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RESEARCH ARTICLE

Evaluation of Bacteriological and Mercury level in Cod (*Gadus morhua*) and Saithe (*Pollachius virens*) Stockfish sold in Benin City, Edo State, Nigeria

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Manuscript Info Abstract	
Manuscript History:	Bacteriological qualities and mercury levels in Cod (Gadus morhua) and
Received: 11 September 2013 Final Accepted: 21 September 2013 Published Online: October 2013	Saithe (<i>Pollachius virens</i>) stockfish sold in Benin City, Edo State, Nigeria was investigated. A total of 120 samples representing the head, filet and backbone were obtained from two markets namely: New Benin and Uselu market in Benin City. Standard aerobic pour-plate techniques were used for
Key words:	bacteria enumeration while mercury concentrations in stockfish samples
Methylmercury, Bacteria,	were determined using atomic absorption spectrophotometer (AAS). Cod
Stockfish, Contamination	stockfish filet had the highest bacterial count while Saithe backbones had the least total viable count Bacteria isolated from stockfish samples were <i>Staphylococcus aureus</i> , <i>Micrococcus spp.</i> , <i>Proteus spp.</i> and <i>Bacillus cereus</i> .
*Corresponding Author	Staphylococcus aureus (44.2%) was the most predominating isolate followed by Bacillus cereus (31.7%), Micrococcus sp. (18.3%) and Proteus sp. (5.8%). The mercury levels in the samples ranged between 0.01 to 0.04 mg/L. No mercury was detected in the backbone of stockfish samples analyzed. Based on the findings of this study, strict hygienic process should be carried out by marketers and buyers to reduce microbial loads. Periodic evaluation of stockfish sold is recommended in order to ascertain that mercury level is not above acceptable limit for safe consumption.
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Introduction

Fish is a low fat, high protein food and contains beneficial fatty acids such as omega-3 fatty acids. Omega-3 fatty acids are important for optimal brain and nervous system development in fetuses and infants. Fish constitute more than 60% of the protein intake in adults especially in rural areas (Adeleye, 1992). Fish flesh is tender and is better digested than beef or other types of animal protein (Adebayo-Tayo et al., 2008). Fish acceptability as an alternative protein source cuts across socio-economic, age, religious, and educational barriers.

Fish is an extremely perishable food. The quality of caught fish and its usefulness for further utilization in processing is affected by the fish capture method. Unsuitable fishing method does not only cause mechanical damage to fish, but also creates stress and conditions which accelerate fish deterioration without any preservative or processing measures (Okonta and Ekelemu, 2005). Spoilage proceeds as a series of complex enzymatic, bacterial and chemical changes that begin as soon as the fish dies (Junaid et al., 2010). Fish processing and preservation is carried out mainly to slow down or prevent the enzymatic, bacterial and chemical deterioration of fresh fish.

Akintola et al. (2006)reported different types of preservation methods; drying, smoking, freezing, chilling and brining. But the most prominent fish preservation method in Nigeria is smoke drying. Drying creates a hard outer layer helping to stop microorganisms from entering the food (Junaid et al., 2010).

Stock fish is gutted, beheaded fish (round, split or fillet) produced by natural or industrial drying, without the addition of salt or other additives (Norwegian Industry Standard for fish, 1998). Stockfish can only be made out of cod (*Gadus morhua*), haddock (*Melanogrammus aeglefinus*), saithe (*Pollachius virens*), ling (*Molva molva*) or Task (*Brosmebrosme*). In Nigeria, cod stockfish and saithe stockfish especially their heads are common in the markets. They are popular because of their rich taste and aroma and are sometimes eaten raw but mostly used in cooking

most native soups that complement the grain staples fufu and garri (Junaid et al., 2010). The name "Okporoko" among the Igbos (Eastern Nigeria) refers to the sound the hard fish makes in the pot and literally translate as that which produces sound in the pot. Stockfish is preserved by drying. During drying, 80% of the water is removed while other nutrients are concentrated. Stockfish is known as one of the richest source of protein, Vitamin B, Iron and Calcium.

Mercury occurs naturally, large amounts enter the aquatic environment from anthropogenic sources (Atuanya et al., 2011). Eating contaminated fish is the major source of human exposure to methyl-mercury. The populations most sensitive to the compound are fetuses, infants, and young children. Consequently, fish consumptions by pregnant women, young children and women of child bearing age is a particular cause of concern because of the likelihood of mercury exposure. Methyl-mercury, an organic compound, is the most toxic form of mercury to which humans are normally exposed. Methyl-mercury bioaccumulates with larger fish, which eat smaller ones, containing much higher levels than non-predatory fish (Physicians for Social Responsibility, 2004).

The objective of our study was to evaluate the bacteriological qualities and mercury content of cod (*Gadus morhua*) and saithe (*Pollachius virens*) stockfish sold in Benin City, Edo State, Nigeria with a view of assessing potential risk associated with their consumption.

Material and Methods

Sample collection. A total of 120 Cod (*Gadus morhua*) and saithe (*Pollachius virens*) stockfish samples which included the head fillets and backbones were purchased from New Benin and Uselu market in Benin City, Edo State, Nigeria. The samples were carried in sterile polyethylene bags, labeled according and taken to the laboratory for bacteriological and mercury analysis.

Inoculation and identification of microbial isolates. One gram of the ground stockfish samples were dissolved in 9ml of distilled water to obtain the stock solution. The stock solution was then serially diluted to obtain 10^{-1} , 10^{-2} and 10^{-5} dilutions. The aerobic colony count of stock fish samples were done by the pour-plate method as described by Cheesbrough (2006). 1.0 ml of 10^{-1} , 10^{-2} , and 10^{-5} dilutions of stockfish samples were plated out on molten nutrient agar plates containing 0.5 ml of antifungal. The plates were swirled gently for even distribution and incubated for 24 - 48 hours at 37^{0} C. The mean counts of bacteria colonies were determined. Bacteria pure isolates were characterized and identified using the criteria in Krieg and Holt (1994).

Total mercury (Hg) determination in stockfish samples. Digestion and filtration of stockfish samples was done according to the method of Atuanya et al., (2011). After filtration, the samples were allowed to stand for few minutes for colour development before digests were taken to a spectrophotometer (UV2100 Spectrophotometer, UNICON) to get the instrument reading for mercury using the method described by Willard et al. (1965).

Results and Discussion

The total bacteria counts. The total bacteria counts of isolates for the stockfish samples are shown in table 1. The isolates from the stockfish head samples were highest $(2.3 \pm 1.1 \times 10^3 \text{ CFU/g})$ for Cod (*Gadus morhua*) stockfish while saithe (*Pollachius virens*) had $1.9 \pm 3.1 \times 10^3 \text{ CFU/g}$. The backbone samples of cod stockfish had a total bacteria count of $1.1 \pm 2.1 \times 10^3 \text{ CFU/g}$ while $1.0 \pm 1.1 \times 10^3 \text{ CFU/g}$ was obtained in saithe stockfish backbone. 2.1 $\pm 0.9 \times 10^3 \text{ CFU/g}$ bacteria count was recorded in the filet of cod stockfish while $1.8 \pm 0.9 \times 10^3 \text{ CFU/g}$ was observed in saithe stockfish. This study shows that all the stockfish samples obtained from Uselu and New Benin markets were contaminated with bacteria. The higher bacteria count obtained in the head samples of both stockfish types compared to other body parts may be due to contamination by sellers and buyers. This is similar to the findings of Junaid *et al.* (2010). It also depicts the preferences of the stockfish head to the backbone and filets. Stockfish heads are more common in Nigerian markets than the backbone and filets. The presence of these bacteria in the stockfish samples might probably make the consumption of these stockfishes hazardous to health as similarly reported by Adebayo – Tayo et al. (2008).

Percentage occurrence of bacteria isolates. The bacterial isolates from both stockfish samples included bacteria of the general *Staphylococcus, Bacillus, Micrococcus* and *Proteus* as shown in table 2. *Staphylococcus aureus* had the highest percentage occurrence (44.2%). This was followed by *Bacillus cereus* (31.7%), *Micrococcus* sp. (18.3%) and *Proteus* sp. (5.8%) which had the least percentage occurrence. Table 3 shows the cultural, morphological and biochemical characteristics of the isolated bacteria. The isolated bacteria in the stockfish samples are similar to those

reported by Atuanya et al. (2011). Contrary to their study, *Proteus* sp. isolated in our present study had the lowest percentage frequency. The presence of these isolated bacteria could be due to contamination. Contamination of samples could also be because of unhygienic environment in which the stockfish are stored. Very often, the stockfish are displayed in open baskets or on tables beside the gutter or refuse dumps. This is in conformity with the report of Akande and Tobor (1992); Adebayo-Tayo et al. (2008) and Junaid et al. (2010).

Mercury levels in stockfish samples. The result of mercury levels in the different stockfish samples is recorded in table 4. The mercury level of cod stockfish {head (0.04 mg/L), filet (0.01 mg/L), backbone (ND)} and saithe stockfish {head (0.03 mg/L), filets (0.01 mg/L), backbone (ND)} are within the food and agricultural organization (FAO) acceptable limits. Although cod and saithe are ocean fish, low-level mercury was obtained. This may be because of their placement in the food chain. Thus, they are recommended for eating by pregnant women, young children and women of childbearing age. However, ocean fish like shark, tuna (fresh, frozen or canned) and mackerel should be avoided. Stockfish should be preferred to wild dried catfish, which had been shown to contain high level of mercury (Atuanya et al., 2011).

TABLE 1. Total viable bacteria loads (CFU/g) in stockfish samples.

	Samples			
Stockfish types	Head (CFU/g)	Filet (CFU/g)	Backbone (CFU/g)	
Cod (Gadus morhua)	$2.3 \pm 1.1 \text{ x } 10^3$	$2.1 \pm 0.9 \ge 10^3$	$1.1 \pm 2.1 \ge 10^3$	
Saithe (Pollachius virens)	$1.9 \pm 3.1 \ge 10^3$	$1.8 \pm 0.9 \text{ x } 10^3$	$1.0 \pm 1.1 \text{ x } 10^3$	

TABLE 2. Percentage occurrence of bacteria isolated.				
Bacteria	Number Isolated	Percentage (%)		
Staphylococcus aureus	53	44.2		
Bacillus cereus	38	31.7		
Micrococcocus sp.	22	18.3		
Proteus sp.	7	5.8		

Charateristics	B 1	B2	B3	B4
Cultural elevation	Low convex	Convex	Convex	Low
Margin	Entire	Smooth	Serrated	Smooth
Colour	Yellow	Cream	White	Yellow
Shape	Circular	Circular	Circular	Circular
Gram stain	+	+	-	+
Cell type	Cocci	Rod	Rod	Cocci
Cell arrangement	Cluster	Single	Single	Single
Biochemical		-	-	-
Catalase	+	+	+	+
Indole	-	-	-	-
Urease	+	+	+	+
Oxidase	-	-	-	-
Coagulase	+	-	-	-
Glucose	А	-	А	А

Key:

B1 = Staphylococcus aureus, B2 = Bacillus cereus, B3 = Micrococcus sp. B4 = Proteus sp.

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	Samples			
Stockfish types	Head (mg/L)	Filet (mg/L)	Backbone (mg/L)	
Cod (Gadus morhua)	0.04	0.01	ND	
Saithe (Pollachius virens)	0.03	0.01	ND	

Table 4 Mercury	levels	(mg/L) in	stockfish	samples.
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Note: ND = Not Detected. FAO limit for heavy metals in fish = 0.50mg/kg.

Conclusion

In view of the results obtained in the stockfish samples analyzed in this study, marketers and buyers to reduce microbial loads should carry out strict hygienic process. Periodic evaluation of stockfish sold should be carried out in order to ascertain that mercury level is not above acceptable limit for safe consumption. Also "human bio monitoring", that is analysis of samples of blood, urine, and so on should be carried out to indicate with certainty to what extent certain populations may be at risk.

Conflict of Interests

The authors declare that they have no conflict of interests.

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