IASMA – E. Mach Foundation - Metabolomic platform
Practical Trials LC-MS/MS

Practical trials for the LC-MS/MS are designed to test the performances of the analytical system in conditions which mimics real experimental ones.
The configuration of the instrument selected for practical trials have to be IDENTICAL to the proposed one.
Samples will be provided by IASMA well in advance in order to allow the definition of the best experimental conditions (both for LC and MS). Experimental conditions can be freely optimized by the applicationist in order to maximize the performances of the analytical system. Practical trials should be completed before the meeting with the IASMA commission.

After the practical trials a one day meeting with the IASMA researchers will be organized at the trial site. The meeting agenda will cover

a) Practical demonstration of the proposed analytical system (hardware + software).
b) Presentation and discussion of the practical trial results.
c) Definition of the data which have to be included in the "Practical Trial Report"

Experimental Protocols
Objective: To evaluate the MS/MS functionalities of the instrument,
To evaluate the procedure necessary for MRM optimisation
To test neutral loss function and possbile cross talking between the quadrupoles
To evaluate the sensitivity of the instrument (limit of detection and quantitation), the linearity range, the dynamic range, the stability of the signal (signal intensity, precision/accuracy of detected mass)
To evaluate post acquisition data treatment software utilities: multi-analyte quantitation, construction of calibration curves, quantitation in real samples.
Material: mix of non-volatile standard in matrix (grape extract) in different concentrations. Information on composition of text mixtures will be sent to the demo center by separate email.
Two vials are provided with mixtures (FM1 and FM2) of reference standard mixtures and details on their content are given (analytes and concentration). These should be used to construct calibration curves for the analytes. Reserpine should also be analysed separately only for the evaluation of the detection limit. Furthermore three vials with real sample (extract of grape) are provided: FM3, FM4, FM5. Quantitative analysis of the target analytes should be performed in these samples.
Mode: Gradient separation on a C-18 column (UP)LC and/or a HILIC column with ESI-MS and acquisition of chromatographic and spectral data (MS and MS/MS). Sequential injections to check dynamic range, linearity and detection limit of standards.

d) Dynamic range: injection of different dilutions of the sample over at least 6 order of magnitude.
e) Linearity: repeated injections (at least 3) of the sample at different dilutions. The trial report has to include calibration curves and peak areas both as Tables and Figures .
f) Detection limit: the detection limit for every standard has to be estimated in MS/MS mode and indicated in terms of injected pg. To check the reliability of the estimated detection limit the report must include the chromatographic trace for each standard at the lowest detectable concentration.

Solvents and materials to be provided:
LC: Water, formic acid, acetic acid, common LC eluents (CH_3CN, MeOH), one C-18 column, one HILIC column, ESI configuration