MALDI mass spectrometry imaging workflows for the ecotoxicological model organisms Daphnia magna, Danio rerio and Eisenia fetida

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Abstract

Enviromental toxicology aims to understand the sources, fate and effects of chemicals released into the environment. A classical approach to understand toxic mechanisms involves the use of scientifically well-characterized model organisms. In ecotoxicological studies targeting aquatic ecosystems, *Daphnia magna* (waterflea) and *Danio rerio* (zebrafish) are established model organisms while the earthworm *Eisenia fetida* has proven to be a suitable candidate for the studies of terrestrial ecosystems. MALDI-MSI is a powerful technique for the visualization of molecules within tissue sections. However, for *E. fetida* and *D. rerio* only few and for *D. magna* no suitable MALDI-MSI workflows have been established. We developed three MALDI-MSI workflows for the analysis of *D. magna*, *D. rerio* and *E. fetida* tissue sections with high-spatial resolution (5-25 μm).

An emphasis was put on the development of cryosectioning protocols that preserve the tissue integrity while maximizing signal intensity during MALDI-MSI measurements. Sectioning of daphnids still represents a challenge as their encapsulating carapaces tend to fracture during cryosectioning leading to disruption of the body integrity. To overcome this hurdle, daphnids were allowed to swim in gelatin-solution to ensure that the cavity within the carapace and its appendices are filled with embedding medium. This strongly contributes to tissue stability during cryosectioning. We were able to obtain coronal sections with preserved tissue integrity. The subsequent MALDI-MSI measurements revealed distinct lipid-distributions in embryos/eggs, the carapace lining and parts of the thoracic legs. Sagittal sections of *D. rerio* fish were sectioned at -15 ∘C. We were able to obtain distinct lipid-distributions showing anatomical structures of the eye, the brain, gill filaments and the liver. Earthworms were anesthetized using 7% MgCl to enhance muscle relaxation and subsequently embedded in 3% CMC. Sectioning was performed at -20 ∘C. The resulting lipid ion images showed prominent histological features such as the typhlosole, colon and muscle-ring.

Keywords: MALDI MS imaging workflows, high spatial resolution in mass & space, histology

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