Identification of Carotenoid Isomers by Mass Spectrometry in Crude and Bleached Palm Oil

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Crude palm oil undergoes refining process prior to human consumption, aiming to removing compounds such as phospholipids, free fatty acids and pigments. It is especially important the removal of carotenoids from palm oil as they are responsible for the dark orange color that would derail its use in a wide range of food and pharmaceutical products such as ice creams, breads, margarines and cosmetics. Most of carotenoids are removed during the bleaching, an adsorption purification step. There are several studies in the literature concerning total carotenoids removal by adsorption, however, those studies do not evaluate how the process influence on each carotenoid isomer. In this context, this work aim to identify the $\alpha\text{-}$ and $\beta\text{-}carotene$ by UHPLC-ESI(+)-MS/MS in crude palm oil and bleached by commercial bleaching earths (acid and neutral), under industrial applied conditions, as described elsewhere. The analytical procedure was developed using a model oil consisting in several acylglycerols standards and $\alpha\text{-}$ and $\beta\text{-}carotene.$ The UHPLC separation was achieved by gradient elution (methanol+formic acid+NH4OH and propanol+formic acid+NH4OH) in a C18 column at 40°C. Samples were solubilized by hexane and chloroform, without any further preparation. The technique was able to separate the isomers α - and β -carotene from acylglycerols, at 3.8 and 3.9 minutes of retention time, respectively. Both carotenoid isomer presented the 444 m/z fragment, however, only the isomer α presented the fragment 388 m/z, being this a diagnostic ion. After the analytical development, crude and bleached palm oil were analyzed. It was observed that crude $^{\text{palm}}$ oils presented both carotenoid isomers, but the $\beta\text{-carotene}$ was found in higher concentrations. The chromatograms of bleached oils present different profiles: oil treated with neutral bleaching earth presented a significant reduction of β - peak, however without reducing the $\boldsymbol{\alpha}$ one; oil treated with acid bleaching earth presented a reduction of both peaks.

Keywords: adsorption, carotenes, palm oil, refining, mass spectrometry

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