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Microbial load and aflatoxin content of street foods sold around Federal University of Technology, Akure (FUTA) campus

Abstract

The microbial density and diversity, as well as the presence of aflatoxin in selected street vended-foods sold in FUTA campus was investigated in this study. Seven selected food samples viz; cooked rice, raw rice, garri, eba, peanut, suya and pap, were purchased from vendors at four popular locations around the school campus. The microbial load and diversity in the foods were analyzed using standard methods. The bacterial counts ranged from 3.1 x 10^4 CFU/ml to 4.9 x10^5 CFU /ml, with peanut sample having the highest count and cooked rice having the lowest count. Bacterial isolates belonging to eleven genera (*Staphylococcus, Streptococcus, Campylobacter, Listeria, Bacillus, Clostridium, Salmonella, Klebsiella, Pseudomonas, Enterobacter, Escherichia*) were obtained from the food samples. Mean Fungal counts of food samples ranged from 3.2 x 10^4 to 1.29 x10^6 SFU/ml. The fungi isolated include species of *Aspergillus*, *Penicillium*, *Fusarium, Cladosporium, Cryptococcus* and *Candida*. The high count of microorganisms coupled with the presence of *Salmonella typhimurium* and aflatoxins in some of the food samples portend danger to the consumers of such foods.

Keywords: Street, vended, foods, hygiene, microbial, aflatoxin

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Introduction

Food is needed for energy, body building and protective and regulatory functions. Food is therefore important to the survival and sustenance of man and animals. According to World Health Organisation, street vended foods (SVF) are ready-to-eat foods and beverages purchased on the street but prepared either at home or on the spot by Vendors or hawkers.[1,2] The patronage of SVFs had been on the increase as a result of urbanization.[3] Increase in patronage of SVFs had been related to the obvious advantages such as been cheap, convenient, sweetened with different flavor and easy accessibility.[3,4] Moreover, individuals especially the working class and Professionals are very busy with one activities or the other during the day and their involvement in long term schedule had endeared many to street vended and fast foods.[5]

Ma *et al*.,[6] reported that SVFs is convenient source of diet, especially in developing countries, and about 2.5 billion people patronise such foods. They are sold in environments where there are numerous consumers.[5] The patronage of SVFs is more common among University students. This is because students take care of their feeding and for time constraints.[7]

The major demerit of SVFs is safety due to the conditions under which such foods are prepared. Foodborne illnesses and fatality as a result of consumption of SVFs were respectively estimated at 600 million people and 400,000 deaths worldwide.[8] The following bacteria Viz; *Salmonella, enteropathogenic*, *Escherichia coli, Shigella, Campylobacter*, and *Aeromonas* have been implicated in foodborne disease.[9] Factors responsible for the outbreak of diseases as a result of consuming street vended foods had been identified as non-existence of good manufacturing practices, unhygienic conditions, and lack of awareness about food safety.[5,10,11] Large proportion of Students of the FUTA depends on SVFs. It is therefore important to access the likely health hazards these students are exposed to as a result of consumption of SVFs. The aim of the present study was therefore to assess the microbial load and diversity as well as the aflatoxin content of SVFs sold in and around FUTA campus.

Materials and methods

Study area

The study area for this research is centered on the **FUTA,** Nigeria (Lat 7°17'-7°19'N and Long 5°7'5°9'E. This area is densely populated and has a high prevalence of local food vendors selling various types of foods. These foods are commonly consumed by students, staff, and residents of the surrounding communities. The area is also home to various markets where these foods are sold, making it a significant hub for food distribution and consumption.

Sample collection

Foods samples Viz; raw rice and Garri were collected from the popular Oja-oba market in Akure, Ondo state. Cooked rice and Eba Food samples were aseptically collected into sterile nylon bag from some restaurants at South-gate, FUTA. Suya was collected from Aluta market along Jibowu hostel and other food samples (pap and peanut) were collected from North-gate, FUTA, Nigeria.

Isolation of microorganisms from food samples

Isolation of microorganisms was achieved by homogenizing food sample, each homogenate (1g) was weighed and transferred into 10 mL of sterile water as a stock solution and serially diluted to 10^{-5}. Diluent (0.1mL) of the serially diluted sample was transferred into Petri dish containing Plate Count Agar (PCA, Lab M) for bacteria and Potato Dextrose Agar (PDA, Lab M) for fungi. Incubation of plates was at 37°C for 24 h and 48 h for bacteria and fungi, respectively. After incubation, colonies were counted using the colony counter (TT-20, Techmel and Techmel, USA) and results were expressed as colony forming unit per gram (CFU g-1) for bacteria and spore forming unit per gram (SFU g-1) for fungi. Pure cultures were further obtained by subculturing discreet colonies.

Identification of microorganisms

Identification of microorganisms was done through Gram stain and batteries of biochemical tests such as catalase, coagulase, urease, sugar fermentation tests and so on.[12,13] Results obtained were interpreted using Bergey's Manual of Systematic Bacteriology.[14] Fungal isolates were identified using colonial and morphological features.

Detection of aflatoxin in food samples

Aflatoxins content of food samples was quantified using High-Performance Liquid Chromatography (HPLC). Food samples (25g) are weighed into 7 different glass bottles and properly labeled. Sodium chloride (5g) was added into the glass bottle and 150ml of (Methanol: Deionized water) in ratio (80:20). The mixture was then mixed evenly on an orbital shaker at 250rpm for 15 minutes. The mixture was then filtered using a pre-folded filter paper and 10ml of the filtrate was added to 60ml of Phosphate Buffer Saline (PBS) and 50ml was taken from the mixture and "cleaned up" using an Immuno-Affinty Clean up Column. While running the column, it was washed with 10mls of deionized water twice. The column is then eluted with 500µl and the 750 µl after a minute with Lichrosome methanol. It was then put into a 5ml volumetric flask and made up to mark with deionized water. Thereafter, 200 µl is injected into the HPLC system and ran thrice. The chromatogram generated was then read.

Stastistical analysis

All experiments were carried out in triplicates. Data were statistically analyzed by one-way analysis of variance (ANOVA) and means were compared with New Duncan's Multiple Range Test (SPSS 20.0 version). Differences were considered significant at $P \leq 0.05$.

Results

Total microbial counts of food samples

The microbial load of SVF samples is shown in Table 1. The number of bacterial isolates obtained varied from one food to the other with peanut having the highest bacteria count of $4.9x10^5$cfu/ml and cooked rice having the least bacterial count of $3.1x10^4$cfu/ml. Similarly, the number of fungal isolates obtained varied from one food to the other with Suya having the highest fungal count of $1.29x10^6$sfu/ml in all dilutions and Eba having the least fungal count of $3.2x10^4$sfu/ml.

Identity and distribution of microbial isolates in food samples

The bacterial isolates were tentatively identified as *Salmonella typhimorium, Esherichia coli, Listeria monocytogenes, Enterobacter* sp., *Staphylococcus aureus, Klebsiella* sp., *Bacillus cereus, Campylobacter jejuni, Pseudomonas aeruginosa, Clostridium perfrigens* and *Streptococcus pyogenes*. *Staphylococcus aureus* and *Bacillus cereus* had the highest distribution across the food samples as seen Table 2. The fungi identified are *Fusarium oxysporium, Aspergillus niger, Aspergillus flavus, Aspergillus fumigatus, Penicillium* sp., *Alternaria* sp., *Cladosporium* sp and *Cryptococcus* sp. *Aspergillus niger* and *Fusarium oxysporium* had the highest distribution across the food samples. (Table 3)

Aflatoxin content of food sample

Raw rice was found to contain Aflatoxin B1 (AFBI) and Pap was also positive for Aflatoxin B1 (AFBI), Aflatoxin B2 (AFB2), Aflatoxin G1 (AFG1), Aflatoxin (AFG2) while Cooked rice, Garri, Eba and Suya had no aflatoxin (Table 4).

Table 1 Microbial load of food samples

Food Sample	Bacterial Count (cfu/g)	Fungal count (sfu/g)
Raw rice	$2.2x10^5$	$9.8x10^4$
Cooked rice	$3.1x10^4$	$4.9x10^4$
Gari	$6.2x10^4$	$6.1 x10^4$
Eba	$3.9 x10^4$	$3.2 x10^4$
Suya	$1.8 x10^5$	$1.3 x10^6$
Pap	$4.8 x10^4$	$1.0 x10^5$
Peanut	$4.9x10^5$	$2.0 x10^5$

Values are means of replicates

Table 2 Frequency of distribution of bacteria in the food samples

Organism	Suya	Peanut	Pap	Cooked rice	Raw rice	Eba	Garri	FOO
Staphylococcus aureus	-	+	-	+	+	+	-	57%
Salmonella typhimorium	-	+	-	+	-	-	+	43%
Klebsiella sp.	+	-	-	-	-	-	-	14%
Escherichia coli	-	-	+	-	+	-	+	43%
Bacillus cereus	-	+	-	+	-	+	+	57%
Streptococcus pyogenes	-	-	+	-	-	-	-	14%
Enterobacter sp	-	-	-	-	-	+	-	14%
Clostridium perfringens	-	-	+	-	-	-	-	14%
Campylobaceter jejuni	+	-	-	-	-	-	-	14%
Listeria monocytogenes	-	-	-	-	+	-	-	14%

Keys: +, Positive result; -, Negative result; FOO, Frequency of Occurrence

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Table 3 Frequency of distribution of fungi in the sample

Organism	Pap	Suya	Peanut	Garri	Eba	RR	CR	FOO
Aspergillus flavus	+	-	-	-	-	+	-	29%
Aspergillus parasiticus	+	-	-	-	-	-	-	14%
Aspergillus niger	+	+	-	+	+	+	+	71%
Fusariumoxysporium	-	-	+	+	+	-	+	57%
Candida sp.	-	+	-	-	-	-	-	14%
Cryptococcus sp.	-	+	-	-	-	-	-	14%
Cladosporium sp.	-	-	-	+	-	+	-	29%
Alternaria sp.	-	-	+	-	-	-	-	14%
Penicillum nutatum	-	-	+	-	+	-	+	43%

Keys: +, Positive result; -, Negative result

CR, Cooked Rice; RR, Raw Rice; FOO, Frequency of occurrence

Table 4 Aflatoxin content of food samples

Sample	RT (min)	Response	Amount(ng/g)	Peak Type	AFL
Raw rice	13.069	23.742	0.514	Ordnr	AFB1
Cooked rice	-	-	-	-	-
Garri	-	-	-	-	-
Eba	-	-	-	-	-
Peanut	-	-	-	-	-
Pap	8.28	12.72	0.266	Ordnr	AFG2
	9.567	29.278	1.144	Ordnr	AFG1
	10.997	208.508	2.211	Ordnr	AFB2
	13.113	1669.371	36.159	Ordnr	AFB1
Suya	-	-	-	-	-

Keys: AFL, Aflatoxin; -, Absence of Aflatoxin

AFB1, AflatoxinB1; AFB2, AflatoxinB2; AFG1, AflatoxinG1; AFG, AflatoxinG2

Discussion

Food vendors involved in street vended foods are known not to adhere to good manufacturing practices.[5] Consequently, such foods are suspects in terms of safety and wholesomeness. In the present study, the bacterial and fungal loads of the food samples ranged between $3.1x10^4$ to $4.9x10^5$ cfu/ml and $3.2x10^4$ to $1.29x10^6$ sfu/ml respectively. This is similar to the report of Ogidi *et al*.,[5] and recently Moges *et al*.[15] The high microbial load had been attributed to the poor handling of foods and unhygienic practices by vendors.[16,17] It has been reported that most street-food vendors lack understanding of the principle of good manufacturing practices.[18] In another study, the hygienic status of food-handlers was also found to contribute significantly to the low quality and safety of SVFs examined in Porto, Portugal.[19] Lack of personal hygiene and dirty environment where such foods are produced coupled with illiteracy level of most vendors has discredited the acceptance of street vended foods.[5]

Microorganisms isolated from the street vended foods are *Staphylococcus aureus, Streptococcus pyogenes, Campylobacter jejuni, Listeria monocytogenes, Bacillus cereus, Clostridium perfringens, Salmonella typhimorium, Klebsiella* sp, *Pseudomonas aeruginosa, Enterobacter* and *Escherichia coli*. Similar groups of microorganisms had been isolated in street vended foods in South Africa, Korea and Ehiopia.[15,20,21] The occurrence of bacterial isolates in the food samples ranged from 14% to 57%. *Staphylococcus aureus* (57%) and *Bacillus cereus* (57%) were the highest in prevalence. The high prevalence of *S. aureus* had been associated to its resistance to heat, drying and radiation.[5,22] Moreover, the unsanitary behaviour of vendors is also a factor.[17] *Staphylococcus aureus* are found on the skin and mucous membranes, and humans are the major reservoir for these organisms.[23,24] Unhygienic activities of food vendors can therefore help spread *S. aureus* to food. *Bacillus cereus* is a spore former and this helps it to survive longer in extremes of temperature.[25] This may also explain the prevalence of *Bacillus cereus* in the food samples. Generally, the presence of *Salmonella*, *Escherichia coli*, *Bacillus cereus*, *Staphylococcus aureus* and molds portends harzards to the consumers of street vended foods.[15,26]

Fungi isolated from the food samples belongs to the genera *Aspergillus*, *Penicillium*, *Fusarium, Cladosporium, Cryptococcus* and *Candida*. Similar group of fungi had been isolated from street vended food.[5,26] The occurrence of these moulds especially *Aspergillus* species, is of public health concern since they produce mycotoxins which are most often not eliminated through processing.[26,27]

Aflatoxin B1, B2 and G2 were present in raw rice and pap out of the food samples analyzed. Mycotoxins, secondary metabolites are produced mainly by the gerera *Fusarium, Aspergillus* and *Penicillium*.[28] Nnam and Eruteya[29] recently reported the presence of aflatoxins in ready-to-eat roasted snack in Port Harcourt, Nigeria. Makun *et al*.,[14] had earlier reported that colonization foods by the above moulds are responsible for the presence of mycotoxins in some foods. It has been reported that chronic exposure to low levels of AFB1 poses a serious health and economic hazard.[30]

From this study, the evaluation of the microbial quality of locally consumed food around the Federal University of Technology, Akure campus revealed the presence of microorganisms of health hazards in the street vended foods. Moreover, the presence of highly toxic aflatoxin B1 (AFB1) especially in pap portends double danger to

the consumers of such food. Based on the data gathered from this study, the necessary body saddled with ensuring food safety is advised to monitor these street-vended foods sold around FUTA campus. Moreover, periodic enlightenment programme should be organized for vendors of these foods to intimate them on the principle of good manufacturing practice to ensure the production of safe and wholesome ready to eat food.

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None.

Conflicts of interest

The authors declare there is no conflict of interest.

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