

Formulation of yogurt-like product from coconut milk and evaluation of physicochemical, rheological, and sensory properties

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ABSTRACT

This study focuses on the addition of tapioca starch as a stabilizer in the formulation of yogurt-like products made from coconut milk. Yogurt-like products supplemented with tapioca starch at 0.5, 1.0, 1.5 and 2.0% (w/w) or without tapioca starch were stored at 4 °C for 14 days, and analyzed for pH changes, lactic acid bacteria viability, syneresis, rheological properties, whiteness index and sensory properties using 9-point hedonic scale. The results showed that the addition of tapioca starch did not affect the number of lactic acid bacteria during storage. Syneresis decreased with increasing concentration of tapioca starch, while the storage modulus (G' or solid-like property) increased when the concentration of tapioca starch increased. Whiteness index was not affected by supplementation of tapioca starch at the end of storage. The sensory evaluation indicated that addition of tapioca starch from 1.0% led to higher liking scores, especially texture attribute. The product made with 1.0% tapioca starch was selected based on sensory scores for proximate analysis. The product contained 71.31% moisture, 1.91% protein, 6.01% carbohydrate, 20.22% fat and 0.55% ash. This study suggested that yogurt-like product made from coconut milk can be obtained using tapioca starch as a stabilizer.

Introduction

Plant-based milk alternatives have gained massive popularity among consumers because of sustainable production compared to bovine milk. Animal welfare, environmental and health concerns are the main drivers towards plant-based milk (Raikos et al., 2020). Cow's milk allergy and lactose intolerance are responsible for some adverse effects in sensitive consumers (Gupta et al., 2010). Cow's milk allergy is one of the most common allergy in infants and children, and many studies suggested that these infants may outgrow their allergenicity (Santos et al., 2010). Lactose intolerance affects 65% of the world's population and between 70 and 100% of East Asia population (Aydar et al., 2020). Coconut (*Cocos nucifera*) is one of the important economic crops, especially in the Southeast Asian countries (Sethi et al., 2016). Recently, much attention has been paid to coconut milk as a milk substitute. It contains 31–35% fat and 3.5–4.0% protein, is high in essential amino acids, calcium, phosphorus, potassium, vitamin C, E and B6 and easily digested (Góral et al., 2018). Coconut oils are rich in medium chain fatty acids, which is clinically proven to have preventive effects against hyperlipidemia, fatty liver, and diabetes (Narayanankutty et al., 2018). Coconut milk is an

oil-in-water emulsion in nature. Coconut proteins (globulin and albumin) and phospholipids help stabilize the emulsion by adhering to the surface of coconut oil droplets as emulsifiers, preventing phase separation (Lu et al., 2019).

Yogurt is one of dairy products consumed worldwide because of various health benefits, including supporting the function of digestive system which primarily links to live microorganisms (Buttriss, 1997). Therefore, consumption of yogurt or other fermented dairy products is recommended as part of a healthy eating regime (Wong et al., 2020). The use of plant-based milk alternatives in fermented products has been extensively studied, especially soy yogurt (Donkor et al., 2007; Rinaldoni et al., 2012; Lee et al., 2018) because of its high protein content and long-term consumption of soy products, especially in Asian countries. Among other plant-based yogurts, oat (Walsh et al., 2010; Raikos et al., 2020; Demir et al., 2021), rice (Cáceres et al., 2019) and almond (Shi et al., 2020), have also been studied and commercially manufactured. Some plant sources including soy and almond, however, are allergenic ingredients, which limit people from consumption of the products. Unlike tree nuts, allergy to coconut is rare and not directly linked with nut allergy (Anagnostou, 2017). Almond, rice, and oats contain minimal

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amounts of proteins compared to cow milk (Mäkinen et al., 2016), hence gelling agents, thickeners or stabilizers must be incorporated for the manufacturing of plant-based yogurts or yogurt-like products.

However, there is a few studies based on fermented coconut milk products. Most of the studies incorporated other protein sources, such as chickpea (Mesquita et al., 2020), which can increase the risk of allergenicity or other additives, which do not meet the growing trend of clean label products (Montemurro et al., 2021). Substitution of cow milk with coconut milk in the manufacture of yogurt with high consumer acceptance and optimal characteristics is a major issue and somewhat challenging. Stabilizers or thickeners are frequently added to achieve the desired textural properties, as well as to reduce syneresis. The aim of this present study is to develop yogurt-like product made from coconut milk with minimal use of additive. From our preliminary studies, it was found that addition of tapioca starch gave smooth texture with less syneresis compared to other tested stabilizers including pectin, xanthan gum and corn starch. In this study, the samples were formulated with tapioca starch at 0.5, 1.0, 1.5 and 2.0% (w/w). The products were analyzed for pH changes, viability of lactic acid bacteria (LAB), physicochemical and rheological characteristics and sensory acceptance during storage at 4 °C.

Materials and methods

Raw materials

UHT coconut milk (Aroi-D, Bangkok, Thailand), tapioca starch (Tongchan, Bangkok, Thailand) and sugar (Mitr Phol, Bangkok, Thailand) were obtained from a local supermarket in Bangkok, Thailand. According to the labeled composition, 100 ml of coconut milk are composed of 18 g fat, 2.5 g carbohydrate and 1.25 g protein. The yogurt starter culture, YF-L812, containing *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus*, was obtained from Chr. Hansen (Hørsholm, Denmark).

Preparation of the starter culture

The stock culture was prepared by inoculating 100 mg lyophilized culture to 100 ml MRS broth (HiMedia, Mumbai, India) and incubating at 37 °C overnight. The stock culture was stored at -45 °C in MRS broth with 40% glycerol (Sah et al., 2016). The culture was preactivated in coconut milk before use following the method described in Chetachukwu et al. (2017) with some modifications by adding 1 ml thawed stock culture to 100 g autoclaved coconut milk with 5% sugar (w/w) and incubating at 43 °C overnight.

Preparation of yogurt-like product from coconut milk

Each sample of yogurt-like product was made using 2500 g of UHT coconut milk prepared in a 3000-ml beaker with addition of tapioca starch at 0.5, 1.0, 1.5, or 2.0% (w/w). The sample without addition of tapioca starch was used as a control. At least three replications for each sample were made. All samples were supplemented with 5% sugar (w/w). The mixtures of coconut milk, tapioca starch and sugar were heated to 90 °C for 3min (Yaakob et al., 2012), cooled to 43 °C, and the culture was added at 1% (w/w) and incubated at 43 °C until pH 4.6 was reached. After fermentation, the obtained products were stored at 4 °C for 14 days. The samples were randomly analyzed for viability of LAB, syneresis, rheological properties, and color on day 1, 7 and 14, while pH changes were observed on day 1 and 14. Sensory evaluation was performed after 24 h storage.

pH changes during storage

The changes in pH of yogurt-like products during storage at 4 °C were determined on day 1 and 14 using a pH meter Model Eutech pH

700 (Eutech Instrument, IL, USA) (Guzel-Seydim et al., 2005).

Viability of LAB

LAB were enumerated on MRS agar. The samples were diluted in a 10-fold series and spread on plates that contained MRS agar and incubated at 37 °C for 24–48 h on day 1, 7 and 14 of storage at 4 °C (Pachekrepapol et al., 2021). The number of LAB was reported as log Colony Forming Unit/ml of yogurt-like product (log CFU/ml).

Syneresis

The yogurt-like products from coconut milk formed in a 50-ml conical plastic centrifuge tube were stored at 4 °C before the analysis on day 1, 7 and 14. Syneresis was determined by the weight percentage of the supernatant after centrifugation at 640g at 4 °C for 10 min using Sorvall Legend XTR Centrifuge (Thermo Scientific, MA, USA). The degree of syneresis (%) was calculated as shown in equation [1] (Keogh and O'Kennedy, 1998).

$$\% \text{ Syneresis} = \frac{\text{Weight of supernatant (g)}}{\text{Weight of product (g)}} \times 100 \quad (1)$$

Rheological properties

Dynamic oscillatory measurements were performed on day 1, 7 and 14 using a controlled stress rheometer (HAAKE Mars 40, Thermo Fisher Scientific, Karlsruhe, Germany) as previously described by (Sah et al., 2016) with slight modifications. The measurement was performed using a plate and cone geometry (35 mm diameter) with 1 mm gap and operated at 4 °C. The sample was pre-sheared for 30 s at a shear rate of 500 s⁻¹ and followed by equilibration for 300 s for structural recovery. The frequency sweep test was performed in the frequency range from 0.1 to 10 Hz at 0.5% strain, which is within the linear viscoelastic range (LVR).

Apparent viscosity was measured by applying a shear rate sweep test to the same sample after equilibration for 300 s. The shear rate logarithmically increased from 0.01 to 100 s⁻¹ (Tan et al., 2018). The analyses were performed on day 1, 7 and 14.

Color measurement

Color of the samples was expressed as L* (lightness), a* (redness), b* (yellowness) color space using a colorimeter, Color Flex EZ (Hunter Associates Laboratory, Inc., VA, USA). Thirty grams of sample was weighed and placed in a 60-mm plastic petri dish. Measurements were carried out on 3 different spots of each sample after calibration using a standard whiteboard. Whiteness index (WI) was calculated through equation [2] (Pan et al., 2019). The analyses were performed on day 1, 7 and 14.

$$WI = 100 - \sqrt{(100 - L^*)^2 + a^{*2} + b^{*2}} \quad [2]$$

Sensory evaluation

Sensory evaluation was performed on 30 untrained panelists using 9-point hedonic scale according to a method described by Santiago-García et al. (2021). The participants were frequent yogurt consumers. They were asked to score the products that were stored at 4 °C for 24 h from 1 (dislike extremely) to 9 (like extremely) on 5 attributes, flavor, aroma, color, texture, and overall acceptability.

Proximate analysis of yogurt-like product

Chemical composition of selected yogurt-like product made from coconut milk was determined following AOAC (2016). Moisture content

was measured by oven drying. Protein content was obtained using the Kjeldahl method with a conversion factor of 6.25. Fat content was analyzed by solvent extraction after acid hydrolysis. The ash content was determined in a muffle furnace. Carbohydrates were calculated by difference. The selection of sample for determination of chemical composition was based on the results obtained from sensory evaluation.

Statistical analysis

One-way analysis of variance was carried out using SPSS 20 (Version 20 IBM®SPSS® Statistics, USA). The level of significant difference was determined at $p < 0.05$. The differences between means were analyzed using Duncan's multiple range test.

Results and discussion

pH changes during storage

The pH levels of all samples decreased during 14-day storage period at 4 °C as shown in Table 1. The reduction in pH during storage was also observed in other non-dairy yogurts (Bernat et al., 2015) and may be attributed to post-acidification during storage by LAB presenting in the samples (Altemimi, 2018). Post-acidification is considered an undesirable process in yogurt products because it shortens the shelf-life and cause some defects including severe acidity and syneresis (Deshwal et al., 2021). The effect of tapioca starch on pH of samples was not observed on day 1 of storage, but the control sample without tapioca starch showed significantly higher pH level compared to other samples on day 14. Although, the pH level of the control (pH 4.52) was statistically significant, the difference of about 0.03 pH unit was rather small, thus further study may be required. However, different results from addition of starch to fermented dairy products were reported by Altemimi (2018). It was found that pH of cow milk yogurt added with 1.0% potato starch (pH 4.6) after 15 day-storage was much higher than pH of the control sample (pH 3.6), which the author described as the reduction of available water when starch was added, thus making it difficult for LAB to metabolize sugar and reducing the amount of lactic acid produced.

Viability of LAB

The number of LAB of yogurt-like products from coconut milk during storage is shown in Table 2. The number of LAB did not change during 14-day storage at 4 °C, except for the sample made with addition of 0.5% tapioca starch, which showed slight decrease in LAB at the end of storage. The number of LAB found in this study is within the established quantitative standards, which vary from 6 to 7 log CFU/g (Damin et al., 2008). The sample made with addition of 2.0% tapioca starch contained lower number of LAB (5.9 log CFU/g) by the end of storage. Previous studies showed various effects of starch depending on types and

Table 1
pH of yogurt-like products from coconut milk made with addition of tapioca starch during storage at 4 °C.

% Tapioca starch	pH	
	Day 1	Day 14
Control	4.57 ± 0.01 ^{aA}	4.52 ± 0.02 ^{aB}
0.5	4.57 ± 0.01 ^{aA}	4.49 ± 0.02 ^{bB}
1.0	4.57 ± 0.02 ^{aA}	4.49 ± 0.01 ^{bB}
1.5	4.58 ± 0.01 ^{aA}	4.50 ± 0.02 ^{abB}
2.0	4.58 ± 0.02 ^{aA}	4.49 ± 0.03 ^{bB}

^{a-b} Different superscripts in the same column were significantly different ($p < 0.05$).

^{A-B} Different superscripts in the same row were significantly different ($p < 0.05$).

Table 2

The number of LAB (log CFU/g) of yogurt-like products from coconut milk made with addition of tapioca starch during storage at 4 °C.

% Tapioca starch	Number of LAB (log CFU/g)		
	Day 1	Day 7	Day 14
Control	6.25 ± 0.03 ^{bA}	6.18 ± 0.12 ^{bCA}	6.17 ± 0.06 ^{bCA}
0.5	6.20 ± 0.04 ^{bA}	6.16 ± 0.04 ^{cAB}	6.09 ± 0.07 ^{cdB}
1.0	6.45 ± 0.05 ^{aA}	6.41 ± 0.10 ^{aA}	6.38 ± 0.09 ^{aA}
1.5	6.27 ± 0.04 ^{bA}	6.31 ± 0.03 ^{abA}	6.32 ± 0.09 ^{abA}
2.0	6.21 ± 0.06 ^{bA}	6.04 ± 0.04 ^{cA}	5.96 ± 0.11 ^{dA}

^{a-d} Different superscripts in the same column were significantly different ($p < 0.05$).

^{A-B} Different superscripts in the same row were significantly different ($p < 0.05$).

concentrations on viability of LAB. Enhancement of LAB viability by addition of 2% modified waxy maize starch was reported by Prasad et al. (2013). Altemimi (2018), on the other hand, found no effect of starch at low concentration ($\leq 0.5\%$), but higher concentration (between 0.75 and 1%) of potato starch negatively affected the viability of bacteria because of lower amount of available water for bacterial viability. Therefore, tapioca starch at 2.0% in our study possibly decrease the viability of LAB, while adding 1.0% tapioca starch seemed to enhance bacterial viability. Other non-dairy yogurt-like products made from chickpea and soy milk were found to contain slightly higher number of LAB of 6.87 and 7.23 log CFU/g, respectively (Wang et al., 2018). These differences in LAB viability may be attributed to higher amount of protein content in chickpea and soy milk, which is essential in the growth of LAB, compared to coconut milk, as well as different strains of LAB used as starter cultures (Deshwal et al., 2021).

Syneresis

Syneresis is a measure of the serum released from the gel when exposed to centrifugal force. It is a quality indicator determining water holding capacity of the product (Dönmez et al., 2017). Syneresis of yogurt-like products made from coconut milk during storage is shown in Fig. 1. The syneresis level of all samples decreased with storage time. Addition of 2.0% tapioca starch resulted in significantly lower degree of syneresis compared to other samples. The sample made with 2.0% tapioca starch had an initial degree of syneresis of 19.34%, and decreased to 9.52% by the end of storage, while the control sample showed the higher levels of syneresis of 23.54% and 13.10%, respectively. The lower degree of syneresis in samples added with tapioca

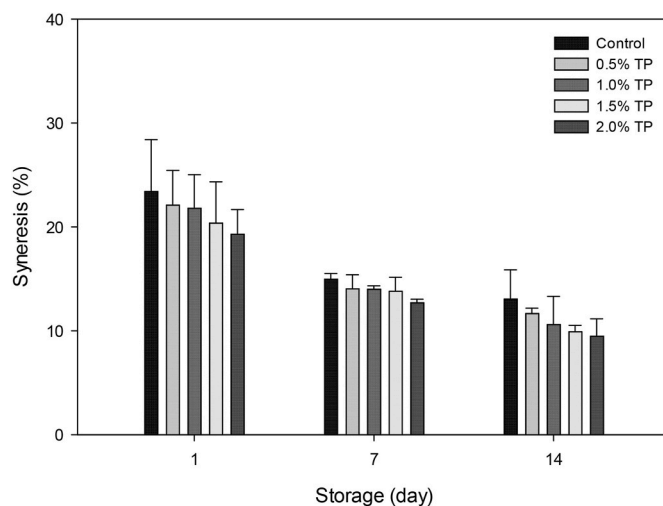


Fig. 1. Syneresis (%) of yogurt-like products from coconut milk made with addition of tapioca starch during storage at 4 °C.

starch may be attributed to an increase in water holding capacity. Many studies have shown a lower level of syneresis when starch was added to yogurts. Lobato-Calleros et al. (2014) observed syneresis reduction from low fat yogurt when tapioca and maize starch were added to the products. Starch is a thickening agent or stabilizer that has been extensively used in the production of yogurt to reduce syneresis because of its swelling property (Wong et al., 2020), leading to substantial quantities of water retained in yogurt matrix (Luo and Gao, 2011). However, much lower syneresis level (~0.7%) of commercial coconut yogurt was achieved using combined thickener and gelling agent such as starch and pectin (Grasso et al., 2020). Other studies on syneresis of non-dairy yogurts have shown different degrees of syneresis. Shi et al. (2020) reported between 26 and 27% syneresis from almond milk yogurts with xanthan gum, pectin and polymerized whey protein used as stabilizers and gelling agents. They also found a reduction of syneresis by 50% after 14-day storage, so these results suggested that the water holding capacity of samples was improved during storage.

Rheological properties

The internal structure of yogurt can be determined by the dynamic rheological measurement. Storage modulus or solid-like property (G') and loss modulus or liquid-like property (G'') values as a function of frequency on day 1, 7 and 14 are shown in Fig. 2a, 2b and 2c, respectively. All samples exhibited viscoelastic characteristics or weak gels, with G' being higher than G'' for the entire frequency range. The G' values increased with an increase in tapioca starch concentration, while the control sample showed the lowest G' values. Similar observations have been reported in yogurts made with addition of maize starch (Wong et al., 2020). The increase in G' values may be attributed to formation of a network gel-like structure by swollen starch granules at high concentrations dominating the rheological properties by increased G' values. The apparent viscosity of the products decreased with increasing shear rate as shown in Fig. 3. The decrease in viscosity during shearing indicated shear-thinning behavior of the samples (Tan et al., 2018). Viscosity of samples increased with increasing tapioca starch concentrations. These results suggested that addition of tapioca starch strengthen the gel network as starch granules swelled and absorbed water in the continuous phase during heating process resulting in enhancement of particle-particle interactions or polysaccharide-protein interactions (Grasso et al., 2020). In addition, tapioca starch was reported to be stable in acidic condition and at high temperature (Mishra and Rai, 2006) suggesting that it is suitable for the formulation of yogurts or yogurt-like products made from plant-based milk.

Color

The resulting whiteness index of yogurt-like products from coconut milk is shown in Table 3. Color is an important visual attribute in food products. Color of yogurts is affected by storage time, shelf-life and color deterioration (Coggins et al., 2010). Addition of tapioca starch did not affect the whiteness on day 1, but it was found that there were significant differences among samples after 7 days of storage. However, at the end of storage the difference in whiteness due to addition of tapioca starch was not found. There was a slight decrease in whiteness of sample made with addition of 1.5% tapioca starch on day 14. Slight change in whiteness of samples may result from Maillard reaction, which slowly occurred in heat-treated coconut milk during storage but the high rate of Maillard reaction was detected after 5 month-storage as reported by Tinchana et al. (2015).

Sensory evaluation

The sensory evaluation is shown in Fig. 4. Yogurt-like products made from coconut milk with 1.0, 1.5 and 2.0% tapioca starch received significantly higher scores for overall acceptability and flavor ($p < 0.05$).

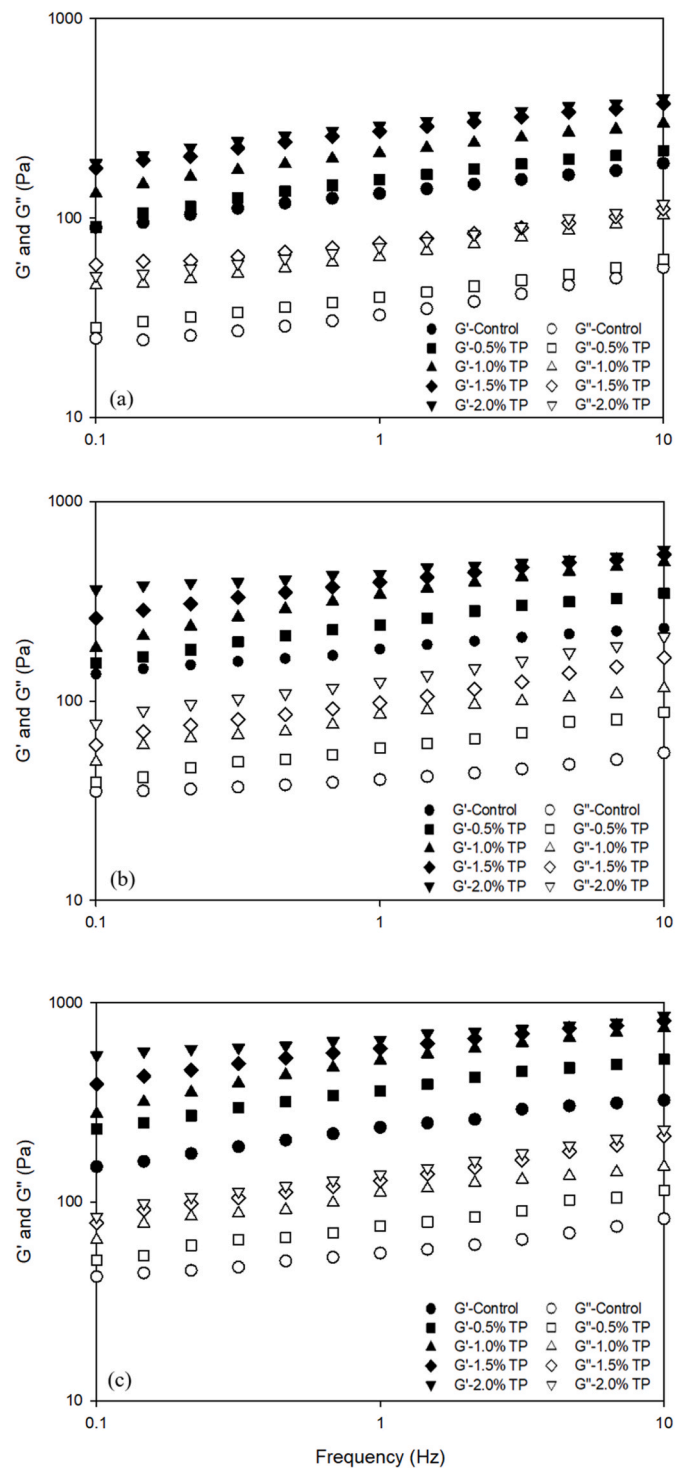


Fig. 2. G' and G'' of yogurt-like products made from coconut milk measured on day 1 (a), day 7 (b) and day 14 (c).

Products made with 0.5, 1.0 and 2.0% tapioca starch gained higher liking scores for aroma attribute. There is no significant difference of color scores among different concentrations of tapioca starch, which may correspond to the non-significant color values measured instrumentally as shown in Table 3. The products made with 1.0, 1.5 and 2.0% received significantly higher scores for texture. These results pointed out that consumers may prefer firmer texture products. Texture of yogurt is one of the major physical characteristics that determine the sensory quality and acceptability of the product. It is generally evaluated using a

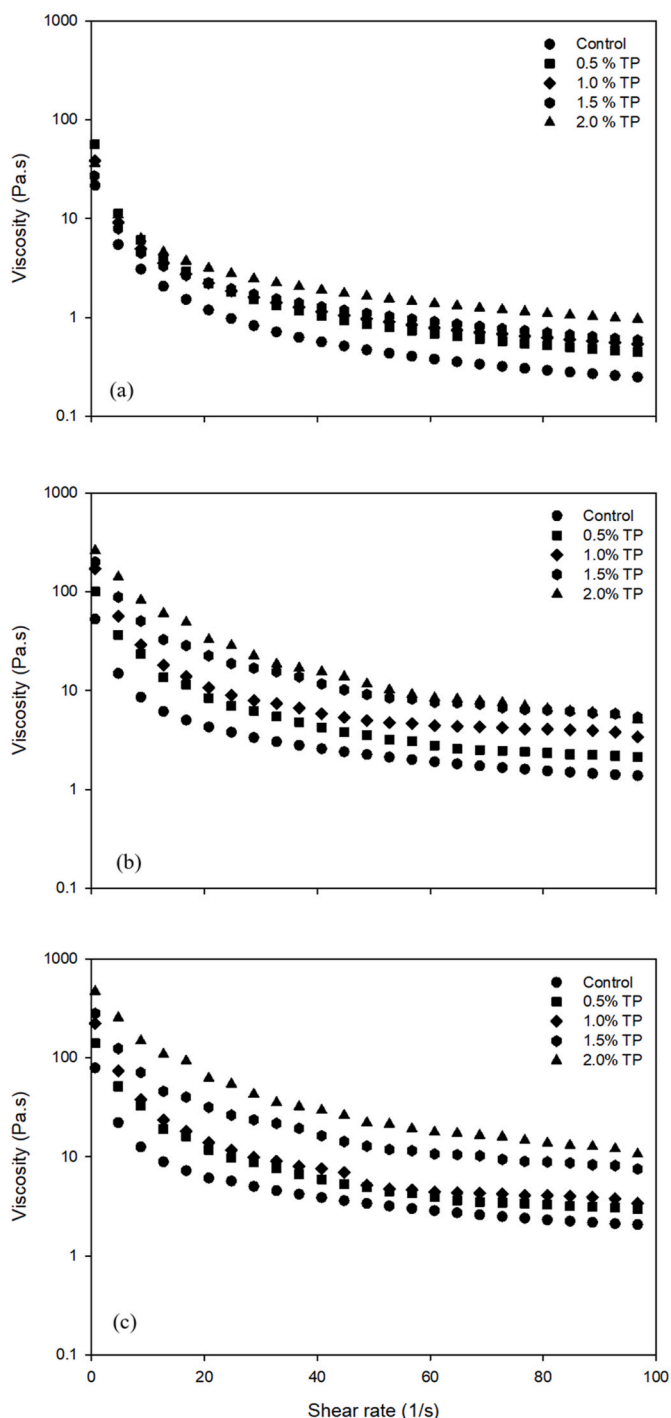


Fig. 3. Apparent viscosity of yogurt-like products made from coconut milk measured on day 1 (a), day 7 (b) and day 14 (c).

spoon or directly on the tongue (Saleh et al., 2020). The results indicated that the addition of tapioca starch from 1.0% may positively affect the sensory properties of yogurt-like products. However, some studies showed an adverse effect of addition of more than 1% of cassava starch to sensory properties of cow milk yogurt (Mwizerwa et al., 2017). The study conducted by Saleh et al. (2020) suggested to add 1% of starch extracted from potato, sweet potato, chickpea, corn, and Turkish beans to improve the sensory properties of products.

Table 3

Whiteness index (WI) of yogurt-like products from coconut milk made with addition of tapioca starch during storage at 4 °C.

% Tapioca starch	Whiteness Index		
	Day 1	Day 7	Day 14
Control	79.73 ± 2.44 ^{aA}	80.15 ± 0.23 ^{abcA}	79.68 ± 0.79 ^{aA}
0.5	80.01 ± 0.39 ^{aA}	80.78 ± 0.42 ^{aA}	78.99 ± 1.54 ^{aA}
1.0	79.53 ± 1.69 ^{aA}	79.78 ± 0.10 ^{bcA}	78.92 ± 1.39 ^{aA}
1.5	80.49 ± 0.94 ^{aA}	80.26 ± 0.46 ^{abA}	78.70 ± 0.25 ^{ab}
2.0	78.81 ± 0.93 ^{aA}	79.52 ± 0.49 ^{cA}	78.26 ± 0.46 ^{aA}

^{a-d} Different superscripts in the same column were significantly different ($p < 0.05$).

^{A-B} Different superscripts in the same row were significantly different ($p < 0.05$).

Proximate analysis of yogurt-like product

Based on the results from sensory evaluation using 9-point hedonic scale, the yogurt-like product made with addition of 1.0% tapioca starch received the highest score in overall acceptability from untrained panelists. The product with 1.0% tapioca starch, therefore, was selected for the proximate analysis. The product with 1.0% tapioca starch contained 71.31% moisture, 1.91% protein, 6.01% carbohydrate, 20.22% fat and 0.55% ash. The protein and fat contents found in this study are higher than reported in commercial coconut yogurt, which contains 0.60 g protein and 4.90 g fat/100 g (Grasso et al., 2020). The differences in protein and fat content may be due to the different manufacturing processes, or addition of water to coconut milk before fermentation in the commercial product, which may dilute fat and protein content.

Conclusion

This study highlights the importance of incorporation of stabilizer or thickener, tapioca starch, for the formulation of yogurt-like product from coconut milk. Addition of tapioca starch modify viscosity, rheological properties and syneresis of the products because of its swelling properties. Based on sensory evaluation, panelists preferred firmer and more viscous products with addition of tapioca starch between 1.0 and 2.0% (w/w). The number of LAB was stable throughout the entire storage and was within the acceptable range. To further raise the value of the described products, increase protein content by adding other protein sources can be performed with emphasis on the influence of textural and sensory properties.

Declaration of competing interest

The authors have declared no conflicts of interest for this article.

Implications for gastronomy

Yogurt is one of important dairy products consumed throughout the world due to its health benefits derived from live microorganisms. However, lactose intolerance, cow's milk protein allergy and environmental awareness have shifted the current consumer trend towards plant-based milk alternatives. Thus, the development of yogurt from plant-based milk has gained high interest because of the new opportunities. This study suggests that the use of tapioca starch as a yogurt stabilizer have led to increased consumer acceptability with improved physicochemical properties with minimal use of food additive. It is an interesting approach, which can be used by the manufacturers of milk alternative products to achieve high quality products with preferred texture, color, flavor, and aroma.

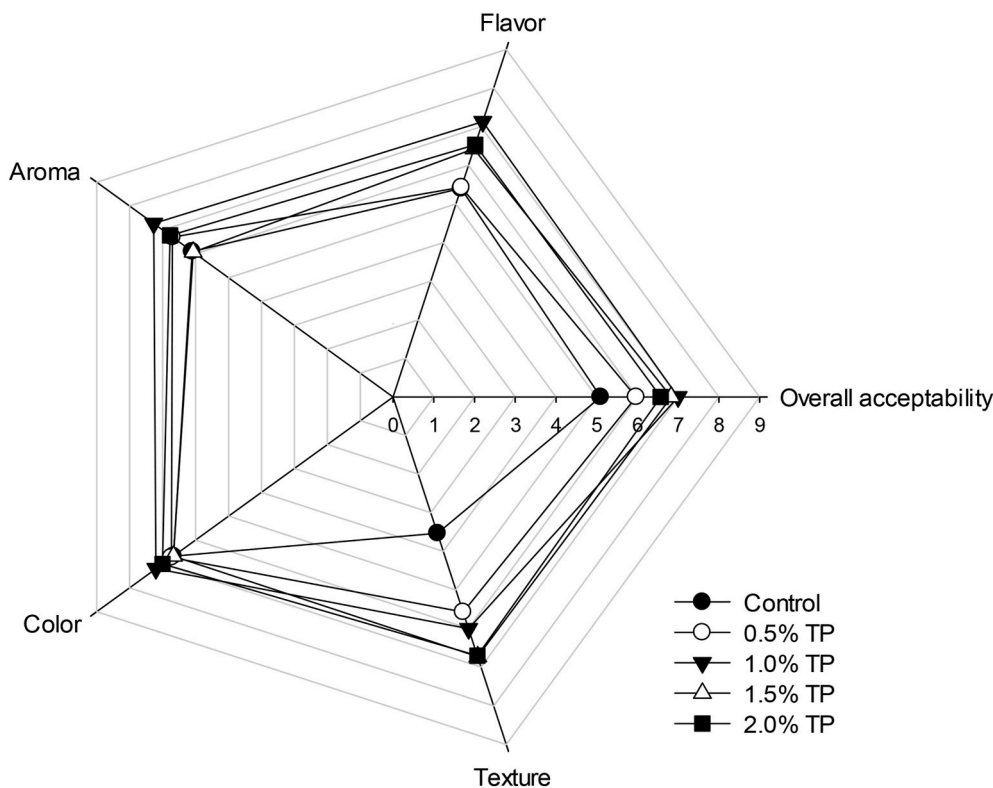


Fig. 4. Sensory evaluation of yogurt-like products from coconut milk made with addition of tapioca starch performed after 24-h storage at 4 °C.

Author's contributions

Dr. Ulisa Pachekrapapol, the principal investigator of the project, developed the research concept, designed experiments and research plan, performed experiments, analyzed and interpreted data, and wrote the manuscript. Yanin Kokhuenkhan and Jarinya Ongsawat assisted in designing experiments, performed experiments and data analyses.

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