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The Effect of Different Processing Methods on the Nutritional Quality and Microbiological Status of Cat Fish (*CLARIAS LEZERA* now (*Clarias gariepinus*)

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INTRODUCTION

- Fish provides 30 to 80% of the total animal protein intake of West Africa people.
- Catfish is high in unsaturated fatty acids, vitamins, proteins, minerals, low in carbohydrates and saturated fat. Smoke-dried fish consumed in Nigerian has appetizing taste and flavour [2].
- Catfish easily loses quality rapidly after catch.
- From literature some preservatives used are potassium sorbate, sodium chloride and sodium citrate.
- These preservatives can inhibit food-borne pathogens like *Staphylococcus aureus, Escherichia coli, Listeria monocytogenes* and toxin production of *Clostridium botulinum*.
- *Black pepper (Piper guineenses)* contains compounds like *piperine* and large amounts of *beta-caryophyllene*.
- In Nigeria it is used in making stew where it adds *pungent* aroma. The peppers have preservative and anti-oxidant properties.
• The various kinds of woods in the tropics are suitable for fish smoking process.

• Heat from the smoke can reduce the water activity of the fish cells. resulting in microbial inhibition which minimises spoilage [4].

• Smoking and the use of preservatives can influence the organoleptic attributes of fish .

• The antimicrobial effect of smoking depends on temperature, humidity, intensity of the smoke, duration, fish microflora and concentration of active components in the smoke [17].
JUSTIFICATION AND OBJECTIVE

- Epileptic power supply and high cost of equipment in Nigeria have made cold storage, canning and freezing non-realistic.

- Therefore traditional technique of smoke-drying is in common use but without preservative addition.

- The utilization of indigenous spices and chemicals like sodium citrate as catfish preservative properties has not been exploited in Nigeria.

- Therefore preservation of smoked fish using chemical preservatives and spices has become imperative.

- This study assessed the effect of sodium citrate and black pepper (*Piper guineenses*) on proximate composition, microbial and organoleptic attributes of smoked catfish during a 6-week storage at ambient temperature.

- Also the outcome is expected to encourage reduction in post–harvest losses and how to improve the nutritional and sensory attributes for consumption.
Methodology

• Fresh catfish \textit{(Clarias gariepinus)} samples were selected, washed, eviscerated, washed with distilled water and paper towel dried and weighed.
• About 300ml distilled water was added to 20g each of ground black pepper and sodium citrate.
• Each mixture was boiled and refluxed for 5 minutes, filtered hot through a sterile cheese-cloth (300 \(\mu\)). The filtrate was cooled to room temperature \((29\pm1^\circ\text{C})\).
• A= Control smoked fillets only, B= fillets previously soaked in Sodium citrate extract before smoking
• C= Fillets soaked in black pepper extract before smoking
• D=Fillets smoked in combination of black pepper and sodium citrate extracts before smoking.
• All samples were soaked in warm \((45\pm1^\circ\text{C})\) spice extracts for 10 minutes, drained and smoke-dried by laying the fish samples over the smoking kiln at 185\(^\circ\text{C}\).

• uniform smoking was achieved by turning samples over at intervals of 15 minutes for 2 hours.
• Some of the smoked samples were stored at \(-20^\circ\text{C}\) and analyzed for Proximate composition, Peroxide value (PV)and thiobarbituric acid (TBA)(for lipid oxidation) were all determined using methods of AOAC (2005) at day one(0), then at two, four and six weeks of storage.
• \textbf{Shelf –life studies}
• Smoke-dried fish was packaged in sterile low-density polyethylene bag, and sealed.
• Samples were kept in cardboard waxed paper boxes and stored at room temperature \((29\pm1^\circ\text{C})\) for the shelf life study at day 0, weeks 2,4 and 6.
Methodology contd ii.

• Lipid oxidation was conducted using the peroxide value (PV) measurement (as milliequivalent peroxide per kg of sample).
• Thiobarbituric acid (TBA) was determined by using UV–spectrophotometer (and absorbance measured at 532nm per kg of fish sample. TBA value was expressed as milligram malonaldehyde/kg of sample.
• **Microbiological evaluation**
  • A 10g representative sample from smoked fish muscle dissolved in sterile distilled water.
  • was homogenized for 60 seconds and serial dilutions (10^{-1} – 10^{-3}) made.
  • One milliliter (1ml) of the diluents was pour-plated on nutrient agar and incubated at 30ºC for 48hrs.
  • Total number of visible bacterial colonies, was determined by the Grid-Membrane Filtration method (GMFM) [25]
• The means of duplicate colony counts were calculated.
Trained panellists evaluated the various samples

The 5 – point descriptive hedonic scale was used for taste, texture, aroma, colour and general acceptability of the samples at 5% level of significance

Ranking scores were: like extremely(5), like moderately(4), neither like nor dislike (3), dislike moderately(2) and dislike extremely (1) respectively.
Statistical Analysis

• Measurements were carried out in triplicates.
• All microbial counts were converted into base-10 logarithms of colony forming units per g of sliced catfish samples (log_{10} CFU/g).
• Data were subjected to analysis of variance (ANOVA) using the General Linear Models procedure of the Statistical Analysis System software of SAS Institute (SAS Institute, Inc., 1990).
• Differences among the mean values of the various treatments were determined by the least significant difference (LSD) test, and the significance was defined at p< 0.05.
• The differences which are equal to or more than the identified LSD values are considered statistically significant.
Table 1. Effect of sodium citrate and black pepper treatments on moisture content (g/100g) of sliced catfish during six weeks storage

<table>
<thead>
<tr>
<th></th>
<th>0</th>
<th>2</th>
<th>4</th>
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<tbody>
<tr>
<td></td>
<td>Time (weeks)</td>
<td></td>
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</tr>
<tr>
<td>A=Control</td>
<td>19.42±0.01</td>
<td>20.41±0.01</td>
<td>19.04±0.03</td>
<td>15.13±0.03</td>
</tr>
<tr>
<td>B=NaC</td>
<td>10.12±0.01</td>
<td>10.68±0.01</td>
<td>13.06±0.01</td>
<td>13.54±0.03</td>
</tr>
<tr>
<td>C=Black pepper (Bp)</td>
<td>16.23±0.01</td>
<td>18.03±0.02</td>
<td>18.01±0.01</td>
<td>17.87±0.01</td>
</tr>
<tr>
<td>D=NaC+Bp</td>
<td>13.51±0.01</td>
<td>15.02±0.01</td>
<td>15.02±0.01</td>
<td>14.64±0.02</td>
</tr>
</tbody>
</table>
Table 2. Effect of sodium citrate and black pepper treatments on protein content (g/100g) of sliced catfish during six weeks storage

<table>
<thead>
<tr>
<th>Sample</th>
<th>0</th>
<th>2</th>
<th>4</th>
<th>6</th>
</tr>
</thead>
<tbody>
<tr>
<td>A=Control</td>
<td>60.52±0.01</td>
<td>61.34±0.02</td>
<td>65.17±0.03</td>
<td>69.13±0.01</td>
</tr>
<tr>
<td>B=NaC</td>
<td>69.30±0.02</td>
<td>70.93±0.05</td>
<td>66.08±0.02</td>
<td>65.54±0.02</td>
</tr>
<tr>
<td>C=Black pepper(Bp)</td>
<td>65.34±0.03</td>
<td>64.05±0.03</td>
<td>63.84±0.02</td>
<td>63.66±0.01</td>
</tr>
<tr>
<td>D=NaC=BP</td>
<td>66.58±0.01</td>
<td>66.15±0.02</td>
<td>66.15±0.02</td>
<td>65.54±0.03</td>
</tr>
</tbody>
</table>
**Protein content**

- The increase in the protein content of the smoked catfish could be attributed to an increase in the dry matter content per unit of weight following sample dehydration during smoking; this is in agreement to the finding of [28].

- Storage time appeared not to have affected (p>0.05) the protein content of smoked catfish.
Table 3. Effect of Sodium citrate (NaC) and black pepper (pg) on fat content (g/100g) of sliced fish during six weeks storage

<table>
<thead>
<tr>
<th>Sample</th>
<th>0</th>
<th>2</th>
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<tbody>
<tr>
<td></td>
<td>0</td>
<td>2</td>
<td>4</td>
<td>6</td>
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<tr>
<td><strong>Time (weeks)</strong></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>A=Control</td>
<td>16.66a±0.02</td>
<td>15.48a±0.01</td>
<td>12.44d±0.02</td>
<td>12.05d±0.01</td>
</tr>
<tr>
<td>B=NaC</td>
<td>15.33c±0.02</td>
<td>14.51c±0.01</td>
<td>15.02a±0.02</td>
<td>15.00a±0.01</td>
</tr>
<tr>
<td>C= Black pepper(Bp)</td>
<td>12.24d±0.02</td>
<td>14.17d±0.02</td>
<td>14.04c±0.02</td>
<td>17.87a±0.01</td>
</tr>
<tr>
<td>D= NaC+Bp</td>
<td>13.51c±0.01</td>
<td>15.02c±0.01</td>
<td>15.02c±0.01</td>
<td>14.64c±0.02</td>
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</table>
Table 4. Effect of Sodium Citrate (NaC) and black pepper (pg) treatments on ash content (g/100g) of sliced fish during six weeks storage

<table>
<thead>
<tr>
<th>Sample</th>
<th>0</th>
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<tbody>
<tr>
<td></td>
<td>Time (weeks)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A=Control</td>
<td>19.42$^{a}$$\pm 0.01$</td>
<td>20.41$^{a}$$\pm 0.01$</td>
<td>19.04$^{a}$$\pm 0.03$</td>
<td>15.13$^{b}$$\pm 0.03$</td>
</tr>
<tr>
<td>B=NaC</td>
<td>10.12$^{d}$$\pm 0.01$</td>
<td>10.68$^{d}$$\pm 0.01$</td>
<td>13.06$^{d}$$\pm 0.01$</td>
<td>13.54$^{d}$$\pm 0.03$</td>
</tr>
<tr>
<td>C= Black pepper (Bp)</td>
<td>16.23$^{b}$$\pm 0.01$</td>
<td>18.03$^{b}$$\pm 0.02$</td>
<td>18.01$^{b}$$\pm 0.01$</td>
<td>17.87$^{a}$$\pm 0.01$</td>
</tr>
<tr>
<td>D= NaC+Bp</td>
<td>13.51$^{c}$$\pm 0.01$</td>
<td>15.02$^{c}$$\pm 0.01$</td>
<td>15.02$^{c}$$\pm 0.01$</td>
<td>14.64$^{c}$$\pm 0.02$</td>
</tr>
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</table>
Figure 1: Effect of sodium citrate (NaC) and black pepper (Pg) treatments on lipid oxidation (peroxide value) in sliced catfish during six weeks storage.
**Peroxide Value**

- The PV in all samples were below the acceptable level of 10–20 meq peroxide/kg fish fat [30].
- This might be due to the extended storage and high temperature exposure.
- Fatty fish are particularly vulnerable to lipid oxidation which can create unpleasant (rancid) taste, smell, alteration in color.

- The various reactions involved in the lipid oxidation are either non-enzymatic or catalyzed by microbial enzymes or by intracellular or digestive enzymes from the fish themselves.
Figure 2: Effect of sodium citrate (NaC) and black pepper (Pg) treatments on lipid oxidation (Thiobarbituric acid value) in sliced catfish during six weeks storage.
Thiobarbituric Acid Value

• All the TBA values obtained were significantly (p<0.05) increased with the storage time.

• However, samples B, C and D showed significantly (p<0.05) lower TBA values with the storage time, when compare with the control.

• TBA assay is a widely used indicator for the assessment of degree of lipid oxidation. The result of TBA assay corroborated that obtained by the PV.

• In this study, TBA values for all the samples were much lower than recommend limit 0.007mg/kg (7 μg/kg) throughout the 6th week storage period. This is contrary to the values reported by [35].
Figure 3: Effect of different treatments on total aerobic plate count of catfish during six weeks storage.
Total Aerobic Plate Count of Microbes

• Sample C (with black had the least value while sample A (control) had the highest value.
• This indicated that dipping of the sliced catfish in the different treatment solutions resulted in drastic reduction of the initial TPC.
• The effects of preservatives on the microbial growth in fish may be depend on also on the degree of microbial contamination.

• By the end of 6th week of storage, TPCs for all of the different treatments were below $6 \log_{10} \text{CFU/g}$, while that of control attained a count of $6.24 \log_{10} \text{CFU/g}$.

• The control TPC is in close proximity to the maximal recommended limit of $7 \log_{10} \text{CFU/g}$ for TPC in raw fish [36].
Table 6. Sensory Evaluation of Sliced Catfish during Six Weeks Storage

<table>
<thead>
<tr>
<th>Sample</th>
<th>Colour</th>
<th>Aroma</th>
<th>Texture</th>
<th>Taste</th>
<th>General Acceptability</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>3.93d ± 0.38</td>
<td>3.58d ± 0.52</td>
<td>3.53d ± 0.57</td>
<td>3.62d ± 0.45</td>
<td>3.96d ± 0.66</td>
</tr>
<tr>
<td>B</td>
<td>4.29c ± 0.72</td>
<td>3.73c ± 0.84</td>
<td>3.62c ± 0.95</td>
<td>4.38c ± 0.86</td>
<td>4.52c ± 0.63</td>
</tr>
<tr>
<td>C</td>
<td>4.37a ± 0.85</td>
<td>4.93b ± 0.49</td>
<td>4.77b ± 0.52</td>
<td>4.57b ± 0.74</td>
<td>5.74b ± 0.84</td>
</tr>
<tr>
<td>D</td>
<td>4.36a ± 0.87</td>
<td>5.40a ± 0.72</td>
<td>4.89a ± 0.64</td>
<td>5.89a ± 0.85</td>
<td>5.88a ± 0.73</td>
</tr>
</tbody>
</table>
Sensory Evaluation of Sliced Catfish during Six Weeks Storage
• There was significant (p<0.05) difference in the sensory attributes of all the samples assessed except in terms of color

• Might be due to individual variations among individuals in responding to the same level of stimuli.

• Sample D (with NaCl+ Black pepper) was most preferred in terms of aroma, texture, taste while sample A (Control) was least preferred.
CONCLUSION

- From this study, sodium citrate and black pepper can be used as preservatives in smoked catfish without adversely affecting:
  - quality in terms of lipid oxidation, microbial load
  - color, and nutritional quality throughout 6 weeks of storage.

- There was significant (p>0.05) reduction in the PV and TBA values in comparison with the control.

- The effect of the spices applied singly and in combination on the catfish showed shelf life extension of all samples during the storage.

- The catfish samples exhibited shelf stability and the microbiological load fell within acceptable level stipulated by microbiological standards.

- This indicates that the samples might be safe for consumption throughout the period of 6 weeks storage.
• Dr K Oni Food Science Department, Federal University, Oye-Ekiti, Nigeria.
FUTURE WORK

• Isolation and Identification of microorganisms associated with smoked fish using molecular methods

• Incorporation of treated smoked fish into fermented cereal as infants complementary food of adequate nutritional quality

• Co-fermentation of cereal/ legume/ fish fillets to produce infant complementary food of improved nutritional quality that can be affordable for low-socio economic mothers in Nigeria
REFERENCES


33. Asiedu MS, Julshamn K, and Lie O (1991) Effect of local processing methods (cooking, frying, and smoking) on three fish species from Ghana: Part I,


APPRECIATION

• THANK YOU FOR YOUR

• ATTENTION!!!!!!