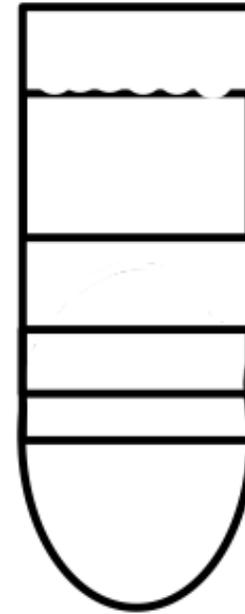




# How to Lex

A Sample De-Multiplexing Plugin for SeqGeq



# Why Would I Want This Tool?

So you've generated multiplexed sample data (many samples sequenced in a single tube simultaneously with sample oligo-tags).

*Cool!*

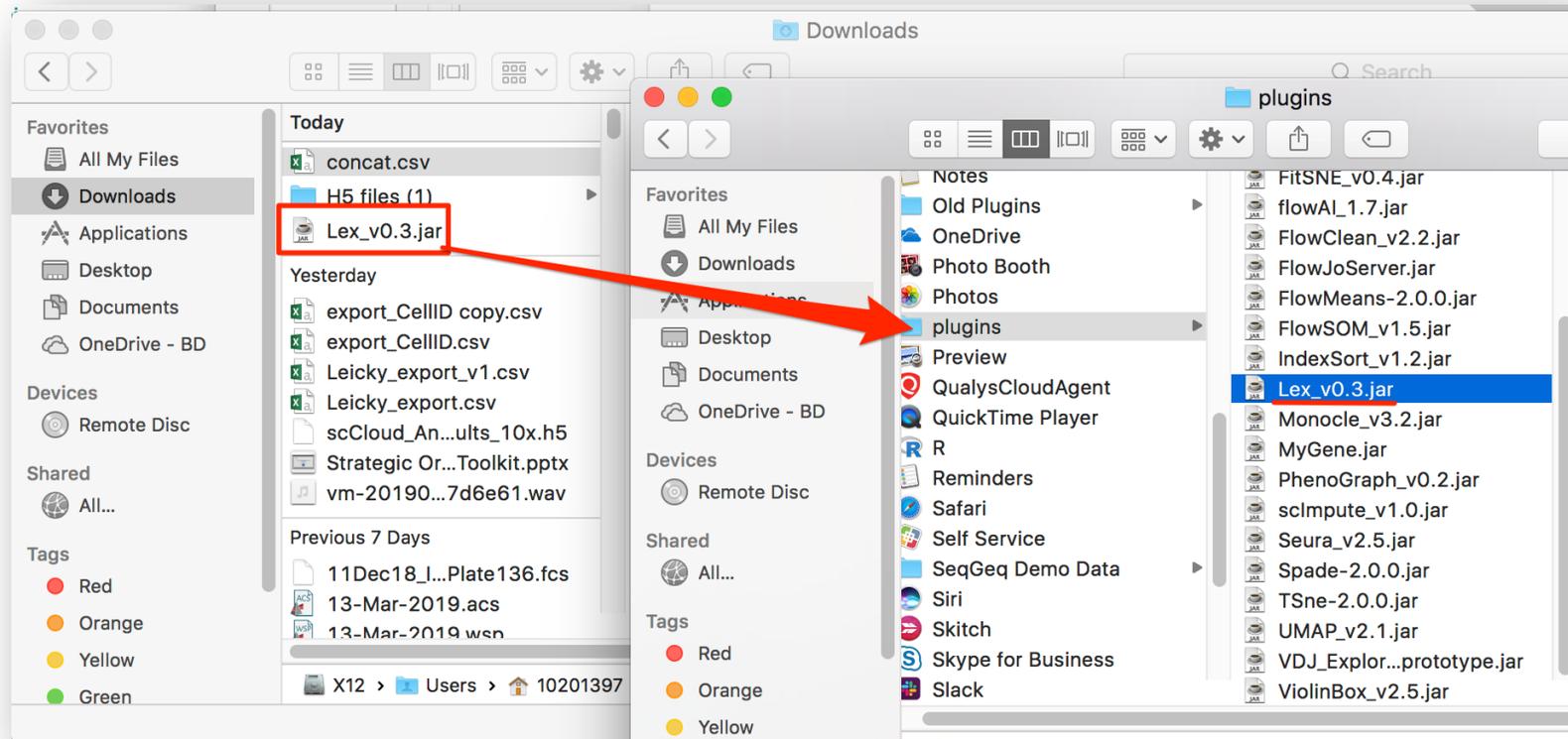
Now you just need to pull those samples apart within downstream analysis.

Whether your data is combined by using **Cell Hashing**, or **BD™'s Sample Multiplexing Kit**, or you've simply concatenated your samples together in SeqGeq - Lex is here to help!

\* Lex now also able to "de-concatenate" combined samples



# Step 1a - Install Lex within your plugins folder, simply by dragging and dropping the plugin JAR into that directory



## Step 1b – Restart SeqGeq



# Step 2 - Load your data into SeqGeq just as you normally would and select the multiplexed data file

The screenshot shows the SeqGeq software interface. The top menu bar includes 'Analyze', 'Genes', 'Workspace', and 'Edit'. Below the menu is a toolbar with various analysis tools. The main workspace is divided into two panes: 'Navigate' on the left and 'Discovery' on the right. The 'Samples' table is visible, showing the following data:

Name	Count	Description
Samples		
All Samples	2	
Combined_SampleMultiplexDemo_RSEC_MolsPerCell.csv	1	
Example_CellHashing.csv	1	

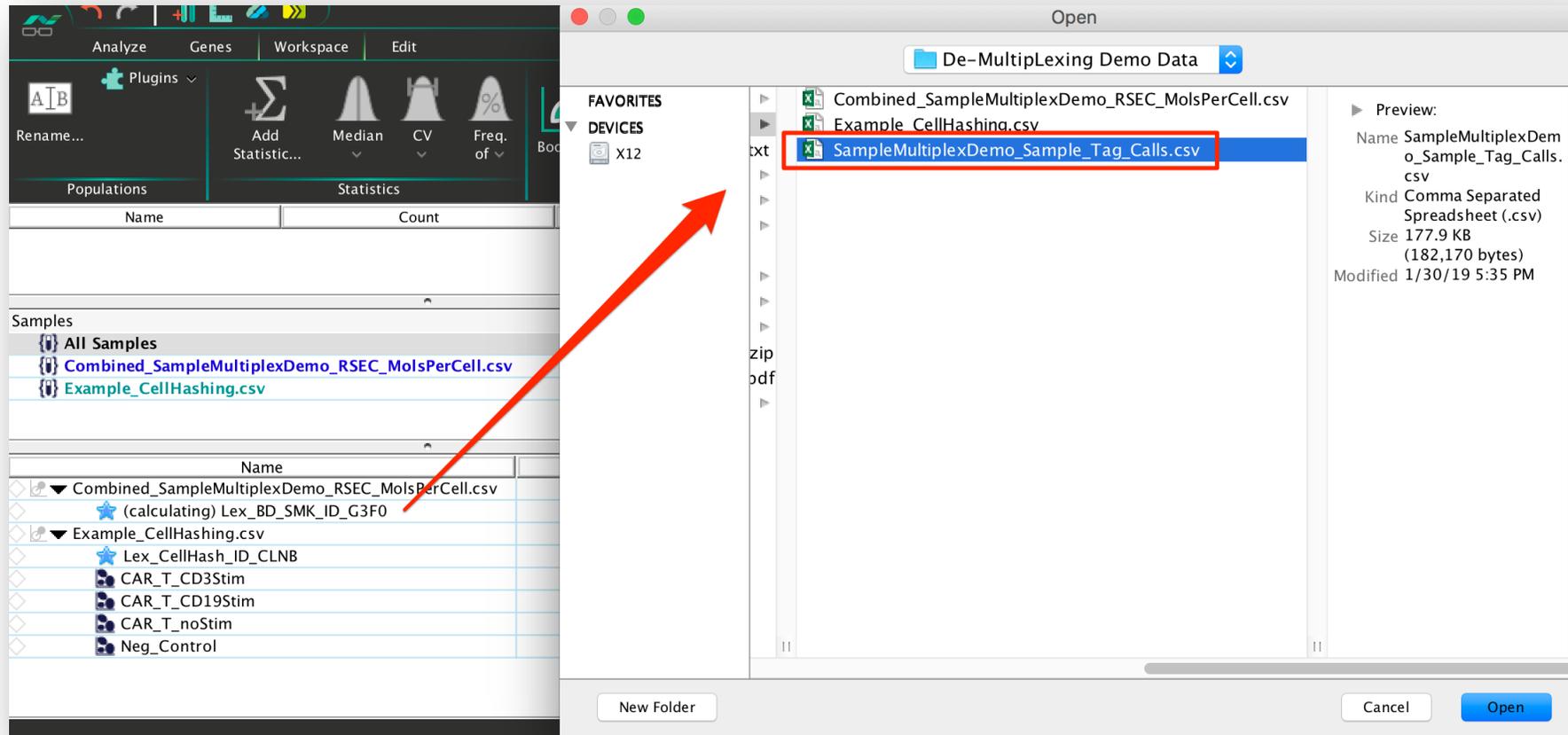
Name	Statistic	#Cells
Combined_SampleMultiplexDemo_RSEC_MolsPerCell.csv		5568
Example_CellHashing.csv		11922







# If you're using BDs SMK you'll need to select the associated Sample Tag Calls CSV file



Once distinguished you can now analyze or otherwise treat “sample subpopulations” in the data matrix just like any other population within the SeqGeq workspace

The screenshot displays the SeqGeq software interface. The top menu bar includes 'Analyze', 'Genes', 'Workspace', and 'Edit'. Below the menu, there are icons for 'Rename...', 'Add Statistic...', 'Median', 'CV', 'Freq. of', and 'Boolean'. The main workspace is divided into two sections: 'Populations' and 'Statistics'.

**Populations Table:**

Name	Count	Description
All Samples	2	
Combined_SampleMultiplexDemo_RSEC_MolsPerCell.csv	1	
Example_CellHashing.csv	1	

**Statistics Table:**

Name	Statistic	#Cells
Combined_SampleMultiplexDemo_RSEC_MolsPerCell.csv		5568
Lex_BD_SMK_ID_G3F0		
Multiplet	3.70	206
PBMCDonor1	39.4	2193
PBMCDonor2	56.7	3158
Undetermined	0.20	11
Example_CellHashing.csv		11922
Lex_CellHash_ID_CLNB		
CAR_T_CD3Stim	58.0	6910
CAR_T_CD19Stim	15.3	1829
CAR_T_noStim	9.59	1143
Neg_Control	16.8	1999



# A Note on DeConcatenating Samples

The DeConcatenation process *only works on concatenated samples*, for the time being.

As a result, if you want to concatenate and then deConcatenate subsets, you'll first need to export them as separate individual samples, then concatenate, in order to effectively use the Lex option to nicely re-annotate the combined populations automatically.



# Resources

- Demo Videos of Lex in Action:

[tinyurl.com/Lex-DeMultiplex](https://tinyurl.com/Lex-DeMultiplex)

[tinyurl.com/Lex-DeConcat](https://tinyurl.com/Lex-DeConcat)

- Documentation:

[docs.flowjo.com/seqgeq](https://docs.flowjo.com/seqgeq)

- Support:

[seqgeq@bd.com](mailto:seqgeq@bd.com)



# Thank You!

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