Prevalence of *Staphylococcus aureus* in some street vended ready-to-eat meat products in Birnin Kebbi metropolis: A potential food safety threat

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**ABSTRACT**

This study was designed to determine the prevalence of *Staphylococcus aureus* in Street vended Balangu, Kilishi and Tsire meat products in Birnin kebbi, Kebbi State, Nigeria. Sixty six (66) samples (Tsire-21, Kilishi-21 and Balangu-24) were purchased from street vendors and transferred for microbiological analyses. Microbiological analyses were conducted according to standard culture procedure involving isolation in general and selective media for isolation and identification of isolates, followed by series of biochemical tests for confirmation of isolates. Data was analyzed and expressed as colony forming units per gram of sample (cfu/g). Results revealed bacterial load and presence of *Staphylococcus aureus* isolated from the RTE-MPs. The total bacterial counts for *tsire*, *balangu* and *kilishi* were found to be 1.34, 1.71 and 2.01×10\textsuperscript{6} cfu/g, respectively. Also, the *Staphylococcus aureus* population isolated from *tsire*, *balangu* and *kilishi* were recorded as 0.14, 0.29 and 0.33×10\textsuperscript{6} cfu/g, respectively. In terms of public health significance, the results of the current study placed the studied RTE-MPs consumed in the study area within unsatisfactory limits since the values are more than log 9×10\textsuperscript{6} recommended to be acceptable in related RTE-MPs. However, the value for *Staphylococcus aureus* was satisfactory since it was found to be within the limits of <20×10\textsuperscript{6} cfu/g recommended be the satisfactory limits for consumption. It was concluded that the high bacterial count observed and the presence of *Staphylococcus aureus* in the RTE-MPs are of public health significance since they could pose health risks. Good manufacturing practices in the production and consumption of RTE-MPs were recommended to improve their safety and quality.

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1. INTRODUCTION

Food safety is a significant and growing public health problem in Nigeria and the world at large, since food-borne diseases are important contributors to the huge burden of sickness and death of humans. The Nigerian Federal Ministry of Health reported 90,000 cases of food poisoning in 2007 [1]. However, that was a gross underestimate since most cases of gastroenteritis are not reported to hospitals or concerned bodies, rather, patients resort to the use of over-the-counter anti-diarrheal drugs as previously reported [2-3]. The World Health Organization (WHO) estimated 200,000 deaths from diarrhea each year in Nigeria (WHO, 2008), as many as 70% of which may be attributable to contaminated food and water.

Presence of pathogens in food products imposes potential hazard for consumers and causes grave losses in both economic and human productivity through food-borne diseases. Symptoms include nausea, vomiting, and abdominal cramps with or without diarrhea. Preventive measures include safe food handling and processing practice, maintaining cold chain, adequate cleaning and disinfection of equipment, prevention of cross-contamination in...
Meat products (RTE-MPs) including intermediate moisture meats like balangu (berbacue), tsire (skewered meat), guru (roasted chicken) and dried meat products like kilishi (jerky), dambun nama (meat floss), banda (dried meat), were reported to be highly appreciated because of their characteristic taste, texture and storage stability [5].

However, according to Aworh [6], a major source of concern, from a public health standpoint as revealed by consumer surveys, is the unhygienic conditions under which meat products are often processed and retailed in Nigeria and other West African countries. FSAI [7] reported that during processing, handling, packaging and storage, these products are liable to contamination by pathogenic biological agents that could result in food poisoning.

Staphylococcus aureus (S. aureus) is a bacterium that causes staphylococcal food poisoning, a form of gastroenteritis with rapid onset of symptoms. S. aureus is commonly found in the environment (soil, water and air) and is also found in the nose and on the skin of humans. Staphylococcal food-borne disease (SFD) is one of the most common food-borne diseases worldwide resulting from the contamination of food by preformed S. aureus enterotoxins [8].

Elsewhere, several studies have documented prevalence of S. aureus in many food products including meat and its products, indicating that consumers are at potential risk of S. aureus colonization and subsequent infection [9]. There are little or no documented records of such studies in Kebbi state. Hence, the aim of this work is to determine the Prevalence of Staphylococcus aureus in some Street Vended Balangu, Kilishi and Tsire meat products in Birnin kebbi, Kebbi State, Nigeria.

2. MATERIALS AND METHODS

2.1 Study area

The study was conducted in Kebbi State, North-West region of Nigeria. The study area is located between Latitudes 11°30’ and 12°16’N, and Longitudes 4°00’ and 6°25’E. Kebbi State lies in the Savanna regions of Nigeria with its characteristic grasslands and isolated hills. The study Area has an annual average temperature of 28.3°C and the maximum daytime temperature could reach a little above 40°C [10]. The area is known for its abundance of livestock especially the popular Sokoto Gudali, Uda and Yankasa sheep, Sokoto red goat, and many poultry species like domestic fowls, guinea fowls and turkeys, and many other breeds of livestock [11]. According to [18], among all the livestock that makes up the farm animals in Nigeria, ruminants, comprising sheep, goats and cattle, constitute the farm animals largely reared by farm families in the country’s agricultural system.

The State has a variety of animal products that are obtained from the vast kinds of animals available in the region of which meat products predominates [12,13]. This could be evident from the many RTE meat joints sighted along major roads in both urban and rural areas of the region [14].

2.1.1 Sampling of ready-to-eat meat products and sample size

A total of sixty six (66) samples of RTE-MPs, comprising of balangu(24), Kilishi(21) and Tsire(21) were purchased from 14 different locations in Birnin Kebbi Metropolis. The samples were obtained from both on the spot processors and street vendors. The samples were carefully collected and packed in polythene bags and stored in ice packed coolers, then transferred to the laboratory for microbiological analyses.

<table>
<thead>
<tr>
<th>Sampling location</th>
<th>Samples by Rte-mp type</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Bal</td>
<td>Killi</td>
</tr>
<tr>
<td>A</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>B</td>
<td>4</td>
<td>3</td>
</tr>
<tr>
<td>C</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>D</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>E</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>F</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>G</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>H</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>I</td>
<td>3</td>
<td>1</td>
</tr>
<tr>
<td>Total</td>
<td>24</td>
<td>21</td>
</tr>
</tbody>
</table>
2.2 Microbiological Procedures

Microbiological analyses were conducted according to the procedures described by [15]. The microbiological analyses involved isolation in general (non-selective) and selective media for isolation and identification of isolates, followed by series of biochemical tests for confirmation of isolates.

2.2.1 Non-selective pre-enrichment using general medium for Total Bacterial Count

Twenty five grams of each RTE-MP sample was picked, minced and homogenized. Then one gram (1g) of the homogenized sample was weighed out and homogenized in 9mls buffered peptone water (LabM, UK) and hand shaken for two minutes to give a dilution of 1:10. A six-fold serial dilution was then prepared by pipetting 1 ml of the homogenate into the first test tube containing 9 ml of buffered peptone water to make the first dilution. From the first dilution, 1ml was taken and pipetted into the second test tube containing 9ml of diluent. The process was repeated until the sixth dilution was made and 1ml from the last dilution was removed and discarded.

The pour plating method was used for colony counting. Petridishes were prepared and labelled according to samples and dilutions selected. For aerobic plate count determination, about 10-12 ml of the molten Nutrient agar (Biomark, India) (cooled to 42-45°C) was poured into each petridish within 15 minutes from the time of preparation of the original dilution. 1 ml each from dilutions 10\(^{-2}\), 10\(^{-3}\) and 10\(^{-6}\) for every sample was respectively poured on a prelabelled petri plates. The media and dilutions were mixed gently swirling clockwise and anticlockwise, to and fro taking care that the mixture did not touch the lid and was allowed to set. The plates were incubated inverted for 48 hrs at 35°C. Distinct colonies on on plates were counted using a digital colony counting chamber (Quebec colony counter, Reichert, USA) and recorded per dilution counted. The actual number of colonies on dishes containing 30-300 colonies was counted and recorded.

2.2.2 Selective enrichment using specific media for pathogen isolation

2.2.2.1 Identification and confirmation of Staphylococcus aureus

The pre-enrichment was carried out by 25g of the RTE-MP into a sterile jar and 225ml of buffered peptone water was added. The jar was capped and shaken thoroughly for two minutes. 1ml of the homogenate was pipetted in 9ml of peptone water and mixed well using a vortex mixer. 1ml of the homogenate was transferred into triplicate plates of Mannitol salt agar (MSA). The plates were incubated in an upright position in the 35-37°C incubator for about 1hrs or until inoculum is absorbed by the medium, then the plates were inverted and incubated for 45-48 hrs. Catalase and coagulate tests were used to confirm S. aureus.

2.3 Biochemical Tests

Identification of isolates was conducted based on established conventional cultural, morphological and biochemical characterizations. The biochemical tests conducted were Gram staining, Mannitol salt ager, catalase and coagulate tests using methods as previously described [16].

2.4 Data analysis

2.4.1 Determination of total bacterial count and prevalence of pathogens in RTE-MPs

The colony forming unit per gram of the samples was calculated and results expressed as colony forming unit per gram of meat sample (cfu/g) using the following expression by FSSAI [15]:

\[
\text{CFU} = \frac{\Sigma C}{N1 \times D} \times 10^{N2}
\]

Where:
- CFU = colony forming units;
- \(\Sigma C\) = sum of all the colonies counted on all dishes retained;
- \(N1\) = number of dishes retained in first dilution;
- \(N2\) = number of dishes retained in the second dilution;
- 0.1 = constant;
- \(D\) = dilution factor corresponding to the first dilution.

3. RESULTS AND DISCUSSION

Table 2 shows the bacterial load and presence of Staphylococcus aureus isolated from some RTE-MPs in Birnin Kebbi, Kebbi State.

<table>
<thead>
<tr>
<th>Rte-MP</th>
<th>No. of samples</th>
<th>Mean TBC</th>
<th>S. aureus pop.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Balangu</td>
<td>24</td>
<td>16.85(\times 10^{4}) cfu/g</td>
<td>0.29(\times 10^{4}) cfu/g</td>
</tr>
<tr>
<td>Kilishi</td>
<td>21</td>
<td>19.82(\times 10^{5}) cfu/g</td>
<td>0.33(\times 10^{4}) cfu/g</td>
</tr>
<tr>
<td>Tsire</td>
<td>21</td>
<td>13.36(\times 10^{6}) cfu/g</td>
<td>0.14(\times 10^{4}) cfu/g</td>
</tr>
<tr>
<td>Total</td>
<td>66</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 2. Bacterial load and presence of Staphylococcus aureus isolated from some RTE-MPs in Birnin Kebbi, Kebbi State.
The variation in colony counts could be as a result of variations in processing and handling methods where tsire, and balangu at post-lethality, are in most cases kept warm around embers which could assist in killing the bacterial population unlike kilishi which are usually displayed in open trays and table tops, making the product more vulnerable to bacterial contamination. The lower counts in tsire could also be due to the presence of spices added during processing which were reported to have a significant anti-microbial and anti-oxidant effects [17,18]. In most cases, balangu is processed without addition of spices but could be added while serving. Previous research works reported comparatively similar results for the products. [19] recorded similar total plate counts in suya samples in the order of ×10^5 and ×10^6 cfu/g. [13] isolated a range of 0.07 – 2.22×10^5 cfu/g from suya collected from different locations.

In terms of public health significance, the results of the current study placed the studied RTE-MPs consumed in the study area within unsatisfactory limits according to [20,23] standards since the values are more than log 9×10^6 recommended to be acceptable in related RTE-MPs. However, the value for Staphylococcus aureus was satisfactory since it was found to be within the limits of <20×10^6 cfu/g recommended by [21] to be the satisfactory limits. In Nigeria, similar results were reported for various RTE-MPs such as [13] who recorded S. aureus range of 0.53 – 1.67×10^7 in balangu. [22] reported 3.1×10^5 of S. aureus isolated from chicken RTE-MP. Higher isolates were recorded by [23] from suya with S. aureus ranging between log 0.0 and 6.25×10^5 cfu/g. The current study revealed that Kilishi had higher TBC, and individual bacterial counts than kilishi and tsire. Incidentally, balangu was the commonly consumed among the identified product in the study area. Figure 1 shows the prevalence of Staphylococcus aureus among tsire, balangu and kilishi.

![Figure 1](image1.png)

Figure 1. Prevalence of Staphylococcus aureus among tsire, balangu and kilishi.

4. CONCLUSION

Staphylococcus aureus was found to be prevalent in the three RTE-MPs studied in the study area, hence, consumers are exposed to risks of infections. It was concluded that the high bacterial count observed and the presence of Staphylococcus aureus in the RTE-MPs are of public health significance since they could pose health risks. Good manufacturing practices in the production and consumption of RTE-MPs were recommended to improve their safety and quality.

AUTHORS’ CONTRIBUTIONS

MIR designed the study, conducted the laboratory experiments, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript. SSM assisted in the literature searches and organization of the manuscript. All authors read and approved the final manuscript.

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