**INTRODUCTION**

The assessment of the ability of drug candidates to reach the desired target(s) in preclinical animal models is an essential step in the pharmaceutical discovery and development process. Typically, labelled probe based methodologies such as whole-body autoradiography (WBA) are used for assessing pharmacokinetic and distribution properties of a drug candidate.

Mass spectrometry imaging (MSI) permits the visualization of molecules in a label free and multiplexed approach. This provides the accurate localization of drug candidates and potential metabolites simultaneously, without the need for labeling. There is also the ability to identify m/z values for use as biomarkers.

Desorption electrospray ionization (DESI) mass spectrometry imaging (MSI) was used to analyse tissue sections from cassette dosed drug metabolism and pharmacokinetic (DMPK) models. The ability to map the distribution of drugs and their metabolites in the liver, alongside endogenous lipid species is demonstrated.

**METHODS**

**Sample Preparation**

In this oral dosing mouse model experiment, one cohort of animals was cassette dosed with four drugs; Terfenadine, Olanzapine, Moxifloxacin and Erlotinib (at 25, 10, 10 and 25 mg/kg respectively) and a second cohort was dosed with only Olanzapine at 25mg/kg. A further cohort was vehicle dosed to act as a control. Animals were euthanized and liver sections taken from each cohort at 2 and 6 hrs post administration.

Liver sections were adhered in the manner displayed in Figure 1.

**Instrument Settings**

DESI MSI analysis was performed using a Prospec DESI source fitted with a Waters sprayer on a Waters Xevo G2-XS Q-TOF. All data were acquired in MS mode. The DESI source can also be fitted onto a Synapt G2-Si (IMS enabled Quadrupole time-of-flight (Q-TOF)). Schematics of both Q-TOF instruments are shown in Figure 2.

**Data Processing**

High Definition Imaging (HDI)v1.4 software was used to define DESI imaging experiments and for subsequent processing/visualization of the imaging datasets from the Xevo G2-XS Q-TOF.

**RESULTS**

Ion images demonstrating the localization of the four drugs as well as endogenous species (mainly lipid species) were generated from a single experiment, as shown in Figure 3.

Within the same experiment it was also possible to detect some of the metabolites from Olanzapine, Terfenadine and Erlotinib. The ion images for terfenadine (m/z 472.32) and for two of its metabolites; hydroxyterfenadine (m/z 488.31) and carboxyterfenadine (m/z 502.29) are displayed in Figure 4. The distribution and signal intensity of ions across the sections were consistent with those shown in the latest publication from Swales et al.

In addition to the information gained for the distribution of the drugs themselves and their metabolites, it is also possible to view the distribution of molecular ions from species endogenous to the sample. Figure 5 shows the varying distribution of endogenous phosphocholine lipid species (as labeled on the individual images).

High Definition Imaging (HDI)v1.4 software also allows for overlay of the drug, metabolite and lipid ion images. Figure 6 shows the RGB overlaid ion images which allows clear visualization of the different distribution of ions within different anatomical microstructures in the liver. This has the potential to help determine the biological and histological impact of the drugs and metabolites within the sample, and provides an extra level of detail not available with other techniques. The level of can be improved by the acquisition of additional imaging datasets at higher spatial resolution, which when using DESI-MI is possible to by using a consecutive tissue section, or if using solvents of a low abrasive nature, on the same tissue section as the 150 µm images pictured here.

**Conclusion**

All of the aforementioned information was obtained from a single acquisition, without the need for any labeling or treatment of the tissue section prior to commencing the MSI imaging experiment. This saves valuable time, cost, and ultimately provides a wealth of information from a single section of tissue.

- Multiple drugs of low molecular weight were detected in both liver and brain tissue sections, independently of any labeling methodology.
- Simultaneous detection of drug molecular ions, their metabolites and endogenous lipid species, all in a single acquisition.
- Image overlay function, for ion images and for optical images.

**References**

1. J.G.Swales & co, Scientific reports, 2016, DOI: 10.1038/srep37648

**Acknowledgements**

We thank Dr Richard J.A. Goodwin and John G Swales from the Drug Safety & Metabolism at Astrazeneca R&D for providing all the samples used in these experiments.

TO DOWNLOAD A COPY OF THIS POSTER, VISIT WWW.WATERS.COM/POSTERS

©2017 Waters Corporation MEC3197