

Quality Assessment of a Nigerian Marine Fish, Mullet (*Liza falcipinnis*) under different Storage Conditions

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Abstract: Quality assessment of a typical Nigerian marine fish specie, *Liza falcipinnis* (Mullet) was carried out at various storage temperatures such as ambient (28°C), Refrigeration (4 °C) and Frozen state (-5 °C) using sensory, microbiological and biochemical method of evaluation. At ambient temperature, the shelf life of the fish specie was estimated to be 12hrs. At this time, the fish was still sensorily acceptable, the bacterial load of the tissue was 6.2×10^4 cfu/g, TMA-N and TVB-N values were 4.8mg/100g and 26.7mg/100g respectively while the pH was 6.5. At refrigeration temperature, the shelf life was estimated to be 6 days. At this time, the fish was still sensorily acceptable, the bacterial load of the tissue was 2.4×10^4 cfu/g, TMA-N and TVB-N values were 6.80mg/100g and 28.80mg/100g respectively while the pH was 7.6. At freezing temperature, the fish was still sensorily acceptable at the estimated shelf life of 3 weeks. The bacterial load of the tissue was 3.2×10^3 cfu/g, TMA-N and TVB-N values were 9.50mg/100g and 39.80mg/100g respectively while the pH was 6.2. The limits of freshness stated here were mostly within the proposed limits of acceptability by some international quality standard organisations for marine fishes. [New York Science Journal 2010;3(8):21-28]. (ISSN: 1554-0200).

Keywords: Fish specie, *Liza falcipinnis*, quality assessment, shelf life and storage temperature.

1. Introduction

Fishes make up a huge and complex group of those vertebrates known in latin word as Pisces. This group comprises in simple terms, the finned gill breathing aquatic vertebrates (Hubbs, 1963). Among the food resources of the world, Fish and Fishery products are very important as sources of protein especially in some countries that are unsuitable for live stock production. At present there are numerous problems confronting the wide field of fisheries and some of which seems to be related to the keeping quality of the fish. Fish is a very perishable food and its high perishability has been the main obstacle in its preservation. According to Huss(1995), the main changes that can occur between capture and consumption of fish can be divided into 3 stages;

- i. The pre-rigor state in which the muscle tissue is soft and pliable.
- ii. The stiff and rigor condition known as rigor mortis whose onset can occur between 1-24hrs depending on the fish specie following the death of fish.
- iii. The post-rigor state in which the fish softens and starts to deteriorate.

It is noteworthy that pollution can also significantly affect the shelf life of the marine fish because the fish species inhabiting polluted waters have already compromised their original healthy status(Adeyemo, 2003).

Generally, in the first couple of hours after death, changes in fish are mainly due to biochemical processes, however after several hours, few bacteria penetrate the flesh where they degrade tissue components producing the unpleasant odours and flavours that are associated with spoilage (Huss, 1995). Time and temperature are the most critical factors to control to ensure that seafood retains high freshness quality as long as possible (Adams and Moses, 2008). At room temperature, seafood are likely to deteriorate very fast, Freezing and storage at low temperatures slows down bacterial growth and deterioration of fish through some enzymatic and chemical changes that progresses slowly (Huss, 1988).

Quality assessment of fishes has more to do with the determination of its shelf life or storage life which is the amount of time that sea foods remain palatable. Different species of fishes have different shelf lives which also varies depending on the oil levels, catch area, season, duration of rigor mortis, intrinsic conditions of the fish and how it was captured and handled (Huss, 1995). The shelf life of most marine fishes have been predicted to range between 2-24 days in ice, 5 days at 5°C and 3 days at 10 °C (Huss, 1995). The shelf life of Croaker (*Pseudotolithus elongatus*) found in Nigerian marine waters have been predicted to be 20 days in ice and 12 hrs at ambient temperature (Ola and Oladipo, 2004). Super chilling at -4°C and below have proven

to extensively extend the shelf life of frozen fish to several weeks because at such temperatures, microbial spoilage is almost impossible, its only chemical and enzymatic changes that can lead to spoilage after a considerable long time (Huss, 1995. Adams and Moses, 2008).

Marketing of Fish in Nigeria is mostly carried out by local fish sellers at ambient temperature, therefore knowledge of spoilage patterns of tropical fishes and their shelf life under ambient conditions is very important. Refrigeration temperatures are also relevant because they are used by most households in Nigeria for temporary storage of fish. Frozen state condition is also important since most fishes consumed in Nigeria are imported and they usually come in frozen state.

In the recent time, modern biotechnology have introduced new techniques that can detect early fish contamination, improve the taste, modify the quality of fish and prolong the shelf life and also impact disease resistance to the fish (William and Michael, 2009).

There are two main methods of assessing fish quality to determine its freshness and shelf life and these are i. Sensory and Non-sensory methods. Sensory methods rely mostly on appearance, odour, texture and taste of the Fish while non-sensory methods use physical, biochemical, chemical and microbiological means (Huss, 1995).

In the present study, quality assessment was carried out on a typical Nigerian marine fish specie, *Liza falcipinnis* (mullet) at different storage temperatures such as ambient (28°C), Refrigeration (4°C) and frozen state (-5 °C) using sensory, Microbiological and Biochemical method of evaluation. The limits of Freshness proposed by some international standard organisations for marine fishes were used as standard limits in this study.

2. Materials and Methods

i. Sample collection:

Fish samples *Liza falcipinnis* (mullet), were collected live from local fishermen at a site located near the Nigerian Institute for Oceanography and Marine Research, Victoria Island, Lagos where the experiment was carried out. The live fishes were killed by banging them on the head with a hammer and the exact time of death of life fishes were noted. The fishes were then taken to the laboratory for proper identification and analysis.

ii. Storage temperature:

The storage temperature employed were -5°C, 4°C and 28°C. A refrigerator with a temperature control setting was used and it was adjusted to give a temperature of -5°C at the upper freezing chamber

and 4°C at the lower refrigeration chamber. The temperature of the laboratory at the time of storage was 28°C and this was taken as the ambient temperature.

iii. Sensory analysis:

The method adopted was a conventional method used by Kremdorf et al,(1979). The characteristics features of the fish such as the colour of the eyes, skin and gills were observed. The odour and texture of the tissue and development of the slime on the surface were also observed as the storage period increased.

iv. Determination of pH:

pH of the fish was determined by the method of Waller (1980). 10g of the fish sample were homogenised with 50mls of distilled water and the pH value of the homogenate was measured by means of a glass electrode pH meter (Munchean 15) that was previously standardised.

v. Microbiological examination:

The bacterial counts on the external surfaces, intestines and tissue were estimated as follows;

a. Skin surface:

A sterilised rectangular wire swab guide measuring 5cm by 2cm was placed on the lateral surface of the fish sample. A sterile cotton wool swab was dipped in 0.10% sterile peptone water and was robbed over the surface of the fish on the area covered by the wire swab guide. The swab was immediately placed in a sterile sample bottle containing 100mls of 0.10% (w/v) peptone water. The bottle was vigorously shaken for 10mins and allowed to stand for 20mins. 10 fold serial dilution of the bacterial suspension in peptone water was prepared in duplicate and viable aerobic bacterial counts were enumerated in standard plate count agar after incubation at 37°C for 48 hrs as described in Slaby et al, 1981.

b. Intestines, Gills and Tissues:

10g of the fish sample was dissected out, blended and mixed properly in a mortar. It was aseptically transferred to a sample bottle containing 90mls of 0.1% sterile peptone water. The bottle was closed and shaken thoroughly for 10 mins. and allowed to stand for 20mins, after which a 10 fold serial dilution was carried out in duplicates and viable aerobic bacterial counts were enumerated in standard plate count agar after incubation at 37°C for 48 hrs as described in Slaby et al, 1981.

c. Identification of Microorganisms:

Morphological characteristics of the various bacterial isolates were noted in the agar plates and after staining reactions and series of biochemical tests, individual microbial species were identified as described in Slaby et al, 1981.

vi. Determination of Trimethyl Amine-Nitrogen (TMA-N).

Trimethyl-amine nitrogen (TMA-N) was determined by a slight modification of Conway Microdiffusion Method (1968). 25g of the fish sample was chopped and mixed thoroughly with 75 mls. distilled water in a 250 ml. beaker. The pH was adjusted to 5.2 by addition of few drops of 2N HCL, this was followed by heating at 70°C and cooling to room temperature. After cooling, the sample was filtered into a conical flask with the aid of whatman No. 1 Filter paper. 2mls. of 0.025N HCL was transferred to the central compartment of the micro diffusion dish with the aid of a pipette, this was followed by the addition of 2mls. of the extract and 0.5mls. of 35% formaldehyde with 1 mls. of saturated K₂CO₃ solution into the outer ring. The dish was covered immediately with a glass plate and the set up was left at room temperature for 24hrs. After this, the HCL in the inner compartment was titrated with 0.025N NaOH using 2-3 drops of methyl red/methylene blue indicator. The result was expressed in mg, TMA-N/100g of fish as described in Conway (1968).

vii. Determination of Total Volatile Bases (TVB-N):

Total Volatile Bases (TVB-N) was determined by a slight modification of Conway Microdiffusion Method (1968). 25g of the fish sample was chopped and mixed thoroughly with 75 mls. distilled water in a 250 ml. beaker. The pH was adjusted to 5.2 by addition of few drops of 2N HCL, this was followed by heating at 70°C and cooling to room temperature. After cooling, the sample was filtered into a conical flask with the aid of whatman No. 1 Filter paper. 2mls. of 0.025N HCL was transferred to the central compartment of the micro diffusion dish with the aid of a pipette, this was followed by the addition of 2mls. of the extract and 1 mls. of saturated K₂CO₃ solution into the outer ring. The dish was covered immediately with a glass plate and the set up was left at room temperature for 24hrs. After this, the HCL in the inner compartment was titrated with 0.025N NaOH using 2-3 drops of methyl red/methylene blue indicator. The result was expressed in mg, TVB-N/100g of fish as described in Conway (1968).

The Statistical Measure used in the quantification of result is Linear Correlation

Correlation coefficient (r)

$$r = \frac{\sum (x - \bar{x})(y - \bar{y})}{\sqrt{\sum (x - \bar{x})^2 \sum (y - \bar{y})^2}}$$

$$y = \text{Tma/Tvb (mg/100g),}$$

$$x = \text{Log TVC (total viable counts) (cfu/g)}$$

$$\bar{x} = \text{Mean of } x, \bar{y} = \text{Mean of } y.$$

Strong and perfect correlation should be near 1 or -1 (0.999 or -0.999)

3. Results:

Population densities and varieties of bacterial isolates from the fish sample under different storage conditions.

The population densities of various bacterial isolates from the fish sample were monitored at different storage temperatures i.e. 28°C, 4°C and -5°C and the experiment was terminated when the spoilage pattern exceeded the proposed international limits for the evaluation of the shelf life of fish and fishery products. Bacterial organisms were isolated from different parts of the fish body such as surface skin, gills, intestines and tissue. At ambient temperature, the experiment was monitored for 18hrs. as shown in table 1. Refrigeration storage (4°C) was monitored for 12 days while frozen state storage (-5°C) was monitored for 5 weeks, the results were shown in tables 2 and 3 respectively. It was generally observed that both the population and types of bacterial flora increased as spoilage progresses except at frozen storage where the reverse was the case.

Values of Trimethylamine (TMA-N), Total volatile bases (TVB-N) and pH recorded at different storage temperatures.

In contrast to the microbiological examination where different organs and tissues of the fish sample were used for the evaluation, only intact tissue of the fish was used for the determination of TMA-N, TVB-N and Ph. and the experiment was monitored for 24 hrs. at ambient temperature storage as shown in table 4, 15 days at refrigeration storage as shown in table 5 and 5 weeks at frozen storage as shown in table 6. As in microbiological studies, the experiment was terminated when the TMA-N and TVB-N values exceeded the proposed international limits for the evaluation of fish and fishery products.

Table 1: Types and Population Densities of Bacteria isolated from the Fish sample (*Liza falcipinnis*) during storage at ambient temperature at 28°C for 18hrs

Storage Time (Hrs.)	Bacteria Isolate	Total Viable Counts (TVC) Cfu/g or Cfu/cm ²
0		
External Surface	<i>Pseudomonas</i> sp., <i>Acinetobacter</i> sp., <i>Lactobacillus</i> sp., <i>Micrococcus</i> sp, <i>Corynebacterium</i> sp,	1.4 x 10 ³ Cfu/cm ²
Tissue	<i>Micrococcus</i> sp.	2 x 10 ² Cfu/g
Gills	<i>Flavobacterium</i> sp, <i>Bacillus</i> sp, <i>Micrococcus</i> sp	1.6 x 10 ⁵ Cfu/g
Intestine	<i>Staphylococcus</i> sp., <i>Flavobacterium</i> sp., <i>Escherichia</i> sp., <i>Bacillus</i> sp., <i>Micrococcus</i> sp.	8.7 x 10 ⁵ Cfu/g
6		
External Surface	<i>Corynebacterium</i> sp, <i>Micrococcus</i> sp., <i>Pseudomonas</i> sp., <i>Flavobacterium</i> sp.	3.8 x 10 ⁴ Cfu/cm ²
Tissue	<i>Micrococcus</i> sp., <i>Bacillus</i> sp., <i>Pseudomonas</i> sp.	2.1 x 10 ³ Cfu/g
Gills	<i>Micrococcus</i> sp., <i>Bacillus</i> sp., <i>Vibrio</i> sp., <i>Pseudomonas</i> sp.	2.8 x 10 ⁶ Cfu/g
Intestine	<i>Vibrio</i> sp., <i>Pseudomonas</i> sp., <i>Escherichia</i> sp., <i>Micrococcus</i> , <i>Flavobacterium</i> , <i>Acinetobacter</i>	6.6 x 10 ⁷ Cfu/g
12		
External Surface	<i>Pseudomonas</i> sp., <i>Flavobacterium</i> sp., <i>Micrococcus</i> sp., <i>Bacillus</i> sp. <i>Acinetobacter</i> sp.	5.4 x 10 ⁵ Cfu/g
Tissue	<i>Bacillus</i> sp., <i>Pseudomonas</i> sp., <i>Micrococcus</i> sp.	6.2 x 10 ⁴ Cfu/g
Gills	<i>Escherichia</i> sp., <i>Vibrio</i> sp., <i>Pseudomonas</i> sp., <i>Bacillus</i> sp., <i>Pseudomonas</i> sp.	3.2 x 10 ⁷ Cfu/g
Intestine	<i>Acinetobacter</i> sp., <i>Pseudomonas</i> sp., <i>Corynebacterium</i> sp., <i>Micrococcus</i> sp., <i>Bacillus</i> sp.	4.2 x 10 ⁸ Cfu/g
18		
External Surface	<i>Vibrio</i> sp., <i>Flavobacterium</i> sp., <i>Escherichia</i> sp., <i>Pseudomonas</i> sp., <i>Micrococcus</i> sp., <i>Bacillus</i> sp.	3.8 x 10 ⁶ Cfu/cm ²
Tissue	<i>Micrococcus</i> sp., <i>Pseudomonas</i> sp., <i>Staphylococcus</i> sp. <i>Vibrio</i> sp.	3.4 x 10 ⁶ Cfu/g
Gills	<i>Vibrio</i> sp., <i>Pseudomonas</i> sp., <i>Bacillus</i> sp., <i>Micrococcus</i> sp., <i>Corynebacterium</i> sp.	4.6 x 10 ⁸ Cfu/g
Intestine	<i>Pseudomonas</i> sp., <i>Micrococcus</i> sp., <i>Vibrio</i> sp., <i>Flavobacterium</i> sp., <i>Escherichia</i> sp., <i>Bacillus</i> sp.	3.5 x 10 ⁹ Cfu/g

Table 2: Types and Population Densities of Bacteria isolated from the Fish sample (*Liza falcipinnis*) during storage at a temperature of 4°C for 12 days

Storage Time (Days.)	Bacteria Isolate	Total Viable Counts (TVC) Cfu/g or Cfu/cm ²
0		
External Surface	<i>Pseudomonas</i> sp., <i>Acinetobacter</i> sp., <i>Lactobacillus</i> sp., <i>Micrococcus</i> sp, <i>Corynebacterium</i> sp,	1.4 x 10 ³ Cfu/cm ²
Tissue	<i>Micrococcus</i> sp.	2 x 10 ² Cfu/g
Gills	<i>Flavobacterium</i> sp, <i>Bacillus</i> sp, <i>Micrococcus</i> sp	1.6 x 10 ⁵ Cfu/g
Intestine	<i>Staphylococcus</i> sp., <i>Flavobacterium</i> sp., <i>Escherichia</i> sp., <i>Bacillus</i>	8.7 x 10 ⁵ Cfu/g

3	sp., <i>Micrococcus</i> sp.	
External Surface	<i>Micrococcus</i> sp., <i>Pseudomonas</i> sp., <i>Micrococcus</i> sp., <i>Pseudomonas</i> sp., <i>Flavobacterium</i> sp.	3.6×10^3 Cf/cm ² 7.7×10^3 Cf/g
Tissue		
Gills	<i>Bacillus</i> sp., <i>Pseudomonas</i> sp., <i>Escherichia</i> sp., <i>Micrococcus</i> sp.	4.8×10^4 Cf/g
Intestine	<i>Escherichia</i> sp., <i>Flavobacterium</i> sp., <i>Vibrio</i> sp.,	3.2×10^6 Cf/g
6		
External Surface	<i>Pseudomonas</i> sp., <i>Flavobacterium</i> sp., <i>Micrococcus</i> sp.,	4.8×10^3 Cf/g
Tissue	<i>Pseudomonas</i> sp., <i>Micrococcus</i> sp. <i>Flavobacterium</i>	2.4×10^4 Cf/g
Gills	<i>Escherichia</i> sp., <i>Vibrio</i> sp., <i>Pseudomonas</i> sp., <i>Bacillus</i> sp., <i>Pseudomonas</i> sp.	1.2×10^5 Cf/g 6.5×10^6 Cf/g
Intestine	<i>Vibrio</i> sp., <i>Pseudomonas</i> sp., <i>Bacillus</i> sp.,	
9		
External Surface	<i>Pseudomonas</i> sp., <i>Micrococcus</i> sp., <i>Corynebacterium</i> sp.	6.5×10^3 Cf/cm ²
Tissue	<i>Pseudomonas</i> sp., <i>Micrococcus</i> sp., <i>Flavobacterium</i> sp.	4.5×10^4 Cf/g
Gills	<i>Micrococcus</i> sp., <i>Pseudomonas</i> sp., <i>Vibrio</i> sp.,	7.2×10^5 Cf/g
Intestine	<i>Escherichia</i> sp., <i>Pseudomonas</i> sp., <i>Vibrio</i> sp.,	3.2×10^7 Cf/g
12		
External Surface	<i>Pseudomonas</i> sp., <i>Micrococcus</i> sp., <i>Acinetobacter</i> sp., <i>Flavobacterium</i> sp.,	1.2×10^4 Cf/cm ²
Tissue	<i>Pseudomonas</i> sp., <i>Micrococcus</i> sp., <i>Flavobacterium</i> sp.	2.6×10^5 Cf/g
Gills	<i>Micrococcus</i> sp., <i>Pseudomonas</i> sp.	2.8×10^5 Cf/g
Intestine	<i>Escherichia</i> sp., <i>Pseudomonas</i> sp., <i>Vibrio</i> sp.,	4.3×10^8 Cf/g

Table 3: Types and Population Densities of Bacteria isolated from the Fish sample (*Liza falcipinnis*) during storage at a temperature of -5°C for 5 Weeks.

Storage Time (Weeks)	Bacteria Isolate	Total Viable Counts (TVC) Cf/g or Cf/cm ²
0		
External Surface	<i>Pseudomonas</i> sp., <i>Acinetobacter</i> sp., <i>Lactobacillus</i> sp., <i>Micrococcus</i> sp, <i>Corynebacterium</i> sp,	1.4×10^3 Cf/cm ²
Tissue	<i>Micrococcus</i> sp.	2×10^2 Cf/g
Gills	<i>Flavobacterium</i> sp, <i>Bacillus</i> sp, <i>Micrococcus</i> sp <i>Staphylococcus</i> sp., <i>Flavobacterium</i> sp., <i>Escherichia</i> sp., <i>Bacillus</i>	1.6×10^5 Cf/g
Intestine	sp., <i>Micrococcus</i> sp.	8.7×10^5 Cf/g
1		
External Surface	<i>Micrococcus</i> sp., <i>Pseudomonas</i> sp., <i>Acinetobacter</i> sp.	2.5×10^3 Cf/cm ²
Tissue	<i>Micrococcus</i> sp., <i>Pseudomonas</i> sp.	1.11×10^4 Cf/g
Gills	<i>Micrococcus</i> sp., <i>Flavobacterium</i> sp.	2.6×10^4 Cf/g
Intestine	<i>Escherichia</i> sp., <i>Flavobacterium</i> sp.	3.5×10^4 Cf/g
2		
External Surface	<i>Pseudomonas</i> sp.	2.7×10^3 Cf/cm ²
Tissue	<i>Pseudomonas</i> sp., <i>Micrococcus</i> sp.	9×10^3 Cf/g
Gills	<i>Flavobacterium</i> sp., <i>Micrococcus</i> sp.	2.7×10^4 Cf/g

Intestine 3	<i>Pseudomonas</i> sp., <i>Flavobacterium</i> sp.,	4.5×10^4 CfU/g
External Surface	<i>Pseudomonas</i> sp., <i>Acinetobacter</i> sp.	3.2×10^3 CfU/cm ²
Tissue	<i>Pseudomonas</i> sp.	3.2×10^3 CfU/g
Gills	<i>Pseudomonas</i> sp., <i>Flavobacterium</i> sp.	3.5×10^4 CfU/g
Intestine 4	<i>Pseudomonas</i> sp., <i>Vibrio</i> sp.	4.2×10^4 CfU/g
External Surface	<i>Pseudomonas</i> sp.	1.5×10^3 CfU/cm ²
Tissue	<i>Pseudomonas</i> sp.	3.7×10^4 CfU/g
Gills	<i>Flavobacterium</i> sp., <i>Pseudomonas</i> sp.	5.8×10^4 CfU/g
Intestine 5	<i>Pseudomonas</i> sp., <i>Vibrio</i> sp.	6.3×10^4 CfU/g
External Surface	<i>Pseudomonas</i> sp.	1.8×10^3 CfU/cm ²
Tissue	<i>Pseudomonas</i> sp.	4.3×10^4 CfU/g
Gills	<i>Flavobacterium</i> sp.,	4.8×10^4 CfU/g
Intestine	<i>Pseudomonas</i> sp., <i>Vibrio</i> sp.	6.5×10^4 CfU/g

Table 4: Values of Trimethylamine (TMA-N), Total volatile bases (TVB-N) and pH recorded at ambient temperature (28°C).

Storage time (Hrs.)	TMA-N (mg/100g)	TVB-N (mg/100g)	pH
0	0.70	19.20	6.8
6	2.0	21.20	5.6
12	4.80	26.70	6.5
18	14.30	83	7.2
24	18.30	102.8	7.9

Table 5: Values of Trimethylamine (TMA-N), Total volatile bases (TVB-N) and pH recorded at refrigeration temperature (4°C).

Storage Time (Days)	TMA-N (mg/100g)	TVB-N (mg/100g)	pH
0	0.70	19.20	6.8
3	4.10	23.30	6.8
6	6.80	28.80	6.9
9	10.20	88.40	7.6
12	12.90	93.90	7.8
15	16.30	97.30	7.8

Table 6: Values of Trimethylamine (TMA-N), Total volatile bases (TVB-N) and pH recorded at Freezing temperature (-5°C).

Storage Time (Weeks)	TMA-N (mg/100g)	TVB-N (mg/100g)	pH
0	0.70	19.20	6.8
1	2.70	24	6.7
2	8.10	35.60	6.9
3	9.50	39.80	6.9
4	10.90	42.20	7.0
5	11.20	48	7.0

4. Discussion:

The quality assessment of a common Nigerian marine fish, *Liza falcipinnis* (Mullet) was carried out in order to determine the storage life (shelf life) of the fish sample under different storage conditions using sensory, microbiological and chemical methods of evaluation.

At ambient temperature storage (28°C) for 18 hours at 6 hourly monitoring interval, the storage life of the fish sample was predicted to be 12 hours, at this stage the fish was still sensorily acceptable. Bacterial load at the fish surface was 5.4×10^5 cfu/cm². The tissue, gills and the intestine had bacterial load of 6.2×10^4 , 3.2×10^8 and 4.2×10^8 cfu/g respectively. The tissue was considered as the reference point for bacterial spoilage because every other part of the fish with the exception of the tissue harbour normal bacterial flora even while the fish is alive. The tissue of a healthy fish is normally considered sterile until bacterial inversion that leads to spoilage. According to Adams and Moses (2008), the normal bacterial load of the surface slime of fish can range from $10^2 - 10^7$ cfu/cm² and the Gills and Intestines can range up to 10^3 and 10^7 cfu/g respectively. The bacterial load of the Fish sample (6.2×10^4 cfu/g) at the predicted shelf life of 12hrs. was within the range of the maximum limit (10^6 cfu/g) recommended by the international commission for microbiological standards of foods (ICMSF, 1978).

The values of Trimethyl Amine Nitrogen (TMA-N) and Total volatile bases (TVB-N) at the predicted shelf life of 12 hrs. at ambient temperature were 4.80 and 26.70 mg/100g respectively and these values were also within the recommended limits of acceptability. According to Connell (1995), TMA-N and TVB-N values in freshly caught fish ranged from 0.5 – 2.0 and 5-20 mg/100g respectively, these values increases as spoilage progressed and the maximum

limits of acceptability ranged from 4.50-7.20mg/100g for TMA-N and 30-35mg/100g for TVB-N. Ola and Oladipo (2004) have also predicted the shelf life of a similar Nigerian marine fish (*Pseudoholitus senegaliensis*) to be 12hrs. using sensory, microbiological and chemical approach.

At refrigeration temperature (4°C), the storage life of the fish sample under investigation was predicted to be 6 days. At day 6, the fish was still sensorily acceptable and the bacterial load of the fish tissue was 2.4×10^4 cfu/g which was still within the recommended maximum limit for acceptability (ICMSF, 1978). The respective TMA-N and TVB-N values of 6.80 and 28.80mg/100g were also within the recommended standard limits for acceptability (Connell, 1995). Huss (1995) stated that the shelf life of most marine fishes at refrigeration temperature (5°C) can last up to 5 days but this depends on the fish specie, oil level of the fish tissue, catch area, intrinsic conditions of the fish and how it was handled since capture.

At freezing temperature (-5°C), the storage life of the fish sample was predicted to be 3 weeks and this was based mainly on the sensory and microbiological evaluation which were within the recommended maximum limits of acceptability. The respective TMA-N and TVB-N values of 9.50 and 39.80 mg/100g were slightly higher than the recommended maximum limits of acceptability (Connell, 1995). Huss (1995) stated that super chilling at -4°C can effectively prolong the shelf life of fish for up to 5 weeks because at such temperature, microbial spoilage is very unlikely but rather chemical and enzymatic changes leads to spoilage. This assertion was also corroborated by Adams and Moses (2008) that microbial spoilage is very unlikely at frozen storage. In the present study, at frozen temperature (-5°C), the microbial load of the fish sample was as low as 3.2×10^3 cfu/g and the microbial flora was dominated by the psychrophilic *Pseudomonas* species which are still capable of growing at such temperatures even though at very slow rate. The sensory and microbiological analysis were still found to be more reliable in this case than the chemical approach. Mhongole (2009) in his quality assessment of Nile perch under different storage conditions found sensory and microbiological assessment methods to be more reliable than the chemical methods.

Generally, there was a strong correlation at ambient and refrigeration temperature between the total viable counts and the TMA-N and TVB-N values but at frozen temperature, no correlation existed and that explained why the sensory and microbiological analysis did not tally with the TMA-N and TVB-N values at frozen temperature. In a

similar study, Hozbor et al, 2006 found a strong correlation between the microbiological changes in sea salmon stored in ice and other quality indices like TMA-N, TVN-N and Histamine but no correlation existed at frozen temperatures (-25°C).

Conclusively, it can be advanced that the shelf life of *Liza falcipinnis* at ambient and refrigeration temperatures were 12hrs. and 6 days respectively and the sensory, microbiological and chemical approach used in the analysis were all in agreement with the recommended international limits for acceptability, however at frozen temperature, no correlation existed between the total viable counts of bacteria and the values of the TMA-N and TVB-N values which were slightly higher than the recommended acceptability limits, microbiological and sensory analysis were however found to be more reliable and subsequently used to predict the shelf life at day 6.

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