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FULL PAPER

Comparison of nutritional composition, chemical preservative, and glutamic acid content of canned food with freshly cooked and home-cooked food products

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Abstract

Thermal preservation using the canning method is a promising alternative for retaining the maximum quality of the foods. Recently, the rising awareness of the nutritional value of canned products has received the attention of various researchers, as canned food can offer both healthy and convenient solutions to consumers. The present study reported the nutritional values and presence of food additives and preservatives in different Ayam Brand™ canned foods in tomato sauce namely sardines (CS), mackerel (CM) and baked beans (CB), canned tuna flakes in water (CTF) and canned tuna in mayonnaise (CTM) compared to the fresh and home-cooked food samples. The canned products, CS and CM contain significantly higher (p<0.05) amounts of Omega-3 but are lower in protein compared to their fresh counterparts. The calcium compositions of canned sardines and mackerels are 10-fold higher than fresh and home-cooked, contributed by the soft and brittles bones which enriched with calcium. The nutritional values of canned products were maintained throughout the storage period throughout the span of 3 years, with insignificant changes (p>0.05). Regardless of canned or fresh food samples, there was no total dietary fiber, benzoic acid, sorbic acid and sulfur dioxide detected in all samples except for the dietary fiber in the baked beans. CS and CTM contained significantly (p<0.05) higher amounts of glutamic acid than their fresh counterparts and the glutamic acid in all canned products ranging from 0.001-0.37 mg/g which is within the acceptable daily intake of 13 g/day. The natural glutamic acid was contributed by the raw materials of fish and tomato sauces themselves. No MSG was detected in all canned products. Therefore, it is safe to consume canned food products that greatly representing fresh and home-cooked foods without compromising with the nutritional values.

1. Introduction

Fish is one of the major sources of protein, providing 17% of animal protein and 7% of total protein consumed by the world's population. Besides protein, fish also contains vitamins, omega-3, high unsaturated fatty acids (HUFA), eicosapentaenoic (EPA), docosahexaenoic (DHA) acids, amino acids, minerals, and carbohydrates, making it a vital food source (Abraha *et al.*, 2018). Nevertheless, owing to their biological composition and a high degree of microbial load on the surfaces, fishes are highly susceptible to deterioration and have a short shelf life (Abraha *et al.*, 2018). Due to the exponential demand for fish meal, bean (*Phaseolus vulgaris* L.) also has been used as an alternative product to replace animal protein in the human diet (Schoeninger *et al*., 2017). In addition, freshly harvested common beans with high moisture content are also highly prone to deterioration and microbial contamination (Manzoor *et al.*, 2019). As a result, a number of food processing and preservation methods have been developed to overcome these problems.

Canning is one of the most efficient thermal processing methods to destroy the pathogenic and spore-forming microorganisms as well as to inactivate undesired spoilage-causing enzymes present in the food (Kapica and Weiss, 2012). Canned foods have a longer shelf life than their fresh counterpart by offering a convenient solution in terms of affordability and accessibility to consumers, especially those who have limited access to fresh food, storage, preparation facilities, time, and food preparation skills. Apart from being convenient, canned foods are also practical and

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recommended for consumers to plan, prepare and add more nutrient-dense foods into their diet without compromising their budget. In addition, canned foods can be stored much longer compared to freshly or home-cooked foods with minimal food waste, thereby, reducing the occurrence of spoilage and preparation costs (Kapica and Weiss, 2012).

The growing awareness of a healthy lifestyle has led consumers to become more particular and concern about their food intake. Canned foods are always thought not nutritious as frozen or fresh foods (Pasha *et al.*, 2014; Perito *et al.*, 2020). Throughout the years, canning processes are optimized and studied to ensure the end products (canned foods) nutrients and quality factors (taste, flavour, and colour) are maximally retained. Raw materials used in the canning process are usually selected at their best quality with good post-handling and storage (Bahurmiz *et al.*, 2018). Several types of research have shown that the canning process may reduce/maintain the nutritional value when comparing to fresh or frozen foods. This finding was supported by Abdullahi *et al.* (2016) where the carbohydrates, proteins, crude fibre, ash, sodium, potassium, and calcium concentrations were significantly higher ($p<0.05$) in canned tomato compared to fresh tomato. In addition, canning also increased the nutritional composition of pumpkins such as carotenoids calcium, iron, magnesium and vitamin K (Barnes-Svarney and Svarney, 2015).

Food additives and preservatives are commonly used in food industries to preserve or enhance the appearance, flavour, and taste of food products. One of the very common food additives is monosodium glutamate (MSG). MSG is commonly used in processed food products such as canned food as a flavour enhancer (Wang and Adhikari, 2018). In food canning, food preservatives such as benzoic acid, sorbic acid, and sulphur dioxide are also commonly added to control the growth of microorganisms (Perito *et al.*, 2020). Despite these advantages, food additives and preservatives have been associated with adverse health effects, thus leading to consumers demand 'clean label' food products (Populin *et al.*, 2007; Sharma, 2014; Silva and Lidon, 2016; Mirza *et al.*, 2017).

This study aimed to assess whether canned food could deliver nutrient levels comparable to fresh and home-cooked food. In this study, different Ayam Brand™ canned foods in tomato sauce namely sardines (CS), mackerel (CM), canned tuna flakes in water (CTF), and mayonnaise (CTM) were analysed to compare their nutritional composition with fresh cooked and home-cooked food samples. An accelerated shelf-life study was also carried out on canned fish products (CS, CM, CTF, and CTM) and baked beans (CB) to examine their freshness and extent of nutrition loss during storage. Chemical tests were also carried out to ensure the absence of chemical preservatives and MSG.

2. Materials and methods

2.1 Samples

For canned samples, five different canned samples from Ayam Brand™ (A.Clouet and Co. Sdn Bhd, Malaysia) were acquired in this study, which was tuna flakes in water, tuna in mayonnaise, baked beans, sardines and mackerel in tomato sauce conserve. Samples had undergone normal processing as commercial Ayam Brand™ products.

The frozen fishes were supplied and stored in a cool chamber upon arrival before being transferred to the production for thawing, beheading, gutting, cut into smaller sizes and cleaned under running tap water. The cleaned fishes were then filled into cans and steamed at 100°C for 20-25 mins. The fresh-cooked samples were filled with the same tomato sauce at the same ratio as in the canned version accord, except that it did not undergo the canning and commercial sterilization process.

For home-cooked samples, fresh sardines and mackerel were purchased from a local fresh market. Upon arrival, the whole fish was beheaded, gutted, and cleaned with running tap water. The fish was cooked with tomato sauce made of commercial tomato ketchup, chilli sauce, onion, garlic, and some oil. For home-cooked tuna in mayonnaise, the preparation involved drained out a can of Ayam Brand™ tuna in water, and mix with commercial mayonnaise and some chopped onion, at the same ratio as in the canned version.

2.2 Accelerated shelf-life study

The accelerated shelf-life study was carried out on all the canned food products. For the accelerated storage study, all the canned food samples were stored for 4.5 months at 55°C which is equivalent to 40.5 months at 25°C. During storage, the samples were collected at regular intervals of 1.5, 3.0 and 4.5 months to fit the kinetic model based on the Arrhenius method. The accelerated test samples were analysed for Omega-3, calcium, protein, and dietary fibre.

Arrhenius equation:

$$t_2 = \frac{t_1}{Q_{10}^{(\Delta T/10)}}$$

Where t_1 = shelf life at temperature T_1, t_2 = shelf life at temperature T_2 and ΔT = different in degree Celsius between T_1 and T_2

2.3 Real-time ageing

The real-time ageing sample was carried out on the canned food products of sardine (TS) and mackerel (CM) in tomato sauce. The samples were stored at room temperature (31°C) for 3.5 years before being subjected to omega-3 analysis.

2.4 Nutrient analysis

2.4.1 Omega 3 (α-linolenic acid, eicosapentaenoic acid and docosahexaenoic acid

Omega 3 fatty acid analysis was carried out by using the method adopted from AOAC and IUPAC 5th Edition IID.19 / IID.25. This method involves the quantification of α-linolenic (ALA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA acid in canned, fresh, and home-cooked of sardines, mackerel and tuna fish. The determination is performed by direct transesterification of fatty acids to the corresponding fatty acids methyl esters (FAMEs) with methanolic potassium hydroxide. Then, FAME was analysed by gas chromatography equipped with a flame ionization detection (GC-FID) system to separate and quantify each FAME component. Besides that, pure FAMEs standards were prepared as internal standards for the identification and quantification of fatty acids by comparing the retention times of samples under the same operating conditions.

2.4.2 Lipid extraction

Lipid extraction from food samples (whole content) was carried out by using the Soxhlet extraction method according to AOAC 920.39. About 2 g of sample was weighed on filter paper, wrapped, and inserted into extraction thimble. The extraction thimble was placed into the Soxhlet extractor, followed by the addition of 150 mL of petroleum ether. The reflux process was then carried out. After the 6 h of reflux process, the solvent was distilled off. The weighing flask containing the extracted lipid was then dried at 100°C, cooled in a desiccator and weighed.

$$\% \text{ of Lipid} = \frac{B - A}{C} \times 100\%$$

Where A = Weight of weighing flask, B = Weight of weighing flask with lipid, C = Weight of sample

2.4.3 Fatty acid methyl ester

The lipid extracted from Soxhlet extraction was methylated and converted into fatty acid methyl ester (FAME). Approximately 0.1 - 0.2 g of extracted lipid extract was transferred into a test tube. The extracted lipid extract was then dissolved in 10 mL of hexane followed by the addition of 100 µL 2N potassium hydroxide. The test tube was capped, vortexed for 1 min and centrifuged. The clear supernatant was then transferred into 2 mL GC vials for injection.

2.4.4 Gas Chromatography analysis (GC)

GC analysis was performed on an Agilent 7890A GC equipped with a flame ionization detector (FID). The automated split injection was performed using an Agilent 7963 autosampler. The column used was 60 m × 0.25 mm ID × 0.15 µm DB-23 (JandW 122-2361). The experimental conditions (inlet temperature, injection volume, split ratio, carrier gas, detector temperature) were 250°C, 1 µL, 1/50, helium gas and 250°C, respectively. The oven temperature was held at 50°C for 1 min, followed by an increment of 25°C per min until it reached 175°C and increment of 4°C per min until it reached 230°C. The temperature of the oven was held for 5 min when it reached 230°C. The pressures of hydrogen, air and nitrogen gases were 40 mL/min, 450 mL/min and 30 mL/min, respectively. Vials containing FAMEs standard and samples were injected into GC-FID. The retention times of the selected FAME standard were used to identify individual FAs and TFAs in the sample. Omega-3 fatty acids were determined by comparing the retention times of standard ALA (C18 3n:3), EPA (C20 5n:3) and DHA (C22 6n:3) with the samples. Each FAs and TFAs were determined by using formula below:

$$\% \text{ of} \frac{\text{FAs}}{\text{TFAs}} = \% \text{ of Total from Chromatogram} \times \% \text{ of Total Lipid Extracted}$$

2.4.5 Calcium

The calcium concentration was determined following the AOAC method 985.35. Approximately 25 to 50 mL aliquot of grounded food sample (whole content) was weighed on the Vycor evaporating dish containing 5 g of filter pulp and dried in an oven at 100°C. After the aliquot was dried, it was heated on a hot plate until smoking ceases. The dish was then placed in 525°C muffle furnace until the ignition was completed (indicated by the absence of black particles). The ash residues were then dissolved by heating in 5 mL of 1 M nitric acid. Dissolved residues were transferred into a 50 mL volumetric flask, treated twice more, and brought to volume with 1 M nitric acid. Lanthanum chloride releaser was then added to aliquots taken from the 50 mL volumetric flask for the quantification of calcium. The calcium was quantified by using atomic absorption spectrometry at the wavelength of 422.7 nm.

2.4.6 Crude protein

Protein was analysed by using the standard Kjeldahl procedure (MS 1194:1991). About 0.7 - 2.2 g of sample (whole content) was weighed and transferred to the Kjeldahl digestion flask. Approximately 8 g of catalyst mixture (a mixture of 96% anhydrous sodium sulphate,

3.5% copper sulphate and 0.5% selenium dioxide) and 20 mL of concentrated sulphuric acid were added. The solution was mixed by gentle swirling. The solution was swirled from time to time and the heating was continued until the solution became clear.

About 50 mL of boric acid solution (2% w/v) and methyl red indicator were then added to the receiving flask. The diluted digest alkaline was made with 50% w/v sodium hydroxide solution. The tap was then closed, and the ammonia was distilled into the boric acid solution. The distillate was then titrated with sulphuric acid or hydrochloric acid standard solution (0.5 N or 0.1 N).

$$\% \text{ Nitrogen} = \frac{(V_2 \times N_2) - (V_1 \times N_1)}{W} \times 1.4007$$

Where V_1 = Volume of NaOH Standard Solution (mL), V_2 = Volume of Standard Acid (mL), N_1 = Normality of NaOH Standard Solution, N_2 = Normality of Standard Acid, W = Weight of sample (g)

2.4.7 Total dietary fibres

The total dietary fibres were determined by using an enzymatic–gravimetric method (AOAC 985.29). Samples were defatted with petroleum ether three times with 25 mL portions (per g of sample) before milling. About 1 g of dried and defatted food sample was placed in a beaker. Approximately 50 mL phosphate buffer (pH 6.0) was added into the beaker followed by 50 µL of heat -stable α-amylose solution. The beaker was then wrapped with aluminium foil and placed in a boiling water bath for 15 mins. The beaker was stirred gently at every 5 min interval. The pH value of the solution was adjusted to 7.5±0.1 by using 0.275 N NaOH after being cooled to room temperature.

About 100 µL of protease solution was then added, followed by incubation at 60°C with continuous agitation for 30 mins. The pH value of the cooled solution was then adjusted to 4.5±0.2 with 0.325 N HCl, followed by the addition of 200 µL of amyloglucosidase and incubation (60°C with continuous agitation for 30 mins). After the incubation, 280 mL of 95% ethanol (preheated to 60°C) was added and the solution was left at room temperature for 60 min for precipitation. The residue was then washed with 20 mL of 78% ethanol three times, 10 mL of 95% ethanol two times and 10 mL of acetone two times. The residue was then decanted under a slight vacuum and filtered through the prepared glass filter crucibles, dried overnight at 105°C, cooled and weighed.

2.5 Chemical preservatives analysis

2.5.1 Benzoic acid and sorbic acid

Approximately 2 g of sample was weighed in a 50 mL volumetric flask, topped up with 50% MeOH and mixed well. The solution was then filtered with filter paper through a 0.45 µm filter disk. The sample extraction was then analysed by using HPLC. The conditions of HPLC were mobile phase (ratio 95:5), flow rate (1.0 mL/min), wavelength (227 nm) with UV detector and volume injection (20 µL). Standard solutions (10 ppm, 25 ppm, 50 ppm and 100 ppm) were injected to get peak area and the standard curve was achieved by using Chemstation Software. The sample extraction (20 µL) was injected, and the peak area obtained was compared with the standard curve.

$$\text{Sodium Benzoate or Potassium Sorbate, mg / kg} = \left(\frac{A}{B}\right) \times \text{Reference Standard} \times \frac{V}{\text{Sample Wt.}}$$

Where A = Peak area obtained from sample, B = Peak area obtained from reference standard, V = Top up volume. The amount of benzoic acid and sorbic acid in sample were calculated as:

$$\text{Sodium Benzoate, mg/kg} = \text{Benzoic Acid, mg/kg} \times 1.1801$$

$$\text{Potassium Sorbate, mg/kg} = \text{Sorbic Acid, mg/kg} \times 1.3397$$

Where 1.1801 and 1.3397 are the conversion factors, respectively

2.5.2 Sulphur dioxide

About 50 g of homogenized sample was weighed into distilling flask followed by addition of 200 mL of distilled water. Approximately 25 mL of 20% phosphoric acid solution and 20 mL concentrated HCl was then added into the flask. The solution was heated and boiled for 15 - 30 mins. The pale colour of the solution was maintained throughout the titration by adding 0.05 N of iodine from the burette into the receiving beaker.

$$\text{Sodium metabisulphite, mg/kg} = \frac{TV \times f \times 0.0016 \times 10^6}{W}$$

Where, TV = Titration value of the iodine used, f = Factor for iodine, W = Total amount of sample used. The amount of sulphur dioxide in sample was calculated as:

$$\text{Sodium metabisulphite, mg/kg} = \text{Sulphur Dioxide, mg/kg} \times 1.4831$$

Where 1.4831 is the conversion factor

2.5.3 Monosodium glutamic acid (MSG) content

The content of glutamic acid was analysed by using the method of AOAC 994.12 and JAOAC, Vol. 71, No. 6, 1988.

2.5.4 Performic acid oxidation

Approximately 100 - 1000 mg of finely grounded sample was weighed into the digestion tube. The tube was placed in an ice bath (0°C) and stirred for 15 mins. After cooling, 5 mL of performic acid was added into the tube, covered, and stirred for 15 mins on a magnetic stirring plate. The tube was returned to the ice bath and

left for oxidization. About 0.84 g of sodium metabisulfite was added into the tube to decompose the performic acid after 6 hrs of oxidization. The tube was stirred for 15 mins to liberate SO_2.

2.5.5 Hydrolysis

About 50 mL of 6M HCl-phenol solution was added to the test solution. The solution was then hydrolysed under reflux at 110 - 120°C using a digestion block. After 24 hrs, the digestion tube was removed from heat, cooled to room temperature and 20 mL of norleucine standard solution was added. The solution was then mixed. The hydrolysates were filtered through sintered glass filter into evaporating flasks. The hydrolysates were evaporated under vacuum at 40°C to 5 mL. Sodium citrate buffer (pH 2.2, 50 mL) was then added into the evaporated test solution and mixed well.

2.6 Glutamic acid content

The glutamic acid content was determined by using HPLC. The response factor, detection and quantification limits of glutamic acid were 2.64, 0.057 μM and 0.168 μM, respectively.

3. Results and discussion

3.1 Nutrients composition of canned, fresh-cooked, and home-cooked food samples

3.1.1 Omega-3

Every species of cold-water fish such as sardines, tuna, salmon, and herring contain a considerable amount of Omega-3 fatty acids and are widely canned according to regional specifications (Singer *et al.*, 2016). Omega-3 fatty acids have various health benefits such as preventing cancer, cardiovascular disease, inflammatory bowel disease, rheumatoid arthritis, psoriasis, and assisting brain development and function. Canning is the most important way to preserve fish since it involves the pasteurization and the sterilization process that helps to prevent the product from microorganism's spoilage (Mesías *et al.*, 2015). Canned foods offer convenience and year-round availability at a reasonable price, and one of the important sources of nutrients particularly proteins and Omega-3 in fish products (Bahurmiz *et al.*, 2017). In this study, the Omega-3 fatty acids content of canned samples was examined to investigate the alteration or loss of fatty acids during canning and storage.

Results in Table 1 shows that canned sardine (CS) and mackerel (CM) were significantly higher in omega-3 compared to fresh cooked and home-cooked samples, respectively. Omega-3 fatty acids in the canned fish were reported as dependent on the species of fish, quality of raw materials, type of liquid/oil used in canning, the process of heat treatment and/or cooking before canning, as well as the time and storage conditions (Teale, 2006). Fish that contain high levels of unsaturated fatty acids are susceptible to oxidative rancidity (Al-Reza *et al.*, 2015) due to the presence of double bonds in the fatty acid structure. These fatty acids could be degraded during cooking, particularly at high temperatures. The retort sterilization in the canning process involves a high-temperature application for an elongated period of time, hence ensuring adequate devitalization of microorganisms but with the possibility of inducing deterioration and loss of heat-sensitive nutrients (Nunes *et al.*, 1992; Mesías *et al.*, 2015).

According to the results in Table 1, the content of omega-3 fatty acids of all canned sardine (CS) was consistently maintained, showed no significant changes throughout the storage periods. Our results are in agreement with Stephen *et al.* (2010), where the canning process minimally deteriorated the omega-3 fatty acids. This is also evidenced by the results obtained where the sample that has been stored for 3.5 years has approximately the same omega 3 content (no significant changes) as the other CS samples. This indicated that omega-3 in the CS was well preserved throughout the years although it has been processed at high temperatures. The canning process that involves a high thermal treatment helps to retard the growth of spoilage microorganisms that may cause food deterioration and nutrients degradation. However, for fresh-cooked and home-cooked, the omega-3 content decreased significantly. This might be due to the thermal degradation of unsaturated fatty acids during cooking.

However, the result of omega-3 content in canned mackerel (CM) is not consistent whereby the content reduced significantly with the increase of storage time up to 4.5 months. The content was reduced further in home-cooked and fresh-cooked mackerel. However, for canned samples stored for 3.5 years, the omega-3 content is significantly higher in comparison to other samples. The reduction of omega-3 content in CM might be attributable to fish tissue cells lysis during salting, followed by oxidation of polyunsaturated fatty acids into free fatty acids, aldehydes, peroxides, and ketones during heat treatment (Al-Reza *et al.*, 2015). Thermal degradation during cooking might also have contributed to the omega-3 reduction in fresh-cooked and home-cooked mackerel. The variation in the result could also be due to the variation of fat content in each fish.

In contrast, canned tuna in water (CTF) and canned tuna in mayonnaise (CTM) showed lower omega-3 content compared to home-cooked tuna. This could be due to the variation of fish species. Studies had shown

Table 1. Nutrients composition of canned, fresh cooked and home-cooked food samples

Sample			Storage time	Omega 3 (g/100 g)	Protein (g/100 g)	Fiber (g/100 g)
Sardine in tomato sauce	Canned (Ayam Brand) (CS)	*Accelerated	0 day	5.15±0.78^{hij}	12.50±0.06^{e}	n/a
			1.5 months	5.00±0.00^{hij}	15.20±0.10^{g}	n/a
			3.0 months	5.00±0.00^{hij}	13.10±0.06^{ef}	n/a
			4.5 months	4.65±0.07^{hi}	18.20±0.23^{i}	n/a
		Real-time ageing	3.5 years	5.35±0.07^{ij}	n.d	n.d
		Fresh-cooked		2.00±0.14^{e}	16.70±0.72^{h}	n/a
		Home-cooked		0.75±0.07^{abc}	8.50±0.50^{b}	n/a
Mackerel in tomato sauce	Canned (Ayam Brand) (CM)	Accelerated	0 day	5.40±0.00^{j}	8.50±0.06^{b}	n/a
			1.5 months	3.80±0.00^{g}	13.10±0.06^{ef}	n/a
			3.0 months	3.60±0.00^{g}	9.70±0.06^{c}	n/a
			4.5 months	1.85±0.07^{de}	15.70±0.32^{g}	n/a
		Real-time ageing	3.5 years	4.55±1.20^{h}	n.d	n.d
		Fresh-cooked		2.70±0.14^{f}	17.70±0.40^{i}	n/a
		Home-cooked		0.60±0.00^{abc}	10.70±0.91^{d}	n/a
Tuna in mayonnaise	Canned (Ayam Brand) (CTM)	Accelerated	0 day	0.60±0.00^{abc}	13.20±0.06^{f}	n/a
			1.5 months	0.85±0.07^{bc}	12.90±0.06^{ef}	n/a
			3.0 months	0.90±0.28^{bc}	13.00±0.12^{ef}	n/a
			4.5 months	0.75±0.07^{abc}	13.50±0.21^{f}	n/a
		Home-cooked		1.30±0.00^{cd}	15.40±0.75^{g}	n/a
Tuna flakes in water	Canned (Ayam Brand) (CTF)	Accelerated	0 day	0.40±0.00^{ab}	25.10±0.06^{k}	n/a
			1.5 months	0.08±0.01^{a}	23.80±0.25^{j}	n/a
			3.0 months	0.07±0.00^{a}	24.60±0.15^{k}	n/a
			4.5 months	0.30±0.00^{ab}	23.20±0.67^{j}	n/a
Baked beans in tomato sauce	Canned (Ayam Brand) (CB)	Accelerated	0 day	n/a	3.50±0.10^{a}	5.60±0.10^{a}
			1.5 months	n/a	3.50±0.10^{a}	5.60±0.10^{a}
			3.0 months	n/a	3.60±0.10^{a}	5.60±0.10^{a}
			4.5 months	n/a	3.90±0.15^{a}	5.53±0.25^{a}

CS: Canned sardines in tomato sauce, CM: Canned mackerel in tomato sauce, CTF: Canned tuna flakes in water, CTM: Canned tuna in mayonnaise, CB: Canned baked beans in tomato sauce, FS: Fresh cooked sardines in tomato sauce, FM: Fresh cooked mackerel in tomato sauce, FTM: Fresh cooked tuna in mayonnaise, HS: Home-cooked sardines in tomato sauce, HM: Home-cooked mackerel in tomato sauce, n/a: Not available, n.d: Not determined. Values are presented as mean±standard deviations. Values with different superscript within the same column are significantly different at $p<0.05$ as determined by Duncan's Test.
*Accelerated shelf-life study was carried out on five canned food products only.

that species, age, sex, catching season and the fishing area could affect the nutritional composition of fishes. The majority of the commercially available canned marine products contain 0.5-3.0 g/100 g of omega-3 fatty acids (Singer *et al.*, 2016). The daily limit of omega-3 fatty acids ranges from a minimum of 100 mg/day to the upper limit of 5g/day for dietary claims (Singer *et al.*, 2016) and all or canned fish meet the minimum omega-3 fatty acids required for the dietary claims.

3.1.2 Crude protein

Seafood included fish is well known as a good source of high nutritional quality proteins. Canning commonly plays a vital role to preserve fish. In this study, canned, fresh cooked and home-cooked fishes were examined to study the effect of the canning process on their protein content. Canned sardine and mackerel showed significantly lower protein content throughout the storage period compared to freshly cooked samples, respectively. This could be attributed to the heating effect during canning, which weakened lipid-protein bonds and caused the protein to undergo hydrolyzation (Castrillón *et al.*, 1996). The cell membrane protein tends to denature under high temperatures of the cooking and retorting process resulted in the degradation of the initial crude protein to more volatile products. The leaching of some extractable soluble protein fragments from the fish muscle immersed in a boiling cooking medium could be contributed to the loss of protein (Al-Reza *et al.*, 2015). These denatured proteins are readily reacting with other compositions such as oxidized lipids, which leads to loss of protein content after the multistep

canning process (Aubourg, 2001).

Minor changes were observed in the protein content of canned sardine, canned mackerel, canned tuna in mayonnaise, canned tuna in water, and canned beans throughout the storage periods. These results indicated that the canning process did not affect the protein content of the samples over time. Our results are in accordance with Farinde *et al.* (2017) who demonstrated that the protein content in both steamed and baked lima beans was similar to the protein content of canned pinto beans. The protein content of fresh-cooked mackerel and sardine (21.6% and 18.1% (wet weight basis), respectively) reported by Bahurmiz *et al.* (2017) was comparable to the protein content in canned tuna in brine, canned tuna in sunflower oil, and canned sardines in olive oil (26.1, 25.0, and 21.7 g/100 g, respectively) observed by Mesías *et al.* (2015). These results indicated that the canning process was able to preserve the protein content during storage. The protein content of canned mackerels (CM) that were stored for 3 months was lower than CM that stored for 1.5 months. These increments of protein content could be due to the canned fish may gain protein by dilution of components or by absorption of material from the cooking medium of the canned food throughout the storage periods (Castrillón *et al.*, 1996).

3.1.3 Total dietary fibres

The dietary fibre is vital for maintaining human health because it turns into a substrate for colonic flora. The gut bacteria could ferment the fibre and result in the short-chain fatty acids production that could maintain colonic metabolism and integrity, prevent colon carcinogenesis, as well as facilitate glucose and cholesterol metabolism (Pedrosa *et al.*, 2015; Samaan, 2017). According to Table 1, dietary fibre was not detected in all samples except for canned baked beans in tomato sauce (CB). As we know, fish products do not have dietary fibre. However, the tomato sauce used to prepare canned fish does. Nevertheless, no dietary fibre was observed in our canned fish products in tomato sauce, which might have been lost during processing (Rickman *et al.*, 2007). In this study, canned baked beans in tomato sauce (CB) had a total dietary fibre of 5.60 g/100 g which is similar to the total dietary fibre of baked beans in tomato sauce reported by Aldwairji *et al.* (2014) which is approximately 5.96 g/100 g. Results also indicated that storage time did not affect the total dietary fibre content of CB.

3.1.4 Calcium

Calcium is an essential mineral present in the human body that provides the rigidity of bones, a vital function in nerve impulses, and muscle contractions. Normally, the average recommended nutrient intake of calcium is about 800-1000 mg/day for adults depending on the country of origin. According to Nurnadia *et al.* (2013), the calcium content in fish samples collected from the west coast of Peninsular Malaysia was ranging from 12.89 to 127.59 mg/100 g which is still safe under the permissible limit. From Figure 1, canned mackerel (CM) and canned sardines (CS) showed significantly (approximately 10-15-fold) higher calcium content compared to their fresh and home-cooked counterparts. This might be due to the effects of heating during the retort process that caused the fish bones to soften, which broke down the complex organic molecules in the fish bones and released the mineral elements (Rickman *et al.*, 2007). Our results are in consistent with Heaney *et al.* (1990), where higher calcium contents were only noticed in canned seafood with bones. Home-cooked tuna in mayonnaise (FTM) showed no significant changes with canned tuna in mayonnaise and canned tuna in water throughout the storage periods.

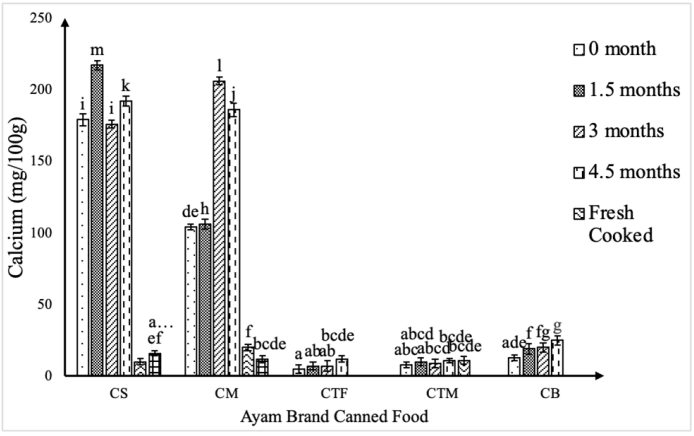

Figure 1. Calcium composition of canned, fresh cooked and home-cooked food samples. CS: Canned sardines in tomato sauce, CM: Canned mackerel in tomato sauce, CTF: Canned tuna flakes in water, CTM: Canned tuna in mayonnaise, CB: Canned baked beans in tomato sauce, FS: Fresh cooked sardines in tomato sauce, FM: Fresh cooked mackerel in tomato sauce, FTM: Fresh cooked tuna in mayonnaise, HS: Home-cooked sardines in tomato sauce, HM: Home-cooked mackerel in tomato sauce. Values are presented as mean±standard deviations, n = 3. Bars with different notations are significantly different at $p<0.05$ as determined by Duncan's Test.

Throughout the storage periods (0 to 4.5 months), the calcium content in canned sardine (CS) and canned mackerel (CM) increased significantly. As discussed earlier, this could be due to the high temperature used during processing and the long storage period, which softened the bones and increased the calcium content. In addition, the filled oil seeped into the fish and salt absorption over the storage periods resulted in increased calcium content (Castrillón *et al.*, 1996). Furthermore,

these results are in accordance with Lukoshkina and Odoeva (2003), which showed that the absorption of the salt added into the cooking medium by the fish muscle during canning and storage periods could increase the ash content. No significant changes in calcium content were noticed in all canned tuna in mayonnaise (CTM), canned tuna flakes (CTF), and canned baked beans during the storage period. These results are in agreement with Bushway *et al.* (1985), Elkins (1979), and Rickman *et al.* (2007) who reported that minerals (except for iron and copper) in canned foods were unaffected during storage. Vafaei *et al.* (2019) also reported that no changes in the calcium content of canned silver carp after one year of storage as compared to fresh cans.

3.2 Chemical preservatives composition (benzoic acid, sorbic acid and sulphur dioxide)

Ready-to-eat fresh food products pose food safety and quality challenges for food distributors. The challenges can be solved by introducing artificial preservatives that help to prolong the shelf-life of food products. However, these preservatives come with detrimental side effects (Mirza *et al.*, 2017). In food canning, food preservatives such as benzoic acid, sorbic acid, and sulphur dioxide are also commonly added to control the growth of microorganisms (Perito *et al*., 2020). Despite the benefits and extensive usage, these preservatives were reported to have adverse effects on human health (Silva and Lidon, 2016).

Table 2. Chemical preservatives (benzoic acid, sorbic acid and sulphur dioxide) content of canned and fresh cooked food samples

Sample	Benzoic Acid (mg/kg)	Sorbic Acid (mg/kg)	Sulphur Dioxide (mg/kg)
Canned Sardines in Tomato Sauce (CS)	n/a	n/a	n/a
Canned Mackerel in Tomato Sauce (CM)	n/a	n/a	n/a
Canned Tuna Flakes in Water (CTF)	n/a	n/a	n/a
Canned Tuna in Mayonnaise (CTM)	n/a	n/a	n/a
Canned Baked Beans in Tomato Sauce (CB)	n/a	n/a	n/a
Fresh Cooked Sardines in Tomato Sauce (FS)	n/a	n/a	n/a
Fresh Cooked Mackerel in Tomato Sauce (FM)	n/a	n/a	n/a
Fresh Cooked Tuna in Mayonnaise (FTM)	n/a	n/a	n/a

CS: Canned sardines in tomato sauce, CM: Canned mackerel in tomato sauce, CTF: Canned tuna flakes in water, CTM: Canned tuna in mayonnaise, CB: Canned baked beans in tomato sauce, FS: Fresh cooked sardines in tomato sauce, FM: Fresh cooked mackerel in tomato sauce, FTM: Fresh cooked tuna in mayonnaise, HS: Home-cooked sardines in tomato sauce, HM: Home-cooked mackerel in tomato sauce, n/a: not available. Values are presented as mean±standard deviations, n = 3. Bars with different notations are significantly different at $p<0.05$ as determined by Duncan's Test.

Based on the result in Table 2, no chemical preservatives were detected in all the canned products. This indicated that the heat treatment during the canning process was sufficient to kill and prevent the growth of undesirable microorganisms, moulds, yeast, and bacteria, and thereby, the addition of chemical preservatives such as benzoic acid, sorbic acid and sulphur dioxide was not required. Therefore, the tested canned products are safe to consume as they are free from chemical preservatives and fulfil the standard requirement mentioned in Food Regulations 1985.

3.3 Addition of MSG: glutamic acid content

According to Food and Drug Administration (2012), MSG is recognized as safe (GRAS) when the daily intake for both free and protein-bound is not exceeded the proposed value of 13 g/d. Research showed that patients who suffer from allergies or severe asthma may have more severe asthma attacks after consuming MSG (Tuormaa, 1994; Populin *et al.*, 2007). There has been increased consumer sentiment in acquiring more natural-tasting food and avoiding additional MSG intake in the past few years. Some countries such as the United States has the label "No MSG" or "MSG free" on the packaging of many processed foods in their market (Kobayashi *et al.*, 2019). Since avoidance is the only treatment, required by FDA, MSG must be listed on the label if it is added. Some of the raw ingredients of processed foods may also contain free glutamic acid, which may naturally be a presence in food such as mushrooms, tomatoes and cheese (Yamaguchi and Ninomiya, 2000).

According to Kobayashi *et al.* (2019), laboratory tests are not able to differentiate between naturally occurring glutamates and/or glutamic acid from added MSG. Hence, in this study, all canned food samples and their respective fresh cooked food samples were examined for their glutamic acid content to show no MSG was added in canned food samples. Results in Figure 2 show that the glutamic acid contents of all food samples ranged from 0.01 mg/g to 0.37 mg/g. This was in accordance with the study reported by Kobayashi *et al.* (2019) who showed that canned fish contained 0.01 to 0.30 g/100 g natural glutamic acid. Populin *et al.* (2007) reported that the natural glutamic acid contents of food without the addition of MSG were between 0.003 mg/g to 1.29 mg/g. In addition, the higher amount of glutamic acid in CS compared to FS might be due to the

processing of tomato sauce (Jongen, 2002; Amerine *et al.*, 2013).

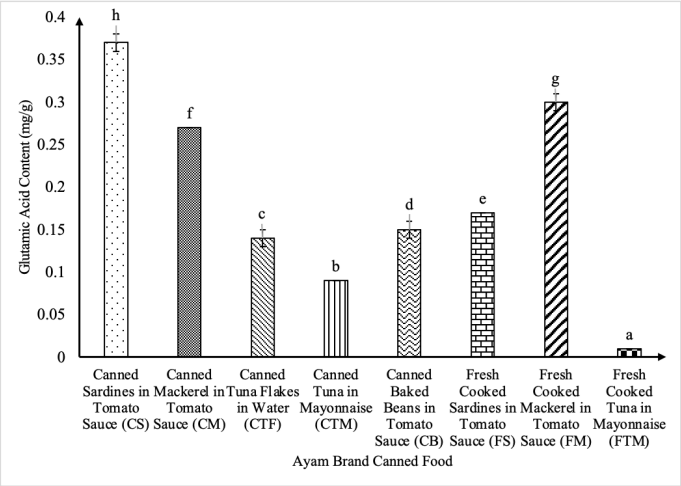

Figure 2. Glutamic acid content of canned and fresh cooked food samples. CS: Canned sardines in tomato sauce; CM: Canned mackerel in tomato sauce; CTF: Canned tuna flakes in water; CTM: Canned tuna in mayonnaise; CB: Canned baked beans in tomato sauce; FS: Fresh cooked sardines in tomato sauce; FM: Fresh cooked mackerel in tomato sauce; FTM: Fresh cooked tuna in mayonnaise; HS: Home-cooked sardines in tomato sauce, HM: Home-cooked mackerel in tomato sauce. Values are presented as mean±standard deviations, n = 3. Bars with different notations are significantly different at $p<0.05$ as determined by Duncan's Test.

According to Jongen (2002), the processing of tomato sauce at the temperature of 104°C for 20 mins will cause an increment in the free amino acids such as glutamic acids due to denaturation and partial hydrolysis of protein (El-Miladi *et al.*, 1969). Tomato is one of the foods that contain large amounts of glutamic acid which is influenced by the ripening of tomato during maturation then contributed to the increase in flavour (Populin *et al.*, 2007). Furthermore, the different amounts of glutamic acid between fresh cooked and canned food samples might be due to the raw materials: the fishes (sardines, mackerel, and tuna). The variation of amino acids in fish such as aspartic acid glycine and glutamic acids is influenced by the fish species, geographical regions, age and diet (Mohanty *et al.*, 2019). All three types of fishes contained glutamic acids in nature, but with varying amounts of glutamic acids (Peng *et al.*, 2013). Hence, artificial MSG was not present in all canned food samples. In conclusion, the results in this study showed no significant differences between the canned food products and the fresh-cooked or home-cooked foods. It is considered that canned foods, representing the fresh-cooked or home-cooked foods, furnish a particular means of delivering easily spoiled foods with their intrinsic nutritional qualities efficiently preserved.

4. Conclusion

In conclusion, Ayam Brand™ canned product maintains nutritional values throughout the storage period (omega 3, calcium, protein, dietary fibre), comparable to fresh-cooked and home- cooked products. The calcium compositions of canned sardines and mackerels are 10-fold higher than fresh and home-cooked contributed by the soft and brittles bones which are enriched with calcium. Storage study showed a promising result, whereby nutrient compositions was consistently maintained throughout the span of 3 years period. None of the Ayam Brand™ canned products contained chemical preservatives such as benzoic acid, sorbic acid, and sulphur dioxide. No MSG was detected in all Ayam Brand™ canned products. Only natural glutamic acid was found in all food samples which may be contributed by the raw materials of fish and tomato sauces themselves. Therefore, canned products were safe to be consumed by the consumer with the nutrient composition more than the fresh and home-cooked style.

Conflict of interest

The authors declare no conflict of interest.

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