Isolation and Antibiotic Susceptibility of *Staphylococcus Aureus* from Fresh Poultry Meat Sold in Keffi Metropolis, Nigeria

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Abstract: Staphylococcus aureus (S. aureus) is an important opportunistic pathogen that causes both human and animals infection. Study on isolation and antibiotic susceptibility of S. aureus from poultry meat sold in Keffi metropolis, Nigeria was carried out. A total of forty (40) samples of fresh poultry meats from different location in Keffi metropolis, Nigeria were collected using sterile swab sticks and S. aureus was isolated from the sample using standard Microbiological method. The antibiotic susceptibility test was carried out using Clinical and Laboratory Standard Istitute (CLSI) protocol. Out of 40 samples of poultry meat, 29 (72.5%) of S. aureus were isolated from the poultry meat samples. The S. aureus isolates were more susceptible to Perfloxacin (86.2%), Gentamycin (82.8%), Ciprofloxacin (82.7%) and Streptomycin (79.3%). Also the isolates were less susceptible to Cotrimoxazole (48.3%), Ceftriaxone (44.8%), Erythromycin (41.4%), Ampiclox (17.8%), Amoxicillin (13.8%) and Cefuroxime (3.5%) respectively. The frequency of isolation of S. aureus from meat samples was high and the isolates were more susceptible to perfloxacin, gentamycin, ciprofloxacin and streptomycin. Further studies on molecular characterization of resistant strains of S. aureus from poultry meat in Keffi should be carried out.

Key Words: Antbiotics, Susceptibility, Staphylococcus aureus, Poultry, Meat.

1. INTRODUCTION

Poultry meat is a good source of animal protein, appealing to consumers very easily due to its sensorial attributes. In terms of safety, poultry meat ranks first or second in food associated with disease in Australia, Canada, and England, while in the United States it is considered a prevalent food vehicle of reported foodborne disease outbreaks (Sams, 2001).

Staphylococcus aureus is a Gram positive, coagulase positive coccoid cell in the family Staphylococcaceae (Songer *et al.*, 2005) commonly found in the breeder house, environment and can be isolated from the litter, dust and feathers of chickens. The bacterium is considered to be a normal resident of the chicken, located on the skin, feathers, respiratory and intestinal tracts.

A staphylococcus infection, or staphylococcosis, refers to a variety of diseases in poultry caused by staphylococci bacteria (Eric *et al.*, 2001). Approximately 20 species have been isolated but only *S. aureus*, is of veterinary importance in breeders. This organism is an important opportunist pathogen that can cause superficial to life threatening illnesses in a variety of animal species. Also *Staphylococcus aureus* is recognized as causing health care associated and community-acquired infections in every region of the world (Hassan, *et al.*, 2013, CDC, 2015).

Toxigenic *S. aureus* in meat posses a potential health hazard to consumers; however the identification of such strains should be used as a part of risk analysis of meat and poultry products (Zouharova and Rysanek, 2008).

Staphylococcus aureus has been a food safety concern for meat producers and food processors for decades because it's widespread in the environment and often detected in air, dust, water, raw meat, other foods, and on environmental surfaces. It survives desiccation and tolerates high levels of salt. *Staphylococcus aureus* is a well-known foodborne pathogen that produces heat-stable enterotoxins during growth on a variety of foods, including meat and poultry products, eggs, cream-filled pastries, potatoes, and some salads (Cunha *et al.*, 2006).

Staphylococcus aureus cells are destroyed by heat but if they have already produced enterotoxins in a food, the toxins will survive approved doses of irradiation and some thermal processes, including

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pasteurization. In some circumstances, ingestion of staphylococci can cause enteritis. *Staphylococcal enterocolitis* occurs occasionally in infants, immunocompromised adults and others receiving large doses of antibiotics. When normal human intestinal flora is depleted or absent, *S. aureus* cells may grow in the intestines and produce enterotoxins that cause profuse diarrhea (Lin *et al.*, 2010). (Augustinsson *et al.*, 2004). The emergence of multidrug resistance in Gram-positive bacteria (pneumococci, enterococci and staphylococci) is a particularly important development. Perhaps the pathogen of greatest concern is *Staphylococcus aureus*, because of its intrinsic virulence, its ability to cause an array of life threatening conditions, and its capacity to adapt to different environmental infections so much that today most dermatoses caused by bacteria can be treated effectively. (Cuny *et al.*, 2010).

Currently, a clinically significant number of staphylococcal species that infect humans and domestic animals exhibit some degree of antimicrobial resistance. In human medicine, methicillin resistance (MR) in *S. aureus* strains has contributed to the scope of multidrug resistance since the early 1960s (Morris *et al.*, 2006). This study was focus on the isolation and antibiotic susceptibility of *S. aureus* from poultry meat sold in Keffi metropolis, Nigeria.

2. MATERIALS AND METHODS

2.1. Materials

The media that were used include; Mannitol salt agar (Oxoid Ltd UK), Nutrient agar (Merck KGaA, Darmstadt, Germany) and Mueller-Hinton agar (Oxoid Ltd UK). The media was prepared in accordance with manufacturer's instruction.

The glass wares used are as follows: Petri dish, conical flask, Cover-slip, Beaker, Slide, Test tube and Bijou bottle. Sterilization was achieved using autoclave at 121° C for 15 minutes or hot air oven at 160° C for 1 hour.

Equipments that were used include; Autoclave, Refrigerator, Incubator, Weigh-balance, Micropipette and Hot air oven.

Reagents that were used include: Hydrogen peroxide, Ethanol, Na₂HPO₄, Magnesium sulphate, Phenol red, NaHCO₃, Calcium chloride (CaCl₂), Iodine reagent, Potassium chloride (KCl), and Potassium iodide and Soluble starch.

2.2. Study Area

This study was carried out in Keffi town Local Government, which is in Northern part of Nigeria, Keffi is about 58km from Abuja, the Federal Capital Territory (FCT), and 128km from Lafia, Nasarawa state capital, the town was situated on latitude 85[°] North longitude 85[°] East (Akwa *et al.*, 2007).

2.3. Sample Collection

A total of forty (40) swabs samples of poultry meat from abattoir and Keffi main markets were collected using sterile swabs sticks and transported to the Microbiology laboratory, Nasarawa State University, Keffi.

2.4. Isolation of Staphylococcus aureus

The *Staphylococcus aureus* was isolated from poultry samples as follow. The swabs samples was resuspended in 5 ml of tryptone soy broth and incubated at 37° C for 24 h and the 24 h culture was streaked on Mannitol salt agar and further incubated at 37° C for 24 h. The tiny pin head golden yellow colonies that green on Mannitol salt agar were presumptively selected as *Staphylococcus spp*.

2.5. Identification of Staphylococcus aureus

The golden yellow colonies that grew on Mannitol salt agar were identified using Gram Staining, catalase, coagulase and phospatase test as earlier described by Cheesbrough (2006).

2.6. Antibiotic Susceptibility Test

The antibiotic susceptility test was carried out as earlier described by Clinical and Laboratory Standards Institutes (CLSI) CLSI (2007) and the result of the susceptibility test was interpreted in

accordance with susceptibility break point as earlier described by CLSI (2014). The antibiobitics disc that were used includes; Pefloxacin (10 μ g), Gentamycin (10 μ g), Ampiclox (30 μ g), Zin, (20 μ g), Amoxacillin (30 μ g), Rocephin (25 μ g), Ciprofloxacin (10 μ g), Streptomycin (30 μ g), Septrin (30 μ g), and Erythromycin (15 μ g) (Optun Lab. Nigeria).

3. RESULTS AND DISCUSSIONS

3.1. Results

Out of forty (40) samples of fresh poultry meat obtained from different location in keffi metropolis, 29 (72.5%) of S. aureus were isolated from the samples. The frequency of isolation of S. aureus decreases in order with location specific; Abattoir (100%) > Main market (70%) > Daddin kowa and High court (60%) respectively as given in Table 1

The *S. aureus* isolates were more susceptible to Pefloxacin (86.2%), Gentamycin (82.8%), Ciprofloxacin (82.7%) and Streptomycin (79.3%) respectively. Also the S. aureus isolates were less susceptible to Cotrimoxazole (48.3%), Cefuroxime (44.8%), Erythromycin (41.4%), Ampiclox (17.3%), Amoxacillin (13.8%) and Ceftriaxone (3.5%) respectively as shown in Table 2

Table1. Frequency (%) isolation of Staphylococcus aureus from fresh poultry meat sold in different location in Keffi metropolis, Nigeria

Location	No. of sample	Frequency (%) S. aureus
Abattoir	10	10 (100.0)
Main market	10	7 (70.0)
Daddin kowa	10	6 (60.0)
Kofar hausa	10	6 (60.0)
Total	40	29(72.5)

Table2. Antibiotic Susceptibility of Staphylococcus aureus from fresh poultry meat sold in different location in keffi metropolis, Nigeria

Antibiotics	Disc content (µg)	No. (%) susceptibility of <i>S. aureus</i> (n= 29)
Pefloxacin (PEF)	10	25 (86.2)
Gentamycin (CN)	10	24 (82.8)
Ampiclox (APX)	20	5 (17.3)
Ceftriazole (CTZ)	30	1(3.5)
Amoxacin (AM)	25	4(13.8)
Cefuroxime (CFX)	10	13(44.8)
Ciprofloxacin (CPX)	30	24(82.7)
Streptomycin (S)	30	23(79.3)
Cotrimoxazole (COT)	30	14(48.3)
Erythromycin (E)	10	12(41.4)

3.2. Discussion

Staphylococcus aureus is an important health care and community acquired infection in every region of the world (Hassan, et al., 2013, CDC, 2015). Studies have shown that toxins produce by S. aureus in meat posses a potential health hazard to consumers (Lee, 2003) and isolatin of such strains is useful as a parts of risk analysis of meat and poultry product (Zouharova and Rysanek, 2008). Studies on isolation and antibiotic susceptibility of S. aureus from fresh poultry meat were evaluated from this study.

The high frequency of isolation of S. aureus from poultry meat observed in this study was not surprising and this is in agreement with other studies reported elsewhere (Gundoga, et al., 2005, Normanno et al., 2005, and Abdalrahman, et al., 2015). The high frequency of isolation of S. aureus isolates from fresh poultry meat may be due to the possible transfer of S. aureus genes encoding antibiotic resistance (Lee, 2005). The isolation frequency of S. aureus observed in study was higher than other study reported by Hassan et al. (2013) and Pu and Ge (2011).

The high susceptibility of S. aureus isolates from fresh poultry meat to pefloxacin, gentamycin and streptomycin observed in this study was expected and however this report was not different from other studies reported by Waters et al. (2011) and Hanson et al. (2011). The high susceptibility of S. aureus to these antibiotics may be due to the fact that some of these antibiotics are costly or are in injectable forms and the possibility of abuse of such antibiotics may be low. The high susceptibility of

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S. aureus isolates to the antibiotics observed in this research is in agreement with other studies earlier described (Zouharova and Rysanek, 2008, Gundoga, et al., 2005, Normanno et al., 2005, and Abdalrahman, et al., 2015).

The low susceptibility of Cefuroxime, Erythromycin, amoxicillin and ampiclox observed in this study was not different from the other studies reported by (Lee, 2005, Hassan, et al., 2013). The low susceptibility of S. aureus to antibiotics mentioned may be due to inappropriate use of antibiotics without doctor's prescription and inclusion of antibiotics in animal feeds as growth promoter (Lee, 2005). The low susceptibility of S. aureus to beta-lactam antibiotics observed in this study may be due to the production of beta-lactam enzymes of the modification of the drug target site (Canton and Valverde, 2008).

From this study it was however observed that the frequency of isolation of S. aureus observed in this study was high and isolates were more susceptible to flouroquinolones and aminoglycosides and in view of this, such drugs may be useful for treatment of Staphylococcal infections, hence further studies on molecular characterisation of resistant strains of S. aureus from poultry meat sold in Keffi should be carried out.

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