

BLENDING PALM FRACTIONS AS CONFECTIONERY FATS: A PRELIMINARY STUDY

NORAZURA AILA MOHD HASSIM*; SIVARUBY KANAGARATNAM* and NOOR LIDA HABI MAT DIAN*

ABSTRACT

The study looked into the mouldability of cocoa butter alternative (CBA) from blended palm fractions namely palm mid fraction (PMF) IV 45, palm kernel oil (PKO) and palm stearin (POs) IV 33 and IV 14 for the production of chocolate bar. PMF, PKO and POs IV 33 and IV 14 were blended in eight different optimised ratios based on solid fat content (SFC) response of more or close to 50% at 20°C, SFC of less than 10% at 37°C and SFC of less than 5% at 40°C to produce a melt in the mouth and mouldable plain chocolate bar. Mouldability and snappability of the chocolate bar were the main parameters of the study. Physicochemical properties of the fat blends namely the fatty acid composition (FAC), triacylglycerol composition (TAG), solid fat content (SFC), crystallisation rate at isothermal temperature of 5°C, 10°C and 20°C, thermal behaviour, polymorphism, crystal morphology and compatibility test with cocoa butter (CB) for selected fat were also determined. Fat F blend with 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14 showed the highest SFC (47%) at 20°C and was able to be moulded and demoulded at 10°C by 45 min. It has the highest rupture tension ($105.28 \text{ g}_f \text{ cm}^{-2}$) and breaking force (5501 g_f) at 10°C. The composition of oleic acid, linoleic acid, total monounsaturated fatty acid, crystal morphology (density) as well as SFC profile at 20°C and crystallisation rate profile at 5°C, 10°C (after 40th min) and 20°C (before 35th min) of fat F were comparable with CB while other FAC, TAG, thermal behaviour and polymorphism were dissimilar to CB. Fat F, 10%-30% was observed to be compatible (no eutectic) with CB and therefore, has the potential as fat for chocolate bar. The addition of 10%-30% of fat F to CB increased the SFC to 57% at 20°C. Fat F (30%) with CB could be demoulded by 30 min and produced snappability (breaking force) of $5032.71 \pm 85.91 \text{ g}_f$ at 20°C after one day stabilisation period. Palm-based fat blend having 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14 and CB has the potential to be used as alternative fat for chocolate bar which can eventually benefit the confectionery industry.

Keywords: blended palm fractions, chocolate bar, confectionery fats, mouldability, rupture tension.

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INTRODUCTION

Fat is an important ingredient for chocolate and other confectionery products as it has a major influence on their organoleptic and physical

properties. It is responsible for the snap, appearance, mouthfeel and flavour release of chocolate confectionery (Rios *et al.*, 2014). Cocoa butter (CB) is the traditional fat for chocolate confectionery. It contains three main fatty acids which are palmitic, stearic and oleic. The primary triacylglycerols (TAG) composition of CB are the 1,3-dipalmitoyl-2-oleoyl glycerol (POP), 1(3)-stearoyl-2-oleoyl-3(1)-palmitoyl glycerol (POS) and disaturated 1,3-distearoyl-2-

* Malaysian Palm Oil Board,
6 Persiaran Institusi, Bandar Baru Bangi,
43000 Kajang, Selangor, Malaysia.
E-mail: azuraaila@mpob.gov.my

oleoyl glycerol (SOS) (Mohamed, 2013). This TAG composition is responsible for the sharp melting of CB between 30°C and 35°C but maintains a solid consistency at 20°C which is desirable for chocolate confectionery (Timms, 2003). Due to difficulty in cultivation of cocoa plants, low productivity and pest attacks, the production of CB is hampered although its demand is high. These scenarios lead to the production of CB alternatives (CBA) and are classified into three groups namely CB equivalent (CBE) having similar physicochemical properties as CB, non-lauric CB replacer (CBR) and lauric-based CB substitutes (CBS) having similar physical properties but different chemical properties from CB (Norazura Aila and Noor Lida, 2017).

There are various methods in producing confectionery fats which include blending, fractionation, hydrogenation and interesterification. Based on various studies, currently, interesterification, supercritical extraction and enhanced fractionation are the focus of CBA research. Palm mid fraction (PMF) has been the focus for interesterification studies, particularly enzymatic, for the production of CBE and has been discussed in many published articles (Wu and Deng, 2005; Borhan *et al.*, 2011; Soekopitojo *et al.*, 2009; Soekopitojo, 2011; Gibon, 2011; Mohamed, 2013; Mutia *et al.*, 2016; Biswas *et al.*, 2018). Supercritical carbon dioxide process has been employed by Nik Norulaini *et al.* (2004) and Zaidul *et al.* (2006; 2007) to further process palm kernel oil (PKO) to obtain CBE and CBR, respectively. Timms (2003) reported that the simplest CBS was produced through hydrogenation of PKO. Calliauw *et al.* (2005) published article describing the process of obtaining high-quality CBS from PKO through a two-stage static fractionation process. Kang *et al.* (2003; 2013) demonstrated the production of CBE with palm stearin (POs) through additional acetone fractionation process. However, interesterification, supercritical extraction, hydrogenation and enhanced fractionation are highly processed methods. We are of the opinion that blending on the other hand is simple and straight forward compared to the above processes.

PMF (IV 45) that is obtained through second stage dry fractionation of palm oil (De Clercq *et al.*, 2012) exhibiting melting points between those of POs and palm olein (POo) (Sakamotoa *et al.*, 2004) has been vastly utilised for blending with other vegetable fats in the production of CBE. These vegetable fats include exotic fats (Talbot, 2009; Elham *et al.*, 2013; Jun *et al.*, 2016) as well as hard stearin and high oleic-high stearic sunflower hard stearins (Bootello *et al.*, 2012; 2013). Having high level of POP makes PMF the fat of choice for blending with other sources of fats containing high POS and SOS TAG composition to mimic TAG composition of CB

(Sonwai *et al.*, 2014). In addition, PMF offers unique melting profile similar to CB. To-date, no study has been found for blending of PKO with other fats for the hard-confectionery application. However, blend of PKO with other fats is found to be suitable for confectionery filling by Biswas *et al.* (2017). As PKO (lauric fat) is rich in lauric and myristic fatty acids, eutectic might be a challenge when it is blended with non-lauric fats. POs has been used extensively as natural alternative to eliminate *trans*-fat especially in formulating hardstock such as margarine and shortening. Few blending studies of POs and other fats for confectionery application have been investigated earlier. These include studies by Jahurul *et al.* (2014b) that blended POs and mango seed fat as a CBR and Cain *et al.* (1995) that blended PKO and palm kernel stearin (PKS) for hard coating application.

Hard PMF having IV 35 has been acknowledged to be useful for hard-confectionery (Braipson-Danthine and Gibon, 2007). However, blended soft PMF (IV 45) with other palm fractions has yet to be proven for hard-confectionery applications. Therefore, in this study, blended PMF IV 45, POs (IV 33 and/or IV 14) and PKO at optimised ratios based on solid fat content (SFC) were investigated in term of physicochemical properties including their mouldability for the hard-confectionery application. POs might be able to increase SFC of the blended fats and enhance crystallisation rate while PKO could minimise tailing of the blended fats.

MATERIALS AND METHODS

Materials

PMF IV 45 and refined, bleached and deodourised (RBD) PKO were obtained from Cargill Palm Product Sdn. Bhd., Malaysia while RBD POs IV 33 was obtained from Sawit Raya Sdn. Bhd., Malaysia. RBD POs IV 14 was provided by Mewah Oils Sdn. Bhd., Malaysia. In this article, RBD PKO, RBD POs IV33 and RBD POs IV14 were simplified as PKO, POs IV33 and POs IV14, respectively.

Experimental Design and Preparation of Fat

Interaction of blended fats namely PMF/ PKO/ POs IV 33/ POs IV 14 were studied using simplex-lattice design. SFC was used as the response parameter. Twenty randomised trials including five replicates were used as inputs before optimisation. Experimental error and fit of the model was determined from the replications. Scheffe model was used to fit a polynomial equation. Analysis of variance (ANOVA) was used to check quality of the model. Optimisation of the blended fats was conducted based on SFC parameter. Targeted SFC

values were more than 45% SFC at 20°C, less than 10% SFC at 37°C, and less than 5% SFC at 40°C. These were carried out using point prediction method.

The PMF, PKO, POs IV 33 and POs IV 14 were then mixed at different ratios as shown in *Table 1* according to the optimised ternary diagram when POs IV 14 was set to zero and optimised ternary diagram when POs IV 14 was set to 2% and 3%. The fats were melted at 80°C for at least 1 hr prior to mixing.

TABLE 1. FAT BLENDS OF PMF/PKO/POs IV 33/POs IV 14 PRODUCED THROUGH MIXTURE DESIGN

Fat blend	PMF (%)	PKO (%)	POs IV 33 (%)	POs IV 14 (%)
A	88	1	11	0
B	89	1	10	0
C	91	1	8	0
D	92	1	7	0
E	96	1	3	0
F	91	1	5	3
G	93	1	3	3
H	95	1	2	2

Note: PMF - palm mid fraction; PKO - palm kernel oil; POs - palm stearin.

Solid Fat Content

SFC of the blended fats was measured according to American Oil Chemists' Society (AOCS) Official Method Cd 16b-93 (AOCS, 2012) using pulsed nuclear magnetic resonance (NMS Minispec from Bruker, Germany). Samples were melted at 80°C for 1 hr and filled into NMR tubes of 0.8 cm (diameter) and 3 cm (height). For this procedure, samples were chilled for 90 min at 0°C, followed by tempering for 40 hr at 26°C, 90 min at 0°C and finally maintained for 60 min at desired test temperatures of 5°C, 10°C, 15°C, 20°C, 25°C, 30°C, 35°C, 37°C and 40°C prior to each measurement.

Fatty Acid Composition (FAC)

FAC of the blended fat was analysed using gas chromatography (GC) based on fatty acid methyl esters (FAME) according to MPOB Test Methods p3.5 (MPOB, 2005). Samples of 0.1 g were weighed and dissolved in 1.8 ml hexane. The sample's solution was then mixed with a vortex mixer. Sodium methoxide solution (100 µl) was added to the samples for methylation process followed by water addition. Mixing was performed in between each process. The sample's solution was left for 1 hr to separate. The clear supernatant was transferred to another vial added with sodium sulphate anhydrous and kept for 15 min before complete clear supernatant (1.5 ml) was injected into GC (Agilent Technologies, Model 7890B). The GC was equipped with a flame ionisation detector (FID).

The injector and detector temperatures were both set at 240°C while the column temperature was set at 185°C. Helium gas (carrier) with flow rate of 1 ml min⁻¹ was used. The peaks were identified by comparing retention times with FAME standards and quantified using peak area normalisation method.

Triacylglycerol Composition (TAG)

TAG composition was performed using ultra high-performance liquid chromatography (U-HPLC) system (1290 Infinity LC System, Agilent Technologies, USA) and Agilent 1260 RI detector (Waters Corp., USA) according to AOCS Official Method Ce 5b-89 (AOCS, 2012). A Cortecs UPLC C18 column (2.1 mm x 150 mm length i.d; 1.6 µm particle size) (Waters Corp., Milford, Massachusetts, USA) maintained at 30°C was used. The mobile phase used was a mixture of acetone-acetonitrile with the ratio of 63.5:36.5 (v/v) at a flow rate of 0.25 ml min⁻¹. Samples (0.1 g) was solubilised in 1 ml of acetone. The chemicals and reagents used were HPLC grade. HPLC analysis was conducted in duplicate and data was recorded as percent areas.

Crystallisation Rate at Isothermal Temperature

Crystallisation rate was measured using pulsed nuclear magnetic resonance (NMS Minispec from Bruker, Germany) according to Chen *et al.* (2015) with modification. Samples were melted at 80°C for 1 hr. NMR tubes of 0.8 cm (diameter) and 3 cm (height) were used in this analysis. Tubes with samples were held for 1 hr at 30°C. The tubes were then placed at 5°C, 10°C and 20°C. SFC readings were taken every 1-5 min.

Thermal Behaviour

Thermal properties of the oil samples were measured using a differential scanning calorimetry (DSC)-7 (Perkin Elmer, Norwalk, CT). Calibration was performed using an indium standard and n-decane. Oil sample of 3-5 mg was hermetically sealed in an aluminium pan. The following temperature programme was used to perform the melting and cooling measurements on each sample: cooled to -40°C at 5°C min⁻¹, held at -40°C for 5 min and then heated to 80°C at 5°C min⁻¹ following temperature programme from Biswas *et al.* (2017) with modification.

Polymorphism

Polymorphism was determined at room temperature using coupled X-ray Diffraction (XRD)-DSC Rigaku TTRAX III (Tokyo, Japan).

The sample holders were flat stainless-steel plates with rectangular holes. The X-ray incident-beam 2θ range was from 1° - 30° with scanning rate of $5^{\circ}\text{C min}^{-1}$ according to Biswas *et al.* (2017) with modification. Samples were melted at 80°C and tempered at 10°C for 30, 45 and 60 min. α , β' and β polymorphs were determined using short spacing characterisations.

Crystal Morphology

Crystal microstructure of the fats were determined using Polarised Light Microscope (PLM) following the method as described by Narine and Marangoni (1999). A small droplet (about $15\ \mu\text{l}$) of melted fat was placed on a glass slide and covered with a glass slip. The sample was first heated to 80°C for 20 min to erase crystal memory. The sample was tempered at 10°C , 20°C and 25°C for 48 hr prior to measurement. A Leica DMLP polarised light microscope (Wetzlar, Germany) equipped with a Linkam THMS 600 temperature controller stage and a JVC 3-CCD colour video camera was used. Temperature was thermostatically controlled by a Linkam TP 94 multiramp temperature programme conching machine and LNP automatic cooling system (Linkam, Tadworth, Surrey, United Kingdom). Liquid nitrogen was used as the coolant. The photomicrograph of the crystal was taken at 50X magnification.

Mouldability

Chocolate bars were produced from the blended fats according to plain chocolate formulation by Timms (2003) as per Table 2. The amount of cocoa powder, sugar, blended fat and lecithin were made constant in order to produce the chocolate bars. These raw materials were then refined using 3-mill refiner and conched using chocolate conching machine. The chocolate bars were cooled at 10°C (Keijbets *et al.*, 2010) for 30, 45 and 60 min. The bars were not subjected to further solidification period. Mouldability of the chocolate bars were determined based on observation of the moulding and demoulding activity (easy demoulding or otherwise). Triplicates of chocolate bars were produced for each blended fat.

TABLE 2. FORMULATION OF PLAIN CHOCOLATE

Ingredients	Weight (%)	Weight (g)
Cocoa powder	20.00	200.00
Blended fat	33.00	330.00
Sugar	46.60	466.00
Lecithin	0.40	4.00
Total	100.00	1 000.00

Source: Timms (2003).

Mechanical Resistance Test (Snappability)

Snappability of the chocolate bars at 10°C was conducted using a Texture Analyser (TA-XTplus, Stable Micro Systems) with a Three Bend Rig probe according to Stroppa *et al.* (2014) and Jorge *et al.* (1999). The setting for pre-test velocity was $3\ \text{mm s}^{-1}$, test velocity was $1.7\ \text{mm s}^{-1}$ and post-test velocity of $10\ \text{mm s}^{-1}$. Rupture force was adjusted to the center of the chocolate bars. Three repetitions were conducted for each chocolate having different fat blends and CB. The values of the force obtained were divided by the cross-section area of each bar and the unit for the rupture tension was reported in $\text{g}_f\ \text{cm}^{-2}$ in order to eliminate the effect of thickness variations of the bars.

Compatibility Test

The results obtained from our preliminary analysis were utilised to select the best blended palm fractions. The selected fat was blended with CB at different ratio (0%-100%). The SFC at 20°C for all ratios were determined according to AOCS Official Method Cd 16b-93 (AOCS, 2012). Mouldability at 10°C and mechanical resistance test at 20°C were conducted for blend of selected fats and CB.

Statistical Analysis

Results were analysed with Minitab version 16.0 (Pennsylvania, USA) using one-way ANOVA. Significance differences ($p \leq 0.05$) among the samples were analysed using the Tukey's *post-hoc* test. All tests were carried out in duplicate or triplicate.

RESULTS AND DISCUSSION

Solid Fat Content

SFC profiles of the fats are shown in Figure 1. CB had a high SFC at low temperature, the SFC then decreased sharply from 20°C - 35°C with 0% SFC at 35°C . Similar finding was reported by Biswas *et al.* (2017) and Kadivar *et al.* (2016) in which some finding showed 0% SFC at 37°C based on its varieties. The sharp melting profile is due to its significant level of SOS and POS TAG (Biswas *et al.*, 2017; Kadivar *et al.*, 2016). All of the eight blended fats PMF/PKO/POs (IV 33 and/or IV 14) showed marginal difference in SFC profiles at all temperatures tested. This was explained by the minor deviation in TAG value of all the fats (Table 4). This trend was also reported by Jahurul *et al.* (2014a) when blending PMF with mango seed fat to produce CBE. Blended PMF/PKO/POs (IV 33 and/or IV 14) had 76%-79% SFC at 5°C .

A steep decrease of SFC in all the fats occurred between 15°C-25°C with zero solid at 40°C. Fat F had the highest SFC at 20°C having value of 47% followed by fats G and H. Higher percentage of trisaturated TAG led to higher SFC (Zhou *et al.*, 2010). Overall, all of the blended fats (PMF/PKO/POs (IV 33 and/or IV 14)) exhibited similar SFC trend as compared to CB with high SFC at low temperature, nearly zero SFC at 35°C-40°C and high melting profile in between. Hence, these blended fats have the potential to be used as confectionery fat.

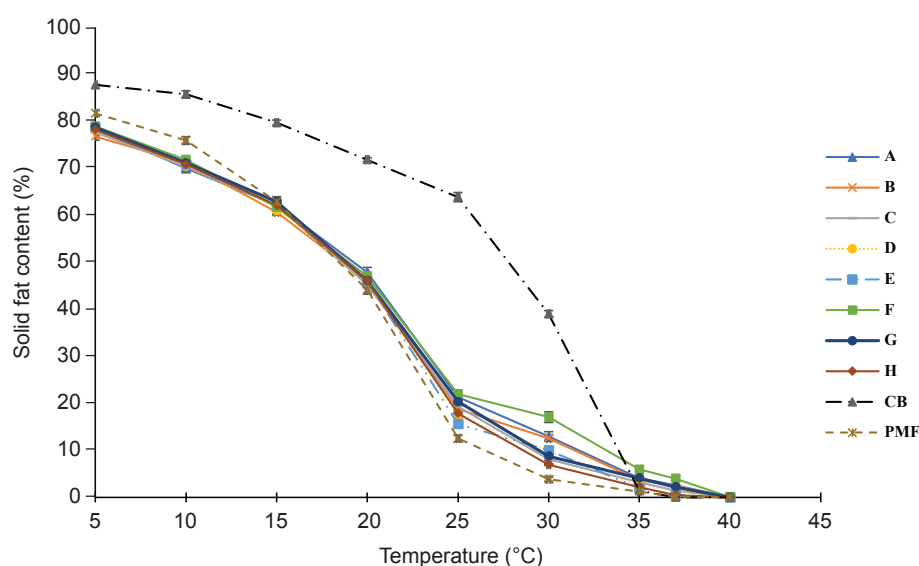
Fatty Acid Composition

FAC of the eight blended fats are tabulated in Table 3. The main fatty acids in CB are palmitic acid (C16:0), stearic acid (C18:0) and oleic acid (C18:1). This has been described in numerous earlier studies, among others were by Żyżelewics *et al.* (2014), Jahurul *et al.* (2013) and Compos and Marangoni (2010). Fatty acid profile of the eight blended fats was more diverse with major fatty acids being palmitic and oleic acids, followed by linoleic acid (C18:2). Other fatty acids were present in small quantity in the fats. Lauric acid (C12:0) and myristic acid (C14:0) values in all blended fats were low (0.70%-1.10% and 1.41%-1.89%, respectively) as only 1% PKO was recommended

after optimisation. Palmitic acid in all fats was significantly different ($p < 0.05$) and observed to be higher than in CB. Stearic acid in all the blended fats (A-H) was not significantly different ($p > 0.05$). It was observed that stearic acid in all fats was significantly lower than in CB. Although the values of oleic acid for all the blended fats were found to be close to one another and in CB, composition of oleic acid in all the fats was examined to be significantly different ($p < 0.05$) compared to each other and CB. Polyunsaturated fatty acid (PUFA) of all blended fats (A-H) was observed to be higher than in CB. The trait of unsaturated fatty acids was that it has lower melting points due to the existence of double bonds (Larsen *et al.*, 2014). In this study, the amounts of oleic and linoleic as well as monounsaturated (MUFA) fatty acids (palmitoleic acid and oleic acid) were comparable to CB.

Triacylglycerol Composition

The TAG composition of all fats is shown in Table 4. TAG (18 types) were found in CB. Saturated-unsaturated-saturated (SUS) TAG namely POS, SOS and POP dominated TAG of CB having 39.26%, 31.11% and 16.18%, respectively. Both PMF and POs were hard fractions of dry fractionation. TAG (16 types) were identified in all fat blends. Major TAG in all fat blends were POP, followed by PLP.



Note: Fat A - 88% PMF, 1% PKO, 11% POs IV 33.
 Fat B - 89% PMF, 1% PKO, 10% POs IV 33.
 Fat C - 91% PMF, 1% PKO, 8% POs IV 33.
 Fat D - 92% PMF, 1% PKO, 7% POs IV 33.
 Fat E - 96% PMF, 1% PKO, 3% POs IV 33.
 Fat F - 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14.
 Fat G - 93% PMF, 1% PKO, 3% POs IV 33 and 3% POs IV 14.
 Fat H - 95% PMF, 1% PKO, 2% POs IV 33 and 2% POs IV 14.
 CB - cocoa butter; PMF - palm mid fraction; PKO - palm kernel oil; POs - palm stearin.

Figure 1. Solid fat content of blended fats, cocoa butter and palm mid fraction.

High content of POP TAG is responsible for the β' crystal formation (Toro-Vazquez *et al.*, 2004). As percentage of PKO was very minor in all fats, LLL, LLM and LMM TAG were not present/detected. The values of POP TAG among the blended fats were comparable to each other having values ranging from 48.52%-50.28%. However, the POP TAG of these blends was significantly different ($p < 0.05$) from CB. The POS and SOS TAG of the blended fats did not differ much from one another having the range of 9.16%-9.58% and 1.08%-1.38%, respectively. However, both of the TAG were significantly different with CB ($p < 0.05$). All of the blended fats had higher unsaturated-unsaturated-unsaturated (UUU) TAG (1.12%-1.34%) compared to CB (0.17%). Saturated-unsaturated-unsaturated (SUU) TAG of the blended fats was higher than CB ranging from 14.52%-15.42%. Despite of this, SUS TAG for all blended fats was found to be higher in CB (88.79%) compared to the blended fats (59.07%-60.84%). Saturated-saturated-saturated (SSS) TAG of the blended fats ranged from 17.06%-18.97% while similar TAG in CB showed a value of 5.51%. Fat F exhibited the highest SSS TAG of 18.45% among all the blended fats. Majority of the fats contained insignificant TAG ($p > 0.05$) which included PLL, OLO, OOO, POO, SOO, POS and SOS.

In the present study, the most significantly different ($p < 0.05$) TAG was PPP. PPP increased progressively with the addition of POs (POs IV 33 and POs IV 14). The addition of POs to PMF improved the SFC due to the presence of high melting fraction (Jahurul *et al.*, 2014a) such as PPP. Further addition of POs IV 14 increased the level of PPP significantly ($p < 0.05$). However, PPP is an undesirable TAG as it provides waxiness to the end product (Cleenerwerck, 2010). The use of POs IV 14 in the formulation was low in percentage for fats F, G and H with values of 3%, 3% and 2%, respectively. However, fats F, G and H also consisted of 5%, 3% and 2% of POs IV 33, respectively. The value of PPP for fats F, G and H increased tremendously compared to fats A-E that have no POs IV 14 in the formulation and only comprised of POs IV 33 of 11%, 10%, 8%, 7% and 3%, respectively. There is potential of using single POs namely POs IV 14 as substitute to POs IV 33 if PPP could be of an assistance for the chocolate mouldability. All of the TAG of the blended fats were not comparable to CB. However, the increased level of PPP in the formulation facilitates crystallisation (Basso *et al.*, 2010) and eventually the mouldability property.

Crystallisation Rate at Isothermal Temperature

Crystallisation rates of all the fats at 5°C, 10°C and 20°C (isothermal temperature) are shown

in Figure 2. At 5°C, CB reached 80% SFC within 10 min and began to plateau at 90% SFC by 20th min. The eight fat blends, on the other hand, did not reach 50% SFC at 10th min crystallisation time. The SFC of all fat blends at 5°C were close to each other. Overlapping of SFC of all fats started to be observed by 35th-40th min of crystallisation time. PMF followed by fat E illustrated the slowest crystallisation rate at 5°C. At 10°C, CB reached 80% SFC by the 15th-20th min crystallisation time. By the 30th-35th min, the SFC of CB begun to plateau. Fats A, B, C, D, F, G and H demonstrated overlapping SFC by the 40th-45th min of crystallisation rate. POs which was part of the formulation played important role in this crystallisation pattern. PPP which is the highest melting fraction in POs influenced the fat crystallisation (Basso *et al.*, 2010; Vereecken *et al.*, 2009) as agent for seeding and crystallisation booster particularly for POP-containing system (Vereecken *et al.*, 2009). PMF and fat E still exhibited the lowest crystallisation rate at 10°C.

At 10th min, CB had the highest SFC compared to other fats at isothermal temperature of 20°C. Fat F showed the highest crystallisation rate from 1st-20th min compared to CB. Fats A, B, C, F, G and H demonstrated higher crystallisation rate than CB from 25th-35th min. At 40th min and above, CB had the highest SFC at 20°C imposing fastest crystallisation compared to other fats. Foubert *et al.* (2006) suggested that the shift of slope in CB may be due to the development of distinct polymorphic form or occurrence of various fractions crystallisation. Unblended PMF fat showed the poorest crystallisation performance at this isothermal temperature with only very minor increase of SFC towards 60th min. Similar result was obtained by Peyronel *et al.* (2016) when rapid crystallisation of PMF started to take place only after 1.5 hr. POP and PPP played important role in determining the rate of crystallisation at isothermal condition of 20°C of all the fats. Higher content of POP in PMF made the crystallisation rate slower. Fat E, having the highest POP content and lowest PPP content, resulted to the lowest crystallisation rate among all of the fats. This result was aligned with the study by Sonwai *et al.* (2014) who reported higher crystallisation rate of PMF when the POP level was low.

In the present study, crystallisation rate of all blended fats was found to have less effect at low temperature of 5°C. All of the blends have close crystallisation rate profile with CB. However, at 10°C and 20°C, crystallisation rate of all the blended fats varied from other another as well as with CB. At 10°C, the crystallisation rate of all the fat blends had similar plateau profile as CB after 40th min. Notwithstanding this, similar crystallisation rate profile was found for all of the fat blends with CB before 35th min at 20°C.

TABLE 3. FATTY ACID COMPOSITION OF VARIOUS FAT BLENDS

Fatty acid	Cocoa butter	Fat blends							
		A	B	C	D	E	F	G	H
Caprylic acid (C8:0)	-	0.12±0.04 ^a	0.14±0.00 ^a	0.16±0.01 ^a	0.17±0.01 ^a	0.11±0.04 ^a	0.17±0.03 ^a	0.17±0.02 ^a	0.15±0.02 ^a
Capric acid (C10:0)	-	0.07±0.01 ^b	0.11±0.00 ^a	0.11±0.02 ^a	0.11±0.01 ^a	0.07±0.01 ^b	0.1±0.00 ^a	0.11±0.00 ^a	0.09±0.00 ^{ab}
Lauric acid (C12:0)	-	0.70±0.05 ^c	1.06±0.05 ^a	1.09±0.05 ^a	1.09±0.08 ^a	0.72±0.08 ^{bc}	1.02±0.02 ^a	1.1±0.00 ^a	0.84±0.01 ^b
Myristic acid (C14:0)	0.10±0.00 ^b	1.63±0.26 ^a	1.80±0.12 ^a	1.57±0.19 ^a	1.74±0.05 ^a	1.8±0.30 ^a	1.89±0.57 ^a	1.59±0.00 ^a	1.41±0.04 ^a
Palmitic acid (C16:0)	25.97±0.00 ^e	49.35±0.10 ^{ab}	49.02±0.07 ^{bc}	49.53±0.30 ^c	48.48±0.20 ^c	48.54±0.13 ^c	46.92±0.48 ^d	49.92±0.02 ^a	49.64±0.08 ^a
Palmitoleic acid (C16:1)	0.70±0.00 ^a	0.17±0.01 ^b	0.15±0.02 ^b	0.19±0.01 ^b	0.16±0.00 ^b	0.17±0.02 ^b	0.17±0.05 ^b	0.17±0.01 ^b	0.17±0.02 ^b
Stearic acid (C18:0)	33.25±0.00 ^a	5.66±0.13 ^b	5.61±0.03 ^b	4.92±0.08 ^b	5.71±0.18 ^b	5.66±0.25 ^b	4.81±0.39 ^b	4.9±0.00 ^b	4.93±0.02 ^b
Oleic acid (C18:1)	32.05±0.00 ^f	34.48±0.08 ^e	34.46±0.09 ^e	34.66±0.06 ^{de}	34.83±0.07 ^{cd}	35.04±0.21 ^{bc}	38.01±0.20 ^a	34.51±0.02 ^{de}	35.2±0.11 ^b
Linoleic acid (C18:2)	3.56±0.00 ^c	6.84±0.09 ^a	6.73±0.01 ^a	6.89±0.20 ^a	6.84±0.07 ^a	6.87±0.01 ^a	6.14±0.09 ^b	6.67±0.01 ^a	6.77±0.04 ^a
Linolenic acid (C18:3)	0.42±0.00 ^a	0.32±0.03 ^b	0.26±0.02 ^{bc}	0.19±0.02 ^{cd}	0.28±0.00 ^b	0.32±0.02 ^b	0.18±0.07 ^d	0.17±0.02 ^d	0.16±0.01 ^d
Arachidic acid (C20:0)	1.23±0.00 ^a	0.54±0.06 ^{bc}	0.51±0.04 ^c	0.53±0.09 ^{bc}	0.47±0.02 ^c	0.65±0.05 ^b	0.44±0.05 ^c	0.54±0.01 ^{bc}	0.5±0.05 ^c
Others	0.32±0.00 ^a	0.14±0.01 ^b	0.14±0.00 ^b	0.16±0.03 ^b	0.14±0.01 ^b	0.14±0.01 ^b	0.17±0.04 ^b	0.15±0.00 ^b	0.14±0.01 ^b
SAFA	62.99±0.00 ^a	58.07±0.14 ^{ab}	58.26±0.10 ^{ab}	57.91±0.32 ^{ab}	57.77±0.13 ^{ab}	57.49±0.23 ^{ab}	55.34±0.12 ^{ab}	51.65±11.55 ^b	57.56±0.13 ^{ab}
MUFA	33.07±0.00 ^a	34.65±0.09 ^a	34.61±0.10 ^a	34.85±0.06 ^a	44.99±7.38 ^a	35.21±0.19 ^a	38.51±0.43 ^a	34.68±0.02 ^a	35.37±0.10 ^a
PUFA	3.46±0.00 ^c	7.15±0.07 ^a	6.70±0.02 ^a	7.08±0.22 ^a	7.12±0.07 ^a	7.19±0.01 ^a	6.32±0.06 ^b	6.84±0.02 ^a	6.92±0.04 ^a

Note: Each value in the table is the mean ± standard deviation of three measurement.

Means within each row bearing different superscript letters are significantly different (p<0.05).

Fat A - 88% PMF, 1% PKO, 11% POs IV 33.

Fat B - 89% PMF, 1% PKO, 10% POs IV 33.

Fat C - 91% PMF, 1% PKO, 8% POs IV 33.

Fat D - 92% PMF, 1% PKO, 7% POs IV 33.

Fat E - 96% PMF, 1% PKO, 3% POs IV 33.

Fat F - 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14.

Fat G - 93% PMF, 1% PKO, 3% POs IV 33 and 3% POs IV 14.

Fat H - 95% PMF, 1% PKO, 2% POs IV 33 and 2% POs IV 14.

SAFA - saturated fatty acid; MUFA - monounsaturated fatty acid; PUFA - polyunsaturated fatty acid;

PMF - palm mid fraction; PKO - palm kernel oil; POs - palm stearin.

TABLE 4. TRIACYLGLYCEROL (TAG) COMPOSITION OF VARIOUS FAT BLENDS

TAG (%)	Fats									
	Cocoa butter	A	B	C	D	E	F	G	H	
OLL	-	0.19±0.00 ^{ab}	0.17±0.03 ^b	0.14±0.01 ^b	0.25±0.01 ^a	0.19±0.00 ^{ab}	0.15±0.02 ^b	0.14±0.00 ^b	0.17±0.03 ^b	
PLL	-	1.48±0.03 ^a	1.26±0.23 ^a	1.22±0.07 ^a	1.53±0.02 ^a	1.50±0.00 ^a	1.20±0.22 ^a	1.14±0.17 ^a	1.23±0.29 ^a	
MLP	-	0.68±0.02 ^a	0.31±0.11 ^{abc}	0.36±0.01 ^{abc}	0.58±0.15 ^{ab}	0.69±0.00 ^a	0.22±0.00 ^{bc}	0.30±0.12 ^{abc}	0.44±0.27 ^{abc}	
OLO	-	0.55±0.00 ^a	0.60±0.00 ^a	0.59±0.01 ^a	0.60±0.00 ^a	0.55±0.01 ^a	0.58±0.00 ^a	0.57±0.02 ^a	0.55±0.03 ^a	
PLO	0.37±0.02 ^e	5.20±0.06 ^{bcde}	5.36±0.01 ^a	5.27±0.00 ^{ab}	5.21±0.02 ^{bcd}	5.25±0.04 ^{bc}	5.19±0.00 ^{bcd}	5.13±0.02 ^d	5.16±0.00 ^{cd}	
PLP	1.61±0.17 ^e	10.88±0.07 ^{ab}	10.38±0.05 ^{bcde}	10.34±0.01 ^{cd}	10.74±0.09 ^{abc}	11.06±0.14 ^a	10.10±0.19 ^d	10.30±0.08 ^{cd}	10.10±0.24 ^d	
MOP	0.22±0.04 ^a	-	-	-	-	-	-	-	-	
MPP	-	0.65±0.01 ^{bc}	0.64±0.01 ^{bc}	0.65±0.01 ^{bc}	0.62±0.00 ^{bc}	0.54±0.00 ^d	0.73±0.02 ^a	0.66±0.00 ^b	0.61±0.02 ^c	
OOO	0.17±0.00 ^b	1.12±0.18 ^a	1.34±0.01 ^a	1.32±0.00 ^a	1.34±0.04 ^a	1.19±0.03 ^a	1.29±0.03 ^a	1.29±0.00 ^a	1.29±0.01 ^a	
SLO	0.18±0.02 ^a	-	-	-	-	-	-	-	-	
POO	1.40±0.01 ^b	12.70±0.76 ^a	13.49±0.02 ^a	13.34±0.10 ^a	13.35±0.18 ^a	13.04±0.06 ^a	13.17±0.08 ^a	13.19±0.10 ^a	13.32±0.12 ^a	
POP	16.18±0.21 ^c	48.97±0.33 ^b	48.58±0.06 ^b	49.20±0.33 ^{ab}	48.92±0.32 ^b	50.28±0.56 ^a	48.76±0.23 ^b	49.35±0.03 ^{ab}	49.61±0.32 ^{ab}	
PLS	2.78±0.01 ^a	-	-	-	-	-	-	-	-	
PPP	4.09±0.03 ^c	4.09±0.03 ^c	4.18±0.00 ^b	3.83±0.03 ^d	3.23±0.00 ^f	2.35±0.02 ^g	4.79±0.03 ^a	4.26±0.02 ^b	3.44±0.02 ^e	
SOO	1.57±0.00 [*]	1.82±0.04 ^a	1.92±0.01 ^a	1.91±0.05 ^a	1.85±0.01 ^a	1.87±0.05 ^a	1.94±0.02 ^a	1.85±0.01 ^a	1.94±0.11 ^a	
POS	39.26±0.01 ^b	9.16±0.12 ^a	9.39±0.24 ^a	9.32±0.02 ^a	9.29±0.08 ^a	9.47±0.16 ^a	9.37±0.02 ^a	9.37±0.06 ^a	9.58±0.22 ^a	
PPS	0.53±0.02 ^d	0.99±0.03 ^{abc}	1.05±0.01 ^{abc}	0.96±0.01 ^{abc}	0.82±0.01 ^{bcd}	0.75±0.17 ^{cd}	1.18±0.08 ^a	1.12±0.14 ^{ab}	0.88±0.04 ^{abc}	
SOS	31.11±0.33 ^a	1.32±0.43 ^b	1.10±0.01 ^b	1.27±0.23 ^b	1.38±0.11 ^b	1.09±0.05 ^b	1.11±0.05 ^b	1.08±0.02 ^b	1.31±0.34 ^b	
SSP	0.80±0.01 ^a	0.20±0.02 ^{bc}	0.22±0.01 ^{bc}	0.28±0.04 ^{bc}	0.28±0.03 ^{bc}	0.18±0.05 ^c	0.23±0.01 ^{bc}	0.25±0.06 ^{bc}	0.36±0.09 ^b	
SLS	0.91±0.11 ^a	-	-	-	-	-	-	-	-	
AOO	0.42±0.01 ^a	-	-	-	-	-	-	-	-	
SOA	2.02±0.03 ^a	-	-	-	-	-	-	-	-	
SSS	0.49±0.01 ^a	-	-	-	-	-	-	-	-	
UUU	0.17±0.00 ^b	1.12±0.18 ^a	1.34±0.01 ^a	1.32±0.00 ^a	1.34±0.04 ^a	1.19±0.03 ^a	1.29±0.03 ^a	1.29±0.00 ^a	1.29±0.01 ^a	
SUU	1.81±0.02 ^b	14.52±0.72 ^a	15.42±0.02 ^a	15.25±0.06 ^a	15.20±0.17 ^a	14.91±0.11 ^a	15.11±0.10 ^a	15.04±0.09 ^a	15.26±0.22 ^a	
SUS	88.78±0.09 ^a	59.45±0.89 ^{bc}	59.07±0.28 ^c	59.79±0.08 ^{bc}	59.60±0.28 ^{bc}	60.84±0.35 ^{bc}	59.23±0.26 ^c	59.79±0.05 ^{bc}	60.50±0.44 ^{bc}	
SSS**	7.12±0.08 ^a	18.97±0.08 ^a	18.05±0.28 ^{bc}	17.64±0.12 ^{cd}	17.80±0.07 ^{bc}	17.06±0.25 ^d	18.44±0.15 ^{ab}	18.03±0.00 ^{bc}	17.06±0.23 ^d	

Note: M - myristic; P - palmitic; S - stearic, O - oleic; L - linoleic; UUU - triunsaturated; SUS - disaturated; SSS** - trisaturated.

* The value is shared with PPP and SOO.

Each value in the table is the mean ± standard deviation of two measurements.

Means within each row bearing different superscript letters are significantly different (p<0.05).

Fat A - 88% PMF, 1% PKO, 11% POs IV 33.

Fat B - 89% PMF, 1% PKO, 10% POs IV 33.

Fat C - 91% PMF, 1% PKO, 8% POs IV 33.

Fat D - 92% PMF, 1% PKO, 7% POs IV 33.

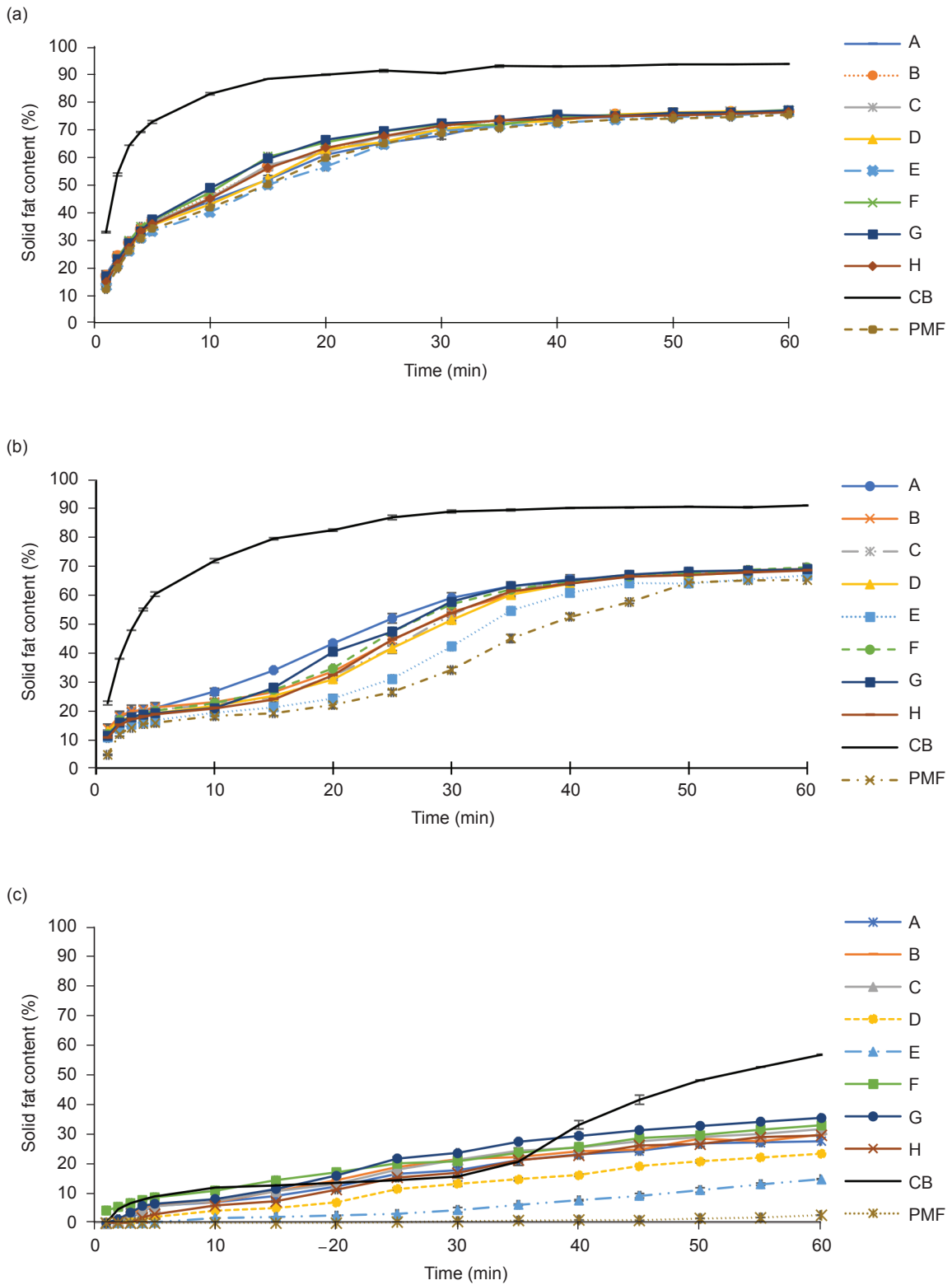
Fat E - 96% PMF, 1% PKO, 3% POs IV 33.

Fat F - 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14.

Fat G - 93% PMF, 1% PKO, 3% POs IV 33 and 3% POs IV 14.

Fat H - 95% PMF, 1% PKO, 2% POs IV 33 and 2% POs IV 14.

PMF - palm mid fraction; PKO - palm kernel oil; POs - palm stearin.



Note: Fat A - 88% PMF, 1% PKO, 11% POs IV 33.
 Fat B - 89% PMF, 1% PKO, 10% POs IV 33.
 Fat C - 91% PMF, 1% PKO, 8% POs IV 33.
 Fat D - 92% PMF, 1% PKO, 7% POs IV 33.
 Fat E - 96% PMF, 1% PKO, 3% POs IV 33.
 Fat F - 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14.
 Fat G - 93% PMF, 1% PKO, 3% POs IV 33 and 3% POs IV 14.
 Fat H - 95% PMF, 1% PKO, 2% POs IV 33 and 2% POs IV 14.
 CB - cocoa butter; PMF - palm mid fraction; PKO - palm kernel oil; POs - palm stearin.

Figure 2. Crystallisation rate of all fats at isothermal temperature of (a) 5°C, (b) 10°C, and (c) 20°C.

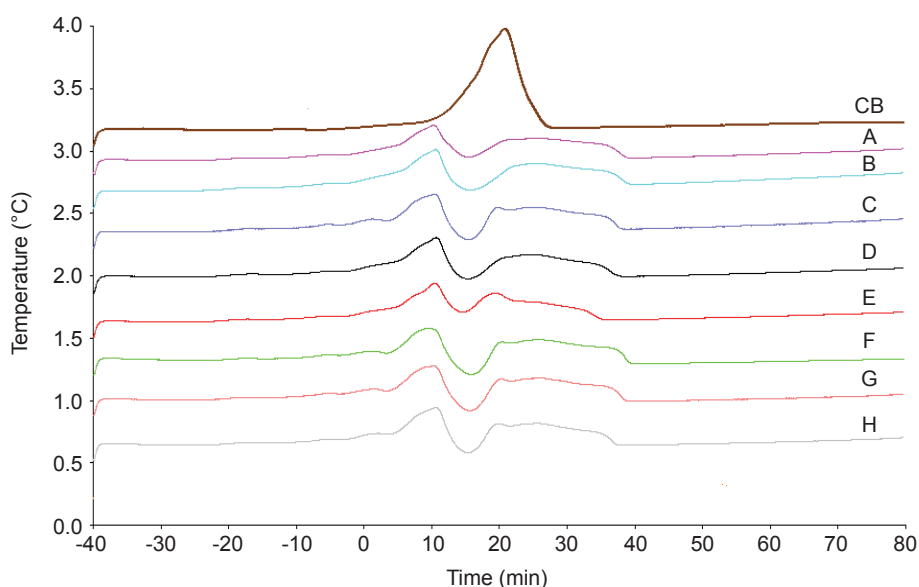
Thermal Behaviour

Thermal melting profiles of all fats are shown in Figure 3. CB produced one sharp thermal melting peak at 22°C which is similar to the finding by Sonwai *et al.* (2014). Two endothermic peaks were observed; low and high melting peaks having sharp and broad peaks, respectively were observed for all the fats. The range of the sharp peaks was from 9.26°C-10.64°C while for the broad peak was from 25.38°C-27.89°C. As all of the fats were PMF dominant, the result was in accordance with the study of PMF by Sonwai *et al.* (2014) in which PMF produced two distinguished peaks. These fats were observed to have small difference in melting temperatures and were mostly influenced by FAC and TAG compositions (Tables 3 and 4). The low melting peaks may consist of UUU and SUU TAG such as OOO, POO and SOO while the high melting peaks may be an overlapping of saturated-saturated-unsaturated (SSU), SUS and SSS TAG such as PPO and POP for the SSU and SUS TAG and PPP for the SSS TAG. It was observed that the higher the level of PMF in the fats (indicated by fats A-H in Figure 3 having PMF in increasing trend), the low melting peaks shifted more to the left producing slightly lower melting temperature. In addition, lesser POs made the melting curves moved to the left having slightly lower melting temperature too. The broad melting peak may suggest tailing or waxiness to the end product as it may consist of PPP TAG. In

general, thermal behaviour of all the fat blends was different from CB. However, they had broad high melting peaks that may assist crystallisation rate and consequently the mouldability property.

Polymorphism

Crystallisation of a molecule in distinctive configuration of crystal packing is defined as polymorphism. The static crystallisation of CB, PMF, PKO, POs IV 33 and IV 14 and fats A-H at 10°C for 30, 45 and 60 min is demonstrated in Figure 4. At 10°C, CB displayed β' polymorphism having two spacings at 4.2 Å and 3.9 Å. Similar findings were reported by Marangoni and McGauley (2003) in which they observed β' polymorphism for CB for temperature between -15°C and 20°C. Three peaks at spacings of 3.8 Å, 4.2 Å and 4.3 Å were detected for individual PMF having high percentage of POP. The spacings of 3.8 Å and 4.2 Å demonstrated β' polymorphism and the spacing of 4.3 Å was approximate of that pseudo- β' configuration (Sonwai *et al.*, 2014). POs IV 33 showed spacings of 4.6 Å, 4.3 Å, 4.2 Å and 3.9 Å indicating β' and β polymorphism. POs IV 14 (high in PPP) showed spacing at 4.5 Å, 4.1/4.2 Å and 3.9 Å indicating β' and β polymorphism. PKO (high in LaLaLa) had β' polymorphism from the spacings of 3.8 Å, 4.2 Å and 4.4 Å which was similar to the finding by Zhou *et al.* (2010).



Note: Fat A - 88% PMF, 1% PKO, 11% POs IV 33.
 Fat B - 89% PMF, 1% PKO, 10% POs IV 33.
 Fat C - 91% PMF, 1% PKO, 8% POs IV 33.
 Fat D - 92% PMF, 1% PKO, 7% POs IV 33.
 Fat E - 96% PMF, 1% PKO, 3% POs IV 33.
 Fat F - 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14.
 Fat G - 93% PMF, 1% PKO, 3% POs IV 33 and 3% POs IV 14.
 Fat H - 95% PMF, 1% PKO, 2% POs IV 33 and 2% POs IV 14.
 CB - cocoa butter; PMF - palm mid fraction; PKO - palm kernel oil; POs - palm stearin.

Figure 3. Melting profiles of blended fats and cocoa butter.

β polymorphism is promoted by trisaturated TAG such as PPP (Nusantoro, 2009; Basso *et al.*, 2010) and SSS (Oleivera *et al.*, 2015). Sato (2001b) and Biswas *et al.* (2017) reported that POP/POS were accountable for β' polymorphism.

Fats A-H (high in POP) tempered at 10°C for 30, 45 and 60 min (Figures 4a, b and c) showed β' polymorphism with spacings of 3.8 Å, 4.1/4.2 Å and 4.3 Å were similar to PMF as PMF was the main fat used in the formulation. Spacing between 3.8-4.3 Å was referred to as having β' polymorphism (Biswas *et al.*, 2017). The amounts of POs IV 33 and 14 were too small to be detected at spacing of 4.5/4.6 Å that represented β polymorphism. The physical structure of fats may also change with time at constant temperature because of polymorphic transformations (Metin and Hartel, 2005). However, the experimental time difference in this study may be too short for the polymorphic transformation. In this study, all blended fats were comparable with CB (β' dominance) in terms of polymorphism at 10°C.

Crystal Morphology

By using PLM, crystal morphology of individual CB, PMF, POs IV 33, POs IV 14, PKO as well as ternary and quaternary fats of PMF, POs IV 33, POs IV 14 and PKO at 10°C, 20°C and 25°C were observed (Figures 5-7). Crystallisation of fat is crucial for fat-based product developments particularly in chocolate confectionery. Crystallisation of fat consists of several steps which include primary crystallisation, development of microstructure, formation of continuous 3-dimensional crystal network and finally formation of solid bridge called sintering between aggregated crystals and aggregates (Delbaere *et al.*, 2016).

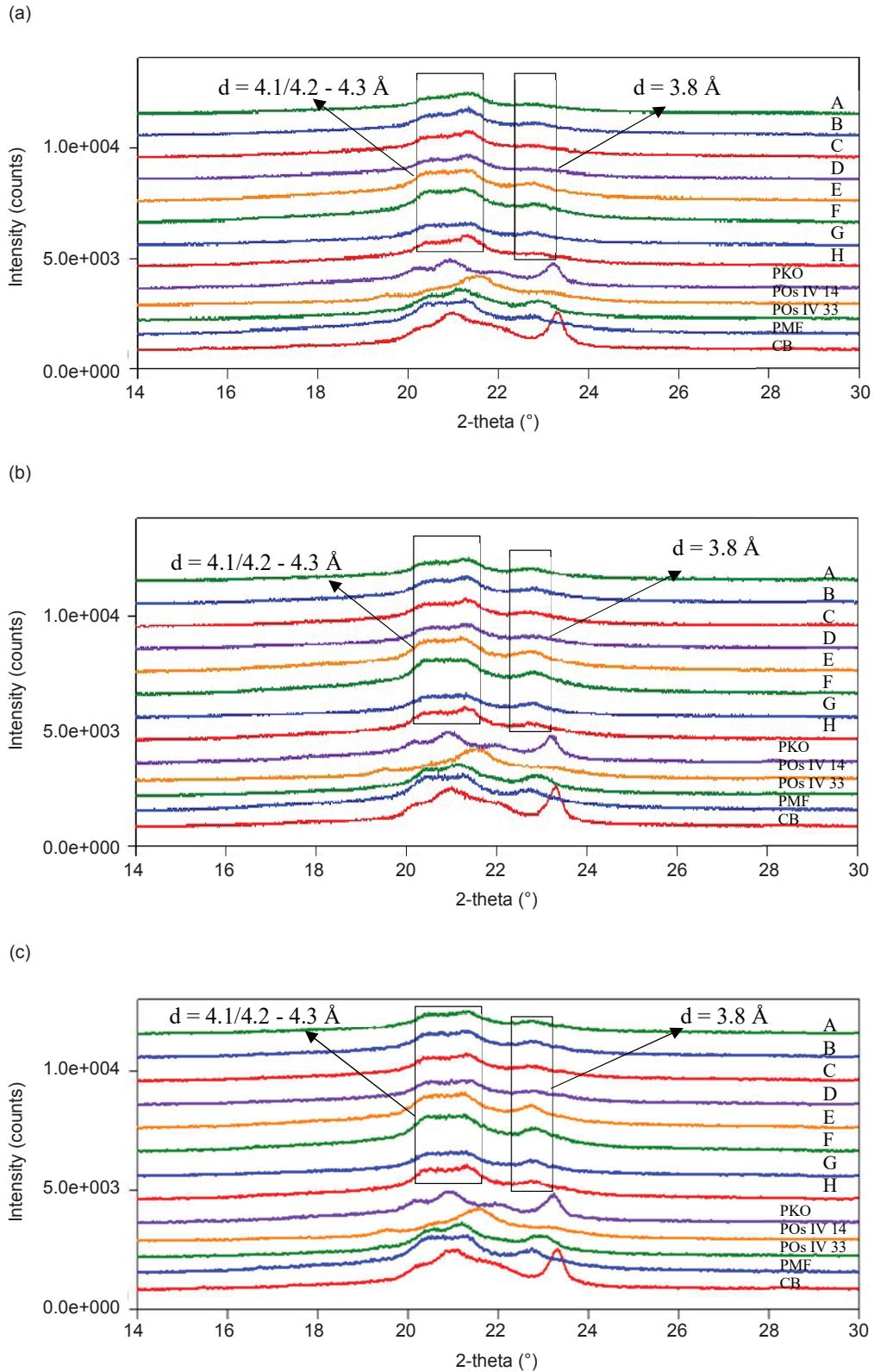
At 10°C, all fats including CB, PMF, POs IV 33, POs IV 14 and PKO showed dense crystal structures. CB and PKO had small granular and well dispersed crystal structures. POs IV 33, on the other hand, had rougher but well dispersed crystal structure. PMF showed unclear spherulitic needle-like crystal structure radiating and branching out from the central nucleus due to low temperature. It could be mistaken with granular crystal structure. All of the blended fats did not show much variation in crystal structure and reflected crystal structure of unclear spherulitic needle-like crystal structure radiating and branching out from the central nucleus and granular. Among the blended fats, fat F had the densest crystal structure due to the addition of POs IV 14. There was a declining trend of crystal packing from fat A to fat E as well as from fat F to fat H indicating decreasing amounts of POs in the fat blends.

At 20°C, all fats including CB, PMF, POs IV 33, POs IV 14 and PKO had slightly lost their crystal packing compared to fats at 10°C. Fats A, B, C,

D and E showed spherulitic needle-like crystal structure radiating and branching out from the central nucleus at 20°C. This structure became clearer when the level of POs lessened. These fats showed a good trend of decreasing POs IV 33 when these crystals became less dense in packing when only 3% of POs IV 33 was added to fat E. Similar scenario was observed by Norizzah *et al.* (2004) when palm kernel olein was added to POs. Structures of evenly distributed small granular crystal were observed in fats F and G due to the addition of POs IV 14. These crystals might be more stable (especially in terms of heat) than other fats as they have denser packing and evenly distributed small crystal structure. Small and dense crystal microstructure might implicate the macroscopic and mechanical attributes of the fat (*i.e.* hardness) as reported by Bootello *et al.* (2018) for their sunflower stearin-based CBE. Fat H having lesser POs IV 14 (2%) was less dense in packing compared to fats F and G.

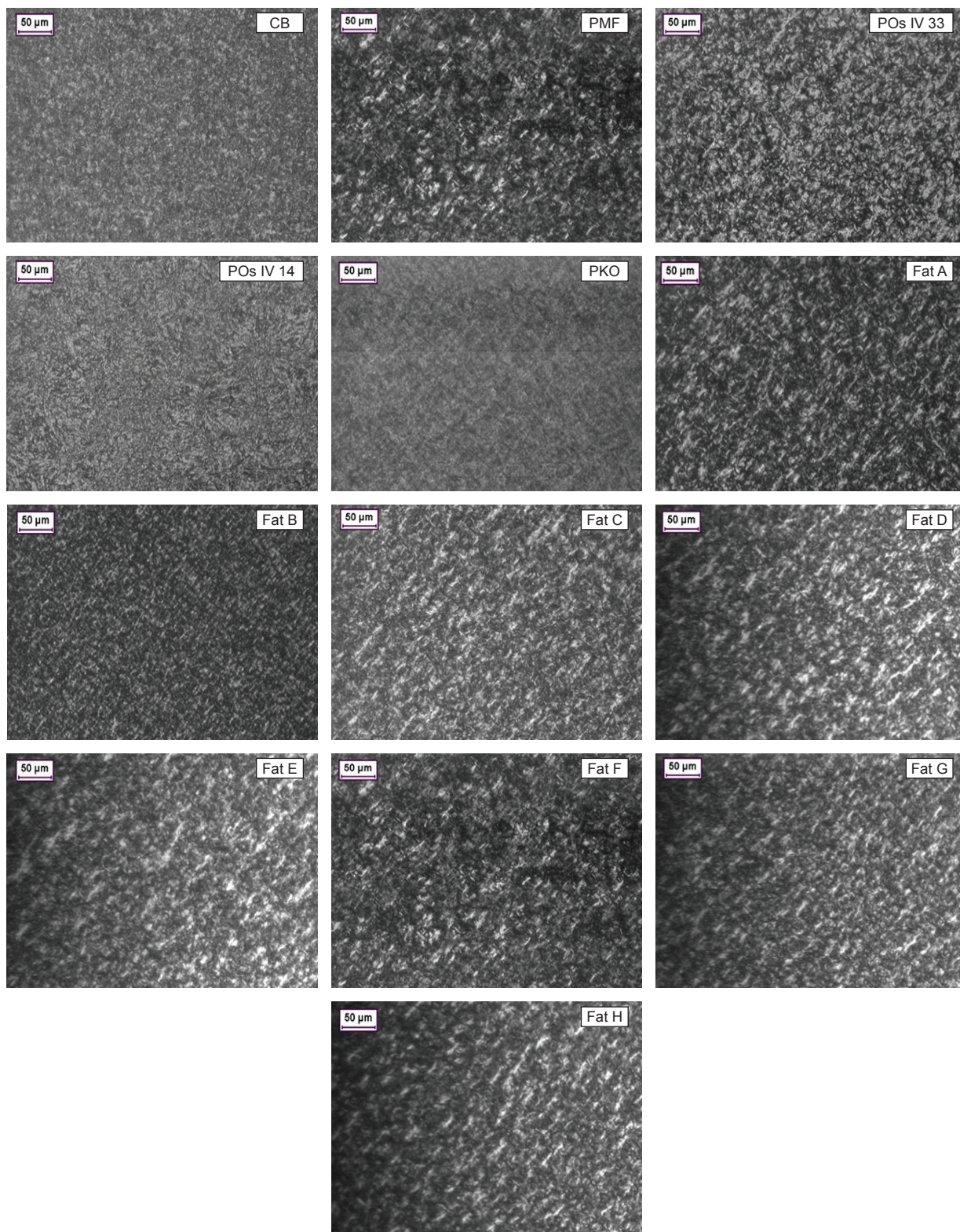
Spherulitic needle-like crystals structure was observed in commercial CB at 25°C. The structure was similar to the findings by Sonwai *et al.* (2014). Bigger and clearer spherulitic needle-like crystals structure was observed in PMF. Granular crystals were observed for POs IV 33 and POs IV 14. Granular crystals of POs IV 14 were observed to have denser packing. The morphology of PKO displayed feather-like crystals. It was observed that the crystal structure of fats A, B, C, D and E were quite similar to one another having spherulitic needle-like crystal structure radiating and branching out from the central nucleus. However, they were different in terms of crystal packing depending on the amounts of POs IV 33. Notwithstanding this, similar crystal structures were found at 20°C with less dense packing. Fats F, G and H also showed spherulitic needle-like crystal structure radiating and branching out from the central nucleus. However, the nucleus of the structure had become larger. The decremental addition of POs IV 33 and IV 14 to PMF led to the development of large and loosely packed crystals. Fat F showed the densest crystal packing compared to other fats. According to Delbaere *et al.* (2016), a denser crystal packing in chocolate reduced overall mobility of fat resulting to the decreased rate of oil migration.

It can be concluded that crystal structure for all fats at temperatures of 10°C, 20°C and 25°C varied from one to another. The disparity might be due to different TAG (Table 4) composition and distinct textural properties of PMF, PKO and POs IV 33 and IV 44 (Çiftçi *et al.*, 2009) which relates to the chemical composition of the fat as well as temperature. In addition, crystal size is dependent on temperature history. The packing (density) of fat F was comparable with CB and therefore, might impose good mouldability property.



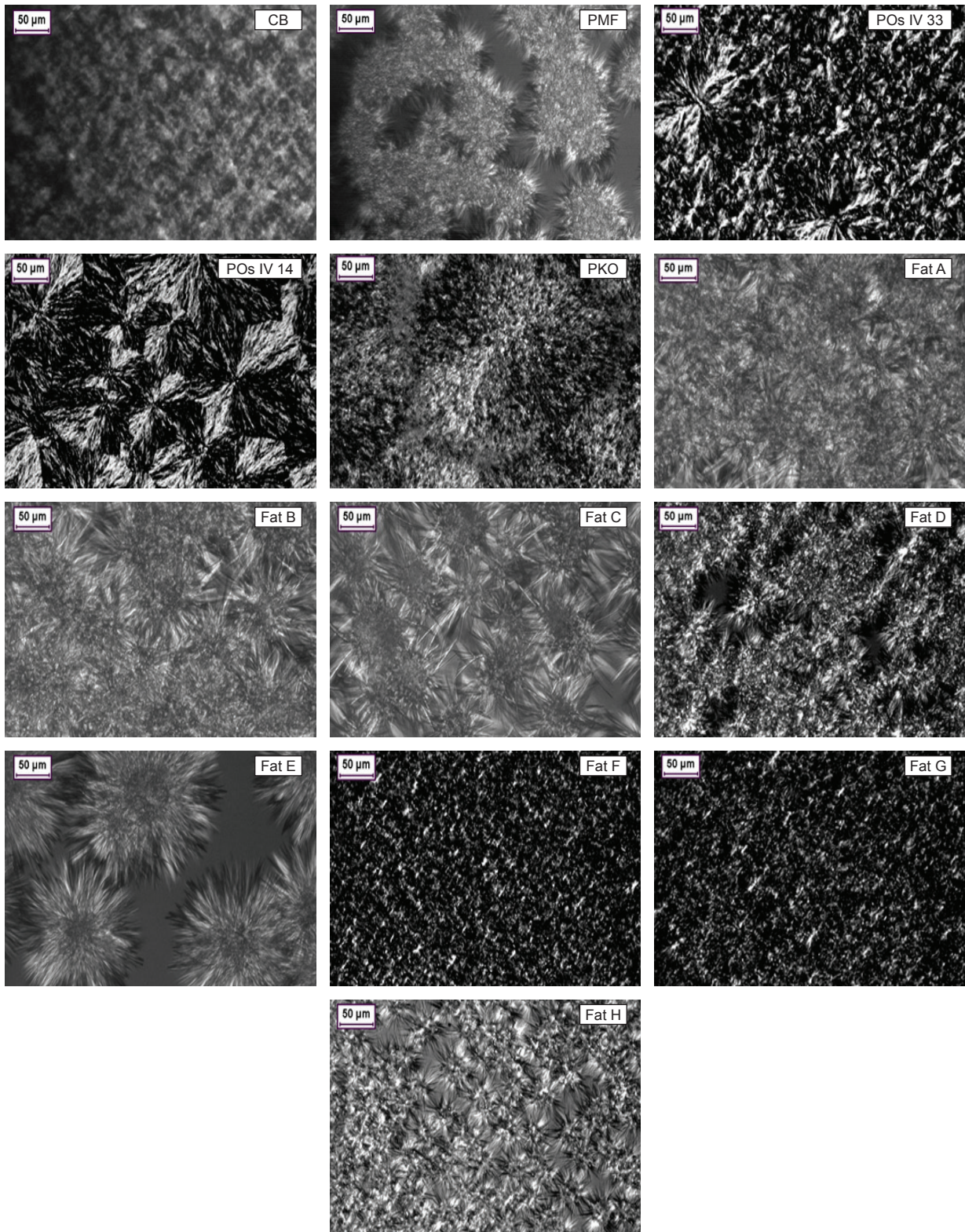
Note: Fat A - 88% PMF, 1% PKO, 11% POs IV 33.
 Fat B - 89% PMF, 1% PKO, 10% POs IV 33.
 Fat C - 91% PMF, 1% PKO, 8% POs IV 33.
 Fat D - 92% PMF, 1% PKO, 7% POs IV 33.
 Fat E - 96% PMF, 1% PKO, 3% POs IV 33.
 Fat F - 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14.
 Fat G - 93% PMF, 1% PKO, 3% POs IV 33 and 3% POs IV 14.
 Fat H - 95% PMF, 1% PKO, 2% POs IV 33 and 2% POs IV 14.
 PKO - palm kernel oil; POs - palm stearin; PMF - palm mid fraction; CB - cocoa butter.

Figure 4. Polymorphism of all fats at (a) 30 min, (b) 45 min, and (c) 60 min.



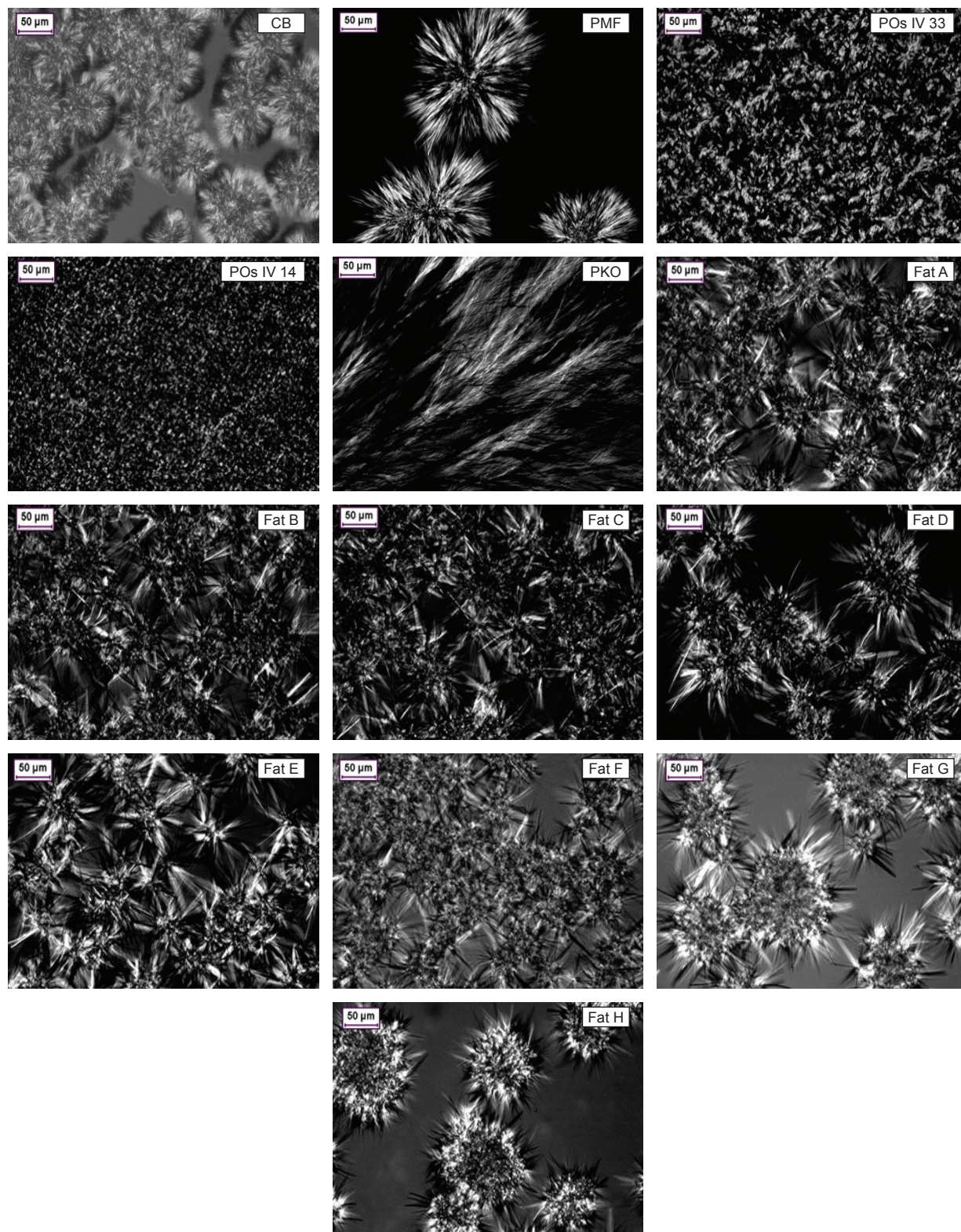
Note: Fat A - 88% PMF, 1% PKO, 11% POs IV 33.
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 Fat C - 91% PMF, 1% PKO, 8% POs IV 33.
 Fat D - 92% PMF, 1% PKO, 7% POs IV 33.
 Fat E - 96% PMF, 1% PKO, 3% POs IV 33.
 Fat F - 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14.
 Fat G - 93% PMF, 1% PKO, 3% POs IV 33 and 3% POs IV 14.
 Fat H - 95% PMF, 1% PKO, 2% POs IV 33 and 2% POs IV 14.
 PMF - palm mid fraction; POs - palm stearin; PKO - palm kernel oil; CB - cocoa butter.

Figure 5. Polarised light micrographs (x20 lens) of cocoa butter, PMF, POs IV 33, POs IV 14, PKO and fats A-H at 10°C.



Note: Fat A - 88% PMF, 1% PKO, 11% POs IV 33.
 Fat B - 89% PMF, 1% PKO, 10% POs IV 33.
 Fat C - 91% PMF, 1% PKO, 8% POs IV 33.
 Fat D - 92% PMF, 1% PKO, 7% POs IV 33.
 Fat E - 96% PMF, 1% PKO, 3% POs IV 33.
 Fat F - 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14.
 Fat G - 93% PMF, 1% PKO, 3% POs IV 33 and 3% POs IV 14.
 Fat H - 95% PMF, 1% PKO, 2% POs IV 33 and 2% POs IV 14.
 PMF - palm mid fraction; POs - palm stearin; PKO - palm kernel oil; CB - cocoa butter.

Figure 6. Polarised light micrographs (x20 lens) of cocoa butter, PMF, POs IV 33, POs IV 14, PKO and fats A-H at 20°C.



Note: Fat A - 88% PMF, 1% PKO, 11% POs IV 33.
 Fat B - 89% PMF, 1% PKO, 10% POs IV 33.
 Fat C - 91% PMF, 1% PKO, 8% POs IV 33.
 Fat D - 92% PMF, 1% PKO, 7% POs IV 33.
 Fat E - 96% PMF, 1% PKO, 3% POs IV 33.
 Fat F - 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14.
 Fat G - 93% PMF, 1% PKO, 3% POs IV 33 and 3% POs IV 14.
 Fat H - 95% PMF, 1% PKO, 2% POs IV 33 and 2% POs IV 14.
 PMF - palm mid fraction; POs - palm stearin; PKO - palm kernel oil; CB - cocoa butter.

Figure 7. Polarised light micrographs (x20 lens) of cocoa butter, PMF, POs IV 33, POs IV 14, PKO and fats A-H at 25°C.

Mouldability

Table 5 shows the mouldability of chocolate bars from all blended fats at cooling time of 30, 45 and 60 min. Cooling temperature of 10°C-15°C is typically used by commercial chocolate manufacturers (CB-based) but cooling temperature of 10°C is normally required to eliminate the sensible and latent heat from the liquid chocolate for easy demoulding in order to produce solid chocolate product. The time needed for cooling and solidification relies upon the temperature of the cooling air which translates as the rate of heat transfer from chocolate product to air (Keijbets *et al.*, 2010). The solidification of chocolate that relates to crystal structure renders the hardness of chocolate (Jahurul *et al.*, 2013) and subsequently influences its mouldability.

Chocolate bars prepared from CB and palm-based CBA are able to be moulded and demoulded between 15-20 min (Keijbets *et al.*, 2010; MPOC, 2006; Bolliger *et al.*, 1998). However, chocolate bars from the blended fats were unable to be demoulded even at 30 min of cooling time. Despite of this, chocolate bars from all blended fats were able to be demoulded at 45 and 60 min of cooling time without fracture. Although polymorphism of all fats observed using XRD that were conditioned for 30, 45 and 60 min at 10°C showed β' dominance, the mouldability outcome varied in which chocolate bars from all of the blended fats were mouldable at 45 and 60 min but was not mouldable at 30 min. Solidification time which relates to crystallisation rate plays important role in this scenario. Figure 2 demonstrates that at 40-45 min of crystallisation at isothermal temperature of 10°C, all fats showed overlapping SFC (approximate value of 65%-70% SFC) and began to plateau indicating equilibrium phase had been reached. Therefore, they may be able to be moulded and demoulded easily. Optimised cooling time for all the fats was at 45 min at isothermal temperature of 10°C due to complete crystallisation. Prolonging the crystallisation time gave similar mouldability result to 45 min crystallisation time. In addition, all of the eight fat blends regardless of formulations were able to produce similar mouldability result. However, crystal morphology among all fats were dissimilar which may influence the snappability. In addition, Wainwright (1996) reported that CBS that has higher crystallisation rate compared to CBR imposes rapid crystallisation and has better manufacturing advantage than CBR.

Mechanical Resistance (Snappability)

Table 6 shows the snappability value (rupture tension and breaking force) of chocolate bars from various blended fats at cooling time of 45 and 60 min at 10°C right after demoulding as the chocolate bars at 45 and 60 min were able to be demoulded without

being fractured. Chocolate bars prepared at cooling time of 30 min were omitted as they fractured during demoulding process at 10°C. The snappability of chocolate bars at 45 and 60 min was significantly different from each other ($p < 0.05$). However, fats A, B, C, D, G and H were insignificantly different from each other ($p > 0.05$) at 60 min. Fat F produced the highest rupture tension and breaking force both at 45 and 60 min of chocolate bars at 10°C while fat E showed the lowest values for both parameters. The rupture tension of chocolate bars of fat F at 45 and 60 min were 105.28 $g_f \text{ cm}^{-2}$ and 107.92 $g_f \text{ cm}^{-2}$, respectively while the breaking force of fat F at 45 and 60 min were 5501 g_f and 5639 g_f , respectively. Fat F had the densest crystal structure compared to other fats at 10°C and had influenced on the snappability value.

The addition, POs IV 33 and POs IV 14 significantly influenced the snappability values when compared to 100% PMF (having the lowest snappability value). Therefore, the addition of POs IV 33 and POs IV 14 influenced the snappability value of the chocolate bar. Increasing trend of snappability values were observed with increasing amount of added POs IV 33 and POs IV 14. However, the increase in snappability value became more prominent when POs IV 14 was added. This also showed that POs IV 33 and POs IV 14 are chemically compatible with PMF. Snappability of chocolate is associated to ingredients in the formulation (Quast *et al.*, 2011) which consequently relates to the mechanical strength of network of fat crystal (Smith, 2009; Marangoni, 2002). Minor changes in the TAG composition can give huge impact on its crystallisation and further development of the fat structure (De Graef *et al.*, 2012; Himawan *et al.*, 2006 and Sato, 2001a). Incompatible fats such as blend of CB with lauric-based CBS showed decreased snappability value due to incompatibility between TAG as has been reported in many articles including Quast *et al.* (2013). Right crystal formation and its homogeneity also influenced snappability as reported by Rowat *et al.* (2011).

According to Timms (2003), chocolate should be crystalline at 20°C and therefore will impart good snappability. However, all of the chocolate using fats A-H had difficulties to retain their shape at 20°C and therefore, the best fat having highest rupture tension and breaking force at 10°C (fat F) was blended with CB.

Compatibility with Cocoa Butter

The compatibility of fat F and CB at 20°C is illustrated in Figure 8. Compatibility of fats refers to eutecticity (softening) when different types of molecular volume, shape and polymorph are found for each fat (Gold *et al.*, 2011). Fat F was found to be fully miscible with CB with no

significant eutecticity observed except for dilution and dissolution effect. However, 70% and 80% addition of fat F to CB showed minor lowering of SFC. Addition of 10%-30% of fat F to CB provided the best mixtures as there were no lowering of SFC observed and they had high SFC as well. Sonwai *et al.* (2014) reported that their blended CBE from mango kernel fat and PMF with CB having a decreasing trend of SFC with different CBE/CB ratio as being fully compatible and does not show

any sign of eutecticity. Jeyarani and Reddy (1999) also reported that their mixtures of kokum fat and mahua stearin which showed an increasing trend of SFC as fully compatible with each other with no major changes in solid phase or melting range other than changes in dilution and dissolution. Fat F (30%) and CB (70%) was able to be demoulded by 30 min and produced snappability (breaking force) of $5032.71 \pm 85.91 \text{ g}_f$ at 20°C after one day stabilisation period.

TABLE 5. MOULDABILITY OF CHOCOLATE BARS AT COOLING TIME OF 30 MIN, 45 MIN AND 60 MIN AT 10°C

Fat	Cooling time 30 min	Cooling time 45 min	Cooling time 60 min
A	Not mouldable	Mouldable	Mouldable
B	Not mouldable	Mouldable	Mouldable
C	Not mouldable	Mouldable	Mouldable
D	Not mouldable	Mouldable	Mouldable
E	Not mouldable	Mouldable	Mouldable
F	Not mouldable	Mouldable	Mouldable
G	Not mouldable	Mouldable	Mouldable
H	Not mouldable	Mouldable	Mouldable

Note: Score reading is average of three replicates.

Fat A - 88% PMF, 1% PKO, 11% POs IV 33.

Fat B - 89% PMF, 1% PKO, 10% POs IV 33.

Fat C - 91% PMF, 1% PKO, 8% POs IV 33.

Fat D - 92% PMF, 1% PKO, 7% POs IV 33.

Fat E - 96% PMF, 1% PKO, 3% POs IV 33.

Fat F - 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14.

Fat G - 93% PMF, 1% PKO, 3% POs IV 33 and 3% POs IV 14.

Fat H - 95% PMF, 1% PKO, 2% POs IV 33 and 2% POs IV 14.

PMF - palm mid fraction; CB - cocoa butter; PKO - palm kernel oil; POs - palm stearin.

TABLE 6. RUPTURE TENSION AND BREAKING FORCE OF CHOCOLATE BARS AT COOLING TIME OF 45 MIN AND 60 MIN AT 10°C

Fat	45 min		60 min	
	Rupture tension ($\text{g}_f \text{ cm}^{-2}$)	Breaking force (g_f)	Rupture tension ($\text{g}_f \text{ cm}^{-2}$)	Breaking force (g_f)
A	$86.81 \pm 11.22^{\text{bcd}}$	$4536 \pm 586^{\text{bcd}}$	$93.57 \pm 2.56^{\text{bc}}$	$4889 \pm 134^{\text{bc}}$
B	$85.45 \pm 7.81^{\text{cde}}$	$4465 \pm 408^{\text{cde}}$	$88.11 \pm 6.26^{\text{bc}}$	$4604 \pm 327^{\text{bc}}$
C	$74.91 \pm 1.61^{\text{def}}$	$3914 \pm 84^{\text{def}}$	$84.25 \pm 4.82^{\text{bc}}$	$4402 \pm 252^{\text{bc}}$
D	$72.63 \pm 1.59^{\text{ef}}$	$3795 \pm 83^{\text{ef}}$	$83.23 \pm 6.72^{\text{bc}}$	$4349 \pm 351^{\text{bc}}$
E	$67.37 \pm 3.69^{\text{f}}$	$3520 \pm 193^{\text{f}}$	$81.97 \pm 7.58^{\text{c}}$	$4283 \pm 396^{\text{c}}$
F	$105.28 \pm 6.51^{\text{ab}}$	$5501 \pm 340^{\text{ab}}$	$107.92 \pm 4.42^{\text{b}}$	$5639 \pm 231^{\text{b}}$
G	$92.06 \pm 1.00^{\text{bcd}}$	$4810 \pm 52^{\text{bcd}}$	$95.08 \pm 0.80^{\text{bc}}$	$4968 \pm 42^{\text{bc}}$
H	$86.33 \pm 3.37^{\text{bc}}$	$4511 \pm 176^{\text{bc}}$	$92.27 \pm 7.77^{\text{bc}}$	$4821 \pm 406^{\text{bc}}$
PMF	$66.26 \pm 4.34^{\text{f}}$	$3462 \pm 227^{\text{f}}$	$91.41 \pm 4.00^{\text{bc}}$	$4776 \pm 209^{\text{bc}}$
CB	$121.44 \pm 8.56^{\text{a}}$	$6345 \pm 447^{\text{a}}$	$162.39 \pm 20.84^{\text{a}}$	$8485 \pm 1089^{\text{a}}$

Note: Score reading is average of three replicates.

Means within each row bearing different superscript letters are significantly different ($p < 0.05$).

Fat A - 88% PMF, 1% PKO, 11% POs IV 33.

Fat B - 89% PMF, 1% PKO, 10% POs IV 33.

Fat C - 91% PMF, 1% PKO, 8% POs IV 33.

Fat D - 92% PMF, 1% PKO, 7% POs IV 33.

Fat E - 96% PMF, 1% PKO, 3% POs IV 33.

Fat F - 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14.

Fat G - 93% PMF, 1% PKO, 3% POs IV 33 and 3% POs IV 14.

Fat H - 95% PMF, 1% PKO, 2% POs IV 33 and 2% POs IV 14.

PMF - palm mid fraction; CB - cocoa butter; PKO - palm kernel oil; POs - palm stearin.

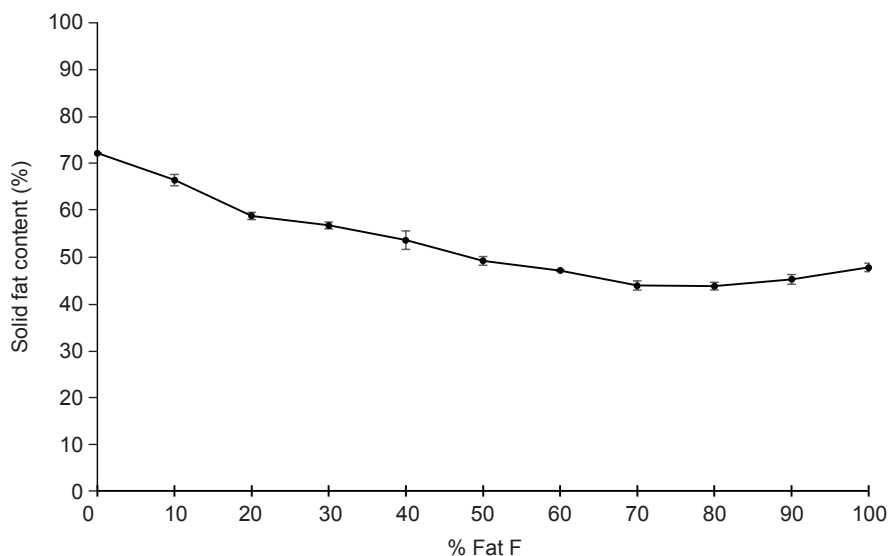


Figure 8. Compatibility of fat F (91% palm mid fraction: 1% palm kernel oil: 5% palm stearin IV 33: 3% palm stearin IV 14) with cocoa butter at 20°C.

CONCLUSION

Eight fat blends namely A) 88% PMF, 1% PKO, 11% POs IV 33, B) 89% PMF, 1% PKO, 10% POs IV 33, C) 91% PMF, 1% PKO, 8% POs IV 33, D) 92% PMF, 1% PKO, 7% POs IV 33, E) 96% PMF, 1% PKO, 3% POs IV 33, F) 91% PMF, 1% PKO, 5% POs IV 33 and 3% POs IV 14 G) 93% PMF, 1% PKO, 3% POs IV 33 and 3% POs IV 14, H) 95% PMF, 1% PKO, 2% POs IV 33 and 2% POs IV 14 of CBA from palm-based fractions (PMF, PKO, POs IV 33 and 14) have been identified through optimised ternary blends. Chocolate bar produced from these blended palm fractions were able to be moulded and demoulded at 45 and 60 min at cooling temperature of 10°C with optimised time of 45 min. The addition of POs IV 33 and IV 14 increased the snappability (rupture tension and breaking force) of all fat blends progressively. Blend of 91% PMF: 1% PKO: 5% POs IV 33: 3% POs IV 14 (Fat F) was observed to have the highest snap value at 10°C. It has comparable oleic acid content, linoleic acid content, monounsaturated fatty acid content, crystal morphology (density) as well as SFC profile at 20°C and crystallisation rate profile at 5°C, 10°C (after 40th min) and 20°C (before 35th min) with CB with dissimilarity in other FAC, TAG, melting behaviour and polymorphism. Therefore, this blend is the most potential CBA to be used as fat for chocolate bar. However, its SFC at 20°C was less than 50% producing a soft chocolate bar. Mixture of 10%-30% of fat blend consisting of 91% PMF: 1% PKO: 5% POs IV 33: 3% POs IV 14 with CB improved the SFC at 20°C and produced SFC of 57%-72% with no eutecticity. Fat blend having 91% PMF: 1% PKO: 5% POs IV

33: 3% POs IV 14 with amount of 30% and CB with amount of 70% was able to be demoulded by 30 min and produce snappability (breaking force) of 5032.71±85.91 g, at 20°C after one day stabilisation period.

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