FOOD

RESEARCH

Physicochemical properties, oxidative stability and sensory acceptance of shortfin scad (*Decapterus macrosoma*) – surimi emulsion sausages

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Abstract

The aim of this study was to identify the physicochemical properties, oxidative stability and sensory acceptability of shortfin scad (*Decapterus macrosoma*) emulsion sausage incorporated with commercial surimi and silver catfish (*Pengasius*) protein hydrolysate. Fish emulsion sausage was prepared with fish mince and surimi ratios as follows: Formulation A (100:0); B (60:40); C (40:60); D (0:100). The proximate compositions of the fish emulsion sausages were determined. An increase in surimi percentage was increased lightness (L*) and decreased redness (a*) values of the sausages significantly (p<0.05). The higher fish mince ratio caused an increase in texture profile and a better-emulsified microstructure with fewer voids compared to the control. The fish protein hydrolysate could help maintain oxidative stability, as it has antioxidant properties. Moreover, the sensory evaluation indicated that all formulations of shortfin scad sausage incorporated with surimi and fish protein hydrolysate can be accepted by consumers, with formulation B had the highest rank as compared to control (100% surimi).

1. Introduction

Emulsion sausages such as frankfurters are popular among both Western and Asian consumers. Emulsion sausage typically has 25-30% fat and produced from beef, pork and chicken (Panpipat and Yongsawatdigul, 2008). The Malaysian market commonly formulates sausage from beef and chicken sources (Huda *et al.,* 2012). Recently, the incorporation of fish mince and surimi in emulsion sausage manufacturing has become widespread, especially for Asian consumers (Panpipat and Yongsawatdigul, 2008). The purpose of adding surimi in sausage manufacturing is to improve texture and decrease costs (Intarasirisawat *et al.,* 2014). Additionally, surimi has the high gel-forming ability, binding and emulsifying properties which make it suitable as a raw material to improve the stability of emulsion products (Intarasirisawat *et al.,* 2014). Fish sausages production from various fish such as Argentine anchovy (*Engraulis anchoita*) (Piotrowics and Mellado, 2015) and Shortfin scad (*Decapterus macrosoma*) (Zakaria and Sarbon, 2018) have been studied.

The measurement of sausages' physicochemical properties is important in determining consumer acceptance. Moisture is the main component in fish sausage, followed by carbohydrate, protein, fat and ash (Santana *et al.,* 2012). According to the Food Act 1985, sausage composition should contain less than 1.7% nitrogen and not more than 30% fat (Ministry of Health Malaysia, 1985). The texture is a vital sensory characteristic that determines the quality and acceptability of fishery products (Dincer and Cakli, 2015). Many studies have been conducted concerning the incorporation of non-meat ingredients to enhance physicochemical and sensory properties to promote a healthier meat sausage, including fish protein hydrolysate (Intarasirisawat *et al.,* 2014; Zakaria and Sarbon, 2018) and ethanolic kiam wood extract (Maqsood *et al.,* 2012).

The oxidative stability of sausage not only plays an important role in determining acceptability but also determines shelf life. The shelf life of sausage products is identified using a microbial growth test, physical tests in terms of colour and texture, and chemical tests through lipid oxidation which occurs during storage (Valencia *et al.,* 2007). Therefore, the oxidative stability and shelf life of sausage products could be improved with the incorporation of natural antioxidant and material

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containing low sources of fat content. The antioxidant properties of fish protein hydrolysate could protect emulsion sausage from lipid oxidation, help maintain the membrane structure of muscle fibres and decrease muscle reduction (Maqsood *et al.,* 2012). Several studies have shown a decrease in PV and TBARS values in sausage with the addition of surimi and protein hydrolysate (Intarasirisawat *et al.,* 2014; Zakaria and Sarbon, 2018).

Fish protein hydrolysate (FPH) is defined by the formation of smaller peptides due to the breakdown of fish protein by enzymatic conversion and consists of 2 to 20 amino acids (Chalamaiah *et al.,* 2012). FPH sources include flesh, skin and waste components (Halim *et al.,* 2016). In addition, FPH functions as an antioxidant, and thus can be used against free radicals and to improve the physicochemical properties and fat absorption, water holding capacity, and emulsifying properties without decreasing nutritional value (Halim *et al.,* 2016; Kang *et al.,* 2018). The addition of FPH enhances structural properties and increases oxidative stability (Ren *et al.,* 2011). Studies on incorporating fish protein hydrolysate in sausages made from Skipjack (*Katsuwonus pelamis*) (Intarasirisawat *et al.,* 2014), Snakehead (*Channa striata*) (Zakaria and Sarbon, 2018) and Channel catfish (*Ictalurus punctatus*) (Ren *et al.,* 2011) have been successfully conducted.

Surimi defined as the concentrated myofibril protein that been extracted from the fish flesh by washing minced meat that has been separated from bones, skin and guts. During the washing process, the fat and any water-soluble content are removed whereas insoluble myofibril protein was isolated (Santana *et al*., 2012). Surimi has suitable properties such as light colour, bland odour, low-fat content and high in myofibrillar proteins. Based on the USDA standards (2017), fish surimi contains nutrition about 76% water, 15% of protein, 6.85% of carbohydrates and 0.9% of fat per 100 g. The functional properties of surimi classified into three major categories which were hydration properties which included solubility and water holding capacity, surface properties and protein-protein interactions (Santana *et al*., 2012).

Shortfin scad is known as 'Selayang', 'Curut' and 'Sardin' in Malaysia. They are a low-value tropical fish that has shown potential in a few studies. Shortfin scad is a good source of protein, vitamin and minerals, making them suitable for use in food products such as *keropok lekor* and fish emulsion sausage (Zakaria and Sarbon, 2018). Ishak and Sarbon (2017) have reported that shortfin scad waste has potential in ACE inhibitory peptide. Additionally, studies have been conducted on the use of shortfin scad as a source of gelatin (Sarbon *et al.,* 2014). The purpose of this study was to identify the physicochemical properties, oxidative stability and sensory acceptability of shortfin scad (*Decapterus macrosoma*) emulsion sausage incorporated with surimi and silver catfish (*Pengasius*) protein hydrolysate.

2. Materials and methods

2.1 Materials

Fresh Silver catfish (*Pengasius sp*) were bought from a local market in Manir, Kuala Terengganu, while Shortfin scad (*Decapterus macrosama*) and surimi were bought from MAPEROW Sdn. Bhd., Terengganu. Silver catfish, shortfin scad mince and surimi were stored in iced condition during transportation to the laboratory. Other ingredients for making sausages, including corn oil, salt, white pepper and sugar, were purchased from a local hypermarket in Kuala Terengganu. All chemicals and enzymes, including Alcalase® 2.4 L, were purchased from Sigma and other reagents used were of analytical grade.

2.2 Sample of preparation

Silver catfish (*Pengasius sp*) were decapitated, eviscerated and washed with excessive water. The bones and skin were removed and the flesh was stored at -40ºC until further use. The flesh was grounded using a food processor (Panasonic, Malaysia) until fine.

2.3 Preparation of silver catfish protein hydrolysate

Silver catfish protein hydrolysate was prepared following a method according to Zakaria and Sarbon (2018). Fish flesh (55 g) was immersed in 33 g of distilled water and pre-incubated in a water bath shaker (Memmert GmbH, Denmark) at 85°C for 20 mins to inactivate the endogenous enzyme in the fish muscle. Then, 20 g of Alcalase was added to the mixture to initiate the hydrolysis reaction. Hydrolysis occurred at 50ºC at pH 7.89 for 84 mins. Next, the mixture was placed into the water bath at 85ºC for 15 mins to terminate the enzymatic reaction before centrifuged (Gyrozen, Korea) at 6000 rpm for 20 mins at 4ºC. The supernatant of hydrolysate was then filtered using filter paper then freeze-dried. The freeze-dried Silver catfish (*Pangasius sp*) protein hydrolysate was subjected to incorporation in the fish sausage in the later stage.

2.4 Development of fish emulsion sausage

Shortfin scad (*Decapterus macrosoma*) sausage incorporated with surimi and silver catfish (*Pengasius*) protein hydrolysate for each formulation was prepared following Zakaria and Sarbon (2018). Firstly, the frozen

flesh of shortfin scad and surimi was thawed overnight in a chiller (4°C). Formulations of fish sausage from shortfin scad with surimi incorporation at different ratios included the following: formulation A (100 fish mince: 0 surimi); B (60 fish mince: 40 surimi); C (40 fish mince: 60 surimi); and D (0 fish mince: 100 surimi), as presented in Table 1. In which, formulation D used as a control in this study. Other ingredients, including vegetable oil (canola oil), were added to the mixture. Then, icy cold water was added during the mixing process to ensure that all ingredients were mixed well to form a homogenized mixture. Next, the isolated soy protein was added and ground for 30 s. Then, the remaining oil was added, followed by corn starch, and was continually homogenized for 30 s. Silver catfish protein hydrolysate and lastly the chilled water was added and further ground for 30 s until the ingredients were homogenous. The mixture was placed into the sausage stuffer and pumped into the cellulose casing. The casing was tied at 10-12 cm for each sausage. The sausages were boiled until cooked. Each sausage was divided into the unit of analysis, then vacuum packed in a polyethylene bag and stored at -4°C for further oxidative stability analysis, while fresh sausages were prepared for proximate, microstructure and sensory analyses.

Table 1. Formulation of shortfin scad (*Decapterus macrosoma*) sausage incorporated with surimi and silver catfish (*Pengasius*) protein hydrolysate

Formulations	A	B	C	D
Ratio	100:0	60:40	40:60	0:100
SC mince: Surimi (%)	63:0	37:26	26:37	0:63
SCPH (%)	3	3	3	3
Vegetable oil (corn oil) (%)	7	7	7	7
Egg white powder (%)	3	3	3	3
Isolated soy protein (%)	2	2	2	2
Sugar (%)	0.5	0.5	0.5	0.5
Salt (%)	1	1	1	1
White pepper (%)	1.3	1.3	1.3	1.3
Garlic (%)	1	1	1	1
Corn flour (%)	5	5	5	5
Sodium trypolyphosphate (%)	0.5	0.5	0.5	0.5
Water + ice (%)	12.7	12.7	12.7	12.7
Total	100	100	100	100

2.5 Physicochemical properties

2.5.1 Proximate analysis

The chemical compositions (moisture, protein, fat and ash) of the fish emulsion sausage were measured following AOAC (2002) methods. Carbohydrates were determined by calculation from the remaining percentage of moisture, fats, protein, ash and fibre. The carbohydrate content was calculated as follows:

Carbohydrate (%) = 100 – [moisture (%) + crude protein (%) + crude fat (%) + ash (%)]

2.5.2 Physical analysis

2.5.2.1 Texture profiles

The texture profiles of the emulsion fish sausages were determined following Intarasirisawat *et al.* (2014) and Ismail *et al.* (2014), using a texture analyser (Stable Micro Systems, Godalming, Surrey, UK) with a load cell of 50 kg. A cylindrical aluminium probe was used. The sausage samples were cut into cylindrical shapes of about 2 cm and placed on the instrument base. The tests were run in two compression cycles. TPA textural parameters were measured at room temperature with the following conditions: pre-test speed of 1.0 mm/s, test speed of 5.0 mm/s, post-test speed of 5.0 mm/s and strain of 50%. Parameters such as hardness, cohesiveness, springiness, chewiness, gumminess and resilience were measured as the force required to compress the sample by a pre-set distance. The texture profiles of each sample at 4ºC were analysed at 0, 6, 12 days. The measurements were conducted in triplicate for each sample.

2.5.3 Colour analysis

Colour analysis of the fish emulsion sausage was conducted following Ismail *et al.* (2014). A colorimeter (Hunter Lab, Model colour Flex, Reston, VIRG, USA) with a port size of 0.50 inch was used to determine the colour of the sausage samples. Sample preparation was conducted by crunching the sausage until it is homogenous and placing the sample into a transparent plastic. The instrument for colour analysis was calibrated using a black and white Minolta calibration plate. The values were reported in terms of the CIE colour profile system, including lightness (L*), redness (a*) and yellowness (b*). The measurements were conducted in triplicate for each sample.

2.5.4 Scanning electron microscopy (SEM)

The microstructure of the shortfin scad (*Decapterus macrosoma*) sausage incorporated with surimi and silver catfish (*Pengasius*) protein hydrolysate was analysed using a Scanning Electron Microscope (SEM) (JEOL JSM -6360LA, Tokyo, Japan) following the method from Zakaria and Sarbon (2018). Firstly, samples with a thickness of 2-3 mm were fixed with 2.5% glutaraldehyde in 0.1 M sodium cacodylate buffer in 2 to 4 hrs and washed with 0.1 M sodium cacodylate buffer at pH 7.2 for 10 min. Then, the samples were post-fixed with 0.1 M sodium cacodylate buffer containing 2% (v/v) osmium tetraoxide for 2 to 4 hrs and underwent rinsing with 0.1 M sodium cacodylate buffer for 10 min. They were then dehydrated in ethanol with serial concentrations from 35 to 100% (v/v). The dried samples were mounted on bronze and sputter-coated with gold (JEOL JFC-1600, Tokyo, Japan) and observed with a

scanning electron microscope at an acceleration voltage of 5 kV.

2.6 Determination of oxidative stability

2.6.1 Peroxide value

The determination of peroxide value was conducted following AOAC (2002). About 5.00 g (±0.05) g of the homogenized sausage was weighed and placed into 250 mL of the conical flask. Approximately 30 mL of the acetic acid-chloroform solution with a ratio 3:2 was added into the conical flask. The flask was swirled until the sausage was completely dissolved. Then, 1 mL of saturated potassium iodide solution was added to the solution. The flask was swirled for exactly one minute and stored in a dark place for about 5 mins. Then, 75 mL of distilled water was added and shaken vigorously to liberate the iodine from the chloroform layer. Next, the solution was titrated with 0.01 N sodium thiosulphate, until the colour changed to light yellow. After that, about 1 mL of 1% (w/v) soluble starch solution as an indicator was added into the mixture, which gives a blue colour. Titration continued until a faint blue colour liberated all the iodine from the chloroform layer. Finally, 0.01 N sodium thiosulphate was added drop-wise until the blue colour disappeared. The volume of titrant used was recorded. The peroxide value was calculated based on the following formula:

$$PV \ (mEqperoxide/kgfat) = \frac{(S - B) \times M \times 1000}{Sample \ Weight \ (g)}$$

Where S = sample titration, B = blank and M = molarity of sodium thiosulphate. Measurements were conducted in triplicate.

2.6.2 Thiobarbituric acid reactive substances (TBARS)

The thiobarbituric acid reactive substance was used following Ismail *et al.* (2014) with slight modification. Firstly, 5 g of sausage sample was weighed and homogenized with 15 mL of distilled water using Polytron homogenizer at 8000 rpm for 10 s. Then, about 1 mL was added with 50 μL of 10% butylated hydroxyanisole, 4 mL of thiobarbituric acid (TBA) and trichloroacetic acid (TCA). Then, the mixture was vortexed and incubated for 15 mins in water bath shaker at boiling temperature to develop colour. The sample was cooled and vortexed again, then centrifuged at 4000 rpm for 12 mins. The absorbance was taken at 531 nm against a blank. The blank contained 2 mL of distilled water and 4 mL of TBA-TCA solution. TBARS were calculated from a standard curve of malondialdehyde (MDA) that was freshly prepared by the acidification of TEP (tetramethoxypropane) in the range of 2 to 10 ppm and expressed as mg of MDA per kg of sample.

2.7 Sensory evaluation

The sensory evaluation method was conducted per Zakaria and Sarbon (2018). The sensory evaluation was conducted by 32 untrained panellists from Universiti Malaysia Terengganu using a 7-point hedonic scale for each sample. The scores ranged from 1 (dislike extremely) to 7 (like extremely). Each sausage was evaluated in terms of the degree of liking for colour, odour, taste, texture and overall acceptance. The sausages were steamed for 20 min, then cut into 3cm long pieces and presented to each panellist. The panellists were instructed to rinse their palate between each sample using water supplied to prevent bias.

2.8 Statistical analysis

All measurement for each sample was conducted in triplicate. Completely randomized design (CRD) was used with one-way analysis of variance (ANOVA) to determine the difference between the mean of samples. The significant differences among the means were identified by using Fischer's Least Significant (LSD). Data analysis was performed using Minitab Statistical Software, version 18.0 (Minitab Inc).

3. Results and discussion

3.1 Proximate composition of shortfins scad emulsion sausage

The proximate compositions of shortfin scad (*Decapterus macrosoma*) sausages incorporated with surimi and silver catfish (*Pengasius*) protein hydrolysate (SCPH) are shown in Table 2. The moisture and carbohydrate contents of shortfin scad sausage significantly increased ($p<0.05$) with a higher ratio of surimi, while protein, fat and ash were significantly decreased with the increased ratio of surimi. There was a significant difference among all formulations (A to D) in terms of moisture, protein, fat, ash and carbohydrates ($p<0.05$).

According to Food Regulations (1983), sausage should contain nitrogen less than 1.7% and not more than 30% fat, while the other levels such as carbohydrate, protein, ash and moisture are not stated. Fish sausage with the highest surimi incorporation (Formulation D) (0% shortfin scad mince) showed higher moisture content compared to the control sample because of the higher moisture content of surimi (77.6%) than shortfin scad mince. Fish sausage with the incorporation of surimi has a lower protein and fat content than the control. In fact, the washing method removed the undesirable components such as fat and sarcoplasmic protein, which contribute to 20-25% of total protein in fish muscles. Basically, surimi has a lower protein and

Table 2. Proximate composition of shortfin scad (*Decapterus macrosoma*) emulsion sausage incorporated with surimi and silver catfish (*Pengasius*) protein hydrolysate

Formulation	Moisture (%)	Protein (%)	Fat (%)	Ash (%)	Carbohydrate (%)
A	67.85±0.04[d]	21.61±0.41[a]	3.89±0.03[a]	2.53±0.12[a]	4.12±0.41[a]
B	68.40±0.05[c]	18.06±0.24[b]	3.24±0.52[ab]	1.79±0.21[b]	8.51±0.61[b]
C	70.30±0.25[b]	16.85±0.10[c]	2.59±0.53[bc]	1.33±0.11[b]	8.93±0.60[b]
D	71.82±0.06[a]	14.09±0.8[d]	2.25±0.58[c]	1.26±0.58[b]	10.58±0.84[c]

Values are expressed as mean±SD, n = 3. Values with different superscript within the same column are significantly different (p<0.05).

fat content than fish mince (Piotrowicz and Mellado, 2015). In addition, fish protein hydrolysate (FPH) improves the protein content in meat products such as sausage. However, the importance of the addition of FPH in the fish sausage was not just due to increasing protein levels, but also its effects on protein characteristics such as generation of peptides, which contributed to antioxidant activity and higher water holding capacity (Cumby *et al.,* 2008).

Ash content was significantly higher in formulation A (100% shortfin scad mince) than formulations B (60% shortfin scad mince), C (40% shortfin scad mince) and D (0% shortfin scad mince) due to the washing process in surimi manufacturing, which reduces the mineral content. Additionally, ash levels depend on the proportions of fish mince and surimi. Normally, the addition of salt contributes to increasing ash content (Huda *et al.,* 2012). The increased carbohydrate content with a greater ratio of surimi incorporation may be due to the addition of cryoprotectant, sorbitol and sugar in surimi manufacturing, which causes higher carbohydrate levels in fish sausage (Chuapoehuk *et al.,* 2001).

These results are in agreement with a study conducted by Huda *et al.* (2012) which showed that Malaysian commercial fish sausage had moisture, fat and ash levels of 67.33-73.36%, 0.93-6.53% and 1.71-2.61%, respectively. Moreover, the protein content obtained in this study was in a similar range as Talang Queenfish sausage with surimi addition at 14.61-19.42% (Yousefi and Moosavi-Nasab, 2014). A (100% shortfin scad mince) study conducted by Zakaria and Sarbon (2018) showed that the protein content (18-83-19.59%) in shortfin scad emulsion sausage with the incorporation of Snakehead (*Channa striata*) protein hydrolysate (CSPH) was in agreement with the results obtained in this study. All the fish sausage formulations in this study showed that the incorporation of surimi caused an increase in moisture and carbohydrates while reducing protein, fat and ash content.

3.2 Texture properties of fish sausage

The texture analysis of shortfin scad sausage incorporated with surimi and silver catfish protein hydrolysate (SCPH) is shown in Table 3. Based on the results, formulation A (100% shortfin scad mince) had the highest hardness, gumminess and chewiness values at day 0, 6 and 12 compared to other formulations (p<0.05).

The decrease of hardness in fish emulsion sausage is a good indicator of a good texture which was due to the

Table 3. Texture profile analysis of shortfin scad sausage incorporated with surimi and silver catfish protein hydrolysate

Storage day	Formulations	Texture parameter					
		Hardness	Springiness	Cohesiveness	Gumminess	Chewiness	Resilience
0	A	9.26±0.26[a]	0.918±0.01[b]	0.697±0.00[a]	6.68±0.21[a]	6.41±0.36[a]	0.33±0.04[c]
	B	7.60±0.34[b]	0.923±0.00[ab]	0.720±.01[a]	5.47±.27[b]	5.05±0.24[b]	0.37±0.01[ab]
	C	7.27±0.27[b]	0.926±0.00[ab]	0.729±0.00[a]	5.65±0.75[b]	4.74±0.22[b]	0.38±0.01[a]
	D	4.86±0.06[c]	0.936±0.01[a]	0.731±0.05[a]	3.78±0.31[c]	3.54±0.33[c]	0.35±0.03[bc]
6	A	8.11±0.38[a]	0.927±0.00[b]	0.735±0.00[a]	5.97±0.25[a]	5.53±0.23[a]	0.36±0.01[b]
	B	7.07±0.10[b]	0.927±0.05[b]	0.736±0.00[a]	5.20±0.06[b]	4.82±0.04[b]	0.37±0.01[ab]
	C	6.93±0.08[b]	0.939±0.02[a]	0.745±0.01[a]	5.16±0.03[b]	4.85±0.04[b]	0.37±0.03[ab]
	D	4.44±0.39[c]	0.939±0.14[ab]	0.724±0.01[a]	3.22±0.31[c]	3.01±0.25[c]	0.40±0.01[a]
12	A	7.78±0.44[a]	0.934±0.00[a]	0.810±0.03[a]	6.03±0.42[a]	5.63±0.43[a]	0.44±0.02[a]
	B	7.06±0.27[ab]	0.946±0.00[a]	0.733±0.00[b]	5.49±0.24[a]	5.31±0.32[a]	0.44±0.01[a]
	C	6.12±0.74[b]	0.936±0.00[a]	0.748±0.00[b]	3.14±0.44[b]	2.89±0.43[b]	0.44±0.01[a]
	D	3.91±0.51[c]	0.836±0.19[a]	0.809±0.05[a]	3.397±0.20[b]	2.04±0.21[c]	0.46±0.06[a]

Values are expressed as mean±SD, n = 3. Values with different superscript within the same column are significantly different (p<0.05).

higher moisture content of surimi. The data showed that the hardness of the fish emulsion sausage improved with the increase of surimi percentage used. The increase in hardness for formulation A (100% shortfin scad mince) was likely due to water loss and increased fat and protein content, which contributed to an increase in gumminess and chewiness. These findings were similar to a study done by Andres *et al.* (2006). In addition, this attribute may lead to reduce elastic properties of the fish emulsion sausage. Thus, affect the quality and consumer acceptance. A study by El-Nashi *et al.* (2015) found that the reduction in the amount of water was due to the lesser protein content in beef sausage Additionally, surimi has good gelling and emulsifying properties, which could enhance the texture properties of fish emulsion sausage. In addition, the functional properties of fish protein hydrolysate might stabilise the matrix of fish sausage and improve sausage hardness. This finding has been supported by the result obtained from the study on the effect of fish protein hydrolysate on the improvement of sausage hardness done by Zakaria and Sarbon (2018). This has been supported by a study from Intarasirisawat *et al.* (2014) which showed the hardness value of fish emulsion sausage increased with increased Skipjack roe protein hydrolysate content. This was probably due to the high moisture content and low-fat content of surimi. Furthermore, the increases in fat content contribute to hardness, increased gumminess and higher chewiness of fish sausage (Andres *et al.*, 2006). On day 6, the hardness value of fish emulsion sausage started to decrease, as well as gumminess and chewiness (p<0.05). The process of proteolysis during storage also will affect the texture of the final product. Enzymes cause proteolysis from microorganisms as well as from indigenous meat enzymes (Riebroy *et al.,* 2005). The softening of texture during storage is probably due to

proteolytic action by muscle endopeptidases (Toldra, 2006).

The results obtained in this study are in agreement with a study conducted by Santana, Huda and Yang (2012), who reported that threadfin bream sausage had a hardness of 4.14-5.85 kg, but that study was not in agreement with this study in terms of chewiness and gumminess. According to Santana *et al.* (2012), the use of surimi powder in a sausage formulation could decrease emulsion stability due to the higher amount of fluid that can be expressed. Furthermore, the drying process used to produce surimi powder contributes to protein denaturation, affecting the matrix of protein. The addition of Skipjack roe protein hydrolysate increased the hardness, chewiness and resilience of fish emulsion sausage (Intarasirisawat *et al.,* 2014). SCPH has an emulsifying activity that could stabilise the matrix of fish emulsion sausage. In general, the incorporation of surimi in fish sausage significantly affected the hardness, gumminess and chewiness since the other parameters were not significantly different during storage (p>0.05).

3.3 Colour properties of fish sausage

Colour changes for L*, a* and b* parameters of shortfin scad (*Decapterus macrosoma*) emulsion sausage incorporated with surimi and SCPH are shown in Table 4. Fish sausage incorporated with surimi and SCPH had significantly increased lightness (L*) when storage time increased (p>0.05). Meanwhile, the redness (a*) value of fish sausage incorporated with surimi and SCPH significantly decreased as storage time increased (p>0.05). However, there were no significant differences in yellowness (b*) between all formulations.

The highest lightness value (L*) of shortfin scad

Table 4. Colour analysis of shortfin scad (*Decapterus macrosoma*) sausage incorporated with surimi and silver catfish (*Pengasius*) protein hydrolysate

Storage time (days)	Formulations	Colour Analysis		
		L*	a*	b*
0	A	60.95±0.38[d]	3.763±0.17[a]	13.86±0.29[a]
	B	67.15±0.45[c]	2.413±0.11[b]	13.55±0.35[a]
	C	70.85±0.58[b]	1.790±0.11[c]	13.59±0.10[a]
	D	79.67±0.18[a]	0.25±0.036[d]	13.80±0.63[a]
6	A	61.64±0.78[d]	2.74±0.12[a]	13.66±0.65[a]
	B	66.11±0.79[c]	2.07±0.13[b]	14.52±0.24[ab]
	C	70.41±0.08[b]	1.60±0.04[c]	14.36±0.46[b]
	D	79.21±1.66[a]	0.59±0.07[d]	12.86±0.08[c]
12	A	59.76±0.87[d]	2.02±0.12[a]	14.23±0.48[a]
	B	66.44±1.29[c]	1.62±0.074[b]	14.45±0.25[a]
	C	71.28±0.25[b]	1.42±0.090[c]	14.71±0.22[a]
	D	78.91±0.23[a]	0.46±0.037[d]	14.67±0.24[a]

Values are expressed as mean±SD, n = 3. Values with different superscript within the same column are significantly different (p<0.05).

emulsion sausage incorporated with surimi and SCPH in Formulation D (0% shortfin scad mince) may be due to the colour properties of surimi and the elimination of heme pigments by washing process which increased the lightness of surimi (Yousefi and Moosavi-Nasab, 2014). Therefore, the incorporation of surimi in fish emulsion sausage caused the increase in lightness value compared to Formulation A. In addition, the decrease in redness (a*) of shortfin scad sausage with the increased ratio of fish mince to surimi was due to the myoglobin content in fish mince since it contributes to 80% of meat colour (Huda *et al.,* 2012). The washing process caused extraction, which contributed to the absence of blood pigments in surimi, thus reducing the reddish colour of fish sausage (Piotrowicz and Mellado, 2015). There were no significant differences in yellowness (b*) of fish emulsion sausage between all samples (p>0.05). The results obtained by a study conducted by Huda *et al.* (2012) indicate that Malaysian commercial fish sausage has lightness (L*) values in the range of 58.73-79.56 and redness (a*) values in the range of -0.58-17.43, respectively. However, the (L*) and (a*) parameters of this study were not in the range with those reported by Piotrowicz and Mellado (2015). The sausage produced with surimi in phosphoric acid had a lighter colour but was higher in gel strength than that produced with surimi washed with phosphoric acid. The colour properties of sausage depend on the fish species, as different species have different colours and lipid oxidation slightly affects the colour of fish emulsion sausage during storage. However, the use of surimi and protein hydrolysate could change the colour properties of fish emulsion sausage.

3.4 Microstructure of fish sausage

The microstructure of fish emulsion sausage developed from shortfin scad emulsion sausage incorporated with surimi and SCPH is shown in Figure 1. The microstructure of Formulation A contained more voids compared to Formulations B (60% shortfin scad mince), C (40% shortfin scad mince) and D (0% shortfin scad mince). As the surimi content in fish emulsion sausage increases, it enhances the structure of fish emulsion sausage, with the evidence of less void's presence on the surface.

The results showed that the higher the surimi ratio, the fewer the voids in the microstructure of fish emulsion sausage. According to Cáceres *et al.* (2008), the voids were known to be due to the presence of fat in the gel matrix. The voids disappear as the surimi ratio increases in fish emulsion sausage. The results show that the fat level of fish sausage with the incorporation of surimi has a lower fat level than the control (Formulation D) (0%

shortfin scad mince). In addition, the low-fat sausage was chewy and less hard than high-fat sausage (Carbello *et al.,* 1996). Formulations B, C and D had a less dense matrix compared to control and were thus softer and chewier than control. The microstructure was also influenced by the composition of moisture content in all formulations. The addition of surimi in fish emulsion sausage contributes to the high moisture content in the range of 67.85-71.82%, leading to a less dense structure. Furthermore, the protein hydrolysate addition in fish emulsion sausage contributes to emulsification, binding and gelling properties, due to the formation of interfacial film around fat globules (Intarasirisawat *et al.,* 2014).
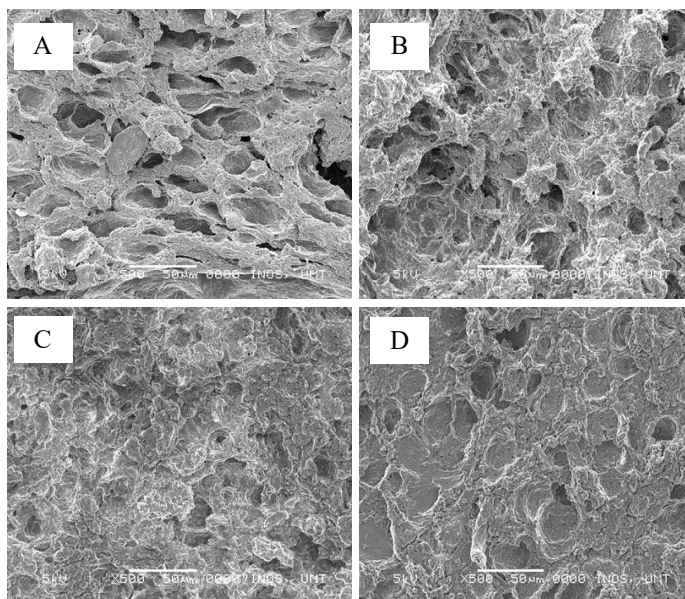

Figure 1. Scanning electron micrograph of shortfin scad sausage incorporated with surimi and silver catfish protein hydrolysate. (A) control (without surimi) 100 fish mince: 0 surimi (B) 60 fish mince: 40 surimi (C) 40 fish mince: 60 surimi (D) 0 fish mince: 100 surimi.

This study's results agree with those of a study conducted by Yousefi and Moosavi-Nasab (2014), which indicated that the surimi addition in sausage leads to better texture since the porousness of the surface of surimi sausage was less compared to control during preservation. In addition, the addition of SCPH providing stability in fish sausage emulsion was similar to studies conducted by Maqsood *et al.* (2012) and Intarasirisawat *et al.* (2014), which reported the addition of tannic acid and Skipjack roe protein hydrolysate had emulsifying properties, yielding stable meat emulsion and antioxidants capable of reducing the destruction of the matrix in fish emulsion sausage.

3.5 Lipid oxidation of shortfin scad sausage incorporated with surimi and silver catfish protein hydrolysate

3.5.1 Peroxide value of fish sausage

Figure 2a shows the peroxide value for shortfin scad sausage incorporated with surimi and SCPH during 12

days of storage (4°C). The initial value of PV at 0 days was significantly decreased when the ratio of surimi increased in shortfin scad emulsion sausage. The sharp increase was noticeable in all samples above 6 days during the storage for all formulations, except formulation D (0% shortfin scad mince), which significantly increased until day 9 then decreased until 12 days of storage. Peroxide value (PV) values were significantly different for all formulations over 12 days of storage (p<0.05).
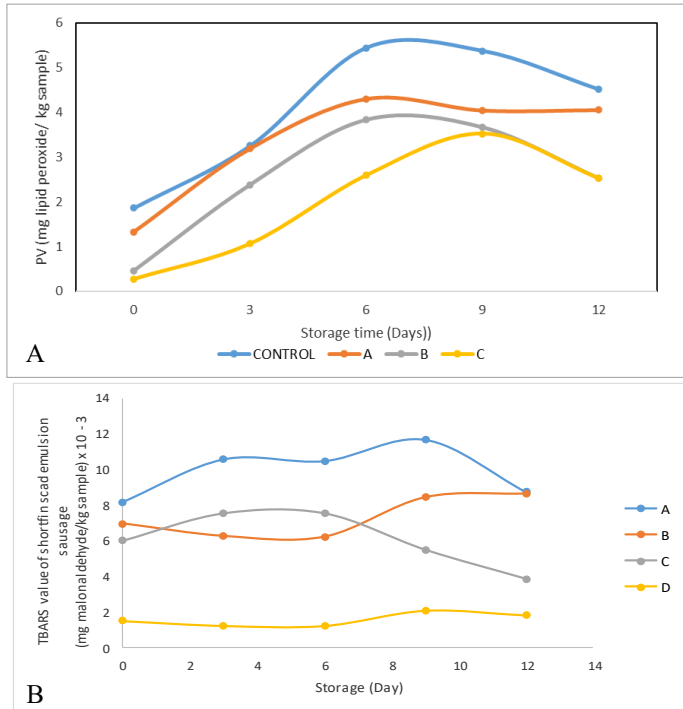

Figure 2. A) Peroxide value and B) TBARS value of shortfin scad sausage incorporated with surimi and silver catfish protein hydrolysate.

The peroxide values obtained in this study were lowered than the range of noticeable rancidity at 20-40 mEq/kg (Pearson, 1976). During the 12 days of storage, formulation A (100% shortfin scad mince) showed the highest peroxide value because of its higher fat content. Formulation D (0% shortfin scad mince) showed the lowest PV value due to the higher surimi ratio and had the lowest fat content because of the washing process. This was also due to the SCPH being incorporated in the shortfin scad emulsion sausage. A study by Andres *et al.* (2006) stated that high-fat content in fish mince (2.4%) compared to surimi (0.9%) contributed to the reduction of PV value in shortfin scad emulsion sausage. Peroxide values increased over time due to lipid oxidation during storage. Lipid oxidation causes rancid odour and flavour in processed meat product because of the autoxidation of unsaturated fatty acids (Olsen *et al.,* 2005). The decomposition of hydroperoxides, which produce decomposition products such as aldehydes and ketones, correlate with the results which showed a decrease in PV values during storage (Chaijan *et al.,* 2006).

The PV values obtained in this study correlate to those in a study conducted by Raju *et al.* (2003), indicating that fish sausages with the addition of niacin as a preservative have PV values of 18.15 to 24.80 mEq/kg, which are within the range of rancidity level (20-40 mEq). The use of SCPH as a natural preservative also lowered the PV values in fish emulsion sausage of the current study, as also found in studies by Intarasirisawat *et al.* (2014) and also Zakaria and Sarbon (2018). The decrease in PV values was noticeable because of the antioxidant activity of fish protein hydrolysate (Zakaria and Sarbon, 2018). In addition, formulation D (0% shortfin scad mince) has the lowest value of PV, which indicates that the use of surimi could reduce oxidation in fish emulsion sausage. Therefore, the use of low-fat source material such as surimi has been proven to decrease lipid oxidation in fish emulsion sausage (Yousefi and Moosavi-Nasab, 2014; Zakaria and Sarbon, 2018).

Meanwhile, during storage, the processes of lipid oxidation contribute to the factors that change the colour, aroma and taste. Lipid oxidation and free radicals produced products that caused the oxidation of myoglobin which produced metmyoglobin (Zakaria and Sarbon, 2018). Based on Table 4, there were slight differences between the L* and a* parameters of fish sausage during storage, perhaps due to the oxidation process. However, the formulations containing more surimi had a high lightness and low redness.

3.5.2 TBARS value of fish sausage

Figure 2b shows the effects of thiobarbituric acid on shortfin scad sausage incorporated with surimi and SCPH. There were significant increases in TBA values from day 0 until day 9 for samples A (100% shortfin scad mince) and D (0% shortfin scad mince), then significant decreases until day 12. The TBA values during storage were significantly different at 0, 6 and 12 days of storage (p<0.05).

The range of rancidity (0.5 to 2.0 mg MDA/kg) was slightly higher than TBARS values from a study by Gray and Pearson (1987). The increase in TBA value for formulation A (100% shortfin scad mince) as compared to formulations B (60% shortfin scad mince), C (40% shortfin scad mince) and D (0% shortfin scad mince) was due to high-fat level in fish mince compared to surimi. The increased amounts of fat and unsaturated fatty acids cause a higher TBA value (Tang *et al.,* 2001). The TBARS values of most of the samples increased during storage up to 9 days followed by a decrease until the end of the storage period. The small increase in TBA value during 12 days of storage was due to the storage condition. Meanwhile, the decrease in TBARS probably

due to TBARS reaction with free amino acids, protein and peptides present in the sausages producing Schiff's base (Maqsood *et al.,* 2012). The results indicate that the control sample has the highest TBARS values, which might be due to the loss of volatile secondary products during storage.

A study conducted by Yousefi and Moosavi-Nasab (2014) reported that sausage from Talang Queenfish and surimi had low TBARS values, which can enhance oxidative stability. As expected, as the fat content is higher in fish flesh than in surimi, peroxides and malonaldehyde are produced with the increasing length of storage. Furthermore, the use of natural antioxidants manages to control lipid oxidation (Maqsood *et al.,* 2012). In addition, a study by Zakaria and Sarbon (2018) also showed that the incorporation of fish protein hydrolysate was successfully retarded lipid oxidation. The antioxidant activity of SCPH reduces the oxidation in sausage.

3.6 Sensory evaluation of fish sausage

Table 5 shows the means scores for the sensory evaluation of shortfin scad emulsion sausage incorporated with surimi and SCPH. Each formulation was evaluated in terms of colour, odour, texture, taste and overall acceptability. Formulation B showed the highest scores in colour, odour, texture, taste and overall acceptability. However, it was found that there were no significant differences between colour, odour, texture, taste and overall acceptability between all formulations with surimi and SCPH added (p>0.05). This showed that the incorporation of surimi and SCPH can be accepted by consumers, as most of the formulations had higher values compared to control.

The result obtained for colour preference showed that the panellist could accept the colour of shortfin scad emulsion sausage with the incorporation of surimi and SCPH. Formulation A had the lowest score for colour preference since it contained the highest amount of fish mince ratio that contributed to the darkest colour as compared to the other formulations. Colour analysis showed the highest fish mince ratio in Formulation A had the lowest lightness (L*) and the highest redness (a*) values. In addition, in terms of odour preference, the

higher addition of fish mince ratio in fish emulsion sausage contributed to a strong fishy odour. However, the lowest score in odour preference was formulation D (0% shortfin scad mince), indicating that the panel preferred a moderate fish mince ratio because fish mince contributes to tenderness and a fishier flavour as compared to surimi. The results obtained for texture preference was 4.69 to 5.09, indicating that the panellists preferred the texture of shortfin scad emulsion sausage incorporation with surimi and SCPH. According to Dingstad *et al.* (2005), a hardness value of 47.3 N (4.8 kg) and above will produce sausage favoured by 60% of consumers. The hardness value obtained in this study was 4.86 to 9.26 kg and this indicated that the texture of sausage was preferred by the consumer. Meanwhile, the taste preference obtained in this study was 4.47 to 5.72. The higher fish mince ratio produced a strong fish flavour compared to the high surimi ratio. The incorporation of surimi could balance the fish flavour. In addition, a major disadvantage of protein hydrolysate in fish emulsion sausage is the occurrence of bitter peptides (Cavalheiro *et al.,* 2014). However, since the amount used for incorporation of SCPH in fish emulsion sausage was only 3%, it did not obviously cause a bitter taste.

The results obtained in this study are in agreement with a study conducted by Intarasirisawat *et al.* (2014) which found that fish protein hydrolysate did not affect the sensory properties of fish emulsion sausage. The results obtained in this study indicated that there were no significant differences in all attributes, in agreement with Ismail *et al.* (2014), who found that duck sausage with the addition of surimi showed no significant differences in terms of the colour, odour, taste, texture and overall acceptability. Therefore, the incorporation of surimi and fish protein hydrolysate in shortfin was accepted by the panellists in terms of colour, odour, texture and taste.

4. Conclusion

In conclusion, the physicochemical properties of fish sausage were influenced by the incorporation of surimi and fish protein hydrolysate. The higher the ratio of shortfin scad mince in fish sausage contributes to greater hardness, gumminess and hardness. Meanwhile, high shortfin scad to surimi ratio reduced the lightness value

Table 5. Sensory acceptability of shortfin scad sausage incorporated with surimi and silver catfish protein hydrolysate

Formulations	Color	Odor	Texture	Taste	Overall acceptability
A	4.47±1.70[a]	4.78±1.74[a]	4.78±1.62[a]	4.81±1.79[a]	4.84±1.55[a]
B	5.44±1.19[a]	5.34±1.07[a]	5.09±1.47[a]	5.72±1.11[a]	5.67±1.18[a]
C	5.16±1.57[a]	5.19±1.36[a]	4.69±1.66[a]	5.16±1.57[a]	5.28±1.44[a]
D	4.59±1.64[a]	4.63±1.48[a]	4.84±1.55[a]	4.47±1.69[ab]	4.63±1.39[ab]

Values are expressed as mean±SD, n = 3. Values with different superscript within the same column are significantly different (p<0.05).

(L*), while increasing redness (a*). The greater hardness of fish emulsion sausage contributed to compact structure and greater voids formation, in agreement with the level of fat content. Meanwhile, the increased addition of surimi with protein hydrolysate in shortfin scad emulsion sausage lowered lipid oxidation, with evidence of reduction of PV and TBARS values below the range of rancidity over 12 days of storage. In addition, the sensory analysis showed that the addition of surimi and SCPH did not affect the sensory properties. Formulation B with a 60% fish mince and 40% surimi ratio was the most acceptable compared to the control and other formulations.

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