Desorption Electrospray Ionization (DESI) for tissue imaging on a Time-Of-Flight (TOF) mass spectrometer.

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In the past few years, Mass Spectrometry Imaging (MSI) has seen a rapid increase interest and utilization in areas such as life sciences (Omics, biomarker discovery), Pharmaceutical (drug distribution/validation in DMPK and toxicology), clinical sciences and many surface analysis techniques. MSI was originally developed using a Matrix Assisted Laser Desorption Ionization (MALDI) mass spectrometer where the sample is prepared by coating it with an ionizable matrix and placed under vacuum where a laser allow the ionization of molecule present on the surface of the sample.

More recently an ambient ionization technique called Desorption Electrospray Ionization (DESI) was introduced and applied to MSI to allow for the direct analysis of surfaces at atmospheric pressure.

The DESI technique uses a charged jet of solvent to deposit micro-droplets onto a surface where analytes are extracted and desorbed into the gas phase at ambient pressure and temperature, before being sampled into the MS inlet and subsequently mass analyzed using a TOF. This technique is compatible with Waters SYNAPT G2-Si or a Xevo G2-XS mass spectrometers.

To perform a DESI MS imaging experiment, the sample, that can be a fresh frozen thin tissue section is directly mounted onto a glass slide from the cryostat or freezer where it was stored, is placed onto the 2D linear moving stage of the DESI source without any other pre-treatment. An optical image is taken and co-registered with the High Definition Imaging (HDI) software (figure 1,B). This optical image is then used to define the area to be imaged (i.e. a rectangle is drawn around the tissue section). The surface of the tissue section is rastered line-by-line using the charged-droplet DESI sprayer with mass spectra collected at predefined X,Y coordinates. The pixel size in the X direction is defined by the speed of the stage movement and acquisition rate of

mass spectra. The Y-direction is defined by the distance between two line of acquisition. Typically, DESI imaging experiments are acquired with pixel sizes of 50 µm or more.

Raw imaging data is subsequently processed and visualized within the HDI software (figure 1,D). A significant advantage of DESI is that the tissue section can be H&E stained directly after MS acquisition (figure 1,E) since DESI is a non destructive technique. This allows the H&E stained optical image to be overlaid with the DESI molecular images from the same tissue section.

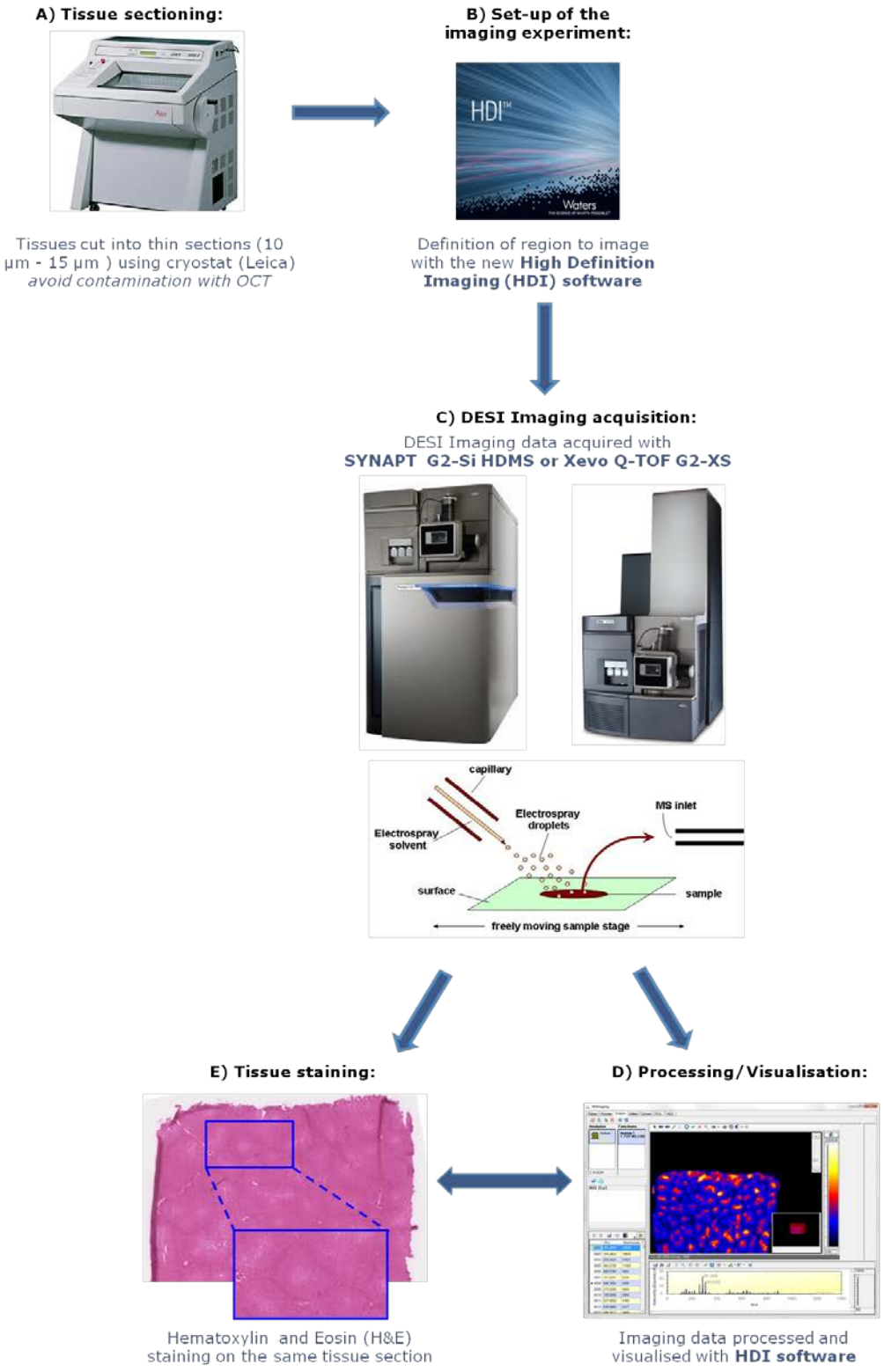

Figure 1. Workflow of a DESI MS imaging experiment .

DESI imaging, being a surface analysis technique, can be applied to a numerous types of samples, varying from animal and human tissue samples, to plant material, pharmaceutical tablets and bacteria grown on agar material.

Figure 2 shows a selection of ion images from a variety of samples acquired using DESI at a range of pixel sizes varying from 100 to 200 microns, measured either in positive or negative ionization mode. Figure 2,A is an ion image of oleic acid in porcine liver, highly concentrated in the center of the liver lobules co-localized with the central vein. Figure 3,B shows the potential to apply DESI imaging to forensics trace evidence analysis by generating molecular images from a fingerprint. Figure 2,C displays the overlay of two small molecules, differentially distributed within a bacterial colony grown on agar material[1]. Figure 2,D illustrates a clinical sample[2] that contains both normal cells and a secondary tumor, the distribution of two lipids m/z 698.51 (PE (O-34:3)) and m/z 773.53 (PG (36:2)) specifically differentiate the tissue type. In this example PE (O-34:3) is specifically localized within the tumor region.

DESI mass spectrometry imaging is a very effective technique that determines the spatial localization and molecular distribution of molecules within a variety of samples under ambient condition.

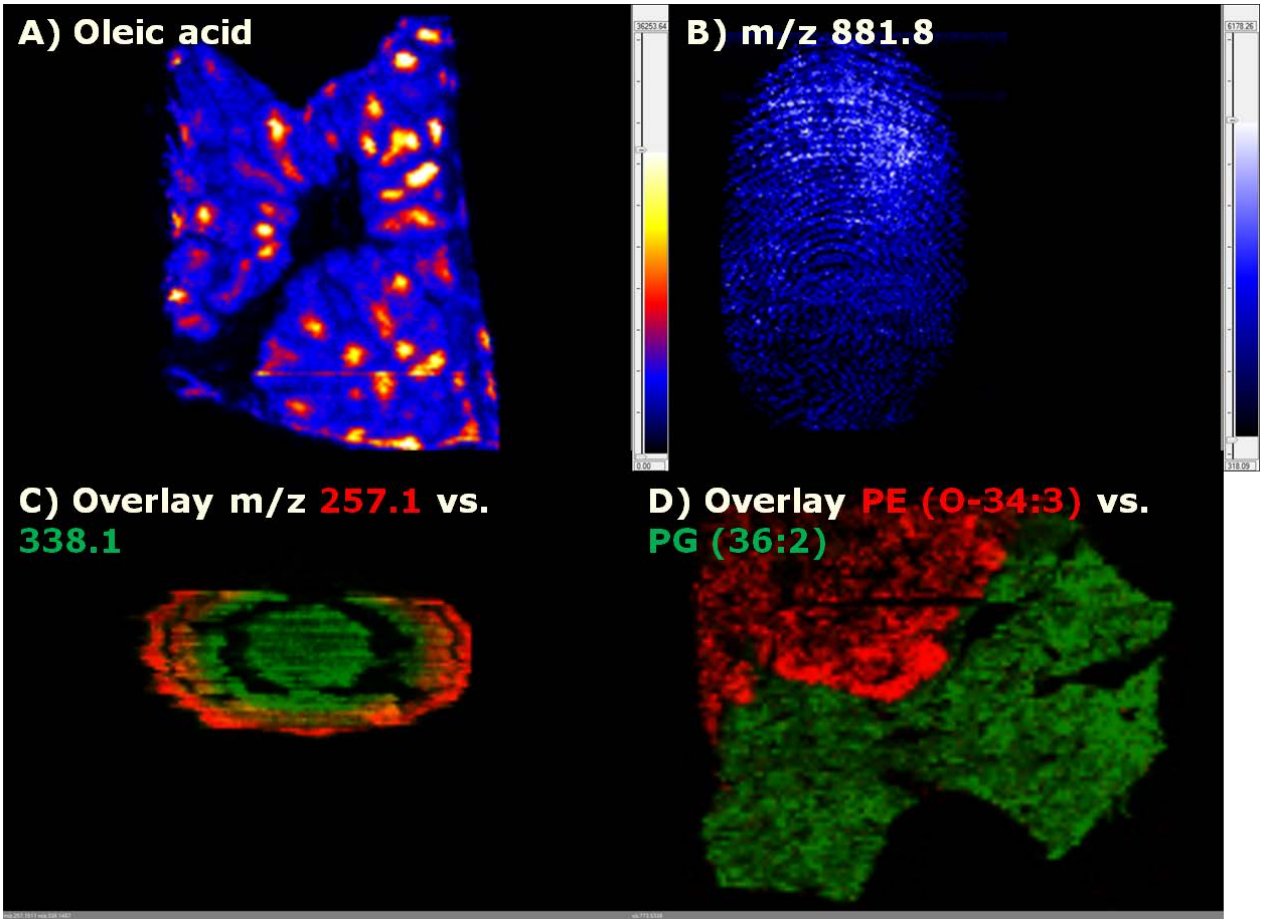

Figure 2. A) ion image of oleic acid in porcine liver, B) m/z 881.8 ion image of a human fingerprint, C) overlay of m/z 257.1 (red) and 338.1(green) from a bacterial colony[1] and D) overlay of ion images m/z 698.51 (PE (O-34:3) (red) and m/z 773.53 (PG (36:2)) (green) from human liver clinical sample[2].

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