ENZYMATIC INTERESTERIFICATION OF FORMULATIONS BASED ON MACAUBA PULP OIL

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ABSTRACT

Enzymatic interesterification is a promising method for obtaining zero trans industrial fats. For many years, fats with high levels of fatty acids were used by the industry, due to their ideal technological characteristics for food formulation, but currently many studies show that the presence of these fatty acids is related to several diseases, leading to the banning of trans fats in several countries. Thus, the objective of this study is to evaluate the triacylglycerol composition and thermal behavior of mixtures after the interesterification of blends of macauba pulp oil and fully hydrogenated macauba pulp oil using the Lipozyme® *TL IM*. Changes were observed after interesterification, such as in the triacylglycerol composition, in which the predominant TAGs were POO (16.09%) in the I90 mixture, while for the I50 mixture the majority TAG was POS (16.52%). Trisaturated TAGs varied from 9.52 to 24.31% and triunsaturated varied from 38.16 to 8.9% among all the samples. The results of the thermal behavior proved that after interesterification were observed changes in the behavior and speed of crystallization and melting in lipid formulations. Therefore, it can be observed that there were changes after interesterification, allowing the obtaining of fats with different functionalities for industrial application.

Keywords: Lipases. Structured lipids. Crystallization.

1 INTRODUCTION

Lipids are essential nutrients in the human diet and promote technological properties in foods. They are mainly composed of triacylglycerols, and have diacylglycerols, monoacylglycerols, free fatty acids, waxes, phospholipids, sphingolipids, glycolipids, terpenes, sterols, tocopherols and carotenoids as minor components.[1,2] In recent years, several studies have proven the negative effects attributed to a diet containing trans fatty acids.[3,4,5,6] Thus, the need for lipid modification methods that meet the demands of the food industry has grown. Currently, structured lipids stand out with great potential, they modify characteristics such as polymorphism, melting point, solid profile, viscosity and consistency and to modifying nutritional properties and have been widely used by the oil and fat industries for the manufacture of margarines, chocolates, infant formulas, food supplements and shortenings due to the replacement of partially hydrogenated fats that contain trans fatty acids.[7,8] With enzymatic interesterification being the process with the greatest potential, with the advantage of minimal generation of byproducts, since lipases are highly regioselective in their reactions.[9] The lipids obtained from this process, called structured lipids, can present adequate physical characteristics without producing trans fatty acids and altering the characteristic regiospecificity of oils and fats.[10]

Macauba (*Acrocomia aculeata*) grows spontaneously in Brazil, being reported in practically all the Brazilian States. It is occurring in greater abundance in Cerrado region and its cultivation is being domesticated. Macauba has several advantages in relation to other oilseed sources, such as high oil productivity, wide adaptability, perennial culture and generation of co-products, such as toxin-free oil and cake.[11] In addition to presenting advantages over palm, which currently represents the most widely used lipid source globally, macauba is adapted to a large part of the national territory and has a productivity potential of more than 20 metric tons fruit/ha/year and 5 ton oil/ha/year, while palm cultivation is restricted to the equatorial region.[12] For these reasons, it is important to study the enzymatic interesterification of macauba oil to obtain new lipid bases with high added value and that meet current demands in legislation and with technological properties for industrial applications. Thus, the objective of this study was to evaluate the physicochemical properties of the lipid bases of macauba pulp oil obtained from the enzymatic interesterification catalyzed by a *sn-1,3* specific lipase in binary blends of OP and OPTH. The following physicochemical properties were analyzed: TAG composition and differential scanning calorimetry (DSC).

2 MATERIAL & METHODS

The presente study was developed at the Fats and Oils Laboratory FEA - UNICAMP. The macauba pulp oil was supplied by the company S. Oleum Soleá Brasil Óleos Vegetais LTDA and physically refined and fully hydrogenated by the company Cargill Agrícola SA. For the enzymatic interesterification reaction, the commercial enzyme Lipozyme® *TL IM*, was supplied by the company Novozymes. For the enzymatic reaction, pulp oil (OP) and fully hydrogenated pulp oil (OPTH) were used and the formulations followed the proportion OP:OPTH (%w/w) of 100:0 (I100); 90:10 (I90); 80:20 (I80); 70:30 (I70); 60:40 (I60) and 50:50 (I50). Interesterification reactions were carried out with the enzyme Lipozyme® *TL IM*. To conditioning the enzyme, soybean oil was used and deaeration was carried out under vacuum, at 70 °C under agitation at 450 rpm, as well as drying at 70 °C under agitation at 450 rpm, under vacuum, with oil replacement of soy every 30 minutes. The exchange process was repeated until a maximum of 0.5% of free fatty acids was obtained in the sample, followed by centrifugation and separation of the oil from the conditioned enzyme. The enzymatic interesterification reaction was carried out for 6h at 70°C, under vacuum and stirring at 350

rpm. Thus, after the mixture (OP:OPTH) reached a temperature of 70 °C, the already conditioned enzyme was added at a percentage of 4%. After 6 hours of reaction, the enzyme was thermally inactivated at 90 °C and followed by filtration, obtaining the interesterified sample.[13] These steps were followed for all formulations. The methods used were triacylglycerol composition (TAG) according to AOCS method Ce 5-86 and thermal behavior by differential scanning calorimetry (DSC) according to the AOCS Cj 1-94 method.[14] All tests were carried out in triplicate and the values were expressed as arithmetic means.

3 RESULTS & DISCUSSION

In the interesterified blends, the majority TAGs varied as the proportion of OPTH (composed of TAGs with high melting point) was increased in the mixtures. Table 1 presents the results for the TAG composition of the mixtures after interesterification, in which the predominant TAGs were POO (16.09%), followed by PLO (13.75%) and OOO (12.71%) for the mixture I90, while for I50 the majority TAG was POS (16.52%), followed by PSS (10.75%) and SOS (9.48%). Figure 1 shows the distribution of TAGs by classes. Among the interesterified samples, it was possible to note that the highest levels were of SUS and SUU TAGs , therefore constituting TAGs with an intermediate melting point, as was expected to occur after interesterification.

Table 1 Triacylglycerol composition of interesterified mixtures

NC	TAG	I100	I90	I80	I70	I60	I50
C48	PPP	1.62	1.18	1.15	1.73	1.71	1.80
C48	PPPoP	1.00	0.58	0.70	0.74	0.66	0.59
C50	PPS	7.9	1.46	2.34	4.68	5.73	7.10
C50	POP	5.56	7.60	7.61	8.17	7.77	6.36
C50	PLP	1.32	4.82	4.55	4.16	3.44	2.78
C50	PPoO	-	1.00	0.90	0.70	0.51	0.40
C52	PSS	-	0.72	1.68	4.09	6.79	10.75
C52	POS	1.23	5.06	8.96	12.77	15.22	16.52
C52	POO	20.13	16.09	14.18	11.28	8.95	6.25
C52	PLS	-	3.01	17.40	5.07	6.79	6.72
C52	PLO	16.44	13.75	2.98	9.08	6.87	5.24
C52	PLL	4.94	4.10	1.80	2.46	1.70	1.20
C54	SSS	-	0.79	2.52	1.38	2.37	4.66
C54	SOS	-	5.56	7.36	5.08	7.33	9.48
C54	SLS	-	12.68	9.14	8.26	7.92	6.32
C54	SOO	1.69	5.12	5.05	6.26	5.38	4.92
C54	OOO	18.00	12.71	6.44	5.87	5.45	5.12
C54	OOL	14.91	3.02	1.53	4.99	2.87	1.84
C54	OLL	5.25	0.61	2.91	1.25	1.41	1.04
C54	LLL	-	0.15	0.81	1.99	1.14	0.90

Abbreviations: NC: number of carbons; Acids - P: Palmitic; L: linoleic; O: Oleic; S: stearic; Po : Palmitoleic.

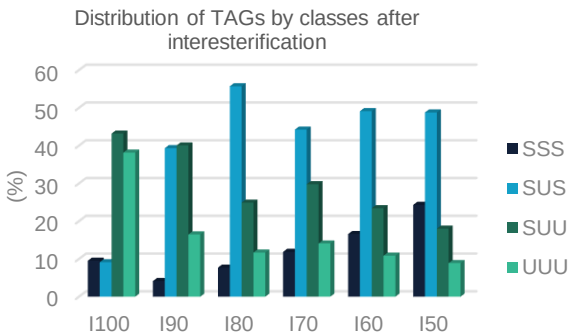

Figure 1 Distribution of TAG classes in interesterified mixtures

SSS: Triacylglycerols trisaturated ; SUS: Triacylglycerols disaturated ; SUU: Triacylglycerols diunsaturated ; UUU: Triacylglycerols triunsaturated

After the enzymatic interesterification process, the crystallization onset temperature of the mixtures decreased. The interesterified mixtures I90, I80, I70, I60 and I50 showed a shift of crystallization peak 1 to the left, which resulted in a lower crystallization onset temperature, with 14; 23.32; 26.87; 30.5 and 34.14 °C, respectively. For the peak 2 the same behavior was observed. The decrease in crystallization temperature was caused by the reduction in the content of trisaturated TAGs. Figures 2 and 3 represents the simple and interesterified mixtures M90 (OP:OPTH simple mixture - 90:10); I90 (OP:OPTH interesterified mixture - 90:10); M50 (OP:OPTH simple mixture - 50:50) and I50 (interesterified mixture OP:OPTH - 50:50) to illustrate the changes occurring after interesterification. Regarding to the crystallization peak temperature, the peaks increased as OPTH was incorporated into the mixtures, as did the final crystallization temperature for crystallization peak temperature 1 and 2. The crystallization enthalpy represents the energy required for crystallization to occur; and with the increase in OPTH in the

interesterified mixtures, an increase in enthalpy for both peaks was observed. The enthalpy ranged from 2.31 to 22.27 (J/g) for the I90 and I50 formulations, respectively, in the peak 1; and 16.71 to 33.93 (J/g) for formulations I90 and I50, respectively, in the peak 2.

For melting, after the enzymatic interesterification process, the presence of two peaks was observed for the mixtures I90 to I50, in which the first melting peak corresponds to the fraction of TAGs with a low melting point and the second peak to the TAGs with a higher melting point. There was an increase in the melting onset temperature and the mixtures I90, I80, I70, I60 and I50 presented, for the peak 1 , temperatures of -37.63 to -9.16 °C, respectively. The Peak 2 presented temperatures varying between 20.81 and 28.69 °C for I90 and I50, respectively. In relation to the peak temperature, there was an increase with the greater proportion of OPTH, as well as an increase in the final melting temperature for peaks 1 and 2. The melting enthalpy decreased, in peak 1 of melting, varying from 58. 23 to 38.12 (J/g) for formulations I90 and I50, respectively. In the peak 2, there was an increase in melting enthalpy, ranging from 18.33 to 41.14 (J/g) for formulations I90 and I50, respectively.

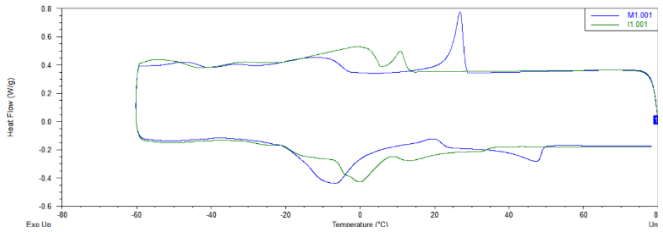

Figure 2 The crystallization and melting profiles obtained by DSC of simple and interesterified mixtures M90 and I90

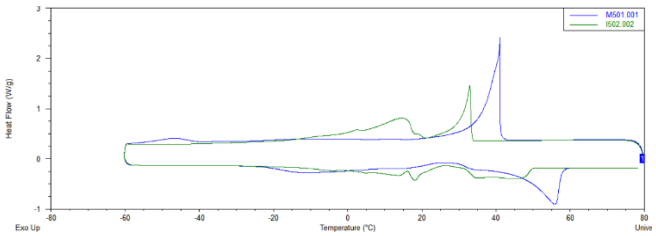

Figure 3 The crystallization and melting profiles obtained by DSC of simple and interesterified mixtures M50 and I50

4 CONCLUSION

After enzymatic interesterification of the OP:OPTH mixtures, there was a reduction in the levels of TAGs UUU and SSS and an increase of the intermediary TAGs SUS and SUU, as well as changes in the melting and crystallization profiles. Therefore, the interesterified lipid bases obtained from macauba pulp oil represent an important alternative for use in the food industry, mainly considering the sustainability of the raw material and process.

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ACKNOWLEDGEMENTS

The authors acknowledge the financial support received from the National Council for Scientific and Technological Development (CNPq), process numbers: 310936/2021-7 and 141039/2021-3, as well as support of Universidade Estadual de Campinas (Unicamp).