

# Effects of Enzymatic Interesterification on the Physicochemical, Polymorphism and Textural Properties of Palm Stearin, Palm Kernel Oil and Soybean Oil Blends

Siti Hazirah Mohamad Fauzi, Norizzah Abd Rashid, and Zaliha Omar

**Abstract**—This study evaluated the effects of enzymatic interesterification (EIE) using Lipozyme TL IM on palm stearin (PS), palm kernel oil (PKO), soybean oil (SBO) and their constituent blends formulated according to a Design Expert 8.0.4 (2010). The effects of EIE on slip melting point (SMP), solid fat content (SFC), triacylglycerol composition (TAG), polymorphism and textural properties were investigated. Palm-based *trans*-free table margarine containing ternary mixture of PS/PKO/SBO [50/22/28 (w/w)], was optimally formulated through analysis of multiple isosolid diagrams and was found to have quite similar SMP and SFC profile as compared to the commercial table margarine.

**Index Terms**—Design expert, enzymatic interesterification, lipozyme TL IM, *trans*-free table margarine formulation.

## I. INTRODUCTION

Palm stearin (*Elaeis guineensis jacquin*) is a natural and cheaper high-melting fraction obtained from palm oil and does not require any hydrogenation process. However, because of its high melting profile ranging from 44 to 56 °C, PS cannot be used directly for production of margarine as it causes low plasticity to the products and does not completely melt at body temperature [1]. Palm kernel oil contains short and medium chain length TAG similar to those present in butterfat, and therefore helps to give the right melting characteristics and ensures the stable beta prime ( $\beta'$ ) crystalline form necessary for production of margarine [2]. To enhance the PS compatibility, its need to be blended and/or interesterified with polyunsaturated soft oil such as soybean oil and sunflower oil in order to impart plasticity to the final IE product [3]–[6].

Intesterification (IE) has received much interest in the edible oil industry as an alternative method to improve the physical properties of fats and oils. It modifies the physical properties of oils by rearranging the distribution of fatty acids (FAs) on the glycerol backbone without changing their chemical composition. Interesterification reaction has been widely used to formulate margarine and shortening fats with zero *trans* fat and improved spreadability and nutritional

properties [4]. Currently, EIE has gained preference over chemical interesterification (CIE) [7]–[11], EIE can provide several benefits such as milder conditions, low side-products since these reactions mimic natural pathways and regioselectivity [12], [13]. Lipozyme TL IM from *Thermomyces lanuginosa* is an immobilised 1, 3-specific lipase and was used in this present study respectively.

In this study, blends of PS, PKO and SBO were modified by EIE. The enzymatic modified blends prepared will be formulated to achieve desirable properties of *trans*-free table margarine fats. Hence, the objectives of this framework were: (i) to modify the physical and chemical properties of PS, PKO and SBO by blending at certain ratios according to the Design Expert version 8.0.4 (2010), (ii) to compare the physicochemical, polymorphism and textural properties of the ternary blends before and after EIE, and (iii) to determine the most suitable interesterified ternary blends for formulation *trans*-free table margarine fats.

## II. MATERIALS AND METHODS

### A. Materials

Refined, bleached, and deodorised (RBD) PS and PKO were obtained from Golden Jomalina Sdn Bhd, Klang, Malaysia, while RBD SBO was purchased from local hypermarket at Shah Alam, Selangor, Malaysia. All the chemicals used were either analytical or high-performance liquid chromatography grade.

### B. Blend Preparation

The PS, PKO and SBO were melted at 60 °C in an oven prior to use. Based on the Simplex-Lattice Mixture Design (Design Expert 8.0.4, 2010), blends were prepared in the following mass (w/w) ratios: NIE were indicated by A (1/0/0), B (0/1/0), C (0/0/1), D (1/1/0), E (1/0/1), F (0/1/1), G (4/1/1), H (1/4/1), I (1/1/4) and J (1/1/1) and EIE were indicated by a (1/0/0), b (0/1/0), c (0/0/1), d (1/1/0), e (1/0/1), f (0/1/1), g (4/1/1), h (1/4/1), i (1/1/4) and j (1/1/1).

### C. Enzymatic Interesterification (EIE)

The EIE was conducted according reported in [14]. The non-interesterified oil is abbreviated as NIE and enzymatically interesterified oil as EIE.

### D. Slip Melting Point (SMP)

The SMP of the NIE and EIE blends was determined according in [14]. Three replicates of this analysis were performed.

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### E. Solid Fat Content (SFC)

The SFC of the blends was measured using a Bruker Minispec pulsed Nuclear Magnetic Resonance (pNMR) spectrometer (Karlsruhe, Germany). Measurements were carried out at 5–50 °C with 5 °C interval [14].

### F. Triacylglycerol Composition (TAG)

The TAG composition of the blends were analysed in a reversed phase high-performance liquid chromatograph (Gilson, Villiersel-Bel, France). A Lichrosphere RP-18 column (250 mm × 4 mm) of 5-µm particle size (Merck, Darmstadt, Germany) with acetone/acetonitrile (75:25% v/v) as the eluent at a flow rate of 1.0 ml/min and a refractive index detector was used.

### G. Polymorphism

The polymorphic forms of fat crystals in the blends were determined by X-ray diffraction, using an Enraf Nonius Model FR592 (Delft, The Netherlands). The short spacings of the β' form are at 4.2 and 3.8 Å and that of the β form is at 4.6 Å [15].

### H. Texture Measurement

A constant speed compression test was determined by Texture Analyser TA-XT2 (Stable Micro Systems, Surrey, England) controlled by a microcomputer. A 25 kg load cell an ebonite penetration cone with an angle of 10 ° was applied to a penetration depth of 10.0 mm at a rate of 1.0 mm/s. Samples were then tempered at 10 °C for 24 h prior to analysis.

### I. Experimental Design

An optimal mixture design consisting three variables was used in this study. The three mixture components are PS represented by  $X_1$ , PKO by  $X_2$  and SBO by  $X_3$ , with the sum of  $X_1 + X_2 + X_3 = 1$ . The SFC at 5–50 °C and SMP were used as responses. These responses were chosen due to their crucial role attribute in indicating acceptability of formulated margarine. Total number of 10 design blends was suggested by the software and ternary diagrams at each temperature were built by mixture design. Optimisation of ternary mixture components for production of *trans*-free table margarine formulation was conducted through graphical multiple responses optimisation tool in Design-Expert 8.0.4 (2010) software.

### J. Statistical Analysis

The linear, quadratic and special cubic models as in (1)-(3) were used to represent the fitted response values. The statistical significance of each equation was determined by variance analysis (ANOVA) at 5%.

$$Y = b_1X_1 + b_2X_2 + b_3X_3 \quad (1)$$

$$Y = b_1X_1 + b_2X_2 + b_3X_3 + b_1b_2X_1X_2 + b_1b_3X_1X_3 + b_2b_3X_2X_3 \quad (2)$$

$$Y = b_1X_1 + b_2X_2 + b_3X_3 + b_1b_2X_1X_2 + b_1b_3X_1X_3 + b_2b_3X_2X_3 + b_1b_2b_3X_1X_2X_3 \quad (3)$$

where  $Y$  is the predictive dependent variable (SFC at 5–50 °C and SMP),  $b$  is the equation coefficients as in [16] and  $X$  is the proportions of pseudo-components.

## III. RESULTS AND DISCUSSIONS

### A. Slip Melting Point (SMP)

As observed in Fig. 1 a) PS had the highest SMP (52 °C), followed by PKO (28 °C) and SBO (7 °C). The highest melting point of PS was due to the presence of long-chain saturated FA. The PKO contains short- and medium-chain saturated FAs, while SBO had the lowest melting point as it contains high amount mono- and polyunsaturated FA. The SMP of both NIE and EIE of the original oils did not much change as much as in binary and ternary blends as shown in Fig. 1 a-c). This could be due to intraesterification which involves reshuffling of FA moieties within the TAGs molecule, and hence the SMP was hardly altered.

The SMP of all EIE binary blends decreased with an increase in the proportion of SBO in PS/SBO and PKO/SBO blends. This could be due to the decrease of highly saturated TAGs presence in PS and PKO, diluted by mono- and polyunsaturated SBO. The effects of diluting factor of SBO before and after EIE can be observed in Fig. 1b. As expected, binary blend of EIE PS/PKO, had lower SMP (37 °C) than NIE (45 °C). This finding has demonstrated that EIE could provide a better alternative for producing new tailored-fats with desired melting profile that meets consumer preferences.

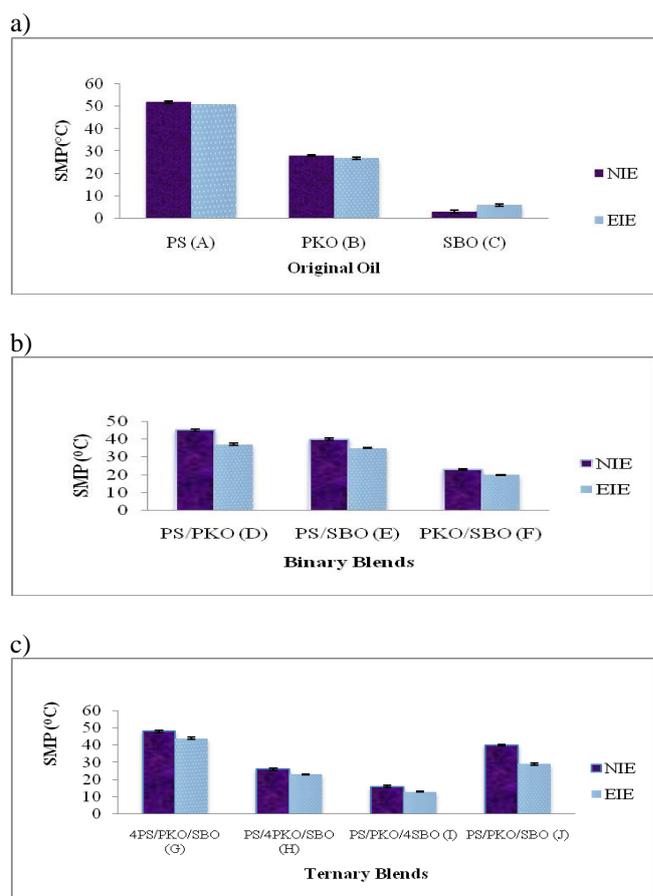


Fig. 1 a)-c). The SMP of non-interesterified (NIE) and enzymatic interesterified (EIE) of original, binary and ternary PS, PKO and SBO blends.

### B. Solid Fat Content (SFC)

The SFC was dependent on both temperature and the proportion of each fat in the blend. As shown in Fig. 2 a), the

EIE lowered the SFC at all analysed temperatures; however the SFC of original oil PS, PKO and SBO before and after EIE did not change as much as binary and ternary blends. This is relatively due to intraesterification which involves reshuffling of FA moieties within the TAG molecule in the same chemical composition of blend, and hence the SFC was hardly altered. This finding agreed with the SMP results.

The binary blends of NIE PS/PKO (Fig. 2 b) ) had the highest SFC (74.44%) at the starting temperature and followed by EIE PS/PKO 72.46% as compared to PS/SBO and PKO/SBO blends. This could be due to chemical composition of PS and PKO that have almost equal proportions of the high-melting of trisaturated ( $S_3$ ) and disaturated-monounsaturated ( $S_2U$ ) TAG in the blends (Table I). The EIE PS/PKO blends tended to have lower SFC curves than NIE PS/PKO relatively due to randomisation and redistribution of high-melting TAG of PS with high and medium-melting TAG of PKO.

The SFC of ternary PS/PKO/SBO blends differed (Fig. 2 c) ), depending on the amount of PS, PKO and SBO in the blends. As expected, blends with high proportions of PS tended to have higher SFC compared to other ternary blends. Overall, the EIE relatively lowered SFC at all analysed temperatures. According to the literatures, this may be due to a decrease of  $S_3$  TAGs by IE reaction [17]-[19]. Principally all the IE blends completely melted (SFC = 0%) at lower temperatures than their NIE blends and this is correspondingly in line with the SMP observation.

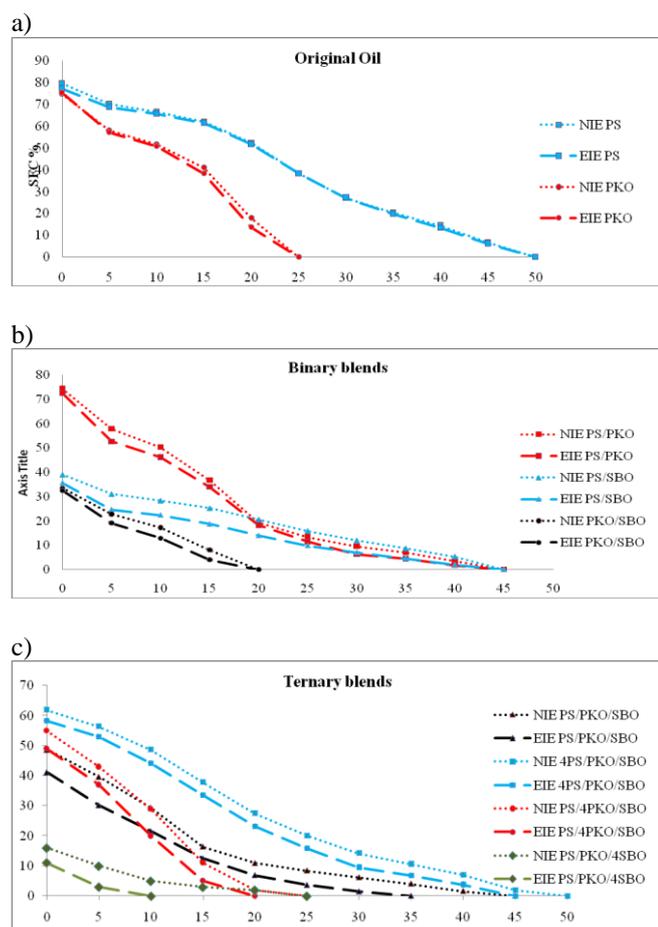


Fig. 2 a)-c). The SFC of non-interesterified (NIE) and enzymatic interesterified (EIE) of original, binary and ternary PS, PKO and SBO blends.

### C. Triacylglycerol Composition (TAG)

TABLE I: TRIACYLGLYCEROL COMPOSITION OF PS, PKO, SBO AND BLENDS BEFORE AND AFTER EIE

TAG	PS		PKO		SBO	
	NIE	EIE	NIE	EIE	NIE	EIE
CLaLa	-	-	9.0	5.2	-	-
CaLaLa	-	-	12.3	2.0	-	-
LaLaLa	-	-	25.5	22.1	-	-
LaLaM	-	-	14.0	14.6	-	-
LaLaO	-	-	5.5	14.5	-	-
LaLaP	-	-	7.5	12.0	-	-
LLL	-	-	-	-	32.0	25.5
LaOM	-	-	4.0	9.5	-	-
LaPM	-	-	3.8	4.0	-	-
OLL	-	-	-	-	30.5	28.0
PLL	0.8	-	-	-	8.0	8.0
LaOO	-	-	2.8	3.0	-	-
LaOP	-	-	5.0	3.0	-	-
LaPP	-	-	0.8	3.0	-	-
OLO	1.0	1.0	-	-	9.8	10.5
PLO	5.0	2.5	-	-	9.5	10.0
MOO	-	-	1.0	1.0	-	-
PLP	7.5	6.0	-	-	1.0	2.5
MOP	-	-	2.3	2.0	-	-
OOO	2.0	1.0	1.2	1.0	3.0	3.0
POO	17.5	17.7	1.0	1.0	3.8	3.0
POP	33.5	35.0	0.5	1.0	0.5	2.0
PPP	18.7	25.0	0.1	1.0	1.0	2.0
SOO	1.0	2.0	-	-	0.5	1.0
POS	5.5	3.0	-	-	0.2	1.0
PPS	3.6	3.5	-	-	0.2	1.0
Others	3.9	3.3	3.7	0.1	0.0	2.5
Total	96.1	96.7	96.3	99.9	100.0	97.5

TAG	PS/PKO		PS/SBO		PKO/SBO	
	NIE	EIE	NIE	EIE	NIE	EIE
CLaLa	5.0	2.0	-	-	4.5	2.0
CaLaLa	6.0	4.8	-	-	5.8	5.0
LaLaLa	8.0	4.8	-	-	11.4	11.4
LaLaM	6.5	3.0	-	-	7.8	7.0
LaLaO	3.0	3.0	-	-	1.9	2.0
LaLaP	3.7	3.0	-	-	5.0	3.0
LLL	-	-	15.0	6.0	10.0	11.8
LaOM	2.0	1.0	-	-	3.0	3.8
LaPM	1.5	1.0	-	-	-	-
OLL	-	-	18.0	11.5	15.8	5.0
PLL	0.5	2.0	5.0	11.0	5.0	5.0
LaOO	1.5	3.0	-	-	2.1	1.0
LaOP	2.0	13.0	-	-	2.5	1.0
LaPP	-	12.1	-	6.0	-	-
OLO	1.0	1.0	5.0	18.7	6.8	16.8
PLO	2.5	1.0	7.3	2.0	6.2	15.0
MOO	3.0	1.0	-	-	-	-
PLP	5.0	4.3	4.5	10.5	-	-
MOP	3.0	2.0	-	-	-	-
OOO	1.5	2.0	2.5	-	2.0	3.0
POO	8.5	6.3	8.0	10.0	2.3	1.0
POP	20.5	8.0	18.5	12.5	0.5	1.0
PPP	10.0	6.1	8.0	5.8	0.5	0.1
SOO	0.8	2.0	1.0	2.0	0.2	0.1
POS	4.0	4.0	4.0	2.0	0.1	0.1
PPS	0.5	3.0	2.5	2.0	0.1	0.1
Others	0.0	6.6	0.7	0.0	6.5	4.8
Total	100.0	93.4	99.3	100.0	93.5	95.2

TAG	4PS/PKO/ SBO		PS/4PKO/ SBO		PS/PKO/ 4SBO		PS/PKO/ SBO	
	NIE	EIE	NIE	EIE	NIE	EIE	NIE	EIE
CLaLa	3.0	2.0	5.3	4.0	3.0	2.0	3.0	2.0
CaLaLa	3.5	4.2	9.0	9.0	3.0	5.0	4.0	4.6
LaLaLa	2.8	4.0	10.1	7.5	4.2	5.0	7.5	4.5
LaLaM	2.0	2.0	8.5	7.5	4.0	5.0	4.5	4.5
LaLaO	3.5	2.0	5.5	7.0	3.5	2.0	2.3	5.5
LaLaP	-	-	5.0	7.0	1.0	4.8	-	5.5
LLL	5.0	5.5	3.0	8.0	15.3	10.5	10.6	2.8
LaOM	2.3	1.0	3.5	1.0	-	1.0	1.0	3.0

LaPM	-	-	3.0	1.0	-	1.0	-	-
OLL	5.0	5.0	4.8	5.0	7.5	8.0	11.5	3.9
PLL	4.0	5.0	3.1	5.0	8.0	8.0	3.0	3.0
LaOO	1.0	1.0	-	-	1.0	-	1.1	4.0
LaOP	1.5	1.0	2.0	1.0	-	-	1.7	5.5
LaPP	-	-	1.9	2.0	-	-	-	-
OLO	3.6	3.0	2.0	3.0	5.5	9.5	5.2	5.0
PLO	4.5	3.0	1.7	3.0	3.0	2.8	6.0	5.0
MOO	-	-	-	-	-	-	-	-
PLP	3.0	5.0	1.3	2.0	4.0	5.0	3.3	7.0
MOP	-	-	-	-	-	-	-	-
OOO	4.0	-	4.0	2.0	4.2	2.5	1.5	5.0
POO	4.8	9.0	4.3	4.0	4.0	3.5	6.0	3.0
POP	10.0	9.5	3.7	3.0	5.8	5.8	15.0	9.0
PPP	11.3	10.	4.1	3.0	5.0	5.5	5.0	3.8
SOO	1.7	2.0	2.0	2.0	2.0	-	1.0	1.5
POS	10.8	8.7	4.2	3.5	4.0	4.0	2.5	2.5
PPS	12.0	8.0	3.5	3.0	4.0	4.0	1.2	1.0
Other	0.7	9.0	4.5	6.5	8.0	5.1	3.1	8.4
Total	99.3	91.0	95.5	93.5	92.0	94.9	96.9	91.6

Notes: C, capric acid; La, lauric acid; Ca, caprylic acid; M, myristic acid; O, oleic acid; P, palmitic acid; L, linoleic acid; S, stearic acid.

Table I shows the TAG composition of PS, PKO and SBO as well as their blends before and after EIE. The main TAG of PS were PPP, POP, PLP and POO where P is palmitic acid, L is linoleic acid and O is oleic acid. Palm kernel oil contained a wide range of medium- chain TAG species such as LaLaLa, LaLaM, CaLaLa, CLaLa, LaLaP, and LaLaO, where La is lauric acid, M is myristic acid, Ca is caprylic acid, C is capric acid and O is oleic acid. While SBO consists mainly of U<sub>3</sub> and SU<sub>2</sub> classes such as LLL, OLL, OLO, PLO and PLL, where P is palmitic acid, L is linoleic and O is oleic acid. The original PS, PKO and SBO oil generated only few changes in TAG after EIE as compared to the NIE. This apparently because of intraesterification did not result in much change in the TAG composition.

The changes of TAG proportions due to IE reaction were more pronounced in binary and ternary blends due to the presence of varied TAG species in the mixtures. Randomisation led to the formation of a new TAG that could not be identified due to the complex interaction of the mixture blends. However, the TAG profile of the IE blends showed an even TAG distribution than the starting blends, as the relative concentration of several TAG increased; while others decreased as shown in Table I. This result is consistent with findings in [20].

#### D. Polymorphism

The polymorphism of original oil, before and after EIE, attained at 20 °C, was shown in Table II. The patterns before and after EIE showed a strong short spacing at ~4.6 Å, indicating dominant tendency of β polymorph for PS together with a very weak short spacing at 4.2 Å and 3.8 Å, indicating presence of small amount of β' polymorph. These results are in agreement with the data previously reported in [21] for binary blends of PS with palm kernel olein (PKOo) and as in [22] for soft PS. The presence of β polymorph in PS blend is related to the higher amount of S<sub>3</sub> TAGs while the β' polymorph was associated with the abundance of asymmetrical TAGs [15], [23].

For PKO, the initial and after EIE of PKO were exclusively stable in β' polymorphism. The presence of β' polymorph in PKO is related to FA of different chain length and TAG composition (Table I). The melting point of

original oil PKO was lower as compared to the melting point of original oil PS and this behavior can be explained according to the dominancy of polymorphism in blend, respectively. The β' crystal needs little energy to melt the fats as compared to the stable ordered-packing of β crystal, as confirmed by the SMP results given in Fig. 1a. However, the polymorphic form of SBO was not able to determine as it was liquid at measured temperature.

In NIE PS/PKO blends containing both mixtures of β and β' crystals in the same proportions. This can be explained due to β-tending of PS and β'-tending of PKO in their nature polymorphism behavior. However, after EIE of PS/PKO blends, the β' polymorph tended to dominate the blends with only small amount of β crystals were observed in EIE blends. While for PS/SBO, before EIE, containing high proportions of β' polymorph compared with only small amount of β polymorph. This could be due to the dilution effect by SBO, as observed by [24] where the β' polymorphic form tends to dominate the more the fat is diluted with a liquid oil, or because the amount of β polymorphic form is too low in concentration to exert any influence.

Polymorphism of ternary blends PS/PKO/SBO showed the dominancy of β' with aggregation of small amount of β-tendency in initial blends and also after the IE process. By EIE, it promotes of stabilising the β' polymorph, since it can promote, according to the compositional characteristics of raw materials, the formation of TAGs with greater variation in chain length corresponding to the mixtures of FAs of PS, PKO and SBO.

Crystals of β' are small and present morphology that is suitable for the plasticity characteristics which are desirable in products such as margarines, shortenings and pastry. Conversely, polymorphic form β tends to produce wide granular crystals, generating sandy products with low potential for aeration, which may compromise the macroscopic properties of some kinds of food [25].

TABLE II: POLYMORPHIC FORMS OF THE NON-INTERESTERIFIED (NIE) AND ENZYMATIC INTERESTERIFIED (EIE) BLENDS

	Polymorphic form(s)	
	NIE	EIE
<b>Original oil</b>		
PS	β >> β'	β >>> β'
PKO	Liquid	β'
SBO	Liquid	Liquid
<b>Binary blends</b>		
PS/PKO	β' + β	β' >> β
PS/SBO	β' >> β	β'
PKO/SBO	Liquid	Liquid
<b>Ternary blends</b>		
4PS/PKO/SBO	β' >> β	β' >>> β
PS/4PKO/SBO	β' + β	β'
PS/PKO/4SBO	Liquid	Liquid
PS/PKO/SBO	β' >> β	β' >>> β

#### E. Textural Properties

Texture plays critical factor in determining the functionality and consumer acceptance of table spreads [26], [27]. The values of compression force equal to zero (Table III) indicated that properties of blends could not be detected,

since the blends were fluid and appear in liquid state at the 10 °C. Original oil of SBO (coded C), binary blends of PKO/SBO (coded F) and ternary blends of PS/PKO/4SBO (coded I) showed values of 0 N/cm<sup>2</sup>. This observation can be explained due to addition of polyunsaturated SBO into the blends thus resulted blends were in fluid.

TABLE III: COMPRESSION FORCE (N/CM<sup>2</sup>) OF PS, PKO AND SBO BLENDS BEFORE AND AFTER EIE AT 10 °C

PS/PKO/SBO	Compression Force (N/cm <sup>2</sup> )		
	Ratios	NIE	EIE
A	(1/0/0)	1250.50±2.10 <sup>a</sup>	872.15±3.01 <sup>b</sup>
B	(0/1/0)	300.25±5.07 <sup>a</sup>	203.74±5.12 <sup>b</sup>
C	(0/0/1)	0±0.00 <sup>a</sup>	0±0.00 <sup>a</sup>
D	(1/1/0)	1034.53±7.19 <sup>a</sup>	635.12±1.82 <sup>b</sup>
E	(1/0/1)	1094.11±2.50 <sup>a</sup>	700.56±4.22 <sup>b</sup>
F	(0/1/1)	0±0.00 <sup>a</sup>	0±0.00 <sup>a</sup>
G	(4/1/1)	1180.41±1.10 <sup>a</sup>	780.93±3.25 <sup>b</sup>
H	(1/4/1)	452.28±2.10 <sup>a</sup>	371.10±1.74 <sup>b</sup>
I	(1/1/4)	0±0.00 <sup>a</sup>	0±0.00 <sup>a</sup>
J	(1/1/1)	711.59±4.56 <sup>a</sup>	482.55±4.81 <sup>b</sup>

Each value represents the mean of 3 observations.

<sup>ab</sup>Comparisons between NIE and EIE blends within a row. Means with different letters are significantly different ( $P < 0.05$ ).

The PS before and after the EIE required the highest force for penetration among all blends analysed. This indicated that PS was the hardest among these blends. The compression force required for PS decreases as the PKO or SBO percentage in the blend is increased. For equi-mixtures of ternary blends which having 33.33% equal proportions of PS, PKO and SBO (PS/PKO/SBO) experienced a prominence decrease with 43.01% lower as compared to the PS. This indicated that the hardness of PS was significantly reduced by blending with PKO and SBO.

Table III also indicated that the EIE promotes a decrease in the compression force values for the blends. Apparently, EIE PS resulted in 30.26% reduction of force required by NIE PS, EIE PKO with 32.14% reduction as compared to the NIE PKO, respectively. This finding can be explained due to the effect of randomisation of TAG by Lipozyme TL IM resulted softer and elastic blends produced as compared to the NIE blends. Overall, the low compression force observed in this experiment following blending and EIE indicated a softer and more pliable texture being obtained, which in turn could have affected the rheological and textural properties, resulting in a product with a more homogeneous and less granular appearance with satisfactory spreadability.

#### F. Optimisation and Fitting Ternary Blends of PS, PKO and SBO for Trans-Free Table Margarine Formulation

The regression coefficient and correlation of the adjusted model to the experimental data in mixture design studies of both the independent and dependent variables were fitted to linear and quadratic models and residuals plots were generated to check the goodness of model fit. The linear model was found the best fitted for SMP and SFC at 5, 10, 15 and 20, 25, 30 °C, while the quadratic model was adequately fitted to the responses of SFC at 35, 40, 45 and 50 °C. The R<sup>2</sup> value obtained through the analysis of response models (SFC

and SMP ternary diagrams) was found to be always greater than 0.92, indicating a great fitting model (Fig. 3).

As mentioned before, optimisation of ternary mixture of PS, PKO and SBO for table margarine formulation was conducted through graphical multiple responses optimisation tool in Design-Expert 8.0.4 (2010) software. This tool is able to predict all possible proportions of blend constituents having similar SFC profile with table margarine and to determine as an area in a ternary diagram. In doing so, it is necessary to enter the SFC range at each temperature suitable for table margarine. The acceptable SFC range for each temperature was selected based on SFC profile of oils extracted from commercial table margarine [2]. Fig. 3 shows possible fat blend formulations for table margarine made from PS, PKO and SBO blends.

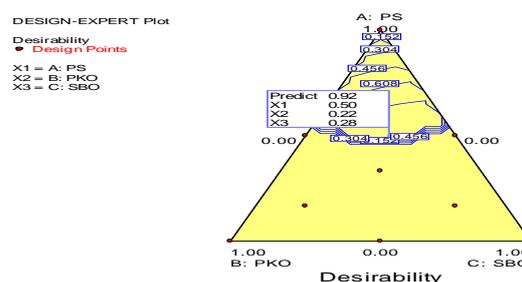


Fig. 3. Enzymatically-interesterified trans-free table margarine formulation with PS/PKO/SBO (50/22/28, w/w).

#### IV. CONCLUSION

Ternary phase diagram of PS, PKO and SBO provided valuable information through analysis of complicated solution behaviors of these three oils and their influence on the physical properties of final product. *Trans*-free table margarine formulation containing PS/PKO/SBO [50/22/28, (w/w)] not only has beneficial effect due to the presence of high proportions of palm fraction but it also free from *trans*-FA. Design-Expert 8.0.4 (2010) software was found to have a valuable tool to optimise the new fat blend formulation using the minimum number of blend preparation. By using this tool, assessment of complicated interactions among the blend components through construction of the corresponding phase diagrams which are critical for optimisation purpose would also be possible.

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