ISSN 0101-2061 Ciência e Tecnologia de Alimentos

Physical properties of structured lipids from lard and soybean oil produced by enzymatic interesterification

Propriedades físicas das misturas e dos lipídios estruturados obtidos a partir de banha e óleo de soja por interesterificação enzimática

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Abstract

The main goal of the present research was to evaluate the physical properties of blends of lard and soybean oil modified by enzymatic interesterification catalyzed by two different commercial (microbial) lipases, viz. from *Candida cylindracea* (AY30™) and from *Mucor circinelloides* (M10™). Pure lard exhibited a softening point of ca. 31.8 °C before interesterification, and this value shifted towards 29.1 °C after interesterification by AY30 lipase and towards 28.8 °C after interesterification by M10 lipase The interesterified lard exhibited lower consistency after reaction with both lipases, and this decrease was more pronounced for the reaction catalyzed by M10 lipase. This result was most likely due to the *sn*-1,3-specificity of M10 lipase. Pure lard displayed a lower SFC after interesterification, and M10 lipase proved to be more effective than AY30 lipase. The non-interesterified lard had a SFC of 31.3% at 10 °C, which was reduced to 23.8 and 19.9% after interesterification with AY30 lipase and M10 lipase, respectively. The lard and soybean oil blends were affected by the enzymatic interesterification and dilution with soybean oil.

Keywords: *structured lipids; lipase; lard; soybean oil; interesterification.*

Resumo

O objetivo deste trabalho foi avaliar as propriedades físicas das misturas entre banha e óleo de soja modificadas pela interesterificação enzimática catalisada por duas diferentes lipases comerciais, a AY30™ proveniente do microorganismo *Candida cylindracea* e a M10™ proveniente do microorganismo *Mucor circinelloides*. A banha apresentou ponto de amolecimento de 31,8 °C antes da interesterificação e este valor foi reduzido a 29,1 e 28,8 °C após a interesterificação com as lipases AY30™ e M10™, respectivamente. A consistência da banha diminuiu após a interesterificação enzimática com ambas as lipases, e a redução foi maior quando o catalisador utilizado foi a lipase M10™. Este resultado se deve à especificidade das posições *sn*-1,3 da lipase M10™. A banha apresentou menor conteúdo de gordura sólida após a interesterificação, e essa redução foi mais efetiva quando a reação foi catalisada pela lipase M10™. A banha não interesterificada apresentou conteúdo de gordura sólida de 31,3% a 10 °C, que foi reduzido a 23,8 e 19,9% após a interesterificação com as lipases AY30 e M10, respectivamente. A banha e suas misturas binárias com o óleo de soja foram afetadas pela interesterificação enzimática e pela diluição com o óleo de soja.

Palavras-chave: *lipídios estruturados; lipase; banha; óleo de soja; interesterificação.*

1 Introduction

Structured lipids (SLs) are defined as triacylglycerols (TAGs) that have been modified by the incorporation of new fatty acids, restructured to change the positions of fatty acids, or synthesized to yield novel TAGs (AKOH, 2002) aiming at obtaining some desirable properties, such as reduced caloric value or modified melting point. They can also present more favorable characteristics, inducing e.g. the immune system, synthesis of eicosanoids, and anti-inflammatory effects (AKOH; MOUSSATA, 1998). SLs can be produced via interesterification reactions of fats, oils, or mixtures thereof, either chemically or enzymatically.

Chemical interesterification is a random reaction that produces complete randomization of the fatty acid moieties in the triacylglycerol backbones (WILLIS; MARANGONI, 1999; BALCÃO et al., 1996). Currently, under the perspective of cost reduction and large scale application, chemical interesterification seems to be the most attractive method. However, under the perspective of producing lipids with very specific compositions aiming at functional and medical applications, enzymatic interesterification is far more interesting (WILLIS et al., 1998; BALCÃO et al., 1996). With this respect, the enzymatic interesterification has the advantage of allowing a greater control of the positional distribution of fatty acid moieties in the final product due to both selectivity and regiospecificity of lipases (WILLIS; MARANGONI, 1999; BALCÃO; MALCATA, 2002; BALCÃO; MALCATA, 1998a, b, c). Lipases naturally catalyze

Recebido para publicação em 11/2/2008
Aceito para publicação em 3/7/2008 (003171)
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the hydrolysis of triacylglycerols into monoacylglycerols, diacylglycerols, free fatty acids and glycerol, under macroaqueous conditions (BALCÃO et al., 1996). In addition to acylglycerol ester hydrolysis, lipases can also catalyze a wide variety of esterification, transesterification, and polyesterification reactions under microaqueous conditions (BALCÃO et al., 1996). The set of transesterification reactions includes acidolysis, interesterification, and alcoholysis (LEE; AKOH, 1998; BALCÃO et al., 1996). Among the currently available methods for modifying lipids, lipase-catalyzed reactions are better than conventional chemical methods since lipases mimic natural pathways, which concern mild reaction conditions, high catalytic efficiency, and the inherent selectivity of natural biocatalysts. Although lipase-catalyzed reactions have been widely studied as useful tools for modification of plant and fish oils in recent years (SENANAYAKE; SHAHIDI, 1999, 2002; XU et al., 2000; PAEZ et al., 2002), related available studies on animal fats, especially lard, are few although some do exist for butterfat (BALCÃO; MALCATA, 2002; 1998; 1997; SAHIN; AKOH; KARAALI, 2005; YANG et al., 2003; NIELSEN et al., 2006). Typical applications of lipase-catalyzed interesterification reactions include the production of cocoa butter substitutes, human milk fat substitutes, partial acylglycerols, modified fish oil products, margarines, structured lipids, and several lipid products (SAHIN; AKOH; KARAALI, 2005; YANG et al., 2003; NIELSEN et al., 2006). Many different types of lipases have been investigated for the enzymatic modification of oils and fats. Commercial lipases are available from microbial, plant, and animal sources. Among those, microbial lipases are the most attractive ones and their utilization has been described extensively (XU, 2000; BALCÃO et al., 1996).

Regarding the physical characteristics of (structured) lipids (SLs) produced via lipase-catalyzed reactions, it is important to know the thermal characteristics, rheology, crystallization patterns, texture, and appearance of a new SL when determining its suitability for the use in certain food applications (OSBORN; AKOH, 2002). The slip melting point procedure measures the temperature at which a column of fat moves in an open capillary when heated. The drop or slip point of a fat usually occurs at a lower temperature than the melting point because the column of fat will begin flowing at temperatures where 5% solid fat still exists (TIMMS, 1984). Some researchers have used the dropping point method to verify the occurrence of complete interesterification (LIST et al., 1995). However, this method may not provide an accurate measurement for all SLs. ROUSSEAU and co-workers (1996) have found that a linear increase in the proportion of canola oil did not lead to a linear reduction in the dropping point. The Solid Fat Content (SFC) greatly influences the suitability of oils and fats for a particular application. SFC is, in this sense, responsible for many product characteristics including general appearance, ease of packing, organoleptic properties, ease of spreading, and oil exudation. SFC can also be used to study the compatibility of fats by determining the changes in the percentage of solids at different fat proportions (RODRIGUEZ et al., 2001; NOOR LIDA; ALI, 1998). The texture of a SL can be measured via cone penetrometry using an INSTRON™ testing machine or a texture analyzer. Hardness and cohesiveness are important textural parameters for spreadable materials and chocolate products. If a SL is brittle and not spreadable, it will have low cohesion values (OSBORN; AKOH, 2002). Lohman and Hartel (1994) clearly demonstrated a link between SFC of fat and hardness in a "chocolate fat".

Human milk contains nearly 3-5% total lipids. More than 98% of milk lipids are TAGs, 90% of which are composed of fatty acids. The structure of human milk fat is unique since it contains mostly long-chain fatty acids such as oleic acid (~30-35%) followed by palmitic acid (~20-30%) and linoleic acid (~7-14%). Palmitic acid accounts for the highest proportion of saturated fatty acids in human milk fat with 70% of these esterified at the *sn*-2 position of the glycerol backbone; the *sn*-1 and *sn*-3 positions of the TAGs in human milk fat are occupied by unsaturated fatty acids unlike vegetable oils, cow's milk, and infant formulas (XU et al., 2002; INNIS et al., 1995; BALCÃO; MALCATA, 2002). Lard is the only fat that has a structure similar to that of human milk fat.

Numerous studies have been conducted on the production of human milk fat substitutes (MUKHERJEE; KIEWET, 1998; YANG et al., 2003; SCHMID et al., 1999; CHRISTENSEN; HOLMER , 1993; YANG; FRUEKIELD, 2003; SCHMID et al., 1999).

The main goal of this research was to characterize the physical properties of the structured lipids produced by enzymatic interesterification of lard and soybean oil blends catalyzed by commercial lipases from *Candida cylindracea* and *Mucor circinelloides* that could be used as SLs resembling human milk fat.

2 Materials and methods

2.1 Materials

The lard and soybean oil were obtained from local commerce (São Paulo, Brazil). Crude commercial lipases from *Candida cylindracea* (AY30™) and *Mucor circinelloides* (M10™) were kindly supplied by Amano Pharmaceuticals Inc. (Nagoya, Japan), which have shown good performance in previous studies involving lipase-catalyzed reactions (BALCÃO et al., 1996; BALCÃO; MALCATA, 1997; BALCÃO; MALCATA, 1998a). All other reagents and solvents were of analytical or chromatographical grade.

2.2 Methods

Reactant blend preparation

Three binary mixtures of lard and soybean oil were prepared. Lard and soybean oil were blended in 80:20, 60:40 and 50:50 (w/w) proportions. The blends were prepared after complete melting of the fats at 70-80 °C for 30 minutes under magnetic stirring, and the mixtures thus obtained were stored under refrigeration at ca. 4 °C.

Performance of interesterification reactions

The interesterification reactions were carried out at the Bioengineering and Biopharmaceutical Chemistry Research

Group Laboratory (GIBQB) at Fernando Pessoa University (Porto, Portugal). All reactions were carried out in accordance with previous experiments by Balcão and co-authors (BALCÃO et al., 1996; BALCÃO; MALCATA, 1998a,b,c; BALCÃO; MALCATA, 2002). Lipase-catalyzed interesterification reactions were carried out in a magnetically stirred cylindrical glass reactor thermostated at 60 °C. For each reaction with a given lipase, thermal equilibration of the appropriate binary mixture (60 °C) was allowed to proceed for ca. 10 minutes. After that, a known amount of crude lipase powder (ca. 1% (w/w)) was added and the reaction was allowed to proceed under mild (ca. 750 rpm) magnetic stirring for 24 hours. The headspace above the reaction mixture was continuously supplied with (anhydrous) nitrogen. Aliquots were withdrawn at regular intervals of time and assayed for fatty acid composition by gas chromatography. At the end of the reaction timeframe, the remaining interesterified blend was filtered using plain filter paper, bubbled with anhydrous nitrogen, and kept in tight polyethylene flasks at –80 °C.

Determination of fatty acid composition

Fatty acids in the triacylglycerols of the interesterified mixtures were converted into fatty acid methyl esters (FAMES) according to the procedure described by Hartman e Lago (1973). Analyses of FAMES were carried out in a Varian GC gas chromatograph (model 3400CX, from Varian Ind. Com. Ltda., Brazil) equipped with a split-injection port, a flame-ionization detector, and a software package for the system control and data acquisition (model Star Chromatography Workstation version 5.5). Injections were performed in a 30 m fused silica capillary column (ID = 0.25 mm) coated with 0.25 μm of CP-Wax 52CB (Chrompack, Chromtech, Minnesota MN, USA) using helium as carrier gas at a flow rate of 1.5 mL/min and a split ratio of 1:50. The injector temperature was set at 250 °C and the detector temperature was set at 280 °C. The oven temperature was initially set at 150 °C for 5 minutes and then programmed to increase up to 215 °C at a rate of 3 °C/min. After drawing up air into the filled syringe (sample volume 1 μL) and inserting the needle into the heated injector, the samples were injected manually after a dwell-time of ca. 2 s. The qualitative fatty acid composition of the samples was determined by comparing the retention times of the peaks produced after injecting the methylated samples with those of the respective standards of fatty acids. The quantitative composition was obtained by area normalization and expressed as mass percentage according to the AOCS Official Method Ce 1-62 (AOCS, 1997a). All samples were analyzed in duplicate and the reported values are the average of the two runs.

Iodine value

The iodine value was calculated from the fatty acid composition, according to the procedure described in the AOCS Official Method Cd 1-25 (AOCS, 1997b).

Softening point

The softening point of the samples was determined using the open tube melting point method according to the AOCS Official Method Cc 3-25 (AOCS, 1997c). These analyses were performed in triplicate.

Consistency

The consistency C of samples was determined via penetration tests using a 45º acrylic cone fitted to a constant speed Texture Analyzer (model TA-XT2, from Stable Micro Systems). The consistency was calculated as a "yield value" (kgf/cm^2), according to the equation proposed by HAIGHTON (1959) (Equation 1).

$$C = \frac{K \times W}{p^{1.6}} \quad (1)$$

where *C* is the yield value (kgf/cm^2), *K* is a constant depending on the cone angle (4700 –undimensional), *W* is the compression force (kgf), and *p* is the penetration depth (cm). Test parameters were as follows: penetration depth: 1.0 cm; speed: 0.2 cm/s; time: 5 seconds. Samples were heated to 70-80 °C in a microwave oven for complete melting of the crystals and conditioned in 50 mL glass beakers (Pyrex, USA). Tempering was allowed to occur for 24 hours in a common refrigerator (5-8 °C) and then for 24 hours in an oven with controlled temperature (5, 10, 15, 20, 25, and 30 °C ± 0.5 °C). The measurements were performed in triplicate, and the reported values are the simple average of the three values.

Solid fat content (SFC)

The SFC was determined by nuclear magnetic resonance using a 20 MHz Maran Ultra Bench Top NMR (Oxford Instruments, England) according to the AOCS Official Method Cd 16b-93 (AOCS, 1999). Two replicates were performed for every sample, and the reported value is the average of the two values.

Statistical analysis

A multiple regression model was applied to some analytical responses softening point and consistency and solid fat contents (HARE, 1974) represented by the following Equation 2:

$$y = \beta_1 x_1 + \beta_2 x_2 + \beta_{12} x_1 x_2 \quad (2)$$

where: y = variable; β = coefficients generated by multipleregression; and x = proportion of the components. The Statistics 7.0 software – StatSoft was used to generate the coefficients for the model and to present their significance levels, determination coefficients, and variance analysis. Significance was evaluated at 5% (p = 0.05)

3 Results and discussion

3.1 Fatty acid composition

The fatty acid composition and distribution within the triacylglycerols in infant formulas have recently gained much attention. Human milk fat substitutes have been developed to mimic human milk fat composition and structure

(AKIMMOTO et al., 1999; CHRISTENSEN; HOLMER, 1993; YANG et al., 2003).

The fatty acid compositions of lard and soybean oil are in accordance with the results published in the specialty literature (O' BRIEN, 1998; CODEX, 1999; DAVENEL et al., 1999; GLÄSER et al., 2004). The fatty acid composition of lard depends on characteristics such as race, sex, diet, and age of the animal. Therefore, the saturation of lard depends mainly on the amount and composition of the fat supplied to the animal (O' BRIEN, 1998).

The major fatty acids in lard were oleic acid, which accounted for 41% of the total fatty acids followed by palmitic acid (25%), linoleic acid (13%), and stearic acid (13%). Saturated fatty acids of lard constitute about 40% of the total fatty acid.

An adequate intake of the essential fatty acids (viz. linoleic and α-linolenic acids) is very important for the newborn babies. Hence, infant formulas must have a ratio of linoleic acid to α-linolenic acid between 5 and 15 (NIELSEN et al., 2006). In human milk, such ratio is ca. 14.4 (NIELSEN et al., 2006). The data displayed in Table 1 clearly shows that only the samples with more than 40% of soybean oil reached a ratio in this range.

Both the Food and Agricultural Organization/World Health Organization (FAO/WHO) and the European Union Committee recommended a minimum polyunsaturated to saturated fatty acid ratio (PUFA/SFA) of 1. The PUFA/SFA ratio of lard and soybean oil blends ranged from 0.3 to 1.6%. Lard has an intrinsically low content of polyunsaturated fatty acids in its composition, but this ratio could be attained via incorporation of 40% soybean oil (Table 1).

Interesterification does not affect the degree of saturation and does not cause isomerization. Our results suggest that interesterification did not cause a significant alteration in the fatty acid profile of the starting blends.

3.2 Iodine value

The iodine value is a valuable parameter in fat analyses which measure the degree of unsaturation without defining the specific fatty acids. The iodine value of the fat blends under study changed as the function of the proportion of soybean oil and were not affected by interesterification.

3.3 Softening point

The results obtained for the softening point of all samples can be found in Figure 1. Pure lard presented a softening point of 31.8 °C, a value that is in close agreement with the results published in Codex Alimentarius (1999), and which describes a large range of temperatures (28 to 40 °C). After interesterification, the softening point of lard was reduced to ca. 29.1 °C and to ca. 28.8 °C following reactions catalyzed by AY30 and M10 lipases, respectively. The addition of soybean oil promoted a decrease in the softening point, and this was probably caused by the incorporation of polyunsaturated fatty acids present in the soybean oil.

Silva and Co-Workers (2006) reported the softening point for human milk fat as 31.5 °C. In the present research, all fat blends exhibited softening points very close to this value, both before and after the enzymatic interesterification reactions. The interesterified blends displayed lower values for the softening points when compared with blends before enzymatic interesterification. These results are in close agreement with the findings of other researchers, which carried out interesterification of tallow with rapeseed oil catalyzed by a *sn*-1,3-specific lipase (FORSSEL et al., 1992; KOWALSKI et al., 2004). The decrease observed in the softening point of interesterified lard and soybean oil blends was most likely caused by a change in the structure of triacylglycerols. Due to the exchange of fatty acids on glycerol backbone and between triacylglycerols, new triacylglycerols are formed and new interrelations among them can appear. The observed decrease in the softening point was higher after the interesterification reaction catalyzed by M10 lipase, when compared to that catalyzed by the AY30 lipase.

In the case of enzymatic interesterification, small changes in the fatty acids esterified at the *sn*-2 position may stem from acyl migration in the triacylglycerol species during prolonged interesterification times, as reported by Xu et al. (1998).

3.4 Consistency

Texture is a critical factor in determining the functionality and consumer acceptability of table spreads (WRIGHT et al., 2001).

Figure 2 presents the results obtained for the consistency of lard before and after the enzymatic interesterification reactions at temperatures ranging from 10 to 25 °C. In Figure 2 the results obtained for the consistency of the blends of lard and soybean oil can be seen.

The consistency decreased as a function of temperature, and this is due to the gradual melting of crystals, leading to a structurally weaker network (ROUSSEAU; HILL; MARANGONI, 1996), which is, in turn, responsible for the plasticity of fats. For the binary blends, the consistency of non-interesterified samples was higher than those of the interesterified ones for all temperatures studied.

None of the samples analyzed exhibited a measurable consistency at 30 °C. A zero value for the consistency physically represents a product with a very low consistency, and so the analytical equipment is not able to detect it. These products generally present themselves as high viscosity fluids.

Crystal patterns (both polymorphism and morphology) of interesterified lard substantially differed from those of native lard. Differences in the crystal pattern and aggregation behavior could have led to an alteration in the structure of the fat crystal network in lard resulting in altered rheological properties such as the hardness index (MARANGONI; ROSSEAU, 1998).

3.5 Solid fat content (SFC)

The altered composition of triacyglycerols in interesterified lard and soybean oil blends were also reflected in the SFC over the temperature range 10-40 °C. Significant reductions in the

Table 1. Fatty acid composition of lard, soybean oil and their bynary mixtures before and after interesterification.

Fatty acid	Lard (Before)	Lard (AY30)	Lard (M10)	80-20 (Before)	80-20 (AY 30)	80-20 (M10)	60-40 (Before)	60-40 (AY30)	60-40 (M10)	50-50 (Before)	50-50 (AY 30)	50-50 (M10)	Soybean oil (Before)	Soybean oil (AY 30)	Soybean oil (M10)
14:00	1.5 ± 0.02	1.5 ± 0.01	1.5 ± 0.00	1.20 ± 0.00	1.0 ± 0.00	1.1 ± 0.00	0.8 ± 0.12	0.8 ± 0.00	0.8 ± 0.00	1.0 ± 0.04	0.9 ± 0.02	0.8 ± 0.01	-	-	-
16:00	25.4 ± 0.19	25.4 ± 0.04	23.7 ± 0.04	23.0 ± 0.07	22.1 ± 0.03	22.6 ± 0.02	19.3 ± 0.06	19.2 ± 0.07	18.9 ± 0.00	20.5 ± 0.01	20.1 ± 0.09	18.8 ± 0.30	12.1 ± 0.01	12.0 ± 0.03	12.0 ± 0.01
16:01	2.4 ± 0.00	2.3 ± 0.00	2.4 ± 0.00	1.9 ± 0.00	1.7 ± 0.00	1.8 ± 0.00	1.3 ± 0.01	1.3 ± 0.00	1.3 ± 0.00	1.3 ± 0.03	1.2 ± 0.00	1.2 ± 0.03	-	-	-
18:00	13.3 ± 0.05	13.3 ± 0.10	12.7 ± 0.02	11.7 ± 0.01	11.8 ± 0.02	11.7 ± 0.01	9.5 ± 0.30	9.5 ± 0.00	8.9 ± 0.00	8.3 ± 0.00	9.1 ± 0.06	8.8 ± 0.06	4.1 ± 0.02	4.1 ± 0.00	4.1 ± 0.00
18:1n-9	41.4 ± 0.01	41.2 ± 0,08	42.9 ± 0.01	37.0 ± 0.02	37.6 ± 0.04	37.3 ± 0.02	32.8 ± 0.07	33.2 ± 0.02	32.7 ± 0.00	30.5 ± 0.02	31.1 ± 0.09	31.8 ± 0.11	22.1 ± 0.03	22.5 ± 0.02	22.5 ± 0.01
18:1n-9 t	2.7 ± 0.01	2.7 ± 0,04	2.5 ± 0.00	2.4 ± 0.00	2.5 ± 0.02	1.5 ± 0.07	2.1 ± 0.09	2.2 ± 0.00	2.2 ± 0.00	2.0 ± 0.02	2.14 ± 0.00	2.1 ± 0.00	1.5 ± 0.05	1.5 ± 0.00	1.5 ± 0.00
18:2 n-6	13.4 ± 0.00	13.6 ± 0.02	14.4 ± 0.02	21.5 ± 0.04	22.0 ± 0.02	22.7 ± 0.04	31.1 ± 0.00	31.3 ± 0.06	32.7 ± 0.00	33.5 ± 0.03	32.9 ± 0.70	34.1 ± 0.15	55.6 ± 0.02	55.1 ± 0.00	55.1 ± 0.00
18:3n-3	-	-	-	1.3 ± 0.00	1.3 ± 0.01	1.2 ± 0.01	3.1 ± 0.00	2.3 ± 0.01	2.3 ± 0.00	2.4 ± 0.02	2.3 ± 0.07	2.4 ± 0.12	4.6 ± 0.01	4.8 ± 0.01	4.8 ± 0.02
18:2/18:3	-	-	-	16.5	16.9	18.9	10.0	13.4	14.2	13.9	14.3	14.2	12.1	11.5	11.5
P/S	0.3	0.3	0.4	0.6	0.6	0.7	1.1	1.1	1.2	1.2	1.1	1.5	1.6	1.5	1.5
IV	59	59	62	72	74	75	90	89	91	103	100	100	115	115	115

Mean value of two replicates ± standard error of the mean; and P/S – ratio of polyunsaturated/saturated fatty acids; IV – iodine value.

SFC occurred as a consequence of enzymatic interesterification. The SFC profile of lard is displayed in Figure 3.

The SFC of lard substantially decreased by the end of the lipase-catalyzed enzymatic interesterification reaction. When using M10 lipase, the reduction in the SFC was more effective when compared to that promoted by AY30 lipase. Non-interesterified lard exhibited a SFC of 31.3% at 10 °C, which was drastically reduced to 23.8 and 19.9% after the interesterification reactions catalyzed by AY30 lipase and M10 lipase, respectively.

It can also be observed that the addition of soybean oil caused a significant decrease in SFC since soybean oil is liquid oil. When lard was blended with 20, 40, and 50% soybean oil, the SFC at 10 °C was reduced to ca. 23.7, 17.9 and 12.5%, respectively. For the binary blends, the interesterification reaction did not promote significant differences between the lipases utilized.

An increase in the temperature caused a reduction in the SFC in all blends due to the melting profile of the crystals. In their studies, Marangoni and Rousseau (1998) found the same behavior for blends between lard and canola oil. According to the literature (DEMAN et al., 1991; MARANGONI; ROUSSEAU, 1998; DAVENEL et al., 1999), lard presents a SFC of ca. 20% at 25 °C, and this value is close to the SFC obtained in the research effort entertained in this study. Lard and soybean oil blends were affected by both enzymatic interesterification and dilution with soybean oil. Considerable changes in the SFC induced by lipase-catalyzed interesterification occurred, and these were lower for the reactions catalyzed by AY30 lipase.

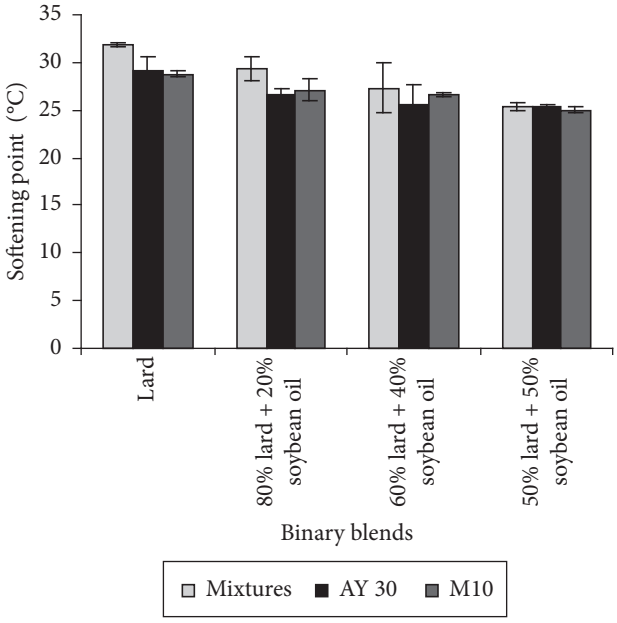

Figure 1. Softening point of lard and soybean and their binary blends before and after enzymatic interesterification.

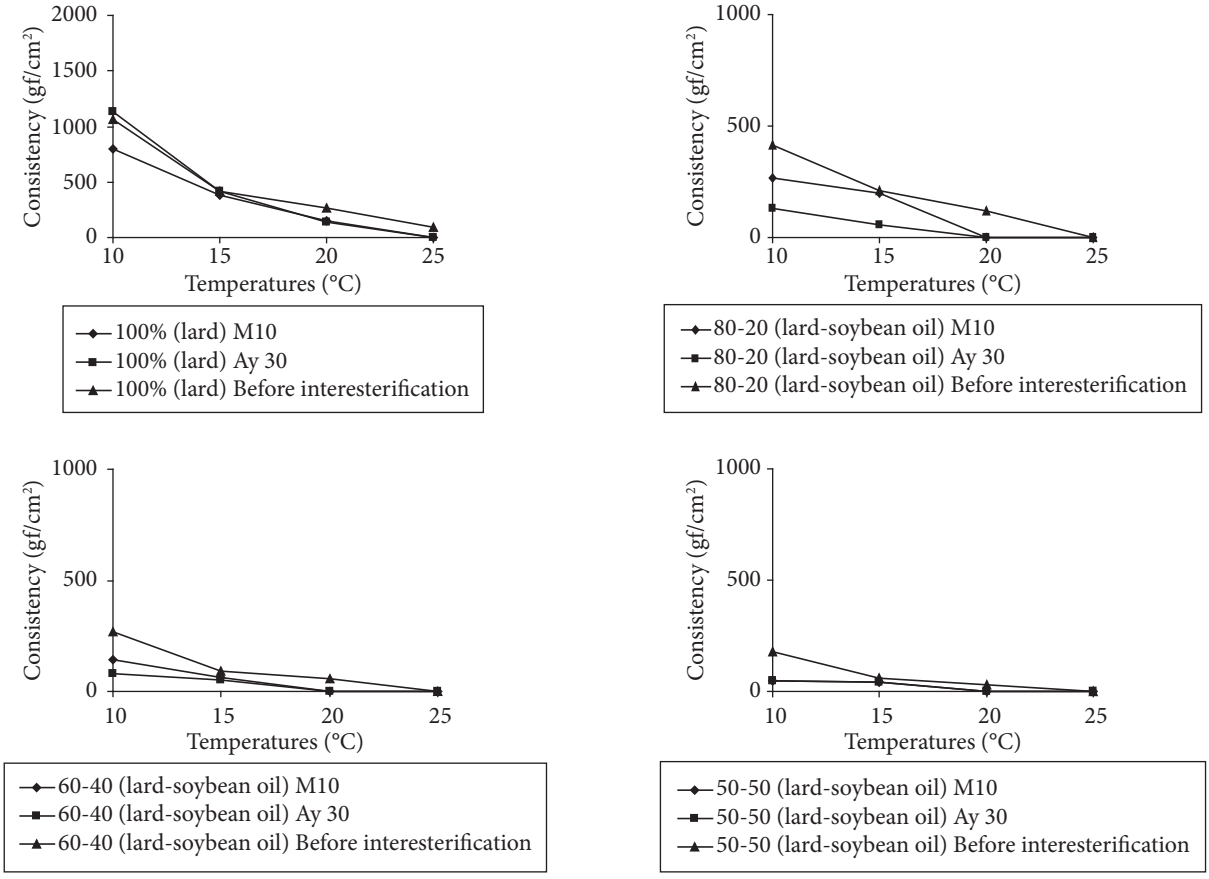

Figure 2. Consistency of lard and their blends with soybean oil before and after interesterification.

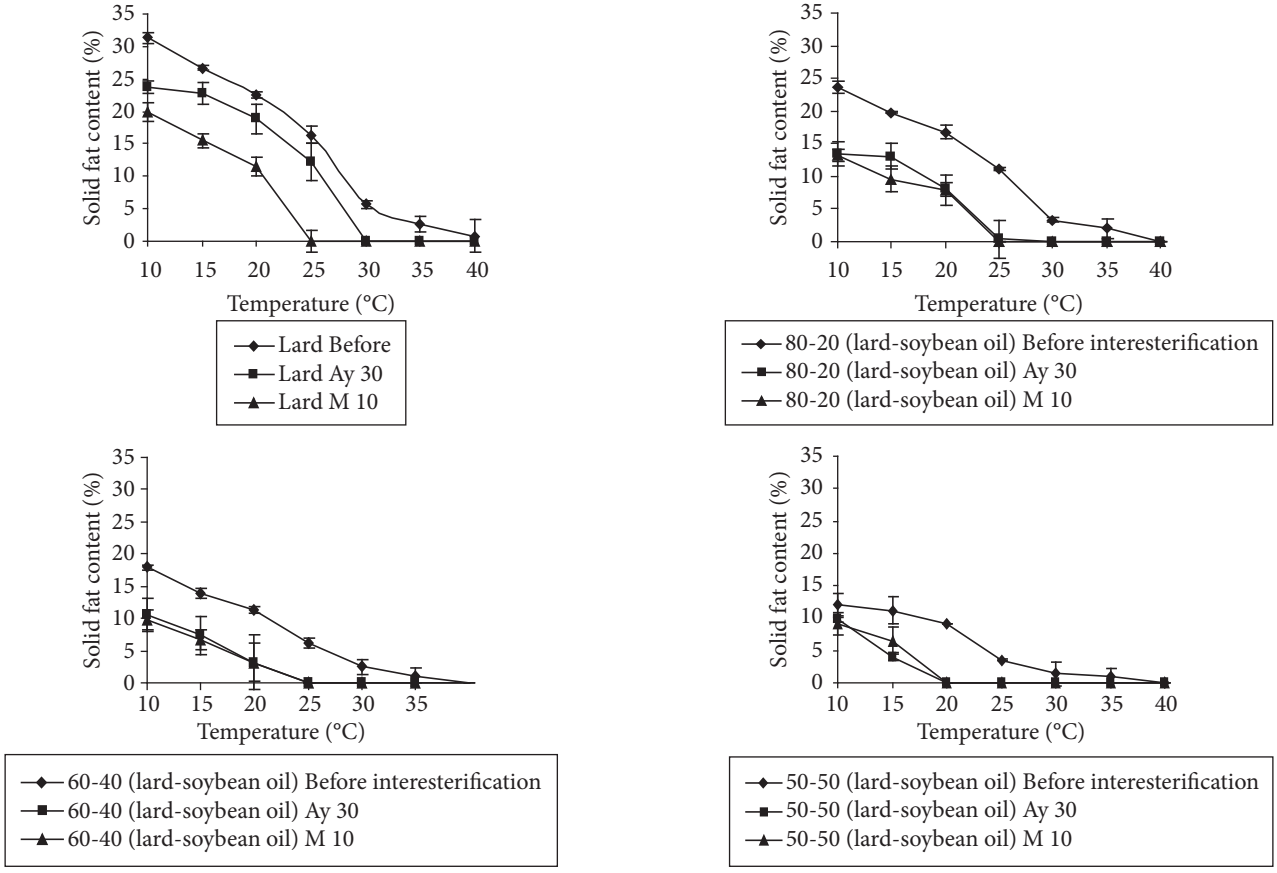

Figure 3. Solid fat content of lard and their blends with soybean oil before and after interesterification.

3.6 Statistical analysis

The statistical analysis reveals that the solid fat content, softening point, and consistency were not dependent on the soybean oil content ($p > 0.05$), but were dependent on the lard content and on the interaction between the two fats ($p < 0.05$). The interaction coefficients β_{12} were negative, showing a monotectic interaction between lard and soybean oil. Interesterified samples presented the same behavior.

4 Conclusions

The addition of soybean oil promoted a generalized decrease in the physical properties of lard, such as softening point, consistency, and solid fat content. The lard and soybean oil blends produced were affected by enzymatic interesterification with a marked decrease in the softening point, consistency, and solid fat content. These reductions were greater for the interesterification reactions catalyzed by the *sn*-1,3-specific M10 lipase.

Acknowledgements

The authors are grateful for the financial support provided by the National Council for Scientific and Technological Development (CNPq) and Brazilian research supporting foundation CAPES, to Victor Manuel Balcão, Fundação Fernando Pessoa, Portugal, for the support provided.

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