EXTRACTIONS, PHYSICOCHEMICAL CHARACTERIZATIONS AND SENSORY QUALITY OF CHICKEN FEET GELATIN

Mohd. Nazri Abdul Rahman & Shariffah Azzainurfina Syed Khair Azlan Jamalulail

School of Food Science and Nutrition,
Universiti Malaysia Sabah, Jalan UMS, 88400 Kota Kinabalu, Sabah, Malaysia.

ABSTRACT. *This study was carried out to determine the percentage of gelatin yield (w/w) extracted from chicken feet (CFG), and to compare the physicochemical properties and sensory qualities of CFG with bovine commercial gelatins (CBG). CFG extraction was based on gelatin type B method through three alkaline treatments by soaking in 4% HCl, 10% NaOH then 60°C thermal hydrolysis for 5 hours. A higher percentage of CFG powder was obtained at 18% w/w, and texture profile analysis showed correlation (r^2=0.98) between bloom strength and the gel hardness. Proximate analysis has shown that the powder of CFG extracted meets the standard as regulated in Food Act 1983 and Food Regulations 1985 with 6.43% humidity, 1.54% ash, 67.40% protein and 0.42% fat. There were no significant differences (p≥0.05) in ash and fat percentage of CFG and CBG as the values are 1.56±0.01 and 1.36±0.14 for ash, also 0.32±0.01 and 0.19±0.03 for fat respectively. Significant differences (p≤0.05) existed in water and protein percentage of the CFG and CBG with values of 6.64±0.20 and 8.03±0.16 for water, also 67.40±0.82 and 88.18±1.90 for protein. Sensory evaluations showed CFG was less acceptable (n=20) compared to CBG for colour, aroma and texture attributes. The score mean value for overall acceptance of CFG compared to CBG is 5.95±0.39 and 6.65±0.49 respectively.*

KEYWORDS. Chicken feet gelatin, halal gelatin, food hydrocolloids, nutraceuticals, pharmaceuticals.

INTRODUCTION

Gelatin is classified as animal protein derived from bone and skin collagen obtained through acid or alkali hydrolysis. Gelatin properties are affected by source, age and type of collagen (Bell, 1989; Gennadios *et al.*, 1994; Johnston-Banks, 1990). Gelatin is widely used in the food industry as an additive for its elasticity, consistency and as a stabilizer in pharmaceutical products and photographic technology. Gelatin has unique physicochemical properties that allow it to form thermo-reversible gel (Jamilah & Harvinder, 2001; Zhou & Regenstein, 2005) at melting point approaching body temperatures (the perception of melting in the mouth) and the ability to dissolve in water (Norziah *et al.*, 2009). Food grade gelatin not only depends on the rheological properties, but also the color, translucence, flavor and solubility (Gimenez *et al.*, 2005a).

Gelatin is known to replace the function of fat (as thickener agent E441, emulsifier, binder and nutrient) in food with no negative impact on the real taste of food products and to have a comparable sensory quality with fat functions which is widely used in the pharmaceutical industry as medication capsule gel (Demirhan *et al.*, 2012). Gimenez *et al.* (2005b) stated gelatin is used as a stabilizing agent in dairy and fermentation products including ice-cream to obtain smooth and fine texture, flexible gel structure and to prevent syneresis. In addition, gelatin is widely used as a dietetic agent in obesity management due to its low calorie content, as well as in baby food because it has high protein content (Riaz & Chaudry, 2004). Gomez-Guillen *et al.* (2011) reported many parts of poultry by-products are used in the preparation of high-value products based on collagen. These include skin and sternum cartilages that produce collagen type I and type II respectively. Meanwhile, Cheng *et al.* (2009) state the *'Silky fowl'* (a type of chicken) feet containing collagen and melanin are potential ingredients in cosmetic industries.

There are two types of gelatin extraction method, using either acid or alkali. Pre-treatment with acids such as hydrochloric acid (HCl), sulphuric acid (H_2SO_4), and phosphoric acid are frequently used for the extraction of gelatin from young collagen and have no complex structure such as pig skin (Hinterwaldner, 1977) and several species of fish within short period between 10 and 48 hours. While pre-treatment with alkali like sodium hydroxide (NaOH) or potassium hydroxide (KOH) (Jackson, 1995; Riaz & Chaudry, 2004) is used for extracting gelatin from matured collagen with complex cross structure such as skin, bones and cartilage of cattle and buffalo, which takes long time from 6 to 20 days (Imeson, 1997) to dissolve the mature and complex collagen.

The main gelatin production is from mammalian sources (Gimenez *et al*. 2005a) such as skin and bones of cattle and pigs (Binsi *et al*. 2009). According to Karim and Bhat (2009) and See *et al.* (2010), about 46% gelatin in the market is made from pig skin, 29.4% from cow skin, 23.1% from bones and 1.5% from other sources such as various species of fish (Lim *et al.* 2001). Issues of halal food by Muslims and vegetarians (Karim & Bhat, 2008) and kosher by Jews and the spread of mad cow disease (*Bovine spongiform encephalopathy*, BSE) in Europe (Demirhan *et al.*, 2012; Binsi *et al*., 2009; Jamilah & Harvinder, 2002) increased efforts to find new gelatin sources, especially in European Countries, India and Pakistan (Riaz & Chaudry, 2004) to meet the demands of Muslim consumers.

Fish gelatin has become a focus of the market (Grossman & Bergman, 1992). However, it has various disadvantages and is less stable than mammalian gelatin (Gomez-Guillen *et al.*, 2011) especially in terms of physicochemical properties such as low viscosity and gelling properties (Badii & Howell, 2006). Fish gelatins also have strong odour (Jamilah & Harvinder, 2002) depending on the fish environment such as the depth of habitat, pollution levels and types of plankton. Strong fishy smell of fish gelatin from fish skin such as *grouper*, *snapper* and *mackerel* (Irwandi *et al.*, 2009) results in limited use in food, pharmaceutical and nutraceutical products. Leuenberger (1991) reported that gelatins from cold fish contain amino acids, and exhibits low melting temperature and gelling properties. In addition, fish gelatin may affect allergic consumers (Hamada *et al.*, 2001; Sakaguchi *et al.*, 1999). In general, gelatin from mammals has the highest quality based on high melting point and gelling characteristics, it is also stronger than fish gelatin due to its high content of hydroxylproline (Gimenez *et al.*, 2005a).

In Malaysia, the use of chicken feet as food is limited due to the presence of small bones and cartilage with no muscle. Cho *et al*., (2006) reported on a study of the suitability of chicken feet as a replacement for cow hide in Korean traditional gellied foods (known as *Jokpyun*). From literature, high cartilage content can produce high yields of gelatin. To date, few studies have reported on the extraction of gelatin from chicken feet. According to Gomez-Guillen *et al.*, (2002), the quality of gelatin is greatly influenced by physicochemical properties and various methods of production, including types of tissue and animal species. Therefore, this study was conducted to determine the percentage yield of chicken feet gelatin (CFG) [w/w] and to compare the physicochemical and sensory quality of CFG with commercial bovine gelatin (CBG).

MATERIALS AND METHODS

Samples

Fresh chicken feet (4 kg) were obtained from poultry slaughterhouse EverGrowth Sdn. Bhd., Menggatal, Kota Kinabalu, cleaned and stored at -27°C. Only the bone and cartilage were used in the gelatin extraction. To minimize the differentiation in the yield of gelatin extracted, only chicken feet of the same age and from the same slaughterhouse were used. While the commercial gelatin powder of bovine source (CBG) was obtained from Halalgel, Kota Bharu, Kelantan.

Sample Preparation

Chicken feet samples were cleaned: skin, fat and cuticles were removed by soaking the samples in boiling water at 100°C for 40 minutes according to Dunn (2003) method which used to extract gelatin from cattle bones. The chicken feet were then dried at 50°C for 18 hours, and 3 kg dried chicken feet was recorded

(Muyonga *et al.*, 2004). After fat removal, the dried chicken feet were soaked in HCl solution (Merck) at concentration of 4% with ratio 1:6 of acid per weight of sample (Dunn, 2003). This process was performed at 27±1°C (room temperature) and the solution was changed at intervals of three days for 9 to 12 days.

Alkali Pre-Treatment

Osein, a spongy material result from the acidic treatment (HCl solution) for the purpose of minerals and non-collagen materials removal; then immersed in 0.2 M NaOH (1:10 w/v) (Badii and Howell 2006; Cheng *et al.*, 2009) (LAB-SCAN) (pH ~ 12.5) for 20 days and the solution was changed at intervals of 3 days. After 20 days in immersion, the samples were washed and soaked in distilled water for 24 to 48 hours and washed 7 times. Then the pH of the samples was neutralized using HCl or H_2SO_4 (Merck) up to pH 5 – 7 (Lim *et al.*, 2001).

Gelatin Extraction

After pre-treatment, the sample was transferred into a beaker and placed in a waterbath at 60°C for 5 hours. Then, gelatin obtained was filtered using Whatman filter paper No. 4 (Jamilah and Harvinder, 2002; Badii and Howell, 2004; Cho *et al.*, 2006). Evaporation was performed using a freeze drier until the moisture content was 10 to 13% (Riaz and Chaudry, 2004). Then the dried chicken feet gelatin (CFG) obtained was weighed.

Physicochemical Parameters

Percentage of Gelatin

CFG extracted was weighed to determine the percentage of gelatin yield according to Cho *et al.* (2006) using the following equation;

Percentage of gelatin (%) = (*a* g) / (*b* g) x 100%

where;

a = weight of dried gelatin at 10 – 13% moisture content (in gram)

b = weight of raw sample (in gram)

Gel Strength and Texture Profiles

Gel strength of the CFG was determined using a method performed by Johnston-Bank (1990). *Bloom* strength was measured in grams using a specific plate to put pressure on the surface of the gel (Schrieber and Gareis, 2007). Almost 6.67% weight per volume (w/v) CFG solution was prepared in accordance with *British Standards (BS 757:1975)* by mixing 7.5 g of dried CFG with 105 ml of distilled water. The solution was left at room temperature for 30 minutes (Johnston-Bank, 1990) prior to heating at 65°C for 20 minutes until the gelatin completely dissolved. It was then stored at 4°C for 16±2 hours for gel strength determination, and kept at 9±1°C for 16 to 18 hours according to the method of Pye (1996) for the determination of the gelatin texture profiles.

Gel strength and gelatin texture profiles were determined using *TAXT2 Texture Profile Analyzer Stable Micro System* with 1.27 cm diameter plate at the speed setting (pre-, run- and post-test) 0.5 mm/s, at 4 mm distance, trigger type Auto-4 g, and probe cylinder of radius 0.5 inches (P/0.5R) for the gel strength determination. While the speed settings (pre-, run- and post-test) for determining texture profiles were set at 2.0 mm/s, 1.0 mm/s and 10.0 mm/s, at distance of 2 mm, trigger type Auto-5 g, at 500 pps data acquisition, and cylinder probe with 4 mm radius (p/4). Both of these tests were determined by the weight of 5 kg load.

Viscosity

Gelatin solution at a concentration of 10% weight per volume (w/v) prepared by dissolving gelatin powder in distilled water and heat up to 60°C. Viscosity was measured using *Brookfield* digital viscometer with spindle No. 1 at speed of 60 rpm, temperature 40±1 °C (Kim *et al.*, 1994).

Colour
Almost 6.67% weight per volume (w/v) gelatin solution was prepared and cooled at 10±1°C for 16 to 18 hours. The colour of the gelatin was determined using the *Hunter Colorimeter* at tri-stimulus L* (bright to dark), a* (red to green) and b* (yellow to blue) (Cho *et al.,* 2006).

pH
pH values were determined at room temperature (27±1°C) by providing gelatin solution at concentration of 6.67% (w/v) by mixing 7.5 g of gelatin powder into 105 ml of distilled water (See *et al.*, 2010).

Melting Point Determination
Melting point of gelatin in the study was determined based on Muyonga *et al.,* (2004). Gelatin solution at concentration of 6.67% was prepared and placed in screw cap test tubes with little air space. Samples were closed tightly and stored in a refrigerator at 7°C for 16 to 18 hours. Then, the sample was transferred into waterbath at 10°C with inverted position, so that air space would move to the bottom. Staged heating was carried out by increasing the temperature by 1°C per minute with the addition of warm water (45°C) at 60 second intervals. The temperature when the gel melted and the air transferred from the bottom up is recorded as the melting point (Muyonga *et al.*, 2004).

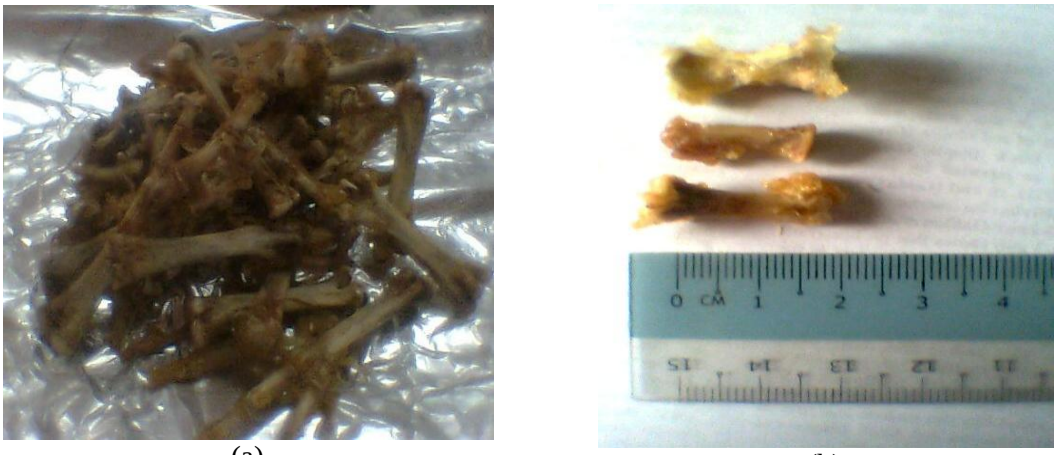
Proximate Analysis
Moisture content, ash and fat of the extracted gelatin was determined according to AOAC International (2000) and Badii and Howell (2006). Protein content was determined using *FOSS instrumentation Kjeltec 2300*, while the fat content was determined using *FOSS SOXTEC 2050* (AOAC International, 2000).

Sensory Quality Evaluation
CFG powder extracted and dried at 10% moisture content (Riaz and Chaudry, 2004) was tested for sensory quality attributes based on odour, colour, texture and overall acceptability; also comparisons were made with CBG as reference samples. No tasting sensory evaluations were carried out. CFG and CBG powder kept in an airtight container with a different number of permutations was served to panelists. Texture attributes were evaluated based on viscosity and clustering (agglomerates) of both samples in the study. Sensory panelists were provided with plastic teaspoons to press on samples for the determination of the texture scale. The hedonic test used 7 point scale (1 = strongly dislike, 2 = dislike extremely, 3 = dislike moderately, 4 = neither like nor dislike, 5 = moderately like, 6 = like very much, and 7 = like extremely) (Aminah, 2000). This testing involved 20 semi-trained panelists' comprising final year students of Food Science and Nutrition program, Universiti Malaysia Sabah.

Statistical Analysis
The data collected from physicochemical and sensory quality were analyzed using Statistical Package for Social Science (SPSS) version 17.0 using one-way analysis of variance (ANOVA) with significance level at $p \leq 0.05$. Paired t-tests were performed to determine the difference between the attributes studied.

RESULTS AND DISCUSSIONS

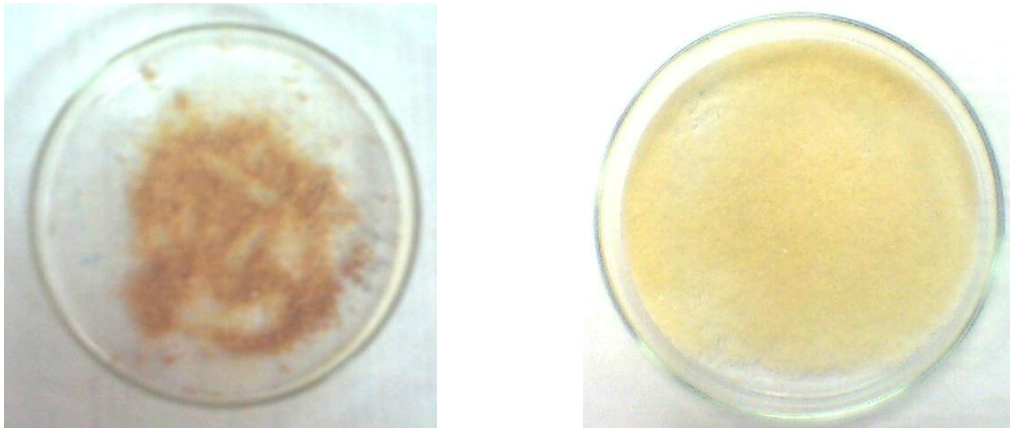
Yield of Gelatin
A total of 678.2 g of wet weight of CFG were obtained from 3 kg of dried chicken feet bone and cartilage which is equivalent to 22.6%. While the dry weight of gelatin (after freeze drying) was 122.1g. The percentage of gelatin extracted **[(dry gelatin / weight chicken feet) x 100]** was 4.1%. And the percentage of dry gelatin powder **[(dry gelatin / gelatin wet weight) x 100]** was 18.0%. Gelatin yield decline occurred due to the removal of cuticle, fats, minerals and water during sample pre-treatment and drying.



(a) (b)

Figure 1. Chicken feet bones obtained for gelatin extractions (a) Dried samples at 50^{o}C for 18 hours; (b) Bones cut to small size, 1 – 3 cm.

Based on the percentage of gelatin yield, CFG extracted had a higher yield (18.0%) compared to gelatin from freshwater fish such as snakehead (16.6%), red tilapia (11.6%) and silver catfish or *Pangasius Sutchi* (10.8%) as reported by previous researchers. However, the percentage of the CFG is 9.8% less than gelatin from catfish (27.8%) (See *et al.,* 2010). According to See *et al.* (2010), the percentage of gelatin derived from freshwater fish depends on animal species. While Songchotikunpan *et al.,* (2008) state that the differences in gelatin percentage obtained were influenced by species and age of animals, proximate composition, collagen content and methods of extraction.



(a) (b)

Figure 2. Gelatin in form of powder used in study (a) CFG powder produced after freeze-dried (b) powder of CBG.

Table 1. Physicochemical Properties and Proximate Analysis of CFG and CBG in studies.

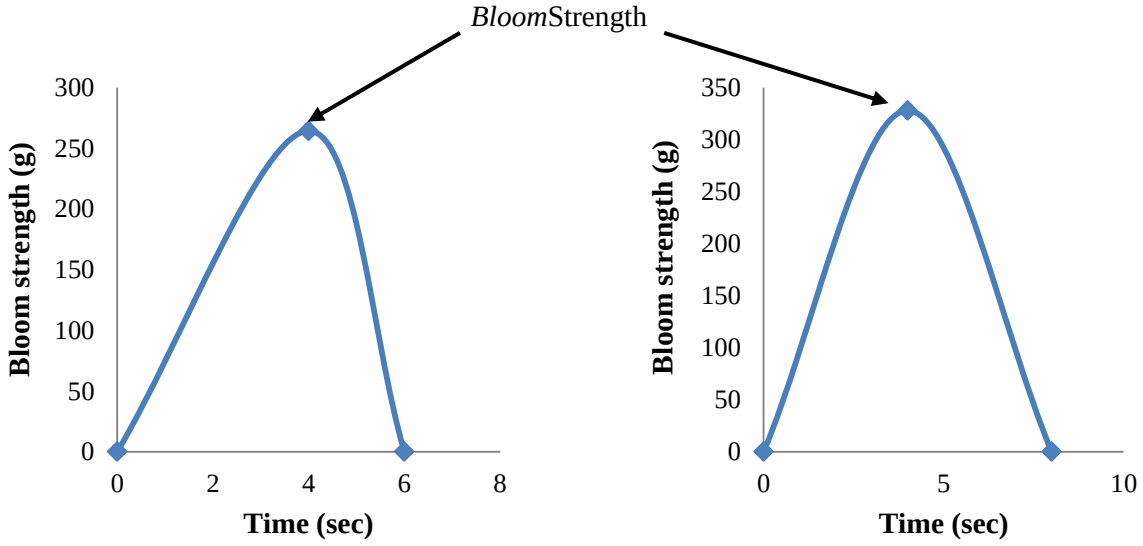
	Gel *Bloom* and Texture Profiles					**Colour**			**Proximate Analysis (%)**			
	Bloom (g)	Hardness (g)	Stickiness (g)	Elasticity (%)	Viscosity (%)	L*	a*	b*	Water	Ash	Protein	Fats
Chicken feet gelatin (CFG)	$264.33^{b} \pm 5.13$	$60.53^{b} \pm 0.68$	$4.82^{a} \pm 0.45$	$14.09^{b} \pm 0.12$	$4.96^{b} \pm 0.12$	$42.94^{a} \pm 0.69$	$2.82^{a} \pm 0.23$	$11.42^{a} \pm 0.20$	$6.64^{b} \pm 0.20$	$1.56^{a} \pm 0.01$	$67.40^{b} \pm 0.82$	$0.32^{a} \pm 0.01$
Commercial bovine gelatin (CBG)	$328.00^{a} \pm 9.17$	$74.21^{a} \pm 0.71$	$1.15^{b} \pm 0.24$	$18.01^{a} \pm 0.04$	$6.32^{a} \pm 0.02$	$34.40^{b} \pm 0.03$	$1.87^{b} \pm 0.12$	$-4.68^{b} \pm 0.08$	$8.03^{a} \pm 0.16$	$1.36^{a} \pm 0.14$	$88.18^{b} \pm 1.90$	$0.19^{a} \pm 0.03$

[a-b]: Different alphabet superscripts in the same column indicate significant difference ($p \leq 0.05$)

According to Tavakolipour (2011), alkali treatment can produce higher percentage of gelatins than acid treatment. This is because the alkali treatment can resolve complex collagen completely, and improve the rheological and physicochemical quality of extracted gelatin. The transformation process converts the collagen structure to gelatin, where the fibrous chain collagen structure is hydrolyzed into tropocollagen units through hydrogen chain and hydrophobic bonds hydrolysis (Martianingsih & Atmaja, 2009). This is because the hydrogen bonds (which stabilize the collagen complex) decompose in the thermal hydrolysis process (Zhou & Regenstein, 2005) using distilled water and produce gelatin. Taking into consideration the report by Tavakolipour (2011), the chicken feet in this study did not produce higher percentage yield than the gelatin from catfish and snakehead extracted by acid treatment as reported by See *et al.,* (2010).

Gel Strength and Texture Profiles

CFG gel *bloom* strength measured at 264.3g compared to 328.0g of CBG (Figure 3 and Table 1) which showed significant difference ($p \leq 0.05$). Based on the value of *bloom* CFG had high level of quality even though the CFG *bloom* gel (264.3g) was lower than CBG (328.0g). CFG was measured at 60.53g hardness**,** which was lower than CBG at 74.21g (Table 1). There was high correlation ($r^2 = 0.98$) between hardness and strength of the *bloom*, as shown by the linear relationship. High correlation occurred because the hardness of gelatin can also be used to determine the strength of the gelatin *bloom* (Muyonga *et al.*, 2004). According to Imeson (1997) gelatin that contains high in glycine, proline and hydroxylproline amino acids (Sarabia *et al.*, 2000; Arnesen & Gilberg, 2002; See *et al.*, 2010) were having high gel strength compared to gelatin with low amino acids (Muyonga *et al.*, 2004; Cho *et al.*, 2006). This shows that CBG had contain high amount of proline and hydroxylproline compared to CFG in this study.



(a) (b)

Figure 3. Gel *bloom* strength (a) CFG compared to (b) CBG at concentration 6.67%.

Gel strength (gel *Bloom*) is a key parameter in determining the quality of gelatin (Cheow *et al.*, 2007) which is determined based on hardness, elasticity, strength, compressive strength and the ability to compress at setting temperature and molecular weight (Ockerman & Hansen, 1998). Gel *bloom* strength values were measured in grams using a special plate to put pressure on the surface of the gel (Schrieber & Gareis, 2007).

According to Johnston-Banks (1983) there are three levels of gelatin quality: low quality (strength 150g), medium (150 - 220g), and high (220 - 300g). Thus, the CFG in this study had a high

level of quality with the gel *bloom* at 264.3g. However, the *bloom* gel for CBG in the study exceeded the value of high quality, which is > 300g and this shows that CFG had lower quality than CBG. In comparison, the quality of CFG is still at higher level than freshwater fish gelatin like cod-cake (90g), hake (110g) (Gomez-Guillen *et al.,* 2002), *Alaska Pollock* (98g) (Zhou *et al.,* 2006), and salmon (108g) (Arnesen & Gildberg, 2007). While it is comparable with other tropical fish gelatin such as catfish (265g) (Yang & Wang, 2009) and *Nile perch* (222 – 229g) (Muyonga *et al.,* 2004).

According to Muyonga *et al.,* (2004) gelatin extracted from bones has crossed structures and more stability than gelatin extracted from animal skin. This is because; collagen from animal skin contains dehydroxylysinonorleucine (deHLNL) while collagen from animal bones contains hydroxylysinoketonorleucine (HLKNL) (Sims *et al.,* 2000; Muyonga *et al.,* 2004b). Generally, HLKNL would change from the divalent to trivalent structure, which is hystidinohydroxylysinonorleucine (HHL) and pyridolines (PYR) after maturity. PYR structure is more stable under heat stress than HHL. PYR content in the bone remains stable even if the extraction is carried out at a high temperature (60°C).

This shows the discrepancy between the qualities of CFG and fish skin gelatin. However, CFG can be proposed to replace the use of fish gelatin in food and nutraceutical products because it is of higher quality than fish gelatin. According to Karim and Bhat (2008), the different value for gelatin gel strength is affected by intrinsic properties such as the composition of the protein (molecular weight distribution), the temperature of their habitats and the extraction process (Songchotikunpan *et al.,* 2008), and amino acid composition and pH of the gelatin extracted (Gudmunsson & Hafsteinsson, 1997). Cho *et al.,* (2004) states that high peptide chains in the gelatin can produce high gel *bloom* strength and are the most suitable for commercialization.

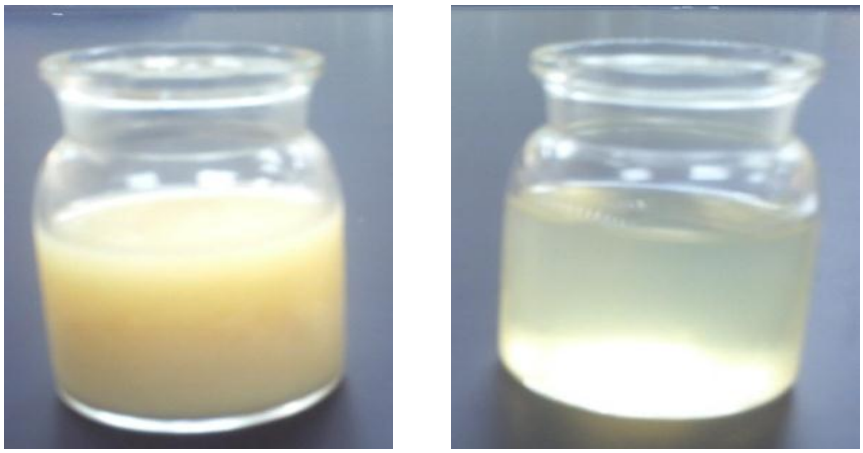
According to Arnesen and Gildberg (2006) thermal hydrolysis process at high temperatures causes denaturation of collagen, followed by the breaking of bonds by electrostatic and hydrogen bonding leads to the gelatin. This is because the hydrogen bonds between water molecules and hydroxyl groups of amino acids are key factors of gel strength. According to Sarabia *et al.* (2000) high hydroxylproline and proline content in gelatin produce high gel *bloom*. Meanwhile, animal species influence the proline and hydroxylproline distribution (See *et al.,* 2010) and mammals are estimated to have higher content of proline and hydroxylproline than freshwater and tropical fish with respective percentages at 30%, 22-25% and 17% (Muyonga *et al.,* 2004).

Viscosity

CFG in the study shows lower viscosity and significant difference ($p \leq 0.05$) compared to CBG in their percentage at 4.96% and 6:32% (Table 1). Differences in viscosity of gelatin are influenced by molecular weight, molecular size distribution and pH (Sperling, 1985; Cho *et al.,* 2006). According to Ockerman and Hansen (1988) and Ward and Courts (1997) the minimum viscosity of gelatin is achieved at pH 6 – 8. While Jamilah and Harvinder (2002) stated gelatin viscosity can be increased by the production of gelatin at pH 3 –10.5.

Colour

CFG has value of L*, a* and b* significantly higher than the CBG with L* values respectively 42.94±0.69 and 34.40±0.03. While a* value for CFG is 2.82±0.23 and 1.87±0.12 for CBG. For values of b*, CFG shows the reading 11:42±0:20 with CBG at -4.68±0.08 (Table 1). According to Ockerman and Hansen (1999) gelatin color is influenced by raw materials and it does not affect the nature and chemical quality of gelatin. Significant colour difference was observed between CFG and CBG (Figure 4). CFG appears cloudy due to imperfect filtration process. According to Muyonga *et al.,* (2004) imperfect filtering produces high turbidity values and affects the value of L*, a* and b*. The turbidity value of bone gelatin is higher than gelatin from animal skins (Muyonga *et al.,* 2004).



(a) (b)

Figure 4. Different colours between two types of gelatin (a) CFG and (b) CBG.

pH Value

The pH value for CFG is higher than the CBG with the respective values 6.15±0.07 and 5.57±0.02 but the difference is not significant ($p \geq 0.05$). The pH value affects the texture profile of gelatin and pH values approaching the isoelectric point (pH 5.0 for gelatin B) result in high *bloom* strength (Gudmundsson & Hafsteinsson, 1997). This shows that CBG has higher gel strength than CFG as close to pH 5.0. Previous studies on the pH value of fish gelatin showed lower values than chicken feet gelatin, such as *black tilapia* (pH 3.9), *red tilapia* (pH 3.1) (Jamilah & Harvinder, 2002), *sin croaker* (pH 3.3), *shortfin scad* (pH 4.9) (Cheow *et al.,* 2007) and *Chinese herring* (pH 4.5) (Norziah *et al.,* 2009). However, the pH value of gelatin is influenced by the type and strength of chemicals used during the pre-treatment (Songchotikunpan *et al.,* 2008).

Melting Point

CFG melting temperatures were lower than CBG in the study with the respective values 26.7°C and 33.3°C. The melting point of gelatin refers to the measured temperature when the gel softens and allows the carbon tetrachloride to decrease and is inter-related with the viscosity and gelatin *bloom* strength (Imeson, 1997). This proves that the viscosity and CFG *bloom* strength are low compared to CBG with the respective values 4.96% and 264.33g and 6.32% and 328.0g (Table 1) and this factor contributed to the low melting temperature. According to Norziah *et al.* (2009) and Haug *et al.* (2004), the melting point of gelatin is influenced by the content of proline and hydroxylproline because both are amino acids that function to stabilize the collagen structure. Low hydroxylproline and proline content contributed to the low melting temperature.

Proximate Analysis

Proximate analysis showed that CFG contains 6.64% water, 67.40% protein, 0.32% fat and 1.56% ash (Table 1). Gelatin is mainly made up of protein and water. Low moisture content increases the shelf life of gelatin, and influences the rheological properties such as elasticity and viscosity of the products. According to Jellouli *et al.* (2011) the presence of ash, fat and other foreign materials (impurities) at a low level is important for determining the quality of gelatin. Proximate analysis is performed on gelatin as a parameter in ensuring the removal of fat, mineral and hydrolysis processes are carried out efficiently (Muyonga *et al.,* 2004). According to Binsi *et al.* (2009) proximate analysis of raw samples and extracted gelatin is necessary, especially for determining the composition of proteins and amino acids that affect the gel *bloom* strength and gelling effects (Gomez-Guillen *et al.,* 2011), especially involving proline and hydroxylproline (Haug *et al.,* 2004).

In comparison, the CFG contains lower water (6.64%) content than CBG (8.03%). According to Ockerman and Hansen (1988) percentage of water content in gelatin is influenced by drying time, humidity, storage room and type of packaging used. Under the Food Act (2011) Malaysia, chicken feet gelatin in the study conforms to the regulations because it contains less than 16%water. High moisture content will damage gelatin cause it to be sticky.

The fat content of CFG is higher than CBG with respective values of 0.32% and 0.19% with a variance of 0.13%. A low fat content will determine the quality of gelatin. According to Muyonga *et al.* (2004) the fat content in the gelatin should be less than 0.5%. Proximate analysis showed that CFG contains 1.56% ash which is higher by 0.2% than CBG (1.36%) in the study. Under the Food Act (2011) Malaysia, the Food Act 1983 and Food Regulations 1985 the percentage of ash for gelatin powder should not exceed 3%, the low percentage of fat and ash showed that gelatin extraction process was done effectively.

Sensory Quality

Based on paired t-test, CFG and CBG powder in the study had significant differences ($p \leq 0.05$) in the odour, texture and overall acceptability attributes. For the attribute of colour, CFG does not show significant difference ($p \geq 0.05$) compared to CBG (Table 2). For odour and texture attributes, CFG has mean score lower than CBG with the mean scores respectively of 5.40±0.50 and 6.75±0.44 for odour attributes and 6.50±0.51 and 6.75±0.49 for the texture attributes. This significant difference shows that CFG has more unpleasant smell than CBG. The mean score for overall acceptability of the hedonic test shows CFG as less acceptable than CBG with the mean scores of 5.95±0.39 and 6.65±0.49 respectively.

Table 2. Score Mean Value for Hedonic Test on CFG and CBG Powder.

Attributes	Chicken Feet Gelatin (CFG)	Commercial Bovine Gelatin (CBG)
Colour	$6.25^{a} \pm 0.76$	$6.45^{a} \pm 0.44$
Odour	$5.40^{b} \pm 0.50$	$6.75^{a} \pm 0.44$
Texture	$6.50^{b} \pm 0.51$	$6.75^{a} \pm 0.44$
Overall acceptance	$5.95^{b} \pm 0.39$	$6.65^{a} \pm 0.49$

$^{a-b}$: Different superscript letters in the same row indicate significant differences ($p \leq 0.05$)

CONCLUSION

Gelatin from the chicken feet was successfully extracted with higher yield at 18% than freshwater fish gelatin of a comparable physicochemical quality with catfish and *Nile perch* but still at low quality compared to mammalian gelatin from bovine source. These results indicate that chicken feet gelatin (CFG) is of a lower quality than commercial bovine gelatin (CBG) but better than fish gelatin. This is because CFG had some weaknesses in physicochemical, proximate analysis and sensory quality compared to CBG in the study. However, these weaknesses should mitigate through other studies and still need improvements in methods and techniques of pre-sample preparation, treatment and extraction such as comparison of chemical extraction method using enzymes. At the same time, the gelatin from the chicken feet can be proposed as an alternative to halal gelatin in the market for food products, nutraceuticals and pharmaceuticals that are more cost effective than fish gelatin that has some physicochemical and sensory quality weaknesses.

REFERENCES

Akta Makanan. 2011. *Peraturan-peraturan Makanan (Akta 281)*. Malaysia: MDC Publishers Sdn. Bhd. Aminah Abdullah. 2000. *Prinsip Penilaian Sensori*. Bangi: Universiti Kebangsaan Malaysia.

AOAC International. 2000. *Official Methods of Analysis*. (17th edition). Gaitherburg: AOAC International.

Arnesen, J. A. & Gildberg, A. 2006. Extraction of Muscle Proteins and Gelatin from Cod Head. *Process Biochemistry*, **41**: 697-700.

Badii, F. & Howell, K. N. 2006. Fish Gelatin: Structure, Gelling Properties and Interaction with Egg Albumen Proteins. *Food Hydrocolloids*, **20**: 630– 640.

Bell, A. E .1989. *Gel Structure and Food Biopolymers*. London: Elsevier Applied Science.

Binsi, P.K., Shamasundar, B.A., Dileep, A.O., Badii, F. & Howell, N.K. 2009. Rheological and Functional Properties of Gelatin from the Skin of Bigeye Snapper (Priacanthus Hamrur) Fish: Influence of Gelatin on the Gel-Forming Ability of Fish Mince. *Food Hydrocolloids*, **23**: 132-145.

Cheng, F. Y., Hsu, F. W., Chang, H. S., Lin, L. C. & Sakata, R. 2009. Effect of Different Acids on the Extraction of Pepsin-Solubilised Collagen Containing Melanin from Silky Fowl Feet. *Food Chemistry*, **113**: 563-567.

Cheow, C. S., Norizah, M. S., Kyaw, Z. Y. & Howell, N. K. 2007. Preparation and Characterization of Gelatins from the Skins of Sin Croaker (*Johnius dussumieri*) and Shortfin Scad (*Decapterus macrosoma*). *Food Chemistry*,**101**: 386-391.

Cho, S. H., Jahncke, M.L., Chin, K. B. & Eun, J. B. 2006. The Effect of Processing Conditions on the Properties of Gelatin from Skate (Raja Kenojei) Skins. *Food Hydrocolloids*, **20**: 810-816.

Cho, S. M., Kwak, K. S., Park, D. C., Gu, Y. S., Ji, C. I., Jang & D. H. 2004. Processing Optimization and Functional Properties of Gelatin from Shark (*Isurus oxyrinchus*) Cartilage. *Food Hydrocolloids*, **18**: 573–579.

Demirhan, Y., Ulca, P. & Senyuva, H. Z. 2012. Detection of Porcine DNA in Gelatin and Gelatin-Containing Processed Food Products-Halal/Kosher Authentication. *Meat Science*, **90**: 686-689.

Dunn, J. M. 2003. *Bovine Bone Sourcing Practices and Bone Gelatin Manufacturing Processes in the United States of America*: Gelatin Manufacture Institute of America.

Gennadios, A., McHugh, T. H., Weller, C. L. & Krochta, J. M. 1994. *Edible Coatings and Film based on Protein*. Lancaster: Technomic Pub. Co. Inc.

Gimenez, B., Gomez-Guillen, M. C. & Montero, P. 2005a. Storage of Dried Fish Skins on Quality Characteristics of Extracted Gelatin. *Food Hydrocolloids*, **19**:958-963.

Gimenez, B., Turnay, Lizarbe, J., M. A., Montero, P. & Gomez-Guillen, M. C. 2005b. Use of Lactic Acid for Extraction of Fish Skin Gelatin. *Food Hydrocolloids*, **19**: 941-950.

Gomez-Guillen, M. C., Gimenez, B., Lopez-Caballero, M. E. & Montero, M. P. 2011. Functional and Bioactive Properties of Collagen and Gelatin From Alternative Sources: A Review. *Food Hydrocolloids*, **25**: 1813-1827.

Gomez-Guillen, M. C., Turnay, J., Fernandez-Diaz, M. D., Ulmo, N., Lizarbe, M. A. & Montero, P. 2002. Structural and Physical Properties of Gelatin Extracted from Different Marine Species: A Comparative Study. *Journal of Food Hydrocolloids*, **16**: 25-34.

Grossman, S. & Bergman, M. 1992. *Process for the Production of Gelatin from Fish Skin*. Patent No. **5**. United States of America.

Gudmundsson, M. & Hafsteinsson, H. 1997. Gelatin from Cod Skins as Affected by Chemical Treatment. *Journal of Food Science*, **62**: 37–47.

Hamada, Y., Nagashima, Y. & Shiomi, K. 2001. Identification of Fish Collagen as a New Allergen. *Bioscience Biotechnology Biochemistry*, **65 (2)**: 285–291.

Haug, I. J., Draget, I. K. & Smidsrod, O. 2004. Physical and Rheological Properties of Fish Gelatin Compared to Mammalian Gelatin. *Food Hydrocolloids*, **18**: 203–213.

Hinterwaldner, R. 1977. *Technology of Gelatin Manufacture*. London: Academic press.

Imeson, A. 1997. *Thickening and Gelling Agents for Food*. (2nd edition). New York: Chapman & Hall.

Irwandi, J., Faridayanti, S., Mohamed, E. S. M., Hamzah, M. S., Torla, H. H. & Che Man, Y. B. 2009. Extraction and Characterization of Gelatin from Different Marine Fish Species in Malaysia. *International Food Research Journal*, **16**: 381 – 389.

Jackson, E. B. 1995. *Sugar Confectionery Manufacture*. (2nd edition). New York: Chapman and Hall.

Jamilah, B. & Harvinder, K. G. 2002. Properties of Gelatins from Skins of Fish – Black Tilapia (*Oreochromismossambicus*) and Red Tilapia (*Oreochromis nilotica*). *Journal of Food Chemistry,* **77**: 81- 84.

Jellouli, K., Balti, R., Bougatef, A., Hmidet, N., Barkia, A. & Nasri, M. 2011. Chemical Composition and Characteristics of Skin Gelatin from Grey Triggerfish (*Balistes capriscus)*. *LWT – Food Science and Technology*, **44**: 1965-1970.

Johnston-Bank, F. A. 1983. From Tannery to Table: An Account of Gelatin Production. *Journal of the Society of Leather Technologists and Chemists*, **68**: 141– 145.

Johnston-Banks, F. A. 1990. *Food gels*. London: Elsevier Applied Science Publishing Co., Inc.

Karim, A. A. & Bhat, R. 2008. Fish Gelatin: Properties, Challenges, and Prospects as an Alternative to Mammalian Gelatins. *Food Hydrocolloids*, **23**: 253 – 576, 656.

Karim, A. A. & Bhat, R. 2009. Fish Gelatin: Properties, Challenges, and Prospects as an Alternative to Mammalian Gelatins. *Food Hydrocolloids*, **23**: 563 – 576.

Kim, S. K., Byun, H. G. & Lee, E. H. 1994. Optimum Extraction Conditions of Gelatin from Fish Skins and its Physical Properties. *Journal of Korean Industrial and Engineering Chemistry,* **5 (3)**: 547–559.

Leuenberger, B. H. 1991. Investigation of Viscosity and Gelation Properties of Different Mammalian and Fish Gelatins. *Food Hydrocolloids,* **5 (4)**: 353–361.

Lim, J. Y., Oh, S. S. & Kim, K. O. 2001. The Effects of Processing Conditions on the Properties of Chicken Feet Gelatin. *Food Science and Biotechnology*, **10(6)**: 638–645.

Martianingsih, N. & Atmaja, L. 2009. Analisis Sifat Kimia, Fizik dan Thermal Gelatin dari Ekstraksi Kulit Ikan Pari (*Himantura Gerrardi*) Melalui Variasi Jenis Larutan Asam. *Prosiding Kimia FMIPA*. Institut Teknologi. 10 November.

Montero, P., Fernandez-Diaz, M. D. & Gomez-Guillen, M.C. 2002. Characterization of Gelatin Gels Induced by High Pressure. *Food Hydrocolloids*, **16**: 197-205.

MS 1500: 2004. *Halal Food – Production, Preparation, Handling and Storage – General Guidelines* (First Revision). Department of Standards Malaysia.

Muyonga, J. H., Cole, C. G. B. & Duodu, K. G. 2004a. Extraction and Physicochemical Characterisation of Nile Perch (*Lates Niloticus*) Skin and Bone Gelatine. *Food Hydrocolloids*, **18**: 581–592.

Muyonga, J.H., Cole, C.G.B., &Duodu, K.G. 2004b. Fourier transform infrared (FTIR) spectroscopic study of acid soluble collagen and gelatin from skins and bones of young and adult Nile perch (*Lates niloticus*). *Food Chemistry*, **86**: 325–332.

Norziah, M. H., Al-Hassan, A., Khairulnizam, A. B., Mordi, M.N. & Norita, M. 2009. Characterization of Fish Gelatin from Surimi Processing Wastes: Thermal Analysis and Effect Of Transglutaminase on Gel Properties. *Food Hydrocolloids*, **23**: 1510-1616.

Ockerman, H. W. & Hansen, C. L. 1988. *Glue and Gelatin.* England: Ellis Horwood Ltd.

Ockerman, H. W. & Hansen, C. L. 1999. *Glue and Gelatine*. Boca Raton: CRC Press.

Pye, J. 1996. Gelatin- The Scientific Approach to Product Quality. *Food Australia*, **48**: 414- 416.

Riaz, M. N. & Chaudry, M. M. 2004. *Halal Food Production*. Boca Raton: CRC Press.

Sakaguchi, M., Hori, H., Ebihara, T., Irie, S., Yanagida, M. & Inouye, S. 1999. Reactivity of the Immunoglobulin E in Bovine Gelatin-Sensitive Children to Gelatins from Various Animals. *Immunology*, **96**: 286–290.

Sarabia, A. I., Gomez-Guillen, M. C. & Montero, P. 2000. The Effect of Added Salts on the Viscoelastic Properties of Fish Skin Gelatin. *Food Chemistry*, **70**: 71–76.

Schrieber, R. & Gareis, H. 2007. *Gelatine Handbook.* Weinhem: Wiley-VCH GmbH & Co.

See, S. F., Hong, P. K., Ng, K. L., Wan Aida, W. M. & Babji, A. S. 2010. Physiochemical Properties of Gelatins Extracted from Skins of Different Freshwater Fish Species. *International Food Research Journal*, **17**: 809 – 816.

Sims, J. T., Avery, N. C. & Bailey, A. J. 2000. *Introduction to Physical Polymer Science*. New York: Wiley.

Songchotikunpan, P., Tattiyakul, J., & Supaphol, P. 2008. Extraction and Electro-Spinning of Gelatin from Fish Skin. *International Journal of Biological Macromolecules*, **42**: 247 – 255.

Sperling, L. H. 1985. *Introduction to physical polymer science*. New York: Wiley.

Tavakolipour, H. 2011. Extraction and Evaluation of Gelatin from Silver Carp Waste. *World Journal of Fish and Marine Sciences*, **3 (1)**: 10 – 15.

Ward, A. G. & Courts, A. 1997. *The Science and Technology of Gelatin*. London: Academic press.

Yang, H. & Wang, Y. 2009. Effects of Concentration on Nano-Structural Images and Physical Properties of Gelatin from Channel Catfish Skins. *Food Hydrocolloids*, **23**: 577-584.

Zhou, P. & Regenstein, J. M. 2005. Effects of Alkaline and Acid Pretreatments on Alaska Pollock Skin Gelatin Extraction. *Journal of Food Science*, **70**: 392- 396.

Zhou, P., Mulvaney, S. J. & Regenstein, J. M. 2006. Properties of Alaska Pollock Skin Gelatin: A Comparison with Tilapia and Pork Skin Gelatins. *Journal of Food Science*, **71**: 313–321.