PHYSICO-CHEMICAL PROPERTIES OF PALM STEARIN, SOYBEAN OIL AND THEIR BINARY BLENDS

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LIST OF ABBREVIATIONS

%	:	Percentage
%v/v	:	Percentage volume over volume
%wt/wt	:	Percentage weight over weight
α	:	Alpha
β	:	Beta
δ	:	Delta
γ	:	Gamma
µm	:	Micrometer
AOAC	:	Association of Analytical Chemists'
C12:0	:	Lauric acid
C14:0	:	Myristic acid
C16:0	:	Palmitic acid
C16:1	:	Palmitoleate
C18:0	:	Stearic acid
C18:1	:	Oleic acid
C18:2	:	Linoleic acid
C18:3	:	Linolenic acid
cm	:	Centimeter
FA	:	Fatty acid
FAC	:	Fatty acid composition
FAME	:	Fatty acid methyl ester
FAMEs	:	Fatty acid methyl esters
GC	:	Gas Chromatograph

HI	:	Hardness index
HLERO	:	Hydrogenated low-erucic acid rapeseed oil
HPLC	:	High-Performance Liquid Chromatography
HPO	:	Hydrogenated palm oil
HSO	:	Hydrogenated soy bean oil
IE	:	Interesterification
IV	:	Iodine Value
KI	:	Potassium iodide
LERO	:	Low-erucic acid rapeseed oil
LLL	:	Trilinolein
LnLnO	:	3-oleo-dilinolein
m	:	Meter
M	:	Molarity
min	:	Minute
mL	:	Mililiter
mm	:	Milimeter
MPOB	:	Malaysian Palm Oil Board
°C	:	Degree celcius
°F	:	Degree Fahrenheit
OLO	:	2-linoleo-diolein
PKO	:	Palm kernel olein
PLL	:	1-palmito--dilinolein
PLP	:	2-linoleo-dipalmitin
pNMR	:	pulsed Nuclear Magnetic Resonance
POO	:	1-palmito-diolein
POP	:	2-oleo-dipalmitin

POs : Palm stearin

POS : Palmito-oleo-stearin

PPP : Tripalmitin

PS : Palm stearin

RBD : Refined, Bleached, Deodourised

RSO : Rapeseed oil

SBO : Soybean oil

SFC : Solid fat content

SFO : Sunflower oil

SMP : Slip melting point

TAG : Triacylglycerol

WHO : World Health Organisation

ABSTRACT

PHYSICO-CHEMICAL PROPERTIES OF PALM STEARIN, SOYBEAN OIL AND THEIR BINARY BLENDS

The objective of this study was to determine the changes in physicochemical properties of palm stearin (PS) and soybean oil (SBO) blends at different mixing ratios in order to identify the suitable blend ratio of PS and SBO for certain food applications. Palm stearin was blended with SBO at different ratios of 70:30, 50:50 and 30:70 PS:SBO. The physicochemical properties of PS, SBO and PS:SBO binary blends were determined based on the major fatty acid composition, triacylglycerol profile, iodine value (IV), solid fat content (SFC), slip melting point (SMP) and hardness index (HI). Results indicated that the addition of SBO into PS caused the changes amount in fatty acid composition (FAC) of PS which cause decreased in the palmitic acid but increased in the linoleic acid. These results was supported with triacylglycerols (TAG) profile where the major TAG for PS which was 2-oleo-dipalmitin (POP) was decreased upon dilution with SBO, and increased the triunsaturated acid namely trilinolein (LLL). Blending PS and SBO resulted the IV increased significantly ($p<0.05$) with the increasing amount of SBO thus decreased the SFC, SMP, and HI for all of the blends. Based on SFC, the blend 50:50 PS:SBO have a product stability and resistance to oiling out, with good melting point below body temperature which might be suitable for product such as margarine that must be melted in the mouth with minimum waxiness in order to have a good oral-melt.

ABSTRAK

CIRI-CIRI FIZIKOKIMIA STEARIN SAWIT, MINYAK SOYA DAN ADUNANNYA

Objektif kajian ini adalah untuk menentukan perubahan ciri-ciri fizikokimia adunan stearin sawit (PS) dan minyak soya (SBO) pada nisbah yang berbeza untuk mengenal pasti nisbah gabungan yang sesuai untuk diaplikasi dalam makanan tertentu. Stearin sawit telah dicampur dengan SBO pada nisbah yang berlainan 70:30, 50:50 dan 30:70 PS:SBO. Ciri-ciri fizikokimia PS, SBO dan adunan PS:SBO telah ditentukan berdasarkan komposisi asid lemak utama, profil triasilgliserol, nilai iodin, kandungan lemak pepejal, nilai takat gelincir dan indek kekerasan. Keputusan menunjukkan penambahan SBO kedalam PS menyebabkan perubahan dalam komposisi asid lemak PS iaitu asid palmitik telah menurun tetapi terdapat peningkatan kandungan asid linolik. Keputusan ini disokong dengan profil triasilgliserol dimana triasilgliserol utama bagi PS iaitu 2-oleo-dipalmitin (POP) telah menurun apabila pencairan dengan SBO manakala asid tidak tepu iaitu trilinolein (LLL) meningkat. Percampuran PS dan SBO mengakibatkan nilai iodin meningkat dengan ketara ($p<0.05$) apabila peratus SBO ditambah kedalam PS, dan kandungan lemak pepejal, nilai takat gelincir, dan indek kekerasan semua adunan menurun. Adunan 50:50 PS:SBO mempunyai kestabilan produk dan rintangan daripada meleleh keluar, dengan takat lebur di bawah suhu badan yang sesuai untuk produk seperti marjerin yang mesti cair dalam mulut tanpa rasa "waxy

CHAPTER 1

INTRODUCTION

1.1 Background of study

Today, partial hydrogenated vegetable oils are used worldwide in the formulation of functional shortenings and confectionary coatings. Unfortunately, partial hydrogenation of vegetable oils results in the production of *trans* fatty acids that once ingested have detrimental effects on metabolic functions, resulting in altered cell membrane integrity and reduced production of biologically active metabolites derived from essential fatty acids (Mensink, 2005; Stender and Dyerberg, 2004).

With the rising demand of natural products and emphasis on nutritional enrichment, blending of vegetable oils and fats has emerged as an economical way to produce edible oils devoid of any chemical treatment and which possess natural flavour and characteristics as well as nutritional value. The advantages of using blending as a means of modifying oils is that it is easy and less cost. More importantly blending of oils serves to improve and enhance the nutritional and qualities of the oils by combining of oils into one, by this therefore improving commercial viability (AbdulKarim *et al*., 2010b).

The blending of palm stearin and soybean oil at different ratios could able to modify the properties of natural fats. Blending can changes the natural physical and chemical characteristics of a fat or oil, thus it offers greater functionality for a large number of product formulations. The blending process of palm stearin which is highly saturated fat with the sunflower oil which is highly unsaturated fat can alter the chemical composition of the fats. The blending and interesterification are

worth to investigate given information available on the utilization of high melting palm stearins obtained from palm oil after fractionation (Pal *et al*., 2001).
Palm stearin obtained from palm oil by fractionation has limited use due to its high melting point. A great deal of work has already been done in utilising palm stearin by interesterification and by blending in mixtures with liquid oils (Majumdar and Bhattacharyya, 1986). The high melting palm stearin fraction with melting point of about 58 °C cannot be used as such in fat-based edible products due to its high melting point (Ghosh and Bhattacharyya, 1997).

One of the potential oils that can be blended with palm stearin is soybean oil. Soybean oil is obtained from soybeans, *Glycina maxima*, which are grown in several countries of the world. Soybeans are native to Eastern Asia, where ancient Chinese literature indicates that soybeans have been important part of their diet for centuries (O’Brien, 2009). Soybean oil has a relatively high content of linoleic acid and linolenic acid (Gunstone, 2011). Driscoll *et al*. (2001) reported that soybean oil is one of the major oils that have been used in emulsion products such as margarine, salad dressings and mayonnaise. Previous studies have shown that blending of soybean with other oils or fats could improve overall properties.

1.2 Problem statements

Trans fatty acid is a major concern of the world. Consumption of high saturated oil is most currently issues in Malaysia. Since Malaysians continue to lead unhealthy lifestyles and high-calorie unhealthy foods, the situation is definitely worse now. Because of this situation, people in Malaysia suffer obesity and heart disease problem. It was reported that obesity prevalence of 14.0% by Mohammad *et al.* (2008). Pang (2006) reported based on the National Health Morbidity Survey (NHMS) that 2.99 million people will have heart disease by year 2010 and 3.56 million, subsequently in 2020. In order to overcome this problem, the need for low saturated fat is in critical demand for the people in Malaysia.

Palm oil and palm products have become important raw materials in the production of solid-fat formulations such as shortening and margarine in view of the current trends. Palm oil is very suitable as a major component in margarine and shortening because it is naturally semi-solid and it can be further modified by fractionation, interesterification, hydrogenation and blending (Nor Aini and Miskandar, 2007).

Soybean oil is widely accepted as healthy oil, low in saturated acids and rich in polyunsaturated acids, especially linoleic acid. However, these oils are also easily oxidised, leading to rancidity and quality deterioration (Chen *et al*., 2011; Waraho *et al*., 2011). Since vegetable oils are liquid at room temperature, their use in foods is limited (Karabulut *et al*., 2003). Soybean oil and other fats and oils containing linolenic acid show reversion phenomenon when exposed to air. Reversion flavour is a particular type of oxidized flavour which develops at comparatively low levels of oxidation (deMan, 1999). Apart from that, high linolenic (C18:3) and linoleic (C18:2) fatty acids are the most important precursors of flavour reversion (O'Brien, 2009).

In many studies, it has been claimed that saturated fatty acids increase blood cholesterol levels, while mono and polyunsaturated fatty acids decrease blood cholesterol levels. Thus, it has been recommended that people should change their diet in this way. So a majority of people has began to consume vegetable oils instead of animal fats (Karabulut *et al*., 2003).

In order to improve the nutritional value and stability margarine and shortening, the blending of palm stearin and vegetable oil such as soybean oil could be possible. The suitable ratio of blending of palm stearin and soybean oil can be used in modifying the nutritional value and physicochemical of margarine and shortening.

1.3 Significance of study

The main purpose of this study was to modify oils and fats by using a process of fat modification method which is non-destructive and green method in order to produce the low saturated oil. The aim of this study was to determine the physicochemical properties of palm stearin and soybean oil blends in different mixing ratios. In addition, it also aimed at finding a blend with oxidatively stable palm stearin and with low melting point by the addition of soybean oil which is high degree of unsaturation and avoiding the presence of *trans* fatty acid.

This study will help the oil producing industry to find the most economically oil blends, with maximum nutrition as well as desirable physicochemical properties. Thus blending was a good choice where edible oils of good characteristics and quality could be achieved. The food value of the oils and blends can also be predetermined to provide the safest food for consumers (Bazlul *et al*., 2010).

1.4 Objectives

The objectives of this study were:

1. To determine the changes in physicochemical properties of palm stearin and soybean oil blends at different mixing ratios.
2. To determine the suitable blend ratio of palm stearin and soybean oil for certain food applications.

CHAPTER 2

LITERATURE REVIEW

2.1 Soybean oil

2.1.1 Chemical and physical properties of soybean oil

Soybean is the dominant oilseed produced in the world, because of its favorable ergonomic characteristics, its high quality protein, and its valuable edible oil. It contributes 47% of all oilseeds produced worldwide in 2008/09 (Gunstone, 2011). Soybean oil is less expensive than corn, safflower, and sunflower oils, yet it has many of the desirable characteristics of these so-called premium vegetable oils. It has high linoleic acid content and low saturated fatty acid content, and thus it is more desirable nutritionally than the more saturated oils (Pryde, 1980).

Soybean oil, palm oil and low-erucic acid rapeseed oil are the main vegetable oils used in industrial shortenings (Danthine and Deroanne, 2003a). Soybean oil contains 12%-15% saturated fatty acids which is mostly palmitic and 85%–88% unsaturated fatty acids which is mostly oleic, linoleic, and linolenic (Ribeiro *et al*., 2009). The essential fatty acid linoleic (18:2 *n-6*) and α-linolenic (18:3 *n-3*) acid account for 89% and 11% of the total essential fatty acid from this source. Soybean oil and canola oil are the only two common plant oils that have a considerable amount of *n-3* linolenic acid (Gunstone, 2011).

The low linolenic (~3%), ultra low-linolenic (~1%), and mid-oleic with low-linolenic acid soybeans have been produced by Pioneer Hi-bred (a division of DuPont), Monsanto, and researchers at Iowa State University (marketed by Asoyia) (Wilkes, 2008). In world vegetable oil consumption, soybean oil (SBO) stands out for its nutritional qualities, permanent supply, considerable economic value and high functionality, making it a particularly interesting raw material for producing special fats (O'Brien, 2009).

Nor Hayati *et al*. (2009) had discovered that soybean oil demonstrated the melting behaviour range at the lowest temperatures (approximately from -40 ºC to 0 ºC). Based on the solid fat content, SBO was almost completely liquid at all temperatures measured. The difference in solid fat content for the soybean oil compared to palm kernel olein was by differences in their fatty acid compositions and their corresponding triacylglycerols species.The soybean oil composition and physical properties are as shown in Table 2.1.

Table 2.1 Soybean oil composition and physical properties

Characteristics	**Typical**	**Range**
Specific gravity, 25/25 ºC	0.9175	0.917 to 0.921
Refractive index, 25 ºC	1.4728	1.470 to 1.276
Iodine value	131	123 to 139
Saponification number	192	189 to 195
Unsaponifiable matter, %	0.6	0.6 to 1.6
Titer, ºC	24	
Melting point, ºC	-22	20 to 23
Solidification point, ºC		16 to 10
Cloud point, ºC	-9	
Cold test, hours	25	
AOM stability, hours	12	12 to 15
Oxidative stability index, (110 ºC), hours	2	2.2 to 3.3
Fatty acid composition, %		
C-14:0 Myristic	0.1	<0.2
C-16:0 Palmitic	10.6	8.0 to 13.3
C-16:1 Palmitoleic	0.1	<0.2
C-17:0 Margaric	0.1	
C-18:0 Stearic	4	2.4 to 5.4
C-18:1 Oleic	23.3	
C-18:2 Linoleic	53.7	
C-18:3 Linolenic	7.6	
C-20:0 Arachidic	0.3	
C-20:1 Gadoleic		
C-22:0 Behenic	0.3	
C-22:1 Erucic		
C-24:0 Lignoceric		
Triglyceride composition, %		
SSS Trisaturated	0.1	
SUS Disaturated	5.6	6.6 to 9.6
SUS Disaturated		5.2 to 9.3
SUU Monosaturated	35.7	14.0 to 32.4
UUU Triunsaturated	58.4	55.2 to 80.3

Notes: S= saturated, U=unsaturated, AOM= active oxygen method
Source: O'Brien (2009).

In general, soybean oil was characterised by a high proportion of polyunsaturated fatty acids up to 65%. High proportion of polyunsaturated fatty acids in SBO was mainly derived from fully unsaturated triacylglycerols. Linoleic (C18:2) acid was predominant fatty acid in SBO, followed by oleic acid (C18:1), palmitic oil (C16:0) and linolenic (C18:3) acids in decreasing order (Nor Hayati, 2009). Triacylglycerols are the primary neutral lipids in soybean oil. Due to high concentration of unsaturated fatty acid in natural soybean oil, nearly all triacylglycerols molecules contain at least two unsaturated fatty acids, and di- and tri-saturates are essentially absent (Gunstone, 2011).

2.2 Palm stearin

2.2.1 Physical and chemical properties of palm stearin

The oil palm (*Elaeis guineensis jacquin*) was originated from South Africa. It was introduced to East Asia as an ornamental plant at the Bogor Botanical Garden in Java, Indonesia in 1848. Another species of oil palm, *Elaeis Oleifera*, originates from Central America and South America. Its oil is more unsaturated, but the oil to bunch ratio is extremely low, making it uneconomical to plant on a commercial scale (Gunstone, 2011).

Palm stearin, the cheaper high-melting fraction from palm oil, can be used as a source of fully natural hard component in the manufacture of solid fat products such as shortenings, margarines and fat spreads. Palm stearin is available in a wide range of melting points and iodine values. However, because of its high melting point (44 ºC–56 ºC), palm stearin poses problems in the manufacture of the solid fat products as it confers low plasticity to the products and does not completely melt at body temperature (Pantzaris, 2000).

Palm stearin, the more saturated fraction of palm oil, is more viable in composition and thus physical characteristics. The wide range in solid fat content is consistent with the wide range in iodine value for this fraction (Gunstone, 2011). The palmitic acid content of the stearins varies from 47% to 74%, while oleic acid ranges from 15% to 37% (Gunstone, 2011). Table 2.2 shows the fatty acid and triacylglycerol composition of palm stearin, soft stearin and palm mid-fraction.

Table 2.2 Fatty acid and triacylglycerol composition of palm stearin and palm midfraction

	Stearin*	**Soft stearin***	**Palm mid-fraction****
	n = 150	**n = 1**	**n = 39**
Fatty acid composition (wt %)			
12:00	0.1-0.6	0.1	0-0.3
14:00	1.1-1.9	1.1	0.8-1.4
16:00	47.2-73.8	49.3	41.4-55.5
16:01	0.05-0.2	0.1	
18:00	4.4-5.6	4.9	4.7-6.7
18:01	15.6-37.0	34.8	32.0-41.2
18:02	3.2-9.8	9	3.6-11.5
18:03	0.1-0.6	0.2	0-0.2
20:00	0.1-0.6	0.4	0-0.6
Iodine value	21.6-49.4	46.7	34.5-54.8
SMP (ºC)	44.5-56.2	47.7	24.3-44.9
Triacylglycerols by carbon number (wt %)			
C46	0.5-3.3	1.2	0-1.6
C48	12.2-55.8	15.3	1.4-11.3
C50	33.6-49.8	42.7	45.5-73.9
C52	5.1-37.3	33.4	19.4-42.0
C54	TR-8.4	7.4	1.7-8.5
C56	ND	ND	0.0.9

Source: Tan *et al*., (1981) ; Tan and Oh (1981)

Key:
ND = not detectable
TR = trace
SMP = slip melting point

According to deMan and deMan (1995) palm stearin is a β tending fat due to its high concentration of triplamitin. It contains 10%-32% of triplamitin, depending on the fractionation conditions. Peck *et al*. (1989) also reported palm stearin was least stable in the β′ form, followed by palm oil. The hydrogenated oils were very stable in the β′ form. Fractionated palm oil characteristics are listed in Table 2.3.

Table 2.3 Fractionated palm oil characteristics

Characteristics	Palm Oil Fraction		
	Whole	**Olein**	**Stearine**
Softening Point, ºC	31-38	19-24	44-56
Titer, ºC	42-46		46-54
Density at 50/25 ºC	0.892-0.893		
Density at 60/25 ºC		0.909-0.903	0.882-0.891
Iodine Value	51-55	51-61	22-49
Saponification Value	190-202	194-202	193-206
Cloud Point, ºC		6-12	
Unsaponifiable Matter, %			0.1-1.0
Solid Fat content (NMR) at:			
10ºC-50ºF	47-56	28-52	54-91
20ºC-68ºF	20-27	3-9	31-87
30ºC-86ºF	6-11	0	16-74
40ºC-104ºF	1-6		7-57
50ºC-122ºF			0-40
Fatty Acid Composition, %			
C-14:0 Myristic	1-1.5	1-1.5	1-2
C-16:0 Palmitic	42-47	38-42	47-74
C-18:0 Stearic	4-5	4-5	4-6
C-18:1 Oleic	37-41	40-44	16-37
C-18:2 Linoleic	9-11	10-13	3-10
Triglyceride Composition, %			
SSS Trisaturated	0.8-9.0	0.1-0.3	22.2
SUS Disaturated	38.5-50.3	37.6-46.1	43.9
SUU monosaturated	31.8-44.4	41.3-49.1	25.6
UUU Triunsaturated	4.8-9.8	6.4-8.4	3.9
Disaturate, %	3.0-7.6	4.7-6.1	4.5
Natural Antioxidant Content, µg/g			

Notes: S= saturated, U=unsaturated, AOM= active oxygen method
Source: O'Brien (2009)

Palm oil can be separated under controlled thermal condition into two components which is solid (stearin) and a liquid (olein) fraction. This fractionation process can be affected either in the dry form or in the presence of a detergent or solvent. The method employed, to a certain extent, determines some of the chemical and physical properties of the oleins and stearins produced, especially the stearins. By varying the fractionation methods and conditions used, a range of stearins with differing chemical and physical properties could be produced, yet keeping the chemical and physical properties of the oleins to within a very narrow range of values (Basiron, 2005).

Basiron (2005) also mentioned under normal fractionation conditions, soft stearins and oleins with cloud points in the range of 8 ºC–10 ºC are produced. Where required, fractionation conditions could be specifically altered to produce stearin or olein of a desired specification for specialised application, but within the domain of the composition of palm oil.

2.3 Fat modification

2.3.1 Blending

For many years, modification of original fats by means of direct blending with other fats, fractionation, hydrogenation and interesterification has been attempted to improve the fat functionalities and thus optimise their application in food products. Of these modification techniques, direct blending of fats is the method of choice as it has been considered to be a cheap and nondestructive technique (Nor Hayati *et al*., 2009). Blending of vegetable oils and fats has emerged as an economical way of modifying the physicochemical characteristics of vegetable oils and fats besides enhancement in oxidative stability (Chu and Kung, 1997).

Blending polyunsaturated oil with more saturated or monosaturated oils is an option to adjust fatty acid levels to optimal level, such as combining high oleic sunflower oil with corn oil or hydrogenated soybean oil with soybean oil (Chu and Kung, 1997; Chu and Kung, 1998; Premavalla *et al*., 1998). Moreover, understanding the physicochemical properties of these blends allows one to design the blends with formulations suitable for use in specific food applications in a more meaningful way (Nor Hayati *et al*., 2009).

According to Bazlul *et al*. (2010), the iodine values for the blends of palm olein, sunflower oil, canola oil and soybean oil were not similar. At the initial stage, all the oil blends was found to be similar iodine values. However, it was anticipated that the iodine value would decrease with exposure of light and air would follow the ratio of the percentage of the mixture of two oils due to the decrease in unsaturation. From the result, among all blends palm olein and sunflower blends show more consistency between initial and final iodine values.

From the other previous study, AbdulKarim *et al*. (2010b) also reported blending has a significant influence on iodine value (IV) where linoleic acids from sesame seed oil tend to migrate into oil blends. Sesame seed oil was characterised by high unsaturation and its difference from palm oil was reflected in its higher iodine value. Increasing amounts of sesame seed oil (from 10% to 20%, 30% and 40%) and decreasing amounts of palm olein (from 90%, 80%, 70% and 60%) in the blends, results in increase in the degree of unsaturation. The iodine value of the oil blends was therefore improved by blending of palm olein with sesame seed oil.

Hasmadi *et al*. (2005) fractionated blend of 80% refined, bleached, deodourised (RBD) palm oil with 20% sunflower oil and 60% RBD palm oil with 40% sunflower oil at three different temperatures. After blending 60% RBD palm oil with 40% sunflower oil, this blend had higher IV value compared with the other liquid fraction blend of 80% RBD palm oil with 20% sunflower oil.

Norizzah *et al*. (2004) reported that the slip melting point increased with an increase in the amount of palm stearin in the blends with palm kernel olein oil due to high amount of high melting triacylglycerols such as oleodipalmitin (POP) and tripalmitin (PPP) in palm stearin (PS). The largest increase in slip melting point was observed in the blend with 40% to 60% of palm stearin added into the palm kernel olein (PKO). The softening points of the blends of PS and PKO increased with the addition of PS in a non linear relationship.

According to Nor Hayati *et al*. (2009), blending of SBO with PKO reduced the complexity caused by the polymorphism transformation, featuring the endotherms that only related to the β′ fat crystals. Blending of PS and PKO contained mixtures of both β and β′ polymorphic forms with β form dominating except in the blends of 80% POs with 20% of PKO.

Appelqvist and Bialek (2005) reported blending of SBO with PKO at an appropriate level is expected to provide a sufficient solid fraction. This leads to an increase in structuring distribution by introducing a fat droplet network in the emulsion system. According to Danthine and Deroanne (2003a) polymorphic stability in the β′ form of the hydrogenated low erucic acid rapeseed oil mixture blend increased with the addition of palm oil or hydrogenated palm oil.

From the microscopic observation, Norizzah *et al*. (2004) discovered that PS consists of a dense network of fine plate-like structure. Addition of palm kernel olein oil to palm stearin resulted in mixture of plate like and spherulitic crystals. Nor Hayati *et al*. (2009) reported there is a trend of increasing solid fat content with increasing percentage of palm kernel oil in the blends. The blends of 60% SBO with 40% PKO had compatible polymorphs in their solid phase, reflecting a good miscibility of the systems. Norizzah *et al*. (2004) also discovered that the solid fat content increased with the increasing amount of PS in the blend with PKO. Addition of PKO to PS lowered the solid fat content at all measured temperatures.

Bazlul *et al*. (2010) revealed that the viscosity, like all other Newtonian fluids, decrease with an increase in temperature due to the destruction of the solidified oil crystals, as the blends are the simple mixing of two oil which were palm olein-soybean oil, palm olein-canola oil and palm olein-sunflower oil in different proportions, it appears to be an average property of the mixture in the blends.

According to Nor Aini *et al*. (1995), blending of sunflower oil or high-oleic safflower oil with double fractionated palm olein improved the fatty acid composition in terms of the saturated, monosaturated, polyunsaturated ratio**.** It showed that blending of soybean oil with underutilised and cheaper yet more stable oil like palm kernel oil at 40% has pronouncedly modified the overall chemical composition of the starting soybean. From other previous study, AbdulKarim *et al*. (2010b) reported the most prominent fatty acids in palm olein were palmitic (38.39%) and oleic (45.77%) acid, while sesame seed oil has abundance of oleic (40.52%) and linoleic (46.03%) acids. Blending of palm olein with sesame seed oil caused the content of palmitic acid to decrease and linoleic acid to increase in the blends. The fatty acid (FA) composition thus was changed by blending.

Blending of 60% SBO with 40% PKO would provide a significant amount of medium chain triacylglycerols (Nor Hayati *et al*., 2009). According to Driscoll *et al.* (2001), emulsion containing a significant amount of medium chain triacylglycerols was found to be more stable when compared with emulsion containing a single long chain triacylglycerols of soybean oil. Blending RBD palm oil with sunflower oil changed the triacylglycerol composition and resulted in lower amounts of monounsaturated triacylglycerols, namely POP and palmitic-oleic-stearic (POS), as well as stearin-oleic-oleic (SOO) and palmitic-linoleic-palmitic (PLP). On the other hand, the amounts of di- and polyunsaturated triacylglycerols, namely oleic-linoleic-oleic (OLO), palmitic-linoleic-linoleic (PLL), oleic-linoleic-linoleic (OLL) and linoleic-linoleic-linoleic (LLL) or linoleic-linolenic-oleic (LLnO), increased (Hasmadi *et al.*, 2005).

Mohamed and Khaled (2012) reported by increasing proportion of black cumin seed oil and coriander seed oil in corn oil, level of polyunsaturated fatty acid decreased while monounsaturated fatty acid content increase. On the other hand, Pal *et al.* (2001) reported that blending with liquid oils like sunflower oil and soybean oil in different proportions, offer nutritionally important fat products with enriched content of essential fatty acids like C18:2 and C18:3. Incorporation of 50% of palm olein to palm stearin reduced the level of saturated fatty acids by 20% and increased in 48% and 50% the levels of polyunsaturated fatty acid and monounsaturated, respectively. But, from a nutritional point of view, blends containing more than 50% of palm olein proved to be interesting to make healthy food and this is due to the content of polyunsaturated fatty acid and monounsaturated fatty acid of these blends (WHO/FAO, 2003). Apart from that, blending sunflower oil with canola oil at different levels led to increase its stability against oxidation and the extent of this phenomenon was basically depend on the mixing ratios (Radwan *et al*., 2010).

It is important to relate changes in chemical properties, due to blending, to predictive changes in physical properties. This is important for optimising a perfect blend to suit individual applications (Baljit *et al*., 2002). AbdulKarim *et al.* (2010a) concluded that simple blending of natural fats and oils can help to achieve desired characteristics without resorting to drastic methods such as chemical or enzymatic modifications, which often are expensive and time consuming.

2.3.2 Interesterification

Chemical interesterification leads to modifications in triacylglycerol composition and, consequently its physical characteristics. The fatty acid interchanges resulting from interesterification lead to changes in the physical characteristics of oils and fats because in nature fatty acid are not randomly distributed between the triacylglycerols present. In most oils and fats, the 2-position of the triacylglycerols molecules is preferentially occupied by unsaturated fatty acid. In its most commonly used form, interesterification produces a truly random distribution of fatty acid that leads to an increased number of triacylglycerol species. This results in changes in the physical characteristics of the oils and fats, such as melting and crystallisation behaviour. The rearrangement process does not change the degree of unsaturation or the isomeric state of the fatty acid as they shift from one position to another (Noor Lida *et al*., 2002).

Interesterification caused considerable rearrangement of triacylglycerol species, reduction of trisaturated triacylglycerol content and increase in monounsaturated and diunsaturated triacylglycerols, resulting in lowering of respective melting points. The interesterified blends displayed reduced solid fat content at all temperatures and more linear melting profiles as compared with the original blends (Ana Paula *et al*., 2009). Chemical interesterification significantly modified the triacylglycerol composition, and consequently the melting point, solid fat content and consistency properties of the sunflower oil and fully hydrogenated soybean oil blends. The interesterified blends displayed characteristics suited to a variety of industrial purposes, offering alternatives to partly hydrogenated fats (Ana Paula *et al*., 2009).

The chemical interesterification process neither affected the degree of saturation nor caused isomerisation of the fatty acid double bonds (Ana Paula *et al*., 2009). Chemical interesterification produces a complete positional randomisation of acyl groups in triacylglycerols by using chemical catalyst. Enzymatic interesterification uses lipases as catalyst. Each type of interesterification possesses advantages and disadvantages. Chemical interesterification is preferred over enzymatic interesterification because of the lower investment and production costs of the finished products (Noor Lida *et al*., 2002).

Interesterification of palm stearin with liquid vegetable oils such as olive oil could yield a good solid fat stock that may impart desirable physical properties, because palm stearin is a useful source of vegetable hard fat, providing β’ stable solid fats (Roberta *et al*., 2010). Roberta *et al*. (2010) reported that blending and chemical interesterification with olive oil is an effective way to modify the physical and chemical properties of palm stearin. Chemical interesterification can be used to improve the physical properties of fat blends of palm stearin with olive oil. Even though chemical interesterification is extremely functional from the technological point of view, replacing partially hydrogenated fats in food products, particularly in shortenings and confectionery products, does pose challenges, because suitable solid fat content curves, plasticity, crystallisation properties and texture are difficult to obtain in the absence of *trans* fatty acids (Ana Paula *et al*., 2009).

Modification of the characteristics of palm oil, sunflower oil, and palm kernel olein according to conventional three-component mixture designs was undertaken by a combination of blending and chemical interesterification techniques. Upon chemical interesterification, extensive rearrangement of fatty acid among triacylglycerols was evident. Concentrations of several triacylglycerols were increased, some were decreased, and several new triacylglycerols formed (Noor Lida *et al*., 2002).

In addition to its β’ tendency palm oil has a natural semisolid consistency which is useful in margarine applications, but one, which can also be hardened further by random interesterification without the subsequent formation of *trans* fatty acid. Palm oil is however a complex mixture of triacylglycerols, with respect to the carbon chain length (Paul and Niall, 2007). Interesterification of blends of palm stearin and palm kernel olein resulted in the formation of β’ polymorphic oil, yielding mixtures with lower slip melting point and solid fat content (Norizzah *et al*., 2004).

Interesterification is used to modify fat properties without recourse to partial hydrogenation. Hardened fats produced by partial hydrogenation contain *trans* isomers, which are now regarded as undesirable by nutritionists and will be increasingly subject to product labeling regulations. Liquid fats can be hardened by interesterification with fully saturated fats (either stearin fractions or fully hydrogenated oils), raising the solid fat content without isomerising any of the fatty acids. The use of interesterification to produce margarine and spreads has increased recently, particularly in Europe (Charlie, 2005).

Randomisation of soybean oil triacylglycerols improved oxidative stability compared to natural soybean oil (Gunstone, 2011). In palm stearin, palm olein and their blends, chemical interesterification increased trisaturated and decreased monosaturated-diunsaturated triacylglycerols, due to the probabilistic distribution of FA (Fabiana *et al*., 2009). There are several advantages to use palm stearin as a component for interesterification with liquid oils to yield a good hardstock, such as availability of the oil, cheap raw material and eliminate the need for hydrogenation (Gunstone, 2011).

Blending and chemical interesterification are an effective way to modify the physical and chemical properties of palm stearin, palm olein and their blends. The chemical interesterification allows to obtain fats with various degrees of plasticity, increasing the possibilities for the commercial use of palm stearin and palm olein (Fabiana *et al*., 2009). The comparison of the blends before interesterification and after interesterification is shown on the Table 2.4.

Table 2.4 Physical characteristics of vanaspati based on blend of palm stearin (POs) with other oil before & after interesterification.

Sample	**Blends before interesterification**			**Interesterification blends**		
	Melting point (ºC)	**Appearance**	**Consistency**	**Melting point (ºC)**	**Appearance**	**Consistency**
PS:SBO						
40:60	41.5	Wet, granular	Soft	35.9	Wet, oily, granular	Very soft
60:40	45.5	Slightly dry, granular	Slightly firm	41.5	Wet, oily, granular	Very soft
80:20	47.7	Dry, granular	Firm	45.6	Wet, oily, granular	Soft
PS:RSO						
40:60	43.6	Wet, granular		38.7	Wet, oily, smooth	Very soft
60:40	45.9	Slightly dry, granular	Slightly firm	43.4	Wet, granular	Soft
80:20	48.5	Dry, granular	Firm	45.6	Wet, oily, granular	Soft
PS:SFO	43.8	Wet, granular	Soft	34.3	Wet, oily, granular	Very soft
40:60	45.3	Dry, granular	Slightly firm	39.3	Wet, oily, granular	Very soft
60:40	48.6	Dry, granular	Firm	45.4	Oily, granular	Soft
80:20	39.0	Dry, granular	Slightly firm	38.5	Granular	Soft

Source: Nor Aini et al. (1997)

Key: PS = Palm stearin SBO = Soybean oil
RSO = Rapeseed oil SFO = Sunflower oil

2.3.3 Fractionation

Fractionation is the economical and second important way of fat modification technique. Solid fats contain either already precipitated or still dissolved solid triacylglycerols, which under controlled conditions of cooling can be induced to full or partial crystallisation. The crystals formed can be removed by, for example, filtration during which the fat or oil is separated into two or more fractions, at least one having a higher melting point than the starting oil and one remaining liquid at temperature of intended uses, not far above that of crystallisation. The lower the separation temperature chosen, the lower will be the melting point (cloud point) of the two fractions (Hoffmann, 1989).

The physico-characteristic of edible oils and fats are closely linked to their triacylglycerol composition. Edible fats and oils are fractionated to provide new materials more useful than natural product. Fractionation processes separate fats and oils into fractions with different melting points. Separation of fat or oil into fractions can also provide two or more useful functional products from the same original product (O'Brien, 2009).

If fat is melted and cooled slowly to below its melting point, the triacylglycerols with a higher melting point than the tempering temperature will eventually form crystalline material, which can be relatively easily filtered off from the liquid part. Today, the most important oil in terms of fractionation is palm because it has a unique fat profile that can be broken down into individual fractions and sub-fractions thereof (Paul and Niall, 2007).

According to Hasmadi *et al.* (2004), in dry fractionation process, the oil as such is partially crystallised by fractionating the melt in a controlled manner at the desired temperature, after which the remaining liquid is separated from the solid fraction by means of a vacuum filter or membrane filter press. Dry fractionation of palm oil is efficient in producing fractions enriched in saturated fatty acids and could be used to produce different fat products (structured fats, spreads), or as an intermediate step in production of high-melting-point stearins appropriate for confectionery fat formulation (Miguel *et al*., 2011). Figure 2.1 illustrated the dry multiple fractionation of palm oil.

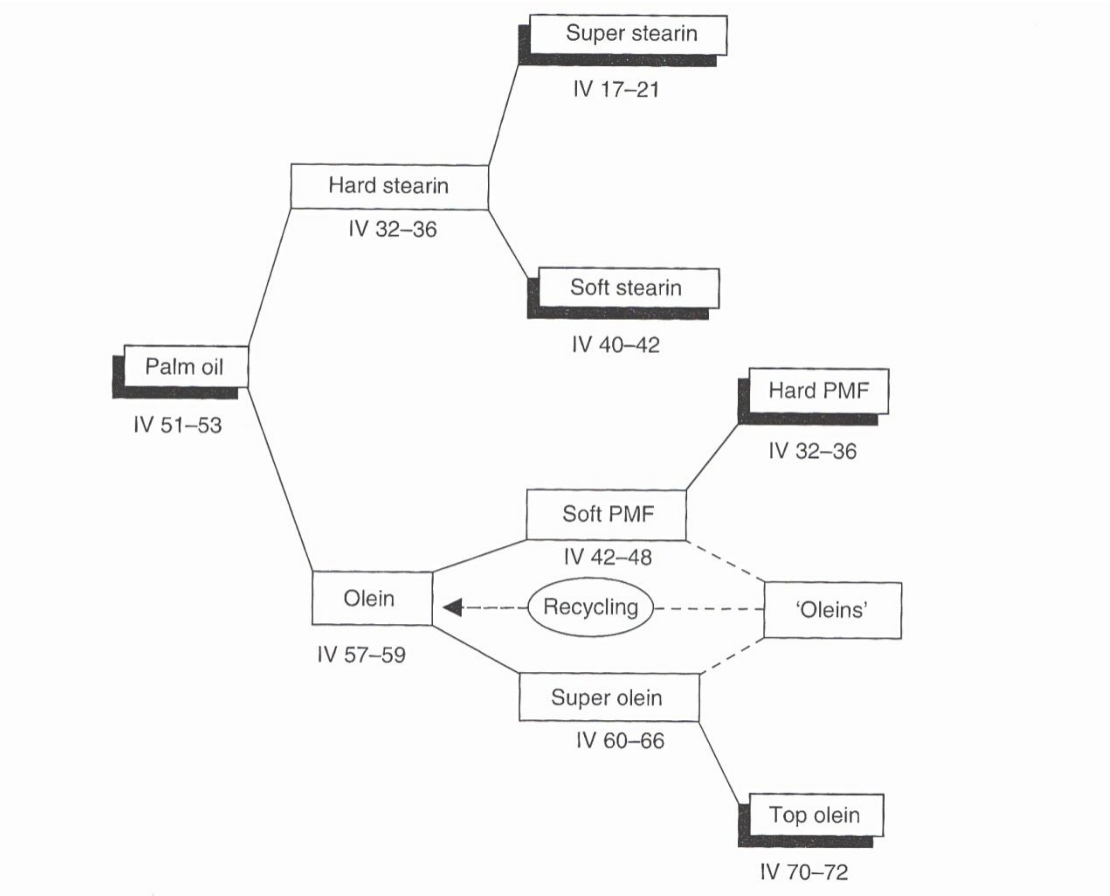

Figure 2.1 Dry multiple fractionation of palm oil. The fraction design hard stearin is more commonly name stearin or palm stearin.
Source: Deffense (1995).

2.3.4 Hydrogenation

Hydrogenation of oils and fats has been used since the beginning of the 20th century to produce hardstocks from a great number of oils, mainly liquid oils. Hydrogenation involves the addition of hydrogen atoms across the unsaturated double bonds of the particular triacylglycerols. Depending on the 'natural' occurring starting point of unsaturation of the oil, the degree of hydrogenation will result in a more saturated fat having a higher melting point than the starting material. The fundamental importance of this aspect cannot be over emphasised, because it enables the production of solid and semi-solid fats (Paul and Niall, 2007).

Hydrogenation of double bonds can be total or partial and the use of a catalyst is necessary to speed up the reaction. The specific surface area of a catalyst determines the speed of reaction, while the surface structure defines the selectivity. Full or partial hydrogenation can be carried out depending upon the solid fat content required for the hydrogenated product. The solid fat content is the amount of solid fat present in the oil at any given temperature (Desci and Koletzko, 1995).

Karabulut *et al*. (2003) reported in the partial hydrogenation, the product is saturated to a certain iodine value. In the selective hydrogenation it is aimed to reach to a certain iodine number and also polyenic fatty acids are converted to monoenic acids. Thus, the production of saturated fatty acids from monoenic fatty acids is avoided. Partial hydrogenation significantly reduces the incidence of offensive odour and flavour development by decreasing levels of the linoleic and linolenic, but the process creates higher melting disaturated triacylglycerols and *trans* isomers, responsible for cloudiness (Gunstone, 2006). The example of combined hydrogenation, interesterification and fractionation are shown in Table 2.5.

Table 2.5 Example of combined hydrogenation, interesterification and fractionation to produce low-trans fat from soybean.

	Iodine value	**Melting point (ºC)**	**Solid fat content (% at ºC)**			
			10	**20**	**30**	**40**
Soybean oil (SBO) feedstock	134	-7	0	-	-	-
Fully hydrogenated SBO (FHSBO)	1	71	95	94	94	93
Blending of SBO and FHSBO (60:40)	81	63	44	42	39	35
Random interesterification of SBO and FHSBO (60:40)	81	53	38	33	20	11
Fractionation of the interesterified oil						
Soft fraction	91	24	25	1	0	0
Hard fraction	63	58	60	58	45	32

Source: Kellens (2000)

During hydrogenation various side reactions occur, some of which have a strong impact on the physical and nutritional properties of the products. Double bond isomerisation and *trans*-fatty acid formation are most important side reactions. The *trans* double bond is a thermodynamically more stable configuration than its *cis* counterpart, and it is produced in significant quantity during partial hydrogenation (Gunstone, 2011).

In a theoretical sense, an oil hardened with perfect preferential selectivity would of all have its linolenic acids (18:3) reduced to linoleic acids (18:2) before any linoleic was reduced to oleic acid (18:1), then all linoleic acids would be reduced to oleic, before any oleic were saturated to stearic acid (18:0). Unfortunately, this does not happen in the actual practice (Karabulut *et al*., 2003). Karabulut *et al*. (2003) have identified, hydrogenation reduced both the iodine value and refractive index of soybean oil. Iodine value and refractive index decreased smoothly during the process due to saturation of double bonds. The iodine number of soybean oil was decreased from 133.6 to 73.3 and the refractive index decreased from 1.4636 to 1.4573 during the process.

The hydrogenation conditions used in that study did not produce large amount of stearic acid, but high amount of *trans* fatty acids are formed (Karabulut *et al*., 2003). Current evidence has implicated the intake of *trans* fatty acids to coronary heart diseases, and the inhibition of arachidonic acid biosynthesis, thus giving rise to issues related to growth necessary for healthy children (Desci and Koletzko, 1995).

2.4 Properties of oils and fats

2.4.1 Fatty acid composition (FAC)

The gas liquid chromatography fatty acid composition analysis provides a rapid and accurate means of determining the fatty acid distribution of fats and oils products. The fatty acid composition result provides a large quantity of information with one analysis, such as identification of individual fatty acids and quantities, saturate/unsaturated levels (calculated iodine value), identification of the unsaturated fatty acid isomers (*cis, trans*, conjugated, positional), provide data to determine the source oil proportions and processing of a blended product, and it applies equally well to refined and unrefined oils (O’Brien, 2009).

The gas chromatography identifies the fats and oils by retention time. O’Brien (2009) mentioned the retention time is indicated on the horizontal axis of the chart and is a qualitative index of the substance, and the area under the curve is in each case a quantitative measure of the component. Separation of fatty acid is based on chain length, degree of unsaturation, as well as the geometry and position double bonds.

From the previous study, Abdulkarim *et al.* (2010b) reported blending of palm olein and sesame seed oil altered fatty acid composition of palmitic and linoleic acids, which results in a significant change in trilinolein, dioleoyl-linoleoyl-glycerol, dipalmitoyl-3-linoleoyl glycerol and dipalmitoyl-3-oleoyl glycerol contents.

2.4.2 Triacylglycerols (TAG)

In general, fats and oils are composed of mixed acylglycerols rather than mixtures of simple acylglycerols. A mixed triacylglycerols has two or three different fatty acid joined to the glycerol. The characteristics of a triacylglycerols depend on the position that each fatty acid occupies on the glycerol molecules (O’Brien, 2009). Fully hydrogenated soybean, corn, canola, and peanut oils are essentially all tristearin due to low level of palmitic fatty acid form stable β-crystals. Cottonseed and palm oils with high palmitic fatty acid levels have mixtures of stearic and palmitic fatty acid when fully hydrogenated form β’ crystals (O’Brien, 2009).

Abdulkarim *et al.* (2010b) reported changes in triacylglycerol profiles will normally affect the physical characteristics of the oils such as the thermal behaviour. Due to the increased in the percentage of low melting TAG, it means that the oil will have a lower melting and crystallising temperatures, a characteristic that is desirable in oils for good storage and functionality.

2.4.3 Iodine value (IV)

The iodine value of oil does not indicate the position of the double bonds or amount of olefinic carbon but rather it provides an overall status of unsaturation of the oils so it is not possible to point out the position of double bond(s) which are more susceptible to oxidation (Knothe and Dunn, 2003). According to Zarinah *et al.* (2012), high IV oil contains a greater number of double bonds than low IV oil.

According Nasirullah *et al*. (1991) slow decrease in iodine value of oil blends may be due to induction period where fat was oxidized slowly showing initiation stage of auto oxidation reaction. Rapid changes in iodine value of oil blends may be attributed to propagation of auto oxidation process where hydroperoxides are formed from free radicals in fatty acids generated in initiation stage or auto oxidation reaction. During the end of storage period slight change in iodine value was observed which might be due to termination stage of reaction.

2.4.4 Solid fat content (SFC)

Solid fat content is the quantity of fat crystals in a fat or fat blend. It has a great influence on the suitability of the fat or fat blend for a particular application. The solid fat content is responsible for many product characteristics in margarines, shortenings and fat spreads, including their general appearance, ease of packing, spreadability, oil exudation and organoleptic properties (Dian *et al*., 2007). According to Narine and Marangoni (1999), the mechanical properties of edible fats can be influenced by a series of factors, including the amount of SFC, the polymorphism of the solid state as well as the microstructure of the network of crystalline particles.

Solid fat content profile reflects the fraction of the fat phase that is solidified at a particular temperature and strongly influences the sensory attributes and the physical stability of the fatty food. For instance, solid fat content at 20 ºC – 37 ºC is of interest to predict the melting behaviour of the confectionery fats that is related to important attributes such as mouthfeel and hardness. In a more complex food system, such as an oil-in-water emulsion, the amount of fat crystals at 5 ºC-10 ºC becomes important, being related to textural contribution by fat crystal networks, or to some extent, emulsion destabilisation due to partial crystallisation of the fat crystals (Boode and Walstra, 1993; Appelqvist and Bialek, 2005).

The palm olein had similar SFC profiles and their SFC increased with increasing amounts of palm stearin in the blends (Fabiana *et al*., 2009). The solid fat content therefore has important implications for the overall melting characteristics of margarines and spreads at various temperatures (Paul and Niall, 2007).

Bazlul *et al.* (2010) reported the solid fat content can be used as a useful tool to compare the resistance of the olein against crystallisation at low temperature. Products with lower solid fat content at sub-ambient temperature (such as 5 ºC or lower) will have better cold stability than those with higher solid fat content at the same temperature. Oil exhibiting a stable β' form tends to be smooth, provide good aeration and have excellent creaming properties (Dian *et al*., 2007).

2.4.5 Slip Melting Point (SMP)

According to Timms (1985), slip melting point is the temperature at which a fat in a capillary tube placed in water becomes soft enough to slip or rise up the tube. The fat will slip in the capillary tube where approximately 5% solid fat is present. The softening point (open tube melting point) or slip melting point is often used to characterise oils and fats and is related to their physical properties, such as hardness and solidification or melting behaviour (Goh and Ker, 1991).

The melting points of triacylglycerols are related to the fatty acid present. For a fatty acid, its melting point depends on the chain length and number and position of double bonds. It increases with increasing chain length and decreases with increasing *cis* unsaturation. The *trans* form has a significantly higher melting point than its *cis* isomer. Polymorphism is an important factor affecting melting point (Gunstone, 2011).

As fats and oils are complex mixture of compounds, they have no definite melting point and pass through gradually softening before becoming liquid (Pomeranz and Meloan, 2003). Miscibility is an important characteristic of specialty fats. When fats of different composition are mixed, they may show eutectic effects. This means that the melting point or solid fat content of the blend is lower than that of individual components (deMan, 1999).

Zarinah *et al*. (2012) reported during slip melting point measurements, the sample temperature is raised and the solid fat melts. The progressive reduction of crystalline matter means that, at a certain temperature, the fat crystal network lacks sufficient cohesion to hold onto its matrix and become sufficiently soft until it suddenly rises. Range of SMP value of palm olein, cotton seed oil and palm stearin blends before interesterification were 50 °C-59 °C while after interesterification, the SMP value was in the range of 44 °C-53 °C.

2.4.6 Hardness Index (HI)

Texture is a hardness measure of a product at a specified temperature. The consistency variation in a temperature range is defined as plasticity (Erickson, 1995). Texture appears as a very important parameter for margarine since these products should have good spread properties under refrigeration and do not settle oils when left under room temperatures (deMan *et al.*, 1991). Hardness of both liquid and solid fractions obtained with the constant speed penetrometer directly reflected trends in their triglyceride composition and solid fat content. According to List *et al.* (1995), texture and spreadability depends on two major factors which are SFC and process condition during cooling and crystallization of emulsion. The yield value parameter was described by (Haighton, 1959), and is considered as an independent parameter. The yield value is used to compare the several hardness index, obtained by penetrometers and other rheological instruments.

CHAPTER 3

METHODOLOGY

3.1 Materials

Soybean oil was purchased from local supermarket. Palm stearin (SMP 47.7 ºC and IV 39.52) was obtained from the Sime Darby Jomalina Sdn. Bhd., Klang, Selangor. All chemical reagents used were either analytical or HPLC grade.

3.2 Blends Preparation

The palm stearin was melted at 60 ºC in an oven prior to use. Palm stearin was blended with soybean oil in varying proportions as indicated in Table 3.1.

Table 3.1 Mixing ratio (%wt/wt) for palm stearin and soybean oil

Ratio (%wt/wt)	
Palm stearin	Soybean oil
0	100
30	70
50	50
70	30
100	0

3.3 Overall Experimental Design

Figure 3.1 illustrated the overall experimental design of this study.

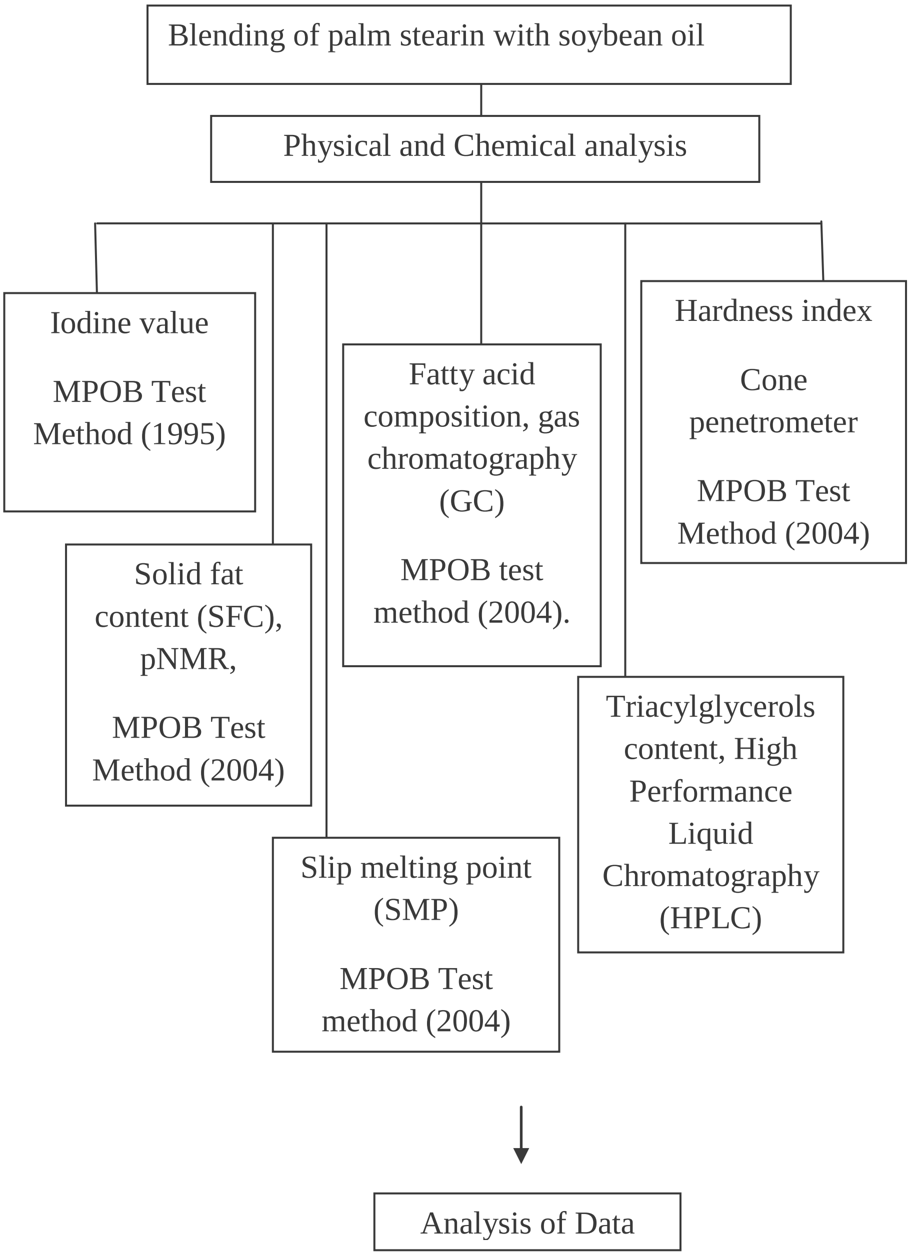

Figure 3.1 Overall Experimental Design

3.4 Methods

3.4.1 Determination of fatty acid composition

Fatty acid composition was determined according to MPOB Test Method (2004). The sample was melted at 60 ºC then it was homogenised thoroughly. Hundred (100) µl aliquot of the test sample was thoroughly mixed by dissolved 0.05 g of sample into 10 ml of petroleum ether, then 0.5 mL of 1M sodium methoxide was added to prepare fatty acid methyl esters (FAMEs). Next, the mixture was stirred vigorously for 30 seconds and was stored for 20 min to form a bilayer. The clear upper layer containing the FAMEs was pipetted off and was injected into a gas chromatograph by using an external standard method, the MPOB Test Method (2004). The oven temperature was set at 150 ºC, and held for 10 min at 250 ºC. The fatty acid components were identified based on the elution order of the reference standard mixture of the fatty acids.

The Agilent 7890N gas chromatograph (GC) fitted with DB-WAX column and flame ionization detector was used for this analysis. The sample was injected into a capillary column and was analysed. The oven temperature was programmed at 250 ºC and the helium carrier gas flow rate will be set at 4.0 ºC/min, column internal diameter between 0.2 mm to 0.8 mm and the length 25 m. The result obtain was calculated quantitatively.

3.4.2 Determination of triacylglycerol profile

The samples were dissolved in chloroform at concentration of 5% (v/v). The sample was filtered through a 0.2 µm nylon membrane. Two µm of the sample was injected. Reversed-phase High-Performance Liquid Chromatography (HPLC) was used. The eluent acetone/acetonitrile (75:25% v/v) were used at a flow rate of 1.0 ml/min and a refractive index detector was used. Identification of triacylglycerols was done by comparison of retention time with those of commercial triacylglycerols standards.

3.4.3 Determination of iodine value

The AOAC Official Method 993.20 (Cyclohexane method) was used to determine the iodine values. The sample was weighed to the nearest 0.001 g into a dry and clean reagent bottle or conical flask. The weight of the sample used was varies according to its expected iodine value as indicated in Table 3.2.

Table 3.2: The amount of sample weight according to expected iodine value

Iodine value expected	Weight to be taken for use
0-5	3.0
5-20	1.0
20-60	0.34
60-80	0.25
80-130	0.15
>130	0.10

Source: Suzanne (2003)

Twenty mL of cyclohexane was added to dissolve the oil. The sample was then swirled to make sure the sample was in liquid form. Next, by using a pipette, 25 mL of Wijs solution was added into the flask contained the sample. The flask was closed with stopper and wrapped with aluminium foil. The sample was kept undisturbed in the dark for 2 hours at 25 ºC$\underline{+}$5 ºC. The flask was removed from the dark storage and 20 mL of 15% potassium iodide (KI) solution was added follow with addition of 100 mL of distilled water.

The sample was titrated with 0.1 M sodium thiosulphate solution. The titration was continued until yellow colour of the solution was almost disappeared. Then, 1 to 2 mL of starch indicator was added and the titration was continued until blue colour disappeared after vigorous shaking. The sample was then analysed in triplicate. A blank test also was carried out under the same condition.

The iodine value of the blends was calculated according to the equation below:

$$\text{Iodine value} = \frac{12.69\ \mathrm{M}\ (Vb - Vs)}{\text{Weight of sample}}$$

M - molarity of sodium thiosulphate solution used

Vb - volume in mL of sodium thiosulphate solution used for blank test

Vs - volume in mL of sodium thiosulphate solution used for sample test

3.4.4 Determination solid fat content

The solid fat content of the blends was determined using the Bruker Minispec PC 120 pulsed Nuclear Magnetic Resonance (pNMR) (Karlsruhe, Germany). The parallel Malaysia Palm Oil Board (MPOB) Test Method (2004) was used for this analysis. The sample tube used was 180mm x 10mm. The sample in the pNMR tubes were melted at 70 ºC in the water bath and the temperature was maintained at 70 ºC for 30 minutes.

Next, the sample tubes were transferred into 0 ºC water bath and were kept at 90 minutes. Then, the sample tubes were transferred again in sequence at the same time interval in batches. The water bath was set to the first measuring temperature. The chilled sample tubes tube with two references tubes was transferred together to pre-equilibrated thermostatic water bath in sequence at fixed intervals accurate to 0.1 ºC. Solid fat content was measured in the temperature range of 5 ºC to 55 ºC.

3.4.5 Determination of slip melting point

Slip melting point was determined according to MPOB Test Method (2004). For sample preparation, the samples were melted in an oven set at 60 ºC to avoid any crystallisation of sample. Three clean capillary tubes were dipped into the completely liquid sample so that the samples were obtained in the capillary tubes. Then the fat columns were chilled at once by hold and rolled the ends of the tubes contained the sample pressed against a piece of ice until the fats had solidified. The open end of the tubes was not allowed to touch the ice. The tubes were placed in a test tube which was held in a beaker that has been equilibrated at 10 ºC±1 ºC in a thermostat water bath. The beaker was transferred to the water bath and hold for 16 hours at 10 ºC±1 ºC.

Then the capillary tube was removed from the test tube and attached with a rubber band to a thermometer, the lower ends of the tube was the same level with the bottom of the mercury bulb of the thermometer. The thermometer was suspended in the beaker with 400 mL of boil distilled water. The bottom of thermometer was immersed in the water. The water was stirred to increase the temperature at a rate 1 ºC per minute and the temperature was slowed down to 0.5 ºC per minute when the slip melting point had reached. The heating was continued until the fat column rose in each tube. The temperature was recorded when the column of fat rise in the tube.

3.4.6 Hardness Index

Fifty grams of the samples was filled in a propylene container (internal diameter = 12 cm, height = 5cm), tempered at 5 ºC overnight. Then the sample was subjected to penetration by using cone penetrometer. The angle of the cone used was 20°, and the time was 5 seconds at temperature 5 ºC. The hardness index (HI) was calculated according to the equation below.

$$\text{Hardness Index (HI)} = \frac{\text{Weight of the cone (g)}}{\text{Depth of penetration (mm)}} \times 0.1$$

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Fatty Acid Composition (FAC)

The percentage of the fatty acid composition in the palm stearin, soybean oil and three binary blends was determined using gas chromatography. Table 4.1 shows the major fatty acid in palm stearin were palmitic acid (60.00%) followed by oleic acid (27.87%). This finding was in agreement with Gunstone (2011) who reported the palmitic acid content of the palm stearin varies from 47% to 74%, while oleic acid ranges from 15% to 37%.

Table 4.1 Fatty acid composition for palm stearin, soybean oil and PS:SBO binary blends.

PS:SBO	C12:0	C14:0	C16:0	C16:1	C18:0	C18:1	C18:2	C18:3
100:0	0.26	1.71	60.00	0.33	4.11	27.87	5.44	0.33
70:30	0.18	1.1	44.55	0.26	4.33	27.7	19.9	1.96
50:50	-	1.2	40.22	0.34	4.29	24.3	26.81	2.89
30:70	-	-	28.06	-	4.47	24.29	38.8	4.31
0:100	-	-	13.21	-	4.84	23.45	52.52	5.98

Note: C12:0 = Lauric acid
C14:0 = Myristic acid
C16:0 = Palmitic acid
C16:1 = Palmitoleate
C18:0 = Stearic acid
C18:1 = Oleic acid
C18:2 = Linoleic acid
C18:3 = Linolenic acid

According to Noh (2009), the increasing of palmitic acid would decrease the liquidity of the oil, which would be undesireable, especially in cooking oil and decrease the IV value of the oil. In order to increase the liquidity and unsaturation of the oil, Noh (2009) suggested increasing the percentage of oleic acid and decreasing the palmitic acid. The major fatty acids in SBO were linoleic acid (52.52%), oleic acid (23.45%) and palmitic acid (13.21%). The percentage of linoleic acid in SBO was the highest compared to other blends. This finding was in agreement with Norhayati (2009) who stated that linoleic (C18:2) acid was the predominant fatty acid in SBO, followed by oleic (18:1), palmitic (C16:0) and linolenic (C18:3) acids in a decreasing order.

Blending of PS with 30, 50 and 70% SBO altered the fatty acid composition of the original oil (PS) that caused the decreasing in percentage of palmitic acid from 60% to 28.06% and oleic acid from 27.7% to 24.3%. On the other hand, the linoleic acid increased from 5.44% to 38.8% and linolenic acid from 0.3% to 4.3%. The fatty acid composition in PS, SBO and PS:SBO binary blends plays a major role in affecting the IV, SFC, SMP, and HI.

4.2 Triacylglycerols profile (TAG)

Table 4.2 shows the triacylglycerol composition of palm stearin (PS), soybean oil (SBO) as well as their blends. The main triacylglycerols in PS which were POP (54.33%) and PPP (31.91%), where P indicates palmitic acid and O, oleic acid. According to Siti Hazirah *et al.* (2013), the main TAG of PS were POP, PPP and POO and with noticeable amounts of POS, PLO and OLO.

Table 4.2 Triacylglycerols composition for palm stearin, soybean oil and PS:SBO binary blends.

PS:SBO	LLL	LnLnO	PLL	OLO	PLP	POO	POP	PPP	POS
100:0	-	-	-	-	-	12.48	54.33	31.91	1.28
70:30	6.66	6.57	6.63	6.96	8.97	11.29	22.87	30.04	-
50:50	16.76	14.47	6.84	10.47	10.61	14.85	20.57	5.43	-
30:70	31.97	23.38	17.11	17.11	0.98	1.39	6.74	1.52	-
0:100	31.87	22.04	28.4	17.69	-	-	-	-	-

Note : Ln = linolenic acid
L = linoleic acid
P = palmitic acid
O = oleic acid
S = stearic acid

The major TAGs of SBO were LLL (31.87%), followed with PLL (28.40%), LnLnO (22.04%) and OLO (17.69%), where L represents linoleic acid, Ln is linolenic acid, O represents oleic acid. According to O'Brien (2009), the predominant TAG in SBO were triunsaturated and followed with monosaturated. The predominant TAGs for SBO were LLL and OLL, with noticeable amount of

OLO, PLO, and PLL (Siti Hazirah *et al.*, 2013). The addition of SBO into the PS had shows the presence of TAGs from SBO which were LLL, LnLnO, PLL, OLO, PLP as compared to the original PS that do not have these type of TAGs.

It was observed the SMP and SFC of the three binary blends of PS:SBO decrease due to the decreased proportion of the high melting TAGs as shown in Table 4.4 and Figure 4.1, respectively. The reduction in the SFC values could be most likely attributed to the decrease in the proportion of the TAGs which are PPP and POP simultaneously with the presence of several types of low melting TAGs such as LLL, PLL and LnLnO.

4.3 Iodine Value (IV)

Table 4.3 shows the IV of the PS and SBO were significantly different ($p<0.05$) with the IV of 37.21 and 129.97, respectively. Soybean oil has higher IV than PS due to the high degree of unsaturation, thus become more vulnerable to oxidation (Bazlul *et al.*, 2010).

Table 4.3 Iodine Value (IV) of palm stearin, soybean oil and PS:SBO binary blends

PS:SBO	Iodine Value (IV)
0:100	37.21 ± 0.06^{e}
70:30	51.08 ± 0.56^{d}
50:50	72.47 ± 1.06^{c}
30:70	93.05 ± 1.12^{b}
0:100	129.97 ± 2.66^{a}

Note: Means within column followed by different superscript a, b, c, d, or e are significantly different at $p<0.05$.

As predicted, the iodine values for the blends increased significantly ($p<0.05$) with the increasing amount of the SBO in the blends content because of the increasing amount of unsaturated fatty acid in SBO which are linoleic acid and oleic acid as indicated in Table 4.1. According to Zarinah *et al.* (2012), high IV oil contains a greater number of double bonds than low IV oil. The lower IV obtained from PS indicated that PS has greater amount of saturated fatty acid which is palmitic acid (60%).

4.4 Solid Fat Content (SFC)

The solid fat content (SFC) is a major concern in food applications, and is generally considered as the fingerprint of oils and fats to characterise the physical and sensory properties of foods such as spreadability, texture, firmness and mouthfeel. Knowing the SFC would be useful for formulating new products and also for authenticating the oils and fats (Aszmil *et al.*, 2007). The SFC for PS, SBO and binary blends at temperature ranging from 5 ºC to 55 ºC are shown in Figure 4.1.

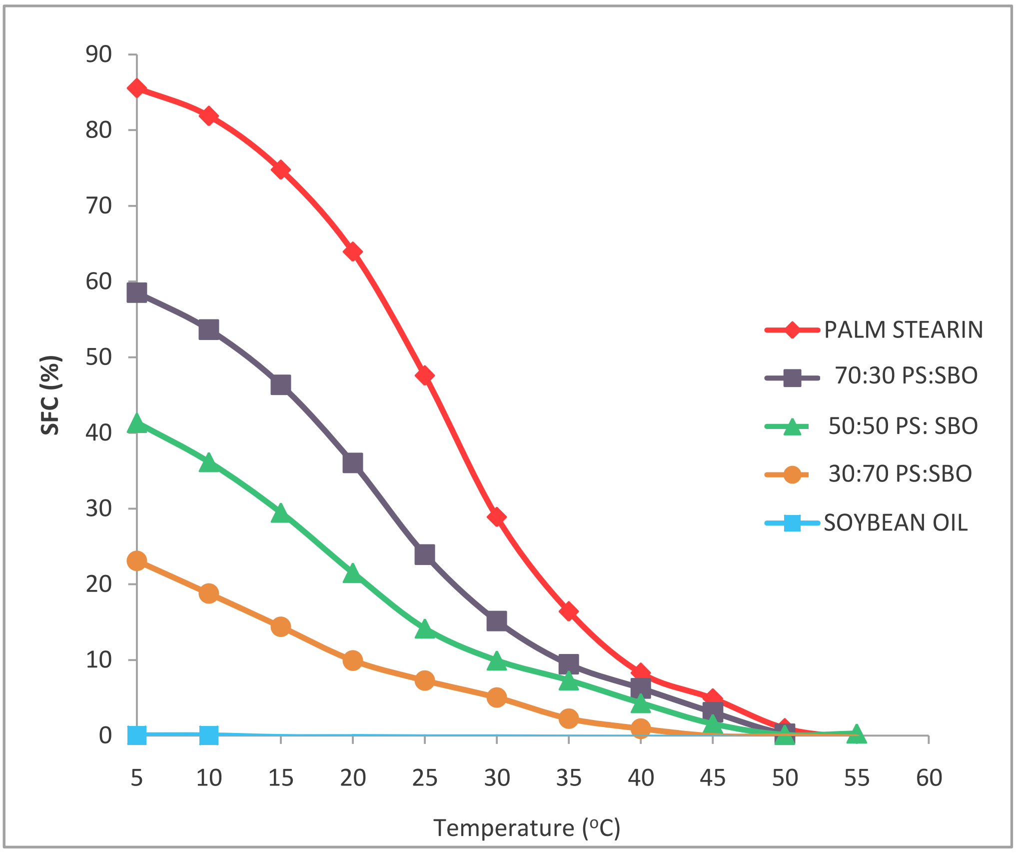

Figure 4.1: Percentage SFC versus temperature (ºC) for Palm Stearin, Soybean oil and PS:SBO binary blends.

The PS had the highest SFC values at all measured temperatures. This is due to the high amount of saturated fatty acids. In contrast, SBO had the lowest SFC as the SBO was almost completely in liquid form at all measured temperatures. This finding agreed with Siti Hazirah *et al.* (2013) that reported SBO is readily liquid at measured temperatures ranging from 0 ºC to 50 ºC as it is very rich in monounsaturated (oleic) and polyunsaturated (linoleic) fatty acids.

As expected, the SFC value decreased with the increasing addition of SBO in the blends. Figure 4.1 illustrates that the PS, SBO and binary blends have a similar SFC profiles. At 5 ºC, all the blends had the highest SFC ranging from 23.08% to 58.53% which decreased until melting completely at the temperature 50 ºC for 70:30 PS:SBO and 50:50 PS:SBO while 30:70 PS:SBO were completely melted at temperature of 45 ºC.

At low temperatures of 4 ºC to 10 ºC, SFC gives an indication of the fat's spreadability at refrigeration temperatures, which should not be greater than 32% at 10 ºC (Nor Lida and Ali, 1996; Wassell and Young, 2007). The 30:70 PS:SBO had SFC of 18.77% which meet the requirement. The other blends which were 70:30 PS:SBO and 50:50 PS:SBO have SFC values exceeding 32% at temperature of 10 ºC, which were 53.63% and 36.16%, respectively. Hence, these two blends might not have good fat's spreadability for use in products such as soft margarine.

On the other hand, the SFC values between 20 ºC and 22 ºC are related to the product's stability and its resistance to oiling-out must not be less than 10% (Norlida and Ali, 1996; Wassell and Young, 2007). Based on the results obtained, the 70:30 PS:SBO and 50:50 PS:SBO blends had SFC values above 10% at 20 ºC which were 36.03% and 21.49%, respectively. Therefore these two blends had product's stability and resistance to oiling-out.

According to Karabulut *et al.* (2004), the lower SFC value of 1% at 37 ºC are regarded as offering the good melting behaviour characteristic at body temperature. Only blend 30:70 PS:SBO had value from 2.2% to 0.9% at temperature between 35 ºC to 40 ºC that could resulting good melting behaviour. The SFC between 35 °C and 37 °C determines the thickness and flavour release properties of reduced fat spread (RFS) in the mouth (Charteris and Keogh, 1991; Krawczyk *et al.,* 1996).

Identification of application for new fats is guided initially by correlating their physical properties with the properties of commercial fats for specific application.

The SFC profile, particularly has important implications for the determination of characteristics of margarines and shortenings at various temperatures (Wassell & Young, 2007). In some systems that contain fats, it is desirable to have a high solid content to promote an adequate crystalline structure in foods with a high content of fats, like margarines and chocolate under room temperature and low solid content at high temperatures, to promote an easy melting in the mouth (Woerfel,1995).

4.5 Slip Melting Point (SMP)

Slip melting point is widely used to characterise the melting and solidification properties of oils and fats. It changes with the chain length of fatty acids, unsaturation ratios, *trans* fatty acid content and the position of the fatty acids in the glycerol backbone (Karabulut *et al*., 2004). Table 4.4 shows a substantial decrease in the melting points when the PS was blended with SBO at different ratios. The PS has significantly ($p<0.05$) highest melting point of 49.2ºC due to presence of saturated fatty acids contain in the PS such as palmitic acid (60%) as shown in Table 4.1. The 30:70 PS:SBO blend has the lowest melting point of 28.03 ºC.

Table 4.4 Slip Melting Point (SMP) of palm stearin, soybean oil and PS:SBO binary blends.

PS:SBO	Slip Melting point (SMP)
100:0	49.16 ± 0.05^{a}
70:30	43.20 ± 0.20^{b}
50:50	35.07 ± 0.12^{c}
30:70	28.03 ± 0.05^{d}
0:100	-

Note: Means within column followed by different superscript a, b, c, or d are significantly different at $p<0.05$.

This SMP result was in agreement with SFC and FAC findings which indicated PS contains long chain fatty acid such palmitic acid (C16:0) and oleic acid (C18:1) resulting in low IV and high SFC throughout the temperatures measured. According to Lai *et al.* (1999), PS with SMP range between 44 ºC and 56 ºC is a very useful source of fully natural hard component for products such as shortening, margarine and other edible fats but at this level PS may not be able to impart the required plasticity and body to the end product.

In contrast, the SMP of SBO cannot be detected since it was in liquid form as SBO contains high unsaturated fatty acid such as linoleic acid (C18:2) and oleic acid (C18:1) resulting in higher IV and lower SFC. From the result obtained, blending of PS with 30%, 50% and 70% SBO had decreased the SMP from 49.2 ºC to 35.1 ºC and 28 ºC, respectively. The decrease in SMP indicates that blending of PS with a liquid oil of lower saturated fatty acid (SBO) was successful in yielding blends with lower melting point compared to the original oil (PS).

As reported by Zarinah *et al*. (2012), during slip melting point measurements, the temperature of the sample is raised and the solid fat will melts. The progressive reduction of crystalline matter means that, at a certain temperature, the fat crystal network lacks sufficient cohesion to hold onto its matrix and become sufficiently soft until it suddenly rises. According to Karabulut *et al*. (2003), SMP is defined as the temperature at which the fats and oils have 4% solid fat.

Karabulut *et al*. (2003) also reported that at body temperature of 37 ºC, the fats and oils should melt almost completely in one’s mouth. The blends 50:50 PS:SBO and 30:70 PS:SBO had melting point below body temperature which were 35.1 ºC and 28.0 ºC, respectively, which means these two type of blends melts below the body temperature. At 35 °C to 40 °C the SFC for blends 30:70 PS:SBO was less than 5%, while blend 50:50 PS:SBO was less than 8% solid fat. The SFC values of less than 1% could result in good organoleptic as the fats melts in the mouth at the body temperature of 37.5 °C. Percentage of fat at the melting point of 30 ºC to 40 °C must not be higher than 1% to avoid a poor mouthfeel due to the 'tailing effect', which is an undesirable waxy aftertaste from incomplete melting of fat in the mouth. The SMP and SFC are related with each other (Alina *et al.*, 2009).

4.6 Hardness Index (HI)

The texture of PS, SBO and their binary blends were evaluated to measure the consistency of the fats. The HI calculated indicates the hardness of the fats at which a product is suitable for a particular use. Table 4.5 shows that PS had significantly the highest ($p<0.05$) HI of 36.20. This agreed with Jeyarani and Yella (2005) who reported that fats with high SFC had high HI. The lowest HI was SBO with 2.56.

Table 4.5 Hardness index for palm stearin, soybean oil and PS:SBO binary blends.

PS:SBO	Hardness Index (HI)
100:0	36.20 ± 4.55^{a}
70:30	11.87 ± 0.66^{b}
50:50	5.12 ± 0.34^{c}
30:70	3.26 ± 0.04^{c}
0:100	2.56 ± 0.03^{c}

Note: Means within column followed by different superscript a, b, or c are significantly different at $p<0.05$

Blending of PS with 30% to 70% SBO had decreased the HI from 36.2 to 3.26 as the addition of the unsaturated fatty acid in SBO contributed to soft texture. This results correlate well with Figure 4.1 as the SFC of the blends decreased with the increasing amount of the SBO. Apart from that, the IV and SMP obtained were related with the HI. The HI was reduced as the SMP decreased due to the high degree of unsaturated of fatty acid in the blends as shown in Table 4.1 and Table 4.4, respectively.

Results indicated that blending of PS with SBO had modified the physical properties of the binary blends in producing softer texture (low HI) compared to the original oil of PS (high HI), hence make it one of the important properties in determining the right blends for producing a good margarine spreadability.

CHAPTER 5

CONCLUSION AND RECOMMENDATIONS

In conclusion, blending palm stearin (PS) and soybean oil (SBO) have modified the physicochemical properties of the binary blends. The addition of SBO into the PS also caused the major changes in the fatty acid composition of the blends. The PS had the highest palmitic acid content which is 60%, but decreased in composition when the SBO was added into the blends caused the increasing amount of linoleic acid thus modified the characteristics of the blends into different physicochemical properties.

From the result obtained, the increasing of the amount unsaturated oils which is SBO into the PS blends had caused the increasing of the IV, whereas the solid fat content (SFC), slip melting point (SMP) and the hardness index (HI) decreased. The blends 50:50 PS:SBO have a product stability and resistance to oiling out, with good melting point below body temperature suitable for product such as margarine that must be melted in the mouth with minimum waxiness in order to have a good oral-melt.

Further study should be carried out an analysis of polymorphism, microstructure, and thermal properties of PS:SBO to obtain a better physicochemical properties of the blends and to achieve better blend formulation for food application such as margarine, soft margarine and shortening.

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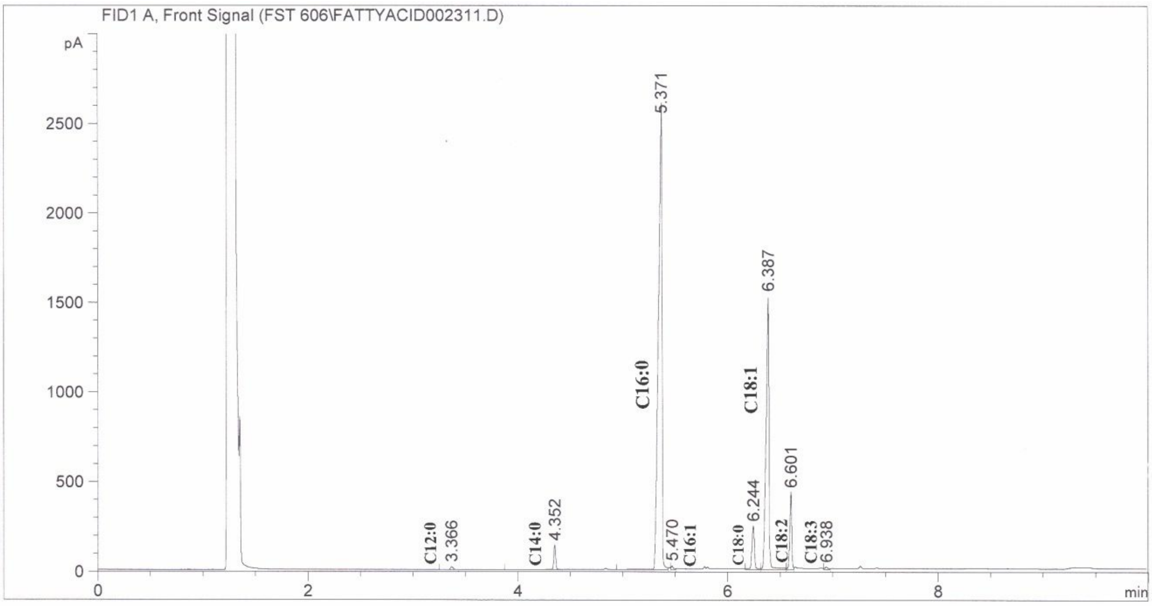
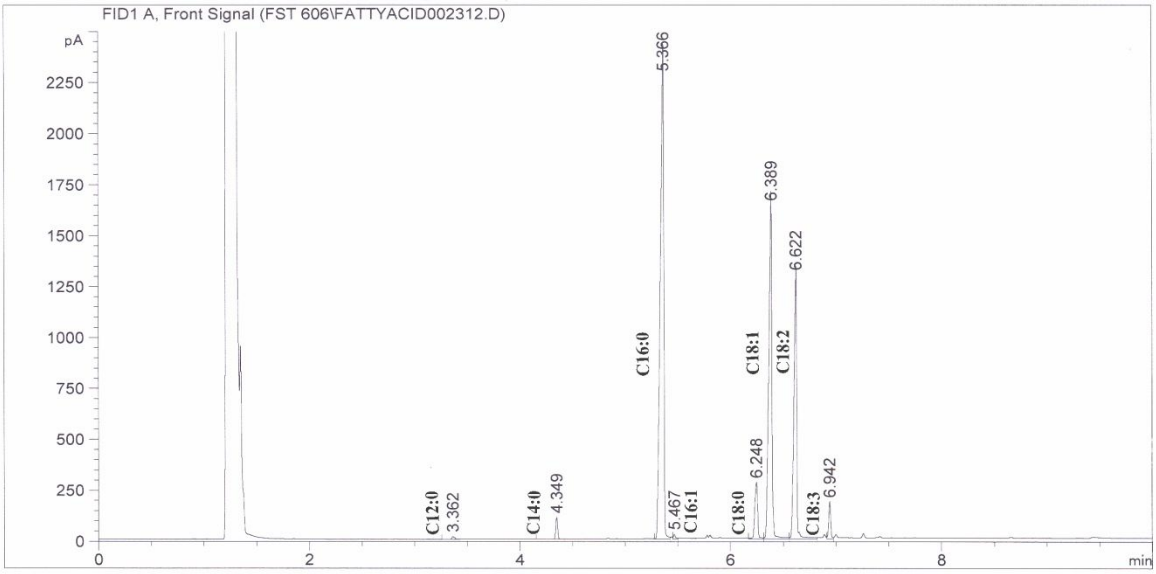
APPENDIX A

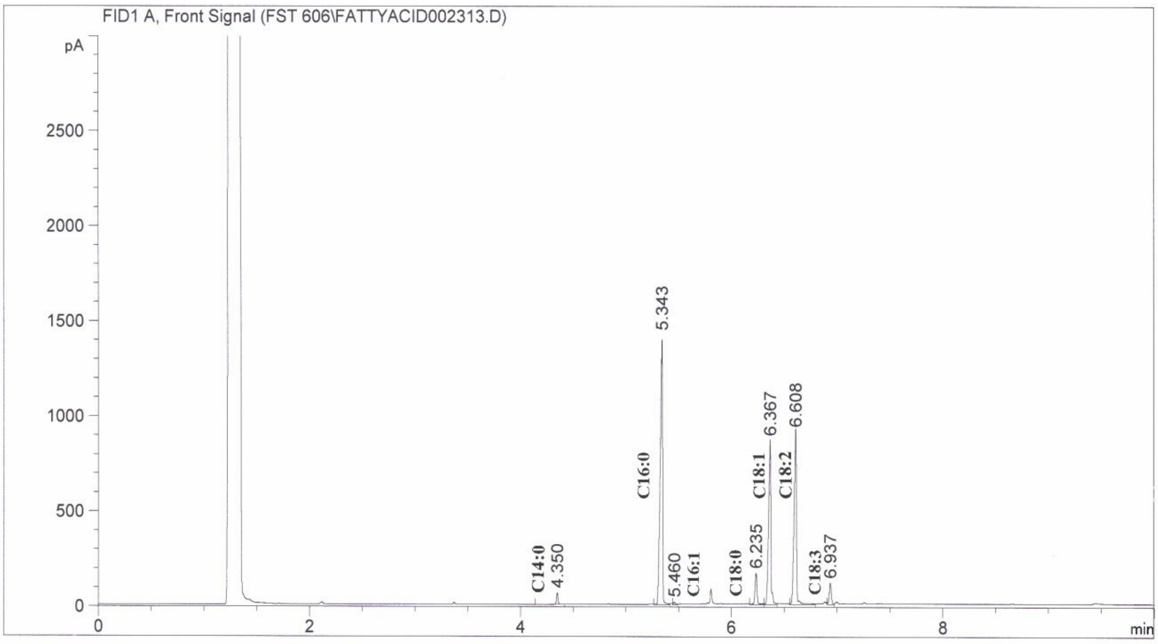
Fatty acid compositions



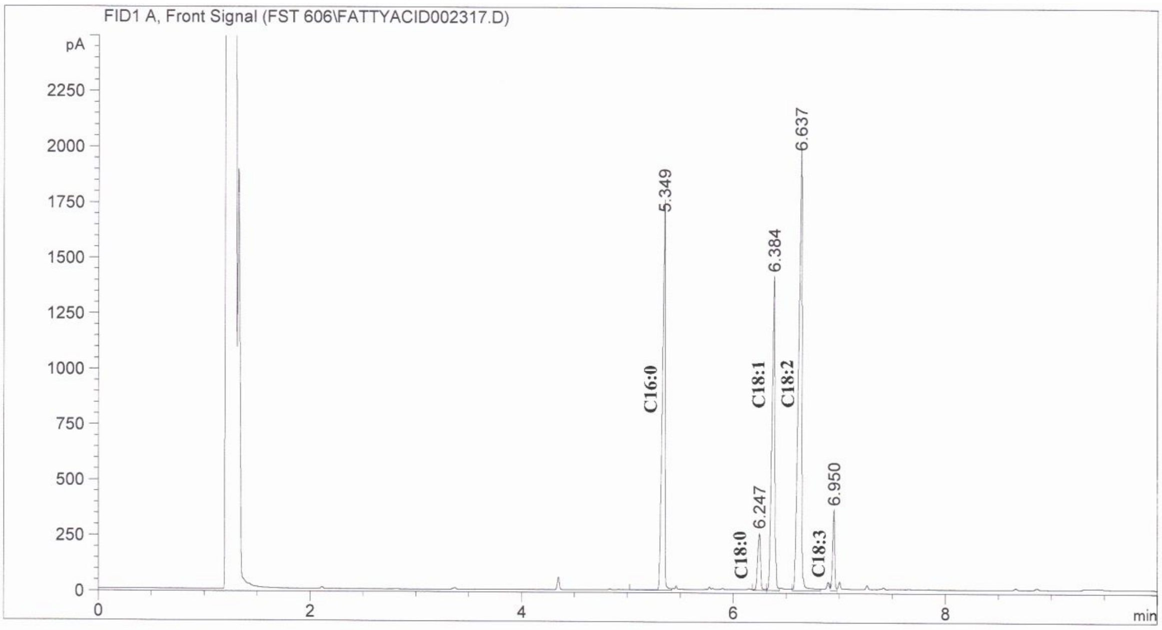
Chromatogram of 100:0 PS:SBO



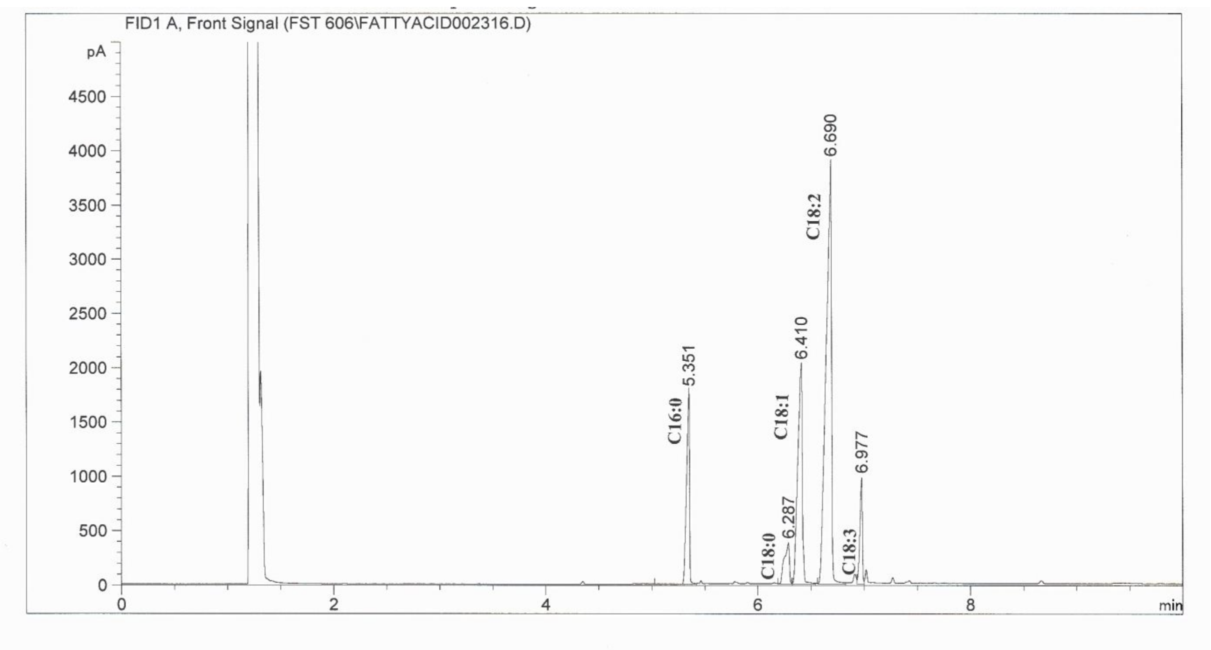
Chromatogram of 70:30 PS:SBO



Chromatogram of 50:50 PS:SBO



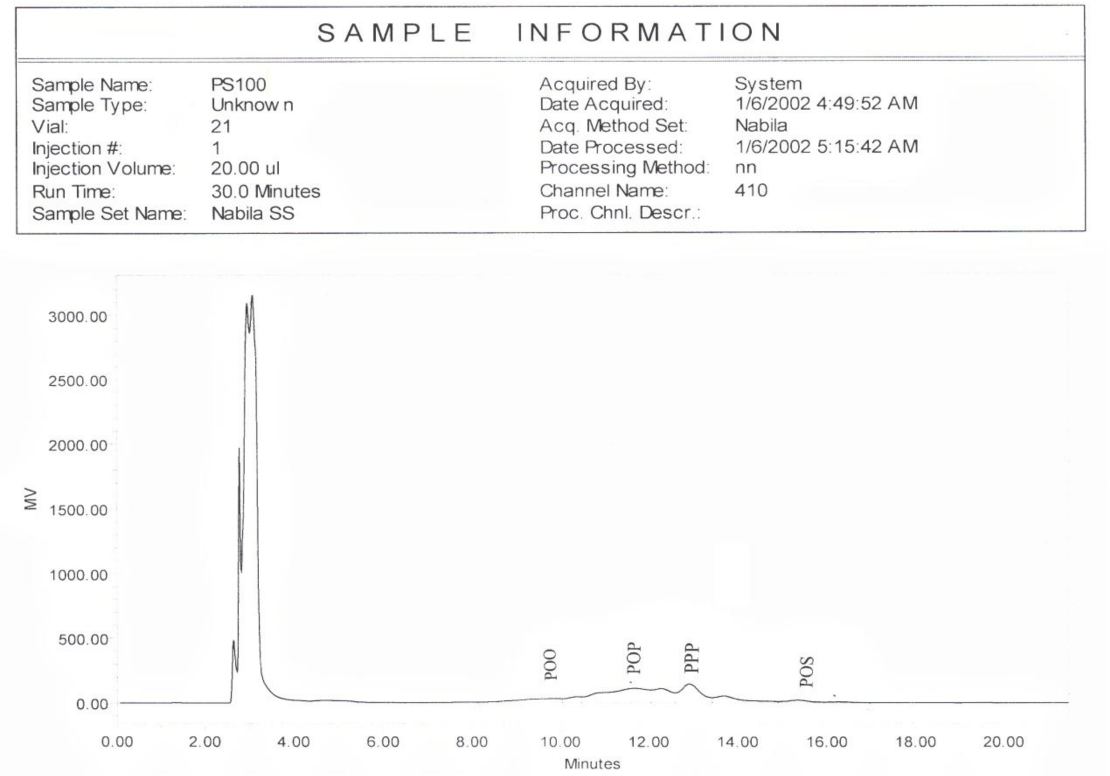
Chromatogram of 30:70 PS:SBO



Chromatogram of 0:100 PS:SBO

APPENDIX B

Triacylglycerol profile

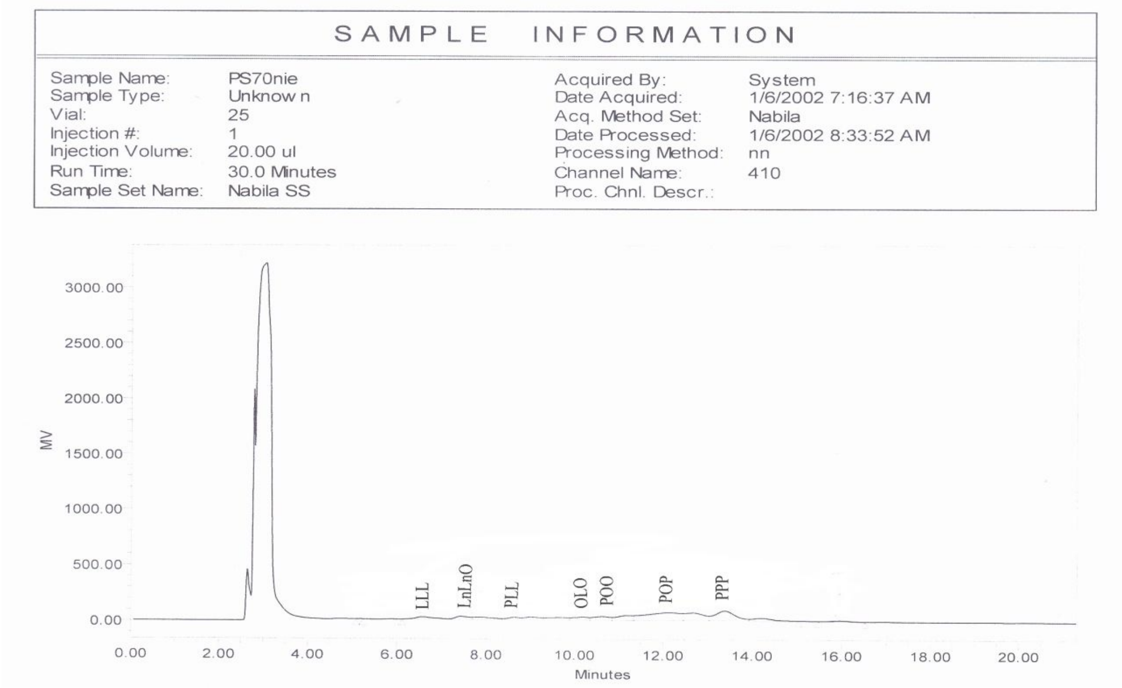


SAMPLE INFORMATION

Sample Name: PS100
Sample Type: Unknow n
Vial: 21
Injection #: 1
Injection Volume: 20.00 ul
Run Time: 30.0 Minutes
Sample Set Name: Nabila SS

Acquired By: System
Date Acquired: 1/6/2002 4:49:52 AM
Acq. Method Set: Nabila
Date Processed: 1/6/2002 5:15:42 AM
Processing Method: nn
Channel Name: 410
Proc. Chnl. Descr.:

Chromatogram of 100:0 PS:SBO

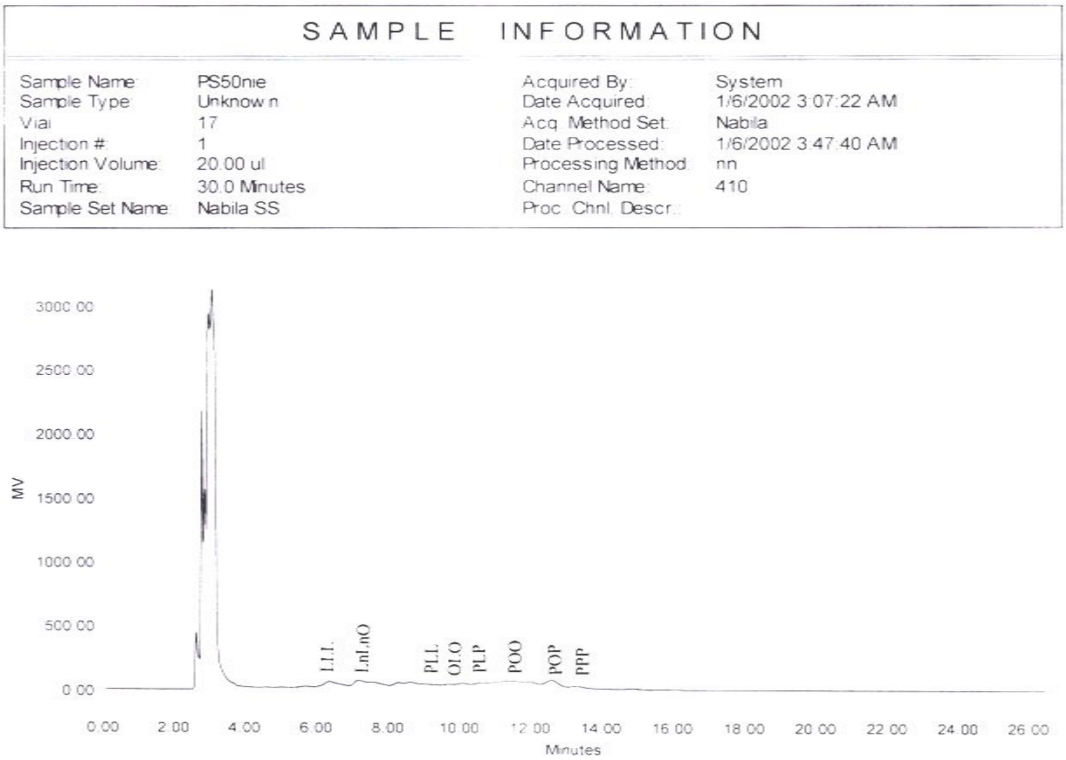


SAMPLE INFORMATION

Sample Name: PS70nie
Sample Type: Unknow n
Vial: 25
Injection #: 1
Injection Volume: 20.00 ul
Run Time: 30.0 Minutes
Sample Set Name: Nabila SS

Acquired By: System
Date Acquired: 1/6/2002 7:16:37 AM
Acq. Method Set: Nabila
Date Processed: 1/6/2002 8:33:52 AM
Processing Method: nn
Channel Name: 410
Proc. Chnl. Descr.:

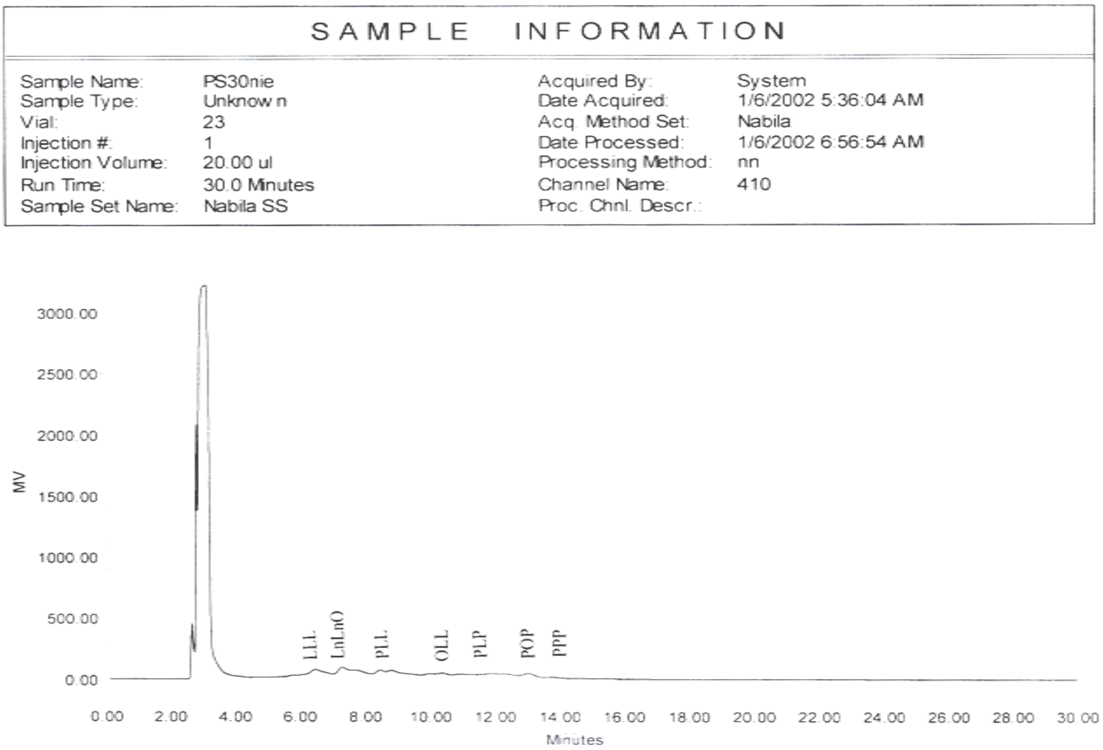
Chromatogram of 70:30 PS:SBO



SAMPLE INFORMATION

Sample Name:	PS50nie	Acquired By:	System
Sample Type:	Unknown	Date Acquired:	1/6/2002 3:07:22 AM
Vial:	17	Acq. Method Set:	Nabila
Injection #:	1	Date Processed:	1/6/2002 3:47:40 AM
Injection Volume:	20.00 ul	Processing Method:	nn
Run Time:	30.0 Minutes	Channel Name:	410
Sample Set Name:	Nabila SS	Proc. Chnl. Descr.:	

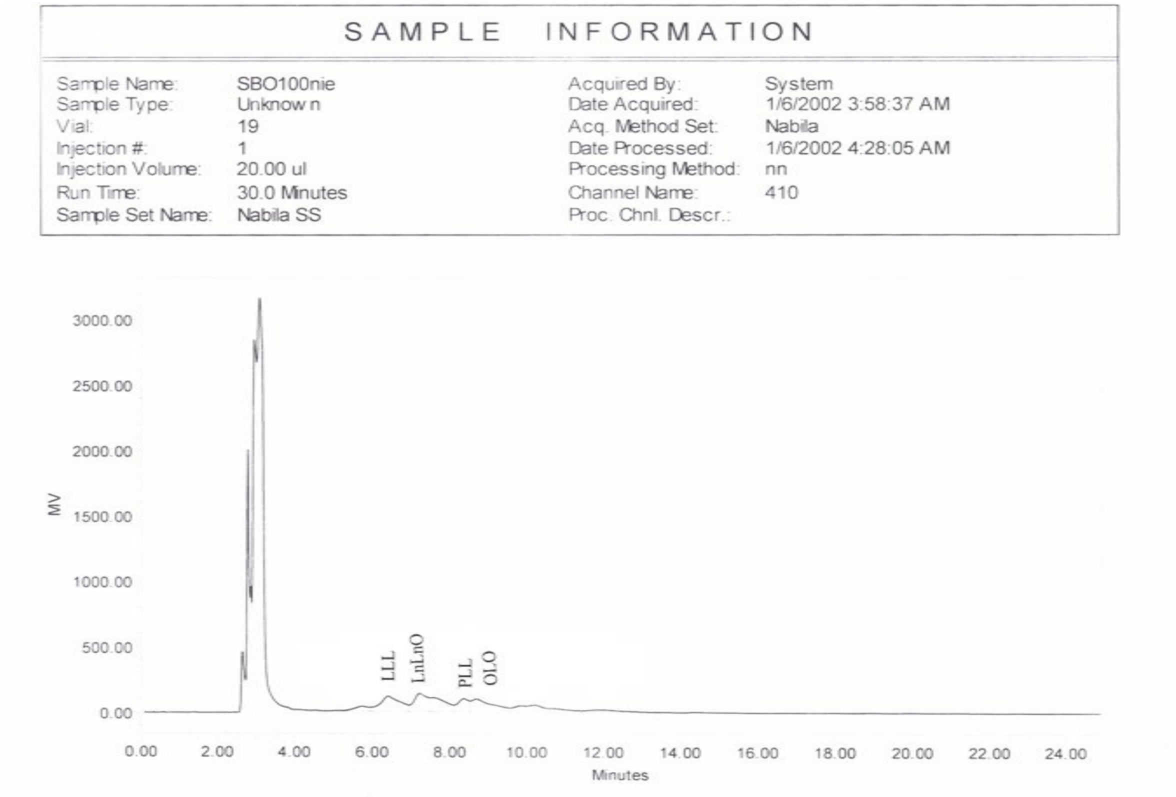
Chromatogram of 50:50 PS:SBO



SAMPLE INFORMATION

Sample Name:	PS30nie	Acquired By:	System
Sample Type:	Unknown	Date Acquired:	1/6/2002 5:36:04 AM
Vial:	23	Acq. Method Set:	Nabila
Injection #:	1	Date Processed:	1/6/2002 6:56:54 AM
Injection Volume:	20.00 ul	Processing Method:	nn
Run Time:	30.0 Minutes	Channel Name:	410
Sample Set Name:	Nabila SS	Proc. Chnl. Descr.:	

Chromatogram of 30:70 PS:SBO



SAMPLE INFORMATION

Sample Name:	SBO100nie	Acquired By:	System
Sample Type:	Unknow n	Date Acquired:	1/6/2002 3:58:37 AM
Vial:	19	Acq. Method Set:	Nabila
Injection #:	1	Date Processed:	1/6/2002 4:28:05 AM
Injection Volume:	20.00 ul	Processing Method:	nn
Run Time:	30.0 Minutes	Channel Name:	410
Sample Set Name:	Nabila SS	Proc. Chnl. Descr.:	

Chromatogram of 0:100 PS:SBO

APPENDIX C

Raw Data

Iodine Value (IV)

Blank Titration: 29.4 mL

	Titration Readings					
PS:SBO	**Volume 1 (mL)**	**Weight 1 (g)**	**Volume 2 (mL)**	**Weight 2 (g)**	**Volume 3 (mL)**	**Weight 3 (g)**
100:0	25.00	0.15	24.70	0.16	25.10	0.15
70:30	25.40	0.10	24.10	0.13	25.00	0.11
50:50	21.90	0.13	22.10	0.13	22.50	0.12
30:70	19.80	0.13	19.80	0.13	20.00	0.13
0:100	18.00	0.11	17.40	0.12	18.00	0.11

Statistical Analysis for Iodine Value (IV)

VAR00001

	sample	N	Subset for alpha = .05 1	2	3	4	5
Duncan[a]	PS100:SBO:0	3	37.2133				
	PS30:SBO70	3		51.0833			
	PS50:SBO50	3			72.4793		
	PS70:SBO30	3				93.0593	
	PS0:SBO100	3					129.9767
	Sig.		1.000	1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Descriptives

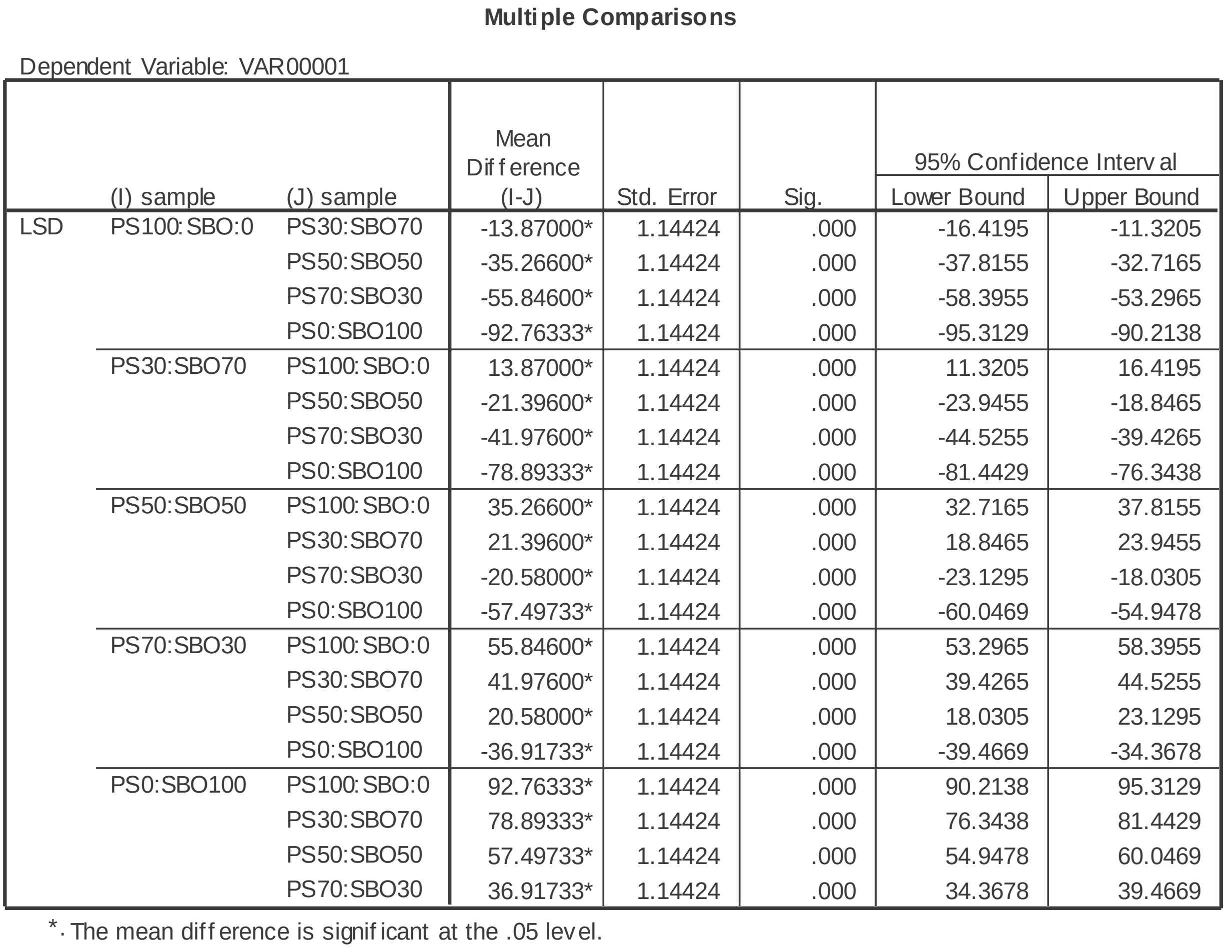
VAR00001

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean Lower Bound	Upper Bound	Minimum	Maximum
PS100:SBO:0	3	37.2133	.06962	.04019	37.0404	37.3863	37.14	37.28
PS30:SBO70	3	51.0833	.56003	.32333	49.6921	52.4745	50.76	51.73
PS50:SBO50	3	72.4793	1.06380	.61419	69.8367	75.1220	71.26	73.21
PS70:SBO30	3	93.0593	1.12699	.65067	90.2597	95.8589	91.76	93.71
PS0:SBO100	3	129.9767	2.66447	1.53833	123.3578	136.5956	126.90	131.51
Total	15	76.7624	33.84560	8.73890	58.0193	95.5055	37.14	131.51

ANOVA

VAR00001

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	16017.708	4	4004.427	2038.984	.000
Within Groups	19.639	10	1.964		
Total	16037.347	14			



Multiple Comparisons

Dependent Variable: VAR00001

	(I) sample	(J) sample	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
						Lower Bound	Upper Bound
LSD	PS100:SBO:0	PS30:SBO70	-13.87000*	1.14424	.000	-16.4195	-11.3205
		PS50:SBO50	-35.26600*	1.14424	.000	-37.8155	-32.7165
		PS70:SBO30	-55.84600*	1.14424	.000	-58.3955	-53.2965
		PS0:SBO100	-92.76333*	1.14424	.000	-95.3129	-90.2138
	PS30:SBO70	PS100:SBO:0	13.87000*	1.14424	.000	11.3205	16.4195
		PS50:SBO50	-21.39600*	1.14424	.000	-23.9455	-18.8465
		PS70:SBO30	-41.97600*	1.14424	.000	-44.5255	-39.4265
		PS0:SBO100	-78.89333*	1.14424	.000	-81.4429	-76.3438
	PS50:SBO50	PS100:SBO:0	35.26600*	1.14424	.000	32.7165	37.8155
		PS30:SBO70	21.39600*	1.14424	.000	18.8465	23.9455
		PS70:SBO30	-20.58000*	1.14424	.000	-23.1295	-18.0305
		PS0:SBO100	-57.49733*	1.14424	.000	-60.0469	-54.9478
	PS70:SBO30	PS100:SBO:0	55.84600*	1.14424	.000	53.2965	58.3955
		PS30:SBO70	41.97600*	1.14424	.000	39.4265	44.5255
		PS50:SBO50	20.58000*	1.14424	.000	18.0305	23.1295
		PS0:SBO100	-36.91733*	1.14424	.000	-39.4669	-34.3678
	PS0:SBO100	PS100:SBO:0	92.76333*	1.14424	.000	90.2138	95.3129
		PS30:SBO70	78.89333*	1.14424	.000	76.3438	81.4429
		PS50:SBO50	57.49733*	1.14424	.000	54.9478	60.0469
		PS70:SBO30	36.91733*	1.14424	.000	34.3678	39.4669

*. The mean difference is significant at the .05 level.

APPENDIX D

Solid Fat Content (SFC)

Blend (PS:SBO)	Temperature (°C)										
	5	10	15	20	25	30	35	40	45	50	55
100-0	85.54	81.85	74.77	63.93	47.54	28.88	16.38	8.26	4.9	0.96	-0.04
70-30	58.53	53.63	46.36	36.03	25.58	15.14	9.42	6.23	3.08	0.23	-0.34
50-50	41.34	36.16	29.47	21.49	14.18	10.03	7.29	4.29	1.54	0.1	0.03
30-70	23.08	18.77	14.47	9.91	7.26	5.01	2.24	0.9	-0.066	-0.117	-0.24
0-100	0.03	-0.04	-0.18	-0.2	-0.23	-0.24	-0.25	-0.31	-0.33	-0.43	-0.54

APPENDIX E

Slip Melting Point (SMP)

PS:SBO	Trial 1	Trial 2	Trial 3	Mean
100:0	49.1	49.2	49.2	49.17
70:30	43.4	43.2	43.0	43.20
50:50	35.0	35.0	35.2	35.07
30:70	28.0	28.1	28.0	28.03
0:100	-	-	-	-

Statistical Analysis for Slip Melting Point (SMP)

VAR00001

			Subset for alpha = .05			
	Samples	N	1	2	3	4
Duncan[a]	PS:70SBO30	3	28.0333			
	PS50: SBO50	3		35.0667		
	PS30: SBO70	3			43.2000	
	PS100: SBO0	3				49.1667
	Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

ANOVA

VAR00001

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	770.007	3	256.669	17111.259	.000
Within Groups	.120	8	.015		
Total	770.127	11			

Descriptives

VAR00001

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
PS100:SBO0	3	49.1667	.05774	.03333	49.0232	49.3101	49.10	49.20
PS30:SBO70	3	43.2000	.20000	.11547	42.7032	43.6968	43.00	43.40
PS50:SBO50	3	35.0667	.11547	.06667	34.7798	35.3535	35.00	35.20
PS:70SBO30	3	28.0333	.05774	.03333	27.8899	28.1768	28.00	28.10
Total	12	38.8667	8.36729	2.41543	33.5503	44.1830	28.00	49.20

Multiple Comparisons

Dependent Variable: VAR00001

	(I) Samples	(J) Samples	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
						Lower Bound	Upper Bound
LSD	PS100:SBO0	PS30:SBO70	5.96667*	.10000	.000	5.7361	6.1973
		PS50:SBO50	14.10000*	.10000	.000	13.8694	14.3306
		PS:70SBO30	21.13333*	.10000	.000	20.9027	21.3639
	PS30:SBO70	PS100:SBO0	-5.96667*	.10000	.000	-6.1973	-5.7361
		PS50:SBO50	8.13333*	.10000	.000	7.9027	8.3639
		PS:70SBO30	15.16667*	.10000	.000	14.9361	15.3973
	PS50:SBO50	PS100:SBO0	-14.10000*	.10000	.000	-14.3306	-13.8694
		PS30:SBO70	-8.13333*	.10000	.000	-8.3639	-7.9027
		PS:70SBO30	7.03333*	.10000	.000	6.8027	7.2639
	PS:70SBO30	PS100:SBO0	-21.13333*	.10000	.000	-21.3639	-20.9027
		PS30:SBO70	-15.16667*	.10000	.000	-15.3973	-14.9361
		PS50:SBO50	-7.03333*	.10000	.000	-7.2639	-6.8027

*. The mean difference is significant at the .05 level.

APPENDIX F

Statistical analysis for Hardness index (HI)

VAR0001

	SAMPLE	N	Subset for alpha = .05		
			1	2	3
Duncan[a]	PS:SBO 0:100	3	2.5600		
	PS:SBO 30:70	3	3.2567		
	PS:SBO 50:50	3	5.1167		
	PS:SBO 70:30	3		11.8650	
	PS:SBO 100:0	3			36.1900
	Sig.		.178	1.000	1.000

Means for groups in homogeneous subsets are displayed.

[a] Uses Harmonic Mean Sample Size = 3.000.

ANOVA

VAR0001

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	2393.728	4	598.432	140.357	.000
Within Groups	42.636	10	4.264		
Total	2436.365	14			

Descriptives

VAR0001

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
PS:SBO 100:0	3	36.1900	4.55688	2.63092	24.8701	47.5099	32.41	41.25
PS:SBO 70:30	3	11.8650	.66183	.38211	10.2209	13.5091	11.35	12.61
PS:SBO 50:50	3	5.1167	.33620	.19411	4.2815	5.9518	4.73	5.34
PS:SBO 30:70	3	3.2567	.03512	.02028	3.1694	3.3439	3.22	3.29
PS:SBO 0:100	3	2.5600	.02646	.01528	2.4943	2.6257	2.54	2.59
Total	15	11.7977	13.19189	3.40613	4.4922	19.1031	2.54	41.25

Multiple Comparisons

Dependent Variable: VAR0001

	(I) SAMPLE	(J) SAMPLE	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
						Lower Bound	Upper Bound
LSD	PS:SBO 100:0	PS:SBO 70:30	24.32500*	1.68595	.000	20.5685	28.0815
		PS:SBO 50:50	31.07333*	1.68595	.000	27.3168	34.8299
		PS:SBO 30:70	32.93333*	1.68595	.000	29.1768	36.6899
		PS:SBO 0:100	33.63000*	1.68595	.000	29.8735	37.3865
	PS:SBO 70:30	PS:SBO 100:0	-24.32500*	1.68595	.000	-28.0815	-20.5685
		PS:SBO 50:50	6.74833*	1.68595	.003	2.9918	10.5049
		PS:SBO 30:70	8.60833*	1.68595	.000	4.8518	12.3649
		PS:SBO 0:100	9.30500*	1.68595	.000	5.5485	13.0615
	PS:SBO 50:50	PS:SBO 100:0	-31.07333*	1.68595	.000	-34.8299	-27.3168
		PS:SBO 70:30	-6.74833*	1.68595	.003	-10.5049	-2.9918
		PS:SBO 30:70	1.86000	1.68595	.296	-1.8965	5.6165
		PS:SBO 0:100	2.55667	1.68595	.160	-1.1999	6.3132
	PS:SBO 30:70	PS:SBO 100:0	-32.93333*	1.68595	.000	-36.6899	-29.1768
		PS:SBO 70:30	-8.60833*	1.68595	.000	-12.3649	-4.8518
		PS:SBO 50:50	-1.86000	1.68595	.296	-5.6165	1.8965
		PS:SBO 0:100	.69667	1.68595	.688	-3.0599	4.4532
	PS:SBO 0:100	PS:SBO 100:0	-33.63000*	1.68595	.000	-37.3865	-29.8735
		PS:SBO 70:30	-9.30500*	1.68595	.000	-13.0615	-5.5485
		PS:SBO 50:50	-2.55667	1.68595	.160	-6.3132	1.1999
		PS:SBO 30:70	-.69667	1.68595	.688	-4.4532	3.0599

*. The mean difference is significant at the .05 level.

A. Personal Profile

Full name	Nabilah binti Abdul Hadi
National IC no	891224-11-5272
Birth date	24th December 1989
Citizenship	MALAYSIA
Place of birth	Terengganu, MALAYSIA
Gender	Female
Correspondence Address	3440, Kampung Duyong Kecil, 21300 Kuala Terengganu.
Telephone no. (H)	09-6232648
Telephone no. (HP)	013-6289593
Email address	nabilah_balqis@yahoo.com

B. Hobbies and interests

I enjoy gardening, reading and exploring new places. I like to spend my time by surfing internet exploring new things and find healthy tips. I love playing with my cats and listen to acoustic music

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C. Academic qualifications

Degree	**Area**	**Institutions**	**Year awarded**
B.SC. (Hons.)	Food Science and Technology	Universiti Teknologi MARA, MALAYSIA	2010-2013
Diploma	Food Technology	Universiti Teknologi MARA, MALAYSIA	2007-2010
S.P.M	Science	Sek.Men.Keb.Bandar	2006

D. Work experience

Post	**Place**	**Year**
Quality control	Pertima Trengganu SDN.BHD.	2010

E. Related experience

Post	Place	Year
Participant	Food Product Development Competition	2009
Participant	Food Transformation Competition	2011
Participant	Food Product Development Competition	2012
Participant	Halal Food Seminar organized by USIM	2012
Participant	Kursus Teknologi Pembungkusan Makanan di UNISZA	2012
Organizer	Hari Nutrisi bersama penduduk Kg Orang Asli Pabai dan Miku	2010

F. Awards

Type	Name of award/awarding organisations	Year
Certificate	Dean’s List Award 2009, UiTM Shah Alam	2009
Certificate	Dean’s List Award 2010, UiTM Shah Alam	2010
Certificate	Dean’s List Award 2011, UiTM Shah Alam	2011
Certificate	Dean’s List Award 2012, UiTM Shah Alam	2012

G. Other relevant information

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