IEomics Manual: For users using Thermo Q-Exactive

Step 1: Install Necessary Programs

* R, version 3.1.0 or higher < <http://cran.rstudio.com/>>
* MSConvert (ProteoWizard) <<http://proteowizard.sourceforge.net/downloads.shtml>>

Step 2: File Conversion in MSConvert

* MS2 for R script
  + Upload .raw
  + Peak Picking: Level: 2-2
  + msLevel: 2-2
  + threshold: absolute .0001 most intense
  + Output: MS2

Step 3: Run the script

* Open the code ([date]\_IEomics.R) and after looking at the notes on the top of the script enter: Ctrl+Shift+S (or Ctrl+A and run) from R-Studio, or open in a text editor and paste into R console.
* A number of pop-up boxes will appear, read the dialogue box and enter the correct information. After the final pop-up box is entered a file will appear in the directory in which you imported the .ms2 files from. Note that pop-up boxes may appear behind all open windows, so minimize all open windows if pop-up boxes are not obvious.

Step 4: Import exclusion list into the Thermo Methods File

* Import the generated exclusion list into the thermo methods file
* Change the exclusion tolerance to 100 ppm (recommended) or the exclusion tolerance desired
* Save the method file under a new name, and make sure to modify the sequence by adding the new method file
* Reinject the sample with the new ddMS2-topN Method with the imported exclusion list
* Repeat from Step 2.