InterCells User's Guide (version 1.0)

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1. About

1.1 General

InterCells is an agent-based Monte-Carlo simulation of user-defined cellular interfaces. The simulation allows for membrane molecules, embedded at intercellular contacts, to diffuse and interact, while capturing the topography and energetics of the plasma membranes of the interface. The simulation was developed following Weikel and Lipowsky[1]. The code is written in Matlab (MathWorks). The simulation is open-source, interactive and modular.

The goal of the simulation is to facilitate an accessible, rapid, yet quantitatively critical feedback for generating experimentally testable hypotheses and the adaptation of working models in an iterative way.

1.2 Requirements

The simulation runs on MATLAB 2012a or newer versions. It runs on a standard PC. Typical runtime of the simulations takes ~5min for 10,000 iterations on a PC with i7 quad processor. Such simulation include two interacting surfaces, each of 400x400 pixels.

1.3 Installing the simulation

All simulation files are provided are available online on Github (https://github.com/ShermanLab/InterCells). These files should be downloaded to the User's computer under a directory that can be accessed by Matlab.

1.4 Licensing and citation

The simulation is freely available under the free GNU General Public License.

The User is requested to cite Neve-Oz et al. [2] in any publication in which the simulation was used.

2. The structure of the simulation

The simulation structure can be divided into multiple layers (Fig. 1). The first layer includes the input parameters and their respective GUI. The second layer includes the core of the simulation, where the physical models and the simulation algorithms are embedded and run. The third layer includes the output of the simulation. The fourth layer includes multiple analysis tools that are provided for quantitative interpretation of the simulation results and for their comparison with experimental data.

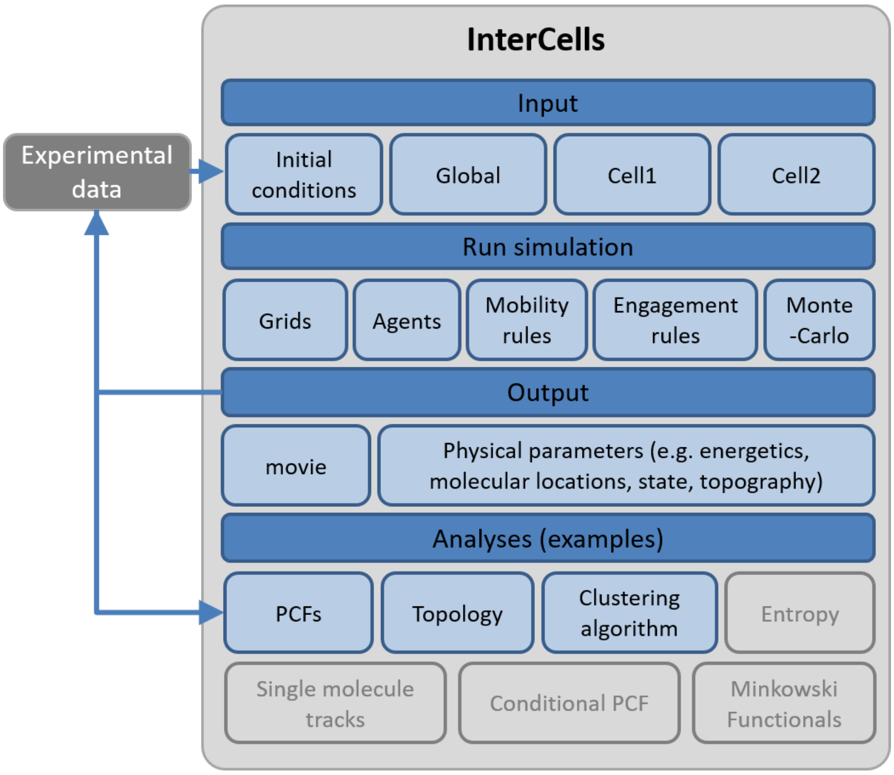


Fig 1. A schematic description of InterCells.

3. Operating the simulation

3.1 Loading and running the simulation

For loading the simulation, the User should type 'start_interface_simulation' and press 'Enter' in the Matlab command line. After running this command, the main window of the simulation GUI appears (Fig. 2). All inputs and outputs of the simulation can be accessed and changed through this main window (Fig. 2). After changing the desired parameters, the simulation can start by pressing the 'Run' button in the window.

3.2 Input

The code uses default parameters for its initial set-up, as described below. The user can modify all simulation parameters through the GUI, as graphically instructed in Figs 3-7 before running the simulation.

3.3 Output

Quantifiable readouts of the numeric simulations include the position and state of individual proteins, the morphology of the PM and their energetics. Visualization tools are provided for showing the simulation results. For instance, live evolution of molecular patterning is provided during the simulation run. The patterns can then be shown for each step individually, or as a movie. All parameters of the simulation are automatically saved into a 'Results' folder.

Examples of simulation outputs for the concrete example of the IS are provided in Neve-Oz, et al [2].

3.4 Analyses

InterCells integrates multiple statistical tools for quantitative analyses and interpretation of the results. Our tools include clustering algorithms and second-order statistics [3,4], and the topology analysis (Fig. 3). These tools are important for the quantitative comparison between results from experiments and from simulations.

3.5 Initial settings

Simulation parameters are divided into global parameters (Table 1), parameters of the plasma membrane of two interacting cells or of two interacting interfaces (Table 2), molecules parameters (Table 3), and analyses parameters.

An initial simulation data is formed after the setup starts. The initial setting is based on default parameters that define the behavior of all the entities in the simulation.

The simulation parameters are drawn from a specific simulation of pattern formation in the immune synapse (SI) between T cells and antigen presenting cells (APCs), as described in detail in Neve-Oz, et al [2]. This manuscript should be cited in reference to the simulation.

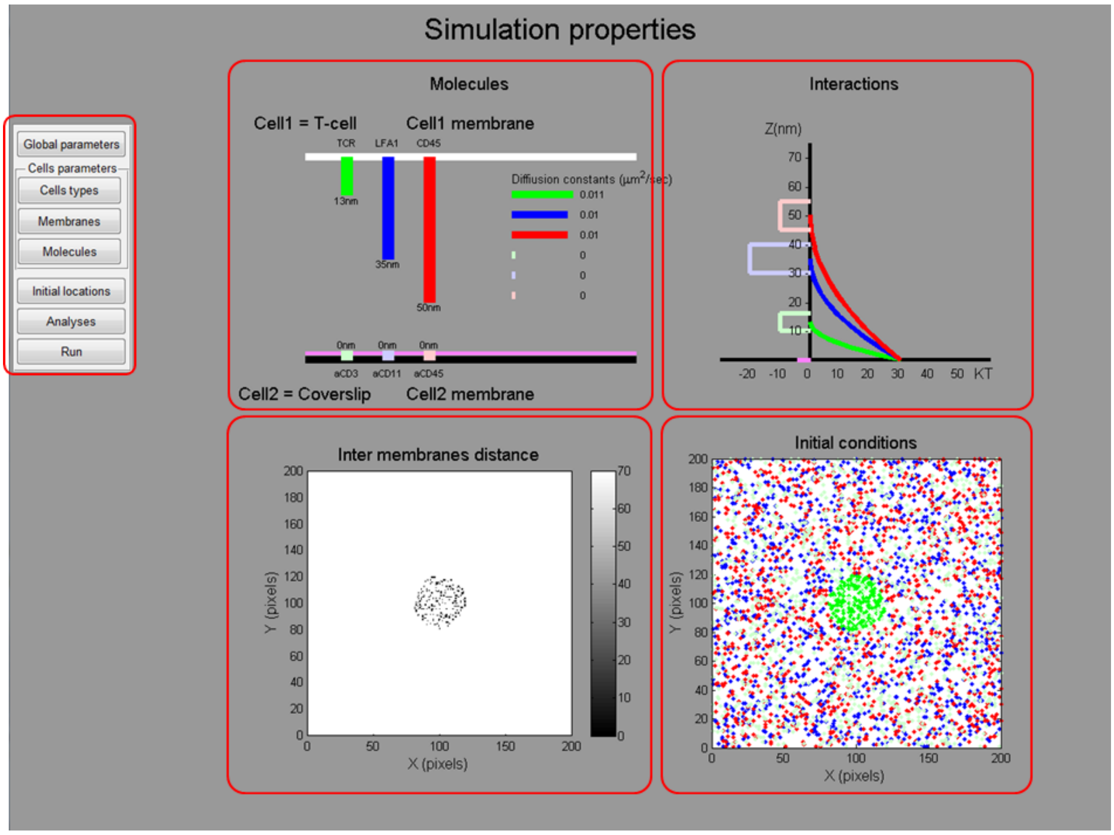
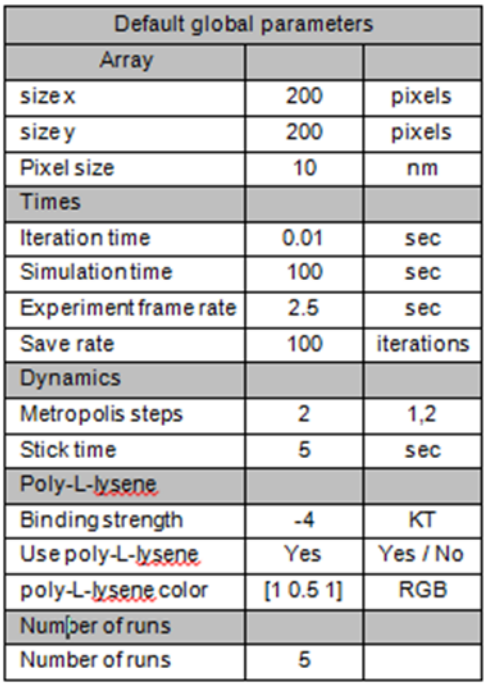


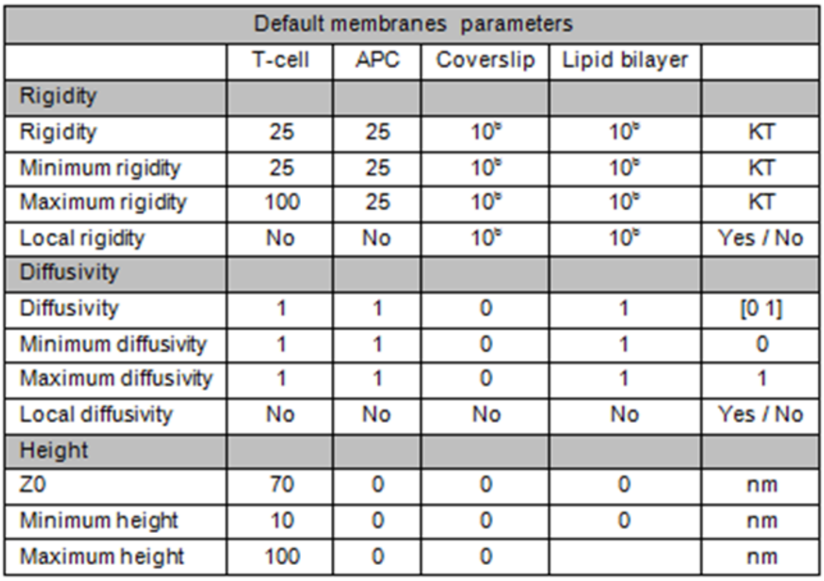
Fig 2. The main window of the GUI. The panel shows the functional parameters of the simulation. The GUI is controlled by the interactive menu at the left (red enclosure).

Table 1. Default Global parameters



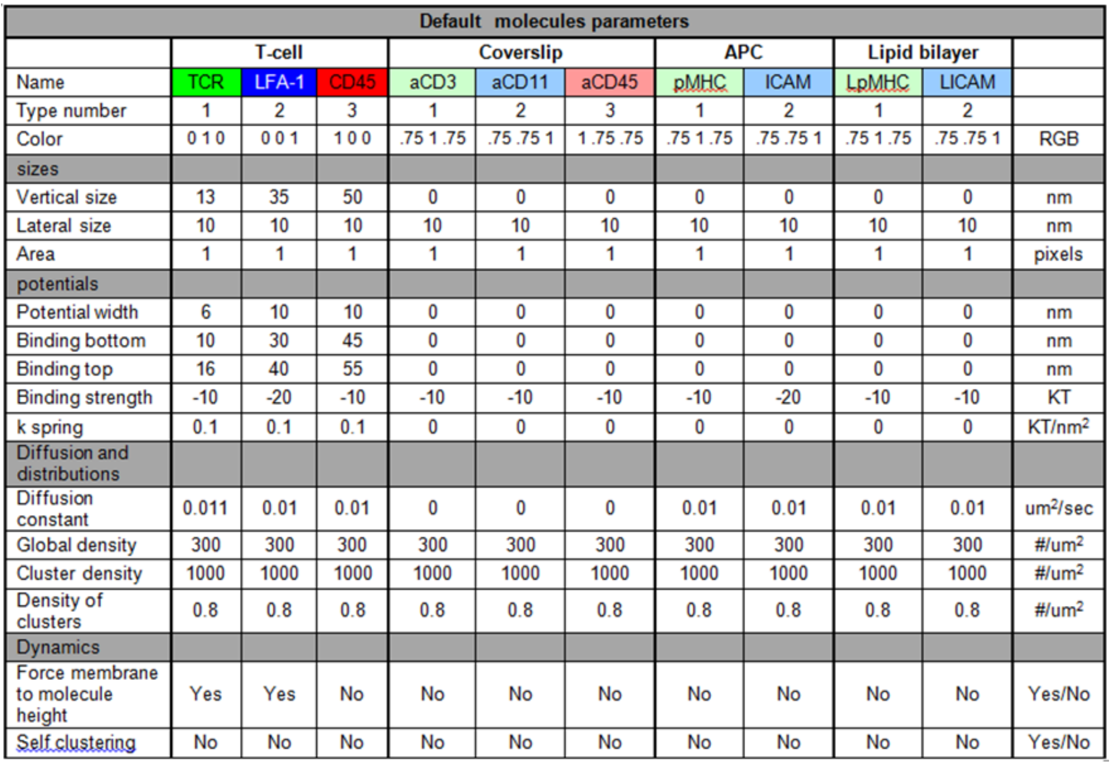
Default global parameters		
Array		
size x	200	pixels
size y	200	pixels
Pixel size	10	nm
Times		
Iteration time	0.01	sec
Simulation time	100	sec
Experiment frame rate	2.5	sec
Save rate	100	iterations
Dynamics		
Metropolis steps	2	1,2
Stick time	5	sec
Poly-L-lysene		
Binding strength	-4	KT
Use poly-L-lysene	Yes	Yes / No
poly-L-lysene color	[1 0.5 1]	RGB
Number of runs		
Number of runs	5	

Table 2. Default Membranes' parameters



Default membranes parameters					
	T-cell	APC	Coverslip	Lipid bilayer	
Rigidity					
Rigidity	25	25	10^6	10^6	KT
Minimum rigidity	25	25	10^6	10^6	KT
Maximum rigidity	100	25	10^6	10^6	KT
Local rigidity	No	No	10^6	10^6	Yes / No
Diffusivity					
Diffusivity	1	1	0	1	[0 1]
Minimum diffusivity	1	1	0	1	0
Maximum diffusivity	1	1	0	1	1
Local diffusivity	No	No	No	No	Yes / No
Height					
Z0	70	0	0	0	nm
Minimum height	10	0	0	0	nm
Maximum height	100	0	0		nm

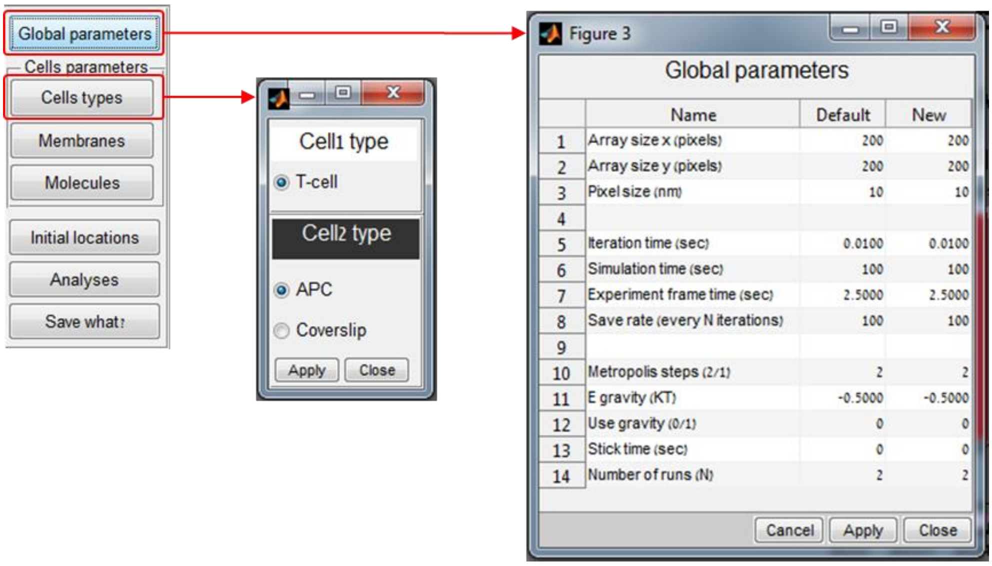
Table 3. Default Molecules' parameters



Default molecules parameters											
	T-cell			Coverslip			APC		Lipid bilayer		
Name	TCR	LFA-1	CD45	aCD3	aCD11	aCD45	pMHC	ICAM	LpMHC	LICAM	
Type number	1	2	3	1	2	3	1	2	1	2	
Color	0 1 0	0 0 1	1 0 0	.75 1 .75	.75 .75 1	1 .75 .75	.75 1 .75	.75 .75 1	.75 1 .75	.75 .75 1	RGB
sizes											
Vertical size	13	35	50	0	0	0	0	0	0	0	nm
Lateral size	10	10	10	10	10	10	10	10	10	10	nm
Area	1	1	1	1	1	1	1	1	1	1	pixels
potentials											
Potential width	6	10	10	0	0	0	0	0	0	0	nm
Binding bottom	10	30	45	0	0	0	0	0	0	0	nm
Binding top	16	40	55	0	0	0	0	0	0	0	nm
Binding strength	-10	-20	-10	-10	-10	-10	-10	-20	-10	-10	KT
k spring	0.1	0.1	0.1	0	0	0	0	0	0	0	KT/nm^2
Diffusion and distributions											
Diffusion constant	0.011	0.01	0.01	0	0	0	0.01	0.01	0.01	0.01	um^2/sec
Global density	300	300	300	300	300	300	300	300	300	300	#/um^2
Cluster density	1000	1000	1000	1000	1000	1000	1000	1000	1000	1000	#/um^2
Density of clusters	0.8	0.8	0.8	0.8	0.8	0.8	0.8	0.8	0.8	0.8	#/um^2
Dynamics											
Force membrane to molecule height	Yes	Yes	No	No	No	No	No	No	No	No	Yes/No
Self clustering	No	No	No	No	No	No	No	No	No	No	Yes/No

3.6 Modifying the simulation parameters and initial conditions

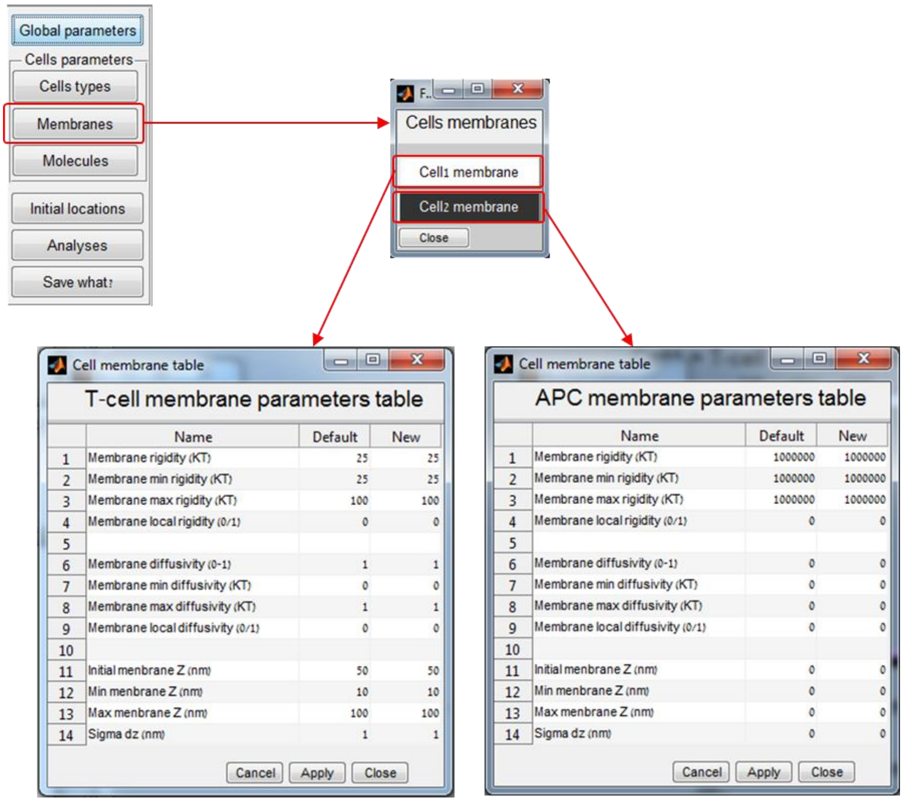
We provide below a graphical explanation on how to modify the simulation parameters before its run (Figs. 3-7).



Global parameters

	Name	Default	New
1	Array size x (pixels)	200	200
2	Array size y (pixels)	200	200
3	Pixel size (nm)	10	10
4			
5	Iteration time (sec)	0.0100	0.0100
6	Simulation time (sec)	100	100
7	Experiment frame time (sec)	2.5000	2.5000
8	Save rate (every N iterations)	100	100
9			
10	Metropolis steps (2/1)	2	2
11	E gravity (KT)	-0.5000	-0.5000
12	Use gravity (0/1)	0	0
13	Stick time (sec)	0	0
14	Number of runs (N)	2	2

Fig 3. Setting the global parameters of the simulation



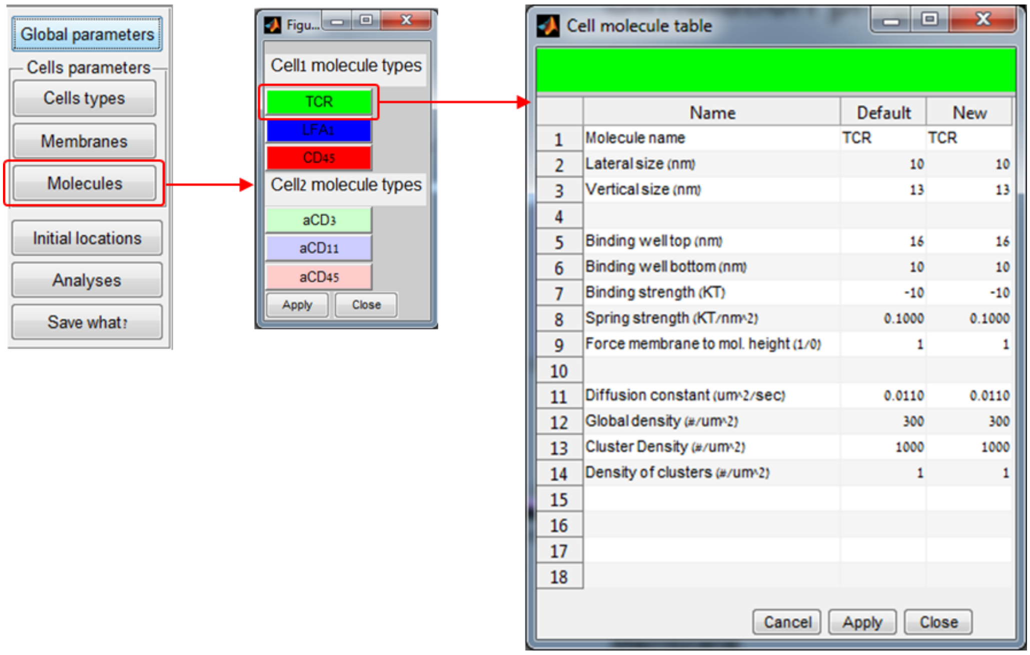
T-cell membrane parameters table

	Name	Default	New
1	Membrane rigidity (KT)	25	25
2	Membrane min rigidity (KT)	25	25
3	Membrane max rigidity (KT)	100	100
4	Membrane local rigidity (0/1)	0	0
5			
6	Membrane diffusivity (0-1)	1	1
7	Membrane min diffusivity (KT)	0	0
8	Membrane max diffusivity (KT)	1	1
9	Membrane local diffusivity (0/1)	0	0
10			
11	Initial menbrane Z (nm)	50	50
12	Min menbrane Z (nm)	10	10
13	Max menbrane Z (nm)	100	100
14	Sigma dz (nm)	1	1

APC membrane parameters table

	Name	Default	New
1	Membrane rigidity (KT)	1000000	1000000
2	Membrane min rigidity (KT)	1000000	1000000
3	Membrane max rigidity (KT)	1000000	1000000
4	Membrane local rigidity (0/1)	0	0
5			
6	Membrane diffusivity (0-1)	0	0
7	Membrane min diffusivity (KT)	0	0
8	Membrane max diffusivity (KT)	0	0
9	Membrane local diffusivity (0/1)	0	0
10			
11	Initial menbrane Z (nm)	0	0
12	Min menbrane Z (nm)	0	0
13	Max menbrane Z (nm)	0	0
14	Sigma dz (nm)	0	0

Fig 4. Setting the Membranes' parameters



	Name	Default	New
1	Molecule name	TCR	TCR
2	Lateral size (nm)	10	10
3	Vertical size (nm)	13	13
4			
5	Binding well top (nm)	16	16
6	Binding well bottom (nm)	10	10
7	Binding strength (KT)	-10	-10
8	Spring strength (KT/nm^2)	0.1000	0.1000
9	Force membrane to mol. height (1/0)	1	1
10			
11	Diffusion constant (um^2/sec)	0.0110	0.0110
12	Global density (#/um^2)	300	300
13	Cluster Density (#/um^2)	1000	1000
14	Density of clusters (#/um^2)	1	1
15			
16			
17			
18			

Fig 5. Setting the Molecules' parameters

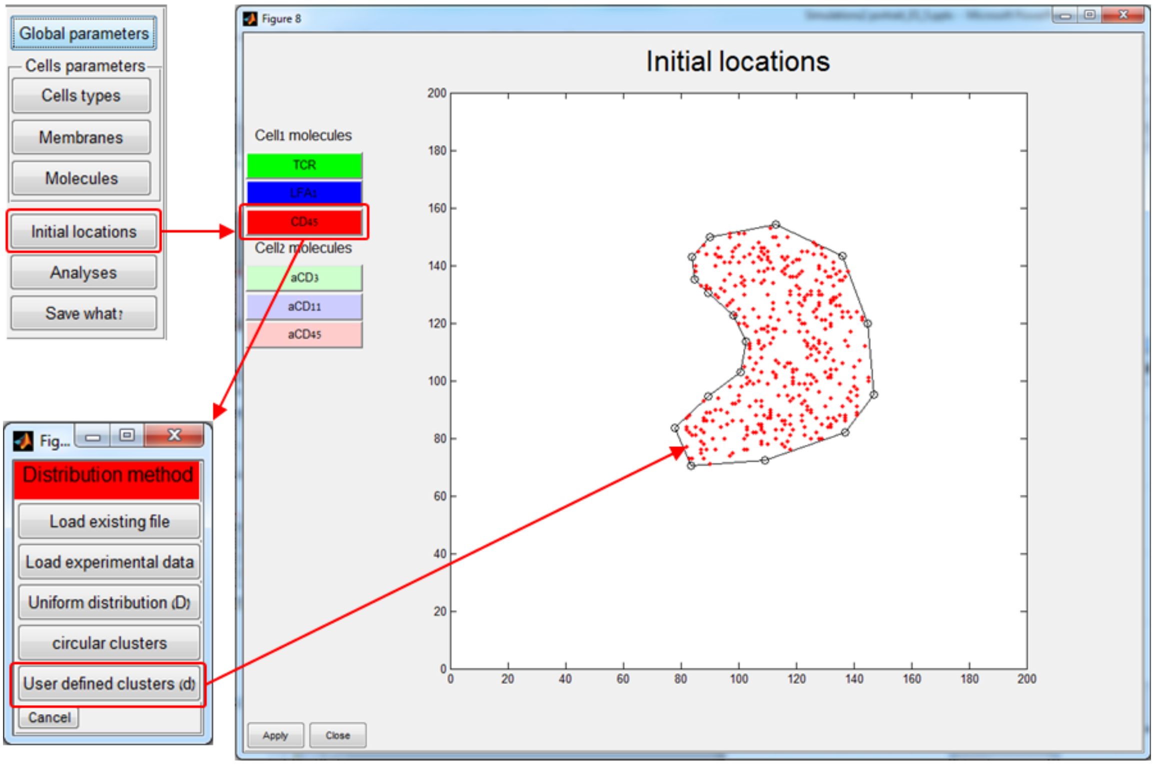
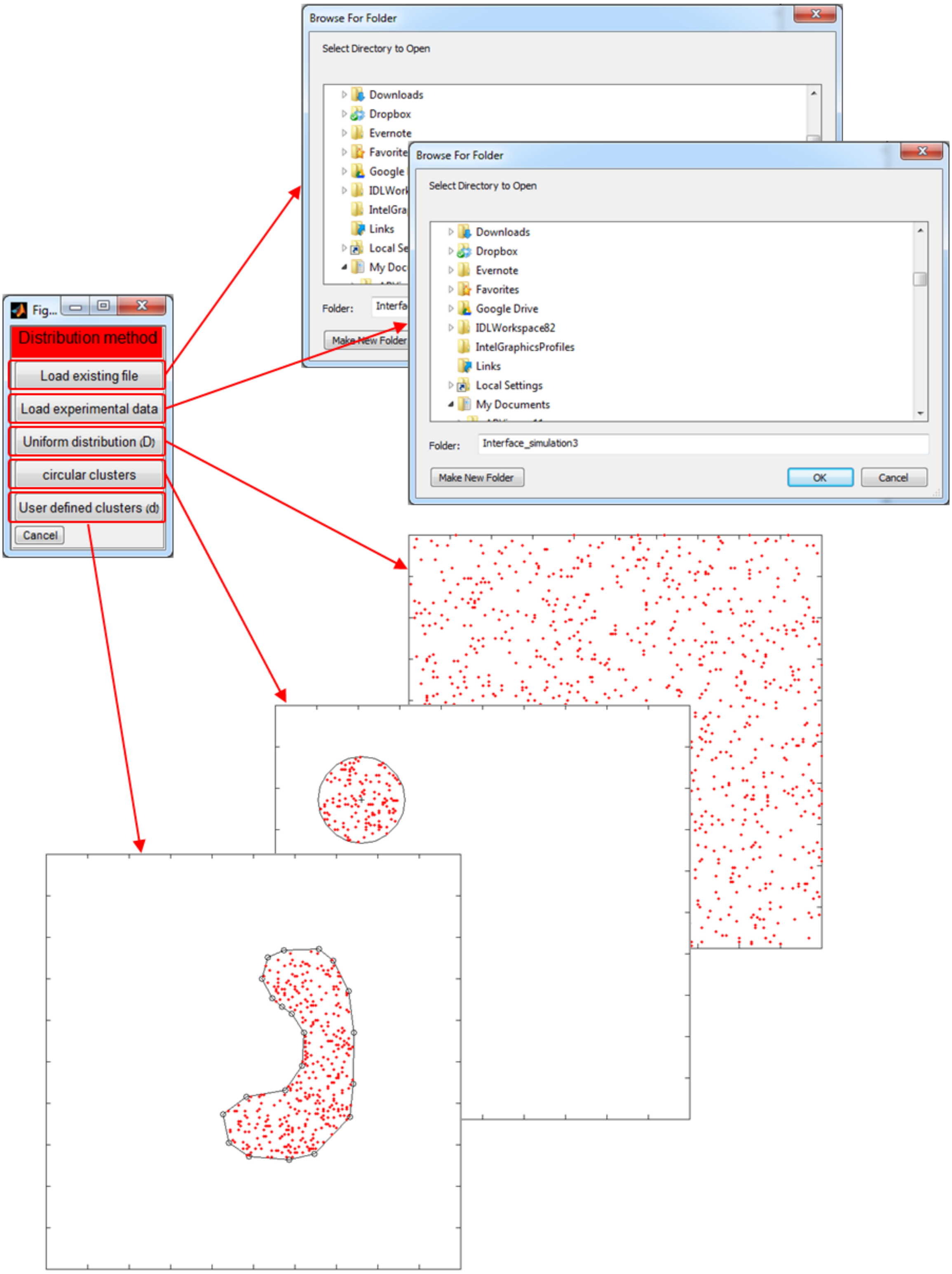


Fig 6. Setting the initial placement of molecules. The placement of each molecule type can be spatially defined in the simulation. For instance, a cluster can be defined by the user via the computer Mouse. Left Mouse clicks in the window set the cluster edges. A single right click of the Mouse sets the last edge. The defined molecules are than distributed randomly within the cluster.



Folder: Interface_simulation3

Fig 7. Setting the initial placement of molecules (continued). Alternative ways to distribute molecules include their uploading from experimental data. This way results in hybrid simulations. Likewise, molecular coordinates generated by any other means can be imported. Otherwise, similar to the instructions in Fig. 6, molecules can be distributed uniformly in circular clusters (rather than in user defined polygons). A uniform distribution of molecules across the simulated field is another useful option, by clicking the 'uniform distribution' button in the menu.

4. References

1 Weikl, T. R. & Lipowsky, R. Pattern formation during T-cell adhesion. *Biophys J* **87**, 3665-3678, (2004).

2 Neve-Oz, Y. S., J. Razvag, Y., Sherman, E. InterCells: a generic Monte-Carlo simulation of intercellular interfaces captures nanoscale patterning at the immune synapse. *Submitted*, (2018).

3 Sherman, E., Barr, V., Manley, S., Patterson, G., Balagopalan, L., Akpan, I., Regan, C. K., Merrill, R. K., Sommers, C. L., Lippincott-Schwartz, J. & Samelson, L. E. Functional nanoscale organization of signaling molecules downstream of the T cell antigen receptor. *Immunity* **35**, 705-720, (2011).

4 Sherman, E., Barr, V. A. & Samelson, L. E. Resolving multi-molecular protein interactions by photoactivated localization microscopy. *Methods* **59**, 261-269, (2013).