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Quality index method (QIM) and quantitative descriptive analysis (QDA) of Nile tilapia (*Oreochromis niloticus*) quality indices

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The aim of this study was to develop specific criteria for evaluating freshness in farmed Nile tilapia (Oreochromis niloticus), eviscerated and stored on ice, by employing sensorial, physicochemical and bacteriological analyses. Sensorial analyses were composed of quantitative descriptive analysis (QDA) for cooked fish and quality index method (QIM) for raw fish evaluation in samples stored for 22 days. Psychrotrophic aerobic heterotrophic bacteria were counted in muscles with and without skin stored for 28 days. Total volatile bases (TVB) were also determined in samples stored for 22 days. TVB analyses were within legal limits during the 22 days. Although psychrotrophic countings remained within acceptable limits until 18 days of storage, increased intensity in the perception of undesired alterations was observed on the 15th day of storage in the Nile tilapia as evaluated by QDA and by QIM. Based on the results of this trial, a shelf-life of 15 days is suggested for farmed tilapia, eviscerated and stored in ice.

Key words: *Oreochromis niloticus*, sensorial analyses, total volatile bases (TVB), psychrotrophic countings.

INTRODUCTION

In the last decades, with overfishing and the decreases in the commercial fish stocks, planned fresh water fish farming began to play an important role in the Brazilian pisciculture/agroindustry. In this context, Nile tilapia (*Oreochromis niloticus*) is an important species for aquaculture due to its great production potential (Sabbag et al., 2007). For instance, the Brazilian Ministry of Fisheries and Aquaculture estimated tilapia production in Brazil to be 253,824.1 tons in 2011 (Brasil, 2011). However, since not all the fishes captured and/or produced are sold at once, there is always the need for storage and therefore the attendant problem of quality

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control, particularly in the determination of fish freshness. In this context, sensorial methods, although old, are still the most effective means of determining, in a quick manner, fish freshness (Martinsdóttir, 1997). Quality index method (QIM) is based on the characteristics of surface, eyes and gills appearance, in addition to the odour of the iced fish (Luten and Martisdóttir, 1997). QIM system is precise because it is adapted for each species and since it also considers many characteristics of the fish, permitting the development of a score system referred to as quality index. This method has been employed for many fish species such as Clupea harengus (Jónsdóttir, 1992), Spaurus aurata (Huidobro e al., 2001), Salmo salar (Sveinsdottir et al., 2002), Merluccius merluccius (Baixas-Nogueras et al., 2003), Sardina pilchardus (Triqui and Bouchriti, 2003), Octopus vulgaris (Barbosa and Vaz-Pires, 2004), Gadus morhua (Esaiassen et al., 2004; Kent et al., 2004; Bonilla et al., 2007), Micropogonias furnieri (Teixeira, 2005), Salvelinus alpines (Cyprian et al., 2008), Litopenaeus vannamei (Oliveira et al., 2009), *Sepia officinalis*, L. (Sykes, 2009), Boops boops, L. (Bogdanovic et al, 2012) among others.

Quantitative descriptive analysis (QDA) is a sensorial method which employs trained evaluators selected for the description and quantification of descriptive sensorial attributes of flavour, odour, texture, appearance and it is statistically supported, representing an important tool in quality control of food processing industries (Stone and Sidel, 1998).

Determination of total volatile bases (TVB) is one of the most widely used methods for evaluating the quality of fish products. It involves evaluation of trimethylamine (TMA), produced by bacterial deterioration, dimethylamine (DMA), produced by autolytic enzymes during frozen storage, ammonia, which is produced by amino acid deamination, nucleotides catabolism and other volatile basic nitrogenated compounds associated with fish deterioration. Although TVB analysis is relatively simple to perform, its main drawback is that the test presents consistent increases only when fish is close to rejection and is therefore not suitable for making prognosis on commercial validity from intermediary data, being only useful, as an indicator of maximum shelf-life period (Contreras-Guzmán, 1994; Huss, 1998).

Tests based on total countings can be useful for measuring raw material conditions, and efficiency of procedures like thermal treatment, processing hygiene conditions, sanitary conditions of equipment and tools and, furthermore, the profile of the binomial, time x temperature, during storage and distribution (Huss, 1997).

Microorganisms that grow in refrigerated food between 0 and 7°C have optimal growing temperature of 20°C and are called psychrotrophic. This microbiota produces a visible growth in 7 to 10 days. Psychrotrophics are considered as a subgroup of mesophiles which are more common in refrigerated food, besides being responsible for food deterioration. Some psychrotrophics can be pathogenic like Aeromonas hydrophila, some strains of Bacillus cereus, Clostridium botulinum type E, B and F, Listeria monocytogenes, Vibrio cholera, Yersinia enterocolitica and some enteropathogenic strains of E. coli, as well as other organisms like Salmonella, Clostridium perfringens type C, some strains of Bacillus cereus and Staphylococcus aureus which grow slowly at temperatures between 7 and 15°C, but are able to grow if temperature abuse takes place during storage (Cousin et al., 2001).

In Brazil the criteria for considering fresh fish suitable for human consumption are determined by different National Laws and Regulations among which are the Regulations for Industrial and Sanitary Inspection of Products of Animal-Origin (RIISPOA) of art. 442 (Brasil, 1997a), Government Directive no. 185 of the Ministry of Agriculture (Brasil, 1997b), and by norms such as those of the Brazilian Association of Technical Standards (ABNT, 1993). Nevertheless, such criteria do not consider diversity among the different species and do not offer sensorial quality scores that could express fish freshness.

The aim of this research was to develop the QIM protocol, as well as to describe sensorial characteristics of cooked flesh, for the fresh water species, Oreochromis niloticus (Nile tilapia) at different periods of storage on ice.

MATERIALS AND METHODS

Sample collection and storage

Tilapias were obtained from a fish farm located in the state of Rio de Janeiro, Brazil. Collection included: 135 male, 4 to 6 months old, with an average weight of 412.1 kg (total of 55.6 kg), in the period of August 2005 to September 2006. After 24 h of depuration, fish were exposed to thermal shock with ice, eviscerated and washed. They were then transported in ice filled isothermal boxes, at the proportion of 1 kg of ice for 1 kg of fish. On reaching the laboratory, the fishes were packed in containers with ice at the proportion of 1 kg of ice for 2 kg of fish, stored and kept in a domestic refrigerator at a temperature of 0.3 ± 0.35°C, until the analyses.

Quantitative descriptive analysis (QDA)

QDA was performed according to the method described by Stone and Sidel (1998) which included recruitment, using a questionnaire; pre-selection by means of a triangular test for salty taste, training and selection of evaluators and further evaluation of the test-product with a sensorial team composed of nine evaluators. During training, cooked samples were offered to the evaluators and the attributes of appearance, odour, taste and texture were assessed by means of an open discussion among evaluators, moderated by a leader. QDA was performed under laboratory conditions where each evaluator examined samples at 1, 8, 15 and 22 days of storage. At day 22, only odour and appearance analyses were performed. Cooked samples, under controlled conditions, were individually presented on disposable plates, served with water and sample evaluation forms.

Table 1. Quality index method (QIM) scheme developed for farmed Nile tilapia (*Oreochromis niloticus*), eviscerated and stored in ice.

Parameter		Characteristics	pt
General aspect	Skin	With brightness, greyish colour, with darker well defined interpolated stripes.	0
		Less intense brightness, stripes less defined	1
		No brightness, loss of stripes definition, faded colour	2
	Scales	Adhered	0
		Scale loss	1
	Fish hardness	Tense	0
		Less tense	1
		Supple	2
	Flesh firmness	Firm	0
		Less firm	1
Eyes	Cornea transparency	Limpid	0
		Slightly opaque	1
		Milky, opaque	2
	Pupil	Black, well delineated	0
		Veiled, still delineated	1
		Veiled, not delineated	2
	Form	Protruding, convex	0
		Flat, even	1
		Concave, hollowed	2
Gills	Odour	Metallic	0
		Blood / Oily	1
		Rancid	2
	Color	Intense red colour	0
		Dark wine colour	1
		Opaque brownish wine colour to discoloured	2
Abdomen	Internal abdominal wall	Bright silver colour with black dots	0
		Bright mother of pearl colour with black dots	1
		Brightless yellowish white, with black dots	2
Muscles	Colour	Bright clear pink	0
		Opaque, old pink, “chicken thigh colour”	1
		Total quality index 0-19	

Quality index method (QIM)

For evaluation of samples with the QIM, selection and training of the team were done according to the methodology used by Sveinsdottir et al. (2003). Whole and raw fish, stored on ice during different time periods of 1, 8, 15 and 22 days were individually presented on a clear colour tray. The trained team, composed of nine evaluators, took part in the evaluation of samples using QIM scheme produced during the training sessions as presented in Table 1.

Determination of total volatile bases

For TVB analyses, 11 specimens of Nile tilapia with average weight of 376.3 g were used. TVB quantifications were done on 1, 4, 8, 11, 15, 18 and 22 days of storage, using the Conway microdiffusion dish method (Brasil, 1981).

Psychrotrophic and heterotrophic bacteria counting

For bacteriological analyses, 11 specimens of Nile tilapia with average weight of 376.3 g were used. Counting was performed on storage days 01, 04, 08, 11, 15, 18, 22 and 28 in muscle samples with or without skin. The methodology for psychrotrophic aerobic heterotrophic bacteria counting was according to the descriptions of Morton (2001) and Cousin et al. (2001). Standard count agar was used and sowed plates were incubated at 7°C for 10 days.

Table 2. Averages (X) and standard deviation (s_X) of intensity in the perception of odour, appearance, taste and quantitative descriptive analysis (QDA) texture in Nile tilapia (*O. niloticus*), eviscerated and stored in ice.

Attributes	Storage period ($X \pm s_X$)			
	1 day	8 days	15 days	22 days
Colour of flesh	0.6^{a} (±1.9)	2.0^{b} (±1.72)	5.6^{c} (±4.90)	10.5^{d} (±3.8)
Orange pigment (I)	0.05^{a} (±0.7)	0.1^{a} (±0.34)	0.8^{a} (±2.32)	2.8^{b} (±4.21)
Brightness (D)	12.4^{a} (±4.3)	11.9^{a} (±4.2)	9.2^{b} (±4.79)	4.0^{c} (±3.08)
Fresh water fish characteristic odour (D)	13.0^{a} (±3.4)	12.2^{a} (±3.9)	9.6^{b} (±5.17)	3.3^{c} (±4.51)
Sea water fish characteristic odour (I)	0.56^{a} (±2.9)	0.1^{a} (±2.35)	2.2^{b} (±4.05)	2.1^{b} (±3.31)
Rancid odour (I)	0.3^{a} (±1.4)	0.3^{a} (±0.82)	1.8^{b} (±3.57)	7.6^{c} (±6.05)
Fresh water fish taste (D)	12.8^{a} (±4.04)	11.8^{a} (±4.21)	7.8^{b} (±5.56)	-
Sea water fish taste (I)	0.6^{a} (±2.62)	1.3^{b} (±2.41)	2.2^{b} (±3.34)	-
Bitter taste (I)	0.2^{a} (±0.30)	0.2^{a} (±0.27)	1.1^{b} (±2.08)	-
Softness (D)	12.9^{a} (±2.88)	12.8^{a} (±2.32)	10.8^{b} (±3.36)	-
Juiciness (D)	12.5^{a} (±2.62)	12.3^{a} (±2.84)	9.9^{b} (±4.05)	-

$^{a, b, c,}$ averages on the same line followed by distinct letters are significantly different ($p<0.05$). (-) Analyses not performed on the 22nd storage day; (I) Undesired attribute; (D) Desired attribute.

Statistical analysis

For statistical treatment of QDA results, One-way ANOVA and Tukey's test ($p<0.5$) were used. Regression analyses were performed on TVB results and on bacterial counts, previously transformed into base 10 logarithms. All statistical tests were done by SAS statistical system (SAS Institute, Inc., 1985).

RESULTS AND DISCUSSION

Quantitative descriptive analysis (QDA)

The sensorial team, composed of nine evaluators, defined eleven sensory attributes of appearance, odour, taste and texture in order to describe the characteristics of cooked flesh of Nile tilapia, eviscerated and stored on ice for 22 days. Average values for perception intensities and the definition of each of the attributes are presented in Tables 2 and 3, accordingly.

Fish stored for 1 and 8 days did not present significant difference ($p>0.05$) in the attributes of "softness", "juiciness", "brightness", "fresh water characteristic odour" and "fresh water fish taste". During this storage period, attributes considered as desirable by the team, such as delicate and mild odour and taste characteristic of fresh water fish, together with softness, juiciness and bright attractive appearance were preserved. This demonstrates that, under adequate conditions, sensorial characteristics of tilapias are conserved in recently captured and stored fishes until the eighth day of storage with the alteration being slight traces of a smell of sea water fish noticed.

In fish stored for 15 days, the team observed loss in sensorial quality of flesh after cooking, presenting lower brightness, softness and juiciness intensity. In this storage period, attributes considered as undesirable began to be noticed, such as "sea water fish odour and taste", reminding of sea-smell, and traces of "bitter taste" and "rancid taste", related to fat oxidation.

Fish stored for 22 days that were analyzed only for appearance and odour, presented sharp undesirable attributes, especially rancid odour and the presence of an orange pigment, both associated with the oxidation process of fatty acids.

"Flesh colour" attribute was important for evaluating freshness in this species of fish as it gradually varies from the first to the last day of storage, from being milky white colour to a dark greyish colour. Besides the initial clear colour, delicate taste also stood out as an attribute of sensorial quality for the cooked flesh of this fish species.

Quality index method (QIM)

The team selected attributes which sensorially characterized Nile tilapia in the different storage periods. Based on this, a QI protocol was developed and used in sample analysis. With the average quality indexes of the different storage periods, a calibration curve was drawn (Figure 1).

The analyzed fish species initially presented bright skin, greyish colour and well defined stripes. Aspects observed in eyes, such as cornea transparency, pupil delineation and shape stood out in the evaluation of freshness. Besides those aspects, odour and gill colour were remarkable aspects that suffer significant alterations during the storage period. Loss of transparency and delineation of pupils were highlighted by the evaluation team, as well as the change in the shape from concave to convex. The gill colour, initially bright red, changed into a brownish wine shade. Gills, initially characterized by a metallic odour, changed to a blood smell with traces of oil and, finally, a rancid odour, then considered as undesirable by the evaluators.

Table 3. Descriptive vocabulary used in QDA of farmed Nile tilapia (*O. niloticus*) eviscerated and stored in ice.

Appearance attributes	**Definition**
Colour of flesh	Colour going from white to light brown during storage period, not considering dark flesh
Brightness	Limpidity of colour, varying from opaque to bright, represented on the scale by “no brightness” to “a lot of brightness”, accordingly
Orange pigment	Clear pigment, associated to fat oxidation
Odour attributes	**Definition**
Characteristic of fresh water fish	Strong fresh water fish odour; fresh water algae
Characteristic of sea water fish	Odour associated with fish stored for a long time in ice or beginning to deteriorate; sea smell
Rancid	Odour associated to deteriorated fat
Taste attributes	**Definition**
Characteristic of fresh water fish	Strong fresh water fish taste; fresh water algae
Characteristic of sea water fish	Taste associated with fish stored for a long time in ice or beginning to deteriorate
Bitter	Taste associated with rancidity – deteriorated fat (not consider dark flesh bitter taste)
Texture attributes	**Definition**
Softness	Force necessary to tear the flesh with the first bite
Juiciness	Amount of humidity in the mass liberated during mastication

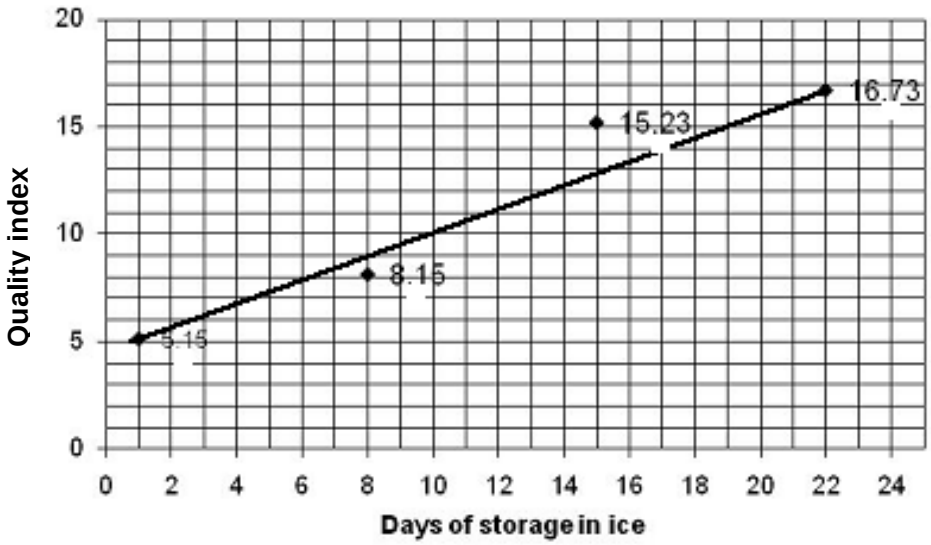

Figure 1. Calibration curve for quality index method of farmed Nile tilapia (*Oreochromis niloticus)* eviscerated and stored in ice for 22 days.

It was apparent based on these results that evaluators find it difficult in differentiating between samples of 15 and 22 days. Based on the results obtained here, it can be concluded that the most important sensorial alterations in Nile tilapia took place during this storage period, when evaluators were able to notice undesirable attributes in the samples obtained at 15 to 22 day of storage.

Similar results were obtained from trials performed by Netto (1984) and Guimarães et al. (1988) who employed

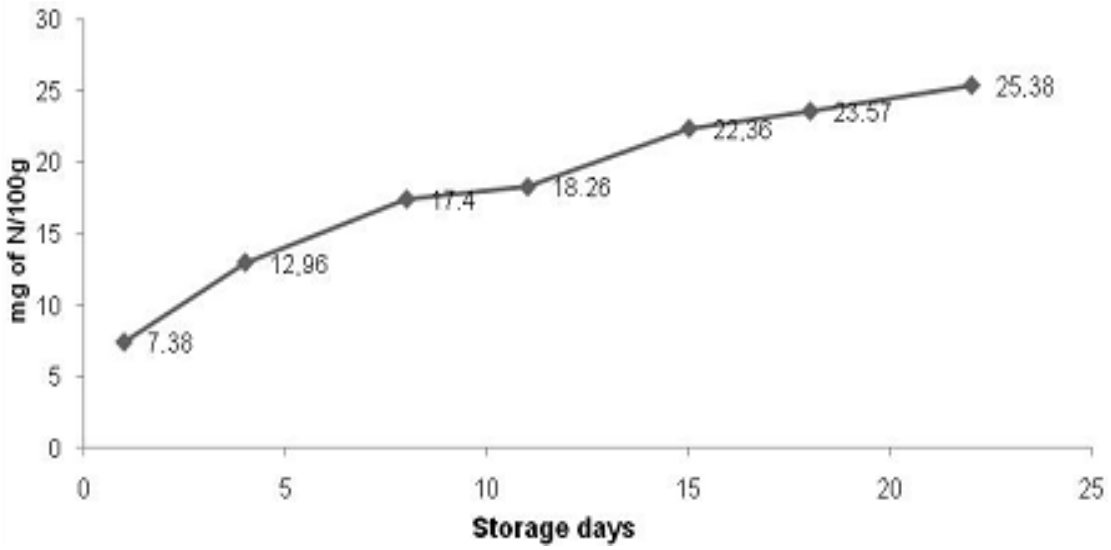

Figure 2. Results of determination of total volatile bases for farmed Nile tilapia (*O. niloticus*) eviscerated and stored in ice for 22.

methodologies that differed from those in the present research, by evaluating sensorial characteristics of whole hybrid tilapia (*Tilapia hornorum* x *O. niloticus*) and whole eviscerated Nile tilapia (*O. niloticus*). These authors observed that deterioration of the prepared fish samples reached unacceptable levels for consumption at 15 and 16 days of storage, respectively.

Albuquerque et al. (2004) used QIM and verified that Nile tilapias, desensitized by two different methods (CO_2 and ice) and stored for 17 days, showed optimal freshness until the storage day (7), developing more significant alterations between the 12 and 17 days of storage. Likewise, Soares and Gonçalves (2012) using QIM observed that the maximum life of the Nile tilapia fillet stored on ice was estimated at 15 days.

Comparing QIM results with QDA, it can be noticed that exactly in the 15 days storage period, Nile tilapia presented loss of sensorial quality. Hence, a QI between 0 and 8 indicates that fish quality can be guaranteed up to 8 storage days, QI between 9 and 15 indicates storage time between 9 and 15 days, and QI between 16 and 19 indicates storage above 22 days which is considered unsuitable for consumption.

Total volatile bases

With the results obtained in this research, it can be noticed that during storage (Figure 2) TVB value did not go beyond acceptable limits for the Brazilian legislation, which is 30 mg of N/100 g of flesh (Brasil, 1997a). Similar findings have been previously reported in trials with Nile tilapia. Guimarães et al. (1988), Sales et al. (1988), Elisabetta et al. (2001), Soccol (2002) and Albuquerque et al. (2004) observed low TVB values when fish were sensorially rejected, in concordance with Beraquet and Lindo (1985) and Contreras-Guzmán (1994), who reported that fresh water fish present low TVB. The need to re-evaluate the acceptable limits of this legal parameter for this species is thus demonstrated.

It was evident on day 4 of the storage as there was a pronounced increase in TVB value, a demonstration of the effect of biochemical events that reduce quality in the initial phases of storage, while bacteria counts are still low, however, their metabolites would be responsible for deterioration in the fish freshness in a second phase as reported by Contreras-Guzmán (1994).

Psychrotrophic aerobic heterotrophic bacteria counting

Figure 3 shows that psychrotrophic bacteria reached the exponential growth phase in samples with and without skin on the 28 day of storage, with respective values of log 9.40 and 7.90. Counts remained within the limits recommended by ICMSF (1986) of 107 UFC/g for aerobes total counts, until the day 18 of the storage in both samples.

According to the description by Huss (1997), tilapia fish is kept under good storage conditions, if bacterial counts results exceeded the acceptable limits only from the 22nd day of storage, and thus showing the good sanitary conditions under which the fishes were handled and kept. The present results was not in accordance with those obtained by Pullela et al. (1998), Martins et al. (2002) and Bartolomeu et al. (2011) who observed higher bacterial counts of above log 3.0 of the psychrotrophic bacteria in

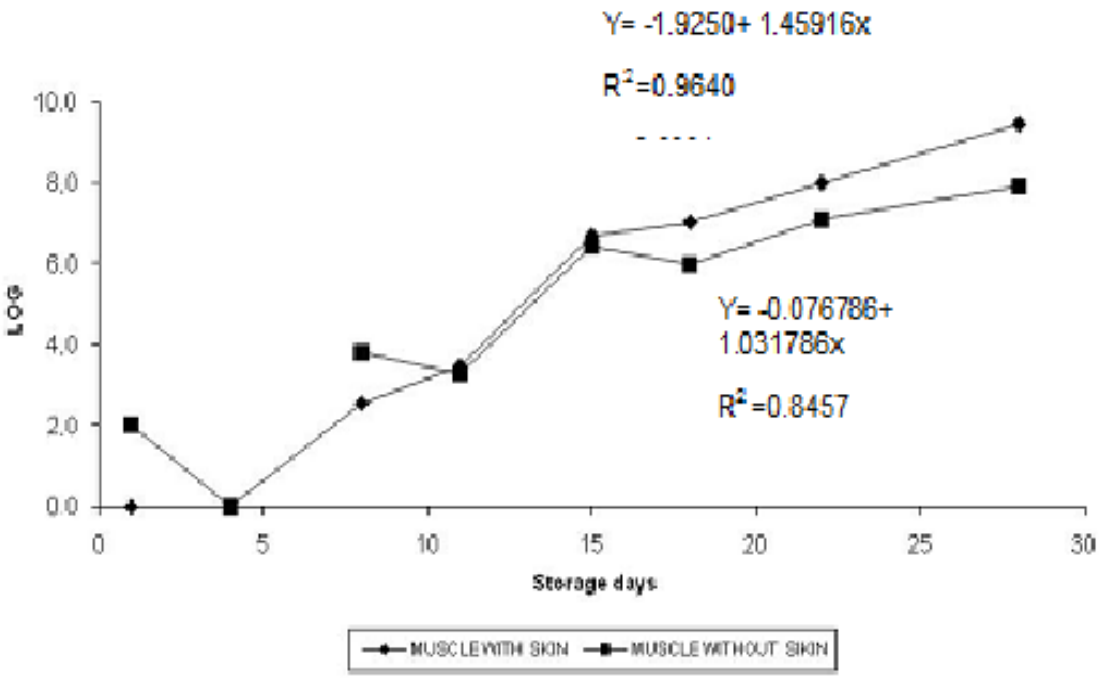

Figure 3. Logarithm results for the counting of psychrotrophic aerobic heterotrophic bacteria in farmed Nile tilapia (*O. niloticus*) eviscerated and stored in ice for 28 days.

a recently captured tilapia, while in the present work, bacterial counts approached these values only at the eighth day of storage on counts that reached this value.

Conclusions

Based on the results obtained with QIM, QIs between 0 and 15 were considered as acceptable values for consumption. Although bacteria counts remained within acceptable limits for human consumption until the 18th day, QDA showed an increase in the perception of undesired attributes from 15th day of storage. A shelf-life of 15 days is suggested for eviscerated Nile tilapia stored on ice.

Conflict of Interests

The authors have not declared any conflict of interests

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