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Supporting Information

1. Experimental Section

1.1 Materials and Methods

Flaxseed cake and camelina cakes were obtained from Savi Italo s.r.l. [Fiorenzuola d'Arda (PC), Italy]. All chemicals and solvents were of analytical grade and purchased from Merck. Immobilized CAL B, PS lipase and AK lipase were purchased from BiCT s.r.l. Protease P Amano-6 SD and Protease M Amano were kindly donated by Amano Enzyme Europe Limited.

1.1.1 Extraction of Residual Oils and Proteins From The Cakes

Residual oils and proteins from Flax (FCO and FCP) and Camelina (CCO and CCP) were obtained following a previously described method [1].

1.1.2 Analyses of Hydrolysates from Oilseed Press Cake Residual Oils

The analyses were carried out by a 1200 Infinity HPLC coupled by a Jet Stream ESI interface with a Quadrupole-Time of Flight tandem mass spectrometer 6530 Infinity Q-TOF detector (Agilent Technologies, USA). The chromatographic separation was performed on an Agilent Poroshell 120 EC-C18 column (3.0 mm x 50 mm, 2.7 µm) with a Zorbax eclipse plus C-18 guard column (4.6 mm×12.5 mm, 5 µm). The mass spectrometric acquisitions were performed in negative mode ionization for the first 17 min of the run to characterize the free fatty acids and monoacyl glycerols (MAGs), while in positive mode ionization for the analysis of diacylglycerols (DAGs) and triacylglycerols (TAGs). The detailed conditions are reported elsewhere [2]. The mass spectrometric identification of diacylglycerols and triacylglycerols was performed according to literature [3].

1.1.3 Analyses of Hydrolysates from Oilseed Press Cake Extracted Proteins

The analyses were performed using a Shimadzu LC chromatograph coupled with an AB Sciex Triple Quad 5500^{TM}. The chromatographic separations were performed on a SIELC Primesep 100 column (3.2 x 100 mm, 3 µm, 100 Å) at LabAnalysis [Casanova Lonati (PV) Italy].

1.2 Biocatalytic Hydrolysis of FCO and CCO – General Method

250mg of immobilized lipase were suspended in 2mL phosphate buffer 50mM pH 7 and activated for 15min at 35°C. Then, to the mixture 50mg of FCO or CCO dissolved in 0.3mL n-hexane (13% v/v) were added. The reaction was performed at 40°C, 550rpm. After 24h the reaction was stopped through filtration of the immobilized enzyme under vacuum through a glass-filter tunnel with fritted disc (medium porosity) fitted with a Buchner flask and washed with n-hexane (3x2.5mL). The aqueous phase was extracted with n-hexane (3x10mL) and the combined organic phases were washed with brine. The organic phase was dried over Na_2SO_4, filtered and the solvent was removed by distillation under reduced pressure on a rotary evaporator to provide the crude as a yellow oil.

1.3 Biocatalytic Hydrolysis of FCP and CCP – General Method

FCP or CCP (20mg) were dissolved in 0.1M phosphate buffer pH 7 (5% w/v) and hydrated during constant stirring for 30min at 35°C. Finally, protease was added in an enzyme-substrate ratio of 1:2 and the reaction was performed at 35°C, 400rpm for 24h. Then, the reaction mixture was heated in a 90°C water bath for 10 min to inactivate the enzyme. Afterwards, the solution was cooled to room temperature and centrifuged at 10.000 rpm for 2min. The supernatant was isolated and characterized as described in 1.1.3.

1.4 Acidic Hydrolysis of FCP and CCP – General Method

Hydrolyses of FCP and CCP were performed according to literature [4].

References

1. Parodi E, La Nasa J, Ribechini E, Petri A, Piccolo O. Extraction of Proteins and Residual Oil from Flax (*Linum usitatissimum*), Camelina (*Camelina sativa*), and Sunflower (*Helianthus annuus*) Oilseed Press Cakes. Biomass Conv Bioref. 2021. doi: 10.1007/s13399-021-01379-z.
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