

# Cocoa Butter Extender from *Simarouba glauca* Fat

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**ABSTRACT:** *Simarouba glauca* is a rich source of fat, having a melting point of about 29°C and consisting of palmitic (12.5%), stearic (27%) and oleic (56%) as major fatty acids. It consists of about 30% of symmetrical monounsaturated-type triacylglycerols and appears to be a good source of fat for preparation of cocoa butter (CB) extender. The stearin fraction (35% yield) obtained by solvent fractionation showed an increased supercooling property and a sudden rise in temperature during solidification compared to native fat as shown by cooling curves. The fraction had a narrow melting range and consisted of a high content (66%) of symmetrical monounsaturated-type triacylglycerols like CB. The fraction was compatible with CB even at 50% substitution. In addition, the fraction did not affect the formation of stable or other polymorphic forms of CB at different tempering conditions. The fraction obtained by dry fractionation also had properties similar to that obtained by solvent fractionation. The conditions of the fractionation determine the yield of stearin, which in turn alters the melting characteristics of the fractions. The stearin obtained after removal of about 60–65% olein was found to be suitable as a CB extender to replace up to 25% of CB in chocolate products.

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**KEY WORDS:** Chocolate, cocoa butter extenders, confectionery fat, fractionation, *Simarouba glauca* fat.

*Simarouba glauca*, D.C. (Simaroubaceae), which is native to El Salvador in Central America and popularly known as acituno, was introduced to India in the early 1960s. It is an evergreen tree with profuse deep green, glossy foliage on its rounded crown and has the habit of growing sprawling branches from almost the ground level in its early stages of growth. The plant begins fruiting at about 5–6 yr of age and yields for up to 50–60 yr. A mature plant yields about 20–50 kg of brown-colored oval-shaped seeds (1). It has been reported that the seeds contain 40% kernels and the kernels contain 60% fat, which is edible. The odorless, greenish-yellow fat melts at 26.4°C, has an iodine value of 52.6, and a saponification value of 190.5. The average values of major fatty acids reported were: oleic (51.1%), stearic (30.4%), and palmitic (12.6%) (2–6). Further, it has been reported that the characteristics of the fat and the fatty acid composition of Indian origin do not significantly differ from those reported from seeds of other countries (5,6). Considering the high fat content in the kernels and moderate iodine value and high content of oleic and stearic acids, the fat has good potential

for use as edible fat or for blending with vanaspati or for use as a cocoa butter (CB) substitute or extender. Preparation of CB extender from *S. glauca* fat by fractionation and its effect on polymorphic behavior of CB are reported in this paper.

## MATERIALS AND METHODS

*Simarouba glauca* seeds were procured from the Assistant Soil Conservation Officer (Keonjhar, Orissa, India). The fat was extracted from the kernels using hexane and refined with alkali. Refined fat was also procured from Regional Research Laboratory (Bhubaneswar, India). Standard triacylglycerol and fatty acid methyl esters were obtained from Sigma Chemical Company (St. Louis, MO).

**Fractionation.** (i) *Solvent fractionation.* Refined *S. glauca* fat (200 g) was dissolved in 400 mL of acetone by heating to about 50°C. The solution was gradually cooled to 18°C and held at this temperature for about 2 h. The partially crystallized mass was filtered through a Büchner funnel under suction to separate stearin and olein fractions. The solvent from the fractions was removed under vacuum. The yield of stearin (Fr. 1) was 35% by weight of the fat. In another experiment, the holding time was increased to obtain stearin of 45% (Fr. 2) yield, and in yet another experiment, the solution was held at 16°C for 1 h to obtain 21% stearin (Fr. 3).

(ii) *Dry fractionation.* *Simarouba glauca* fat (200 g) was melted by heating to ca. 45°C to destroy all crystal nuclei. The fat was cooled gradually to 25°C and held at this temperature for 3 h. The solid and liquid fractions were separated by filtration to yield 38% stearin (Fr. 4).

**Cooling curves.** Solidification characteristics of the samples were determined by cooling curves obtained by using Shukoff's flask according to the procedure described by Wilton and Wode (7). The method is simple, quick, and provides information regarding how crystallization proceeds during cooling, though the crystals are not completely transformed into the most stable form. The method is also sensitive to impurities and an admixture of foreign triacylglycerols and it has been used to assess both the quality of CB from different countries of origin (8) and palm oil fractions as CB replacers (9). The temperature rise is an index of amount of crystallization formed during the experiment, the time taken shows the rate of transition of  $\alpha$  to  $\beta'$  (9,10), and the minimum temperature reached on the curve indicates the supercooling property of the sample.

**Differential scanning calorimetry (DSC).** A Mettler (Zurich, Switzerland) differential scanning calorimeter was used to obtain melting and crystallization characteristics of

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the samples. The heat flow of the instrument was calibrated using indium. The PT-100 sensor was calibrated using indium, zinc, and lead. To ensure homogeneity and to destroy all crystal nuclei, the samples were heated to 60°C. About 15 mg of molten sample was accurately weighed into a standard aluminum crucible and the cover crimped in place. An empty aluminum crucible with pierced lid was used as a reference. For melting characteristics, the samples were stabilized according to an IUPAC method (11), which includes keeping the samples at 0°C for 90 min, 26°C for 40 h, followed by 0°C for 90 min prior to introduction into the DSC cell. Thermograms were recorded by heating at a rate of 2°C/min from -5 to 50°C. The peak temperatures, enthalpy ( $\Delta H$ ), and the percentage liquid at various temperatures were recorded directly using a TC-10A data processor (Mettler). The solid fat content (SFC) was calculated from percentage liquid, and the melting profiles were drawn by plotting SFC against temperature. Though SFC obtained by DSC is not the same as those obtained by pulsed nuclear magnetic resonance, the data give enough information on melting characteristics of different fats/fractions. In addition, the endotherms obtained by DSC provide information regarding the presence or absence of mixed crystals.

For crystallization behavior, the samples were maintained at 60°C for 5 min and immediately cooled to -10°C at 5°C/min. The cooling exotherms, crystallization peak temperatures, and  $\Delta H$  of crystallization were recorded. Also, the samples were immediately heated at 10°C/min to record the melting peak(s) of the crystals generated under a slow (5°C/min) rate of cooling.

To determine the different types of polymorphic forms and their phase transitions, the samples were tempered at different conditions in separate experiments as follows: (i) 60°C for 5 min and keeping at 15°C for 5 h; (ii) 60°C for 5 min and keeping at 20°C for 12 h; and (iii) 60°C for 5 min and keeping at 25°C for 5 h. After each tempering condition, the samples were kept at 0°C for 30 min prior to introduction into the DSC cell. The samples were then heated from -5 to 50°C at 10°C/min to record melting peak temperatures of the crystals obtained under different tempering conditions. (iv) To find out the phase transition of lower polymorphs, the samples were subjected to the following conditions: Held for 5 min at 60°C to destroy all crystal nuclei, cooled rapidly at 20°C/min to -10°C, and immediately heated at different rates separately: 10°C/min, 5°C/min, 2.5°C/min to record the melting peak temperatures of the crystals generated.

**Fatty acid composition.** The fatty acid composition of the samples was determined by analyzing the fatty acid methyl esters by gas chromatography. The methyl esters were prepared using 14% BF<sub>3</sub>/methanol (12) and were analyzed using a Shimadzu GC-9A (Kyoto, Japan) equipped with a flame-ionization detector operating under the following conditions: Column 2.4 m × 0.3 cm stainless steel packed with 15% diethylene glycol succinate coated on chromosorb W (60–80 mesh); column temperature 180°C; injector temperature 200°C; carrier gas, nitrogen, 15 mL/min and 20 mL/min hy-

drogen. The peaks were identified by comparing the retention times with those of authentic standards and reported as relative percentage of individual fatty acids.

**Triacylglycerol composition.** The triacylglycerol composition of the samples was determined by high-performance liquid chromatography using a Shimadzu system controller LC-10A and refractive index detector RID-10A. A C-18 column (3.9 × 300 mm; 5 μm particle size) maintained at 36°C was used. The mobile phase was a mixture of acetone/acetonitrile (63.5:36.5, vol/vol) at the flow rate of 1 mL/min (13). The samples were purified by passing through a silica gel column and eluting pure triacylglycerols with hexane. The dried samples were dissolved in chloroform, and 10 μL was injected. The peaks were identified by comparing the retention times with those of authentic standards and reported as relative percentage of individual triacylglycerols in the sample.

The saponification value, iodine value (Wij's), and unsaponifiable matter of the fat were determined according to the procedures described by American Oil Chemists' Society (14).

## RESULTS AND DISCUSSION

*Simarouba glauca* fat is semisolid at 25°C with a melting peak temperature of *ca.* 29°C (Table 1), iodine value of 57.6; saponification value of 193; and unsaponifiable matter of 1.0%. The fat crystallizes slowly without any significant supercooling or rise in temperature during solidification, as shown by cooling curves (Fig. 1). The fat is soft, consisting of low SFC at all temperatures (Table 1). It consists of 12.5% palmitic, 27% stearic, and 56% oleic as major fatty acids constituting about 30% of monounsaturated and disaturated (SUS)-type triacylglycerols (Table 2). The physical and chemical properties of the fat were in agreement with the literature (2–6). Due to these chemical properties, the fat has good potential for preparation of CB extender/replacer.

The stearin fraction (35%, Fr. 1) obtained by solvent fractionation of the fat showed considerable improvements in physical properties, making it comparable to CB (Figs. 1, 2, and Table 1). Fr. 1 showed increased supercooling characteristics and sudden rise in temperature during solidification (Fig. 1), which indicates rapid crystallization after nucleation (10), compared to original fat and CB. The results in Table 2 revealed that the percentage of low-melting (diunsaturated) triacylglycerols (SUU type) is reduced in Fr. 1 compared to the original fat and showed higher proportions of monounsaturated (SUS)-type triacylglycerols like CB, which are responsible for improvements in the above-mentioned physical properties.

DSC cooling curves showed that Fr. 1 crystallized at low temperature (11°C) compared to CB (13.6°C) (Table 3), confirming the above results that the fraction showed increased supercooling properties. The onset of crystallization and enthalpy is gradually increased as the proportion of CB in the mixture is increased (Table 3).

Fr. 1 showed a narrow melting range like CB with a high SFC at normal ambient temperature (20–25°C) and no solids

**TABLE 1**  
**Solids Fat Content of *Simarouba glauca* Fat Fractions and Their Mixtures with Cocoa Butter Obtained by Differential Scanning Calorimetry (DSC) After Long Stabilization (26°C, 40 h)**

Sample <sup>a</sup>	% Solids at °C					Melting peak			Temp. (°C)	$\Delta H$ (J/g)
	20	25	30	32.5	35	37.5	40			
Cocoa butter (CB)	98.0	96.0	72.4	22.3	2.6	0.3	0	32.5	128	
SG fat	81.0	65.0	36.8	10	1.3	0.9	0.6	29.0		
Fraction 1	89.0	81.0	60.0	31.6	4.1	0.9	0.7	33.2	58.5	
Fraction 1 + CB (50:50)	91.0	80.0	54.7	20.0	1.3	0.4	0.3	32.7	74.0	
Fraction 1 + CB (25:75)	99.0	88.5	67.2	32.6	1.5	0	0	33.3	92.2	
Fraction 1 + CB (75:25)	90.0	77.0	56.7	26.0	2.3	0.9	0.7	33.1	62.0	
(5) <sup>b</sup> + MF (85:15)	82.4	69.7	46.0	12.0	0.5	0.1	0.1	31.5	61.3	
CB + MF (85:15)	86.0	73.3	32.0	4.1	0.8	0	0	30.8		
Fraction 2	70.0	59.0	46.0	24.0	3.0	0.4	0.2	33.2	41	
Fraction 3	94.0	86.3	73.7	52.5	17.1	1.7	0.1	34.6	73	
Fraction 4	83.0	76.3	57.2	29.7	3.1	0	0	33.4	62.3	
Fraction 4 + CB (50:50)	93.0	84.3	61.0	25.2	1.0	0	0	33.0	83.7	
Fraction 4 + CB (25:75)	94.7	88.7	68.1	32.0	1.8	0	0	33.3	100.3	
Fraction 4 + CB (75:25)	90.4	78.5	60.0	30.0	2.1	0.4	0.3	33.3	69.1	
(13) <sup>c</sup> + MF (85:15)	83.2	70.0	44.4	10.3	0.6	0.1	0.1	31.8	68.3	

<sup>a</sup>MF = milk fat; Fraction 1, 2, and 3 = stearin of 35, 45, and 21% yields, respectively, obtained by solvent fractionation of *S. glauca* (SG) fat; Fraction 4 = 38% stearin obtained by dry fractionation of SG fat.

<sup>b</sup>(5) = Fraction 1 + CB (25:75), blended with MF at 85:15 level.

<sup>c</sup>(13) = Fraction 4 + CB (25:75), blended with MF at 85:15 level.

at body temperature (Fig. 2), which are essential characteristics for a CB extender.

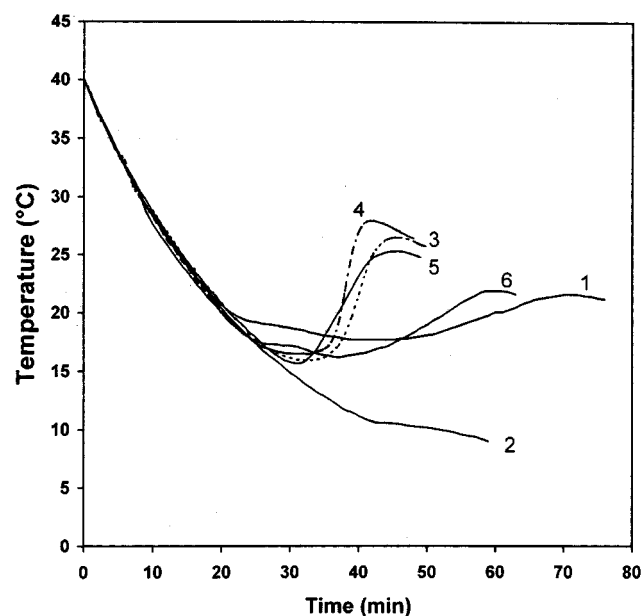
Fr. 1 was found to be compatible with CB even at 50% level as revealed by cooling curves and melting characteristics (Figs. 1, 2). The enthalpy of melting of the blends containing Fr. 1 and CB increased as the proportion of CB increased in the mixture (Table 1). The blend containing 25% Fr. 1 and 75% CB (Blend 1) showed melting profile and  $\Delta H$  of crystallization similar to those of CB alone (Fig. 2, Table 3). Also, Blend 1 showed better tolerance toward milk fat compared to CB alone as it showed less softening effect when milk fat was incorporated (Table 1).

Fr. 1 had higher concentrations of two oleodistearin (StOSt) triacylglycerols than those present in CB (Table 2), which impart temperature resistance and quick setting of fat and chocolate (15,16). These results therefore revealed that the stearin fraction obtained from *S. glauca* fat after removal of about 65% olein fraction is suitable for use in chocolate products as CB extender. Also, the stearin (yield 38%, Fr. 4) obtained by dry fractionation showed similar properties compared to those of Fr. 1 obtained by solvent fractionation (Figs. 1, 2, and Table 1).

The results also revealed that the SFC could be decreased or increased depending on the yield of the stearin (Fr. 2 and Fr. 3, yields 45, and 21%, respectively) (Table 1). It can be seen from Table 2 that StOSt triacylglycerols also increased as the yield of stearin was lowered. Thus, a range of CB extenders with varying melting characteristics could be prepared from *S. glauca* fat by altering the fractionation conditions.

**Polymorphic behavior.** To find out the effect of replacing CB with Fr. 1 on tempering of chocolate products, the polymorphic behavior and phase transition of various forms of Fr. 1 and its mixtures with CB were studied under different tempering conditions.

(i) **Rapid cooling:** When rapidly cooled (20°C/min), the crystals of CB showed a single melting peak at 20.5°C (Fig. 3). On the other hand, the crystals obtained from Fr. 1 showed two melting peaks: the lower at very low temperature (12.2°C) and the next form at a higher temperature (24.6°C) (Fig. 3, Table 4). The difference in melting temperatures between these two forms is large unlike in CB. This explains the



**FIG. 1.** Shukoff's cooling curves for (1) cocoa butter (CB); (2) *Simarouba glauca* fat; (3) Fraction 1; (4) Fraction 3; (5) Fraction 4; (6) Fraction 1 + CB (50:50). Fraction 1, 2, and 3 = stearin of 35, 45, and 21% yields, respectively, obtained by solvent fractionation of *S. glauca* fat; Fraction 4 = 38% stearin obtained by dry fractionation of *S. glauca* fat.

**TABLE 2**  
Triacylglycerol Composition (wt%) of *S. glauca* Fat, Its Fractions, and CB<sup>a</sup>

Sample	SOA	SOS	POS	POP	SOO	POO	AOO	OOO	Others
SG fat	1.7	21.1	15.5	2.6	29.5	12.2	2.3	11.8	3.3
Fraction 1	2.7	38.3	14.4	2.0	10.5	4.7	0	0	0
Fraction 2	2.2	28.2	15.7	2.3	24.5	11.1	1.5	11.0	3.5
Fraction 3	2.9	49.3	14.4	2.0	8.9	4.0	0	0	0
Fraction 4	2.5	31.1	15.8	2.4	23.5	10.0	1.5	10.2	3.0
Cocoa butter	1.0	30.0	39.4	17.1	5.7	4.0	0	0	2.8

<sup>a</sup>A = arachidic; O = oleic; P = palmitic; S = stearic acids. See Table 1 for other abbreviations.

behavior of Fr. 1 during solidification as observed by cooling curves. Since the lowest form has a very low melting point, the fraction showed increased supercooling property (i.e., low temperature at minimum point on the curve), and due to rapid transition to its next form, which is at a high temperature, there was a sudden rise in temperature on the curve (Fig. 1). The mixture containing 50:50 of Fr. 1 and CB showed two melting peaks, whereas that containing 25% of Fr. 1 in CB showed only a single peak like CB at the same temperature (Fig. 3, Table 4).

(ii) *Slow cooling.* The results revealed that the crystals obtained from all the samples under a slow (5°C/min) rate of cooling were similar compared to those obtained under a rapid rate of cooling, indicating that there was no further phase transition during this process (Table 4).

To find out the phase transition of lower polymorphic forms to the next higher forms, the samples were rapidly (20°C/min) cooled and then heated at different rates (10, 5, and 2.5°C/min). It can be seen from the Table 4 that Fr. 1 showed only one melting peak in the range of 23.4–25.3°C under slow rates (5 and 2.5°C/min) of heating, unlike those obtained under rapid rates of heating (Fig. 3 and Table 4). Fr. 1 and its mixtures with CB (50:50 and 25:75) showed a major peak at 20.4 and 20°C respectively, under 5°C/min rate of heating similar to CB. However, at 2.5°C/min rate of heating, CB showed peaks similar to those obtained at 5°C/min, whereas the mixture (50:50) of Fr. 1 and CB showed a major peak at 23.4°C, and that containing 25% Fr. 1 (Blend 1) showed two peaks like CB, the higher one at 23.3°C (Table 4).

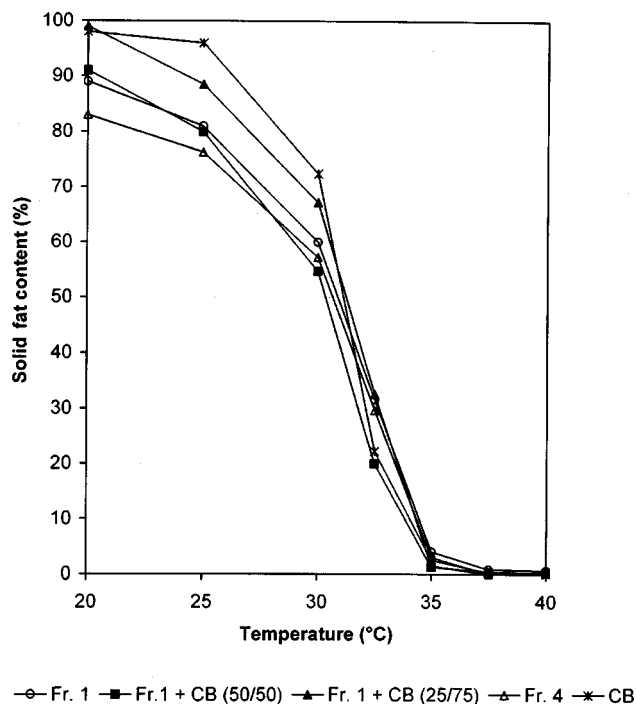
**TABLE 3**  
DSC Crystallization Temperatures and Enthalpy of Crystallization Obtained Under Slow (5°C/min) Rate of Cooling<sup>a</sup>

Sample	Crystallization	
	Peak temp. (°C)	Enthalpy (J/g)
SG fat	8.4	24
Fraction 1	10.1	32
Fraction 1 + CB (50:50)	11.9	54
Fraction 1 + CB (25:75)	12.2	52
Fraction 2	11.7	37
Fraction 2 + CB (50:50)	11.9	55
Fraction 3	11.9	57
Fraction 4	11.9	35
Fraction 4 + CB (50:50)	11.7	48
Fraction 4 + CB (25:75)	12.5	54
CB	13.6	58

(iii) *Tempering at 15°C.* Tempering at 15°C for 5 h, Fr. 1, CB and Blend 1 (25:75) of these two showed a single peak at 26.6°C (Table 4), indicating Fr. 1 did not affect the formation of Form IV of CB at 15°C.

(iv) *Tempering at 20/25°C.* Fr. 1 showed a major peak corresponding to the most stable form after tempering at 20°C for 12 h with a small shoulder at 26°C, whereas all the blends of Fr. 1 and CB showed a single peak corresponding to the most stable form (Form V in case of CB) (Table 4). The samples tempered at 25°C for 5 h also showed the peak corresponding to the most stable form. These results therefore indicated that Fr. 1 did not affect the formation of the stable form (Form V) of CB at 20 or 25°C. The melting peak temperatures obtained under these conditions were similar to those obtained after long stabilization (26°C for 40 h), and hence these are the most stable forms (Tables 1 and 4).

These results therefore revealed that *S. glauca* fat fraction crystallized similar to CB under different tempering condi-

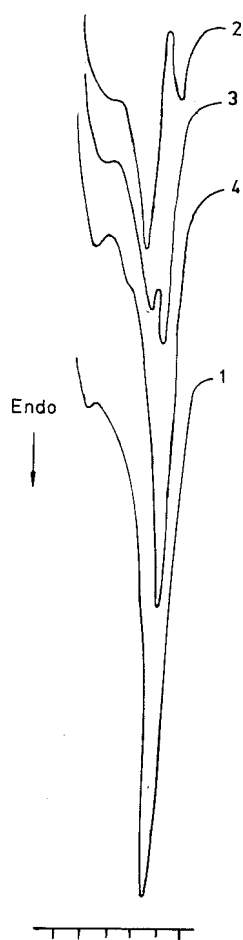


**FIG. 2.** Melting profiles of Fraction 1, CB, and their mixtures and Fraction 4. See Figure 1 for abbreviations.

**TABLE 4**  
**Melting Peak Temperatures of Various Polymorphic Forms Obtained from *S. glauca* Fat Fractions and Their Mixtures with CB Under Different Tempering Conditions as Obtained by DSC<sup>a</sup>**

Rate of heating (°C/min)	Rapidly (20°C/min) cooled			Slowly (5°C/min)	Tempered at		
				cooled	20°C-12 h	25°C-5 h	15°C-5 h
	10	5	2.5	10	10	10	10
CB	20.5	18.0*	18.0	19.9	31.4	32.6	26.6
SG fat	11.2	ND	ND	10.3	10.4	11.4	ND
Fraction 1	12.2	23.4	25.3	12.5	26.0*	35.0	26.6
	24.6			24.6	34.9		
Fraction 1 + CB (50:50)	13.9	17.2	23.4	12.9	32.8	31.8	ND
	19.8	20.4		19.3			
Fraction 1 + CB (25:75)	13.8*	17.0*	17.2	19.7	31.6	24.4	26.3
	20.3	20.0	23.3			33.3	
Fraction 4	13.5	ND	ND	13.3	34.5	29.0	ND
	24.1			24.8	27.0*	35.5	
Fraction 4 + CB (50:50)	14.8	ND	ND	13.8	32.6	23.4*	ND
	19.6			19.5		33.3	
Fraction 4 + CB (25:75)	20.0	ND	ND	20.0	32.1	33.3	ND

<sup>a</sup>Shoulder; ND, not determined; see Table 1 for other abbreviations.



**FIG. 3.** Differential scanning calorimetry heating (10°C/min) curves obtained after rapid (20°C/min) cooling from 60 to -10°C; (1) CB; (2) Fraction 1; (3) Fraction 1 + CB (50:50); (4) Fraction 1 + CB (25:75); See Figure 1 for abbreviations.

tions and the formation of the most stable form is similar to that of CB. The main difference is the very low melting point of the least stable form in case of Fr. 1 unlike that of CB. This suggests that the lowering of temperature may be necessary during manufacture of chocolate to generate crystal nuclei compared to CB. However, the mixtures containing Fr. 1 and CB, especially Blend 1, showed supercooling and other properties similar to those of CB alone. Fr. 1 also did not affect the formation of various crystal forms including the most stable form of CB under different tempering conditions. Similarly, the fraction obtained by dry fractionation of the fat (Fr. 4) showed properties similar to those of Fr. 1. It can be concluded from these results that the stearin fraction obtained by solvent or dry fractionation processes of *S. glauca* fat is suitable for use in chocolate products as a CB extender to replace up to 25% CB without affecting the desirable qualities of the product.

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