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Blending of Butter Oil with Refined Palm Oil: Impact on Physicochemical Properties and Oxidative Stability

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ABSTRACT

Blending of edible fat with vegetable oils is a common practice in many countries to improve the physical and nutritional quality. Physicochemical properties and oxidative stability of Butter Oil (BO), Refined Palm Oil (RPO) and their blends were investigated. The liquefied BO and RPO were mixed in proportions of 10:0, 8:2, 6:4, 4:6, 2:8 and 0:10 (w/w) to create 6 treatments. The results showed that total saturated fatty acids decreased, while oleic and linoleic acids as unsaturated fatty acids, β-sitosterols and tocepherols contents increased by increasing the proportion of RPO in BO. Blending of BO with RPO reduced the PLLn triglyceride, while PLP, PLO, OLnL and LOO triglycerides contents increased (P, L, Ln and O represent palmetic, linoleic, linolenic and oleic acids, respectively). Solid Fat Content (SFC) oil blends was lower than both RPO and BO at 0.0 and 10°C, while at 25°C, the SFC of oil blends was close to the pure BO. A major increase in induction period was observed at the proportion of 20 and 40% RPO in BO, after which the increases were not significant. Addition of RPO caused slight increase in both Peroxide Value (PV) and ρ-anisidine value (ρ-AV) of BO compared with pure BO until day 12, after which blending of BO with 20% RPO had the lowest PV and ρ-AV. Therefore, blending of BO with 20% RPO was sufficient to increase the induction period and depress the secondary oxidation compared with BO, PO and other blends.

Key words: Palm oil, butter oil, fatty acids profiles, triglycerides, tocopherols fractions, oxidative stability

INTRODUCTION

Butter oil is one of the highest economic sources of dietary fat. It imparts organoleptic properties such as creamy mouthfeel, buttery aroma, palatability and desirable texture to the food (Augustin and Versteeg, 2006). Nutritionally, butter oil has the highest cholesterol and hypercholesterolemic fatty acids percentage such as myristic and palmetic acids (Rousseau *et al*., 1996). Physically, poor spreadability is a major disadvantage of butter compared to margarines at refrigerator temperature (Wright *et al*., 2000). Lipid oxidation of fat and fat-containing food is a major quality problem during processing and storage because it affects the flavor, color, texture and the nutritive value of foods (Nawar, 1996; Frankel, 1998; Guillen and Cabo, 2002).

Modification of physical, chemical and nutritional properties of milk fat can be achieved by numerous techniques. Blending and chemical interesterification of fats have been used to modify physiochemical and nutritional properties of natural fats (Rodrigues and Gioielli, 2003). Blending butterfat with vegetable oils can lead to spreads that harmonize nutrition and offer desirable

organoleptic attributes as well as lowered overall costs of production (Hui, 1996; Rousseau *et al.*, 1996). Warner and Knowlton (1997) reported that fatty acid profile of milk fat and natural antioxidants can be improved by blending with vegetable oils. The rate at which fatty acids are oxidized increases with the degree of unsaturation and decreases with the presence of lipid-soluble antioxidants. Phytosterols are commonly found in foods such as wheat germ, soybeans, palm oil and corn oil, which cannot be synthesized in the human body (Lopez Ortiz *et al.*, 2006; Rozner and Garti, 2006). Blending milk fat with vegetable oils affected solid fat content. Shen *et al.* (2001) found that the Solid Fat Contents (SFC) of blends (milk fat, hydrogenated coconut and cottonseed oils) were close to the weighted averages of the oil components at temperatures below 15°C. However, from 15 to 25°C, blends of milk fat with hydrogenated coconut oils exhibited SFC lower than those of the weighted averages of the oil components by up to 10% less solid fat. Blends containing milk fat and phytosterol esters give a softer consistency than pure milk fat at low temperatures. Despite the higher SFC, phytosterol esters caused a decrease in the SFC of milk fat (Rodrigues *et al.*, 2007).

Palm oil is a natural product and has been consumed for many decades. It is now used worldwide in manufacturing of wide varieties of food products, because of its numerous advantageous properties, such as its high thermal and oxidative stability and its plasticity at room temperature (Lida *et al.*, 2002; Mamat *et al.*, 2005; Wan Rosnani *et al.*, 2007). Palm oil and its products are ideally suited to be used in many food product formulations including margarine soft cheeses, processed cheese, ice cream and milk powder. The use of PO can be maximized by employing modification processes such as fractionation, blending, interesterification and hydrogenation (Abdul Azis *et al.*, 2011). In addition, palm oil contains a high proportion of palmitic acid (~44%) as well as considerable quantities of oleic (~39%) and linoleic acids (~10%) which give it a higher unsaturated fatty acid content than butter fat. Also, it contains a high amount of the phytosterols, tocotrinols as well as tocopherols. Vitanin E (tocopherols and tocotrienols) acts as a potent antioxidant. The tocopherols constitute 18-22% of the vitamin E content of palm oil while the tocotrienols constitute 78-82% (Edem, 2002; Tan *et al.*, 2009). Therefore, the aim of this study was to investigate the impact of blending butter oil with refined palm oil on some physicochemical properties and oxidative stability of blends.

MATERIALS AND METHODS

Materials

Butter oil (BO): Fresh butter was obtained from processing milk unit, Faculty of Agriculture, Cairo University, Giza, Egypt. Butter oil (Anhydrous milk fat) was prepared by the method of Amer *et al.* (1985). Fresh butter was melted at 60°C, removing the top oil layer, filtering through glass wool and drying the resulting oil over anhydrous sodium sulphate. The oil was then refiltered under vacuum through Whatman 41 filter paper to obtain clear oil (~99.5% milk fat), flushed with nitrogen and stored at -20°C until used.

Palm oil: Refined Palm Oil (RPO) was obtained from the extracted oils and derivatives company (Safola Misr Company), 10th of Ramadan City, Egypt. The stock was flushed with nitrogen and stored at -20°C until used.

Standards: Cholesterol ($C_{27}H_{46}O$), β-sitosterols ($C_{29}H_{50}O$), α-tocopherol ($C_{29}H_{50}O_2$), γ-tocopherol ($C_{28}H_{48}O_2$) and δ-tocopherol ($C_{27}H_{46}O_2$) were obtained from MERCK, Darmstadt, Germany.

Butter oil and palm oil blends: Formulated oil blends were prepared after complete melting of BO and RPO at 70±0.5°C for 10 min. The liquefied BO and RPO were mixed in proportions of 10:0; 8:2, 6:4, 4:6, 2:8 and 0:10 (w/w) to create 6 treatments. Two represent the original oils and four were binary blends. All treatments were stored at -20°C until analyzes.

Methods:

Fatty acids composition: The method recommended by AOAC (2005) was used for preparation of fatty acid methyl esters. The methyl ester of the fatty acids compounds were analyzed with a Perkin Elmer Auto System XL (GC) gas chromatography Equipped with Flame Ionization Detector (FID), Fused silica capillary column ZB-Wax (60 m×0.32 mm i.d). The oven temperature was programmed in two stages as follows: first held at 40°C for 5 min and then from 40 to 220°C at rate 3°C min^{-1}. Detector and injector temperatures were generally 250 and 230°C, respectively. The carrier gas (helium) flow rate was 1 mL min^{-1}.

Triglycerides composition

Sample preparation: Triglycerides were extracted from 0.5 g of each samples by dissolved in acetone (HPLC grade), sonicated, then transferred to measuring flask 10 mL up to the volume with acetone and filtered with 0.45 μ disposal polytetrafluroethylene (PTFE) syringe filter (Wolff *et al*., 1991).

Identification of triglycerides profile: The preparations above for triglycerides were performed on the liquid chromatograph HPLC 9knauer, Germany) equipped with RI detector. The HPLC column used was a Gemini-Nx 5 u, C18, 250×4.6 mm. The temperature kept constant at 40°C, flow rate was 1 mL min^{-1} and mobile phase was a mix HPLC grade acetone and acetonitrile. Triglycerides were identified according to the soy oil standard.

Sterols fractions

Sample preparation: To 1 g of the sample, methanolic solution of 10 mol L^{-1} potassium hydroxide (9:1) was added and refluxed for 30 min. The samples of oil were dissolved in 10 mL of isopropanol prior to the saponification and an aliquot of 1 mL was used. After cooling 5 mL of deionised water and 10 mL of n-hexane were added and intensively shaken for 20 min. The organic layer was separated, washed with deionised water to the neutral reaction and dried with anhydrous sodium sulphate. The hexane solution was evaporated and residue was dissolved in 1 mL of methanol for HPLC analysis (Borkovcova *et al*., 2009).

Identification of sterols fractions: The sterol fraction obtained by above method was dissolved in methanol and 20 μL solution was injected. Analysis was performed on the liquid chromatograph HPLC (Knauer, Germany) with UV detector at 250 nm. Gemini-Nx 5u, C18, 250×4.6 mm column was used under the following conditions: Isocratic elution with mobile phase of methanol and water (95:5) mixture at flow rate 0.7 mL min^{-1}, column temperature was set up at 35°C. For sterols identification standard solutions of cholesterol and β-sitosterol were analyzed under the same conditions to compare retention times. Data were collected and evaluated by software claritychrome (Knauer, Germany) according to cholesterol and β-sitosterol as external standard.

Tocopherols fractions: Tocopherols (α-, γ-and δ-form) of the tested samples were prepared according to the method of Amaral *et al*. (2005). Samples (~300 mg) were accurately weighed in

glass screw cap tubes and homogenized with 2 mL ethanol by Vortex mixing (1 min), 100 μL of butylated hydroxy toluene (10 mg mL^{-1}) was added for protection from oxidation. Subsequently, 4 mL hexane was added and again Vortex mixed for 1 min. Two milliliter saturated NaCl aqueous solution was added, the mixture was homogenized (1 min), centrifuged at 5000 xg for 2 min and the clear upper layer was carefully transferred to another glass screw cap tube. The sample was re-extracted twice with hexane. The combined extracts were dried under a nitrogen stream, at room temperature. The samples were transferred to micro centrifuge tubes with 1.5 mL of hexane and finally, dehydrated with anhydrous sodium sulphate. The extract was centrifuged (10000 g, 20 sec), transferred into a dark injection vial and analyzed by HPLC.

Identification of tocopherols fractions: Analysis was carried out at room temperature on the liquid chromatograph HPLC (Knauer, Germany) equipped with UV detector at 250 nm. Gemini-Nx 5u, C18, 250×4.6 mm column was used. Mobile phase was a mixture of hexane and isopropanol (99:1 v/v) at flow rate 1.5 mL min^{-1}. The concentration of α, δ and γ-tocopherols in the samples were obtained by comparing their peak areas with the peak area of standards in relation to concentration.

Standard preparation: Appropriate volumes of the stock solutions of the tocopherol standards were mixed to obtain a mixed tocopherol standards working solution and dilute of with methanol to give a solution containing 1 μg mL^{-1} of each tocopherol. Then the solution was filtered with 0.45 μ disposal PTFE syringe filter.

Solid fat content (SFC): The Solid Fat Content (SFC) was determined by Nuclear Magnetic Resonance (NMR, Model:MARAN-SFC, Company: Resonance Instruments Ltd.) according to the method described by IUPAC (1987). The tested samples were measured at 0, 10, 20, 25, 30, 35, 40°C. The sample in NMR tube was melted at 70°C for 30 min and than chilled 0°C for 90 min and held at the measuring temperature for 60 min prior to measurement.

Oxidative stability

Induction period: The resistance to oxidation (Induction period) of the tested samples was determined under accelerated conditions (100°C, Oxygen flow at 18 L h^{-1}) using 5 g lipid in Rancimat 679 (Metrohm Ltd B, CH. 9100 Herisau, Switzerland) according to Mendez *et al.* (1996). Oxidative stability was defined as the point of maximum change of the rate of oxidation.

Peroxide and ρ-anisidine values: BO, PO and its blends (150 g) were stored in 250 mL conical flask in an electrical oven at 65°C. Peroxide (PV) and ρ-ansidine values (ρ-PV) were determined at regular intervals for 18 days as described by Egan *et al.* (1981).

RESULTS AND DISCUSSION

Fatty acids composition: Table 1 depict the fatty acid composition of the BO, RPO and their blends. From these data, it could be noticed that, BO was characterized by short-chain (SCFAs) medium-chain (MCFAs) and long-chain (LCFAs) fatty acids. In PO, SCFAs and MCFAs, except myristic fatty acid, were not found, while LCFAs were the most abundant. Moreover, BO contained a higher portion of SFAs (66.51%) and lower portion of USFAs (33.47%) than RPO (52.12 and

Table 1: Fatty acids profile of butter oil, refined palm oil and their blends

	Treatments (%)					
Fatty acids	BO	BO/PO_{20}	BO/PO_{40}	BO/PO_{60}	BO/PO_{80}	RPO
SCFAs						
C_4	1.97	1.68	1.17	0.74	0.49	0.00
C_6	1.25	0.93	0.51	0.41	0.25	0.00
C_8	0.74	0.54	0.44	0.27	-	0.00
Total	3.96	3.15	2.12	1.42	0.74	0.00
MCFAs						
C_{10}	1.54	1.21	0.82	0.64	0.52	0.00
C_{12}	1.90	1.86	1.39	1.05	0.69	0.00
$C_{14:0}$	11.18	9.75	7.64	5.15	3.11	1.06
$C_{14:1}$	0.74	0.51	0.45	0.29	0.12	0.00
Total	15.36	13.33	10.30	7.13	4.44	1.06
LCFAs						
$C_{16:0}$	34.70	37.78	39.64	39.90	40.92	43.13
$C_{16:1}$	1.46	1.36	1.38	1.13	0.72	0.15
$C_{18:0}$	13.23	9.96	8.99	9.33	7.92	7.93
$C_{18:1}$	27.10	29.04	31.02	32.81	36.63	37.56
$C_{18:2}$	1.93	3.27	4.59	6.46	8.03	9.55
$C_{18:3}$	1.71	1.66	1.41	1.32	0.21	0.21
$C_{20:3}$	0.53	0.48	0.51	0.47	0.39	0.38
Total	80.66	83.55	87.54	91.42	94.82	98.91
SFAs	66.51	63.71	60.60	57.49	53.90	52.12
USFAs	33.47	36.32	39.36	42.48	46.10	47.85

BO: Butter oil: RPO: Refined palm oil: BO/PO_{20}: Mixture of butter oil with 20% palm oil, BO/PO_{40}: Mixture of butter oil with 40% palm oil, BO/PO_{60}: Mixture of butter oil with 60% palm oil, BO/PO_{80}: Mixture of butter oil with 80% palm oil, SCFAs: Short chain fatty acids, MCFAs: Medium chain fatty acids, LCFAs: Long chain fatty acids, SFAs: Saturated chain fatty acids, USFAs: Unsaturated chain fatty acids

47.85%, respectively). Palmatic acid as a SFAs and oleic acid as a USFAs were the most abundant in both BO and RPO, although palmatic and oleic acids of RPO were significantly higher as compared to the original BO. Linoleic acid was also significantly lower in BO (1.53%) as compared to RPO (9.55%). Such results have been reported by other researchers as well Grall and Hartel (1992), Lopez *et al.* (2006), Samet-Bali *et al.* (2009) and Abdul Azis *et al.* (2011). Addition of the RPO would therefore, decrease the concentrations of the SCFAs, MCFAs and SFAs, while would increase the concentrations of the LCFAs and USFAs by a ratio related to the concentration of the added RPO.

Triglycerides composition (TGs): There are three types of TGs: (1) the carbon numbers are 26-34; (2) the carbon numbers are 36-46 and (3) the carbon numbers are 48-54 (Jensen and Newburg, 1995). The third type of TGs (C_{50} to C_{54}) of BO, RPO and their blends according to the reference of soybean oil TGs are presented in Table 2. Major differences in long-chain TGs between BO and RPO were observed. Total long-chain TGs of RPO (38.7) was higher than those of BO (23.2). The prominent TGs of RPO were OLnL (8.81), PLP (10.3) and PLO (9.99). RPO also contained appreciable amounts of LSS (2.3) and LOO (4.5) TGs but it contained low amounts of PLL (1.26), LLO (0.67) and PLLn (0.9) TGs (P, L, Ln, S and O represent palmetic, linoleic, linolenic, stearic and oleic acids, respectively). Similar observations were found by Gunstone (2002) and Jeyarani *et al.* (2009). In the same time, BO contained high amount of PLLn (10.3) and low

Table 2: Triglycerides composition of butter oil, palm oil and their blends

	Treatments (%)					
Triglycerides	BO	BO/PO_{20}	BO/PO_{40}	BO/PO_{60}	BO/PO_{80}	RPO
PLP	2.80	4.10	5.10	8.20	9.01	10.30
PLO	3.10	5.20	7.16	7.89	8.81	9.99
LLO	1.90	1.02	0.80	0.81	0.72	0.67
OLnL	2.82	2.96	4.22	5.61	7.06	8.81
PLLn	10.30	7.72	6.50	4.41	1.73	0.90
LOO	0.80	1.20	1.90	2.30	3.10	4.50
LSS	-	0.90	1.10	-	2.00	2.30
PLL	1.50	1.40	1.00	0.80	-	1.26
Total	23.20	24.50	27.80	30.00	32.40	38.70

BO: Butter oil, RPO: Refined palm oil, BO/PO_{20}: Mixture of butter oil with 20% palm oil, BO/PO_{40}: Mixture of butter oil with 40% palm oil, BO/PO_{60}: Mixture of butter oil with 60% palm oil, BO/PO_{80}: Mixture of butter oil with 80% palm oil, O: Oleic acid, P: Palmetic acid, L: Lenolenic acid, S: Stearic acid, Ln: Lenolenic acid

Table 3: Fractions of sterols and tocopherols contents of butter oil, refined palm oil and their blends

	Treatments					
Unsaponificable matters	BO	BO/PO_{20}	BO/PO_{40}	BO/PO_{60}	BO/PO_{80}	RPO
Sterols (mg/100 g oil)						
Cholesterol	231.0	188.40	142.4	95.10	51.60	8.50
β-sitosterol	0.15	5.97	12.5	19.01	25.73	31.50
Tocopherols (mg/100 g oil)						
α-Tocopherol	2.31	5.90	12.1	16.70	25.20	31.50
γ-Tocopherol	0.60	5.50	11.3	15.40	21.80	27.60
δ-Tocopherol	0.10	1.10	2.9	3.20	4.90	6.10
Total	2.31	5.90	12.1	16.70	25.20	31.50

BO: Butter oil, RPO: Refined palm oil, BO/PO_{20}: Mixture of butter oil with 20% palm oil, BO/PO_{40}: Mixture of butter oil with 40% palm oil, BO/PO_{60}: Mixture of butter oil with 60% palm oil, BO/PO_{80}: Mixture of butter oil with 80% palm oil

amounts of PLP, PLO, OLnL and LOO compared with RPO. The addition of RPO to BO, therefore, reduced the TGs composition, namely PLLn and total long-chain TGs, while PLP, PLO, OLnL and LOO increased. These results are coincident with those reported by Mamat *et al.* (2005).

Sterols fractions: As shown in Table 3, the total contents of cholesterol in BO and RPO were 231.0 and 8.5 mg/100 g, respectively. These values are in agreement with Alonso *et al.* (1997), Contarini *et al.* (2002) and Sundram *et al.* (2003). Molkentin (2006) reported that the average cholesterol content in bovine milk fat obtained from butter was 265.5 mg/100 g with a variation of 204.3-382.4 mg/100 g. However, **β**-sitosterol as the major phytosterols in palm oil (Alonso *et al.*, 1997; Sambanthamurthi *et al.*, 2000; Sundram *et al.*, 2003), was 31.5 mg/100 g for PO, while was 0.15 mg/100 g for BO. These results mean that, BO had highest cholesterol content and lowest **β**-sitosterol content, while RPO had highest **β**-sitosterol content and lowest cholesterol content. Therefore, the addition of PO to BO caused a gradual increase in **β**-sitosterol and gradual decrease in cholesterol compared with pure BO; the changes were proportional to the addition level. With the addition of 20.0, 40.0, 60.0 and 80.0% RPO, the **β**-sitosterol elevated by 12.93, 27.03, 41.12 and 55.64%, respectively compared with pure BO (0.33%).

Tocopherols fractions: Tocopherols content of BO, RPO and their blends which determined by HPLC as another component of the unsaponifiable fraction are presented in Table 3. It is evident from the results that, BO had the lowest tocopherols fractions as compared to RPO. In particular, α-, γ- and δ-tocopherols of BO were 2.31, 0.6 and 0.1 while these of PO were 25.2, 27.6 and 4.9 mg/100 g oil, respectively. Also, it could be seen that the total tocopherols in BO was accounted 3.01 mg/100 g fat, while the total tocopherols in RPO was 65.2 mg/100 g. A similar observation was found by Lusas and Riaz (1996) and Alonso *et al.* (1997). In most cases, therefore, the addition of RPO to BO will result in an increase in the tocopherol content of the BO. These results are coincident with those reported by Dolde *et al.* (1999), Maduko *et al.* (2008) and Schwartz *et al.* (2008). Our observations show that with the addition of 20, 40, 60 and 80% RPO, total tocopherols content of the resultant blends were elevated by 5.9, 12.1, 16.7 and 25.2 mg/100 g oil, respectively.

Solid fat content: Solid Fat Content (SFC) of Butter Oil (BO), Refined Palm Oil (RPO) and their blends at 0.0 to 40°C are presented in Table 4. The results show that, SFC of pure BO was lower than those of PO at 0.0 to 40°C. In particular, SFC of BO was 58.2, 46.5, 20.2, 12.7, 6.7, 1.6 and 0.0 while that was 69.4, 56.5, 28.6, 14.8, 9.7, 6.2 and 3.4% for RPO at 0.0, 10, 20, 25, 30, 35 and 40°C, respectively. Similar observations were found by Berger (2003), Rodrigues *et al.* (2007) and Ozvural and Vural (2008). At 0.0 and 10°C, however, blends of BO with RPO were exhibited SFC lower than both PO and BO. Timms (1984) reported when two fats are mixed, the SFC of the blends may be lower than the weighted average of the component fats. Sometimes, the SFC can be lower than that of either fat. Inversely, at 20, 30 and 35°C, the addition of RPO to BO caused slight increase in SFC compared with pure BO; the increasing was proportional to the addition level. Rousseau and Marangoni (1999) have also observed an increased for chemically interesterified butter fat with canola oil above 10°C. However, at 25°C, the SFC of all oil blends was close to that pure BO. At 40°C, SFC was not observed in BO and blends of BO with 20.0, 40.0 and 60.0% RPO. Shen *et al.* (2001) have a similar observation in blends of milk fat with hydrogenated coconut and cottonseed oils. Also, they reported the absence of strong eutectic effects in blends of milk fat with hydrogenated coconut and cottonseed oils suggested that these blends had compatible polymorphs in their solid phases.

Oxidative stability: The time before a dramatic increase in the rate of lipid oxidation is a measure of oxidative stability and is referred to as the induction time (Coppin and Pike, 2001). In the

Table 4: Solid fat content of butter oil, refined palm oil and their blends at 0.0 to 40°C

	Solid fat content (%)					
Temperature (°C)	BO	BO/PO_{20}	BO/PO_{40}	BO/PO_{60}	BO/PO_{80}	RPO
0.0	58.2	53.7	48.6	44.8	46.5	69.4
10	46.5	43.8	38.9	37.8	42.8	56.5
20	20.2	21.1	22.1	22.9	24.0	28.6
25	12.7	12.4	12.1	12.5	12.7	14.8
30	6.7	6.8	7.3	7.9	8.4	9.7
35	1.6	2.2	2.6	3.3	4.1	6.2
40	0.0	0.0	0.0	0.0	0.7	3.4

BO: Butter oil, RPO: Refined palm oil, BO/PO_{20}: Mixture of butter oil with 20% palm oil, BO/PO_{40}: Mixture of butter oil with 40% palm oil, BO/PO_{60}: Mixture of butter oil with 60% palm oil, BO/PO_{80}: Mixture of butter oil with 80% palm oil

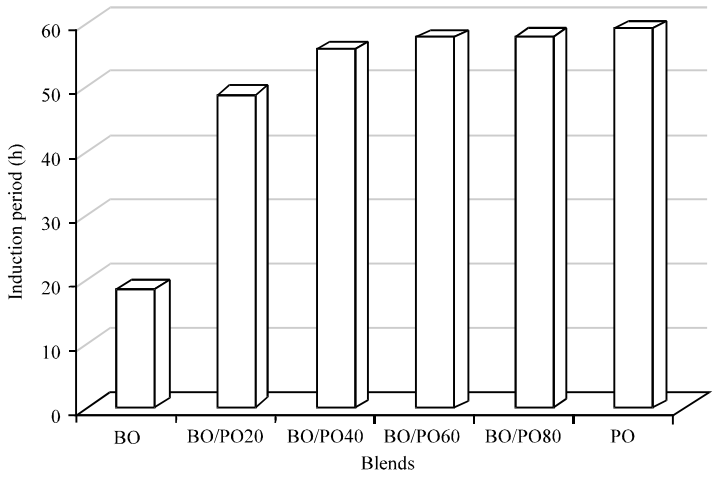

Fig. 1: Induction period of butter oil: refined palm oil and its blends by Rancimat method at 100°C, BO: Butter oil, RPO: Refined palm oil, BO/PO_{20}: Mixture of butter oil with 20% palm oil, BO/PO_{40}: Mixture of butter oil with 40% palm oil, BO/PO_{60}: Mixture of butter oil with 60% palm oil, BO/PO_{80}: Mixture of butter oil with 80% palm oil

Rancimat method, the changes in Induction Periods (IP) of BO as affected by addition of RPO are presented in Fig. 1. In general, the IP of BO was lower than those of RPO. Addition of the RPO caused a significant increase in the IP; the increasing rate, however, was more pronounced in BO contains 20 and 40% RPO. Blends contain 20 and 40% RPO increased the IP of BO 1.62 and 2.0 time, respectively. Based on the fact that fat with high degree of USFAs, it has a greater potential form oxidation (Scott *et al*., 2003), these results were not expected. However, the higher stability of RPO than BO could be attributed to the RPO has high natural antioxidant, i.e, **α-**, **β-**, **γ-** and **δ-**tocopherols (Table 3) carotenoids, phenolic compounds and sterols, which increased in BO with the addition of RPO. Chaiyasit *et al*. (2007) have reported that edible oils contain polar lipids, such as mono-and diacylglycerols, phospholipids, sterols, tocopherols and free fatty acids that are not completely removed by the refining process. Palm oil, like other vegetable oils, contains tocols that can act as natural biological antioxidants. However, it is distinct in that it contains a high level of tocotrinols as well as tocopherols can act as natural biological antioxidants (Ab Gapor *et al*., 1989; Edem, 2002).

Peroxide Value (PV) and **ρ-**ansidine value (**ρ-**AV) of BO, RPO and their blends as other indicators of oxidation rate during oxidation process at 65°C are presented in Fig. 2 and 3. At day 1, PV and **ρ-**AV of BO were lower than these of RPO. During oxidation process, the oxidation rate of BO (PV and **ρ-**AV) was lower than those of PO until day 14, while that of BO was higher than those of RPO from day 16th onwards. The higher oxidation rate of BO in secondary oxidation than RPO may be able to be attributed to BO has high content from free fatty acids (0.3) than PO (0.1). Free fatty acids act as prooxidants in edible oil (Miyashita and Takagi, 1986; Mistry and Min, 1987). They have hydrophilic and hydrophobic groups in the same molecule and prefer to be concentrated on the surface of edible oils. Mistry and Min (1987) have reported that free fatty acids decrease the surface tension of edible oil and increase the diffusion rate of oxygen from the headspace into the oil to accelerate oil oxidation. Also, BO may has higher content from mono-and diacylglycerol than RPO, which have hydrophilic hydroxyl groups and hydrophobic hydrocarbons, decrease the surface tension of edible oil and increase oxidation rate (Mistry and Min, 1988).

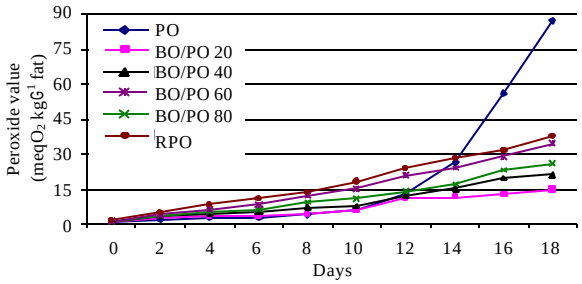

Fig. 2: Peroxide value of butter oil, refined palm oil and their blends during oxidation process at 65°C, BO: Butter oil, RPO: Refined palm oil, BO/PO_{20}: Mixture of butter oil with 20% palm oil, BO/PO_{40}: Mixture of butter oil with 40% palm oil, BO/PO_{60}: Mixture of butter oil with 60% palm oil, BO/PO_{80}: Mixture of butter oil with 80% palm oil

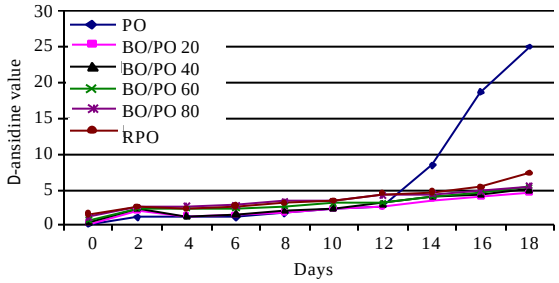

Fig. 3: Para-ansidine value of butter oil: refined palm oil and their blends during oxidation process at 65°C, BO: Butter oil, RPO: Refined palm oil, BO/PO_{20}: Mixture of butter oil with 20% palm oil, BO/PO_{40}: Mixture of butter oil with 40% palm oil, BO/PO_{60}: Mixture of butter oil with 60% palm oil, BO/PO_{80}: Mixture of butter oil with 80% palm oil

On the other hand, the oxidation rate of BO was influenced by addition of RPO. In particular, addition of RPO caused significant increase in both PV and ρ-AV of BO compared with pure BO; the increase was proportional to the addition level of RPO until day 10th for BO contains 20 and 40% RPO and day 12th for that contains 60 and 80% PO. On day 14th onwards, positive correlation was found between PV and ρ-AV of BO and addition level of RPO. This means that, BO with 20% RPO was the most stable to the secondary oxidation followed by that contain 40, 60 and 80% PO during oxidation process at 65°C. This results may be attributed to addition of high level of RPO may induce an imbalance between the presence of reactive oxidants and the antioxidant defense mechanism. For example, soybean oil oxidation decreased with addition of 5 to 10 ppm phospholipids and higher amount of phospholipids acted as prooxidants (Choe and Min, 2006). Yoon *et al.* (1987) reported that phospholipids acted as antioxidants only in the presence of Fe^{2+} by chelating iron. In purified soybean oil, which did not contain any metals, phospholipids worked as prooxidants. From these results could be concluded that BO was more stable in primary oxidation than RPO, while RPO was more stable in secondary oxidation than BO. Also, BO contains 20% RPO was the most stable blend.

CONCLUSION

Blending of BO with RPO can increase the levels of unsaturated fatty acids, bioactive lipids and natural antioxidants, which improve the nutritional quality and oxidative stability of the blend. However, the addition of 20.0% RPO to BO was sufficient to depress the lipid peroxidation to the lower limit. At low temperature (≤10°C), blending of BO with RPO reduce the SFC in the blend, which give a softer consistency than both BO and RPO.

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