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Physico-chemical properties of Brazilian cocoa butter and industrial blends. Part I - Chemical composition, solid fat content and consistency

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RESUMEN

Propiedades físico-químicas de la manteca de cacao de Brasil y mezclas industriales. Parte I - Composición química, contenido de grasa sólida y consistencia.

En este trabajo se presenta un estudio comparativo de las propiedades primarias de mantecas de cacao, representativas de las mezclas industriales, y de la manteca de cacao original de diferentes zonas geográficas de Brasil. Las muestras fueron evaluadas de acuerdo a la composición de ácidos grasos, composición de triglicéridos, distribución de los ácidos grasos en las moléculas de triglicéridos, punto de fusión, contenido de grasa sólida y consistencia. Los resultados permitieron diferenciar las muestras por su composición química, propiedades de resistencia térmica, características de dureza, así como en materia de adecuaciones tecnológicas y los usos potenciales en las regiones de clima tropical.

PALABRAS CLAVE: Composición química – Contenido de grasa sólida – Dureza – Manteca de cacao – Punto de fusión – Resistencia térmica.

SUMMARY

Physico-chemical properties of Brazilian cocoa butter and industrial blends. Part I Chemical composition, solid fat content and consistency .

A comparative study of the primary properties of six cocoa butter samples, representative of industrial blends and cocoa butter extracted from fruits cultivated in different geographical areas in Brazil is presented. The samples were evaluated according to fatty acid composition, triacylglycerol composition, regiospecific distribution, melting point, solid fat content and consistency. The results allowed for differentiating the samples according to their chemical compositions, thermal resistance properties, hardness characteristics, as well as technological adequacies and potential use in regions with tropical climates.

KEYW-ORDS: Cocoa butter – Chemical composition – Hardness – Melting point – Solid fat content – Thermal resistance.

1. INTRODUCTION

Cocoa butter is the most important component used in chocolate and confectionery manufacturing. It is obtained from the cocoa fruit (*Theobroma cacao*), commonly cultivated in equatorial areas, mainly in western Africa, South America, Central America and eastern Asia (Shukla, 2006). Cocoa butter represents the continuous phase of this product, serving as a dispersant matrix for solid cocoa particles, sugar and milk. In addition, it is responsible for several quality attributes of the final product, such as hardness and brittleness at room temperature (*snap*), brightness, contraction during demolding and complete melting at mouth temperature with rapid flavor release at taste. It is the most expensive ingredient in chocolate manufacturing, corresponding to 25 to 36% of the cost of the finished product (Timms, 2003).

Cocoa butter presents a unique chemical composition, responsible for its unique sensorial attributes. The types of fatty acids and their positional distribution of triacylglycerol (TAG) molecules determine the physical properties of cocoa butter and its general characteristics for industrial use. The main fatty acids found in cocoa butter are palmitic, stearic and oleic acids, whose levels correspond to approximately 25, 36 and 35% of total the fatty acid content, respectively. The TAGs in cocoa butter are predominantly composed of 1,3-dipalmitoyl-2-oleoyl-glycerol (POP), 1-palmitoyl-2-oleoyl-3-estearoyl-glycerol (POSt) and 1,3-diestearoil-2-oleoyl-glycerol (StOSt) and the oleic acid occupies, preferibly, the *sn-2* position of the structure. This relatively uniform triacylglycerol composition confers a thermal and structural behavior to cocoa butter which is comparable to a pure compound (Subramaniam, 2000).

Interrelationships among the chemical compositions and physical properties of cocoa butter exert a great influence on the quality and acceptance of chocolate and confectionery products. Sensorial attributes, shelf life and stability during storage are directly dependent on the chemical composition and properties of the fat phase. Therefore, the solid profile, consistency, microstructure, polymorphism and melting and crystallization behaviors of cocoa butter are reflected as primary factors related to its

functionality for industrial applications (Norberg, 2006; Awad and Marangoni, 2006).

Variations in cocoa butter composition can result in unacceptable physical properties, such as excessive softness and high susceptibility of the final product to *fat bloom*. In addition to these drawbacks, difficulties during processing (especially at tempering) or incompatibility of the fat phase with the climate conditions of the producing and consuming areas are also consequences of slight changes in the TAG profile (Chaiseri and Dimick, 1995; El-Mallah and Megahed, 1998).

Studies indicate that the fatty acid profile and TAG composition of cocoa butter and its ascribed properties, such as melting point, solid profile and consistency, depend on geographical areas of culti-vation and their different climate conditions, espe-cially the temperature (Gunstone and Harwood, 2007; Tucci *et al.*, 1996). Data on chemical compo-sition reveal that cocoa butter produced in South America contains levels of oleic acid significantly higher than those from Africa and Asia. Studies re-veal that Brazilian cocoa butter presents lower mo-nounsaturated TAG and higher diunsaturated TAG contents, when compared to species produced in other countries. As a result, its crystallization behavior is, more often, unsatisfactory (Lipp and Anklam, 1998). In Brazil, however, regional climate differences and transport/storage conditions require the use of cocoa butter of superior hardness and with high thermal resistance. A commercial, although partial solution in practice for this problem is the use of cocoa butter blends obtained from mixtures of the almonds from several countries, in order to get a more consistent product (Timms, 2003). The fundamental physico-chemical characteristics of these industrial *blends* and their performance in relation to the cocoa butter produced in Brazil are not disclosed in the scientific literature, although the expressive national volume of production of this raw material represents a strategic product from an economic point of view.

The objective of this work was to perform a comparative study of the chemical and physical attributes of six different cocoa butter samples. Three different industrial standard blends and cocoa butter samples obtained from trees cultivated in three different Brazilian regions were investigated. The samples were characterized according to their fatty acid composition, triacylglycerol composition, regiospecific distribution, melting point, solid fat content and consistency.

2. MATERIALS AND METHODS

2.1. Material

Raw-materials. Samples from three different Brazilian industrial blends were evaluated. The samples were obtained directly from the processing plants of cocoa almonds and represent industrial standard cocoa butter commercially available in Brazil, composed of mixtures of cocoa butter from different geographic origins to ensure the constant physical properties required for use in tropical areas. The industrial standard samples named Blend A, Blend B and Blend C were provided by three different industries, which, together, are responsible for the processing of more than 70% of the cocoa beans in Brazil (Codevasf, 2009). The geographical origin of the almonds and the blending proportions, however, were not reported by the suppliers. Three national cocoa butter samples originating from different Brazilian states (Bahia, Amazonas and Espírito Santo) were selected and labelled Braz A, Braz B and Braz C, respectively. These samples were obtained from regional producers, between January-April 2010, through a Brazilian agency responsible for regional cocoa production politics. The samples represent the harvest in the three main microclimates for cocoa farming in Brazil (Codevasf, 2009). All cocoa butter samples were produced by cold pressing and deodorized. They were stored in polyethylene jars in a cold chamber (10 °C), until use.

2.2. Analytical Methods

Fatty acid composition. The Analyses of fatty acid composition were performed in duplicate in a capillary gas chromatograph (CGC Agilent 6850 Series GC System, Santa Clara, California, USA), after esterification using Hartman and Lago's method (Hartman and Lago, 1973).The fatty acid methyl esters were separated according to AOCS Ce 2-66 method (AOCS, 2004) in a DB23 Agilent capillary column (50% cyanopropyl-methylpolysiloxane), with the following dimensions: length: 60m, internal diameter: 0.5 mm and 0.25 mm film thickness. Analysis conditions: oven temperature program: 110 °C for 5 minutes, 110 °C-215 °C (5 °C/min), 215 °C for 24 minutes; detector temperature: 280 °C; injector temperature: 250 °C; carrier gas: helium; split ratio 1:50; injected volume: 1.0 µL. The qualitative composition was determined by comparing peak retention times with the respective standards for fatty acids. The quantitative composition was performed by area normalization, expressed as mass percentage. The average of two injections is reported.

Triacylglycerol composition. An analysis of the composition of TAGs was performed in a CGC Agilent 6850 Series GC System (Santa Clara, California, USA) capillary gas chromatograph. A DB17HT Agilent Catalogue: 122-1811 capillary column (50%-phenyl-methylpolysiloxane, with 15 m length × 0.25 mm internal diameter, coated with film of 0.15 µm) was used. Conditions: split injection, ratio of 1:100; column temperature: 250 °C, programmed to 350 °C at the rate of 5 °C/min; carrier gas: helium at a flow rate of 1.0 mL/min; injector temperature: 360 °C; detector temperature: 375 °C; injected volume: 1.0 mL; sample

concentration: 100 mg in 5mL of tetrahydrofurane. TAG groups were identified by comparing retention times, following the procedures of Antoniosi Filho *et al.* (1995). The analyses were performed in duplicate for each sample.

Regiospecificdistribution. Nuclear magnetic resonance spectroscopy, the high-resolution carbon-13 (^{13}C RMN) of acyl chains of TAGs was used as a quantitative method for the regiospecific analysis of cocoa butter samples (Silva *et al.*, 2010).The analyses were performed in a Bruker Advance DPX 300 Nuclear Magnetic Resonance spectrometer (RMN)(Silberstreifen, Rheinstetten, Germany). The determination of ^{13}C was made at 75.8 MHz frequency with a 5 mm-multi nuclear probe operating at 30 °C, according to Vlahov's methodology (Vlahov, 1998; Vlahov, 2005). Results are given as saturated, oleic and linoleic fatty acid composition at *sn-2* e *sn-1,3* positions of TAGs. Analyses were performed in triplicate.

Melting point. The melting temperature was determined in duplicate for each sample, using an open capillary tube, according to AOCS procedures, Cc 3-25 Method (AOCS, 2004). Analyses were performed in triplicate.

Solid fat content. Solid fat content profiles were determined using Nuclear Magnetic Resonance (NMR) spectrometry (Bruker pc120 Minispec, Silberstreifen, Rheinstetten, Germany) and TCON 2000 high precision dry baths (0–70 °C) (Duratech, Carmel,USA), according to the AOCS Cd 16b-93 method: direct method, sample reading in series at temperatures of 10, 15, 20, 25, 30, 35 and 40 °C, with tempering for cocoa butterand/or special fats (AOCS, 2004). The analyses were performed in duplicate for each sample.

Consistency. The consistency was determined with the TA-XT Plus Texture Analyzer equipment (Stable Micro Systems, Surrey, UK), controlled by a microcomputer. The samples were warmed an oven at 60 °C, to completely melt the crystals, then placed in beakers of 50 mL. Conditioning was performed in an incubator for 24 hours at 5 °C to induce cocoa butter crystallization and then for 24 hours at temperature readings: 20, 25 and 30 °C. An acrylic cone with an apex angle of 60° and no truncation was used in the experiments. Tests were operated in the following conditions: displacement in the sample equal to 10 mm; probe velocity of 2 mm/s; penetration time of 5 s (Campos, 2005; Rodrigues *et al.*, 2007). From these conditions, compressive force in g_f was obtained. Penetration data were converted into y*ield value*, according to Haighton (1959):

$$C = \frac{K \times W}{p^{1,6}}$$

Where: C = *yield value*, in g_f/cm^2 ; K = factor depending on cone angle (equal to 2815 for a cone

of 60°); W = compression force in (g_f); p = penetration depth (mm/10). Samples were analyzed in triplicate and the results correspond to the calculated average.

2.3. Statistics Analysis

The statistical analyses were based on the Analysis of variance (ANOVA) and TukeyTest to determine the significant differences of the means at a probability level of 5% ($p < 0.05$) using Statistica 8.0 (Statsoft, USA).

3. RESULTS AND DISCUSSION

3.1. Fatty acid composition

Table 1 shows the fatty acid composition obtained for all six cocoa butter samples. The results express the average of two determinations and are in accordance with the limits found in the literature, which indicate the following characteristic range for palmitic, stearic and oleic acids: 24-27%, 34-36% and 32-35%, respectively (Subramaniam, 2000; Awad and Marangoni, 2006; Gunstone and Harwood, 2007). These fatty acids were the predominant species in all samples, with the total varying from 93.4 to 95.2%. According to Shukla (2006), differences in fatty acid composition of cocoa butter can result in iodine values varying from 34 to 40. Therefore, the observed values for the analyzed samples are in agreement with the technical literature. Blend C showed the highest amount of saturated fatty acids (63.9%), while the lowest content was verified for Braz C sample (62.4%).

The different samples presented similar contents of each fatty acid. In general, Brazilian cocoa butter samples showed slightly superior contents of oleic and linoleic acids compared to the values found for the industrial blends. In a detailed study, Lipp *et al.* (2001) characterized the fatty acid composition of cocoa butter samples originating from several countries. South American cocoa butter presented linoleic acid contents higher than 3%; Asian cocoa butter showed percentages inferior to 2.5% and African samples had values between these two limits. The authors emphasized that very small differences, particularly with respect to the linoleic acid content, reflect the geographical origin of the cocoa butter. This information corroborates the current study. The lower percentages of linoleic acid obtained for the blends are possibly due to the mixtures of cocoa butter from other countries.

In accordance with the literature, Braz B and Braz C samples presented the lowest percentages of palmitic acid among the samples, which attributes a lower content of this fatty acid to Brazilian cocoa butter when compared to cocoa butter produced in Ghana, The Ivory Coast and Malaysia (Gunstone and Harwood, 2007).

Table 1
Fatty acid composition (%) of the cocoa butter samples

Fatty acid (%)	Blend A	Blend B	Blend C	Braz A	Braz B	Braz C
C14:0 - myristic	0.1 ± 0.0	0.1 ± 0.0	0.1 ± 0.0	0.1 ± 0.0	0.1 ± 0.0	0.1 ± 0.0
C16:0 - palmitic	25.8 ± 0.1	25.3 ± 0.1	26.7 ± 0.0	25.6 ± 0.2	24.3 ± 0.1	24.8 ± 0.0
C16:1 - palmitoleic	0.3 ± 0.0	0.3 ± 0.0	0.3 ± 0.0	0.3 ± 0.0	0.2 ± 0.0	0.3 ± 0.0
C17:0 – margaric	0.2 ± 0.0	0.2 ± 0.0	0.2 ± 0.0	0.2 ± 0.0	0.3 ± 0.0	0.3 ± 0.0
C18:0 - stearic	35.6 ± 0.3	36.1 ± 0.3	35.6 ± 0.1	35.3 ± 0.2	35.4 ± 0.4	35.1 ± 0.1
C18:1- oleic	33.4 ± 0.1	33.5 ± 0.1	33.0 ± 0.2	33.9 ± 0.2	33.7 ± 0.3	33.9 ± 0.2
C18:2 - linoleic	3.0 ± 0.1	2.9 ± 0.1	2.7 ± 0.0	3.1 ± 0.1	3.2 ± 0.0	3.2 ± 0.1
C18:3 - linolenic	0.2 ± 0.0	0.2 ± 0.0	0.2 ± 0.0	0.2 ± 0.0	0.2 ± 0.0	0.2 ± 0.0
C20:0 - arachidic	1.1 ± 0.0	1.2 ± 0.0	1.2 ± 0.0	1.1 ± 0.0	2.1 ± 0.1	1.8 ± 0.1
C20:1 - gadoleic	–	–	–	–	0.1 ± 0.0	0.1 ± 0.0
C22:0 - behenic	0.2 ± 0.0	0.2 ± 0.0	0.2 ± 0.0	0.2 ± 0.0	0.4 ± 0.0	0.3 ± 0.0
C24:0 - lignoceric	0.1 ± 0.0	0.1 ± 0.0		–	–	0.1 ± 0.0	0.1 ± 0.0
Σ Saturated	63.1 ± 0.0[a]	63.2 ± 0.0[a]	63.9 ± 0.1[b]	62.6 ± 0.0[c]	62.6 ± 0.0[c]	62.4 ± 0.0[d]
Σ Unsaturated	36.9 ± 0.1[a]	36.8 ± 0.0[a]	36.1 ± 0.1[b]	37.4 ± 0.1[c]	37.4 ± 0.1[c]	37.6 ± 0.1[d]

-: not detected. Means ± standard deviation of the replications. Means followed by the same letter within a line indicate significant difference (p < 0.05) by Tukey's test.

The percentages of saturated and unsaturated fatty acids, for samples Blend A and Blend B and for samples Braz A e Braz B showed no significant difference (p < 0.05) among themselves, according to the Tukey Test.

3.2. Triacyglycerol composition

More recent studies on the variation of the chemical compositions of cocoa butter have been focused on TAG composition analyses (Timms, 2003). The results obtained for the six samples are shown in Table 2. For Brazilian samples, 12 different TAGs were found. The industrial blends indicated the presence of 11 TAG species, since POL was not detected. POP, POSt and StOSt were predominant, ranging from 79.8 to 87.4% of the total TAG composition, in accordance with literature information, which indicates a range of 80 to 85% for these symmetrical compounds of saturated-unsaturated-saturated type (Norberg, 2006; Lipp et al., 2001). The main asymmetrical TAGs were the PStSt, POO and StOO species.

The industrial blends showed more POP and POSt than the Brazilian samples, although StOO contents were higher in the national samples. According to Lipp and Anklam (1998), higher percentages of POO and StOO characterize a cocoa butter of superior softness. Higher POO and POL percentages, particularly, were observed for the Braz C sample from Espírito Santo. Since these TAGs correspond to two out of three species of major unsaturetion degrees found in cocoa butter samples, it is possible to assume that this peculiarity in its chemical composition favors differentiated solidification behavior compared to the other samples.

Considering only the TAG species of "saturated fatty acid – oleic acid – saturated fatty acid" type, the Brazilian samples contained percentages ranging from 81.3 to 85.2%; for the blends, these values lay between 87.1 and 88.5%. According to Gunstone and Harwood (2007), this parameter can be used as an indication of the cocoa butter origin and the values presented in Table 2 indicate lower amounts in Brazilian raw materials.

In general, the physical properties of a fat are more easily interpreted using the classification of the TAGs in groups according to their unsaturation degree: S_3 (trisaturated), S_2U (disaturated-mo-nounsaturated), U_2S (monounsaturated-diunsaturated) and U_3 (triunsaturated), instead of the individual TAG species (Neff et al., 2001). For all cocoa butter samples studied, S_3, S_2U and U_2S types were found, but no U_3 was detected. The origin of the samples of cocoa butter show to be statistically significant (p < 0.05) for the different classes of triacylglycerols, according to Table 2.

S_3 varied from 1.4 to 3.1% among all the samples, in accordance to published data for cocoa butter of different origins (Loisel et al., 1998). Higher S_2U contents were observed for the industrial blends while Brazilian samples were characterized by higher percentages of U_2S (from 6.8 to 10.3%), due to the major contributions of POO, POL and StOO species. Lonchampt and Hartel (2004) and Shukla (2006), analyzing cocoa samples from different origins, found that Brazilian cocoa butter showed the highest content of U_2S, which corroborates the results here presented and also confirms that cocoa butter from other countries were used in the production of the industrial standard blends.

Table 2
Triacylglycerol (TAG) composition (%) of samples, for individual TAGs and TAG classes

TAG (%)	Blend A	Blend B	Blend C	Braz A	Braz B	Braz C
PPSt	0.2 ± 0.0	0.9 ± 0.1	0.9 ± 0.0	0.9 ± 0.0	0.2 ± 0.0	0.5 ± 0.0
POP	20.0 ± 0.2	17.3 ± 0.2	21.6 ± 0.5	19.0 ± 0.1	18.4 ± 0.2	17.3 ± 0.0
PLP	2.2 ± 0.1	1.7 ± 0.0	2.0 ± 0.1	2.3 ± 0.1	1.9 ± 0.0	1.6 ± 0.1
PStSt	1.6 ± 0.2	0.5 ± 0.0	2.2 ± 0.1	1.9 ± 0.3	0.4 ± 0.0	1.6 ± 0.1
POSt	43.8 ± 0.2	41.5 ± 0.1	43.1 ± 0.2	41.2 ± 0.2	39.4 ± 0.3	40.8 ± 0.4
POO	3.2 ± 0.1	3.0 ± 0.0	3.4 ± 0.1	4.7 ± 0.3	2.1 ± 0.0	5.2 ± 0.2
PLSt	3.7 ± 0.0	3.1 ± 0.1	3.4 ± 0.1	2.9 ± 0.0	2.6 ± 0.0	4.2 ± 0.2
POL	–	–	–	0.3 ± 0.0	0.3 ± 0.0	0.9 ± 0.0
StOSt	22.8 ± 0.3	26.6 ± 0.3	21.4 ± 0.4	21.5 ± 0.6	27.2 ± 0.5	21.7 ± 0.2
StOO	1.5 ± 0.0	2.9 ± 0.2	1.0 ± 0.0	3.8 ± 0.1	4.3 ± 0.1	4.1 ± 0.1
StLSt	–	1.4 ± 0.0	–	1.2 ± 0.0	2.0 ± 0.0	0.6 ± 0.0
StOA	1.0 ± 0.0	1.1 ± 0.1	1.1 ± 0.0	0.3 ± 0.0	1.3 ± 0.0	1.5 ± 0.1
$\Sigma\,S_3$	1.8 ± 0.1^a	1.4 ± 0.1^b	3.1 ± 0.1^c	2.8 ± 0.0^d	1.6 ± 0.0^e	2.0 ± 0.0^f
$\Sigma\,S_2U$	94.5 ± 0.1^a	94.8 ± 0.2^b	93.0 ± 0.4^c	88.4 ± 0.4^d	91.7 ± 0.5^e	87.7 ± 0.4^f
$\Sigma\,U_2S$	3.7 ± 0.0^a	3.9 ± 0.0^b	3.8 ± 0.3^c	8.8 ± 0.1^d	6.8 ± 0.4^e	10.3 ± 0.2^f

P = palmitic, St = stearic, O = oleic, L = linoleic and A = arachidic acids. S_3 = trisaturated,
S_2U = disaturated-monounsaturated and U_2S = monounsaturated-diunsaturated triacylglycerols. -: not detected.
Means $\pm$ standard deviation of the replications. Means followed by the same letter within a line indicate significant
difference ($p < 0.05$) by Tukey´s test.

U_2S, with melting points between 27 and 42 °C, are responsible for the structuring of the products containing cocoa butter as the fat base. The contribution of U_2S is also important when considering their sensorial properties at room temperature. As a consequence, the ratio between S_2U and U_2S amounts in cocoa butter is selectively associated with an increase in thetechnological functionality or improved ability of the sensorial attributes of this raw-material (Bessler and Orthoefer, 1983). According to Lipp and Anklam (1998), cocoa butter with lower S_2U contents and higher U_2S levels, as for example the Brazilian samples evaluated in this study, can lead to unsatisfactory crystallization characteristics. However, the technical literature does not indicate the limits of these TAG classes suitable for adequate crystallization behavior in different climate regions.

3.3. Regiospecific distribution

A detailed knowledge of the chemical composition of cocoa butter should also contemplate the determination of the regiospecific distribution of the TAGs, which pinpoints the fatty acid substitution in the three positions of the glycerol chain, recognized as *sn-2* and *sn-1,3* (Segall *et al.*, 2005). The method is based on the detection of resonances of the carbonyl carbon groups of the fatty acids in the TAG molecule (Jie and Mustafa, 1997; Vlahov, 1998; Silwood and Grootveld, 1999).

The fatty acid distribution at the *sn-2* and *sn-1,3* positions of the cocoa butter samples are shown in

Table 3. One of the most peculiar composition characteristics of cocoa butter is the major presence of 2-oleyl-glycerol TAGs of palmitic and stearic acids, which occur as symmetrical molecules containing one unsaturated fatty acid at the central position. Information on quantitative differences in this distribution, for the same kind of sample, is rarely found in the literature (Segall *et al.*, 2005; Wang *et al.*, 2006). The results in Table 3 indicate that, for cocoa butter, the *sn-2* position is predominantly characterized by thepresence of unsaturated fatty acids, especially oleic (C18:1) and linoleic (C18:2) acids. The method did not detect any saturated fatty acid for this position. A hypothesis to be considered in this case is that the method of analysis by ^{13}C RMN is not sensitive enough to detect small amounts of saturated fatty acids. Oleic acid percentages for this position varied from 86.4 to 91.7%, and ranged from 8.3 to 13.6% for linoleic acid. The highest percentages of linoleic acid were observed for two national samples (Braz A and Braz B). For blends, however, these contents were similar. For the *sn-1,3* positions, the presence of saturatec fatty acid was dominant, ranging from 91.7 to 96.1%, and can be assigned to the sum of palmitic and stearic acids, according to Tables 1 and 2. Small contents of oleic acid were also observed in the *sn-1,3* position, but linoleic acid was not detected. These findings confirm the TAG composition given in Table 2, where this fatty acid was found in very small amounts (Brazilian samples) or not detected at all (blends) at these positions in the TAGs. All samples were statistically

Table 3
Fatty acid distribution (%) at *sn-2* e *sn-1,3* positions
of triacylglycerols, for the cocoa butter samples

Sample	sn - 2 (%)		sn -1,3 (%)	
	C 18:1	C18:2	Saturated	C18:1
Blend A	91.0 ± 0.5[a]	9.0 ± 0.5[a]	95.4 ± 0.4[a]	4.6 ± 0.4[a]
Blend B	91.7 ± 0.1[b]	8.3 ± 0.1[b]	96.1 ± 0.1[b]	3.9 ± 0.2[b]
Blend C	90.4 ± 0.0[a]	9.6 ± 0.1[c]	93.6 ± 0.1[c]	6.4 ± 0.1[c]
Braz A	86.4 ± 0.0[c]	13.6 ± 0.0[d]	91.7 ± 0.0[d]	8.3 ± 0.0[d]
Braz B	91.1 ± 0.1[b]	8.9 ± 0.2[a]	94.6 ± 0.3[e]	5.4 ± 0.4[e]
Braz C	89.6 ± 0.1[d]	10.4 ± 0.1[e]	95.8 ± 0.1[f]	4.2 ± 0.1[f]

Means ± standard deviation of the replications. Means followed by the same letter within a column indicate significant difference (p < 0.05) by Tukey´s test.

distinguishable (p <0.05) in term of the distribution of fatty acids at the *sn-1,3* position, confirming the statistical results presented in Table 2.

Although studies on the regiospecific distribution of cocoa butter are scarce in scientific literature, the findings obtained in the present study show agreement with the results published by Takagi and Ando (1995), through chiral phase liquid chromatography. They found oleic and linoleic acid percentages equal to 87.5 and 7.4%, respectively for the *sn-2* position . For the *sn-1,3* positions, the saturated fatty acid content corresponded to 95.3%, while for oleic acid a content of 3.9% was found. Unfortunately, the authors did not report on the origin of the cocoa butter evaluated. Kosugi *et al.* (2004) reported a collaborative action among laboratories to study the regiospecific distribution of cocoa butter, using several lipases. For the *sn-2* position, linoleic acid contents ranged from 7.9 to 10.0%; for oleic acid, the values varied from 81.6 to 86.9%, close to the results obtained in the present study. The use of ^{13}C NMR spectroscopy, therefore, proved to be an effective tool for the determination of the regiospecific distribution of cocoa butter TAGs, and in combination with the TAG composition found, it allowed researchers to validate 1,3-dipalmitoyl-2-oleoyl-glycerol (POP), 1-palmitoyl-2-oleoyl-3-stearoyl-glycerol (POSt) and 1,3-diestearoil-2-oleoyl-glycerol (StOSt) as the main TAGs in cocoa butter.

3.4. Melting point

According to Shukla (2006), cocoa butter is characterized by a narrow melting temperature range (32 to 35 °C) due to its relatively uniform chemical composition. The melting points determined for the six samples are presented in Table 4. Values varied from 33.0 to 33.4 °C for the Brazilian cocoa butter samples and were higher for the blends (34.3 to 34.6 °C). The sample Blend C showed no significant difference (p < 0.05) compared to the other samples of blends. Among the Brazilian samples, only sample Braz C differed statistically (p < 0.05) from the others in relation to the melting point.

These results show good correlation with the behavior expected from the TAG compositions (Table 2). The mathematical relationships between the melting point and the levels of S_2U and U_2STAGs for the samples were linear and given by the equations: S S_2U (%) = 4.1483*mp (°C) –48.666; R^2 = 0.8574; and S U_2S (%) = –4.0785*mp (°C) + 144.21; R^2 = 0.9253; where mp = melting point.

S_2U, was represented mainly by the POP, POSt and StOSt species, which have melting points from 35.3 to 41.2 °C. The melting point of U_2S triacylglycerols, however, is significantly lower. For the StOO and POO species, which make up almost all of this TAG class in the evaluated samples, the melting points are, respectively, 23.7 and 19.2 °C, so that these components are liquid at room temperature (Bockisch, 1998). The increase inU_2S triacyglycerol contents, therefore, contributes to a decrease in the melting point of cocoa butter. The melting points for the Brazilian samples are lower, according to the increase in the percentage of the U_2S class. Braz A, Braz B and Braz C samples, with melting points of 33.3; 33.4 and 33.0 °C, showed U_2S contents equal to 8.8; 6.8 and 10.3%, respectively. Likewise, the industrial blends with U_2S percentages varying from 3.7 to 3.9% showed

Table 4
Melting point (°C) of the cocoa butter samples

Melting point (°C)					
Blend A	Blend B	Blend C	Braz A	Braz B	Braz C
34.3 ± 0.1[a]	34.6 ± 0.1[b]	34.4 ± 0.1[a,b]	33.3 ± 0.0[c]	33.4 ± 0.3[c]	33.0 ± 0.0[d]

Means ± standard deviation of the replications. Means followed by the same letter within a line indicate significant difference (p < 0.05) by Tukey´s test.

thehighest melting points, since these samples also have lower contents of TAGs with a lower melting point. The better technological performance of the industrial blends, in comparison to the cocoa butter produced in Brazil, can be attributed to this characteristic. The chocolates produced with the blend show superior thermal resistance, required in regions of hotter climates and with temperature fluctuations during storage and transportation.

3.5. Solid fat content

The solid fat content (SFC) is a parameter that expresses the solid/liquid mass relation of a fat at different temperatures, affecting physical properties like consistency and stability and also important sensorial attributes (Beckett, 2008). Figure 1 presents the solid curves of cocoa butter samples and two distinct behaviors can be distinguished. All the industrial blends and the sample Braz B, showed very similar solid profiles. At 10 °C, the SFC of these samples corresponded to 85.4; 84.5; 83.9 and 82.5%, for Blend C, Blend A, Blend B and Braz B samples, respectively. Braz A and Braz C samples were characterized by lower SFC values, and at 10 °C they were 77.4% (Braz A) and 76.6% (Braz C). At 25 °C, the SFC for the first group ranged from 58.9 to 64.7%; for Braz A and Braz C samples, however, the SFC was 53.7% and 52.3%, respectively. At this temperature, regarding the SFC values, only the samples Blend A and Blend B did not differ statistically from one another($p < 0.05$).

In general, the SFC values at a specific temperature for this fat increase according to S_2U levels, the predominant TAG class in cocoa butter (Shukla, 2006). This correlation could explain the differences found for Braz A and Braz C samples, which had S_2U contents lower than 90%. Blend C had a lower S_2U content compared to Blend A and Blend B, the later with higher SFC. Since this sample presented a high content of S_3 (3.1%), represented by the PPSt and PStSt species, with melting points significantly higher (62.9 and 65.2 °C, respectively), which could justify their superior hardness (Bockisch, 1998). Considering only the Brazilian cocoa butters, the Braz B sample from Amazonas State was the only one that showed a solid profile similar to the blends used as an industrial standard in Brazil. This superior hardness for Braz B can be a consequence of its high S_2U content compared to the other Brazilian samples, according to Table 2. An increase of only 0.7 and 0.9% in oleic and linoleic acid contents can be related to a decrease of approximately 10% in the SFC value at 25 °C, a result that was also found by Timms (2003).

Shukla's (2006) studies showed that, at 25 °C, the SFC of the cocoa butter samples from Ghana, India, Nigeria, Ivory Coast, Malaysia and Sri Lanka presented variations from 74.8 to 83.7%, while for Brazilian cocoa butter, this percentage was significantly lower (Shukla, 2006). He found an SFC of 53.3%, close to the values determined for the Brazilian samples in this work. Similar observation was reported by Beckett (2008)and by Yamada *et al.* (2005), when comparing cocoa butter properties from different countries. According to Subramaniam (2000), the use of raw material with an excess of liquid fat can undermine the stability of chocolate and analogous products during storage. Therefore, the solid curves obtained for Braz A and Braz C samples in this study confirm that national
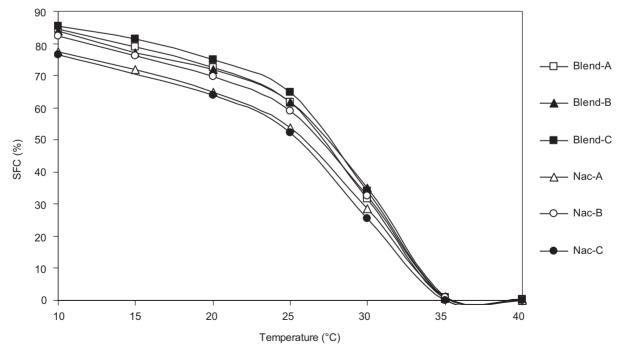

Figure. 1
Solid fat content profiles of the cocoa butter samples.

cocoa butter is considered a softer raw material and needs to be added to the cocoa butter of other origins to confer high SFC at several temperatures, when used in tropical regions.

The solid curve provides indications about the technological performance of cocoa butter. An SFC between 20 and 25 °C qualifies the cocoa butter hardness. The temperature range in which an expressive decline in SFC is evident represents the heating resistance, while the fast melting between 32 and 35 °C is responsible for cooling and creaminess sensation during tasting (Beckett, 2008). One of the more important and practical parameters used in the industry for evaluating cocoa butter quality is the difference between the SFC at 25 and 35 °C (named ΔS). The presence of solid fat at temperatures above 35 °C is recognized as a waxy feeling and is easily detected during tasting. So, in order for cocoa butter to be used in chocolate manufacturing in tropical climates, it should be hard and brittle at room temperature (SFC higher than 50% at 25 °C) and present good melting properties in the mouth (high ΔS value), and no waxy residual (no SFC above 35 °C) (Luccas, 2001). Considering these requirements, all samples meet the hardness requirements for use in warm climates, due to their SFC values at 25 °C. The ΔS values found are 61.2; 60.6; 63.8; 52.9; 58.1 and 52.2%, for Blend A, Blend B, Blend C, Braz A, Braz B and Braz C samples, respectively. The higher ΔS values found for the industrial blends ensure adequate melting properties at body temperature. The evaluated samples showed no waxy residual, and therefore proved to be appropriate regarding the sensory attributes.

3.6. Consistency

According to Gunstone and Harwood (2007), cocoa butter is hard and brittle at 20 °C, turning soft between 30 and 32 °C. Consistent data for the six cocoa butter samples are presented in Table 5 as a function of the temperature, represented as y*ield value* (g_f/cm^2). Consistency of the samples diminished substantially with the increase in temperature, due to gradual crystal melting and consequent destruction of the crystalline network, conferring plasticity to cocoa butter (Deman, 1983).

According to Haighton (1959), a fat is considered as "very hard" when its y*ield value* is above 1500 g_f/cm^2. The values in Table 5 indicate that at 20 and 25 °C, all cocoa butters are extremely hard and that at 20 °C, the hardness of the Brazilian samples is higher than the industrial blends. This behavior does not comply with the tendency found for the melting points and solid curves and should be attributed to the microstructural entanglement (crystal number, size and morphology) developed during the crystallization of these samples at this specific temperature (Campos *et al.*, 2010). At 25 °C, however, the y*ield values* of the blends are two or three times the values measured for Brazilian cocoa butter, thus supporting for their preferential use in tropical climates.
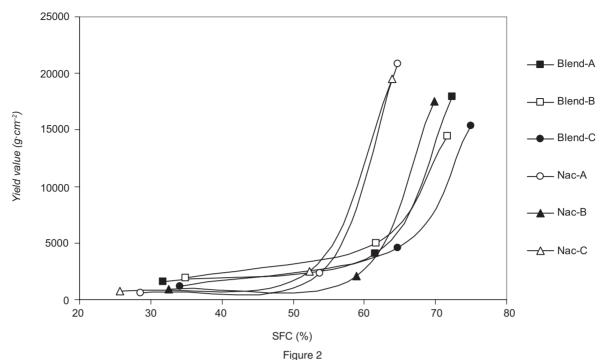
At 30 °C, the Blend A and Blend B samples are still classified as "very hard", while Blend C and Braz B samples, with y*ield values* between 1000 and 1500 g_f/cm^2, can be considered as "hard and at the spreadability limit". Braz A and Braz C samples are qualified as "plastic and spreadable" raw material (Haighton, 1959). According to Jeyarani and Reddy (2005), fats scored as hard at room temperature are suitable ingredients for the manufacturing of food products with firm textures, like chocolate. The consistency values correlated well with the results obtained for the solid profiles for all the six samples. According to Beckett (2008), the relative hardness of cocoa butter is particularly affected by the S_2U/U_2S ratio, which corresponds to 25.7; 24.4; 24.6; 10.0; 13.6 and 8.6, for Blend A, Blend B, Blend C, Braz A, Braz B and Braz C samples, respectively. The S_2U contents, SFC values and y*ield values* of the Brazilian samples presented a tendency to increase according to the average ambient temperature increase of the cultivation location in the sequence Espírito Santo, Bahia and Amazonas states. Lehrian *et al.* (1980) reported similar results in studies for cocoa butter obtained from trees cultivated in controlled microclimates. This kind of influence of environment conditions on vegetal oil and fat crop production has been extensively studied. In the

Table 5
Yield value (g_f/cm^2) of cocoa butter samples

Sample	Yield value (g_f/cm^2)		
	20 °C	25 °C	30 °C
Blend A	17936.71 ± 314.77[a]	4106.10 ± 188.11[a]	1567.81 ± 24.36[a]
Blend B	14413.72 ± 189.23[b]	4999.24 ± 251.28[b]	1950.03 ± 26.78[b]
Blend C	15343.80 ± 216.27[c]	4594.72 ± 175.55[c]	1144.02 ± 11.00[c]
Braz A	20877.39 ± 293.63[d]	2307.48 ± 142.57[d]	557.44 ± 64.93[d]
Braz B	17484.23 ± 180.85[e]	2077.56 ± 185.32[e]	923.51 ± 61.96[e]
Braz C	19535.33 ± 190.12[f]	2505.35 ± 160.88[f]	737.55 ± 20.06[f]

Means ± standard deviation of the replications. Means followed by the same letter within a column indicate significant difference (p < 0.05) by Tukey's test.

Figure 2
Relation between *yield value* and solid fat content of the cocoa butter samples at 20, 25 and 30 °C.

technical literature, there is a consensus that the closer to the equator the plant is cultivated, the lower the unsaturation degree and hardness of its fat components (Tucci *et al.*, 1996).

Recent studies have investigated the relationships between solid profiles and consistency of fats. SFC is an important indicator of hardness in fats and can be used for predictive purposes (Ribeiro *et al.*, 2009). Figure 2 presents the relationship between the y*ield value* and SFC of the six cocoa butter samples at the temperatures corresponding to the consistency evaluation. It can be seen that the increase in consistency started at SFC values between 50 and 60%. According to the solid profiles (Figure 1), this range corresponds to temperatures between 20 and 25 °C, limits usually set for cocoa butter hardness qualification (Gunstone and Harwood, 2007). A subsequent increase in SFC was reflected in an exponential increase in sample consistency, which can be attributed to the formation of a crystalline structure characterized by different structural levels related to individual TAGs, crystalline units and clusters (Shi *et al.*, 2005).

5. CONCLUSIONS

The study on the chemical composition, solid fat content and consistency properties of Brazilian cocoa butter samples and industrial blends, used by Brazilian manufacturers and composed of cocoa butter from different countries, allowed for researchers to differentiate them according to their physico-chemical characteristics and their potential use in industrial processes in tropical regions. In general, the blends exhibited more adequate attributes in terms of thermal resistance and hardness, which are necessary and compatible for their use under the different climate conditions of tropical regions. With regard to Brazilian samples, cocoa butter from the Amazonas State presented characteristics similar to the industrial blends*,* mainly the hardness profile. Cocoa butter from the Bahia and Espírito Santo States showed considerable softness that will give rise to technological problems when used in warm climates, thus justifying the requirement of blendingit with cocoa butter from other countries. In Part II of this study the same samples are evaluated for their crystallization kinetics, microstructure and polymorphism and the results are explained in terms of the chemical profile presented here.

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