

Supplementary Information to:

Quantitative chemical biosensing by bacterial chemotaxis in microfluidic chips

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This supplementary information contains:

- Supplementary Figures S1-S6
- Supplementary Tables S1, S2
- Metropolis script

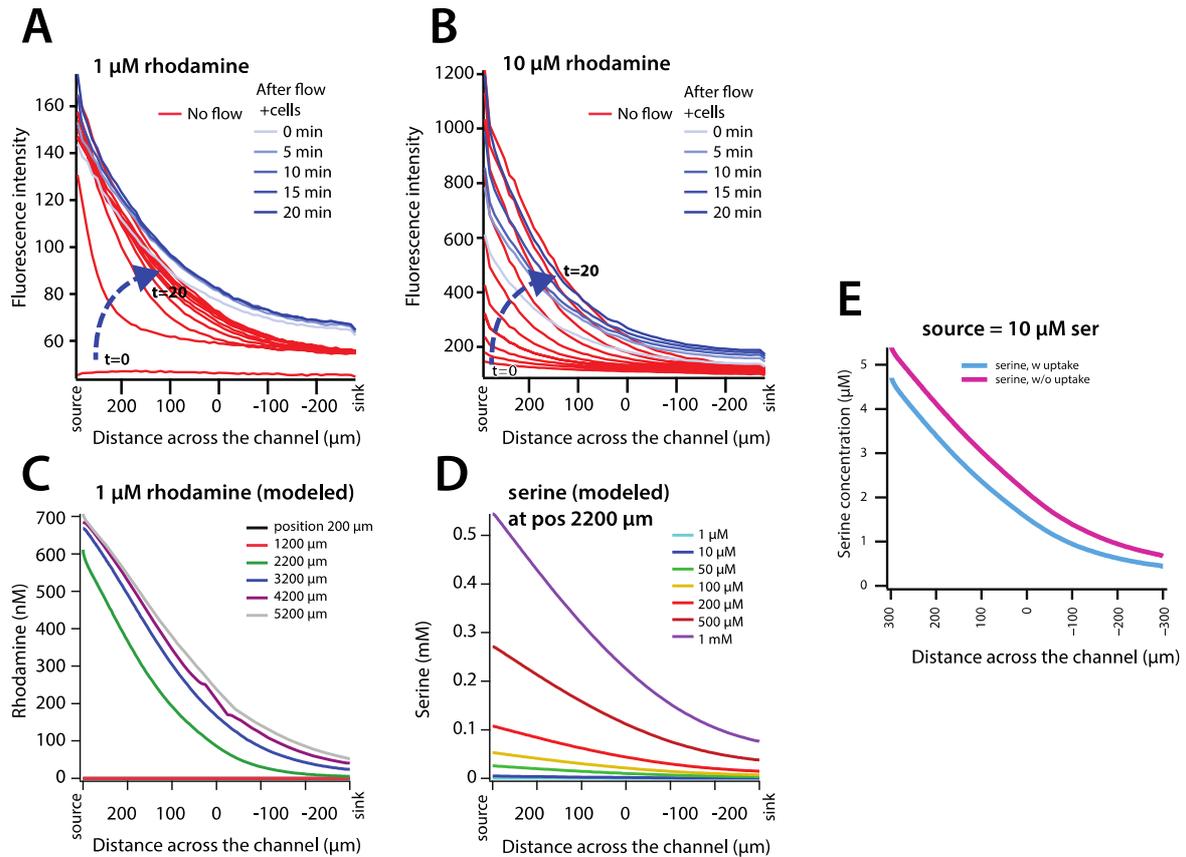


Figure S1. Gradient formation across the observation (middle) channel of the chip. Fluorescence profile formation across the channel without flow with (A) 1 μM or (B) 10 μM rhodamine in the source channel during 20 min (red lines, every 2 min), and after starting *E. coli* cell flow in the observation channel for another 20 min (blue lines, every 5 min). (C) Modeled rhodamine steady-state gradients at different transects along the observation channel. (D) Modeled serine gradients across the observation cell channel at position 2200 μm for seven source concentrations, as indicated, at the flow conditions computed as in Figure 1C. (E) Modeled effect of serine metabolism of *E. coli* on the established gradient at 10 μM serine source concentration and position 2200 μm in the observation channel. Note that for ease of comparison to the chip design, the distance across the channel is represented from 300 to -300 μm .

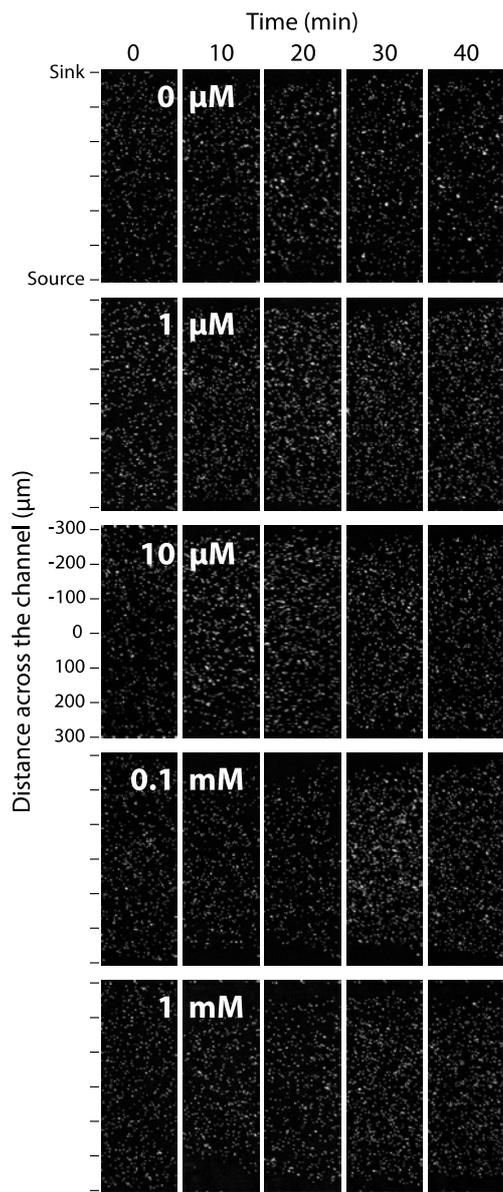


Figure S2: Non-chemotactic cell distribution of *E. coli* $\Delta fliC$ -mcherry toward serine. Images showing the distribution of $\Delta fliC$ -mcherry at the 2100-2200 μm location on the chip over time (0 - 40 min) as a function of the indicated serine concentration. Top is sink channel, bottom is source channel.

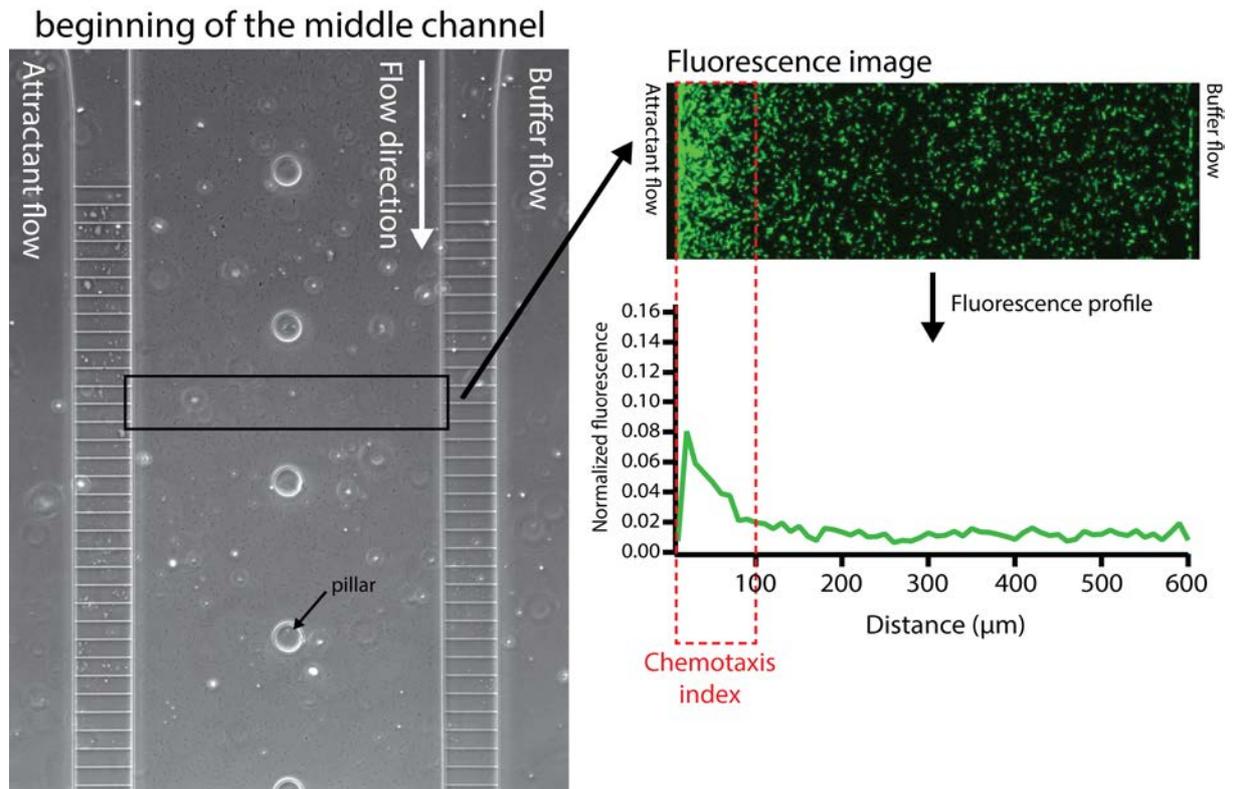


Figure S3: Chemotaxis index measurement setup. Chemotaxis response was quantified in a zone of 600 x 100 microns at a distance of 400 microns from the beginning of the filters. Fluorescence intensity profiles were extracted from the fluorescence images using ImageJ and normalized by the total fluorescence in the zone of measurement. The chemotaxis index was calculated as the proportion of fluorescence in the 100 μm segment closest to the source of attractant compared to the total fluorescence across the channel.

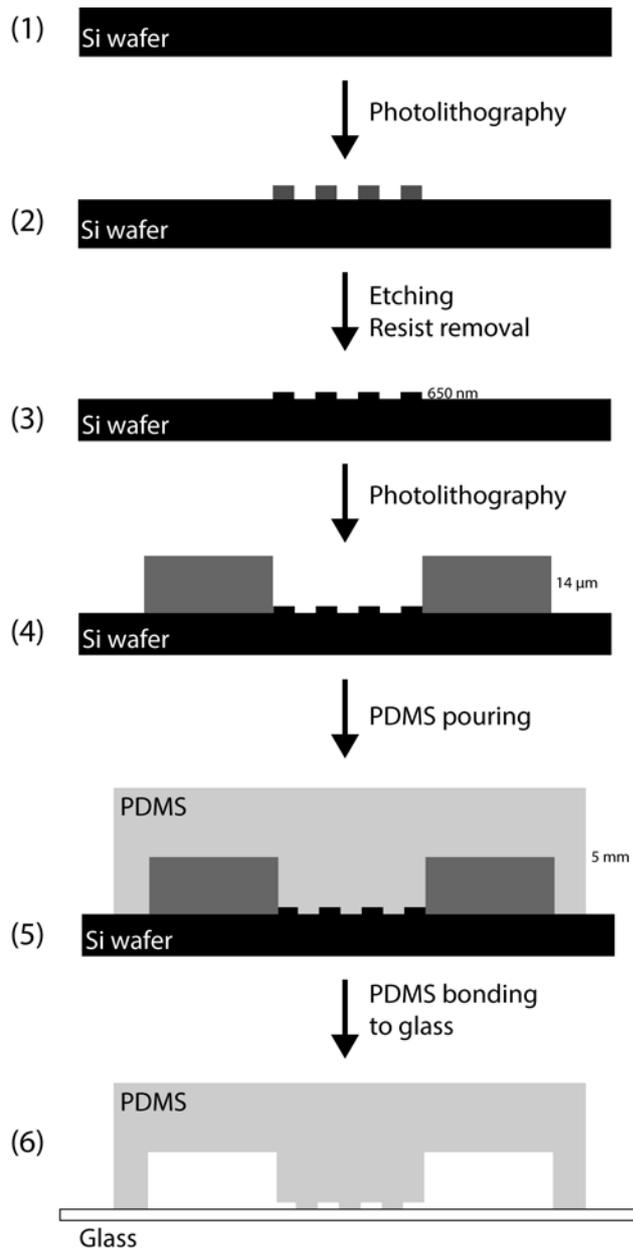


Figure S4: Microfluidic chip fabrication procedure. The fabrication procedure starts with a silicon wafer (1). A photolithography process produces a layer of resist at the filter position that protects this zone during the etching step (2). The etching results in the formation of the negative of the 650 nm high channels of the filters (3). A second step of photolithography produces the mold of the channels with a resist layer of 14 microns high (4). This inverted mold is used multiple times to produce the PDMS chips, by pouring PDMS on it and let polymerize (5). Once polymerized, the PDMS is peeled off the inverted mold and, after punching holes for the inlets, is bonded to the glass slide by a plasma treatment (6).

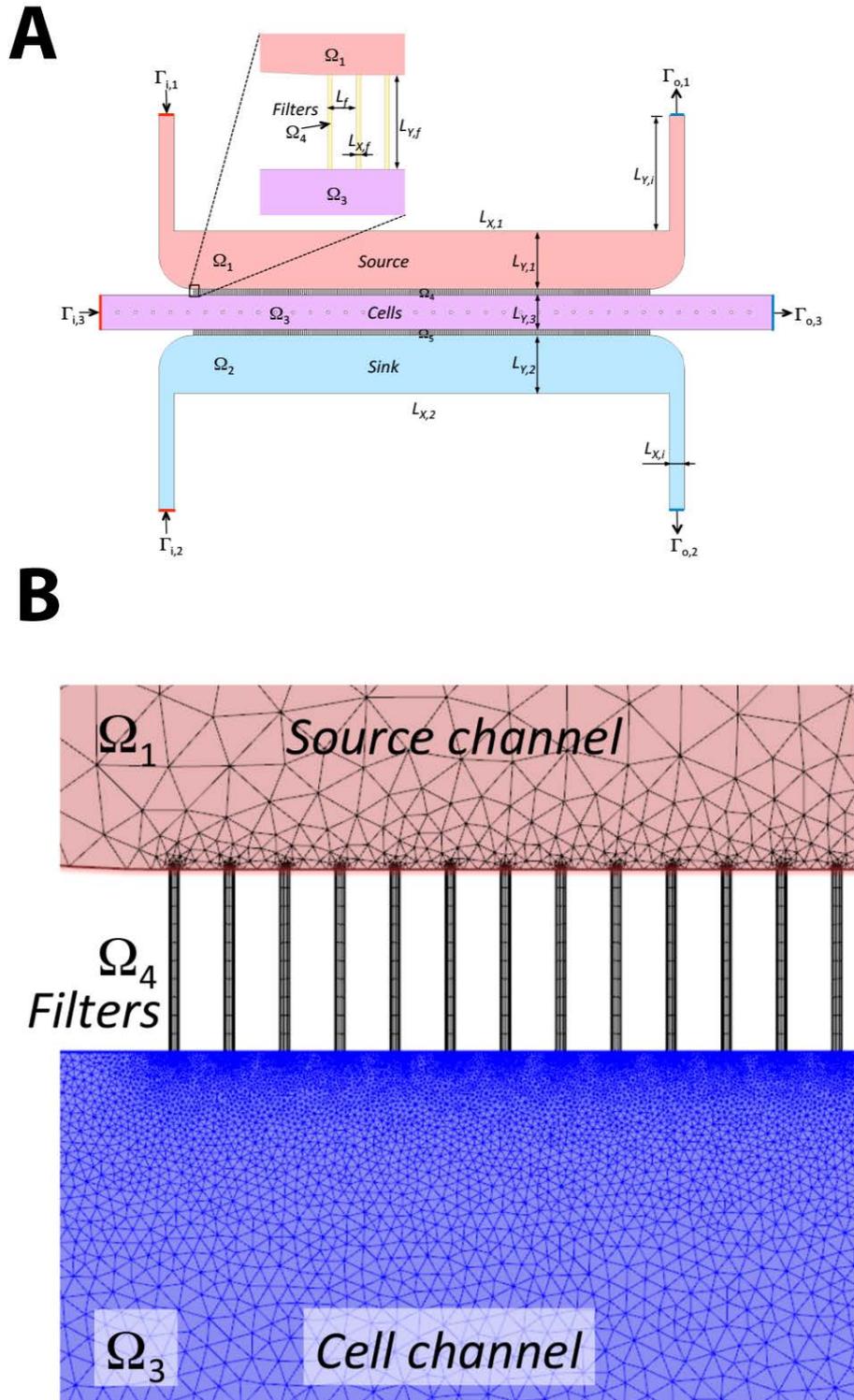


Figure S5. (A) Model geometry, dimensions, domains and boundaries. Ω_1 : Source domain (fed with chemoattractant solution), Ω_2 : Sink domain (fed with water), Ω_3 : Cells domain (fed with a suspension of cells), Ω_4 and Ω_5 : Filter domains (separate the cells from source and sink channels). $\Gamma_{i,1}$, $\Gamma_{i,2}$, $\Gamma_{i,3}$: Inflows, $\Gamma_{o,1}$, $\Gamma_{o,2}$, $\Gamma_{o,3}$: Outflows. The geometry dimensions are listed in Table S1. (B) Finite element mesh detail in the neighborhood of the filter region.

Table S1. Parameters of the continuum steady-state model

Description	Symbol	Value	Units	Source
<i>Geometry</i>				
Height (z-direction)				
- channel	h	14	μm	experimental
- filter		0.25	μm	
Cell channel				
- width	$L_{Y,3}$	0.6	mm	experimental
- length	$L_{X,3}$	11.5	mm	
Source and sink channels				experimental
- width	$L_{Y,1}, L_{Y,2}$	1	mm	
- length	$L_{X,1}, L_{X,2}$	9	mm	
Inlet/outlet channels				experimental
- width	$L_{X,i}$	0.26	mm	
- length	$L_{Y,i}$	2	mm	
Filters				experimental
- width	$L_{X,f}$	5	μm	
- length	$L_{Y,f}$	100	μm	
Filter spacing	L_f	30	μm	experimental
<i>Flow</i>				
Water viscosity (at 20°C)	μ	0.001	Pa s	-
Water density	ρ	1000	kg m^{-3}	-
Flow rate source/sink channels inlet	$F_{in,l}$	0.25	$\mu\text{L min}^{-1}$	experimental
Flow rate cell channel inlet	$F_{in,m}$	0.003	$\mu\text{L min}^{-1}$	experimental
<i>Solutes</i>				
Diffusion coefficient ^{a)}				
- rhodamine B	D_S	3.6×10^{-10}	$\text{m}^2 \text{s}^{-1}$	(Culbertson <i>et al.</i> , 2002)
- serine		8.9×10^{-10}		(Ma <i>et al.</i> , 2005)
Serine concentration in inflow	$c_{S,i}$	1, 10, 20, 50, 100, 200, 500, 1000	$\mu\text{mol L}^{-1}$	experimental
Maximum serine uptake rate	v_{max}	338	$\text{nmol mmol}^{-1} \text{min}^{-1}$	(Kayahara <i>et al.</i> , 1992)
Michaelis-Menten half-saturation coefficient	K_m	6	$\mu\text{mol L}^{-1}$	(Kayahara <i>et al.</i> , 1992)
<i>Cells</i>				
Basic motility coefficient	D_X	3×10^{-9}	$\text{m}^2 \text{s}^{-1}$	estimated
Chemotaxis sensitivity coefficient	χ_0	8×10^{-4}	$\text{cm}^2 \text{s}^{-1}$	estimated after (Chen <i>et al.</i> , 1998)
Receptor-ligand dissociation constant	K_C	25–30	μM	estimated after (Kalinin <i>et al.</i> , 2009)
Cell concentration in inflow	$c_{X,i}$	10	mmol L^{-1}	experimental
Maximum cell density	$c_{X,max}$	$10 \times c_{X,i}$	mmol L^{-1}	b)

a) corrected for 20 °C

b) To calculate the biomass in mM we use a typical elemental formula of *E. coli* cells of $\text{CH}_{1.8}\text{O}_{0.5}\text{N}_{0.2}$ that corresponds to a molecular dry weight of 24.6 g/C-mol biomass. At an estimated individual *E. coli* cell weight of 300 fg and a starting suspension of $8 \cdot 10^8$ cells per mL, this corresponds to 10 C-mmol biomass L^{-1} .

Table S2. Parameters, variables and functions used in the Metropolis model

Description	Symbol in script	Symbol in main text	Value	Units	Source
Counter for simulation steps		s	0, 1, 2, ..., 1200	steps	1200 corresponds to 60 min
Distance per time step of simulation	d		20	μm	Chosen
Grid width	h	L_y	600	μm	Channel width
Grid length	l	L_x	2500	μm	Channel length
Cell simulation window position			2000–2500	μm	
Random angle	r	θ	rand(0:360)	degrees	
Chemotactic sensitivity	ksi_0	ξ_{\square}	$1 \cdot 10^5$	-	(Chen <i>et al.</i> , 1998)
Receptor-ligand dissociation constant	c_{dis}	K_C	0.03	mM	(Kalinin <i>et al.</i> , 2009)
Free parameter Gibbs-Boltzmann equation	beta	β	-0.000013	-	Fitting
Correction factor	alpha	α	0.4	-	Fitting
<i>Gradient</i>					
Local serine concentration in y-direction for inlet concentration	$g(y)$	$c_s(p)$		mM	
0 mM ser			0	mM	Definition
0.001 mM ser			$-0.0006/(0.715 \cdot \exp(-0.0056 \cdot y) + 0.684) + 0.00093$	mM	Fitting
0.01 mM ser			$-0.009/(1.027 \cdot \exp(-0.005 \cdot y) + 1) + 0.0095$	mM	Fitting
0.1 mM ser			$-0.09/(1.34 \cdot \exp(-0.005 \cdot y) + 1.04) + 0.092$	mM	Fitting
1 mM ser			$-0.9/(1.5 \cdot \exp(-0.0054 \cdot y) + 1.03) + 0.9$	mM	Fitting
<i>Mean gradient</i>					
Local serine starting concentration for inlet concentration	c	$c_{s,0}$		mM	
0 mM			1	mM	Arbitrary
0.001 mM ser			0.00028	mM	Fitting
0.01 mM ser			0.00236	mM	Fitting
0.1 mM ser			0.0268	mM	Fitting

1 mM ser			0.279	mM	Fitting
<i>Adaptation function</i>		$a(t)$	$1/(1 + e^{-k*(\frac{s}{100} - s_0)})$		Logistic function
steepness	k	k			
mid-sigmoidal inflection point on <i>simulation</i> -axis	s_0	s_0			
0 mM ser			1		Definition
0.001 mM ser			k=2.5; $s_0=0.01$	-	Fitting
0.01 mM ser			k=2; $s_0=0.1$	-	Fitting
0.1 mM ser			k=1.5; $s_0=2$	-	Fitting
1 mM ser			k=1, $s_0=3$	-	Fitting

Metropolis script

```
function metropolis(x,y,g,d,c_dis,ksi_0,beta,alpha,a,c)

#x,y; starting position on the (x,y) grid within cell accumulation window position
#Number of simulation steps: 1200 (corresponding in the experiments to 60 min; so ~3 s equivalent to
each simulation step). This function is called at each time step.
#Number of cells: 5000
#Number of simulations: 5
#sind(r): sinus in degrees of random angle
#cosd(r): cosinus in degrees of random angle
#rand(): random number between 0 and 1 from a uniform distribution
#h=600 (grid width)

## function H ##
    r = rand(0:360);

    Hx = ksi_0*(c_dis*c/(c+c_dis)^2)^alpha*(g(y)/c);
    Hy = ksi_0*(c_dis*c/(c+c_dis)^2)^alpha*(g(y+d*sind(r))/c);

## metropolis algorithm ##

    if sind(r)<=0
        if y <= -d*sind(r)
            y = 0
            x = x + d*cosd(r)
        else
            y = y + d*sind(r)
            x = x + d*cosd(r)
        end
    else
        if exp(-beta*a*(Hy-Hx)) >= rand()
            if y >= h - d*sind(r)
                y = h
                x = x + d*cosd(r)
            else
                y = y + d*sind(r)
                x = x + d*cosd(r)
            end
        end
    end

    return [x,y]
end
```

Supplementary references

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