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FULL PAPER

Comparative study on the physicochemical characteristics of chicken sausage incorporated with sutchi catfish *(Pangasius hypophthalmus)* gelatin, carrageenan and pectin

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Abstract

Gelatin was extracted from sutchi catfish (*Pangasius hypophthalmus*) skin in hot water (50°C) after pre-treatment in alkaline and acid solutions. The gelatin was then incorporated in sausage and compared with carrageenan and pectin in terms of sensory as well as physicochemical characteristics including moisture, protein, fat, process yield, cooking loss, folding test, purge loss, microstructure and texture profile analysis. Results indicated that sausage incorporated with sutchi catfish gelatin contain a higher amount of protein (17.48%) compared to others. The addition of sutchi catfish gelatin increased the moisture content and give a better folding with reduced cooking loss. Microstructural observation revealed a compact structure in sausage added with sutchi catfish gelatin which is much better compared to other sausage formulations. The sausage produced from sutchi catfish skin gelatin was equally acceptable as control and sausage with carrageenan. Control and carrageenan added sausages were progressively harder during storage while those containing pectin had a very soft texture. Hence, the addition of sutchi catfish gelatin improved much in physicochemical characteristics of the chicken sausage mainly in the protein content, folding test, moisture, process yield, cooking loss and microstructure. Therefore, sutchi catfish skin gelatin can be used as a future gelling agent in food.

1. Introduction

Gelatin, a hydrocolloid formed from collagen hydrolysis is a multifunctional ingredient used in foods, pharmaceuticals, cosmetics, and photographic films as a gelling agent, stabilizer, thickener, emulsifier and film former (Boran and Regenstein, 2010). The functional properties, thermostability and film-forming ability of gelatin are highly dependent on its characteristics which are related to the species origin. The quality of fish gelatin is determined mainly by its bloom strength and heat stability (Gomez-Estaca *et al*., 2009).

Sutchi catfish is a common farm-raised, warm-water fish. The skin of catfish has been studied for potential production of gelatin and it was found that the gels produced are relatively thermally non-degradable and characterized by a good gelling ability (Gomez-Guillen *et al.,* 2011). Gelatin from catfish also exhibited higher gel strength and protein content compared to other fish sources (See *et al*., 2010; Mahmoodani *et al*., 2014). Furthermore, the addition of sutchi catfish gelatin into gummy resulted in higher gumminess value than those added with commercial bovine gelatin (Normah and Muhammad Fahmi, 2014).

Gelatin derived from fish sources and the application of fish gelatin in meat-based food product such as sausage is still limited. Therefore, this study was carried out to compare the effect of sutchi catfish gelatin, carrageenan and pectin incorporation into sausage in terms of physicochemical characteristics, sensory and microstructure.

2. Materials and methods

2.1 Materials

Sutchi catfish (*Pangasius hypophthalmus)* aged 6 months and weigh 600 to 1000 g was obtained from a local freshwater fish supplier in Selangor. Chicken was purchased from a local market. Commercial pectin, carrageenan and analytical grade chemicals including sodium chloride (NaCl), sodium hydroxide (NaOH) and acetic acid (CH_3COOH) were purchased from Meilun Food Chemical Sdn. Bhd. Selangor, Malaysia.

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2.2 Gelatin extraction

Gelatin was extracted according to Normah and Muhammad Fahmi (2015). All preparation steps were performed in cold solution (5°C) prior to extraction. The gelatin powder was properly kept in an airtight bottle until used for sausage production and analysis.

2.3 Production of chicken sausage

The chicken meat was minced in a commercial food processor (Moulinex, Paris, France). Three different sausage formulations were prepared consisting of ingredients as shown in Table 1. After mincing, dry ingredients comprising of white pepper powder, salt, tapioca flour and sodium tripolyphosphate (STPP) were added. Sutchi catfish gelatin was diluted first with 5 mL of hot water (90°C) to dissolve the gelatin before adding into the mixture. Pectin and carrageenan were prepared in the same manner. To form an emulsion, cold water was added during mixing. The emulsion was subsequently stuffed into 27 mm diameter reconstituted sausage casings to form approximately 8 cm long chubs. Then, the sausages were heat-processed in a temperature-controlled water-bath (Haake, Karlsruhe, Germany) at 90°C until the internal temperature reached 74°C. The sausages were then cooled in tap water for 20 mins and then vacuum packed in polyethylene bags.

2.4 Chemical composition

Sausages were submitted to chemical analysis for moisture, crude protein and fat according to the AOAC (2002). All measurements were performed in triplicate.

2.5 Color

For color measurement, sausage was cut to approximately 2.5 cm thickness and 3 cm diameter and placed on a white plate. The color was measured directly on the internal part of the raw sausages using a Minolta Chroma Meter 2002 colorimeter (CR-400/410, Konica Minolta Holdings, Inc., Tokyo, Japan) based on the CIE (L*, a* and b*) system. The equipment was calibrated using white tile before measurements.

2.6 Process yield

The sausages were cooked in the boiling water (100ºC) for 4 mins followed by 10 mins draining. The process yield was determined by calculating the weight difference between samples before and after cooking (Serdaroglu, 2006).

$$\text{Process yield (\%)} = \frac{\text{Cooked sausage weight}}{\text{Uncooked sausage weight}} \times 100$$

2.7 Cooking loss

Cooking loss is equal to percent weight loss when the sausage had been cooked in boiling water (100°C) for 4 minutes. Cooking loss was calculated according to Sampaio *et al.* (2004) as follows:

$$\text{Cooking loss percent (\%)} = \frac{\text{Uncooked sausage weight} - \text{cooked sausage weight}}{\text{Uncooked sausage weight}} \times 100$$

2.8 Folding test

Folding test was performed according to Lanier (1992). The sausage was boiled and then cut into 3 mm thickness. The test specimen was held between thumb and forefinger to observe the way it broke and then evaluated according to the following scale: 1: breaks by finger pressure; 2: cracks immediately when folded into half; 3: cracks gradually when folded into half; 4: no crack shown after folding in half and 5: no crack shown after folding twice. Triplicate measurements were performed.

2.9 Sensory analysis

Sensory analyses were conducted by 30 panelists comprising of students and staff of Faculty of Applied Sciences, Universiti Teknologi Mara (UiTM) who had experienced in sensory evaluation of foods but received no specific training relevant to these products. Panelists were asked to indicate how much they like or dislike the samples in terms of color, taste, texture, juiciness and overall acceptability. A 9-point hedonic scale (9: like extremely; 1: dislike extremely) were used by the panelists to indicate their degree of acceptability. Freshly prepared samples were served to the panelists. The sausages were boiled in water for 3 mins, drained and

Table 1. Ingredients used in chicken sausage preparation (g/100 g sausage)

Ingredients	Control	Sutchi catfish gelatin	Carrageenan	Pectin
Fresh chicken	66.55	66.55	66.55	66.55
White pepper powder	0.55	0.55	0.55	0.55
Salt	1.85	1.85	1.85	1.85
Cold water	15.62	15.62	15.62	15.62
Hot water	5.00	5.00	5.00	5.00
Tapioca flour	10.23	4.39	4.39	4.39
Sodium tripolyphospate	0.20	0.20	0.20	0.20
Sutchi catfish gelatin	-	5.84	-	-
Carageenan	-	-	5.84	-
Pectin	-	-	-	5.84
Total (%)	100	100	100	100

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hold on a warm tray in covered plates for no longer than 30 mins. Warm 2.5 cm-long and 3 cm diameter pieces of the sausages were placed on a white polystyrene plate and presented to the panelists in 3-digit codes and random order for evaluation.

2.10 Microstructure

Microstructure was determined according to the method by Nafiseh *et al.* (2010). Small pieces of sausages which were 0.5 cm in diameter and 0.2–0.3 cm thick were used for microstructure analysis. After preparation of samples, dehydration and sputter-coating with a layer of gold/palladium in a metallizer, micrographs of the samples were obtained with scanning electron microscope (SEM 505; Philips, Noord-Brabant, Eindhoven, The Netherlands).

2.11 Purge loss

Purge loss was measured every two weeks during the 42 days of storage at -18°C. Prior to measurement, the sausage was first thawed for 4 hrs at 4°C. The sausage was weighed before and after opening of the package. Before weighing, the sausage was dried with paper towels. Purge loss was expressed as percentage of the initial sausage weight (Yang *et al*., 2007) and calculated as:

$$\text{Purge loss (\%)} = \frac{W_i - W_t}{W_i} \times 100$$

Where W_i was initial weight of sausage before opening the package and W_t was the weight after opening the package and dried with paper towels.

2.12 Texture profile analysis (TPA)

Texture profile analysis was determined every two weeks during 42 days of storage at -18°C. Sausages were thawed for 4 hrs at 4°C prior to analysis. Two repeated measurements were taken for each replicate and mean values were reported. Samples of 1.5 cm thick and 1.7 cm in diameter was cut from the center of the sausage and compressed twice to 30% of their original height between flat plates using a TAXT2i Texture Analyzer (Stable Micro Systems, Surrey, UK) interfaced with a computer, using the software. The head was operated at 0.5 mm/s. Hardness, springiness, cohesiveness and chewiness were determined.

2.13 Statistical analysis

Data were reported as means ±standard deviation. Statistics on the whole data was performed with the analysis of variance (ANOVA) procedure of SAS (Release 9.1, SAS Institute Inc., Cary, NC, USA, 2004) software. Significant differences at ($p<0.05$) were used to determine the differences among the means.

3. Results and discussion

3.1 Chemical composition

Moisture, protein and fat content of chicken sausage produced with the addition of sutchi catfish gelatin, carrageenan and pectin are shown in Table 2. The sausages containing hydrocolloids had significantly higher ($p<0.05$) moisture content compared to the control. Gelatin has been known for its capacity to improve the gelling and binding properties even used at a low level can stabilize shrinkage and increase the cooking yield of sausage (Jridi *et al.,* 2015). This is beneficial because high moisture content encourages film formation thus enhancing the sausage quality (Duxbury *et al.*, 2009). However, sausage incorporated with gelatin and carrageenan showed no significant difference ($p>0.05$). The moisture content of sausage produced in this study with the presence of hydrocolloids varied from 67% to 70%. The moisture content of sausages from previous studies ranged from 65% to 78% (Martinez *et al.,* 2004).

The addition of sutchi catfish gelatin significantly increased ($p<0.05$) the protein and fat content of the sausage compared to others. Similar findings were reported where the protein content in frankfurter increased when the levels of skin gelatin and wheat fiber mixtures were increased Choe *et al.* (2013). The high fat content of gelatin incorporated sausage was due to the gelatin which derived from the animal-based product in contrary to carrageenan and pectin which are from plant. *Pangasius sutchi* catfish skin has been shown to contain higher fat content than three other freshwater fishes in the previous study, as a result, the gelatin fat content was much higher than others (See *et al*. 2010). According to Malaysian Food Regulation 1985, the maximum allowable fat content in the meat product is 30%. In general, the fat content of the sausages in this study was in the range of 4.89% to 11.07% which is below the maximum fat content allowed by the Malaysian Food Regulation 1985.

3.2 Color

Table 2 shows the color value of the interior of chicken sausage samples. All sausages added with the hydrocolloids was significantly ($p<0.05$) lighter than the control where sausage added with pectin was the lightest. The effect of pectin and inulin as fat reducer on the color and textural properties of low-fat cooked sausages was studied (Álvarez and Barbut, 2013). It was stated that hardness and lightness were reduced. For a* value, there is no significant difference ($p>0.05$) in redness for all the samples. While for the b* value, sausage added with gelatin was more yellowish than others. This is due to the high-fat content in the sausage.

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Table 2. Physicochemical characteristics and sensory of chicken sausage produced with the addition of sutchi catfish gelatin, carrageenan and pectin

Parameters	Control	Sutchi catfish gelatin	Carrageenan	Pectin
Moisture (%)	62.98 ±0.412^{c}	68.05±0.676^{b}	67.24±0.698^{b}	70.46±0.875^{a}
Protein (%)	13.15±0.513^{c}	17.48±0.992^{a}	14.38±0.261^{b}	13.62±0.240^{bc}
Fat (%)	9.16±0.338^{b}	11.07±0.688^{a}	4.89±0.523^{d}	6.03±0.385^{c}
Color				
L*	66.08±1.872^{c}	71.34±0.685^{b}	70.12±0.606^{b}	74.02±1.040^{a}
a*	0.42±0.160^{a}	0.45±0.100^{a}	0.43±0.074^{a}	0.36±0.130^{a}
b*	13.62±0.420^{b}	15.27±0.934^{a}	14.42±0.510^{ab}	13.76±0.373^{b}
Process yield (%)	97.59±0.167^{a}	96.71±0.536^{a}	96.26±0.673^{a}	94.16±0.798^{b}
Cooking loss (%)	2.41±0.167^{c}	3.29±0.536^{bc}	3.74±0.722^{b}	5.84±0.797^{a}
Folding test	4.00±0.330^{ab}	4.33±0.335^{a}	3.78±0.191^{b}	1.22±0.190^{c}
Sensory attributes				
Color	6.27±1.23^{a}	5.97±1.47^{a}	6.20±1.09^{a}	5.60±1.16^{a}
Taste	6.48±1.19^{a}	5.77±1.07^{b}	5.90±1.30^{ab}	4.87±1.04^{c}
Texture	6.13±1.48^{a}	5.90±1.35^{a}	6.53±1.14^{a}	4.60±1.67^{b}
Juiciness	5.93±1.44^{ab}	5.63±1.20^{b}	6.33±1.18^{a}	4.77±1.89^{c}
Overall acceptability	6.50±0.97^{a}	5.97±1.89^{a}	6.27±1.08^{a}	4.47±1.01^{b}

Values are means ±standard deviation. Means within each row with different superscripts are significantly different at $p<0.05$.

3.3 Process yield, cooking loss, and folding test

The process yield indicates the yield retain after the sausage had been cooked, while the cooking loss is the water lost after cooking (Sampaio *et al.,* 2004). Moisture content and cooking loss are positively related whereas process yield, and cooking loss is negatively related (Serdaroglu, 2006). All the samples including control showed significantly higher ($p<0.05$) process yield than those incorporated with pectin. A previous study suggested that adding gelatin as a binder in turkey meat sausages has been considered to be advantageous by improving cooking yield (Pereira *et al.,* 2011).

Sausage added with gelatin had significantly lower ($p<0.05$) cooking loss than pectin. Gelatin and dietary fiber reduced cooking loss by forming hydrogen bond with water (Hoogenkamp, 2014). Additionally, the results of the folding test of the sausage was in the range of 1.22 to 4.33 in which sausage with sutchi catfish gelatin showed significantly higher value ($p<0.05$) than other hydrocolloids but not significantly different ($p>0.05$) from the control (Table 2). Pectin by itself decreased firmness and elasticity and it was suggested that pectin alone was not suitable to be used in meat product, however, its combination with other hydrocolloids gave better results (Solheim and Ellekjaer, 2001).

3.4 Sensory analysis

Color is one of the important sensory attributes closely linked to consumer acceptance. Table 2 showed no significant difference ($p>0.05$) in the acceptance for color among all the samples. As for taste, panelists accepted the control sausage more than sausage formulated with any of the hydrocolloids. Besides, sausage containing sutchi catfish gelatin was significantly ($p<0.05$) more acceptable than those containing pectin for taste, texture and juiciness attributes. Overall, sausage with pectin incorporation was least acceptable among the others. This was in agreement with the previous studies where frankfurter incorporated with pectin recorded the lowest penetrometer value (Candogan and Kolsarici, 2003).

3.5 Microstructure

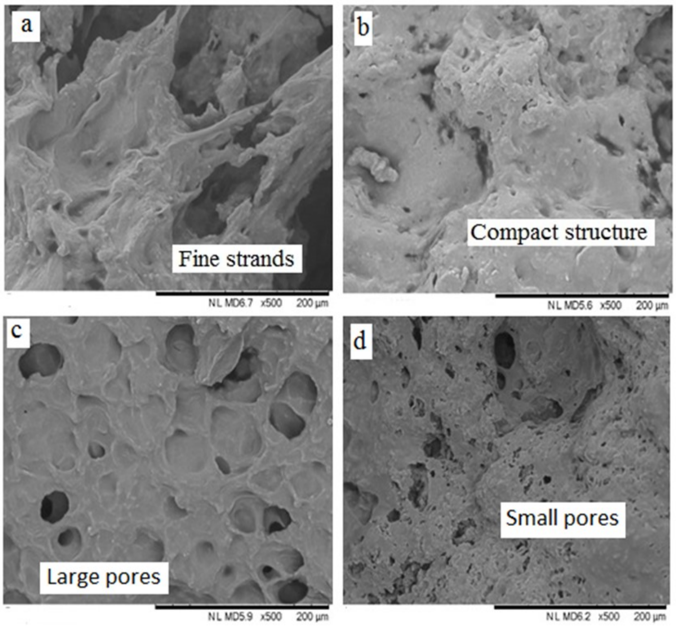

Figure 1. Scanning electron microscopy micrographs of chicken sausages formulated with different hydrocolloids; control (a), sutchi catfish gelatin (b), carrageenan (c) and pectin (d) at 500x magnification

Micrographs of the sausage samples are shown in Figure 1. Fine strands are observed in control (Figure 1a). Different pore sizes also exist with a relatively smooth shell surrounding them. According to Gordon and Barbut (1990) and Carballo *et al.* (1995) sausages containing carrageenan are low in fat but high in moisture forms numerous holes of smaller size compared to control. Similarly, smaller holes and compact structure in the sausages incorporated with gelatin from sutchi catfish as shown in Figure 1b may have been related to the high amount of fat that could not spread, thus accumulated in a point. Sausage incorporated with carrageenan (Figure 1c) and with added pectin (Figure 1d) revealed spongier structure as compared to control sausage and those with gelatin having apparently more cavity formation. This is due to the variation in the gel matrix and water retention in the developed network, thus, the arrangements and aggregation of protein filaments contributed to the different water holding capacity as well as to the textural properties (Riebroy *et al.,* 2005).

3.6 Purge loss

The percentage of purge loss every two-week intervals for 42 days between four different formulations of chicken sausage is shown in Figure 2. Purge loss was higher for all the treated samples compared to control. In general, the formulation with lower moisture contents had lower purge loss percentages. As a result of higher WHC, the purge loss percentages in the control sausage and sausage incorporated with carrageenan were significantly lower ($p<0.05$) than sausage incorporated with sutchi catfish gelatin and pectin. Hence, sausage incorporated with sutchi catfish gelatin exhibited lower purge loss percentage than the pectin and higher than control sausage and carrageenan by day 42. This is due to the inability of sutchi catfish gelatin in holding water than the control sausage and sausage incorporated with carrageenan. This result contradicts with the studies reported that the water holding capacity of the turkey meat sausage increase when the percent of cuttlefish gelatin added was increased (Jridi *et al.,* 2015). This result suggested that the different concentration of gelatin used in the formulation affect the WHC of the sausage.

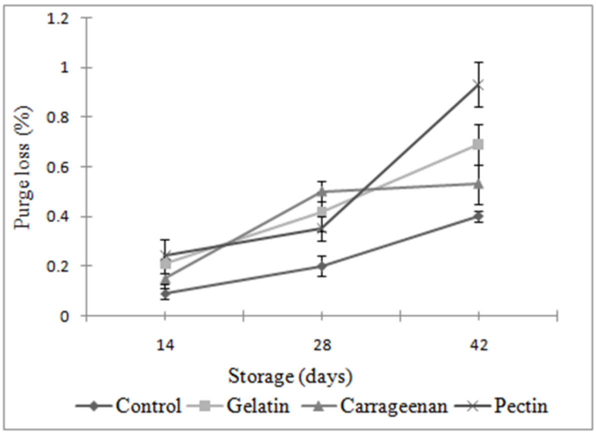

Figure 2. Purge loss of chicken sausage incorporated with sutchi catfish gelatin, carrageenan and pectin

3.7 Texture profile analysis (TPA)

The sausages of all formulations harden with storage time (Figure 3a). All the sausages added with hydrocolloids had lower hardness value than the control because treated sausages contain higher amount of moisture than control. Sausage added with gelatin and pectin showed a slight difference in hardness value during storage. This is due to the role of pectin that can retard the migration of water from cooked meat sausage (Bodner and Sieg, 2009). Gelatin is commonly used in canned meat products to prevent juice lost during storage (Keeton, 2001). The increase in hardness is likely due to water loss from the product (purge) during refrigerated storage and was also reported for low-fat beef

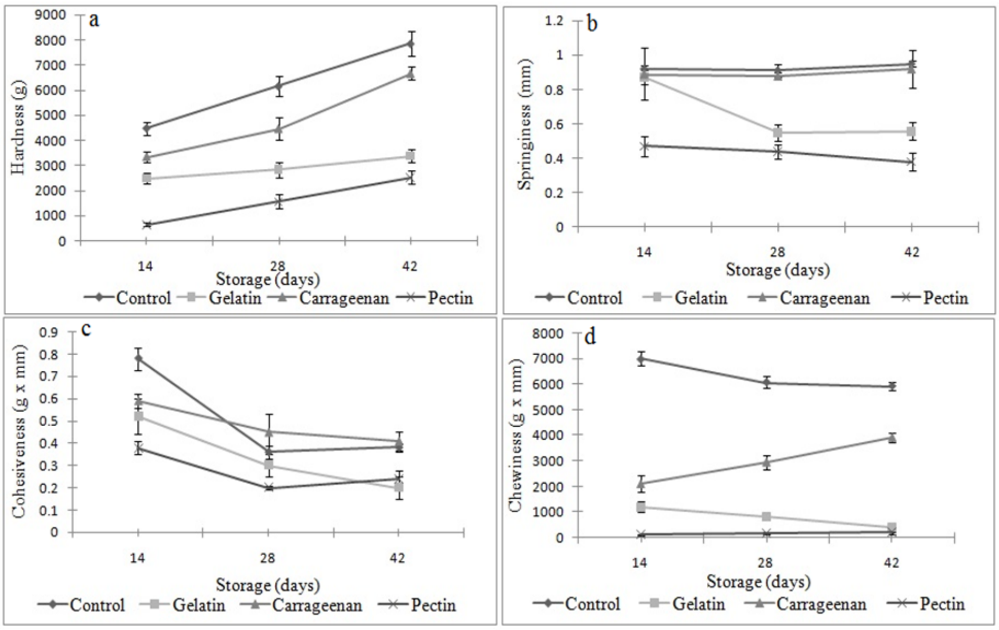

Figure 3. Hardness (a), springiness (b), cohesiveness (c) and chewiness (d) of chicken sausage incorporated with sutchi catfish gelatin, carrageenan and pectin

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frankfurters (Candogan and Kolsarici, 2003).

Only sausages added with gelatin and pectin had a decrease in springiness value during storage (Figure 3b). As for control and sausage added with carrageenan, there was not much difference. For the cohesiveness, sausage incorporated with gelatin had lower cohesiveness value compared to control sausage and carrageenan (Figure 3c). Jridi *et al.* (2015) reported that cuttlefish gelatin addition produced low cohesiveness of turkey sausages compared to the sausage without cuttlefish gelatin. For chewiness, sausages added with pectin and gelatin had lower chewiness values than control and carrageenan. This is due to its higher moisture content (Figure 3d). Chewiness for carrageenan incorporated sausage tends to increase compared to others due to the influence of fat level on the texture of sausage. Carrageenan and water addition to sausages containing low fat caused a significant increase in chewiness (Candogan and Kolsarici, 2003).

4. Conclusion

Sausage incorporated with sutchi catfish gelatin has good properties based on its physicochemical characteristics, therefore, sutchi catfish gelatin can be used in sausage production since it contributes to the similar properties and characteristics as other binding agents.

Conflict of interest

The authors declare no conflict of interest.

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