Workpackage 1: Sample Preparation

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The objective is to enhance the performance of MALDI and SIMS mass spectrometry towards their imaging capabilities. Peptides, proteins, lipids, polymers and oligosaccharides are mapped with MALDI with the help of MALDI matrices, while lipids are mapped with SIMS with and without coating. In all the cases these compounds can be used either as calibrants or to adjust the mass spectrometer parameters.

The compounds are separated in six classes:
- Matrices for MALDI (Matrix Assisted Laser Desorption Ionization) and (ME-)SIMS: this first class of compounds are the matrices which make possible the ionization of:
- peptides and proteins
- lipids
- oligosaccharides
- Peptides and proteins: this second class of compounds can be utilized as calibrants in MALDI mass spectrometry, as well as to test the efficiency of matrices or mass spectrometer configuration (source, analyzer, detector) and performances.
- Lipids
- for SIMS: this third class of compounds can be used in SIMS to check the SIMS spectrometer parameters (primary ion source efficiency, analyzer, detection).
- for MALDI-MS: these compounds occur in biological systems ubiquitary and are very important for adjusting SMALDI ion sources for generating high quality images of biological samples.
- Oligosaccharides: they are the fourth class of substances and from biological interest. They can be used as calibrants and for MALDI mass spectrometry, as well as to test the efficiency of matrices or mass spectrometer configuration (source, analyzer, detector).
- Oligonucleotides: this fifth class of compounds can be used as calibrants and to test the mass spectrometer configuration for the measurement of samples derived from DNA and RNA samples.
- Polymers: the sixth class of compounds can be used as calibrants and to test the mass spectrometer configuration for the measurement of polymers.

All these compounds, as well as the reference spectra, are available all along the duration of the project to characterize and test the ion desorption methods, instrument performances in terms of sensitivity, mass resolution and/or spatial resolution.

The second deliverable reviews the choice and the characterization of reference biological samples which will be utilized along the program as references for SIMS and/or MALDI mass spectrometry and mass spectrometry imaging, as well as their most utilized samples supports, sample preparation methods, sample storage, etc…One important accomplishment is that silicon wafers have been agreed by all the involved partners to become the reference sample support for the future of the project. This material brings together several important and necessary features, flatness, conductivity, purity, adhesion of samples, as well as a moderate price and an excellent reproducibility.

Nevertheless, some of the other sample supports, which were previously utilized by the different partners, will still continued to be utilized for particular cases (e.g. optiTOF plates for the fixation of whole-body tissue sections). Concerning the standard samples, a general agreement has been found during the meeting held in Gif-sur-Yvette in December 2006 for rat brain tissue sections and several well known and well characterized cell lines.

Characteristic spectra and images of the reference biological samples are provided In a report. Some examples are given below:

Different sample treatments protocols utilized by the partners are described and discussed. The aim of these treatments is to obtain, for molecular imaging, and in both MALDI and SIMS, the best sensitivity, with the lowest spatial delocalization of the compounds. In MALDI, the application of a matrix at the surface of the sample is mandatory, and different methods are now supplanting the original method in which the tissue surface was manually coated with an air sprayer. Some of these methods can also be transposed to SIMS in which the addition of a matrix on the surface is not always utilized.

At CNRS-Gif, a prototype of an automatic spotter has been developed in collaboration with a French company (Siliflow). The droplets are delivered by a patented piezoelectric ejector, having different nozzle diameters (80 & 150 µm), with different ejected droplet volume (1 & 6 nL, respectively). The ejector is connected to a syringe pump and all the sucking, ejection and rinsing steps are fully automated and computer controlled. The precise positioning of the droplet on the sample is performed by a computer controlled plate. This system is presently able to deposit several droplets of ~400 µm diameter (depending on the tissue, the sample support, etc) at the same position.