Enrichment of anhydrous milk fat in polyunsatured fatty acid residues from linseed and rapeseed oil through enzymatic interesterification

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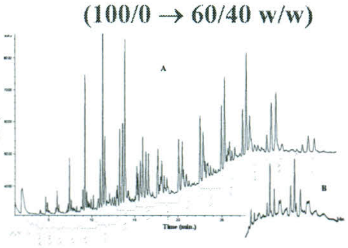
Introduction

The interesterification, or ester exchange, between two fats leads to the rearrangement of acyl moieties in both. The use of a sn-1,3-specific lipase confines the exchange of fatty acid residues to the sn-1 and sn-3 positions of triacylglycerides (TAG), generating products with characteristics that cannot be obtained through a chemical process or a blending. Such reactions require mild conditions with no solvent needed and they yield no unhealthful *trans* fatty acids, justifying the stepped-up interest of enzymatic interesterification for the production of margarines and other food fats.

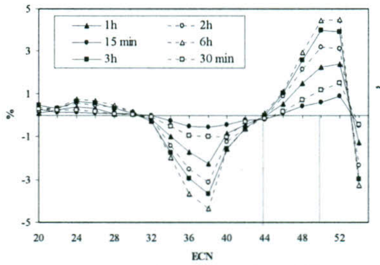
The aim of this work was to use enzymatic interesterification to enrich anhydrous milk fat (AMF) with unsaturated fatty acid C_{18} residues from linseed oil (LO) and eventually from rapeseed oil (RO) through some binary blends and one ternary blend. For that, the 1,3-specific lipase from *Thermomyces lanuginosa* (Lipozyme TL IM) was used in solvent-free batch and micro-aqueous reactions and fat blends with different mass ratios were tested. The evolution of TAG profiles, of interesterification degree (ID) and of free fatty acids (FFA), was followed along the reactions. Determination of dropping points (DP) and solid fat contents (SFC) enabled a rheological characterization of the products. The end products were also characterized for their oxidative stability and their textural properties.

Binary blends AMF/LO (100/0 → 60/40 w/w)

Experimentation and results

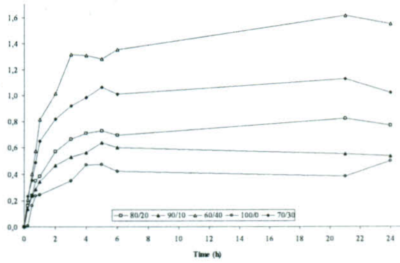


GC chromatograms of AMF (A) and of LO (B). The braces indicate the peaks with the same equivalent carbon number (ECN) and this number is indicated below.



Variations of TAG (%) related to the noninteresterified blend (NIE), along the interesterification reaction for the blend AMF/LO 80/20 (w/w).

ECN 30, 32 and 44 had no or low evolution. A decrease of ECN 34-42 and 54 occurred (corresponding respectively to TAG with medium chains acyl moieties and with tri-C18), together with an increase of ECN 46-52 (TAG with intermediate species).

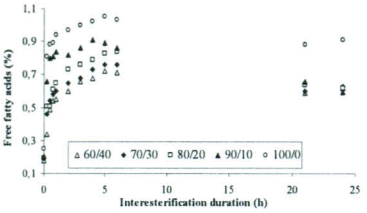


Interesterification degree as a function of time

$$ID = \left(\frac{\text{Area ECN50}}{\text{Area ECN44}}\right)_{IE} - \left(\frac{\text{Area ECN50}}{\text{Area ECN44}}\right)_{NIE}$$

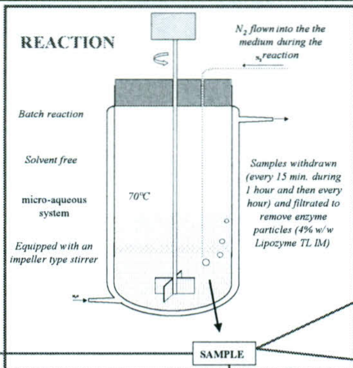
where ECN50 and ECN44 are the peaks with ECN equal to 50 and 44, corresponding respectively to a highly variable and the most stable group of peaks during the reaction, for interesterified (IE) and NIE samples.

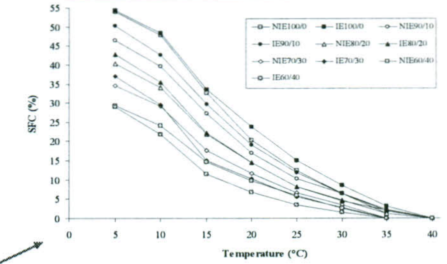
The interesterification occurred fast during the first hour of reaction and reached a quasi-equilibrium state after 4-6 hours with no significant further evolution until 24 h.



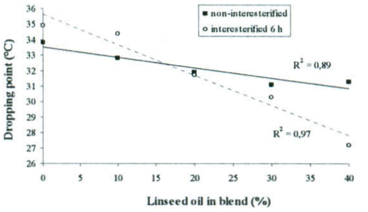
Free fatty acids as a function of reaction time

The contents of FFA reached again a maximum after 4-6 h of reaction and were in all cases inferior to 1%. The more AMF was contained in the blends the higher were the concentrations of FFA.





SFC (%) profiles for NIE and IE 6 h blends



Dropping points (DP) as a function of LO in the blend, for NIE and IE 6h blends

DP = temperature at which the sample flows and sections an IR beam.

- 100/0 and 90/10: slight increase of the DP temperatures after interesterification, explained by a slightly denser crystal network.
- The other blends: decrease of DP observed, the more important as LO content is high → softer ?

		Blends				
		100/0	90/10	80/20	70/30	60/40
Oxidation induction time OIT (h)	NIE	28,6 +/- 0,7	17,6 +/- 0,2	9,7 +/- 0,2	9,4 +/- 0,3	7,0 +/- 0,2
	IE 24 h	27,6 +/- 0,6	18,0 +/- 0,1	9,8 +/- 0,5	9,2 +/- 0,0	6,2 +/- 0,2

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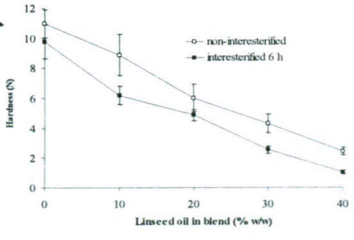
Oxidation induction times determined by the rancimat method

OIT of the IE products were not significantly different from those of the NIE blends. Nevertheless, they diminished clearly with the LO content (poor stability from 20% LO)



Hardness, defined by maximum penetration force (in N), as a function of LO in the blend, for NIE and IE 6 h blends.

In average, the IE products were 30% softer than the NIE.

Ternary blend AMF/RO/LO (70/20/10 w/w)

		ID	FFA (%)	DP (°C)	% SFC			OIT (h)	Hardness (N)
					10°C	15°C	30°C		
Blend 70/20/10 (w/w)	NIE	0	0,2	31,6 +/- 0,8	29,8	16,5	3,3	21,8 +/- 0,3	4,5
	IE 6 h	0,7	0,7	29,5 +/- 0,1	29,8	15,6	2,5	/	/
	IE 24 h	0,6	0,6	30,4 +/- 0,4	30,3	16	3	21,8 +/- 0,3	2,9

Data determined for the ternary blend AMF/R/LO 70/20 /10 (w/w)

The product presents the spreadable character of a good shortening and the oxidative induction time is appreciably improved

Conclusions

A fast batch reaction led to the enzymatic interesterification of AMF with LO, giving rise to different products enriched in unsatured C_{18} and especially in $C_{18:3}$, with physical properties dissent from those obtained with the single blend of oils. While interesterifying AMF alone or with 10% LO yielded a harder fat, rheological data for the other blends demonstrated that softer products were obtained. The properties of the products derived from the blends 70/30 and 60/40 were compatible with those of a butter-based spread, however their oxidative stability was low. Thus, a ternary blend composed of AMF/RO/LO gave satisfactory rheological and oxidative properties, fulfilling the requirements for a marketable spread and moreover offering increased potential health benefits, due to its enriched content in polyunsatured fatty acids.

Reference: **M. Aguedo, E. Hanon, S. Danthine, M. Paquot, G. Lognay, A. Thomas, M. Vandenbol, P. Thonart, J-P. Wathelet and C. Blecker. (2007). Enrichment of anhydrous milk fat in polyunsatured fatty acid residues from linseed oil and rapeseed oil through enzymatic interesterification. Submitted in Journal of Agricultural and Food Chemistry.**