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Full-Length Research Paper

Microbial Examination of Jollof Rice Served in Plateau State University Community for Pathogenic Organisms

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ABSTRACT: Food is an important tool for promoting health and avoiding illness. Poorly prepared and contaminated street food may be the source of foodborne illnesses. Jollof rice was tested for microbiological safety on the Plateau State University campus and in the surrounding community in Bokkos, Nigeria. Jollof rice samples were obtained from seven (7) different food vendors and tested using standard methods. Ten pathogenic species were isolated, including five bacteria and five fungi. The isolated bacteria are *Escherichia coli, Bacillus cereus, Staphylococcus* species, *Bacillus lincheniformis, and Bacillus firmus,* while the isolated fungi are *Aspergillus niger, Aspergillus fumigatus, Cladosporium* species, *Penicillium* species, and *Trichophyton* species. According to the results, *Escherichia coli* had the highest percentage of isolated bacteria (30.7%), followed by *Bacillus cereus* and *Staphylococcus* species (23.1% each), while *Bacillus firmus* had the lowest percentage (7.7%). *Aspergillus fumigatus, Cladosporium, Penicillium*, and *Trichophyton* species all had a 14.3% prevalence among the fungi. Among fungi, Aspergillus niger has the highest percentage of occurrence (42.8%). Food sellers selling prepared foods on the street must adhere to strict public health regulations and follow food sanitary procedures.

Keywords: Bacteria, fungi, microbial, Plateau State University

INTRODUCTION

Rice is a significant staple food, particularly in Nigeria and Sub-Saharan Africa (Jemikalajah, 2018). Nigeria is the world's second-largest rice importer and Africa's largest (Abbas et al., 2018; Nzeka, 2019). As a result, unlike in the past, when rice was only consumed during special ceremonies or religious holidays, rice is now a staple of Nigerian cuisine, and it is widely processed/cooked ready-to-eat by sellers across the

country (Christopher et al., 2022). A nutritious rice supper is ideal for a variety of dietary requirements. It has a protein content comparable to wheat, corn, and sorghum and is primarily composed of carbohydrates. It also has a low fat content. In comparison to other cereals, it has a high digestibility and a good supply of potassium, vitamin E, and vitamins B (thiamine, niacin) (Christopher et al., 2022). Rice farming is best suited to nations and regions with low labor costs and abundant rainfall because it is labor- and water-intensive (Okobia and Orogu, 2021).

Food contamination in Nigeria is currently a problem due to microorganisms, pesticides, herbicides, and other chemicals used for weed control during production and insect control during storage. Foodborne infections affect the majority of countries, including Nigeria, and food safety has become a global issue (Oku and Alagoa, 2019).

The pathogenic contamination of processed/cooked ready-to-eat jollof rice by street vendors is causing major concern at Plateau State University. As a major source of income for many people, particularly women, as well as a chance for self-employment and the development of business skills with little capital investment, street vended foods also provide a cheap, convenient, and frequently nutritious source of food for the poor in urban and rural areas (Mohammed and Shehasen, 2020). However, this industry poses a number of health risks (Henry et al., 2017). *Escherichia coli, Klebsiella* spp., *Proteus* spp., *Salmonella* spp., *Pseudomonas* spp., and *Enterobacter* spp. contamination of ready-to-eat jollof rice from sold foods is of primary concern due to the risk that these organisms will cause an outbreak of food-borne illnesses like gastroenteritis, dysentery, and typhoid fever (Oluyege *et al*., 2009). There is no documented evidence of microorganisms associated with jollof rice sold around the Plateau State University, Bokkos Nigeria. This research was carried out to investigate the pathogenic organisms found in jollof rice sold in several restaurants in the Plateau State University Bokkos campus.

MATERIALS AND METHODS

Study area

The study was carried out at Plateau State University's Bokkos campus. Bokkos Local Government Area is situated 77 kilometers from Jos, the capital of Plateau state. There are eight districts in Bokkos, and the two main languages are *Ron-kulere* and *Mushere.*

Sample collection

Jollof rice samples were obtained from seven (7) different food vendors, placed in separate sterile cellophane bags, and delivered within an hour to the microbiological laboratory for detailed analysis.

Preparation of media

In accordance with manufacturer's instructions, all the media—Blood agar (BA), MacConkey Agar (MCA) and Sabouraud Dextrose Agar (SDA)—were made and weighed. Before usage, they underwent a 30-minute autoclave at 121°C (Cheesbrough, 2000; Jemikalajah, 2018).

Isolation of bacteria

In eight sterile test tubes, 9ml of distilled water was used for serial dilution. To ensure thorough mixing, 1ml of the homogenized jollof rice samples was added to the first tube of distilled water. Using a sterile pipette, one milliliter (1ml) was taken out of the first test tube and put into the second test tube. For the remaining test tubes, this procedure was repeated.

Using a sterile pipette, an aliquot of 0.1 ml was taken from test tubes 6 and 8 (0.001 and 0.00001, respectively) and inoculated into the sterile petri dishes with already prepared agar. For 24 hours, the culture plates were incubated at 37°C. Colonies were harvested using a wire loop after incubation and sub-cultured in petri dish plates with recently made agar (Jemikalajah, 2018).

Identification of bacteria

Colonies were distinguished on the bases of cultural characteristics such as colony size, shape, colour, consistency and haemolytic characteristics (Cullimore, 2010). The bacterial growth was sub-cultured onto BA and MCA slant and incubated at 37ºC for 24 hours. Gram staining, microscopy, and different biochemical tests such as coagulase, catalase, Oxidase, glucose, maltose, sucrose, Mannitol, lactose, urease, and indole were performed to confirm the different bacterial species. The organism was morphologically compared with bacteria identification atlas for further confirmation (Christopher *et al*., 2022; Ibrahim *et al*., 2020; Cullimore, 2010; Chesshrough, 2003).

Isolation of fungi

Sabouraud Dextrose Agar (SDA) was used for culture, and infected plates were cultured at room temperature (25°C) for four days to isolate fungi (Ibrahim *et al*., 2020).

Identification of isolates

To identify the organism, the various isolates were macroscopically inspected, then biochemically tested. The growing section of the fungal mycelia on the

Sabouraud Dextrose Agar medium was cut, placed on a grease-free microscope slide, and covered with a cover slip. A x10 magnification was used to magnify the mycelium for examination and identification with the aid of atlas for the identification of fungi (Watanabe, 2010).

Data analysis

Data obtained were analyzed using R Console software (version 3.6.1). The percentage of occurrence of bacterial and fungal isolates from jollof rice were compared using Pearson's Chi Square (χ^2) test. Level of significance was set at P-value < 0.05.

RESULTS

Results of the biochemical testing is displayed in (Table 1). The result indicates that most of the isolates produce either acid-gas (AG) or acid (A) during the triple sugar iron agar (TSIA) test. Majority of the isolated bacteria were positive for catalase and glucose tests. The percentage of bacterium isolates is displayed in (Table 2). *Escherichia coli* (30.7%) was the most frequently isolated bacterium from jollof rice, followed by *Bacillus cereus* and *Staphylococcus* species (23.1% each), while *Bacillus firmus* (7.7%) was the least frequently recovered bacterium from the samples of jollof rice analyzed. Therefore, there was a significant difference ($\chi^2 = 15.477$, df = 4, P = 0.003807) in the prevalence of bacteria isolated from jollof rice.

The morphological characteristics of the fungus species isolated for this study were shown in (Table 3). *Aspergillus Niger* (3), *Aspergillus fumigatus* (1), *Cladosporium* (1), *Penicillium* (1), and *Trichophyton* species (1) are the fungal species isolated. Table 4 displays the common species of fungi isolated from jollof rice. In this investigation, *Aspergillus niger* (42.8%) was most isolated, whereas *Aspergillus fumigatus*, *Ciadosporium, Penicillium*, and *Trichophyton* species all had a prevalence of 14.2%. Hence, there was a very high significant difference ($\chi^2 = 32.85$, df = 4, P < 0.0001) in the prevalence in the occurrence of fungi isolated from jollof rice.

DISCUSSION

Jollof rice samples were collected from eight restaurants in the Plateau State University (PLASU) Bokkos neighborhood and analyzed for microbial contamination. This study's findings indicate that jollof rice is associated with a variety of pathogenic bacteria and fungi. This backs up a similar investigation conducted in Abraka, Delta State, Nigeria, where rice samples were found to be contaminated in varying amounts by bacterial and fungal isolates (Jemikalajah, 2018).

Ten different pathogenic organisms were isolated, identified, and confirmed from jollof rice sold on the PLASU campus. There were five types of bacteria and five types of fungi.

The findings of this study revealed a significant variation in the prevalence of bacterial isolates from various jollof rice samples collected on the PLASU campus. This implies that jollof rice sold on campus may have a different microbial flora. Furthermore, poor grain quality, contaminated water, improper vendor handling and processing of food, contamination caused by storage facilities, and other factors may all contribute to contaminated jollof rice. This result is compared to others obtained in Nigeria, including Ozoro (Delta State), Gombe Metropolis, and Ekpoma (Edo State), where several species of microorganisms were isolated from ready-to-eat street foods (Osatohanmwen et al., 2019; Ibrahim et al., 2020; Okobia and Orogu, 2021).

Escherichia coli in cooked and ready-to-eat street food illustrates secondary contamination because it is known to be present in the gastrointestinal tract of warm-blooded animals but not in the environment as a natural flora (Osatohanmwen *et al*., 2019). As already mentioned, using dirty water might cause pollution. This finding contrasts with another study in the United Kingdom, where *Escherichia coli* was not found present in ready-to-eat burgers and pastry meat in the United Kingdom (Meldrum *et al*., 2006). *Escherichia* coli belongs to the *Enterobacteriaceae* genus.

Meningitis, nosocomial pneumonia, dysentery, diarrhea, gastroenteritis, and urinary tract infections can all be caused by a specific strain. Foodborne illnesses caused by the *Escherichia coli* 0157H7 strain, which causes a serious and potentially fatal condition known as hemorrhagic colitis, characterized by bloody diarrhoea and excruciating abdominal pain, can also be caused by the *Enterohaemorrhagic Escherichia coli* (EHEC) subunit (Peters et at., 2017). The discovery of *Staphylococcus* species in the jollof rice being sold on the PLASU campus is consistent with similar findings where the organism was found to be responsible for contaminating the ready-to-eat items examined (Hartati *et al*., 2020; Oranusi, 2013).

The occurrence of *Staphylococcus* species in food products is primarily due to human interaction, and as the bacterium naturally lives on the skin and nasal passages of humans, this reflects poor hygiene habits of the vendors (Okobia and Orogu, 2021). It is quite a concern for the public's health that *Staphylococcus aureus* is present. The majority of *Staphylococcus aureus* strains are recognized as being harmful, as it is frequently documented, mostly as a result of the heat stable enterotoxins that they create in direct proportion to their inoculum level (Peters *et al*., 2017). The large significant difference in the percentage of fungi isolated from the

Table 1: Result of Biochemical Tests on isolated bacteria.

Restaurant	Gram Reaction	Catalase	Coagulase	Indole	Citrates	Tsia	Mannitol	Xylose	Dulcitol	Arabinose	Sucrose	Glucose	Urease
1	Gram -ve rods	+	+	+	-	AG	+	-	-	.	v	+	-
	Gram +ve	+	+	-	+	A	-	-	+	+	+	+	-
2	rods												
	Gram +ve	+	+	+	-	AG	+	-	-	.	v	+	-
	rods												
3	Gram +ve	+	-	-	-	AG	+	+	-	-	v	+	-
	cocci												
	Gram +ve	+	+	-	+	A	-	-	-	-	v	+	-
	rods												
4	Gram +ve	+	-	-	-	A	-	+	-	-	-	+	-
	cocci												
	Gram +ve	+	+	-	+	A	+	+	.	+	+	+	-
	rods												
5	Gram -ve rods	+	+	+	+	AG	+	+	-	.	v	+	-
	Gram +ve	+	-	-	-	A	-	-	+	+	+	+	-
	rods												
6	Gram -ve rod	+	-	+	+	AG	+	+	-	-	v	+	-
	Gram +ve	+	-	-	-	-	+	-	-	-	-	-	-
	cocci												
7	Gram +ve	+	+	-	+	A	+	-	-	-	-	+	-
	rods												

Key
+ = indicate a positive result; - = Indicate a negative result
A= Indicate acid production; AG = Indicate Acid Acid gas production
V = Variable

Table 2: Percentage occurrence of bacteria isolates from Jollof rice.

Name of organism	NO. of organism	Percentages
Escherichia Coli	4	30.7%
Bacillus cereus	3	23.1%
Staphylococcus species	3	23.1%
Bacillus lincheniformis	2	15.4%
Bacillus firmus	1	7.7%

$\chi^2 = 15.477$, df = 4, p-value = 0.003807

Table 3: Morphological Features of Fungi Isolated from Different Jollof Rice.

Restaurant	Septation	Colour	Types of spores	Suspected organism
1	Septate	White	Conidiospore	*Asperginus niger*
2	Septate	Blue-green	Conidia in chain	*Aspergillus fumigatus*
3	Non- septate	Green	Conidiospore	*Penicillum* spp
4	Septate	White	Conidiospore	*Aspergillus niger*
5	Septate	white	Conidiospore	*Aspergillus niger*
6	Septate	Dark	Conidiospore	*Cladosporium species*
7	Septate	White	Conidiospore	*Trichophyton* spp.

Table 4: Percentage occurrence of fungi isolated from different Jollof Rice.

Name of Organism	No. of organism	Percentages
Aspergillus niger	3	42.8%
Aspergillus fumigatus	1	14.3%
Cladosporium species	1	14.3%
Penicillium species	1	14.3%
Trichophyton species	1	14.3%

$\chi^2 = 32.85$, df = 4, P < 0.0001

various types of jollof rice analyzed suggests that, among other things, *Aspergillus* species are the main fungal pathogens identified in this study. This outcome is comparable to the outcome in another study conducted in Amassoma, Bayelsa State, Nigeria, where *Aspergillus* species were isolated from ready-to-eat jollof rice (Oku and Alagoa, 2019).

According to other investigations conducted in Nigeria, fungi have reportedly been isolated from jollof rice that is ready to consume (Odu and Akano, 2012; Oranusi and Braide, 2012; Oranusi *et al*., 2013).

The presence of fungi in the samples could be due to improper storage, which caused the foodstuffs to become damp, allowing the fungi to grow in the ready-to-eat food sold in the university community. Aflatoxin, a key metabolite produced by fungi, has been shown to be extremely harmful to humans as well as all domestic and laboratory animals (Oku and Alagoa, 2019).

Due to the identified food pathogens that were isolated from the analyzed food samples, the study's findings are quite concerning.

Due to the lack of essential infrastructure and services, difficulty in regulating most vending operations due to their diversity, mobility, and ephemeral nature, and constant risk to the general public, those who consume these things are always at risk (Peters et al., 2017). Along the entire food chain of food production, processing, and storage, workers who touch food play a crucial role in ensuring food safety (Jemikalajah, 2018; Oku and Alagoa, 2019).

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Conclusion

The findings of this investigation on the jollof rice sold on the Plateau State University campus revealed that all food samples contained bacteria and fungi. The most common microorganism isolates were *Escherichia coli, Staphylococcus aureus, and Aspergillus species*. The presence of these organisms in the prepared, ready-to-eat street food items under investigation was extremely dangerous. As a result, it is critical that relevant public health and food safety agencies organize food safety and hygiene training and instruction for food vendors, particularly compliance with the hazard analysis and critical control points principles (HACCP) during food preparation, packaging, and serving to consumers.

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