

## COMPARATIVE ASSESSMENT OF QUALITY OF TILAPIA (*Oreochromis niloticus*) AND CATFISH (*Clarias gariepinus*) STORED IN ICE FISH BOX

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Received: 06-04-16

Accepted: 27-07-16

### ABSTRACT

The quality of two species of fish, (Tilapia) *Oreochromis niloticus* and (Catfish) *Clarias gariepinus* stored in the Ice fish box was assessed with fish to ice (1:1, 2:1, 3:1 and 4:1) for 36 and 30 hours respectively. The parameters assessed were: PH, Rigor Index, TVB-N, Sensory Evaluation, Temperature, Relative Humidity and Microbial Analysis. The lowest average temperature and relative humidity for all the treatments were 5.7<sup>0</sup>C and 87.7%, and 5.2<sup>0</sup>C and 93.3% for Tilapia and Catfish respectively. Biochemical analysis revealed pH value for Tilapia decreased significantly ( $p < 0.05$ ) within the storage period for all the treatments. From an initial value of 6.90 to between 6.50-6.80 at the end of storage period (36 hours), for Catfish there was a significant difference ( $p < 0.05$ ) from an initial of 7.1 to 7.6. This increase was steady across all treatment at the end of the storage period (30 hours). The TVB-N value for Tilapia increased significantly within the storage period for all the treatments from an initial value of 5.29mg to 7.22mg, 8.82mg, 12.10mg and 13.28mg respectively for 1:1, 1:2, 1:3, 1:4. Similar trend was reported for catfish from an initial value of 0.98mg to 16.56mg. These values were significantly different ( $p < 0.05$ ). The microbial load did not increase significantly for all the treatment within the storage period. Total Viable Count (TVC) ranged from an initial of  $1 \times 10^2$  to  $3 \times 10^2$  cfu/g and an initial of  $1 \times 10^2$  to  $4 \times 10^2$  cfu/g for Tilapia and Catfish respectively. The sensory assessment and organoleptic evaluation of both species were satisfactory at the end of the storage period. However, overall assessment indicates that Tilapia stored longer in the Ice fish box than the Catfish.

**Key words:** Organoleptic assessment, Storage, Quality, Fish, Ice

### INTRODUCTION

Fish farming development has been viewed as a measure of increasing and improving food security and as a means of supplementing income to families. Aquaculture in Africa is carried out at subsistence level with little or no surplus production to be sold at the rural market (Banze and Oddsson, 2005). In Nigeria with an estimated population of 160million

people, the fish protein demand is put at 1.5million metric tonnes as against the current production of 500 metric tonnes (Raufu et al. 2009).

FAO (1999) described catfish as a disease resistant fish species which can tolerate extreme environmental conditions. This has contributed to increase in production in both commercial and subsistence scale in

Nigeria. Tilapia is an important fish in Nigeria due to its prolific nature and high market demand.

Post-harvest fish losses are a major concern and occur in most fish distribution chains throughout the world. Not only do losses constitute lost income to fisher folks, processors and traders but they also contribute to food insecurity – a loss of fish means less fish available for the consumer. (Yvetteet *al.* 2011).

Small-scale fisher folks account for more than half of total fish production in the world. The sector is a major source of food fish, income and employment to many millions of people, especially in developing countries. Despite their importance in terms of poverty alleviation and food security, they face a host of challenges among which is post-harvest fish losses. (Shinkafi, *et al.* 2010).

Reducing losses is therefore an important development goal in the fish industry. These losses are often caused by biochemical and microbiological spoilage changes that occur in fish after death. A live fish has natural defence mechanisms that help to prevent spoilage. However, once a fish dies, its defence mechanisms stop and enzymatic, oxidative and microbiological spoilage begins to cause quality deterioration. Fish enter rigor motis when ATP levels in muscle reach a minimum after death and myosin and actin are interconnected irreversibly (Huss, 1995).

To ensure the product quality is retained during production and storage, it is important to have insight into specific issues such as rigor motis, TVBN, PH, Sensory evaluation and microbial analysis. To

prevent losses associated with these factors, Ice-Fish Box was designed and fabricated by Nigerian Stored Products Research Institute (NSPRI) for transportation and storage of fish in fresh form. This study therefore, assessed the quality of two common species of fish; Tilapia (*Oreochromis niloticus*) and Catfish (*Clarias gariepinus*) stored in the Ice-Fish Box.

## **MATERIALS AND METHODS**

100kg each of live Tilapia and Catfish were obtained from a farm in Ijebu-Ode, Ogun State Nigeria during early hours (7.00-8.00 am) and was immediately transported in a fish tank to the laboratory. They were weighed and divided into lots according to sizes. The average weight was 800g and the length was 30 cm. Ten samples Catfish were randomly selected and after cold shock, the fish were stored in ice in an insulated box (530mm×380mm×270mm) in four treatments (fish: ice treatment 1:1; 2: 1; 3:1 and 4:1). Each box can accommodate weights of up to 18kg. The treatments were in three replicates. The fish boxes were cleaned beforehand for the experiment. Samples were obtained from the ice box at six hours interval for the four treatments. The samples obtained were used for visual examination, organoleptic assessment, biochemical tests, microbiological assessment and rigor index. The same procedures were followed for Tilapia (100kg). The quality of the fish was assessed with the following parameters: pH, Rigor Index, TVB-N, Sensory Evaluation, Temperature, Relative Humidity and Microbial Analysis.

### **pH**

pH was determined according to the method of Abelti (2013) with slight modification.

Ten grams of *Clarias gariepinus* flesh from the upper, middle and lower region were blended, homogenized in 50 mL of distilled water and the mixture filtered using Whatman Filter Paper No 1. The homogenate was allowed to attain a room temperature after 5 minutes. The pH was measured using a Jenway 3310 pH meter at room temperature after calibration using standard buffers of pH 7 and 4. The readings were carried out at the initial stage (fresh) and at various intervals of six hours for the period of the experiment for various treatments of fish to ice (1:1, 2:1, 3:1 and 4:1)

### **Rigor Index**

Rigor Mortis Index calculation was carried out according to Adoga *et al.*, (2010). Fish not exceeding a total length of 30 - 35cm were used for the experiment. This is to give room for proper determination of deflection length in the course of the measurement of rigor- index. Pre-rigor measurements were immediately determined shortly after death. Rigor-index was measured at selected time intervals (within 28 hours) from each treatment. Rigor index was measured by placing the fish on a table with half of its body (tail part) kept out of the table. The following equation was used to calculate the Rigor Index.

$$\text{Rigor index} = \frac{D - D1}{D} \times 100$$

Where D represents the distance of the base of the caudal fin from horizontal line of the table in pre-rigor state and D1 represents the distance of the base of the caudal fin from horizontal line of the table during rigor state

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### **Total Volatile Base – Nitrogen (TVB-N)**

TVB-N was determined using the methods of Antonacopoulos (1968); FAO/WHO/Codex Alimentarius (1968) ; Jinadasa(2014). Ten grams of blended *Clarias gariepinus* flesh was homogenized with 20 ml of 7.5% trichloroacetic acid for 2 minutes. The homogenized flesh was filtered through Whatman Number 1 filter paper to obtain clear extract. 25 ml of the extract was made alkaline with 6 mls of 10 % NaOH solution and 20 mls of distilled water. The solution was quantitatively transferred into the distillation tube and placed in the distillation flask of semi auto distillation apparatus. The receiving flask contained 25 ml of 4% boric acid and few drops of mixed indicator (methyl red/methylene blue 2:1). The steam distillation procedure continued until 100 ml of distillate had been collected. The obtained basic solution was titrated against

0.05M H<sub>2</sub>SO<sub>4</sub> to the endpoint indicated by a green to pink colour change. The TVB-N content was determined after blank correction, which was also determined by steam distillation with 25 ml of distilled water sample. TVB-N content was expressed as mgN/100 g of fish flesh.

$$\text{TVB - N (mg /100g)} = 14 \text{ mg mol} \times a \times b \times 300 / 25\text{mL}$$

Where: a = volume of sulphuric acid used (ml) b = molarity of sulphuric acid

### **Sensory Evaluation (Raw)**

Quality of the stored fish was determined using visual examination based on Quality Index Method (QIM). Whole fish samples were examined physically for general appearance of skin, consistency of flesh, odour and colour of the gills, colour and form of eyes. QIM is based on the significant sensory parameters for raw fish when using many parameters and a score system from 0 to 4 demerit points. QIM is using a practical rating system, in which the fish is inspected and the fitting demerit point is reached. The scores for all the characteristics are then summed up to equal to an overall sensory score, the so called quality index (Jonsdottir, 1992).

### **Sensory Evaluation (cooked)**

Sensory evaluations were conducted on the cooked samples. Attributes such as flavour, odour, taste, texture, and appearance were evaluated by five trained panellists for all the treatments during the storage period. A 5 point (Values 5-1) Likert's Scale (Scale of Summated Rating) was used for the scoring procedure. A cut-off point of 3 was established, above the cut-off point was regarded as good and below was regarded poor.

### **Temperature and Relative Humidity Measurement**

A Tinytag Ultra 2 digital data logger was used in monitoring the temperature and relative humidity (R/H) of the loaded fish boxes in different treatments and the ambient. They were calibrated before each use.

### **Microbial analysis**

Initial sample was taken and cultured using serial dilution. Total viable counts (TVC) were determined on Nutrient agar for bacteria for 18-24 hours at 37°C for bacteria, Eosin Methylene Blue(EMB) for *E.coli*, Salmonella shigella agar (SSA) for *Salmonella shigella spp*, Mannitol salt agar (MSA) for *Staphylococcus spp.*, MacConkey agar (MCA) for coliforms and Potato dextrose agar (PDA) for mould/fungi in depth for 72hrs at 37°C. All the reagents are Oxoid Grade England. Microbiological data were expressed as number of colony-forming units per gram (cfu/g). Distinct and discrete colonies from each plate were then picked by means of a sterile wire loop and sub-cultured on a freshly prepared media in order to get it in pure form. The pure isolates were subjected to gram techniques in order to ascertain their gram reaction (Positive or Negative). The isolates were further subjected to biochemical, microscopically and sugars fermentation test (Sneath *et al.* 1986 ; Basby *et al.* 1998).

### **Statistical Analysis**

The various treatments, period of storage, microbial and chemical analyses results were subjected to statistical analysis using one way analysis of variance (SPSS 20). The correlation between different parameters in the study was carried out by the Pearson's test.

## RESULTS

**Table 1:** Quality Index Method (QIM)

Attributes	Scores
<b>General appearance</b>	
Skin 0 - bright shining, 1 - bright, 2 - dull.	
Blood spot on gill cover	0 - none, 1 - small, 2 - big, 3 - very big.
Stiffness	0 - stiff, 1 - elastic, 2 - firm, 3 - soft.
Belly	0 - firm, 1 - soft, 2 - belly burst.
Smell	0 - fresh, 1 - neutral, 2 - musty/sour, 3 - stale meat/rancid.
<b>Eyes:</b>	
Clarity	0 - clear, 1 - cloudy.
Shape	0 - normal, 1 - plain, 2 - sunken.
<b>Gills:</b>	
Colour	0 - characteristic red, 1 - faded, discoloured.
Smell	0 - fresh, seaweed/metallic, 1 - neutral, 2 - sweaty/slightly rancid, 3 - sour stink/stale, rancid.

**Table 2:** pH of four treatments of *Oreochromis niloticus* in NSPRI ice fish box during the storage period

Duration							
Treatments	0Hr	6Hrs	12Hrs	18Hrs	24Hrs	30Hrs	36Hrs
1:1	6.90 <sup>a</sup>	6.90 <sup>a</sup>	6.75 <sup>b</sup>	6.50 <sup>c</sup>	6.60 <sup>c</sup>	6.80 <sup>ab</sup>	6.80 <sup>ab</sup>
1:2	6.90 <sup>a</sup>	6.81 <sup>ab</sup>	6.60 <sup>c</sup>	6.70 <sup>ab</sup>	6.60 <sup>ab</sup>	-	-
1:3	6.90 <sup>a</sup>	6.80 <sup>b</sup>	6.50 <sup>c</sup>	-	-	-	-
1:4	6.90 <sup>a</sup>	6.70 <sup>b</sup>	6.50 <sup>c</sup>	-	-	-	-

**Table 3:** pH of four treatments of *Clarias gariepinus* in NSPRI ice fish box during the storage period

Duration							
Treatments	0Hr	6Hrs	12Hrs	18Hrs	21Hrs	24Hrs	30Hrs
1:1	7.10 <sup>b</sup>	6.30 <sup>c</sup>	6.90 <sup>d</sup>	7.01 <sup>b</sup>	7.10 <sup>a</sup>	7.50 <sup>a</sup>	7.60 <sup>a</sup>
1:2	7.10 <sup>c</sup>	6.50 <sup>d</sup>	7.20 <sup>c</sup>	7.50 <sup>b</sup>	7.70 <sup>a</sup>	-	-
1:3	7.10 <sup>b</sup>	6.40 <sup>c</sup>	7.30 <sup>a</sup>	-	-	-	-
1:4	7.10 <sup>a</sup>	6.40 <sup>b</sup>	-	-	-	-	-

**Table 4:** TVB-N of four treatments of *Oreochromis niloticus* in NSPRI ice box during the storage period

Duration							
Treatments	0Hr	6Hrs	12Hrs	18Hrs	24Hrs	30Hrs	36Hrs
1:1	0.98 <sup>g</sup>	7.50 <sup>f</sup>	10.74 <sup>e</sup>	13.25 <sup>d</sup>	13.64 <sup>c</sup>	16.37 <sup>b</sup>	16.56 <sup>a</sup>
1:2	0.98 <sup>e</sup>	8.02 <sup>d</sup>	12.18 <sup>c</sup>	15.81 <sup>b</sup>	17.92 <sup>a</sup>	-	-
1:3	0.98 <sup>c</sup>	8.77 <sup>b</sup>	12.91 <sup>a</sup>	-	-	-	-
1:4	0.98 <sup>c</sup>	8.82 <sup>b</sup>	13.13 <sup>a</sup>	-	-	-	-

**Table 5:** TVB-N of four treatments of *Clarias gariepinus* in NSPRI ice box during the storage period

Duration							
Treatments	0Hr	6Hrs	12Hrs	18Hrs	21Hrs	24Hrs	30Hrs
1:1	5.29 <sup>e</sup>	5.40 <sup>d</sup>	5.46 <sup>e</sup>	7.04 <sup>f</sup>	9.19 <sup>c</sup>	12.65 <sup>b</sup>	13.28 <sup>f</sup>
1:2	5.29 <sup>e</sup>	5.43 <sup>d</sup>	6.55 <sup>c</sup>	7.67 <sup>b</sup>	12.10 <sup>a</sup>	-	-
1:3	5.29 <sup>c</sup>	6.99 <sup>b</sup>	8.82 <sup>a</sup>	-	-	-	-
1:4	5.29 <sup>a</sup>	7.22 <sup>a</sup>	-	-	-	-	-

## DISCUSSION

Changes in the catfish were observed as early as 1 hour. The three observed treatments showed a rigor index of about 25% two hours after death. The fourth treatment (4:1) was excluded further from the experiment because the ice melted before death could be achieved. Since the fish were placed alive in the fish boxes and left to die by cold shocking, a considerable amount of stress was experienced. Rigor index increased gradually with the passage of time to about 57% after 6 hours. This was the highest rigor index obtained in the course of the experiment. This is not in tandem with reports by Ihuahi, *et al*(2010) who reported a rigor index of about 78% in ice storage after spiking the fish, this could be attributed to different methods of killing employed in both work.

The rigor index (51%) was maintained for another 2 hours before relaxation. Almost complete relaxation (Thaw Rigor) was

achieved after 18 hours with an index of 23% without any sign of spoilage. This study revealed variations in the treatments. The rigor index obtained in treatment 2 with mean fish to ice ratio of 2:1 was 65%. Fish to ice ratio of 1:1 followed closely with the rigor index of 57%. In the last treatment (3:1) the rigor index was about 50%. All of these were recorded 6 hours after death. Full rigor (100%) was not recorded in this experiment.

In *Tilapia*, full (100%) rigor was observed in most samples in this study at four hours. Hossain *et al.*(2005) observed full rigor in their study. Rate of rigor was not found to be significant in all treatment as they all entered into rigor regardless of the weight of ice utilised.

### Sensory evaluation for *Tilapia* (fresh):

Quality Index (QI) of the attributes had an initial score of 0 and 4 at 6 hours, 5 at 12, 18 and 24 hours respectively, 6 at 30 hours, and

7 at 36 hours for 1:1 treatment. 2:1 treatment QI mean of the attributes were 5 at 6 and 12 hours, and 6 at 18 hours, 3:1 treatment QI was 6 at 6 hours and 7 at 12 hours, 4:1 QI was 6 at 6 hours and 7 at 12 hours respectively.

A high correlation ( $R^2 = 0.932$ ) for 1:1, ( $R^2 = 0.866$ ) for 2:1, ( $R^2 = 1$ ) for 3:1 and 4:1, shows that the attributes gradually increases with storage time as it is assumed in the Quality Index Method that the scores for all parameters increase with storage time (Martinsdottir *et al.*, 2001). The maximum storage time in ice is defined as the day when the fish is unfit for human consumption.

#### **Sensory evaluation for catfish (fresh):**

The Quality Index (QI) of attributes had an initial score of 0, 5 at 6 hours and remains at 5 till 28 hours of termination for 1:1. For 2:1 treatment, QI of the attributes was 5 at 6, 12 and 18 hours respectively. For 3:1 treatment, QI of the attributes was 5 at 6 and 12 hours respectively. For 4:1 treatment, QI of the attributes was 5 at 6 hours. The constant QI scores may be due to the fact that *Clarias gariepinus* did not deteriorate faster with time or according to Olafsdottir *et al.* (1997), each fish species has its own characteristic sensory attributes of flavour, appearance, odour and texture which changes with time and temperature after harvest.

#### **Sensory evaluation (cooked):**

There was no significant difference in all the treatments in both the catfish and the tilapia fish ( $p > 0.05$ ) during the period of storage. This indicates that the fish was acceptable by the panel and therefore fit for consumption at all tested levels.

#### **Microbial Analysis**

The microbial analysis revealed that the total viable count of the initial and four treatments 1:1, 2:1, 3:1 and 4:1 at different terminating points of 36, 24, 12 and 6 hours respectively shows a value ranging between  $1 \times 10^2$  cfu/g -  $12 \times 10^2$  cfu/g) and shows the absence of pathogenic organisms such as *Salmonella* spp., *Shigella* spp., *E. coli*, *Pseudomonas* spp. etc. while the control at 18 hour had a count of  $64 \times 10^6$  cfu/g. This means that the bacterial load is high in the control which indicates spoilage at the point of termination. However, no growth was recorded on SSA, EMB MCA and MSA for all the treatments at different termination periods. This shows the hygienic nature of Tilapia handling absence of cross contamination and absence of faecal contamination.

For Catfish, the total viable count of the initial and four treatments 1:1, 1:2, 1:3, 1:4 at different terminating points of 30, 21, 12 and 6 hours respectively shows a value ranging cfu between  $1 \times 10^2$  -  $12 \times 10^2$  cfu/g while the control at 18 hour had a count of  $70 \times 10^6$  cfu/g. However, the TVC in all the treatments were insignificant. The bacterial load is high in the control, which indicates spoilage at time of termination. No growth was recorded on SSA, EMB, MCA and MSA for all the treatments at different termination periods. This indicated the hygienic nature of fish handling and absence of faecal contamination as observed in the treatment of Tilapia. Catfish from all the treatments were devoid of pathogenic microorganisms that could be harmful when consumed. The mid pH value, cold storage and condition of the fish were favourable factors that contributed to slow microbial growth. Also, the variation in pH and change in storage temperature may have

contributed to the growths in microorganisms which are favourable factors necessary for increased bacterial counts as the storage time increases. The organisms identified from the control of *Clarias gariepinus* are *B. alvei*, and *B. pumillis* while *Proteus vulgaris* was isolated from the control of *Oreochromis niloticus* which are all of medical importance. (Shinkafi *et al.*, 2010)

The presence of the isolated organism was not surprising since according to Draser and Hill (1976), fish lives in water habitat full of micro-organism. Okpokwasili and Alapiki (1990) confirmed that bacteria flora associated with the Nigerian water culture include the genera, *Bacillus*, *Lactobacillus*, *Staphylococcus*, *Escherichia*, *Micrococcus*, *Proteus* and others.

*Oreochromis niloticus* and *Clarias gariepinus* from all the treatments and the freshly killed were devoid of pathogenic organisms that could be harmful when consumed. However, these increases are microbiologically insignificant (F.D.A, 1992)

The two species for all the treatments showed that the length of storage has an effect on the biochemical parameters. However, they were all below the safe limit and this indicates freshness as supported by Rigor index, sensory evaluation, pH and microbial analysis. There is a correlation between the microbial, chemical sensory attributes, storage time and conditions. *Oreochromis niloticus* cannot be kept beyond 12 hours at ambient conditions; however it can be well preserved for 36 hours under refrigerated conditions. This implies live *Oreochromis niloticus* can be transported without incidence for 6 hours and can still retain its freshness and

acceptability in ice up to 36 hours. Based on these findings, NSPRI should popularise the Ice-Fish Box for adoption by the fish farmers and other stakeholders. Policy makers and private organisations should collaborate with NSPRI to ensure the availability of this technology in urban and rural communities.

We hereby acknowledged the West Africa Agricultural Productivity Programme (WAAPP) for funding this research and the contributions of the following members of staff of NSPRI during the designing and execution of this research: Borisade, T. J., Agboola, A.A., Afolabi, A.A., Solagbade, A.M., Udefi, I.O., Nwanade, C.F., Ajayi, A.O., Adeniyi, B.M., Olatunde, I.G., Usanga, O.E., and Abiose, R.O.

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