Bacteriological Quality Assessment of Sliced Pineapple (Ananas comosus) in Birnin Kebbi and Jega Metropolis, Kebbi State, Nigeria

By


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ABSTRACT
Bacteriological quality assessment of ready-to-eat sliced pineapple was demonstrated with the aim to determine the possible bacteria associated the sliced pineapple locally sold in Birnin Kebbi and Jega metropolis. A total of forty (40) sliced pineapple fruit samples over a period of two months (May and June) were purchased from distinct vending points of Birnin Kebbi (Sample A1 to A20) and Jega (Sample B1 to B20). The total standard plate count of the samples ranges from $2.0 \times 10^3$ to $4.8 \times 10^4$ cfu/g, in which on morphological and biochemical characterization revealed the presence of Escherichia coli, Salmonella typhi, Staphylococcus aureus, Staphylococcus epidermis, Bacillus subtilis and Pseudomonas aeruginosa, with highest frequency of 23 and percentage occurrence of 57.5% for Staphylococcus aureus while a frequency of 8 and percentage of 20% was found for Pseudomonas aeruginosa. Based on the findings, the pineapple fruits were not fit for human consumption, hence the need for proper, adequate and effective law on environmental sanitation governing the selling of sliced and ready-to-eat food hawking at different points of the metropolis and the state at large.

Keywords: Bacteriological, quality, ready-to-eat, pineapple and fruit.

INTRODUCTION
Fruits and vegetables remain an important requirement for sustenance of life. The demand therefore cannot be over emphasized (Muller, et al., 2012). Fresh fruit and vegetables are important components of a healthy and balanced diet; their consumption is encouraged in many countries by government health agencies to protect against a range of illnesses such as cancers and cardiovascular diseases. However, fruits and vegetables, and in particular leafy greens that are consumed raw, are increasingly being recognized as important vehicles for transmission of human pathogens that were traditionally associated with foods of animal origin (Berger et al., 2010). Despite the increased importance of fresh produce as a vehicle for...
human pathogens, there is currently limited knowledge about where in the supply chain contamination occurs or about the mechanism by which human pathogens colonize and survive on or in fruits and vegetables (Gal-Mor et al., 2014).

Pineapple is a leading edible member of the family bromeliaceae which embraces about 2,000 species, mostly epiphytic and many strikingly ornamental (Jothi et al., 2014). The plant has a peduncle (stem) on which the fruit develops, it also resembles a pinecone, hence its name. Pineapple is native to South America, but its exact origin is unknown. The general composition of an average pineapple consists of the following: water content (85%), carbohydrate (13%), protein (0.3%), fat (0.1%), ascorbic acid (2.6mg). This nutritional content appears to be capable of supporting the growth of bacteria (Feng et al., 2017).

Pineapple is the third most important tropical fruit in world production after Banana and citrus (Baruwa, 2013), contributing to over 20% of the world production of tropical fruits. In countries producing the pineapple, nearly 70% is consumed as fresh fruit. In Nigeria, pineapple production is the main source of income for many farmers. Until recently, about 80% of the fruits came from small farms man-aged under mixed cropping systems (Adegbite et al., 2014). Seventy percent (70%) of the pineapple produced in the world is consumed as a fresh fruit in the country of origin. This worldwide production has developed since early 1900s when pineapple was first taken to Europe and then distributed throughout the world’s tropics (Bartholomew et al., 2003).

Large investigations on prevalence of pathogenic bacteria in fruits and vegetables were conducted in the UK, Ireland, Germany and the Netherlands in 2007. In developing countries such as Nigeria, continues use of untreated wastewater and manure as fertilizers for the production of fruits and vegetables is a major contributing factor to contamination (Tsado et al., 2013). Thus, despite their nutritional and health benefits, outbreaks of human infections associated with the consumption of fresh or minimally processed fruits and vegetables have increased in recent years (Nestle, 2013).

In Nigeria for instance, street vending of handy ready-to-eat sliced fruit and vegetables has recently become very common and the market is thriving (Obi, 2014). As a result, several cases of typhoid fever outbreak have been associated with eating contaminated fruits and vegetables grown in or fertilized with contaminated soil or sewage (Berger et al., 2010). This increases in fruits and vegetables-borne infections which may have resulted from increased consumption of contaminated fruits and vegetables outside the home as most people spend long hours outside the home.

MATERIALS AND METHODS

Description of the Study Area

Birnin Kebbi Town is the State capital of Kebbi State and also the administrative headquarter of Gwandu Emirate and Birnin Kebbi Local Government Area. Kebbi State was created on 27th August in 1991 from the old Sokoto State, the state is located in the North Western part of Nigeria between the latitude 11.67810N and longitude 4.06950 E, with annual rainfall of about 800mn, and average temperature of about 26°C, ranging from 21°C in winter to 40°C between April and June, with a total population of 3,802,500 people according to
National Population Census (NPC) estimate (Traditional State of Nigeria, 2010). Jega town is the head quarter of Jega local government of Kebbi state. The local government is located in the Sudan and Guinea savanna zone of the central part of the state. The area is relatively bounded by Birnin Kebbi local government to the north, Kalgo local government to the west, Aliero local government to the east and Maiyama local government to the south. Geographically, the area is located within the latitude of 12° 11′ 24″ N and longitudes 4° 23′ 50″ E with inhabitants that are predominantly Hausa, Gimbanawa and Fulani. The major occupations of the inhabitants are farming (Crop production and Animals rearing) and trading with very small percentage as civil servants (Yandaki and Sulaiman, 2008).

Sample Collection
A total of forty (40) sliced pineapple fruit samples were sporadically collected over a period of two (2) months from 40 different vending points of Birnin Kebbi (A1 to A40) and Jega (B1 to B40) metropolis. Each study area was visited twice, which yielded a total of ten samples per each visit. All the samples were wrapped using sterile polythene bags and transported immediately to the laboratory for analyses.

BACTERIOLOGICAL ANALYSES

Determination of Bacterial Load
For enumeration of bacteria present in each sample, 10-fold serial dilutions of each pineapple fruit sample were made and 0.1 ml of 10⁰, 10¹, 10² diluents were pipetted into sterile Petri-dishes and molten nutrient agar (45°C) was added and swirled thoroughly to allow even distribution. After an incubation of 24 hours at 37°C, the colonies were counted using a colony counter (Stuart Scientific, UK) and recorded as cfu/g.

Isolation and Identification of Bacteria
The isolates were characterized and identified based on colonial morphology, cultural characteristics and biochemical tests as described by (Cheesbrough, 1989; Manga and Oyeleke, 2008).

RESULTS
The standard heterotrophic plate count (Table 1 and 2) of the sliced pineapple fruit samples revealed the highest count of 4.8 x 10⁴ cfu/g for sample B20 and the lowest of 2.0 x 10³ cfu/g for sample A3 and B16 respectively. Based on morphological and biochemical characteristics, a total of six (6) bacteria were identified (Table 3) as follows: *Escherichia coli*, *Staphylococcus epidermis*, *Staphylococcus aureus*, *Salmonella typhi*, *Bacillus subtilis* and *Pseudomonas aeruginosa*. Figure 1 was a result showing *Staphylococcus aureus* with the highest occurrence of 23 and frequency of 57.5%, while *Pseudomonas aeruginosa* has the lowest occurrence of 8 and frequency of 20%.
Table 1: Total Standard Bacterial Plate Count of Sliced Pineapple Fruit Samples from Birnin Kebbi Metropolis (A1 up to A20)

<table>
<thead>
<tr>
<th>Sample codes</th>
<th>Bacterial load (cfu/gram)</th>
<th>Sample codes</th>
<th>Bacterial load (cfu/gram)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1</td>
<td>$3.2 \times 10^4$</td>
<td>A11</td>
<td>$2.3 \times 10^4$</td>
</tr>
<tr>
<td>A2</td>
<td>$4.3 \times 10^4$</td>
<td>A12</td>
<td>$4.7 \times 10^4$</td>
</tr>
<tr>
<td>A3</td>
<td>$2.0 \times 10^3$</td>
<td>A13</td>
<td>$2.2 \times 10^4$</td>
</tr>
<tr>
<td>A4</td>
<td>$3.1 \times 10^4$</td>
<td>A14</td>
<td>$3.4 \times 10^4$</td>
</tr>
<tr>
<td>A5</td>
<td>$2.9 \times 10^4$</td>
<td>A15</td>
<td>$4.1 \times 10^4$</td>
</tr>
<tr>
<td>A6</td>
<td>$3.9 \times 10^4$</td>
<td>A16</td>
<td>$3.6 \times 10^4$</td>
</tr>
<tr>
<td>A7</td>
<td>$3.6 \times 10^4$</td>
<td>A17</td>
<td>$2.6 \times 10^4$</td>
</tr>
<tr>
<td>A8</td>
<td>$2.8 \times 10^4$</td>
<td>A18</td>
<td>$4.5 \times 10^4$</td>
</tr>
<tr>
<td>A9</td>
<td>$3.7 \times 10^4$</td>
<td>A19</td>
<td>$3.3 \times 10^4$</td>
</tr>
<tr>
<td>A10</td>
<td>$3.4 \times 10^4$</td>
<td>A20</td>
<td>$2.8 \times 10^4$</td>
</tr>
</tbody>
</table>

Table 2: Total Standard Bacterial Plate Count of Sliced Pineapple Fruit Samples from Birnin Jega Metropolis (B1 up to B20)

<table>
<thead>
<tr>
<th>Sample codes</th>
<th>Bacterial load (cfu/gram)</th>
<th>Sample codes</th>
<th>Bacterial load (cfu/gram)</th>
</tr>
</thead>
<tbody>
<tr>
<td>B1</td>
<td>$4.4 \times 10^4$</td>
<td>B11</td>
<td>$3.7 \times 10^4$</td>
</tr>
<tr>
<td>B2</td>
<td>$3.8 \times 10^4$</td>
<td>B12</td>
<td>$3.1 \times 10^4$</td>
</tr>
<tr>
<td>B3</td>
<td>$3.9 \times 10^4$</td>
<td>B13</td>
<td>$4.0 \times 10^3$</td>
</tr>
<tr>
<td>B4</td>
<td>$4.2 \times 10^4$</td>
<td>B14</td>
<td>$4.4 \times 10^4$</td>
</tr>
<tr>
<td>B5</td>
<td>$3.8 \times 10^4$</td>
<td>B15</td>
<td>$4.1 \times 10^4$</td>
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<tr>
<td>B6</td>
<td>$4.4 \times 10^4$</td>
<td>B16</td>
<td>$2.0 \times 10^3$</td>
</tr>
<tr>
<td>B7</td>
<td>$3.1 \times 10^4$</td>
<td>B17</td>
<td>$3.9 \times 10^4$</td>
</tr>
<tr>
<td>B8</td>
<td>$3.2 \times 10^4$</td>
<td>B18</td>
<td>$4.6 \times 10^4$</td>
</tr>
<tr>
<td>B9</td>
<td>$2.1 \times 10^4$</td>
<td>B19</td>
<td>$3.3 \times 10^4$</td>
</tr>
<tr>
<td>B10</td>
<td>$2.4 \times 10^4$</td>
<td>B20</td>
<td>$4.8 \times 10^4$</td>
</tr>
</tbody>
</table>

Table 3: Biochemical Characterization of the Isolates from Sliced Pineapple Fruit Samples

<table>
<thead>
<tr>
<th>Isolates</th>
<th>Gr</th>
<th>Indole</th>
<th>Mr</th>
<th>Vp</th>
<th>Citrate</th>
<th>Identified organisms</th>
<th>Percentage (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td><em>Escherichia coli</em></td>
<td>52.5%</td>
</tr>
<tr>
<td>2</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td><em>Staphylococcus epidermidis</em></td>
<td>35%</td>
</tr>
<tr>
<td>3</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td><em>Staphylococcus aureus</em></td>
<td>57.5%</td>
</tr>
<tr>
<td>4</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td><em>Salmonella typhi</em></td>
<td>42.5%</td>
</tr>
<tr>
<td>5</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td><em>Bacillus subtilis</em></td>
<td>30%</td>
</tr>
<tr>
<td>6</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td><em>Pseudomonas aeruginosa</em></td>
<td>20%</td>
</tr>
</tbody>
</table>

Keys: Gr= Gram reaction, Mr= Methyl red, Vp= Vogues Proskauer.
DISCUSSION

The bacterial loads/numbers counted which ranged from $20 \times 10^3$ to $47 \times 10^4$ cfu/g, may be due an exposure by the vendors of the pineapple fruits to contaminants, this conformed with the report of Bukar et al., 2010; Oranusi and Olorunfemi 2011, that the consumption of sliced/peeled ready to eat fruits directly from street vendors or hawkers potentially increase the risk of food-borne diseases caused by a wide variety of pathogens, because it is difficult to attest to the hygiene of these vendors or to the sanitary conditions at points of processing as well as the packaging materials.

The kinds of organisms demonstrated (Escherichia coli, Staphylococcus epidermidis, Staphylococcus aureus, Salmonella typhi, Bacillus subtilis and Pseudomonas aeruginosa) in this study, was in line with what have previously been isolated from fruits and vegetables in other studies, both in Nigeria and elsewhere (Olayemi, 1997; Omemu et al., 2005; Tambekar and Mundhada, 2006; Uzeh et al., 2009). However, some of these organisms are enteric (Escherichia coli, salmonella typhi and Pseudomonas aeruginosa), and may likely cause gastroenteritis, diarrhea, cholera, typhoid fever etc.

The highest frequency for Staphylococcus aureus may be due to its been part of the microbial flora of the skin, that can be found at any fruits sold at street, which must always be assessed by the consumers before deciding to purchase.

CONCLUSION

The bacterial loads cfu/gram and their genera obtained in this study revealed that the sliced pineapple fruits locally sold within the study areas has potentials of causing public health problems which may eventually cause diseases like cholera, bacteremia, gastroenteritis, typhoid fever, etc., as such, it is concluded that the sliced pineapple fruits vending at different points of the study areas are unhygienic and unfit for human consumption.
RECOMMENDATIONS
1. To prevent the introduction and invasion of pathogenic bacteria to pineapple fruits, the origin of the water uses for irrigation should be adequately sanitized and maintained.
2. All irrigation spaces should be monitored routinely for possible contaminants.
3. Local manure used as fertilizer should be treated either by composting or aging to eliminate pathogenic microorganisms and farmers should be educated on the need to allow sufficient amount of time between the final manure application and harvest.
4. Processors/vendors should also practice hygienic measures to ensure that they do not serve as causative agents of microbial contamination.
5. Sterile utensils and surfaces should be used in slicing the pineapple fruit to forestall the possible contaminants from invading the fruit.
6. There should be law compelling vendors in Nigeria, to transport/sell sliced pineapple fruits in cool temperature and controlled carts similar to those used for the transportation/sales of yogurts and other soft drinks to avoid any foreign substances from attacking the fruit.

REFERENCES
Bartholomew, D.P., Paul, R. E. and Rorbach, K.G. (2003). The pineapple Botany, Production and Uses, University of Hawaii Manoa Honolulu, USA:
http://bookshop.cabi.org/Uploads/Books/PDF/978085995038/].


