

Effect of Retort Processing on the Nutritional Composition of Ready-To-Eat Chicken Pepes Sausage

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Abstract

Pepes is traditional Indonesia cuisine and it made from chicken mixed with spices and wrapped with banana leaves. Recently, ready-to-eat products such as sausage were popular in market because it offered convenience to consumers. Ready-to-eat chicken pepes sausage provide minimal preparation requirements, nutritious and make it a good option for quick snack and meal solution. The objective of this study was to formulate the chicken pepes sausage by incorporating two different plant-based protein and compare the nutritional composition of before and after retort was conducted toward chicken pepes sausage. Therefore, two formulations of chicken pepes sausage were formulate, which were: F1 (pea protein isolate) and F2 (soy protein isolate). The sausages undergo retort processing at 121 °C for 25 minutes. This study focuses on analyse of moisture, protein, fat, carbohydrate, crude fiber, ash and energy based on AOAC: moisture analyzer for moisture content, Kjeldahl for protein content, Soxhlet for fat content, Different method for carbohydrate content, crude fiber analysis for crude fiber content, dry ashing for ash content and Atwater system for energy. Results showed that the nutritional properties of chicken *pepes* sausage are significantly changed by retort processing which F1 (pea protein isolate) has protein (10.58±0.05%), carbohydrate (35.23±1.04%), and energy (213.06±3.93%) while F2 (soy protein isolate) has protein (10.88±0.02%), carbohydrate (31.41±0.48%), and energy (201.20±1.90%). Thus, it can conclude the retort processing can reduce some nutritional properties such as moisture, protein, fat, ash and crude fiber in chicken *pepes* sausage.

1. Introduction

Pepes is traditional Indonesia cuisine but it also gaining popularity in Malaysia. *Pepes* are defined as a food that is wrapped in banana leaves and then steamed and grilled [1]. *Pepes* involved many spices such as garlic, onion, turmeric, cumin, fennel and ginger that contribute to pleasant aroma and taste. Each spices offers a unique taste and has some beneficial substances that can lower cholesterol, type 2 diabetes, cancer, inflammation and mental health problems [2]. Spices not only enhance the aroma and taste of food but it also having positive health effect.

Currently, sausage has become a popular choice among consumers because it offers convenience for busy individuals or families looking for time-saving. Sausages often require minimal preparation and come in variety of flavours and sizes. Ready-to-eat product such as sausage in high demand, particularly in towns and cities where employee in government establishment, public and industrial sectors consume them as meal substitute or to quench hunger [3]. Addition, sausages are cost-effective compared to other protein sources. Chicken sausage

offers high-quality protein alternative because chicken has higher protein (22.8-24.2%) compared beef (20-21.9%) and duck (19.4%) [4]. Chicken also contains high unsaturated fatty acids which are essential for developing treatment and preventative approaches to cardiovascular disease [5]. People seeking balanced fats and high protein in their diet can consider chicken as moderate source due to chicken contains essential amino acids required for muscle growth and chicken has lower levels of saturated fats than red meat that better for cholesterol management. Chicken sausage absorbs the rich flavours of the *pepes* that make it more appealing to consumers who enjoy authentic flavour.

Furthermore, the development of ready-to-eat chicken *pepes* sausage to enhance shelf-life of products by applying retort processing. Retort processing is thermal preservation technique utilized to extend the shelf life of food product by subjecting them to elevated temperatures under controlled conditions. Retorting 120 °C is the standard temperature for thermal sterilization that facilitate the elimination of harmful contaminants including *Clostridium botulinum* [6]. This prolongs the shelf life of ready-to-eat products for 2 years without requiring refrigeration. However, retort processing cause degradation of food quality which is lead to changes in nutritional composition of food such as nutrient losses, particularly in sensitive bioactive component. Urban consumer is increasingly interested in trying products that mix traditional flavours with the mainstream form like sausage which are typically popular in Western market. By combining *pepes* and sausage formulations can cater to both traditional flavour enthusiasts and those seeking convenience of meal options.

The aims of this research are to formulate the ready-to-eat chicken *pepes* sausage by incorporation two different types of plant-based protein and compare the nutritional composition of before and after retort conducted towards chicken *pepes* sausage. The nutritional properties were analysed including moisture, protein, carbohydrate, fat, crude fiber, ash and energy.

2. Materials and Method

2.1 Materials and Preparation of Pepes Paste

The ingredients and recipe used were obtained from Rasa.My blog [7]. The ingredients were purchased from local suppliers. The ingredients such as 50 g of onion, 20 g of garlic, 10 g of dried chilli, 10 g of lemongrass powder, 10 g of fennel, 10 g of cumin, 10 g of coriander powder, 10 ml of water, 8 g of ginger and 5 g of turmeric powder were grinded into paste. The paste was cooked with hot cooking oil. Then, 65 g of coconut milk, 7 g of salt and 14 g of sugar were added into paste.

2.2 Preparation of Chicken Pepes Sausage

Chicken breast mixed with egg and *pepes* paste [7]. The steamer was placed on stove and boiled the water until achieved 100°C. The mixture was steamed for 20 minutes. The mixture was grinded with fixed ingredients which are barley, wheat flour, tapioca flour, maltodextrin, black pepper, oregano and seasoning. There are two formulations with different plant-based protein isolate. Pea protein isolated used in formulation 1 and soy protein isolate used in formulation 2. The mixture was stuffed into plastic casing which diameter 15 mm and long 10 cm that can ensure consistent size and shape of sausage. Then, mixture was steamed at 100 °C for 20 minutes. Table 1 shows the ingredients of chicken *pepes* sausage.

Table 1 The ingredients of chicken *pepes* sausage

Ingredients	Formulation 1 (F1)	Formulation 2 (F2)
	Quantity, g	Quantity, g
Chicken breast	220	220
Pepes paste	40	40
Egg	15	15
Barley	15	15
Wheat flour	20	20
Tapioca flour	10	10
Pea protein isolate	7	-
Soy protein isolate	-	7
Maltodextrin	5	5
Black pepper	0.5	0.5
Oregano	0.15	0.15
Seasoning	8	8

2.3 Retort Processing

The sausage was packed in retort pouch (7cm x12cm) and sealed hermetically. The material of retort pouch used is co-extruded nylon and polyethylene that resistant with high temperature and protects food from oxygen, moisture and contaminants. The water was filled into retort machine. The stainless-steel plate and pouches were arranged into the retort machine. Temperature was set at 121 °C for 25 minutes [8]. The lid was closed when the retort machine reaches at 100 °C. After the processing time, the retort machine was allowed to cool naturally to ambient temperature, and the pouches were removed once the cooling was complete.

2.4 Nutritional Content Analysis

2.4.1 Moisture Content

Moisture content in the sample was determined by using moisture analyzer according to Association of Official Analytical Chemists (AOAC). The amount of 5 g of sample was placed on an aluminium pan or filter paper and heated at 200 °C. The instrument automatically weighed the sample and determined the percentage of solid when the moisture is exerted out of it.

2.4.2 Protein Content

According to AOAC, Kjeldahl method used to determined protein content in the sample based on nitrogen determination. The amount of 1 g of sample was weighed and transferred into the flask. The 2 tables of catalyst and 12 ml sulfuric acid were added. The sample was digested into the digestion tubes at 420 °C for 60 minutes. The flask heated until the sample clear and no precipitate remain and cooled the sample. Then, the amount 10 ml distilled water added into flask. Next, the sample was immersed in 30 ml standard acid which is 40% boric acid. The amount 50 ml sodium hydroxide was poured into sample and a steam generator distilled the sample. For titration that using 1.0 N hydrochloric acid. The amount of acid used to colour change recorded. Equation (1) to obtain percentage of the nitrogen while equation (2) to determine the percentage of protein.

$$\% \text{ Nitrogen} = \text{Normality HCL} \times \frac{\text{corrected acid volume}}{\text{weight sample}} \times \frac{14 \text{ g N}}{\text{mol}} \times 100 \quad (1)$$

$$\% \text{ Protein} = \% \text{ Nitrogen} \times \text{Protein factor} \quad (2)$$

2.4.3 Carbohydrate Content

The carbohydrate content calculated by different method and the total carbohydrate can be determined by equation (3).

$$\text{Carbohydrate} = 100\% - (\text{fat} + \text{protein} + \text{moisture} + \text{ash}) \quad (3)$$

2.4.4 Fat Content

Fat content in the sample was determined by using Soxhlet method according to Association of Official Analytical Chemists (AOAC). The amount 2 g sample was weighed on filter paper. The sample was placed in the thimble with cotton wool and the thimble was placed into an extraction beaker. Hexane was added into extraction beaker. The thimble was inserted in a Soxhlet extractor and the extraction beaker and the extraction were started. After extraction done, the extraction beaker was heated at 105 °C for 1 hour if some residue solvents are not removed. After that, extraction was cooled for 1 hour in desiccator. The flask and extracted fat were weighted. Fat content was calculated by equation (4).

$$\% \text{ fat} = (\text{weight of flask \& extracted fat} - \text{weight of empty flask}) \times \frac{100}{\text{weight of sample}} \quad (4)$$

2.4.5 Crude Fiber Content

Crude fiber content in the sample was determined by using crude fiber analysis. The amount of 2 g of sample was transferred into conical flask with 200 ml of 0.128 M of sulfuric acid. Then, the flask placed on a hot plate and boiled it for 30 minutes. The flask was rotated to ensure the proper boiling of sample. Next, the sample was

filtered to drain the acid solution and washed the flask with hot water to remove the acid residue. the amount of 200 ml of 0.313 M sodium hydroxide was poured into conical flask washing the filtrate. The flask was boiled for 30 minutes and filtered the sample with hot water. The filtered collected in crucible and put in the hot oven at 230 °C for 2 hours and cooled in desiccator. The crucible contain fiber was weighted. The crucible was placed inside at furnace at 550 °C for 2 hours and cooled it in desiccator. The crucible was weighed. Total crude fiber can be calculated by using formula equation (5).

$$\text{Crude fiber \%} = \frac{(\text{weight crucible with fiber} - \text{weight crucible with ash})}{\text{weight sample}} \times 100 \quad (5)$$

2.4.6 Ash Content

Based on AOAC, ash content in the sample was calculated by using dry ashing. The crucible and lid were placed in furnace at 550 °C 3 hours. The crucible was transferred into a desiccator and cooled it. The amount 2 g of sample was put into crucible and placed on hotplate. The crucible was heated until the smoking stopped. The crucible was weighed. The crucible was transferred into muffle furnace for 3 hours at 550 °C. Then, crucible transferred to desiccator and cooled it. The crucible was weighed. The total ash could be determined by using the equation (6).

$$\text{Ash \%} = \frac{\text{weight after ashing}}{\text{weight before ashing}} \times 100 \quad (6)$$

2.4.7 Energy

Energy in the sample was determined by using Atwater system and the energy can be computed by using the formula in equation (7).

$$\text{Energy, kcal / g} = (\text{carbohydrate} \times 4) + (\text{protein} \times 4) + (\text{fat} \times 9) \quad (7)$$

2.4.8 Statistical Analysis

The result shown the mean \pm standard deviation of triplicate tests. Two-way ANOVA was performed to assess significant differences between formulations and between before and after retort processing was conducted [9]. Statistical significance was confirmed at the 95% confidence level using *p*-value.

3. Result and Discussion

3.1 Nutritional Composition

There are two formulation which F1 (chicken pepes sausage with pea protein isolate) and F2 (chicken pepes sausage with soy protein isolate). The nutritional properties of two formulations were compared. A comparison was made between before and after retort processing was conducted toward chicken pepes sausage. The nutritional composition was analysed includes moisture, protein, carbohydrate, fat, crude fiber ash and energy.

3.1.1 Moisture Content

The highest moisture content value was obtained on chicken *pepes* sausage F2 for before retort processing and the lowest was obtained on F1 for before retort processing. Furthermore, significant differences ($p < 0.05$) were obtained on F1 and F2. This might be due to different plant-based protein used. Pea protein isolate and soy protein isolate affect the moisture in sausage due to their unique water-binding. The water-binding properties can be attributed to several mechanism such as large pores in the matrix can enhance the capacity to absorb and hold water. According to [10] soy protein isolate has lower bulk density than pea protein isolate that showed the pore sizes of soy protein large. The larger pore in soy protein matrix allows it to trap more water molecules that lead to a higher moisture content. Moreover, the highly hydrophilic nature of the isolated protein in soy protein isolate enables efficient hydrogen bonding with water molecules that helps to enhance water retention.

Retort processing can cause the moisture content of meat product decrease. This study reported that moisture content of both formulation decrease after retort processing was applied which formulation 1 (50.32% to 47.57%) and formulation 2 (51.29% to 51.56%). Retort process of chicken *pepes* sausage required high temperature (121 °C). The high temperatures can cause evaporation of water, resulting in a dryer product [11]. From Table 2, it can conclude the moisture content of chicken *pepes* sausage significantly decrease after retort

process. [12] also found that a significant decrease in moisture content during retort process of dried beef rendang production. Table 2 shows moisture content for chicken *pepes* sausage.

Table 2 Moisture content for chicken *pepes* sausage

Analysis	Moisture, %	
	Before	After
F 1	50.32±0.87 ^{a**}	47.57±0.69 ^{a*}
F 2	51.29±0.29 ^{b**}	50.73±0.68 ^{b*}

[F1 (formulation 1) and F2 (formulation 2)]

Values with different letters (a,b) in the same column are significantly difference ($p<0.05$)

Values with different symbol (*, **) in the same row are significantly different ($p<0.05$)

3.1.2 Protein Content

Chicken meat is good source of protein with less fat and cholesterol. The lowest protein content value was obtained on chicken *pepes* sausage F1 for before retort processing and the highest was obtained on F2. The significant different ($p<0.05$) were showed on F1 and F2. Difference of protein content due to ingredients in formulations has different plant-based protein isolate. Plant-based protein isolate used as a binder in sausage to improve texture by providing elasticity and firmness. Soy protein showed higher protein because it derived from soybean which contains 42% protein [13]. Pea protein derived from peas that consist 20-24% of protein [14].

Previous study was reported that chicken sausage contains 13.73% of protein content [31]. [22] also found chicken sausage consists 14.56% protein. [32] also reported that commercial chicken sausage contains 11.89% protein while chicken sausage incorporated with baby corn contains 14.53% protein. Compared with previous research, this study obtained lower range of protein content due to different types of meat and protein source in other ingredients. This study reported that retort processing appears reduce protein content in formulation 1 (10.88% to 10.58%) and formulation 2 (11.19% to 10.88%). Retort processing involves high temperature cause proteins to denature lead to reduced solubility that make it harder to extract and measure the protein content during analytical testing. From Table 3, it can conclude the protein content of chicken *pepes* sausage significantly decrease after retort processing. Table 3 shows protein content for chicken *pepes* sausage.

Table 3 Protein content for chicken *pepes* sausage

Samples	Ash content, %	
	Before	After
F1	10.88±0.02 ^{a**}	10.58±0.05 ^{a*}
F2	11.19±0.02 ^{b**}	10.88±0.02 ^{b*}

[F1 (formulation 1) and F2 (formulation 2)]

Values with different letters (a,b) in the same column are significant different ($p<0.05$)

Values with different symbol (*, **) in the same row are significantly different ($p<0.05$)

3.1.3 Carbohydrate

The result of this study showed that carbohydrate in formulation 1 found to be increasing by retort processing which increasing from 31.56% to 35.23% and carbohydrate in formulation 2 also increase (30.14% to 31.41%). The significant different ($p<0.05$) was obtained on before and after retort process was conducted toward both formulations of chicken *pepes* sausage. The enhancement of carbohydrate content related to the loss of soluble solid which it can increase the concentration of carbohydrate content of chicken *pepes* sausage. Leaching during the retort processing may result in the loss of protein and other compound during the cooking process [14]. This might be due to Millard reaction occur during retort processing. [14] also reported that carbohydrate content of wing beans increasing by sterilization process. Accumulation of carbohydrates as a result of the gelatinization of starch. This sausage might include starchy component such as tapioca flour and wheat flour that gelatinize during the retort process. [32] found chicken sausage with different vegetable contains 8.75% to 11.09% of carbohydrate. [25] also reported the range of carbohydrate level of commercial chicken sausage in Malaysia between 6.69% to 21.59%. Carbohydrate content in this study not in the range due to starch-based ingredients used in formulation. Table 4 shows carbohydrate content for chicken *pepes* sausage.

Table 4 Carbohydrate content for chicken *pepes* sausage

Samples	Carbohydrate content, %	
	Before	After
F1	31.56±1.00 ^{b*}	35.23±1.04 ^{b**}
F2	30.14±0.29 ^{a*}	31.41±0.48 ^{a**}

[F1 (formulation 1) and F2 (Formulation 2)]

Values with different letters (a,b) in the same column are significant different (p<0.05)

Values with different symbol (*, **) in the same row are significantly different (p<0.05)]

3.1.4 Fat Content

According to Malaysia Food Regulation 1958, sausage should not contain more than 30% of fat. [22] reported fat content of chicken sausage with differs amount of hempseed (0%, 10% and 20%) were 11.77%, 11.50% and 10.30% respectively. [31] also found chicken sausage with 0% and 2% banana peel powder were 9.18% and 7.68%. this study obtained lower fat content than previous study due to different ingredient used in formulation. Table 5 indicate the fat content in formulation 2 decrease after retort was applied from 3.68% to 3.56% and fat content in formulation 1 also reduce after retort was applied from 3.42% to 3.32%. Reduction of fat content due to lipid oxidation and changes in fatty acids compositions [15]. High temperature accelerates the rate of oxidation by increasing the energy available for chemical reaction that can lead to the breakdown of unsaturated fatty acid into hydroperoxides, aldehydes and other secondary oxidation products. Heat also can degrade antioxidants which reducing their ability to inhibit lipid oxidation. High temperature also can breakdown the longer fatty acid chains into shorter and produce more volatile compounds that can affect flavour and aroma profiles. Table 5 shows fat content for chicken *pepes* sausage.

Table 5 Fat content for chicken *pepes* sausage

Samples	Fat content, %	
	Before	After
F1	3.42±0.04 ^{a**}	3.32±0.03 ^{a*}
F2	3.68±0.15 ^{b**}	3.56±0.01 ^{b*}

[F1 (formulation 1) and F2 (formulation 2)]

Values with different letters (a,b) in same column are significantly different (p<0.05)

Values with different symbol (*, **) in the same row are significantly different (p<0.05)

3.1.5 Crude Fiber Content

High fiber diets are usually lower in fat and energy density and help to retain a healthy body [16]. The result of this study showed the range of crude fiber in chicken *pepes* sausage were 0.49% to 1.7% which higher than previous research. According to [17], chicken sausage contains 0.21% of crude fiber while [18] found chicken sausage has 0.12% crude fiber. This might be incorporation of fiber sources from barley grains that used in this study. Barley grain is rich source of fiber approximately 17.3% which is higher than in many cereals [19]. The high fiber content in barley contributes to reducing the risk of chronic disease such as cardiovascular. Fiber can improve the texture of food depending on the type of fiber used and the food product it is added to. [20] found use of apple pulp as fiber to enhance texture and nutritional profile of chicken nugget. [21] also reported use of orange peel fibers to improve texture of a bologna-type products. [22] reported hempseed meal increase the fiber content and texture profile of chicken sausage. Table 6 shows crude fiber content for chicken *pepes* sausage.

Table 6 Crude fiber content for chicken *pepes* sausage

Samples	Crude fiber content, %	
	Before	After
F1	1.70±0.71 ^{b**}	0.92±0.20 ^{b**}
F2	0.86±0.50 ^{a**}	0.49±0.07 ^{a**}

[F1 (formulation 1) and F2 (formulation 2)]

Values with different letters (a,b) in same column are significantly different (p<0.05)

Values with symbol (**) in the same row are no significantly different (p<0.05)]

3.1.6 Ash Content

Chicken meat has low mineral. Chicken breast contains 1.10% to 1.14% of ash content [23] and commercial boiler contains 1.30% ash content [24]. Minerals such as calcium, potassium, iron, zinc and magnesium are essential nutrients that contribute to various physiological functions including enzyme activation and immune function. Based on Table 7, the range ash content of before and after retort was conducted toward chicken *pepes* sausages were 3.32% to 3.83%. Compare with previous study, ash content of this study higher than ash content in previous study. [25] reported that the range of ash content in commercial chicken sausages were 2.17% to 3.30%. Another study by [26] found the percentage of ash in chicken sausage with addition of spinach is 2.95%. The difference of total ash of this study and previous study that might be due to the types of meat used such as mechanical deboned meat or traditional hand deboned meat and ingredients in formulation [25]. However, spice used in formulations such as turmeric, ginger, and cumin contain varying amount of minerals. [27] found ginger contains 68.28 mg/100 g calcium, 8.42 mg/ 100 g iron, 128.58 mg/ 100g potassium and 102.67 mg/ 100 g magnesium while turmeric consists 14.07 mg/ 100g calcium, 19.09 mg/ 100 g iron, 7.08 g/ 100g potassium and 93.9 mg/ 100g magnesium. Akinwande and Olatunde [28] also reported zinc level in garlic (66.08 mg/ kg) higher than onion (38.44 mg/ kg) while ferum level in onion (118.15 mg/kg) higher than garlic (30.16 mg/ kg). Table 7 shows ash content for chicken *pepes* sausage.

Table 7 Ash content for chicken *pepes* sausage

Samples	Ash content, %	
	Before	After
F1	3.83±0.15 ^{a**}	3.31±0.60 ^{a**}
F2	3.69±0.32 ^{a**}	3.41±0.49 ^{a**}

[F1 (formulation 1) and F2 (formulation 2)

Values with different letters (a,b) in same column are significantly different ($p < 0.05$)

Values with symbol (**) in the same row are no significantly different ($p < 0.05$)

3.1.7 Energy

According to Table 8 shows that energy in F1 increase after retort processing applied from 200.54 kcal/g to 213.06 kcal/g. Energy in F2 also increase from 198.49 kcal/g to 210.20 kcal/g after retort processing. During retort processing, the energy content of F1 and F2 increased significantly ($p < 0.05$). Enhancement of energy after retort because retort processing involves high temperature and pressure that cause water in food evaporated. This can concentrate the remaining solids such as carbohydrate, fat and protein that increasing the density of calories per unit weight of food [29]. Retort processing cause starch gelatinization which is crystalline structure of starch granules is damaged resulting them more digestible. Digestive enzymes can more readily hydrolyze gelatinized starches which increase the rate and amount of glucose absorption [30]. Protein hydrolyzes into amino acids and small peptides through enzymatic digestion which these amino acids are absorbed into bloodstream and transported to various tissues for energy production and muscle repair. Protein is not directly converted into energy in the form of glucose but it contributes 4 kcal per gram. Carbohydrate provides 4 kcal per gram because it easily converted into glucose and it offer the body an immediate and efficient energy source.

Table 8 Energy content for chicken *pepes* sausage

Samples	Energy content, kcal/g	
	Before	After
F1	200.54±3.93 ^{b*}	213.06±3.93 ^{b**}
F2	198.49±1.05 ^{a*}	201.20±1.90 ^{a**}

[F1 (formulation 1) and F2 (formulation 2)

Values with different letters (a,b) in the same column are significantly different ($p < 0.05$)

Values with different symbol (*, **) in the same row are significantly different ($p < 0.05$)

4. Conclusion

In conclusion, the development of ready-to-eat chicken *pepes* sausage by incorporation two different types of plant-based protein were successfully in providing convenient, nutritious and delightful meal options. For 25 minutes, the sausage undergoes retort processing at 121 °C to prolong the shelf life of the sausage product. This process allows for the sausage suitable for commercial distribution by ensuring that it can be store at room temperature for period of time without spoilage. Therefore, findings revealed that retort processing significantly influences the nutritional properties of chicken *pepes* sausage. Retort processing led to changes in various

nutritional parameter including moisture, protein, carbohydrate, fat and energy due to heat exposure, water loss and chemical reactions. This study found moisture, protein and fat of chicken *pepes* sausage lower than previous study which might be due to difference formulation and processing that can affect nutrition of sausage. Some of nutrients such as carbohydrate, crude fiber and ash were obtained higher than previous study due to presence of fiber in barley and addition of spice incorporation to mineral content. Energy of chicken *pepes* sausage in range with previous study. Heat degradation and protein denaturation cause some nutrient such as moisture, protein, fat, ash, crude fiber was decrease after retort processing. Carbohydrate and energy after retort were increase because moisture loss that affect concentration of nutrient. Retort processing helps the sausage achieves an extended shelf life and maintain safety. Therefore, retort processing can affect the nutritional properties of chicken *pepes* sausage. The recommendation may apply for future studies such as optimizing formulation for better moisture retention and shelf-life extension or conducted microbial stability testing under different storage conditions. Therefore, retort processing can affect the nutritional properties of food.

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Conflict of Interest

Authors declare that there is no conflict of interests regarding the publication of the paper.

Author Contribution

The authors confirm contribution to the paper as follows: **study conception and design, data collection, analysis and interpretation of results and draft manuscript preparation:** Arni Fazlin Amir, Norhayati Muhammad. All authors reviewed the results and approved the final version of the manuscript.

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