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Abstract

Idea of Rapid Preparation of Fatty Acid Methyl Ester Using In Situ Derivatization from Fresh Horse Mussel †

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Abstract: The analysis of the fatty acid (FA) profile requires multiple preparation steps, which are lipid extraction followed by derivatization of the FA into a fatty acid methyl ester (FAME). The procedures are time-consuming, and generally require large volumes of sample sizes and solvents. This report proposes a technique for the preparation of FAME from fresh horse mussels without a step of lipid extraction. A rapid in situ derivatization using *N*,*N*-dimethylformamide dimethyl acetal (DMF-DMA) methylation followed by alkali-transesterification was examined. In this method, acylglycerols and free fatty acids (medium to long-chain FA) of the sample are targeted to convert into FAME. Direct alkali-transesterification of the fresh sample gave only 58.7% FAME with 12.4% triglyceride and 21.1% FFA. The alkali in situ method showed low conversion efficiency due to the initial sample containing high contents of moisture and FFA (75.11% of the fresh sample and 14.3% of total oil, respectively). The reaction was developed by using two steps in situ derivatization. A 50 mg sample was methylated with 1 mL of DMF-DMA (100 °C, 15 min), followed by transesterified with 10 mL of 1% (*w*/*v*) NaOH in methanol (60 °C, 3 min). The conversion into FAME was monitored using size-exclusion HPLC with evaporative light-scattering detection. The column was a 100 Å Phenogel with toluene and 0.25% acetic acid as a mobile phase. The FAME yield of 79.9% with 7.8% triglyceride and 8.5% FFA was obtained. The two steps in situ derivatization gave a promising result with the higher conversion with lower FFA. It is a simple and rapid (less than 20 min) method that requires a low volume of sample and solvent for FAME preparation. However, increasing the conversion efficiency as well as the variety of samples should be further studied.

Keywords: fatty acids; fresh sample; in situ transesterification; methylation; *N*,*N*-dimethylformamide dimethyl acetal

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