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Caricake: Development of vitamin A-rich cake sandwich using papaya (*Carica papaya*) leaves

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Abstract

Based on the Estimated Average Requirement, 58% to 81% of vitamin A deficiency is relatively prevalent among Filipino school-aged children. Even though papaya leaves are rich in vitamin A, they are considered agricultural waste in the Philippines. Papaya leaves were used to make a vitamin-A-rich cake sandwich. To test product acceptance, ground papaya leaves were added to the cake sandwich foundation in 5%, 6%, and 7% ratios to flour. Five professional food validators deemed one cake sandwich for a children's liking. A 5-point hedonic face scale was used to evaluate the cake sandwich by 53 6-9-year-olds. Descriptive statistics followed. Using the 5-point hedonic face scale, 53 Filipino children aged 6 to 9 evaluated CCake2 with an average score of 4.5 (Extremely Like) for its appearance, taste, aroma, texture, and overall acceptability. In HPLC nutrient testing, 35 g of CCake2 has 120 ug of vitamin A. Therefore, CCake2 is the best cake sandwich high in vitamin A for Filipino school-age children.

Keywords: Filipino Children; Vitamin A; Cake sandwich; Papaya leaves

1. Introduction

Vitamin A is a crucial nutrient for the proper functioning of the eyes and in the maintenance of cellular processes such as growth, epithelial integrity, generation of red blood cells, immunity, and reproduction [1]. Consequently, night blindness is one sign of deficiency in this vitamin; when it becomes severe, the cornea dries up, which damages the cornea and retina [2]. According to the Expanded National Nutrition Survey (ENNS) conducted by the Food and Nutrition Research Institute, 66.9% of the Filipino school-aged children at 6-9 years old have inadequate dietary intake of vitamin A [3]. Filipino school-aged children have a high prevalence of inadequate intakes of vitamin A at 58%-81% based on the Estimated Average Requirement (EAR) [4]. The clinical signs of vitamin A deficiency include xerophthalmia, night blindness, bitot's spots, conjunctival xerosis, and keratomalacia [5]. These clinical manifestations may be reversible or irreversible depending on the degree of deficiency [6].

The country's strategies to address vitamin A deficiency include vitamin A supplementation for young children, food fortification of food staples and processed foods, micronutrient powder supplementation, and dietary diversification [3]. In line with this, the ENNS also reported that rice and other cereal products are among the most commonly consumed food groups in school-aged children at 51.7% [3].

Even though these strategies were made to combat vitamin A deficiency in the country, it is still prevalent, particularly among the vulnerable population groups: young children, women of reproductive age, and the elderly. Additionally, school-aged children are among the population groups that had the highest percentage of inadequate dietary vitamin A intake. Since school-aged children frequently eat snacks, the researchers developed a vitamin A-rich cake sandwich that would aid in increasing the vitamin A levels of this particular age group. Then, the researchers determined the acceptability of the cake sandwich among school-aged Filipino children.

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In the Philippines, a Food Fortification law mandates the addition of nutrients to processed foods and food products in accordance with the Recommended Dietary Allowances (RDA) to prevent and minimize nutritional deficiency problems [7]. Fortification is the purposeful addition of micronutrients to a food in order to increase the nutritive quality of the food. In the market, only a few snacks have naturally high vitamin A content, as others are fortified with vitamin A. Thus, development of snacks naturally containing vitamin A is essential for children.

According to the Philippine Statistics Authority, papaya is an abundant fruit crop in the Philippines [8]. Other than the papaya fruit, the leaves and flowers can be eaten. However, according to a study by Alara et al. [9], papaya leaves are usually disregarded and considered as an agricultural waste even though they contain various health benefits to humans. In addition, papaya leaves, aside from being a food ingredient, are traditionally used as herbal medicine to treat different illnesses such as malaria, dengue, and jaundice, acting as an immunomodulator because of their antiviral properties [10]. Hence, papaya leaves can also be considered a functional food that contains compounds such as papain and chymopapain that can reduce the likelihood of infections and diseases.

Further, papaya leaves have been found to have a provitamin A beta carotene content of 303.55 mg per 100 g of leaves which is considered as a high source of vitamin A [11]. Additionally, the leaves contain ascorbic acid, flavonoids, cyanogenic glucosides, vitamin A, C and B vitamins and minerals [13]. According to the Food and Nutrition Research Institute (2019), young papaya leaves contain 16,130 of beta carotene and 1344 of Retinol Activity Equivalent (RAE). On the other hand, boiled young papaya leaves contain 10,180 µg beta carotene and 848 µg RAE, indicating that the leaves are abundant in Vitamin A. Thus, the study aimed to utilize papaya leaves in developing a cake sandwich rich in vitamin A.

Therefore, the researchers answered the following questions:

- How much papaya leaves should be added to the cake sandwich to ensure that it contains an adequate amount of vitamin A for Filipino children aged 6-9 years?
- What is the acceptability of the vitamin-A rich cake sandwich using papaya leaves among Filipino children aged 6-9 years?
- How many servings is required to meet at least 30% of Recommended Nutrient Intake (RNI) in Filipino children aged 6-9 years?

Generally, the study aimed to develop a vitamin-A rich cake sandwich using papaya leaves and determine its acceptability among Filipino children aged 6-9 years.

The product aimed to aid in providing the children with their recommended nutrient intake for vitamin A. The study is aligned with the research agenda of the Philippine Plan of Action for Nutrition (PPAN) 2019-2022, specifically on product and technology development. The study would benefit food product developers and food technologists, who may use this recipe for mass production. It would be beneficial for Filipinos and local bakers if they are encouraged to use economically and locally accessible plants such as papaya leaves for food innovation as they are high in vitamins and minerals. Likewise, local government units and non-government organizations might consider this study to produce cake sandwiches in nutrition programs for children. Lastly, the findings in this study would provide significant information for future researchers who would investigate papaya leaves' nutritional benefits and properties.

2. Material and methods

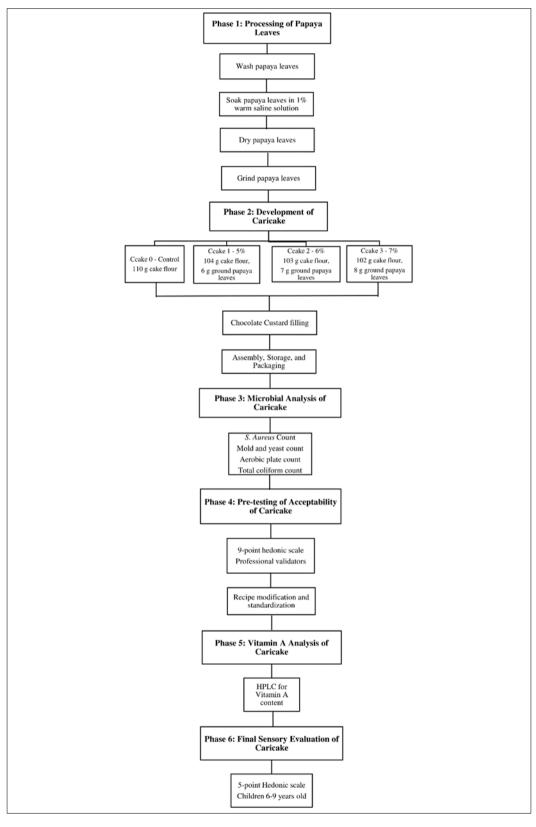


Figure 1 Development of CariCake

2.1. Study Design

The study utilized quantitative research in an experimental approach to determine the acceptability and nutrient content of CariCake. In this study, the 9-point hedonic rating scale was used for the professional validators and 5-point hedonic face scale for the target participants who are Filipino children aged 6-9 years old to evaluate the product's sensory characteristics and its acceptability. The product underwent microbiological analysis to ensure there are no pathogens present and it was also subjected to nutrient analysis to determine its vitamin A content.

2.2. Study Setting and Population

The production of the CariCake was held at a residential area in Barangay Santo Cristo, Quezon City. The microbiological testing was done at Qualibet Testing Services, and nutrient analysis of Vitamin A content was done at SGS Philippines, Inc. The product was evaluated in Potrero Elementary School by using a 5-point hedonic face scale for 50 children ages 6-9 years old with 10 children allotted as a replacement for those who withdrew from the sensory evaluation. A total of 53 children participated in the sensory evaluation of the product.

The study applied purposive sampling in the recruitment of participants. Purposive sampling is a nonprobability sampling in which the gathered participants are based on the own judgment of the researchers [14]. It is determined by the criteria set by the researchers based on the objective of the study. According to a study by Kwak and Kim, the confidence interval of the population data set is frequently increased by a sample size of 30 or more, which allows justification for assertions against the findings [15]. The sample is more likely to be representative of the population set if the sample size is larger. Thus, the study had 60 participants.

The eligibility requirements for the 60 children (sample size of 50 with 10 as replacement) are as follows:

Table 1 Inclusion and Exclusion Criteria for Recruitment of Participants

Inclusion Criteria	Exclusion Criteria
Male or female	Below or older than 6 to 9 years old
Children aged 6 to 9 years old	Not fully immunized
Fully immunized	Not enrolled in Potrero Elementary School
Enrolled in Potrero Elementary School	Has current health-related problems
Has no current health-related problems	Has food allergies in any of the following: papaya leaves,
Does not have food allergies in any of the following: papaya leaves, egg, sugar, lemon juice, cake flour, cornstarch, milk powder, salt, vegetable oil, cocoa powder, and whole milk.	egg, sugar, lemon juice, cake flour, cornstarch, milk powder, salt, vegetable oil, cocoa powder, and whole milk.

The recruitment process is done by allowing the advisers of their respective sections to select eligible participants. Class advisers chose the students based on the inclusion and exclusion criteria. Students who passed the inclusion criteria were selected and given an informed consent form (ICF) for the legally authorized representative (LAR). During the day of the sensory evaluation, signed informed consent form (ICF) was collected from the selected students. The students then proceeded to the classroom wherein the sensory evaluation was conducted.

2.3. Materials for Cake Sandwich

The ingredients stated in Table 2 were acquired from the local market. The primary raw ingredient, papaya leaves, was collected from the researchers' backyard in Brgy. Coloong 1, Valenzuela City, Philippines.

The equipment used for this food product development consists of an electric hand mixer, blender, wire whisks, flat sheet 12x16x1" cake pan, measuring spoons and measuring cups, food weighing scale, a convection oven for baking the cake, food processor for papaya leaves, and for storing of the CariCake, plastic wrappers, and opaque plastic containers were used and stored in the refrigerator.

Table 2 Ingredients for CariCake

Ingredients	HH Measure	EP Weight				
Cake Sandwich Base (10' x 15' flat sheet tray) Yield: 16 servings Serving Size: 30 g						
Egg Whites	8 egg whites	250 g				
White Sugar	80 g	80 g				
Lemon Juice	¼ tsp	1.25 g				
Cake Flour	110 g	110 g				
Cornstarch	7 g	7 g				
Milk Powder	7 g	7 g				
Salt	¹⁄8 tsp	0.625 g				
Vegetable Oil	75 ml	75 ml				
Whole Milk	105 ml	105 ml				
Chocolate Custard Filling Yiel	d: 60 servings Serving Size: 5	g				
Egg Yolks	4 egg yolks	72 g				
Whole Milk	1 ⅔ cups	416 ml				
Cocoa Powder	5 tbsp	75 g				
White Sugar	75 g	75 g				
Vanilla Extract	2 tsp	10 g				
Cornstarch	3 tbsp	45 g				
Butter	2 tbsp	30 g				

2.4. Preparation of CariCake

2.4.1. Formulation of CariCake

CariCake was prepared by gathering the required ingredients and equipment. A cake base and chocolate custard filling were prepared using a standard recipe in which the cake contains ground papaya leaves. The cake sandwich without the ground papaya leaves served as the control. Three formulations were made for the cake, which contained a 5%, 6%, and 7% ratio of ground papaya leaves to flour, as seen in Table 3. The method for formulating ground papaya leaves to flour ratios was adapted from the study of El-Gammal et al. on Moringa leaves powder [16]. Although there was a minimal difference in the amount of papaya leaves per formulation, the three formulations were carried out to determine the difference in the acceptability of CariCake.

Table 3 Recipe Formulation of CariCake

Yield: 16 servings Serving Size: 35 g						
Samples	Base: Cake Flour	Ground Papa	aya Leaves	Filling		
		Percentage	Grams			
CCake0	110 g	0%	0 g	Chocolate Custard		
CCake1	104.5 g ~ 104g	5%	5.5 g ~ 6g	Chocolate Custard		
CCake2	103.4 g ~ 103g	6%	6.6 g ~ 7g	Chocolate Custard		
CCake3	102.3 g ~ 102 g	7%	7.7 g ~ 8g	Chocolate Custard		

The vitamin A content of the papaya leaves was derived from the Papaya, young lvs, boiled in the Philippine Food Composition Tables. This item contains 848 ugRAE/100 g, which was multiplied by the formulation percentage [17]. The MenuEval Plus was utilized to determine the vitamin A content of each formulation per serving.

2.4.2. Processing of Papaya Leaves

Similar studies on the preparation of other leaves were used as references in the processing of papaya leaves. Papaya leaves were processed by washing and soaking in a 1% saline solution to remove any other foreign materials on the leaves. This method of processing was adapted from the study of Mishra et al., on Moringa leaves [18]. Further, in a study by Czarnowska-Kujawska et al., fresh spinach and kale were weighed, blended, and combined with the other key ingredients [19].

2.4.3. Step-by-step Procedures

Phase 1 - Processing of Papaya leaves

- Wash and dry papaya leaves.
- Remove the papaya leaves from the stem.
- Soak the papaya leaves in 1% warm saline solution for 90 seconds at 37 °C.
- Pat dry the papaya leaves.
- Once the papaya leaves have dried, ground into small bits using a food processor.

Phase 2 -Development of CariCake

- Preheat the conventional oven to 120 °C.
- Place the egg whites in a bowl and allow them to settle to room temperature.
- Add the milk and oil in a separate bowl and whisk.
- Sift the cake flour, cornstarch, milk powder, and salt. Add in ground papaya leaves and combine together with a whisk until the batter becomes thick yet flowy and smooth.
 - For the control, CCake0, add 110 grams cake flour.
 - For CCake1, add 6 grams of ground papaya leaves and 104 grams of cake flour.
 - For CCake2, add 7 grams of ground papaya leaves and 103 grams of cake flour.
 - For CCake3, add 8 grams of ground papaya leaves and 102 grams of cake flour
- Beat the egg whites until the foam forms, then add the lemon juice and beat again until fine air bubbles form.
- Add the sugar gradually, in 3 batches, while beating at a medium speed. Scrape the sides of the bowl from time to time and beat until stiff peaks form.
- Fold ¹/₃ of the egg whites into the thick batter using a spatula until they are combined.
- Pour the remaining batter onto the egg whites from the side of the bowl to prevent the egg whites from deflating. Using a spatula, cut down the middle and fold over gently. Repeat until combined well, and the batter is voluminous.
- Line one 12x16x1 inches flat cake sheet pan with parchment paper. Pour the batter into the pan and smoothen the surface with a spatula. Gently tap the pans on the counter a few times to remove air bubbles.
- Place the pans in the middle rack of the oven and bake for 25 minutes at 120 °C.
- Once the cake pans are out of the oven, repeatedly drop it from a height of about 10 cm to the countertop to avoid shrinking.
- Allow it to cool for 15 minutes before taking it out of the cake sheet pan.

Chocolate Custard Filling

- Boil milk and half of the sugar in a pot. Reduce the heat to medium.
- In a bowl, whisk the egg yolks. In a separate bowl, combine cornstarch and remaining sugar. Whisk together egg yolks and the sugar cornstarch mixture.
- Add 1 cup of warm milk and 5 tablespoons of cocoa powder to the egg yolk mixture, then add the mixture to the pot of warm milk.
- Stir until custard thickens and keep stirring for 3-5 minutes. Cook custard until it reaches a temperature of 185 °F.
- Turn off the heat, and add butter. Let the mixture cool, and add vanilla extract.
- Strain custard to remove any lumps.
- Transfer to a bowl and cover tightly using a plastic wrap.

Assembling the cake sandwich

- Once the cake is cooled down, gently remove the cake out of the pans and place on a flat surface.
- Cut the cake into small rectangles and spread the chocolate custard filling
- Cover the cake by putting the remaining cake on top to make a sandwich. Wrap in individual plastic and store in an opaque container in the refrigerator.

2.5. Tool Development

In this study, the 5-point hedonic face scale used in a study by Singh-Ackbarali & Maharaj was utilized and modified to determine the acceptability of CariCake in terms of appearance, aroma/smell, texture/consistency, taste, and overall acceptability among Filipino children aged 6-9 years old [20]. This criteria is according to the sensory attributes that are commonly used in evaluating food [21]. Faces of the hedonic scale were rearranged from most pleasant to least pleasant, as opposed to Singh-Ackbarali and Maharaj's facial scale [20]. In addition, the facial figures in the modified hedonic face scale were changed.

The 9-point hedonic rating scale used in the book, 'Understanding Food: Principles and Preparation', by Brown was adapted for sensory evaluation by professional validators [22]. Instead of using a tabular format, the validators' responses were entered using a line break. Then, based on the specified values of 1-9, a check mark was placed on each line break. In addition to this, a space for comments and suggestions was provided.

The letters written to Ms. Singh-Ackbarali and Dr. Amy Brown requesting permission to use and modify the hedonic rating scales were approved for use in the present study.

2.6. Nutrient Analysis

To determine the vitamin A content of the cake sandwich, a vitamin A analysis was performed using HPLC method. High Performance Liquid Chromatography (HPLC) is the method of choice when analyzing vitamin A in food samples [23]. Therefore, the cake sandwich selected by the five professional validators was subjected to HPLC testing at SGS Philippines, Inc., following the initial sensory evaluation.

2.7. Data Collection

After the development of CariCake, all cake samples underwent microbiological testing for *Staphylococcus aureus* (*S. aureus*), Mold and Yeast Count (MYC), Aerobic Plate Count (APC), and Total Coliforms Count (TCC) in Phase 3. In Phase 4, the CariCake samples were validated by 2 Registered Nutritionist-Dietitians, 1 Bakery Owner, and 2 Food Technologists, who were each given 3 samples of the product.

In phase 5, CCake2, the most acceptable product as evaluated by the validators, was subjected to vitamin A content analysis. Sensory evaluation was done by children aged 6-9 years using a 5-point hedonic face scale in phase 6. The data gathered from the evaluation was compiled using Google Sheets.

The sensory evaluation was conducted for two days, of which one day was allotted for the validators' evaluation, and once recipe modification was done based on their comments and suggestions, one day was allotted for data collection with the eligible children. The information sheet and informed consent form for the professional validators and the LAR were distributed prior to the sensory evaluation. The parents or guardians of eligible children had ample time to decide whether their children would participate in the study.

For the product and test site preparation for the children, product samples were prepared during the day of the evaluation. The sensory evaluation site for eligible children was conducted at a classroom in Potrero Elementary School in Malabon, Potrero. The researchers on-site ensured that any odors that could impair the evaluation were eliminated. As for lighting, the light bulb, windows, and doors were opened to achieve illumination as close to natural light as possible to prevent visual distortion of the product that could influence the outcomes of the evaluation. Each participant was given a folder, which included the sensory evaluation form and a pencil. Product samples were presented in polyethylene plastic sheets with bottled water as a palette cleanser. After the sensory evaluation, participants were given a gesture of appreciation.

Two batches of sensory evaluation were done with 30 participants per batch. A registered nurse and a school teacher were present during the sensory evaluation, yet the researchers ensured that the presence of authority did not affect their evaluation by locating them at a distance from the participants.

Before conducting the sensory evaluation, the briefing process was demonstrated through an audio-visual presentation. Included in the content of the briefing procedure was an introduction to the researchers and the study (objectives, safety, and confidentiality). Instructions and procedures on how the children will evaluate the product were also included in the audio-visual presentation. The verbal assent of the eligible children was acquired through voice recording.

Each participant was given one product sample and bottled water for the actual sensory evaluation. The participants were asked to follow the specific guidelines during the sensory evaluation:

- Look at the product and rate how it looks.
- Try to smell the product. Then, evaluate the aroma/smell.
- Take a small portion of the cake sandwich, and then chew the cake sandwich slowly and steadily until it becomes a fine paste. Evaluate the consistency.
- Drink some water.
- Take a small portion of the cake sandwich, and then chew the cake sandwich slowly and steadily until it becomes a fine paste. Swallow, then evaluate the taste.
- Drink some water.
- Repeat steps 1,2,3, 4 and 5 and consider its overall acceptability.
- Drink some water.
- Conclusion of sensory evaluation
- Ask the participants if they can still consume another slice of the cake sandwich to determine whether they can still consume one.

Prior to and during the actual sensory evaluation, the researchers assisted the participants in reading and understanding the sensory evaluation form. During the sensory evaluation, no external suggestions or remarks were made in order to prevent favorable responses from the participants that could potentially impact the validity of the evaluation.

2.8. Data Encoding, Editing and Analysis

2.8.1. Data Encoding

The data gathered from the nutrient content, chemical, and microbiological analysis were encoded using Google Sheets. The three formulations of the cake sandwich were coded as CCake1 for 5%, CCake2 for 6% and CCake3 for 7% of ground papaya leaves content in the flour. The pre-testing results from the validators were coded as V1, V2, V3, and so on for each of the trials. The three formulations of CariCake were placed in separate sheets. Control numbers were also given to the participants of the study, ranging from P1 to P60. The results of the nutrient analysis, microbiological analysis, and sensory evaluation were compiled in a tabular format.

2.9. Statistical Analysis

The sensory evaluation data gathered from children aged 6-9 years old were collected by the researchers. Descriptive statistics was used to evaluate the acceptability of the CariCake based on the sample's attributes in quantitative analysis. Measures of central tendency showed the participants' average score to discern the CariCake's acceptability.

2.10. Study Limitations

The study was limited to the development of CariCake, a cake sandwich rich in vitamin A. In addition, its shelf-life and packaging were not focused on. Considering that the cake sandwich is rich in vitamin A, the study was not able to evaluate its ability to increase vitamin A status in young children. The study focused on the acceptability of CariCake in terms of its appearance, aroma/smell, texture/consistency, taste, and overall acceptability among Filipino children aged 6-9 years old.

2.11. Ethical Considerations

Prior to the data collection, permission was initially obtained from the Department of Education School Division to gather data from Potrero Elementary School. Subsequently, permission from the school's principal was also acquired. Since the target participants were unable or have decreased ability to provide informed consent, the permission and the consent form of a legally authorized representative (LAR) was acquired. Food validators also received an informed

consent form before sensory evaluation. Since the target participants were between 6 to 9 years old, a verbal assent was issued. The nature and purpose of the study, as well as the researchers' request for permission, were played through a recording device before sensory evaluation. Then, the participants' verbal assent was documented using a recorder.

This research was conducted in accordance with current national ethical standards. The informed consent form included an information sheet that contains the researchers' introduction, the purpose of the research, type of research intervention, components and methods of the study, risks and inconveniences, study duration, possible benefits, compensation, voluntary participation, confidentiality, the right to refuse or withdraw from the study, and the researchers' contact information. Then, the informed consent form for LARs and Validators was processed for review by the University of Santo Tomas - Graduate School (UST-GS). In addition, specific, detailed explanations and clear examples of what the participants were required to perform were provided to the participants. All questions regarding participation were answered until they fully understood.

The information sheet and informed consent form for LAR contain all the necessary information in the study and it was discussed to the participants prior to the signing of the informed consent form. The cake sandwich was developed per batch, in which the first batch was subjected to microbiological testing to ensure its safety for consumption and evaluation by professional validators. Following the approval of the validators, the second batch of cake sandwiches was prepared for consumption and evaluation by the selected participants. Any allergens present in the product were discussed to the participants.

Moreover, data collection was done using a 5-point hedonic face scale for children and validators respectively; thus, validators and participants took part in the study for 10 to 20 minutes. Any individual that may exhibit an allergic reaction to the cake sandwich's ingredients was excluded from the study. All information collected was kept with utmost confidentiality, and it was not used for any other purpose outside of the study.

The researchers handled and safeguarded the personal information of all study participants following the Data Privacy Act of 2012 and its 2016 implementing rules and regulations (IRR). The acquisition, retention, and processing of personal information corresponded to the principles of transparency, legitimate purpose, and proportionality. Participant data would be kept for 3 years following its publication. After three years, electronic data sheets would be deleted irreversibly, and printed data sheets would be shredded and discarded.

2.11.1. Risks

No significant adverse reactions have been documented in relation to the consumption of papaya leaves in its various forms, concentrations, and durations across published clinical studies regarding the toxicity of papaya leaves. However, mild gastrointestinal disturbances and rashes may arise as these were the most commonly reported side effects from the consumption of papaya leaves. Therefore, a registered nurse was present during the sensory evaluation with the eligible children in order to monitor and provide first aid to any allergic reactions or potential adverse effects associated with consumption of papaya leaves. To ensure the safety of the children, a follow-up call was conducted to the assigned school teacher to verify that no instances of food poisoning, rashes or irritation, and gastrointestinal disturbances had occurred following the sensory evaluation. In the event of an emergency or severe adverse reaction to the food product prompting hospitalization, the researchers would reimburse all hospital expenses, including transportation fees, prescription medications, and check-up costs

3. Results

Table 4 illustrates the difference in vitamin A content among the four formulations of the Caricake using the FNRI Menu Eval Plus. The computation of vitamin A was done to determine if adding papaya leaves increased the overall vitamin A content of the product. The initial computation was done in consideration of the vitamin A content of the other ingredients in the cake sandwich and its interpretation as vitamin-A rich. Additionally, the percent increase of the four formulations was adapted from the study of El-Gammal et. al (2016). The 1% increase in the formulation of the cake sandwich was estimated considering the acceptability of the product. The CCake with papaya leaves reached 30% of the NVR.

Serving Size: 3				
Formulations	Vitamin A content from Young Papaya Leaves (ugRAE)	Overall Vitamin A content (ugRAE)	Percent Contribution of vitamin A (%)	Interpretation
CCake0	0	116	29	Vitamin A Source
CCake1	51	118	30	Vitamin A-rich
CCake2	59	119	30	Vitamin A-rich
CCake3	68	119	30	Vitamin A-rich

Table 4 Computation of Vitamin A Content of Four Formulations

Reference: Codex Alimentarius Commission (2013)

Table 5 Nutrient Analysis of the Chosen CariCake for Vitamin A

Parameters	Results
Vitamin A (High Performance Liquid Chromatography)	344 ug RAE/100 g
Vitamin A (Per Serving)	120 ug RAE/35 g

Table 5 illustrates the nutrient analysis of vitamin A content per 100 grams for CCake2 was performed at SGS Philippines, Inc. High-Performance Liquid Chromatography was used to determine the overall vitamin A content of the Cake Sandwich. The results show that the cake sandwich has 1,145 IU of vitamin A or 344 ug RAE per 100 grams of cake sandwich while one serving (35 grams) of cake sandwich has 120 ug RAE of vitamin A.

Table 6 Initial Sensory Evaluation of Five Validators

Attributes	Formulations				
	CCake1	CCake2	CCake3		
Appearance	5.6	6.4	5.2		
Aroma	6.8	7.4	6		
Texture	6.4	6.8	5		
Taste	5.8	6.2	5		
Overall Acceptability	6	6.4	5		
Mean	6	7	5		
Interpretation	Like slightly	Like moderately	Neither like nor dislike		

Table 6 shows the findings of the sensory evaluation scores performed by five professional validators. The objective of the initial sensory evaluation of CariCake was to assess the acceptability and identify which formulation should be provided to children aged 6 to 9 years old. Among all of the formulations, CCake2 received the highest rating in all attributes.



Figure 2 Actual Picture of CariCake2

The professional validators commented that the appearance and flavor of CCake2 are preferable to the other two formulations. Among the other formulations, CCake2 has the least bitter flavor and a light texture. Furthermore, the validators suggested adding more cocoa powder to the chocolate custard filling to improve the flavor of CCake2.

Attribute	Mean	Mode	Frequency of mode, n = 53	Interpretation
Appearance	4.8	5	43	Extremely Like
Aroma	4.5	5	38	Extremely Like
Texture	4.5	5	38	Extremely Like
Taste	4.6	5	40	Extremely Like
Overall Acceptability	4.5	5	37	Extremely Like

Table 7 Sensory Evaluation of School-aged Children (6-9 years old) on CCake2

Table 7 demonstrates the results obtained from the 53 school-aged children after evaluating the product using the 5point hedonic face scale. The scoring description ranged from (5) - Extremely Like to (1) - Extremely Dislike to examine the characteristics of the product, such as its Appearance, Aroma, Texture, Taste, and Overall Acceptability. The overall acceptability of the product had a mean score of 4.5.

Table 8 Demographic Distribution of School-aged Children (6-9 years old)

Demographic Profile	n	%				
Age						
6 years old	3	5.7				
7 years old	18	34.0				
8 years old	15	28.3				
9 years old	17	32.0				
Sex						
Male	21	39.6				
Female	32	60.4				

Table 8 displays the demographic distribution of school-aged children who participated in the study at Potrero Elementary School. Majority of the participants consisted of children aged 7 years, which constituted 34% of the total population. On the contrary, 60.4% of the total population consisted of female participants, indicating that there were more females than males.

For	Analysis Name							
mul atio ns					Mold & Yeast Count, CFU/g		S. aureus Count, CFU/g	
	Result	Interpretation*	Result	Result Interpretation*		Interpretation*	Result	Interpretation*
CCake1	1.1 x103	Acceptable	< 3	Acceptable	< 100	Acceptable	< 10	Acceptable
CCake2	170 est.	Acceptable	< 3	Acceptable	< 100	Acceptable	< 10	Acceptable
CCake3	45 est.	Acceptable	< 3	Acceptable	< 100	Acceptable	< 10	Acceptable

Table 9 Microbiological Test of Three Formulations

References: Philippine Food and Drug Administration (FDA). Circular-No.-2013-010 (2013); Salfinger, Y., & Tortorello, M. L. (Eds.). (2015). Compendium of methods for the microbiological examination of Foods. https://doi.org/10.2105/mbef.0222

*CFU/g = Colony Forming Unit per gram; MPN/g = Most Probable Number per gram; Est.= Estimated; **MPN/g is equivalent to CFU/g; ***Maximum allowable number of defective units (CFU/g): APC - 10⁶; TCC - 10³; MYC - 10⁴; S. aureus - 10⁴

Table 9 displays the results of the microbiological analysis conducted by Qualibet Testing Services Corporation among the three CariCake samples. The microbiological tests done on the cake samples included Aerobic Plate Count (APC), Total Coliform Count (TCC), Mold & Yeast Count (MYC), and *S. aureus* count. Based on the FDA Circular No. 2013-010 on the Assessment of Microbiological Quality of Processed Foods (2013), the acceptable levels of colony forming units per gram (CFU/g) in baked goods is 104 for Aerobic Plate Count (APC), 102 for *S. aureus*, 50 for Total Coliform Count (TCC), and 102 for Mold & Yeast Count (MYC).

Overall, CCake2 showed an estimated 170 CFU/g for the Standard Plate Count, less than 3 CFU/g for the Total Coliform Count, less than 100 CFU/g for the Yeast and Mold Count, and 10 CFU/g for the *S. aureus* Count, indicating that all cake sandwiches were safe for consumption. In HPLC nutrient analysis, CCake2 at 35 g yielded 120 ug of vitamin A. Furthermore, the 9-point hedonic rating scale showed that CCake2 was chosen by the validators due to its high ratings in the characteristics of appearance, aroma, flavor, taste, and overall acceptability. In addition, the sensory evaluation of CCake 2 by 53 children aged 6 to 9 provided favorable results, with an average score of 4.5 (Extremely Like) for appearance, taste, fragrance, mouthfeel, flavor, and overall acceptability.

4. Discussion

4.1. Microbiological Analysis

The microbiological analysis was conducted among three CariCake samples by Qualibet Testing Services Corporation, which included an Aerobic Plate Count (APC), Total Coliform Count (TCC), Mold & Yeast Count (MYC), and *S. aureus* count. For each analysis, four microbiological test methods were conducted. The pour plate method was used for Aerobic Plate Count (APC), the Three-Tube MPN technique for Total Coliform Count (TCC), the spread plate method for Mold & Yeast Count (MYC), and the Compact Dry technique for *S. aureus*. All methods conducted were based on the guidelines of the Compendium of Methods for the Microbiological Examination of Foods [24].

According to the FDA Circular No. 2013-010 on the Assessment of Microbiological Quality of Processed Foods [25], the acceptable level of microorganisms determined in baked goods (including eggs and dairy) is 104 for Aerobic Plate Count (APC), 102 for *S. aureus*, 50 for Total Coliform Count (TCC), and 102 for Mold & Yeast Count (MYC). On the contrary, the maximum allowable number of defective or marginally acceptable units is 106 for Aerobic Plate Count (APC), 104 for *S. aureus*, 103 for Total Coliform Count (TCC), and 104 for Mold & Yeast Count (MYC). Based on the results of the microbiological analysis, all CariCake samples were within the acceptable range for consumption.

4.2. Sensory Evaluation

The 9-point hedonic rating scale by Amy Brown was adapted in the study to collect data on the sensory evaluation for professional validators [22]. Meanwhile, the 5-point hedonic face scale by Singh-Ackbarali & Maharaj was also adapted in the study to determine the acceptability of CariCake among school-aged children [20]. From the 60 participants, 2 participants withdrew during the collection of assent and 5 were excluded from the study for not meeting the inclusion

criteria, garnering a total of 53 participants. Based on the evaluation of the school-aged children, the overall acceptability averaged at 4.5, indicating that the product was favorable. Due to the descriptive nature of the questions, descriptive statistical analysis was utilized.

In terms of the sensory evaluation of the validators, CCake2 received the highest ratings for all characteristics. The formulation also received the highest average grade for overall acceptability, 6.4 (Like Very Much). Therefore, CCake2 underwent mass production and nutrient analysis, as it was the formulation chosen for 53 children between the ages of 6 and 9. Some suggested that the product's appearance could be enhanced. Meanwhile, in appearance, the cake sandwich's packaging should be modified to make it more appealing. Next, the aroma, the majority of panelists agreed that the cake sandwich's aroma does not need to be modified as it smells sweet and milky. In addition, in terms of the product's texture, the majority of panels noted that it melts effortlessly in the mouth. Some have reported that it feels soft and spongy. In terms of taste, the majority of the validators praised the product's overall flavor. The cake sandwich was commended for not being overly sweet and having well-balanced flavors. However, some validators suggested adding additional cocoa powder to the cake sandwich's filling, which the researchers applied in mass production. The overall assessment of the product's acceptability is positive for all samples.

4.3. Vitamin A Content Analysis of CariCake

As seen in Table 6, each CariCake was computed for vitamin A to ensure that it meets the 30% of NRV to be considered vitamin A rich. After the evaluation of the validators, they chose CCake2, which was subjected to vitamin A content analysis. Based on Table 9, CCake2 has a vitamin content of 1,145 IU or 344 ug per 100 g of cake, and it contains 120 ug of Vitamin A per serving of 35 g. This indicates that the CCake2 is vitamin A rich as it met the 30% of NRV, and compared to the computed vitamin A, there was only a little difference among the CCake variations.

Vitamin A is considered relatively unstable during cooking and storage due to light, heat, and oxygen [26, 27]. Regardless of the preparation and baking of the product, the vitamin A content of CCake2 was retained. This shows that there was a minimal loss of vitamin A during preparation and baking, as there was only a minimal difference when the vitamin A computation was compared to the nutrient analysis of the CCake2.

The results of the vitamin A analysis correspond to the computation of CCake2 using MenuEvalPlus. The ground papaya leaves contributed an increase in the overall vitamin A content of the CariCake to reach 30% of the Recommended Nutrient Intake of Filipino children aged 6-9 years old. Although other ingredients used in the formulation were fortified, such as flour and cooking oil, as mandated in the Philippine Food Fortification law or Republic Act No. 8976, other ingredients, including milk, eggs, and butter, may have contributed as well to the vitamin A content of the product.

5. Conclusion

In conclusion, the researchers developed a vitamin A-rich cake sandwich and conducted a sensory evaluation on 53 Filipino children aged 6-9 years old to determine the product's acceptability. The researchers determined that 7 grams of ground papaya leaves should be added to 103 g of cake base for it to be a rich source of vitamin A for Filipino children aged 6 to 9 years old.

The researchers also determined the acceptability of the product's sensory attributes based on its appearance, aroma, texture, taste, and overall acceptability. The initial sensory evaluation, which consisted of 2 registered nutritionist-dietitians, 2 food technologists, and a bakery owner, showed that the CCake2 is the most suitable for mass production. Using a 5-point hedonic face scale with 53 Filipino children ages 6 to 9, the participants of the sensory evaluation classified the product as excellent, with an average score of 4.5.

Finally, the researchers evaluated the results of the cake sandwich's nutrient analysis, which was performed by SGS Philippines, Inc. According to the results, the cake sandwich contains 120 ug of vitamin A per serving, which is 30% of the age group's recommended daily allowance (400 ug). Thus, 1 serving of the product is considered to be rich in vitamin A. However, other key ingredients used in the formulation may have contributed to the vitamin A content of the product in contrast to the contribution of vitamin A content of ground papaya leaves.

In light of the findings of the study, it is recommended that school-aged children aged 6-9 years old should consume 35 grams of Caricake to help them achieve their recommended intake of vitamin A. Additionally, several limitations were identified, which may have affected the outcome of the study. In accordance with the Philippine Food Fortification Law or R.A. 8976, food items such as flour and oil are required to be fortified with vitamin A. These ingredients were substantially used in the development of CariCake. Other ingredients used in the development of the cake sandwich

included milk, eggs, and butter, which naturally contain vitamin A. Therefore, other ingredients used to make the cake sandwich have added to the overall vitamin A content of the cake sandwich.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors declare that there is no conflict of interest.

Statement of ethical approval

The present research work was ethically reviewed and approved by the University of Santo Tomas - Graduate School (UST-GS) and was conducted in accordance with the current national ethical guidelines.

Statement of informed consent

Informed consent and verbal assent were obtained from all individual participants included in the study.

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