

Supplementary Information

Spatial presentation of biological molecules to cells by localized diffusive transfer

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Table S1: Upper limits of spot sizes given no reaction with the cells for diffusion rates representative of different solute types in a system with a pillar diameter of 200 μm and a gap of 100 μm between the pillar tip and the cell substrate.

Diffusion Rate:	4.0E-10 m^2/s	6.4E-11 m^2/s	4.4E-11 m^2/s
Spot size:	1016 μm	667 μm	606 μm

Table S2: Parameters used in FEM for diffusion and cellular uptake of solutes in Figure 3B-E and Table S1

	Parameter	Value(s)	Units
Constants	Diffusion coefficient retained solute (D_r)	0	m^2/s
	Cell layer thickness (cell)	0.01	mm
	Gel coating thickness (gel)	0.02	mm
	Pillar height (h)	2	mm
	Michaelis-Menten turnover constant (kcat)	1400	1/min
Figure 3B	Pillar diameter (d)	0.02, 0.2	mm
	Separation gap (gap)	0.05, 0.1, 0.2	mm
	Diffusion coefficient free solute (D_c)	4E-10	m^2/s
	Gel hindrance coefficient (D_{gel})	0.9	
	Enzyme concentration (E)	1	mM
	Michaelis-Menten constant (K_m)	0.1	mM
Figure 3C	Pillar diameter (d)	0.2	mm
	Separation gap (gap)	0.1	mm
	Diffusion coefficient free solute (D_c)	4E-10, 6.4E-11, 4.4E-11	m^2/s
	Gel hindrance coefficient (D_{gel})	0.9, 0.6, 0.4	
	Enzyme concentration (E)	10, 1, 0.1, 0.01, 0.001	mM
	Michaelis-Menten constant (K_m)	0.1	mM
Figure 3D	Pillar diameter (d)	0.2	mm
	Separation gap (gap)	0.1, 0.2	mm
	Diffusion coefficient free solute (D_c)	4E-10	m^2/s
	Gel hindrance coefficient (D_{gel})	0.9	
	Enzyme concentration (E)	1	mM
	Michaelis-Menten constant (K_m)	0.1	mM
Figure 3E	Pillar diameter (d)	0.2	mm
Table S1	Separation gap (gap)	0.1	mm
	Diffusion coefficient free solute (D_c)	4E-10, 6.4E-11, 4.4E-11	m^2/s
	Gel hindrance coefficient (D_{gel})	0.9, 0.6, 0.4	
	Enzyme concentration (E)	1	mM
	Michaelis-Menten constant (K_m)	0.1	mM

Table S3: Parameters used in FEM for diffusion and cellular uptake of solutes in Figure 3G-I

	Parameter	Value(s)	Units
Constants	Diffusion coefficient retained solute (D_r)	0	m^2/s
	Cell layer thickness (cell)	0.01	mm
	Separation gap (gap)	0.1	mm
Figure 3G	Reservoir height (h)	4, 2, 1	mm
	Diffusion coefficient unreacted dye (D_c)	$4E-10$	m^2/s
	Gel hindrance coefficient (D_{gel})	0.9	
	Consumption rate (k)	0.001	1/s
Figure 3H	Reservoir height (h)	4, 2	mm
	Diffusion coefficient unreacted dye (D_c)	$4E-10$, $6.4E-11$, $4.4E-11$	m^2/s
	Gel hindrance coefficient (D_{gel})	0.95, 0.9, 0.8 0.6, 0.45 0.4, 0.25	
	Consumption rate (k)	0.001, 0.00001	1/s
Figure 3I	Reservoir height (h)	4, 2, 1	mm
	Diffusion coefficient unreacted dye (D_c)	$4E-10$	m^2/s
	Gel hindrance coefficient (D_{gel})	0.9	
	Consumption rate (k)	0.001	1/s

Table S4: Equations used in FEM of transport of diluted species with assumed time-dependence including diffusion and cellular uptake of solutes in Figure 3 and Table S1 with parameters specified in Table S2 and Table S3

Figure 3B-E	In-gel diffusion coefficient	Gel compartment	$D_c * D_{gel}$
	Conversion rate (unreacted)	Cell layer	$-(c * E * k_{cat}) / (K_m + c)$
	Conversion rate (reacted)	Cell layer	$(c * E * k_{cat}) / (K_m + c)$
Table S1	In-gel diffusion coefficient	Gel compartment	$D_c * D_{gel}$
	Conversion rate (unreacted)	Cell layer	0
	Conversion rate (reacted)	Cell layer	$(c * E * k_{cat}) / (K_m + c)$
Figure 3G-1	In-gel diffusion coefficient	Gel compartments	$D_c * D_{gel}$
	Consumption from media	Cell layer	$-k * c$
	Consumption to cells	Cell layer	$k * c$