

Enrichment of PUFA in Nile perch free fatty acids by selective enzymatic esterification and subsequent analysis using HPLC-ELSD

Abstract:

PUFA from oil extracted from Nile perch viscera were enriched by selective enzymatic esterification of the free fatty acids (FFA) or by hydrolysis of ethyl esters of the fatty acids from the oil (FA-EE). Quantitative analysis was performed using RP-HPLC coupled to an evaporative light scattering detector (RP-HPLC-ELSD). The lipase from *Thermomyces lanuginosus* discriminated against docosahexaenoic acid (DHA) most, resulting in the highest DHA/DHA-EE enrichment while lipase from *Pseudomonas cepacia* discriminated against eicosapentaenoic acid (EPA) most, resulting in the highest EPA/EPA-EE enrichment. The lipases discriminated between DHA and EPA with a higher selectivity when present as ethyl esters (EE) than when in FFA form. Thus when DHA/EPA were enriched to the same level during esterification and hydrolysis reactions, the DHA-EE/EPA-EE recoveries were higher than those of DHA/EPA-FFA. In reactions catalysed by lipase from *T. lanuginosus*, at 26 mol% DHA/DHA-EE, DHA recovery was 76% while that of DHA-EE was 84%. In reactions catalysed by lipase from *P. cepacia*, at 11 mol% EPA/EPA-EE, EPA recovery was 79% while that of EPA-EE was 92%. Both esterification of FFA and hydrolysis of FA-EE were more effective for enriching PUFA compared to hydrolysis of the natural oil and are thus attractive process alternatives for the production of products highly enriched in DHA and/or EPA. When there is only one fatty acid residue in each substrate molecule, the full fatty acid selectivity of the lipase can be expressed, which is not the case with triglycerides as substrates.