

SimulatorGUI

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0 Getting Started

0.1 Quickstart

0.1.1 Introduction

The Simulator GUI is software that allows you to combine biochemical models with a three dimensional cell geometry. Further, the simulator provides an interface for specifying initial conditions and conducting virtual experiments, as well as supporting multiple methods for visualizing the state of the simulation. There is also support for modifying certain simulation parameters after the simulation has been loaded; currently, this is limited to inserting and modifying extracellular concentration gradients.

The included visualization methods are:

- 3D geometry of cells with color coded overlays for biochemical compound/species concentrations.
- Time Traces for biochemical compounds/species, with values obtained from simulation start through to current time.
- 2D views of user specified slices of cells. These 2D views support color coded concentration maps, line trace profiles and concentration tracking (vs time) in user specified cell regions.

0.1.2 Overview

All simulations begin in the Simulation Window. This is where the simulation is started and stopped, and where the 3D geometry of the entire space can be viewed. There is also the possibility to apply color coded overlays corresponding to the concentrations of the various biochemical species.

If you want to:

- View concentration data vs time:
 - Select a cell (shift+left click) from the Simulation Window and then proceed to the Time Trace Window.
- View a slice of a cell or the entire simulation space:
 - Select the cell (or the entire simulation space) from the Simulation Window. Select to view in the Slice Selection Window, use the sliders and buttons to align the slice, and then click to view it in the Slice Window.
- Modify or insert a gradient:

- This is done from the Slice Window. Select a cell (or the entire simulation space) in the Simulation Window, select to view it in the Slice Selection Window, pick a slice to view, and then press Add a Concentration Profile
- Track the concentration of some species in an arbitrary region of the cell:
 - This can be accomplished from the Slice Window. The menu option Regions of Interest contains several methods for this.
- View a more realistic rendering of the cell geometry:
 - This is done from the Simulation Window. Select the option Use Alternate Draw Scheme.

0.1.3 How Things Fit Together

This software supports varying levels of spatial resolution, and depending on the level of detail selected, different sets of functionality are made available.

- The Simulation Window contains the most high level functionality and is also where items are selected for closer inspection. All investigations into the state of the experiment will start here, and this is accomplished by selecting either the entire simulation space or an individual cell. Because of the difficulty in selecting specific points or regions in 3D, anything which requires interacting with a specific point in space (inserting a gradient, viewing the concentration of a region or volume element, etc) is NOT done here. While perhaps not immediately obvious, the ability to select individual cells becomes very useful when simulating a crowded, multi-cellular environment. Being able to isolate 1 out of 2 cells is, perhaps, of limited utility. On the other hand, being able to select a single cell is practically required when the total number of cells is in the dozens or hundreds.
- The Slice Selection Window gives functionality that is analogous to moving through the Z-stack of a microscopy data set. While we offer the ability to view color coded representations of species concentrations within this view, this functionality is intended only to provide context when determining which slice to examine further (i.e which slice to look at in the Slice Window).
- The Slice Window is where the most granular operations take place. This is where you insert extracellular concentration gradients, view concentration profiles (along lines drawn by the user with the mouse), and create and view regions of interest. It should be noted that in order to insert an extracellular concentration gradient, it is necessary to be viewing a slice that contains extracellular space. As of this writing, the only way to do so is to have selected View Slice of Entire Region in the Simulation Window.

- The Time Trace Window is provided to view the net concentration, as a function of time, of one or more of the species present within a cell. Only one cell may be specified at a time, and this is done through the Simulation Window (shift + left click).

0.2 Setting Things Up

At startup you will be asked to specify a "simulation description". A simulation description contains everything that is needed to run a simulation. Not only references to the biochemical model and the geometry, but also the initial conditions and parameters of the model. If you cancel this dialog you will be able to specify the model and geometry you would like to simulate. In either case, the option to review and edit the initial biochemistry and parameters of the simulation will be offered.

0.3 Simulation Parameters

There are two primary windows associated with setting initial parameters: one to set the initial biochemistry, and one to modify the potts model parameters (for those simulations which involve dynamic morphologies).

0.3.1 Initial Biochemistry

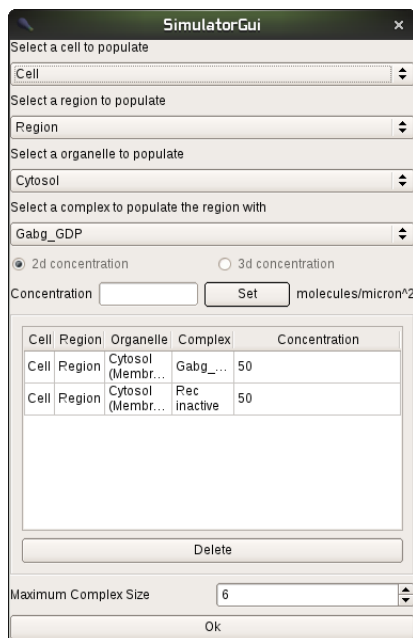


Figure 1: Set Initial Biochemistry

This window allows you specify the starting concentrations for all complexes in the model that do not contain state labels "Don't Care" or trans-bindings. You have to specify which cell, which region within the cell, and which organelle

within that region should be populated with a given complex. For freely diffusing (soluble) complexes you have the option to create membrane pores for the complex instead of the complex itself by clicking on the "Create membrane pore" button. The pores will be created on the membrane enclosing the selected organelle. A membrane for which the "Pore imports" button is selected will allow the transport of the selected complex into the compartment. See Appendix B - Membrane Pores for more information on pores.

0.3.2 Potts Model Parameters

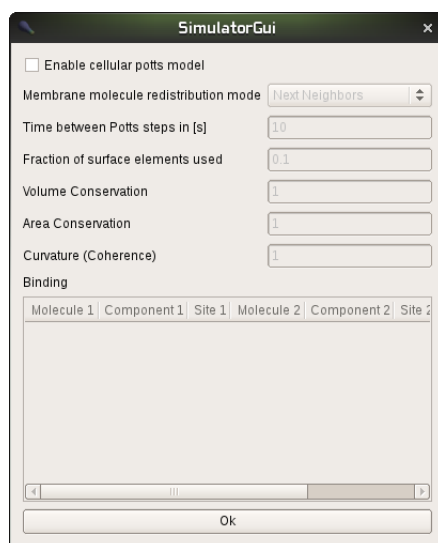


Figure 2: Set Potts Model Parameters

This window allows you to specify the potts model parameters, it will only be shown if every cell contains only the default 'cytosol' organelle, since potts dynamics for cells with additional organelles is not supported by the simulator. The options you can set here include enabling and disabling the potts model, time between potts steps, and setting the weights for volume and area conservations, curvature coherence, and surface fraction. For more information on the potts model, see Appendix B - The Potts Model.

0.3.3 Save and Begin

The last step of setting up is saving the settings of the simulation as a "Simulation Description" this allows to rerun the simulation with the same settings at a later time. This step is optional; if you do not want to save the settings, cancel the file dialog.

1 Program Overview and Window Explanation

Once the setup of the biochemistry and potts model (if applicable) is complete, the simulation will open several windows: The Simulation Window (start and stop the simulation), The Slice Selection Window (select slices of cell[s] for further analysis), and the Time Trace Window (view time series data for every cell and every chemical species).

1.1 The Simulation Window

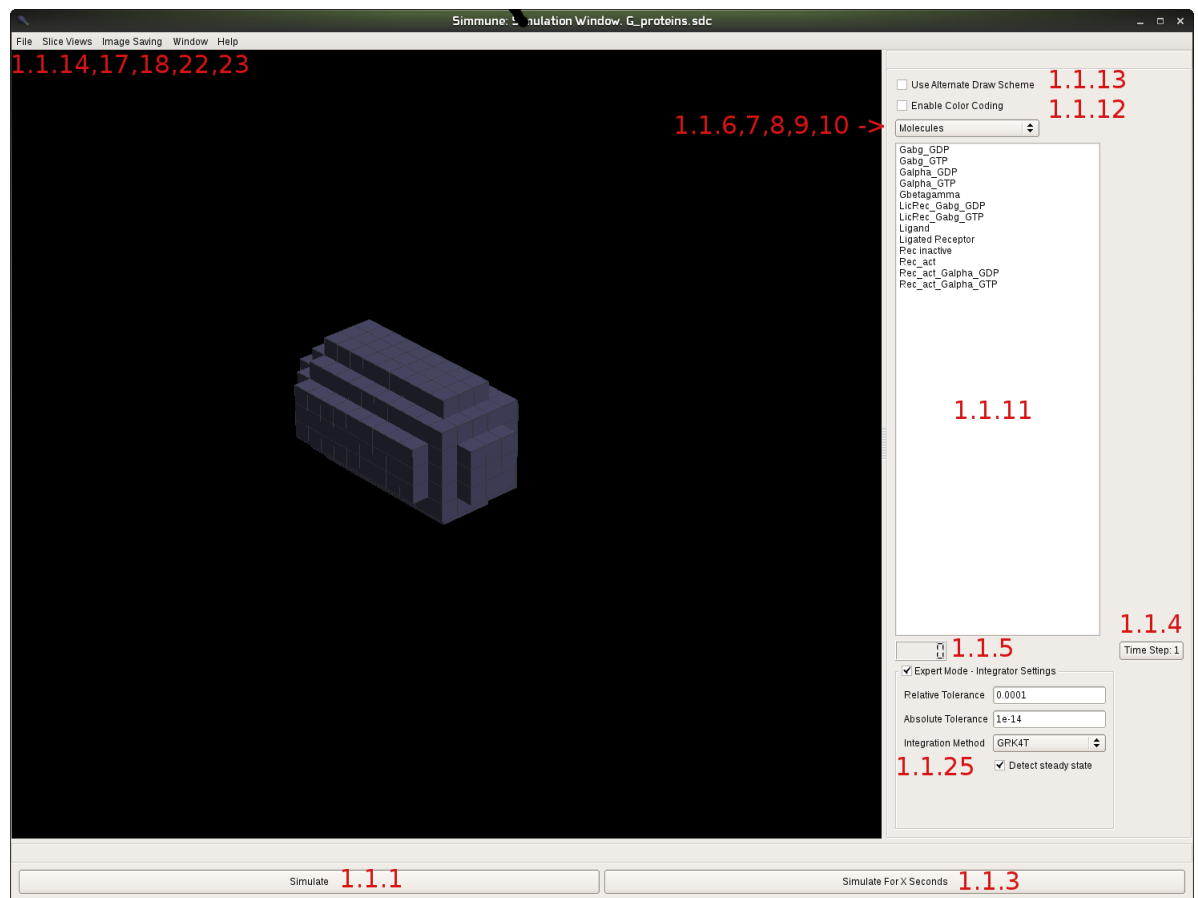


Figure 3: The Simulation Window

This window is where the simulation is managed. Primarily, this means starting and stopping the simulation, viewing graphical representations of the

entire simulation compartment, and selecting cells for further analysis.

1.1.1 Simulate Button

Starts and resumes the simulation. When pressed, the simulation begins and access to the interface will be blocked (excepting options to pause the simulation).

1.1.2 Pause/Quit the Simulation

When the simulation is in progress, a message box will appear and block access to all other parts of the simulation interface. The box offers two options, one to pause the simulation and one to quit the simulation. Clicking Pause will cause the simulation engine to stop after the current time step has been completed, at which point you will be able to interact with the interface again. Clicking Quit will cause the program to exit after a prompt for confirmation.

1.1.3 Simulate For X Seconds Button

Set the simulation to run for a number of seconds. When the countdown (displayed on the button) reaches 0, the simulation will automatically pause. Clicking this button again before X seconds have passed will reset the countdown to whichever value was just entered.

1.1.4 Time Counter

Displays the number of seconds the simulation has run for.

1.1.5 Time Step

Displays the current value of one time step (in seconds). When clicked, provides a prompt to change the value.

1.1.6 View Mode Combo Box

Dropdown combo box for displaying complexes, molecules, complexes containing molecules, and patterns. Please see those sections for further details.

1.1.7 View Complexes

When selected, a list of all Complexes defined in the modeler will appear in the Named Species List. Clicking on an item and enabling color coding will display the cells using a color gradient to indicate the surface concentration of the selected complex. This color gradient is calculated on a per-face basis.

1.1.8 View Molecules

When selected, a list of all Molecules, the components used to define a complex, will appear in the Named Species List. Clicking on an item and enabling color coding will display the cells using a color gradient to indicate the surface concentration of the selected molecule. This color gradient is calculated on a per-face basis.

1.1.9 View Complex with Molecule

When selected, a list of all Molecules will appear in the Named Species List. Clicking on an item and enabling color coding will display the cells using a color gradient to indicate the surface concentration of all **complexes which contain the selected molecule**. This color gradient is calculated on a per-face basis.

1.1.10 View Patterns

When selected, a list of all patterns will appear in the Named Species List. Clicking on an item and enabling color coding will display the cells using a color gradient to indicate the surface concentration of every complex which **matches the selected pattern**. This color gradient is calculated on a per-face basis. For more information on patterns, please see the Modeler documentation.

1.1.11 Named Species List

When Complexes, Complexes Containing Molecule, Patterns, or Molecules are selected, a list of all known species corresponding to the selected type will be displayed here. Left clicking on a species will select it and, when combined with color coding, allow for visualizations of its surface concentration.

1.1.12 Enable Color Coding

If this box is checked then a color gradient (visible in the display) will be used to indicate the relative surface concentrations of the species selected from the Named Species List. The scale runs from 'Cold' (purple, lower concentration) to 'Hot' (red, higher concentration), to represent the concentration of a selected species.

1.1.13 Use Alternative Draw Scheme

Renders (in the display) the cells using a draw method that represents the cell surface based on the same membrane orientation that the simulation engine uses to simulate membrane diffusion. This is in contrast to rendering the cells based on the positions of the volume elements, and the result is a smoother (softer edges) appearance.

1.1.14 3D Display Area

The space containing the 3D rendering of the simulation. The controls are:

Pan - Arrow Keys

Zoom in and out - Z and X keys

Change viewing angle - hold left or right mouse button, and drag.

Select a cell (used for viewing slices and time traces for a cell) - Shift + Left click on a cell.

1.1.15 Slice Views Menu

Menu containing options to view a slice of a cell and to view a slice of the entire simulation compartment.

1.1.16 View a Slice of a Cell

When a cell is selected, click this to display the cell in the Slice Selection Window. Please note that selecting a cell has no effect on either the Slice Selection Window or the Slice Window until this is pressed.

1.1.17 Slice View of Entire Region

Whereas View a Slice of a Cell displays only the selected cell, this functionality displays the entire simulation compartment in the Slice Selection Window.

1.1.18 File Menu

Drop down menu containing an 'Exit' button. Clicking 'Exit' will prompt for quitting the simulation.

1.1.19 Image Saving Menu

This menu has three sections. Options for automated image saving of the entire scene, options for automated image saving of a selected cell, and an option for creating a single image of the entire scene.

1.1.20 Options For Entire Scene

Contains two menu options which govern automatically (after every time step) writing the contents of the display to an image file. Selecting the first option in the drop down menu, which reads Automatic Image Saving is... off/on, toggles whether or not an image is saved. If the menu option reads "... off", then NO image is being saved; if the menu option reads "... on", then an image IS being saved. The second option in the drop down menu displays the file name and directory in which the images are being saved. By default, there is no location set and saving will not take place. When no location is set, clicking on EITHER

of these two options will prompt you for a save location (i.e if no location is set and you click to turn ON automatic saving, you will be prompted to provide a location for saving the image). The chosen filename is the base save name for the image files. Each image will be saved as FILENAMExxx-xxx.png, where FILENAME is the name you provided, and xxx-xxx is the simulation time when the image was taken. Because '.' is a reserved char on some system, a '-' is used in place of a decimal point in the time.

1.1.21 Options For Selected Object

Use is identical to Options For Entire Scene, but instead of saving an image of everything in the display area, only the cell selected when image saving is turned on will be saved. To save a different cell, turn off image saving (by clicking the item that reads "Image Saving is now ...on"), select a new cell (and a new location, if desired) and then turn image saving back on.

1.1.22 Take Snapshot

Saves an image of the display area. You will be prompted to select a location to save the image to.

1.1.23 Window

This menu allows you to bring up windows that aren't being shown. Each name in the menu refers to one of the program windows (corresponding to the name in the window title).

1.1.24 Help

Menu contains the 'about' option. Clicking 'about' will provide information about who developed the program and which third party libraries were used.

1.1.25 Expert mode - Integrator settings

This window allows to change the internals of the integration algorithm of the system of differential equations. The defaults are suitable for most situations. For a description for the parameters please refer to R. Weiner, B. A. Schmitt, H. Podhaisky, ROWMAP—a ROW-code with Krylov techniques for large stiff ODEs, Applied Numerical Mathematics, Volume 25, Issues 2-3, Special Issue on Time Integration, November 1997, Pages 303-319, ISSN 0168-9274, DOI: 10.1016/S0168-9274(97)00067-6. and E.Hairer G. Wanner Solving Ordinary Differential Equations II Stiff and Differential-Algebraic Problems, 2nd. Ed. 1996, Springer Series in Computational Mathematics, Springer, page. 110 The check box "Detect steady state" will cause the integrator to test if a steady state is reached, and if so advance the time without further integration. For the purpose of this function, a steady state is reached if all components of the right

hand side of the differential equation scaled by their variable are smaller than the relative tolerance.

1.2 The Slice Selection Window

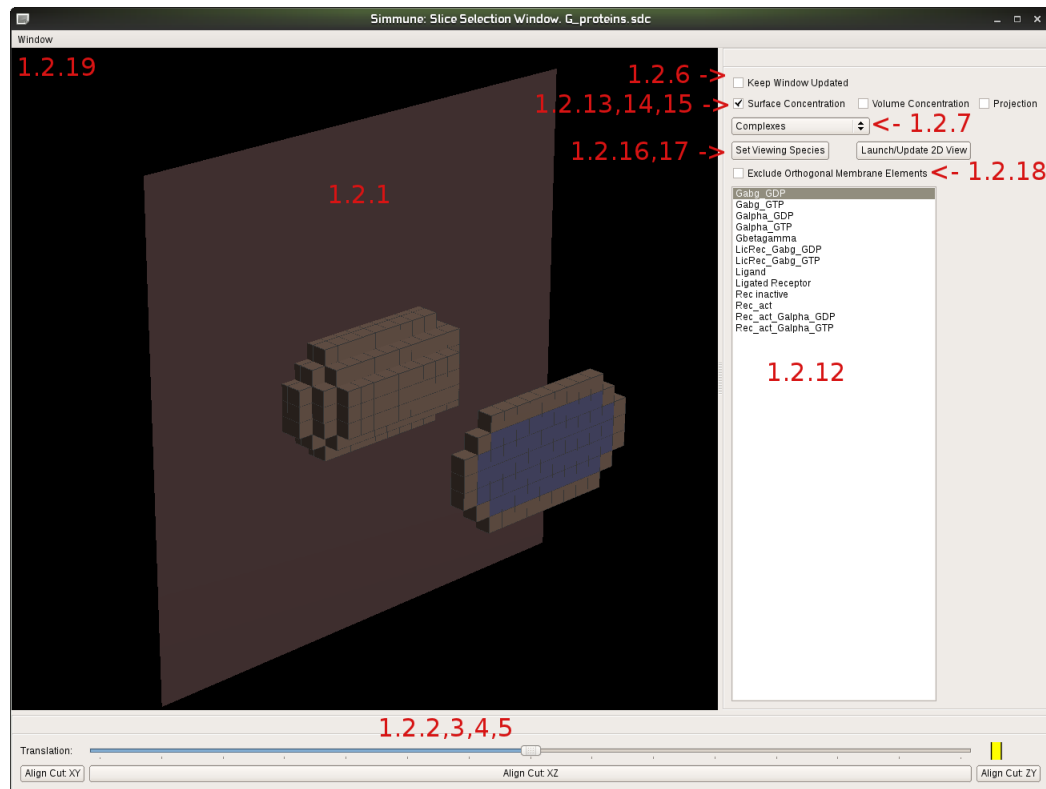


Figure 4: The Slice Selection Window

1.2.1 3D Display Area

The 3D Display Area contains the 3D rendering of the selected cells/region, the cutting plane and the section of the cell/region defined by its intersection with the cutting plane. The same controls as for changing the point of view in the Simulation Window apply here. If you don't see anything in the display area, then you need to do one of the following. (1) From the Simulation Window, you should select a cell and click the button labeled View a slice of a cell, or (2) click the button labeled Slice View of Entire Region.

1.2.2 Align Cut XY

1.2.3 Align Cut XZ

1.2.4 Align Cut YZ

These buttons are used to align the cutting plane with the coordinate planes. The intersection of the cutting plane with the cell(s) produces a slice. The cell(s), cutting plane, and slice are all visible in the display area.

1.2.5 Translation Slider

Moving this slider will move the cutting plane in a direction parallel to its normal. The cell(s), cutting plane and slice are all visible in the display area.

1.2.6 Keep Window Updated

When this box is checked, the cell morphology, visible in the display area, will be updated every time the simulation completes a step. The default behavior is to leave the morphology as it was last received from the Simulation Window, either through viewing a cell or the entire simulation compartment.

1.2.7 View Combo Box

Dropdown combo box for displaying complexes, molecules, complexes containing molecules, and patterns. Please see those sections for further details.

1.2.8 View Complexes

When this box is checked, a list of all complexes defined in the modeler will appear in the Named Species List. When a complex is selected (left click on it from the named species list), the distribution of that complex can be analyzed further by selecting a view mode and clicking either Update Display or View Current Slice. A view mode is one of Surface Concentration, Volume Concentration, or Projection.

1.2.9 View Complexes Containing Molecule

When this is selected, a list of all molecules will appear in the Named Species List. When a molecule is selected (left click on it from the named species list), the distribution of all **complexes which contain that molecule** can be analyzed further by selecting a view mode and clicking either Update Display or View Current Slice. A view mode is one of Surface Concentration, Volume Concentration, or Projection.

1.2.10 View Molecules

When this box is checked, a list of all molecules defined in the modeler will appear in the Named Species List. When a molecule is selected (left click on it from the named species list), the distribution of that complex can be analyzed further by selecting a view mode and clicking either Update Display or View Current Slice. A view mode is one of Surface Concentration, Volume Concentration, or Projection.

1.2.11 View Patterns

When this is selected, a list of all patterns will appear in the Named Species List. When a pattern is selected (left click on it from the named species list), its distribution can be analyzed further by selecting a view mode and clicking either Update Display or View Current Slice. A view mode is one of Surface Concentration, Volume Concentration, or Projection. For more information on patterns, please see the Modeler documentation.

1.2.12 Named Species List

Displays a list of species corresponding to whether Complexes, Molecules, Patterns, or Complexes Containing Molecules was chosen. Left click on a displayed item to select it. Once selected, Update Display and View Current Slice are used for further analysis of the selected species and its distribution within the cell or cell slice.

1.2.13 Surface Concentration

When Update Display or View Current Slice is clicked, then the corresponding visualization will use surface concentrations for displaying the distribution of, and calculating any data corresponding to, the selected species.

1.2.14 Volume Concentration

When Update Display or View Current Slice is clicked, then the corresponding visualization will use volume concentrations for displaying the distribution of, and calculating any data corresponding to, the selected species.

1.2.15 Projection

When Update Display or View Current Slice is clicked, then the corresponding visualization will use a projection for displaying the distribution of, and calculating any data corresponding to, the selected species.

1.2.16 Update Display

When clicked, the slice and cell(s) in the display will be rendered using a color gradient. The gradient reflects the relative local concentration/amount of the

species (ranges from Purple, least, to Red, most) selected in the named species display list, and is calculated based on the selected view mode. A view mode is one of Surface Concentration, Volume Concentration, or Projection. Note that changing the selected species or view mode will not be reflected in the display until this button is pressed.

1.2.17 View Current Slice

When clicked, the slice is displayed in the Slice Window. The species data for the slice is based on whether molecules, complexes, or complexes containing molecules were selected. See The Slice Window and Technical Notes - Slice Window Calculations for details.

1.2.18 Exclude Orthogonal Membrane Elements

When this box is checked, any calculations which involve membrane elements will include only those membrane elements which are parallel to the cutting plane. See Technical Notes - Slice Window Calculations for details.

1.2.19 Window Menu

This menu allows you to bring up windows that aren't being shown. Each name in the menu refers to one of the program windows (corresponding to the name in the window title).

1.3 The Slice Window

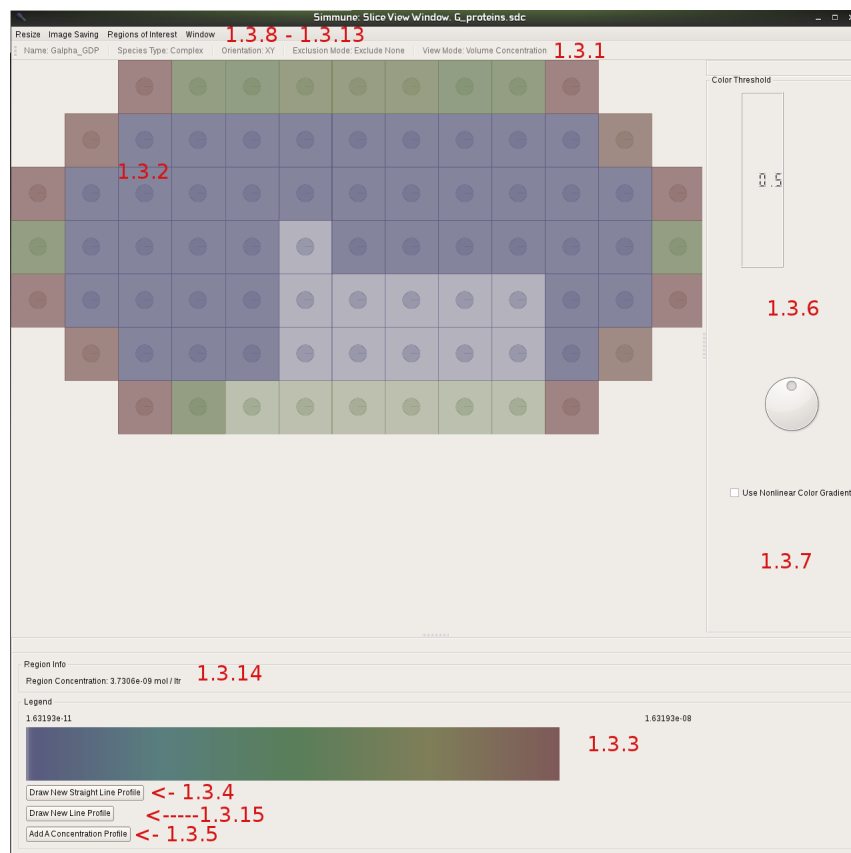


Figure 5: The Slice Window

1.3.1 Info Bar

Displays all of the settings selected in the Slice Selection Window. These settings are used in constructing and displaying all of the information accessible through this window. From left to right, these are: the name of the species, the species type, the orientation of the plane, the exclusion mode, and the view mode. The name of the species is the text of the item selected in Named Species List. The species type is one of: Complex, Molecule, Complex With Molecule or Pattern. The orientation of the plane is either XY, ZY, or XZ. The exclusion mode is either Exclude Orthogonal or Exclude None, and corresponds to whether Exclude Orthogonal Membrane Elements is checked (Exclude Orthogonal) or not (Exclude None). The View Mode is one of Surface Concentration, Volume

Concentration, or Projection. For additional information on each setting please see the corresponding item in this document documentation under the section Slice Selection Window.

1.3.2 Display Area

A 2D representation of the slice selected in the Slice Selection Window. Each square represents a separate volume element. If your slice is more than one volume element thick, then each square will represent up to two volume elements. The colors used for each square are shown using a gradient going from purple, representing those regions with the lowest concentration/amount of the selected species, to red, representing those regions with the largest concentration/amount of the selected species. For details on slices (what's contained in a slice; what does it mean to have a slice thickness greater than one) see Appendix B - Obtaining A Slice.

1.3.3 The Color Legend

The color gradient and values, directly above the gradient, show the mapping used to convert concentration/amount to a color. In the case depicted here, any region with a concentration of less than .00028013 molecules per square micron (seen above the purple color) will be rendered using a flat purple color whereas regions with a concentration greater than .28013 (shown at the opposite end of the gradient) will be red. Clicking on the legend will produce a tool tip showing what concentration/amount that color represents. The range used to produce the color gradient can be changed using the color threshold dial.

1.3.4 Draw New Straight Line Profile

Clicking this button allows the user to draw a line between two points in the display area in order to produce a graph showing the values contained in the intersected volume elements vs distance along the line. In order to draw the line, click the button and then left click, hold, and drag the cursor between any two points in the display area (the line will be drawn as you drag). The first click on the display area will set the start point for the line and releasing the left mouse button will set the end point. If your line lies entirely in one plane, then you will be presented with the option of giving your line a thickness and given the choice between two sampling types: gaussian and box. For additional information, please see Technical Notes - Line Profiles for more detailed information on how these graphs are generated.

1.3.5 Add A Concentration Profile

Brings up the Concentration Profile Wizard. This will allow you to add a gradient or constant concentration, for any diffusable species, to the extra-cellular space of the simulation compartment. See the Concentration Profile Wizard section for details.

1.3.6 Color Threshold

Moving the dial will change the values mapped to purple and red for the color gradient. Moving the dial counter clockwise will lower the threshold (causing everything to shift towards the red end of the spectrum) and vice versa for moving the dial clockwise. The number changing as you move the dial, ranging between 0 and 1, is a numerical representation of the color threshold. For details on what this number means, see Technical Notes - The Color Threshold and Dial

1.3.7 Use Non Linear Color Gradient

Uses an exponential function to determine how concentrations or amounts are mapped to colors in the color gradient.

1.3.8 Fit Slice To Window Menu

Menu with an option to fit the slice to the menu. Click this to change the scale of the image drawn in the display area. The slice will be expanded/shrunk to fit the available space.

1.3.9 Save Image

Saves, to a .png file, an image of everything appearing in the display area. A window will appear to prompt you for the file name and directory.

1.3.10 Regions of Interest

This menu contains options for specifying a region of a slice, and then viewing time plots of the concentration/amount of some species within that region. The portion of the slice that is included in this region is referred to as a 'Region of Interest'. A region of interest can be designated in two ways: by taking all of the points that **intersect a line** (see Line Saving), or by taking all of the points that lie within the region **intersected and surrounded by a line** (see Region Saving). For each time step, a single data point is obtained by averaging the concentrations/amount for all of the points in the region of interest.

1.3.11 Area Region Saving

A 'region' is defined as the space inclosed by an arbitrary outline, and this section covers how to set them, track them, and graph them.

The button labeled 'Set New Region' allows for the specification of new regions of interest. Once pressed, left click and drag the mouse around an area of the slice. As the mouse is moved, a line is drawn to indicate the shape of the region. To stop drawing, release the mouse button. Once released, the area of the slice that will be included in the region is drawn with a lighter shade (see 1.3.11 in the picture). Because only one region can be set at a time, clicking this button will overwrite the previous region data (after being prompted for confirmation).

However, both a region and a line area **can** be designated and tracked at the same time.

The button labeled 'Region Saving is ... On/Off' governs whether or not region data is being saved. Clicking it toggles saving on and off, and the current state is reflected in the button text. When region data is being saved, it reads 'Region Saving is ... On'. When region data is not being saved, it reads 'Region Saving is ... Off'.

The button labeled 'Show Region Graph' will display a graph of the concentration/amount data for all time steps in which tracking was turned on. This means that when 'Region saving is ... Off' is being displayed in the menu, no data will be gathered for subsequent time steps.

1.3.12 Line Region Saving

Setting a line allows for the creation of an area of interest which contains only those points that have been specifically clicked on or moused over. The definition of an area of interest is covered in the section Areas of Interest. The process of setting, tracking, and graphing an area of interest is the same regardless of how the area is defined (as a line or a region). See Region Saving for details on these processes.

1.3.13 Window Menu

This menu allows you to bring up windows that aren't being shown. Each name in the menu refers to one of the program windows (corresponding to the name in the window title). Click on the name of a closed window to bring it back.

1.3.14 Average Concentration of Region ...

Displays the concentration of a region. The concentration is calculated as an average over all of the concentrations in all of the volume elements included in the region. Like all of the information present in this view, it is as up to date as the last time View Slice was clicked.

1.3.15 Draw New Line Profile

After clicking this button a line profile with an arbitrarily chosen path can be generated. Volume elements are only included in the profile when the cursor gets close to their center. 'Close' is defined as within the shaded circle (which will appear after the button is pressed) that is drawn in the center of each volume elem

1.4 Concentration Profile Wizard

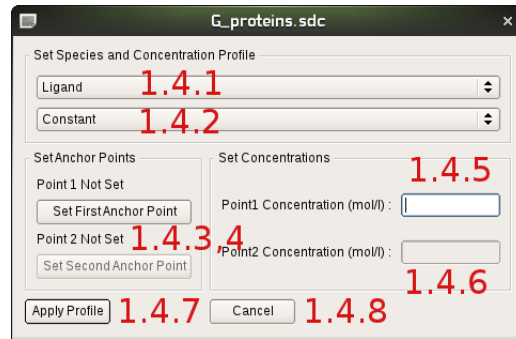


Figure 6: The Concentration Profile Wizard

The Concentration Profile Wizard is used to insert a concentration, of a diffusible species, into every **extra cellular volume element**. There are two types of concentration profile: a constant concentration profile and a linear gradient

A constant concentration profile applies the same concentration to every extra-cellular volume element.

A linear gradient interpolates two concentrations linearly between two points. Concentrations outside the range determined by the pair of points are extrapolated with a cutoff at 0. The concentration in a voxel not on the line connecting the control points is evaluated by projecting the point onto the line and using the projected coordinate to determine the voxel's concentration. The following figure illustrates the process:

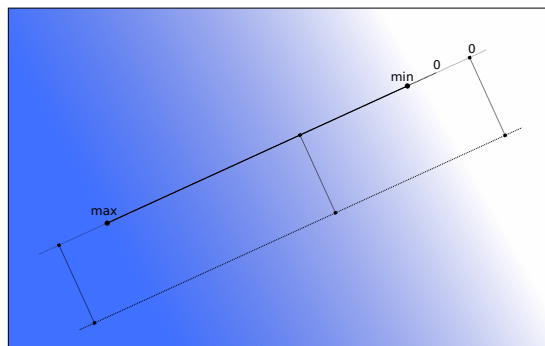


Figure 7: Linear Gradient

1.4.1 Species Selection

This combo box contains all of the diffusable species in the current simulation. Whichever species is selected when 'Apply Profile' is clicked will be the species used for the concentration profile.

1.4.2 Profile Selection

This combo box contains the three different profile types: constant, linear gradient. Whichever profile type is selected when 'Apply Profile' is clicked will be used for applying the concentration profile. See the Concentration Profile Wizard overview for details on the different profile types.

1.4.3 Set First Anchor Point

Clicking this button will dismiss the wizard so that you can select the first anchor point. An anchor point is selected by clicking on any of the volume elements that are visible in the slice view window. For linear gradients, the anchor points are used to fit the function used to apply the gradient.

1.4.4 Set Second Anchor Point

Identical in use to Set First Anchor Point, but enables the setting of the second anchor point. If the concentration profile is set to Constant, then this button will be disabled (and unused).

1.4.5 Concentration at Anchor Point 1

Specify the concentration for anchor point one. The gradient will be calculated so that the concentration at the first anchor point is equal to the value entered here. This input accepts scientific notation.

1.4.6 Concentration at Anchor Point 2

As Concentration at Anchor Point 1, but sets the concentration used for the second anchor point. If the concentration profile is set to Constant, then this field will be disabled (and unused).

1.4.7 Apply Profile

Applies a concentration profile as defined through the options chosen in this wizard. If an option hasn't been specified (i.e, unchosen anchor point or anchor point concentration), then clicking this button will do nothing.

1.4.8 Cancel

Discards all choices and returns to the simulation without applying a concentration profile.

1.5 The Time Trace Window

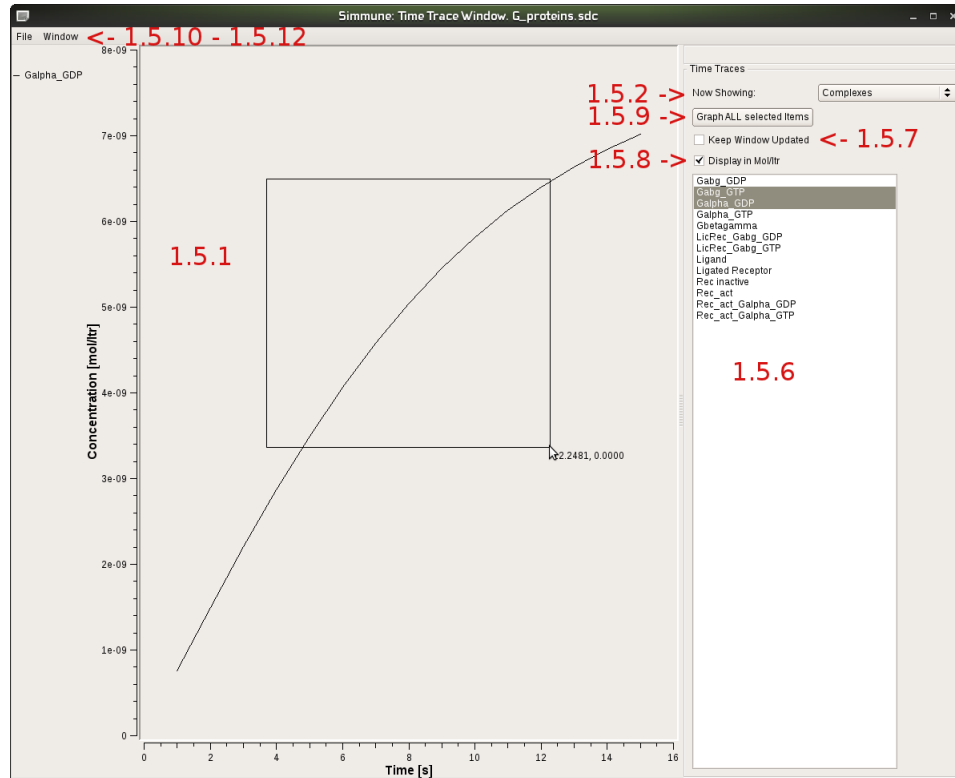


Figure 8: Time Trace Window

This window is used to display graphs of concentration/molar amounts of a species vs time. For a cell, selected in the simulation window, the molar amount of a species is summed over all volume elements and then, if concentration is selected, transformed into $\frac{\text{mol}}{\text{ltr}}$ by dividing by the total volume of the cell in liters. The final graph is shown with respect to time (in seconds), with $t = 0$ being the start of the simulation. **This window is disabled when no cell is selected.**

1.5.1 Graph Display Area

This is the space the graph occupies. Whichever cell is currently selected in the Simulation Window will be the cell whose data is graphed in this window. When the simulation begins, nothing is shown here. To change this, first select

a cell from the Simulation Window and then check one of molecules, patterns, complexes, or complexes containing molecule and then select whichever species you'd like to graph from the list in the Named Species List. Left clicking in this area will let you draw a box around a portion of the graph you'd like to zoom in on. Once the box is drawn (left click once, release left click, move the mouse to see the box being drawn, and then left click again to finish), the graph and axes will be adjusted to reflect the portion of the graph that is within the zoom box. The graph can be panned in the +/- X and Y directions using the left and right (X axis), and up and down (Y axis) arrow keys. **Right clicking in the graph area will reset the graph to the default view (un-zooms and un-pans everything).**

1.5.2 View Mode Combo Box

Contains options for displaying lists of Complexes, Molecules, Complexes With Molecules, and Patterns.

1.5.3 View Complexes

Choosing between these options will cause a list of all named complexes to appear in the Named Species List.

1.5.4 View Molecules

Choosing between these options will cause a list of all named molecules to appear in the Named Species List.

1.5.5 View Complexes with Molecule

Choosing between these options will cause a list of all named complexes containing a molecule to appear in the Named Species List.

1.5.6 Named Species List

When Complexes, Complexes Containing Molecule, or Molecules are selected, a list of all known species corresponding to the selected type will be displayed here. Left clicking on a species will select it and cause a graph of the concentration (or amount) of that species vs time to appear in the graph display area. You can graph multiple species by shift+left clicking them and then selecting Graph ALL Selected Items.

1.5.7 Keep Window Updated

When this box is checked, the graph will automatically be updated after each simulation step.

1.5.8 Display in Mol/Ltr

When this box is checked, the graph will use a concentration in $\frac{mol}{ltr}$ as the units for the y-axis. By default, the box is unchecked and the units are amount of molecules in the selected cell.

1.5.9 Graph ALL Selected Items

When multiple items are selected, then clicking this will graph all of those items.

1.5.10 File Menu

Contains an option for writing the plot to file.

1.5.11 Write Plot To File

Writes the contents of the current plot to file. When selected, a prompt will appear asking for a save location and a save name. In the location selected, a file will be generated for every species being displayed in the plot. The files will be named `<GIVEN_FILE_NAME><SPECIES>`, where `<GIVEN_FILE_NAME>` is the name entered in the file prompt, and `<SPECIES>` is the species that is being saved. For example, if `Galpha_GDP` and `Galpha_GTP` are being plotted when this option is selected and `March_1st_` was entered as a file name, then two files would be generated. One would be labeled `March_1st_Galpha_GDP` and the other would be labeled `March_1st_GalphaGTP`. The file format is:

```
#<SDC_file>
#<view_mode>
<x_axis_label><TAB><y_axis_label>
<x_axis_value 0><TAB><y_axis_value 0>
<x_axis_value 1><TAB><y_axis_value 1>
```

...

Where: `<SDC_file>` is the Sim Description File used to load the simulation, `<view_mode>` is one of `Molecule`, `Complex`, `Complex With Molecule`, or `Pattern`, `<x_axis_label>` is the label of the x axis, `<TAB>` is the 'tab' character, `<y_axis_label>` is the label of the y axis, `<x_axis_value N>` and `<y_axis_value N>` are the N'th data points on the graph.

1.5.12 Window Menu

This menu allows you to bring up windows that aren't being shown. Each name in the menu refers to one of the program windows (corresponding to the name in the window title). Click on the name of a closed window to bring it back.

2 Technical Notes

2.1 Obtaining A Slice

2.1.1 What's included in a slice?

When the cutting plane is through the middle of a volume element (the plane moves in increments of half the length of a volume element) then the resulting slice will be one volume element thick. This slice will include all volume elements intersected by the plane as well as all of the membrane elements attached to those volume elements. When the cutting plane is on the border between volume elements **and Surface Concentration is NOT selected**, then the slice will be two volume elements thick and will contain all volume elements bordering the plane as well as all of the membrane elements attached to those volume elements.

2.1.2 What does it mean to have a slice be more than two volume elements thick?

When a slice is more than one volume element thick there are some special considerations that need to be made when converting the slice into the 2D format seen in the Slice Window. Most importantly, there will be a number of pairs of volume elements whose coordinates will differ only with respect to the normal of the cutting plane. These pairs will be lumped together (hereafter referred to as a stack), have their differing 3rd coordinate thrown out and be represented with a single square in the Slice Window. While the effect of this on the various settings is detailed in later sections, the following picture should help illustrate what is meant by stacks.

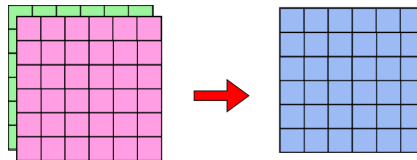


Figure 9: Cutting Plane Between Volume Elements

The left side is a view of some of the volume elements from two adjacent cells (one cell in pink and the other in green), with the cutting plane lying parallel to and on the border of the green and pink cell sections. Note that, since the plane lies on the border between volume elements, each point in the resulting slice (seen to the right of the arrow) will contain data sampled from two volume elements. Each set of two volume elements would constitute a separate stack.

2.2 Slice Selection Window Options and their Impact in the Slice Window

2.2.1 Surface Concentrations

When the Surface Concentration box is checked, the value used to represent the intensity of the selected species will be a 2D concentration in $\frac{molecules}{\mu m^2}$. The concentration, C , in an arbitrary volume element is defined as follows:

$$C = \frac{\sum_{j=1}^6 MA_j}{\sum_{j=1}^6 SA_j} \quad (1)$$

MA_j is the molecular amount in the j th membrane element

SA_j is the surface area of the j th membrane element

If Exclude Orthogonal Membrane Elements is checked, then the only membrane elements which contribute to the concentration are those parallel to the cutting plane.

2.2.2 Volume Concentrations

When the Volume Concentrations box is checked, the value used to represent the intensity of the selected species will be a 3D concentration in mol/ltr . The concentration, C , is defined as follows:

$$C = \frac{\sum_{i=1}^n ((\sum_{j=1}^6 MA_{ij}) + MA_i)}{\sum_{i=1}^n V_i} \quad (2)$$

n is the number of volume elements in the local stack

MA_{ij} is the molecular amount in membrane element j of volume element i

MA_i is the molecular amount in the volume of volume element i

V_i is the volume of volume element i

Since it is possible to be both intracellular and membrane bound, this option will sum over both the volume of a volume element as well as the membrane elements of a volume element. If Exclude Orthogonal Membrane Elements is checked, then only those membrane elements which are parallel to the cutting plane will be counted.

2.2.3 Projections

When the Projection box is checked, the value used to represent the intensity of the selected species will be a molar amount in mol . The amount, A , is defined as follows:

$$A = \sum_{i=1}^n ((\sum_{j=1}^6 MA_{ij}) + MA_i) \quad (3)$$

n is the number of volume elements in the local stack

MA_{ij} is the molecular amount in membrane element j , volume element i

MA_i is the molecular amount in the volume of volume element i

This is calculated in the same way as the volume concentration, except no normalization is made for volume.

2.3 Data Visualizations Within the Slice Window

2.3.1 Line Profiles

The process for generating a concentration profile from a line differs depending on whether or not all of the intersected volume elements lie on the same axis (i.e. whether they form a vertical/horizontal line).

If your line looks something like this:

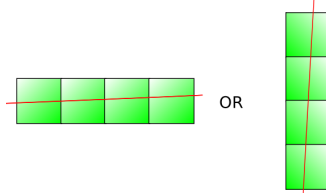


Figure 10: The volume elements are parallel to an axis

then you have the option for giving the line a 'thickness'(with a unit of 'volume elements') and a function type. The thickness determines the number of volume elements that will be sampled in a direction perpendicular to the line, as the following diagram illustrates:

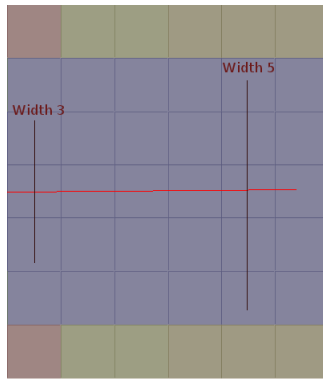


Figure 11: Line thickness 3 and 5

The function can be either 'Box' or 'Gaussian'. If 'Box' is chosen then the concentration is simply an average of the concentrations of the included volume elements (1, 3, or 5). If 'Gaussian' is chosen, and the thickness is either 3 or 5, then the concentrations are weighted using a Gaussian distribution. The function is formed such that the full width at half maximum is equal to the thickness and the total number of points sampled is chosen such that the error, $1 - \sum Weights$, is $< .05$.

Regardless of thickness or function type, the X coordinate of each point represents the distance from the center of each volume element (mapped to the line) to the point first clicked when forming the line. The Y coordinate is the amount/concentration (in the intersected volume element[s]), as calculated above. Adjacent data points are then connected with a linear interpolation. This means that if, for example, the line were to cross over a membrane boundary where the concentration of molecule A was zero on one side and non zero on the other, that the resulting graph would NOT look like a step function. Rather, it would have a sloped line connecting the two points.

If, in contrast, your line looks something like this (the dots and circle are not shown in the actual display, but are used here for clarity):

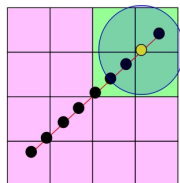


Figure 12: The volume elements are not parallel to an axis

then the graph is formed by creating a number of reference points along the line and sampling nearby volume elements. The reference points (represented as dots, above) start at the beginning of the line and have a distance between them equal to half the length of one side of a volume element. Volume elements whose center is within one side length (the blue shaded circle represents the coverage of the sample area) of a point are sampled (the volume elements highlighted in green are the ones being sampled by the yellow dot) and the final concentration/amount for the point satisfies the following equations:

$$Y = \sum_{i=1}^n w_i * C_i \quad (4)$$

$$\sum_{i=1}^n w_i = 1 \quad (5)$$

$$\forall i \in [1, n], \quad w_i \geq 0 \quad (6)$$

where

n is the number of volume elements being sampled

C_i is one of the n volume elements being sampled

w_i is inversely proportional to the distance from C_i to the sample point

The graph will start at $X = 0$, the first sample point, and will have an associated

concentration/amount as calculated above. The X coordinates is calculated as:

$$X_i = i * SL \quad (7)$$

$$SL \text{ is the length, in } \mu\text{m, of a volume element} \quad (8)$$

$$i \in [0, n] \quad (9)$$

$$n \text{ is the number of sample points} \quad (10)$$

2.3.2 The Color Threshold and Dial

The dial readout in the Slice Window displays a number that, through moving the dial, will run between 0 (purple) and 1(red). This number influences how the species concentrations/amounts are normalized in order to get a color. The normalized concentration/amount, N_i , for an arbitrary volume element, V_i , is

$$N_i = \frac{C_i}{2.0 * DR * MAX} \quad (11)$$

Where:

C_i is the concentration/amount of the selected species in V_i

DR is the value displayed in the dial readout

MAX is the maximum of all C_i

N is clamped to $[0,1]$

From this, we can see that when the dial is in the middle, where $DR = .500$, the deepest red will be seen where $C_i \geq MAX$. If the dial was moved so that $DR = .250$, then red would be seen wherever $C_i \geq \frac{MAX}{2}$.

2.4 Membrane Pores

2.4.1 Membrane Pores in a Simmune simulation

The current version of Simmune contains a simple implementation of membrane pores that can be defined for any membrane (cell cytoplasmic membrane or nuclear membranes) in a cell geometry. The pores have a length and diameter of 40 nanometers. The definition of a pore type in a simulation description requires:

- 1) Specifying the complex that will be transported.
- 2) Specifying whether the pore imports or exports.
- 3) Specifying the surface density of the pores in the membrane.

For importing and exporting, the volume region bounded by a membrane is considered the inner volume. The units for surface density are pores per square micron membrane surface.

2.4.2 Pore Transport

In a simulation, the complexes are assumed to travel through the pores as if they would perform diffusion through an interface channel with the same dimensions (diameter, length), however only in one direction (specified by the import/export setting). Technically, this is equivalent to simulating diffusion into a volume whose concentration for the diffusing species is clamped to 0. For a given concentration C of a complex with diffusion coefficient D allowed to travel through a pore with surface density m in a membrane with total surface S the flux per second through the pores would thus be

$$F = C * m * S * D * S_p / 40nm \quad (12)$$

with the pore surface

$$\begin{aligned} S_p &= 20nm * 20nm * pi. \\ &\text{with } (pi = 3.141527\dots). \end{aligned} \quad (13)$$

Note that the simulator will not check whether the chosen pore density will result in a total pore surface larger than the surface of the membrane containing the pore in order to permit the modeler to experiment with high transport rates. At a pore density of $10000/4 * pi$ (~ 796) pores per square micron the total pore surface is equal to the membrane surface. For quantitative analysis of transport through nuclear membrane pores, see e.g. K. Ribbeck, D. Grlich, "Kinetic analysis of translocation through nuclear pore complexes", The EMBO Journal (2001) 20, 1320 - 1330.

2.5 Cellular Potts Model

We use a cellular potts model (CPM) [1] to simulate dynamic cell morphologies. During each morphology update, which occurs with a user defined frequency, a

subset of the volume elements that are adjacent to the cell's surface are randomly chosen. The energy cost for the extension of the element in a randomly chosen direction and the retraction of the element are evaluated. The change with the lower cost is chosen and will be accepted if the cost is smaller than 0 or with a probability $\exp(-E)$ for a cost E . A volume element of one cell can extend into a volume element occupied by another cell, the total cost will be the sum of the costs for extending one volume element and retracting the other.

Our CPM utilizes four different energy terms. The relative strengths s_x of the terms are tunable as part of a simulation definition. The total cost E is given by

$$E = s_v E_v + s_a E_a + s_b E_b + s_c E_c.$$

The energy terms contributing to the total cost are defined below. Their proportionality constants have been chosen to result in realistic behaviour of the E-cadherin receptor mediated adhesion model for relative strengths close to one.

Volume

The volume term models the effects of an impermeable cell membrane. The potential energy of a cell with volume V and an equilibrium Volume V_0 is proportional to the squared volume difference. The energy cost for osmotic swelling of the cell, given the new V_{new} and old V_{old} cell volumes, is thus

$$\Delta E_v \propto (V_{new} - V_0)^2 - (V_{old} - V_0)^2 = V_e (n+1 - n_0)^2 - (n - n_0)^2 = V_e (2(n - n_0) + 1),$$

where V_e is the volume of a single volume element, n the number of volume elements, and n_0 the number of volume elements at which the cell is in equilibrium.

Area

The area term models energy cost of changes to the cells surface area. The area change ΔA is approximated by change of the area of the surface of the subset of the cubic grid that is covered by the cell

$$\Delta A = A_e \Delta s.$$

with Δs being the change of the number of surface elements and A_e the area of a single surface element. In analogy to the Volume term we model the energy cost for extending the membrane out of its relaxed state by a harmonic potential. The energy cost for changing the morphology is thus

$$\Delta E_a \propto (\Delta A)^2 + 2\Delta A A_e (s - s_0).$$

Binding

The binding term models the energy cost of the breaking of intercellular bonds due to changes of the morphology. The contribution of each type of bond is

proportional to the number of bonds that are broken by the morphology change

$$\Delta E_b \propto \sum_{\square} \sum_{\text{Bond}} A_{\square} C_{\text{Bond}},$$

where the first sum is over all surface elements that are removed and the second sums over all intercellular bonds. The area of a surface elements is given by A_{\square} and the concentration of a bond type by C_{Bond} .

Coherence

The bending resistance of the cell membrane is modeled by a phenomenological term that depends only on the number of direct neighbors of the changing volume element. The relative cost $E_{rel} \propto E_c$ for extending a volume element is given by

Number of Neighbors	1	2	3	4	5
Relative Cost	1	0	-1	-10	-25

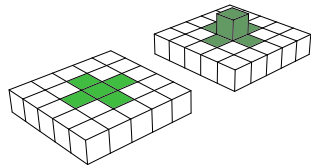
This penalizes the formation of small, thin extensions while it favours closing of small invaginations. The cost terms for retracting a volume element have the opposite cost favouring the retraction of a small protrusion while penalizing the formation of invaginations. The asymmetric nature of the cost terms allow the cell to sample it’s surroundings while keeping the cell’s surface from becoming very rough.

Redistribution of Membrane Molecules

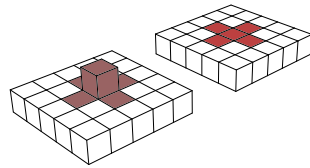
Performing simulations with dynamic morphology raises a question, that has to our knowledge so far not been discussed extensively: How are the concentrations of membrane molecules affected by changes of the morphology? The answer to this question will depend on several factors, the time scales of the biochemical, diffusive and morphological processes as well as which phenomena are actually modeled. The last question is the most delicate to answer. The lipid bilayer strongly resists changes of its area, yet cells can significantly change their apparent surface when migrating. Thus there has to be a process adding lipid bilayer to the apparent cell surface. The details of this process are not well understood [2, 3].

We implement four different algorithms for updating surface concentrations following changes of the membrane morphology. Two of the four algorithms conserve the number of molecules on the membrane by redistributing molecules.

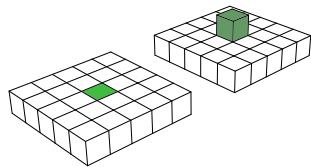
Both redistribute the molecules from a membrane element that is being removed into newly created membrane elements that have a common edge with the neighboring static membrane elements. Membrane elements that are scheduled for removal, which do not have neighbors that are static redistribute their contents into all their neighbors. Newly created membrane elements which do not have neighbors that are static, draw their contents from all neighbors. In each



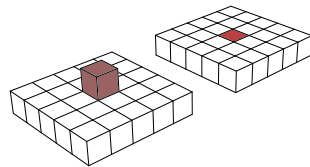
(a) Illustration of the surface elements used for the redistribution of membrane molecules when extending a volume element with the scheme conserving the number of molecules, using the neighbors of changing elements.



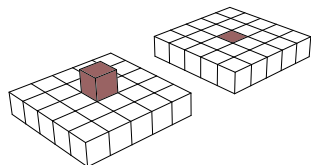
(b) Illustration of the surface elements used for the redistribution of membrane molecules when retracting a volume element with the scheme conserving the number of molecules, using the neighbors of changing elements.



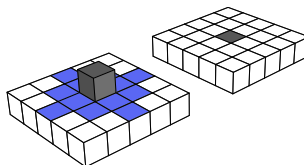
(a) Illustration of the surface elements used for the redistribution of membrane molecules when extending a volume element with the scheme conserving the number of molecules, using only changing elements.



(b) Illustration of the surface elements used for the redistribution of membrane molecules when retracting a volume element with the scheme conserving the number of molecules, using only changing elements.



(a) Illustration of the surface elements used for calculating the concentration average when conserving concentrations.



(b) Illustration of the neighbors of changing surface elements, for which the concentration instead of particle number is conserved, when the concentration conservation is enabled for an extended neighborhood.

case the number of molecules transferred between membrane unequal elements is weighted to conserve mass.

The mass conserving algorithms differ in that one of the two includes the static neighbors as targets for redistribution. This reduces concentration peaks that e.g. would be created by the first conservative algorithm in situations where the number of removed membrane elements is large e.g. 5 and only one or two new membrane elements are created.

For the other two algorithms we consider the cell surface an approximation to the actual membrane, where parts of the membrane - including surface molecules - might be excluded from the apparent cell surface, or added to the membrane by exocytosis. Instead of keeping the number of molecules constant this algorithm preserves the average concentration between surface elements incident on a volume element that has been added or removed from the cell volume. The difference between the concentration conserving algorithms lies in the way how local changes of the surface area ¹ induced by changes of the geometry in the neighborhood of an element are handled. By default, changes of the area of a surface element would lead to a change of the concentration to conserve mass. However we can attribute these changes to the change of the approximated membrane as well and therefore, instead of the particle number, the concentration is conserved.

References

- [1] J. A. Glazier and F. Graner, *Phys Rev E Stat Phys Plasmas Fluids Relat Interdiscip Topics* 47 (3), 2128 (1993).
- [2] M. B. Hallett, C. J. von Ruhland and Sharon Dewitt, *Nature Reviews Molecular Cell Biology* 9, 662 (August 2008).

¹these changes stem from the approximation of a smooth membrane cf. Supplementary Text 7a

- [3] R. R. Kay, P. Langridge, D. Traynor and O. Hoeller, *Nature Reviews Molecular Cell Biology* 9, 455-463 (June 2008)