Abstract

For a specific detection of primary metabolites in microalgae different analytical platforms were compared. Therefore, methods for each platform were developed. As primary metabolites lipids and carbohydrates were analyzed.
A specific method for the detection of lipids as triacylglycerols was developed for UPLC-MS/MS. The method was tested qualitatively with model substances. As model substances a mixture of five different TAG was used. For the detection of fatty acids, lipids were derivatized to fatty acid methyl esters and analyzed by GC-MS. Both, the extraction protocol and the analytical method were established for the detection of FaA and tested, if the process is suitable for the detection in microalgal cultures.

Specific methods for the detection of carbohydrates were developed for the analytical platforms of UPLC-HRMS, UPLC-MS/MS and HPAEC-PAD. These methods were tested with respect to their individual performance in qualitative and quantitative assessment of model substances and complex extracts from microalgal cultures. As model compounds different monosaccharides and disaccharides were tested. For example, glucose, galactose, fucose and rhamnose which are known compounds for the monosaccharide profile of microalgae were tested. Furthermore, other monosaccharides were analyzed, because only few information about the specific composition of the sugar profile for different microalgae is known and some algae may include other monosaccharides. The disaccharides were used to detect not fully hydrolyzed sugars.

Comparison of the methods and of the analytical platforms was done with reference to separation, limit of detection, linearity and influence of matrix substances. For all developed methods linearity R^2 of 0.99 was detected. LOD ranged between > 100 pg $\cdot$ μL^{-1} for UPLC-HRMS and < 10 pg $\cdot$ μL^{-1} for UPLC-MS/MS. Baseline separation for most of the analyzed sugars was only achieved with anion exchange chromatography. To determine the specific carbohydrate composition polysaccharides were hydrolyzed to monosaccharides under acidic conditions.
Additionally, extraction protocols were tested for possible sources of contamination and application to the extraction of microalgae.

In a second project (page 52) passive samplers (SPATT bags, consisting of hydrophobic resins) were used to absorb toxins from microalgae species in Arctic waters. Methanolic extracts from these resins were analyzed for dinoflagellate toxins and the detected toxins were used as chemotaxonomic markers for different harmful microalgae species. Detection was done with an established multi method for the detection of lipophilic toxins and another more specific method for azaspiracids.