

THE PREVALENCE OF *S. aureus* AND *E. coli* IN JOLLOF RICE SOLD IN VARIOUS EATERIES IN OZORO

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ABSTRACT

The prevalence of *Staphylococcus aureus* and *Escherichia coli* in jollof rice was carried out to determine the microbial quality of jollof rice sold in various eateries in Ozoro, Delta State. Eleven (11) samples were collected from various eateries and brought to the laboratory for analysis. Six (6) bacteria species was identified; *Escherichia coli*, *Klebsiella* spp., *proteus* spp., *citrobacter jejuni*, *Streptococcus pyogenes* and *Staphylococcus aureus*. The total heterotrophic plate count ranges from 1.2×10^5 CFU/ML to 6.2×10^5 CFU/ML. *E. coli* and *Staphylococcus aureus* has the highest percentage occurrence of 27.27% while *Klebsiella* spp., *Proteus* spp., *Citrobacter jejuni* has the least percentage occurrence of 9.09%. These micro-organisms isolated are pathogenic and are toxic when ingested in contaminated food.

INTRODUCTION

Safe food is a basic human right despite the fact that many foods are frequently contaminated with naturally occurring pathogenic microorganisms which cannot be detected organoleptically (seen, smelled or tasted) but can cause diseases death especially if the way they are conserved during exposition for sale provides condition for those micro-organisms to grow and reach considerable levels of contamination (WHO, 2000).

There are reasons why people eat away from home. These include among others absence from home while travelling, studying, while at work or need for a change in terms of food type or location and as such, many people resort to buying street vended food which may be poorly processed. These situations however, have resulted to the transfer of food sanitary measures and poor food handling from individuals/ families to the food vendors who rarely enforce such practices (Musa and Akande 2002).

Rice is the grain with the second highest worldwide production after maize (corn). Since a large portion of maize crops are grown for purposes different from human consumption rice is probably the most important grain with regards to human nutrition and caloric intake, providing more than one fifth of the total calories consumed worldwide by humans. In Africa, rice has been used to improve nutrition quality, boost food security, foster rural development and support sustainable land care (Chomvarin *et al.*, 2006). It is basically grown as an annual plant, and its cultivation is well-suited to countries and regions with low labour cost

and high rain fall, as its cultivation is very labour-intensive and requires plenty of water.

A number of observation studies have shown that these foods are sometimes held at improper temperatures, excessively handled by food vendors and sold at very dirty surroundings (WHO, 2001, 2003; Muide and Kuria, 2005; Ghosh *et al.*, 2007). In addition, the vendors practice poor personal hygiene and reports of foods vendors being carriers and therefore could serve as a potential source of transmission of enteric fevers are many. Most of the vendors have either no formal education or few years of schooling and therefore, lack of knowledge on proper food handling and their role in the transmission of pathogens (Mensah *et al.*, 2002). At the same time, most people who use these food services are more interested in its convince than the question of microbiological quality and hygiene. The microbiological quality of food indicates the amount of microbial contaminant it has, a high level of contamination indicates low quality of food storage and its handling and more likely to transmit infection and the reverse is true (Anonymous, 1988). Thus, concerns have been raised by the food and agricultural organization (FAO) and others about these foods serving as a potential source of food poisoning outbreaks (Chaleravarty and Canet, 2002). Ready – to – eat foods refer to foods that do not require further significant preparation other than reheating or completion of a cooking process (FEHD, 2001; FSAI 2001). It has been reported that ready-to-eat take away foods account for a large volume of sales of the food service volume output (Powers and Barrow, 1999).

The microbiological quality of ready – to – eat rice is said to be influenced by a number of factors such as cuisine type, rice type, cooking serving methods and management/ food handling (FEHD, 1995). In 2009, De Bess *et al.*, (2009) reported that 32% of food handlers in ready-to-eat (enters in Washington had no knowledge of food safety practices and of prevention of food borne disease. This may result in the transmission of food-borne pathogens to the people consuming such foods. As a consequence, Nichols *et al.*, (1999) demonstrated that the microbiology quality of ready-to-eat rice from some eateries is low compared to some others. Due to the number of outbreaks of food poisoning from ready-to-eat food centers, it has been suggested that laws should be enacted on the establishment of ready-to-eat food centers, and that the staff of such centers should be trained on proper hygiene procedures as well as the transmission of food-borne disease, especially in developing countries.

MATERIALS AND METHODS

Study area

This research was conducted in Ozoro, Delta State of Nigeria. Ozoro is the local Government headquarters of Isoko North Local Government of Area Delta State. The people are Isoko speaking and hospitable. Their main activities are food crop, farming accompanied by some hunting. They are also engaged in trade of food crop for cash to meet the other basic house hold needs. The region experience higher rainfall and humidity most of the year.

Collection of samples

Several samples of jollof rice were obtained from different fast food centers. Samples were collected in a sterile container when freshly cooked and was taken under aseptic condition to the laboratory for microbiological analysis.

Materials

The following materials were used for the experiment Nutrient Agar, Blood Agar, weighing balance, conical flask, test tube, measuring cylinder, petri dish, slides, microscope, wire loop, auto clave & filter paper.

Method

Several dilutions were prepared and spread on standard bacteriological culture media for enumeration of the different microbial counts. The plates were incubated in duplicate to ensure accuracy.

Total bacterial count

Nutrient agar plates were used to determine the total bacterial plates count and the plates were incubated for 24 hours at 37⁰c. After incubation, the number of colonies on each plate was counted and averages were computed for each sample.

Microbiological method

All the media were according to manufacturer's specification.

Gram staining: Preparation and fixing of smear

A colony from purified sub culture was indicated and emulsified in sterile distilled water and a thin preparation was made on the slides. Its evenly spread to cover an approximated area of about 15-20mm in diameter on the slide. After the smear was made, it was left on a dry area to dry; making sure it was protected from dust and sunlight. The smear was that fixed using gentle heat by rapidly passing the slide with the smear uppermost three (3) times through the flame of a Bunsen burner. The slide was then placed on the back of the hand just to make sure too much heat was not used which can affect or even kill the micro-organism, the smear was then allowed to cool before staining was done.

Staining

The glass slide containing the smear was placed on the staining rock and covered with crystal violet stain and allowed for 60 secs. The iodine was wasted with distilled water. The water also tipped off and the smear was covered with lugol's iodine for boxes. The smear was decolorized rapidly with acetone and washed immediately with clean water. The back of the slide was wiped clean and placed on a draining rack for the smear to air-dry. Using a lens to check the staining and to see the distribution of the material.

Biochemical test

Citrate utilization test

Using a sterile wire loop a colony from purified sub-cultures was isolated and stabbed straight down into the slanted agar medium. The wire loop was removed, flame sterilized, and the inoculum was streaked on the surface of the stand. The test tube was covered tightly with a screw cap and labeled according for 24hrs at 37⁰c. it was then removed and observed for fermentation shown by a change in colour from green to blue.

Indole test

Using a sterile wine loop a colony from the purified sub-cultures was isolated and inoculated into bijou bottle, was then flamed. Sterilized and covered highly with a screw cop and labeled accordingly, it was incubated for 48 hours at 37⁰c. the bijou bottle were then shaken gently and left standing for 10 mins. Examination for positive result was done by the formation of red color in form of a ring on the surface layer of the culture media.

Motility test

Using a sterile needle, the nutrient agar medium was inoculated to make 5 stables of the test organisms to the depth of 1-2 cm of the bottom of the tube. Then test tubes were then incubated at 37⁰c for 24 hours. The line of incubation was examined for cloudiness showing the organisms are motile.

Oxidase test

The oxidize test is used in the identification of pseudomonas species as it produces the enzyme cytochrome oxidize. Piece of filter paper was placed in a clean petri dish and 25 drops of the oxidize reagent (pamino dimethy iodine) added to it. Using a sterile wire

loop a from the purified sub culture nutrient agar was removed and a smear was made on the filter paper was left for losses after which it was observed for the development of a blue-purple color as a positive oxidize test.

RESULT AND DISCUSSION**Result****Table 1: Biochemical Characteristics and Identification of Organisms.**

CULTURAL MORPHOLOGY	GRAM	CATALASE	OXIDASE	MOTILITY	GAS	GLUCOSE	LACTOSE	H ₂ S	ACID	CITRATE	INDOLE	ISOLATES
Cocci	+	+	-	-	-	+	+	-	+	+	-	<i>S. aureus</i>
Cocci	+	-	-	-	-	+	+	+	+	-	-	<i>S. pyogenes</i>
Rod	-	+	-	+	+	+	+	+	+	+	-	<i>Citrobacter</i>
Rod	-	+	-	+	+	+	+	-	+	+	-	<i>Klebsiella</i>
Rod	-	+	-	+	+	+	+	+	+	-	+	<i>E. coli</i>
Rod	-	+	-	+	+	+	-	+	+	+	-	<i>Proteus</i>

Table 2: Shows the total bacteria count of isolated organism.

Samples	Numbers isolated
A	2.7 X 10 ⁵
B	2.9 X 10 ⁵
C	4.2 X 10 ⁵
D	3.4 X 10 ⁵
E	5.5 X 10 ⁵
F	2.4 X 10 ⁵
G	1.2 X 10 ⁵
H	4.8 X 10 ⁵
I	3.7 X 10 ⁵
J	1.3 X 10 ⁵
K	6.2 X 10 ⁵

Table 3: Percentage occurrence of bacteria isolates.

Name of organism	% occurrence of Bacteria
<i>Klebsiella spp</i>	9.09
<i>E. coli</i>	27.27
<i>Proteus spp</i>	9.09
<i>Citrobacter jejuni.</i>	9.09
<i>Staphylococcus. Aureus</i>	27.27
<i>Streptococcus pyogene</i>	18.18
Total	100

DISCUSSION

The result of the study shows the presence of bacteria on jollof rice in the samples studied. However, the significant difference observed in the level of microbial contamination in jollof rice could be associated with inadequate handling and processing by vendors, contamination caused by storage facilities, either poor hygiene or poor quality of grains and water used.

Similarly, the extensive mixing during processing could have introduced contaminants via food handlers, cooking utensils and from the environment.

The isolation of *Klebsiella spp.*, *Staphylococcus aureus*, *E. coli*, *Proteus spp.*, *Citrobacter jejuni* (table 1) correlate with the findings of Oranusi *et al.*, 2013, Tauro *et al.*, 2008 in which these organisms were implicated in ready to eat food. According to Bibeki (2001), contamination

of food items by specific species of micro-organisms is largely due to the presence of these organisms and their entrance in to food as a result of poor hygiene and sanitation.

E. coli and *Staphylococcus aureus* has the highest percentage occurrence of 27.27% while *Klebsiella spp.*, *Proteus spp.*, *Citrobater jejuni* has the least percentage occurrence of 9.09% in Table 3.

It is necessary that food must be free from contamination as much as possible. The presence of *Staphylococcus aureus* is largely as a result of human contact and this suggested poor hygiene practices of the food handlers since the organism is a normal flora of the skin and nasal passage (Gorret, 1988; Nichol *et al.*, 1999). *E. coli* is especially of fecal origin and has been implicated in numerous food-borne diseases (Eni, *et al.*, 2010; Oranusi *et al.*, 2007). However its presence is an indication of possible fecal contamination of food, water or food workers and poor hygiene processing practice (Little *et al.*, 1998; Tambeker *et al.*, 2007).

CONCLUSION AND RECOMMENDATIONS

Conclusion

The prevalence of *E.coli* and *Staphylococcus aureus* demonstrates a potential health risk as these organisms are pathogenic and have been implicated in food borne disease. Standard eateries in Ozoro are beginning to enjoy high patronage but the microbial quality of jollof rice they offer is rather questionable and needs to be improved. This indicates inadequate processing and poor handling practices which can pose health risk to the consumers. It is mandatory that food most free from contaminations as much as possible.

Recommendations

It is hereby recommended that good manufacturing practices (GMP) and hazard analysis critical control point (HACCP) application in the chain of food production and processing as well as health education to improve the knowledge of food vendors on food safety and hygiene practices should be encouraged.

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