[] General

Manual of

P R O T O N . E X E

(c) Heribert Cypionka, Univ. Oldenburg

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PROTON.EXE is a portable freeware app that simulates
metabolic processes in order to explain pH effects one might
find in cell suspensions. This includes uptake and reduction
of an electron acceptor, oxidation of an electron donor, and
the electron transport-coupled proton translocation. At the
same time changes of the membrane potential, ΔpH and ATP
conservation are simulated.

[] Installation

Download the newest version (proton.zip) from

www.microbial-world.com/freewarelist.htm

There is no installation required, just unzip the file
'proton.exe'. Proton does not change any system parameters.

[] Decimal separator

For correct processing of floating point numbers it is
recommended to set the point as decimal separator under 'Regional
Settings'.

[] Removal

There is no deinstallation required, just delete the file
'proton.exe'.

[] Freeware

Proton is a test program. It is freeware that you use on
your own risk.

The author does not guarantee that it is working correctly and
will not take any responsibility for any damage users might
experience by using it.

[] Open a file

Click on File -> Open and select a Proton parameter File
(*.H+). Only files following exactly the structure decribed
below will be read correctly.

[] File format of H+ files

The parameters are saved in a text file that can also be
loaded as CSV file with Excel or compatible programs. The
semicolon ";" is used as separation character.

The data format is:

Headline 1: Copyright
Headline 2: Experiment description (= headline above data
table)
Next 61 lines: Parameter number (01 ... 61): Parameter name
; Parameter value

Data: Copy of the calculated data panel shown on the right
side

File Example:

H+ Proton Parameter and Data File, (c)1986-2021 Heribert
Cypionka
Description: Demo
 1: Cell suspension OD ; 10.0000
 2: Cells per ml (OD=1) [*10^8] ; 3.2800
 3: Volume/cell [*10^-15 l] ; 0.3870
...
61: Active Channels (Code 1+2+4+8) ; 15.0
(Order, name and units of listed data:)
Time (s); pH (mV); Acc.(out); Acc.(red); ATP cons.; MP(mV);
pHi; pmf (mV)
(Calculated data:)
  0;-9,30;990,00;0,00;0,00;-130;7,70;-162
  2;-9,30;970,00;0,00;0,00;-130;7,70;-162
...
(Number of lines depends on the steps that have been
calculated accoto 'Step' and 'Stop after (sec)')

[] Save Data

Lets you select a folder and file name (*.H+) to save your
parameters in the format described above. Instead of '.H+'
you could also name your file with '.txt' or '.csv'. The
files can be read by text editors or spread sheet programs

(such as Microsoft Excel).

[] Exit Program

Quits without checking whether data are saved.

[] Options

- Allows for omitting symbols and grid in the graphical
plot.

- Allows for plotting membrane potential and pH(i) instead
of electron acceptor and reduced acceptor

[] Cell volume and surface

If one assumes the cell to be a rod with rounded ends one
can describe it as a cylinder with two hemispherical caps at
the ends. Then the following parameters to calulate cell
volume and surface can be applied:

Assume: Cell length   len = 1.5 µm
        Cell width   wid = 0.5 µm
                      pi = 3.14

Radius   r = wid/2 = 0.5 µm/2 = 0.25 µm
Circular area   a = pi r^2 = 3.14 * 0.25^2 = 0.196
µm^2
Circumference   cir := 2 pi * r = 2* 3.14 * 0.25 =
1.57 µm

Cylinder height   cylh = len -2r =1.5- 2*0.25 = 1 µm
Cylinder mantle area cyla = cir * cylh = 1.57 µm^2
Cylinder volume   cylvol = cylh * a = 1*0.196 =0.196
µm^3

The two caps add to a sphere, therefore:
Cap area   capa = 4 * pi * r^2 = 4* 3.14 * 0.25^2
= 0.785 µm^2
Cap volume   capvol = (4/3) * pi * r^3 = 1.333*
3.14 * 0.25^3 = 0.785 = 0,0654 µm^3

For the complete cell follows:

Cell volume   cellvol = cylvol + capvol = 0.196 +
0.065 = 0.261 µm^3
Cell surface   cella = cyla + capa = 1.57 + 0.785
µm^2
                            = 0.785 * 10^-8 cm^2

[] Membrane capacity

Based on Hodgkin & Nakajima (1972) J. Physiol. 221:121-136

Constant: 1 µF/cm^2 = 1.036*10^-11 mol or 6.24*10^12 e-
per V and cm^2

F = 96.5 C/mol = 96 500
1 farad = 1 coulomb / 1 volt
1 coulomb = 1 A * s

[] Calculate data

- proton.exe calculates results once per second. If 'Step'
is set to values >1 sec, the calculations are unchanged,
but only part of the results are shown.

- The amounts added are assumed to be nmol. This corresponds
to µmol per Liter or µM for a reaction volume of 1 ml.

- Transport and biochemical reactions rates (v) are
described by a maximum rate (vmax, nmol per sec), a Km value
(concentration at half maximum rate) and the current
substrate concentration [S] according to

$$v = \frac{vmax * [S]}{[S] + Km}$$

For intracellular processes, Km and [S] refer to the
cytoplasmic volume.

- Responses of the pH electrode and the ATPase rate (both
1/sec)
are assumed to follow first-order kinetics.

- For the pH electrode the values depend on the difference
to the final (real) mV signal according to:

displayed signal = current signal + R*(final value - current
signal)

 with R = Response of the pH electrode per sec (0...1)

The current ATPase activity is assumed to be proportional to
the
difference from the initial transmembrane pH gradient (t=0,
which
is assumed to be in equilibrium) according to:

H+ uptake = A . (∆pH − ∆pH0)

with A = ATPase H+ uptake rate per sec (0...1)
and ∆pH resp. ∆pH0 being the current or initial proton
gradient. (The membrane potential is not considered as it
is assumed to be neutralised chemically.)

- Some additional data (electron acceptor and donor amount,
ATP conserved, membrane potential and the cytoplasmic pHi)
are calculated as well.

[] Graphics

There is a simple graphical plot (with adjustable scales)
showing the simulated pH curve and (optionally) changes of
the electron acceptor, reduced acceptor and ATP
concentrations. Instead of the oxidised and reduced electron
acceptor concentrations, display of membrane potential and
∆pH can be selected under 'Options'. For a more
sophisticated display please save the simulation as H+ (or csv)
file and read data with a spreadsheet program.

[] Tutorial

Several tutorial videos are available under
www.microbial-world.com/proton.htm

[] Finally

Please be patient when playing with proton.exe. Chemiosmotic
processes are not always easy to understand, but they are
among the the most common processes in every living cell. It
took me more than 30 years to design the current simulation.
Just contact me, if you have questions or ideas how to
improve it.

Good luck and enjoy!

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www.microbial-world.com