

Plate Analyzer Quick Tutorial

J. Paul Robinson, Purdue University Cytometry Labs

Quick facts:

Plate Analyzer - Copyright Purdue University Cytometry Lab (MULTICORE #4) 1.8 x64 licenced to Licensed to J. Paul Robinson

C:\Data files\DEMO\Data for Bindley presentation\Plates\20100615J-AM1J-p0058b (152624)
 C:\Data files\DEMO\pap files\32 drugs with names for 58.pap

Callouts and functions:

- Load data icon
- History box see below
- Undo actions BUT it undoes all
- Load logic map (PAP)
- Debarcode CyTOF data
- Name of logic map used
- # of cells analyzed in this file
- Click this and then go to options, data and select the percentage of cells to load from the file - default checked is 10% (goes up in 10s)
- Create a new logic map (protocol)
- Check web for updates
- Save Logic map
- Data file being analyzed

History window content:

```

C:\software\Plate-An\workspace\project.pap
C:\Data Res\Biodemiler\data\Parameters\Biodemiler - 1 stauraspine-reverse.pap
C:\Data Res\Special data - Nuclei compiled plates\10 Compounds for GenoSo JC1\Barringer GFP.pap
C:\Data Res\Special data - Nuclei compiled plates\10 Compounds for GenoSo JC2\simple project with drug names.pap
C:\Data Res\DEMO Data for Bindley presentation\Plates\20100615-AM1J-p0058b.pap
C:\Data Res\DEMO Data for Bindley presentation\Plates\20100617-AM1J-p0058a.pap
C:\Data Res\DEMO Data for Bindley presentation\Plates\58 only for manual gating.pap
C:\Data Res\Germany-Dr. Heering\Parameters\Heeringer.pap
C:\Data Res\Special data - Nuclei compiled plates\10 Compounds for GenoSo JC1\96 384 well plate.pap
C:\Data Res\Special data - Nuclei compiled plates\10 Compounds for GenoSo JC1\132 drugs with names new.pap
C:\Data Res\Special data - Nuclei compiled plates\10 Compounds for GenoSo JC1\132 drugs with names.pap
C:\Data Res\Biodemiler\data\Parameters\Biodemiler - 1 stauraspine-reverse.pap
C:\Data Res\Biodemiler\data\Parameters\Biodemiler - 2 stauraspine-reverse.pap
C:\Data Res\Biodemiler\data\Parameters\Biodemiler map\Biodemiler stauraspine.pap
C:\Data Res\Biodemiler\data\Parameters\Biodemiler - 1 stauraspine-reverse.pap
C:\Data Res\Peter Koutzk\Param Res\Res\32-4.pap
C:\Data Res\Peter Koutzk\Param Res\Res\32-2.mn
C:\Data Res\DEMO Data for Bindley presentation\Plates\20100615-AM1J-p0058b
C:\Data Res\Param data\Biodemiler\1
C:\Data Res\Biodemiler\data\stauraspine-fcs-1\1_un gated
C:\Data Res\Germany-Dr. Heering\Analysis\Plate_Heeringer_Analysis_Plate_Heeringer
C:\Data Res\Biodemiler\data\Sunamb_cyt_2
C:\Data Res\BNI1-GFP - modified data set
C:\Data Res\Special data - Nuclei compiled plates\10 Compounds for GenoSo JC1
C:\Data Res\Peter Koutzk\Full data set
C:\Data Res\BNI1_GFP_P1-BNI1_GFP_P1_Analysis-Plate 0001
C:\Data Res\Witas\Tuhini
C:\Data Res\Peter Koutzk\print_illustrations_files
C:\Data Res\Peter Koutzk\Peter_data\renamed Res
C:\Data Res\Peter Koutzk\Peter_data\small 3 row data set
    
```

New Parameter

DNA(Ir193)D / DNA(Ir193)D =

Max Value: 10
Min Value: 1

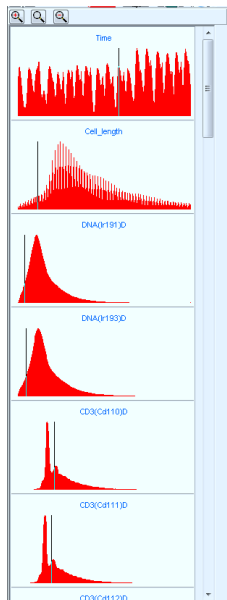
Buttons: Ok, Cancel

Create new parameter: You can click this and get the box on the left. Now you can select any parameter in the list mode file, and add, subtract, divide or multiply with any other parameter creating a new parameter (think ratios)...and make sure you add point 1 or 1 to the "Mn" value since you don't want to divide by zero!

Toolbar icons: GATE, Inclusive, Exclusive, Either not, neither, CTRL, Create a drug dose response curve, Distance, Create a graph from anything (link it to a drug container etc)

Callouts:

- Click this icon before clicking on the graphic area you want to gate
- Inclusive
- Exclusive
- Either not
- neither
- This is used to identify the positive control well or wells (will average) or statistical analysis modes
- Create a drug dose response curve
- Create a drug container which will hold all your dose response curves
- Create a distance measure based on selections in OPTIONS
- Create a graph from anything (link it to a drug container etc)



The + and - change the scale of the histograms

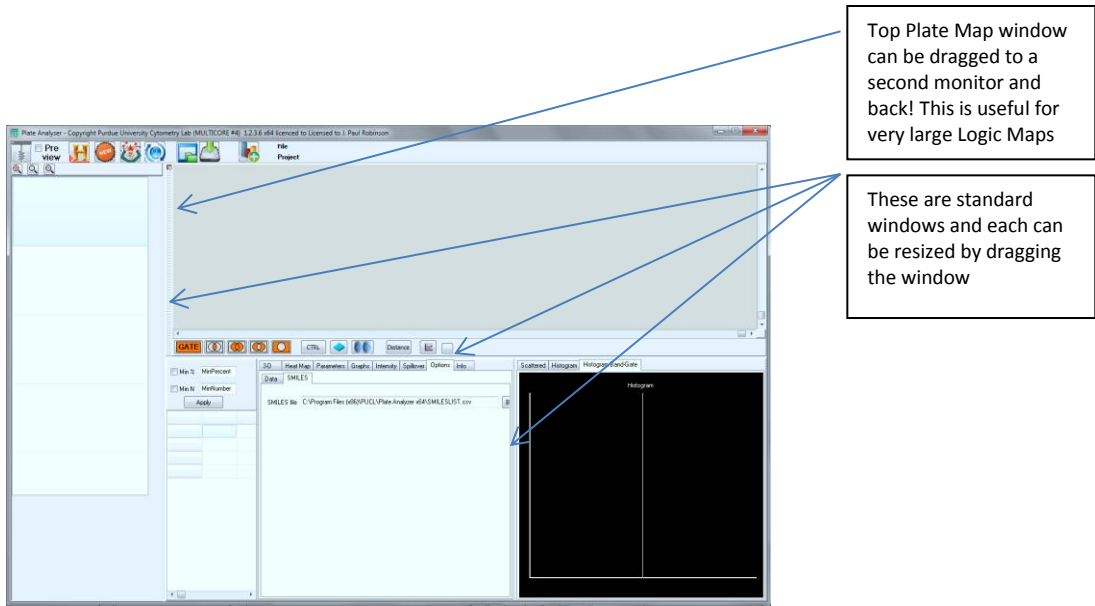
The slider moves the histograms up or down
The + and -

The cursor is active when the PRISM button is checked (see to the right)

The PRISM box shows all the parameters collected in the list mode file and there are 3 options, ++ (take the positive side of the cursor (from the left box), -- take the negative side of the cursor, and == none

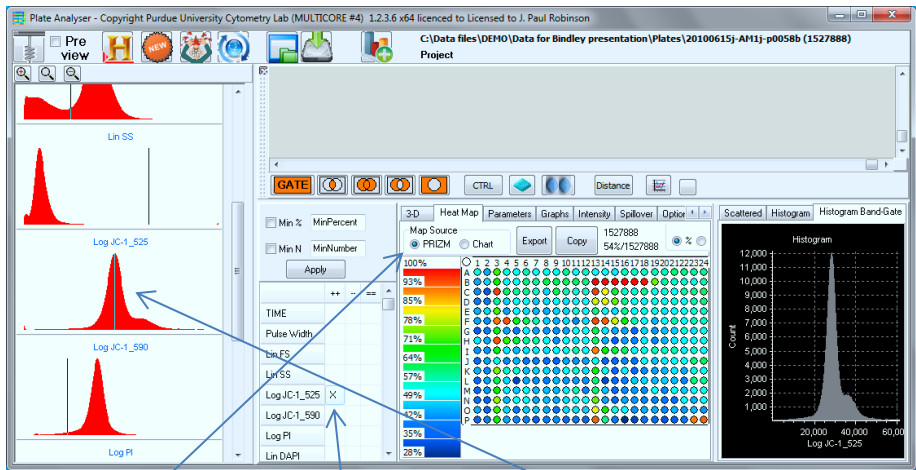
| | ++ | -- | == |
|--------------|----|----|----|
| Time | | | |
| Cell_length | | | |
| DNA(Ir191)D | | | |
| DNA(Ir193)D | | | |
| CD3(Cd110)D | | | |
| CD3(Cd111)D | | | |
| CD3(Cd112)D | | | |
| CD3(Cd113)D | | | |
| CD3(Cd114)D | | | |
| CD45(Ir115)D | | | |

Options: Also has mode to read SMILES code from a CSV file. You can load any file (it must be a specific format see later).



Functions

Creation of Drug containers

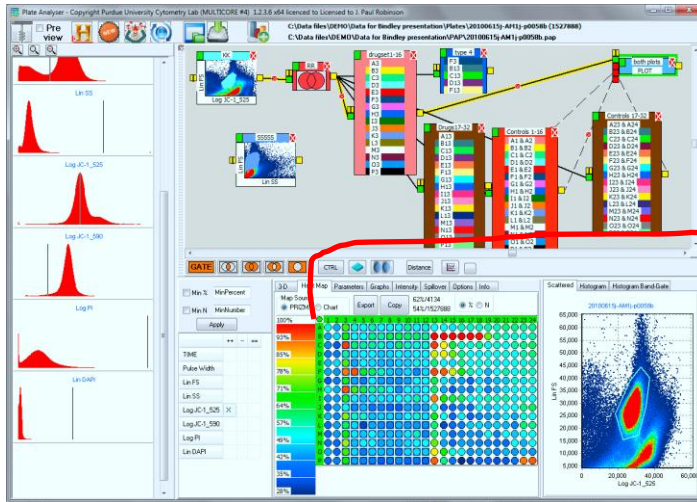


STEP 1: Click the **PRISM** Button

STEP 2: Click on one of the prism parameters such as ++ (positive) signal or (--) negative signal.

STEP 3: Move the cursor for the parameter you just selected (here Log JC1-525). As you move the cursor, the heat map will respond

Create Gates:



STEP 1: Identify a plot type you want to gate. Click **GATE** then draw your gate on the plot

STEP 3: Once you have the gate drawn, a new ICON pops up in the logic map – you can right click the title on this ICON and rename it.

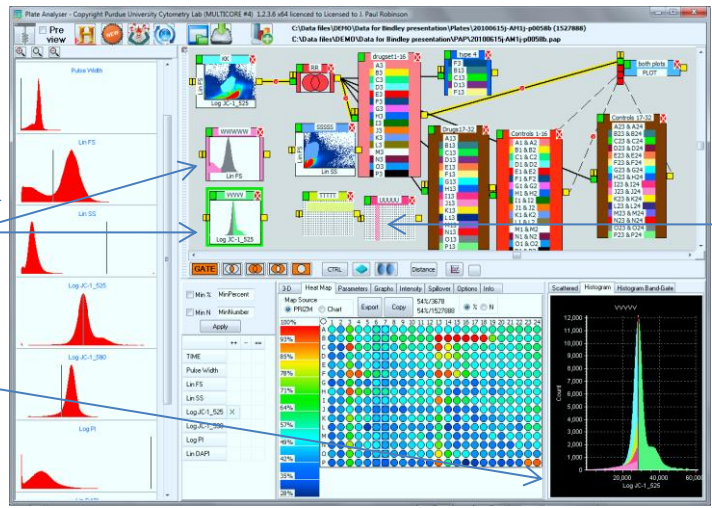
STEP 2: To draw the gate, first select a place on the plot you want the gate to start, move the left mouse, move the mouse, click the left button and do this until you are near the end, and right button will close the gate.

THE SECRET BUTTON: If you want to see the result of a single well, or multiple wells, you must click the small white button (here) and the top and side bars become GREEN. Whenever the green bars are there, you can view any single or multiple wells

More Gates

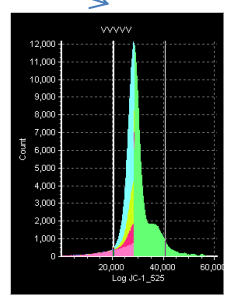
STEP 1: Select either the right or left side of the left histograms. Click **GATE**, then click the mouse on the side you want. An ICON pops up showing the direction of the positive cells.

STEP 2: whenever you select a gate icon in the logic map, this will appear in the lower right box.



STEP 1: To gate specific wells, just highlight the wells you want to gate (you can use any combination by using windows CTRL or SHFT Keys, and an **ICON** of this selection appears on the Logic Map

STEP 1: Select **HISTOGRAM BAND GATE** and you can set two cursors. If you click the **GATE** button, then inside the two cursors, you will create a band gate .



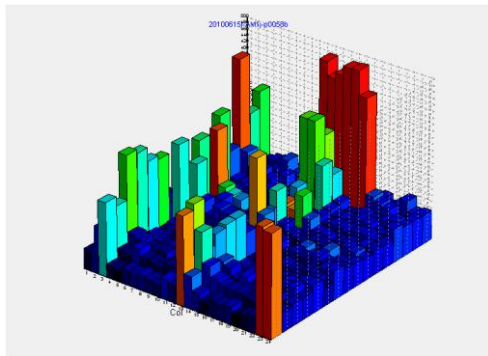
The **SAVE LAYOUT** and **OPEN Layout** are linked to the data files – if you have drug names these will be linked and a small file will be written to the data directory.

Updated Options (Feb 2013)

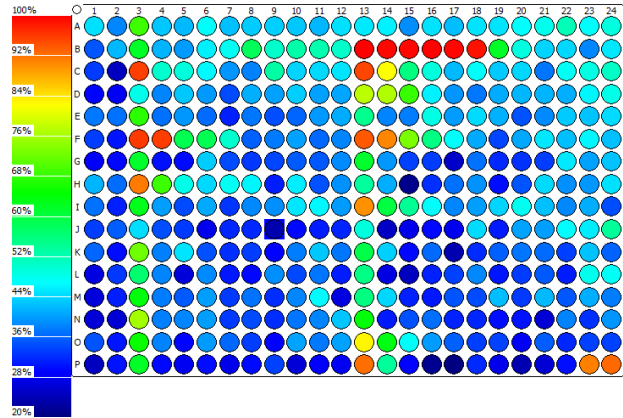


Data Output Opportunities

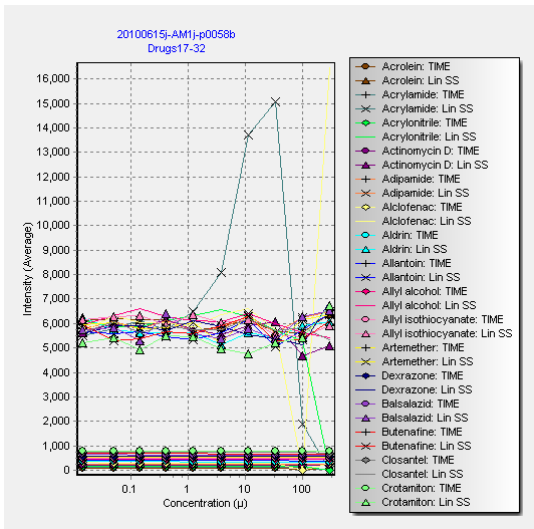
3D output graph



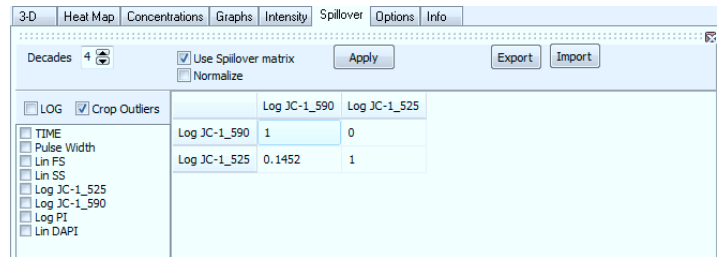
Heat-map type display



Any single parameter across time or concentration

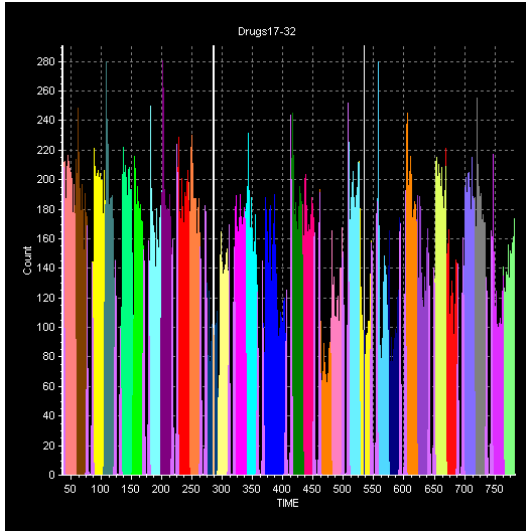


Spillover table data

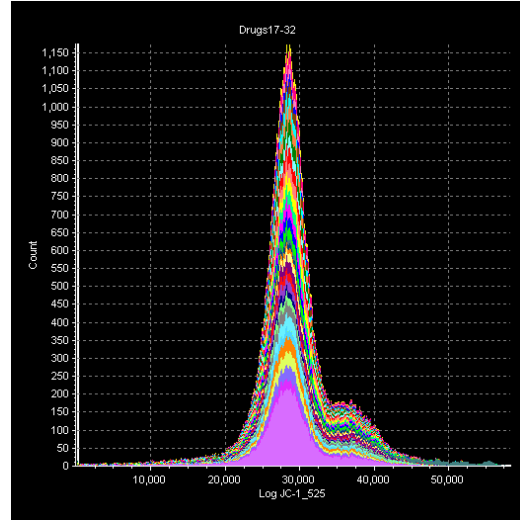


Types of Graphic Output

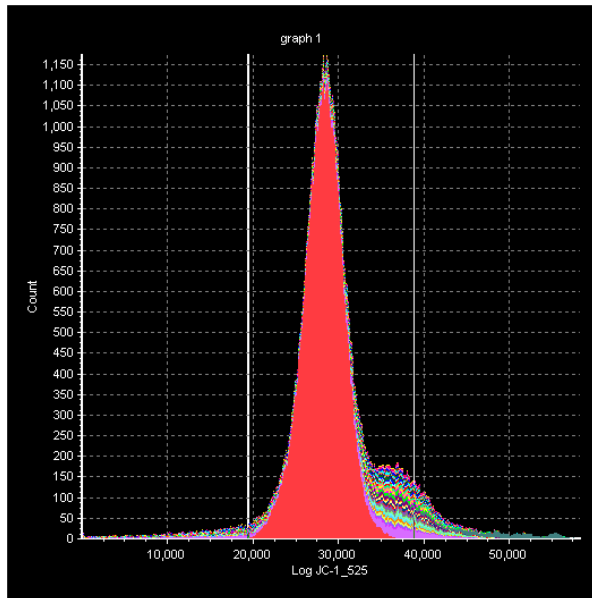
Single Parameter Histogram



Single Parameter Histogram



Gated Histogram (2 cursors gate between)



2 Parameter dotplot

