

Recommendations for the preparation of cryostat sections for MALDI imaging

General remarks:

For good results, the preparation of the tissue sections is crucial.

Only cryosections are suited for MALDI imaging.

The tissue must not be embedded in supporting material such as optimal cutting temperature polymer (OCT). OCT contains polymers, which are detected in the mass spectrometer. These polymer signals may suppress the analyte signals completely. Homogenous tissue such as brain, liver kidney, tumors can be cut in a sufficient quality without embedding. Samples which contain unconnected tissue such as e.g. zebrafish need to be embedded for good quality sections. In this case the samples can be embedded in water or PBS buffer.

Freezing the tissue in liquid nitrogen usually makes the tissue brittle. For "normal" analyses freezing the tissue in a freezer or in the cryostat will be sufficient.

Please clean the blade of the cryostat with ethanol prior to preparation to remove OCT traces that may stick to the blade from earlier preparations.

The sections have to be prepared onto conductive slides (Bruker Daltonik Part No. 237001 "Glass slides for MALDI imaging")

On unstained tissue it may be impossible to distinguish histological features. To correlate the MALDI image with histological features it is therefore recommended to use a suited staining protocol on reference sections not used for MALDI imaging.

Histologically stained samples:

Some histological stains are compatible with MALDI imaging. This can help to correlate features from the stained samples directly with the MALDI image.

For detailed information please refer to:

Chaurand P, Schwartz SA, Billheimer D, Xu BJ, Crecelius A, Caprioli RM
Anal. Chem. (2004), 76(4): 1145-1155

Required Materials:

Cryostat

Conductive slides or a MALDI target

Optional: Artists brush for small sections or forceps for large sections

Two petri dishes

70% and 96% Ethanol (HPLC-grade)

Preparation:

- Appropriate thickness of the section is 10 to 20µm, if possible 10 to 12µm should be used. A recent paper states that for spraypreparations the thinner the section the better the spectra (Thin Sectioning Improves the Peak Intensity and Signal-to-Noise Ratio in Direct Tissue Mass Spectrometry; Yuki Sugiura, Shuichi Shimma, Mitsutoshi Setou *J. Mass Spectrom. Soc. Jpn. (2006), 54 (2), pp 45-48*).
- Place the conductive slides and the brush inside the cryostat and wait until they reached the cryostat temperature.
- Cut a tissue section.
- Transfer the section onto the cold (frozen) slide.
There are two possibilities to do this: Either the section is picked up with the artists brush or the forceps and transferred to the slide, or the tissue is picked up directly with the frozen slide from the blade.
(The often used method to transfer the section to a warm slide directly off the blade will result in significantly less intensity in the mass spectra!)
- Inside the cryostat, place the cold slide with the section on your hand to warm it. (If you omit this step, humidity in the air will condensate on the slide once you take it out the cryostat. This must be avoided.) Keep the slide on your hand until it appears to be fully dry.
- Take the warm slide out of the cryostat.

If possible, put the slide for five minutes in the vacuum of a desiccator (Brain sections should be placed in the desiccator for up to 45 min).

Prepare one petri dish with 70% Ethanol (HPLC grade) and one petri dish with 96% or pure Ethanol (HPLC grade). Wash the slide one to two times gently for 15 to 30 seconds in the 70% Ethanol and once for 15 seconds in the pure Ethanol. If small peptides (e.g. neuropeptides) are in the focus of interest keep the washes extremely short, for drugs or lipids skip the washing steps completely.

Desiccate the slide. The sample is now ready, it can be stored for a short time in the desiccator at room temperature or frozen down for longer storage or shipment. To facilitate the thawing procedure please pack the each slide in a separate container or wrap the individual slides in aluminium foil prior to freezing.