RESEARCH ARTICLE

Sokoto Journal of Veterinary Sciences

(P-ISSN 1595-093X: E-ISSN 2315-6201)

http://dx.doi.org/10.4314/sokjvs.v17i1.8

Anosa *et al*./*Sokoto Journal of Veterinary Sciences*, **17**(1): 62 - 70.

Occurrence and antibiogram of *Staphylococcus aureus* and methicillin-resistant *Staphylococcus aureus* from table eggs in Nsukka, Enugu State, Nigeria

GN Anosa[1], EV Ezenduka[2]*, KC Gabriel[1], A Ngene[1] & UU Eze[1]

[1.] *Department of Veterinary Medicine, University of Nigeria, Nsukka, Nigeria*

[2.] *Department of Veterinary Public Health and Preventive Medicine, University of Nigeria, Nsukka, Nigeria*

*Correspondence: Tel.: +2347061032840; E-mail: ekene.ezenduka@unn.edu.ng

Publication History:
Received: 20-11-2018
Accepted: 29-01-2019

Abstract

Staphylococcus aureus is one of the leading causes of foodborne diseases worldwide and is associated with consumption of various foods of animal origin, including eggs and other poultry products. Eggs get contaminated with food poisoning pathogens when they make contact with dirty surfaces, litter materials as well as clothing and hands of poultry workers. This study aimed to reveal the presence of methicillin resistant *Staphylococcus aureus* (MRSA) in table eggs in Enugu, Nigeria and to evaluate the antimicrobial susceptibilities of the isolates. A total of 290 eggs comprising of 58 composite samples (5 eggs/composite) from either the eggshell or egg contents were analyzed for *S. aureus*. Each isolate was tested against 15 antimicrobial agents. Seventy-five (75) isolates of *S. aureus* were recovered from the 58 composite samples, 51 from eggshells and 24 from egg contents. Of the 75 isolates of *S. aureus* tested for antibiotic susceptibility, frequency of resistance was very high to penicillin (96%) and amoxicillin (96%) but low to ciprofloxacin (2.7%) and gentamicin (4%). A high number of multi-drug resistant isolates were observed, with as much as 60 (80%) of the isolates being resistant to 8 or more antibiotics. The study confirmed a high occurrence of multi-drug resistant *S. aureus* in table eggs, including MRSA. An improvement in hygienic practices in poultry farms and retail outlets and the control of the use of antibiotics in poultry production is recommended.

Keywords: Antibiogram, *Staphylococcus aureus*, MRSA, Table eggs, Nigeria

Introduction

Egg, a product of poultry production is a common source of protein in the tropics. The hen's egg is very rich in nutrients, making it one of the most valuable and perfect foodstuffs. Eggs can fully meet the requirements of humans for all nutrients necessary for their development and life functions (McIntosh, 2018).

Eggs can, however, be contaminated by micro-organisms, especially bacteria (Jahantigh & Dizaji, 2015). The contamination can occur when passing through the vent, but many researchers suggest that the contamination occurs mostly within a short period after lay, due to contact with dirty surfaces (Manickam *et al*., 2017). Several factors have been incriminated in egg contamination: among these factors are litter materials, egg crates, feaces of the birds (Al Monami *et al*., 2018), egg packing and storage. Other sources of contamination are clothes

and hands of poultry workers, dust and the environment. Human handling of food products, as well as infection/colonization of farm animals and workers, has been described as important mechanisms of contamination of eggs with *Staphylococcus aureus* (Hiramatsu *et al*., 2014). Increasing numbers of micro-organisms on the eggshell consequently increase the risk of microbial egg penetration and egg content contamination (Manickam *et al.*, 2017). Some of these contaminant organisms cause food poisoning directly, as in the case of *Salmonella* infection when they are consumed, or indirectly through their toxins, as seen in the case of *S. aureus*.

S. aureus is an ubiquitous pathogen that is associated with various disease conditions including skin and soft tissue infections, osteomyelitis, sepsis and is of public health concern (Kechrid *et al.,* 2011). Apart from causing infections, *S. aureus* is also associated with toxin-mediated food poisoning. *S. aureus* is the third most common source of confirmed food poisoning in the world (Akbar & Anal, 2013). Available information indicates that *Staphylococcus* and some other Gram-positive bacteria are mostly found on the egg surface likely due to their better adaptation to survival and development in dry environment and ready availability in dust, faeces and soil in the poultry house because of their ubiquity in nature (Pyzik & Marek, 2012).

Numerous *S. aureus* enterotoxins have been described, and the ingestion of these neurotoxins causes a rapid onset of nausea and vomiting within 1-6 h. Abdominal cramping, prostration, diarrhea and at times, fever are also observed (Kechrid *et al.,* 2011). Several studies have described point-source foodborne outbreaks attributed to a variety of foods (Leong *et al.*, 2014).

Due to the relatively rapid progression of *S. aureus* food poisoning and an equally rapid return to normal state of health almost always, it is speculated that most cases are unreported (Shawish & Al-Haman, 2016). This could be responsible for the dearth of information on *S. aureus* food poisoning, especially in Nigeria and Africa. The continuing emergence of multi-drug resistant pathogenic organisms is a global economic and public health concern. Infections involving these multi-drug resistant organisms have been associated with frequent treatment failure and increased the severity of disease (Finch & Hunter, 2006). The use of antibiotics in human hospitals and for animal production may result in the increased occurrence of bacterial resistance (Akbar & Anal, 2013). Antibiotics are also used as antimicrobial growth promoters in animal production (Gutierrez *et al.*, 2017), especially in poultry production. In Nigeria, there is widespread misuse/abuse of antibiotics, particularly in poultry production. MRSA are isolates of *S. aureus* which have acquired genes encoding antibiotic resistance to all penicillins, including methicillin. Antibiotic resistance may be an inherent trait of the organism that renders it naturally resistant, or it may be acquired by means of mutation in its' own DNA, or acquisition of resistance-conferring DNA from another source (Haddad *et al*., 2018). MRSA are resistant to all currently available beta-lactam antibiotics, including penicillins; cephalosporins; carbapenems; and their derivatives (Foster, 2017). The organism has been considered as an emerging problem in veterinary settings, and various studies have reported MRSA in different species of animals and veterinary professionals (Dittmann *et al.*, 2017; Momtaz *et al*., 2013). MRSA has been detected in a variety of foods (pork, beef and chevon) of animal origin from countries in North America, Europe and Asia (Hadjirin *et al.*, 2015; Benedetti *et al.,* 2010). There is a dearth of information on the occurrence of MRSA in eggs after it was first reported in chickens in Korea (Normanno *et al.*, 2007). The incidence of foodborne illness is increasing worldwide possibly due to a change in commercial food production involving minimal processing and consumer demands for ready to eat meals. Foods, including eggs, are indiscriminately sold by street vendors in Nigeria with little or no recourse to public health concerns.

This study aimed to reveal the presence of *S. aureus* and MRSA in table eggs in Enugu, Nigeria and furthermore to evaluate the antimicrobial resistance or sensitivity pattern of this pathogen.

Materials and Methods

Study area

Enugu state is a largely agricultural state located in South Eastern Nigeria. This area has a typical tropical climate characterized by two distinct seasons, the wet and dry seasons with mean temperatures of at least 18°C (64°F). Eggs are an important source of protein in Enugu state. Poultry farms in Enugu state are intensively reared mostly in deep litter and sometimes battery cage. Eggs are sold in crates either directly from the farms or retail outlets ranging from market to supermarkets or side road stalls

Sample collection

A total of 290 eggs were collected randomly from 13 poultry farms (n = 65) and 45 retail outlets (n = 225)

in the study area, over a period of 4 months (January – April 2016). Eight major farms from Enugu metropolis and 5 major farms from Nsukka metropolis were sampled. Five crates were systematically selected from the crated eggs in each farm, depending on the number of crates available at the time of visit. Every fifteenth egg from each crate was then selected, making a total of five eggs from each farm. Same systematic random sampling was used in selecting eggs from 45 retail outlets; 30 outlets from Enugu metropolis and 15 from Nsukka Metropolis. Five eggs per outlet, make up a composite sample. Overall, 58 composite samples were analyzed for the presence of *S. aureus*. Forty-five samples made up of 5 eggs per composite were analyzed from retail outlets, while 13 samples made up of 5 eggs per composite were sampled from the poultry farms.

Sampling of eggs

Unwashed eggs were collected in sterile polythene bags and transported to the laboratory for further studies. Sterile cotton swabs moistened with sterile buffered peptone water (BPW) were used to swab the entire surface area of the eggshell and inoculated into 10ml BPW in screw-capped bottles and incubated at 37^{o}C for 24 h.

To collect the egg contents, the surface of the eggs was sterilized by immersion in 70% alcohol for 2 min, air-dried in a sterile chamber for 10 min, and then cracked with a sterile knife. Each egg content was then thoroughly mixed in sterile conical flax over a vortex mixer, and 25 ml (from each composite sample) was inoculated into 225 ml of BPW and incubated at 37^{o}C for 24 h.

Isolation and Identification of S. aureus

Following pre-enrichment in BPW, the cultures (eggshell or contents) were streaked onto mannitol salt agar (MSA) a selective media for *S. aureus* and incubated at 37^{o}C for 48 hours. Yellowish colonies with yellowish background on MSA were considered presumptive *S. aureus* and identified with Gram stain and biochemical tests including catalase, coagulase (tube coagulation), and hemolysis test using 5% sheep blood agar.

Identified *S. aureus* isolates were subcultured on Nutrient agar (Oxoid®) at 37^{o}C for 24 h.

Detection of methicillin-resistant S. aureus

The isolates were identified as methicillin-susceptible (MSSA) or resistant (MRSA) using Oxacillin resistant screening agar base (ORSAB®, Oxoid). The *S. aureus* isolates were streaked onto ORSAB and incubated at 37^{o}C for 24 h. Blue colonies were considered presumptive MRSA isolates. The subcultured isolates that showed no growth were considered MSSA.

Presumptive MRSA colonies were finally confirmed by the PBP-2′ latex agglutination test (Oxoid, ® UK).

Antibiogram of MRSA and other Staphylococcus isolates

Antimicrobial susceptibility tests were carried out by the disc diffusion method (Bauer *et al.*, 1966) following the guidelines of the Clinical and Laboratory Standards Institute (CLSI, 2011), using *Staphylococcus aureus* ATCC 25923 as the reference standard. The following antibiotics were used: Oxacillin (Ox 1µg), Neomycin (N 30µg), Doxycycline hydrochloride (D 30µg), Rifampicin (RD 5µg), Amoxicillin-clavulanic acid (AMC 30µg), Gentamicin (CN 30µg), Tetracycline (TE 30µg), Erythromycin (E 15µg), Vancomycin (VA 30µg), Amoxicillin (AML 10µg), Sulphamethoxazole–trimethoprim (SXT 25µg), Nitrofurantoin (F 300µg), Penicillin G (P 10µg), Ciprofloxacin (CIP 5µg), Streptomycin (S 10µg) (Oxoid UK). Briefly, a suspension of each isolate equivalent to 0.5 McFarland turbidity standards (1x 10^{8}cfu/ml) was prepared and spread on Mueller-Hinton agar (Oxoid, UK) plates. The antibiotic discs were aseptically placed on to the inoculated Mueller-Hinton agar plates and incubated at 37^{o}C for 24 h. The susceptibility to the antibiotics was observed as a clear zone of inhibition around the discs. The zone of inhibition was interpreted as recommended by the CLSI (2011).

Statistical analysis

Data from the study were analyzed in GraphPad Prism Statistical software version 5.02 (www.graph-pad.com). The frequency of contamination of eggs by *S. aureus* according to the source of eggs (i.e., retail outlet/farm) and location in egg (i.e., eggshell/egg content) were compared using the Fisher's exact test. A probability value of less than 5% was considered significant.

Results

Occurrence of S. aureus in/on eggs from retail outlets and farms

Table 1 shows that out of the 75 positive isolates of *S. aureus* from 58 composite samples, 51(87.9%) were isolated from eggshells while 24(41.4%) isolates were from egg contents. From the retail outlets, 41(91.1%) *S. aureus* isolates were from egg shell while 18(43.9%) isolates were from the egg contents. From the 13 Farms sampled, 10(76.95%)

and 6(46.15%) *S. aureus* organisms were isolated from egg shells and egg contents respectively. There was a significant (P = 0.0001) association between the part of egg sampled and contamination with *S. aureus.*

Susceptibility of Staphylococcus aureus isolates to antimicrobial agents

The antimicrobial susceptibility test was done using *Staphylococcus aureus* ATCC 25923 as the reference standard and the reference inhibition zones for each antimicrobial stated (Tables 2 and 4). All the intermediate inhibition zones were regarded as resistant. The results indicated a high rate of resistance to penicillin (96%), amoxicillin (98.7%), and erythromycin (98.7%), while lower rates were observed against ciprofloxacin (18.7%) and gentamicin (5.3%) (Table 2). A high number of multi-resistant isolates were observed, with as much as 80% (n = 60) of the isolates being resistant to 8 or more different antibiotics (Table 3). The predominant resistance patterns amongst the *S. aureus* isolates were: D-RD-SXT-AML-F-P-VA-E-S-TE-OX-AMC (n = 6) and RD-SXT-AML-F-P-VA-E-S-TE-OX-AMC (n = 4). Most (68%) of the *S. aureus* isolates had unique resistance patterns.

Susceptibility of MRSA isolates to antimicrobial agents

Out of a total of 75 *S. aureus* isolates, 43(57.3%) were MRSA and 14(18.7%) were MSSA. The MRSA isolates expressed complete (100%) resistance to oxacillin (Figure 1), a high rate of resistance to penicillin (95.3%), amoxicillin (86%), nitrofurantoin (93%), and vancomycin (90.6%). However, a low rate of resistance to ciprofloxacin (30.3%) and gentamicin (3%) was detected (Table 4).

Discussion

S. aureus was isolated from both eggshell and egg contents from the sampled eggs, and the isolates were resistant to commonly used antibiotics in poultry production in the study area. It is a well-known fact that contaminated food is an important source of transmission for pathogenic bacteria. It is the main source of enteric diseases in the developing countries and is a major cause of morbidity and mortality (Akbar & Anal, 2013). *S. aureus* is considered as one of the most important pathogenic species causing illness (Akbar & Anal, 2013). *S. aureus* presence in food indicates poor hygiene and improper storage conditions.

Table 1: Incidence of *S. aureus* in/on eggs from retail outlets and farms in Enugu, Nigeria

Source	No of Eggs	No of composite samples	Shell Positive	Content Positive
Retail outlet	225	45	41(91.1)	18(43.91)
Farms	65	13	10(76.95)	6(46.15)
Total	290	58	51	24

Table 2: Frequency of susceptibility of *Staphylococcus aureus* isolates to antimicrobial agents and their CLSI breakpoints

Antimicrobial agent	Number (%) of isolates (n = 75)		CLSI breakpoint values (mm)		
	Resistant (%)	Sensitive (%)	S	I	R
Neomycin	25 (34)	50 (67)	≥15	13-14	≤12
Doxycycline	41 (54.6)	34 (45.3)	≥16	13-15	≤14
Gentamycin	4 (5.3)	71 (94.7)	≥15	13-14	≤12
Rifampicin	66 (14.7)	9 (12)	≥20	17-19	≤16
Amoxicillin	74 (98.7)	1 (1.3)	≥20	-	≤19
Ciprofloxacin	14 (18.7)	61 (81.3)	≥21	16-20	≤15
Sulpha/trimethoprim	64 (85.3)	11 (14.7)	≥16	11-15	≤10
Tetracycline	56 (74.7)	19 (25.3)	≥19	15-18	≤14
Streptomycin	65 (86.7)	10 (13.3)	≥18	14-17	≤13
Penicillin	72 (96)	3 (4.0)	≥29	-	≤28
Oxacillin	71 (94.7)	14 (5.3)	≥13	11-12	≤10
Vancomycin	62 (82.6)	13 (17.3)	≥17	15-16	≤14
Nitrofurantoin	64 (85.3)	11 (14.7)	≥17	15-16	≤13
Amoxicillin/Clauv	57 (76)	18 (24.0)	≥20	-	≤19
Erythromycin	74 (98.6)	1 (1.3)	≥23	14-22	≤13

S=sensitive; I= intermediate and R=resistant

Table 3a: Antibiogram profile of *Staphylococcus spp* isolated from eggs

Resistance pattern	Frequency	No of antibiotics
SXT-P-E-S	1	4
AML-F-OX-AMC	1	4
RD-AML-F-P-OX	1	5
D-AML-P-TE-OX	1	5
RD-SXT-F-E-S-OX	1	6
RD-AML-F-P-S-TE	1	6
RD-AML-P-E-OX-AMC	1	6
RD-AML-P-VA-OX-AMC	1	6
AML-F-P-E-S-OX-AMC	1	7
AML-F-P-VA-E-OX-AMC	1	7
D-RD-AML-F-VA-S-AMC	1	7
RD-AML-F-P-VA-OX-AMC	2	7
RD-AML-P-VA-E-OX-AMC	2	7
D-AML-F-P-E-S-TE-OX	1	8
D-SXT-AML-F-P-VA-S-OX	1	8
N-RD-AML-P-VA-E-S-OX	1	8
RD-AML-F-P-VA-E-S-OX	1	8
RD-SXT-AML-P-E-S-TE-OX	1	8
D-AML-P-VA-E-S-OX-AMC	1	8
RD-AML-F-P-E-S-OX-AMC	2	8
D-SXT-AML-P-E-S-OX-AMC	1	8
RD-AML-F-P-VA-E-TE-AMC	1	8
RD-AML-P-VA-E-S-OX-AMC	1	8
RD-F-P-VA-E-S-OX-AMC	1	8
RD-SXT-AML-F-P-VA-E-S-OX	2	9
N-RD-AML-F-P-VA-S-OX-AMC	1	9
RD-AML-F-P-VA-E-OX-AMC	1	9
D-RD-AML-F-P-VA-E-OX-AMC	1	9
RD-AML-F-P-VA-E-S-OX-AMC	1	9
D-RD-AML-F-P-VA-E-S-AMC	1	9
RD-SXT-AML-P-VA-E-S-TE-OX	1	9
AML-F-P-VA-E-S-TE-OX-AMC	1	9
RD-SXT-AML-F-P-VA-E-OX-AMC	1	9
RD-SXT-AML-F-P-VA-TE-OX-AMC	1	9
RD-SXT-AML-P-E-S-TE-OX-AMC	1	10
RD-AML-F-P-VA-E-S-TE-OX-AMC	1	10
D-RD-SXT-AML-P-VA-E-S-TE-OX	1	10
D-RD-SXT-AML-F-P-E-S-OX-AMC	1	10
RD-SXT-AML-F-P-VA-E-S-OX-AMC	2	10
SXT-AML-F-P-VA-E-S-TE- OX-AMC	1	10
RD-SXT-AML-F-P-VA-S-TE-OX-AMC	1	10
RD-SXT-AML-F-P-VA-E-TE-OX-AMC	1	10
N-RD-SXT-AML-F-P-VA-E-S-TE-OX	1	11
D-RD-SXT-AML-F-P-VA-E-S-TE-OX	1	11
D-RD-AML-F-P-VA-E-S-TE-OX-AMC	1	11
N-D-RD-AML-P-VA-E-S-TE-OX-AMT	1	11
D-RD-AML-F-P-VA-E-S-TE-OX-AMC	3	11
D-RD-SXT-AML-F-P-VA-E-S-OX-AMC	1	11
D-SXT-AML-P-VA-CIP-E-S-TE-OX-AMC	1	11
D-RD-SXT-AML-F-P-VA-E-TE-OX-AMC	1	11

Table 3b: Antibiogram profile of *Staphylococcus spp* isolated from eggs

D-RD-SXT-AML-P-VA-E-S-TE-OX-AMC	2	11
RD-SXT-AML-F-P-VA-E-S-TE-OX-AMC	4	11
N-D-RD-SXT-AML-F-P-VA-E-S-TE-OX	1	12
D-RD-SXT-AML-F-P-VA-E-S-TE-OX-AMC	1	12
D-SXT-AML-F-P-VA-E-CN-S-TE-OX-AMC	1	12
D-RD-SXT-AML-F-P-VA-E-CN-S-TE-AMC	1	12
D-RD-SXT-AML-F-P-VA-E-S-TE-OX-AMC	6	12
N-D-RD-SXT-AML-F-P-VA-E-S-TE-OX-AMC	1	13
N-D-RD-SXT-AML-F-P-VA-E-CN-S-TE-OX-AMC	1	14
N-D-RD-SXT-AML-F-P-VA-CIP-E-S-TE-OX-AMC	1	14

D=Doxycycline; RD=Rifampicin; N=Neomycin; SXT=Sulphoquinoxaline/Trimetoprine; AML=Amoxycillin; F=Nitrofurantoin; P=Penicillin; VA=vancomycin; CIP=Ciprofloxacin; E=Erythromicin; CN=Gentamycin; S=Streptomycin, TE=Tetracycline; OX=Oxacillin; AMC=Amoxicillin-Clavulinic acid

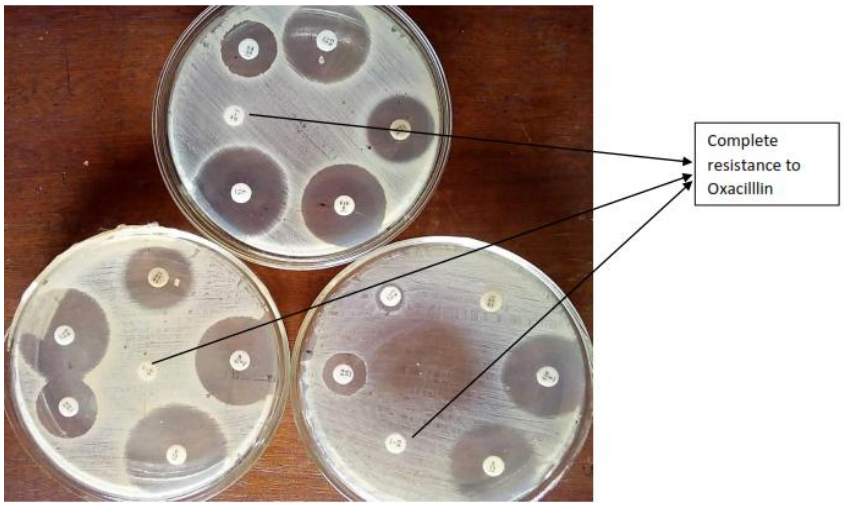

Figure 1: Antimicrobial susceptibility test of MRSA isolates

The high level of contamination of eggs with *S. aureus* could be attributed to the poor hygienic practices observed in the farms and retail outlets and the extended storage of eggs at room temperature before consumption. The average consumer in Nigeria may not be aware of the consequences or likelihood of food poisoning from eggs and other foods. The farmers and retailers take advantage of this situation; in many situations, the eggs are not properly cleaned and are kept under unhygienic conditions at room temperature for extended periods of time. An important factor influencing quantitative bacterial contamination of eggs is the temperature at which they are stored (Stepien-Pysniak, 2010), which was confirmed in the study of Al Monami *et al.* (2018), who stated that the group of eggs that had higher contamination were the ones not stored at refrigerated temperatures. This allows microorganisms to multiply during transportation and handling. The higher number of isolates of *S. aureus* obtained from the eggshell (54) when compared to egg contents (24) both on the farms and retail outlets, could be generally attributed to contamination of eggs when picked by the farm workers. Eggs may also get contaminated when passing through the cloaca (a highly contaminated area) at the point of lay, which is sometimes noticed as visible stains on the shell (Board & Tranter, 1995). Similarly, the significantly higher contamination of eggs from retail outlets than farms agrees with the work of Al Monami *et al*. (2018), who also recorded high contamination rate of eggs from retail outlets. This may be partly due to the egg handling by retailers and costumers alike when trying to purchase the eggs, also from clothes of retailers and the crates used in packaging the eggs. The contamination of the egg contents in this study is worrisome and could be attributed to the possibility of vertical transmission of organisms to the egg content, originating from systemic infection of the ovaries or ascending infection from the cloaca via the vagina to the oviduct (Keller *et al*., 1995). It has also been suggested that egg contents can be contaminated when laid, by contraction due to sudden drop in temperature, thereby moving contaminants through the shell (Padron, 1990).

The high frequency of resistance shown to the beta-lactam antibiotics including penicillin (96%) is in accordance with the natural resistance of *S. aureus* to these antibiotics. In contrast, the low resistance shown to ciprofloxacin (2%) and gentamicin (3%) could be explained by the limited use of these two

Table 4: Frequency of susceptibility of antimicrobial agents amongst isolates of MRSA and their CLSI breakpoints

Antimicrobial agent	Number (%) of isolates (n = 43)		CLSI breakpoint values (mm)		
	Resistant (%)	Sensitive (%)	S	I	R
Neomycin	15 (34.9)	28 (65.1)	≥15	13-14	≤12
Doxycycline	29 (58.1)	18 (41.9)	≥16	13-15	≤14
Gentamycin	3 (7)	40 (93)	≥15	13-14	≤12
Rifampicin	39 (90.7)	4 (9.3)	≥20	17-19	≤16
Amoxicillin	37 (86)	6 (14)	≥20	-	≤19
Ciprofloxacin	13 (30.3)	30 (69.7)	≥21	16-20	≤15
Sulpha/trimethoprim	36 (82.4)	7 (16.3)	≥16	11-15	≤10
Tetracycline	31 (72.1)	12 (27.9)	≥19	15-18	≤14
Streptomycin	35 (81.4)	8 (18.6)	≥18	14-17	≤13
Penicillin	41 (95.3)	2 (4.7)	≥29	-	≤28
Oxacillin	43 (100)	-	≥13	11-12	≤10
Vancomycin	39 (90.6)	4(9.4)	≥17	15-16	≤14
Nitrofurantoin	40 (93)	3 (7)	≥17	15-16	≤13
Amoxicillin/Clauv	37 (86)	6 (14)	≥20	-	≤19
Erythromycin	36 (83.7)	7 (16.3)	≥23	14-22	≤13

S=sensitive; I= intermediate and R=resistant

antibiotics in the study area due to their relatively high cost. Gentamicin like all other aminoglycosides is poorly absorbed orally. It requires the parental administration to reach a minimum inhibitory concentration in the systemic circulation. This could limit the use of gentamicin and subsequent development of resistance. The high resistance shown to vancomycin (81.3%), which is also widely considered to be the drug of choice for MRSA, has serious clinical implications. In June 2002, the first clinical isolate of *S. aureus* completely resistant to vancomycin was isolated from a patient in the United States (CDC, 2002). Some other workers (Gutierrez *et al.*, 2017) did not observe *S. aureus* resistance to vancomycin in poultry meat samples. Otalu *et al.* (2011), reported that 100% *S. aureus* isolates from poultry meat were resistant to tetracycline and 61.5% resistant to methicillin in Nigeria. They also reported 15.4% and 38.5% resistance against ciprofloxacin and gentamicin. Our results showed a high incidence of multi-resistant *S. aureus* with 80% of the isolates being resistant to 8 or more different antibiotics. These bacteria can reach immune-compromised people through eggs and cause infections, and result in failure of antibiotic therapies. This high prevalence of multi-resistant isolates can be explained by the uncontrolled use of antimicrobial agents in the treatment of bacterial infections or as growth promoters. In Nigeria, there is poor regulation of the use of antibiotics and other drugs in livestock production (Ezenduka *et al.*, 2011). The farmers have unlimited access to these agents which are invariably of substandard quality.

The high rate (37%) of MRSA infection in table eggs from the study area poses a serious public health risk. The presence of MRSA in foods particularly poultry meat has been well documented in Germany 34.5% (Nkang *et al*. 2010), Netherlands 16% (Cuny, 2011), China 2.3% (De Boer *et al*., 2009), and England 7.3% (Citak & Duman, 2011). Foodborne illness due to MRSA has been documented causing severe symptoms, illustrating the potential impact that this pathogen can have on human health and in some individuals whose normal flora has been depleted by antibiotic treatment, MRSA in foods could cause staphylococcal enterocolitis (Crago *et al.*, 2012).

In conclusion, the present study confirmed the presence of *S. aureus* including MRSA in both egg shell and most importantly, egg content from both retail and farm eggs in the study area. A high occurrence of multi-resistance to some antibiotics was also recorded in those isolates from commercial table eggs, a potential threat to consumer health. An improvement in hygienic practices in poultry farms and retail outlets and the control of the use of antibiotics in poultry production is recommended. Also, eggs should not be eaten raw and should be properly cooked before consumption.

Conflicts of Interest

The authors declare they have no conflict of interest.

References

Akbar A & Anal AK (2013). Prevalence and antibiogram study of *Salmonella* and *Staphylococcus aureus* in poultry meat. *Asian Pacific Journal of Tropical Medicine*, **3**(2): 163-168.

Al Momani M, Janakat S & Khatatbeh M (2018). Bacterial contamination of table eggs sold in Jordanian markets. *Pakistan Journal of Nutrition*, **17**(1): 15-20.

Bauer AW, Kirby WM, Sherris JC & Turck M (1966). Antibiotic susceptibility testing by a standardized single disc method. *American Journal of Clinical Pathology*, **45**(4): 493–496.

Benedetti V, Cremonesi P, Ferrari S, Castiglioni B, Fabbi M, Vicari N, Garbarino C, Battisti A, Franco A, Feltrin F & Luini M (2010). *Staphylococcus aureus* (MRSA) from bovine milk samples. *Large Animal Review*, **16**(2): 67–70.

Board RG & Tranter HS (1995). The Microbiology of Eggs. In: *Egg Science and Technology* (WJ Stadelman, OJ Cotterill, editors). *New York, Food Products Press* - The Haworth Press, Inc. Pp 81-104.

CDC (2002). *Staphylococcus aureus* with reduced susceptibility to vancomycin – United States. *Morbidity and Mortality Weekly Report*, **51**: 26.

Citak S & Duman T (2011). *Staphylococcus aureus* and coagulase – negative *Staphylococcus* from raw chicken samples in Turkey: Prevalence and antimicrobial resistance. *Journal of Food Agriculture and Environment*, **9**(1): 156-158.

CLSI (2011). *Performance Standards for antimicrobial disc susceptibility testing, twenty first Information Supplement.* CLSI document M100-S21 Vol 31(1). Clinical Laboratory Standard Institute, Wayne, PA, Pp 68-83

Crago B, Ferrato C, Drews SJ, Svenson LW, Tyrrell G & Louie M (2012). Prevalence of *Staphylococcus aureus* and methicillin-resistant S. aureus (MRSA) in food samples associated with foodborne illness in Alberta, Canada from 2007 to 2010. *Food Microbiology*, **32**(1): 202-205.

Cuny C, Layer F & Witte W (2011). *Staphylococcus aureus* and MRSA in thawing liquid of broiler chicken carcasses and their relation to clonal lineages from humans. *International Journal of Medical Microbiology*, **301**(S1): 117.

De Boer E, ZwartKruis – Nahuis JT, Wit B Huijsdens XW, de Neeling AJ, Bosch T, van Oosterom RA, Vila *A* & Heuvelink AE (2009). Prevalence of Methicillin–resistant *Staphylococcus aureus* in meat. *International Journal of Food Microbiology*, **134**(1-2): 52-56.

Dittmann KK, Chaul LT, Lee SH, Corassin CH, Fernandes de Oliveira CA, Pereira De Martinis EC, Alves VF, Gram *L &* Oxaran V (2017). *Staphylococcus aureus* in some Brazilian dairy industries: Changes of contamination and diversity. *Frontiers in Microbiology*, 10: 3389/fmicb.2017.02049.

Foster TJ (2017). Antibiotic resistance in *Staphylococcus aureus*. Current status and future prospects, *Fems Microbiology Reviews*, **41**(3): 430-449.

Ezenduka EV, Oboegbulam SI, Nwanta JA & Onunkwo JI (2011). Prevalence of antimicrobial residues in raw table eggs from farms and retail outlets in Enugu State, Nigeria. *Tropical Animal Health and Production*, **43**(1): 557-559.

Finch R & Hunter PA. (2006). Antibiotic resistance-action to promote new technologies: report of an EU Intergovernmental Conference held in Birmingham, UK. *Journal of Antimicrobial Agents and Chemotherapy*, **58**(10): i3 - i22

Gutierrez LL, Martinez AB & Mahecha HS (2017)**.** Methicillin resistant *Staphylococcus aureus* isolated from meat raw in Cartagena, Colombia. *Revista Facultad Nacional de Agronomia*, *Medellín* **70** (1): 8091-8098.

Haddad O, Merghni A, Elargoubi A, Rhim H, Kadri Y & Mastouri M (2018), Comparative study of virulence factors among methicillin resistant *Staphylococcus* aureus clinical isolates, *BMC Infectious diseases* **18**(1): 560

Hadjirin NF, Lay EM, Paterson GK, Harrison EM, Peacock SJ & Parkhill J (2015). Detection of livestock-associated meticillin-resistant *Staphylococcus aureus* CC398 in retail pork, United Kingdom. EuroSurveillance, **20**(24): 21156.

Hiramatsu K, Katayama Y, Matsuo M, Sasaki T, Morimoto *Y* & Sekiguchi A (2014). Multi-drug-resistant *Staphylococcus aureus* and future chemotherapy. *Journal of Infections and Chemotherapy*, **20**(10): 593-601.

Jahantigh M & Dizaji RE (2015). Antimicrobial drug resistance pattern of *Escherichia coli* isolated from chickens farms with colibacillosis infection. *Open Journal of Medical Biology*, **5**(4)**:** 159-162.

Kechrid A, Perez-Vazquez M, Smaoui H, Hariga D, Rodríguez-Baños M, Vindel A, Baquero F, Cantón R & Del Campo R (2011). Molecular analysis of community acquired methicillin – susceptible and resistant *Staphylococcus aureus* isolates recovered from bacteraemic and osteomyelitis infections in children from Tunisia. *Clinical Microbiology and Infection*, **17**(7): 1020–1026.

Keller LH, Benson CE, Krotec K & Eckroade RJ (1995). *Salmonella enteritidis* colonization of the reproductive tract and forming and freshly laid eggs of chickens. *Infection and Immunity*, **63**(7): 2443-2449.

Leong D, Alvarez-Ordóñez A & Jordan K (2014). Monitoring occurrence and persistence of *Listeria monocytogenes* in foods and food processing environments in the Republic of Ireland. *Frontiers in Microbiology*, **5**: 436

McIntosh, J (2018). Everything you need to know about eggs. https://www.medicalnewstoday.com/,retrieved 25-8-2018.

Momtaz H, Dehkordi FS, Rahimi E, Asgarifar A & Momeni M (2013). Virulence genes and antimicrobial resistance profiles of *Staphylococcus aureus* isolated from chicken meat in Isfahan province, Iran. *The Journal of Applied Poultry Research,* **22**(4): 913–921.

Manickam R, Samuel MR & Ponnusamy P (2017). Isolation and antibiogram of *Staphylococcus* species from eggs of Japanese quail in an organized farm. *International Journal of Science, Environment and Technology*, **6**(2): 1275-1282.

Nkang AO, Okonko IO, Lennox JA, Eyarefe OD, Abubakar MJ, Ojezele MO, Babalola ET, Ogunnusi TA, Onajobi BI & Amusan TA (2010). Assessment of the efficacies, potencies, and bacteriological qualities of some of the antibiotics sold in Calabar, Nigeria. *African Journal of Biotechnology*, **9**(41): 6987–7002

Normanno G, Corrente M, La Salandra G Dambrosio A, Quaglia NC, Parisi A, Greco *G*, Bellacicco *AL*, Virgilio S & Celano GV (2007). Methicillin-resistant *Staphylococcus aureus* (MRSA) in foods of animal origin product in Italy. *International Journal of Food Microbiology*, **117**(2): 219-222

Otalu OJ, Junaidu K, Chukwudi OE & Jarlath UN (2011). Multi-drug resistant coagulase positive *S. aureus* from live and slaughtered chickens in Zaria, Nigeria. *International Journal of Poultry Science,* **10**(11): 871-875.

Padron M (1990). *Salmonella typhimurium* penetration through the eggshell of hatching eggs. *Avian Diseases*, **34**(2): 463-465.

Pyzik E & Marek A (2012). Characterization of bacteria of the genus *Staphylococcus* isolated from the eggs of Japanese quail (*Coturnix coturnix japonica*). *Polish Journal of Veterinary Sciences,* **15**(4): 767-772.

Shawish RR & Al-Humam NA (2016). Contamination of beef products with staphylococcal classical enterotoxins in Egypt and Saudi Arabia. *GMS Hygiene and Infection Control*, 10.3205/dgk000268.

Stepien–Pysniak D (2010). Occurrence of Gram-negative bacteria in hen's eggs depending on their source and storage conditions. *Polish Journal of Veterinary Sciences,* **13**(3): 507-513.