Thermal Analysis of Lipid Crystallization and Water Ice on Coconut Milk Emulsions: Effect of NaCl Concentrations

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***Abstract*—The aim of this study was to investigate the effect of NaCl concentrations at low temperature (50 to -80°C) on lipid crystallization and freezing water of coconut milk emulsions. Phase change behavior of coconut milk emulsions has been evaluated using differential scanning calorimetry at heating/cooling rate of 2°C/min. Unhomogenized coconut milks (34% fat content) showed an overlapping peak of fat crystals with an onset at 8°C and a large exothermic peak of freezing water at -20°C during cooling. Reheating sample leads to water ice melt at 0°C and subsequently a melting of coconut fat with an endset of 25°C. Commercial coconut milks (18 – 20% fat content) exhibited more than three peaks of crystallization due to a variety of oil droplet sizes in emulsions. To study the effect of salt, NaCl (0 – 4%) were then added to coconut milk (20% fat content) containing 5%w/w acacia gum. Homogenized coconut milk emulsions showed lower crystallization and melting temperatures of water ice in the presence of NaCl. However, it does not affect phase change of fat crystals. Freezing depression could be seen only on aqueous phase of its emulsion.**

***Index Terms*—coconut milk, crystallization, differential scanning calorimetry, melting**

I. Introduction

Coconut milk, a milky white oil-in-water emulsion, naturally extracted from endosperm of coconut. Fresh coconut milk contains about 30 - 35% fat and solid non-fat referring to carbohydrate, protein, vitamin, and etc. [1]. Coconut fat became a majority composition in coconut milk. It contains more than 90% saturated fatty acid. 70% of total fatty acid is short or medium chain fatty acid that consists of primarily 44 - 60% lauric acid, followed by 15 – 20% myristic and 4 - 8% caprylic acid [2]-[4]. The remaining are oleic and linoleic acid which are 6% of monounsaturated fatty acid, and 2% of polyunsaturated fatty acid [5].

Commercially, coconut milk was treated by thermal process to ensure safety from pathogen. During storage while exporting, coconut milks experience cooling conditions with a wide range of temperature. This would lead coconut milk to undergo heating and cooling cycles. The change in temperature causes the crystallization of coconut milk which accelerates emulsion instability and induces an undesirable solid-like coconut cream on top at low temperature storage. To monitor the crystallization behavior, differential scanning calorimetry (DSC) is one of an effective methods, normally used to study phase transition of emulsion that associated with the energy change and heat release [6]-[8]. However, only rare research has been found to investigate phase change behavior of coconut milk emulsion. Studied by [9], the researchers have been determined the DSC thermograms of 10% fat coconut milks containing several food emulsifiers which experience heat-cool cycles and its effect on emulsion instability. Our experiment, however, would like to investigate phase change behavior of fresh and commercial coconut milks which was aimed to determine the crystallization and subsequent melting profiles to be better understanding the effect of low temperature storage and effect of added NaCl onto 20% fat to depress freezing of stabilized coconut milk emulsions.

II. Materials and Methods

A. Materials

Fresh coconut milk was purchased from local market in Bangkapi district, Bangkok, Thailand. It was extracted directly from the fresh grated coconut meat without adding water under hydraulic press. The fat content was determined using Babcock method (adapted from AOAC official method 989.04) [10].

Commercial coconut milks (UHT and can) from different manufactures in Thailand were selected by top market share and used in this experiment. They contain fat content between 18–20%. NaCl (99.5%) was purchased from Rungsup Chemical Ltd., Thailand. Acacia gum, purchased from Jumbo Trading CO.,Ltd., Thailand, was used to stabilize coconut milk emulsion.

B. Light Microscopic Method

Fresh coconut milk was diluted by distilled water at a ratio of 1:20. The emulsion structure was observed and taken the photo using a microscope Eye-piece camera (AM 423X, ANMO Electronic Corporation, Taiwan), connected with a digital microscope (Nikon microscope

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YS2-H, Nikon Corporation, Japan). The magnification is at 1200x (30x from eyepiece magnification with a 40x objective lens).

C. Emulsion Preparation

Fresh coconut milk (32.74 ± 2.07 % fat content) was diluted using distilled water to contain final fat content of 20%. 5% w/w of acacia gum (ACG) was used as stabilizer by adding 30% ACG-solution into coconut milk solution prior homogenization. To study the effect of NaCl concentrations onto the phase transition using differential scanning calorimetry (DSC), NaCl solutions was added to obtain final concentrations of 0%, 2% and 4% w/w. The emulsions were then homogenized using rotor-stator homogenizer (Ultra-Turrax IKA® T-18 basic, IKA-werke GmbH & Co. KG, Germany) at 17,500 rpm for 3 min. Fresh prepared samples were then determined thermal properties using DSC measurement. All measurements were done at least in duplicate.

D. Differential Scanning Calorimetry

Thermal analysis on coconut milks including fresh, UHT and canned coconut milks were investigated using differential scanning calorimetry (DSC-1, Metller-Toledo (Thailand), Inc.). Samples were weighed (14-15 mg) into aluminum pans and hermetically sealed. All samples were run against empty pan as the reference. The calibration was done with the standard procedure according to manufacturer user manual. Prior measurement, samples were heated to 50°C in the DSC chamber to ensure the complete melt with absence of coconut fat crystal in the coconut milk samples. DSC profile was then applied to the samples by (1) cooling process from 50°C to -80°C at 2°C/min, and then (2) kept constant at -80°C for 5 min before (3) reheated to 50°C at the same rate. The heat flow was recorded and expressed in form of DSC thermal curve as a function of temperature. During measurement, nitrogen gas was purged and flowed with 200 cc/min. Each DSC scan was made with a new sample to ensure the same thermal history.

The manufacturer's software program (STARe Evaluation Software version 10.00, Mettler-Toledo AG, copyright© 1993 - 2011) was used to plot thermal curve and to analyze the thermal data. The transition temperatures were identified to each thermal curve, based on the temperature at which a change of the value of heat flow occurred. The onset temperature (temperature at which the extrapolated basis line intersects the extrapolated slop in the first transition state.) and the endset temperature (temperature at which the extrapolated leading edge of the last peak intersects with the basis line), the peak temperature (temperature at which the temperature is at peak or highest point of the curve). The crystallization and melting temperatures were determined according to the onset and endset temperatures on the cooling and heating processes, respectively.

III. Results and Discussion

A. DSC Profile of Fresh Coconut Milk

Differential scanning calorimetry was conducted on unhomogenized fresh coconut milk (32.74 ± 2.07 % fat content) at 2°C/min.

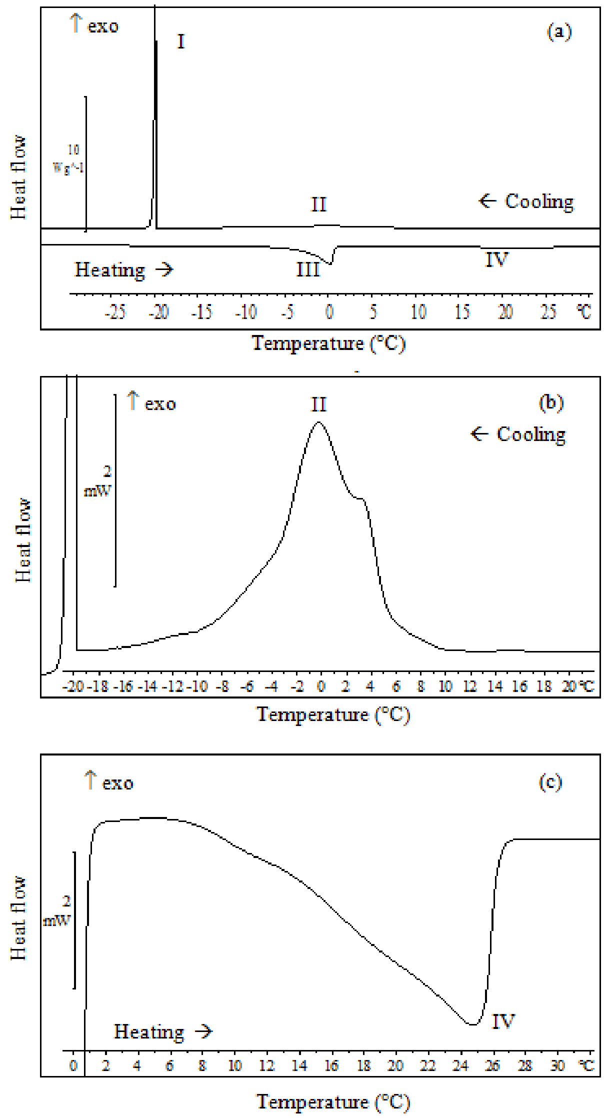

Figure 1. DSC curves of fresh coconut milk. Samples were cooled from 50°C to -80°C at 2°C/min and subsequent heating at the same rate; where (a) represents all peaks, (b) represents peak II when magnified, and peak (c) represents peak IV when magnified.

Fig. 1a showed the DSC profile during cooling and heating process. A multiple peaks can be classified into two peaks of crystallization (I and II) and two peaks of melting processes (III and IV). The peak II and IV has been magnified to the width bar of 2 mW, and determined to present peak details during cooling (Fig.1b) and heating (Fig. 1c) processes, respectively. During cooling, a multiple crystallization peaks have been shown with dramatically one high exothermic peak (I) of heat flow (Fig. 1a), with extrapolated onset of -20.01°C to endset of -20.47°C. This would represent the freezing process of water. This was similar to the results reported by [9].

The results in phase change profile were similar to Tungsuphoom and Coupland (2009) [9]. The researchers stated that coconut milk showed two endothermic peaks and two exothermic peaks where the crystallization of oil in coconut milk was higher than water and water was melted at 0 °C before coconut fat. However, the transition temperatures could not be comparable because difference cooling and heating rate was used.

Before the freezing of water, there was a small overlapping peak (II) at the beginning of crystallization (Fig. 1b). This could express a crystallization of fat and oil in coconut milk emulsion. An overlapping could be explained by the nucleation of different oil droplet sizes. Stated by [11], the intermediate oil droplet sizes crystallized for multiple peaks where heterogeneous coarse emulsion crystallized at higher temperature than homogeneous fine emulsion. Oil droplet size of fresh coconut milk was then investigated to confirm the non-uniform particles using light microscopic method. The optical micrograph (Fig. 2) showed that a boarded band of individual large and small oil droplets found in fresh coconut milk at room temperature. Another explanation would be the nucleation of different free fatty acid of coconut milk. Our previous study founded that coconut oil induce at least two peaks on DSC curves owing to saturated medium chain and unsaturated long chain fatty acid [12], consequently.

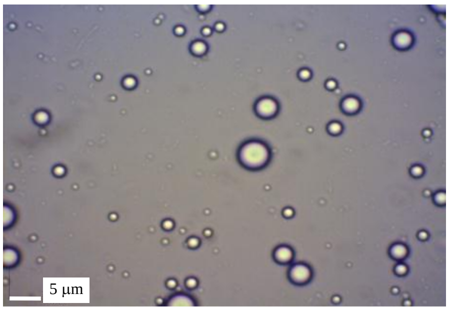

Figure 2. Optical micrograph (1,200x) at dilution 1:20 of fresh coconut milk. The width of the bar corresponds to 5 µm.

During heating, there was a high endothermic peak (III) corresponded to melting of free water in emulsion. A small peak (IV) after water melting showed only one non-smoothed peak due to melting of coconut fat. The effect of oil droplet sizes would diminish and was not keen to be seen (Fig. 1c).

Considering the average of transition temperatures, when cooled coconut milk from 50°C, coconut milk emulsion starts to crystallize at 5.55± 0.03°C (onset) with endset temperature at – 4.99± 0.25°C (peak II), then the freezing of water occurred. When reheated samples the melting of water has been first achieved. The melting of coconut fat in sample was then occurred at 11.53±1.17°C (onset) to 26.26± 0.04°C (endset).

B. *DSC Profile of Commercial Coconut Milk*

The investigation on thermal properties has also been done on two brands of UHT coconut milks and a canned coconut milk manufactured in Thailand. Generally, the results showed that a high exothermic of freezing water was presented at the same onset temperature (-17 to -21°C) (data not shown). Multiple crystallization peaks of oil droplets in commercial coconut milk emulsions was clearly seen (Fig. 3a). Throughout sterilizing process and storage, flocculation and coalescence in emulsion are generally occurred. Different oil droplet size distribution is consequently obtained. This would affect of unpredictable multiple overlapping peaks in samples. More than three crystallization peaks has been exhibited (Fig. 3a). Extrapolated onset of UHT-1, UHT-2 and canned coconut milks are 2.03±0.39°C, 1.41±0.39°C and 4.51±0.01°C, respectively. The endset, however, showed relatively similar which are -14.96±0.94°C, -13.60 ±0.87°C and -12.72±0.24°C for UHT-1, UHT-2 and canned coconut milks, respectively.

Concerning to the cooling effect on coconut milk quality, samples showed a wide range of crystallization temperature with relatively high temperature of onset. This would result in a quick freezing and packing of coconut cream in emulsion above ice freezing point. This will also accelerate emulsion instability [9] and will induce an undesirable appearance in coconut milk product.

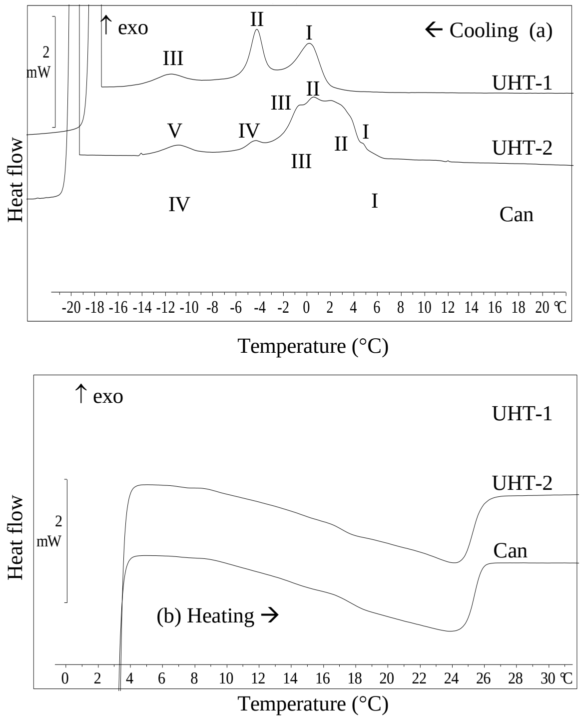

Figure 3. DSC curves of commercial coconut milks; (a) during cooling and (b) during subsequent heating processes.

Completely crystallized coconut milks (-80°C) was then reheated to study the melting behavior at the same rate of 2°C/min. The result was curved from endset temperature of water ice melt at approximately 4°C to endset temperature of oil droplets (Fig. 3b). Surprisingly, even the crystallization process of commercial coconut milks was different but the same melting curves were obtained. No different between samples has been detected by DSC profiles. All samples showed one melting peak starting to melt immediately above ice melting point and ended at the same temperature. Extrapolated endset of samples are 25.69±0.02°C, 26.07±0.08°C and 25.90

±0.02°C for UHT-1, UHT-2 and canned coconut milks, respectively. These results could lead to the assumption that either the differences in free fatty acid composition or in oil droplet sizes of an emulsion, are not effect to the melting temperature of fat crystals in coconut milks. Coconut milks experienced the same cooling rate could produce similar fat crystals morphology and consequently provided the same melting profile with an identical endset temperature at 25°C in all samples. These were also correlated to our previous study, where the coconut oil structure is reversible under heat-cooled processes [2]. The same microstructure, detected by oscillation test, has been observed before and after heat-cooled cycle.

Notice that coconut milk emulsion crystallized above ice point. It would lead to the confirmation that an undesirable crystallized coconut cream would occurred during low storage and transportation while exporting.

C. *Effect of NaCl Concentrations*

To study the effect of NaCl concentrations, coconut milk was diluted to a final fat content of 20%. The addition of 5% w/w acacia gum before homogenization was to stabilize emulsion and to avoid phase separation during DSC measurement. The results showed that without NaCl free water in coconut milk emulsion froze at higher temperature than those containing NaCl (Fig. 4).

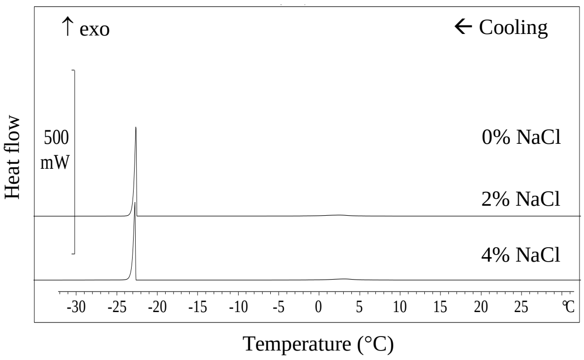

Figure 4. DSC curves of freezing water in coconut milk emulsions at different NaCl concentrations during cooling process at 2°C/min.

In presence of NaCl, a freezing point of water in emulsion depressed from -16°C to -22°C (Table I). It lowers freezing temperature due to colligative property of dissolved NaCl [6]. The freezing point depression, however, showed no different between different concentrations of NaCl. Increasing in NaCl from 2% to 4% exhibited the same freezing point of water in emulsion (Fig. 5). However, all samples showed the same crystallization peak of oil droplets. Onset temperatures of samples are similar and closed to 5°C (Table I). This could lead to a conclusion that the addition of salt does not effect to the crystallization of coconut oil.

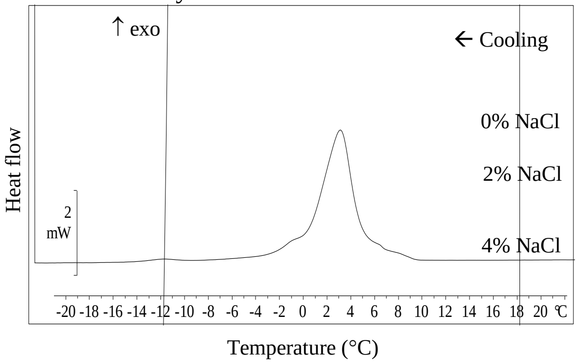

Figure 5. DSC curves of lipid crystallization in coconut milk emulsions at different NaCl concentrations during cooling process at 2°C/min.

When reheating, coconut milk emulsions exhibited more than two peaks of melting (Fig. 6). In the absence of NaCl, coconut milk emulsion showed a smooth single peak of melting of water ice. When adding 2% NaCl, the melting peak was shifted to lower temperature. Increasing NaCl concentrations (4%), more peaks have been observed. Also, peak enthalpy (heat flow) was lowering by the addition of NaCl. These could be suggested that faster melt of water ice was induced by the presence of NaCl in emulsion.

At 4% w/w of NaCl, at least three endothermic peaks have been exhibited. A small peak at temperature below freezing point of water has been observed. The presence of NaCl, however, does not effect to the melting of oil droplets in all coconut milk samples. The melting profile of oil droplets at high temperature still remains the same. This could be because NaCl was dissolved only in aqueous phase of emulsion (dissolved in water) but not in dispersed phase of oil droplets.

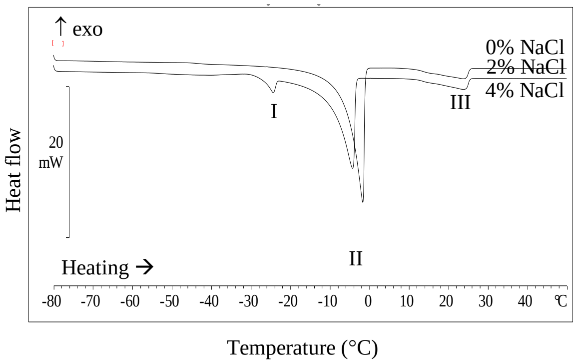

Figure 6. DSC melting curves of coconut milk emulsions containing different NaCl concentrations during heating process at 2°C/min

TABLE I. TRANSITION TEMPERATURES (°C) OF ACACIA GUM-STABILIZED COCONUT MILK EMULSIONS CONTAINING DIFFERENT SODIUM CHLORIDE CONCENTRATIONS DRUING COOLING AND SUBSEQUENT MELTING AT 2°C/MIN

NaCl concentration	Transition temperature (°C)								
	Crystallization				Melting				
	onset	endset	Peak I	Peak II	onset	endset	Peak I	Peak II	Peak III
0%	5.06±0.01	-16.76±2.62	3.12	-16.30	-2.08±0.04	25.88±0.03	-	1.05	24.29
2%	4.58±0.02	-22.62±0.49	2.42	-22.23	-5.88±0.01	25.49±0.02	-	-1.63	23.82
4%	4.89±0.28	-21.85±1.77	3.14	-21.44	-10.02±0.33	25.66±0.24	-24.36	-4.45	24.09

IV. CONCLUSIONS

Differential scanning calorimetry has been done on coconut milk and its emulsion containing NaCl at 2°C/min. All samples showed multiple crystallization peaks of oil droplets with an onset temperature closed to 5°C. A single peak with relatively high enthalpy has been observed which corresponds to water freezing process. When sample was reheated, they showed similar melting profile and is independent on oil droplet size or types of coconut milks. High endset of melting temperature at 25°C has been detected in all samples. To decrease onset crystallization and lower endset melting temperature, NaCl was added. The addition of salt, however, affect only on an aqueous phase. This can be concluded that NaCl have an ability to depress freezing point of water, but does not effect to the crystallization and melting of oil droplets in coconut milk emulsions. The effect of salt onto crystallization of a bulk phase of coconut milk has been still controversial.

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