

Fats and Oils for a Healthier Future  
**Macro, Micro and  
Nanoscales**



**PROFESSOR DR. TAN CHIN PING**

Fats and Oils for a Healthier Future

# Macro, Micro and Nanoscales

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## ABSTRACT

This book reviews major research outcomes produced and published by the author's research group over the last 12 years on topics related to fats and oils at the macro-, micro- and nanoscales from the scientific and technological perspective. Today, the food industry expects fats and oils to perform in many different applications, and consumers want them to deliver various functions and healthy benefits. At the macroscale level, the continuous search for new oilseed crops with more advantageous oil compositions may lead to the development of excellent candidates that can eventually reach commercial acceptance. More than 10 different types of oilseeds have been rigorously studied for their physicochemical properties and their potential for use in food and non-food applications. In addition, constant formulation development related to fats and oils serves important functions in food and beverage products, from texture and product stability to flavour.

At the microscale level, the extraction of valuable lipid bioactive compounds, the mitigation of 3-monochloropropane-1,2-diol (3-MCPD) esters in refined palm oil and the thermal analysis of fats and oils products using the differential scanning calorimetry (DSC) technique are the primary focuses of my research team. The extraction of phospholipids from palm-pressed fibre provides an opportunity to recover valuable lipid compounds from by-products of the palm milling industry. The quality of refined palm oil plays a pivotal role in meeting consumer demands for high-quality finished food products. Investigation of the formation of 3-MCPD esters and modification of the refining processes used in the production of refined palm oil are two tasks that are key to maintaining the status of palm oil as the major vegetable oil on the world market. In addition, a few DSC methods have been developed to enable

researchers to measure changes in the thermal properties and quality of various fat and oil products.

At the nanoscale, the range below 100 nm is important because at this small size, the laws of physics and chemistry for a bulk system change, resulting in novel properties that enable researchers to produce new materials with the exact characteristics they desire. The food industry has traditionally dealt with macro- or microscale lipid substances, but the introduction of the nanoscale may spur a paradigm shift within the field. The study of both fundamental and applied aspects of functional lipid nanodispersions has received increasing attention in recent years. Dispersion or high-energy emulsification methods are frequently used for the formation of functional lipid nanodispersions. Studies on the optimisation of the formulation and processing parameters have been the primary focus of my research team. The application of functional lipid nanodispersions as formulations for active delivery and targeting in various food systems is also an active and interesting area of study.

The research findings presented in this book are a compilation of publications from many Masters and PhD dissertations supervised by the author. The major findings presented in this book are not expected to be comprehensive, rather, this book is intended to appraise the reader of the wide variety of opportunities and challenges involved in the effort to understand the physicochemical properties and functionalities of fats and oils and subsequently improve the application of fats and oils in food products.

## **Introduction**

Fats and oils are the third main class of macronutrients required in human nutrition. They are basic components of man's caloric diet and have been used since time immemorial as food, as food ingredients and in the preparation of other foods. Today, fats and oils serve as raw materials for cooking oils, salad oils, margarine, shortening and other specialty products that have become essential ingredients in food products prepared in the home, in restaurants and by food manufacturers. Spurred by income and population growth in many developing countries, as well as the rapidly expanding food industry in Asia, the global growth in the consumption and trade of fats and oils is outpacing most other food commodities. The total use of 17 major fats and oils in 2012/2013 was predicted to climb to 187.7 million tons from the 183 million tons in 2011/2012 (Mielke, 2013). At the same time dietary use of fats and oils was forecast to climb to 164.2 million tons from 159.7 million tons. The great nutritional importance of fats and oils in feeding the ever-increasing world population and the technological advances achieved in this field have led to higher levels of production and improvements in the product quality and versatility of these commodities over the past three decades.

Today, it is well known that fats and oils are important components of our diet where at least a minimum intake is essential. However, there are many problems associated with an excessive intake of dietary fat, including obesity, cardiovascular diseases and certain forms of cancer. To ensure a healthier future for the next generation, it is essential for us to understand fats and oils at the macro-, micro- and nanoscales from a scientific and technological perspective.

## **THE OUTLOOK FOR FATS AND OILS AT THE MACROSCALE LEVEL**

### **Potential New Oilseeds for Food and Non-food Applications**

Oil-bearing fruits, nuts and seeds have been grown and used as food for many centuries. Approximately 250,000 plant species are known, with 4,500 species having been examined for their oil (Orthofer, 1996). Although nearly all higher plants contain oils to a significant extent, only approximately forty of these are suitable as raw materials for human consumption (Salunkhe, 1992). The largest source of vegetable oil at present is the seeds of annual plants which produce oils, such as, soybean, cottonseed, peanut, sunflower, safflower, corn and canola or rapeseed (Orthofer, 1996). These plants are cultivated in temperate climatic regions (Sonntag, 1979). Many of these oil-bearing seeds are sources of not only oil but also protein, and in many cases, they are more valuable as a protein source. Other oil-bearing seeds such as peanuts and corn are by-products of crops grown primarily for other purposes (O'Brien, 1998). A second source of vegetable oils is the oil-bearing fruits and nuts of perennial trees which produce oils such as coconut, palm, palm kernel and olive oil (Padley, 1994). These perennial oil-bearing trees are grown in tropical regions. The oils from palm and olive trees are extracted from the fruit rather than the seed of the fruit.

Due to the high demand for and price of vegetable oils, interest in novel sources of vegetable oils has recently increased. To cater to the increasing demand for vegetable oils, non-conventional oilseeds are being considered because their constituents exhibit unique chemical properties and this may result in an increase in the supply of edible oils for various food and non-food applications (Nyam et al., 2009a, Lim et al., 2010, Nehdi et al., 2012a, Nehdi et al., 2012b,

Nehdi et al., 2013, Sbihi et al., 2013, Nehdi et al., 2014a, Nehdi et al., 2014b). No oil from any source has been found to be suitable for all purposes because oils from different sources generally differ in their compositions. Thus far, a large number of seeds have been analysed, and some of these seeds have been cultivated as new oil crops. Over the last five years, the physicochemical properties and chemical compositions of oils extracted from more than ten varieties of plant seeds (bitter melon, Kalahari melon, kenaf, pumpkin, roselle, pitaya, *Acacia senegal*, garden cress, colocynth, bitter and sweet lupins, *Chamaerops humilis* palm and *lantoro*) have been investigated by my research group using established methods (Table 1).

The chemical compositions of oils extracted from the bitter melon, Kalahari melon, kenaf, pumpkin and roselle seeds have been evaluated by Nyam et al. (2009a). Improved knowledge of the composition and properties of the bitter melon, Kalahari melon, kenaf, pumpkin, and roselle seeds will assist in efforts to achieve industrial application of these plants. The Bitter melon (*Momordica charantia* L.), also known as bitter gourd, is a monoecious climbing vine. It is a tropical crop that is grown throughout Asia for food and medicinal purposes (Chakravarty, 1990). The bitter melon's seeds contain an oil in which the major fatty acid is eleostearic acid. The Kalahari melon (*Citrullus lanatus*) is an important source of water in the Kalahari during the dry months of the year when no surface water is available (Van Wyk and Gericke, 2000). For the last few years, the kenaf (*Hibiscus cannabinus* L.) plant has been cultivated in Malaysia. Kenaf has been reported to be a suitable source for livestock and horse feed because it is high in protein (more than 20%) and has potential to replace alfalfa (Daham, 2005). Pumpkin (*Cucurbita pepo* L.) seed oil is a common salad oil in Austria and is produced in Slovenia, Hungary and the southern parts of Austria.

**Table 1** Twelve selected oilseeds

<b>Oilseed</b>	<b>Scientific name</b>	<b>Reference</b>
Bitter melon seed	<i>Momordica charantia</i> L.	Nyam et al. 2009a
Kalahari melon seed	<i>Citrullus lanatus</i>	Nyam et al. 2009a
Kenaf seed	<i>Hibiscus cannabinus</i> L.	Nyam et al. 2009a
Pumpkin seed	<i>Cucurbita pepo</i> L.	Nyam et al. 2009a
Roselle seed	<i>Hibiscus sabdariffa</i> Linn.	Nyam et al. 2009a
Pitaya seed	<i>Hylocereus undatus</i> and <i>Hylocereus polyrhizus</i>	Lim et al., 2010
<i>Acacia senegal</i> seed	<i>Acacia Senegal</i> (L.) Willd.	Nehdi et al., 2012a
Garden cress seed	<i>Lepidium sativum</i> Linn.	Nehdi et al., 2012b
Colocynth seed	<i>Citrullus colocynthis</i> (L.) Schrad	Nehdi et al., 2013
Bitter and sweet lupins seeds	<i>Lupinus albus</i> L.	Sbjiti et al., 2013
<i>Chamaerops humilis</i> palm seed	<i>Chamaerops humilis</i> L. var. <i>argentea</i> André	Nehdi et al., 2014a
<i>Lamitoro</i> seed	<i>Leucaena leucocephala</i> (Lam.) de Wit	Nehdi et al., 2014b

The vitamin E content in pumpkin seeds is very high (Murkovic et al., 1996). Roselle (*Hibiscus sabdariffa*, Linn.) seed is a valuable food resource (Al-Wandawi et al., 1984; Balami, 1998) because of its protein and caloric content in addition to its substantial amounts of fibre and valuable micronutrients (Omobuwaju, Sanni and Balami, 2000).

Table 2 presents the fatty acid compositions of the crude seed oils of the bitter melon, Kalahari melon, kenaf, pumpkin and roselle seeds. All the oil samples contained high levels of total unsaturated fatty acids (consisting predominantly of oleic and linoleic acids). The oleic and linoleic acid contents were found to be 65.6% for bitter melon seed oil, 81.6% for Kalahari melon seed oil, 74.8% for kenaf seed oil, 73.5% for pumpkin seed oil and 64.1% for roselle seed oil. Among these oils, the Kalahari melon seed oil had the highest content of linoleic acid (63.1%). The presence of such high levels of linoleic acid suggests that these oils may serve as good sources of essential fatty acids. In addition, these oils seem to be a good source of lipid-soluble bioactives (Table 3). Based on this study, the presence of phenolics, tocopherols and sterols at the estimated levels may be of nutritional importance in the application of these seed oils.

**Table 2** Relative percent composition of fatty acid in bittermelon, Kalahari melon, kenaf, pumpkin and roselle seed oils.

Fatty acids	Oil type				
	Bittermelon	Kalahari melon	Kenaf	Pumpkin	Roselle
C <sub>14:0</sub>	-	-	-	-	0.2
C <sub>16:0</sub>	1.5	12.4	20.3	19.1	21.4
C <sub>16:1</sub>	-	-	0.4	-	0.3
C <sub>18:0</sub>	32.4	7.5	3.8	7.4	5.0
C <sub>18:1</sub>	1.5	17.1	37.1	42.8	26.2
C <sub>18:2</sub>	2.6	63.1	36.6	30.4	39.4
C <sub>18:3</sub>	-	1.1	0.3	-	-
C <sub>18:3n9</sub>	61.5	-	-	-	-
C <sub>20:0</sub>	-	-	0.6	-	1.4
C <sub>20:1</sub>	-	0.3	0.5	0.4	0.6
C <sub>21:0</sub>	-	-	0.5	-	-
C <sub>22:1</sub>	-	-	-	-	0.3
C <sub>22:2</sub>	-	-	-	-	0.3
C <sub>24:0</sub>	-	-	0.6	-	5.1
SAT	33.9	19.9	25.8	26.5	33.0
MONO	1.5	17.3	37.9	43.1	27.4
POLY	64.1	64.3	36.9	30.4	39.7

(Source: Nyam et al. 2009a)

**Table 3**  
a. Sterols and squalene of oilseeds (mg/ 100 g, mean  $\pm$  SD)<sup>A</sup>

Phytosterol	Oil type				
	Bittermelon	Kalahari melon	Kenaf	Pumpkin	Roselle
Squalene	12.95 $\pm$ 3.98 <sup>b</sup>	160.32 $\pm$ 14.37 <sup>c</sup>	3.69 $\pm$ 0.37 <sup>a</sup>	590.69 $\pm$ 26.58 <sup>d</sup>	14.51 $\pm$ 6.79 <sup>b</sup>
Cholesterol	nd	nd	nd	nd	14.25 $\pm$ 0.75
Ergosterol	nd	nd	nd	22.40 $\pm$ 1.46	nd
Campesterol	35.03 $\pm$ 2.20 <sup>b</sup>	130.41 $\pm$ 2.73 <sup>e</sup>	58.08 $\pm$ 2.16 <sup>c</sup>	22.60 $\pm$ 3.90 <sup>a</sup>	102.35 $\pm$ 3.94 <sup>d</sup>
Stigmasterol	24.06 $\pm$ 1.17 <sup>b</sup>	25.87 $\pm$ 0.28 <sup>c</sup>	23.30 $\pm$ 1.68 <sup>b</sup>	18.02 $\pm$ 1.18 <sup>a</sup>	38.53 $\pm$ 2.43 <sup>d</sup>
Sitosterol	405.21 $\pm$ 2.69 <sup>c</sup>	485.41 $\pm$ 4.11 <sup>e</sup>	289.87 $\pm$ 2.40 <sup>b</sup>	210.98 $\pm$ 3.66 <sup>c</sup>	602.42 $\pm$ 1.32 <sup>d</sup>
Sum	477.25	802.01	371.25	864.69	772.06

nd not detected.

<sup>A</sup>Means in the same row with different letters are significantly different (\*P < 0.05).

b. Tocopherols of oilseeds (mg/ 100 g, mean  $\pm$  SD)<sup>A</sup>

Tocopherol/	Oil type				
	Bittermelon	Kalahari melon	Kenaf	Pumpkin	Roselle
$\alpha$	44.22 $\pm$ 6.82 <sup>c</sup>	25.94 $\pm$ 2.68 <sup>b</sup>	20.01 $\pm$ 1.01 <sup>b</sup>	15.19 $\pm$ 1.84 <sup>b</sup>	3.68 $\pm$ 0.23 <sup>a</sup>
$\beta$	19.66 $\pm$ 1.62 <sup>b</sup>	3.27 $\pm$ 0.30 <sup>a</sup>	0.79 $\pm$ 0.01 <sup>a</sup>	nd	3.19 $\pm$ 0.01 <sup>a</sup>
$\gamma$	53.93 $\pm$ 7.35 <sup>c</sup>	70.56 $\pm$ 0.66 <sup>b</sup>	63.86 $\pm$ 3.63 <sup>a</sup>	61.32 $\pm$ 1.17 <sup>a</sup>	70.65 $\pm$ 2.98 <sup>a</sup>
$\delta$	17.21 $\pm$ 0.04 <sup>b</sup>	9.33 $\pm$ 0.93 <sup>b</sup>	nd	4.14 $\pm$ 0.33 <sup>a</sup>	4.60 $\pm$ 0.50 <sup>a</sup>
Sum	135.02	109.10	84.66	80.65	82.12

nd, not detected.

<sup>A</sup>Means in the same row with different letters are significantly different (\*P < 0.05).

(Source: Nyam et al. 2009a)

Industrial processes for the extraction of edible oils from oilseeds generally involve a solvent extraction step, sometimes preceded by pressing. In the past, safety considerations with respect to the use of organic solvents have prompted attempts to develop aqueous extraction methods (Hagenmaier et al., 1975). Supercritical fluid extraction using CO<sub>2</sub> is another popular technology for rapid, contamination-free extraction in the food and pharmaceutical industries. Supercritical carbon dioxide (SC-CO<sub>2</sub>) extraction has also been used to concentrate minor constituents of various oilseeds and other products (Mendes et al., 2005, Ibanez et al., 2002, Catchpole et al., 1997, Montanari et al., 1999, de Franca and Meireles, 2002). Meanwhile, Ultrasound-assisted extraction provides a mechanical effect, allowing greater solvent penetration into the sample matrix and increasing the contact surface area between the solid and liquid phases, whereby as a result, the solute quickly diffuses from the solid phase to the solvent (Rostagno et al. 2003, Jin et al., 2013). Recently, interest in aqueous extraction, SC-CO<sub>2</sub> extraction and ultrasound-assisted extraction has been revived because of the need for environmentally cleaner alternative technologies for oil extraction. For these reasons, improvements in oilseed processing that facilitate obtaining products of richer composition, such as aqueous enzymatic oil extraction, supercritical CO<sub>2</sub> extraction and ultrasound-assisted extraction, are becoming more common. Recently, all three extraction techniques have been used to extract oil from Kalahari melon seeds and roselle seeds (Nyam et al., 2009b, 2009c, 2010a, 2010b, 2011, Wong et al. 2014a). In addition, because of the presence of high levels of bioactive compounds, the Kalahari melon seed oil has been encapsulated in powder form and the effects of both the processing and formulation on the stability of the microencapsulated oil have been thoroughly

studied (Ng et al., 2013a, 2013b, 2014, Razmkhah et al., 2013, Wong et al., 2014b).

Pitaya exists in a number of varieties; three varieties that have been commercialised are the *Hylocereus undatus* (Red Pitaya), which has red-skinned fruit with white flesh, *Hylocereus polyrhizus*, which has red-skinned fruit with red flesh, and *Hylocereus megalanthus* (Yellow Pitaya), which has yellow-skinned fruit with white flesh (Arcadio, 1986; Barbeau, 1990). High oil contents (18.3 - 28.4%) have been found in two species of pitaya (*Hylocereus cacti*): *Hylocereus undatus* (white-fleshed) and *Hylocereus polyrhizus* (red-fleshed) (Lim et al., 2010). The principal fatty acids in *H. undatus* seed oil (WFSO) and *H. polyrhizus* seed oil (RFSO) are palmitic (C16:0), oleic (C18:1) and linoleic (C18:2) acids. This study also demonstrated that pitaya seed oil can serve as a potential source of natural antioxidants, such as phenols, tocopherols and sterols (Lim et al. 2010). Both  $\alpha$ -tocopherol and  $\gamma$ -tocopherol were detected in both seed oils while the RFSO was found to contain a significantly ( $P < 0.05$ ) higher amount of  $\alpha$ -tocopherol than the WFSO. Both RFSO and WFSO were found to contain an abundance of  $\beta$ -sitosterol: 676 and 548 mg per 100 g, respectively. In addition, seven phenolic acid compounds were identified in the WFSO and RFSO, namely, gallic, vanillic, syringic, protocatechuic, *p*-hydroxybenzoic, *p*-coumaric and caffeic acids. Protocatechuic acid is the major phenolic acid in both seed oils.

Due to its high oil content and significant level of bioactive compounds, *Hylocereus polyrhizus* (red-fleshed) seed oil has been spray dried into an encapsulated oil powder (Lim et al., 2012). Six matrices were used for the wall material of the spray-dried encapsulated pitaya seed, and their effects on the physiochemical properties of the microencapsulated oil powder were determined. A study on oil retention revealed the following ranking for the

effectiveness of the preferred wall materials for the encapsulation of pitaya seed oil: sodium caseinate > whey protein > gum Arabic. Although gum Arabic demonstrated lower microencapsulation efficiency it was found to be the most effective encapsulant for the prevention of lipid oxidation in the encapsulated oil and exhibited strong oxidative stability. With regards to the two saccharides used in this study, lactose was found to be significantly superior to maltodextrin in retarding the oxidation of spray-dried pitaya seed oil powder.

*Acacia senegal* is a leguminous multipurpose African tree species that belongs to the subgenus *Aculeiferum* (Arce and Banks, 2001). The species grows to 2–15 m in height with a flat or rounded crown (Chiveu et al., 2008). In previous studies (Kafi and Sabahalkhair, 2010, Aoki et al., 2007 and Abd-Razig et al., 2007), only the gum Arabic that can be naturally obtained from the *A. senegal* tree had been studied to determine its uses and applications in the food industry and non-food industries. Nehdi et al. (2012a) studied the potential of the *A. senegal* seed oil. The chemical composition, major physicochemical properties and thermal stability of the oil extracted from *Acacia senegal* seeds were evaluated. The major fatty acids in the oil are oleic acid (43.62%) followed by linoleic acid (30.66%) and palmitic acid (11.04%) (Table 4). The observed physicochemical characteristics of the oil indicate that this oil can be successfully applied in the coating industry and for deep frying. According to these results, *A. senegal* seed oil has physicochemical properties, a fatty acid composition and thermal characteristics that may be interesting for specific applications in several segments of the food industry and non-food industries.

**Table 4** Fatty acid profile (% of total fatty acid) of selected oilseeds

Fatty acid	<i>A. senegal</i> seed oil	<i>Lepidium</i> <i>sativum</i> oil	Sweet lupin ( <i>Lupinus albus</i> L.) seed oils	Bitter lupin ( <i>Lupinus albus</i> L.) seed oils	<i>C. humilis</i> seed oil	<i>L.</i> <i>leucocephala</i> seed oil.
<b>SFA</b>						
C10:0	nd	0.04	nd	nd	2.40	nd
C11:0	nd	0.07	nd	nd	nd	nd
C12:0	0.06	0.04	nd	nd	21.27	nd
C14:0	0.19	0.11	0.06	0.10	7.17	0.058
C15:0	nd	0.05	0.07	0.06	0.067	0.033
C16:0	11.04	9.03	7.50	7.39	9.96	13.21
C17:0	0.07	0.05	0.05	nd	0.10	0.11
C18:0	2.07	3.28	1.74	1.83	3.57	5.92
C20:0	2.06	3.92	1.04	1.13	0.19	1.87
C21:0	0.09	nd	nd	nd	nd	nd
C22:0	nd	0.99	3.44	3.20	0.04	1.86
C23:0	0.07	nd	nd	nd	nd	nd
C24:0	0.91	nd	nd	nd	nd	nd

<b>MUFA</b>							
C14:1	nd	0.04	nd	nd	0.016	nd	nd
C15:1	nd	0.04	nd	nd	0.019	0.022	0.43
C16:1	0.19	0.26	0.42	0.31	0.10	0.058	0.072
C17:1	0.07	0.04	0.07	0.06	46.28	38.99	23.01
C18:1	nd	22.51	48.72	4.76	0.36	0.16	0.48
C20:1	0.92	12.22	4.59	1.74	nd	nd	nd
C22:1	nd	4.51	1.96	nd	nd	nd	nd
<b>PUFA</b>							
C16:2	nd	0.04	nd	nd	nd	nd	nd
C16:3	nd	0.05	nd	nd	nd	nd	nd
C18:2	30.66	11.17	20.90	21.55	15.15	51.65	1.23
C18:3	2.78	30.11	8.95	7.69	0.11	nd	nd
C20:2	2.04	0.41	0.42	2.16	nd	nd	nd
C20:3	2.19	0.45	nd	1.65	nd	nd	nd
C20:4	0.78	nd	nd	nd	nd	nd	nd

SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; nd, not detected.

(Source: see Table 1)

Vegetable oils such as palm, rapeseed, soybean and sunflower oil are the most commonly used raw materials for biodiesel production, but these oils are expensive and are also used for food production. Thus, non-food oils have been evaluated as potential renewable resources for biodiesel production (Ruan et al., 2008 and Ruan et al., 2012). One possible alternative for biodiesel production is the oil from the seeds of the garden cress (*Lepidium sativum* L.). Garden cress is another fast-growing annual herb which is native to Egypt and west Asia but is also widely cultivated in temperate countries throughout the world for various culinary and medicinal uses (Gokavi et al., 2004). Garden cress can be sown and harvested several times throughout the year, although January, February and November are the most suitable months of the year to sow garden cress in a Mediterranean climate (Tuncay et al., 2011). These growth features make the *L. sativum* seed an attractive potential source of oil, hence justifying its study for potential industrial uses. In another study, *L. sativum* seed oil extracted from plants grown in Tunisia was analysed to determine its potential for use as a raw material for biodiesel production (Nehdi et al. 2012b). The oil content of the seeds was found to be 26.8% and to consist predominantly of polyunsaturated (42.2%) and monounsaturated (39.6%) fatty acids (Table 4). *L. sativum* seed oil has been extracted and chemically converted via an alkaline transesterification reaction into its fatty acid methyl esters. Based on the observed physicochemical properties, the methyl esters of *L. sativum* seed oil have been demonstrated to be a good potential alternative to imported petroleum diesel.

Another potential alternative oil is the non-conventional oil from the seeds of the *Citrullus colocynthis* (L.) Schrad, commonly known as colocynth, bitter apple, egusi melon or ground melon. It is a species of the genus *Citrullus* of the Cucurbitaceae family and

exists in a large number of varieties that are generally regarded as melons (Mabaleha, Mitei, & Yeboah, 2007). *C. colocynthis*, which is native to tropical Asia and Africa (Dane et al., 2007), is now widely distributed in the Saharo-Arabian phylogeographic region of Africa, in the Mediterranean region, eastward through Iran to India and in other parts of tropical Asia. This perennial herb is a drought-tolerant species and can survive in arid environments, even under severe stress conditions. Therefore, *C. colocynthis* can be considered as a serious oilseed crop candidate for medicinal, food and non-food applications. Nehdi et al. (2013) have studied the physicochemical properties, fatty acids, tocopherols, thermal properties, and <sup>1</sup>H NMR and FTIR profiles of the non-conventional oil extracted from the *Citrullus colocynthis* (L.) Schrad seeds and compared them with those of conventional sunflower seed oil. The oil content of the *C. colocynthis* seeds is approximately 23%. The predominant fatty acids in the oil are linoleic acid (66.7%) followed by oleic acid (14.8%), palmitic acid (9.7%), and stearic acid (7.4%). The tocopherol content is 121.85 mg per 100 g, with  $\alpha$ -tocopherol as the major component (95.5%). Thermogravimetric analysis demonstrated that the oil remained thermally stable up to 286.57 °C and then began to decompose in four stages, namely, at 377.4 °C, 408.4 °C, 434.9 °C and 559.2 °C. This study revealed that *C. colocynthis* seed oil possesses physicochemical properties, antioxidant properties, fatty acids and tocopherol compositions that may be of interest for both food and non-food applications. In addition, because of its high content of linoleic acid and wide range of absorbance in the UV-B region, it can be used for the restoration of the stratum corneum permeability barrier of the skin through topical application, indicating that it may be beneficial to be included in the formulation of sunscreen products with a high SPF factor.

Lupins are members of the family Fabaceae. White lupin, which is native to West Asia and the eastern Mediterranean region of southern Europe, is an annual, erect, branched, bushy, and more or less pubescent herbaceous plant. Today, white lupin is a traditional minor pulse crop that is grown around the Mediterranean and the Black Sea as well as in the Nile valley. It is also cultivated elsewhere in Africa, North and South America (Lim, 2012). Lupins are of particular interest because they are one of the richest known sources of protein (Jul et al., 2003 and Chew et al., 2003) with significant antioxidant properties and health benefits for humans and animals (Vicenti et al., 2009 and Chiofalo et al., 2012). There are two basic types of lupins: the bitter and sweet varieties. In a recent study, a full characterisation and comparison of the oils extracted from the seeds of bitter and sweet *L. albus* lupins was undertaken (Sbihi et al. 2013). The results indicated that the bitter and sweet lupin seeds contained 8% and 12% oil, respectively. The total tocopherol contents of the bitter and sweet lupin seed oils were found to be 184.70 and 317.01 mg per 100 g of oil, respectively. Oleic acid, linoleic acid and linolenic are the major fatty acids contained in these oils (Table 4). Their fatty acid composition makes bitter and sweet lupin seed oils very interesting from a nutritional point of view.

The worldwide production, utilisation and industrialisation of date palms have been continuously increasing, as date fruits have earned a position of great importance in human nutrition because of their rich content of essential nutrients (Al-Shahib and Marshall, 2003; Chandrasekaran and Bahkali, 2013). The *Chamaerops humilis* palm has traditionally been used as a medicinal plant. The presence of tannins, flavonoids, saponins, sterols and terpenoids in the leaves, the spadices and the heart of the palm is the origin of the therapeutic virtues of this species (Hasnaoui and others,

2013). Furthermore, the fruit has also been used as an astringent agent because of its bitterness and high tannin content (Merlo and others, 1993). *Chamaerops humilis* L. var. *argentea* André (*C. humilis*) date palm seeds are an underutilised source of vegetable oil, and recently, a major study describing their physicochemical characteristics to elucidate the potential uses of this seed or seed oil has been reported (Nehdi et al. 2014a). The oil content of the seeds is approximately 10% and is predominantly composed of oleic acid (38.7%), lauric acid (21.3%), linoleic acid (15.2%), palmitic acid (10.0%) and stearic acid (7.2%) (Table 4). The tocol (tocopherols and tocotrienols) content is 74 mg per 100 g, with  $\delta$ -tocotrienol as the major contributor (31.9%). This study revealed that *C. humilis* date palm seed oil can be classified as a natural nutritional dietary product that presents a host of health benefits.

The genus *Leucaena* belongs to the family Fabaceae and the subfamily Mimosoideae and includes approximately 32 species. *Leucaena leucocephala* (Lam.) de Wit Syn. *Acacia leucocephala* (Lam.), *Mimosa leucocephala* (Lam.) and *Leucaena glabrata* (Rose) are the most important species of this genus (Pandey and Kumar, 2013). *Leucaena leucocephala* (Lam.) de Wit (coffee bush), also known as *lamtoro* in Malaysia, is a strictly tropical species that requires warm temperatures for optimal growth. It is drought tolerant and can withstand up to 7 months of dry season. *L. leucocephala* produces a gum similar to gum Arabic, which is used in ice creams, cosmetics, and in the pharmaceutical industry (Lim, 2012). Recently, *L. leucocephala* oil extracted from the seeds of this plant was analysed to determine its potential uses (Nehdi et al. 2014b). The study revealed that *L. leucocephala* seed oil is of good quality and has a pleasant odour. The oil content of the seeds is approximately 5.4% and predominantly consists of linoleic acid (51.7%), oleic acid (21.4%), palmitic acid (13.2%) and stearic

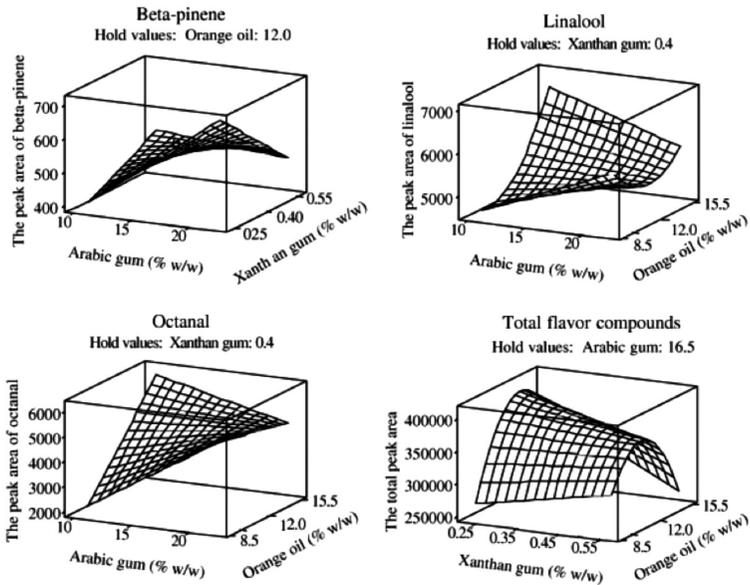
acid (5.9%) (Table 4). This oil has high vitamin E activity because of its concentration of  $\alpha$ -tocopherol (175.5 mg per 100 g). These characteristics, along with its absorbance in the UV range, suggest that *L. leucocephala* could be used as a component in natural cosmetic formulations, moisturisers, lipsticks and sunscreens.

## Development of Fat and Oil Products

Most flavour compounds present in various food products are lipid soluble. The first perceptions of food products are formed when flavour compounds are released from the foods and transported to the oral cavity and the nose to trigger the appropriate senses. However, the long-term stability of flavour compounds in food products has become a major concern in the food industry, especially in the beverage industry, because of the complex interactions among key food ingredients (e.g., polysaccharides and proteins). Hence, various studies have been conducted to formulate emulsion-based beverages using natural food emulsifiers and to understand the interactions between emulsion compositions and flavour compounds. Beverage emulsions are oil-in-water (O/W) emulsions that are typically prepared in concentrated form and then diluted several hundred times prior to consumption as either carbonated or non-carbonated soft drinks.

The effects of various hydrocolloids on the physicochemical properties and stability of orange-oil beverage emulsions have been investigated in a series of studies (Mirhosseini et al. 2007a, 2008a, 2008b, 2008c, 2008d, 2009a, 2009b, 2009c). For example, studies have been conducted to investigate the effects of pectin and carboxymethylcellulose on the physical stability, turbidity loss rates, cloudiness and flavour-release profiles of orange-oil emulsions during storage (Mirhosseini et al., 2008c). Moreover, the effects of gum Arabic, xanthan gum and orange oil on flavour

release from diluted orange-oil emulsions have been investigated using response surface methodology (Mirhosseini et al. 2008b). Figure 1 illustrates how significant ( $p < 0.05$ ) interaction effects of independent variables influence the release of certain volatile flavour compounds.



**Figure 1** Response surface plots showing the significant ( $p < 0.05$ ) interaction effects of independent variables on the release pattern of some of target volatile flavor compounds (Source: Mirhosseini et al., 2008b)

In addition, the effects of glycerol and vegetable oil on the physicochemical properties of gum-Arabic-stabilised orange-oil emulsions have been studied (Mirhosseini et al., 2008e). In beverages, glycerol acts as a humectant, solvent and sweetener, whereas vegetable oil is one of the major emulsion components

in the emulsion system. The presence of vegetable oil in the emulsion formulation alters the physicochemical properties of the emulsion. Vegetable oil is typically added to the formulation of a beverage emulsion to induce a desirable cloudiness in the finished emulsion-based product. To study the flavour release of key volatile compounds in orange-oil emulsions, solid-phase microextraction with headspace analysis has been developed (Mirhosseini et al., 2007b, 2008f).

In line with this, propylene glycol alginate and sucrose esters have been used to produce modified-starch-stabilised soursop flavour emulsions (Cheong et al. 2014a). The results indicated that propylene glycol alginate may be used in combination with modified starch to modulate the physical stability of a soursop emulsion at low and room temperatures. In addition, sucrose monoesters were found to be unsuitable for emulsion stabilisation in low-pH conditions, either in a mixture or as the sole emulsifier. More recently, the effect of prime emulsion components as a function of the equilibrium headspace concentration of soursop flavour compounds was investigated by Cheong et al. (2014b). This study demonstrates the importance of understanding the effects of interactions among emulsion components for the development of an optimal beverage emulsion with a desirable flavour-release profile. The interactions between modified starch and protein were found to have antagonistic effects on the flavour release from the emulsion matrix. Prior to these two studies, optimisation of the equilibrium headspace of volatile flavour compounds in Malaysian soursop had been performed using solid-phase microextraction and two-dimensional gas chromatography time-of-flight mass spectrometry (GC×GC-TOFMS) (Cheong et al., 2010, 2011).

Previous studies of cookie filler (CF) product development resulted in the application of various hydrogenated fats and oils to

achieve acceptable structural stability (O'Brien, 2004). However, lately the use of hydrogenated fats and oils has been discouraged because of the resultant formation of *trans* fatty acids, which have been demonstrated to increase the level of low-density lipoproteins and decrease the level of high-density lipoproteins (Martin et al., 2005). In the absence of hydrogenation, the required properties of the fat portion of a CF product can be obtained by blending lauric oils with liquid oils or hard fats, such as coconut- and palm-based fats and oils (Martin et al., 2005). The potential of a fat blend consisting of palm mid-fraction, palm stearin and virgin coconut oil for the development of CF products was recently investigated by Masni et al. (2013). In general, blends of coconut- and palm-based fats and oils that do not require hydrogenation and/or esterification demonstrate great potential in the development of healthier fat-based food products.

O/W emulsions, which consist of oil droplets dispersed in an aqueous continuous phase, have been proven to enhance the bioavailability of fat-soluble bioactive compounds and protect them from environmental destruction (Cornacchia et al., 2011). Coenzyme Q10 (CoQ10), also known as ubiquinone, ubiquinol and ubidecarenone, is a lipid-soluble compound that is synthesised endogenously in the human body (Miles et al., 2002, Balakrishnan et al., 2009). The important role of CoQ10 in various clinical contexts is well established. A CoQ10 formulation in a  $\kappa$ -carrageenan-coated O/W emulsion was developed recently (Chan et al. 2013). In this study, we examined the solubility of CoQ10 in various carrier oils and the effects of the emulsifier type on the formation and stability of CoQ10-loaded O/W emulsions. CoQ10 was found to be significantly ( $p < 0.05$ ) more soluble in medium-chain oils (coconut oil and palm kernel oil) compared with other vegetable oils. Irrespective of the oil used, the results indicated that complexes

of sodium stearyl lactylate and  $\kappa$ -carrageenan yielded the most stable CoQ10-loaded O/W emulsions, with smaller and narrower particle size distributions. Both macroscopic and microscopic observations indicated that an O/W emulsion stabilised with SSL/ $\kappa$ -carrageenan was the only type of emulsion that exhibited no signs of coalescence, flocculation or phase separation throughout the investigated storage period.

In another study, a virgin-coconut-oil-based emulsion was developed and proved to be a stable emulsion with good textural properties that is suitable for use as a novel food supplement to promote increased consumption of virgin coconut oil among consumers (Khor et al., 2014). This newly developed virgin coconut oil emulsion was compared with four commercial emulsion products.

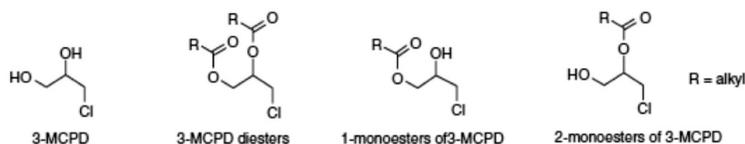
## **THE OUTLOOK FOR FATS AND OILS AT THE MICROSCALE LEVEL**

### **Extraction of Lipid Bioactive Compounds and Mitigation of 3-monochloropropane-1,2-diol (3-MCPD) Esters**

Palm-pressed fibre (PPF) is a by-product of palm oil milling and is generally used to generate the electricity supply for the mill and small housing estates around the mill (Basiron and Simeh, 2005). It typically constitutes approximately 15%, by weight, of a fresh fruit bunch and contains a high level of phospholipids (PLs) (Choo et al., 2004). A central composite design (CCD) was employed to study the effects of ultrasound-assisted extraction (UAE) conditions — namely, amplitude, cycle and sonication time — on the extraction yield of PLs from PPF (Chua et al., 2009).

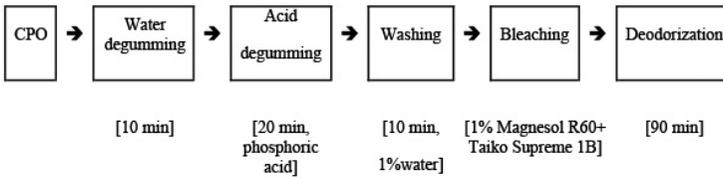
Consequently, a combination of an amplitude of 20%, a cycle of 0.2 W/s and a sonication time of 30 min was predicted to provide the highest PL-extraction efficiency. Under these optimal conditions, the response values obtained for the overall extraction efficiency and individual extraction yield of phosphatidylethanolamine and phosphatidylcholine were 110 (mg/g), 12,570 (mg/kg) and 5,426 (mg/kg), respectively. The best solid-phase extraction (SPE) method for the purification of PLs from PPF was determined in an earlier publication (Chua et al. 2008). UAE has also been used to extract other non-lipid bioactive compounds from various local herbs (Thoo, 2013, Ho et al. 2014)

Free 3-MCPD has been demonstrated to be carcinogenic in animal studies and induces infertility and the malfunction of certain organs, such as the kidney. 3-MCPD is found in refined edible oils in the form of esters (bound 3-MCPD, Figure 2) with fatty acids (Zelinková et al., 2006), but no toxicological data are available for the chloroesters. Detection of 3-MCPD esters in human breast milk (Zelinková et al., 2008) suggests that bioaccumulation of esterified 3-MCPD is possible in bodily lipids, leading to the exposure of infants to this hazard. Full hydrolysis of these chloroesters in vivo would result in significant exposure to free 3-MCPD, exceeding the provisional maximal tolerable daily intake (PMTDI) of 2 µg per kg of body weight set by the JECFA (JECFA, 2002). The formation of 3-MCPD esters in refined oils is associated with high-temperature processing, primarily during the deodorisation step, with the highest level reported in refined palm oil (4.5 to 13 mg/kg) (Franke et al., 2009). There are no facile solutions to this problem because other stages of refining, such as degumming and bleaching, could activate the formation of 3-MCPD ester precursors.



**Figure 2** Reduction of 3-MCPD esters formation in refined palm oil via optimization of physical refining

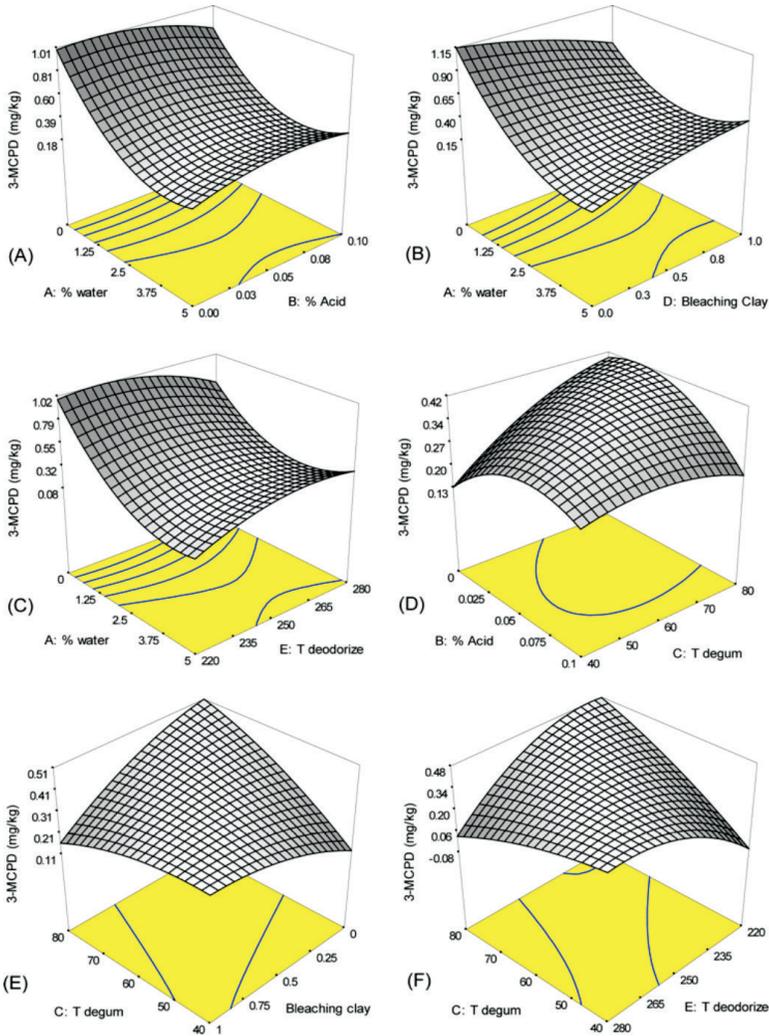
The presence of 3-MCPD esters in most refined oils is an important issue in the edible oil industry. Palm oil has been reported to contain the highest level of 3-MCPD esters compared with other oils. Contamination with 3-MCPD esters during the physical refinement process of palm oil has been studied, including the analytical aspects, processing factors and related precursors that contribute to their formation, for the mitigation of chloroesters during the refinement process (Zulkurnain et al., 2012, 2013). The effects of the degumming and bleaching processes on the reduction of 3-MCPD ester formation in palm oil refined from poor-quality crude palm oil using a D-optimal design have been studied relative to the minor components of the palm oil that are likely to be the precursors of these 3-MCPD esters (Zulkurnain et al., 2012). Zulkurnain et al. (2012, 2013) modified the refinement process for palm oil to produce a refined, bleached and deodorised (RBD) palm oil with a reduced level of 3-MCPD esters and equivalent RBD palm oil quality for all ranges of crude palm oil as the feed oil. Figure 3 illustrates the process flow for palm oil refinement according to this study.



**Figure 3** Modified process flow for palm oil physical refining  
 (Source: Zulkurnain et al., 2013).

The modified refinement process was optimised for minimal 3-MCPD ester formation and acceptable quality of the refined palm oil using response surface methodology (RSM) with five processing parameters: water dosage, phosphoric-acid dosage, degumming temperature, activated clay dosage and deodorisation temperature. The effects of the interactions among the various parameters were examined using the generated response surface plots for the 3-MCPD ester level (Figure 4). The removal of chloroester precursors was accomplished primarily by increasing the water dosage, whereas the reduction in the level of 3-MCPD esters was a compromise between the oxidative stability and colour of the refined palm oil. The optimisation resulted in an 87.2% reduction of 3-MCPD esters.

## Fats and Oils for a Healthier Future

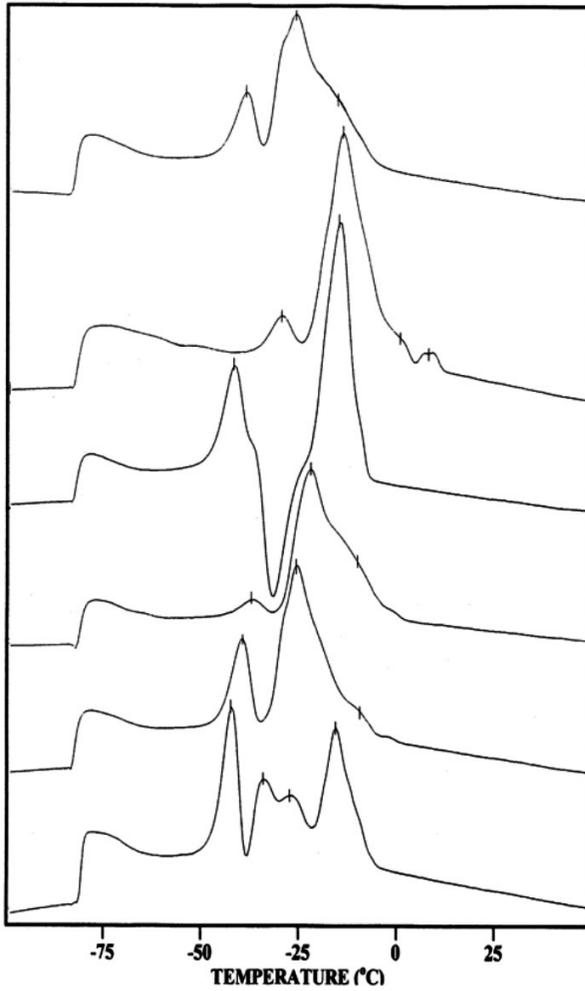


**Figure 4** Estimated surface responses of 3-MCPD levels as a function of (A) water and acid dosage; (B) water and clay dosage; (C) water dosage and deodorization temperature; (D) acid dosage and degumming temperature; (E) degumming temperature and clay dosage; and (F) degumming and deodorization temperature with other factors held at center points (Source: Zulkurnain et al., 2013).

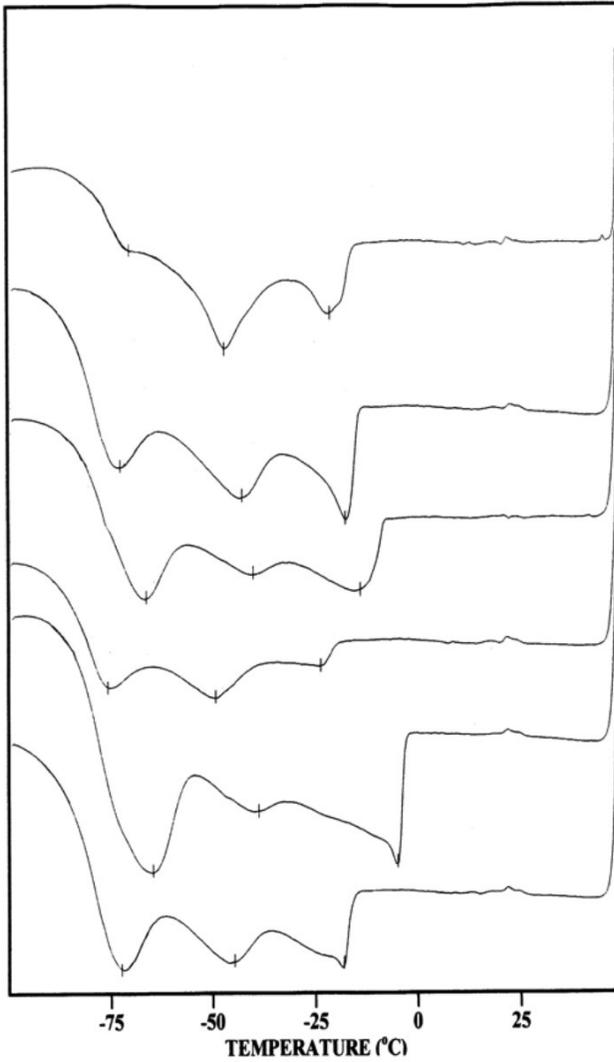
## **Application of DSC in the Analysis of Fats and Oils**

To use fat and oil materials efficiently, it is important to understand the complex structures and properties of these major food components. Heat-related phenomena in fats and oils are of fundamental importance in elucidating their physical and chemical properties. In the field of fats and oils, one major area of application that is eminently suitable for study via DSC is the various melting or crystallisation profiles of vegetable oils. To date, the number of fat and oil compounds whose thermal properties have been studied via DSC is extensive. In most such studies, DSC data have been applied to complement the results obtained using other analytical instruments, such as nuclear magnetic resonance (NMR), X-ray diffraction (XRD), high-pressure liquid chromatography (HPLC) and gas chromatography (GC). For example, Tan and Che Man (2000) have studied the melting and crystallisation behaviour of various vegetable oils via HPLC, GC and DSC.

It is well known that the properties of fats and oils are profoundly influenced by their physicochemical interactions, particularly those among triacylglycerols (TAGs). TAGs are the major chemical species present in fats and oils. DSC is particularly suitable for studying the physicochemical interactions among TAGs because these techniques readily produce phase-equilibrium diagrams which provide a wealth of physicochemical information. Recently, Nehdi and co-workers (Nehdi 2011a; 2011b; 2013; Nehdi et al. 2010) evaluated the thermal properties of various plant oils extracted from selected seeds using DSC, while Tan and Che Man (2000) studied the thermal properties of seventeen different vegetable oils using DSC. The latter study characterised the DSC melting and crystallisation profiles of seventeen edible oils. Figures 5 and 6 present the melting and crystallisation profiles of five different edible oils.



**Figure 5** Differential scanning calorimetry melting curves of corn oil, peanut oil, sesame oil, safflower oil, soybean oil and sunflower oil; from top to bottom (Source: Tan and Che Man, 2000).



**Figure 6** Differential scanning calorimetry crystallization curves of corn oil, peanut oil, sesame oil, safflower oil, soybean oil and sunflower oil, from top to bottom (Source: Tan and Che Man, 2000).

Tan and Che Man (2000) demonstrated that if edible oils yield identical DSC scans at the same scan rate, the DSC technique promises to offer a sensitive, rapid and reproducible fingerprinting method for quality-control purposes. However, the results reported in most scientific literature indicate that one critical limitation of DSC is the dependence of the thermal transition on the scanning rate. Tan and coworkers (Tan and Che Man 2002a; Tan and Che Man 2002b; Che Man and Tan 2002) focused their study on how the measured thermal properties of edible oils are influenced by variations in the DSC scanning rate. In general, an edible oil sample behaves differently depending on the heating/cooling rate of the DSC. These studies concluded that accurate comparisons of calorimetric experiments on vegetable oils can only be performed when the DSC experiments were conducted at the same scanning rate. The use of slow scan rates is advisable in that it minimises instrumental lag in output response.

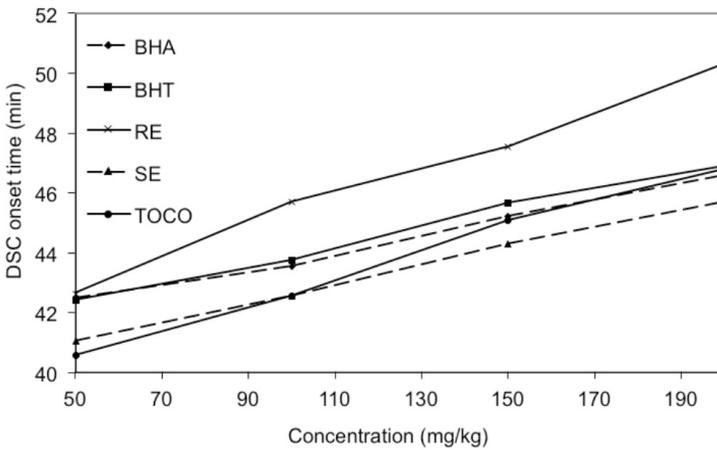
The application of statistical techniques to DSC data is a common indirect method of determining various quality parameters of fat and oil products. Tan and Che Man (1999) have developed a simple and reliable DSC method for monitoring oxidation in three different types of heated oils (corn oil, RBD palm olein and soybean oil). In their study, the heated oils produced simple DSC curves after cooling, each with a single, well-defined crystallisation peak. Based on calibration and validation analyses, their study revealed that a single DSC cooling curve could be used to predict the total polar compounds (TPCs), free fatty acid (FFA) and iodine value (IV) of a heated oil using stepwise regression analysis. In addition, in two separate papers, Tan et al. (2001a; 2002a) have also studied the effects of microwave heating on the DSC cooling and/or melting profiles of edible oils. The influence of the microwave power (low-, medium-, and high-power settings) and the heating time on the lipid

deterioration induced during the microwave heating of the vegetable oils was evaluated. These papers concluded that the DSC method could be employed as an indicator of time and microwave power during microwave heating.

The transfer of an oxygen molecule to an unsaturated fatty acid requires energy (it is an exothermic process). Therefore, the oxidative stability of edible oils can also be established via DSC. The application of DSC for accelerated oil-stability tests has been investigated by several researchers. We have also conducted a comparative study to determine the oxidative stability of twelve different vegetable oils using DSC and an oxidative stability instrument (OSI) (Tan *et al.*, 2002b, 2002c). The DSC technique is based on an isothermal condition, with purified oxygen as the purge gas. The isothermal DSC technique for the direct determination of the oxidative stability of vegetable oils was established, and a comparative study with an OSI has been performed. The DSC cell temperature was set to four different isothermal temperatures: 110, 120, 130, and 140°C. A dramatic increase in evolved heat was observed, with the appearance of a sharp exothermic curve during the initiation of the oxidation reaction. In this method, the oxidative induction time ( $T_0$ ) can be automatically determined via extrapolation of the downward portion of the DSC oxidation curve to the time axis. The results indicated a good correlation ( $P < 0.01$ ) between the DSC  $T_0$  and OSI values.

In our laboratory (Tan *et al.*, 2001a), the isothermal DSC method was applied to obtain kinetic data for the lipid oxidation of ten different vegetable oils. The temperature dependence of the rates of lipid oxidation revealed highly significant correlations when analysed using the DSC method. In addition, based on the *Arrhenius* equation and activated complex theory, the reaction rate constants ( $k$ ), activation energies ( $E_a$ ), activation enthalpies

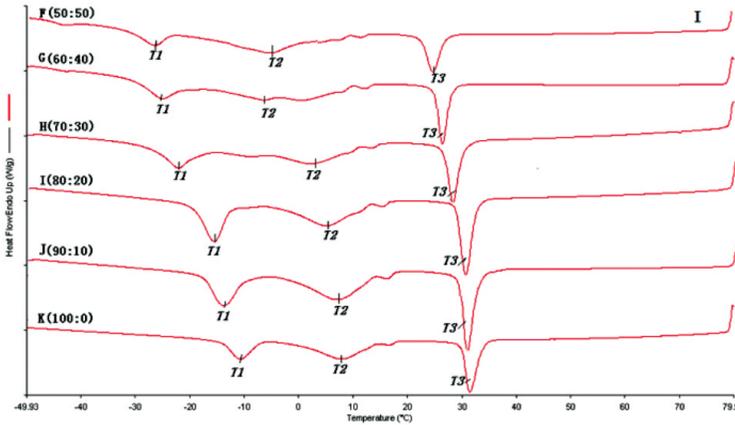
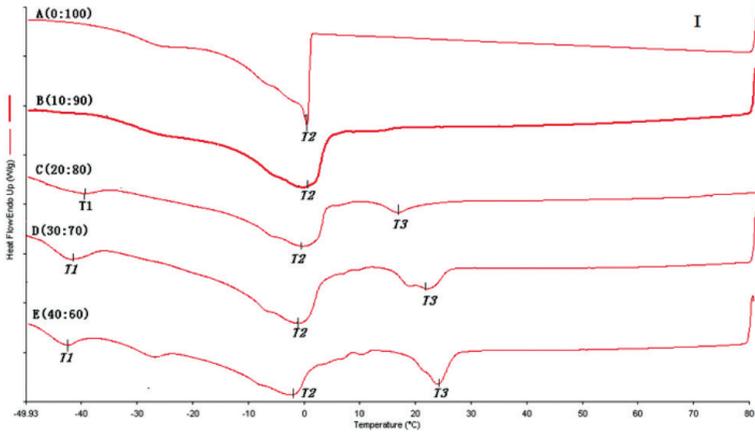
( $\Delta H$ ) and activation entropies ( $\Delta S$ ) were calculated to evaluate the oxidative stability of the vegetable oils. The  $E_a$ ,  $\Delta H$ , and  $\Delta S$  values for all vegetable oils ranged from 79 to 104 kJ mol<sup>-1</sup>, from 76 to 101 kJ mol<sup>-1</sup> and from -99 to -20 J K<sup>-1</sup> mol<sup>-1</sup>, respectively. Based on the same principle, a simple DSC method for measuring the antioxidant activity in RBDPOo was developed in our laboratory (Tan *and Che Man*, 2002c). In the cited study, the oxidation temperature was 150°C, and the oxygen flow at a rate of 50 ml/min. In this method, the thermal changes that occur during the oxidation of the oil are recorded. Generally, the results indicate that the antioxidants act primarily by increasing the induction period ( $T_{on}$ ) for lipid oxidation. Figure 7 presents the  $T_{on}$  values for five natural antioxidants added to RBDPOo at various concentrations. The observed relationship between the increase in induction time with increasing antioxidant concentration is best fitted by a linear or polynomial equation. These calorimetric results indicate that DSC is a valuable technique in the development and optimisation of antioxidant systems for various fats and oils. This work can contribute to the selection of appropriate antioxidants (or combinations of antioxidants) at the optimum levels in various fats and oils.

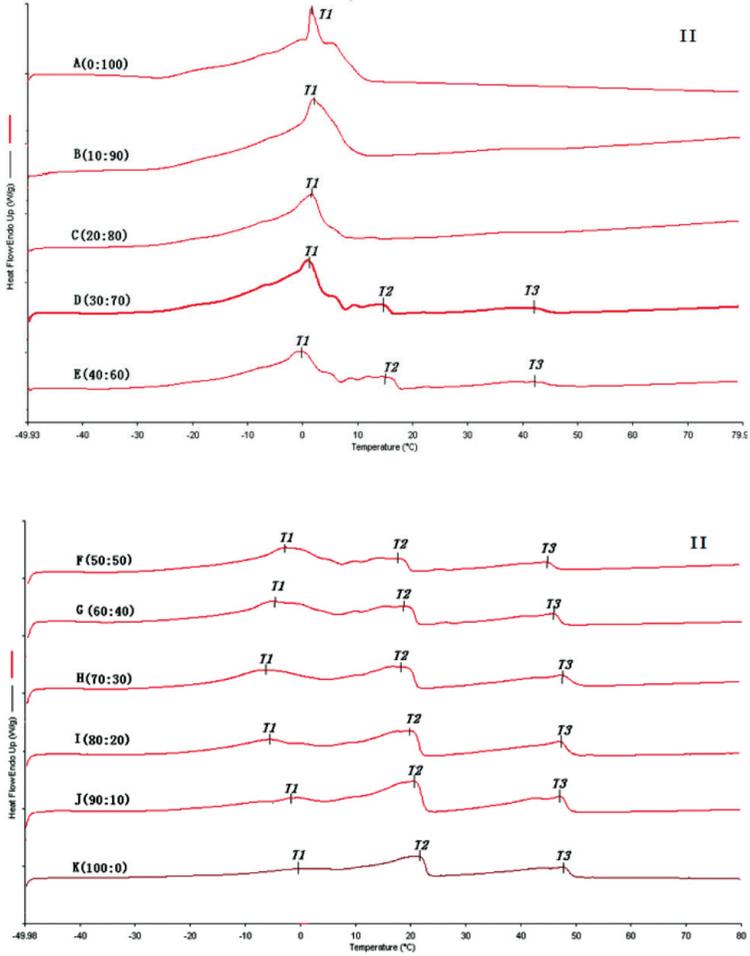


**Figure 7** Scatter plot showing DSC induction time versus antioxidant concentration Abbreviations: BHT, butylated hydroxytoluene; BHA, butylated hydroxyanisole; RE, rosemary extract; SE, sage extract; TOCO, tocopherol.

DSC is also frequently used to monitor the phase transitions of various oil blends. Previous reports (Nor Hayati et al., 2007; Nor Hayati et al., 2009) have indicated that binary blends of palm kernel olein with soybean oil could improve the oxidative and physical stability of soybean-oil-based emulsions to a certain extent. In these studies, DSC is one of the important tools that have been used to evaluate the developed fat/oil blends. For example, DSC has been used to monitor the thermal behaviour of fat blends consisting of palm mid-fraction, palm stearin and virgin coconut oil (Masni et al., 2013). Saberi, Tan and Lai (2011) have also monitored the phase behaviour of palm oil in blends with palm-based diacylglycerol. More recently, our group (Ng et al. 2014) has studied the crystallisation and melting curves obtained via DSC for palm-olein-based diacylglycerol, palm super olein and various

blends thereof (0-100 wt% palm-olein-based diacylglycerol, in 10 wt% increments). These DSC thermal curves are presented in Figure 8. The different characteristic curves of the crystallisation and melting profiles indicate the compositions of the diacylglycerol contents in each of the oil blends.





**Figure 8** DSC crystallization (I) and melting (II) cruves of palm olein-based diacylglycerol (K), palm super olein (A) and their binary blends (B–J) (Source: Ng et al., 2014)

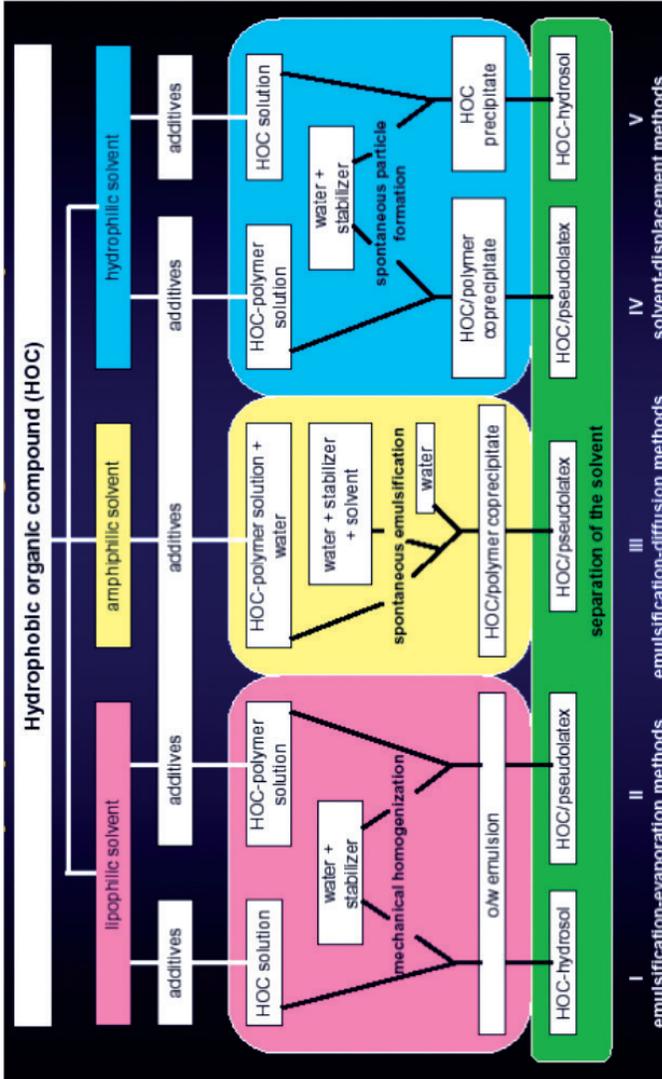
## **THE OUTLOOK FOR FATS AND OILS AT THE NANOSCALE LEVEL**

### **Preparation, Characterisation and Stability of Functional Lipid Nanodispersions**

Functional lipids, such as  $\beta$ -carotene, tocopherols, phytosterols, astaxanthin, lutein, lycopene, natural antioxidants and numerous other compounds, are widely used as active ingredients in various food products. However, the poor water solubility of functional lipids has made their use in food formulations problematic. Most functional lipids are nearly insoluble in water or exhibit very low water solubility. The solubility of functional lipids in food formulations is a major consideration in the food industry. Moreover, functional lipids with low water solubility may be prone to reduced bioavailability. For these reasons, it is important to find solutions to this problem. Nanotechnology provides a good opportunity to improve the solubility of such active ingredients and to increase their bioavailability. Today, nanotechnology is a new frontier in the field of food science and it offers some of the most exciting prospects for technological innovation.

The modern history of nanotechnology began when Richard Feynman, a Nobel laureate, delivered a speech titled “There’s Plenty of Room at the Bottom” in 1959. Today, food scientists are just beginning to try to understand the physicochemical properties of small-sized materials (a leap from the microscale to the nanoscale). Over the past decade, nanotechnology has been explored primarily as a result of the development of new tools that have made the characterisation of nano-sized materials practical and also as a result of various new methods for the preparation of these materials. In the field of pharmaceuticals and medicine, drug particles in the nanometer range exhibit a substantial increase in solubility

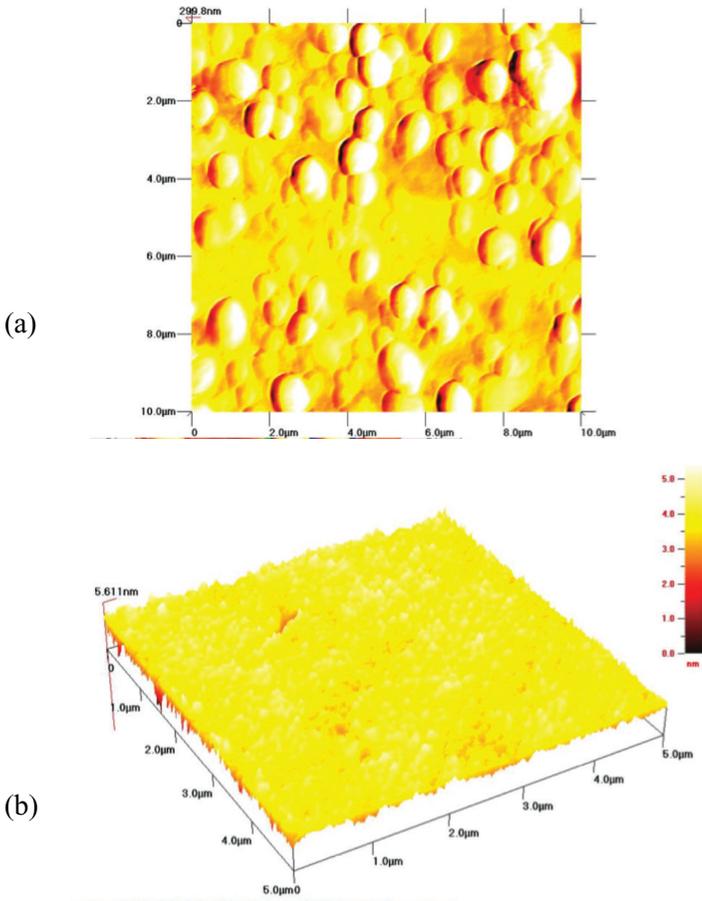
in water, which should lead to improved bioavailability (Grau et al., 2000, Muller et al., 1999 and Trotta et al., 2001). Therefore, over the past decade, considerable effort has been devoted to the preparation of drug nanoparticles of narrow size distributions. The emulsification–evaporation method, a process of emulsification followed by solvent evaporation, is the most widely used technique for the preparation of drug-containing nanoparticles (Kwon et al., 2001). In 2005, this preparative technique was used to prepare  $\beta$ -carotene nanodispersions for food application (Tan and Nakajima, 2005). In the last ten years, this paper has become one of the pioneer publications quoted in the field of food nanotechnology, with more than 110 citations thus far (from [www.scopus.com](http://www.scopus.com)). In addition to the emulsification–evaporation technique, lipid nanoparticles can also be prepared using the emulsification–diffusion and solvent displacement techniques. Figure 9 provides a schematic diagram illustrating all three techniques.



**Figure 9** Precipitation and condensation processes for the preparation of organic nanoparticles. (Source: Horn and Rieger, 2001)

High-pressure homogenisation is extensively used in the food, pharmaceutical and biotechnology industries to emulsify, disperse, mix and process various products (Floury et al., 2000 and Floury et al., 2002). High-pressure homogenisers have frequently been used to prepare various functional lipid nanodispersions in our laboratory (Cheong, et al., 2008, Cheong and Tan, 2010, Cheong et al., 2010, Leong et al. 2009, 2011a, 2011b, 2011c, Anarjan et al., 2010, 2011a, 2012, 2013). In 2008, my team prepared an  $\alpha$ -tocopherol nanodispersion using a high-pressure homogeniser via the emulsification-evaporation technique. This resulted in an  $\alpha$ -tocopherol nanodispersion with a size distribution of 90-120 nm being successfully prepared. In general, it has been demonstrated that high-pressure homogenisers can be used as suitable equipment for the production of  $\alpha$ -tocopherol nanodispersions with narrow size distributions. In 2010, the first paper concerning palm-based functional lipid nanodispersions was published by my team (Cheong and Tan, 2010). In this study, average particles ranging from 95 to 130 nm and from 140 to 210 nm in size were obtained in nanoemulsions that contained palm-based tocopherols and tocotrienols and in nanoemulsions that contained palm-based carotenoids, respectively. However, this study indicated that increasing the energy input beyond moderate pressures (20–80 MPa) and a few cycles (1–3) led to “over-processing” of the droplets. In summary, increasing the operating pressure and applying multiple operating cycles did not necessarily result in the desired fine dispersions. Figure 10 (a and b) presents atomic force microscope (AFM) images of palm-based functional lipid nanodispersions stored for 12 weeks at 4°C. These AFM images reveal the morphologies of the palm-based carotenoid and tocopherol–tocotrienol nanodispersions prepared under the specified homogenisation conditions (40 MPa and two cycles).

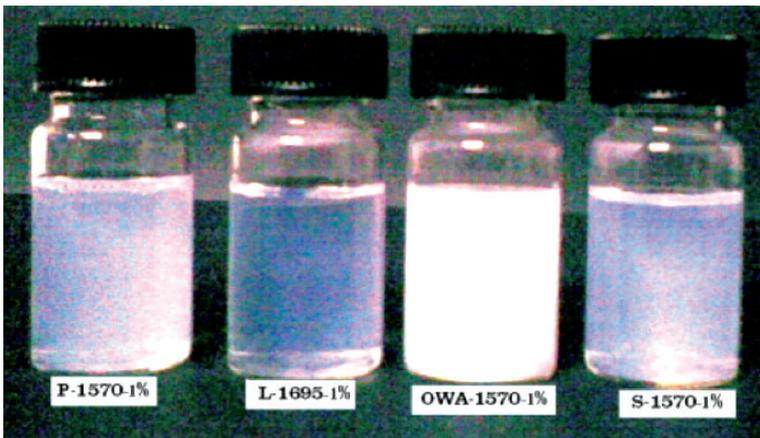
The effect of polyoxyethylene sorbitan esters and sodium caseinate on the physicochemical properties of palm-based functional lipid nanodispersions was evaluated in another study(Cheong et al., 2010).



**Figure 10** AFM images of: (a) carotenoid nanodispersions and (b) tocopherol–tocotrienol nanodispersions sample prepared by the emulsification–evaporation technique (40 MPa, two cycles) (Source: Cheong and Tan, 2010).

Both the emulsification–evaporation and solvent displacement techniques have been used to prepare phytosterol nanodispersions/microemulsions, by Leong et al. (2009, 2011a). Phytosterols, or plant sterols, are a group of naturally occurring functional lipid and steroid alcohols that are found exclusively in plants and are essential constituents of the cell membranes in plants. Over the past decade, the food industry has introduced phytosterols into a number of fat-derived food products, turning these low-nutritional-value products into more expensive, value-added, premium functional food products. Incorporating phytosterols into food products is no simple task, as these sterols are water insoluble and only barely soluble in fats and oils. The high melting temperature of pure phytosterols allows for only very small amounts of phytosterol enrichment. Using the solvent displacement technique, the production of phytosterol microemulsion has been successfully optimised using 5 homogenisation cycles, a homogenisation pressure of 400 bar and an evaporation temperature of 44.5 °C. The parameters for the production of water-soluble phytosterol nanodispersions using the emulsification–evaporation technique have also been optimised (Leong et al., 2011a). Generally, we have successfully optimised the processing parameters for the production of phytosterol-containing nanodispersions by using a mixing time of 15.25 min, a mixing speed of 7,000 rpm and homogenisation pressure of 42.4 MPa, while keeping the formula parameters constant. A more in-depth study of the effects of various sucrose fatty acid esters on the particle characteristics and flow behaviour of phytosterol nanodispersions has been conducted by my research team (Leong et al., 2011b). Sucrose fatty acid esters, commonly called sugar esters, are non-ionic small-molecule emulsifiers that contain a hydrophilic sucrose group and a lipophilic fatty acid group. In this study, sucrose stearate (S-1570), sucrose palmitate (P-1570), sucrose laurate

(L-1695) and sucrose oleate (OWA-1570) were used. Figure 11 shows the phytosterol nanodispersions prepared with 0.5% w/v of phytosterols and 1.0% w/v of various types of sucrose fatty acid esters. In general, the phytosterol nanodispersions prepared via sucrose fatty acid esters at particle sizes of <100 nm exhibited higher clarity. Phytosterol nanodispersions with mean particle sizes ranging from 2.8 to 259.9 nm were successfully produced.



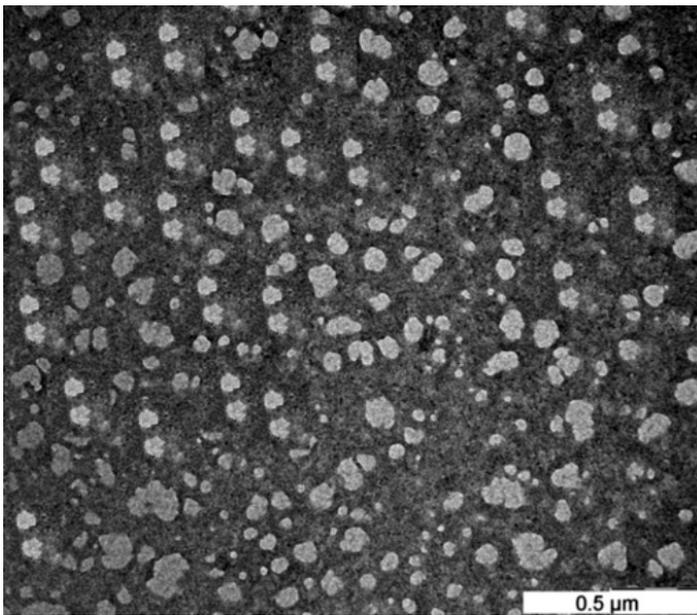
**Figure 11** Phytosterol nanodispersions prepared by 0.5% w/v of phytosterols and 1.0% w/v of different types of sucrose fatty acid esters (Source: Leong et al., 2011b)

It is vital to determine the optimal phase ratio and to explore the primary and secondary homogenisation conditions to produce the smallest possible particle size and a desirable monomodal particle size distribution profile; these factors are also related to the conservation of the costs and energy involved in the production of nanodispersions. We have thus demonstrated the feasibility of producing phytosterol nanodispersions using the emulsification–evaporation technique, although the solvent displacement technique

can also be used to produce phytosterol dispersions with larger particle sizes. In another study, the effects of several factors were examined: four different types of organic phases (hexane, isopropyl alcohol, ethanol and acetone), the organic-to-aqueous phase ratio and conventional homogenisation vs. high-pressure homogenisation (Leong et al., 2011c). The particle sizes produced using the emulsification–evaporation technique were as low as 50 nm.

Astaxanthin (3,3'-dihydroxy- $\beta,\beta'$ -carotene-4,4'-dione) is a ketocarotenoid derived from the oxidation of  $\beta$ -carotene. Its structure contains both ketonic and hydroxylic functional groups, which are responsible for its exceptional antioxidant properties in free-radical scavenging and in singlet-oxygen quenching (Ribeiro et al., 2005). Astaxanthin can be used as a supplement or colorant ingredient in food formulations and as a “nutraceutical”. However, it cannot be readily absorbed by the human body because of its poor bioavailability. The low bioavailability of functional lipids of this type is attributable to their poor water solubility. In a series of 13 papers, we have explored the possibility of increasing the bioavailability of astaxanthin via a nanotechnological approach (Anarjan et al., 2010, 2011a, 2011b, 2012, 2013a, 2013b, 2014a, 2014b, Anarjan and Tan, 2013a, 2013b, 2013c, 2013d, 2013e). This is the most comprehensive research project conducted for the preparation, characterisation and stability evaluation of astaxanthin nanodispersions. A representative TEM image of an optimal astaxanthin nanodispersion is presented in Figure 12. The optimally formulated astaxanthin nanodispersion contained relatively well defined but rather polydisperse quasi-spherically shaped particles, as evidenced by the TEM observation. The observations closely corresponded with the results obtained in the dynamic-light-scattering particle-size analysis.

The influence of the processing conditions — namely, the pressure degree of the high-pressure homogeniser (20–90 MPa), the number of passes through the homogeniser (0–4) and the evaporation temperature (16–66 °C) — on the physicochemical properties of the prepared astaxanthin nanodispersions were evaluated using a three-factor central composite design (Anarjan et al., 2011a). Multiple-response optimisation predicted that three passes through the high-pressure homogeniser at 30 MPa for the preparation of the astaxanthin nanoemulsion followed by the removal of the solvent from the system via evaporation at 25 °C yielded astaxanthin nanodispersions with the optimal physicochemical properties.



**Figure 12** Transmission electron micrograph of optimum astaxanthin nanoparticles in water (optimum astaxanthin nanodispersion)

The efficacy of select polysorbate and sucrose ester emulsifiers as well as four select polysaccharides in the preparation of astaxanthin nanodispersions has been investigated (Anarjan and Tan, 2013a, 2013b, Anarjan et al., 2013a, 2014b). In the case of small-molecule emulsifiers, the results indicated that the emulsifiers with higher HLB values (higher hydrophilicities) and shorter fatty acid chains produced nanodispersions with smaller particle sizes. Polysorbate 20 was found to be the most appropriate small-molecule emulsifier for use in the preparation of astaxanthin nanodispersions. It was also found that generally, hydrocolloids produced nanodispersions with larger average particle sizes, higher PDIs and less astaxanthin content than did proteins and small-molecule-stabilised nanodispersions. GA was identified as the most suitable polysaccharide for use in the preparation of nanodispersions.

The design of food emulsions and dispersions for the delivery of lipophilic bioactive compounds should include an estimate of their bioavailability to support the claimed effect. The cellular uptake of astaxanthin from nanodispersions has been investigated by my team using colon carcinoma cells (HT-29) as a model for human colon epithelial cells (Anarjan et al., 2011). The cellular uptake of astaxanthin was found to increase with decreasing particle size and with *trans* to *cis* isomerisation. The mean particle size of the nanodispersions exhibited the most significant effect (i.e., the lowest *p* value and highest *F* value) compared with the other predictors. Between the *cis* isomerisations of astaxanthin, the 13-*cis* isomerisation affected the cellular uptake of astaxanthin from nanodispersions more strongly than did the 9-*cis* isomerisation.

The stability of optimally formulated astaxanthin nanodispersions under various environmental and storage conditions has been evaluated. The nanodispersions demonstrated

some physical instability against intense heat treatment, acidic pH and the addition of calcium ions as well as under simulated *in vitro* gastric and intestinal conditions. Chemical instability for nanodispersions stored at various temperatures and under various lighting and atmospheric conditions has also been monitored (Anarjan and Tan, 2013c, 2013e). The astaxanthin degradation rates under all conditions were observed to obey first-order kinetics with relatively high coefficients of determination ( $R^2 > 0.95$ ). In the final part of this project, the stability for astaxanthin nanodispersions diluted in orange juice and skim milk as model food systems and in deionised water as a control were evaluated (Anarjan and Tan, 2013d). In general, the nanodispersions were found to be more stable in the model foods than in water.

## CONCLUSION

In this book, the author has examined various aspects of fats and oils at the macro-, micro- and nanoscales. Fats and oils, together with proteins and carbohydrates, constitute one of the three major classes of food nutrients. In addition to being the most concentrated source of dietary energy, fats and oils contribute to the texture characteristics and flavour reception of a wide variety of foods. The characterisation and exploitation of all the roles played by fats and oils in a dietary context require comprehensive understanding of their physicochemical properties. Continuous improvements in our understanding of the physicochemical properties of this major class of food nutrients will play an important role in the sustainable development of fats and oils as a major agriculture commodity. In addition, the various safety and health issues related to fats or oils are also closely related to the basic chemical components of various refined fats and oils. Recent safety issues, such as the presence of 3-MCPD esters in refined edible fats and oils, should also be

addressed through in-depth studies of the interactions of basic components of unrefined oils with external precursors.

Oilseeds are important sources of oils with economical, nutritional, and industrial importance. Non-conventional oilseeds which are being explored due to their unique physicochemical properties may augment the supply and consumption of fats and oils in the future. The study of major and minor components of oilseeds is useful to facilitate the effective use of both oils and their minor constituents. Based on the number of recent publications in this field, it is clear that research in this area will continue to flourish. The subtle difference between a fat and an oil is that a fat is solid at room temperature whereas an oil is liquid. Beyond this subtle difference, the functionality of fats and oils is determined by their physicochemical properties and their degree of processing. There have been considerable advances in our understanding of the physicochemical and functional properties of fats and oils over the past few decades. The broad diversity of the physicochemical and functional properties exhibited by fats and oils is the result of the product formulations and processing conditions used to create them. However, the development of new product formulations, the improvement of existing products and the establishment of efficient processing operations for the fats and oils industry will require a more systematic and rigorous approach than has been used previously.

Many novel physicochemical properties of fats and oils may be determined through the development of advanced analytical instruments such as the DSC. The DSC thermal profiles of fats and oils constitute an interesting area of research. DSC can be applied as a common analytical tool that is capable of detecting many different physicochemical properties of fats and oils. The versatility of DSC will allow us to gain a deeper knowledge, at the molecular level, of

the mechanisms underlying the macroscopic processes related to fats and oils. Furthermore, DSC may also become a unique tool for quality control, as its sensitivity to structural changes can provide useful insight for analysis.

The era of nanotechnology that we are now entering presents an unprecedented opportunity to understand the emergence of new properties of fats and oils on the nanometer scale. In future, nanotechnology will play an important role in Malaysian research and innovation. In the field of fats and oils, the use of nano-sized functional lipids in specific foods may lead to attractive advantages, such as better solubility, higher bioavailability, improved properties, or better quality of the final product. Although the potential for the development of novel food ingredients using nanotechnology is apparent, the attendant risks must be mitigated. Understanding the long-term safety consequences of nanotechnology products is critical. It will thus be necessary to develop *in vitro* and *in vivo* tests to predict human reactions to nano-sized functional lipids.

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## **BIOGRAPHY**

**Tan Chin Ping** was born in Klang, Selangor. He received his degrees, Bachelor of Food Science and Technology with First Class Honours in 1998 and PhD in Food Processing in 2001, from Universiti Putra Malaysia (UPM). During his PhD studies, he was awarded the AOCS Honored Student Award in 2000 and IUFOST's Student Travel Grant a year later. Tan began his career at the Department of Food Technology, Faculty of Food Science and Technology, Universiti Putra Malaysia in 2001 in the area of food processing and fats and oils technology. He then served as a JSPS Postdoctoral Fellow at the National Food Research Institute in Japan from September 2002 to August 2004. He was promoted to Associate Professor in 2008 and full professor in 2012. Currently, he is the head of the Department of Food Technology. He is currently leading one of the major research programs at UPM, Fats and Oils Technology. He is also the leader of the fats and oils research group in the Faculty of Food Science and Technology, UPM. To date, he has published one joint-edited book, seven book chapters and over 190 scientific articles in peer-reviewed journals, has filed five patents and has presented more than 200 papers at various national and international conferences. Furthermore, Prof. Tan's research impact can be observed from the citation data provided by Scopus and Google Scholar. His published works have been cited more than 2300 times, and his h-index is 27 as of 1 October 2014 (from [www.scopus.com](http://www.scopus.com)).

As a lecturer at the Department of Food Technology, he has been assigned to teach various undergraduate and postgraduate courses in three different academic programs, namely, the Bachelor of Food Science and Technology, BS (Food Studies) and Master of Food Technology programs. These courses include principles of food processing and preservation, chemistry and technology for

plant- and animal-based products, product development, the sensory evaluation of food, fats and oils chemistry and technology and food innovations. By virtue of his teaching and research experience in food science and technology, Tan has been appointed to the Expert Review Panel for the BS (Food Science and Technology) program at Monash University; the Board of Advisors for the School of Hospitality, Tourism and Culinary Arts (SHTCA) at KDU University College; and the Programme Advisory Panel for Food Science at Tunku Abdul Rahman University College. He is also an external examiner for the SHTCA at KDU University College, KDU College Penang, Nottingham University and UIA, among others.

His areas of research specialisation are palm oil, food nanotechnology, food emulsions and the extraction of bioactive compounds from various agricultural by-products. In these research areas, he is responsible for managing 20 research projects for various government and private agencies with a total funding of more than RM7.5 million. In addition, Tan has secured more than 20 consultation projects from various private companies and government agencies. These projects are focused on new product development, the development of value-added processes and products and quality and safety issues related to fats and oils. In relation to international research grants, his research work on carotenoid nanodispersions has been awarded approximately SAR2 million (RM1.7 million) by the Kingdom of Saudi Arabia.

Tan has supervised 23 PhD and 14 MSc students. Of these, 9 PhD and 10 MSc students have since graduated. In addition, he has also been involved with the co-supervision of more than 35 PhD and 30 MSc students, of whom 22 PhD and 20 MSc students have graduated. The supervision of these students has spanned various departments and faculties. Many of the PhD and MSc students with whom he has worked most closely have won various national

and international awards, such as the international Alltech Young Scientist Award, the Merck Young Scientist Award, IUFoST's Student Travel Grant, ICoFF's Student Travel Grant and the Student Award for AOCS's Edible Applications Technology Division.

At the national level, Tan is the Chairperson for SIRIM's Working Group on Eco-labelling Food Grade Lubricants and participates on the expert panels of many working groups related to coconut products. In addition, he is also the external assessor for promotion to the Associate Professor position at public universities, an assessor of many research proposals submitted to government agencies and a member of the project monitoring team for various TechnoFund projects managed by the Ministry of Science, Technology and Innovation of Malaysia. In addition, he also serves on the expert panel of the Expert Working Committee for Food Commodities, established under the Advisory Committee for Food Regulations 1985.

He has also provided research-based consulting advice to a range of food companies and the Food Quality and Safety Division of the Ministry of Health, Malaysia. Tan also assesses food safety issues related to various lipid-based products. The quality and safety of various heated oils, such as frying and microwave-heated oils, are his main research focuses in this area. His has been recognised by the Ministry of Health as a major food analyst for lipid-related food products. To date, he has written more than 10 classified reports related to food safety issues concerning fat and oil products for the Ministry of Health. He has also presented five expert review reports to the Minister of Health during the 6<sup>th</sup> to 10<sup>th</sup> Meetings of the National Food Safety and Nutrition Council.

Tan has received more than 40 national/international research awards throughout his research career. In recognition of his high-quality research, he was awarded the Young Scientist Award, the

Excellent Researcher Award and the Scientific Paper Award by UPM in 2006, 2011 and 2013, respectively. He was also the recipient of the 2010 ProSPER.Net Scopus Young Scientist Award, was a TWAS Young Affiliate for 2010-2014 and was awarded the German S&T Fellowship in 2010. He was recognised as one of the Top Research Scientists Malaysia (TRSM) by the Academy of Sciences Malaysia (ASM). He is currently one of the members of the Young Scientists Network of the Academy of Sciences Malaysia.

Tan has the opportunity to contribute internationally through his research activities. He has been one of the scientific advisors to the International Foundation for Science (IFS) in Sweden since 2008, in the area of food science and technology. Since 2011, he has been appointed as a Visiting Professor at the College of Science, King Saud University, Kingdom of Saudi Arabia. In an effort to share his expertise, he has been frequently invited to give talks by various agencies in Malaysia, Indonesia and Japan. Tan has consistently assisted many foreign agencies in evaluating and improving research proposals received from various developing countries around the world. In addition, he is member of the editorial boards of a number of journals (PeerJ, International Food Research Journal, the Scientific World Journal and Journal of Engineering) and has organised workshops, symposia and conferences in the fields of food nanotechnology, palm oil, food safety and food science and technology. As an academic, he has frequently contributed to the scientific community through his role as a regular reviewer of many manuscripts submitted to international refereed journals. Tan is the only Malaysian on the advisory board of the International Symposium of Agriculture and Food Applications of Nanoscale Science (ISAFANS).

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My gratitude to the late Professor Dato' Dr. Yaakob Che Man, my academic advisor, PhD supervisor and mentor, without whose encouragement and guidance I would perhaps not be sharing these fruits of knowledge with you today.

Having spent my entire working life at Universiti Putra Malaysia, I have had the opportunity to witness its development since my undergraduate days. I am forever grateful to Universiti Putra Malaysia for nurturing me all these years.

To my Dean, Prof. Dato' Dr. Mohd. Yazid Abd Manap, I say thank you. To all the former Deans of the Faculty of Food Science and Technology and Heads of the Department of Food Technology, I bow and doff my cap to all of you. Additionally, I wholeheartedly register my sincere thanks to all the staff members of the Faculty of Food Science and Technology for the support each has accorded me, which, in one way or the other, has propelled me to my current status.

I am grateful to all my postgraduate students, too numerous to mention, who bore the brunt of my development. I say, thank you. Your contributions at the various stages of my academic career are highly acknowledged.

During the past 13 years or so, I have met people who have inspired me and made my work pleasant when interacting with them. Many of you have supported me in various ways, and your kindness cannot go unacknowledged. However, it is not possible to individually thank each and every one of you. So, I say thank you all for always being there for me.



Tan Chin Ping

## LIST OF INAUGURAL LECTURES

1. Prof. Dr. Sulaiman M. Yassin  
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22 July 1989
2. Prof. Ir. Abang Abdullah Abang Ali  
*Indigenous Materials and Technology  
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30 August 1990
3. Prof. Dr. Abdul Rahman Abdul Razak  
*Plant Parasitic Nematodes, Lesser  
Known Pests of Agricultural Crops*  
30 January 1993
4. Prof. Dr. Mohamed Suleiman  
*Numerical Solution of Ordinary  
Differential Equations: A Historical  
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11 December 1993
5. Prof. Dr. Mohd. Ariff Hussein  
*Changing Roles of Agricultural  
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5 March 1994
6. Prof. Dr. Mohd. Ismail Ahmad  
*Marketing Management: Prospects  
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6 April 1994
7. Prof. Dr. Mohamed Mahyuddin Mohd.  
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*The Changing Demand for Livestock  
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20 April 1994
8. Prof. Dr. Ruth Kiew  
*Plant Taxonomy, Biodiversity and  
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9. Prof. Ir. Dr. Mohd. Zohadie Baraie  
*Engineering Technological  
Developments Propelling Agriculture  
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11. Prof. Dr. Abdul Salam Abdullah  
*Natural Toxicants Affecting Animal  
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12. Prof. Dr. Mohd. Yusof Hussein  
*Pest Control: A Challenge in Applied  
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9 July 1994
13. Prof. Dr. Kapt. Mohd. Ibrahim Haji  
Mohamed  
*Managing Challenges in Fisheries  
Development through Science and  
Technology*  
23 July 1994
14. Prof. Dr. Hj. Amat Juhari Moain  
*Sejarah Keagungan Bahasa Melayu*  
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15. Prof. Dr. Law Ah Theem  
*Oil Pollution in the Malaysian Seas*  
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16. Prof. Dr. Md. Nordin Hj. Lajis  
*Fine Chemicals from Biological  
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21 January 1995
17. Prof. Dr. Sheikh Omar Abdul Rahman  
*Health, Disease and Death in  
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*Fish Health: An Odyssey through the Asia - Pacific Region*  
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*Chromosome Distribution and Production Performance of Water Buffaloes*  
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20. Prof. Dr. Abdul Hamid Mahmood  
*Bahasa Melayu sebagai Bahasa Ilmu-Cabaran dan Harapan*  
10 June 1995
21. Prof. Dr. Rahim Md. Sail  
*Extension Education for Industrialising Malaysia: Trends, Priorities and Emerging Issues*  
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*The Diminishing Tropical Rain Forest: Causes, Symptoms and Cure*  
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*Management of Highly Weathered Acid Soils for Sustainable Crop Production*  
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*Pesticide Usage: Concern and Options*  
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*Microbial Fermentation and Utilization of Agricultural Bioresources and Wastes in Malaysia*  
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28. Prof. Dr. Wan Sulaiman Wan Harun  
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