

Quality of Sardine (*Sardinella* sp.) *Peda* during Storage as Affected by Salting Pre-Treatment Process and Chitosan

Normalina Arpi^{a,c*}, Murna Muzaifa^{a,c}, Hasanuddin^b, Mira Intania^a

^a Department of Agricultural Product Technology, Faculty of Agriculture, Universitas Syiah Kuala, Banda Aceh, 23111, Indonesia

^b Department of Plant Protection, Faculty of Agriculture, Universitas Syiah Kuala, Banda Aceh, 23111, Indonesia

^c Halal Research Center, Universitas Syiah Kuala, Darussalam, Banda Aceh, 23111, Indonesia

Corresponding author: *normalina.arpi@unsyiah.ac.id

Abstract— *Peda* is an Indonesian fermented whole fish prepared by adding salt before the fermentation and drying process. The storage of fermented fish products will lead to a change in the quality due to microorganism growth. This research aimed to determine the chemical, microbial and sensory qualities of *peda* sardines during four months of storage as affected by the salting pre-treatment process and the use of chitosan. The treatment consisted of P0K1=fish added 20% salt for 6 hours then washed + no chitosan added; P0K2=fish added 20% salt for 6 hours then washed + 2% chitosan; P1K1=fish soaked in freshwater for 6 hours + no chitosan; P1K2=fish soaked in fresh water for 6 hours + 2% chitosan; P2K1=fish left at ambient temperature for 6 hours + no chitosan; P2K2=fish left at ambient temperature for 6 hours + 2% chitosan. The *peda* salt content were 10.52-12.54% after fermentation (0 months) and increased significantly ($P < 0.05$) to 14.11-18.51% after four months, lactic acid bacteria count were 10.47–11.37 log cfu/g (0 month) and increased significantly ($P < 0.05$) to 13.15–14.26 log cfu/g (4 months). The treatment of P1K2 was able to preserve a strong *peda* aroma until 4 months. A total of 10 volatile components in *peda* with the highest peak were identified and grouped as alcohol, ketone, alkane, and carboxylic acid. Undecanone was the most prominent aroma compound in sardines *peda*, with a total area of 6.23%. The treatment of P1K2 resulted in the overall best quality of *peda*.

Keywords— Chitosan; fermented fish; *peda*; pre-treatment; undecanone.

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I. INTRODUCTION

Fish is an important food group for providing adequate nutrition for humans. The variety of nutrients in fish makes fish an important nutrient source that is easily accessible worldwide [1]. Both fresh and processed, Fisheries products are in high demand in markets, but tend to deteriorate in storage. Therefore, it is necessary to process or preserve these products to maintain the quality of the fish, increase economic value, and provide variation and other alternatives for the product's uses. Fish preservation includes a simple chilling, salting, smoking, fermenting, drying, using natural or synthetic additives, and canning. Some of the most common ways fish are preserved in Indonesia include smoked fish, salted fish, dried fish, and other fermentation products, namely *peda*, fish sauce, and shrimp paste [2], [3].

Peda is one of Indonesia's traditional fermented fish, which is very popular on the island of Java. This product is fermented traditionally by using salt, which is pink-colored

and has a distinctive aroma. It has a gritty texture that most consumers prefer. The salt concentration used in the process reaches 15–30% (w/w). The concentration of salt in fermented *peda* fish greatly affects the quality of *peda* fish. Salt will affect the type of microbes that are active during the fermentation process. The fermentation occurs spontaneously and the high salt concentration allows for the desired fermentative microorganism to thrive and inhibits spoilage and pathogen microorganism growth [4]–[6].

However, the storage of fish fermented products for a long time will lead to changes in the quality due to the growth of microorganisms, and this unwanted process will occur more easily if handled inappropriately [7]. The deterioration will be visible in the texture and aroma produced. *Peda* stored for more than three months will produce white spots due to the growth of halophilic bacteria [8]. Therefore, it is necessary to find a way to delay the deterioration.

Chitosan has been proposed as natural preservative in keeping the quality and extending the shelf life of fish products [9]–[11]. Chitosan is a linear polysaccharide

consisting of β -(1 \rightarrow 4)-linked glucosamine and N-acetyl-D-glucosamine that is obtained from deacetylation of chitin. Chitosan is present in the exoskeleton of crustaceans and insects and in the cell walls of most fungi and some algae [12], [13]. Due to its several unique properties, chitosan has been extensively studied for applications in many fields. The broad antimicrobial activity of chitosan against fungi and bacteria has been reported in many studies [12]–[14]. However, the effectiveness is highly dependent on the type of target microorganism [14]. *Peda* is a different product from previous studies and is thought to have a different consortium of microorganisms. The effectiveness of using chitosan as a preservative on *peda* storage is not yet known. This study aimed to determine the effect of salt pre-treatment and the addition of chitosan on the chemical, microbial, and sensory qualities of *peda* during four months of storage.

II. MATERIALS AND METHOD

A. Sardine samples

Sardines (*Sardinella* sp.) weighing 160-165 g/fish, and measuring 25-28 cm in length were acquired from TPI (fish auction site) Lampulo in Banda Aceh, Indonesia. The sardines were immediately put in an icebox and taken to the laboratory within 30 minutes for *peda* production.

B. Sardine preparation and *peda* treatment

The sardines were eviscerated, washed, divided into three groups, and prepared for pre-treatment (P). Three different types of pre-treatment were carried out, P0=fish was added 20% salt for 6 hours then washed; P1=fish soaked in fresh water for 6 hours, and P2=fish were left at ambient temperature ($27\pm 28^\circ\text{C}$) for 6 hours (these fish were left in the basin and covered with a cloth). After that, 20% salt was added to P0, P1, and P2. The addition of salt was done by sprinkling salt onto fish, which were arranged layer by layer, then the containers were covered with a cloth and the fish fermented for 2 days.

After fermentation, the liquid formed was drained and the fish were sundried for 4 hours to deplete the remaining water. Furthermore, the *peda* fish were dipped in a chitosan solution

(K) for 3 minutes and dried again for 1 hour. Chitosan solution is made by dissolving 2% chitosan in 1% acetic acid [15]. Table 1 shows sardine *peda* treatments. The *peda* were then wrapped in opaque paper and stored at ambient temperature for 0, 2, and 4 months.

TABLE I
SARDINE *PEDA* TREATMENTS (PK)

P0K1	fish added 20% salt for 6 hours then washed + no chitosan
P0K2	fish added 20% salt for 6 hours then washed +2% chitosan
P1K1	fish soaked in fresh water for 6 hours + no chitosan
P1K2	fish soaked in fresh water for 6 hours + 2% chitosan
P2K1	fish left at ambient temperature for 6 hours + no chitosan
P2K2	fish left at ambient temperature for 6 hours + 2% chitosan

Note: S=Storage time at ambient temperature, S0=0 month, S1=2 months, S3=4 months

C. Product analysis

The analysis carried out on the *peda* is a chemical, microbiological, and sensory analysis. These analysis carried out included salt content, moisture content, crude protein, and total lipid content [16], total bacteria and lactic acid bacteria enumeration [17], pH and total titratable acidity [16], total volatile base (TVB) [18], aroma identification with GCMS [19] and descriptive sensory test [20].

D. Data analysis

The data obtained were expressed as mean \pm standard deviation, and analyzed by using SPSS statistical program (version 22.0) for Windows. Data were subjected to analysis of variance (ANOVA). Significant differences among mean value were determined with Least Significant Difference (LSD) at level $P < 0.05$.

III. RESULTS AND DISCUSSION

A. Chemical characteristics of sardine *peda*

The chemical analysis results of fermented sardine *peda* are summarized in Table 2.

TABLE II
CHEMICAL CHARACTERISTICS OF SARDINE *PEDA*

Parameter (%)	Storage time (mo)	Treatments*					
		P0K1	P0K2	P1K1	P1K2	P2K1	P2K2
NaCl	0	11.86 \pm 0.35 ^{aA}	12.54 \pm 0.28 ^{aA}	10.52 \pm 0.72 ^{aA}	10.71 \pm 0.92 ^{aA}	11.73 \pm 0.13 ^{aA}	11.10 \pm 0.33 ^{aA}
	2	14.98 \pm 0.37 ^{bA}	14.94 \pm 0.83 ^{bA}	12.34 \pm 0.1 ^{aA}	13.61 \pm 0.30 ^{bA}	13.27 \pm 0.69 ^{abA}	12.95 \pm 0.02 ^{aA}
	4	16.10 \pm 0.05 ^{cA}	18.51 \pm 0.62 ^{cB}	17.73 \pm 3.1 ^{bB}	14.11 \pm 1.8 ^{bA}	15.59 \pm 0.18 ^{bA}	16.96 \pm 1.39 ^{bB}
MC	0	56.19 \pm 0.79	54.29 \pm 0.05	61.10 \pm 0.79	57.98 \pm 0.45	59.02 \pm 1.48	55.40 \pm 0.08
	2	50.58 \pm 1.23	48.64 \pm 0.32	50.14 \pm 0.71	49.09 \pm 0.78	49.91 \pm 0.34	47.58 \pm 1.00
	4	47.82 \pm 0.53	44.72 \pm 1.93	45.54 \pm 0.97	44.69 \pm 0.11	46.76 \pm 1.58	43.89 \pm 1.13
CP	0	26.24 \pm 0.03	24.38 \pm 0.95	23.68 \pm 1.92	25.27 \pm 0.16	23.41 \pm 0.51	24.58 \pm 0.72
	2	27.25 \pm 0.22	27.91 \pm 1.54	25.83 \pm 1.52	29.09 \pm 4.26	28.03 \pm 0.30	30.49 \pm 1.61
	4	27.86 \pm 0.33	29.79 \pm 0.07	29.20 \pm 1.64	31.88 \pm 1.73	30.18 \pm 0.40	31.46 \pm 1.85
TL	0	1.91 \pm 0.62	2.94 \pm 0.92	3.07 \pm 0.26	2.54 \pm 0.01	2.41 \pm 0.21	1.40 \pm 0.19
	2	2.33 \pm 0.62	2.41 \pm 0.23	3.08 \pm 0.47	2.58 \pm 0.74	2.65 \pm 0.04	2.19 \pm 0.61
	4	2.88 \pm 0.62	2.67 \pm 0.54	3.78 \pm 0.78	2.27 \pm 0.35	2.99 \pm 0.78	2.47 \pm 0.35

Note: * See Table 1 for Treatments. MC = Moisture content, CP = Crude protein content, TL = Total lipid content. Same superscript lowercase letters (a-c) in a column indicate no significant differences in the column ($P > 0.05$) and same superscript capital letters (A-B) in a row indicate no significant differences in the row ($P > 0.05$).

The salt content of the sardine *peda* varied from 10.52–18.51%. *Peda* moisture content ranged from 43.89–61.10%, whereas the crude protein and total lipid varied from 23.41–31.88%, and 1.40–3.78%, respectively. There was no interaction effect between treatments (P0K1, P0K2, P1K1, P1K2, P2K1, and P2K2) and storage time (0, 2, and 4 months) on the moisture, crude protein, and total lipid contents. The *peda* salt content was lower than those of *peda* reported by Thoriq [21], *hout-kasef* and *lanhouin*, types of fermented fish published by Gassem [22] and Kindossi [23]. However, the *peda* salt content conformed to that of *peda* [24] and *lanhouin* [25]. Sardine *peda* moisture contents, however, were higher than those reported for *peda* [24] and fermented fish from India, *telesech* [26] and *shidal* [27], and slightly lower than that of *lanhouin* [25]. It was proposed that salt and moisture contents differences of diverse fermented fish products could be the result of variations in the drying process, and the amount and quality of salt used during processing [25]. It is recommended to manage the moisture of fermented fish at 50–70%, for a longer shelf life [28].

Sardine *peda* crude protein contents were comparable to those of India's fermented fish reported by Roy [26] and Gupta [29], but much lower than that of the Africa's *lanhouin* (around 50 g/100g DM) [23]. Total lipid content of the sardine *peda* were lower in comparison to 11–16% lipid content of fermented fish reported by Kindossi [23], Roy [26] and Gupta [29]. During fish fermentation, complex biochemical reactions increase which extensively change the initial attributes of the fish tissue. Proteolysis or protein degradation, lipolysis, and lipid oxidation are some of important biochemical changes [28]. The variation in chemical characteristics of various fish fermented products could be associated with the fish species, environmental conditions, salt content, and different processing technologies [22].

B. Total bacteria and lactic acid bacteria (LAB)

Peda storage for 4 months showed a change in total bacteria and lactic acid bacteria. Figure 1 illustrates the change in total bacteria, which ranged from 9.8–10.33 log cfu/g at the start of *peda* storage, increased significantly ($P < 0.05$) to 11.62–12.47 log cfu/g after 2 months, and increased again to 13.29–14.08 log cfu/g after 4 months. The total bacteria of the sardine *peda* was higher than those of most reported fermented fish which ranged from 3–9 log cfu/g [23], [26], [29] [30], [31]. These reported low total bacteria amounts were mostly from fresh fermented fish products that had not been stored for a long time. Microbial count in fermented foods depends on many factors, such as type of raw materials, duration of exposure to open atmosphere before fermentation, mode of fermentation, type of packaging material and condition, duration and condition of storage [29], [32].

The high total bacteria count of sardine *peda* might be associated with high LAB count which varied from 10.47–11.37 log cfu/g after fermentation (0 months) to 13.15–14.26 log cfu/g after 4 months of storage, although *peda* of all treatments at the 4 months storage time had the highest NaCl content, up to 18.51%. It is suggested that some LAB genera are halophilic, which withstand high NaCl concentrations, including halophilic lactococci, such as *Tetragenococcus* and

Vagococcus, which have been isolated from different fermented fish [28], [32].

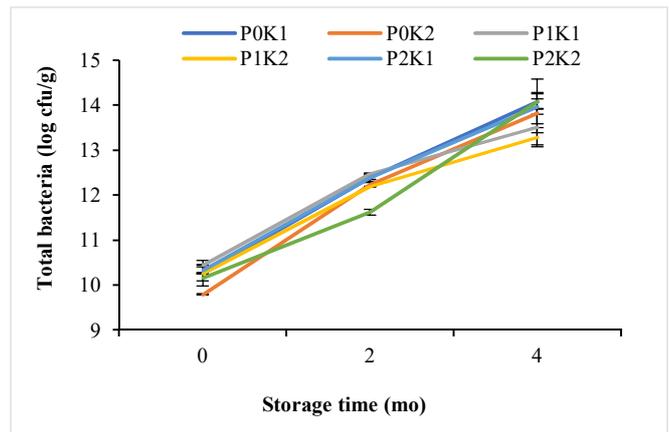


Fig. 1 Total bacteria of sardine during storage

The trend of increasing total bacteria in all treatments during storage was relatively similar, except for treatments dipped in chitosan (P0K2, P1K2, and P2K2), regardless of the pre-treatment. Total bacteria and the significant ($P < 0.05$) increase of bacteria count during 2 and 4 months of storage in these treatments, was relatively lower than the others. On the contrary, the addition of chitosan to the *peda* (P0K2, P1K2, and P2K2) seemed to affect the LAB count, and the increase of LAB count during storage higher than that without chitosan. This shows that the antibacterial properties of chitosan have a different effect on the Gram-negative and Gram-positive groups. Two studies have stated that the antibacterial capacity of chitosan is limited to Gram-negative and less effective in the group of Gram-positive bacteria [33], [34].

The increase of total bacteria count during storage could be facilitated by the availability of nutrients, such as free amino acids and other soluble non-nitrogenous substances derived from proteolysis occurring during fish fermentation and storage. However, spoilage microorganisms are inhibited by salt at levels higher than 6–8% [32]. NaCl in the sardine *peda* samples were around 10–18%. Salt engages an important role in preservation by constraining the growth of putrefactive and pathogenic microorganisms. It is suggested that the dominant microorganisms in the fermented fish products appear not to be those putrefactive or spoilage microorganisms but may be linked to the protein degradation and involve a profound and complex role in developing some aroma and flavor of the fermented products [32]. It has been recorded by Waisundra [35] that LAB and Gram-positive microorganism involved in *peda* fermentation using 1:3 salt-to-fish ratio resulting in *peda* that has a shelf life of 4–6 weeks.

The roles of LAB in fermentation have been reported to repress spoilage microorganism, convert protein, oligopeptide and/or peptide to amino acids, and improve sensory qualities, the aroma, and flavor characteristics [31], [36]. Moreover, Wang [37] indicated that there is a high interaction between microorganisms and unique flavor contributed by esters, acids, ketones, and aldehydes in fermented products. However, Xu [28] stated that the role of microorganisms in the fish fermentation process has not been fully clarified and needs further study.

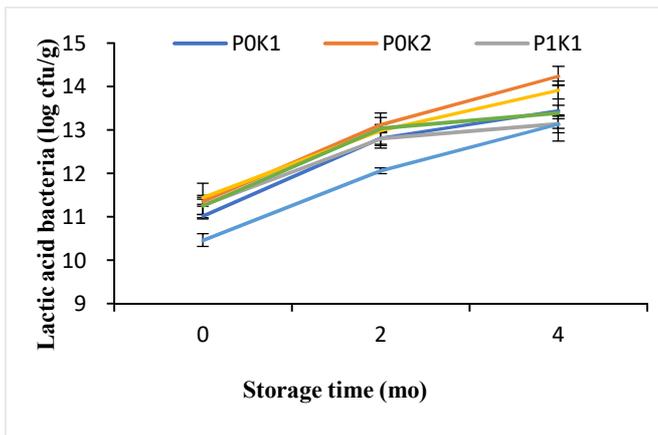


Fig. 2 Total lactic acid bacteria of sardine peda during storage

C. pH and total titratable acidity

The pH and total titratable acidity of sardine *peda* ranged from 5.94–8.18 and 0.23–1.17%, respectively. The pH of *peda* was slightly higher than those pH 6.2–6.5 of fermented fish reported by Gassem [22], Martin [24], Bao [31], Talab [38] but was most comparable to the pH 6.5–7.8 recorded by Kindossi [25], Han [30], Anihouvi [39], and Nicomrat [40]. The total titratable acidity of *peda* were lower than that reported by Nicomrat [40] and higher than those published by Gupta [29]. There was a concurrent significant ($P < 0.05$) increase in pH and total titratable acidity (Fig. 3 and Fig. 4), which has been also reported to develop during the fermentation of other high-protein food [39].

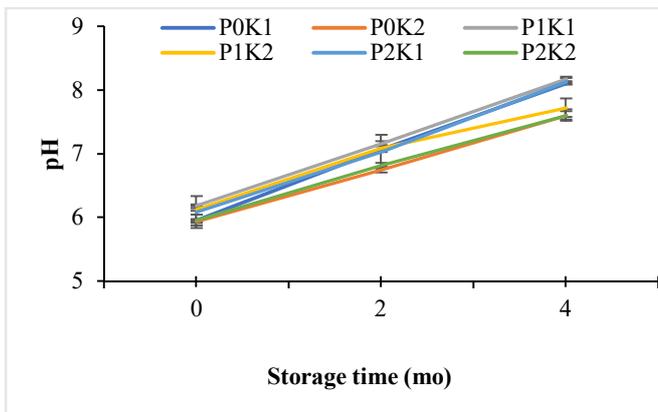


Fig. 3 pH value of sardine peda during storage

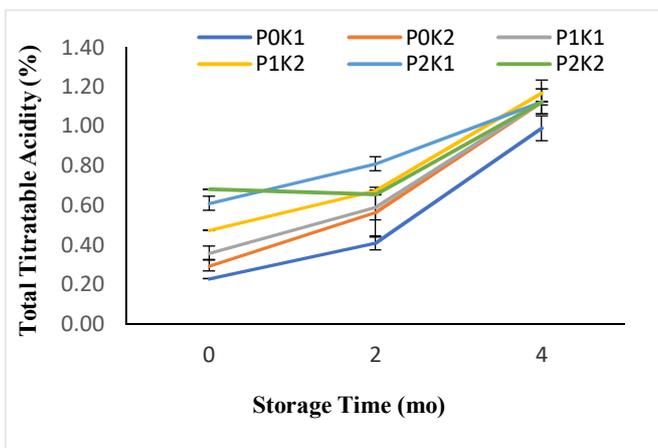


Fig. 4 Total titratable acidity of sardine peda during storage

The initial pH of *peda* after fermentation at 0 months of storage was around a pH of 6 and increased to approximately pH 7 after 2 months of storage and to $\text{pH} \pm 8$ after 4 months. The increase in pH during storage was affected by the continuing enzymatic and microbial activity after processing. The pH rose conceivably due to microbial proteolytic activity during fermentation and the storage resulted in the formation of nitrogenous compounds that accumulate in the fermented product [23], [28]. All sardine *peda* samples with chitosan (K2) had a pH lower than those without chitosan since the beginning and during storage. These *peda* samples with low pH and the changes in pH during storage were related well with total titratable acidity, LAB, and their changes (Fig. 2 and Fig. 4).

The higher pH value in *peda* samples were associated with a lower total titratable acidity. Total titratable acidity and LAB count of *peda* samples dipped in chitosan (K2) were higher along with lower pH compared to those *peda* without chitosan (K1), except for the sample with pre-treatment fish added 20% salt for 6 hours and dipped in chitosan (P0K2) that the total titratable acidity was lower. This is possibly because the P0K2 sardine *peda* had the highest NaCl content of 15.33%. Different salt concentrations affect the content of organic acids, because various enzymes were activated and the activity and type of microbes changed at different salt levels [28].

D. Total Volatile Base Nitrogen (TVBN)

The pre-treatment of the sardines by adding 20% salt for 6 hours before fermentation resulted in low TVBN fresh *peda* between 13.25 mg N/100g (P0K2) and 26.52 mg N/100g (P0K1) for the pre-treatment with 2% chitosan, and the pre-treatment without chitosan added, respectively. However, TVBN of other sardine *peda* samples were between 128.75–584.54 mg N/100g, and after 4 months of storage, it was mostly above 300 mg N/100g. This final TVBN range was higher than that of fresh Indian fermented fish *telesech* published by Roy [26], but lower than that of African fermented fish *lanhouin*, which had a TVBN of 453.6–618.6 mg N/100g [23]. The high TVBN may result from the long exposure of the fish in open conditions, without salt, which makes them prone to enzymatic and microbial activity and produce higher amounts of low molecular nitrogenous products [26]. Autolytic enzymes might have caused degradation of proteins, resulting in the formation of volatile bases. During fish fermentation, salt-soluble and water-soluble proteins degrade into peptides and amino acids by microbial or indigenous proteases, increasing free amino acids and non-protein nitrogen causing the TVBN to increase [28], along with the volatile compounds, which one of the volatile compounds account for specific aroma for the fermented fish [41]. High TVBN content has been associated with a high degree of fermentation that usually has no apparent ammonia-like odor in the fermented product.

The odor of ammonia may probably be masked by the characteristic strong odor of fermented fish [29], as in *peda*, the strong odor is indicated by the increasing sensory *peda* aroma value during *peda* storage and the value becomes the highest after 4 months of storage (Fig. 6c). The allowable range of TVBN in fermented fish product is difficult to

explain because it varies depending on the species, sex, age of the fish, catching region and season of fishing [41].

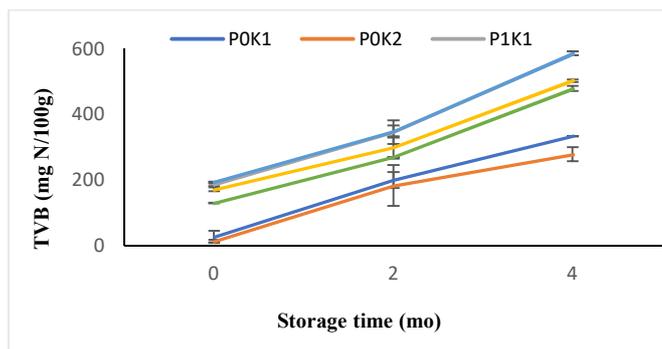


Fig. 5 Total Volatile Base Nitrogen (TVBN) sardine peda during storage

E. Sensory Quality Profile of Peda during Storage

Analysis of the sensory quality of *peda* during storage was carried out by a descriptive test of five sensory components: color, *peda* aroma, gritty texture, *peda* taste and salty taste. Panelists were asked to provide an objective assessment of each sensory component according to its intensity on a scale of 1 to 5 with the following explanation: 1 = very weak, 2 =

weak, 3 = neutral, 4 = strong and 5 = very strong. The sensory profile of the *peda* during storage can be seen in Fig 6.

Figure 6 shows the sensory sardine *peda* at the beginning of storage (0 months) which has a relatively similar color profile, gritty texture, *peda* aroma, *peda* taste and salty taste. All treatments had characteristic color, *peda* aroma, *peda* taste and strong salty taste. The color of the sardine *peda* fish used in this study was a characteristically bright fish. The color of commercial *peda* fish varies depending on the raw material (type of fish) used. *Peda* fish from mackerel produces a pale to red-brown color [42]. The salty taste in this study was one of the dominant sensory characteristics of *peda* fish that stands out, arising from the addition of salt in the manufacturing process [4], [43]–[45]. In addition to the salty taste, "*peda* taste" was also the main characteristic of *peda* fish which refers to its distinctive delicious taste. According to Giyatmi and Irianto [4], the taste of *peda* was slightly sour so that its combination with salty taste produces a distinctive taste of this specific process of producing the fermented fish *peda*. The appearance of the distinctive aroma and taste of fermented fish products indicates that physicochemical changes have occurred in the raw materials [45].

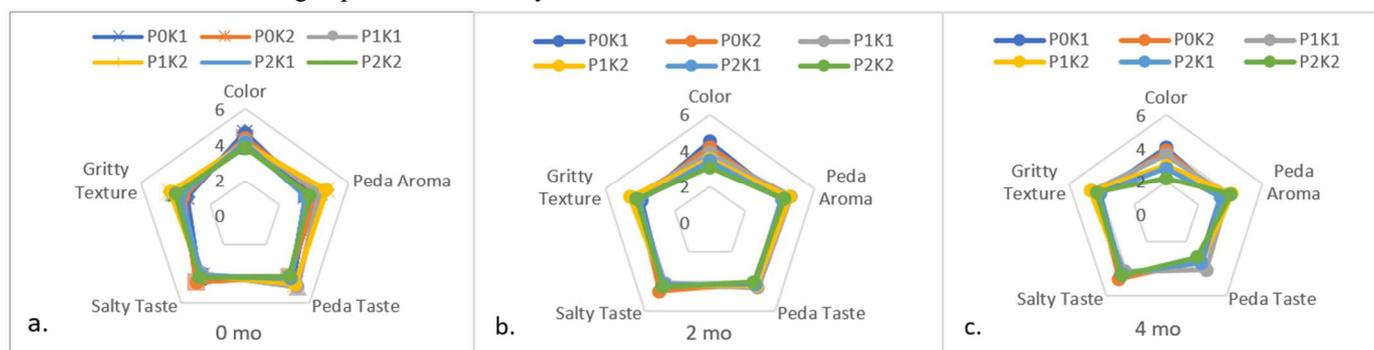


Fig. 6 Descriptive sensory value of *peda* at (a) 0 months, (b) 2 months, and (c) 4 months of storage at ambient temperature

The distinctive taste and aroma arise due to a salting process that selects certain microorganisms so that they can grow. During the fermentation process, there is a degradation of fatty acids and proteins into simpler compounds, which is done by microorganisms and the enzymes they produce which become aroma precursors. The main precursors of volatile compounds are free fatty acids (FFA) produced by lipolytic enzymes and free amino acids (FAA) produced by proteolytic enzymes [45], [46]. Another feature of *peda* is its unusual and peculiar texture, which may best be described gritty. The gritty texture of *peda* fish can be influenced by the concentration of salt used in making *peda* [5], [47]. Several factors can affect the speed of salt penetration into the fish meat, namely the fat content of the fish, thickness of the meat, temperature of the fish, and concentration of the salt solution [48].

After storage for 2 and 4 months, it was generally seen that there was a change in the intensity of the various sensory parameters of the sardine *peda*. Intensity of color and strength of taste consistently decreased significantly ($P < 0.05$) for most of the treatments, and the intensity of the aroma fluctuated after 2 and 4 months of storage, while the gritty texture increased significantly ($P < 0.05$). The treatment of making *peda* without the addition of chitosan (P0K1, P1K1,

P2K1) tends to have a stronger color than with the addition of chitosan (P0K2, P1K2, P2K2).

In terms of aroma, the treatment of making *peda* by soaking fish in fresh water for 6 hours and the adding chitosan (P1K2) was able to maintain a strong aroma of *peda* until it was opened after 4 months of storage. The general decrease in intensity of aroma and taste during storage is thought to be influenced by the activity of microorganisms and the enzymes they produce [45], [46].

Furthermore, on the sensory salty taste and gritty texture, all treatments with added chitosan (P0K1, P1K1, P2K1) had a stronger salty taste and a stronger gritty texture than without the addition of chitosan. The intensity of the salty taste and the feeling of gritty texture tend to increase with increasing storage time. The increasing gritty texture and saltiness during storage is thought to be because the salt molecules have completely absorbed into the fish flesh. The salt in fish meat will increase the saltiness of the pepper, affect the activity of the water, and improve the texture of the *peda*. Lopez-Caballero et al. [49] stated that the activity of texture formation is one of the functional properties of chitosan.

F. Identification of volatile compounds of Sardine Peda

The volatile aroma components of sardine *peda* were analyzed by GC-MS from samples of fresh *peda* fish (without storage) which had strong sensory characteristics and low TVBN, experimental name P1K2. The results of the identification of volatile aroma components found in sardine *peda* is shown in Figure 7.

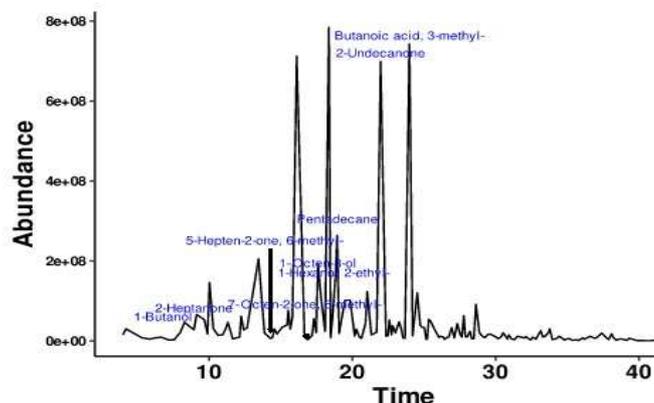


Fig. 7 Volatile aroma compounds of sardine *peda*

The profile of the aroma component of *peda* fish can be established by comparing the unknown MS spectrum with library data, as shown in Table 3. Aroma of *peda* consisting of 10 volatile components with the highest peaks, consisting of 1-Butanol, 2-Heptanone, 5-Hepten-2-one,6-methyl-, 7-Octen-2-one,6-methyl-, 1-Octen-3-ol, 4-Heptanol,2,6-dimethyl-, 1-Hexanol,2-ethyl-, Pentadecane, Undecanone and Butanoic acid,3-methyl-.

TABLE III
VOLATILE COMPOUND OF SARDINE *PEDA*

No	Retention Time	Compounds	% Area	Group
1	8.287	1-Butanol	0.65	Alcohol
2	8.941	2-Heptanone	2.53	Ketone
3	13.864	5-Hepten-2-one,6-methyl-	1.82	Ketone
4	16.653	7-Octen-2-one,6-methyl-	3.54	Ketone
5	17.598	1-Octen-3-ol	5.84	Alcohol
6	18.092	4-Heptanol-2,6-dimethyl-	2.19	Alcohol
7	18.835	1-Hexanol,2-ethyl-	1.62	Alcohol
8	18.942	Pentadecane	4.24	Alkane
9	21.967	2-Undecanone	6.23	Ketone
10	23.978	Butanoic acid,3-methyl-	3.55	Carboxylic Acid

The ten dominant aroma compounds identified in fresh *peda* fish were grouped as alcohol, ketone, alkane and carboxylic acid. Some of these groups are commonly found in fermented fish products such as shrimp paste [50], *miso* [51]; salt-fermented fish and shrimp paste [52] and *lanhouin* [53]. The main flavor compounds of fermented fish products that have been detected using GC-MS are mainly acids, aldehydes, hydrocarbons, alcohols, ketones, esters, nitrogen-containing compounds, and furans [28]. Microbial metabolism was reported to be very important for flavor formation of fermented fish products [54].

In this study, the highest aroma compound in the sardines *peda* was undecanone with a total area of 6.23%. The highest percentage of area compared to other components indicates that this component is a key aroma in the formation of the desired sardine *peda* aroma. Undecanone (which is a ketone

compound), also known as methyl nonyl ketone is the main constituent of *Zanthoxylum armatum* DC essential oil. This compound is applied widely in perfumery, preservation, and pharmacological activity and is the starting material for the synthesis of new molecules. In addition, this compound is known to have good antioxidants, anti-inflammatory properties, and antibacterial activity [55]. Undecanone in fermented *peda* fish is thought to originate from lipid metabolism. Xu et al. [28] reported that lipolysis of triglycerides and phospholipids by microbes and indigenous enzymes result in the development of free fatty acids (FFA). Phospholipid lipolysis contributes greatly to the release of FFA during fish fermentation. FFA does not only directly affect the formation of taste compounds, but is also a methyl ketone precursor, secondary alcohols, esters and lactones. Undecanone was reported as a key compound in *miso*, a Japanese fermented fish product. Giri [51] also reported undecanone to be one of the key compounds in fish oil [56].

The second largest volatile compound detected in *peda* fish after undecanone was 1-octen-3-ol. It belongs to the group of alcohols and is widely present in mushrooms [57]. Generally, alcohol is present as a result of yeast metabolism of sugar. In addition, aroma could also be formed by the decomposition of secondary hydroperoxides fatty acids. It has been identified as a lipid oxidation product in fish oil, which is an important flavor contributor due to its low odor threshold [51], [54]. So far, 1-Octen-3-ol has been reported as the most volatile characteristic of fresh sardines and the aroma of other fatty fish. 1-Octen-3-ol contributes to the grass and mushroom aroma and considered a product of oxidation linoleic acid and arachidonic acid [54], [58]. Several recent studies have also shown that 1-octene-3-ol is also found in Bigeye tuna (*Thunnus obesus*) [59], Shanghai smoked fish [60], fish sauce [61], grass carp [62] and tilapia sausage [63].

IV. CONCLUSION

The quality of sardine *peda* during storage due to various salting and chitosan treatments resulted in varying chemical, microbiological and sensory qualities. The moisture content of all treatments decreased during storage, while the NaCl, protein, total amount of lipid increased. The trend of increasing total bacteria in all treatments during storage was relatively similar, except for treatments dipped in chitosan. The higher pH value of sardine *peda* samples was associated with lower total titratable acidity. Total titratable acidity and LAB count of *peda* samples dipped in chitosan were higher along with lower pH compared to those sardine *peda* without chitosan.

The sensory sardine *peda* at the beginning of storage (fresh sardine *peda*) had a strong characteristic color, *peda* aroma, *peda* taste and salty taste. During storage, color and *peda* taste tended to continue to decrease across all treatments. Aroma sardine *peda* increased after 2 months of storage and decreased again after 4 months, while the salty taste and gritty texture increased through both storage periods. The treatment of making *peda* without the addition of chitosan tended to have a stronger color than with the addition of chitosan. In terms of aroma, soaking fish in fresh water for 6 hours and adding chitosan (P1K2) was able to preserve a strong *peda* aroma for 4 months of storage. A total of 10 volatile components in sardine *peda* with the highest peak were

identified as 1-Butanol, 2-Heptanone, 5-Hepten-2-one, 6-methyl-, 7-Octen-2-one, 6-methyl-, 1-Octen-3-ol, 4-Heptanol-2,6-dimethyl-, 1-Hexanol, 2-ethyl-, Pentadecane, Undecanone and Butanoic acid, 3-methyl-. The aroma compounds were grouped as alcohol, ketone, alkane, and carboxylic acid. Undecanone was the strongest aroma compound in sardines *peda* with a total area of 6.23%. Further analysis is needed to determine the effect of all treatments on the volatile components and other metabolites of *peda* sardines.

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