**First time user:**

*If you have previously run a python program, you can proceed to the next section. Note: all instructions are written for implementation on a Windows Computer*

Without a user interface, we run Python scripts through the command prompt. You might also hear the command prompt referred to as a “terminal”, as this is the name Apple uses. Your computer has a command prompt built in, you can simple type “command prompt” in your search bar, and you will see the option appear. However, using a different command prompt, named Anaconda, is a nice feature because it can work separately from your computer’s processing. This means you can install programs and make changes that will not be permanent to your computer.

1. The first thing to do is install Anaconda (<https://www.anaconda.com/products/distribution>). Anaconda comes built with python, which is a nice feature. Simply select “download” and follow the on-screen prompts

Graphical user interface, text, application, chat or text message

Description automatically generated

1. Once installed, you can access the Anaconda command prompt by searching from the task bar:

Graphical user interface, application

Description automatically generated

1. Open the anaconda prompt:

Shape, rectangle

Description automatically generated

1. Navigate to the folder where the program is stored. For example, if the path to my folder is C:\Users\lawashburn\documents\LiP-MS:

I see that I am already in the folder C:\Users\lawashburn (this is what is written on the screen when you first open anaconda)

I can change the directory to documents by typing:

cd documents

Press enter

And then I can change the directory again to the LiP-MS folder by typing:

cd LiP-MS

It should look like this:

Text

Description automatically generated

1. From here we can jump into step 1 of the first set of instructions by typing:

python LiP-MS\_v3pt1.py

***General anaconda/command prompt notes:***

* To rerun the last command, simple press the up button and the last command will appear, and press enter to run

**Using the program:**

1. Open up a terminal, navigate to the folder where the program is housed, and type:

python LiP-MS\_v3pt1.py

1. The first step will prompt you to input your working directory. This is the path to a folder where you would like all results exported to.

*\*When entering .csv file paths, make sure there are no quotation marks surrounding the path*

1. Input path to proteingroup.csv and peptidegroup.csv files

*\*Example files are packaged with the distribution for reference*

1. A message will appear that will let you know that the first stage of processing is complete
2. Open these files from your working directory
3. Add a column entitled “p value”
4. In the first row of that column, add this formula:

=TTEST(A2:C2,E2:G2,2,2)

1. Double click the bottom right corner of the box to apply that formula to all the cells in that column
2. Click “find and replace”. Select to search for “#DIV/0!” and leave the replace field empty. Click replace all.
3. Save, ensuring file is still saved as .csv

*\*An example file is packaged with the distribution as “MCIvCtrl\_nofilter\_updated.csv” for reference*

1. In terminal, again navigate to folder housing the program, and type:

python LiP-MS\_v3pt2.py

1. Follow onscreen prompts as before
2. All output data will appear in the working directory specified

**Troubleshooting:**

***Converting to .csv format:***

The easiest way to accomplish this is to open the file of interest in Excel. From there go to:

File > Save As > CSV (Comma delimited) (\*.csv)

Please note, .csv files can only be one sheet. If you have multiple sheets in your Excel file, all but the first sheet will be lost when converting file types.

***Getting file path:***

(Windows) In File Explorer, highlight the file or folder of interest. In the top “home” toolbar, click copy path. The path is now copied to the clipboard.

Note: when adding path to terminal prompt, make sure no quotations around path

***Error message:***

An error message will occur if the files you are trying to access are open. Close all necessary files prior to running.

You will also likely get an error message if you do not follow the template formatting. Please see all sample data in the supplied folder so the templates match.

***File names:***

You will see some errors if you do not follow proper file naming convention:

* No spaces or special characters in any file or folder name: you can use “\_” in place of a space
* Files/folder names cannot begin with a number, must begin with a letter