

Estimating the Efficiency of Fatty Acid Extraction from Fungal Biomass by FTIR Spectroscopy

Kristin Forfang^{1,2}, Achim Kohler¹, Odd Ivar Lekang¹, Volha Shapaval^{1,2}

¹Dept. Mathematical Sciences and Technology, Norwegian University of Life Sciences
Drøbakveien 31, 1430 Ås Norway

²Nofima AS, Osloveien 1, 1430 Ås, Norway

During the last decades, the growing global demand for fuel and food resources has increased the interest in microbial production of lipids. This growing interest imposes requirements for thorough investigations of methods used for extraction and quantification of lipids from microbial sources (yeast, bacteria and filamentous fungi). Analysis of the fatty acid content is commonly done by gas chromatography (GC) after extracting lipids from the biomass and converting the fatty acids into fatty acid methyl esters (FAME) by transesterification. A wide range of lipid extraction protocols are presented in literature, including the well-known protocols described by Folch *et al.* [1] and Bligh & Dyer [2]. These methods are widely cited, but numerous modifications have been introduced through the years depending on the sample origin, some being presented as modifications while others are still being referred to as the original methods. The application of different methods to new sample material can cause considerable variation in the amount and composition of fatty acids extracted [3]. This variation can be explained both by variation between extraction methods and inequalities in sample structure, like water content and the nature of cell membranes and lipids.

By FTIR spectroscopy, cell compounds can be probed in situ. While FTIR spectroscopy does not allow performing a direct quantitative fatty acid analysis, it is an excellent tool for fingerprinting the biochemical composition of biomass. In the present work, we compare different lipid extraction protocols on filamentous fungi and the biochemical fingerprints of the biomass obtained by FTIR spectroscopy before and after extraction. We show that applications of known extraction protocols [1, 2] to fungal biomass are not able to extract all lipids and hence underestimate the fatty acid yield. Further, we evaluate the use of FTIR spectroscopy as a rapid quality control (QC) method for determination of lipid extraction efficiency in filamentous fungi. The developed FTIR-based QC approach can easily be transferred to other biomass sources such as yeasts, bacteria and algae.

References

- [1] J. Folch, M. Lees, G.H. Sloane Stanley, *J Biol Chem* 226, 497-509 (1957).
- [2] E.G. Bligh, W.J. Dyer, *Canadian Journal of Biochemistry and Physiology* 37, 911-917 (1959).
- [3] S.J. Iverson, S.L.C. Lang, M.H. Cooper, *Lipids* 36, 1283-1287 (2001).