SENSORY EVALUATION AND MICROBIAL IDENTIFICATION ON SOME SMOKED FRESHWATER FISH SPECIES TREATED WITH SELECTED VEGETABLE OIL

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ABSTRACT

Vegetable oils which are oil extracted from edible plants have been used in a wide range of application in processed food. The study investigated the effect of groundnut, soybean and castor oil treatment on sensory quality, and microbial identification on smoked Clarias gariepinus, Heterotis niloticus and Proptopterus annectens. Forty-two (42) fish samples of mean weight of 650g were grouped into batches and immersed in 5 % brine solution at ambient temperature of 30±2 ^{0}C. The batches were smoked using smoking kiln. Batch (A), the control. B, C and D were immersed in 30 ml, of groundnut, soybean and castor oil respectively. The batches tagged A were the controls from each species of fish. Subscripts 1, 2 and 3 used to indicate fish species from C. gariepinus, H. niloticus and P. annectens respectively. Smoked fish samples were subjected to microbial identification and sensory analysis. Results were subjected to analysis of variance (ANOVA). Significant($p<0.05$) difference existed between taste, appearance and odour. The microbes identified include Shigella species, Proteus species, Penniselium species, Rhizopus species, Bacillus species, Aspergilus species, Fusarium species and Mucor species. Appearance recorded highest mean score (6.61) for sample B_1, the least (4.40) appearance was recorded for C_3. Sample B_1 recorded highest (6.00) acceptability, the least (5.00) was recorded for C_2 Oil treatments have shown lower microbial population below control level and have also improved smoked fish sensory parameters. Groundnut oil was most effective in improving the sensory parameters. Soybean oil recorded least microbial identification.

Keywords; *Smoked fish, Oil treatment, Storage duration, Sensory quality and Microbial count.*

INTRODUCTION

The world fish production stands at 184.6 million tons, out of which 146.3 million tons is used for human consumption (FAO, 2022). The remaining quantity is however used as non-food purpose and discarded as waste material. Idris (2010) stated that fish protein is indispensable to many people in developing countries, such as Nigeria, where the staple diet or food consist primarily of starchy foods. As important as fish is, high degree of fish spoilage still occur in Nigeria and serves a major constraint to the development of fishing industry in Nigeria (Akinpelu *et al.,* 2013). Blackwell (2014) characterized fish as a very perishable product and processing is therefore necessary to assure safety and prolonged shelf life of fish. Davies and Davies (2009) reported that about 30-50% of fish harvested are wasted in Nigeria due to poor handling. Traditional processing methods of fish including smoking were originally developed to preserve fisheries products by lowering the water activity (WA) which prevents the growth of spoilage bacteria (Kose, 2010). Different types of preservation methods employed to arrest fish spoilage include: drying, smoking, freezing, and brining (Kumolu-Johnson *et al.,* 2011). Fish smoking is particularly relevant in the artisanal fisheries sector in that it prolongs the shelf-life of the fish, enhances flavour, increases utilization of the fish as well as increases protein availability to rural people (Kiin-Kabari *et al.,* 2011). *Clarias gariepinus,* one of the species of catfish is a highly nutritious fish that contain high amount of vitamins, proteins, minerals, little saturated fat and is low in carbohydrates (Idris, 2010). *C. gariepinus* is a very important fresh water fish in Nigeria as it enjoys wide acceptability in most parts of the country because of its unique taste, flavour and texture (Ayeloja *et al*., 2011). *Heterotis niloticus*, also called African bony-tongue This commercial fish is a highly preferred source of food because of its high protein content and hardy flesh, thus forming a very important component in the diet of many Nigerians. The species has excellent flesh quality and good source of amino acids (Monentcham *et al.*, 2009). *H. niloticus* is one of the species that has great potentials for commercial aquaculture but associated with low acceptability by consumers due to its poor taste, reducing its market

value (Amuneke *et al.,* 2020). *Protopterus annectens* also called African lungfish is widely distributed in Nigeria inland waters and grow large to the relish of riparian communities (llozumba, and Ezeife, 2009). Groundnut oil is an organic oil derived from peanuts. It has neutral flavour and odour and does not absorb odours from other foods (Sulaiman *et al*., 2012) The major fatty acids in the oil are palmitic, oleic and linoleic acids (Merck, 2015). Soybean oil is used as a salad and cooking oil, as well as to make margarine, shortening, mayonnaise, and a wide range of processed foods (Merck, 2015). The wide range of application of soybean oil in food systems has been attributed to its high viscosity which makes it stick to surfaces for long and its safety to humans (Matthews, 2010). Castor oil is a vegetable oil obtained from castor oil plant. It contributes to 0.15 % vegetable oil produced in the world (Patel *et al.,* 2016). It has a long history of use commercially for the manufacture of soap, lubricants and coatings. The medicinal use of castor oil is minor (<1 %) (McKeon *et al.,* 2016). However, little information of its food uses is available. The reasons for fish smoking varied but in Nigeria, the process has proven relevant to prolonging shelf-life, enhancing flavor, storing for lean season and increasing protein availability for people throughout the year (Kumolu-Johnson and Ndimele, 2011). Oil is not only used as flavouring agent, but also as a preservative, due to its antioxidant, antifungal and antibacterial activity. The study examined the use of three oils (Groundnut, Castor and Soybean oils) on smoked *Clarias gariepinus, Heterotis niloticus and Protopterus annecterns* stored under ambient temperature with the optimal variant taking into consideration the sensory, microbial identification

MATERIALS AND METHODS

The Study Area

The study was conducted in Makurdi, Benue State, Nigeria Benue State, Nigeria located on longitude 7.3 ^{0}N and latitude 8.54 ^{0}E.

Collection and Measurement of Fish Samples.

Forty-two (42) freshly caught fish samples with mean weight of 650 g comprising of three different species (*Clarias gariepinus, Heterotis niloticus* and *Protopterus annectens*) were bought from fishermen at fish landing site, Wadata, Makurdi, Benue State. The fish samples were conveyed to the Department of Fisheries and Aquaculture, Joseph Sarwuan Tarka University, Makurdi, Benue State, Nigeria in an ice-filled plastic bowl covered with a sack bag to prevent deterioration.

Fish samples were thoroughly washed with clean water. The length and weight of the fish samples were taken using a measuring tape and using sensitive scale (Atom A 122 Electronic kitchen digital weighing scale, model SF: 400A). Fish samples were eviscerated and washed thoroughly with clean water. 8kg each of the fish species were divided into 2 kg each for oil treatment and control.

Sample Preparation and Smoking

All the batches of the fish samples were immersed in 5 % brine solution for 30 minutes within ambient temperature of 30±2 ^{0}C. The fish samples were transferred into different clean baskets for proper draining of water prior to smoke-drying in the smoking kiln. 8kg of each of the fish species were divided into four equal parts of 2kg each for three oil treatments and control (without oil). The fish samples were smoke-dried in the smoking kiln for twelve hours for two days using firewood following the method describe by (Iheanacho *et al*., 2017). Each fish smoked species divided into four batches (A B C and D) for the control and treatments. The 3 fish species tagged with subscripts 1,2 and 3

Subscripts 1 (*Clarias gariepinus*) was divided into samples A_1B_1 C_1, D_1.

Subscripts 2 *(Heterotis noclioticus*) was divided into samples A_2, B_2, C_2, D_2.

Subscripts 3 (*Protopterus annectens*) was divided into samples A_3, B_3, C_3, D_3.

Samples with letter A (The controls) from each of the fish species. The fish samples tagged with letters B, C, and D were those treated with groundnut oil, soybean oil and castor oils respectively.

The fish samples in Batch A (control) was not treated with oil while the other three batches B, C and D were immersed in 30 mL of groundnut oil, 30mL of soybean oil and 30 mL of castor oil respectively, dried on laboratory table and stored for 8 weeks under ambient temperature.

Microbial Identification

Identification of microbial species in smoked fish was done following the procedures described by Onyeagba (2004). Microscopic examination follows the incubation period where the colonies were observed on the plates and their size, colour, texture, and any other distinctive features. In the microscopic

examination process, wet lactophenol cotton blue mounts of microbial colonies were observed under a microscope and the structures such as hyphae, spores (conidia), conidiophores and any other characteristic features were examined as staining technique of lactophenol cotton blue stain or potassium Hydroxide (KOH) was used to enhance visualization of microbial structures.

Sensory Evaluation of the Smoked Fishes Samples

Sensory evaluation was carried out using a panel of 20 members consisting of students and members of staff in Fisheries and Aquaculture Department, Joseph Sarwuan Tarka University, Makurdi, Nigeria. Panelists was chosen based on their familiarity and experience with smoked fish for sensory evaluation. Fishes treated with the oils and without oil were represented in coded form and were randomly presented to the panelists. The panelists were provided with portable water to rinse their mouths between evaluations. Each sensory attribute was rated on a 7-point Hedonic scale

Statistical Analysis

The GENSAT Statistical Program (Rothamsted Experiment Station, 2007) was used for data analysis. Data were subjected to analysis of variance (ANOVA) and difference of mean were separated by the Fisher's least significant difference (LSD) test at ($P<0.05$).

RESULTS

The various microbes identified in the smoked fish samples are shown in Table 1. The microbes identified from the smoked fish samples include *Shigella species, Proteus species, Penniselium species, Rhizopus species, Bacillus species, Micrococcus species, Streptococcus species, Aspergilus species, Fusarium species and Mucor species.* Among these microbes, Mucor species was the most identified (10 times) in the smoked fish samples. It was detected in all fish samples except sample C_1 and A_2. *Aspergilus species* the least (once) in the smoked fish samples as it was only detected in smoked *P. annectens* sample treated with groundnut oil. Among the samples of fish analysed, smoked *P. annectens* recorded the highest number of microbes (9 species) as its untreated sample (A_3) and sample treated with groundnut oil (B_3) recorded six (6) different species of the microorganisms.

Table 1: Effect of Groundnut, Soybean and Castor oils on identification of Microbes on Smoked *Clarias gariepinus*, *Heterotis niloticus* and *Protopterus annectens*

Samples/ Treatment	Microbes										
	Shigella Spp	***Klebsiella Spp***	***Proteus Spp***	***Penniselium spp***	***Rhizopus spp***	***Bacillus Spp***	***Micrococcus Spp***	***Streptococcus Spp***	***Aspergilus spp***	***Fusarium spp***	***Mucor spp***
C. gariepinus											
A_1		+	+		+		+				+
B_1	+	+				+					+
C_1		+				+	+			+	
D_1		+	+			+					+
H. niclotnicus											
A_2		+	+		+	+				+	
B_2		+	+					+			+
C_2	+	+			+						+
D_2		+			+	+	+				+
P. annectens											
A_3	+		+		+	+	+				+
B_3			+		+	+		+	+		+
C_3			+	+		+		+			+
D_3	+		+	+							+

KEY: + =Mean of microbial presence. A_1 = Smoked *C. gariepinus* without oil treatment (Control – 1); B_1 = Smoked *C. gariepinus* with groundnut oil treatment; C_1 = Smoked *C. gariepinus* with soybean oil treatment; D_1 = Smoked *C. gariepinus* with castor oil treatment; A_2 = Smoked *H. niloticus* without oil treatment (Control – 2); B_2 = Smoked *H. niloticus* with groundnut oil treatment; C_2 = Smoked *H. niloticus* with soybean oil treatment; D_2 = Smoked *H. niloticus* with castor oil treatment; A_3 = Smoked *P. annectens* without oil treatment (Control – 3); B_3 = Smoked *P. annectens* with groundnut oil treatment; C_3 = Smoked *P. annectens* with soybean oil treatment; D_3 = Smoked *P. annectens* with castor oil treatment

Sensory attributes of freshly smoked *C. gariepinus*, *H. niloticus* and *P. annectens* analyzed vary between treatments as mean scores showed significant ($p< 0.05$) differences among oil treatments for appearance, texture, taste, odour and general acceptability respectively (Table 2). Appearance recorded the highest mean score (6.61) for sample B_1 while the least appearance score (4.40) was recorded for C_3. Mean sensory scores for texture of smoked fish ranged between (4.30) in sample B_1 to (6.20) in sample D_3. The mean sensory scores recorded for taste of smoked fish ranged from (4.50) to (6.35) with sample B_1 scoring highest (6.35) and sample D_3, the least (4.50). Odour of smoked fish samples recorded least value of (5.4) for sample C_3 and highest value of 6.00 for sample A_2. The range of 5.00 – 6.00 was recorded for general acceptability of the smoked sample with sample B_1 scoring highest (6.00) while the least value (5.00) was recorded for C_2, D_2 and D_3

Table 2: Effect of Groundnut, Soybean and Castor oils on the Sensory Quality of Smoke-dried *Clarias gariepinus*, *Heterotis niloticus* and *Protopterus annectens*

Parameters	Samples												LSD
	Smoked Clarias gariepinus				*Heterotis niloti cus*				*Protopterus annectens*				
	A_1	B_1	C_1	D_1	A_2	B_2	C_2	D_2	A_3	B_3	C_3	D_3	
Appearance	6.30^{b}	6.65^{a}	5.85^{c}	5.20^{d}	6.20^{b}	6.30^{b}	5.60^{c}	5.00^{d}	5.90^{c}	5.00^{d}	4.40^{e}	4.45^{e}	0.27
Texture	5.80^{ab}	6.20^{a}	5.90^{ab}	5.55^{bc}	5.20^{de}	5.85^{ab}	5.90^{ab}	5.45^{cd}	4.50^{f}	5.00^{e}	4.60^{f}	4.30^{f}	0.41
Taste	6.20^{ab}	6.35^{a}	5.85^{bc}	5.80^{bc}	5.50^{cd}	5.80^{bc}	5.00^{ef}	5.15^{def}	4.85^{fg}	5.30^{de}	4.55^{fg}	4.50^{g}	0.50
Odour	5.90^{c}	5.95^{c}	5.60^{f}	5.95^{c}	6.00^{b}	6.25^{a}	5.70^{e}	5.90^{c}	5.55^{f}	5.60^{f}	5.40^{g}	5.75^{d}	0.09
General acceptability	5.70^{b}	6.00^{a}	5.75^{a}	5.70^{b}	5.55^{b}	5.55^{b}	5.00^{c}	5.00^{c}	5.60^{b}	5.20^{c}	5.10^{c}	5.00^{c}	0.30

KEY:

Values are means ± standard deviations of triplicate determinations. Means with different superscript in the same row are significantly ($p<0.05$) different. A_1 = Smoked *C. gariepinus* without oil treatment (Control – 1); B_1 = Smoked *C. gariepinus* with groundnut oil treatment; C_1 = Smoked *C. gariepinus* with soybean oil treatment; D_1 = Smoked *C. gariepinus* with castor oil treatment; A_2 = Smoked *H. niloticus* without oil treatment (Control – 2); B_2 = Smoked *H. niloticus* with groundnut oil treatment; C_2 = Smoked *H. niloticus* with soybean oil treatment; D_2 = Smoked *H. niloticus* with castor oil treatment; A_3 = Smoked *P. annectens* without oil treatment (Control – 3); B_3 = Smoked *P. annectens* with groundnut oil treatment; C_3 = Smoked *P. annectens* with soybean oil treatment; D_3 = Smoked *P. annectens* with castor oil treatment

DISCUSSION

The microorganisms isolated and identified from the smoked fish samples can be said to be normal flora of the fish e.g. *Bacillus sp.* (Ola and Oladipe, 2004). In agreement with the study of Renitta *et al.* (2021) for Biochemical, Microbiological and Sensory Quality of underutilized Marine gastropod smoked products from *Chicoreus ramosus* and *Volegalea cochlidium*, pathogenic bacteria like *E. coli, salmonella*, and vibrio were not enumerated in all smoked fish samples throughout the storage period. Identical results were also reported by Ratsimba *et al.* (2019) in a smoked meat product of Madagascar. Among the oil treatments applied to the smoked fish samples, soybean oil was most effective in consistently recording the lowest number of the microbes as only 4 different species of the microbes were detected across the three smoked fish samples treated with soybean oil. This suggests that soybean oil confers the most antimicrobial tendency than groundnut oil and castor oil. The quality of smoked products is dependent on several factors, including, the quality of the fish at the time of smoking, the preparation of the raw material, the nature of wood and the type of the smoking procedure employed (Da Silva, 2002). The microorganisms isolated in this study have been reported in some fish species. Akinwumi and Adegbehingbe (2015) studied the microbiological analysis of three of smoked fish obtained from Ondo State, Nigeria and isolated *Streptococcus aureus, Bacillus species, Proteus species, Aspergillus fumigatus and Aspergillus niger* in smoked herring, tilapia and catfish. Adelaja *et al.* (2013) confirmed that the presence of A*spergilus spp* in the smoked fish investigated could be as a result of handling processes during smoking and cross contamination during storage. The microbial flora associated with fish could be from the environment in which the fish are harvested and not specific to a particular species (Teugels and Audenerde, 2003).

The presence of biological and chemical (biotoxins) hazards has posed serious safety concern in food systems (Akinwumi and Adegbehingbe, 2015). Huss *et al.* (2000) reported that these hazards are present in fish and shellfish pre-harvest and are, therefore, difficult or impossible to control. However, several techniques exist in the prevention of the growth of pathogenic microorganisms during distribution and storage of processed fish. Cheng-An *et al.* (2009) recommended that the processed fish should be exposed to a drying temperature that will provide insufficient moisture content for the growth of micro-organisms.

The mean sensory scores had an average score of 5.52 on a scale from 1 to 7 (7 – point Hedonic scale), thus indicating smoked fish is liked very much by sensory panelists. For a good consumer acceptance, the perceivable sensory attributes are appearance, odour, taste, and texture, and the synthesized judgments of all quality attributes provide the overall quality of the product (Assogba *et al.*, 2019). The oil treatment may have acted as an enveloping layer against the adverse interaction between the fish system and the environment resulting in the improved sensory quality of the fish products. Osibona *et al.* (2019) observed that containing materials play a vital role in the enhancement of smoke-dried fish. The organoleptic properties of the smoked *Clarias gariepinus* were assessed, and *C. gariepinus* had the highest (best) result in appearance, texture and taste when treated with 30ml of groundnut oil. The untreated smoked *C. gariepinus* sample (The control) had the lower score compared to the sample treated with groundnut oil (B_1) in all the assessments. This could probably be due to the absence of oil on it. This shows that application of oil on smoked fish improves quality and this is in agreement with Kumolu *et al.* (2015) who revealed that smoked fish samples treated with ginger oil had lower mould count and improved quality. Smoked *H. niloticus* and *P. annectens* generally showed improvement on all sensory parameters assessed for groundnut oil treatment in the present study. This revealed that groundnut oil is most effective in improving the sensory qualities of smoked fish than soybean oil and castor oil. The secondary products for oil oxidation are ketones and aldehydes. The off-flavour of soybean oil comes from low ketones or aldehydes (wang *et al.*, 2016). The superior effect of groundnut oil on sensory quality of smoked fish compared to soybean oil and castor oil could be attributed to the tendency of groundnut oil relinquish beany flavour upon heating application. According to Sanders (2003) crude groundnut oil has a bland, slightly beany, nut-like flavour which is removed during heating to produce oil that is odourless. This observation is in agreement with the claim of Wang and Chen (2016) who stated that heated foods containing groundnut oil have excellent sensory properties (flavour, crispness, mouth feel) as well as superior fry life.

Also, the improved odour of smoked fish sample containing groundnut oil could have been informed by the presence of good proportion of flavour compound in the oil. Wang and Chen (2016) stated that peanut oil contains the optimum linoleic acid needed for production of compounds contributing to fried food flavour. The high rating with regards to appearance of smoked fish treated with oil may have resulted from the combined effect of the oil treatment and the firewood used in the smoking process. Fire wood used in smoked fish product offers golden brown colour to fish till the end of the smoking termination period which might be as a result of phenolic and hemicellulose compound present in smoked hardwood materials (Umar *et al.*, 2018). This is in agreement with the study conducted by Adibe *et al.* (2018) who evaluated the organoleptic properties of *Clarias gariepinus* smoked with natural and artificial spices. The observations in the present study are also in agreement with the reports of Taniya and Kannan (2016) and Iheagwara (2013). The general increase in texture scores across all fish samples treated with soybean oil is buttressed by the claim of Wang *et al.* (2022) which reported that chewiness and springiness (texture properties) of ice cream increased with soybean oil body (a lipid-storing organelles in soybean which consists of neutral lipid drop, mainly phospholipids and basic proteins) substitution, by prolonging the residence time of ice cream in the mouth. Again, the general increase in odour mean scores across all smoked fish sample treated with castor oil may be as a result of richness of saturated fatty acids in castor oil (Pereira *et al.*, 2010). Castor oil is rich in fatty acid which enhances flavour and odour. Castor oil substituted at 5 % level into goat milk increased sweet flavour from (4.6) to (4.7) (Pereira *et al.*, 2010).

According to Umar *et al.* (2018), fish could be smoked using fire wood without any fear of acceptability by the consumers. The better preference for acceptance of groundnut oil reported by the sensory panelists in the present study disagrees with earlier report by Adadu *et al.* (2021) which stated that viscosity of castor oil is high and it shines brighter than so many oils and put forward that castor oil has higher level of acceptance based on appearance (67%) than those treated with groundnut oil (33%). The origin of the castor oil and groundnut oils used in the present investigation coupled with the sense of judgment of the evaluators could have accounted for the difference. The quality of the oils, rancidity and oxidation might also be responsible for the observed results

CONCLUSION AND RECOMMENDATION

The oil treatments have also shown a good potential in controlling microbial population and pathogens in the smoked fish by maintaining microbial counts at safe level during storage.

Smoked fish sensory parameters (taste, appearance, flavor and texture and general acceptability) have also shown improvement with the application of the selected oils.

It was evident that among the oils treatments applied to the experimental smoked fish, groundnut oil was most effective in improving most of the sensory parameters evaluated.

Groundnut oil is recommended for pre-treatment of fish prior to processing and storage especially when consumer's palatability and nutrient retention is desired.

When demand is placed on smoked fish product microbial safety, soybean oil should be used to treat smoked *C. gariepinus, H. niloticus* and *P. annectens*

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