Prevalence and antibiotic susceptibility profile of *Staphylococcus aureus* from milk and milk products in Nasarawa State, Nigeria

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**Abstract**

This study determined the prevalence and antibiotic susceptibility profile of *Staphylococcus aureus* isolated from milk and milk products in Nasarawa State, Nigeria. A total of 180 samples comprising of fresh raw milk, bulk milk, Nono, and Kindirmo were collected over a period of 6 months. Standard microbiological procedures were employed in the isolation, identification, characterization and determination of the antibiogram of *S. aureus* from the milk samples. Characterization was achieved by morphological, biochemical characteristics using conventional methods and Microgen® STAPH-ID kits. The isolates were tested for susceptibility or resistance to a panel of 11 commonly used antibiotics using the agar disc diffusion technique. Out of the 180 milk samples examined, nine (9) *S. aureus* were isolated giving a prevalence of 5.0%. The occurrence of *S. aureus* was higher in Nono (12.1%) and Kindirmo (10.6%) than in fresh raw milk (5.9%). The kind of water (well water) used for cleaning utensils at the Nono and Kindirmo selling points was found to be a risk factor associated with the occurrence of *S. aureus* in the products. All of the isolates were resistant to cefoxitin (100%), ampicillin (100%), and amoxicillin/clavulanic acid (100%). The isolates displayed various rates of resistance to erythromycin (22.2%), sulphamethoxazole/trimethoprim (22.2%), and tetracycline (44.4%). Five (5) antibiotic resistance patterns were recorded among the isolates an indication of different levels of use and misuse of antibiotics in the areas studied. The detection of *Staphylococcus aureus* in fresh and fermented milk in the areas studied suggests that consumption of dairy products especially those produced using traditional methods, constitute a hazard to consumers. It is recommended that since compliance with basic hygiene requirements is not guaranteed, hazard analysis and critical control points (HACCP) concepts should be seen as a part of an effective total hygiene concept at the selling points.

**Keywords:** Antibiotic susceptibility profile, Milk, Nasarawa State, Nigeria, Prevalence, Staphylococcus aureus
Introduction

Raw milk and milk products of various types are consumed as supplement to normal meals in homes and even for commercial purposes (Maduka et al., 2013). Traditionally produced milk products especially those produced from raw milk under neglected hygienic conditions, are potential vehicles for the transmission of different foodborne pathogens especially toxigenic *Staphylococcus aureus* (Kadariya et al., 2014). *Staphylococcus aureus*, including those associated with animals have been frequently recovered from raw milk and milk products worldwide (Peton & Le Loir, 2014). Despite the fundamental roles that milk and milk products play in human nutrition, they serve as vehicles for the transmission of many bacterial pathogens to man. For example, in Europe, milk and other dairy products are found to be responsible for 5% of staphylococcal outbreaks (Bianchi et al., 2014). Health risk to consumers can be associated with milk due to the presence of zoonotic pathogens and antimicrobial drugs residues (Vyletĕlova et al., 2011). Milk quality can be lowered by a number of factors such as contamination during and after milking, and the presence of udder infections (Adams & Moss, 2011). Pathogenic microorganisms in milk can be derived from the cow itself, the human hand during handling, or the environment (Eson et al., 2005). Antibiotic use is commonplace in human medicine and animal production. The extensive use of antibiotics in both human medicine and agriculture particularly in disease prevention and growth enhancement in animal production is a considerable cause of the selection pressure and prevalence of antibiotic resistant bacteria (Jamali et al., 2015). The use of antimicrobial agents is associated with the risk of inducing resistance to antimicrobial agents in bacterial pathogens and transmission of resistance bacteria to humans via the food chain (Jamali et al., 2015).

Food contamination with antibiotic resistant pathogens poses a major public health threat as the antibiotic resistance determinants can be transferred to other bacteria of human clinical significance (Jamali et al., 2014). Contamination of food products with microorganisms influence considerably, the safety of the products, endanger the health of consumers, lower their shelf life resulting in foodborne infections, intoxications, and economic losses due to food spoilage (Shiferaw & Ahmad, 2016). Milk is considered as a good substrate on which *Staphylococcus aureus* grow and produce enterotoxins (Korpysa-Dzirba & Osek, 2011).

*Staphylococcus aureus* has been reported as one of the most common causative agents of food poisoning associated with the consumption of raw milk and milk products (Spanu et al., 2012). Contamination of food stuff occurs directly from infected food-producing animals or may result from poor hygiene during production processes. Contamination could also occur during the storage and retail of foods since humans also harbours microorganisms (Vázquez-Sánchez et al., 2012).

The hygienic standard of milk may be assessed based on the level of contamination with *S. aureus* and studying the antibiotic susceptibility profile and risk factors associated with the occurrence of this bacterial pathogen in foods like milk and milk products will provide valuable information on planning effective control and preventive measures of the pathogen. Considering the aforementioned points, this study was conducted with the aim of isolating, characterizing and determining the antibiotic susceptibility profile of *Staphylococcus aureus* from milk and milk products in parts of Nasarawa State, Nigeria.

Materials and Methods

Study area

This study was carried out in Nasarawa State, Nigeria. Two (2) Local Government Areas were selected in each of the three Senatorial Zones in the State viz: Akwanga and Wamba (Nasarawa North); Lafia and Doma (Nasarawa South); Keffi and Nasarawa (Nasarawa West) (Figure 1). The State has thirteen (13) Local Government Areas and is bounded to the north by Kaduna State, Plateau State to the northeast, Taraba State to the southeast, Benue State to the south, Kogi State to the southwest, and the Federal Capital Territory (FCT), Abuja, to the west (Figure 2). These positions were taken using Taiwan-made Etrex® high sensitive Geographic Positioning System (GPS) receiver. Nasarawa State is situated between latitude 70° 40 and 90° 40, and between longitude 70° 0 and 90° 30.

Study design

Sample size

The sample size was determined using the 12.6% prevalence of *S. aureus* as reported by Umaru et al. (2013). The sample size was determined by using the equation described by Naing et al. (2006):

\[ n = \frac{Z^2 \cdot P \cdot (1-P)}{d^2} \]
Where $n$ is the sample size;
$P$ is the prevalence from a previous study = 12.6% = 0.126;
$Z$ is the standard normal distribution at 95% confidence interval = 1.96;
$d$ is the absolute desired precision at 5% = 0.05.

Therefore,
$$n = \frac{(1.96)^2 \times 0.126 \times (1-0.126)}{(0.05)^2} = 169 \text{ samples}$$

**Sample collection**

For the purpose of this study, the sample size was rounded up to 180. Thirty cow milk and milk products samples were randomly collected from each of the Local Government Areas selected for this study from May to October, 2017.

Proportionate sampling method was used in collecting fresh raw milk samples from lactating cows at the accessible Fulani settlements. This was done by taking 50% of the number of lactating cows in a herd at the settlements. On the whole, 34 fresh raw milk samples were collected. Herds were visited during milking time where composite fresh cow milk samples were collected directly from the milking cows and placed into sterile bottles. Each sample (30ml) was collected into sterile screw-capped plastic bottles and labelled appropriately.

Fourteen bulk milk samples were collected from the accessible Fulani settlements in each town selected for this study. The bulk fresh milk samples were collected after the milk has been collected and pooled. Twenty-two nono and locally-pasteurized milk (*kindirmo*) samples were randomly purchased from vendors in the towns selected for this study.

All samples were placed in separate sterile plastic bags to prevent spillage and cross contamination. Samples were stored in a cool box with ice blocks and then transported to the Biochemical Laboratory of the Institute for Agricultural Research (IAR), Samaru, Zaria and the Postgraduate Students’ Laboratory of the Department of Microbiology, Ahmadu Bello University, Zaria, for proximate and microbiological analyses respectively.

**Isolation and identification of Staphylococcus aureus**

All samples were inoculated with the aid of a sterile wire loop onto the surface of prepared Baird-Parker agar plates (Oxoid, Basingstoke, England) supplemented with 5% egg yolk tellurite emulsion (Baird-Parker, 1962). Representative colonies were selected after incubation at 37°C for 24 h based on the appearance of presence of black colonies on the medium which occur as a result of the ability coagulase-positive staphylococci to reduce tellurite, and clear zones of lypolysis which is due to the lecithinase of staphylococci that break down the egg yolk. Discrete colonies were further sub-cultured on to freshly prepared plates of the selective media and nutrient agar plates for biochemical tests and identification (Patrick *et al.*, 2013). *Staphylococcus aureus* ATCC 25923 strain was used as a positive control.
Observation of colony morphology and characteristics
Presumptive morphological identification of the colonies was done by observing their individual appearance on the selective media that was used for the isolation and Gram reaction.

Biochemical tests
The conventional biochemical tests carried out to identify the suspected *S. aureus* colonies were Catalase test, slide coagulase test, haemolysis on blood agar, and DNase test (Japoni et al., 2004).

Microgen* staphylococci identification (STAPH identification) kits
The Microgen Staph-ID system comprises of a single microwell test strip containing 12 standardized biochemical substrates which have been selected on the basis of extensive computer analysis i.e. each well contains dehydrated substrates, namely: nitrate, sucrose, tetrazolose, mannitol, N-acetyl glucosamine, mannose, turanose, N-acetyl glucosamine, β-glucosidase, β-glucuronidase, urease, arginine, and 1-pyrolidonyl-α-naphthylamide (www.microgenproducts.com UK). A colour change occurs if the individual substrates are metabolised by the organism during incubation, or after addition of specific reagents.

Assessment of the risk factors associated with the occurrence of *Staphylococcus aureus* in Milk and Milk Products
Structured questionnaires were used in investigating some risk factors that are associated with the occurrence of *Staphylococcus aureus* in milk and milk products.

*Staphylococcus aureus* ATCC 25932 obtained from the National Veterinary Research Institute (NVRI), Vom, Plateau State, Nigeria was included in each batch analysis as the quality control standard strain.

Determination of minimum inhibitory concentration (MIC) of vancomycin
The MIC of vancomycin against the isolates was determined by broth microdilution method as recommended by the CLSI (2016). Standard powders of the antibiotic (vancomycin) were obtained from GlaxoSmithKline Pharmaceutical Companies. The MIC of vancomycin against the isolates was determined using the procedure described by Wayne (2002). Readings were taken and recorded according to the guidelines of CLSI (2016).

Statistical analysis
The Chi-square test was used to determine the significant differences between the occurrence of *Staphylococcus aureus* in the different sample types analyzed, and the occurrence of *Staphylococcus aureus* in the different sampling areas, and the association between some risk factors and the occurrence of the bacterium in milk and milk products. P-values of 0.05 were considered significant for all comparisons.

Results
Table 1 show the distribution of *Staphylococcus aureus* in fresh and fermented milk samples collected from the study areas. Although there were variations in the occurrence of the bacterium in the different sample types, no significant association ($X^2 = 0.646$) was found between its occurrence and the different samples collected.

Table 2 show the occurrence of *Staphylococcus aureus* in fresh and fermented milk samples collected from the different Local Government Areas studied. No significant association ($X^2 = 1.233$) was found between the occurrence of the bacterium and the different Local Government Areas sampled.

The results of the risk factors associated with the occurrence of *Staphylococcus aureus* in fresh and fermented milk are presented in Table 3. A significant association ($X^2 = 62.219$) was found between the occurrence of the bacterium in *nono* samples collected and the kind of water used for cleaning utensils at the product’s selling points. Similarly, a significant association ($X^2 = 63.119$) was found between the occurrence of *Staphylococcus aureus* in *kindirmo* and the kind of water used for cleaning utensils at the product’s selling points. Higher occurrence of the bacterium
was found in nono and kindirmo samples collected from selling points with high street activities (mainly vehicular movement) compared to samples collected from selling points with low street activities. The results antibiotic susceptibility profile of the nine (9) *Staphylococcus aureus* isolated from fresh and fermented milk in parts of Nasarawa State using 11 antibiotics are as presented in Table 4. The results show that, 66.7% of the isolates were susceptible to sulphamethoxazole/trimethoprim, 88.9% of the isolates were susceptible to vancomycin, seven 77.8% were susceptible to chloramphenicol, 44.4% were susceptible to erythromycin, 100% were susceptible to gentamicin and ciprofloxacin, 88.9% were susceptible to imipenem, while 33.3% were susceptible to tetracycline (Table 3). However, 100% of the *S. aureus* isolates were resistant to cefoxitin, ampicillin, and amoxicillin/clavulanic acid, 44.4% were resistant to tetracycline, 22.2% were resistant to both erythromycin and sulphamethoxazole/trimethoprim. All the nine (9) *S. aureus* isolates obtained in this study were resistant to cefoxitin, an indication that they are all methicillin-resistant *Staphylococcus aureus* (MRSA) strains. The antibiotic susceptibility profile of the *S. aureus* isolates showed that, they were highly susceptible to gentamicin (100%), ciprofloxacin (100%), imipenem (88.9%), vancomycin (88.9%), and chloramphenicol (77.8%).

The results of the antibiotic resistance patterns of the nine (9) *Staphylococcus aureus* isolates obtained from the fresh and fermented milk samples examined are as presented in Table 5. Five (5) antibiotic resistance phenotypes were obtained, all from the multiple resistance types with varying combinations of three (3), (4), and five (5) antibiotics. No antibiotics resistance phenotype was found with a single or two antibiotics as all of the isolates were found to be resistant to three (3) antibiotics and above. The highest frequency (3) isolates showing resistance to a combination of antibiotics was a found in a combination of five (5) antibiotics.

**Discussion**

The results obtained from this study revealed that, *Staphylococcus aureus* were present in fresh and fermented milk in parts of Nasarawa State, Nigeria. This is of public health significance since it is a commonly recovered pathogen in outbreaks of food poisoning attributed to dairy products (Junaidu et al., 2011). The occurrence of *Staphylococcus aureus* (5.0%) in the study area is an indication of defective or absence of public health measures and poor sanitary habits among the people that are concerned with milking, milk handling, transportation and sales as these have been documented to be factors that predisposes milk to contamination with pathogens (Akram et al., 2013).

**Table 1:** Occurrence of *Staphylococcus aureus* in Relation to the Type of Milk Samples Collected from Parts of Nasarawa State, Nigeria

| Type of Milk Sample | No. Examined | No. Positive (%) | \( X^2 \) | p-value |
|---------------------|--------------|------------------|-----------|---------|
| Nono                | 66           | 4(6.06)          |           |         |
| Bulk Milk           | 14           | 1(7.14)          |           |         |
| Fresh Milk          | 34           | 1(2.94)          |           |         |
| Kindirmo            | 66           | 3(4.55)          |           |         |
| Total               | 180          | 9(5.0)           |           |         |

**Table 2:** Prevalence of *Staphylococcus aureus* in Fresh and Fermented Milk in relation to the Local Government Areas sampled

| LGAs               | No. examined | No. Positive (%) | \( X^2 \)- value | p-value |
|--------------------|--------------|------------------|------------------|---------|
| Nasarawa           | 35           | 1(2.86)          |                  |         |
| Keffi              | 29           | 2(6.89)          |                  |         |
| Akwanga            | 28           | 2(7.14)          |                  |         |
| Wamba              | 28           | 1(3.57)          | 1.233            | 0.942   |
| Lafia              | 31           | 2(6.45)          |                  |         |
| Doma               | 29           | 1(3.45)          |                  |         |
| Total              | 180          | 9(5.0)           |                  |         |

Key: LGAs – Local Government Areas
Table 3. Risk Factors Associated with the Occurrence of *Staphylococcus aureus* in Fresh and Fermented Milk in Parts of Nasarawa State, Nigeria

| Risk factor                                             | No. Examined (%) | No. Positive (%) | $\chi^2$ | $p$-value | Odds Ratio (95%CI) |
|---------------------------------------------------------|------------------|------------------|---------|-----------|-------------------|
| Washing of teats and udder before milking               |                  |                  |         |           |                   |
| Yes                                                     | 6(17.65)         | 0(0.0)           | 0.221   | 0.638     | 0.964(0.898-1.036) |
| No                                                      | 28(82.35)        | 1(2.94)          |         |           |                   |
| Total                                                   | 34(100.0)        |                  |         |           |                   |
| Milkers washing their hands before milking              |                  |                  |         |           |                   |
| Yes                                                     | 2(5.88)          | 0(0.0)           | 0.064   | 0.800     | 0.969(0.910-1.031) |
| No                                                      | 32(94.12)        | 1(3.13)          |         |           |                   |
| Total                                                   | 34(100.0)        |                  |         |           |                   |
| Cleanliness of milk storage containers                  |                  |                  |         |           |                   |
| Satisfactory                                            | 4(21.43)         | 0(0.0)           | 0.431   | 0.512     | 0.900(0.732-1.107) |
| Unsatisfactory                                          | 10(71.43)        | 1(10.0)          |         |           |                   |
| Total                                                   | 14(100.0)        | 1(7.14)          |         |           |                   |
| Source of water for cleaning utensils at the Nono selling points |                  |                  |         |           |                   |
| Tap                                                     | 0(0.0)           | 0(0.0)           | 62.219  | 0.000     |                   |
| Vendors                                                 | 48(72.73)        | 1(2.08)          |         |           |                   |
| Well                                                    | 18(27.27)        | 3(16.67)         |         |           |                   |
| Total                                                   | 66(100.0)        | 4(6.06)          |         |           |                   |
| Source of water for cleaning utensils at the Kindirmo selling points |                  |                  |         |           |                   |
| Tap                                                     | 0(0.0)           | 0(0.0)           | 63.119  | 0.000     |                   |
| Vendors                                                 | 42(63.64)        | 1(2.38)          |         |           |                   |
| Well                                                    | 24(36.36)        | 2(8.33)          |         |           |                   |
| Total                                                   | 66(100.0)        | 3(4.55)          |         |           |                   |
| Street activities at the Nono selling points            |                  |                  |         |           |                   |
| High                                                    | 64(96.96)        | 4(6.25)          | 0.133   | 0.715     | 0.938(0.880-0.999) |
| Low                                                     | 2(3.03)          | 0(0.0)           |         |           |                   |
| Total                                                   | 66(100.0)        | 4(6.06)          |         |           |                   |
| Street activities at the Kindirmo selling points        |                  |                  |         |           |                   |
| High                                                    | 65(98.48)        | 3(4.62)          | 0.048   | 0.826     | 0.954(0.904-1.006) |
| Low                                                     | 1(1.52)          | 0(0.0)           |         |           |                   |
| Total                                                   | 66(100.0)        | 3(4.55)          |         |           |                   |

The use of traditional method of *nono* and *kindirmo* production also exposes the products to bacteria found on the hands and clothes of the people that are concerned with the production and also in the containers used. The unsanitary conditions of the places where the products are marketed might have also contributed to their contamination.

The percentage occurrence of *Staphylococcus aureus* in fresh and fermented milk in parts of Nasarawa State, recorded in this study was 5.0%, which was lower than the 12.6% and 12.14% recorded by Umaru et al. (2013) and Usman & Mustapha (2016), in studies conducted to determine the occurrence of *Staphylococcus aureus* in fresh and fermented milk in Kaduna and Zaria. It was also lower than the 8.7% prevalence recorded by Okpo et al. (2016) in in parts of Kaduna State, Nigeria. Higher rates of occurrence of *S. aureus* at 32%, 56%, 25.53%, and 55.26% were reported by Patrick et al. (2013), Gundogan & Avci (2014), Jahan et al. (2015), and Chaalal et al. (2016) in Kenya, Turkey, Bangladesh, and Algeria, respectively.
Table 4: Antibiotic susceptibility profile of *Staphylococcus aureus* isolated from fresh and fermented milk in parts of Nasarawa State.

| Antibiotics               | Disc Conc. (µg) | N = 9 |  |  |  |
|---------------------------|-----------------|-------|---|---|---|
|                           |                 | S (%) | I (%) | R (%) |
| Ampicillin                | 10              | 0(0.0) | 0(0.0) | 9(100.0) |
| Amoxicillin/ clavulanic acid | 30              | 0(0.0) | 0(0.0) | 9(100.0) |
| Cefoxitin                 | 30              | 0(0.0) | 0(0.0) | 9(100.0) |
| Gentamicin                | 30              | 9(100.0) | 0(0.0) | 0(0.0) |
| Chloramphenicol           | 30              | 7(77.8) | 2(22.2) | 0(0.0) |
| Vancomycin                | 30              | 8(88.9) | 1(11.1) | 0(0.0) |
| Ciprofloxacin             | 5               | 9(100.0) | 0(0.0) | 0(0.0) |
| Erythromycin              | 15              | 4(44.4) | 3(33.3) | 2(22.2) |
| Imipenem                  | 10              | 8(88.9) | 1(11.1) | 0(0.0) |
| Tetracycline              | 30              | 3(33.3) | 2(22.2) | 4(44.4) |
| Sulphamethoxazole/ Trimethoprim | 25 | 6(66.7) | 1(11.1) | 2(22.2) |

Table 5: Antibiotic resistance pattern of *Staphylococcus aureus* isolated from fresh and fermented milk in parts of Nasarawa State Nigeria

| No. of Antibiotics | Resistance Pattern | No. (%) of Isolates | LGA |
|--------------------|--------------------|---------------------|-----|
| 3                  | Amp, Amo, Fox      | 4(44.4)             | NS, KF, AK, AK |
| 4                  | Amp, Amo, Fox, Tet | 2(22.2)             | LF, WM |
| 5                  | Amp, Amo, Fox, Tet, Sul | 1(11.1) | KF |
| 5                  | Amp, Amo, Fox, Ery, Tet | 1(11.1) | LF |
| 5                  | Amp, Amo, Fox, Ery, Sul | 1(11.1) | DM |

AMP = Ampicillin, Amo = Amoxicillin/clavulanic acid, Fox = cefoxitin, Tet = Tetracycline, Ery= Erythromycin, Sul = Sulphamethoxazole/trimethoprim, NS = Nasarawa, KF = Keffi, AK = Akwanga, WM = Wamba, LF = Lafia, DM = Doma

This portrays *Staphylococcus aureus* as a bacterial pathogen of public health significance as it relates to food safety. The paucity of information on *S. aureus* in milk and foods in general in the study area made it difficult to make any comparison and to assess the level of *S. aureus* in dairy products in the areas studied.

The isolation of *Staphylococcus aureus* from fresh and fermented milk is a cause for public health concern because many people in the area consume the products. The findings of this work lend credence to the assertion that, dairy products are one of the major vehicles for the transmission of *S. aureus* to man (Jahan et al., 2015). No significant difference (p>0.05) was found in the occurrence of *Staphylococcus aureus* in fresh and fermented milk with respect to the different sample types collected in the course of this study, indicating that, the milk samples might have been exposed to the same levels of contamination. This may be due to similar handling procedures employed during milking, milk collection, and processing and production of fermented milk (*nono* and *kindirmo*). This trend of occurrence of *S. aureus* is in contrasted with previous report documented by Umoh (1989) that fermented foods are not good media for the survival and growth of *S. aureus*. The occurrence of the organism in these processed foods implies recontamination during and/or after processing. Proper heat treatment and refrigeration can minimise the chances of contamination with *S. aureus* (Kivaria et al., 2006). It has been observed that, during the heat treatment of milk to make *kindirmo*, the temperature does not rise up enough to achieve effective pasteurisation. The occurrence of *S. aureus* in fresh and bulk milk in this study may be attributed to the presence of sub-clinical mastitis in the lactating cows, poor sanitary practices during milking, and unclean milking utensils (Kivaria et al., 2006). The main source of *S. aureus* in milk is the udder of infected cows which could be transferred via the milkers hands, milking utensils, towels, and the environment (Radostitis et al., 1994). *S. aureus* can adapt to and survive in the udder of cow and establish chronic and sub-clinical infections. From the udder, it is shed into the milk which serves as a
primary source of infection to individuals who drink unpasteurised milk (Adams & Moss, 2011). It should be noted that the use of water of unsatisfactory microbiological quality for milking and manufacturing of milk products is associated with the risk of contamination of milk with S. aureus (Kivaria et al., 2006). The use of unclean water for washing the teat and udder of cow could lead to wet, dirty udder at milking time. Cross contamination can be avoided if the hands of milkers and milking utensils are washed adequately with detergent and clean water in between milking each cow and using the utensil. Hand washing is basic component of infection control (Larson et al., 2003). Most (71.43%) of the containers used as ‘bulk tanks’ at the Fulani settlements were probably of unsatisfactory cleanliness which could have resulted in milk contamination with microorganisms. A significant association (p<0.05) was found between the kind of water used at the nono and kindirmo selling points and the occurrence of Staphylococcus aureus in the products. Majority of the nono and kindirmo selling points were using water purchased from vendors for washing utensils. Water contamination often occur in the storage containers and if the contaminated water gain access to milk or is used for rinsing containers, microorganisms present in the water will contaminate the milk (Kivaria et al., 2006). The results indicate that the microbiological quality of milk product is influenced by factors associated with water quality. No significant association (p>0.05) was found between the occurrence of Staphylococcus aureus in nono and kindirmo and street activities (mainly vehicular movement) at the selling points, although the samples collected from the selling points with high street activities were found to be more contaminated compared to the samples collected from the selling points with low street activities. Many of the selling points were located in or close to motor parks, shops, and other non-dairy activities which put the products at a high risk of contamination. The susceptibility of the isolates to gentamicin, ciprofloxacin, and chloramphenicol was in consonance with the findings of Okpo et al. (2016) and Rodrigues et al. (2017) in parts of Kaduna State, Nigeria and Brazil, respectively. The high performance of these antibiotics to could be attributed to their small molecular sizes – a factor that enhances their solubility in diluents thus enhancing their penetration power through the cell wall into the cytoplasm of the target organism where they exert their effects (Okpo et al., 2016). This agrees with the assertion of Maillard (2002) who opined that, the high efficacy of antibiotics may be attributed to their molecular sizes. High level of susceptibility (88.9%) of the S. aureus obtained in this study to vancomycin was observed. None of the isolates was found to be resistant to vancomycin. This finding is not surprising because vancomycin is rarely used in the treatment of diseases in livestock and in routine chemoprophylaxis in the study area which could lead to resistance among bacteria as a result of selective pressure. This result is in consonance with the findings of Suleiman et al. (2012) and Rodrigues et al. (2017) who opined that, the non-use of vancomycin for routine chemoprophylaxis and therapy in an area can result in S. aureus exhibiting high susceptibility to it. This finding also agrees with the results of Alian et al. (2012) in Iran who recorded 0% resistance among S. aureus isolated from dairy products. However, this finding contrasted starkly with that of Umaru et al. (2013) and Usman & Mustapha (2016), who reported 42.6 % and 66.7% resistance of S. aureus to vancomycin in Kaduna and Zaria, respectively. The disparity between the findings of the present study and the aforementioned could be as a result of contamination of the milk with vancomycin-resistant S. aureus derived from human sources. The present study observed that all the nine (9) S. aureus isolates were resistant to ampicillin and amoxicillin/clavulanic acid. This could be attributed to the contamination of milk products (nono and kindirmo) with S. aureus strains that are resistant to β-lactam drugs derived from human sources. S. aureus resistant to one β-lactam drug can develop resistance to β-lactams because they have the same mechanism of activity (Suleiman et al., 2012). This finding is not surprising because, outside the hospital environment, people have easy access to various antibiotics at any drug store without any prescription from qualified personnel. This agrees with the findings of Anueyiagu & Isiyaku (2015) who reported 100% resistance of S. aureus isolated from dairy products in Jos, Plateau State, Nigeria, and Jahan et al. (2015) in Bangladesh. The relatively high frequency of resistance to tetracycline as observed in this study could be attributed to tetracycline being the most commonly available antibiotic that is used as a growth promoter and routine prophylaxis in livestock management in Nigeria (Olatoye, 2010). This finding is a cause for concern considering the fact that, tetracycline is a first-line drug in Nigeria. This is one drug that people with cases of gastro-intestinal infections in most developing countries readily purchase over-the-counter for self-medication (Chigor et al., 2010). This was in consonance with the findings of Usman &
Mustapha (2016), and Tessema (2016), who reported 55.5% and 40% resistance of *S. aureus* isolated from dairy products in Kaduna State, Nigeria, and Ethiopia respectively. This trend is a cause for concern in human medicine and livestock disease management and production generally due to the existing emergence of bacterial strains that are resistant to major antibiotics. The use of antibiotics in food animals have been established to promote the spread of antibiotic-resistant bacteria via the food chain to humans resulting in human infections (Phillips et al., 2004). The relatively high level of resistance to erythromycin could be a reflection of the frequent use and misuse of the antibiotic in the study area. Higher levels of resistance of 76% and 85.7% among *S. aureus* isolated from dairy products have been reported Mirzaei et al. (2012) and Anueyiagu & Isiyaku (2015) in Iran and Kaduna State, Nigeria, respectively. The relatively high level of resistance to sulphonamide/trimethoprim in this study is baffling considering the fact that, the drug is not routinely used in veterinary practice in Nigeria. This suggests cross contamination of the dairy products by handlers with the drug-resistant strains of the pathogen. Mixed fermentation is known to occur in dairy products like *nono* and *kindirmo* and as the fermentation process is uncontrolled and that different organisms can occur at different times, transfer of determinants of antibiotic resistance can occur between organisms. Food is an important medium through which the transfer of determinants of antibiotic resistance among bacteria occurs. Such transfer can occur by means of residues of antibiotics in foods, through the transfer of antibiotic-resistant foodborne pathogens, or through the ingestion of drug-resistant strains of the original food microflora, and transfer of antibiotic-resistance determinants in bacteria (Pereira et al., 2009).

Multidrug resistance is defined as resistance of an isolate to three or more antibiotics in different classes (Magiorakos et al., 2011). This finding is in consonance with the findings of Umaru et al. (2013), Anueyiagu & Isiyaku (2015), Tessema (2016), and Chaalal et al. (2016) who reported cases of multidrug resistance among *S. aureus* isolated from dairy products in Zaria, Jos, Ethiopia, and Algeria respectively. The isolates were resistant to a combination of three (3), four (4), five (5), and six (6) of the antibiotics tested. Isolates obtained from Keffi, Akwanga, and Lafia, showed higher frequencies of multi-drug resistance. Multi-drug resistance in *S. aureus* may be attributed in part, to the spread of mobile genetic elements like plasmids, transposons, and integrons that may confer resistance to numerous antimicrobial agents (Zhao et al., 2001). According to Aarestrup (1995) and Levin et al. (1997), determinants of multi-drug resistance are capable of being disseminated in a region or between regions as a result of antibiotic selective pressure in either livestock or humans. Empirical evidence abounds which indicate that drug-resistant strains of bacteria can be transmitted to humans via food (Khachatourians, 1998).

The five (5) antibiotic resistance patterns among the *Staphylococcus aureus* isolates recorded in this study, varied with the nine (9) and 25 antibiotic resistance patterns recorded among *S. aureus* isolated from dairy products by Usman & Mustapha (2016), and Shiferaw & Ahmad (2016) in Kaduna State, Nigeria and Bahir Dar, Ethiopia, respectively. The disparity in the antibiotic resistance patterns of *Staphylococcus aureus* isolates recorded in the present study and the one recorded in Ethiopia could be as a result of the different levels of use and misuse of antibiotics in the two different areas.

The detection of *Staphylococcus aureus* in fresh and fermented milk in parts of Nasarawa State, Nigeria, suggests that consumption of dairy products especially those that are produced using traditional methods constitute a hazard to consumers as the transmission of pathogens via foods has been well documented. The antibiotic susceptibility profile of the *S. aureus* isolates revealed high performance of gentamicin, ciprofloxacin, imipenem, vancomycin, and chloramphenicol, while relatively high levels of resistance to tetracycline was recorded. This is of public health concern because tetracycline is a commonly used antibiotic. The data obtained in this study suggests that, selection pressure imposed by the use of antibiotics whether therapeutically in human and veterinary medicine, or in routine chemoprophylaxis in livestock production is a key driving force in the promotion of antibiotic resistance in *S. aureus*. It is therefore recommended that since compliance with basic hygiene requirements is not guaranteed, hazard analysis and critical control points (HACCP) concepts should be seen as a part of an effective total hygiene concept at the selling points. There is need for frequent education of the *nono* and *kindirmo* sellers on the aspects of milk hygiene and handling practices, which will help in no small measure in improving the quality standards of the products at the selling points. This can be achieved through teaching and training programmes using participatory approach method. There is need for relevant authorities to educate the public on the
dangers of indiscriminate purchase and use of antibiotics.

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Conflicts of Interest
The authors declare no conflict of interest.

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