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Effect of Post-slaughter Time Intervals on the Quality of the African Catfish, *Clarias gariepinus* (Burchell, 1822)

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ABSTRACT

Effect of post-slaughter time intervals on the quality of fresh and hot smoke-processed African catfish, *Clarias gariepinus* products was investigated. A total of one hundred and four live catfish (average weight 700.0±7.0 g) was slaughtered and smoked. Ten freshly slaughtered fish samples were selected for organoleptic assessment at 0, 4, 8 and 12 h post-slaughter respectively. Three fresh and smoked fish samples each were selected for chemical and microbial analyses using standard analytical procedures. Microbial load on both fresh and smoked fish samples increased significantly ($p < 0.05$) with increase in post-slaughter time interval (PSI) with lowest value of 5.52±0.03 and highest value of 5.87±0.03 Log CfU g⁻¹ for fresh fish samples and lowest and highest values of 2.04±0.02 and 3.09±0.10 Log CfU g⁻¹ respectively for smoked fish samples. Bacteria isolates identified from fresh samples included: *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Bacillus spp* and *Staphylococcus aureus*; while *E. coli*, *S. aureus* and *Bacillus spp*. were isolated from smoked fish samples. Protein and ash contents of fresh fish samples increased progressively with increasing PSI while moisture content decreased; and lipid content remained unchanged. Protein, lipid and ash contents of smoked *C. gariepinus* significantly ($p < 0.05$) decreased with increased PSI while moisture content increased significantly ($p < 0.05$) with increasing PSI. This study suggests that *C. gariepinus* should be smoked immediately after slaughter for best quality hot-smoked fish products.

Key words: Hot-smoked fish, *Clarias gariepinus*, post-slaughter interval, microbial load, processing, preservation

INTRODUCTION

Fishery resources are important sources of dietary protein in most coastal communities in Nigeria and the world at large (Widjaja *et al.*, 2009). Fish supplies a good balance of protein, vitamins and minerals with very low carbohydrate content; hence its role in nutrition is recognized. Research confirms that fish is a rich source of essential nutrients required to supplement both infant and adult human diets (Abdullahi *et al.*, 2001; Adeparusi *et al.*, 2003).

In spite of the importance of fish in human nutrition and health, an estimated 40% of total fish landing in Nigeria is lost as post harvest losses. Eyo (2001) estimated that 20 to 50% of the fish produced in the remote coastal centers and hinter land of many tropical countries perish before they reach consumers due to the poor handling, preservation and processing practices adopted by the

artisanal fishermen, fish farmers and fisheries entrepreneurs. In addition, significant quality is lost through the absence of adequate technology and know-how to prevent losses in many tropical countries. According to Clucas (1990), the rate of fish spoilage depends on handling during processing, acidity level, species of fish, weather conditions, mode of storage and temperature during transportation while Daramola *et al.* (2007) reported that chemical breakdown of protein, fat and water contents contribute to rapid spoilage of fish. Hood *et al.* (1983) also reported that microbial load increases with increasing temperature and results in rapid fish spoilage while Bernacsek (1993) noted that variations in product quality arise from differences in the freshness of the raw material and preparation of fish prior to smoking.

A number of processing techniques are in operation in Nigeria, these include: chilling, freezing, salting, canning, drying and smoking (Kumolu-Johnson *et al.*, 2010), however, smoking is the most popular method of fish processing (Bako, 2005). Fish smoking is particularly relevant in the artisanal fisheries sector in that it prolongs the shelf-life of the fish, enhances flavour and increases utilization of the fish in addition to reducing wastes when catches are good as well as increasing protein availability to rural people (Jallow, 1995).

Clarias gariepinus is a very important fresh water fish in Nigeria (Idodo-Umeh, 2003) and enjoys wide acceptability in most parts of the country because of its unique taste, flavour and texture. It is widely distributed and extensively cultivated in ponds, but is still undervalued. This informs the choice of *Clarias gariepinus* for the present study. Akande and Ola (1992) observed that a 3 h post-slaughter holding of fish at ambient temperatures prior to processing still resulted in good quality hot-smoked fish products; thus it is not necessary to keep the fish for less than 3 h post-slaughter at ambient temperatures for this study. This study was carried out to assess the effect of post-slaughter time intervals on organoleptic, microbial, and chemical characteristics of raw and smoke-processed *C. gariepinus*, with an overall objective of determining appropriate post-slaughter time interval suitable for the production of good quality smoke-dried products of *C. gariepinus*.

MATERIALS AND METHODS

One hundred and four live *C. gariepinus* of approximately five months of age and average weight of 700.0 ± 7.0 g were collected early in the morning from concrete tank of Palm Royale Foods fish farm at Odogunyan, Ikorodu, Lagos State, Nigeria. The size of the culture tank was $8\text{m} \times 7\text{m} \times 7\text{m}$ and the water temperature at the time of collection was $27 \pm 2^\circ\text{C}$. Thereafter, the fish were transferred by road within 90 min to the Nigeria Institute for Oceanography and Marine Research (NIOMR) laboratories using eleven 30 L capacity plastic bowls with < 10 fish in each bowl. The fish were later kept in a holding tank ($7\text{m} \times 7\text{m} \times 6\text{m}$) for twenty-four hours. The water temperature in holding tank at the time of storage was $27^\circ\text{C} \pm 2$. Fifty-two fish were randomly selected, stunned by a hard hit on individual fish head and slaughtered using a sharp knife, washed with clean portable water and allowed to drip. Fish were later spread in the laboratory at ambient temperature ($27^\circ\text{C} \pm 2$). Twelve samples from the slaughtered *C. gariepinus* were filleted immediately to determine their proximate composition and microbial load, while forty samples (ten at different time intervals of 0, 4, 8 and 12 h, respectively) were collected for organoleptic assessment. Fifty-two samples of *C. gariepinus* were selected for smoking, with 13 samples each slaughtered at four time intervals ranging from 0 h (immediately after slaughter), 4, 8 and 12 h post-slaughter. Immediately after the fish were slaughtered for each treatment, they were immersed in 8% brine prior to smoking.

Nigerian Institute for Oceanography and Marine Research (NIOMR) smoking kiln was used at a temperature of $35\pm 10^{\circ}\text{C}$ for the first two hours and then at $100\pm 10^{\circ}\text{C}$ for another four hours, totaling 6 h (Obileye and Spinelli, 1978; Horner, 1997). Charcoal and *Anogeissus leiocarpa* were used to generate heat and smoke. *A. leiocarpa* was used because it is a hardwood believed to generate smoke with greater preservative potential than smoke from soft wood (Eyo, 2001). After smoking, three samples each were randomly selected from each treatment for the determination of proximate composition and microbial load, while the remaining ten samples for each treatment were used for organoleptic assessment. This study was carried out between March and August, 2008.

Proximate analysis: Crude protein, moisture, ash and fat contents of the fresh and smoked fish were carried out in triplicate in accordance with the AOAC (1995). The protein content was obtained through the determination of total nitrogen by micro Kjeldahl's method and the value of nitrogen obtained was multiplied by 6.25 to obtain the crude protein value.

Moisture content of samples was determined in accordance with AOAC (1995). The fat content was determined by extraction with petroleum ether by soxhlet's method.

Microbiological analysis: Total viable microbial count was determined using routine microbiological procedures described by Olutiola *et al.* (1991) and Fawole and Osho (2002) and identified using Bergey's Manual of Determinative Bacteriology.

Organoleptic (sensory) assessment: Sensory evaluation was carried out by a ten-man trained panel from NIOMR using a 5-point hedonic scale modified from Eyo (2001) and Tobor (1985). The following grades were allotted depending on the condition of the fish:

- $8\leq 10$ = Very good
- $6\leq 8$ = Good
- $4\leq 6$ = Fair
- $2\leq 4$ = Bad
- ≤ 2 = Very bad/unacceptable

Eyes, gills, skin, odour and flesh were examined for raw samples; while odour, flavour and texture were examined for smoked samples using the five-point hedonic scale.

Statistical analysis: Analysis of variance (ANOVA) was carried out using F-test to determine the treatments' level of significance and treatment means were separated using Duncan Multiple Range Test (DMRT) at 95% confidence value i.e., ($p < 0.05$).

RESULTS AND DISCUSSION

The results of the microbiological study (Table 1) indicated that Total Viable Count (TVC) of fresh fish samples at 0 h interval recorded the lowest TVC of 3.37×10^5 (Cfu g^{-1}) and TVC value increased significantly ($p < 0.05$) up till 12 h post slaughter. This is in accordance with the report of Hood *et al.* (1983) and Colakoglu *et al.* (2006) that microbial load increases with handling, duration of storage and temperature. This study established that fish left at ambient up to 8 hours post-slaughter still had TVC values within the maximum recommended bacterial count for good quality fish product which is 5×10^5 ($5.7 \log_{10}$ Cfu g^{-1}) according to the International Commission on

Table 1: Total microbial count of raw and cooked *C. gariepinus*

Time (h)	Fresh samples		Smoked samples	
	TVC (Cfu g ⁻¹)	Log Cfu g ⁻¹ ± SD	TVC (Cfu g ⁻¹)	Log Cfu g ⁻¹ ± SD
0	3.37×10 ⁵	5.52±0.03 ^a	1.10×10 ²	2.04±0.02 ^a
4	3.49×10 ⁵	5.54±0.01 ^b	2.25×10 ²	2.34±0.03 ^b
8	5.50×10 ⁵	5.74±0.02 ^c	3.14×10 ²	2.50±0.02 ^c
12	7.31×10 ⁵	5.87±0.03 ^d	1.24×10 ³	3.09±0.10 ^d

Means with different superscripts in the same column indicate significant difference at p<0.05

Table 2: Bacterial isolates from raw and smoked fish samples

Samples	Raw samples					Smoked samples		
	<i>Escherichia coli</i>	<i>Klebsiella pneumoniae</i>	<i>Staphylococcus aureus</i>	<i>Pseudomonas aeruginosa</i>	<i>Bacillus spp</i>	<i>Escherichia coli</i>	<i>Staphylococcus aureus</i>	<i>Bacillus spp.</i>
0	+	+	+	+	+	+	+	+
4	+	+	+	+	+	+	+	+
8	+	+	+	+	+	+	+	+
12	+	+	+	+	+	+	+	+

+: Presence, -: Absence

Microbiology Safety for Foods (ICMSF, 1986). This value was however exceeded in fresh fish samples left at ambient temperature for 12 h post slaughter with the value of 7.31×10⁵ of 5.869 log₁₀ (Cfu g⁻¹), yet the fish was not totally unacceptable as it has not exceeded the maximum recommended bacterial counts for marginally acceptable products which is 10⁷ (7log₁₀ Cfu g⁻¹) (ICMSF, 1986). Microbial quality evaluation confirms the presence of various psychrophilic microorganisms indicated in fish spoilage (Oksuztepe and Inanli, 2007) including *Klebsiella pneumoniae*, *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Bacillus spp*. This study also establishes that smoking reduces TVC in *C. gariepinus* with a lower value of 1.10×10⁵ (Cfu g⁻¹) recorded for fish smoked immediately after slaughter and values increasing significantly (p<0.05) at successive four hours of 4, 8 and 12 post-slaughter. The highest TVC which is 1.24×10³ obtained from *C. gariepinus* smoked at 12 h post-slaughter interval was still below the minimum recommended bacterial counts for good quality fish products. This means that *C. gariepinus* still gives good quality products if held for up to 12 h after slaughter prior to smoking.

Escherichia coli, *Klebsiella pneumoniae*, *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Bacillus spp*. were isolated from fresh fish samples (Table 2) while only *Escherichia coli*, *Staphylococcus aureus* and *Bacillus spp* were isolated in the smoked fish products (Table 2). This conforms to the report of Fernandez *et al.* (1998) that heat treatment during hot smoking of fish would destroy or inactivate most of pathogens found in fish.

Table 3 presents the proximate composition of fresh and hot-smoked *C. gariepinus*. The highest moisture content (78.32±0.50%) was recorded in freshly slaughtered *C. gariepinus* (0 h) with values decreasing significantly (p<0.05) with increase in post slaughter intervals. However, the lowest moisture value of 44.88±0.61% was recorded for fish smoked immediately after slaughter with values increasing significantly (p<0.05) with increase in post-slaughter intervals. The observed loss of moisture after smoking was due to moisture reduction through heat application during hot smoking (Kumolu-Johnson *et al.*, 2010) and is in agreement with the findings of Salan *et al.* (2006) and Kumolu-Johnson and Ndimele (2001) that spoilage of fish

Table 3: Mean proximate composition (%) of *Clarias gariepinus* parameters

Time (h)	Raw fish				Smoked fish			
	Moisture	Protein	Lipid	Ash	Moisture	Protein	Lipid	Ash
0	78.32±0.50 ^a	18.01±0.34 ^d	1.11±0.35 ^a	1.13±0.04 ^b	44.88±0.61 ^d	37.40±1.10 ^a	10.87±0.47 ^a	3.93±0.17 ^a
4	78.30±0.12 ^a	19.00±0.21 ^c	1.10±0.30 ^a	1.04±0.02 ^b	47.67±0.21 ^c	35.32±0.21 ^b	9.75±0.16 ^b	3.00±0.50 ^b
8	74.91±0.12 ^b	19.70±0.03 ^b	1.44±0.21 ^a	2.88±0.12 ^a	49.89±0.54 ^b	34.29±0.33 ^c	8.90±0.04 ^c	2.59±0.44 ^c
12	72.16±0.16 ^c	21.99±0.35 ^a	1.68±0.39 ^a	2.97±0.12 ^a	53.73±0.78 ^a	31.80±0.82 ^d	8.50±0.56 ^d	2.20±2.20 ^d

Means±SD with different superscript in the same column indicating significant difference at p<0.05

Table 4: Mean sensory score for raw and smoked *Clarias gariepinus*

Time (h)	Fresh fish samples				Smoked fish samples			
	Eyes	Gills	Skin	Odour	Flesh	Odour	Flavour	Texture
0	8.20±0.43 ^a	9.00±0.45 ^a	8.40±0.67 ^a	8.80±0.91 ^a	8.60±1.03 ^a	8.60±0.60 ^a	8.60±0.83 ^a	8.40±0.94 ^a
4	7.00±0.32 ^b	7.20±1.11 ^b	7.40±0.37 ^b	7.00±0.23 ^b	7.00±0.88 ^b	7.00±0.54 ^b	7.80±0.58 ^b	7.00±0.58 ^b
8	6.80±0.33 ^c	6.40±0.74 ^b	6.00±0.44 ^c	5.80±0.43 ^c	5.60±0.32 ^c	6.60±0.71 ^b	7.00±0.28 ^c	6.20±0.67 ^c
12	6.00±0.65 ^d	3.80±1.27 ^c	5.00±0.32 ^d	3.60±0.85 ^d	5.80±0.25 ^d	5.20±0.63 ^c	6.00±0.30 ^d	6.00±0.91 ^c

Mean±SD with different superscript in the same column indicates significant difference at p<0.05

resulting from the action of enzymes and bacteria can be slowed down by the addition of salt as well as reduction in moisture through sun drying or smoking. The percentage protein contents of fresh fish samples increased significantly (p<0.05) with increase in post slaughter interval, while it decreased significantly (p<0.05) with increased post slaughter intervals for smoked samples. This was probably due to moisture loss and an increase in dry matter content per unit of fish weight following sample dehydration (Omojowo *et al.*, 2009). Results in the present study reveal that *C. gariepinus* has similar ash contents between 0 and 4 h post-slaughter, followed by 8 and 12 h post-slaughter. It was observed that the ash content of the smoked fish decreased significantly (p<0.05) from 0 to 12 h post slaughter. Hours of delay after slaughter however, had no significant (p<0.05) effect on the lipid of fresh *C. gariepinus*, however, percentage lipid contents of the smoked fish samples significantly (p<0.05) decreased with increases in post slaughter intervals. It was observed that lipid content of smoked fish was higher than the values of fresh fish samples. This agrees with the view of Doe and Olley (1983) and Huda *et al.* (2010) that fish processing often results in the concentration of nutrients like crude protein and fat and that the quality of fish products is influenced by fish species differences, processing (including smoking), storage practices and duration of storage (Widjaja *et al.*, 2009; Kayim and Can, 2010). In general, the proximate composition and chemical quality of experimental fish reduced as post-slaughter interval prior to smoking increased in agreement with Ahmed *et al.* (2010).

Table 4 shows the mean sensory scores for fresh and smoked *C. gariepinus*. There was a significant (p<0.05) effect of post slaughter interval on the sensory quality of fresh and hot-smoked *C. gariepinus*. *C. gariepinus* retained most of its original freshness up to 4 h post slaughter, with the eyes transparent, clear and protruding with white cornea and dark pupil; the gills retaining a bright red colour and fresh odour, the skin bright, shining with slime and a firm belly. The flesh remained firm, flexible and elastic while the odour was fresh and sea weedy. This result agrees with the findings of Akande and Ola (1992) that African catfish *Clarias gariepinus* retained most of its original freshness up to 3 h post-slaughter. Similarly, the fish smoked immediately after slaughter (0 h) displayed very good physical attributes, with a fresh sea weedy, fishy odour and delicious

flavour and sweet taste while the texture was firm and tender. Akande and Faturoti (2003) reported similar results for Bonga (*Ethmolosa fimbriata*). Fish delayed for 4 h before smoking had good fishy odour, sweet flavour and slightly firm texture. However, *C. gariepinus* started losing its physical attributes when delayed for over 8 h after slaughter before smoke-processing. Similar findings were reported by Akande and Ola (1992) that deterioration was rapid in *Clarias gariepinus* delayed for 7 and 9 h and left at ambient temperature after slaughter prior to smoking. Fish smoked after being delayed for 8 and 12 h post-slaughter still displayed good physical attributes. The odour was fair, not offensive, the texture and flavour were good and the fish still had a sweet flavour and slightly firm texture. This agrees with the report of Sveindsdottir (2006) that smoke compounds play a dominant role in colour development as they cause the reactions of the Maillard type. Maga and Chen (1985) also reported that smoke contributes to overall smoky aroma of fish. This study also agrees with the report of Bernacsek (1993) that variations in product quality stem from the differences in the freshness of the raw material and the preparation of the fish prior to smoking.

CONCLUSION

This study confirms that fish quality deteriorates progressively with increasing post slaughter interval and hot smoking of fish inactivates and destroys pathogens normally associated with unprocessed fish. The study also confirms that fresh fish quality was still within acceptable limits when fresh fish is held up to 12 h post-slaughter at ambient temperatures before processing. However, delaying fresh *C. gariepinus* beyond 12 h post slaughter at ambient temperatures prior to smoke-processing should be avoided as the quality of the resulting product is reduced with total viable bacterial count in excess of the recommended level of 5×10^5 ($5.7 \log_{10}$ Cfu g⁻¹) for good quality product. Smoking in this study reduced the incidence of micro-organisms, hence guaranteeing the safety of the product. For premium quality hot-smoked products, *C. gariepinus* should be smoked immediately after slaughter.

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