Assessment of the Bacteriological Quality of Cat Fish (Clarias gariepinus) Sold at 8 Miles Market, Cross River State, Calabar

1 Andy, I. E., 1Bassey, I. U., Lennox, J. A, and 1Obo, V. U.

Abstract- The quality of fish and its products are one of the major concerns of consumers given the unhygienic environment where they are harvested and sold. This research is aimed at isolating the bacteria flora that are associated with catfish as well as its safety when consumed. Ten (10) samples were collected from different locations of 8 miles market and were subjected to standard microbiological analysis. The results obtained revealed a significantly high mean bacterial count of 2.08x10^7 cfu/ml for fresh fish skin swabbed with a lower count of 1.89x10^7 cfu/ml for the washed dry fish samples, while the washed dry fish samples recorded a mean count of 3.50x10^5 cfu/ml. The decrease in bacterial load of the dry fish sample can be attributed to the preservative techniques that were used to preserve the fish. The bacterial distribution in the cat fish revealed 27.2% for Bacillus spp, 18.2% for Micrococcus spp, 18.2% for Serratia mercescens, 9.1% for Salmonella spp, 9.1% for Pseudomonas spp, 9.1% for Escherichia coli and 9.1% for Enterobacter spp. The results from this study reveals the dangers that may result from handling of both fresh and dry catfish and the health implications of consuming improperly cooked or processed fish. Hence, the need to process this species of fish properly before consumption.

Index Terms- Catfish (Clarias gariepinus), bacteriological quality, health implications

I. INTRODUCTION

Fish and fishery products are generally regarded as high risk commodity in respect of pathogen contents, natural toxins and other possible contaminants (Yusuf et al., 2008). Food borne infections are a major cause of illnesses and death worldwide (Adak et al., 2005). Food borne disease most often results from the ingestion of bacteria toxins and possibly microorganisms present in food.

Wafaa et al. (2011) stated that, fish and sea foods constitute an important food component for a larger section of the world population after meat and poultry as staple animal protein foods, where fish forms a cheap source of protein. There has been an increase in the awareness about the nutritional and health benefits of fish consumption over the years (Amusan et al., 2010). More people have turned to fish as an alternative to red meat. Fishery products, with their diverse great importance (health and nutrition) can also act as a source of various foodborne diseases because of the presence of contaminating pathogenic microorganisms found on them.

Fish meat deteriorates more quickly than other muscle foods, particularly when poorly handled. This spoilage is primarily bacterial in nature, but other factors such as enzymatic break down of the tissues contribute to spoilage. About 30% of landed fish are lost through microbial activity alone (Ghaly et al., 2010; Huisint, et al., 1996). Even with the improved food safety, progress is still uneven and food borne outbreaks from microbial contamination, chemicals and toxins are still common in many countries (WHO, 2007). Among all the food borne disease outbreaks reported globally, seafoods accounts for up to 8% of all outbreaks (Huss, 2003).

The microbial flora associated with fish is sometimes a reflection of their aqueous environment (Arafat, 2013). Water being a natural habitat for a wide range of microorganisms including bacterial, protozoa and fungi (Sumer et al., 2014), fish taken in or harbor these organisms from its environment. These organisms may be pathogenic to fish as well pathogenic to humans when ingested. Bacteria such as Pseudomonas fluorescens, Aeromonas hydrophila, Edwardsiella tarda, Vibrio spp are ubiquitous in the aquatic environment (Gilmour et al., 1976; Allen et al., 1983). Pathogenic bacteria such as E. coli, Salmonella, Shigella are most times introduced into water bodies through human or animal feaces. When fishes from these environments are ingested they could pose a great risk to the health of the consumers.

At the point of harvesting Catfish (C. gariepinus), contamination may occur through the equipment used in harvesting the fishes and may result in its spoilage. Catfish as one of the exceptionally perishable foods often face a lot of
rejections as they are most times priced and sold on some basic criteria such as freshness, which is also seen as the most important attribute of Catfish quality. Hence, this study was carried out to assess the bacteriological quality of catfish harvested and sold within calabar metropolis.

II. MATERIALS AND METHODS

Sample Collection and Source
The samples were collected using aseptic techniques early in the morning. Fresh and dry fish samples belonging to catfish (Clarias gariepinus) were bought from 8 miles, Calabar Municipal Local Government Area of Cross River State and placed in food grade sterile container. These were placed in pre-cool cooler and transported to the laboratory within one hour for bacteriological analysis.

Preparation of Sample
The samples were aseptically removed from the polyethylene bag and a sterile swab sticks were used to swab the surface of the fish and also the gills. These were inoculated in both nutrient agar and MacConkey agar. Twenty five grams the skin and gills each were cut with sterile scalpel and homogenized separately in peptone water. Tenfold serial dilutions were carried out up to $10^{-5}$. One ml of $10^{-5}$ dilution was plated in duplicate on nutrient and MacConkey agar using the spread plate technique.

Ten grams (10g) of the unwashed dry fish sample was homogenized into 90ml of distilled water using a sterile blender. A tenfold serial dilution of the sample was then carried out. From the tenfold serial dilutions of the homogenate, 1ml of $10^{-4}$ dilution was plated in duplicate on nutrient agar and MacConkey agar using the spread plate technique.

Ten gram (10g) of the washed dry fish sample was homogenized in 90ml of distilled water and a tenfold serial dilution was carried out of which 1ml of the dilution factor $10^4$ was plated in duplicate using spread plate techniques on MacConkey agar and Nutrient agar. The plates were incubated at room temperature for 24hours for bacteriological growth.

Bacteria Colony Count
Enumeration of the colonies was carried out after incubation and plates with the dilution factor that gave between 30-300cfu were counted and expressed as colony forming unit per gram (cfu/g) of the samples. Discreet colonies were subcultured into fresh agar plates aseptically to obtain pure culture of the isolates.

Characterization and Identification of the Isolates
The isolates where identified in terms of their colony characteristics, microscopy and biochemical tests.

Statistical Analysis
The statistical analysis was carried out using one-way ANOVA to determine the significant difference of the isolates in each sample. The hypothesis was tested at 95% confidence interval and conclusion was drawn.

III. RESULTS
The results of the assessment of the bacteriological quality of cat fish (Clarias gariepinus) sold at 8 miles market, cross river state, Calabar are presented in tables below. The results obtained from the total heterotrophic mean count of bacteria in fresh cat fish shows that the skin recorded the highest mean count of $2.08 \times 10^7$cfu/ml followed by the gills with bacterial count of $1.89 \times 10^7$cfu/ml while flesh + skin had the least count of $1.82 \times 10^7$cfu/ml (table 1).

Table 2 presents the total heterotrophic mean of the bacteria in the dry fish, unwashed and wash fish samples. The results obtained shows that unwashed fish samples recorded highest mean counts of $1.22 \times 10^6$cfu/ml compare to the washed sample with bacteria counts of $3.50 \times 10^5$cfu/ml. Table 3 shows the frequency of occurrence of bacterial isolates. Bacillus sp (27.2%) was the most predominant while Enterrobacter, Salmonella, E. coli and Pseudomonas (9.1%) were the least organisms isolated.

| TABLE 1 |
| Total Heterotrophic Bacteria counts in Fresh Catfish |

<table>
<thead>
<tr>
<th>FISH PART</th>
<th>MEAN PLATE COUNT</th>
<th>CFU/ML ($10^5$)</th>
<th>LOG 10 CFU/ML ± SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>GILLS</td>
<td>189.00</td>
<td>$1.89 \times 10^7$</td>
<td>7.2755 ± 0.0351</td>
</tr>
<tr>
<td>SKIN</td>
<td>208.33</td>
<td>$2.08 \times 10^7$</td>
<td>7.3176 ± 0.0398</td>
</tr>
<tr>
<td>FLESH + SKIN</td>
<td>182.33</td>
<td>$1.82 \times 10^7$</td>
<td>7.2564 ± 0.0767</td>
</tr>
</tbody>
</table>

http://dx.doi.org/10.29322/IJSRP.8.11.2018.p8138
TABLE 2
Total Heterotrophic Bacteria counts in Dry Catfish

<table>
<thead>
<tr>
<th>SAMPLE</th>
<th>MEAN COUNT</th>
<th>PLATE</th>
<th>CFU/ML (10⁴)</th>
<th>LOG 10 CFU/ML ± SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>UNWASHED</td>
<td>122.33</td>
<td>1.22 x 10⁶</td>
<td>6.0840 ± 0.0693</td>
<td></td>
</tr>
<tr>
<td>WASHED</td>
<td>35</td>
<td>3.50 x 10⁵</td>
<td>5.5422 ± 0.0499</td>
<td></td>
</tr>
</tbody>
</table>

TABLE 3
Frequency of Occurrence of the Bacterial Isolates

<table>
<thead>
<tr>
<th>Bacterial isolates</th>
<th>Number of occurrence</th>
<th>% occurrence</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bacillus spp</td>
<td>3</td>
<td>27.2%</td>
</tr>
<tr>
<td>Enterobacterspp</td>
<td>1</td>
<td>9.1%</td>
</tr>
<tr>
<td>Micrococcus spp</td>
<td>2</td>
<td>18.2%</td>
</tr>
<tr>
<td>Serratia mercescens</td>
<td>2</td>
<td>18.2%</td>
</tr>
<tr>
<td>Salmonella spp</td>
<td>1</td>
<td>9.1%</td>
</tr>
<tr>
<td>Pseudomonas spp</td>
<td>1</td>
<td>9.1%</td>
</tr>
<tr>
<td>E. coli</td>
<td>1</td>
<td>9.1%</td>
</tr>
<tr>
<td>Total</td>
<td>11</td>
<td>100%</td>
</tr>
</tbody>
</table>

Figure 4.1 shows a bar chart of fresh catfish that depicts the mean occurrence of the different parts of fish that was sampled. Each bar represents the part of the fish analyzed. Looking at the chart, the highest bar and value shows the highest counts of bacteria in the skin of fresh catfish. It shows a comparative analysis of bacterial counts between the various part of the fresh catfish sample. The bacteria counts with the largest \( \log_{10} \) value were more predominant in the skin with over 7.32 \( \log_{10} \) cfu/ml followed by the flesh + skin which had the least \( \log_{10} \) value of 7.26 \( \log_{10} \) cfu/ml, while the gills had a \( \log_{10} \) value of 7.27 a little above that of the flesh + skin. Together, the three parts of the fish accounts for over 7.32 \( \log_{10} \) cfu/ml of the bacterial isolates inherent in the fresh catfish.

In figure 4.2 a bar chart representing the dry cat fish is displayed, the highest bar and value stands for unwashed dry cat fish bacterial isolates.

Figure 4.1: Comparative Analysis of Bacterial Counts between the Gills, Skin and Flesh + Skin of Fresh Catfish Samples
IV. DISCUSSION

The consumption of fresh African catfish (*Clarias gariepinus*) is on the increase in both rural and urban centers of Nigeria (Emikpe et al., 2011; Adedeji et al., 2012). Fish is an important food commodity in the international trade but deteriorate rapidly when storage facilities are lacking. (Adedeji et al., 2012).

From the results of this study, it was discovered that bacterial loads of the catfish skin showed a slight difference from other parts of the fish. This can be compared with the work done by Adebayo-tayo *et al.*, (2012). Comparing the fresh and dry catfish, it was discovered that bacterial loads of the fresh catfish were higher compared to that of the dry fish as the fresh catfish recorded a counts of $2.08 \times 10^7$ cfu/ml against unwashed samples of dry fish with a count of $1.22 \times 10^6$ cfu/ml (Table 1 and 2). The results showed that, the unwashed dry-cat fish had higher counts compared to the washed samples. The low counts in the dry fish generally may be due to the lack of water in the fish as it is subjected to much heat in the course of drying. Drying by heat causes considerable reduction of microorganisms.

Base on the frequency of occurrence, *Bacillus spp* recorded the highest percentage of 27.2%. The presence of these organisms might be associated with the habitat in which it was caught. (Draser and Hill,1990, Shinkafi and Ukwaja,2010). *Bacillus* is known to cause a number infectious disease such as septicemia, wound and foodborne infections, meningitis, respiratory and urinary tract infections. (Morales *et al.*,2004; Shinkafi and Ukwaja,2010; Bassey *et al.*, 2015).

*Pseudomonas* is a soil bacterium that requires a high water activity for growth, it is known to cause food spoilage (meat, fish), and this is done by secreting lipases and proteases that causes off-odors and formation of slime, (Shinkafi and Ukwaja, 2010). *Serretia mercescens*, has been associated with infection such as lower respiratory tract infections and urinary tract infections. (Shinkafi and Ukwaja, 2010).

*Salmonella* is reported to be a very high pathogenic bacterium, (Mahmuda *et al.*, 2010) it had been reported to cause enteritis and systemic disease (Shinkafi and Ukwaja,2010). The detection of these pathogenic microorganisms in the catfish samples analyzed may be from the source in which it was harvested and the environment where the fishes are sold. Wafaa *et al.*, (2012) stated that the potential source of *Salmonella* in seafood is likely due to poor water quality, farm run off and fecal contamination from wild animals or livestock. Worthy of note in this study is the presence of both spoilage and pathogenic bacterial species that where isolated form the fish samples analyzed. The most prominent of these group of bacteria isolated include, *Bacillus spp*, *Micrococcus spp* and *Serretia mercescens*. These organisms are pathogenic and can be become harmful if consumed without proper processing. Food poisoning is often associated with some these bacterial species isolated from the samples analyzed. In a similar study by Bassey *et al.*, (2016), *Chromobacterium violaceum* and *Micrococcus luteus* had the highest prevalence of 18.18% in leachate and air samples while *C. violaceum* and *Staphylococcus aureus* had the lowest incidence of 1.76% and 1.87% in Ikot Effanga Mkpa river and stream water samples respectively. The high distribution of these organisms mostly in streams and rivers of Ikot Effanga Mkpa is a clear indication of the volume of wastes washed into the water bodies during the wet season and could possibly affect the microbiological quality of catfish harvested from this water body (Bassey et al., 2016; Eja *et al.*, 2010). These bacteria species including *Escherichia coli*, *Salmonella* sp and *Pseudomonas* sp have been reported by Bassey and Andy, (2015) as Beta-lamase producing organisms with high potentials of degrading beta lactam antibiotics at will. The study confirmed medicinal plants extracts can be used for both ESBL detection and treatment of E. coli, Klebsiella pneumonia and proteus mirabilis isolated from various sources (Bassey and Andy, 2015).

The method of harvesting, preserving and handling also contribute immensely to the high bacterial loads of the catfish as statistical analysis using one way ANOVA revealed significant differences in bacterial loads between the different parts of both the fresh and dry catfish sampled (p<0.05). It is pertinent to note here that beside source contamination of fishes, improper

![Figure 4.2: Bacterial Counts for Unwashed and Wash Dry Catfish Sample](http://dx.doi.org/10.29322/IJSRP.8.11.2018.p8318)
handling and environmental organisms from point of sale can as well increase the bacterial loads in catfish sold in open markets hence, the need to process fish and fish products properly before consumption.

V. CONCLUSION

Based on the result from the study, it can be concluded that the fresh cat fish habours a greater percentage of bacteria compare to dry fish. This study shows that the bacteriological quality and safety of catfish is influenced by habitat, harvesting tools and handling (display) of this fish for buyers which eventually encourage cross contamination. It is therefore suggested that contamination and re-contamination can be minimized by avoiding exposure, increase in hygiene of the retailers and adequate measures should be taken while harvesting, preserving and processing the fish before consumption.

REFERENCES


AUTHORS

First Author – Andy, I. E, Department of Microbiology, University of Calabar, Calabar Cross River State, Nigeria. P.M.B 1115. Email:bazero2007@gmail.com Mobile: +2348057222778

Second Author – Bassey, I. U, Department of Microbiology, University of Calabar, Calabar Cross River State, Nigeria. P.M.B 1115. Email:bazero2007@gmail.com Mobile: +2348057222778

Third Author – Lennox, J. A, Department of Microbiology, University of Calabar, Calabar Cross River State, Nigeria. P.M.B 1115. Email:bazero2007@gmail.com Mobile: +2348057222778

Fourth Author – Obo, V. U., Department of Microbiology, University of Calabar, Calabar Cross River State, Nigeria. P.M.B 1115. Email:bazero2007@gmail.com Mobile: +2348057222778