

How-to-Seurat

Seurat is an extremely popular and powerful pipeline for analyzing single cell RNA Sequencing (scRNA-Seq) data developed and maintained by the Satija lab.(1)

We've implemented this tool as a plugin in SeqGeq (v1.1.0+) in order to make the features there available for our users and simplify the process of producing results from the Seurat pipeline as simple as possible.

After downloading the plugin and placing it in your SeqGeq Plugins directory you still need to perform a few steps to get this tool running on your system:

- a) Connect your R application and SeqGeq Plugins directory to SeqGeq through the Diagnostics section of the preferences there (Seurat was tested using R versions 3.4.1 and 3.4.2):

On Windows -

SeqGeq: Diagnostics



◀ Show All ▶

Settings

Open Engine Interaction Window

Total Memory = 367.5 MB
Max Memory = 1799.5 MB
Free Memory = 192.4 MB

Local Website:

Local Contact:

Write default values to local preferences file

Watch Sample Cache Threshold

Miscellaneous

Select Equivalent Parameters

Action Delay (in ms)

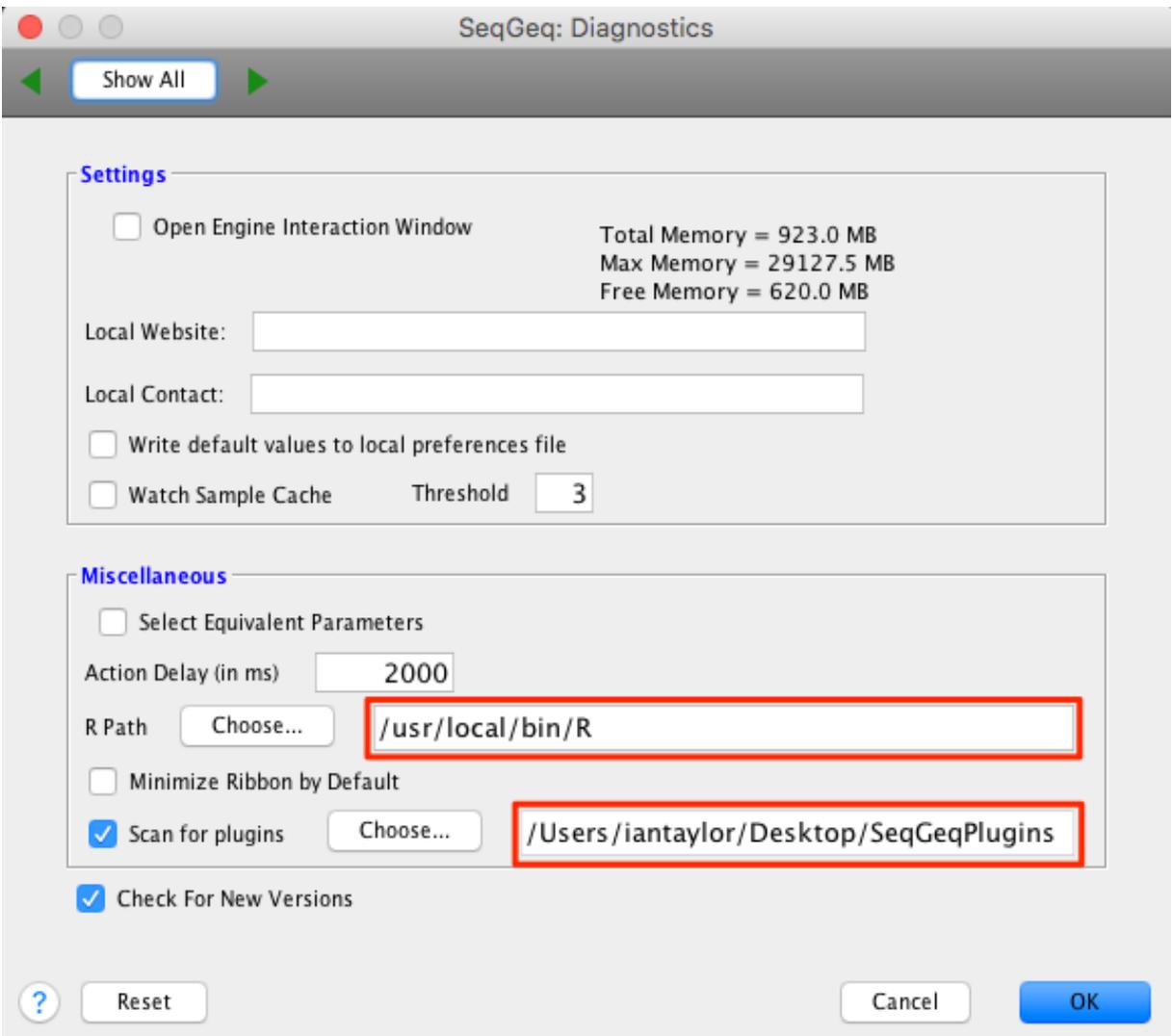
R Path

Minimize Ribbon by Default

Scan for plugins

Check For New Versions

On Mac -

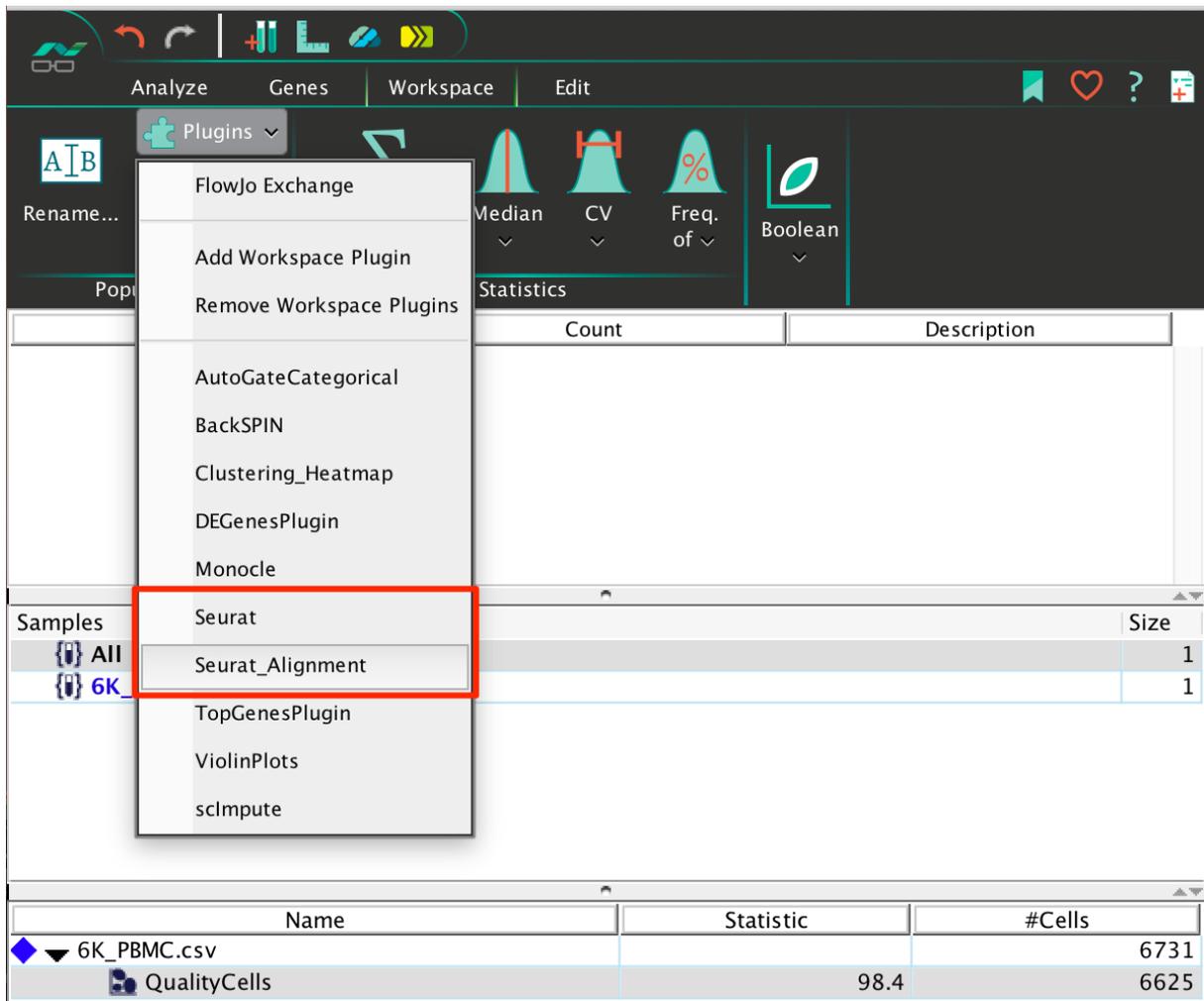


b) Instal the Seurat R packages using the following commands there:

```
install.packages('Seurat')  
library(Seurat)  
install.packages('tidyverse')  
install.packages('Impute')  
install.packages('ape')  
install.packages('lubridate')
```

c) Restart SeqGeq.

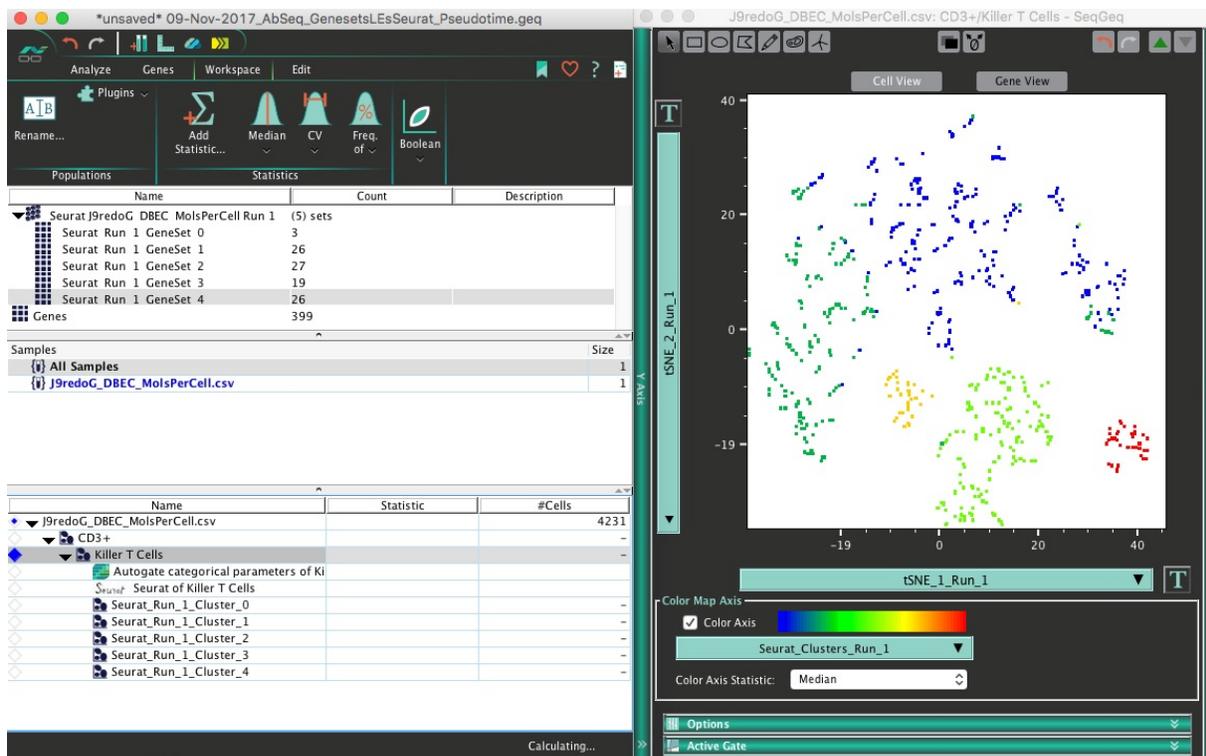
Once the plugin is correctly connected to SeqGeq and dependencies made available in R, you can run either avenues of the plugin by selecting a population on which you'd like the Seurat plugin to analyze, going to the Workspace tab, and selecting "Seurat_Workflow" or "Seurat_Alignment" within the Plugins there:



After choosing the plugin avenue you'll be prompted to enter some options. Mousing over the various fields there will bring up tool tips describing what the various options change, with regard to the analysis.

The Seurat Alignment option will allow you to align disparate samples, such that their parameters will be normalized to one another in copies of the expression matrices. This will make samples from different collections more amenable to concatenation.

The Seurat Workflow option will perform QC, normalize data for dimensionality reduction, perform PCA guided tSNE mapping in R. After the Seurat Workflow option of the plugin runs, it will generate a pair of tSNE parameters within your data, as well as a set of graph based clusters from a K Nearest Neighbors algorithm (and corresponding categorical parameter), and gene sets from differentially expression for each cluster:



If you have any questions or concerns regarding this or any other plugin for SeqGeq, we would welcome your feedback:
seqgeq@flowjo.com

References:

1. Seurat is developed and maintained by the Satija lab, and is released under the GNU Public License (GPL 3.0)