Quality parameters of pacu (*Piaractus mesopotamicus*) and tambaqui (*Colossoma macropomum*) gutted and stored on ice for different periods

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Abstract

The purpose of this work was to assess the quality of pacu (*Piaractus mesopotamicus*) and tambaqui (*Colossoma macropomum*) gutted and stored on ice for a period of 19 days. Color, texture, pH, acid value, peroxide value, thiobarbituric acid reactive substances, total volatile bases and counts of aerobic mesophilic heterotrophic and psychrotrophic bacteria assessments were performed in the meat of tambaqui and pacu gutted and stored on ice for different periods. We concluded that tambaqui and pacu, when gutted and stored on ice, continue to be fit for consumption, up until the 18th and 11th day of storage, respectively.

Introduction

The Brazilian production of fish in 2010 was of 479 399 tons of farmed fish, which represents an increase of 15.3%, when compared to the 2009 production, of which 27% (337,353 tons) were represented by continental aquaculture (Brazil, 2012). Among the most farmed Brazilian native species are the fishes known as round fishes, among which the most important ones are pacu and tambaqui (Brazil, 2012). Production of these species, which was of 14,821 tons in 2001 (Borguetti *et al.*, 2003), reached 97.179 tons in 2010 (Brazil, 2010), showing an increase of 336% in almost ten years due to the adoption of more intensive management practices.

Due to fish production and consumption expansion, the quality of fresh fish has been a constant challenge for both industry and consumers (Mischke *et al.*, 2012). The degree of freshness characterizes the quality and safety of food, both for immediate consumption and for processing and subsequent commercialization. Bacteriological, physical, chemical and biochemical methods have been used to evaluate the freshness, quality and possible commercial validity of fish (Massa *et al.*, 2012; Borges *et al.*, 2013).

Therefore, the purpose of this study was to determine the quality parameters of the muscle of gutted freshwater fish species *Piaractus mesopotamicus* (Pacu) and *Colossoma macropomum* (Tambaqui), soon after capture and at different times of storage on ice, correlating such results to the bacteriological, physical and chemical analyses.

Material and Methods

Collection and storage of samples

Pacu (*Piaractus mesopotamicus*) and tambaqui (*Colossoma macropomum*), coming from the feeding tanks, were fished with a dragnet, on a farm located in the municipality of the state of Rio de Janeiro, Brazil, with appropriate criteria to minimize animal stress. A total of 18 individuals of each species were assessed. These animals were caught in three batches in the months of September, October and November 2010, respectively. In each batch, 6 specimens were collected from each species, aged from 8 to 10 months and weighing 2.0 ± 0.1 kg, for purposes of bacteriological, physical and chemical analyses.

The fishes were submitted to depuration for 24 to 36 hours. In the process of cleaning the fish kept in tanks suitable for this process step and in fast running water, according to the methodology described by Pillay (1974), after 24 h. The fish were placed in masonry reservoir with tap water and high flow. In this process the live fish undergo an external cleaning and digestive tract. Immediately after fished, were made numb, gutted manually and washed. They were then placed in isothermal boxes with ice (one kilogram of ice for every two kilograms of fish) coated with a thin protective plastic film and sent to the Laboratory. These containers were kept in a domestic refrigerator (average temperature 0.5ºC ± 0.5°C) for analytical procedures, and the scale ice was replaced daily for 19 days. Samples were removed from the dorsal muscle of one single fish. Each homogenized sample was analyzed in analytical triplicate, and the mean

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was calculated. The experiment was conducted in three different batches, featuring a triple experiment.
Bacteriological analysis

For the purpose of the bacteriological analysis, two portions of different areas of the fish muscle were collected with the aid of sterile forceps and scissors, forming a 10 g sample. Then, each portion was transferred under sterile conditions into the Stomacher envelope (Seward brand model 80, New York, 2008), and 90 mL of 0.1% w/v saline peptone water (110582 buffered peptone solution and sodium chloride, Frankfurter, Germany) was added. This solution was used to prepare other dilutions under sterile conditions for bacteriological analyses.

The methodology was applied three times in the muscle samples, both for Aerobic Heterotrophic Mesophilic Bacteria Count (AMHBC) and Aerobic Psychrotrophic Heterotrophic Bacteria Count (APHBC), according to Cousin *et al.* (2001). At the end of incubation, the plates with the dilutions that showed better conditions for colony count, containing 25 and 250 colony forming units (CFU), were selected (Swanson *et al.*, 2001). This procedure was repeated periodically over 19 days.

Physical and chemical analyses

All physical and chemical analyses were performed in analytical triplicate periodically for 19 days, on days 1, 4, 6, 8, 11, 13, 15, 17 and 19 of storage for two, both species of fish. The following analyses were performed according to the methodology proposed by AOAC (2000): pH (digital potentiometer method (Digmed DM 22 equipped with a glass electrode and calibrated with buffer solutions 4 and 7), determination of Total Volatile Bases (TVB-N) in meat (based on the micro-diffusion method), determination of acid value (AV) and peroxide value (PV).

Thiobarbituric acid reactive substances (TBARS) were determined using the methodology proposed by Tarladgis *et al.* (1960) as modified by Monteiro *et al.* (2012). Results were expressed as mg of malonaldehyde per kg of muscle (mg MA/kg).

To instrumentally determine the muscle color and brightness, color parameters L^* (luminosity ranges from 0 (black) to 100 (white)), a* (-a = green, + a = red) and b^* (-b = blue + b = yellow) were used, previously calibrated to a white tile standard, by using the colorimeter CR 400/410 (Minolta Co. Ltd., Osaka, Japan). Two fragments of the tambacu muscle fillet (50 mm diameter) were removed and stored on ice in metallized packaging. At the time of the analysis, fillets were exposed to the atmosphere at room temperature for a period of 30 minutes, after which the reading was performed on the surface of both sides of each piece of sample, totaling four readings, resulting in the average color and brightness of the day of storage, according to the methodology described by Macagnano *et al.* (2005).

The instrumental texture profile was performed in the TA-XT Express - Texture Technologies Corp.. texturometer (Stable Micro System Ltd., Vienna Court, UK) in triplicate, in which samples (20 mm diameter, 20 mm length) were subjected to the Texture Profile Analysis (TPA) Bourne (2002) to calculate hardness, adhesiveness, springiness, cohesiveness and resilience parameters which were automatically calculated using the Texture Expert® software. The conditions used were: a) pre-test speed = 1.0 mm/sec.; b) test speed = 1.0 mm/sec.; c) post-test speed = 1.0 mm/sec.; d) distance to which the device compressed the sample was 9.6 mm, equivalent to 40% compression, e) contact force = 5.0 N. The probe used was SMS P/36. All measurements were made at room temperature with the samples previously exposed to the atmosphere for a period of 30 minutes.

Statistical analysis

Descriptive measures expressed as mean and standard deviations were used for bacteriological, physical and chemical analysis parameters. Data for bacteriological analysis was expressed as log CFU/g.

For the results of quality attributes such as: pH, TVB-N, AMHBC, APHBC, peroxide value (PV), acid value (AV), TBARS, instrumental color parameters L^*, a^* and b^*, and instrumental texture parameters of hardness, adhesiveness, springiness, cohesiveness and resilience, Principal Component Analysis (PCA) was applied on the correlation matrix. Quality attributes (Y) and storage times (X) were analyzed in the covariance matrix. Pearson's correlation was used to determine the correlation between all quality attributes.

Also, linear regression analysis (Granato *et al.*, 2013) was used in relation to bacterial counts, physical and chemical values. Results were organized in tables with the means and standard deviations for each product. PCA statistical calculations were performed in the 2012.5 version of the XLSTAT program for Windows (Adinsoft, Paris, France.).

Results and Discussion

Correlations between bacteriological, physical and chemical analyses

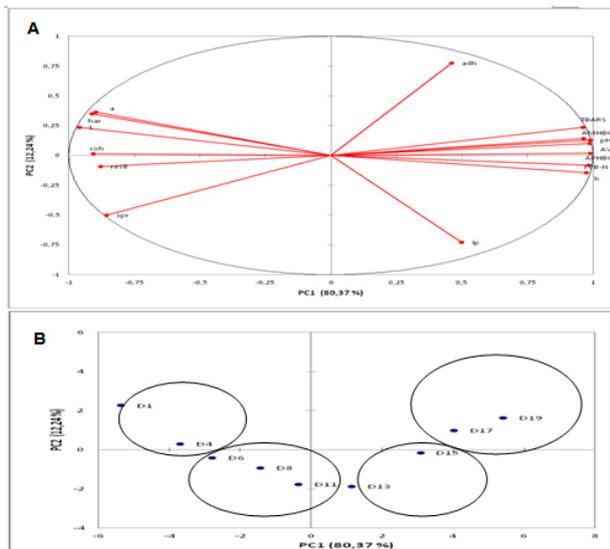Changes in the quality of tambaqui and pacu and

Figure 1. Principal Component Analysis applied to the results of quality parameters assessed in tambaqui (*Colossoma macropomum*) gutted and stored on ice: (A); pH; TVB-N = total volatile bases, L* = brightness, a*; b*; har = hardness, adh = adhesiveness; spr = springiness; coh = cohesiveness; resil = resilience; AV = acid value; PV = peroxide value; TBARS = thiobarbituric acid reactive substances; AMHBC = Aerobic Mesophilic Heterotrophic Bacteria Count and APHBC = Aerobic Psychrotrophic Heterotrophic Bacteria Count. (B) D1 = day 1, D4 = day 4, D6 = day 6, D8 = day 8, D11 = day 11, D13 = day 13, D15 = day 15, D17 = day 17 and D19 = day 19.
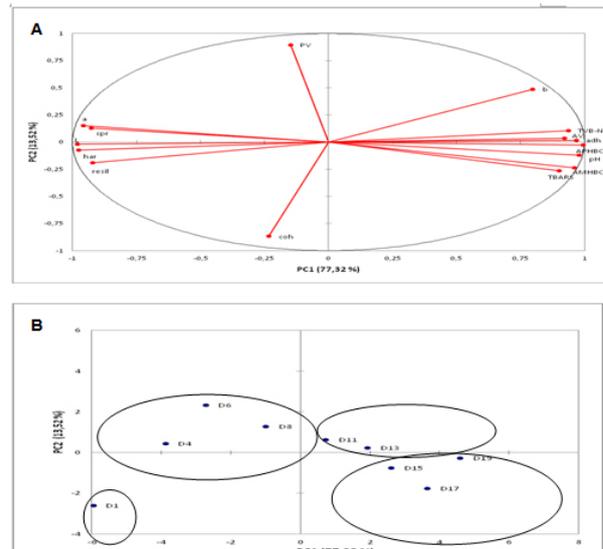

Figure 2. Principal Component Analysis applied to the results of quality parameters assessed in pacu (*Piaractus mesopotamicus*) gutted and stored on ice: (A); pH; TVB-N = total volatile bases, L* = brightness, a*; b*; har = hardness, adh = adhesiveness; spr = springiness; coh = cohesiveness; resil = resilience; AV = acid value; PV = peroxide value; TBARS = thiobarbituric acid reactive substances; AMHBC = Aerobic Mesophilic Heterotrophic Bacteria Count and APHBC = Aerobic Psychrotrophic Heterotrophic Bacteria Count. (B) D1 = day 1, D4 = day 4, D6 = day 6, D8 = day 8, D11 = day 11, D13 = day 13, D15 = day 15, D17 = day 17 and D19 = day 19.

the relationship between the assessed characteristics or parameters can be observed graphically with the use of Principal Component Analysis (PCA). However, in the PCA of this study, the correlation between the parameters pH, TVB-N, instrumental texture parameters, as to know, hardness, adhesiveness, springiness, cohesiveness and resilience and color parameters L, a* and b*, acid value, peroxide value, TBARS, mesophilic count (AMHBC) and psychrotrophic count (APHBC) was determined for each fish species. PCA was applied to the average value results.

In Figure 1A, it is possible to observe the PCA results of the variables used in evaluating the quality of tambaqui and Figure 1B represents the correlation of samples between different days of tambaqui storage on ice (days 1, 4, 6, 8, 11, 13, 15, 17 and 19). In PCA, assessed quality parameters are shown in these graphs represented as vectors, while samples are represented by dots. The first principal component (PC1) explained 80.37% of the variability and the second principal component (PC2) explained 12.24% of the total variability.

As far as pacu is concerned, PCA results of variables used in the quality assessment are shown in Figure 2A and Figure 2B represents the correlation of samples between different days of storage on ice

(days 01, 04, 06, 08, 11, 13, 15, 17 and 19). Therefore, these graphs explain 77.32% in PC1 and in PC2 explained 13:52%.

When Santos (2011) studied the bacteriological, physical and chemical quality parameters of *Cynoscion acoupa* gutted and stored on ice, he obtained results similar to this research, in which the first principal component (PC1) explained 76.32% of the total variability between samples of different storage periods and the second principal component (PC2) explained 14.04% of the total variability.

In PCA, the length of vectors corresponds to the relative importance of each quality parameter in the discrimination of samples, so long vectors suggest parameters in which samples differ more from each other. However, in this study, we can see, both in Figures 1A and 1B, that all vectors present very similar lengths, which means that their importance in the discrimination of samples was similar.

The spatial separation between samples is related to the degree of similarity or difference among them. For tambaqui, we can see, in Figure 1A, that the relative position between vectors also results in important information in PCA, thus close vectors indicate attributes that are likely to have a linear correlation between them. Quality parameters pH and TVB-N, instrumental color parameter b*, adhesion,

acid value, peroxide value, TBARS, mesophilic count (AMHBC) and psychrotrophic count (APHBC) confirm the existence of a positive linear correlation in PC1. However, there are vectors in PC1 which show a negative linear correlation between them, what happens in the quality attributes of resilience, springiness, hardness, cohesiveness, instrumental brightness L* and instrumental color a*. For pacu, Figure 2B shows a positive linear correlation in PC1 for attributes TVB-N, pH, AMHBC, APHBC, instrumental color b*, acid value, TBARS and adhesiveness. While the negative linear correlations in PC1 were related to the attributes peroxide value, springiness, resilience, hardness, cohesiveness, instrumental brightness and instrumental color a*.

Figures 1B and 2B show four groups of samples. Days 1 and 4 of tambaqui storage are included in negative PC1 quadrant and positive PC2 quadrant and day 01 of pacu storage is in the negative PC1 quadrant and negative PC2 quadrant. Samples stored on these days show similar quality attributes, which means that they are considered to be fresh samples. Day 6 and 8 tambaqui samples are included in negative PC1 and negative PC2 quadrants and days 4, 6 and 8 pacu samples are in negative PC1 and positive PC2 quadrants. This means that these samples are different from the others, and they are not considered fresh samples. Days 11, 13 and 15 tambaqui samples are included in positive PC1 and negative PC2 quadrants and days 11 and 19 pacu samples are in positive PC1 and positive PC2 quadrants, which indicates that they are similar to each other and different from the rest. Finally, tambaqui samples collected on storage days 17 and 19 are located in positive PC1 and positive PC2 quadrants and pacu samples collected on storage days 15, 17 and 19 are in positive PC1 and negative PC2 quadrants, standing out from the other samples.

As the statistical result of Multivariate Principal Component Analysis (PCA) in this research, Santos (2011), when he studied PCA in yellow weakfishes eviscerated and stored on ice, different storage days were well separated between PC1 and PC2 quadrants.

By relating Figure 1B's PCA with data obtained in this study, during the first days of tambaqui gutted and stored on ice (day 1 to day 4), all quality attributes remained unchanged. Changes began on day 6. However, mean values of all quality attributes varied significantly ($p < 0.05$) during the 19 days of storage on ice as described in Tables 1 and 2.

pH, AMHBC and APHBC values showed that species were unfit for consumption from the 19th day of storage, respectively, under the laws of the Brazilian Department of Agriculture (Brazil, 1997) of 6.5, the

Food Agriculture Organization (FAO, 1997) and the International Commission on Microbial Specifications for Foods (ICMSF, 1986) of 10^7 log CFU/g. TVB-N varied significantly ($p < 0.05$), from 8.82 to 13.86 mg per 100 g, but did not reach the limit that turns the product unfit for consumption according to the legislation of the Brazilian Ministry of Agriculture (Brazil, 1997) of 30 mg per 100 g of fish muscle. The value did not reach the limit because the precursor of the major volatile bases, monomethylamine, dimethylamine and trimethylamine, is trimethylamine oxide (TMAO) that is missing or appears in small amounts in freshwater fish (Leitão and Silveira, 1992; Rodrigues et al., 2013).

So, in this study, the authors stipulated the commercial validity of tambaqui (C. macropomum) gutted and stored on ice as day 18, using the bacteriological analyses and pH as criteria. This study is in agreement with Almeida et al. (2006) whose results on the research with tambaqui stored on ice showed a regular increase of muscle's pH and TVB-N with storage time but is not in agreement with the commercial validity period for human consumption conditions, which was set at 43 days.

Therefore, by relating figures 1A and 1B, we confirmed that tambaqui samples with over 18 days of storage on ice showed high pH, AMHBC and APHBC, adhesiveness, TBARS and instrumental color a* (the meat tends to be greenish).

As for pacu gutted and stored on ice, Borges et al. (2013) studied the commercial validity, through AMHBC, APHBC and pH, and set it to the 11th day. However, comparing the results found by these authors with the PCA results of this study in Figures 2A and 2B, pacu samples stored for more than 11 days on ice showed high pH, TVB, AMHBC and APHBC, adhesion, acid value, TBARS and instrumental color b* (the meat tends to be yellowish).

These results are not in accordance with the findings of Poulter and Nicolaides (1985) who determined that the commercial validity of pacu fished in rivers, gutted and stored on ice (Colossoma macropomum) was of 40 days. Tropical fish coming from its natural habitat should present a lower microbial load in comparison to the fish coming from excavated tanks, which have a high population rate and are given feed, which favors the reduction of the commercial validity of fish produced under farming conditions (Lima et al., 2011).

Given these facts, there are various factors to consider for the commercial validity of fish, including slaughter method, concentration of endogenous enzymes and initial microbial contamination, fish handling at the time of slaughter and storage

Table 1. mean scores and standard deviation of quality parameters, pH, TVB-N, AMHBC, APHBC, hardness, adhesiveness, springiness, cohesiveness, chewiness, resilience, acid value, peroxide value, TBARS, instrumental color L, a* and b* of tambaqui (*Colossoma macropomum*) gutted and stored on ice for 19 days.

PARAMETER	SAMPLE/DAY	1	4	6	8	11	13	15	17	19
pH	MÉAN (SD)	5.80 (0.03)	5.83 (0.03)	5.90 (0.01)	5.97 (0.04)	6.08 (0.01)	6.17 (0.01)	6.31 (0.06)	6.43 (0.04)	6.57 (0.03)
TVB	MÉAN (SD)	3.78 (1.26)	5.04 (1.26)	5.04 (0.00)	6.30 (0.00)	7.56 (2.18)	7.56 (1.26)	8.82 (0.00)	8.82 (0.00)	10.08 (0.72)
AMHBC	MÉAN (SD))	3.32 (0.55)	3.94 (0.55)	4.08 (0.12)	4.93 (0.85)	5.11 (0.52)	5.30 (0.64)	5.89 (0.38)	6.92 (0.63)	8.23 (0.04)
APHBC	MÉAN (SD)	1.81 (1.71)	2.84 (0.67)	3.17 (0.51)	3.94 (0.24)	5.30 (1.40)	5.91 (0.86)	6.70 (0.37)	7.35 (0.54)	9.22 (0.48)
ACID VALUE	MÉAN (SD)	0.02 (0.00)	0.04 (0.00)	0.08 (0.02)	0.24 (0.00)	0.36 (0.00)	0.45 (0.00)	0.60 (0.10)	0.70 (0.05)	0.95 (0.03)
PEROXIDE VALUE	MÉAN (SD))	0.0 (0.00)	0.01 (0.00)	0.04 (0.00)	0.08 (0.00)	0.16 (0.00)	0.16 (0.00)	0.16 (0.00)	0.08 (0.02)	0.12 (0.03)
TBARS	MÉAN (SD)	-	-	-	0.0889 (0.02)	0.0982 (0.01)	0.1567 (0.01)	0.2449 (0.01)	0.3798 (0.07)	0.3985 (0.11)
L	MÉAN (SD)	65.80 (1.04)	64.37 (0.41)	61.87 (1.03)	60.92 (0.65)	60.30 (0.55)	57.97 (0.91)	57.48 (0.77)	57.19 (0.59)	54.05 (0.42)
a*	MÉAN (SD)	3.70 (0.23)	2.50 (0.41)	1.93 (0.62)	1.87 (0.29)	1.82 (0.46)	1.29 (0.35)	1.28 (0.04)	1.17 (0.62)	1.02 (0.11)
b*	MÉAN (SD)	13.94 (0.77)	15.14 (0.06)	15.53 (0.41)	15.67 (0.46)	15.96 (0.27)	16.78 (0.07)	16.94 (0.16)	17.48 (0.33)	18.99 (1.48)
HARDNESS	MÉAN (SD)	1807.41 (895.75)	1724.59 (9.11)	1650.71 (38.32)	1473.57 (98.64)	1407.41 (360.56)	1408.55 (328.17)	1360.02 (116.61)	1351.94 (487.33)	1342.91 (48.81)
ADHESIVENESS	MÉAN (SD)	-46.00 (0.80)	-47.00 (0.74)	-47.00 (0.55)	-47.00 (0.95)	-47.00 (0.87)	-47.00 (3.25)	-49.00 (0.16)	-46.00 (0.99)	-46.00 (1.80)
SPRINGINESS	MÉAN (SD)	0.49 (0.04)	0.49 (0.00)	0.49 (0.00)	0.49 (0.00)	0.49 (0.01)	0.49 (0.06)	0.47 (0.01)	0.46 (0.06)	0.43 (0.04)
COHESIVENESS	MÉAN (SD)	0.55 (0.07)	0.50 (0.04)	0.50 (0.06)	0.49 (0.04)	0.46 (0.07)	0.45 (0.04)	0.45 (0.02)	0.43 (0.04)	0.35 (0.08)
RESILIÊNCE	MÉAN (SD)	0.16 (0.05)	0.16 (0.00)	0.16 (0.00)	0.16 (0.00)	0.16 (0.03)	0.13 (0.00)	0.13 (0.00)	0.13 (0.00)	0.13 (0.01)

Table 2. Models of average regression equation (y) of pH, TVB-N, mesophilic counts, psychrotrophic counts, acid value, peroxide value, TBARS, brightness, color a*, color b*, hardness, adhesiveness, springiness, cohesiveness and resilience in terms of days of storage on ice (x) and their respective coefficients of determination (R²) and probability levels (p > F) in gutted tambaqui.

Parameters	Regression model	R²	Prob > F
pH	y = 5.9260 + 0.0425.x	0.8857	0.0001
TVB	y = 2.9760 + 1.1214.x	0.9230	0.0001
AMHBC	y = 2.1513 + 0.7490.x	0.8210	0.0001
APHBC	y = 2.1513 + 0.7490.x	0.8210	0.0001
HARDNESS	y = 17774.4520 − 51.9812.x	0.8250	0.0003
ADHESIVENESS	y = −47.0000 − 0.0363.x	0.0144	0.7416
SPRINGINESS	y = 0.5093 − 0.0062.x	0.7564	0.0011
COHESIVENESS	y = 0.5586 − 0.0184.x	0.9331	0.0001
RESILIENCE	y = 0.1700 − 0.0045.x	0.7576	0.0011
ACID VALUE	y = 0.0452 + 0.1638.x	0.8953	0.0001
PEROXIDE VALUE	y = 0.3611 + 0.4300.x	0.6079	0.0132
TBARS	y = 0.8543 + 0.5746.x	0.9607	0.0001
L BRIGHTNESS	y = 66.5293 − 1.1962.x	0.9447	0.0001
COLOR a*	y = 3.2247 − 0.2765.x	0.8130	0.0009
COLOR b*	y = 14.4540 + 0.2912.x	0.7287	0.0017

conditions (Özogul and Özogul, 2004).

By associating the value of correlation between variables and the PC1 factor of tambaqui and pacu, variables with values greater than 95% (p < 0.05) were highlighted for purposes of identification of relevant attributes. In particular, for tambaqui, the most important quality attributes, with loading, were: pH (0.989), TVB-N (0.985), AMHBC (0.965), APHBC (0.990), acid value (0.987), instrumental brightness L (-0.961) and instrumental color b* (0.975). For pacu, pH, AMHBC, APHBC and adhesiveness showed higher significance (0.980, 0.960, 0.994 and 0.968, respectively) indicating a greater influence on correlations.

Pearson's correlation in PCA

Pearson's correlation coefficients were used in this study to measure the correlation between various quality attributes of tambaqui and pacu gutted and stored on ice, namely pH, TVB-N, mesophilic bacterial count (AMHBC), psychrotrophic bacterial count (APHBC), instrumental color and texture parameters, acid value, peroxide value and TBARS. However, twenty one significant correlations (p < 0.05) were found between the quality attributes of tambaqui and seventeen were found between the quality attributes of pacu. Nevertheless, there are relevant correlations that explain the lack of merit of certain attributes.

There were significant positive correlations in tambaqui between the AMHBC and APHBC results with: pH (respectively 0.979 and 0.989), TVB-N (0.952 and 0.986, respectively), acid value (respectively 0.978 and 0.992) and TBARS (respectively 0.967 and 0.952), positive correlations between pH and TVB-N (0.967) and between AMHBC and APHBC (0.978). Such correlations lead to the interpretation that as microbial load increases in fish during storage on ice, there is an increase of TVB-N and hence pH.

Authors like Geromel and Foster (1989), Burt and Hardy (1992) and Gonçalves (2006) state that it is clear that, soon after capture, fish undergoes a series of biochemical, physical, chemical and bacteriological changes that begin with the enzymatic action of the muscle. Therefore, this research shows that, with the increase of microbial load in fish, the action of aerobic mesophilic heterotrophic bacteria and aerobic psychrotrophic heterotrophic bacteria caused significant physical and chemical changes in fish stored on ice, leading to complete deterioration.

There also is a significant positive correlation (p < 0.05) between rancidity variables, as acid value and TBARS, and attributes related to the commercial validity of tambaqui (AMHBC, APHBC, pH and TVB-N). This means that, the the longest the storage time and the closest to the end of the commercial validity of tambaqui, the more hydrolytic rancidity of triglycerides and oxidative rancidity of unsaturated fatty acids is found in fish. Therefore, the following positive correlations with loadings were found: AV

x pH (0.996), AV x TVB (0.976), AV x AMHBC (0.978), AV x APHBC (0.992), AV x TBA (0.977), TBA x pH (0.983), TBA x AMHBC (0.967) e TBA x APHBC (0.952).

The results of this study corroborate those reported by Perez-Alonzo *et al.* (2003) who proved that *Brama brama* species stored on ice was gradually damaged by hydrolytic and oxidative rancidity until the product quality was compromised after 19 days of storage.

There was a positive correlation between instrumental color b* and APHBC (0.955), TVB-N (0.956) and a negative correlation between instrumental color b* and instrumental brightness L (-0.976). When APHBC and TVB-N increase, the b* value increases, indicating a tendency to yellow color of tambaqui meat and there is a meat brightness decrease. The meat color, resulting from the presence of various pigments, may be affected by biological factors such as growth of microbial load, muscle pH changes, lipid oxidation, muscle temperature, relative moisture (Cichoski and Terra, 1996). The color is critically evaluated by consumers, and is often the fundamental basis for acceptance or even rejection, being used as an attribute for quality and freshness of fish (Macagnano *et al.*, 2005).

Regarding pacu, there was a significant positive correlation between AMHBC and APHBC (0.961) and between these attributes and pH (0.977 and 0.987, respectively). Hardness, springiness and adhesiveness correlated significantly (p < 0.05), with bacterial counts, with respective loadingS -0.960, -0.962 and 0.974. This demonstrates that the process occurs in cascade with the increase of mesophilic bacteria, as they produce alkaline metabolites (TVB, ammonia and amines) that increase the pH of meat, leading to the denaturation of proteins which, consequently, reduce the water retention capacity of the food matrix. These physical and chemical changes alter the texture of meat, and the sensory quality tends to decrease. These results confirm the principles described by Hyldig and Nielsen (2001) who associate the texture of fish meat with pH and microbe increase. They reported that, when pH increases, the meat is softer, less elastic and non-arranged, while the meat of the fish freshly caught has low pH and is still firm.

As far as pacu is concerned, there were no significant correlations between attributes related to rancidity and the other quality attributes used in this research. This means that the bad odor that results as the pacu storage time increases is related to the formation of nitrogen compounds coming from protein degradation due to the increase of AMHBC and APHBC (Huss *et al.*, 2004) and not to the process of lipid rancidity.

There was a significant negative correlation (p < 0.05) between bacterial count and instrumental brightness L (-0.977) and instrumental color a* (-0.960) and a positive correlation between a* x L (0.972). With these data, it was possible to determine that the higher the bacterial count in fish due to storage time on ice, the less bright and the more greenish the meat of this fish is.

Pavlidis *et al.* (2006) studied the effect of storage time on the coloration of fish species *Pagrus pagrus*, *Pagrus caeruleostictus* and *Dentex gibbosus* bred in captivity and captured. They showed that the ice storage time slightly affected the color of the dorsal area of all fishes and that there also was a strong reduction in color from day 3 to day 7 in both the dorsal and ventral skin. These authors concluded that, the longer the storage period, the more changes the color of the fish in all parts of the animal body. The same thing happened in this study in which pacu muscle significantly lost brightness and instrumental color a*, ranging respectively from 69.48 to 59.50 and from 2.49 to 0.81.

Conclusions

Based on the results of the various quality parameter analyses, and especially the results of bacteriological and pH analyses, we may conclude that tambaqui (*Colossoma macropomums*), when gutted and stored on ice, remains fit for consumption until the 18th day of storage, and can be eaten without any risk to the health of consumers.

Based on the comparisons with other articles on the subject, the results of the various quality parameter analyses, and especially the correlation between the analyses of the findings of this study, we may conclude that pacu (*Piaractus mesopotamicus*), when gutted and stored on ice, remains fit for consumption up to the 11th day of storage, and can be eaten without risk to consumers' health.

References

Almeida, N. M., Batista, G. M., Kodaira, M. and Lessi, E. 2006. Alterações post-mortem em tambaqui (*Colossoma macropomum*) conservados em gelo. Ciência Rural 36: 1288-1293.

Borges, A., Conte-Junior, C. A., Franco, R. M. and Freitas, M. Q. 2013. Quality Index Method (QIM) developed for pacu *Piaractus mesopotamicus* and determination of its shelf life. Food Research International 54: 311–317.

Borghetti, N. R .B., Ostrensky, A. and Borghetti, J. R. 2003. Aquicultura: uma visão geral sobre a produção

de organismos aquáticos no Brasil e no mundo. Curitiba: Grupo Integrado de Aquicultura e Estudos Ambientais (GIA).

Bourne, M. C. 2002. Food texture and viscosity: concept and measurement. 2nd ed. London: Academic Press.

Brazil 1997. Portaria 185 de 13 de maio de 1997, que aprova o Regulamento Técnico de Identidade e Qualidade de peixe fresco (inteiro e eviscerado). Dowloaded from *http://www.cda.sp.gov.br/www/legislacoes/popup. php?action=view&idleg=670.*

Brazil 2010. Consumo Per Capita Aparente de Pescado no Brasil 1996-2009. Dowloaded from *http://www.sepaq. pa.gov.br/files/u1/ anuario_da_pesca_completo.pdf.*

Brazil 2012. Boletim Estatístico da Pesca e Aquicultura 2012. Dowloaded from *http://www.mpa.gov.br/mpa/ seap/Jonathan/mpa3/ docs/folder%20consumo%20.*

Burt, J. R. and Hardy, R. 1992. Pelagic fish. London: Fishing news book.

Cichoski, A. J. and Terra, N. N. 1996. Características sensoriais em carne. Higiene Alimentar 10: 32-43.

Cousin, M. A., Jay, J. M. and Vasavada, P. C. 2001. Psychrotrophic microorganisms. In: Dowens, F. P. and Ito, K. (Eds.). Compendium of Methods for the Microbiological Examination of foods, p. 159-164. Whashigton: American Public Health Association – APHA.

FAO 1997. Aseguramiento de la calidad de los productos pesqueros. Documento tecnico de pesca 334. Dowloaded from: *http://www.fao.org/DOCREP/003/ T1768S/T1768S00.HTM.*

Geromel, E. J. and Forster, R. J. 1982. Princípios fundamentais em tecnologia de pescado. São Paulo: Fundação Tropical de Pesquisas e Tecnologia.

Gonçalves, A. A. 2006. Ocorrência de off-flavor em pescado: um problema a ser resolvido em peixes marinos. Revista Aquicultura e pesca 18: 30-31.

Granato, D., Araujo, V. M. and Jarvis, B. Observations on the use of statistical methods in food science and technology. DOI: 10.1016/ j.foodres.2013.10.024

Huss, H. H., Ababouch, l. and Gram, P. 2004. Assessment and management of seafood safety and quality. Fisheries Technical Paper, No. 444. Rome: FAO.

Hyldig, G., Nielsen, D. 2001. A review of sensory and instrumental methods used to evaluate the texture of fish muscle. Journal Texture Studies 32: 219–242.

ICMSF (International Commission on Microbial Specifications for Foods) 1986. Microorganisms in Foods. 2. Sampling for microbiological analysis: Principles and specific applications. 2nd ed. New York: Blackwell Scientific Publications.

Leitão, M. F. M. and Silveira, N. F. 1992. Influência da temperatura ambiental na natureza e potencial deteriorador da microbiota bacteriana de peixes em ambientes lacustres Tropicais. Coletânea ITAL 23: 85-96.

Lima, R. P., Paludo, D., Soavinski, R. J., Silva, K. G. and Oliveira, E. M. A. 2011. Levantamento da distribuição, ocorrência e status de conservação do Peixe-Boi Marinho (*Trichechus manatus*, Linnaeus, 1758) no litoral nordeste do Brasil. Natural Resources 1: 41-57.

Macagnano, A., Careche, M., Herrero, A., Paolesse, R., Martinelli, E., Pennazza, G., Carmona, P., D'amico, A. and Di Natale, C. 2005. A Model to predict fish quality from instrumental features. Sensors and Actuators B 111: 293–298.

Massa, A. E., Manca, E. and Yeannes, M. 2012. Development of Quality Index Method for anchovy (*Engraulis anchoita*) stored in ice: Assessment of its shelf-life by chemical and sensory methods. Food Science and Technology International 18: 339-351.

Mischke, C. C., Greenway, T. E., Griffin, M. J. and Wise, D. J. 2012. Effects of Fry Age-at-Stocking on Growth and Survival of Channel Catfish. Journal of the World Aquaculture Society 43: 135–139.

Monteiro, M. L. G., Mársico, E. T., Teixeira, C. E., Mano, S. B., Conte Junior, C. A. and Vital, H. C. 2012. Validade comercial de filés de Tilápia do Nilo (*Oreochromis niloticus*) resfriados embalados em atmosfera modificada e irradiados. Ciência Rural 42: 737-743.

Özogul, Y. and Özogul, F. 2004. Effects of slaughtering methods on sensory, chemical and microbiological quality of rainbow trout (*Onchorynchus mykiss*) stored in ice and MAP. European Food Research Technology 219: 211-216.

Pavlidis, M., Papandroulakis, N. and Divanach, P. A. 2006. method for the comparison of chromaticity parameters in fish skin: preliminary results for coloration pattern of red skin Sparidae. Aquaculture 258: 211–219.

Perez-Alonso, F., Arias, C. and Auborg, S. P. 2003. Lipid deterioration during chilled storage of Atlantic pomfret (*Brama brama*). Journal of Lipid Science and Technology 105: 661-667.

Pillay, T. V. R. 1974. Aquaculture and fishery products development. In Kreuzer, R., Surrey. and W. B. (eds.). Fishery products. p. 83-98. Rome: Fishing News (Books) Ltd.

Poulter, N. H. and Nicolaides, L. 1985. Studies of the iced storage characteristics and composition of a variety of Bolivian freshwater fish. Journal Food Technology 20: 451-465.

Rodrigues, B. L., Álvares, T. S., Costa, M. P., Sampaio, G. S. L., Lázaro de la Torre, C. A., Mársico, E. T. and Conte Junior, C. A. 2013. Concentration of biogenic amines in rainbow trout (*Oncorhynchus mykiss*) preserved in ice and its relationship with physicochemical parameters of quality. Journal of Aquaculture Research & Development 4: 174-178.

Santos, A. P. B. 2011. Índices químicos, sensoriais e microbiológicos para avaliação do frescor de pescada amarela (*Cynoscion acoupa*) armazenada em gelo. Pirassununga, Brazil: Universidade de São Paulo, MSc thesis.

Swanson, K .M. J., Petran, R. L. and Hanlin, J. H. 2001. Culture Methods for Enumeration of Microorganisms. In: Dowens, F. P. and Ito, K. (Eds.) Compendium of Methods for the Microbiological Examination of foods, 4º ed, p. 387-409. Washington: American Public Health Association – APHA.

Tarladgis, B. G., Watts, B. M., Younathan, M. T. and Dugan, L. A. 1960. Distillation method for the quantitative determination of malonaldehyde in rancid foods. The Journal of the American Oil Chemists' Society 37: 403.