

**Development of functional beverage from Sapodilla (*Manilkara Zapota L.*) fruit**

Lim, W.S., \*Rabeta, M.S. and Uthumporn, U.

*Food Technology Division, School of Industrial Technology, Universiti Sains Malaysia, 11800 Minden, Penang, Malaysia***Article history:**

Received: 14 September 2017

Received in revised form: 8 November 2017

Accepted: 10 November 2017

Available Online: 12 November 2017

**Keywords:**Sapodilla juice,  
Total phenolic content,  
Ascorbic acid content,  
Total titratable acidity,  
Microbial stability,  
Sensory analysis**DOI:**[https://doi.org/10.26656/fr.2017.2\(2\).227](https://doi.org/10.26656/fr.2017.2(2).227)**Abstract**

This study started with the development of juice from sapodilla (*Manilkara zapota L.*) fruit. Among three formulations, sapodilla juice with the combination of 50% pure sapodilla juice, 25°Brix, and 0.40% of titratable acidity have gained the highest score in the hedonic sensory test, with overall acceptability ranging from “like slightly” to “like moderately”. Formulated sapodilla juice and pure sapodilla juice were analysed for their total phenolic and ascorbic acid contents, pH, total soluble solid and titratable acidity. The formulated sapodilla juice has lower pH (3.35), higher titratable acidity content (0.40%) and total soluble solid (25°Brix) than pure sapodilla juice. The total phenolic (469.82 mg GAE/L) and ascorbic acid contents (3.60 mg/100 mL) of formulated sapodilla juice which consists only 50% of sapodilla juice showed the lower value than the pure sapodilla juice. Formulated sapodilla juice with lower pH will be less susceptible to enzymatic browning. In microbiology total plate count, no colony formed on the formulated juice, whereas the mean number of colony forming units (CFU) in pure juice was 169318.18 CFU/ml juice stored in room temperature (28°C) for a week. These results revealed that the formulated juice had better microbial stability than pure juice.

**1. Introduction**

In our previous study, we have successfully developed a low calorie fruit bar from sapodilla (*Manilkara Zapota L.*) fruit (Rabeta *et al.*, 2016). In this study, sapodilla was selected to be used for the development of a beverage. Sapodilla is also commonly known as “sapota” in India and “chiku” in Malaysia. According to the Malaysian Food Pyramid, individuals are encouraged to consume at least 2 servings of fruit per day (Ministry of Health Malaysia, 2013). Studies have also revealed that a high consumption of fruits and vegetables is related to a lower incidence of degenerative diseases (Hossain *et al.*, 2012).

It has also been found that these health benefits can be obtained through the consumption of a beverage derived from fruits or vegetables containing a variety of bioactive compounds (Percival 2011). Thus, fruit juices have become an important part of the modern diet (Hossain *et al.*, 2012). Consumer demand for healthy drinks increases not only the popularity of nutrient-rich beverages but also people’s willingness to experience new and exotic flavours (Martinez *et al.*, 2012).

Additionally, phenol has good antioxidant, anti-

mutagenic, and anti-cancer properties, and directly influences radical-scavenging potential (Zhang and Humauzu 2004; Shanmugapriya *et al.*, 2011). Minerals can aid in the normal functioning of biological systems and can prevent deleterious effects such as decreased cellular immunity and behavioural and cognitive alterations (Kwong *et al.*, 2004; Kulkarni *et al.*, 2007). Vitamin C is one of the antioxidant nutrients involved in strengthening the body’s defence against potential damage from free radicals (Wilson *et al.*, 2011).

One advantage of processing is that it increases the shelf life of fruits. Sapodilla has a short shelf life due to its high moisture content (69.05 to 75.7%) and total soluble solids (TSS) content (17.4 to 23.7°Brix) (Morton, 1987). The fruits are also easily bruised, which leads to waste (Ganjyal *et al.*, 2004). To prolong shelf life, sapodilla can be preserved using sugar and citric acid to make products such as jam, jellies or juices (Ahmed *et al.*, 2011). The demand for high quality fruit juice has obviously increased in size in recent years because of consumer attention to outstanding quality (Unluturk and Atilgan, 2015).

Sapodilla, *Manilkara zapota L.*, is a fast growing fruit-bearing tropical evergreen tree that belongs to the

\*Corresponding author.

Email: [rabeta@usm.my](mailto:rabeta@usm.my)

family *Sapotaceae* (genus *Manilkara*) (Singh *et al.*, 2011) and is typically cultivated for its fruits (Ahmed *et al.*, 2011). It can be found throughout the tropical regions around the world. Sapodilla originated in Central and South America, where the largest population of native trees still exists from the Yucatan Peninsula of Mexico to Costa Rica (Ganjyal *et al.*, 2004).

Although sapodilla is a popular fruit in Malaysia, it remains relatively understudied due to its limited commercialisation and production (Kementerian Pertanian, 1981; Balerdi *et al.*, 2005). From 1976 to 1981, the amount of land planted with “chiku” increased from 443 to 863 hectares (Kementerian Pertanian, 1981). Additionally, information regarding changes in physical and chemical characteristics of the fruit during postharvest storage is limited, though there are reports on some aspects of postharvest treatment (Balerdi *et al.*, 2005). The main objective of this study was to develop a beverage from sapodilla fruit with some nutritional benefits and acceptable by consumers due to its sensory attributes.

## 2. Materials and methods

### 2.1 Raw materials

Sapodilla fruits were freshly harvested in October 2013 from Kuala Kurau, Perak Darul Ridzuan. Sapodilla (*Manilkara zapota*) was identified by Dr. Rahmad Zakaria, botanist of the Herbarium Unit, School of Biological Sciences, Universiti Sains Malaysia. The voucher number of sapodilla is USM. Herbarium 11475. Healthy ripe sapodilla without any physical damage was used to produce fruit juice.

### 2.2 Sample preparation

A 1 kg of ripe sapodilla fruits were washed with running water and their skin was peeled off manually using a knife without damaging the pulp and then blended in a blender (Panasonic MX-7995, Malaysia). Both the glass bottle and cap were sterilized in boiling water for 5 min and then drained and dripped dry. Fruit juice was extracted using a muslin cloth, and the weight of the extracted juice was recorded. An amount of water was weighed to be the same as the weight of extracted juice (1:1 ratio) and added to the extracted juice. The initial soluble solid content in degrees Brix (B1) was measured using a handheld refractometer, and the total weight of the diluted juice (W1) in grams was recorded. The initial titratable acidity of the diluted juice was determined. Each sample was titrated with 0.1 N of sodium hydroxide (NaOH) to an endpoint pH of 8.2. The mixture was warmed to dissolve all the sugars, and the juice was pasteurized at 85°C for 3 min. Then, the caps were closed and filled while hot. Lastly, the juice was blast frozen (-18°C) prior to freeze drying and grinding

into powder.

### 2.3 Sensory evaluation of Sapodilla fruit juice

A seven-point hedonic scale (1=dislike very much to 7=like very much) sensory evaluation on sapodilla fruit juice was performed. The sensory evaluation was conducted by 32 semi-trained panelists, consisting of the undergraduates and postgraduates of Food Technology. Sapodilla fruit juice samples with 3 different formulations (A, B and C) were prepared freshly and presented to the panelists with a three-digit numerical identifier and in random order. The samples were all served in a room with booths and normal lighting. Panelists were instructed to rinse their mouths before tasting each of the samples. Panelists evaluated the samples based on the attributes which were colour, aroma, taste and overall acceptability.

### 2.4 pH

The pH value of the sapodilla juice was measured using a pH meter (Mettler Toledo/320k). The pH meter is calibrated using the buffer solution before collecting the pH reading of the sample tested.

### 2.5 Titratable acidity content

An amount of sapodilla juice sample was measured and titrated with 0.1N NaOH to an end point of pH 8.2. The tittered value was recorded. Three readings were taken for each sample and the percentage of acid on the basis of citric acid was calculated according to the following equation:

$$\text{Percentage of acid (\%)} = \frac{\text{Volume of NaOH used} \times 0.1\text{N NaOH} \times \text{milliequivalent factor} \times 100}{\text{grams of sample}}$$

### 2.6 Total soluble solid content

Total soluble solid of the juice was determined using an Atago hand refractometer (ATAGO, Japan). It is done by placing the homogenized samples onto the prism surface of a hand refractometer. By looking through the eyepiece, the reading at the boundary line where the blue and white colors meet was taken. The results obtained were expressed in °Brix.

### 2.7 Total phenolic content

Total phenolic content was determined with the modified method of (Taga *et al.*, 1984). A 100 µL (10<sup>-6</sup>/micro) of sapodilla juice was mixed with 2 mL of 2% aqueous sodium carbonate solution. After 3 min, about 100 µL of 50% Folin-Ciocalteu's phenol reagent was added to the mixture. After 30 min of incubation at room temperature (26°C), absorbance was measured at 750 nm against a blank.

Total phenolic content was calculated on the basis of the calibration curve of gallic acid and expressed as

gallic acid equivalents. In this analysis, gallic acid was used as standard. Gallic acid with a series of concentrations (0, 100, 200, 300, 400 and 500 µg/mL) was prepared in order to construct a calibration curve of absorbance against concentrations. The equation obtained from calibration curve was used to determine the total phenolic content of sample tested.

### 2.8 Ascorbic acid content

The determination of ascorbic acid content using 2,6-dichloroindophenol (DCPIP) titrimetric method. For dye standardization, 5ml of ascorbic acid standard and 5 ml of extract phosphoric acid extract were added to a conical flask. The ascorbic acid standard solution was titrated with dye solution as quick as possible until pink color is formed. The pink color should be last for 15 seconds.

The dye factor is determined using the following formula,

$$\text{Dye Factor} = \frac{0.5}{\text{Titre (ml)}}$$

For sample extraction preparation, 10ml of juice sample is taken and diluted to 100ml with phosphoric acid extract solution. For titration part, an aliquot of 3 to 5 ml extracted sample are taken and titrated with standard dye solution. The sample is titrated quickly until pink color is formed and lasted for 15 seconds. The titer value is taken.

The concentration of ascorbic acid was calculated according to the following equation,

$$\text{Ascorbic Acid} = \frac{\text{Titre} \times \text{dye factor} \times \text{final volume} \times 100}{\text{Extract aliquot use for estimation} \times \text{volume of sample use for estimation}}$$

### 2.9 Microbiological total plate count

A 10g of fruit juice was added to 90 mL of 0.1 % buffered peptone water to achieve  $10^{-1}$  sample dilution. Serial dilutions of  $10^{-2}$  to  $10^{-6}$  were prepared by transferring 1mL of previous dilution to 9 mL of 0.1 % buffered peptone water. All dilutions were shaken for 25 times. Then, about 1 mL of each dilution was pipetted into appropriately marked petri dishes. The plate count agar was poured onto the petri dishes by using pour plate technique. Petri dish with agar was swirled clockwise and counter-clockwise gently to avoid spillage on dish lid. Agar was leaved to solidify and then incubated at 37 °C for 48 h. Duplicate plate count was performed.

The plates with 25-250 Countable Forming Unit (CFU) and spreader-free were counted and the total CFU was calculated and expressed as CFU/g sample. The calculation of CFU following the equation:

$$N = \frac{\Sigma c}{[(1 \times n_1) + (0.1 \times n_2)] \times d}$$

Where  $N$  = Number of colony forming units (CFU) per ml or g of product;  $c$  = Total colonies on all plates counted;  $n_1$  = Number of plates counted at the first dilution;  $n_2$  = Number of plates counted at the second dilution and  $d$  = Dilution of the first dilution.

### 2.10 Statistical analysis

The standard curves Gallic Acid Equivalence (GAE) in the determination of total phenolic content and radar spider chart for sensory evaluation were accomplished by using Microsoft Office Excel 2007. Statistical test of data was performed by using the SPSS Statistics 17.0 (SPSS Inc., Chicago, IL, USA). Comparison of means with one-way ANOVA was carried out by Duncan's multiple-range test at a level of significance of 0.05. The differences between means at  $p < 0.05$  were considered as significant difference.

## 3. Results and discussion

### 3.1 Sensory evaluation of formulated Sapodilla juice

Results of sensory attributes are shown in Table 1. Appearance scores of three formulated sapodilla juice were  $4.97 \pm 1.15$  (A),  $5.81 \pm 1.09$  (B), and  $4.97 \pm 1.33$  (C). The sensory evaluation of the color preferences revealed that no significant different ( $p > 0.05$ ) between formulation A and C; but formulation B was significantly different ( $p < 0.05$ ) from the others two formulations. The color and appearance of food products, especially fruits and vegetables serve as the first impression are typically the primary indicators of perceived quality (Gambaro *et al.*, 2001; Lawless and Heymann, 2010). Hedonic test, which accesses consumers' acceptance or preference, is a discriminative analysis in which, the panelists are required to give a numbers scores to describe the extent of their like and dislike of the sensory characteristics of foods according to the scale provided in the questionnaires (Lawless and Heymann, 2010).

Table 1. Sensory attributes preference of formulated sapodilla juice with 7-points of hedonic scale

| Sensory Attributes | Formulation       |                   |                   |
|--------------------|-------------------|-------------------|-------------------|
|                    | A                 | B                 | C                 |
| Visual acceptance  | $4.97 \pm 1.15^a$ | $5.81 \pm 1.09^b$ | $4.97 \pm 1.33^a$ |
| Aroma              | $4.66 \pm 1.29^a$ | $5.25 \pm 1.02^a$ | $3.81 \pm 1.44^a$ |
| Taste              | $4.72 \pm 0.00^a$ | $5.66 \pm 0.90^b$ | $3.75 \pm 1.14^c$ |
| Overall acceptance | $4.66 \pm 1.29^a$ | $5.69 \pm 0.74^b$ | $3.81 \pm 0.74^c$ |

Values are mean  $\pm$  standard deviation ( $n = 32$ ). Values with different superscript letters in the same row are significantly different at  $p < 0.05$ .

Sensory evaluation scores on the taste of formulated sapodilla juices were formulation  $4.72 \pm 0.00$  (A),  $5.66 \pm$

0.90 (B), and  $3.75 \pm 1.14$  (C). These results revealed that the three formulations were significantly different ( $p < 0.05$ ) in terms of taste acceptability. Formulation B was the most preferred, followed by formulation A and C. Formulation B secured the highest score among the three samples. Sugar and organic acid were the major chemical compounds that influence consumer preference (Saliba-Colombani *et al.*, 2001). The sweetness-to-sour ratio ( $^{\circ}\text{Brix}/\text{acid}$ ) is often used to express the quality of the fruit juice (Kulkarni *et al.*, 2007). Thus, determining the right  $^{\circ}\text{Brix}$  values for the formulated juice is crucial. The sensory evaluation results showed that  $25^{\circ}\text{Brix}$  value was the suitable  $^{\circ}\text{Brix}$  value because of its highest panelists' acceptability score.

### 3.2 pH

In this experiment, the pH of the pure juice was 5.36 whereas that of the pH of the formulated juice was  $3.53 \pm 0.17$  (Table 2). Compared with the pure juice ( $\text{pH } 5.51 \pm 0.20$ ), the formulated juice was expected to be less susceptible to enzymatic browning because its pH value was  $< 4$ . The pH value (5.36) of sapodilla juice reported by (Kulkarni *et al.*, 2007) was lower than the pH value of pure sapodilla juice in this experiment.

Table 2. Physico-chemical and total phenolic content of pure juice and formulated juice

| Sample                           | Pure Juice          | Formulated Juice    |
|----------------------------------|---------------------|---------------------|
| pH                               | $5.51 \pm 0.20^a$   | $3.35 \pm 0.17^b$   |
| Titrateable acidity (%)          | $0.08 \pm 0.02^a$   | $0.40 \pm 0.00^b$   |
| Total soluble solid (Brix)       | $14.40 \pm 1.06^a$  | $25.00 \pm 0.20^b$  |
| Total phenolic content (mg/L)    | $469.82 \pm 3.89^a$ | $222.45 \pm 4.24^b$ |
| Ascorbic acid content (mg/100mL) | $5.52 \pm 0.23^a$   | $3.60 \pm 0.24^b$   |

Values are mean ( $n=3$ )  $\pm$  SD. Values with different superscript are significantly different at  $p < 0.05$

Polyphenol oxidase (PPO) causes the enzymatic browning of fruits and vegetables (Cortez *et al.*, 2013). PPO is less reactive below the pH range of 5 and 7, which is the optimum pH range for PPO activity (Martinez and Whitaker, 1995). Hence, sapodilla is susceptible to enzymatic browning.

To order reduce the occurrence of enzymatic browning in the sapodilla juice, food acid was added to adjust the pH value (Almeida and Nogueira, 1995). Our major concern is to prevent microbial growth. The formulated juice will have lower pH than the pure juice because citric acid was added. The pH value of the juice will be lower with the increase in the amount of organic acid present (Tasnim *et al.*, 2010).

### 3.3 Titrateable acidity

In this experiment, the titrateable acidity of the pure juice was  $0.08 \pm 0.02\%$ , whereas that of the formulated

juice was  $0.40 \pm 0.00\%$  (Table 2). The titrateable acidity of the formulated juice was higher than of the pure juice because of the citric acid. Tasnim *et al.* (2010) explained that total acidity of the juice will be influenced by the presence of a mixture of organic acid. The titrateable acidity was calculated on the basis of citric acid because citric acid is the major organic acids in the sapodilla fruit (Lee *et al.*, 2013). The titrateable acidity of the sapodilla fruit was found to peak during its mature green stage (5.89%) and the lowest value was found during the over-ripe stage (4.95%) (Brito and Narain 2002). Kulkarni *et al.* (2007) reported that sapodilla juice contained 0.16% of titrateable acidity, which was calculated on the basis of citric acid.

Titrateable acidity measures the total acid concentration in food where and is determined by neutralizing the acid present in a pre-weight sample using a standard base (Boulton, 1980; Mitcham *et al.*, 1996). Organic acids could originate from biochemical processes or microbiological activity and are found to be widely distributed in the fruits (Lee *et al.*, 2013). The titrateable acidity of the sapodilla differs throughout different stages of maturity, acidity was observed to decrease with an increase in ripeness (Pawar, 2011). According to Abdul-Karim *et al.* (1987), the titrateable acidity of the sapodilla fruit reaches its maximum level at 3.9 months, and then a gradual decrease in the titrateable acidity could be observed thereafter.

### 3.4 Total soluble solid content

In this experiment, the total soluble solid content of the pure sapodilla juice was  $14.40 \pm 1.06^{\circ}\text{Brix}$  whereas for the formulated juice was  $25.00 \pm 0.20^{\circ}\text{Brix}$  (Table 2). Total soluble solid is expressed in  $^{\circ}\text{Brix}$  value, which represents the percent weight of sucrose in sucrose solution (Redd *et al.*, 1986). The place of growth and the species of sapodilla influence the total soluble solid content in sapodilla, which varies at different stages of maturity (Abdul-Karim *et al.*, 1987). Total soluble solid content decrease with age, where the highest was found during the half-ripe stage, and the lowest was found during the fully ripen stage (Pawar, 2011). The  $^{\circ}\text{Brix}$  value of the sapodilla fruit could reach as high as 21.40 during the half-ripe stage and as low as 15.80 during the fully ripened stage (Brito and Narain, 2002).

### 3.5 Ascorbic acid content

In this experiment, the ascorbic acid content of the pure sapodilla juice was  $5.52 \pm 0.23$  mg/100 mL, whereas the formulated juice, which contained 50% of sapodilla juice had  $3.60 \pm 0.24$  mg ascorbic acid/100 mL (Table 2). Ascorbic acid content in the juice is determined through 2, 6 dichloroindophenol titrimetric method, in which the result is recorded when the sample solution changes from colorless to pink (Nielsen, 2014).

The different cultivars of sapodilla will influence the quality parameters of sapodilla (Suhasini *et al.*, 2013). For instance, the Mexican and Indian varieties of sapodilla have different ascorbic acid contents (Lakshminarayana and Moreno-Rivera, 1979). The ascorbic acid content of Mexican sapodilla (8.9mg/100 g to 41.4 mg/100 g) was higher than that of Indian sapodilla (0.1 mg/100 g to 11.9 mg/100 g) (Lakshminarayana and Moreno-Rivera, 1979). Additionally, varying ascorbic acid contents were found in sapodilla fruits at different ripening stages (Pawar, 2011). Sapodilla fruits with different the lowest degree of maturity had the higher ascorbic acid content (Brito and Narain, 2002).

Over-ripe sapodilla fruit is not a good source of ascorbic acid (Pawar, 2011). Although ascorbic acid increases with ripening, the acid decreases during the over-ripe stage (Broughton and Wong, 1979). The ascorbic acid content of sapodilla fruit can reach as high as 21.7 at its mature green stages but exist in trace amount at its over ripen stage (Brito and Narain, 2002). The ascorbic acid content of ripe sapodilla fruit can be as low as 0.06 mg per 100 g of edible portion (Ganjyal *et al.*, 2003). Thus, the ascorbic acid content in the fruit decreases throughout the ripening process (Pawar, 2011).

### 3.6 Total phenolic content

In this experiment, the mean value of the total phenolic content of pure juice was  $469.82 \pm 3.89$  mg GAE/L, whereas that in the formulated juice, which consists of 50 % of pure sapodilla juice, was found to be  $222.45 \pm 4.24$  mg GAE/L (Table 2). Lee *et al.* (2013) showed that the total phenolic content of sapodilla is 166.47 mg GAE/kg, which was lower than those of the pure sapodilla juice and formulated sapodilla juice tested in this study.

The total phenolic content of the samples used in this study was lower than the total phenolic content of tropical fruits, such as custard apple, cempedak and Longkong reported by (Lee *et al.*, 2013). The total phenolic content of custard apple (*Annona reticulate*), cempedak (*Artocarpus integer*) and Longkong (*Aglaia dookkoo*) are 712.20 mg GAE/kg, 384.46mg GAE/kg and 589.08 mg GAE/kg, respectively (Lee *et al.*, 2013). The total phenolic content of the pure sapodilla juice and formulated sapodilla juice in the study were also markedly lower than that total phenolic content of sapodilla juice (1346 mg GAE / kg) as reported by (Kulkarni *et al.*, 2007).

The polyphenol of unripe sapodilla fruit had been reported as catechin, gallic acid, epicatechin, chlorogenic acid, 4-hydroxycatechin and gallic acid which contribute to the astringency of sapodilla (Lakshminarayana *et al.*, 1969; Ma *et al.*, 2003; Shui *et*

*al.*, 2004). The highest polyphenol concentration was found during the early stage of maturity, as characterized by astringent, which is the predominant taste at this stage (Lakshminarayana *et al.*, 1969). The polyphenol concentration of the fruit decreases from the unripe stage to the overripe stage (Shui *et al.*, 2004). Sapodilla develops sweetness in taste as the content of polyphenol decreases with developing maturity in fruit size and non-polyphenolic constituent content (Lakshminarayana *et al.*, 1969).

In the present, a standard curve of gallic acid in a series of concentrations against the absorbance was initially plotted to express the absorbance, which is recorded as milligram gallic acid equivalent (GAE) / L of juice (Georg *et al.*, 2005). Gallic acid is one of the standards used in expressing a number of phenolic compounds of the extract or sample derived from fruits and vegetables (Kulkarni *et al.*, 2007).

Different extraction methods and analyses, as well as the complex nature of the compound, could lead to a large variation in the total and free phenolic acid concentration detected in fruits of the similar and different species (Tomás-Barberán and Espín, 2001). Additionally, variation in the total phenolic content of fruits of the same species in different studies may have also been influenced by the degree of maturity of the fruits (Lozano, 2006).

For instances, proanthocyanidins are believed to be the compounds that contribute to the high antioxidant activity in sapodilla. The astringent property of unripe sapodilla is due to the high proanthocyanidins content, which decreases as the fruit reaches maturity (Wang *et al.*, 2012). In addition, the grown environment, handling method during storage, species, and cultivar also influence the total phenolic content of fruits (Tomás-Barberán and Espín, 2001).

### 3.7 Microbiological total plate count

In this experiment, no colony formed on the formulated juice whereas the mean number of colony forming units (CFU) in pure juice was  $1.6 \times 10^5 \pm 8035.30$  CFU/ml juice stored in room temperature (28° C) for a week. These results show that the formulated juice had better microbial stability than the pure juice. No population growth was found on the agar plate of formulated juice, which could be attributed to its pH of 3.35. Foods with a pH lower than 4 are known as high-acid food and considered to be less susceptible to most microbial and bacterial growth than other food (Ray, 2004).

The microbial spoilage of fruits and fruit products are normally caused by mould, yeast and aciduric bacteria as fruits had pH < 4.5 (Dilbaghi and Sharma, 2007). Organic acid is an antimicrobial agent that can be

used to control the growth of microorganisms in foods by increasing the acidity of food (Raybaudi-Massilia et al., 2009).

Microorganisms react differently toward pH that allows their growth (Ray, 2004). Generally, most bacteria prefer to grow at a pH close to neutrality (pH 6.5 to 7.5); for yeast pH 4.0 to 6.5 and mold grow at a wider range with a pH range of 4.0 to 6.8 (Dilbaghi and Sharma, 2007). When the pH drops below 5, microbial growth will be slow and some of the bacteria will even die and injured (Marriott, 1997; Ray, 2004).

Total plate count is a generic test that indicates the number of aerobic and mesophilic microbes that exist in the food; however, the test does not give any information on the types of microorganism that grew (Marriott, 1997; Morton, 2001). Hence, the colonies grow on the agar plate might be different from each other (Marriott, 1997). It is able to give some information regarding the raw material, processing and storage conditions, thus it is able to provide information about the shelf life stability of the food as well as access contamination, sanitary quality, food safety and organoleptic acceptability (Morton, 2001).

#### 4. Conclusion

Sapodilla fruit had the potential to be developed into a fruit juice product. By developing into a fruit juice product can help in prolonging the shelf life of the sapodilla fruit and increased its availability at the marketplace.

#### Conflict of Interest

We declare that we have no conflict of interest.

#### Acknowledgments

Short term grant (304/PTEKIND/6312124) and RU grant of Universiti Sains Malaysia (1001/PTEKIND/812176) contributed to the funding of this research.

#### References

- Abdul-Karim, M.N., Tarmizi, S.A. and Bakar, A.A. (1987). The physico-chemical changes in ciku (*Achras sapota* L.) of Jantung variety [in Bahasa Malaysia]. *Pertanika*, 10(3), 277-282
- Ahmed, T., Burhannuddin, M., Haque, M.A. and Hossain, M.A. (2001). Preparation of Jam of Sapota (*Achras zapota*). *The Agriculturist*, 9(1 and 2), 1-7
- Almeida, M.E.M. and Nogueira, J.N. (1995). The control of polyphenol oxidase activity in fruits and vegetables. *Plant Foods for Human Nutrition* (Formerly *Qualitas Plantarum*), 47(3), 245–256. <https://doi.org/10.1007/BF01088333>
- Balerdi, C.F., Crane, J.H. and Maguire, I. (2008). Sapodilla Growing in the Florida Home Landscape. Horticultural Sciences Department Document HS1. Florida Cooperative Extension Service, Institute of Food and Agricultural Sciences, University of Florida. Retrieved from: <http://edis.ifas.ufl.edu/pdffiles/MG/MG05700.pdf>
- Boulton, R. (1980). The general relationship between potassium, sodium and pH in grape juice and wine. *American Journal of Enology and Viticulture*, 31(2), 182–186.
- Brito, E.S.D. and Narain, N. (2002). Physical and chemical characteristics of sapota fruit at different stages of maturation. *Pesquisa Agropecuária Brasileira*, 37, 567-572. <https://doi.org/10.1590/S0100-204X2002000400020>
- Broughton, W.J. and Wong, H.C. (1979). Storage conditions and ripening of chiku fruits *Achras sapota* L. *Scientia Horticulturae*, 10(4), 377–385. [https://doi.org/10.1016/0304-4238\(79\)90098-0](https://doi.org/10.1016/0304-4238(79)90098-0)
- Cortez, J.T., Méndez, C.H.H., Duch, E.S., Vargas, M.D.L.V.Y. and Pereira, S.S. (2013). Purification and Partial Characterization of Polyphenol Oxidase from Sapodilla Plum (*Achras sapota*). *Food and Nutrition Sciences*, 4(7), 727 - 734. <https://doi.org/10.4236/fns.2013.47093>
- Dilbaghi, N. and Sharma, S. (2007). Food and industrial microbiology. Food Spoilage, Food Infections and Intoxications Caused by Microorganisms and Methods for Their Detection. Retrieved from: <http://nsdl.niscair.res.in/jspui/handle/123456789/386>
- Gambaro, A., Gimenez, A. and Burgueño, Juan. (2001). Sensory and instrumental evaluation of strawberry yogurt color. *Journal of Sensory Studies*, 16(1), 11-22. <https://doi.org/10.1111/j.1745-459X.2001.tb00287.x>
- Ganjyal, G.M., Hanna, M.A. and Devadattam, D.S.K. (2003). Processing of sapota (sapodilla): Drying. *Journal of Food Science*, 68(2), 517–520. <https://doi.org/10.1111/j.1365-2621.2003.tb05704.x>
- Ganjyal, G.M., Reddy, N., Yang, Y.Q. and Hanna, M.A. (2004). Biodegradable packaging foams of starch acetate blended with corn stalk fibers. *Journal of Applied Polymer Science*, 93(6), 2627–2633. <https://doi.org/10.1002/app.20843>
- Georg, S., Brat, P., Alter, P. and Amiot, M.J. (2005). Rapid Determination of Polyphenols and Vitamin C in Plant-Derived Products Rapid Determination of Polyphenols and Vitamin C in Plant-Derived Products. *Society*, 53(5), 1370–1373.
- Hossain, M.A., Rahman, M. and Shabuz, M.Z.R. (2012). Quality of Industrially Processed Fruit Juices: An

- Assessment Using Multivariate Framework. *Dhaka University Journal of Science*, 60(2), 169–173. <https://doi.org/10.3329/dujs.v60i2.11488>
- Kementerian Pertanian. (1981). Keluasan pelbagai tanaman di Semenanjung Malaysia. Kuala Lumpur: Government Printer.
- Kulkarni, A.P., Policegoudra, R.S. and Aradhya, S.M. (2007). Chemical composition and antioxidant activity of sapota (*Achras sapota* Linn.) fruit. *Journal of Food Biochemistry*, 31(3), 399–414. <https://doi.org/10.1111/j.1745-4514.2007.00122.x>
- Kwong, W.T., Friello, P. and Semba, R.D. (2004). Interactions between iron deficiency and lead poisoning: epidemiology and pathogenesis. *The Science of the Total Environment*, 330(1-3), 21-37. <https://doi.org/10.1016/j.scitotenv.2004.03.017>
- Lakshminarayana, S., Mathew, A.G. and Parpia, H.A.B. (1969). Changes in polyphenols of sapota fruit (*Achras zapota* L.) during maturation. *Journal of the Science of Food and Agriculture*, 20(11), 651–653. <https://doi.org/10.1002/jsfa.2740201105>
- Lakshminarayana, S. and Moreno-Rivera, M.A. (1979). Proximate characteristics and composition of sapodilla fruits grown in Mexico. *Annual Meeting of the Florida State Horticultural Society*, 92, 303-305
- Lawless, H.T. and Heymann, H. (2010). Sensory Evaluation of Food: Principles and Practices, 2<sup>nd</sup> ed., p. 8-9. New York: Springer. <https://doi.org/10.1007/978-1-4419-6488-5>
- Lee, P.-R., Tan, R.-M., Yu, B., Curran, P. and Liu, S.-Q. (2013). Sugars, organic acids, and phenolic acids of exotic seasonable tropical fruits. *Nutrition and Food Science*, 43(3), 267–276. <https://doi.org/10.1108/00346651311327927>
- Lozano, J.E. (2006). Fruit Manufacturing. New York: Springer, Boston. <https://doi.org/10.1007/978-0-387-30616-2>
- Ma, J., Luo, X.D., Protiva, P., Yang, H., Ma, C., Basile, M J. and Kennelly, E.J. (2003). Bioactive novel polyphenols from the fruit of *Manilkara zapota* (Sapodilla). *Journal of Natural Products*, 66(7), 983–986. <https://doi.org/10.1021/np020576x>
- Marriott, N. (1997). Essentials of Food Sanitation. New York: Chapman and Hall. <https://doi.org/10.1007/978-1-4615-6045-6>
- Martinez, M.V. and Whitaker, J.R. (1995). The biochemistry and control of enzymatic browning. *Trends in Food Science and Technology*, 6(6), 195–200. [https://doi.org/10.1016/S0924-2244\(00\)89054-8](https://doi.org/10.1016/S0924-2244(00)89054-8)
- Martinez, R., Torres, P., Meneses, M.A., Figueroa, J.G., Pérez-Álvarez, J.A. and Viuda-Martos, M. (2012). Chemical, technological and in vitro antioxidant properties of mango, guava, pineapple and passion fruit dietary fibre concentrate. *Food Chemistry*, 135 (3), 1520-1526. <https://doi.org/10.1016/j.foodchem.2012.05.057>
- Ministry of Health Malaysia (MOH). (2013). Retrieved from <http://www.my.health.gov.my>
- Mitcham, B., Cantwell, M. and Kader, A. (1996). Methods for determining quality of fresh commodities. *Perishables Handling Newsletter*, 85, 1–5.
- Morton, J.F. (1987). Fruits of warm climates. Retrieved from <http://www.cabdirect.org/abstracts/19876763104.html;jsessionid=61405C300416EC9A053AEFA02FB3A2A9>
- Morton, R. (2001). Aerobic plate count for the Microbiological Examination. Retrieved from [https://scholar.google.com/scholar?q=Compendium+of+Methods+for+The+Microbiological+Examination+of+Foods+Morton%2C+R.D.+2001+and+btnG=andhl=enandas\\_sdt=0%2C5#0](https://scholar.google.com/scholar?q=Compendium+of+Methods+for+The+Microbiological+Examination+of+Foods+Morton%2C+R.D.+2001+and+btnG=andhl=enandas_sdt=0%2C5#0)
- Nielsen, S. (2014). Food analysis. New York: Springer Science and Business Media.
- Pawar, C.D., Patil, A.A. and Joshi, G.D. (2011). Physico-chemical parameters of sapota fruits at different maturity stages\*. *Karnataka Journal of Agricultural Sciences*, 24(3).
- Percival, S.S. and Findley, B. (2011). What's in Your Tropical Fruit?. Retrieved from [http://edis.ifas.ufl.edu/document\\_fs144](http://edis.ifas.ufl.edu/document_fs144)
- Rabeta, M.S., Tee, L.Y. and Leila, M. (2016). Development of Fruit Bar Using Sapodilla (*Manilkara Zapota* L.). *Journal of Food Processing And Preservation*, 41(2), 1-7. <https://doi.org/10.1111/j.1541-4337.2009.00076.x>
- Ray, B. (2004). Fundamental Food. Retrieved from [http://books.google.com/books?hl=en&lr=&id=zYPFZby2wtcC&oi=fnd&pg=PA1&dq=Fundamental+Food+Microbiology&ots=qvHkiBdUCbandamp;sig=WtdhhPDq1al7P\\_S\\_jcef1xLo9t8](http://books.google.com/books?hl=en&lr=&id=zYPFZby2wtcC&oi=fnd&pg=PA1&dq=Fundamental+Food+Microbiology&ots=qvHkiBdUCbandamp;sig=WtdhhPDq1al7P_S_jcef1xLo9t8)
- Raybaudi-Massilia, R.M., Mosqueda-Melgar, J., Soliva-Fortuny, R. and Martín-Belloso, O. (2009). Control of pathogenic and spoilage microorganisms in fresh-cut fruits and fruit juices by traditional and alternative natural antimicrobials. *Comprehensive Reviews in Food Science and Food Safety*, 8(3), 157–180.
- Redd, J.B., Hendrix, C.M. and Hendrix, D.L. (1986). Quality Control Manual for Citrus Processing Plants: Regulation, Citrus Methodology, Microbiology, Conversion Charts, Tables, Other. United States: Intercit Inc.
- Saliba-Colombani, V., Causse, M., Langlois, D., Philouze, J. and Buret, M. (2001). Genetic analysis of organoleptic quality in fresh market tomato. 1.

- Mapping QTLs for physical and chemical traits. *Theoretical and Applied Genetics*, 102(2–3), 259–272. <https://doi.org/10.1007/s001220051643>
- Shanmugapriya, K., Saravana, P.S., Payal, H., Mohammed, S.P. and Binnie, W. (2011). Antioxidant Activity, Total Phenolic And Flavonoid Contents of Artocarpus Heterophyllus And Manilkara Zapota Seeds and Its Reduction Potential. *International Journal of Pharmacy and Pharmaceutical Science*, 3(Suppl. 5), 256-260.
- Shui, G., Wong, S.P. and Leong, L.P. (2004). Characterization of antioxidants and change of antioxidant levels during storage of Manilkara zapota L. *Journal of Agricultural and Food Chemistry*, 52(26), 7834–7841. <https://doi.org/10.1021/jf0488357>
- Singh, M. and Shivhare, Y. (2011). Manilkara Zapota: An Overview. *Herbal Tech Industry*, 8(6), 13-16.
- Suhasini, J., Kanamadi, V.C., Swamy, G.S.K., Shirol, A.M. and Chavan, M. (2013). Performance of sapota (*Achras sapota* L.) varieties and hybrids under Ghataprabha command area. *Karnataka Journal of Agricultural Sciences*, 25(4).
- Taga, M.S., Miller, E.E. and Pratt, D.E. (1984). Chia seeds as a source of natural lipid antioxidants. *Journal of the American Oil Chemists' Society*, 61 (5), 928–931. <https://doi.org/10.1007/BF02542169>
- Tasnim, F., Anwar Hossain, M., Kamal Hossain, M., Lopa, D. and Formuzul Haque, K.M. (2010). Quality assessment of industrially processed fruit juices available in Dhaka city, Bangladesh. *Malaysian Journal of Nutrition*, 16(3), 431–8.
- Tomás-Barberán, F.A. and Espín, J.C. (2001). Phenolic compounds and related enzymes as determinants of quality in fruits and vegetables. *Journal of the Science of Food and Agriculture*, 81(9), 853–876. <https://doi.org/10.1002/jsfa.885>
- Unluturk, S. and Atilgan, M.R.(2015). Microbial Safety and Shelf Life of UV-C Treated Freshly Squeezed White Grape Juice. *Journal of Food Science*, 80(8), M1831-M1841. <https://doi.org/10.1111/1750-3841.12952>
- Wang, H., Liu, T., Song, L. and Huang, D. (2012). Profiles and  $\alpha$ -amylase inhibition activity of proanthocyanidins in unripe Manilkara zapota (Chiku). *Journal of Agricultural and Food Chemistry*, 60(12), 3098–3104. <https://doi.org/10.1021/jf204715q>
- Zhang, D. and Hamauzu, Y. (2004). Phenolics, ascorbic acid, carotenoids and antioxidant activity of broccoli and their changes during conventional and microwave cooking. *Food Chemistry*, 88(4), 503-509. <https://doi.org/10.1016/j.foodchem.2004.01.065>