Coconut Milk and Coconut Oil: Their Manufacture Associated with Protein Functionality

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Abstract: Coconut palm (*Cocos nucifera* L.) is an economic plant cultivated in tropical countries, mainly in the Asian region. Coconut fruit generally consists of 51.7% kernel, 9.8% water, and 38.5% shell. Coconut milk is commonly manufactured from grated coconut meat (kernel). Basically, coconut milk is an oil-in-water emulsion, stabilized by some proteins existing in the aqueous phase. Maximization of protein functionality as an emulsifier can enhance the coconut milk stability. In addition, some stabilizers have been added to ensure the coconut milk stability. However, destabilization of emulsion in coconut milk brings about the collapse of the emulsion, from which virgin coconut oil (VCO) can be obtained. Yield, characteristics, and properties of VCO are governed by the processes used for destabilizing coconut milk. VCO is considered to be a functional oil and is rich in medium chain fatty acids with health advantages.

Keywords: coconut milk, coconut proteins, emulsion stability, oil-in-water emulsion, virgin coconut oil

Introduction

Coconut (*Cocos nucifera* L.) is monocotyledon palm from the *Palmaceae* family (Ohler, 1999). *Cocos nucifera* L. is generally called as a coconut palm and is one of the most useful trees in the world. Well-known products of coconut palm include coconut oil, coconut milk, coconut water and coconut meat. Coconut milk is generally extracted from grated coconut meat after pressing or squeezing with or without the addition of water. Coconut milk has been used as a major ingredient for several cuisines such as curries and desserts (Tansakul & Chaisawang, 2006). Besides serving as a food ingredient, coconut milk is used for the production of virgin coconut oil (VCO), for which collapse of coconut milk emulsion is required. Coconut milk emulsion stability is generally governed by some proteins in the aqueous phase (Peamprasart & Chiewchan, 2006). Thus, to maximize the yield of VCO, the emulsion of coconut milk must be collapsed to a high degree, in which oil can be released and separated effectively. To obtain VCO from the wet extraction process, destabilization of coconut milk emulsion has been implemented via several processes such as physical extraction, fermentation, and enzymatic extraction (Raghavendra & Raghavarao, 2010). VCO is commonly manufactured from coconut meat (wet kernel) by natural or mechanical means without or with the application of heat. Chemical refining, bleaching, or deodorizing methods are omitted. Therefore, the nature of resulting VCO is not changed (Villarino, Dy, & Lizada, 2007). VCO or coconut oil consists of medium chain fatty acids (MCFAs), mainly lauric acid. VCO is not similar to other vegetable oils because of its high MCFAs content (Dayrit, 2014). Because of high stability and various health benefits, VCO has become the subject of consumer and processor interest (Carandang, 2008). This review covers characteristics and functional properties of coconut proteins, especially their role in emulsifying or stabilizing coconut milk. In addition, a summary of production, quality, and applications of VCO, mainly by induction of emulsion collapse, is revisited.

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Coconut

Coconut (*Cocos nucifera* L.) is economically important and generally used in many traditional foods of Pacific and Asian regions (DebMandal & Mandal, 2011). Asia is the major coconut producer all over the world and 90% of the world's total coconuts are cultivated in Indonesia, Philippines, India, Sri Lanka, and Thailand. About 70% of coconuts are consumed domestically, and over half of the crop is consumed fresh (Grimwood, 1975). The edible coconut products are mostly obtained from meat (solid endosperm) and water (liquid endosperm) (Grimwood, 1975). Coconut has been also used as traditional medicine, crafting material and fuel. In general, fruits take about one year for the entire development. First, the husk and shell grow and cavity of embryosac enlarges considerably. This cavity is filled with liquid. After about four months, the husk and shell become thicker. The solid endosperm begins to form against the inner wall of the cavity after six months. This first layer is thin and gelatinous. About eight months later, the soft white endocarp becomes hard and dark brown. The fruit becomes mature within 12 months (Ohler, 1999). The mature coconut (MC) fruit (about 12 months) contains 35% husk (fibrous coat of fruit), 12% shell (inner hard coat of fruit), 28% meat (solid endosperm), and 25% water (liquid endosperm; Grimwood, 1975). A cross-section of a coconut is illustrated in Figure 1.

To obtain the edible portion, coconut is subjected to removal of the shell, followed by paring and draining of water. Subsequently, coconut meat can be collected manually and grated with the aid of rotary wedge cutter machine (Senphan & Benjakul, 2015). The composition of the mature kernel is dependent on cultural practices, variety, maturity of the nut, and geographical location. Patil, Benjakul, Prodpran, Senphan, and Cheetangdee (2017) reported that different maturity stages had the marked impact on the chemical composition of coconut meat and milk. Proximate compositions of MC meat are listed in Table 1. Coconut meat can be consumed fresh. Moreover, coconut meat is grated, mixed with or without water and pressed to extract the coconut milk (Grisingha, 1991).

Coconut proteins

Apart from oil, coconuts also contain proteins with moderately well-balanced amino acid profile in term of nutritive value (Gonzales & Tanchuco, 1977; Gunetileke & Laurentius, 1974;

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Table 1–Proximate composition of mature coconut kernel.

Composition (%)						
Moisture	Protein	Oil	Crude fiber	Ash	Carbohydrates	References
44.0	3.6	38.1	3.1	1.3	9.9	Dendy and Timmins (1973)
42–48	4.0	36.0	2.0	–	7.20	Grimwood (1975)
35.37	5.5	44.01	3.05	0.77	6.57	Balachandran et al. (1985)
36.0	4.5	41.5	–	1.1	16.9	Chakraborty (1985)
40.9	3.8	35.2	–	–	–	Kwon et al. (1996)
61.07	3.95	20.86	–	1.14	13.05	Patil et al. (2017)

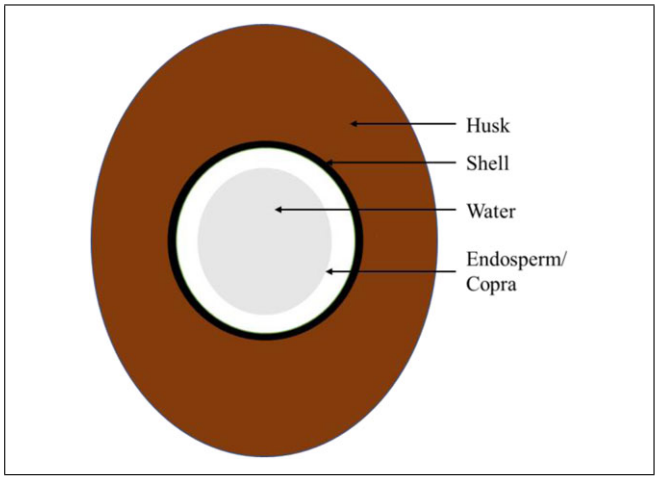

Figure 1–Coconut fruit cross-section.

Kwon, Park, & Rhee, 1996; Rasyid, Manullang, & Hansen, 1992). To recover or extract coconut proteins, protein isolates from coconut skim milk were prepared by ultrafiltration, salt precipitation, isoelectric precipitation, and heat coagulation (Capulso, Gonzales, & Celestino, 1981; Raghavendra & Raghavarao, 2010).

Isolation and fractionation

Several procedures for isolation of protein from coconut skim milk have been developed. Those include isoelectric (p*I*) precipitation, heat coagulation, combined isoelectric precipitation as well as heat coagulation, and co-precipitation with a calcium salt. High yield of protein was achieved by heat coagulation followed by pI precipitation. Similarly, Capulso et al. (1981) studied the effect of heat coagulation, isoelectric precipitation and simultaneous pH and heat coagulation on the recovery of coconut proteins from skim milk. Eighty-four percent of proteins in the skim milk were precipitated with HCl at pH 4 and further coagulated by heat at 90 °C for 30 min. Proteins were extracted using alkaline extraction process from coconut milk press cake using saturated Na_3PO_4 and the yield of 47% was obtained (Chambal, Bergenståhl, & Dejmek, 2012).

Coconut proteins are generally classified according to their solubility and amino acid composition (Rasyid et al., 1992). Coconut proteins can be fractionated into five fractions using different solvents. Water, sodium chloride, isopropanol, acetic acid, and sodium hydroxide soluble fractions are designated as albumin, globulin, prolamin, glutelin-1, and glutelin-2 fractions, respectively. The predominant proteins in coconut endosperm or kernel are classified as globulin (salt-soluble) and albumin (water-soluble), which account for 40% and 21% of total protein, respectively (Balachandran, Arumughan, & Mathew, 1985; Kwon et al., 1996). Distribution of proteins in defatted coconut meal, classified based on solubility, is shown in Table 2. For protein content in coconut skim milk, 75% is accounted for globulin, whereas the remaining (25%) is albumin (Garcia, Arocena, Laurena, & Tecson-Mendoza, 2005). Globulin fraction of coconut has a high level of charged amino acids. Those are aspartic acid, glutamic acid, arginine, and lysine (Kwon et al., 1996; Patil & Benjakul, 2017). The albumin fraction has higher proportions of amino acids with polar side chains. The relative proportion of each protein fraction affects the functional properties and the nutritional quality. The differences in maturation stage, fertilizer, climate, starting material, and so on, also result in varying proportion of various proteins in coconut meat (Patil & Benjakul, 2017).

Characteristics

Molecular weight (MW) of five coconut protein fractions (i.e., albumin, globulin, prolamin, glutelin-1, and glutelin-2) from defatted coconut flour analyzed by SDS-PAGE with reducing agent (β-mercaptoethanol) was reported by Kwon et al. (1996) and Sringam (1997). The albumin fraction had MW ranging from 18 to 52 kDa. MW of globulin fraction was below 60 kDa. Cocosin as the major protein (~65%) with MW of 55 kDa was observed in the endosperm of coconut (Garcia et al., 2005). Patil and Benjakul (2017) also documented that both albumin and globulin fractions contained major protein with MW of 55 kDa. Prolamin fraction had MW with the range of 17 to 56 kDa, whereas the glutelin-1 fraction had MW ranging from 14 to 100 kDa. Coconut globulin consist of two major types, named 11S and 7S globulin. Cocosin, a globulin, is one of seed storage proteins identified as 11S globulin, accounting for 86% of the total globulin (Balasundaresan, Sugadev, & Ponnuswamy, 2002). Cocosin is generally hexameric quaternary in structure, of which the MW is about 300 to 360 kDa and each subunit has MW of 55 kDa. The subunits consist of the basic (22 to 24 kDa) and acidic (32 to 34 kDa) polypeptides linked via disulfide bridge. Basic and acidic chains are dissociated under the reducing conditions (Garcia et al., 2005). The 7S coconut globulin is a type of vicilins, which are characterized as trimer having an oligomeric MW of 150 to 190 kDa, with each single chain subunit of about 55 kDa (Garcia et al., 2005). The coconut 7S globulin is unglycosylated and lack of sulfur-containing amino acids (Carr, Plumb, Parker, & Lambert, 1990). Protein pattern of coconut milk at different stages of maturity under reducing conditions showed several major protein bands with MW of 55, 33, 31, 25, 21, 20, 18, and 16 kDa. However, nonreducing condition showed six protein bands with MW of 55, 46, 33, 25, 18, and 16 kDa (Patil et al., 2017).

The coconut protein can be separated as high MW (HMW) and low MW (LMW) fractions by Sephadex G-200 column using 0.95 M NaCl in 0.01M Na_2HPO_4 (pH 8.2) as elution buffer (Hagenmaier, Cater, & Mattil, 1972). The HMW (150 kDa) and

Table 2–Distribution of proteins in defatted coconut meal.

Fraction	Extraction solvents	Samson et al. (1971)	Kwon et al. (1996)	Sringam (1997)	Patil and Benjakul (2017)
Albumin	Water	30.6[a]	21.0	22.7	19
Globulin	NaCl (1–0.5 M)	61.9	40.1	46.1	36
Prolamin	Isopropyl alcohol (70%)	1.1	3.3	2.0	2
Glutelin-1	Glacial acetic acid (50%)	–	14.4	12.5	10
Glutelin-2	NaOH (0.1 M)	4.7	4.8	1.2	4
Unextractable protein	Residue	1.8	16.4	13.4	–

[a]Values are expressed as % for individual fraction.

the LMW (24 kDa) were found approximately 84% and 14% of the total proteins, respectively. Kwon et al. (1996) separated the major fractions of protein (albumin and globulin) from defatted coconut flour using Sephadex G-200 column and found that the albumin was separated into two major peaks with MW of 12 and 141 kDa, whereas one minor peak had MW about 27 kDa. The globulin showed five peaks with MW of 186, 120, 46.7, 21.4, and 14.6 kDa, respectively.

Amino acid compositions

Coconut proteins generally provide good nutritional value with a relatively balanced amino acid profile (Gonzales & Tanchuco, 1977; Gunetileke & Laurentius, 1974; Kwon et al., 1996; Rasyid et al., 1992). Those proteins contain a high amount of essential amino acids (71% to 77%) and a digestibility of 86% to 94% (Hagenmaier, Mattil, & Cater, 1974; Molina & Lachance, 1973). In coconut skim milk, the limiting amino acids are methionine, isoleucine, threonine, and tryptophan (Hagenmaier, Lopitakwong, & Verasestakul, 1975). Amino acid composition of three coconut protein fractions, as well as coconut flour in comparison with Food and Agriculture Organization (FAO) amino acid scoring, are shown in Table 3. Generally, coconut proteins have comparatively high level of glutamic acid (17.0% to 27.2%), arginine (14.2% to 17.9%), and aspartic acid (5.6% to 8.9%) but are deficient in methionine (1.2% to 2.9%; Kwon et al., 1996). Most amino acid levels are lower in the albumin fraction, except glutamic acid, arginine, and lysine, which are higher than those found in glutelin-1 and globulin fractions. The coconut globulin contains a high amount of essential amino acids including valine and phenylalanine but has less glutamic acid, lysine, and arginine than the albumin (Kwon et al., 1996). The leucine and phenylalanine of globulin fraction are comparable to those guided by FAO, while the globulin and glutelin-1 fractions show higher valine content. Threonine, cysteine, and methionine seemed to be the limiting amino acids for coconut proteins (Kwon et al., 1996).

Functional properties

Functional properties of coconut proteins depend strongly on their solubility. The solubility of coconut proteins is generally low between pH 4 and 5, and is increased when pHs are above or below such pHs. The proteins of coconut endosperm from different regions were reported to have different solubility (Balachandran et al., 1985), associated with different amino acid profiles. The minimum solubility of major protein components of coconut protein isolate, coconut skim milk, and the extracts of coconut endosperm was observed between pH 4 and 5, known as a range of isoelectric point of those proteins (Balasubramaniam & Sihotang, 1979; Gonzales & Tanchuco, 1977; Hagenmaier et al., 1974; Kwon & Rhee, 1996). Nevertheless, the maximum solubility was reported at pH 10.3 (Balasubramaniam & Sihotang, 1979). Foaming capacity of coconut protein isolate was also affected by pH. At pH 2 and 11, foam expansion was highest but foam stability was low (Gonzales & Tanchuco, 1977). Proteins in coconut milk play a profound role in emulsion stability. Onsaard, Vittayanont, Srigam, and McClements (2006) stated that proteins isolated from coconut skim milk effectively stabilized emulsions that are fairly viscous. However, the lower efficacy of the proteins extracted from coconut cream was observed, compared to whey protein isolate, by either producing small oil droplets by the homogenizer or avoiding droplet aggregation to obtain a stable emulsion (Onsaard et al., 2006). In general, ionic strength, pH, and especially temperature drastically influence emulsifying properties of coconut proteins (Kwon & Rhee, 1996; Onsaard et al., 2006). Proteins form a protective barrier film around oil droplets, in which repulsion (*e.g.*, electrostatic and steric) between the oil droplets prevent their coalescence. Effects of sonication (120 W, 20 kHz and 250 W, 20 kHz) on the stability of sunflower oil-in-water emulsions prepared by coconut milk protein was studied by Lad and Murthy (2012). The emulsion containing coconut milk protein (1.2%) with the application of ultrasound was very stable. Solubility and emulsification properties of a crude freeze-dried alkaline protein extract (APE) was studied by Chambal, Bergenståhl, and Dejmek (2013). Solubility and emulsification properties of APE increased at pH above and below 3 to 4. Rodsamran and Sothornvit (2018) studied physicochemical and functional properties of protein concentrate from a by-product of coconut processing. Protein powders from milk cake showed higher oil and water absorption capacities. However, protein powders from oil cake showed better emulsifying and foaming properties. Patil and Benjakul (2017) fractionated albumin and globulin from defatted coconut meat and comparatively studied emulsifying properties of these protein fractions. Globulin fraction was more competent as an emulsifier in the oil-in-water emulsion as compared to albumin. The differences in emulsifying property of coconut proteins (albumin fraction and globulin fraction) were possibly related to varying amino acid compositions. Variation in the distribution of amino acids and the proportion of nonpolar and polar amino acids, mainly on the surface of the protein, determine emulsifying property. Generally, hydrophobic proteins with nonpolar side chains exhibits high emulsifying properties (Patil & Benjakul, 2017).

Thermal property

Coconut proteins have been shown to be highly sensitive to heat. They undergo denaturation and coagulation upon heating to 80 °C (Kwon et al., 1996). Differential scanning calorimetric studies of raw undiluted coconut milk revealed several endothermic transitions in the range of high temperature (80 °C to 120 °C). This result reflects the varying thermal denaturation behavior and complex protein composition of coconut proteins (Kwon et al., 1996; Seow & Goh, 1994). The exposure to heat at high temperatures for a long time, results in denaturation and precipitation of proteins in the coconut milk. The denaturation of coconut

Table 3–Amino acid composition of three major protein fractions and coconut flour.

Amino acid (g/100g of protein)	Albumin	Globulin	Glutelin-1	Coconut flour	FAO[a]
Isoleucine	2.8	4.1	3.7	4.2	–
Leucine	3.9	6.5	6.5	7.4	7.0
Lysine	5.1	3.5	3.5	4.7	5.5
Methionine	1.2	2.9	2.1	1.8	3.5
Phenylalanine	2.7	5.9	4.6	5.1	6.0
Tyrosine	3.0	3.7	3.1	1.8	–
Threonine	3.3	3.3	3.2	2.5	4.0
Tryptophan	–	–	–	–	1.0
Valine	3.5	7.5	6.7	5.4	5.0
Histidine	1.8	1.9	1.9	1.8	–
Aspartic acid	5.6	8.9	8.3	9.3	–
Proline	2.7	3.4	3.2	3.6	–
Serine	3.1	5.0	3.9	5.3	–
Glutamic acid	24.5	17.5	17.0	22.4	–
Glycine	4.0	4.9	4.5	5.1	–
Alanine	2.9	4.1	3.9	4.8	–
Arginine	17.9	15.0	14.2	12.3	–

[a]Value guided by food and agriculture organization (FAO).

protein by heat is enhanced at the acidic and basic pH regions (Onsaard, Vittayanont, Srigam, & McClements, 2005). However, coconut protein is more resistant to heat denaturation when salts, polyols, and sugars are presented (Seow & Goh, 1994).

Coconut milk

Coconut milk can be prepared at home from grated meat by squeezing with hand, whereas industrial or commercial scale employs the screw press or hydraulic to extract the milk. Basically, coconut milk is an oil-in-water emulsion, in which continuous phase is water and oil is dispersed phase (Figure 2). The oil droplets in coconut milk emulsion are surrounded by a film of interfacial active protein and emulsion stability is depending on these proteins (Dendy & Timmins, 1973). The composition of coconut milk is generally depending on that of the coconut meat used for extraction. The efficiency of extraction and composition of coconut milk from coconut meat are governed by operation parameters such as the temperature of added water and the pressing condition (Grisingha, 1991). The difference in the water: coconut meat ratio, ranging from 1:1 to 20:1, had no effect on oil and protein extraction into coconut milk (Dendy & Timmins, 1973). Thungkao (1988) also documented that protein contents were not affected by temperatures (30 °C, 55 °C, and 80 °C) used for coconut milk extraction when the grated coconut meat and water ratio of 1:1 was employed. Nevertheless, the fat content of the coconut milk extracted at 55 °C was the highest, while those of coconut milk extracted at 30 °C and 80 °C were not significantly different. Grisingha (1991) compared the oil and protein extractability in coconut milk prepared using three different methods including (1) twice pressing with water adding in the second time, (2) twice pressing with water adding in both times, and (3) once pressing with water adding. Protein and fat contents of extracted coconut milk were not significantly different. Coconut milk extraction from a fresh coconut is the most important step in wet or aqueous processing. The wet process is a promising alternative method to the traditional mechanical pressing of copra to manufacture the oil (Seow & Gwee, 1997). In this case, the breakdown of emulsion is crucial for the effective recovery of both protein and oil.

Stability

Coconut milk is naturally stabilized by proteins and phospholipids (Monera & Del Rosario, 1982). The aqueous phase of coconut milk emulsion contains some proteins, which act as an emulsifier to stabilize oil droplets (Peamprasart & Chiewchan, 2006). Hydrophilic and hydrophobic groups of these molecules can minimize the interfacial tension among two phases and promote the dispersion of oil droplets in the aqueous phase, thereby enhancing emulsion stability (Monera & Del Rosario, 1982). Hydrophobic domains or nonpolar side chains of the proteins were able to interact with hydrocarbon chains on fatty acids. This interaction can promote physical entrapment of oil. The interactions between oil droplets depend on the quantity and quality of the proteins (Patil & Benjakul, 2017). When repulsive forces are dominant, the oil droplets have a tendency to persist as individual entities, thus forming a stable emulsion. Tangsuphoom and Coupland (2005) investigated the colloidal stability of coconut milk as affected by homogenization and heat treatment. Both non-homogenized and homogenized samples were subjected to heat at different temperatures from 50 °C to 90 °C for 1 hr. Homogenization minimized the primary emulsion oil droplets size from 10.9 to 3.0 μm.

Processing operations, which tend to produce smaller globules, are expected to yield more stable emulsion (Onsaard et al., 2005). Coconut milk fat structure affected by homogenizing pressure was investigated by Chiewchan, Phungamngoen, and Siriwattanayothin (2006). Homogenized sample had smaller oil droplet size than nonhomogenized counterpart. Homogenization can reduce droplet size by the high shear force applied to dispersed phase (Floury, Desrumaux, & Legrand, 2002). Smaller oil droplet size was achieved at higher homogenizing pressure and was associated with more stable emulsion. The effect of sonication on homogenization of coconut milk was reported by Iswarin and Permadi (2012). Ultrasonic treatment (7 W for 25 min) was an effective technique for reducing fat globule size up to 3.64 μm. Reduction in fat globule size by ultrasound was caused by cavitation effect (Iswarin & Permadi, 2012).

Proteins act as emulsifiers, which stabilize the oil droplets in coconut milk (Senphan & Benjakul, 2015). Emulsifiers perform two roles in the stability of emulsion: (1) lower the interfacial tension between water phases and oil; and (2) form a mechanically cohesive interfacial film surrounding oil droplets, thus preventing coalescence. Patil et al. (2017) carried out a comparative study to evaluate the physicochemical properties and emulsion stability of coconut milk obtained from the coconut at three different maturity stages. Stability of coconut milk emulsion depended on

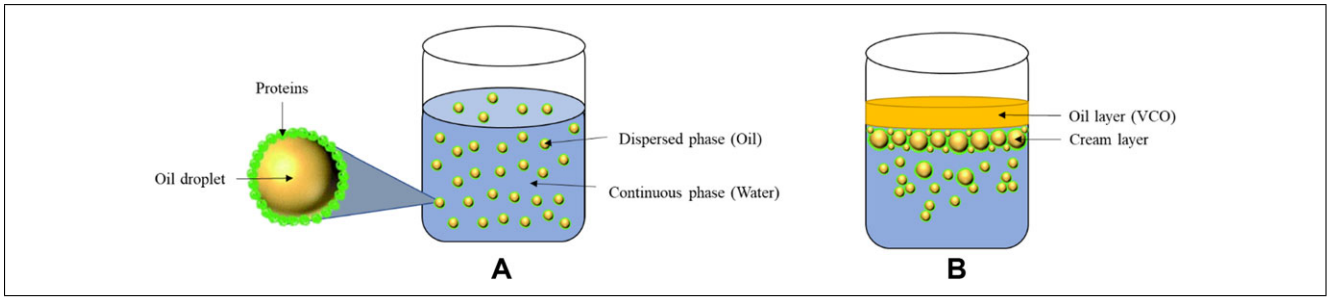

Figure 2–Coconut milk model oil-in-water emulsion. (A) Stable emulsion and (B) instable emulsion.

intrinsic factors, mainly pH and protein content. pH can affect the net charge of proteins surrounding the oil droplets. At p*I*, net charge on the protein is zero. Therefore, repulsion of protein film surrounding the oil droplets is lower. As a result, emulsion stability of coconut milk is decreased. High protein content can lead to efficient localization of protein films at the oil–water interphase. As a consequence, the stability of coconut milk emulsion is increased (Patil et al., 2017). To increase the shelf-life of coconut milk, heat treatment has been introduced. However, such a harsh treatment can induce instability of emulsion in coconut milk. Bao, Wang, and Li (2004) suggested the optimal conditions to prepare sterilized coconut milk drink as follows: coconut: water ratio 1:10, pH 6.5, sugar 4%, homogenization at 20 to 25 MPa and sterilization at 121 °C for 20 min. The combined effect of the amount of emulsifier, emulsifier types and sonication time on the droplet size of the emulsion to stabilize coconut milk was studied by Jena and Das (2006). Emulsifiers (maltodextrin and gum acacia) were added to coconut milk at different emulsifier/fat ratios (4, 2.75, and 1.5). Droplet size of coconut milk treated with ultrasound (about 2 to 2.5 min) was decreased with increasing emulsifier/fat ratio. Jena and Das (2006) also documented that distribution of particle size modeling by Rosin–Rambler–Sperling–Bennet relation could be a promising tool for prediction of uniform distribution and average droplet size of sonicated coconut milk. Linear regression equations provided a suitable model to predict the sonication time required to obtain the certain degree of reduction in droplet size. Effect of coconut sugar (10% to 30%) and stabilizing agents, namely Montanox 60 (0.6% to 1.0%) and carboxymethyl cellulose (CMC, 0.6 % to 1.0%) on physical properties of sterilized high-fat coconut milk (30%) was studied by Jirapeangtong, Siriwatanayothin, and Chiewchan (2008). Coconut sugar, as well as stabilizing agents, had marked effect on both rheological properties and emulsion stability of coconut milk with high-fat. An emulsion containing sugar required a higher concentration of stabilizing agents to stabilize the colloidal system. For the production of high stability sweetened coconut milk, 0.8% to 1.0% of Montanox 60 and CMC were recommended. The effect of surface-active stabilizers (whey protein isolate [WPI], sodium caseinate, Tween 20, or SDS at concentration of 0 to 1 wt%) and homogenization on the microstructure and colloidal stability of coconut milk was elucidated by Tangsuphoom and Coupland (2008). Coconut milk added with small-molecule surfactant (Tween 20 and SDS) either before or after homogenization (10 MPa) completely displaced the interfacial coconut proteins and produced a stable emulsion. Coconut milk added with stabilizers (sodium caseinate and WPI) prior to homogenization competed with coconut proteins to adsorb at the newly-formed oil–water interface, thus yielding a stable emulsion. However, stability and oil–water interface of non-homogenized coconut milk was not affected by the addition of stabilizers. Coconut milk stabilized by different emulsifiers (WPI, sodium caseinate, Tween 20, or SDS at concentration of 0 to 1 wt%) was subjected to various cooling (5 °C for 24 hr), freezing (−10 and −20 °C for 24 hr) and heating treatments (70 °C, 90 °C, and 120 °C for 1 hr) and the changes in microstructure and bulk properties were monitored by Tangsuphoom and Coupland (2009). The coconut milk added with 1 wt% stabilizer (WPI or sodium caseinate) had smaller oil droplets (0.4 μm) and were stable against chilling at 5 °C. Sodium caseinate added sample was stable against freeze-thawing (−10 °C or −20 °C), whereas WPI emulsion was unstable. The microstructure of sodium caseinate stabilized coconut milk emulsion was not changed by heating (70 °C, 90 °C, or 120 °C) for 1 hr. However, oil droplets of WPI stabilized coconut milk flocculated and coalesced when subjected to heat at 90 °C or 120 °C for 1 hr. No marked change was observed in droplet size of the emulsion heated at a temperature of 70 °C. Small-molecule surfactants added to coconut milk showed better emulsion stability against heat treatments but were completely unstable upon freeze-thawing because of their thin interfacial film surrounding oil droplet which was less efficient to protect oil droplets against coalescence (Tangsuphoom & Coupland, 2009). Sucrose esters can be used as a good alternative to petrochemically synthesized Tweens for preparation of coconut milk emulsions with improved stability. Ariyaprakai, Limpachoti, and Pradipasena (2013) compared emulsifier and interfacial properties of sucrose ester (monostearate) with Tween 60 for application in coconut milk. Sucrose ester had a moderately good capacity to minimize the interfacial tension between the oil-water interface of coconut milk. Sucrose ester also showed a marked effect on the thermal properties of coconut milk. The complex between coconut protein and sucrose ester could protect coconut milk against freeze and heat damages (Ariyaprakai et al., 2013).

Instability

The freshly prepared coconut milk appears stable and homogenous. However, coconut milk is physically unstable and is prone to phase separation into two distinct phases (cream phase and aqueous phase) after a few hours (Seow & Gwee, 1997). There are three main mechanisms that can contribute to emulsion instability: (1) creaming, (2) flocculation, and (3) coalescence (Walstra, 1987). Creaming is due to differences in density between the two phases, which leads to phase separation (Beydoun, Guang, Chhabra, & Raper, 1998). In coconut milk, cream separates from the aqueous phase within 5 to 10 hr of production (Seow & Gwee, 1997). The separated milk, however, can be easily re-homogenized by shaking (Escueta, 1980). Flocculation is the aggregation of oil droplets due to weak repulsive forces and strong attractive forces between oil droplets (Verwey, 1947). During flocculation, oil droplet of the dispersed phase will be attached to each other, but retain

their individual structural integrity. This results in the separation of cream from the aqueous phase (McClements & Demetriades, 1998). However, during coalescence, protein films surrounding the oil droplets are disrupted and two oil droplets will form a single larger droplet. The severe coalescence brings about the separation of oil from emulsion, including coconut milk. The main reason for coconut milk emulsion instability is the low surface activity and poor emulsifying properties of coconut proteins (Monera & Del Rosario, 1982). The rate of emulsion collapse is strongly affected by environmental conditions (pH, temperature, etc.), processing condition and composition. The coconut milk was poorly stable over the pH range of 3.5 to 5 but exhibited stability maxima at pH 6.5 as well as pH 1.5 to 2 (Monera & Del Rosario, 1982). In coconut milk, oil droplet size and pH are the most paramount factors affecting the emulsion stability. Coconut milk emulsion can be destabilized by adjustment of their pH between pH 5.6 and 3 (Marina, Man, Nazimah, & Amin, 2009c). Raghavendra and Raghavarao (2010) studied the coconut milk emulsion destabilization at different temperature and pH levels. Coconut milk emulsions were very unstable at pH 7 to 8 and pH 3 to 6. Because proteins have polar groups, their intra- and intermolecular interaction are directly affected by changes in pH of the emulsion. The low pH of coconut milk could enhance the destabilization of the emulsion, by lowering the repulsion of protein film surrounding the oil droplets (Patil et al., 2017). Acetic acid (25%, w/v) disrupt the coconut milk emulsion because coconut milk proteins were plausibly coagulated and precipitated at pH 4 (Zakaria et al., 2011). Furthermore, protein denaturation was observed in coconut milk when heated at a higher temperature. Coconut proteins were reported to coagulate and denature at 80 °C or higher temperature (Kwon et al., 1996; Raghavendra & Raghavarao, 2010). Thermal denaturation of coconut proteins influences the surface charge of oil droplets and causes droplets aggregation in coconut milk. This results in unstable coconut milk emulsion. Coconut milk is an abundant source of oil. Therefore, to obtain coconut oil, emulsion must be destabilized at a high degree.

Virgin coconut oil

VCO is the purest form of coconut oil with natural characteristics coconut smell and taste. At low temperature, VCO is solidified but when liquefied, it becomes colorless like water (Marina et al., 2009c). VCO exhibits good digestibility mainly due to medium chain fatty acids (MCFAs). MCFAs are burned up immediately after consumption and therefore the body uses it instantly to make energy, instead of storing it as body fat (Patil, Benjakul, Prodpran, Senphan, & Cheetangdee, 2016). Lauric acid is converted into a very valuable compound known as monolaurin, which has antibacterial and antiviral properties (DebMandal & Mandal, 2011). VCO is rapidly gaining popularity because of high stability and various health advantages (Carandang, 2008). VCO also possesses antioxidant properties that boost the immune system. Therefore, consumption of VCO may help protect the body from infections. VCO does not undergo any hydrolytic and atmospheric oxidation as confirmed by its low peroxide value as well as very low free fatty acid content (Marina et al., 2009c; Patil et al., 2016).

Production

Dry extraction. Coconut is served as a raw material for coconut oil production. Dry and wet materials are generally known as dry coconut (copra) and wet coconut, respectively. Both raw materials can be used for extraction of oil. Dry processing is the most commonly applied for extraction. In this process, clean ground copra is pressed by screw press, wedge press, or hydraulic press to release the coconut oil, which is subsequently subjected to refining processes, namely degumming, bleaching, and deodorizing.

Wet extraction. Recently, wet process has become very popular to produce coconut oil or VCO and does not need the refining process. Two major steps are involved in the extraction of VCO by a wet process; first, the extraction of an emulsion (coconut milk) from the coconut meat, and second, the breaking of this emulsion to separate oil and protein components (Gunetileke & Laurentius, 1974). Wet extraction is superior as no high heat treatment or chemical is used and the oil obtained has been called as VCO. Alteration of VCO is negligible (Marina et al., 2009c). VCO has a fresh coconut smell that can be mild to intense, dependent upon the process used for extraction of oil. The separation of VCO can be further enhanced by several methods.

Physical extraction. Gravitational separation is mostly related to the slow creaming process of an oil-in-water emulsion. In general, centrifugation is used to accelerate this creaming process, in which higher rotation frequencies are allowed to separate the cream effectively. Centrifugation process is desirable to the simple gravitation method (Nour, Mohammed, Yunus, & Arman, 2009). Gravitational separation may be very slow due to the closeness between oil droplets and the aqueous phase, or due to attractive forces holding the oil droplets together (Nour et al., 2009). Gravitational separation is time-consuming, although centrifugal separation is accomplished within a short time. It is possible to break down the emulsions by centrifugation in order to separate dispersions of fine oil droplets. Consequently, the coalesced disperse phase is separated as VCO from the water phase (Coulson & Richardson, 1991; Nour et al., 2009).

Chilling and thawing techniques have been used to destabilize oil-in-water emulsion. Gunetileke and Laurentius (1974) reported that the protein and oil can be separated from coconut cream obtained from centrifugation of coconut milk by chilling at 10 °C for 4 hr, followed by thawing at 40 °C. The emulsion was centrifuged (3585 x *g* for 10 min) prior to chilling and thawing for close packing of coconut oil droplets (cream). Coconut milk was subjected to chilling at various temperatures (5 °C, 10 °C, 15 °C, and 20 °C) for 6 hr, followed by thawing at room temperature (29 °C ± 2 °C). During thawing process, oil droplets coalesce and form the large size droplets. The cream was centrifuged at 4880 x *g* for 15 min to obtain oil and the highest oil recovery (92%) was obtained at 5 °C (Raghavendra & Raghavarao, 2010). Similarly, VCO was obtained from the chilling method by centrifugation of coconut milk at 3600 x *g* for 10 min and the cream was removed from the upper layer. Subsequently, the cream was chilled at 5 °C for 24 hr and thawed in a water bath at 50 °C. Oil recovery of 86.62% was obtained from chilling and thawing process (Mansor et al., 2012). Raghavendra and Raghavarao (2010) documented that combined treatments (the use of Aspartic protease at 37 °C, followed by chilling and thawing) on coconut milk yielded the highest oil recovery (94.5%).

Fermentation process. Natural fermentation process is the conventional method to produce VCO, where coconut milk is allowed for fermentation using microorganisms (Marina, Man, & Amin, 2009b). Coconut milk can be fermented with normal flora, allowing the oil to separate on the top portion within 24 to 48 hr. The separated oil can be collected. Fermentation enhances the breakdown of the emulsion, probably by microbial proteases. The contamination with microorganisms can take place because coconut milk is the abundant source of moisture, carbohydrates, and

proteins. This environment can promote the growth of microorganisms (Tansakul & Chaisawang, 2006). Distilled water was added to fresh coconut milk at 1:1 ratio. Baker's yeast (*Saccharomyces cerevisiae*) of 2.0 g was added to 1 L of the mixture as an inoculum for the fermentation process. The mixture was then allowed to stand at room temperature for 36 hr. As the water and oil layers became separated, the top layer of oil was simply decanted (Mansor et al., 2012). Nevertheless, coconut milk may spoil by some microorganisms, resulting in a low quality of VCO (generally in yellow color) with oil recovery of 65% (Mansor et al., 2012). Therefore, the major drawbacks of fermentation process are fermented odor and low oil recovery (Raghavendra & Raghavarao, 2010). In addition, during the fermentation process, lipolytic enzymes in the presence of water could produce high free fatty acid (FFA). Coconut milk emulsion can also be destabilized by adjustment of pH between pH 3 and 5.6 and added with bacterial cultures (Chen & Diosady, 2003). Man, Karim, and Teng (1997) extracted VCO via an induced fermentative process using 5% inoculum (*Lactobacillus plantarum*) at 70 °C for 6 hr under semicontrolled conditions. The yield of VCO was 95.06%. The temperature of 45 °C, pH of 5, inoculum (*Lactobacillus plantarum* 2%), fermentation time of 48 hr, and anaerobic conditions was found as an optimum condition for the induced fermentation process of VCO (Satheesh & Prasad, 2014).

Enzymatic extraction. Enzymes can be used in the aqueous extraction process of oil. Enzymatic pretreatment has been known as a potential means to obtain the high yield of oil (Marina et al., 2009b). VCO can be extracted from coconut milk by using enzymatic hydrolysis process (Senphan & Benjakul, 2016). Enzymatic extraction is the most promising method among all processes for extracting oil from coconut milk (Tano-Debrah & Ohta, 1997). Enzymatic hydrolysis, particularly mediated by proteases, effectively destabilize the coconut emulsion and release the oil (Rahayu, Sulistyo, & Dinoto, 2008). Coconut milk proteins play a role as the emulsifier to stabilize the oil droplets in the emulsion. After enzymatic hydrolysis of those coconut proteins, the emulsion was unstable with concomitant release of oil from the emulsion (Patil & Benjakul, 2017). The use of enzyme can shorten the extraction time of VCO. Furthermore, higher yield of VCO with prime quality was attained (Senphan and Benjakul (2015). VCO obtained by enzymatic hydrolysis method has safety and is more beneficial than the oil produced from copra by the traditional method because the latter is often infected via aflatoxin or insects producing molds related with toxicity problem during production (Handayani, Sulistyo, & Rahayu, 2009). VCO aqueous extraction involved the mixture of enzymes such as 0.075% (w/v) pectinase, 0.05% (w/v) protease, and 0.05% (w/v) amylase. The process resulted in high extraction yields (76.4%) of oil, as compared with a nonenzymatic process, in which the yield was less than 20% (Barrios, Olmos, Noyola, & Lopez, 1990). Coconut milk was added with papain (0.1%, w/w) and left to stand for 3 hr at 55 °C. The mixture was subsequently subjected to centrifugation at 4900 x *g* for 25 min to collect the oil and the recovery was 65% (Mansor et al., 2012). Enzyme concentration and substrate, pH, incubation time, and temperature affected the hydrolytic reaction (Handayani et al., 2009). These factors determined extraction yield of oil differently (Rahayu et al., 2008). Protease efficiency in enhancing extraction yield of oil was found in descending order as follows: alkaline protease > neutral protease > acid protease. Raghavendra and Raghavarao (2010) documented that coconut milk subjected to hydrolysis using papain showed 60.09% oil yield. Neutrase 1.5 MG (0.3%, w/w) and Viscozyme L (0.6%, w/w) at pH of 7, the temperature of 60 °C and total incubation time of 30 min were used to achieve the maximum extraction yield of oil (Sant'Anna, Freitas, & Coelho, 2003). Enzyme mixture (α-amylase, cellulase, protease, and polygalacturonase) at 1% (w/w) and pH 7.0 with extraction temperature of 60 °C were used for VCO extraction by Man, Asbi, Azudin, and Wei (1996). The 73.8% of oil recovery with fine quality of oil were gained. Senphan and Benjakul (2015) used proteases from shrimp hepatopancreas as an alternative for commercial enzymes to reduce the cost of VCO production. Senphan and Benjakul (2015) reported that VCO extracted with aid of crude protease extract (from hepatopancreas from Pacific white shrimp; 10 unit/g protein) for 6 hr at ambient temperature had the maximum yield of oil (92.39%). Patil et al. (2016) extracted VCO from coconut milk with three different maturity stages including immature coconut (IMC), MC, and overlay mature coconut (OMC) using Alacalase at a level of 0.5% (v/v). Highest oil recovery of 95.64% was obtained in OMC, followed by MC (84.45%) and IMC (61.06%). Among all wet extraction processes, the enzymatic extraction has been known to be less time consuming and effective. Moreover, the maximum yield of VCO could be attained Senphan and Benjakul (2015).

Table 4–Essential composition and quality parameters of virgin coconut oil (VCO) appointed by Asian Pacific Coconut Community (APCC).

Serial no.	APCC parameters	APCC standards
1.	Moisture (%)	Max 0.1
2.	Refractive index at 40°C	1.4480–1.4492
3.	Relative density	0.915–0.920
4.	Specific gravity at 30°C/30°C	0.915–0.920
5.	Iodine value (g I_2/100 g oil)	4.1–11
6.	Saponification value (mg KOH/g oil)	250–260
7.	Free fatty acid (%)	Max 0.2
8.	Peroxide value (meq O_2/kg)	Max 3

Quality of VCO

Essential composition and quality parameters of VCO appointed by Asian Pacific Coconut Community (APCC) standards are enlisted in Table 4. Different types of raw materials, namely incubated and desiccated coconut meat, incubated coconut milk as well as freeze-thawed coconut milk affected physicochemical properties of VCO (Marina et al., 2009b). However, no drastic differences in overall VCO quality were observed. Physical and chemical qualities must comply with the Philippine standards for VCO and the Codex standard for coconut oil (Dia, Garcia, Mabesa, & Tecson-Mendoza, 2005). Mansor et al. (2012) characterized VCO obtained from different methods including chilling, fermentation, fresh-drying, and enzyme treatment. Various methods slightly affected the quality but the difference was not significant. Marina et al. (2009c) reported the chemical properties of commercial VCO available in Indonesia and Malaysia. Chemical properties of VCO was not changed among the samples. The FFA, peroxide, iodine, and saponification values reported for commercial VCO samples were in accordance with the specification guided by Codex standard (2003) for refined coconut oil. Senphan and Benjakul (2015) also stated that VCO extraction aided by Alcalase (10 unit/g protein) or crude protease extract (from the hepatopancreas of Pacific white shrimp) at 60 °C for 90 min had no influence on the resulting VCO quality. Patil et al. (2016) studied characteristics as well as the quality of VCO as influenced by maturity stages.

Maturity stages of coconut had no profound effect on oxidative stability and quality of VCO.

Marina, Che Man, Nazimah, and Amin (2009a) reported the fatty acid composition of commercial VCO available in Malaysia and Indonesia. Lauric acid (46% to 48%) was dominant fatty acid and the content was within the standard limit for VCO according to Asian and Pacific Coconut Community (APCC, 2003) and Malaysian Standard (2007). VCO samples obtained by different processes had differences in fatty acid compositions (Mansor et al., 2012). Lauric acid (with the range of 46.36% to 48.42%) was found in all VCO samples. However, VCO separated from coconut milk with three different maturity stages had a similar fatty acid composition (Patil et al., 2016).

Uses of VCO

VCO is gaining popularity as a functional oil with increasing public awareness (Marina et al., 2009b). VCO serves as a significant source of energy in the diet (Boateng, Ansong, Owusu, & Steiner-Asiedu, 2016). VCO provides lubricating action in dressing and enhances food flavor (Carandang, 2008). In addition, medium chains are similar to the fats presented in mother's milk, which provides immunity for babies against disease. Similar advantageous effects are also found in adults (Maria & James, 2013). VCO possesses anti-inflammatory, antimicrobial, and antioxidant properties and boosts the immune system (Carandang, 2008). VCO also showed high antimicrobial activity and inhibited various pathogenic bacteria for example *Listeria monocytogenes* (Wang & Johnson, 1992). It was also reported that coconut oil in combination with menhaden oil was able to reduce mammary tumor in animal study (Craig-Schmidt, White, Teer, Johnson, & Lane, 1993). Effect of VCO on LDL oxidation in cholesterol, blood coagulation factors, and lipid levels fed Sprague–Dawley rats were studied by Nevin and Rajamohan (2008). Antioxidant levels were higher and also reduced the triglyceride and cholesterol levels in VCO fed animals. VCO, without bile, can easily digest and goes directly to the liver for conversion into energy (DebMandal & Mandal, 2011). VCO has been using to treat fat malabsorption patients, as it contains medium chain fatty acid (Carandang, 2008). The effect of consumption of VCO on HDL cholesterol and waist circumference (WC) in coronary artery disease (CAD) patients was studied by Cardoso, Moreira, de Oliveira, Raggio Luiz, and Rosa (2015). Diets rich in VCO decrease WC and increase HDL-cholesterol concentrations, thus supporting the secondary prevention for CAD patients. VCO increases the metabolism and therefore support weight management (Liau, Lee, Chen, & Rasool, 2011). Protective effect of VCO against liver damage in albino rats challenged with the anti-folate combination, trimethoprim–sulfamethoxazole (TMP–SMX), was studied by Otuechere, Madarikan, Simisola, Bankole, and Osho (2014). The active components of VCO had protective effects against the toxic effects induced by TMP-SMX administration, mainly in the liver of rats. Arunima and Rajamohan (2014) studied the effects of VCO in comparison with olive oil and sunflower-seed oil on the synthesis and oxidation of fatty acids and the molecular regulation of fatty acid metabolism in normal rats. VCO had the beneficial effects on lipid parameters by decreasing lipogenesis and enhancing the rate of fatty acid catabolism, thus reducing coronary heart disease. VCO can be used for cooking and frying because of its high resistance against rancidity development (Patil et al., 2016). Gani, Benjakul, and Nuthong (2017) reported that VCO (5%) can be used as an alternative to other vegetable oils in surimi gel, as it contains MCFAs, therefore, health advantages can be claimed.

Conclusion

The information on coconut proteins and their role in emulsion stability could provide the better understanding of destabilization or enhancement of coconut milk emulsion. Hence, coconut milk emulsion can be stabilized or collapsed to obtain the desired products, named coconut milk and oil, respectively. To stabilize coconut milk, additional stabilizer can be employed to work in conjunction with coconut proteins. The combined methods should be developed to enhance destabilization of coconut milk, in which the shorter processing time and lower cost can be achieved to manufacture VCO. The applications of VCO as ingredient and exploitation of its unique property can lead to the new products with desired characteristics.

Authors' Contributions

Umesh Patil gathered the information from literature and drafted the review article. Soottawat Benjakul helped with the editing of the review article.

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