**Installing the Embedding Add-in:**

**The Idiot-Proof Guide**

1. This is a set of instructions to install the “Embedding” add-in in JMP (MUST be JMP Pro 15 or higher), which gives easy access to useful dimensionality reduction algorithms like t-SNE and UMAP, as well as Wanderlust, Leiden, and t-Space. If you haven't already, the first step is to install R version 4.0 (or the latest version) and Rstudio. Once these are installed, in the R console enter this command: install.packages("Rtsne"). At the y/n prompt, answer "n". Then, enter the command: install.packages("umap"). At the y/n prompt, answer "y". You now have UMAP and tSNE packages installed, that will later be called from JMP. Make sure you have some version of MATLAB installed and operational as well. For Matlab, you will also need to install the Bioinformatics and Parallel Computing toolbox add-ons. Also make sure you have the Xcode application installed if you are using a mac.
2. Next, we will add UMAP/tSNE capability to JMP. Download the attached file called “UMAP\_RtSNE050820ecb0511.jmpaddin”. Double-click the"UMAP\_RtSNE050820ecb0511.jmpaddin" icon, and click install. Graphical user interface, text, application

   Description automatically generated . With JMP open, click "view" at the top of the screen and go down to "Add-ins". Graphical user interface, application, Word

   Description automatically generatedClick on this, and you should see that UMAP\_RtSNE is installed. Within, click on the blue link after "Home Folder" to get to your personal "com.jmp.umaptsne1" folder within your hidden Addins directory: Graphical user interface, application

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3. Next, unzip and open the "com.jmp-3.zip " file attached to this email. Search for a subdirectory within titled "scripts" which should contain 3 .jsl files:Graphical user interface, text, application

   Description automatically generated Drag “scripts” into your personal "com.jmp.umaptsne1" folder, which should then look like this:Table

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4. There should be a file within "scripts" titled "JSL\_Utils.jsl". Still working within your personal "com.jmp.umaptsne1" folder, drag this file out of "scripts".Table

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5. Finally, download the attached file titled “embedding\_noremovedup050820ecb.jsl” and use it to replace the file currently in your personal“com.jmp.umaptsne1” folder with the same name:Text

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6. At this point, we now have the base package operational with UMAP and t-SNE capabilities, which we will see in the drop-down add-ins menu in JMP. To begin adding the rest of the functionality, we must install a number of dependencies. First, install the R packages “igraph”, “ggplot2”, “devtools”, “remotes”, “assertthat”, and “RANN” using the lines of code: install.packages(“packageName”) (change the directional quotes to nondirectional) followed immediately by library(packageName) for each of these packages.
7. Once this is done, we will install several packages through Bioconductor. Still in R, run this code: Text

   Description automatically generated. Follow this immediately with library(“BiocManager”).
8. Use the line of code: BiocManager::install(“packageName”) for the packages “DESeq2”, “fgsea”,”openxlsx”,”edgeR”, and “DelayedMatrixStats”. One final package, “seqGlue”, can be installed with the line: remotes::install\_github(“scfurl/seqGlue”). If these cannot be done, see step 10. Keep in mind that Bioconductor is not compatible with the Apple M1 version of R 4.1.0. Instead, try the Intel version. Also, if you choose to install a new version of R, you must redownload all R packages.
9. For the leiden algorithm to run, we need a package “leidenbase”, which is installed with the R line: devtools::install\_github(‘cole-trapnell-lab/leidenbase’).
10. THIS STEP ONLY IF LEIDENBASE DOWNLOAD AND/OR BIOCONDUCTOR PACKAGES DOWNLOAD FAILED: If you see an error that compilation of the package has failed, follow this link: <https://github.com/cole-trapnell-lab/leidenbase> . Once there, scroll down to the section “Tested Conditions”. Find the section pertaining to your computer operating system, which should contain links to downloads of the updated compilers necessary. Once these are updated using the provided user-friendly installers, run the same line of code as in the previous step, and this should now work.

* The following set of steps was done on Mac. First, click on the link for the FORTRAN compiler, shown here:Graphical user interface, text, application, email

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* Then, scroll to the section of your latest OS, and under Assets, download and open the attached files (one or two, depending on your version of macOS). In my case(macOS 11), the two packages were gfortran-ARM…pkg and gfortran-ARM….tar.xz. The .pkg file is a user-friendly installer that will walk you through the steps (you may need to right click to open the .pkg file successfully). The tar.xz file, once opened, creates a directory in your computer to store the FORTRAN compiler information.Graphical user interface, application

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1. In addition, there are 2 folders and 1 matlab .m file you need to drag into your computer’s MATLAB directory, under “Documents” in your Finder if you are using Mac, titled “cyt3-mac”,“tSpace\_Lab”, and “tspace\_pipe\_og.m”, respectively. They are attached in this email.
2. For the wanderlust and tSpace algorithms, which interface with MATLAB, there is an additional necessary script that I’ve attached here, called “MATLABfuncs\_install-3.R”. To get this done, simply open this file in RStudio and run it from top to bottom.
3. At this point, everything should be up and running and you should now have a host of algorithms available for your use, including UMAP, tSNE, wanderlust, tSpace, and leiden. NOTE: this set of instructions has been tested with Mac, and should also work for PC, but this has not yet been tested. Here’s a screenshot of the interface, which allows you to select rows and columns (Parameters) to run the algorithms on.Graphical user interface, application

   Description automatically generated**Congratulations, you’re done!!!**

\*NOTE: the FindMarkers addin is not complete yet, and so within the GUI you can ignore this.