VALIDATION OF LC-MS/MS BASED MULTI-TOXIN METHODS – A SUGGESTION FOR REDUCING THE WORKLOAD AND FOCUSING ON THE DATA THAT ARE ESSENTIAL

M. Sulyok, D. Stadler, D. Steiner, R. Krska

University of Natural Resources and Life Sciences, Vienna, Department IFA-Tulln *michael.sulyok@boku.ac.at

Most of the guidelines that are available on proper method validation have been designed for assays targeting only one or very few analytes. In case of multi-analyte analysis, following these guidelines without any modification is impractical in view of the. To give an example, the validation data discussed in this work – targeting 540 fungal metabolites in 7 matrices with 7 individual samples per matrix and concentration, with additional measurements for the intermediate precision – comprised half a million chromatograms.

In addition, it is unrealistic to expect that compliance to all performance criteria (e.g. a target range of 70-120% for recovery) might be obtained for all analytes covering a diverse set of target substances. Consequently, guidelines for multi-analyte determination (such as SANTE/11945/2015 for pesticides residues) allow for some flexibility as considers both performance criteria and experiments that are foreseen for method validation.

This presentation tries to identify the part of the experiments that might be skipped during validation in order to reduce the workload. In particular, matrix effects as well as recoveries of the extraction step were independent of the concentration, which would allow for spiking at a single high level, thus facilitating data evaluation. In addition, recoveries of the extraction step have been found not to be significantly different between similar matrices from a given matrix group (grains, nut, dried fruits), which suggests that spiking experiments are not essential for method transfer to a similar matrix. Finally, the numerical values determined for the limits of detection and limits of quantitation following the EURACHEM guide were almost for mot analytes identical in figs and maize, questioning the necessity of carrying out the related experiments for each new matrix.

However, we found that a significant fraction of the method uncertainty derives from relative matrix effects, indicating that there is the need for an additional effort to characterize these effects as an essential part of the validation process. This is not foreseen in current guidelines and might lead to an underestimation of the methods uncertainty.