Research Journal of Microbiology

ISSN 1816-4935

Academic Journals Inc.

www.academicjournals.com

Research Journal of Microbiology 6 (4): 356-365, 2011
ISSN 1816-4935 / DOI: 10.3923/jm.2011.356.365

Antibiotic Resistance of Aerobic Mesophilic Bacteria Isolated from Poultry Faeces

E.O. Adeleke and B.O. Omafuvbe
Department of Microbiology, Obafemi Awolowo University, Ile-Ife, Osun State, Nigeria

Corresponding Author: B.O. Omafuvbe, Department of Microbiology, Obafemi Awolowo University, Ile-Ife, Osun State, Nigeria

ABSTRACT

This study was aimed at enumerating, isolating and identifying the aerobic mesophilic bacteria associated with poultry faeces obtained from the Obafemi Awolowo University Teaching and Research Farms, Ile-Ife, Nigeria. The second aim was to study the antibiotic sensitivity patterns of the associated bacteria. The aerobic mesophilic bacteria were enumerated, isolated and identified phenotypically following standard microbiological methods. The antibiotic sensitivity patterns of the isolated bacteria against amoxicillin, augmentin, ceftriaxone, chloramphenicol, ciprofloxacin, erythromycin, gentamycin, nitrofurantoin, ofloxacin, pefloxacin, streptomycin, tetracycline, cotrimoxazole were also determined. The total aerobic count of bacteria isolated ranged from 6.15 to 8.64 log cfu g^{-1} of cockerel faecal sample and 7.18 to 7.67 log cfu g^{-1} of layer fecal sample. Bacteria associated with the faecal samples were identified as *Alcaligenes faecalis, Corynebacterium kutseri, Staphylococcus aureus, Bacillus alvei, Proteus morganii, Corynebacterium ulcerans, Salmonella arizonae, acinetobacter mallei, Staphyloccus* sp. *Escherichia coli, Aeromonas* sp. and *Pseudomonas fluorescens*. *C. kutsceri, C. ulcerans* and *A. faecalis* showed 100% resistance to all the antibiotics tested. Eleven of the isolates showed multiple antibiotics resistance. The quinolones (ofloxacin, ciprofloxacin and pefloxacin) were the most effective of all the antibiotics used. The Multiple Antibiotics Resistance (MAR) index of the bacterial isolates ranged from 0.1 to 1. All the bacterial isolates showed high level (>0.2 MAR index) antibiotics resistance except *Aeromonas* sp. (2D2) which showed a low-level antibiotics resistance. Using two-way clustered analysis, the relatedness of antibiotics resistance pattern was highest in *C. kutsceri* and *C. ulcerans*. The microorganisms isolated from this study are of public health importance and their high level of resistance to commonly used antibiotics in human and veterinary medicine make them a great risk to human and animal.

Key words: Poultry faeces, bacteria isolates, multiple-antibiotics-resistance, quinolones

INTRODUCTION

Poultry is a major fast growing source of meat in the world today, representing a quarter of all the meat produced in the year 2000. The modern poultry industry can produce market ready broiler chickens in less than six weeks. This accomplishment is done through genetic selection, improved feeding and keen health management practices involving usage of antibiotics as therapeutic agents to treat bacterial diseases in intensive farming systems (Apata, 2009). Acquired resistance against frequently used antibiotics has been observed since the introduction of these Antimicrobial agents

in human and veterinary medicine (Smith, 1999). The usage of antibiotics is a major factor in emergence, selection and dissemination of antibiotic resistant microorganisms in both veterinary and human (Tollefson and Flynn, 2002). The rise in antibiotics resistance has been reported in the past two decade (Kapil, 2004). Antibiotic resistance still remains a global problem today. In intensively reared food animals, antibiotics are administered for therapeutic purpose and as Antimicrobial growth promoters (AMGPs) to the whole flock rather than individuals (Van-den-Boogaard and Stobberingh, 1999). Hence, the antibiotic selection pressure for resistance in bacteria in poultry is high and consequently their faecal flora contains a relatively high proportion of resistant bacteria (Van den Bogaard *et al*., 2001). Resistant strains from the poultry gut readily soil poultry carcasses when they are being sacrificed and as a result poultry meats are often contaminated with multi resistant bacteria. Therefore, resistant faecal coliforms from poultry can infect humans both directly and via food, colonizing the human intestinal tract and also contributing resistant genes to human endogenous flora (Van den Bogaard *et al*., 2001). Gene transfer occurs majorly *in vivo* between gastrointestinal tract bacteria and between gastrointestinal tract bacteria and pathogenic bacteria, as identical resistance genes are present in diverse bacterial species from different hosts (Scott, 2002). In light of this, there is probability that most pathogenic bacteria that threaten human health may soon be resistant to all known antibiotics (Mathur and Singh, 2005). Certain antibiotics, however are critical to human infections caused by multidrug resistant pathogens, or because alternative therapies are less effective or are associated with side effects (Akond *et al*., 2008). The determination of the effectiveness of Antimicrobial agents against specific pathogens-either human or animal source- is essential for proper therapy (Prescott *et al*., 2005).

In Nigeria, there has been an increase in poultry production since government regulation on the importation of poultry meat. This in turn has led to increase in the poultry manure production especially in urban areas (Ayeni, 2011). Poultry feces are the excretory product released as a result of digestion of food taken in by poultry birds (Adegunloye, 2006). Poultry industries play a prominent role in everyday production of poultry manure. A typical broiler and layer have been reported to produce estimated manure of about 0.17 ft^3/finished animal (f-a) and 0.0031 ft^3/day animal (d-a), respectively (ASAE, 2005). Poultry manure, is an inevitable byproduct of the poultry industries that is very useful as a source of organic matter and fertilizer for crop and pasture production (Ogejo, 2008).

The aim of the present study was to identify the aerobic mesophilic bacteria associated with poultry (cockerels and layers) faeces and to study their antibiotic resistant patterns for possible recommendation on the antibiotic of choice in the poultry industries.

MATERIALS AND METHODS

Studies on the antibiotic resistance of aerobic mesophilic bacteria isolated from poultry faeces obtained from Obafemi Awolowo University Teaching and Research farms, Ile-Ife, Nigeria was carried out in Microbiology Laboratory, Department of Microbiology, Obafemi Awolowo University, Ile-Ife, Nigeria between 1st February and 27th August, 2010.

Samples: Fifteen samples each of fresh poultry faecal droppings from cockerels and layers were obtained between the hours of 7 and 8 a.m. over a period of five weeks (1st February to 8th

March, 2010) from the poultry unit of the Obafemi Awolowo University Teaching and Research Farms, Ile-Ife, Nigeria. The samples were collected aseptically in sterile McCartney bottles and transported to the laboratory within 30 min for analysis.

Isolation and Identification of bacteria strains: Ten grams of faeces sample were homogenized with 90 mL of maximum recovery diluent (MRD, Oxoid) to obtain a 1:10 dilution. Successive decimal dilutions were carried out with sterile MRD. Aliquots (1000 μL) of appropriately diluted sample homogenates were pour-plated in duplicate using nutrient agar. The agar plates were allowed to set and incubated at 37°C for up to 48 h. The colony forming units of the bacteria on the plates were enumerated and representatives of the different colonies were selected according to their morphological characteristics and purified by successive sub culturing on nutrient agar and identified phenotipically based on standard methods (Harrigan and McCance, 1976; Buchannan and Gibbons, 1985).

Testing for resistance to antibiotics: The bacterial isolates were tested for resistance to 14 antibiotics produced by FONDISC (Fondoz Laboratories Ltd., Nigeria). These were: augmentin (30 μg), ceftriazone (30 μg), nitrofurantoin (200 μg), gentamycin (10 μg), cotrimoxazole (25 μg), ofloxacin (5 μg), amoxicillin (25 μg), ciprofloxacin (10 μg), tetracycline (30 μg), pefloxacin (5 μg), ofloxacin (5 μg), streptomycin (10 μg), chloramphenicol (30 μg) and erythromycin (5 μg). This testing was performed using the standard disc diffusion method (Clinical Laboratory and Standards Institute, 2006). The antibiotics susceptibility pattern of the isolates was interpreted using Progressive Diagnostics Manufacturers (PDM) Interpretative Chart.

Multiple antibiotics resistance indexing of isolates: The Multiple Antibiotic Resistance (MAR) index is defined as a/b where 'a' represents the number of antibiotics to which the particular isolate is resistant and 'b' the number of antibiotics to which the isolate is exposed (Krumperman, 1983). MAR index values higher than 0.2 are considered to have originated from high-risk sources where antibiotics are often used. MAR index values of less than or equal to 0.2 indicates a strain originated from sources where antibiotics are seldom or never used.

Statistical analysis: A two-way clustered analysis of multi-variance was used to estimate overall similarities of the bacterial resistance using their zones of inhibition. Correlation method of the similarity measure was used on the Paleontological statistics software package for education and data analysis (Hammer *et al*., 2001).

RESULTS

The total aerobic mesophilic counts ranged from 7.16±0.10 to 7.67±0.05 log cfu g^{-1} and 6.14±0.09 to 8.64±0.04 cfu g^{-1} in layers and cockerel faeces, respectively (Table 1). A total of 15 strains of bacterial belonging to the genera *Alcaligenes, Corynebacterium, Staphylococcus, Bacillus, Proteus, Pseudomonas, Escherichia, Acinetobacter* and *Aeromonas* were isolated and characterized phenotipically. The occurrence pattern of the isolates in the faeces samples is shown in Table 1.

The antibiotic susceptibility pattern and Multiple Antibiotics Resistance (MAR) index of both Gram-positive and Gram-negative bacteria isolates from poultry feces are shown in Table 2 and 3, respectively. The MAR index ranged from 0.6 to 1.0 and 0.1 to 1.0 for gram positive and gram negative bacteria isolates respectively. All the bacterial isolates showed high level MAR index (>0.2) except a strain of *Aeromonas* sp. which had a value of 0.1 (Table 3). The percentage antibiotic susceptibility pattern of the Gram positive bacteria isolates showed 100% resistance to streptomycin,

Table 1: Total aerobic mesophilic bacteria count and their occurrence in poultry faeces

Faecal sample	Bacteria count (log cfu g^{-1})	Occurrence of bacteria isolates
Cockerel faeces	8.27±0.07	*A. faecalis* (C2), *A. faecalis* (C9), *P. morganii*, *A. mallei* (C3), *Staphylococcus* spp., *A. faecalis* (1D4)
Layer faeces	7.16±0.10	*P. morganii*, *C. ulcerans*, *A. mallei* (C3), *A. mallei* (1D2), *E. coli*
Cockerel faeces	8.64±0.04	*C. kutsceri*, *S. aureus*, *S. arizonae*
Layer faeces	7.36±0.07	*S. aureus*, *P. morganii*, *S. arizonae*, *A. mallei* (1D2), *Staphylococcus* sp. *P. fluorescens*
Cockerel faeces	7.02±0.15	*A. faecalis* (C2), *C. kutsceri*, *S. aureus*, *B. alvei*, *A. faecalis* (C9), *A. mallei* (C3), *Staphylococcus* sp. *A. faecalis* (1D4)
Layer faeces	7.29±0.06	*C. kutsceri*, *B. alvei*, *C. ulcerans*, *S. arizonae*, *Staphylococcus* sp.
Cockerel faeces	6.14±0.09	*A. faecalis* (C2), *S. aureus*, *A. faecalis* (C9), *C. ulcerans*, *A. mallei* (C3), *A. faecalis* (1D4), *Aeromonas* sp. (2D2), *Aeromonas* sp. (2D5)
Layer faeces	7.62±0.03	*A. faecalis* (C2), *B. alvei*, *P. morganii*, *C. ulcerans*, *S. arizonae*, *Aeromonas* sp. (2D2), *Aeromonas* spp. (2D5)
Cockerel faeces	7.90±0.02	*A. faecalis* (C2), *S. aureus*, *Staphylococcus* sp., *P. fluorescens*, *Aeromonas* sp. (2D5)
Layer faeces	7.67±0.05	*A. faecalis* (C2), *C. kutsceri*, *B. alvei*, *P. morganii*, *S. arizonae*, *A. mallei* (1D2), *A. faecalis* (1D4), *E. coli*

Values represent the mean of three determinations±standard deviation, Isolate codes are in parenthesis

Table 2: Antibiotic susceptibility pattern of gram positive bacteria isolated from poultry faeces

ISOLATE*	AMX	OFL	STR	CHL	CEF	GEN	PEF	COT	CPX	ERY	MAR INDEX
Corynebacterium kutsceri (C5)	R	R	R	R	R	R	R	R	R	R	1.0
Staphylococcus aureus (C7)	I	S	R	R	R	R	I	R	S	R	0.6
Staphylococcus spp. (1D3)	R	I	R	R	R	R	I	R	I	R	0.7
Corynebacterium ulcerans (L4)	R	R	R	R	R	R	R	R	R	R	1.0
Bacillus alvei (C8)	R	S	R	R	R	R	S	S	S	R	0.6

Antibiotics	Susceptible	Intermediate	Resistant
Amoxicillin (AMX)	≥ 28	22-27	≤ 21
Augmentin (AUG)	≥ 26	23-25	≤ 22
Ceftriaxone (CEF)	≥ 16	13-15	≤ 12
Chloramphenicol (CHL)	≥ 24	21-23	≤ 20
Ciprofloxacin(CPX)	≥ 22	17-21	≤ 16
Erythromycin(ERY)	≥ 26	21-25	≤ 22
Gentamycin (GEN)	≥ 20	19	≤ 18
Nitrofurantoin(NIT)	≥ 18	14-17	≤ 13
Ofloxacin (OFL)	≥ 22	16-21	≤ 15
Pefloxacin (PEF)	≥ 22	16-21	≤ 15
Streptomycin (STR)	≥ 26	23-25	≤ 22
Tetracycline (TET)	≥ 26	23-25	≤ 22
Cotrimoxazole(COT)	≥ 26	23-25	≤ 22

R = Resistant, S = Susceptible, I = Intermediate, MAR INDEX = Multiple Antibiotic Resistance Index; *Isolate code in parenthesis

Table 3: Antibiotic susceptibility pattern of gram negative bacteria isolated from poultry faeces

ISOLATE*	AUG	CRO	NIT	GEN	COT	OFL	AMX	CPX	TET	PFX	MAR INDEX
Escherichia coli (1D7)	R	R	R	R	R	S	R	S	R	I	0.7
Alcaligenes faecalis (C9)	R	R	R	R	R	R	R	R	R	R	1.0
Proteus morganii (L3)	R	R	R	R	R	R	R	R	R	I	0.9
Salmonella arizonae (L8)	R	I	R	R	R	S	R	S	R	S	0.6
Acinetobater mallei (C3)	R	R	R	R	R	S	R	I	R	R	0.8
Acinetobacter mallei (1D2)	R	R	R	R	R	S	R	S	S	S	0.6
Alcaligenes faecalis (1D4)	R	R	R	R	S	S	S	S	S	S	0.4
Alcaligenes faecalis (C2)	R	S	R	R	R	I	R	R	R	I	0.7
Aeromonas sp. (2D2)	S	S	R	S	S	S	S	S	S	S	0.1
Pseudomonas fluorescens (2D4)	R	R	R	R	R	I	R	S	R	I	0.7
Aeromonas sp. (2D5)	R	R	I	R	R	S	S	S	S	S	0.4

R = Resistant; S = Susceptible; I = Intermediate; *Isolate code in parenthesis

Table 4: Percentage antibiotics susceptibility pattern of gram positive bacteria isolated from poultry faeces

Antibiotics (%)	AMX	OFL	STR	CHL	CEF	GEN	PEF	COT	CPX	ERY
Resistant	80	40	100	100	100	100	40	80	40	100
Intermediate	20	20	0	0	0	0	40	0	20	0
Sensitive	0	40	0	0	0	0	20	20	40	0

AMX = AMOXYCILLIN (25 µg), OFL = OFLOXACIN (5 µg), STR = STREPTOMYCIN (10 µg) CHL = CHLORAMPHENICOL (30 µg), CEF = CEFTRIAZONE (30 µg), GEN = GENTAMYCIN (10 µg) PEF = PEFLOXACIN (5 µg), COT = COTRIMOXAZOLE (25 µg), CPX = CIPROFLOXACIN (10 µg), ERY = ERYTHROMYCIN (5 µg)

Table 5: Percentage antibiotics susceptibility pattern of gram negative bacteria isolated from poultry faeces

Antibiotics (%)	AUG	CRO	NIT	GEN	COT	OFL	AMX	CPX	TET	PFX
Resistant	91	73	91	91	82	18	73	27	64	18
Intermediate	0	9	9	0	0	18	0	9	0	36
Sensitive	9	18	0	9	18	64	27	64	36	46

AUG = AUGMENTIN (30 µg), CRO = CEFTRIAZONE (30 µg), NIT = NITROFURANTOIN (200 µg) GEN = GENTAMYCIN (10 µg), COT = COTRIMOXAZOLE (25 µg), OFL = OFLOXACIN (5 µg) AMX = AMOXICILLIN (25 µg), CPX = CIPROFLOXACIN (10 µg), TET = TETRACYCLINE (30 µg), PFX = PEFLOXACIN (5 µg)

chloramphenicol, ceftriazone, gentamycin and erythromycin; 80% resistance to amoxicillin and cotrimoxazole and 40% resistance to ofloxacin, pefloxacin and ciprofloxacin (Table 4). While the Gram negative bacterial isolates showed above 70% resistance to augmentin, nitrofurantoin, ceftriazone, gentamycin cotrimoxazole and amoxicillin; above 60% resistance to tetracycline; below 30% resistance to ciprofloxacin and below 20% resistance to ofloxacin and pefloxacin (Table 5). The two-way clustered analysis of the Gram positive bacteria isolates showed that the antibiotics susceptibility pattern of *Staphylococcus* sp. (4) and *Bacillus alvei* (6) were the most related followed by *Corynebacterium kutsceri* (2) and *Corynebacterium ulcerans* (5) (Fig. 1). The two-way clustered analysis of the Gram negative bacteria isolates deduced that the antibiotic sensitivity pattern of *Acinetobacter mallei* (6) and *Pseudomonas fluorescens* (11) were the most related (Fig. 2).

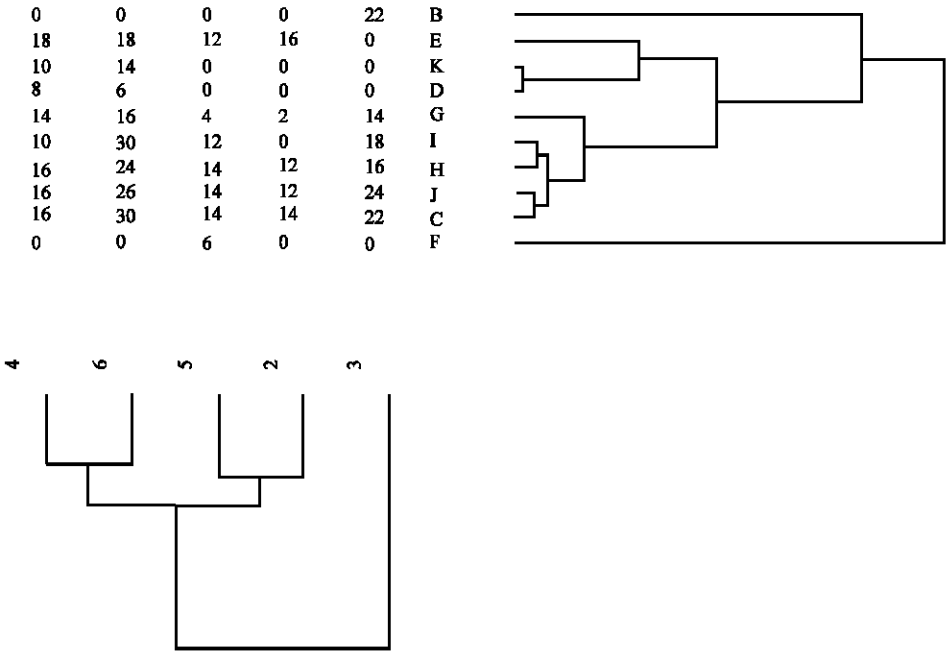

Fig. 1: Two-way clustered analysis of the gram positive bacteria isolated from poultry faeces. B = Amoxicillin; E = Chloramphenicol; K = Erythromycin; D = Streptomycin; G = Gentamycin; I = Cotrimoxazole; H = Pefloxacin; J = Ciprofloxacin; C = Ofloxacin; F = Ceftriazone; 2 = *Corynebacterium kutsceri*; 3 = *Staphylococcus aureus*; 4 = *Staphylococcus* sp.; 5 = *Corynebacterium ulcerans*; 6= *Bacillus alvei*

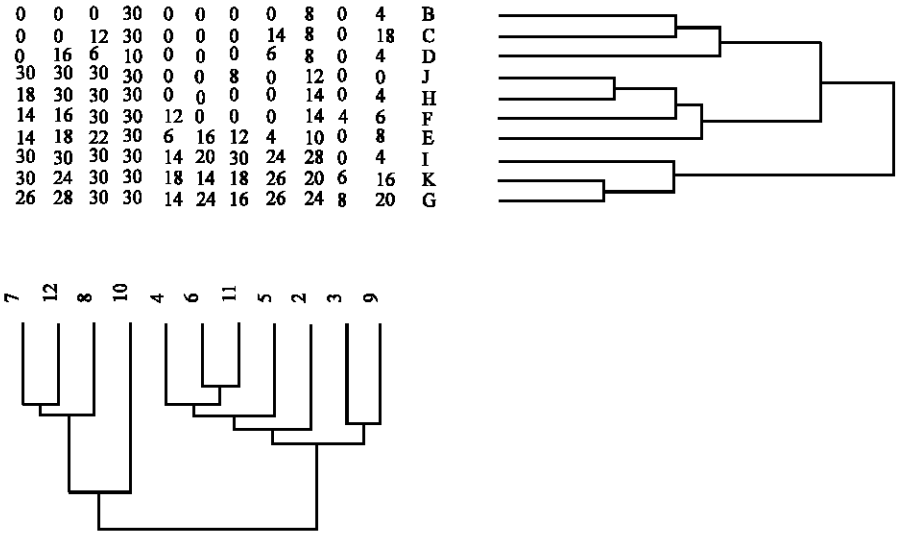

Fig. 2: Two-way clustered analysis of the gram negative bacteria isolated from poultry faeces. B = Augmentin; C = Ceftriazone; K = Pefloxacin; D = Nitrofurantoin; G= Ofloxacin; I = Ciprofloxacin; H = Amoxycillin; J = Tetracycline; E = Gentamycin; F = Cotrimoxazole 2 = *Escherichia coli*; 3 = *Acaligenes faecalis*; 4 = *Proteus morganii*; 5 = *Salmonella arizonae*; 6 = *Acinetobacter mallei*; 7 = *Acinetobacter mallei*; 8 = *Acaligenes faecalis*; 9 = *Acaligenes faecalis*; 10 = *Aeromonas* sp.; 11 = *Pseudomonas fluorescens*; 12 = *Aeromonass* sp.

DISCUSSION

The cockerel faeces in this study contained slightly higher bacterial population than those of layer faeces, This seems to correspond with the reports of Adegunloye (2006). The average bacterial load (8.15 log cfu g^{-1}) of cockerel faeces in this study is slightly higher than values previously reported for poultry faeces in Nigeria (Adegunloye, 2006). This might be attributed to constant contact between feed, poultry fowl and faecal droppings (Vellinga and Van Loock, 2002). The bacteria isolated from the faecal samples were identified as *Alcaligenes faecalis*, *Corynebacterium kutsceri*, *Staphylococcus aureus*, *Bacillus alvei*, *Proteus morganii*, *Corynebacterium ulcerans*, *Salmonella arizonae*, *Acinetobacter mallei*, *Staphylococcus* sp. *Escherichia coli*, *Aeromonas* sp. and *Pseudomonas fluorescens*. Presant finding on the bacterial composition of poultry faeces is different from those of Adegunloye (2006) who only reported *Staphylococcus aureus*, *Staph. epidermidis*, *Bacillus cereus* and *E. coli*. Our identification was however based on phenotypic characters, authentic identity of species, should be confirmed by molecular identification. Many of the isolated bacteria are normal flora of intestinal tract of poultry (Esposito and Leone, 2007) while a few have been implicated in poultry diseases (Islam *et al*., 2003; Simon, 2005). The distribution of bacterial isolates between the layers and cockerel faecal samples was similar in this study. The observation of more Gram negative bacteria (11 strains) than Gram-positive bacteria (5 strains) in this study is in agreement with the findings of Chopra and Roberts (2001).

In this study, all the bacteria isolates showed high level of antibiotics resistance except a strain of *Aeromonas* sp. This result is in agreement with Cloud *et al*. (1985) and Muhammad *et al*. (2010) who reported that the abuse and misuse of antimicrobial agents for growth promotion and prevention of diseases has impressed a selective pressure that causes discovery of more resistance bacteria. This is true with the bacteria associated with poultry faeces in this study. Hence, the antibiotic selection pressure for resistance by bacteria in poultry is high and as a result their fecal flora contains high proportion of resistant bacteria. Salehi and Bonab (2006) reported that the resistance of bacteria to existing antimicrobial agents is widespread and of great concern to poultry veterinarians. The use of antimicrobial in animal feed can also lead to selection of antimicrobial resistant zoonotic enteric pathogens, which can be transferred to human through the consumption of contaminated food, or by direct animal contact.

According to Nandi *et al*. (2004) Gram-positive bacteria especially *Corynebacterium* sp. that has been found to be associated with poultry litter serves as a major reservoir of class 1 integrons (in-1). *Corynebacterium kutsceri* and *Corynebacterium ulcerans* showed 100% resistance to all the antibiotics used in this study. *C. ulcerans* is a veterinary pathogen, which has been implicated in pharyngeal infection mimicking classical diphtheria in humans (Gubler *et al*., 1990). *Alcaligenes* are apparently saprophytic inhabitants of the intestinal tract of vertebrates, which are involved in decomposition and mineralization processes of poultry products (Holt, 1981). Three strains of *Alcaligenes faecalis* (C9, C2, 1D4) showing resistance of 100, 70 and 40%, respectively to the antibiotics used were encountered in this study. The percentage of antibiotics susceptibility in *Salmonella* sp. isolated in this study corresponds with reports on *Salmonella* strains isolated from broiler flocks in Canada and India (Suresh *et al*., 2000).

All the isolates except *Aeromonas* sp. (2D2) showed MAR index > 0.2, inferring that they have arisen from high-risk sources of contamination where antibiotics are often used. This is an indication of a high presence of antibiotics selective pressure, which agrees with the report of Suresh *et al*. (2000). The MAR pattern of *E. coli* and *P. fluorescens* showed the same multiple antibiotic resistance pattern hence they have the identical MAR index (0.7), this is suggestive that

both strains have a common origin (Kasper *et al*., 1990). Similar pattern of MAR was also observed for the *Corynebacterium* species. The development of multiple antibiotics resistance may be as a result of transfer of R factor (borne on plasmids) and *E. coli* are noted to carry multiple plasmids which can carry any number of multiple resistant genes (Sumathi *et al*., 2008).

The two-way clustered analysis of the Gram positive bacterial isolates showed that the antibiotics susceptibility pattern of *Staphylococcus* sp. and *Bacillus alvei* are the most related followed by *Corynebacterium kutsceri* and *Corynebacterium ulcerans*. This establishes the observed MAR index (1.0) for *Corynebacterium* sp. in this study and it is in agreement with previous report by Kasper *et al*. (1990). The relatedness of *Proteus morganii* and *Acinetobacter mallei* to *Salmonella arizonae* and *Acaligenes faecalis* is close but distantly followed by their relatedness to that of *Staphylococcus aureus*. Similar relatedness is observed in the resistance pattern of Erythromycin to Streptomycin and Ciprofloxacin to Ofloxacin. This is also used to show the relatedness in Gram negative bacterial isolates. From the analysis it can be deduced that two-way clustered analysis is a method that can be used as a taxonomic tool.

CONCLUSION

The results obtained in this study revealed that poultry (cockerel and layer) faeces contain bacteria with multiple antibiotic resistance patterns suggestive of possible horizontal gene transfer among non related bacterial isolates. The multiple antibiotic resistance index of the bacteria isolates suggest that they have arisen from sources of high level of antibiotics selective pressure resulting from non-specific, misuse or abuse of antibiotics. The cross infection of these bacteria in humans or as secondary pathogen might be of serious health concern. Therefore, there is need for a national policy which will take into cognizance the rational use of antibiotics and standard antibiotics test before veterinary antibiotics therapy is administered in poultry industry.

REFERENCES

ASAE, 2005. Manure production and characteristics. ASAE Standard D384.2 MAR2005. American Society of Agricultural Engineers-The Society for Engineering in Agricultural, Food, and Biological Systems 2950 Niles Rd., St. Joseph, MI 49085-9659, USA.

Adegunloye, D.V., 2006. Microorganisms associated with poultry faeces. J. Food Agric. Environ., 4: 41-42.

Akond, M.A., S. Alam, S.M.R. Hasan, S.N. Uddin and M. Shirin, 2008. Antibiotic resistance of *Vibrio cholera* from poultry sources of Dhaka, Bangladesh. Adv. Biol. Res., 2: 60-67.

Apata, D.F., 2009. Antibiotic resistance in poultry. Int. J. Poult. Sci., 8: 404-408.

Ayeni, L.S., 2011. Integrated plant nutrition management: A panacea for sustainable crop production in Nigeria. Int. J. Soil Sci., 6: 19-24.

Buchannan, R.E. and N.E. Gibbons, 1985. Bergey's Manual of Determinative Bacteriology. Vol. 1. 9th Edn., Williams and Wilkins, Baltimore, USA.

Chopra, I. and M. Roberts, 2001. Tetracycline antibiotcs: Mode of action, applications, molecular biology and epidemiology of bacterial resistance. Microbiol. Mol. Biol. Rev., 65: 232-260.

Clinical and Laboratory Standards Institute, 2006. Performance standards for antimicrobial susceptibility testing; sixteenth informational supplement. M100-S16, Vol. 26 No. 3, pp: 1-37. http://www.sld.cu/galerias/pdf/servicios/medicamentos/nccls.jan2006parte01.pdf.

Cloud, S.S., J.K. Rosenberger, P.A. Fries, R.A. Wilson and E.M. Odor, 1985. *In vitro* and *in vivo* characterization of avian *Escherichia coli* Serotypes, metabolic activity and antibiotic sensitivity. Avian Dis., 29: 1084-1093.

Esposito, S. and S. Leone, 2007. Antimicrobial treatment for Intensive Care Unit (ICU) infections including the role of the infectious diseases specialist. Int. J. Antimicrob. Agents, 29: 494-500.

Gubler, J.G., J. Wust and T. Krech, 1990. Classical pseudo-membranous diphtheria caused by *Corynebacterium ulcerans*. Schweiz Med. Wochenschr, 120: 1812-1816.

Hammer, O., D.A.T. Harper and P.D. Ryan, 2001. Paleontological statistics software package for education and data analysis. Palaeontologia Electronica, 4: 9-9.

Harrigan, W.F. and M.E. McCance, 1976. Laboratory Methods in Food and Dairy Microbiology. 1st Edn., Academic Press, London, pp: 25-29.

Holt, J.G., 1981. The Shorter Bergey's Manual of Determinative Bacteriology. 8th Edn., Williams and Wilkins Co., Baltimore, USA.

Islam, M.R., B.C. Das, K. Hossain, N.S. Lucky and M.G. Mostafa, 2003. A study on the occurrence of poultry diseases in Sylhet region of Bangladesh. Int. J. Poult. Sci., 2: 354-356.

Kapil, A., 2004. The challenge of antimicrobial resistance: Need to contemplate. Indian J. Med. Res., 121: 83-91.

Kasper, C.W, J.L. Burgess, I.T. Knight and R.R. Colwell, 1990. Antibiotic indexing of *Escherichia coli* to identify high risk sources of faecal contamination of foods. Applied Environ. Microbiol., 46: 165-170.

Krumperman, P.H., 1983. Multiple antibiotic resistance indexing of *Escherichia coli* to indentify high-risk sources of fecal contamination of foods. Applied Environ. Microbiol., 46: 165-170.

Mathur, S. and R. Singh, 2005. Antibiotic resistance in food lactic acid bacteria-A review. Int. J. Food Microbiol., 105: 281-295.

Muhammad, M., L.U. Muhammad, A.G. Ambali and A.U. Mani, 2010. A survey of early chick mortality on small-scale poultry farms in jos, central Nigeria. Int. J. Poult. Sci., 9: 446-449.

Nandi, S., J.J. Maurer, C. Hofacre and A.O. Summers, 2004. Gram positive bacteria are a major reservoir of class 1 antibiotic resistance integrons in poultry litter. Proc. Natl. Acad. Sci., 101: 7118-7122.

Ogejo, J.A., 2008. Manure production and characteristics. Animal Manure Management Home and Cooperative Extension System. http://www.extension.org/pages /Manure-Production-and-Characteristics.

Prescott, L.M., J.P. Harley and D.A. Klein, 2005. Microbiology. 6th Edn., McGraw-Hill Co., New York, London.

Salehi, T.Z. and S.F. Bonab, 2006. Antibiotics susceptibility pattern of *Escheichia coli* strains isolated from chickens with colisepticemia in Tabriz Province, Iran. Int. J. Poult. Sci., 5: 677-684.

Scott, K.P., 2002. The role of conjugative transposons in spreading antibiotic resistance between bacteria that inhabit the gastrointestinal tract. Cell Mol. Life Sci., 59: 2071-2082.

Simon, M.S., 2005. Handbook on Poultry Diseases. 2nd Edn., American Soybean Association, USA.

Smith, D.W., 1999. Decreased antimicrobial resistance after changes in antibiotics use. Pharmacotherapy, 19: 129-132.

Sumathi, B.R., R.G. Amitha and G. Krishnappa, 2008. Antibiogram profile based dendogram analysis of *Escherichia coli* serotypes isolated from bovine mastitis. Vet. World, 1: 37-39.

Suresh, T., D. Srinivasan, A.A.M. Hatha and P. Lakshmanaperumalsamy, 2000. The incidence, antibiotics resistance and survival of *Salmonella* and *Escherichia coli* isolated from broiler chicken retail outlets. Microbes Environ., 15: 173-181.

Tollefson, L. and W.T. Flynn, 2002. Impact of antimicrobial resistance on regulatory policies in veterinary medicine: Status report. AAPS Pharmsci., 4: 150-159.

Van den Bogaard, A.E., N. London, C. Driessen and E.E. Stobberingh, 2001. Antibiotic resistance of faecal *Escherichia coli* in poultry, poultry farmers and poultry slaughterers. J. Antimicrob. Chemother., 47: 763-771.

Van der Boogaard, A.E. and E.E. Stobberingh, 1999. Antibiotics usage in animals-Impact on bacterial resistance and public health. Drugs, 58: 589-607.

Vellinga, A. and F. Van-Loock, 2002. The dioxin crisis as experiment to determine poultry-related *Campylobacter enteritis*. Emerg. Inf. Dis., 8: 19-22.