Cell GUI V3.09 Tutorial, Part 1: Fields and buttons.

Part 1 of the Tutorial presents general explanations of the Cell GUI program as well as specific descriptions of the fields and buttons of the program interface. This part provides enough information to start using the Cell GUI program, although "Cell GUI V3.09 Tutorial, Part 2: Examples" offers assistance for better understanding of the program and the data it generates.

1. GENERAL EXPLANATIONS

Cell GUI was designed to quantitatively examine relationships between ionic concentrations, membrane potential, and volume of a generic cell. The simulator applies the entered starting conditions and produces calculations within a series of subsequent discrete steps. The results of a simulation are written to a tab delimited text file and also displayed on the simulator interface. Three membrane permeant ions – Na^+, K^+, and Cl^- - are considered, as well as an impermeant neutral osmolyte and an impermeant internal anion with a mean valence that the user chooses. The user defines external and internal concentrations of ions and osmolytes, temperature, cell volume, membrane capacitance, transmembrane ionic conductances and water permeability, the rate of the Na^+/K^+-ATPase and two cation-Cl^- cotransporters – NKCC and KCC. The user also determines the duration of the set of calculations and their frequency (the time step between iterations). The model operates on the principle that the cell is in a large extracellular volume. Thus, in general, external concentrations remain as set at the start of a simulation by the user, while intracellular concentrations can change depending on driving forces and conductances. Extracellular concentrations can change, however, if the user allows the buildup of an extracellular molecule (see below).

2. THE INTERFACE

The interface serves a dual input-output purpose. *First,* it is a place where the user enters initial parameters of the system. *Second*, it is the place where the final results of the calculations are displayed. Most of the fields in the interface are active, i.e. can be used by user for inputs, but there are also informational fields for displaying certain parameters and properties tightly associated with active inputs. Some parameters (temperature, cell membrane capacitance, ionic conductances, and transporters rates) remain the same during calculations; some others (ionic concentrations, membrane potential, cell volume, and transporters activities) may change, and in most cases are expected to change. Data displayed on the interface are truncated to a few digits, since their purpose is to present a general picture of the system's current conditions. More accurate results giving the parameter values over time are written to a text file generated by the program, making it simple to export the data to various graphing and analytical suites. The interface allows the user to control the flow of the calculation (time step, number of iterations) and chose the time scale in the data file. Scientific XeY notation is used for very large and very small numbers; for instance 1e-10 (or 1E-10) means 10^{-10}.

Note: The program is not protected from erroneous inputs; it is the user's responsibility to avoid, for instance, an input of a negative concentration or too large a negative buildup that will lead to a negative concentration.

2.1 The upper left and central portions of the GUI: **Concentrations**

Outside K+ Field (active): User defined K^+ concentration outside of the cell, in mM/L; can be defined as 0 in calculations concerning only one cation

Outside Na+ Field (active): User defined Na^+ concentration outside of the cell, in mM/L; a new value of external Na^+ concentration could appear at the end of calculations, if the simulation included an Na^+ buildup; must be more than 0

Outside Cl- Field (informational): The Cl^- concentration outside the cell, in mM/L. This field is calculated by the simulator from the user defined outside K^+ and Na^+ concentrations to maintain electroneutrality, i.e. $[Cl^-]_o = [K^+]_o + [Na^+]_o$. A new value of external Cl^- concentration could appear at the end of calculations, if Na^+ buildup was set up, because it is automatically associated with equal Cl^- buildup

Outside Neutral Osmolyte Field (active): User defined concentration of the external impermeable electrically neutral osmolyte, in mM/L; could be changed due to osmolyte buildup

Outside Buildup Na+ Field (active): User defined rate of changes in external Na^+ concentration, in mM/s. Both increase and decrease of external Na^+ concentration is permitted

Outside Buildup Cl- Field (informational): The rate of change in external Cl^- concentration, in mM/s; always equal to Na^+ buildup

Outside Buildup Neutral Osmolyte Field (active): User defined rate of change in external concentration of neutral osmolyte, in mM/s

Inside K+ Field (active): User defined K^+ concentration inside the cell, in mM/L; a new value of external K^+ concentration could appear at the end of calculations as result of the system searching for equilibrium; can be defined as 0, but only if external K^+ concentration is 0

Inside Na+ Field (active): User defined Na^+ concentration inside the cell in mM/L; a new value of external Na^+ concentration could appear at the end of calculations; must be more than 0

Inside Cl- Field (informational): The Cl^- concentration inside the cell in mM/L. This field is calculated by the simulator using the user defined inside concentrations of K^+, Na^+, and the impermeable intracellular anion (An^-) as well as mean valence of An^- (z) to maintain electroneutrality, i.e. $[Cl^-]_{in} = [K^+]_{in} + [Na^+]_{in} - [An^-]_{in}*z$. The new value resulting from calculations may not be exactly in accordance with the equation partly due to truncation of displayed values and partly because of small deviation from electroneutrality when $Em \neq 0$

Charged Osmolyte Field (active): User defined intracellular membrane-impermeant anion (An^-) concentration inside the cell, in mM/L; the value could change if the cell volume changed and/or if there was a buildup of An^-

Mean Valence Field (active): User defined mean valence of the An^-, should be entered as a positive number, since the program interprets it as an average number of negative charges per one An^- molecule; remains unchanged during calculations

Inside Buildup Na+ Field (informational): The rate of change in internal Na^+ concentration associated with the buildup of An^-, in mM/s. Equal to the buildup of An^- multiplied by the mean valence of An^- (z)

Inside Buildup Neutral Osmolyte Field (active): User defined rate of change in internal concentration of neutral osmolyte, in mM/s

Inside Buildup Charged Osmolyte Field (active): User defined rate of change in intracellular membrane-impermeant anion (An^-) concentration of neutral osmolyte, in mM/s

Adjust Conc Button: Adjusts the user defined inside concentrations in order to establish balanced osmolarity between the inside and outside of the cell ($[Cl^-]_{in} + [A^-]_{in} + [K^+]_{in} + [Na^+]_{in} + [osm]_{in} = [Cl^-]_o + [K^+]_o + [Na^+]_o + [osm]_o$) while maintaining user defined inside concentration ratios.

Note: More than one adjustment may be needed, but more than 3 subsequent adjustments have no practical effect; additional adjustment has no more effect at all when the message “No adjustment necessary” appears

2.2. The upper right portion of the GUI: **General features**

Em (mV) Field (active): User can define initial membrane potential of the cell, in mV; the field can be used as an input, but is generally used to display Em at the end of calculations

Volume (L) Field (active): User defined initial volume of the cell, in liters; the value may be changed as a result of calculations

Temp (Celsius) Field (active): Allows the user to define the temperature at which the simulation is to take place in degrees Celsius

RT/zF per dec (mV) Field (informational): Nernstian coefficient used in calculations, in mV (for log10); associated with the temperature

Capacitance (F) Field (active): User defined cell membrane capacitance, in Farads

2.3. The middle part of the GUI: **Defaults Buttons**

Save as Defaults Button: Saves current user defined or recently calculated values on the upper part of the interface (concentrations, buildups, membrane potential, and cell volume) as the defaults

Restore to Default Button: Restores saved default values to the respective fields on the upper part of the interface; also resets the Spent ATP, Voltage by the Pump, and Current Time to 0.

2.4. The lower left portion of the GUI: **Conductances**

K+ Conductance (ions/(s*V)) Field (active): User defined conductance for K^+ ions across the cell membrane, in ions per volt per second; can be defined as 0 and this is true for other conductances

Na+ Conductance (ions/(s*V)) Field (active): User defined conductance for Na^+ ions across the cell membrane, in ions per volt per second

Cl- Conductance (ions/(s*V)) Field (active): User defined conductance for Cl^- ions across the cell membrane, in ions per volt per second

R Input (MOhm) Field (informational): The total resistance of the cell membrane, in MΩ. Calculated from the user defined conductances for K^+, Na^+, and Cl^-

Volume τ (s) Field (active): User defined membrane water permeability, for convenience presented as a time constant of volume changes, associated with osmotically demanded movement of water. A large time constant implies a low water permeability, and vice versa. The

value of 1e8 and larger (i.e. longer than 3 years) is interpreted in the program as no water permeability at all.

2.5. The lower central portion of the GUI: **Transporters**

Rate of the Na/K-ATPase (cycles/s) Field (active): User defined maximum number of cycles that the all pumps in the cell can perform in a second (assuming that $[Na^+]_{in} \rightarrow \infty$)

Spent ATP (1/s) Field (informational): The amount of ATP used by the Na^+/K^+-ATPase; calculated from the pump rate and the current inside Na^+ concentration; accordingly, could be changed if inside Na^+ concentration was changed

Number per cycle… K, Na Fields (active): User defined number of K^+ and Na^+ ions carried in one cycle by the Na^+/K^+-ATPase

Voltage by the Pump (mV) Field (informational): Transmembrane voltage generated by Na^+/K^+-ATPase; calculated from pump activity (i.e. Spent ATP which in turn depends on rate of the pump and internal Na^+ concentration), difference in transfer of Na^+ and K^+ per cycle, and the transmembrane resistance to cations

Rate of the Na,K,2Cl-Cotrans (cycles/s) Field (active): User defined standard number of cycles the $Na^+,K^+,2Cl^-$-cotransporter performs in a second (assuming that $([K^+]_o*[Na^+]_o*[Cl^-]_o*[Cl^-]_o) / ([K^+]_{in}*[Na^+]_{in}*[Cl^-]_{in}*[Cl^-]_{in}) = 10$)

Rate of the K,Cl-Cotrans (cycles/s) Field (active): User defined standard number of cycles the K^+,Cl^--cotransporter performs in a second (assuming that $([K^+]_o*[Cl^-]_o) / ([K^+]_{in}*[Cl^-]_{in}) = 10$)

2.6. The lower right portion of the GUI: **Calculation setting**

Time Step (sec) Field (active): User defined length of each step within the calculation

Number of Iterations Field (active): User defined number of consecutive repetitions of the calculations, each time using results of the previous time step as an input

Duration of Calculation Field (informational): Shows the total duration of a calculation, in seconds if Detailed Log is chosen, and in minutes if General Log is chosen. Calculated from the user defined time step length, number of iterations and whether a Detailed or General Log has been chosen

Detailed Log Radio Button: Having this button selected while running a simulation performs a single set of calculations with defined number of iterations and writes the results from every iteration to the data file

General Log (100 lines) Radio Button: Having this button selected while running a simulation performs 100 sets of calculations, each with defined number of iterations, writing only the end result of each set to a file

Current Time Field (informational): Shows the current time within the simulation that can combine several calculations

Reset Time Button: Resets the simulation time to 0

2.7. The bottom portion of the GUI

Output File Name Field: User defined file name that data will be written to. There are no restrictions for the file name and its extensions. Warning: The program will overwrite existing file upon calculation if the name is not changed before the calculation.

Calculate Button: Runs a simulation with user defined values and parameters, writes the results to a file and display on the interface

3. RUNNING THE SIMULATION

To run the simulation, set all parameters and properties and click "Calculate." Cell GUI will generate a data file containing intracellular values as a function of time in the same directory that it resides and update changed values in windows of the interface. Note that if invalid values are defined (such as too long time step), the simulation may still run and the file may still be generated, but results will also be invalid.