

## Characterization of gelatins from Nile tilapia skins preserved by freezing and salting

### Caracterização das gelatinas obtidas a partir de peles de tilápia do Nilo conservadas pelos métodos de congelamento e salga

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#### Abstract

After Nile tilapia skin was preserved using the methods of freezing and dry salting, characteristics of skin gelatin were evaluated with regard to yield, rheological features and physical and chemical properties. Preservation was performed after filleting, at which time skins were either frozen (-18°C) for 7 days or salted (25°C) for 7 days. Although no differences ( $p > 0.05$ ) were observed with respect to humidity, protein, lipid, ash and calcium levels, gelatin from salted skins had a higher concentration of iron relative to frozen skins. Further, twenty-three fatty acids were detected in salted skins compared with merely three found in skin derived gelatin. Of amino acids found, glycine, alanine, proline and arginine were the most abundant. Hydroxyproline abundance in salted and frozen skin gelatin were 8.76% and 8.71%, respectively. In addition, salted skin gelatins had a greater accumulation of saturated fatty acids and lower rates of monounsaturated fatty acids. Salted skin gelatin had the highest yield ( $18\text{g} \times 100\text{g}^{-1}$ ), gel strength (200 g) and viscosity (19.02 mPas) when compared to the yield ( $17\text{g} \times 100\text{g}^{-1}$ ), gel strength (12.7g) and viscosity (9.16 mPas) of frozen skins. Results show that gelatin from dry salted skin had the best yield and also had relatively better rheological properties, more iron, and better coloration relative to gelatin obtained from frozen skins of Nile tilapia.

**Key words:** Gelatin from fish skin. Amino acid profile. Gel strength. *Oreochromis niloticus*.

#### Resumo

O objetivo deste estudo foi avaliar os métodos de conservação de peles de tilápia do Nilo (por congelamento e salga a seco) sobre o rendimento, propriedades reológicas e físico-químicas da gelatina obtida a partir destas peles. Para tanto, após a filetagem, peles de tilápia foram distribuídas em dois

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métodos de conservação: peles congeladas (-18°C) por 7 dias e peles salgadas (25°C) por 7 dias. Não foram observadas diferenças ( $P>0,05$ ) nos teores de umidade, proteína, lipídeos, cinzas e cálcio entre gelatinas; porém a gelatina de peles salgadas apresentou maior teor de ferro. Foram encontrados 23 ácidos graxos nas peles e apenas três nas gelatinas, sendo que gelatinas de peles salgadas apresentaram maior somatório de ácidos graxos saturados e menor de ácidos graxos monoinsaturados. Os aminoácidos encontrados em maiores proporções foram glicina, alanina, prolina e arginina, sendo que os valores de hidroxiprolina encontrados para a gelatina de pele salgada foi de  $8,76 \text{ g} \times 100\text{g}^{-1}$  e na gelatina de peles congeladas,  $8,71 \text{ g} \times 100\text{g}^{-1}$ . A gelatina de pele salgada apresentou maior rendimento ( $18 \text{ g} \times 100\text{g}^{-1}$ ), força de gel (200 g) e viscosidade (19,02mPas) em relação a gelatina de pele congelada ( $17 \text{ g} \times 100\text{g}^{-1}$ , 12,7g e 9,16 mPas). Conclui-se que a gelatina extraída a partir de peles conservadas pela salga a seco apresentou melhor rendimento, melhoria nas propriedades reológicas, maior teor de ferro e melhor coloração, em relação a gelatina obtida de peles congeladas de tilápia do Nilo.

**Palavras-chave:** Gelatina de pele de peixe. Perfil de aminoácidos. Força de gel. *Oreochromis niloticus*.

## Introduction

Gelatin is a common polymers and is widely utilized in food, pharmaceutical and cosmetic industries, due to its functional and technological properties (KARIM; BHAT, 2009). Approximately 95% of commercial gelatin is derived from mammals, mainly from pig and cow hides. The other 5% comes from the bones of these animals (TABARESTANI et al., 2010), even though several social and cultural mores limit the consumption of gelatin derived from swine and cattle (KARIM; BHAT, 2009). Fish skin may serve as an alternative source of gelatin used commercially. Further, it has received a lot of attention as an alternative to bovine sources of gelatin due to the occurrence of bovine spongiform encephalopathy (HOSSEINI et al., 2013) and foot-and-mouth disease, both which cause severe harm to humans (CHO et al., 2005).

The production of gelatin from fish skin is attractive because it is an important byproduct of the fish industry (HOSSEINI et al., 2013). Fish gelatin is different when compared to gelatin derived from mammals, mainly because the melting point of fish gelatin is lower. This results in fish gelatin dissolving quickly in the mouth without requiring chewing (KARIM; BHAT, 2009). The characteristics of gelatins and their resulting applications are affected by factors including collagen features of the prime matter, type of pre-treatment, extraction process and skin preservation methods (JOHNSTON-BANKS, 1990).

Mammal collagen is more stable and has greater preservation efficiency than fish-extracted collagen, which is more likely to deteriorate (FERNÁNDEZ-DÍAZ et al., 2003; GIMÉNEZ et al., 2005a; TABARESTANI et al., 2010). Fast microbial degradation and losses occur after the filleting and skin removal processes through the autolytic activities of proteases in the skin (GIMÉNEZ et al., 2005a; LIU et al., 2010) as a part of the process of collagen hydrolysis (INTARASIRISAWAT et al., 2007). Throughout the process, collagen is broken down into simple structures, and, subsequently, a loss of quality in prime matter occurs, reducing the final quality of the gelatin (FERNÁNDEZ-DÍAZ et al., 2003; LIU et al., 2008). Further studies on the different methods of fish skin preservation will be required to establish a method resulting in gelatins of higher stability that maintains commercially attractive properties throughout the extraction process.

The present work evaluated two currently utilized preservation methods for treating tilapia skins, freezing and dry salting. Analysing yield, and the rheological, physical and chemical properties of gelatin derived from fish skins treated by these methods.

## Materials and Methods

Raw skins of Nile tilapia (*Oreochromis niloticus*) were used. Fish (weighting between 600g and 800g)

were caught in earth tanks, stunned in a solution of ice water and slaughtered by cutting at the gills by the staff of the fish farm. Tissue residues were removed manually by trained employees of the farm. The raw material was refrigerated upon arrival at the laboratory. Skins were packed in ice coolers during the cleaning process. The skins were weighed and distributed in two treatment groups corresponding to the method used to preserve the skins. Group one was designated the freezing treatment set and group two would be preserved by dry salting. For each treatment, 3 kg of skin was processed.

During the cleaning of skins, temperature was maintained at 0-5°C for skins that would be later frozen and around 25°C for skins that would be later salted. Clean skins and solid wastes were weighted. Batches of skins from group one were frozen at -18°C, whereas skins for salting were maintained at ± 25°C for seven days. The salting process was carried out in perforated plastic trays alternating layers of skin and salt at a ratio of salt:skin equal to 1:6 (w/v).

Frozen skins were thawed in a refrigerator for 12 h and salted skins were desalted by four, 10 min washes. To wash, each batch was washed for 10 min, weighed and immersed in water at a ratio of raw material/water of 1:6 (w/v) at 24°C ± 2°C. Further, pH was adjusted between 2.8 and 3.2 with 10N H<sub>2</sub>SO<sub>4</sub> for 1 hr. The skins were then washed with tap water and a skin sample was used for the boiling test which checks the skin's internal pH (ZHANG et al., 2005).

Gelatin extraction was performed as previously described in Bordignon et al. (2012). After pH stabilization (between 4.8 and 5.2) was reached, gelatin was extracted in a water bath for an hour at 50°C ± 2°C. The concentration of total solids was measured with digital refractometer (Atago 0-85% Brix). After extraction, both the gelatin solution and the non-dissolved skins were filtered through a Buchner funnel with wet cellulose pulp (20mm) and Whatman filter paper n.4. A sample was taken to use

for molecular profiling. The remaining solution was concentrated in a multistage evaporator to remove part of the water present in the gelatin solution, which was later gelled, ground and dried in a forced air dryer at 25°C for 24 hours and then at 50°C for 4 hours (Figure 1).

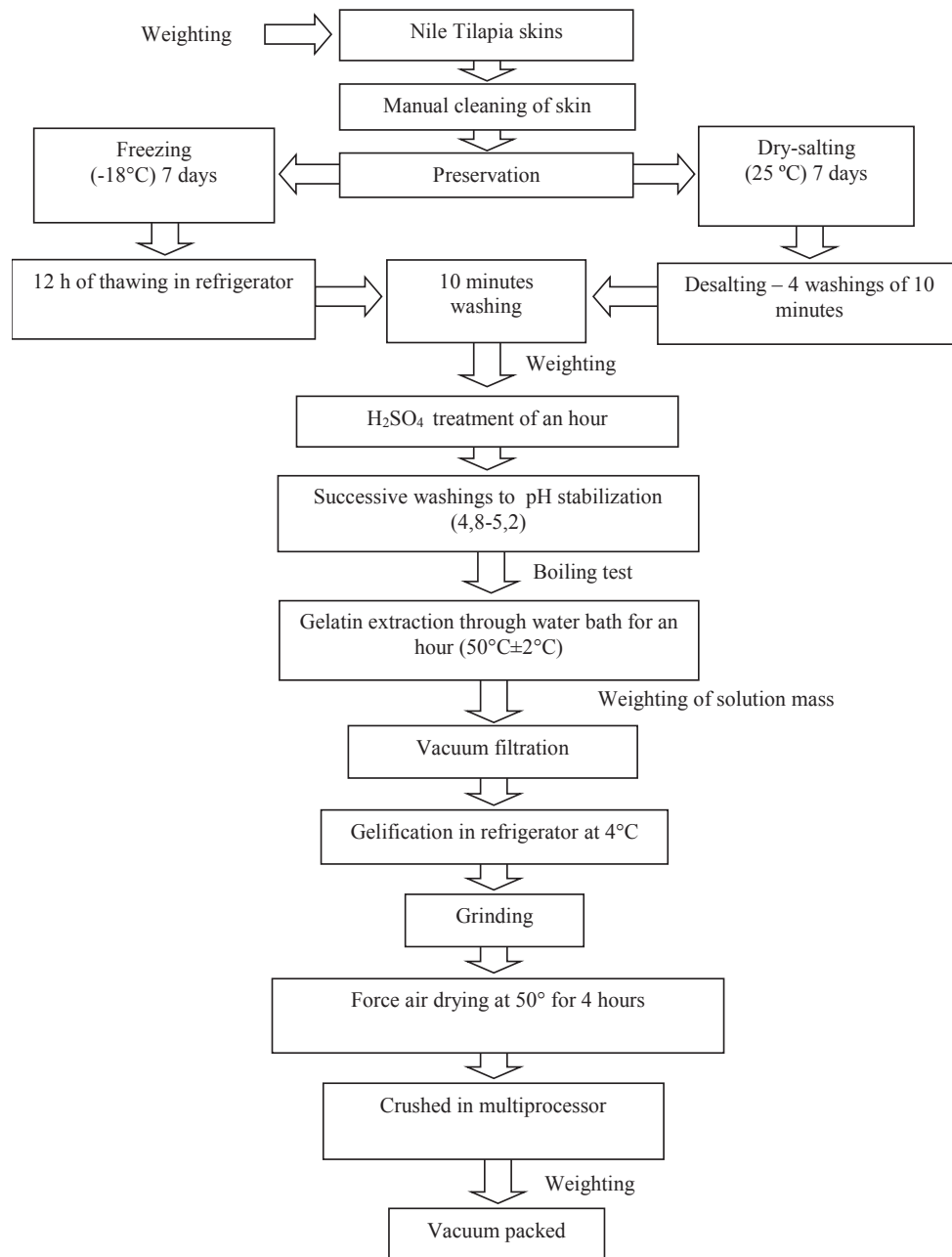
Gelatin extraction yield was calculated by the ratio proposed by Nagarajan et al. (2012) as shown in the following equation:

$$\text{Yield (\%)} = \frac{\text{Weight of dried gelatin (g)}}{\text{Weight of dry skin (g)}} \times 100. \quad (1)$$

After seven days, frozen and salted skins coupled to dry gelatin were analyzed to measure moisture, protein, ash, minerals, amino acids, fatty acids and hydroxyproline levels. Approximate composition of moisture, protein, ash and fat were determined by AOAC methods (2005). Ca and Fe levels were analyzed following Eijsink et al. (1997) using an atomic absorption spectrophotometer with flame atomization (AA240FS - VARIAN).

Gelatin pH was measured using a concentrated solution of 6.67% gelatin dissolved in 100 ml of deionized water with a pH-meter (Model FE 20-K, Mettler Toledo). Total lipids were extracted following method described by Bligh and Dyer (1959) and trans-esterified according to ISO methodology (1978), with separation by gas chromatography. A 2.0µL aliquot was injected into the fused-silica capillary column CP-7420 (Select FAME) (100m x 0.25mm i.d. x 0.25µm of cyanopropyl) at 165°C for 18 min and increased to 235°C for 24.5 min.

Fatty acids were identified by comparing retention times with Sigma standards (USA) through co-elution spiking of the standard with sample rates and comparing to Equivalent Chain Length (ECL). Percentage of fatty acids was determined by integration of peak areas using the Software Varian Workstation Star 5.0 and data was given as relative area percentages (STRÁNSKY et al., 1997).

**Figure 1.** Flowchart outlining the procedure used for the extraction of gelatin from Nile tilapia skins.

The composition of amino acids was determined with the Waters-PICO-TAG amino acid auto-analyzer using high-performance liquid chromatography (Waters 501, measuring 250x4.6mm) equipped with amino acid analyzing software. Each sample was hydrolyzed with 6N hydrochloric acid at 110°C for 24 hours (WHITE et al., 1986).

Hydroxyproline content was determined by hydrolysis of the gelatin solution in hydrochloric acid with boiling under constant reflux, hydrated and diluted. It was then oxidized with hydrogen peroxide and measured by spectrophotometer (SPEIGHT, 2005).

Viscosity was analyzed by measuring flow time of 100 $\mu$ L of gelatin solution (6.67%) prepared at 60°C, using a pipette viscosity-meter. Gel strength was obtained according to the British Standard (BS 757:1975) by texture analyzer TAX-T2 (Stable Micro System). The color of the gelatin solution was analyzed with a Hunter Ultrascan Sphere Spectrocolorimeter (Minolta Cr-300 series), using the L\*, a\* and b\* scale, following methods outlined by Hunter (1975).

The experimental design was completely randomized and was comprised of two treatments of six replicates each. Each set of treatments was comprised of either gelatin extracted from frozen skins or gelatin extracted from salted skins. Results for each of the parameters, including yield, centesimal and mineral composition, bloom, viscosity and color, were analyzed by analysis of variance (ANOVA) and means were compared by F-test at 5% probability to determine significant differences using a Statistical Analysis System (SAS, SAS Inst. Inc. Cary, NC USA).

## Results and Discussion

The overall yield of gelatin extracted from salted skins (18 g  $\times$  100g<sup>-1</sup>) was greater ( $p < 0.05$ ) than that extracted from frozen skin gelatin (17 g  $\times$  100g<sup>-1</sup>). Yield variations are due to the chemical composition of the skin, as well as the age of animals, species, extraction method (JONGJAREONRAK et al., 2006; MUYONGA et al., 2004), loss of extracted collagen or by incomplete hydrolysis (JAMILAH; HARVINDER, 2002), temperature or storage conditions of the skins. Lower yield rates may be related to the degree of covalent crosslinks, which may reduce the solubility of collagen (FOEGEDING et al., 1996). Possibly, freezing and thawing of the skins degraded the fibrillar structure of collagen, which may have led to the lower observed yield of gelatins obtained from frozen skins. Fernández-Díaz et al. (2003) reported that freezing of fish skins negatively affected the molecular composition

and rheological properties of the resulting gelatin. Apparently, the salt provided a greater protection to collagen fibers and enhanced the preservation of other properties of the skin. This resulted in an increased yield for the extraction, when compared to the preservation through freezing. Another observation of note is that salt in skins that remained after desalting washes may have interfered in the extraction of collagen. Chlorides are commonly used to solubilize myofibrillar proteins, thus helping to remove muscle that remains adhered to skin and may also predispose collagen for better extraction (GIMÉNEZ et al., 2005b). According to Giménez et al. (2005b), fish skins washed with NaCl resulted in a higher yield gelatin extractions (16g  $\times$  100g<sup>-1</sup>) than unwashed skins using the same solutions (13g  $\times$  100g<sup>-1</sup>).

Yields resulting from gelatin extractions in the current study were similar to those from frozen skins of the Nile tilapia performed by Bueno et al. (2011) and fresh skins of the Nile tilapia by Songchotikunpan et al. (2008), with yields of 18.3g  $\times$  100g<sup>-1</sup> and 18.1 g  $\times$  100g<sup>-1</sup>, respectively.

Mean pH values for gelatin extracted from frozen and salted skins were 6.71 and 6.48, respectively, featuring type A gelatin with a pH range between 6.0 and 9.5, and type B gelatin with a pH range between 4.7 and 5.6 (NINAN et al., 2011).

There was no difference ( $p > 0.05$ ) in the chemical composition of gelatins derived from frozen or salted skins (Table 1) with respect to humidity, crude protein, lipids and ashes. The two types of gelatin averaged 11.80g  $\times$  100g<sup>-1</sup> humidity, 85.06g  $\times$  100g<sup>-1</sup> protein, 0.03g  $\times$  100g<sup>-1</sup> lipids and 2.44g  $\times$  100g<sup>-1</sup> ashes. High crude protein rates in the gelatins and low rates for humidity, lipids and ashes, reveal the efficient removal of nutrients during the collagen extraction and gelatin preparation processes, and were also reported by both Jongjareonrak et al. (2010) and Silva et al. (2014). Variations in the chemical composition of gelatin mainly occurs as a result of differences in



extraction methods and the protein composition of skins (GÓMEZ-GUILLÉN et al., 2011). Gelatin extracted from the skins of *Rachycentron canadum* and croaker revealed  $88 \text{ g} \times 100\text{g}^{-1}$  crude protein

(SILVA et al., 2014). Songchotikunpan et al. (2008) reported  $7.3\text{g} \times 100\text{g}^{-1}$  humidity,  $89.4\text{g} \times 100\text{g}^{-1}$  protein,  $0.3 \text{ g} \times 100\text{g}^{-1}$  lipids and  $0.4\text{g} \times 100\text{g}^{-1}$  ashes in the gelatin from the skins of Nile tilapia.

**Table 1.** Chemical composition and mineral content of gelatin extracted from Nile tilapia skins.

	Gelatin		F-Test	CV (%)
	Frozen skins	Salted skins		
<b>Chemical composition</b>				
Moisture (g.100g <sup>-1</sup> )	11.92±2.78	11.68±1.01	0.03 <sup>ns</sup>	16.79
Protein (g.100g <sup>-1</sup> )	84.47±5.59	85.65±2.31	0.18 <sup>ns</sup>	4.77
Fat (g.100g <sup>-1</sup> )	0.04±0.03	0.02±0.24	2.72 <sup>ns</sup>	10.95
Ash (g.100g <sup>-1</sup> )	2.37±0.55	2.51±0.29	0.21 <sup>ns</sup>	17.30
<b>Minerals</b>				
Calcium (mg.100g <sup>-1</sup> )	117.17±10.77	106.64±36.56	45.76 <sup>ns</sup>	32.28
Iron (g.100g <sup>-1</sup> )	0.27±0.08	0.50±0.04	2.50*	69.92

ns not significant ( $p>0.05$ ) by F test. \* Significant ( $p<0.05$ ) by F test.

Similar rates were observed regarding the ash content of frozen ( $2.37\text{g} \times 100\text{g}^{-1}$ ) and salted ( $2.51\text{g} \times 100\text{g}^{-1}$ ) skins. The minor differences were likely due to the fact that the latter underwent successive washings to remove salt excess, which may have affected the composition of the gelatin. However, ash rates were below the maximum limits recommended for gelatin at  $2.6\text{g} \times 100\text{g}^{-1}$  (JONES, 1977).

In the case of minerals (Table 1), the calcium content of both frozen and salted skins was similar ( $p>0.05$ ) even though levels of iron were significantly higher ( $p<0.05$ ) in gelatin derived from salted skins ( $0.50\text{g} \times 100\text{g}^{-1}$ ) when compared to that from frozen ( $0.27\text{g} \times 100\text{g}^{-1}$ ). As a rule, the mineral composition of gelatins is similar to that of skins. The current study showed that the salted skin preservation method affected levels of iron in skin derived gelatin. Contrasting rates were reported for herrings, in which the salting process decreased observed levels of several minerals (Ca, P, Mg, K, Cu, Zn, Se and Fe) when compared to the *in natura* prime matter (POLAK-JUSZCZAK, 2016).

Twenty-three fatty acids were detected in frozen and salted tilapia skins, but merely three were found in gelatins (Table 2). Several fatty acids had high concentrations in skins and in gelatins, with special reference to palmitic (16:0), stearic (18:0) and oleic (18:1n-9) acids. Levels of fatty acids in gelatins were lower than those in skins, differences which were likely due to the hydrolysis process of fatty acids in skins. Further, gelatin lipid content from both groups ( $0.03\text{g} \times 100\text{g}^{-1}$  and  $0.02\text{g} \times 100\text{g}^{-1}$ ) could be considered fat-free due to observed values that were lower than  $0.5\text{g} \times 100\text{g}^{-1}$  rates (CHEOW et al., 2007).

Although salted skins have an accumulation of polyunsaturated fatty acids (PUFAs), they were not detected in gelatins. Gelatins from salted skins had a greater accumulation of saturated fatty acids (SFAs) and a lower amount of monounsaturated fatty acid (MUFA) when compared to gelatins from frozen skins, demonstrating the composition of skin fatty acids can affect gelatin content. Tilapia skins stored in ice for 18 days had  $36.30\text{g} \times 100\text{g}^{-1}$  SFAs,  $35.67\text{g} \times 100\text{g}^{-1}$  MUFAs and  $23.79\text{g} \times 100\text{g}^{-1}$  PUFAs (SAE-LEAW et al., 2013), which are rates similar to those found in the current study.

**Table 2.** Composition of fatty acids ( $\text{g} \times 100\text{g}^{-1}$ ) in gelatins and skins of Nile tilapia preserved by either freezing or dry-salting methods.

Fatty acids	Skins		Gelatin	
	Frozen	Salted	Frozen	Salted
14:00	2.10	2.29		
14:1n-9	0.22	0.23		
16:00	24.69	24.24	45.12	46.67
16:1n-9	0.57	0.62		
16:1n-7	4.76	4.61		
16:1n-5	0.20	0.20		
17:00	0.40	0.41		
17:1n-9	0.21	0.24		
18:00	8.54	8.69	18.90	29.16
18:1n-9	33.46	30.82	35.96	24.15
18:1n-7	3.42	3.98		
18:2n-6	9.54	10.14		
18:3n-6	0.50	0.48		
20:00	0.54	0.56		
18:3n-3	2.31	2.09		
20:1n-9	0.56	0.48		
21:00	0.22	0.24		
20:2n-6	0.58	0.70		
20:3n-6	1.60	2.63		
20:5n-3	0.35	0.43		
24:1n-9	0.83	1.09		
22:4n-3	0.75	0.95		
22:6n-3	0.74	0.75		
Sum of saturated fatty acids (SFA)	36.49	36.43	64.02	75.83
Sum of monounsaturated fatty acids	44.23	42.27	35.96	24.15
Sum of polyunsaturated fatty acids (PUFA)	16.37	18.17		
PUFA/SFA ratio	0.45	0.50		
Sum of n-3 fatty acids	4.15	4.22		
Sum of n-6 fatty acids	12.22	13.95		

Table 3 shows the composition of amino acids of skins and gelatins. Glycine, alanine, proline, arginine and glutamic acid are the amino acids detected in skins and gelatins in high proportions. The composition of amino acids in gelatins is mainly determined by species (ALFARO et al., 2015).

Glycine was the amino acid with the highest abundance in gelatins, a finding corroborated by

Jongjareonrak et al. (2010). This is due to the type of structure forming the collagen chain, where at every two amino acid positions, a glycine residue is present within the collagen helix, followed by proline and hydroxyproline (NELSON; COX, 2011).

Hydroxyproline levels in gelatin extracted from salted and frozen skins of Nile tilapia were 8.76g

$\times 100\text{g}^{-1}$  and  $8.71\text{g} \times 100\text{g}^{-1}$ , respectively. Amino acid levels are proportional to the resistance of gelatin gel (BENJAKUL et al., 2012). Proline and hydroxyproline are particularly involved in gel formation and determine the rheological properties and thermal stability of gelatin. The greater the proline and hydroxyproline content of gelatin, the greater its susceptibility for the formation of more compact helices and, consequently, the higher its melting temperatures and the better its viscoelastic properties (HAUG et al., 2004). Gelatin from the

skin of the fish *Pangasianodon gigas* revealed 87 hydroxyproline residues per 1000 amino acids (JONGJAREONRAK et al., 2010), whereas gelatins extracted from the skin of the tilapia contained 79 hydroxyproline residues per 1000 amino acids (SARABIA et al., 2000).

Commercial gelatins from mammal and tuna skin contained 113 and 119 alanine, 342 and 336 glycine and 127 and 107 proline residues per 1000 amino acid residues, respectively (GÓMEZ-ESTACA et al., 2009).

**Table 3.** Amino acids in Nile tilapia gelatin and skins preserved by both freezing and dry-salt methods.

Amino acids	Skins		Gelatin	
	Frozen	Salted	Frozen skins	Salted skins
Aspartic acid	18.4	17.0	54.8	53.2
Glutamic Acid	29.8	27.6	95.6	92.8
Serine	11.1	9.8	34.0	33.1
Glycine	67.6	58.1	225.2	222.0
Histidine	2.7	2.5	7.1	6.5
Arginine	26.6	23.3	83.5	82.0
Threonine	8.5	7.8	25.2	24.5
Alanine	30.2	26.3	100.7	100.4
Proline	38.3	32.5	125.9	123.7
Tyrosine	2.3	2.3	4.4	4.3
Valine	70.0	6.2	18.8	17.1
Methionine	5.5	4.8	16.7	16.5
Cystine	3.3	1.9	7.0	3.7
Isoleucine	4.1	3.7	9.9	9.6
Leucine	9.9	8.8	25.5	25.1
Phenylalanine	6.7	5.9	19.2	19.0
Lysine	10.3	9.3	31.2	30.7
Total	282.3	247.8	884.7	864.0

Residues/1000 amino acid residues.

When viscosity and gel strength were assessed, gelatin viscosity from salted skins (19.02 mPas) was significantly ( $p < 0.05$ ) greater compared to gelatin from frozen skins (9.16 mPas) (Figure 2a). The difference is directly proportional to rates of gel strength or bloom, which was significantly ( $p < 0.05$ ) greater (200.10 g) in the gelatin from salted skins than in the gelatin from frozen ones (12.70 g) (Figure 2b). Therefore, skin preservation by the dry

salt method provides gelatins with greater viscosity rates and greater gel strength.

Gelatins with low viscosity rates produce weak gels, whereas high viscosity rates provide consistent and extensible ones (SILVA et al., 2014). The above is supported by this analysis showing that gelatin with greater viscosity also had greater gel strength. Gel strength is the most relevant functional characteristic of gelatin and varies depending



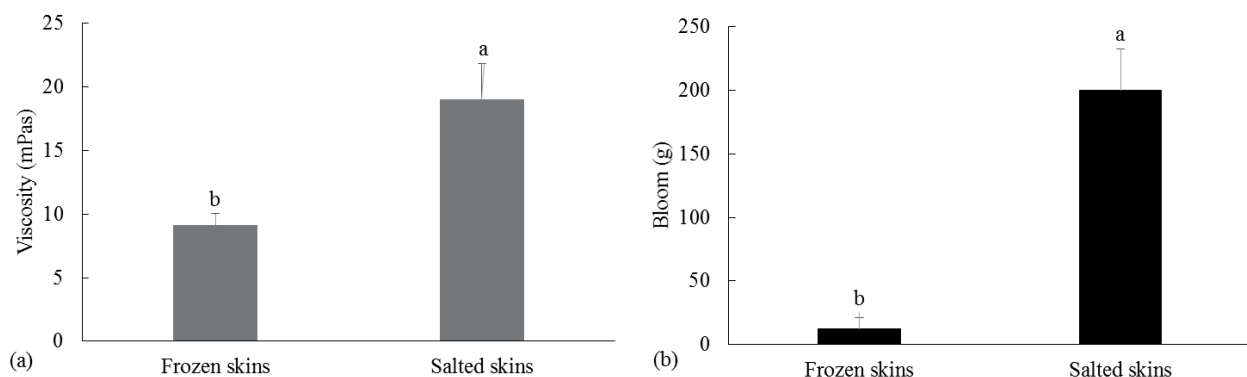
on concentration, conditions of preparation and the presence of other hydrocolloids (SILVA et al., 2014). As a rule, higher rates of gel strength correspond to higher quality gelatin (BUENO et al., 2011). Commercially usable gelatin strength values fall between 100g and 300g (GÓMEZ-GUILLÉN et al., 2011). Results show that the skin preservation method affects the gel strength of fish gelatins, possibly due to the degradation of alpha chains of collagen (FERNÁNDEZ-DÍAZ et al., 2003; LIU et al., 2008).

Whereas collagen is composed of three alpha chains, gelatin is comprised of alpha chains, beta components (dimers of alpha chains), high molecular weight polymers including gamma components (trimmers of alpha chains) and some fragments of smaller molecular weight (LIU et al., 2008). Gelatins with greater amounts of alpha chains have higher gel strength (SHI et al., 2002). It seems that skin freezing at  $-18^{\circ}\text{C}$  for seven days degraded alpha chains, resulting in reduced gel strength. Gelatin from the fresh skin of the fish *Platichthys flesus* has a greater gel strength when compared to gelatins from frozen skins ( $-12$  and  $-20^{\circ}\text{C}$ ), due to fragments of greater molecular weight and greater

proportions of alpha versus beta and gamma in gelatins derived from fresh skins. On the other hand, gelatins from frozen skins had more fragments with low molecular weight and smaller amounts proportions of components alpha in relation to beta and gamma (FERNÁNDEZ-DÍAZ et al., 2003). These were also trends that were confirmed by data in the current analysis (Figure 2).

On the other hand, the dry salt preservation method may improve gel strength when compared to that with fresh skins, as this study demonstrates. Liu et al. (2008) showed that the drying of skins provided gelatins with greater gel strength (256g) when compared with gelatin from frozen skins (246g) of *Ictaluru punctatus*. The improvement is due to the higher proportion alpha chains observed in the gelatin of dry skins. The authors found that fresh skins have many fragments of low molecular weight, providing a lower gel strength when compared to the gelatin extracted from dried skins (LIU et al., 2008). However, Giménez et al. (2005a) did not detect any difference in the gel strength of gelatins from *Solea vulgaris* skins stored for 160 days employing air-drying, ethanol, ethanol-glycerol mix and sea salt.

**Figure 2.** Data showing viscosity (a) and bloom (b) of gelatin extracted from frozen and salted skins of Nile tilapia. Mean values of six determinations ( $\pm$  SD) are given. Different letters indicate significant difference ( $p < 0.05$ ) by F-test.



There was no difference ( $p > 0.05$ ) in luminosity averages ( $L^*$ ) when the color of powder gelatins extracted from frozen and salted skins of the Nile

tilapia was compared (Table 4). However, differences ( $p > 0.05$ ) in averages was seen between chromes  $a^*$  and  $b^*$ , where gelatin extracted from salted skins

had the greatest means for chrome a\* (0.56) and b\* (9.23). Results show that gelatin from frozen skins had a more greenish color when compared to that of gelatin extracted from salted skins. Further, gelatin extracted from salted skins had a deeper yellowish color when compared to that of gelatin from frozen skins. Gelatin color depends on extracted prime matter and on whether it involves first, second or other extractions (NINAN et al., 2011). For instance,

gelatin from the skin of the African catfish (*Clarias gariepinus*) revealed rates of 57.60 for luminosity, 0.23 for chrome a\* and 2.66 for chrome b\* and were a light yellowish color (ALFARO et al., 2014). The color of most commercial gelatins ranges between light yellow and dark amber (JAMILAH; HARVINDER, 2002). It should be understood that the color of gelatin does not influence its functional characteristics (CHEOW et al., 2007).

**Table 4.** Color of dry gelatin extracted from Nile tilapia skins.

	Gelatin		F Test	CV (%)
	Frozen skins	Salted skins		
L*	37.54 ± 4.05	39.67 ± 2.25	1.05 <sup>ns</sup>	8.50
a*	-1.10 ± 0.56	0.56 ± 0.63	18.85*	8.50
b*	5.27 ± 1.60	9.23 ± 1.47	16.50*	21.22

L\*: luminosity a\*: intensity of red color; b\*: intensity of yellow color.

<sup>ns</sup>Not significant (p > 0.05) by F-test. \*Significant (p < 0.05) by F-test.

## Conclusions

Gelatin extracted from the dry-salted skins of the Nile tilapia had better yield, better rheological properties (gel strength and viscosity), higher iron levels and better color in relation to gelatin obtained from frozen Nile tilapia skins. In fact, sodium chloride is a highly efficient preserver for skins and, consequently, results in the production of quality gelatins from the skins of the Nile tilapia.

## Declaration of conflicting interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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