Sample Preparation Procedure for

Meat
Organic Solvent Extraction (OSExtr)

General Information

This protocol for meat, provided by CVUA Stuttgart, is based on the short description of *Post & Dikler* [2] and was further developed by P. Stoll within the scope of a student internship for biotechnology at the University of Applied Sciences Esslingen [4]. It was used to create and validate a reference database for muscle meat described before [1, 3].

A comprehensive collection of reference spectra (msp) and single spectra generated using this protocol is available for exchange on the MALDI-UP homepage (https://maldi-up@ua-bw.de).

Field of Application

Muscle meat of mammals, birds and reptiles in natural form, raw, frozen or heated (cooked, grilled). Also applicable for liver and kidney.

Chemicals and Material

- 1.5 ml reaction tubes and tips
- Silica beads (0.1 mm diameter)
- Micropestle, fitted for the tubes used
- Benchtop centrifuge
- HCCA Matrix solution (please see »Tipps and Recommendation«, page 2)
- OS Solvent (acetonitrile 50 %, water 47.5 % and trifluoroacetic acid 2.5 %)

References

[1] Hiller, E., Männig, A., Rau, J. (2017): Tierartendifferenzierung bei Fleisch – Mit MALDI-TOF-MS von der Datenbank zur Validierung. Deutsche Lebensmittelrundschau 113: 12–16.

[2] Post, A., & Dikler, S. (2010): Identification of fish species by protein profiling using MALDI-TOF mass spectrometry. Poster at FACSS 2010, 583.

[3] Rau, J., Hiller, E., Männig, A., Dyk, M., Wenninger, O., Stoll, P., Wibbelt, G., Schreiter, P. (2020b). Animal Species Identification of Meat using MALDI-TOF MS. In preparation.

[4] Stoll, P., & Rau, J. (2015): Tierartendifferenzierung von Fleisch mittels MALDI-TOF MS; 44. Deutscher Lebensmittelchemikertag 14.–16.09.2015 Karlsruhe. https://maldi-up.ua-bw.de/docs/CVUAS_Stoll_Rau_Tierarten_MALDITOFMS_20150914.pdf

Meat:

Organic Solvent Extraction (OSExtr)

Extraction Procedure

- Add silica beads (about 5 mm high) into a 1.5 ml reaction tube
- Add 100–200 µl OS
- Add about 3 mm^3 muscle tissue
  For the best results please use material from the inner part of the tissue to avoid possible oxidation and contamination on the surface
- Homogenize/grind the sample using a micropestle
- Mix thoroughly by vortexing for 10–20 seconds
- Centrifuge at 12,000–14,000 rcf in a benchtop centrifuge for 2 minutes
- Pipet 1 µl supernatant onto a target sample spot (We recommend spotting the supernatant in duplicate or triplicate)
- As soon as the sample spot has dried, overlay the sample with 1 µl HCCA matrix solution (to prevent oxidation reactions leading to peak shifting)
- Allow the sample spot to air dry before analysis ➔ MALDI measurement

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