Electrochemistry/MS – a Powerful Tool in Drug Metabolism

Martin Evebergh, Agnieszka Kraj, Arleen Kennedy, Nico Reinholds, Jean-Pierre Chervet
Antec, Zoeterwoude, The Netherlands; Antec (USA), Boston, USA

Introduction
Electrochemistry (EC) in combination with mass spectrometry creates a powerful tool to simulate various oxidation and reduction processes in the human body. Electrochemistry is a complementary technique to traditional in vivo or in vitro metabolism studies, and delivers the oxidative metabolic fingerprint of a drug molecule in a very short time. Mass spectrometry delivers selective and sensitive detection and allows for unambiguous identification of all generated metabolites. Electrochemistry and mass spectrometry can then be collected for supplementary research such as NMR.

Methods
A preparative electrochemical cell (µPrepCell, Antec) equipped with a Glassy Carbon (GC) or Magic Diamond™ (BDD) working electrode was used for synthesis of metabolites. Typically, 2 mM solution of the drug was used in experiments with MS detection. 250 µM solution of the Verapamil in 250 mM ammonium formate, pH 7.4 in acetonitrile (ACN):vH2O was used as a model drug. The electrochemically synthesized metabolites of Verapamil were collected off-line, followed by MS analysis of the collected fractions (Figure 2C). The flow rate used in the synthesis experiments was 50 µL/min. The electrochemical cell was switched OFF confirms that the generated metabolites can then be collected for supplementary research such as NMR.

Results
For the formation of GSH adducts, 50 µM GSH solution was prepared based on scanning voltammetry. An LTQ Orbitrap XL (Thermo, USA) or HCT ion trap (Bruker Daltonics, Germany) mass spectrometer equipped with electrospray (ESI) source was used to monitor the oxidation or reduction products.

Conclusions
Using the ROXY™ EC system on-line with MS results in fast generation of metabolites (seconds to days), and in vitro methods, phase II reactions as well as reactive metabolites. Amiodarone and Verapamil were successfully used as model drug to mimic the oxidative metabolic pathway in the human liver by on-line EC/MS. Phase I and II metabolites, which were already known from the literature as detection products in vivo, were generated in the EC reactor cell and on-line identified by MS.

References