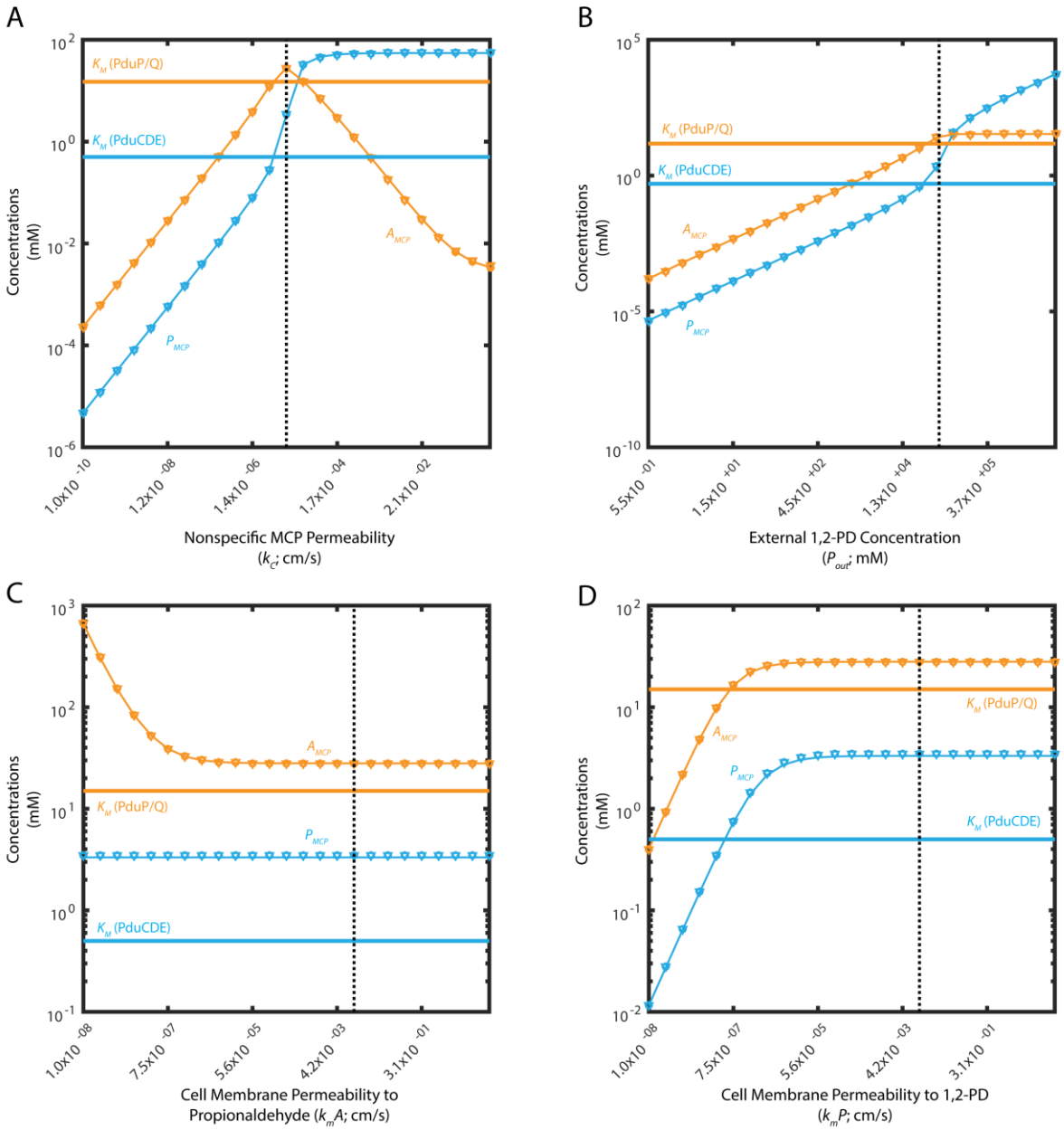

Figure S1. Comparison of analytical solution assuming constant concentrations in the MCP (solid lines) and numerical solutions from the edge (circles) and center (triangles) of the MCP for 1,2-PD (blue) and propionaldehyde (orange). The baseline parameter values are shown with a black dashed line. The K_M of the PduCDE and PduP/Q enzymes are plotted in blue and orange lines, respectively.

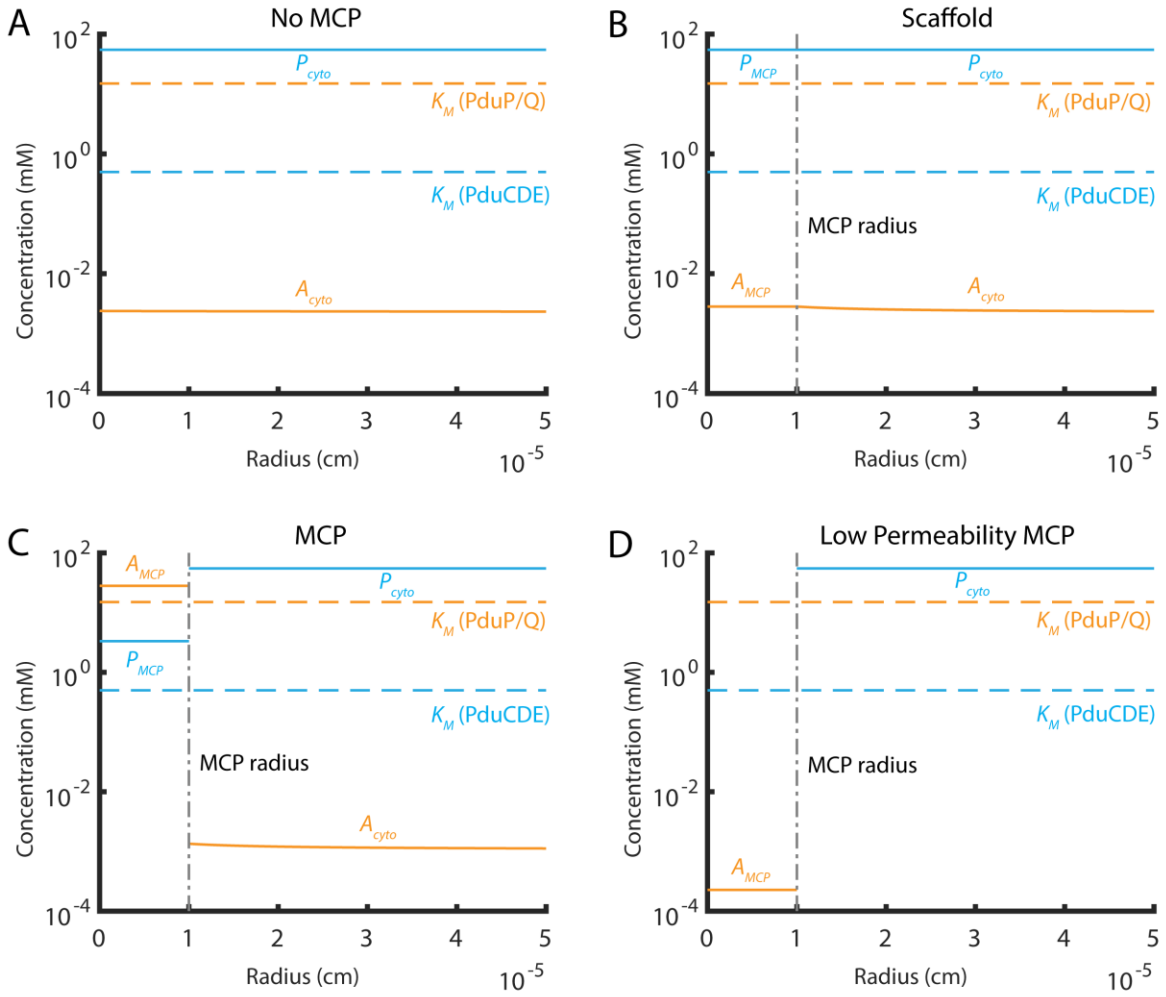

Figure S2. Concentration profiles as a function of *r* for a cell with (A) no MCPs; (B) a scaffold with no diffusion limitation ($k_c = 10^{10}$); (C) MCPs ($k_c = 10^{-5}$); and (D) sparingly permeable MCPs ($k_c = 10^{-10}$). 1,2-PD in the MCP (P_{MCP}) and in the cytosol (P_{cyto}) are plotted in blue and propionaldehyde in the MCP (A_{MCP}) and in the cytosol (A_{cyto}) in orange. The K_M of the PduCDE and PduP/Q enzymes are plotted in blue and orange dashed lines, respectively.

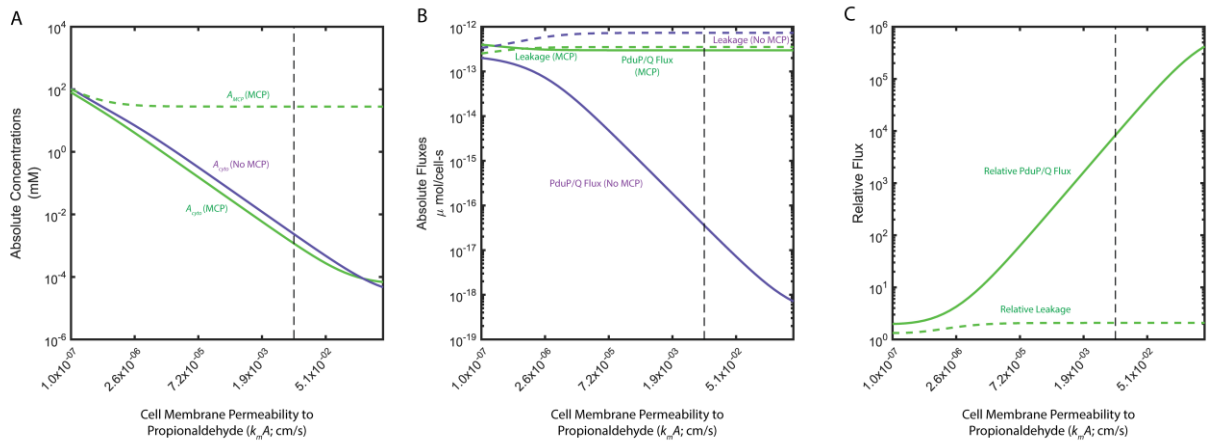

Figure S3. (A) Cytosolic aldehyde concentration (A_{cyto}) with and without MCPs and MCP aldehyde concentration (A_{MCP}) with MCPs; (B) relative carbon flux through PduP/Q ($flux_{MCP}/flux_{NoMCP}$) and relative aldehyde leakage rate ($leakage_{NoMCP}/leakage_{MCP}$); and (C) relative flux through the PduP/Q enzymes (with MCPs/without MCPs) and relative propionaldehyde leakage across the cell membrane (without MCPs/with MCPs) as a function of k_mA. The baseline k_mA value is shown with a black dashed line.

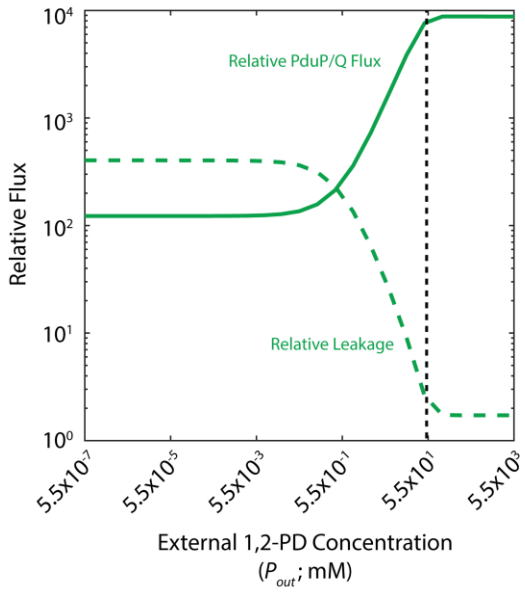

Figure S4. Relative flux through the PduP/Q enzymes (with MCPs/without MCPs) and relative propionaldehyde leakage across the cell membrane (without MCPs/with MCPs) as a function of external 1,2-PD concentration. The baseline external 1,2-PD concentration is shown with a black dashed line.

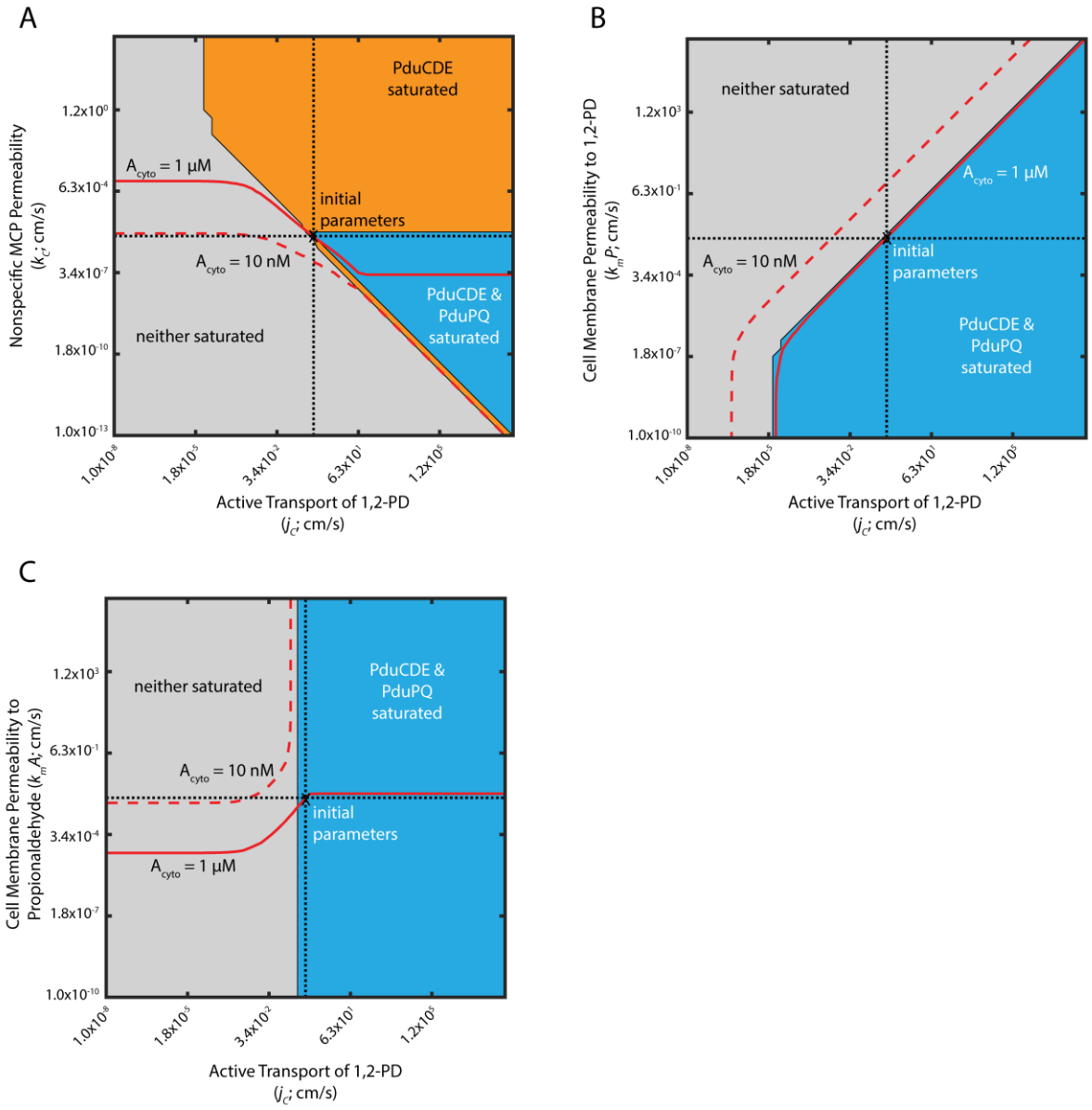

Figure S5. Saturation phase spaces of PduCDE and PduP/Q with respect to (A) j_c and k_c, (B) with respect to j_c and k_mP, and (C) with respect to j_c and k_mA when P_{out} is 0.5 mM. Regions of saturation (concentration of substrate > K_M of the appropriate enzyme) are plotted in blue when both enzymes are saturate, orange when only PdyCDE is saturated, and in grey when neither enzyme is saturated. Red solid lines indicate when A_{cyto} is 1 µM; red dashed lines indicate when A_{cyto} is 10 nM. Black dashed lines indicate the baseline parameter values used in the model of the Pdu MCP.

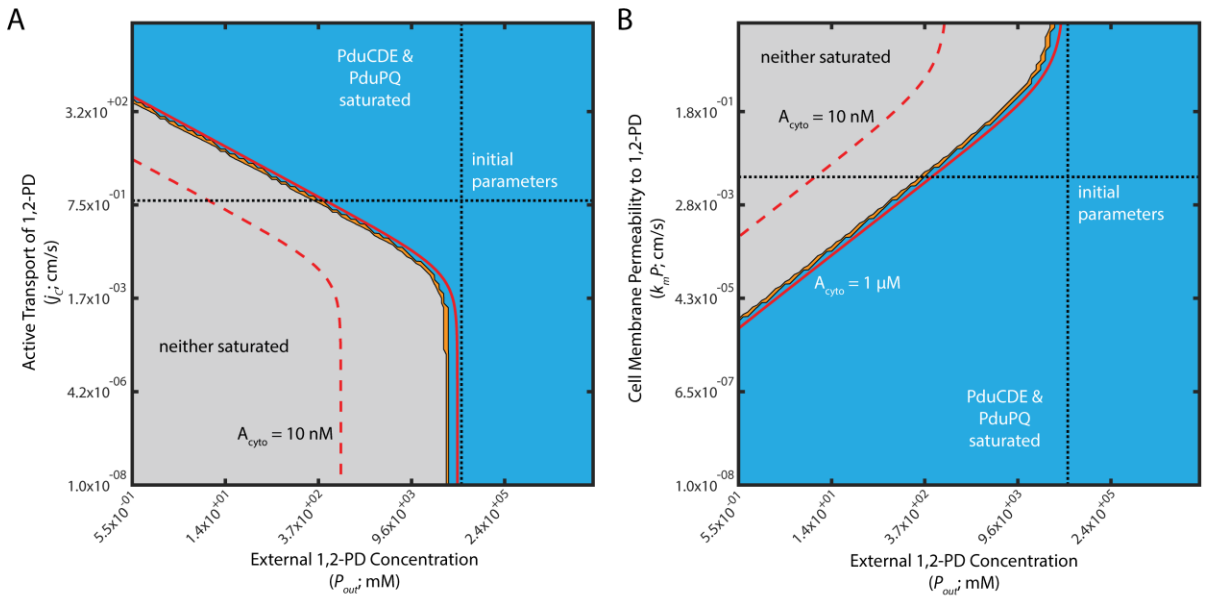

Figure S6. Saturation phase spaces of PduCDE and PduP/Q with respect to (A) P_{out} and j_c, and (B) with respect to P_{out} and k_mP. Regions of saturation (concentration of substrate > K_M of the appropriate enzyme) are plotted in blue when both enzymes are saturate, orange when only PdyCDE is saturated, and in grey when neither enzyme is saturated. Red solid lines indicate when A_{cyto} is 1 μM; red dashed lines indicate when A_{cyto} is 10 nM. Black dashed lines indicate the baseline parameter values used in the model of the Pdu MCP.

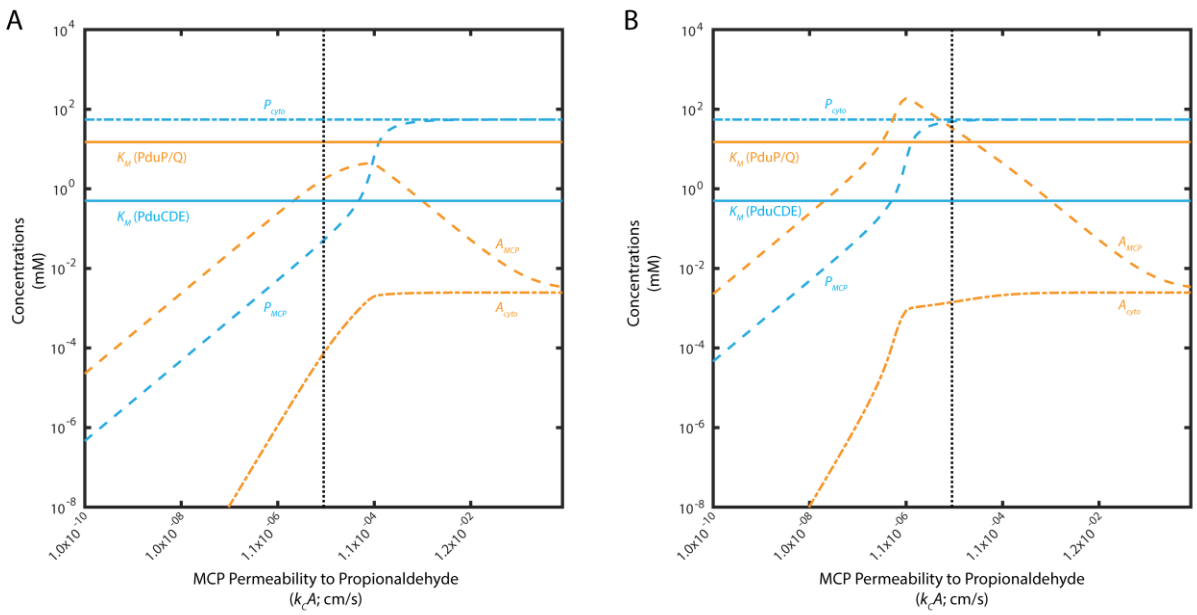

Figure S7. Mean concentrations of 1,2-PD and propionaldehyde in the MCP (P_{MCP}; A_{MCP}) and cytosol (P_{cyto}; A_{cyto}) as a function of (A) k_cA when k_cP = 0.1 x k_cA and (B) k_cA when k_cP = 10 x k_cA. K_M of PduCDE and PduP/Q are shown as solid lines. The baseline permeabilities are shown with a black dashed line.

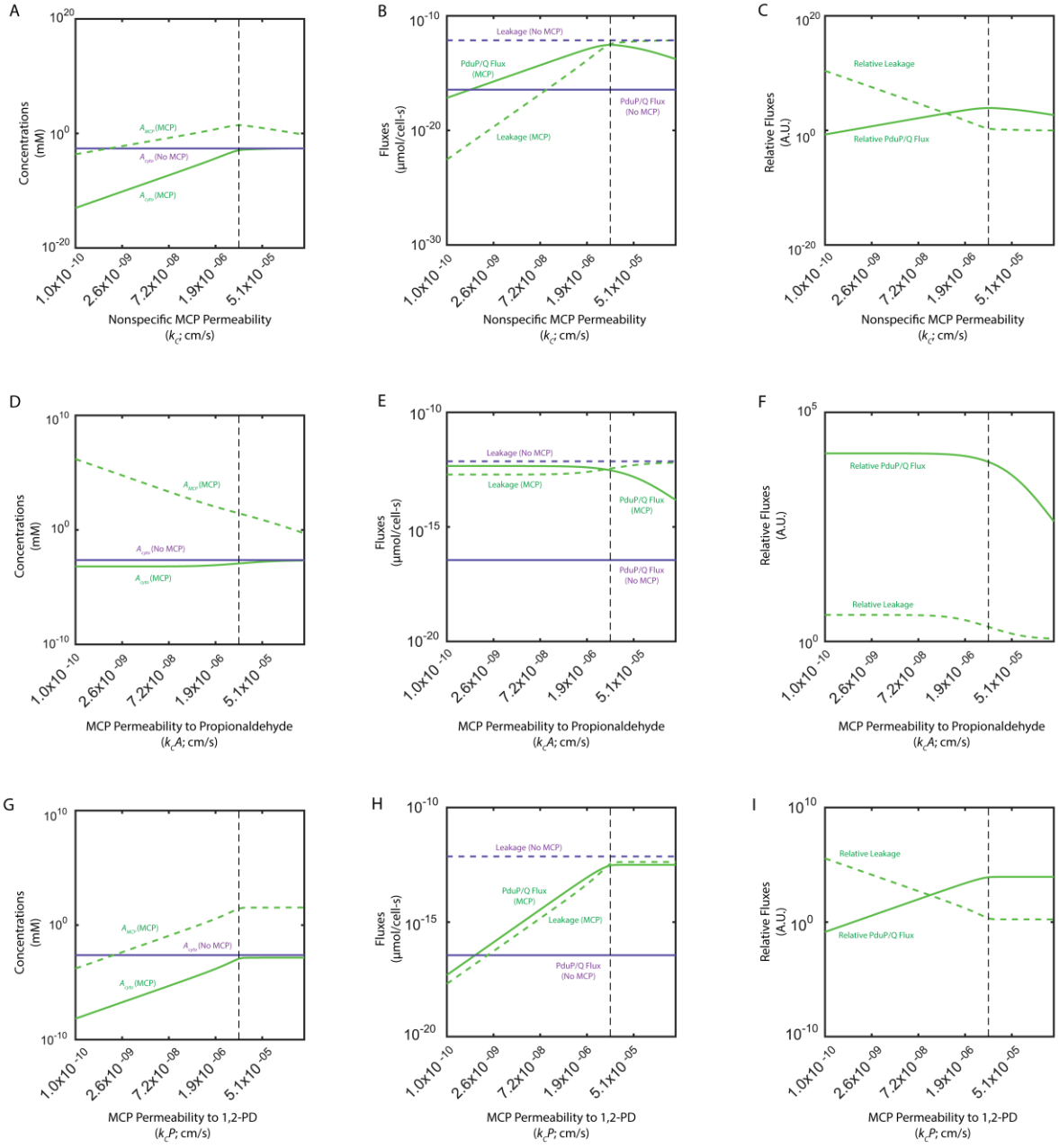

Figure S8. (A,D,G) Cytosolic aldehyde concentration (A_{cyto}) with and without MCPs and MCP aldehyde concentration (A_{MCP}) with MCPs; (B,E,H) absolute flux through PduP/Q and aldehyde leakage from the cell with and without MCPs; and (C,F,I) relative carbon flux through PduP/Q ($flux_{MCP}/flux_{NoMCP}$) and relative aldehyde leakage rate ($leakage_{NoMCP}/leakage_{MCP}$); as a function of (A,B,C) $k_c = k_cA = k_cP$; (D,E,F) k_cA; and (G,H,I) k_cP. The baseline permeabilities are shown with a black dashed line.

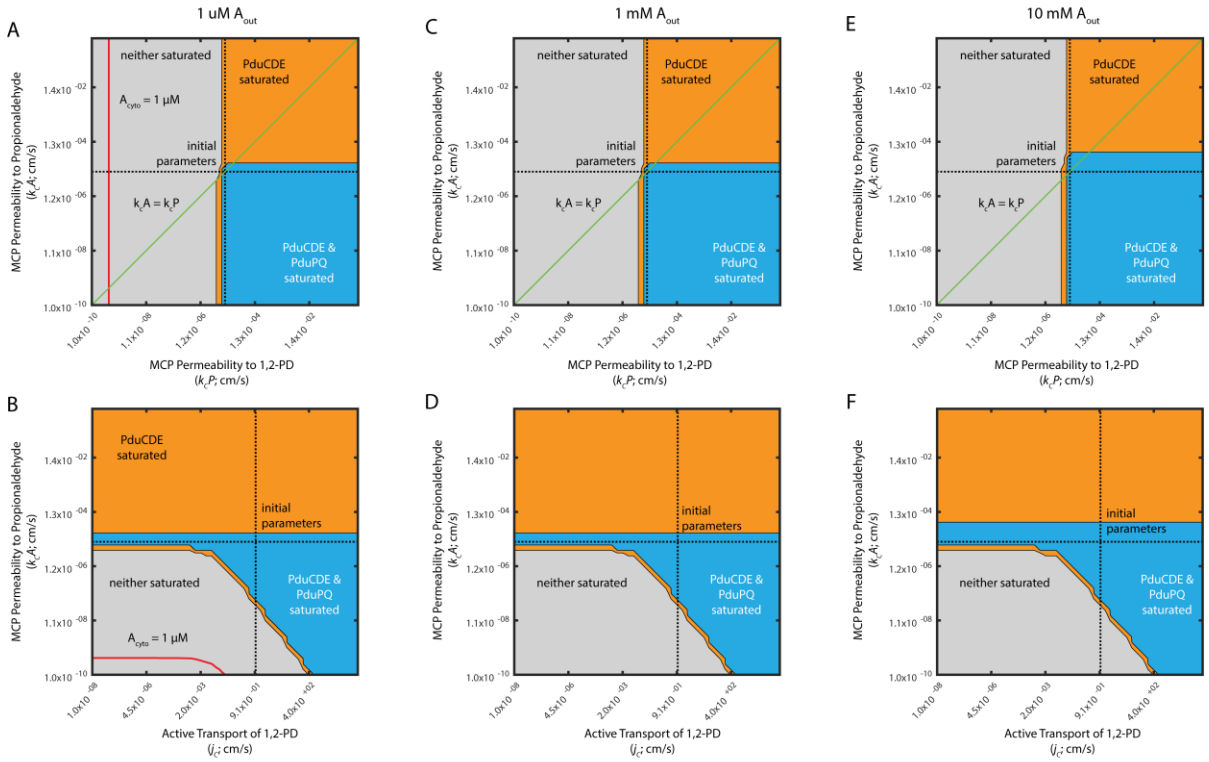

Figure S9. (A, C, E) Saturation phase space of PduCDE and PduP/Q with respect to k_cA and k_cP and (B, D, F) saturation phase space of PduCDE and PduP/Q with respect to k_cA and j_c for external propionaldehyde concentrations of (A, B) 1µM, (C, D) 1 mM, and (E, F) 10 mM. Regions of saturation (concentration of substrate > K_M of the appropriate enzyme) are plotted in blue when both enzymes are saturated, orange when only PduCDE is saturated, and in grey when neither enzyme is saturated. Red solid lines indicate when A_{cyto} is 1 µM; red dashed lines indicate when A_{cyto} is 10 nM. Black dashed lines indicate the baseline parameter values used in the model of the Pdu MCP. Green line in (A, C, E) indicates when $k_cA = k_cP$.