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Food Science

DEVELOPMENT OF VITAMIN C BUTTERFLY PEA JUICE BEADS BY REVERSE SPHERIFICATION TECHNIQUE

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Abstract

The edible beads of calcium lactate for encapsulating vitamin C butterfly pea juice were transformed by frozen reverse spherification technique. This juice consisted of 1 L blue butterfly pea tea, 20 g erythritol, 50 ml lime juice and 1.2 g ascorbic acid. The influence of sodium alginate concentration solutions at 0.5% w/v, 1.0% w/v, and 1.5% w/v on the physicochemical characteristics, for example, length, width, sphericity ratio, color (*L*, *a*, and *b* values), pH, and brix were investigated. The 9-point hedonic scale, just about right, and acceptance tests of three samples were also examined. The Duncan's multiple range test of data was performed on the experiment.

The findings revealed that the application of higher concentration of sodium alginate was significantly ($P<0.05$) leading to higher *L* and *a* values of the bead, nevertheless, lower *b* value, brix, preference score of appearance, sweetness, bead membrane texture, overall liking and acceptance of the bead. The most of the respondents was male (67%) aged ranging between 21 to 35 years. The sensory results indicated that hedonic score in term of appearance, sweetness, bead membrane texture and overall liking of the vitamin C juice bead immersed at the lowest concentration (0.5% w/v) was the highest. Additionally, the respondents 41 from 50 panelists or 82% accepted this product.

Keywords: Beads, Reverse spherification, Sodium alginate, Physicochemical properties, Sensory Attributes and Just about right scale

Introduction

Spherification is a modern cuisine technique that involves creating semi-solid spheres with thin membranes out of liquids. This product provides a burst-in-the-mouth effect with the liquid inside, when chewing a sphere membrane. There are two commonly types of spherifications namely basic and reverse spherification. Basic spherification is where the liquid/beverage containing sodium alginate is submerged in a water bath containing calcium, whereas reverse spherification is where the liquid/beverage containing calcium is dropped into a water bath containing sodium alginate (Sullivan, 2019). When these two solutions come into contact, the beads or spheres are created and formed. Calcium lactate and sodium alginate are the two basic components used for this technique in this research. Sodium alginate is naturally taken from seaweed, while calcium lactate is a type of salt that widely uses in food industry. Sodium alginate is used as a binder, stabilizer, thickening agent and viscosity inducing agent. The mixing of alginate with the calcium salt, it intensifies the viscosity of liquid and transform into strong gel due to alginate polymer cross-linking (Patomchaiviwat et al., 2022).

Vitamin C, also known as L-ascorbic acid, is a water-soluble vitamin that is an essential nutrient for human health, which frequently applies in foods and beverages. It is interesting to combine vitamin C in the beverage, such as butterfly pea juice, in order to enhance health benefits, including improve immune function, extend product shelf life, improve skin health and promote wound healing as shown in this research. Mukherjee et al. (2008) reported that the butterfly pea popularly used in traditional medicine, particularly as a supplement to enhance cognitive functions and alleviate symptoms of numerous ailments including fever, inflammation, pain, and diabetes. Erythritol is a sugar alcohol that is commonly used as a sugar substitute. This sweetener naturally presents in low amounts in calorie, approximately 0.24 calories per gram, while sugar contains 4 calories per gram.

It is essential to use high-quality ingredients and employ the innovative technology, such as frozen reverse spherification technique, to ensure that the final product is safe, nutritious, and appealing for consumers. Therefore, this research aims to apply all these mentioned materials, namely, sodium alginate, calcium lactate, vitamin C, butterfly pea flower, and erythritol to study and develop the healthy edible beads. The physicochemical and sensory properties of the edible beads in different sodium alginate concentrations were also investigated.

Research Objectives

1. To study the effect of sodium alginate concentrations at 0.5% w/v, 1.0% w/v, and 1.5% w/v of vitamin C butterfly pea juice beads
2. To study the physicochemical properties of vitamin C butterfly pea juice beads
3. To study the sensory evaluation of vitamin C butterfly pea juice beads

Literature Review

1. Reverse spherification technique

The spheres can be made of different sizes and have been given names like caviar when they are small, eggs, gnocchi and ravioli when they have larger size. The resulting spheres have a thin membrane and are filled with the flavored liquid, which is flexible and can be carefully manipulated (Dhrubo Jyoti Sen, 2017). The Reverse spherification technique consists of submerging a liquid with calcium content in a bath of sodium alginate. When the liquid drops into the bath, a thin coat of gel forms around the droplet as the calcium reacts with the sodium alginate. Previous study revealed that the reverse spherification technique was effective in protecting the probiotics in the bead during storage and simulated gastric conditions (Siripatrawan and Kaewklin, 2016).

2. Vitamin C

Vitamin C, or ascorbic acid, is an essential micronutrient for human, which must be taken daily through food or supplements. It is required for human body like the biosynthesis of collagen, L-carnitine, certain neurotransmitters, and protein metabolism (Li, 2007). Furthermore, vitamin C is also an important physiological antioxidant within the body (Carr and Frei, 1999). Thus, the lack of vitamin C results in impaired immunity and higher susceptibility to infections. The recommended dietary allowance for vitamin C is shown in Table 1.

Table 1: Recommended dietary allowances (RDAs) for vitamin C adequate intake (Institute of Medicine, 2000)

Age	Male (mg/day)	Female (mg/day)	Pregnancy (mg/day)	Lactation (mg/day)
0–6 months	40	40		
7–12 months	50	50		
1–3 years	15	15		

Age	Male (mg/day)	Female (mg/day)	Pregnancy (mg/day)	Lactation (mg/day)
4–8 years	25	25		
9–13 years	45	45		
14–18 years	75	65	80	115
19+ years	90	75	85	120
Smokers	Individuals who smoke require 35 mg/day more vitamin C than nonsmokers.			

The RDAs for vitamin C is based on its known physiological and antioxidant functions in white blood cells and are much higher than the amount required for protection from deficiency. Previously, it was reported that daily supplementation with vitamin C improved skin elasticity and reduced the appearance of wrinkles in middle-aged women (Pullar, 2017).

3. **Butterfly pea juice**

Butterfly pea juice, also known as *Clitoria ternatea* juice, is a beverage made from the flowers of the butterfly pea plant. The bright blue color of this juice is unique. It is commonly consumed in Southeast Asian countries due to several health benefits. Moreover, it is widely used in traditional medicine in order to reduce fever, headache, and constipation symptom. Several studies reported that the butterfly pea extract had anti-inflammatory and antioxidant properties (flavonoids and anthocyanin), which could potentially protect against chronic diseases such as cancer and cardiovascular disease (Singh et al., 2022). It also could improve cognitive function and memory in rats as described in the research of Damodaran et al. (2018).

4. **Erythritol**

Erythritol, an ingredient category called sugar alcohol, is a common artificial sweetener. Low amounts occur naturally in fruits, vegetables, fermented foods and beverages. It is used as a zero-calorie sweetener to help replace calories from carbohydrates. In addition to providing sweetness, erythritol also helps foods retain moisture. When used as a sweetener, erythritol levels are typically more than 1,000-fold greater than levels found naturally in foods, which are not required to be listed individually on nutrition facts labels (Witkowski et al., 2023).

Methodology

1. **Materials**

Distilled water, sodium alginate, calcium lactate, erythritol and ascorbic acid (vitamin C) were obtained from Krungthepchemi Co., Ltd. (Bangkok, Thailand), which were food grade. Fresh lime and dried butterfly pea flower were purchased from a local market in Nonthaburi, Thailand.

2. **Preparation of vitamin C butterfly pea juice**

The dried butterfly pea flower (8 g) was added in the boiling water (1.2 L) and then stirred and pressed the flowers against the pot for 3 minutes. After that, the blue tea was strained with a mesh strainer in order to discard the flowers. The twenty grams of erythritol was put in the one litre of warmed blue tea. Next, the fresh limes were cut and then squeezed the water out by hands. The lime juice about 50 ml and ascorbic acid 1.2 g was mixed in the butterfly pea juice.

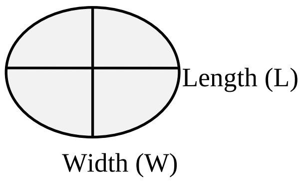
3. **Preparation of juice beads by reverse spherification technique**

Calcium lactate 0.06 g was dissolved and mixed with 1 L vitamin C butterfly pea juice and they were poured into sphere silicone mould. After that, they were frozen at -15°C for 24 hr. The frozen liquids were immersed into the bath containing various concentrations (0.5-1.5% w/v) of sodium alginate solution for 5 min to form the bead membrane. The obtained beads were then removed from the bath and transferred to three distilled water baths to rinse off any excess sodium alginate. This reverse spherification method was slightly modified in accordance to the research of Kamprawet and Vatthanakul (2018). The vitamin C juice beads, soaked in different sodium alginate solutions including 0.5% w/v, 1.0% w/v, and 1.5% w/v, were investigated and compared the physicochemical and sensory properties in this study.

4. Physicochemical properties determination

4.1 Sphericity ratio

The sides of beads were measured by using a digital Vernier. The length (L) and width (W) of the bead were determined. The findings were represented as the mean of ten measurements. Sphericity ratio was calculated by using the equation in the same way as described in the research of Kamprawet and Vatthanakul (2018). If the ratio was close to 1, it meant that the bead was round due to the little difference between their length and width.



$$\text{Sphericity ratio} = \frac{Length\ of\ bead\ (mm)}{Width\ of\ bead\ (mm)}$$

4.2 Weight

The bead was weighed with a Mettler analytical balance. The findings were reported as the mean of six measurements.

4.3 Color

The color of samples was measured with a portable colorimeter as lightness (L), redness (a), and yellowness (b). The colorimeter was calibrated against a standard white tile (L = 94.81, a = −0.53, and b = 5.672. Measurements were conducted in triplicate. The L, a, and b values represent as described by Samard and Ryu (2018).

4.4 Acidity and sweetness (brix)

The pH and sweetness measurements of the vitamin C juice inside the beads (i.e., before and after burst of bead) were examined by using a calibrated digital pH meter and a calibrated digital refractometer, at 25°C, respectively. The results were averaged of six treatments.

5. Sensory Evaluation

A 9-point hedonic scale was commonly used to assess the acceptability of food product. The criteria used were: blue color, appearance, sweetness, sourness, flavor, bead membrane texture and overall liking on a scale of 1 to 9, where 9 represented "like extremely" and 1 represented "dislike extremely." The 3-scale just about right test was used for measuring attribute intensity and acceptability simultaneously (Carvalho et al., 2017). The attributes were designed as continuous line scale with three descriptive principles, low intensity (much too weak) on the left end, just-about-right or acceptance at the center, and high intensity (much too strong) on the right end, score ranging from 1 to 3, respectively. If the net score is less than -20, the attribute intensity should be increased. On the contrary, if it is higher than 20, the attribute intensity should be decreased. The net score between -20 and 20 means that the attribute intensity is just about right. Moreover, the acceptance of vitamin C juice beads was inquired.

A panel of 50 untrained students and staff aged ranging from 21 to 35 years, Food Business Management Department, Panyapiwat Institute of Management, attended in the sensory evaluation. Every panelist was served three beads from three different sodium alginate concentration solution s (0.5% w/v, 1.0% w/v, and 1.5% w/v). Samples were blind coded with random three-digit numbers and the order of serving samples was randomized so that each sample occurred equally. One cup of water was served to rinse.

Statistical Analysis

Vitamin C juice bead data: percentage, mean and standard deviation were analyzed by using IBM SPSS software version 24.0 (IBM, Armonk, NY, USA). Significant differences among treatments were determined at $p<0.05$ using Duncan's multiple range test.

Results and Discussion

The physicochemical properties of vitamin C butterfly pea juice beads at three different sodium alginate concentration solutions, including 0.5% w/v, 1.0% w/v, and 1.5% w/v, presented in **Table 2**. There was no significant difference in the length, width and sphericity ratio of all three samples, which could be due to using the same size of sphere silicone mold. In addition, the weight of the bead, soaked in the lowest concentration solution at 0.5% w/v (15.06 g), was significantly ($p<0.05$) lighter than those of the rest concentration solutions (16.67 g to 16.77 g), which might be attributed to the lower forming of the bead membrane texture.

The color parameters of all samples were significantly different ($p<0.05$) by the immersion of sodium alginate concentration solution as shown in **Table 2**. The application of higher sodium alginate concentrations brought to the higher *L* and *a* values of the beads, however, the lower in *b* value. The pH of the vitamin C juice before and after bead bursting, ranging from 5.23 to 5.32, did not differ significantly ($p<0.05$), which was in accordance to the pH of energy drink in the previousresearch of Patomchaiviwat et al. (2022). As regards to sweetness (brix), the increasing of sodium alginate concentrations from 0.5 % w/v to 1.5 % w/v was significantly different leading to the lower sweetness of the vitamin C juice.

Table 2: Physicochemical properties of vitamin C butterfly pea juice beads

Properties	Sodium alginate concentration (% w/v)		
	0.5	1.0	1.5
Length (mm)	34.20 ± 1.00^{ns}	33.77 ± 1.57^{ns}	34.60 ± 1.00^{ns}
Width (mm)	26.51 ± 1.00^{ns}	26.03 ± 1.00^{ns}	26.81 ± 1.00^{ns}
Sphericity ratio	1.29 ± 0.97^{ns}	1.30 ± 0.98^{ns}	1.29 ± 1.00^{ns}
Weight (g)	15.06 ± 0.97^{b}	16.67 ± 0.98^{a}	16.77 ± 1.00^{a}
L	34.48 ± 1.52^{b}	35.12 ± 1.52^{a}	35.37 ± 1.62^{a}
a	-0.42 ± 1.52^{b}	-0.07 ± 0.97^{a}	0.05 ± 0.87^{a}
b	16.02 ± 1.00^{a}	15.00 ± 1.00^{b}	14.74 ± 1.00^{c}
pH	5.32 ± 0.97^{ns}	5.23 ± 1.12^{ns}	5.27 ± 1.15^{ns}
Brix (%)	7.85 ± 1.15^{a}	6.30 ± 1.23^{b}	6.44 ± 1.50^{b}

Values are means ± standard deviation
Different letters (a–c) in the same row are significantly different at $p<0.05$

The three samples of vitamin C butterfly pea juice beads were immersed into different sodium alginate concentration solutions at 0.5 % w/v, 1.0 % w/v, and 1.5 % w/v, respectively. The results indicated that the fifty respondents who participated in the sensory evaluation of this research were female 33% and male 67%, aged ranging from 21 to 35 years. The sensory attributes of three juice beads were significantly different ($p<0.05$) dependent on sodium alginate concentration solutions (0.5% w/v to 1.5% w/v) as indicated in Table 3. The highest overall liking score (6.58) was the sample at 0.5% w/v sodium alginate concentration, compared to those two concentrations (1.0% w/v and 1.5% w/v). The increasing of sodium alginate concentrations from 0.5% w/v to 1.5 % w/v was leading to the lower score of sweetness, bead membrane texture and overall liking ($p<0.05$). Moreover, the preference score of blue color, sourness, and flavor scores showed no statistically significant differences at 0.05 level among three samples.

Table 3: Sensory attributes of vitamin C butterfly pea juice beads

Attributes	Sodium alginate concentration (% w/v)		
	0.5	1.0	1.5
Blue color	4.20 ± 0.30^{ns}	4.07 ± 0.57^{ns}	4.00 ± 1.00^{ns}
Appearance	7.06 ± 0.75^{a}	6.67 ± 0.83^{b}	6.77 ± 1.60^{b}
Sweetness	6.32 ± 0.01^{a}	5.53 ± 1.74^{b}	4.63 ± 1.32^{c}
Sourness	5.20 ± 1.02^{ns}	5.43 ± 1.23^{ns}	5.37 ± 1.24^{ns}
Flavor	5.02 ± 0.52^{ns}	5.27 ± 0.22^{ns}	5.37 ± 0.72^{ns}
Bead membrane texture	6.02 ± 0.00^{a}	5.00 ± 0.10^{b}	4.74 ± 0.50^{c}
Overall liking	6.58 ± 1.45^{a}	5.30 ± 1.23^{b}	5.44 ± 1.80^{b}

Values are means ± standard deviation
Different letters (a–c) in the same row are significantly different at $p<0.05$

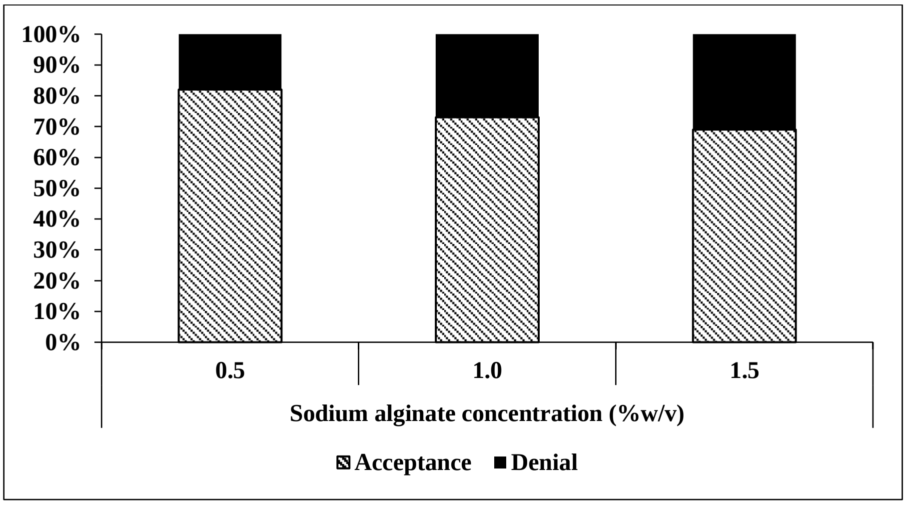
The just about right (JAR) score of the sample at 0.5% w/v sodium alginate concentration provided the different consideration amongst three samples as demonstrated in **Table 4**. The results reveled that all three vitamin C butterfly pea juice beads were too dark, and the blue color intensity of the beads needed to be decreased, while the sourness and flavor of three samples were too low, and considered to be increased. Besides, the appearance intensity of three beads was just about right. For the sweetness, the samples at sodium alginate concentrations of both 1.0% w/v and 1.5% w/v were not enough and needed to be increased, while the bead membrane texture of these samples were too tough to chew and needed to be softer. This might be due to these membrane beads were quite thick and sticky, especially the sample at the highest concentration (1.5% w/v). It might be explained that the higher sodium alginate concentration lead to the stronger membrane bead, which the respondent felt difficult to chew. Patomchaiviwat et al. (2022) also described that increasing the sodium alginate concentration resulted in a slight increase in shell thickness.

Interestingly, the bead at the lowest sodium alginate concentration solution (0.5% w/v) presented just about right consideration in appearance, sweetness and bead membrane texture, which be recommended not to be improved (**Table 4**). The result of just about right scale was in accordance to the 9-hedonic scale that respondents preferred sensory attributes of the sample at the lowest concentration to those of the rest samples at 1.0% w/v and 1.5% w/v concentration.

Table 4: Just-about-right (JAR) consideration of vitamin C butterfly pea juice beads

Sodium alginate concentration (%w/v)	Attributes	Intensity (%)			Net score	Consideration
		Too weak	JAR	Too strong		
0.5	Blue color	3	63	34	31	Decrease
	Appearance	15	75	10	-5	JAR
	Sweetness	15	70	15	0	JAR
	Sourness	36	64	0	-36	Increase
	Flavor	35	65	0	-35	Increase
	Bead membrane texture	19	67	14	-5	JAR
1.0	Blue color	0	54	46	46	Decrease
	Appearance	7	70	23	16	JAR
	Sweetness	44	56	0	-44	Increase
	Sourness	40	60	0	-40	Increase
	Flavor	44	53	3	-41	Increase
	Bead membrane texture	0	54	46	46	Decrease
1.5	Blue color	6	60	34	28	Decrease
	Appearance	13	74	13	0	JAR
	Sweetness	37	52	11	-26	Increase
	Sourness	49	51	0	-49	Increase
	Flavor	35	61	4	-31	Increase
	Bead membrane texture	0	48	52	52	Decrease

The percentage of acceptance score of three vitamin C juice beads at different sodium alginate concentration solutions (0.5% w/v to 1.5% w/v) displayed in Picture 1. The beads, soaked in the lower sodium alginate concentration solution, indicated more acceptance score. Remarkably, the sample at 0.5% w/v concentration showed the highest acceptance score (82%) than those two concentrations. On the other side, few panelists about sixteen percent (8 from 50 panelists) did not accept this product (0.5% w/v concentration) by reasoning of dark blue color, little sourness and odorless of the bead. They mentioned that the product needed to further developed and improved.



Picture 1: The acceptance of vitamin C butterfly pea juice beads

Conclusion

The frozen reverse spherification technique was used to practically prepare edible vitamin C beads of calcium lactate for encapsulating juice comprising 1 L blue butterfly pea tea, 20 g erythritol, 50 ml lime juice and 1.2 g ascorbic acid. The beads were created by freezing calcium lactate-containing vitamin C drinks and dropping them into sodium alginate solution.

The comparison of three juice beads immersed with different sodium alginate concentration solutions revealed that the utilization of lower concentration of sodium alginate, the higher *b* value, brix, preference score of sweetness, bead membrane texture, overall liking and acceptance of the bead. The vitamin C butterfly pea juice bead immersed at the lowest concentration (0.5% w/v) was outstandingly the highest preference score of appearance, sweetness, bead membrane texture and overall liking. The respondents liked this bead slightly. Eighty-two percent of respondents (41 from 50 panelists) accepted the product. However, the additional study on the development of this bead is needed.

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UTILIZATION OF WASTE SWEET PICKLED MA-MUANG-BAO SYRUP FOR PRODUCTION OF GUMMY

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Abstract

This research aims to use the waste syrup from sweet pickled Ma-Muang-Bao production for gummy development. The gummy basic recipe was chosen from three commercial gummy recipes by using rank sum differences method. The gummy recipe that gained the highest rank sum difference was chosen for further experiment. In order to develop the gummy recipe and investigate the factors that affected the sensory attributes of the gummy, the experiment was designed by using 2 x 2 factorial, two levels of the waste pickled mango syrup's total soluble solid (23°brix and 33°brix) and citric acid (0.5% and 1%) were examined by using the sensory evaluation. For the sensory evaluation, consumer preference test and just about right test were conducted by using a total of 30 panelists. The sample that had highest preference score was chosen and further improved for its better attributes according to the just about right score. Consumer acceptance and buying decision was examined by a total of 50 panelists.

The results revealed that the rank sum differences of the overall liking of gummy recipe B which contained 8% of gelatin, 31% of sugar, 30% of glucose syrup, 1% of citric acid and 30% of waste pickled mango syrup had the highest score on texture and overall liking but not significantly lower than gummy recipe A on taste and flavors. The study on the factors that affected the gummy sensory found that the total soluble solid of the waste syrup (brix) was the main factor that affected the pickled mango flavor, sweetness and saltiness of the gummy while yellow color, gumminess and sourness of the gummy were depended on citric acid content. Gummy recipe B made from 23° brix of waste pickled mango syrup and 0.5% of additional citric acid (T1), had the significantly highest liking score on overall preference, yellow color, pickled mango flavor, sweetness, saltiness and sourness. The consumer acceptance test revealed that the majority of the panelists very liked the gummy color (8.02 ± 0.68), the gumminess (8.34 ± 0.48), the sweetness (8.24 ± 0.49) and overall preference (8.56 ± 0.40) while they were moderately like the gummy flavor (7.86 ± 0.48), sourness (7.87 ± 0.37) and saltiness (7.76 ± 0.58). The majority of panelists (98%) accepted this product and 95% of them decided to purchase the developed gummy at 35 THB per 70 g (7 pieces). The researchers suggest that further market research should be performed to develop effective product positioning strategies.

Keywords: gummy jelly, sweet pickled mango, syrup, value-added product

Introduction

Gummy is made with a base of gelatin. Customers of all age groups, especially children, prefer gummies due to their attractive texture, appearance, taste, and flavor that is easy and enjoyable to bite. Gummies represent approximately 18% of global sugar confectionery market value. The global gummy market is continue growing and is expected to reach 17.7 billion US dollar by 2025, growing at a CAGR of 3.5% during the forecast period of 2019 – 2025 (Grand View Research, 2019; Dataintelo, 2022). The rising demand driven by people of all ages since gummies are available in a wide range of flavors and varieties, making them an attractive option for all customer. A report by Grand View Research revealed that flavor of the gummy is an important factor driving the consumers' preferences for gummies. Gummies prepared from exotic fruits, vegetables, liqueur can attract a large number of consumers.

Ma-Muang-Bao, Bao mango, Tiny Mango or Mini Mango (*Mangifera indica L. Var.*) is a small size light weight mango which is widely known for its sour taste, unique flavor, crisp and firm texture. It provides high mangiferin, phenolic compounds, calcium, vitamins A, B and C which is good for health. Bao mangoes abundantly grown in Songkhla province, especially at Singha Nakhon, Krasae Sin, Ranot and Sathing Phra District. It is an important economic crop of Thailand that is exported to various countries. In order to preserve the oversupply mango, various forms of mango products have been developed.

Thai style sweet pickled Bao mango or Ma-Muang-Bao Chae-Im (GI, GI65100190) is one of a mango product which is a well-known geographical indication product and also a famous souvenir of Songkhla province, Southern part of Thailand. Sweet pickled Bao mango is first pickling in brine and end up in syrups containing a high concentration of sugar. A syrups used for mango preservation is abundantly discharged as a food processing waste, more than 1000 liters per day, which can create environmental problems if it is discarded in open lands and drains without waste water treatment (Uthairungsri et.al., 2019). It can also contribute to greenhouse gas emissions, water pollution, and other forms of environmental degradation. However, this waste syrup has a unique mango aroma and flavor and contained high sugar and acid contents that can be utilized to develop a variety of new product. Thus, it will be helpful to the producer to reduce the amount of waste syrup that consequently reduce the environmental problem, the cost of water treatment and can create new revenue streams from the production of new value-added product.

In this study, the waste syrup generated from sweet pickled Ma-Muang-Bao production was used as raw material for the production of gummy as a value added alternative of using these by-products. Since SPM syrup had unique aroma and flavor and contained high sugar and acid contents, it was suitable to be developed to sorbet. In addition, qualities and properties of sorbet were evaluated.

Research Objective

1. To develop gummy from the sweet pickled Ma-Muang-Bao waste syrup.
2. To study the consumer's acceptance toward the developed gummy

Literature Review

1. Ma-Muang-Bao

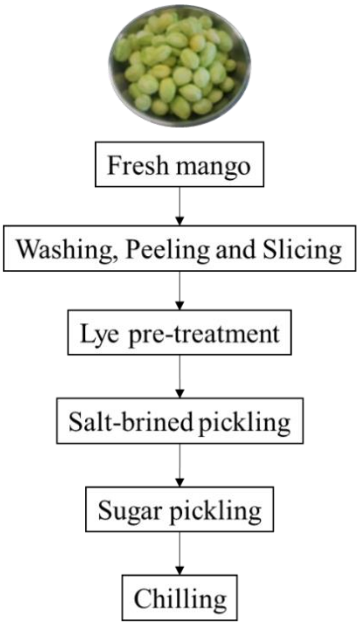
Ma-Muang-Bao, Bao mango, Tiny Mango or Mini Mango (*Mangifera indica* L. Var.) is a variety of mango in the Anacardiaceae family which has a smaller fruit when compared to other mango varieties. It is known for its small size, oval-shaped, with a small seed and thin skin. Its size varies from 4-6 centimeters and weighs between 50-60 grams as show in **Picture 1.**

Bao mangoes abundantly grown in Songkhla province, especially at Singha Nakhon, Krasae Sin, Ranot and Sathing Phra District. Generally, Bao mangoes are harvested when they are young (20 - 50 days after flowering) which is indicated by their smooth light green color and soft thin layer seed coat. Bao mangoes are harvested from March to April in Thailand, and the peak season is from September to October. It can produce up to 400-600 fruits per tree per year (Office of the Permanent Secretary for Ministry of Agriculture and Cooperatives, 2020). Yong Bao mango has a sour taste, unique aroma and flavor and white firm crispy texture which can be eaten fresh, cooked or used in a variety of processed forms, such as brine or sweet pickle, juice, jam, or dried mango slices.

Picture 1: Ma-Muang-Bao fruit

2. Sweet pickled mango

Sweet pickled mango or Ma-Muang Chae-Im is a traditional preserved mango of Thailand. In order to control the microorganisms and extend the mango shelf life, with ensuring its safety and quality, the water content in mango is reduce by using the osmosis dehydration. This phenomenon is involving with the diffusion of water from fruit or vegetable to the solution (generally sugar syrup or brine) and the migration of sugar or salt towards the fruits and vegetables. The water remove from lower concentration of solute to higher concentration through semi permeable membrane until the concentration of the solute reaches the equilibrium condition in both sides of membrane (Tiwari 2005). The advantage of this preservation is that it can retain vitamin and minerals, color, flavor and taste of mango. There are three types of osmotic agent use in sweet pickled mango including lye, brine and sugar. Accordingly, sweet pickled mango has crispy texture with sour-sweet and salty taste. The process of sweet pickled mango has been shown by the following flow diagram.



Picture 2: Flow diagram for sweet pickled mango process

For sweet pickled mango process, after washing, peeling and halving, the mangoes are lye treated by 1.5% CaCl for 10-12 hours for the crispiness of the mango. After pre-treatments, the fruit pieces are pricked in brine-salt at a concentration of about 5-10% for 10-12 hours to reduced water, prevent oxidative and non-enzymatic browning. The mangoes are then moved and immersed in sugar solution at 30 – 45 °Brix for 24 hours to reduce more water. The sugar is added periodically to maintain the sugar concentration level at 45-50 °Brix and immersed for additional 24 hours.

3. Gummy

Gummy or dry jelly is a type of soft confectionery products that has a chewy gel-like structure with soft texture. It contains 5 to 10% gelatin, 16 to 20% moisture, up to 75% sweetener solids and 0 to 2% acids. It has various shape and size such as bears, worms, or fruits, depending on its mold. It is made from a mixture of three main ingredients including fruit juice, gelling agents and sugars (in the form of sucrose and/or glucose syrup) combined with acid, flavorings, and colorings (Marfil et al., 2012). Gummy is made by boiling of gel components and sugars at high temperatures. After boiling, the mixture was mixed with the food acid, flavor and color agents. The obtained mixture is poured into gummy molds. The gummy molds are placed in a cool and dry place (Hay, 2016).

3.1 Gelling agent

Gelatin usually use as a gelling agent in gummies. It is a high molecular weight polypeptide made from animal collagen (Atik et. al., 2022). However, there are also plant-based gummies that use alternative ingredients such as carrageenan or pectin (De Mars et.al., 2001; Song et.al., 2022)

Gelatin is a water soluble hydrocolloid that provide firm and elastic nature in gummy products at concentrations 5-10 wt %. Gelatin has unique gel-forming ability. It can dissolve in hot water and remains in a solution state at temperatures higher than 40 °C. Gelatin gel system is formed in the presence of sugar and glucose syrup. Generally, gelatin is in the form of tablets, granules or powders. Based on the pre-treatment of gelatin production, there are two different types of gelatin:

acid treatment gelatin (type A gelatin) and alkaline treatment gelatin (type B gelatin). Both type of gelatin may be used in gummy formulations but its isoelectric point is different. Alkaline treatment gelatin has a higher crosslinking degree, resulting in a slower degradation rate compared to the acid treatment gelatin (Hartel, von Elbe, & Hofberger, 2018).

The role of gelatin in gummy jelly was to form a gelling network (Guo et al., 2003). In gelling process, heating at temperatures above the gelling temperature, gelatin will dissolve into colloids solution, but at the temperature below 35 to 40°C, the solution will reform into gel network. The thermoreversible change of gelatin between solution and gel is called the sol–gel transition. However, long time heating of gelatin gel can resulted in the decomposition and it would not be reformed after cooling. (Bigi, Panzavolta, & Rubini, 2004).

The gelling process can be affected by many factors, such as gelatin bloom index, concentration, pH, and temperature which leading to a critical attribute of gummies (Banerjee & Bhattacharya, 2012). For example, the stiffness of the gelatin gel is depended on the bloom index. High bloom index resulted in high strength of a gel. Acid can weaken the gelatin gel network by inhibit the formation of hydrogen bonds. Moreover, the incorporating of hydrocolloids or inulin into gelatin can enhance the gel strength (Ge et al., 2021).

3.2 Sugar

Gummy is a kind of sweet that have high sugar content (including sucrose and glucose syrup). Gummy contains 55 to 75% of sugar (Zhang, Y., & Barringer, S., 2018). Sugar plays an important role on the gel structure and sensory properties of the gummy, but also the rheological properties, the mechanical properties and the water activity of the gummy which involving in shelf life and microbiological risks (Tau and Gunasekaran, 2016).

The presence of sugars in gelatin gels structure can enhance gel strength and thermal stability. Furthermore, sugar provides mouthfeel to the gummies and increases the mass of the product. However, soluble sugar (°Brix) has no effect on the gelling temperature of the gelatin gummy until it reached 80 or higher when the gelling temperature decreased probably due to the extrusion of sugar from the gelatin network (Ge et al., 2021).

Gummies require an extra-fine granulated grade of sugar which can dissolve easily in gel solution and in order to prevent crystallization of the sugar in finished product, glucose syrup can be used. In addition, glucose syrups can help reduce water activity of the gummy and prevent microbial growth.

Methodology

1. **Preparation of waste sweet pickled mango syrup**

Ma-muang Bao was pickled in sugar syrup which contained 45% of sugar for 24 hours, after that adjust the syrup total soluble sugar to 45 °Brix and continued the pickling for additional 24 hours. The waste sweet pickled Bao mango syrup (WBMS) was by-products from sugar pickling step of sweet pickled mango production. The syrup was filtered through a white cloth sheet before the chemical properties (pH, total soluble solid and salt content) were analyzed.

2. **The selection of gummy recipe**

In order to select the most favorite gummy recipe, three commercial gummy recipes (A, B and C) were compared. The recipes were prepared with 5 main ingredients; gelatin, sugar, glucose syrup, citric acid and sweet pickled Bao mango syrup (WBMS). Each gummy recipe has the same list of ingredients but different in ratio according to **Table 1.**

Table 1: Ingredients of three gummy recipes (Total weight 100 grams)

Ingredients	Recipes (%)		
	A	B	C
Gelatin	6	8	10
Sugar	31	31	31
Glucose syrup	35	30	7
Citric acid	-	1	2
WBMS	28	30	50

Initially, the gelatin was poured into WBMS and stirred to let the gelatin absorb the liquid for 5 minutes. The sugar and glucose syrup were mixed in a saucepan and bring to a simmer. After the sugar melt, the gelatin mixture was added and stirred with high heat until the temperature reached 80 ^{0}C. When all the ingredients were completely dissolved, the saucepan was removed from the heat. In case the citric acid was present in the recipe, stirred it in and let the mixture sit for 5-10 minutes until the foam rise to the surface. The foam was scoop off by a spoon. The gummy mixture was then filled into the mold. After that, the molds were kept at 4°C for 6-24 hours before unmolding.

Ranking for preference test of the three difference gummy recipe on overall liking, taste flavors and texture were conducted by using 30 panelists. The differences between rank sum were calculated and compared with the critical values of differences between rank sum at $p = 0.05$ (critical value= 18.2, N= 30, Sample = 3) as method described by Christensen et al. (2006). Gummy with significantly maximum rank sum difference in overall liking was chosen as the most favorite gummy recipe.

3. Development of gummy from waste sweet pickled Bao Mango syrup

To examine the appropriate ratio of total soluble solid (23 and 33 °Brix) and citric acid content (0.5% and 1%) of WBMS using in four gummy treatments (T1, T2, T3 and T4) on gummy characteristics, the experiment was planned by the 2x2 factorial experiments in RCBD at a 95 percent confidence level as shown in **Table 2**. The sensory quality in terms of appearance, color, flavor, sweetness, sourness, saltiness, gumminess and overall preferences was assessed by hedonic preference test (9-point hedonic scale; 1 = “dislike extremely,” 9=“like extremely”) using 30 panelists who is a student in Panyapiwat Institute of Management, Nonthaburi, Thailand. The Just about right test (JAR) on the four samples were also conducted by using 30 panelists. The panelists were asked to evaluate the intensity of yellow color, mango flavor, sweetness, sourness, saltiness and gumminess attributes using the 3-scale of Just About Right (1= “not enough” 2= “just about right” 3= “too much”). The JAR data were evaluated using net score (% Too much - % Not enough). If the net score is less than -20, the attribute intensity should be increased and If the net score is higher than 20, the attribute intensity should be decrease. The net score between -20 to 20 means that the attribute intensity is just about right.

Table 2: Gummy recipes with difference total soluble solid and citric acid content of WBMS

Treatment	Citric (%)	Total soluble solid (%)	pH
T1	0.5	23	3.48
T2	1	23	3.33
T3	0.5	33	3.44
T4	1	33	3.31

4. Consumer Acceptance Test

A sensory evaluation and consumer acceptance test of the developed gummy recipe was conducted with 50 target consumers. The selected consumers were asked to evaluate the developed recipe in terms of the following parameters; appearance, color, flavor, sweetness, sourness, saltiness, gumminess and overall preferences of the gummy, using the 9-point hedonic scale (1 = "dislike extremely," 9="like extremely"). The experimental data were expressed as mean ± standard deviations. For consumer acceptance test, the consumers were asked for the acceptance and purchasing intention on the developed recipe. The data were expressed as percentage.

Results and Discussion

1. The selection of gummy recipe

The chemical properties of WBMS (pH, total soluble solid and salt content) were analyzed and shown in **Table 3**. It was found that WBMS contained lower total soluble solid than the initial syrup (45°Brix) but higher than normal fruit juice that usually has a Brix around 10-25, depended on the type of fruit juice (Serpen, 2012). According to the sweet pickling step, the osmosis diffusion of salt (from brine pickling step) and organic acid from pickled mango to the syrup cased the syrup has higher acid and salt content than initial syrup as shown in **Table 3**.

For the comparison of three WBMS gummy recipes (A, B and C), the WBMS was used to substitute fruit juice in each gummy recipe with different ratio of gelatin, glucose syrup, juice and citric acid.

Table 3: Chemical properties of the waste sweet pickled Bao mango syrup (WBMS)

Properties	Total soluble solid (%)	Salt (%)	pH
WBMS	31.0 ± 1.6	8.16 ± 0.62	3.47 ± 0.01

The rank sum differences (Max –Min) results of three gummy recipe shown that the consumer preference towards all of the three recipes were significantly different at a 95 percent confidence level (**Table 4**). The rank sum differences of the overall liking of recipe B which contained 8% gelatin, 31% sugar, 30% glucose syrup, 1% citric acid and 30% WBMS had the significantly highest score, accordingly, it was chosen as the most favorite gummy recipe. On the other hand, the rank sum differences of its taste and flavor were not significantly difference from recipe A. Also, its texture was not significantly difference from recipe C.

Table 4: The sum of ranking differences (SRD) on gummy recipes

Attributes	Rank Sum difference		
	A	B	C
Taste	74 ^{a}	66 ^{a}	40 ^{b}
Flavors	75 ^{a}	64 ^{a}	41 ^{b}
Texture	38 ^{b}	66 ^{a}	76 ^{a}
Overall liking	54 ^{b}	79 ^{a}	47 ^{b}

Note: Rank sum with the different superscripts in the same rows are significantly different (P = 0.05) using Christensen's critical value.

2. Development of gummy from waste sweet pickled Bao Mango syrup

The result from the selection of gummy recipe revealed that the ranking score in taste and flavor of gummy recipe A was higher than gummy recipe B and C, respectively. This might because the recipe A has lower amount of WBMS and citric acid than recipe B and C. Since the WBMS contained sugar, salt and organic acid, so its concentration might have affected the gummy taste and flavor.

Table 5: Consumer liking scores on difference recipe of gummy from WBMS

Attributes	T1 (23 Brix, 0.5%Citric)	T2 (23 Brix, 1%Citric)	T3 (33 Brix, 0.5%Citric)	T4 (33 Brix, 1%Citric)
Yellow color	7.50±0.51 ^{ab}	7.90±0.84 ^{a}	7.33±0.71 ^{b}	7.83±0.91 ^{a}
Appearance	7.63±1.07 ^{ns}	7.67±0.66 ^{ns}	7.57±0.68 ^{ns}	7.60±0.72 ^{ns}
Pickled mango flavor	7.67±0.76 ^{a}	7.57±1.04 ^{a}	6.20±1.94 ^{b}	6.17±1.93 ^{b}
Gumminess	8.13±0.68 ^{ns}	7.83±0.79 ^{ns}	8.07±0.64 ^{ns}	7.80±0.61 ^{ns}
Sweetness	8.20±0.71 ^{a}	8.00±0.59 ^{a}	7.47±0.63 ^{b}	7.23±0.68 ^{b}
Sourness	7.97±0.76 ^{a}	6.40.±1.00 ^{c}	7.40±0.67 ^{b}	6.53±1.43 ^{c}
Saltiness	7.60±0.93 ^{a}	7.43±0.63 ^{a}	6.57±0.86 ^{b}	6.50±0.86 ^{b}
Overall preference	8.07±0.87 ^{a}	7.53±0.82 ^{b}	7.63±0.67 ^{b}	7.03±1.10 ^{c}

Note: Means ± standard deviation with the different superscripts in the same rows are significantly different ($p \leq 0.05$) using Duncan's test.

In order to examine the appropriate concentration of the WBMS's brix (sugar concentration) and the additional citric acid on the gummy sensory attributes, two level of total soluble solid (23 and 33 °Brix) and citric acid content (0.5% and 1%) were planned by the 2x2 factorial experiments in RCBD into four treatments including sample T1 (23 Brix, 0.5% citric), sample T2 (23 Brix, 1% citric), sample T3 (33 Brix, 0.5% citric) and sample T4 (33 Brix, 1% citric).

The results in **Table 5** found that all gummy recipes were not statistical significant in the appearance and gumminess at the 95 confidence level. However, gummy T1 and T4 were significantly different from gummy T2 and T3 in the overall preference attribute while gummy recipe T2 and T3 were not significantly different from each other in the overall preference attribute.

The difference of sensory preference on each gummy recipe involving two factors; brix and citric acid content. Result from the investigation of the relationships and interactions between brix and citric acid content on the sensory attributes shown in **Table 6**.

Table 6: The tests of between-subjects effects of gummy

Attributes	Interaction significant (Sig.)		
	Brix	Citric	Brix*Citric
Yellow color	0.40 ns	0.00*	0.72 ns
Appearance	0.65 ns	0.82 ns	1.00 ns
Pickled mango flavor	0.00*	0.81 ns	0.90 ns
Gumminess	0.69 ns	0.03*	0.89 ns
Sweetness	0.00*	0.07 ns	0.89 ns
Sourness	0.24 ns	0.00*	0.06*
Saltiness	0.00*	0.44 ns	0.74 ns
Overall preference	0.00*	0.00*	0.84 ns

The results in **table 6** revealed that there was a significant interaction between brix and citric acid content on panelist preference on sourness. However, there was no correlation between brix and citric on yellow color, appearance, pickled mango flavor, gumminess, sweetness, saltiness and overall preference. The level of both WBMS's brix and citric acid added in gummy had no effect on the appearance of the gummy. Both factors significantly the main factors that affected on overall preference of the gummy. The WBMS's brix was the main factor that affected on pickled mango flavor, sweetness and saltiness of the gummy. This result was supported by Zhang, and Barringer (2018) who found that sugar content has a large effect on flavor release. The volatile compounds significantly decreased in concentration when sugar was increased. Accordingly, the gummy with 23°brix WBMS might had strong pickled mango flavor than the with 33°brix WBMS and resulted in higher flavor preference score of gummy recipe A and B in **Table 5**. While the panelist's preference on yellow color, gumminess and sourness of the gummy was depended on citric acid. This is corresponding with the result reveled by Ge et.al. (2021) that citric acid weakened the gummy texture due to their restrictive effect on the formation of hydrogen bonds on gelatin gummy network.

Gummy T1, which contained 23° brix of WBMS and 0.5% of additional citric acid, had the significantly highest liking score on overall preference, and sourness from panelist ($p \leq 0.05$). It also had the highest liking score on yellow color, pickled mango flavor, sweetness and saltiness but not significantly different from gummy T2 which contained 33° brix of WBMS and 1% of additional citric acid. It grained very much like on overall preference, gumminess and sweetness. However, the panelist gave it moderately like on yellow color, appearance, pickled mango flavor, sourness and saltiness.

Table 7: Just-about-right (JAR) consideration of gummy formulas

Recipe	**Attributes**	**Intensity (%)**			**Net Score**	**Consideration**
		Too low	**JAR**	**Too high**		
T1	Yellow color	33.3	56.7	10.0	-23.3	Increase
	Pickled mango flavor	13.3	70.0	16.7	3.3	JAR
	Gumminess	10.0	86.7	3.3	-6.7	JAR
	Sweetness	13.3	86.7	3.3	-10.0	JAR
	Sourness	10.0	80.0	10.0	0.0	JAR
	Saltiness	3.3	90.0	6.7	3.3	JAR
T2	Yellow color	20.0	70.0	10.0	-10.0	JAR
	Pickled mango flavor	16.7	56.7	26.7	10.0	JAR
	Gumminess	16.7	76.7	6.7	-10.0	JAR
	Sweetness	23.3	50.0	26.7	3.3	JAR
	Sourness	10.0	43.3	46.7	36.7	Decrease
	Saltiness	16.7	73.3	10.0	-6.7	JAR
T3	Yellow color	23.3	76.7	0.0	-23.3	Increase
	Pickled mango flavor	10.0	50.0	40.0	30.0	Decrease
	Gumminess	6.7	90.0	3.3	-3.3	JAR
	Sweetness	10.0	46.7	43.3	33.3	Decrease
	Sourness	26.7	46.7	26.7	0.0	JAR
	Saltiness	10.0	66.7	23.3	13.3	JAR
T4	Yellow color	16.7	76.7	3.3	-13.3	JAR
	Pickled mango flavor	13.3	40.0	46.7	33.3	Decrease
	Gumminess	16.7	73.3	10.0	-6.7	JAR
	Sweetness	16.7	40.0	43.3	26.7	Decrease
	Sourness	20.0	30.0	50.0	30.0	Decrease
	Saltiness	23.3	43.3	33.3	10.0	JAR

The Just-About-Right scales (JAR) in **table 7** was applied in the questionnaire to evaluate the optimal sensory attributes of gummy recipe including yellow color, pickled mango flavor, gumminess sweetness, sourness and saltiness that have to be improved. The gummy attributes with percentage of intensity more than the cut-off point (70% of JAR) were accepted by consumer and were not needed to be improved. The improving of brownie attributes with percentage of intensity less than 70% were decided by the net score value. The attribute with net score of ±20 was considered JAR which mean the attribute was accepted and not need to be improved. The attribute intensity should be improved by decreasing the intensity of the attribute if the net score was less than -20 while increasing if the net score was higher than 20.

For gummy T1, the results of consideration showed that the intensity of yellow color was too low and should be improved while other attributes were accepted by the panelists. Accordingly, 0.01% of yellow color (Food grade) was added to enhance yellow color of the gummy.

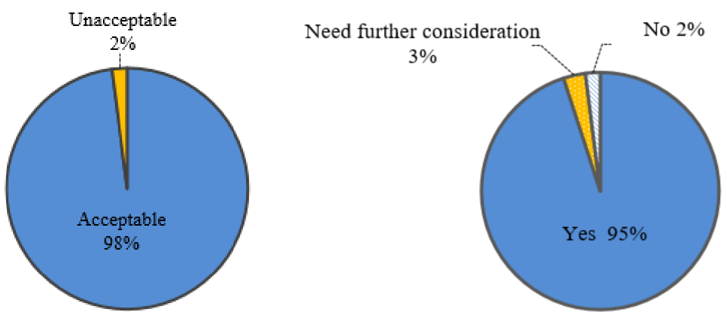
3. Consumer Acceptance Test

The sensory evaluation of the developed gummy recipe B, which contained 8% gelatin, 31% sugar, 30% glucose syrup, 0.5% citric acid, 30% WBMS (23°Brix) and 0.01% of yellow color, was represented in **Table 8**. The results showed that the majority of the panelists very liked the gummy color, the gumminess, the sweetness and overall preference while they were moderately like the gummy flavor, sourness and saltiness.

Table 8: Hedonic score of the developed gummy from waste sweet pickled Bao Mango syrup

Attributes	Hedonic score ± Standard deviation
Color	8.02 ± 0.68
Flavor	7.86 ± 0.48
Gumminess	8.34 ± 0.48
Sweetness	8.24 ± 0.49
Sourness	7.87 ± 0.37
Saltiness	7.76 ± 0.58
Overall preference	8.56 ± 0.40

For the consumer acceptance test, the majority of panelists (98%) accepted this product. About two percent denied this product because of the pickled mango flavor. The majority of panelists (95%) decided to purchase the developed product at 35 THB per 70 g (7 pieces) while three percent of them need more information of the gummy nutrition. Two percent of panelists considered not purchasing this product because of the g pickled mango flavor (**Picture 3**).



(a) (b)

Picture 3: The acceptance (a) and the buying decision (b) of the developed gummy from waste sweet pickled Bao Mango syrup

Conclusion

In conclusion, the developed gummy from waste sweet pickled Bao Mango syrup recipes contained 8% gelatin, 31% sugar, 30% glucose syrup, 0.5% citric acid, 30% WBMS (23^{o}Brix) and 0.01% of yellow color. The WBMS's brix was the main factor that affected on pickled mango flavor, sweetness and saltiness of the gummy while the yellow color, gumminess and sourness of the gummy was depended on citric acid. The sensory evaluation revealed that the majority of the panelists very liked the gummy color (8.02 ± 0.68), the gumminess (8.34 ± 0.48), the sweetness (8.24 ± 0.49) and overall preference (8.56 ± 0.40) while they were moderately like the gummy flavor (7.86 ± 0.48), sourness (7.87 ± 0.37) and saltiness (7.76 ± 0.58). The majority of panelists (98%) accepted this product and 95% of them decided to purchase the developed gummy at 35 THB per 70 g (7 pieces).

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