Processing and export of Sciex 6500+ MS direct infusion MRM data

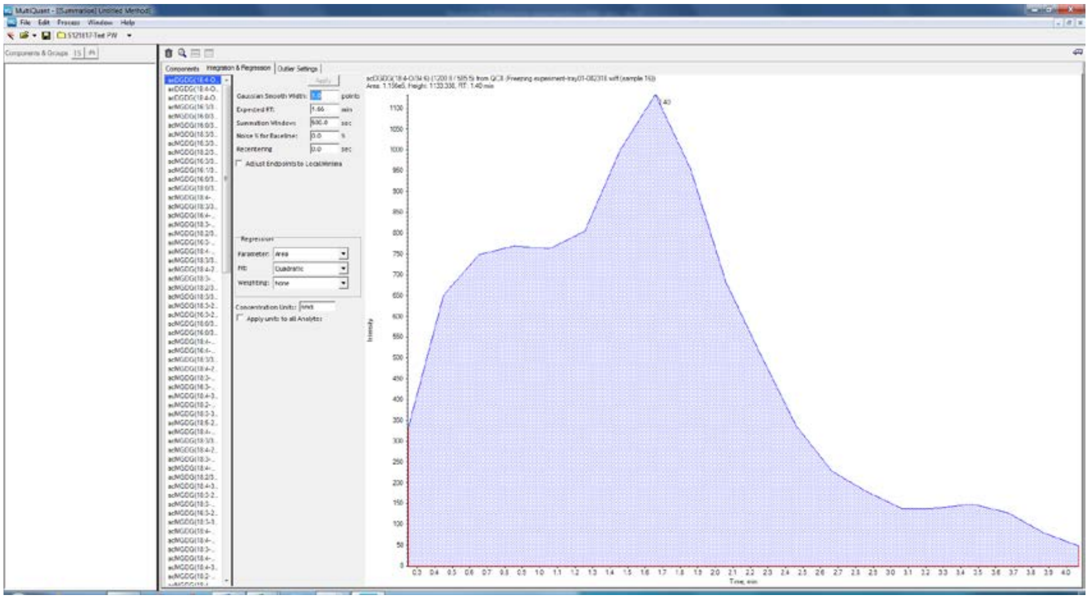
by Yu Song, Kansas Lipidomics Research Center, Kansas State University

A. Establish quantitation method

(1) Open MultiQuant → File → New Quantitation Method → Select Sample The sample you choose should be one that is likely to contain all the analytes of interest.

(2) Review component information → Set Integration & Regression parameters (adjust parameters for each compound or set default values for all compounds)

Set integration defaults: Edit → User Integration Defaults → For direct infusion data in 6500+, choose <u>Summation</u> integration algorithm, and choose the retention time(s) and summation window(s) so that the entire time that you would like to sum is covered in all samples, as indicated by shading in the intensity vs time profile.



Expected RT: 1.66 min

Summation Window: 600.0 sec

Noise % for Baseline: 0.0 %

Recentering 0.0 sec

☐ Adjust Endpoints to Local Minima

Regression

Parameter: Area

Fit: Quadratic

Weighting: None

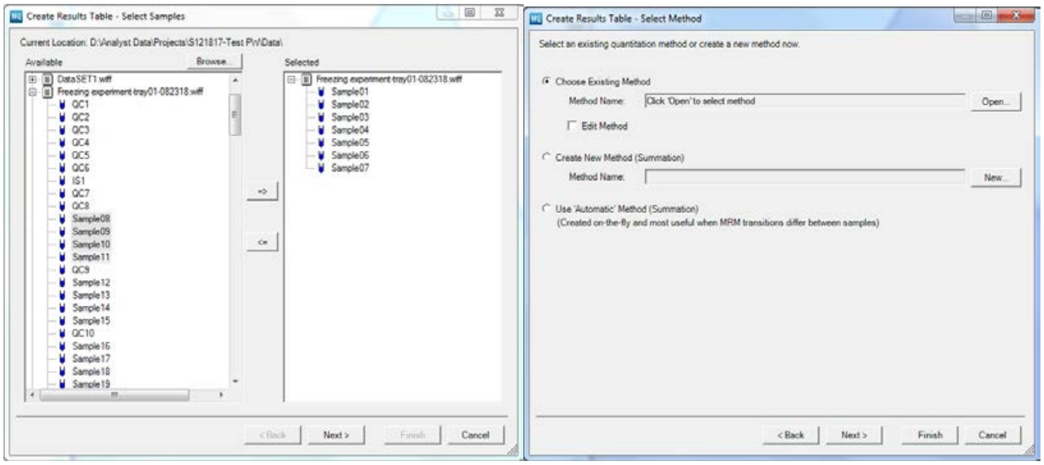
☐ Apply units to all Analytes

Area: 1.156e5, Height: 1133.338, RT: 1.40 min

(3) Save the quantitation method as “name.qmethod” file (continued on next page).

B. Data processing

(1) File → New Results Table → Select (all) Samples → Next → Select an existing method (the established quantitation method) → Finish


Create Results Table - Select Samples

Current Location: D:\Analyst Data\Projects\S121817-Test PW\Data\

Available:
- DataSET1.wiff
- Freezing experiment-tray01-082318.wiff
  - QC1
  - QC2
  - QC3
  - QC4
  - QC5
  - QC6
  - IS1
  - QC7
  - QC8
  - Sample08
  - Sample09
  - Sample10
  - Sample11
  - QC9
  - Sample12
  - Sample13
  - Sample14
  - Sample15
  - QC10
  - Sample16
  - Sample17
  - Sample18
  - Sample19

Selected:
- Freezing experiment-tray01-082318.wiff
  - Sample01
  - Sample02
  - Sample03
  - Sample04
  - Sample05
  - Sample06
  - Sample07

Create Results Table - Select Method

Select an existing quantitation method or create a new method now.

● Choose Existing Method

Method Name: Click 'Open' to select method

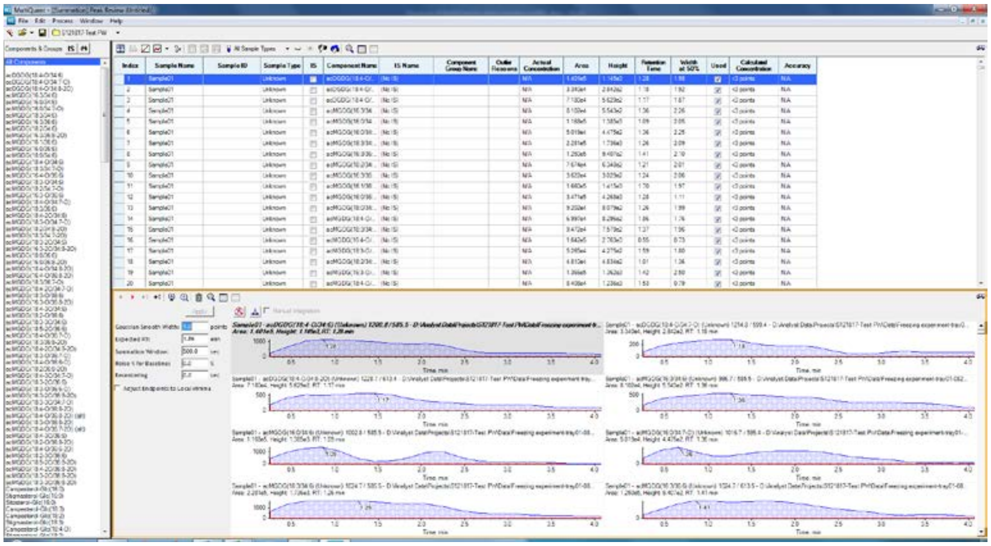
☐ Edit Method

○ Create New Method (Summation)

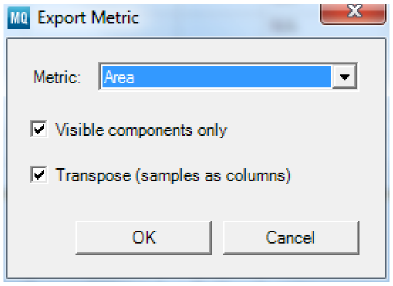
Method Name:

○ Use 'Automatic' Method (Summation)
(Created on-the-fly and most useful when MRM transitions differ between samples)

(2) Review compound list, check results table, check peak integration to make sure all peaks are integrated, i.e., are shaded.



(3) File → Export → Result Table-Metric→ select "Area", select "Transpose" → generate a text file that can be opened in Excel (by choosing to view all files).


Export Metric

Metric: Area

☑ Visible components only

☑ Transpose (samples as columns)

(4) Verify that compounds are listed in the order that you have in your MRM data upload file. Remove rows 2 and 3 from the output and paste the intensity data from column B rightward into cell AA2 in the MRM data upload file.