Fjrs.futa.edu.ng
FUTA Journal of Research in Sciences
ISSN: 2315 – 8239 (Print); E-ISSN: 2489 - 0413

FUTA Journal of Research in Sciences, Vol. 14(1), April, 2018: 112-120

METHICILLIN RESISTANT *STAPHYLOCOCCUS AUREUS* IN COMMERCIAL LAYERS AND POULTRY FARM ATTENDANTS IN NORTH CENTRAL, NIGERIA.

[1]M.I. Bale , [1]D.O.Kolawole, [2]S.K. Babatunde , [1]E.O. Ajiboye, [1]R.M. Adedayo, [1]M. Adetumbi and [1]A.T. Ajao.

[1] Microbiology Unit, Department of Biosciences and Biotechnology, College of Pure and Applied Sciences, Kwara State University Malete, Kwara State, Nigeria.

[2] Microbiology Unit, Department of Biological Sciences, Kings University Odeomu, Osun State Nigeria

Corresponding Author's:muribaale@gmail.com; 08035442428

ABSTRACT

Lately, infections caused by antibiotic resistant microorganisms especially methicillin resistant *Staphylococcus aureus* (MRSA) in man and animals is on the increase. This study therefore assessed the existence of MRSA in layers birds and its attendants in three (3) medium sized commercial poultry farms in Asa, Nigeria. A total number of 180 cloacal swabs were collected from egg producing layer birds from three selected farms, nasal swabs was also taking from 29 volunteered poultry attendants. They were all screened for *Staphylococcus aureus* using standard microbiological procedures. Antimicrobial susceptibility testing of the isolates to cefoxitin and other conventional antimicrobial sensitivity discs of some antibiotics commonly prescribed in the study area was done using the disc's diffusion method. A total of 126 (70%) *Staphylococcus aureus* was detected from layers and 8 (6.3%) were MRSA. The resistance profile of the 8 MRSA isolates shows that all the isolates (100%) were resistant to cefuroxime, cefotaxime, cloxacillin and augmentin while 25%, 50%, and 75% are resistant to ofloxacin, gentamicin and erythromycin respectfully. Also 13 (44.8%) of the poultry workers carried *Staphylococcus aureus* in their nostrils and 5 (38.5%) carried MRSA. The antibiotic sensitivity against the 5 MRSA shows 100% resistance to cefuroxime, cefotaxime, cloxacillin and augmentin, 20% resistance to both gentamicin and erythromycin, while 100% susceptibility against ofloxacin was recorded. The detection of MRSA in both the birds and poultry attendants in the study area is of great public health importance.

Key words; Antibiotic sensitivity, LA-MRSA, Public health, Methicillin

INTRODUCTION

The increased rate of usage of antimicrobial agents for infections treatment in both human and animal has prompted the rise in antimicrobial resistance in bacterial organisms among which is *S. aureus* (Bager *et al*., 1997; Casey *et al.,* 2007). *S. aureus* is a gram positive catalase, coagulase positive coccoid bacterial belonging to family Staphylococcaceae (Songer and Post., 2005).This organism is carried asymptomatically in the nose and mucous membrane of about 33% of human population and has also been isolated in the skin, feather, respiratory and intestinal tracts of poultry birds (Casey *et al.,* 2007). It is a major opportunistic pathogen in both man and animal responsible for a wide range of community acquired and nosocomial infections, pneumonia, toxic shock syndrome, food poisoning, post-operative wound infections, bacteremia, septic shock syndrome, and staphylococcal scalded-skin syndrome among many others (Martineau *et al*., 2000). This pathogen has also been implicated in many poultry infections which include bumble foot, gangrenous dermatitis, tenosynovitis,

omcephalitis and bacterial chondronecrosis with osteomyelitis (BCO) (Capital *et al.,* 2001; Suleiman *et al.,* 2013).

Many poultry farms use different antimicrobial agents in sub-therapeutic/therapeutic doses for disease prevention, growth promotion, and treatment of bacterial diseases (Waters *et al*., 2011).They are also used freely as feed or water additives without supervision from regulatory agency which has prompt the development of resistance to the drugs been used (Kabir *et al*., 2004; Otalu *et al*., 2011).This indiscriminate practice is usually worse in many developing countries including Nigeria, where there are no proper legislation and strict laws guiding the use of antimicrobials in food-producing animals which has led to increase in resistance to most of the antimicrobials agent especially the beta-lactams (penicillin and cephalosporin) used in food animals production, animal feeds and growth promotion (Wulf and Voss, 2008).

Methicillin resistant *S. aureus (*MRSA) evolved from methicillin-susceptible *S. aureus* (MSSA) by acquisition of *SSCmec* elements containing a *mec* gene (*mec* A, more rarely *mec C*), which codes for an additional penicillin binding protein that has low affinity for β-lactam antibiotics and therefore mediates resistance to nearly all compounds from this antibiotic class (Cuny *et al.,* 2015). So far at least 11 different structural types of *SCCmec* are known (Palavecino, 2014). MRSA is a major causes of hospital associated *S. aureus* (HA-MRSA), community acquired *S. aureus (*CA-MRSA) and livestock associated *S. aureus* (LA-MRSA) (Layer *et al.,* 2012).

LA-MRSA is acquired by physical contact and exposure to contaminated dust in the poultry farm. This strain of MRSA can be transmitted from animal to man and vice versa and it is spreading at an alarming rate more than HA-MRSA and CA-MRSA and It has also been discovered in countries with low prevalence of MRSA like Netherland and Denmark (Van-Cleef *et al*., 2011). Methicillin resistance was first observed in *S. aureus* isolates from chickens in Korea (Lim *et al.,* 2010) and later detected in broiler chickens in Belgium (Persoons *et al*., 2009).

The occurrence of this pathogen is a public health concern because it could be a source of environmental contamination and transmission could also occur through the poultry products like eggs and meat (Hardy *et al.,* 2012; Suleiman *et al.,* 2013*)*. Nasal colonization has been found in 77% - 86% of human working in MRSA – positive stable environment and this colonization may be as a result of constant exposure and contact with the animal (Cuny *et al*., 2009; Van den Broek *et al*., 2009).

In Nigeria, egg is one of the major poultry products that is on a high demand due to its uses in baking, making of dough nut, pastries, egg custard, milk shake and egg pasta. With the current production of about 1 billion egg per annum (PAN., 2015), the demand on it production has lead to the increase in the rearing of the layers chicken across the country both in small and large scale. The location of many of these farms in close proximity to the human dwelling could be a source of environmental contamination and public health emergency (Li *et al.,* 2014).

This research therefore aimed to investigate the occurrence of MRSA in poultry birds and its attendants in three commercial egg producing farms in Asa Local Government Area Kwara State, Nigeria.

MATERIALS AND METHODS

Study area

An informed consent was obtained from the farms owners for the use of their birds and from the farm workers for participation in the study. This research was carried out in three (3) different well managed commercial poultry farms in Ballah, Otte Etile and Lasoju in Asa Local Government Area, Kwara State, North Central Nigeria.

Isolation and identification of *S. aureus*

A total of 180 cloacal swabs was collected from 180 apparently healthy egg producing layers of age 34weeks, 41weeks and 38 weeks old layers birds in farms in Ballah, Otte Etile and Lasoju respectfully between the first week and fourth week of March, 2016. Also, nasal swabs was also taking from 29 volunteered poultry attendants in the three selected farms in the same period.

S. aureus was isolated and identified from the samples using conventional methods which includes initial incubation in Brain heart infusion broth (as described by Persoons *et al* 2009) without the addition of antibiotic supplements. After overnight incubation at 37°C, 1µL of the broth was streaked onto Mannitol salt agar (MSA)

(Oxoid, UK) and incubated for 24-48hours at 37°C.This is then followed by Gram stain, catalase, and coagulase test (Cheesbrough, 2004). Isolates that produce characteristics golden yellow colonies on MSA and were later characterised as Gram positive cocci in clusters, catalase positive and coagulase positive were considered as *S. aureus.*

Antimicrobial susceptibility test

Antimicrobial susceptibility test was done for the catalase and coagulase positive *S. aureus* according to Clinical and Laboratory Standards Institute standard (CLSI, 2014). Each of the *S. aureus* isolates was inoculated separately into nutrient broth (Oxoid, UK) and before it was incubated at 37°C for 24 hours before they were tested. Turbidity of the culture was adjusted to correspond with that of a barium sulphate (0.5 McFarland) standard. About 0.1 ml of the nutrient broth culture was later inoculated onto Mueller Hinton agar plates after which it spread over the surface with sterile cotton swabs. Antibiotic discs were then placed on the surface of each plate by means of antibiotic disc dispenser and incubated at 37°C for 24 hours. Zone of inhibition diameters were measured and compared with the standards (CLSI., 2014). The panel of antibiotics disc used were ceftazidime (30 µg), cefuroxime (30 µg), gentamicin (10 µg), erythromycin (30 µg), ofloxacin (5µg), cloxacillin (5 µg) and augmentin (30 µg).

Methicillin resistance screening

This was done using cefoxitin (30 µg) disks (Oxoid Ltd.UK), this disc was applied with sterile forceps, and after which the agar plates were incubated for a full 24 hours at 35°C aerobically. The zone of inhibition diameter of each plate of the isolate was measured and compared with the interpretative standard (CLSI., 2014). *S. aureus* ATCC 43300 was used as control.

RESULTS AND DISCUSSION.

Out of a total figure of 180 cloacal samples that were collected from all the three farms, 60 samples each from Ballah, Otte Etile and Lasoju. Microbiological procedures used shows that 34, 45 and 47 are isolated as *S. aureus* from farm in Ballah, Otte Etile and Lasoju respectfully. Further confirmatory methods shows that 1, 4 and 3 were screened as methicillin resistant *S. aureus* in layer birds in Ballah, Otte Etile and Lasoju respectfully (Table1).

Many study has shown the presence of *S. aureus* in various parts of poultry birds (Suleiman *et al.*, 2013), Poultry dropping (Olonitola *et al.*, 2015) and poultry products (Pesavento *et al.*, 2007; Lin *et al.*, 2009; Waters *et al* 2011; Mohamed, 2013; Thaker *et al.*,2013). In this study the overall prevalence rate of *S. aureus* is 70%, this is low compare to study by Adeyeye *et al*. (2013) who reports 98% prevalence, however it is worth noted that they conducted their own study on broilers and not on layers in the active stage of egg production. All the three selected farms have layers that carries *S. aureus* in their cloaca. This is agreement with reported by Misic *et al.* (2015) who state that *S. aureus* is carried asymptomatically on the skin and mucous membrane of animals.

The total number *S. aureus* detected in each farm differs from each other, farm in Ballah has a prevalence rate of 56.7%, while farm in Otte Etile has 83.3% and Lasoju farm has 84.5% respectively (Table 1).

A total of 29 poultry workers consent to the taking of their nasal swab, out of these 13 (44.8%) show growth of *S. aureus*.

Table 1: Number and percentage distribution of *S. aureus* and methicillin resistant *S. aureus* detected in poultry Layers.

Farm	Total sample		*S.aureus*		MRSA	
	Number	Percentage	Number	Incidence rate	Number	Incidence rate
Ballah	60	33.3	34	56.7	1	0.3
Otte Etile	60	33.3	45	83.3	4	8.9
Lasoju	60	33.3	47	84.5	3	6.4

Legend; MRSA=methicillin resistant *S. aureus*,

Healthy human asymptomatically carriers *S. aureus* in their nose, throat, skin, nail and contribute substantially to normal microflora (Ekhaise *et al.*, 2008; Bien *et al*., 2011). Nasal carriage of *S. aureus* has been demonstrated as significant risk factor in nosocomial and community acquired infections in several populations. Exposure to livestock has also been demonstrated as a serious risk factor in the transmission and contamination with methicillin resistant *S. aureus* (Persoons *et al*., 2009; Smith *et al*., 2009; Van Cleef *et al*., 2011). The percentage of farm workers that is, 44.8% confirmed to be harboring *S. aureus* in their nostrilsis in the same ranges as reported by Smith *et al*. (2009) who also reported 45% carriage among pig farm labourers, carriage of 25-30% has also been reported in pig farm in Netherlands (Van den Broek *et al*., 2009). The nasal colonization is a potential public health hazard as individuals carrying this pathogen could be transmitting it within the community (Lim *et al*., 2010).

All the *S. aureus* isolated from poultry birds are resistant to cefuroxime, ceftazidime and augmentin, 53% are resistant to gentamicin, 46% to erythromycin which is in the same range as reported by Geidam *et al.* (2012) and 28% to ofloxacin. Ofloxacin is the best drug in this farms which may be attributed to low usage in the area.

The antibiotic profile against the 13 *S. aureus* in poultry workers shows that 100% are resistant to ceftazidime, and cefuroxime, this could be attributed to the fact that cefuroxime effectiveness on *S. aureus* is enhanced when combine with clavulanic acid (Jalil *et al*., 2008). 46% were resistant to gentamicin, 23% to erythromycin, 92% to augmentin and 15% to ofloxacin. Again ofloxacin has the lowest resistant profile perhaps due to reduction in its abuse in this area. The number of methicillin resistant *S. aureus* detected in layer bird is 8 (6.3%) while that detected in the workers is 5 (38.5%). The prevalence rate of MRSA in poultry farm attendants varies in each farm, with workers in Otte Etile has the highest prevalence rate of 60%, Lasoju has 50% while Ballah farm workers has no individual carrying MRSA in their nostrils. These differences in nasal colonization could be as a result of many factors like provision of good clothing for the farm workers, wearing of gloves and nose mask, maintenance of personal hygiene, isolation of sick birds and assess to rodents among others. De Boer *et al.* (2009) on the prevalence of MRSA in raw retail meat products which include pork, beef, veal, lamb and poultry indicated that 11.9% of the samples were contaminated with MRSA.

The high rate of nasal colonization of MRSA in poultry attendants in farm in Otte Etile may be attributed to high prevalence rate of MRSA in the cloacal of poultry birds in their farm which could have been the source of infection for workers who could have get infected by the birds or the contaminated environments and they could in turn transmitting it between themselves. Many researchers have also reported transmission of MRSA from animals and contaminated environments to humans (Juhasz-kaszanyitzky *et al.*, 2007; Huber *et al.*, 2010; Garcia *et al.*, 2011; Friese *et al.*, 2013).

The resistance profile of the 8 MRSA isolates from poultry birds shows that all the isolates (100%) were resistant to cefuroxime, cefotaxime, cloxacillin and augmentin while 25%, 50%, and 75% are resistant to ofloxacin, gentamicin and erythromycin respectfully (Table 4).

Furthermore, 13 (45%) of the poultry workers carried *S. aureus* in their nostrils and 5 (17%) carried methicillin resistance *S. aureus*. The antibiotic sensitivity against the 5 MRSA shows 100% resistance to cefuroxime, cefotaxime, cloxacillin and augmentin, 20% resistance to gentamicin and erythromycin and 100% susceptibility against ofloxacin (Table 4).

Table 2: Number and percentage distribution of *S. aureus* and methicillin resistant *S. aureus* detected in poultry workers.

Farm	Total sample		*S.aureus*		MRSA	
	Number	Percentage	Number	Incidence rate	Number	Incidence rate
Ballah	12	41	4	33.3	0	0
Otte Etile	8	28	5	62.5	3	60
Lasoju	9	31	4	44.4	2	50

Legend; MRSA=methicillin resistant *S. aureus*

Table 3: Resistant profile of various antibiotics to *S. aureus* in poultry layers poultry and farm attendants

	poultry	layer	birds		Poultry	farms	attendants	
Antibiotics	Ballah	Otte Etile	Lasoju	Total	Ballah	Otte Etile	Lasoju	Total
FOX	1	3	3	8	0	3	2	5
CAZ	34	34	47	126	4	5	4	13
CRX	34	45	47	126	4	5	4	13
GEN	11	25	29	65	1	3	2	6
ERY	8	21	27	56	0	2	1	3
CXC	34	43	47	124	4	5	4	13
OFL	5	12	18	35	0	1	1	2
AUG	33	43	47	123	4	5	3	12

Legend; FOX= cefoxitin, CAZ=cefuroxime,CRX=cefotaxime GEN=gentamicin, ERY= erythromycin, CXC= cloxacillin, OFL=ofloxacin, AUG= augmentin,

Table 4: Susceptibility profile of various antibiotics to Methicillin resistant *S. aureus* isolate from poultry layers and poultry farm attendants

Antibiotics	MRSA isolates from layers								MRSA isolates from attendants				
	1	2	3	4	5	6	7	8	1	2	3	4	5
CAZ	R	R	R	R	R	R	R	R	R	R	R	R	R
CRX	R	R	R	R	R	R	R	R	R	R	R	R	R
GEN	S	R	R	S	R	S	S	R	R	S	S	S	S
ERY	S	R	R	S	R	R	R	R	S	R	S	S	S
CXC	R	R	R	R	R	R	R	R	R	R	R	R	R
OFL	S	S	R	S	R	S	S	S	S	S	S	S	S
AUG	R	R	R	R	R	R	R	R	R	R	R	R	R

MRSA= methicillin resistant *S. aureus* isolate, S= susceptible, R=resistance, CAZ=cefuroxime, CRX=cefotaxime GEN=gentamicin, ERY= erythromycin, CXC= cloxacillin, OFL=ofloxacin; AUG= augmentin,

Table 5; Multiple antibiotic resistance among MRSA isolated from layer birds and poultry farm attendants.

	Poultry layers		Poultry farm attendants	
Number of antibiotics	Number of MRSA	Percentage	Number of MRSA	Percentage
4	2	25	3	60
5	2	25	2	40
6	2	25	-	-
7	2	25	-	-

Of all the antibiotic used in this study ofloxacin shows the highest level of potency to MRSA in both layers (75% susceptibility) and poultry attendants (100% susceptibility), thus it could be recommended as the drug of choice in the treatment of MRSA infections. This is in agreement with result obtained by Chibuike *et al.* (2013); Adetayo *et al.* (2014) and Bale, (2016). Fluoroquinolone has been reported as one of the drug for the treatment of MRSA infections in Nigeria (Okonko *et al.*, 2009, Okon *et al.*, 2013; Bale, 2016) and it use in animal production in Nigeria is limited, due to the high costs, however possible exposure to less potent and adulterated antibiotics by humans sharing the same environments with the farm animals may be responsible for the possible emergence of resistance to these less misused antibiotics (Nsofor and Iroegbu, 2012).

Multiple antibiotic resistance (MAR) profile shows that all the MRSA from both birds and farm attendants were multidrug resistant. MAR profile among MRSA from bird shows that 2 (25%) isolates were resistant to 4, 5, 6 and 7 antibiotics while MAR among MRSA isolated from workers shows that 3 (60%) and 2 (40%) isolate are resistant to 4 and 5 antibiotics respectfully (Table 5).

This study shows high rate of both cloacal colonization in poultry birds and nasal colonization of multi drug resistant methicillin resistant *S. aureus* in poultry farms attendants. Resistance to the antibiotic could be attributed to the constant use of different drugs as food supplements for prophylaxis and treatment of bird's diseases and infections (Persoon *et al.*, 2009).

CONCLUSION

The detection of methicillin resistant *S. aureus* in both the cloacal of the poultry birds and in the

nostrils of the poultry workers in the farms under study and the fact that all the MRSA detected are multidrug resistant shows that the level of abuse of beta-lactam antibiotics is on the high side and this may be attributed to lack of proper supervision, monitoring and evaluation of antibiotics that is been used in the poultry farm across the country. Therefore, there is need for proper education and counseling of the farm owners and workers on the danger pose by this trend in order to minimize the development of resistance and prevent public health emergency. Government also need proper drug formulations and strict monitoring of the poultry farms across the country in order to prevent the spread of resistance microorganisms from poultry birds and its products to human being.

REFERENCES

Adetayo, T. O., Deji-Agboola A. M., Popoola M.Y., Atoyebi T.J. and Egberongbe K. J. (2014) Prevalence of methicillin resistant *S. aureus* from clinical specimens in Ibadan, Nigeria *The International Journal Of Engineering And Science* (IJES) 3 (9) 2319 – 181

Adeyeye J. O, and Adewale A. O. (2013) Incidence of Methicillin-Resistant *S. aureus* (MRSA) In a Small Poultry in South West, Nigeria. *Journal of Agriculture and Veterinary Science* 5 (3) 53-55

Bager F, Madsen M, Christensen J and Aarestrup, FM (1997). Avoparcin used as growth promoter is associated with the occurrence of vancomycin resistant *Enterococcus faecium* on Danish poultry and pig farms. *Preventive Veterinary Medicine*, 31, 95-112.

Bale M.I (2016). Molecular and phenotypic characterization of methicillin resistant *S. aureus* in apparently healthy individuals in Malete rural Community M.Sc Thesis Department of Biosciences and Biotechnology Kwara State University Malete p 99-144

Bien, J O S, and Przemyslaw B (2011) Characterization of Virulence Factors of *S. aureus*:Novel Function of Known Virulence Factors That Are Implicated in Activation of Airway Epithelial Proinflammatory Response *Journal of Pathogens* 13

Capita R, Alonso-Calleja C, Garcia-Fernandez MC, Moreno B.(2002). Characterization of *S. aureus* isolated from poultry meat in Spain. *Poultry Science* 81:414-421.

Casey, AL.; Lambert, P A. and Elliot, T.S.J. (2007) Staphylococci. *International Journal of Antimicrobial Agents*, 29, Pp23–32.

Cheesbrough, M. (2004). District laboratory practice in tropical countries part 2, *Cambridge University Press, United Kingdom*, Pp. 434.

Chibuike I, Reginald A O, Solomon U C, Ifeanyi A U C J, Chinenyenwa J N, Nnadozie J and Kelechi U O (2013) Prevalence and Antibiotic Susceptibility Patterns of Methicillin Resistant *S. Aureus* (MRSA) Isolated from Healthy Inhabitants of Uturu Rural Communities, Abia State, *Nigeria Journal of Natural Sciences* 3 (10),

Clarence J. Fernandes, Lorna A. Fernandes and Peter C (2005) Cefoxitin resistance as a surrogate marker for the detection of methicillin-resistant *S. aureus. Journal of Chemotherapy* 55, 506–510

Clinical Laboratory Standard Institute (CLSI) (2014). Performance Standards for Antimicrobial Susceptibility Testing; Twenty-Fourth. This document provides updated tables for the Clinical and Laboratory Standards Institute antimicrobial susceptibility testing standards M02- A11, M07-A9, and M11-A8. 30(1)

Cohn L.A. and middleton J.R (2010). A veterinary perspective on methicillin-resistant staphylococci. *Journal of Veterinary Emergency Critical Care*. 20 pp 31-45.

Cuny, C.; Nathaus, R.; Layer, F.; Strommenger, B.; Altmann, D.; Witte, W. (2009). Nasal colonization of humans with methicillin-resistant *S. aureus* (MRSA) CC398 with and without exposure to pigs. *PLoS ONE*, *4*, e6800.

Cuny C, Lothar H. Wieler and Wolfgang Witte (2015) Livestock Associated MRSA: The Impact on Humans *Antibiotics*, *4*, 521-543

Cuny, C., Friedrich, A., Kozytska, S., Layer, F., Nubel, U., Ohlsen, K., Strommenger, B., Walther, B.,Wieler, L. and Witte, W. (2010). Emergence of Methicillin-Resistant *S. aureus* (MRSA) in different animal Species. *International Journal of Medical Microbiology*, 300 (1):109–117

De Boer, E. J. Zwartkruis-NahuisT. M. Wit B, Huijsdens X.W., D E Neeling A.J, Bosch T, Van Oosterom R.A.A., Vila A., Heuvelink A.E. (2009): Prevalence of methicillin-resistant *S. aureus* in meat. *International Journal of Food Microbiology* 134 pp52–56

Ekhaise F.O, Ighosewe O.U, Ajakpovi O. D. (2008).Hospital Indoor Airborne Microflora in Private and Government Owned Hospitals in Benin City, Nigeria. *Journal of Medical Science* 3 (1): 19-23.

Friese A, Schulz J, Zimmermann K, Tenhagen B-A, Fetsch A, Hartung J, *et al*. (2013) Occurrence of Livestock-Associated Methicillin-Resistant *S. aureus* in Turkey and Broiler Barns and Contamination of Air and Soil Surfaces in Their Vicinity. *Applied Enviromental Microbiology*.;79 (8):2759-66.

García-Álvarez L, Holden MT, Lindsay H, Webb C.R, Brown D.F, Curran MD, Walpole E, Brooks K, Pickard D.J, Teale C, Parkhill J, Bentley S.D, Edwards G.F, Girvan E. K, Kearns A. M, Pichon B, Hill R. L, Larsen A. R, Skov R. L, Peacock S. J, Maskell D. J, Holmes M. A. (2011). Methicillin-resistant *S. aureus* with a novel *mec A* homologue in human and bovine populations in the UK and Denmark: a descriptive study. *Lancet Infectious Disease*.

Geidam Y. A, Ibrahim U. I, Grema H. A, Sanda K A, Suleiman A and Mohzo D. L (2012). Patterns of antibiotic sales by drug stores & usage in poultry farms: a questionnaire-based survey in Maiduguri, Northeastern Nigeria. *Journal of Animal and Veterinary Advances*,11 (16):2852-2855.

Geidam Y.A, Zakaria Z, Abdul Aziz S, Bejo S.K., Abu J & Omar S (2012). High prevalence of multidrug resistant Bacteria in selected poultry farms in Selangor, Malaysia. *Asian Journal of Animal and Veterinary Advances* 7 (9): 891- 897.

Hardy Zielicka, A.; Zarowna, D.; Szych, J.; Madajczak, G. and Sadkowska-Todys, M. (2012): Ensuring safety of home-produced eggs to control salmonellosis in Poland: lessons from an outbreak in September 2011. Euro Surveill. 17 (47). pii: 20319.

Howden B.P, Davies J..K, Johnson PDR, Stinear T.P, Grayson ML (2012) .Reduced Vancomycin Susceptibility in *S. aureus*, Including Vancomycin-Intermediate and Heterogeneous Vancomycin-Intermediate Strains: Resistance Mechanisms, Laboratory

Detection, and Clinical Implications. *Clinical Microbiology Review* 23(1):99–139.

Huber H, Koller S, Giezendanner N, Stephan R, Zweifel C. (2010). Prevalence and characteristics of meticillin-resistant *S. aureus* in humans in contact with farm animals, in livestock, and in food of animal origin, Switzerland. *European Surveill*ance,15 (16):19542

Jalil A, Niazi I.D, Khan S.U. (2008) Evaluation of restoration of sensitivities of resistant *Staphylococcus aureus* isolated by using cefuroxime and clavulanic acid in combination.*J Ayub Med Coll Abbottabad*. 20 (2):28-30

Juhasz-Kaszanyitzky, E., Janosi S., Somogyi P., Dan A., Van Der Graaf-L, Bloois A.V, Van Duijkeren E., Wagenaar J.A. (2007). MRSA transmission between cows and humans. *Emergency Infectious Disease* 13:630-632.

Kabir J, Umoh V.J, Audu O.E, Umoh J.U and Kwaga J. K. P (2004). Veterinary drug use in poultry farms and determination of antimicrobial drug residues in commercial eggs and slaughtered chickens in Kaduna State, Nigeria. *Food Control*,15: 99-105.

Layer, F.; Cuny, C.; Strommenger, B.; Werner, G.; Witte, W. Aktuelle Daten und trends zu (2012) Methicillin-resistant *S. aureus* (MRSA). *Bundesgesundheitsbl*, *55*, 1377–1386

Li, S.; Li, J.; Qiao, Y.; Ning, X.; Zeng, T.; Shen, X. (2014) Prevalence and invasiveness of community-acquired methicillin-resistant *S. aureus:* A meta-analysis. *Indian Journal of Pathology and Microbiology*, *57*, 418–422.

Lim, SK; Nam, HM; Park, HJ; Lee, HS; Choi, MJ; Jung, SC; Lee, JY; Kim, YC; Song, SW and Wee, SH (2010). Prevalence and characterization of methicillin-resistant *S. aureus* in raw meat in Korea. *Journal of Microbiology and Biotechnol*ogy, 20: 775-778.

Lin J, Yeh KS, Liu HT and Lin J H (2009). *S. aureus* isolated from pork and chicken carcasses in Taiwan: prevalence and antimicrobial susceptibility. *J Food Prot* ;72(3):608-611.

Martineau, F., F.J. Picard, N. Lansac, C. Menard, P.H Roy, M. Ouellette and M.G. Bergeron, (2000). Correlation between the resistance genotype determined by multiplex PCR assays and the antibiotic susceptibility patterns of *S. aureus* and *S. epidermidis. Antimicrobial Agents and Chemotherapy*, 44: 231 238.

Mohamed Karmi (2013). Prevalence of methicillin-resistant *S. aureus* in poultry meat in Qena, Egypt *Veterinary World*, 6 pp 2231-0916

Misic A. M, Meghan F. D, Amanda S T, Brendan P H, Pam Tolomeo, Baofeng H U, Irving N, Ebbing L, Daniel O M and Elizabeth A G (2015) The shared microbiota of humans and companion animals as evaluated from *S.* carriage site *Microbiome* 3 pp52-7

Nicole M. Broekema, Tam T. Van, Timothy A. Monson, Steven A. Marshall, and David M. Warshauer (2008). Comparison of Cefoxitin and Oxacillin Disk Diffusion Methods for Detection of *mec A*-Mediated Resistance in *S. aureus* in a Large-Scale Study *Journal of clinical microbiology*, 47 (1) p.217–219

Nsofor, C. A., and Iroegbu C. U. (2012). Antibiotic resistance profile of *Escherichia coli* isolated from apparently healthy domestic livestock in south-east Nigeria. *J. Cell An. Biol.* 6:129–135

Okonko IO, Soleye FA, Amusan TA, Ogun AA and Kwaga TA (2009). Incidence of multi drug resistant (MDR) organisms in Abeokuta, southwestern Nigeria. *Global Journal of Pharmacology*, 3: 69-80.

Olonitola Olayeni Stephen, Nicole Fahrenfeld and Amy Pruden (2015) Antibiotic resistance profiles among mesophilic aerobic bacteria in Nigerian chicken litter and associated antibiotic resistance genes *Poultry Science* 00:1–8

Okon, K.O, Shittu AO, Usman H, Adamu, N, Balogun S.T, Adesina, O.O (2013). Epidemiology and Antibiotic Susceptibility Pattern of Methicillin-Resistant *S. aureus* Recovered from Tertiary Hospitals in Northeastern, *Nigeria Journal of Medicine and Medical Sciences*. 4(5) pp. 214-220,

Otalu O JR, Kabir J, Okolocha EC, Umoh VJ.(2011) Multi-drug Resistant Coagulase Positive *S. aureus* from Live and Slaughtered Chickens in Zaria, Nigeria. *International Journal of Poultry Science*.;10 (11):871-5.

Palavecino, E.L. (2014) Clinical, epidemiologic, and laboratory aspects of methicillin-resistant *S. aureus* infections. *Methods Mol. Biol*., 1085, 1–24.].

Pesavento G, Ducci B, Comodo N and Lo Nostro A (2007). Antimicrobial resistance profile of *S. aureus* isolated from raw meat: a research for methicillin resistant *S. aureus* (MRSA). *Food Control*,18: 196-200.

Persoons, D., Van Hoorebeke, S., Hermans, K., Butaye, P., De Kruif, A., Haesebrouck, F and Jeroen, D (2009). Methicillin-resistant *S. aureus* in poultry. *Emerging Infectious Diseases* 15: 452-453

Persoons D, Van Hoorebeke S, Hermans K, Butaye P, De Kruif A, Haesebrouck F, *et al*., (2013) Methicillin-resistant *S. aureus* in poultry. *Emergency Infectious Dis*ease 15 (3):452-3.

Poultry Association of Nigeria (2015) available at www.cpafrica.com/2015/10/15/ nigeria-n220bn-annually -on-eggs-production.

Quinn P.J., Carter M.E., Markey B.K., Carter G.R. (2000). *Staphylococcus species*. *Clinical Veterinary Microbiology Mosby, Edinburgh*. Pp. 118–126

Smith TC, Male MJ, Harper AL, Kroeger JS, Tinkler GP, Moritz ED, Capuano AW, Herwaldt LA, Diekema DJ. (2009) Methicillin-resistant *S. aureus* (MRSA) strain ST398 is present in midwestern U.S. swine and swine workers.*PLoS One.*;4 (1):e4258

Stephen T Odonkor and Kennedy K Addo (2011) Evaluation of Three Methods For Detection of Methicillin Resistant *S. aureus* (MRSA) *International Journal of Biological & Medical Research* 2 (4): 1031 – 1034

Suleiman A, Zaria LT.; Grema, HA. and Ahmadu, P. (2013): Antimicrobial resistant coagulase positive *S. aureus* from chickens in Maiduguri, Nigeria. Sokoto *Journal of Veterinary Sciences*. 11(1): 51-55.

Songer J. G and Post K. W (2005). Veterinary microbiology. 1st Edition Bacterial and fungal agents of animal disease. Illustrated edition. (KW editor), *Elsevier Saunders, St.Louis*, 35-42.

Thaker, H.C., Brahmbhatt, M.N. and Nayak, J.B. (2013) Isolation and identification of *S. aureus* from milk and milk products and their resistance patterns in Anand, Gujarat. *Veterinary World* 6 (1), 10-13.

Van Cleef BA, Monnet DL, Voss A, Krziwanek K, Allerberger F, Struelens M, (2011) Livestock-associated methicillin-resistant *S. aureus* in humans, *Europe. Emergency Infectious Disease*.;17:502–5.

Van Den Broek, I.V.; Van Cleef, B.A.; Haenen, A.; Broens, E.M.; Van Der Wolf, P.J.;Van Den Broek, M.J.; Huijsdens, X.W.; Kluytmans, J.A.; Van De Giessen, A.W. and Tiemersma, E.W (2009). Methicillin-resistant *S. aureus* in people living and working in pig farms. *Epidemiology and Infection137*, 700–708.

Van Duijkeren E, Box AT, Heck ME, Wannet WJ and Fluit AC (2004). Methicillin-resistant Staphylococci isolated from animals. *Veterinary Microbiology*, 103: 91-97.

Waters AE, Contente-Cuomo T, Buchhagen J, Liu CM, Watson L, Pearce K *et al*., (2011). Multidrug resistant *S. aureus* in US meat and poultry.*Clinical Infectious Diseases*,

Winstanley T, Courvalin P (2011). Expert systems in clinical microbiology. *Clinical Microbiology Review.*;24 (3):515-56.