Automatic molecular annotation of mass spectrometry imaging data

Jan H. Kobarg1; Nikolaus Kessler2; Wiebke Timm3; Janina Oetjen1; Klaus Steinhorst1; Stefan Schiffler5; Shannon Cornett5; Aiko Barsch2; Heiko Neuweger3; Alice Ly4; Dennis Trede1

1SCILS, Bremen, Germany 2Bruker Daltonik, Bremen, Germany 3Bruker Daltonics Inc., Billerica, MA, USA

ASMS 2019, TP 409

Introduction
Mass spectrometry imaging (MSI) is used in untargeted metabolomics studies to investigate how compounds are localized across tissue samples. Modern MS instrumentation enables measurement of analytes at high mass resolving power with both high mass accuracy and high lateral resolution.

We present a novel software workflow for the identification of signals using data from the high-speed, high spatial resolution timsTOF flex instrument (see Fig. 1).

Methods

Sample preparation: Fresh frozen mouse brain specimen was sectioned to 10 μm thickness, and mounted onto conductive glass slides. DHB matrix was applied using a HTX TM sprayer.

MSI acquisition: Data were collected in positive ionization mode from 300 to 1000 Da at a pixel size of 20 μm on a timsTOF flex MALDI-QTOF instrument.

ROI information: SCiLS Lab Pro (version 2020a) was used to automatically create segments based on molecular similarity with bisecting k-means.

Analyte list matching: MetaboScape 5.0 was used to detect and combine isotope peaks and adducts with T-ReX2 algorithm. Annotations were then assigned based on chemical formula and matched against Lipid MAPS Structure Database.

Results

In total, the imaging experiment comprised of 123,300 pixels and 630 peaks. The spectra were segmented according to their molecular similarity. Representative segments matching anatomical features were converted into regions of interests (ROIs, see Fig. 2).

From these regions, 2500 spectra were averaged into 5x5 blocks yielding 100 spectra with average signal intensities. For feature extraction, deisotoping, and ion deconvolution, mass tolerances were set to 5 mDa and 250 mDa for isotopic pattern fits.

The MetaboScape T-ReX2 algorithm found 114 unique features. Chemical formula could automatically be assigned for 110 of these. Furthermore, 32 formulae could be matched against lipids in the LIPID MAPS Database.

Conclusion

This workflow provides annotations to the morphological topography in untargeted metabolomics studies.

Lipid annotation automated with MetaboScape 5.0 featuring T-ReX2 feature extraction technology and confident annotation quality scoring.

Visualization of annotated signals with SCiLS Lab Pro completing the intuitive workflow and confirming lipid distributions map to the expected localizations.

Imaging MS