

ABSTRACT

Lipids are basic biomolecules found in all organisms. They have a key function as structural molecules forming cell membranes, and, in the form of fats and oils, energy is also stored. There are a huge number of lipid types that have other functions, for example, in cell signaling, enzyme support, protection against stress, and others. Microscopic algae are the main primary producers in both freshwater and marine ecosystems. Therefore, algal biosynthesis has a fundamental effect on the trophic networks of aquatic ecosystems and ultimately on humans. The environment affects the ecophysiology of algae, which is reflected in their biochemical composition, i.e. in the composition of their lipids. This work is focused on two groups of lipids, namely glycerolipids, which consist of fatty acids, and carotenoids, which are photosynthetic pigments with antioxidant and photoprotective properties. In these groups of lipids, the target substances were further selected, namely polyunsaturated fatty acids and the carotenoid fucoxanthin belonging to xanthophylls. These substances are important not only in the ecophysiology of algae, but they are valuable substances that have positive effects on the human organism and, with the help of optimized cultivation, could be obtained from algae for industrial production under certain conditions.

Although much attention has been paid to this research direction for a long time, there are still many gaps in our knowledge, some of which this Ph.D. thesis tries to fill. Its core is formed by seven scientific publications as well as several outputs from applied research. The work deals with different species and groups of algae with different origins and ecology. In addition to the selected target substances, the unifying element is also the methodological approach, i.e. the implementation of extensive manipulative multi-parametric experiments. In this way, it is possible to monitor the influence of individual biological factors (geographical origin or taxonomic affiliation) or abiotic factors (temperature, light intensity or composition of the culture medium) on the ecophysiology of selected algae strains manifested at the level of selected target substances.

The greatest attention was paid to the freshwater flagellated alga *Hibberdia magna* (Chrysophyceae), in which we studied in detail its reactions to temperature and lighting with regard to the productivity of biomass and selected valuable substances. Fucoxanthin productivity was optimal under different conditions than biomass and polyunsaturated fatty acid productivity. This work presents the first insight into the biotechnological potential of this poorly explored species, which can serve as a model organism for future studies of photoautotrophic chrysophytes.

In another study, we compared the alga *Hibberdia magna* with the species *Chlorochromonas danica*, which belongs to the same class but differs in its trophic strategy (mixotrophy). The presence of light stimulated the increase in fucoxanthin productivity, the content of which was minimal in the dark. Both species had similar fucoxanthin productivity under optimal conditions, which was comparable to other fucoxanthin-producing strains. Both organisms were positively evaluated as candidates for the multi-target biorefinery concept.

In the third paper, we focused on freshwater diatoms, another group that is not so well known in terms of manipulative culture experiments and lipid composition. Based on the incubation of 11 newly isolated strains from different climatic zones (tropical, temperate, polar) at different temperatures, we found clear trends in their fatty acid profiles. Surprisingly, the geographic origin of the strains had a comparable influence as the temperature of cultivation, and taxonomic affiliation was similarly significant. The most sensitive to temperature changes were the polar strains, which were also characterized by the highest average proportion of polyunsaturated fatty acids compared to the temperate and tropical strains.

In another work, we tested an Antarctic strain of the green alga from the genus *Monoraphidium* to produce polyunsaturated fatty acids on a pilot scale. From the point of view of the year-round use of cultivation capacities in the temperate zone, the cultivation of psychrotolerant strains can be advantageous. The tested strain was isolated from a frozen lake and, in addition to the shifted growth optimum to low temperatures, it was also characterized by low light requirements and an increased content of polyunsaturated fatty acids compared to other members of this genus. Cultivation on a thin-layer platform in the winter season proved that this strain is promising.

Other organisms studied in terms of the influence of conditions on lipid composition were five strains from the Haptophyta group. Using reversed-phase high-performance liquid chromatography (NARP-HPLC) with atmospheric pressure chemical ionization (APCI), we characterized the composition of their triacylglycerols at different salinity levels, with an emphasis on 18-carbon polyunsaturated fatty acids. Salinity had an effect on the proportion of saturated and unsaturated fatty acids, as well as their composition in terms of regioisomers.

The other two works were devoted to methodological aspects of algal biotechnology. It involved optimizing the extraction and purification of fucoxanthin from the biomass of the marine diatom *Phaeodactylum tricornerutum* using high-performance countercurrent chromatography (HPCCC), which consistently preserved its biological activities. Furthermore, we focused on the comparison of different approaches for the early detection of contamination of algal cultures using the example of a culture of the diatom *Phaeodactylum tricornerutum* contaminated with the alga *Chlorella* sp. The use of PCR and qPCR methods proved to be the most reliable, detecting contamination already at a level of around 75 cells/ml, and therefore appear to be the best approach for the early detection of contamination in algal cultures.

Last but not least, the work also includes outputs from applied research. The first of them is the presentation of a functional prototype of the cultivation device, which was developed as part of the doctoral study and with its help experiments were carried out, which are published as part of the main results of this work. Furthermore, the outputs from the semi-operational cultivations of the algae *Hibberdia magna* and *Phaeodactylum tricornerutum* are presented. This work was carried out in cooperation with the Czech company Algamo s.r.o., which deals with the commercial cultivation of microalgae.