

SeqGeq's Violin Plot Plugin - How To:

Violin and box plots are popular ways of illustrating expression patterns between genes of interest and across different populations or samples in single-cell RNA sequencing analyses. In SeqGeq these features have been implemented as a plugin with some dependencies in R.

Installation

With regard to the R dependencies, this plugin has been tested in the R environment 3.4.2, and requires the path to R be specified in SeqGeq's Diagnostics preferences, as well as the ggplot2 package within R. This package can be installed in R simply by copy/pasting the following command there:

```
install.packages("ggplot2")
```

As with all plugins for SeqGeq you'll need the JAR file for the Violin plot plugin to be placed in the plugins folder referenced within the Diagnostics preferences of SeqGeq.

Running the Plugin

Once the necessary dependencies have been put into place, users can select a population of interest from the Samples area of SeqGeq's workspace, click on the "Plugins" option within the Workspace tab of SeqGeq's workspace, and choose the Violin Plot plugin there:

The screenshot shows a software interface with a 'Plugins' dropdown menu open. The menu items are: FlowJo Exchange, Add Workspace Plugin, Remove Workspace Plugins, AutoGateCategorical, Clustering_Heatmap, DEGenesPlugin, Export_Population, GeneAnnotation, Monocle, Seurat, Seurat_Alignment, Seurat_Workflow, TopGenesPlugin, VDJ_Explorer, **ViolinBox** (highlighted with a red box), and scImpute. The background interface includes a top navigation bar with 'Analyze', 'Genes', 'Workspace', and 'Edit' tabs. Below the navigation bar are icons for 'Median', 'CV', 'Freq. of', and 'Boolean'. A table with columns 'Count' and 'Description' is visible. On the left, a 'Samples' panel shows a tree view with 'All' and 'Lupu'. At the bottom, a table shows 'Statistic' and '#Cells' for various cell types.

Statistic	#Cells
97_v2.csv	6937
B Cells	344
T Cells	1287
CD4+	311
CD8+	350

This will bring up a dialog in which researchers can select their genes of interest, as well as make adjustments to the violin plot settings:

Violin
 Box

Select parameters (genes) to be plotted

Collections: All

Parameter Sets

All (30177)
UpRegulated CD4 (12 of 12 Valid)

Parameters

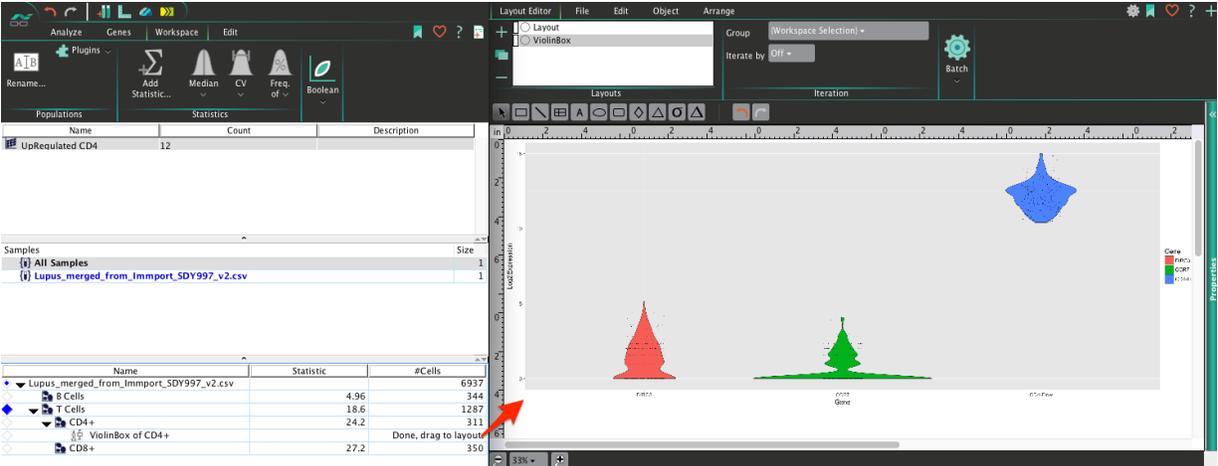
CD4-Flow
BIRC3
CCR7
CD4.y
CD40LG
DGKA

Flip Trim Jitter PDF Background

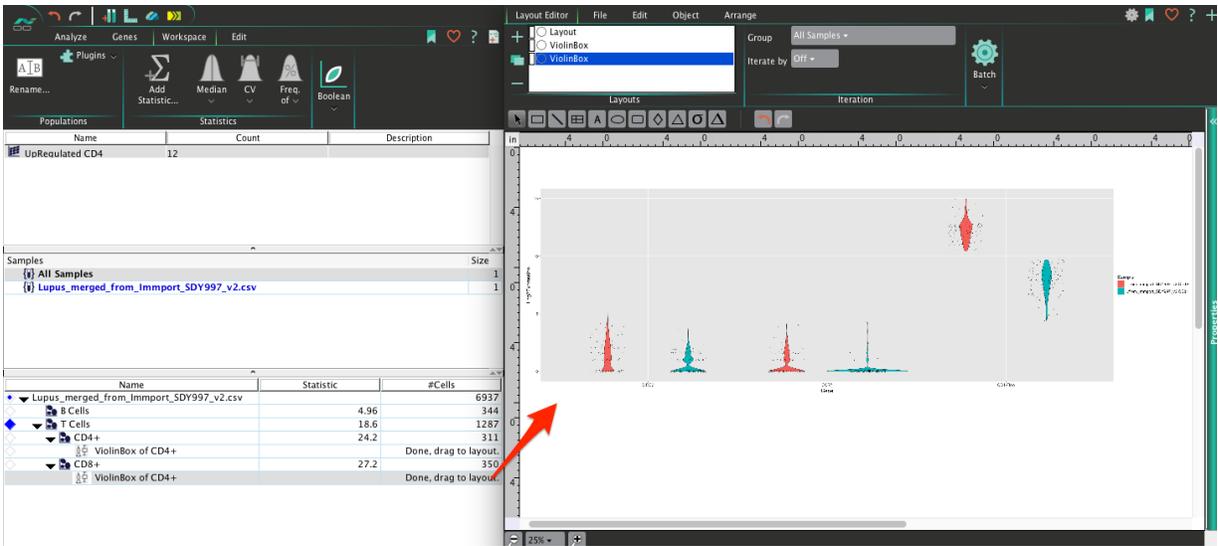
Figure height: 1000 Violin width: 200 Legend width: 200
Max name length: 30 Legend size: 100
Jitter dispersion: 10 Jitter dodge: 90 Jitter size: 100
Legend position: Right

Cancel OK

Clicking OK will calculate the plot selected, add a ViolinBox plugin node to the population selected, and create a new layout within SeqGeq's Layout Editor, in order to see the Violin plot displayed there:



In order to compare genes from this initial Violin plot across numerous populations, simply drag the initial plugin node to the other populations of interest:



Help Resources

SeqGeq documentation is available at: docs.flowjo.com/seqgeq

For additional assistance with all things SeqGeq related, feel free to reach out for technical support: seqgeq@flowjo.com