ijlpr 2022; doi 10.22376/ijpbs/lpr.2022.12.1.L105-111

International Journal of Life science and Pharma Research
ISSN 2250-0480

Occurrence Of Multi Drug Resistant Bacteria From Raw Chicken Meat Of South India Retail Markets.

Sharmiladevi Natarajan*[A], Balaji Nagarajan[A], Ramar DineshKumar[A], Mythili Ravichandran[B], Prathaban Muniasamy[C], Varadharaju Chandrasekar[D]

[A] Department of Microbiology, Karpagam Academy of Higher Education, Coimbatore, Tamil Nadu, India
[B] Department of Microbiology, K S Rangasamy College of Arts and Science, Tiruchengode, Tamil Nadu, India
[C]department of Microbiology, Pondicherry University, Puducherry.
[D] Department of Microbiology, Bioline Laboratory and Research Institute, Coimbatore, Tamil Nadu, India

Abstract: Antibiotic resistance and its dissemination from poultry food to human beings further sequentially to the environment is becoming an ever-increasing distress worldwide. Nowadays a small ratio of meat sample from the rural area itself may be found positive with Multi Drug Resistant (MDR) isolates. This study is aimed to enumerate the prevalence of antibiotic resistant isolates in raw chicken meat collected from retail shops (fresh meat) and supermarkets (frozen meat) situated in different areas of Coimbatore, South India. Also, the study focused to exhibit the severity of consuming poultry food with the antibiotic residues unknowingly. Resistant isolates from raw chicken meats was collected during the period of December 2020 to February 2021 and screened for resistant attributes against antibiotics as per clinical settings using VITEK® 2 Compact. The identified MDR isolates was submitted to the National Database of Antibiotic Resistant Organisms (NDARO). Out of 22 chicken meat samples collected, a total of 50 pathogens were isolated. Among them, 2 frozen chicken meats showed no bacterial growth, and other chicken meat samples was found to have resistant bacteria like MDR *E.coli, Salmonella* spp. and *Staphylococcus spp*. This study confirmed the presence of MDR isolates in raw chicken meat and it is the first study from South India to evidence the presence of MDR in raw chicken samples. This study also suggests that antimicrobial stewardship programs and monitoring practices needs more concentration in the veterinary antibiotics usage in poultry and Livestock. Hence it is an alarm of poultry stock which leads to risk on public health and ecological system of South India.

Keywords: Multi drug resistant; Poultry food; antibiotic resistance; Antibiotic Susceptibility Test; NDARO

***Corresponding Author**

SharmilaDevi Natarajan , Department of Microbiology, Karpagam Academy of Higher Education, Coimbatore, Tamil Nadu, India

Received On **22 April, 2021**
Revised On **24 November, 2021**
Accepted On **18 December, 2021**
Published On **7 January, 2022**

Funding This research did not receive any specific grant from any funding agencies in the public, commercial or not for profit sectors.

Citation Sharmiladevi Natarajan , Balaji Nagarajan, Ramar DineshKumar, Mythili Ravichandran, Prathaban Muniasamy , Varadharaju Chandrasekar , Occurrence Of Multi Drug Resistant Bacteria From Raw Chicken Meat Of South India Retail Markets..(2022).Int. J. Life Sci. Pharma Res.12(1), L105-111 http://dx.doi.org/10.22376/ijpbs/lpr.2022.12.1.L105-111

1. INTRODUCTION

The uncontrolled usage of antimicrobials in poultry and livestock for the purpose of therapeutic and preventive practices leads to the conversion of poultry food products as reservoirs of MDR microorganisms. The main reason for irresistible antibiotics usage in poultry is for the purpose of market demand and industrialism rather than a quality food to consumers.[1] Nowadays, the rapid growth of antibiotic resistance in the environment, poultry, livestock and clinical settings is a serious concern worldwide and has become a challenging force to the research community.[2] The main concern about chicken is that it is a well-known, preferable food for the majority of the population worldwide.[3] Without any discrimination, both rural and urban sector people are using excessive level of antibiotics in the poultry food either knowingly or unknowingly. It was repeatedly reported in many findings that gram positive and gram negative bacteria like *Escherichia coli, Klebsiella pneumoniae, Pseudomonas spp., Enterobacter spp., Staphylococcus aureus, Streptococcus pneumoniae, Salmonella* spp., become MDR through gene dissemination and in turn evolving among humans and animals through mishandling and unhygienic practices.[3-5] Apart from this, spread of the MDR *Salmonella* spp. among human beings through poultry products is highly evidenced in literature. Moreover, it has been documented as a leading cause of food borne infection since 1997.[6,7] This public health study intends to exhibit current status of resistant as well as MDR bacteria prevalence in raw chicken meat collected from retail shops of South India. This study points out the evolving AMR bacteria as a nightmare and it's spread through the food, cycle and environment.

2. MATERIALS AND METHODS

2.1. *Study plan, Area and Sampling*

A poultry food investigational study was carried out in Coimbatore, South India from December 2020 to February 2021 with the collaboration of the Department of Microbiology, Karpagam Academy of Higher Education and Bioline Laboratory (Coimbatore, South India). A total of 50 raw chicken meat samples were collected from retail shops from different populated areas of the selected region. Randomly fresh and frozen raw chicken samples were collected aseptically from five different populated areas of Coimbatore. The meat samples were transported to the Department of Microbiology, Karpagam Academy of Higher Education in sterile condition in a thermocol box for further microbial analysis.

2.2 *Bacterial Isolation and Characterization*

25 grams of raw chicken meat samples were thoroughly homogenized with 250 ml of 1% buffered peptone water for 2 minutes to get fine suspension. 0.1 ml of homogenized sample was transferred aseptically on to each of the selective media including Blood Agar (Hi-Media, India), MacConkey Agar (Hi-Media, India), and Mannitol Salt Agar (Hi-Media, India). The plates were incubated for 24 hours at 37◦C. The isolated colonies were identified using standard morphological characteristics, staining techniques, and biochemical properties . Representative isolates were further identified by VITEK® 2 Compact (Software version 6.01, bioMerieux, France) for further confirmation.[1]

2.3 *Antimicrobial Susceptibility Test*

The antimicrobial susceptibility test for the bacterial isolates was performed by using Kirby Bauer Disk diffusion method as per CLSI guidelines. The antibiogram included the following panel of respective antibiotics Penicillin (Ampicillin, Benzylpenicillin, Oxacillin), Aminoglycosides (Amikacin, Gentamicin), Cephalosporins (Cefoperazone/Sulbactam, Cefepime, Ceftazidime, Cefuroxime, Cefuroxime Axetil, Ceftriaxone), Tetracyclines (Tigecycline, Minocycline, Sulfonamide, Trimethoprim/Sulfamethoxazole), Quinolones (Nalidixic Acid, Levofloxacin, Ciprofloxacin), Sulfonamides, Nitrofurantoin, Beta lactam combinations (Piperacillin/Tazobactam, Ticarcillin/ Clavulanic Acid, Amoxicillin/Clavulanic Acid), Cefoxitin Screen, Linezolid, Daptomycin, Teicoplanin, Vancomycin, Rifampicin, Monobactam **(**Aztreonam**),** Carbapenems (Imipenem, Meropenem, Ertapenem, Doripenem), Polymyxins (Colistin), Macrolides (Erythromycin), Clindamycin, Inducible Clindamycin Resistance, Daptomycin, Vancomycin, Rifampicin, Linezolid which are selectively tested against gram positive and gram negative pathogens using VITEK® 2 Compact (Software version 6.01, bioMerieux, France). The *E. coli* isolate ATCC 25922 and *S. aureus* isolate ATCC 25923 were used as reference organisms for quality control of antimicrobial susceptibility testing (AST). The data was analyzed and the results were interpreted using percentage analysis.

2.4 *AST data submission to NCBI*

The confirmed MDR isolates in this study with their phenotypic Antimicrobial Susceptibility Test – antibiogram data was submitted to National Database of Antibiotic Resistant Organisms (NDARO) for the purpose of Antimicrobial Resistance data surveillance and stewardship of resistant pathogens present in the poultry products.

3. STATISTICAL ANALYSIS

AST data was analysed to determine the resistance attributes of isolates using percentage analysis. One-way ANOVA was performed using SPSS version 16 for comparison of different groups. The values represented as mean and standard deviations (mean ± SD). P <0.05 was considered statistically significant in this study.

4. RESULTS

The present study included a total of 32 raw chicken meat samples from different geographical regions of Coimbatore, South India. Among them, 30 samples were fresh chicken meat from retail shops and remaining two samples were frozen chicken meat samples from supermarkets. All fresh raw chicken meat samples from retail shops showed that bacterial growth, except the frozen samples collected from supermarkets, where they did not show any bacterial growth. From the 30 fresh raw chicken meat samples, 50 isolates were found including gram positive and gram-negative bacteria. Out of that 50 isolates, *E.coli* was the most predominant (28%). Subsequently, other commonly found bacteria were *Salmonella spp.* (18%), *Pseudomonas* spp. (18%), *Staphylococcus* sp. (18%), *Klebsiella* spp. (10%) and *Enterobacter* spp. (8%) respectively. In the current study, among the bacterial isolates, gram negative bacteria prevalence is higher than gram positive bacteria significantly, "*p* value" <0.05 (Table 1).

Table 1: Percentage distribution of bacterial isolates from raw chicken sample					
Bacterial Isolates Raw chicken meat	**Count (n)**	**Prevalence (%)**	**Gram Negative**	**Gram Positive**	***P* value**
Escherichia coli	14	28%	41	9	0.18
Klebsiella spp.	5	10%			
Salmonella spp.	9	18%			
Pseudomonas spp.	9	18%			
Enterobacter spp.	4	08%			
Staphylococcus aureus	9	18%			

Note: Clinically important pathogens are encountered

The resistant attributes were analysed for all the observed isolates using percentage analysis, thus the MDR bacteria characteristics were also observed based on the resistant characters shown against three different classes of antibiotics. In this study, the *E. coli* resistant attributes towards the following antibiotics Gentamicin 50%, Levofloxacin 50%, Ciprofloxacin 50%, Trimethoprim/Sulfamethoxazole 100% and the 100% sensitive attributes towards Amikacin, Cefoperazone/Sulbactam, Cefepime, Ceftazidime, Imipenem, Meropenem, Doripenem, Tigecycline, Minocycline, Colistin, Aztreonam, Ticarcillin/Clavulanic Acid. *Salmonella* spp. resistant attributes towards the following antibiotics with 100 % resistance on Amikacin, Gentamicin, Cefuroxime Axetil, Ceftriaxone, Trimethoprim/Sulfamethoxazole and 100 % sensitive towards Ampicillin,Cefoperazone/ Sulbactam, Cefepime, Nalidixic Acid, Imipenem, Meropenem, Tigecycline, Nitrofurantoin, Colistin, *Staphylococcus sp.* resistant characters shown Penicillin (Benzyl Penicillin), Tetracycline; Macrolides (Clindamycin and Erythromycin) with 33.3%, 100%, 33.3% and 33.3% respectively. The least frequent observed isolates *Pseudomonas* spp., resistance characters shown 100% resistance with Trimethoprim Sulfamethoxazole and Tigecycline and 100% sensitive towards Aminoglycoside (Amikacin, Gentamicin), Cephalosporins(Cefoperazone/Sulbactam, Cefepime, Ceftazidime), Carbapenem(Imipenem, Meropenem), Tetracycline (Minocycline), Polymixin (Colistin), Beta lactam combinations (Piperacillin/Tazobactam, Ticarcillin/Clavulanic Acid**)**. The least number isolates *Klebsiella* spp. and *Enterobacter* spp. shown 0.0% resistance to all antibiotic classes notably. The most concerned isolate *Salmonella* spp. in the poultry sector the chicken sample was observed with 100% resistance towards Aminoglycosides (Amikacin, and Gentamicin), Cephalosporins (Cefuroxime Axetil) and Sulfonamides (Trimethoprim Sulfamethoxazole) (Table 2). *Staphylococcus* spp., *E.coli* and *Salmonella* spp., showed MDR attributes against three classes of antibiotics (Table 3). No Vancomycin-Resistant *Staphylococcus aureus* (VRSA) was found in this study. Similarly no Carbapenem and Polymyxin resistant Gram negative bacteria were found in this study. The figure 1 represents the frequency of MDR pathogens found in this study, similar to previous reports the *E. coli* and the *Salmonella* sp. were equally observed with 50% of MDRcharacters among all other studied isolates and the *Staphylococcus* sp. was found with 33.3% MDR frequency with this preliminary antimicrobial resistance study on chicken meat products.

Table: 2 Antibiotic resistances attributes of bacterial isolates in percentage based on AST (VITEK) report.						
Antibiotics	***E. coli* (n=14)**	***Enterobacter sp.*(n=5)**	***Klebsiella sp.*(n=9)**	***Pseudomonas sp.*(n=9)**	***Salmonella sp.*(n=4)**	***Staphylococcus sp.*(n=9)**
Penicillin						
Ampicillin	NA	NA	NA	NA	0(0.0%)	NA
Benzylpenicillin	NA	NA	NA	NA	NA	**33.3%**
Oxacillin	NA	NA	NA	NA	NA	0(0.0%)
Aminoglycoside						
Amikacin	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	**100%**	NA
Gentamicin	**50%**	0(0.0%)	0(0.0%)	0(0.0%)	**100%**	0(0.0%)
Cephalosporins						
Cefoperazone/ Sulbactam	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	NA
Cefepime	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	NA
Ceftazidime	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	NA	NA
Cefuroxime	NA	NA	NA	NA	NA	NA
Cefuroxime Axetil	NA	NA	NA	NA	**100%**	NA
Ceftriaxone	NA	NA	NA	NA	**100%**	NA
Quinolone						
Nalidixic Acid	NA	NA	NA	NA	0(0.0%)	NA
Levofloxacin	**50%**	0	NA	0	NA	0
Ciprofloxacin	**50%**	0(0.0%)	0(0.0%)	NA	0	0
Carbapenem						
Imipenem	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	NA
Meropenem	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	NA
Ertapenem	NA	NA	NA	NA	0	NA
Doripenem	0(0.0%)	0(0.0%)	NA	NA	NA	NA
Tetracycline	NA	NA	NA	NA	NA	**100%**
Tigecycline	0(0.0%)	0(0.0%)	0(0.0%)	**100%**	0(0.0%)	0(0.0%)
Minocycline	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	NA	NA
Sulfonamide	NA	NA	NA	NA	NA	NA

Trimethoprim/ Sulfamethoxazole	**100%**	0(0.0%)	0(0.0%)	**100%**	**100%**	0(0.0%)
Macrolides						
Clindamycin	NA	NA	NA	NA	NA	**33.3%**
Erythromycin	NA	NA	NA	NA	NA	**33.3%**
Inducible Clindamycin Resistance	NA	NA	NA	NA	NA	**Negative**
Nitrofurantoin	NA	NA	NA	NA	0(0.0%)	0(0.0%)
Polymixin						
Colistin	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	NA
Monobactam						
Aztreonam	0(0.0%)	0(0.0%)	0(0.0%)	NA	NA	NA
Beta lactam combinations						
Piperacillin/ Tazobactam	NA	NA	NA	0(0.0%)	0(0.0%)	NA
Ticarcillin/ Clavulanic Acid	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)	NA	NA
Amoxicillin/ Clavulanic Acid	NA	NA	NA	NA	0(0.0%)	NA
Cefoxitin Screen	NA	NA	NA	NA	NA	Negative
Linezolid	NA	NA	NA	NA	NA	0(0.0%)
Daptomycin	NA	NA	NA	NA	NA	0(0.0%)
Teicoplanin	NA	NA	NA	NA	NA	0(0.0%)
Vancomycin	NA	NA	NA	NA	NA	0(0.0%)
Rifampicin	NA	NA	NA	NA	NA	0(0.0%)

Note: NA – antibiotics are not tested for that isolate as per guidelines.

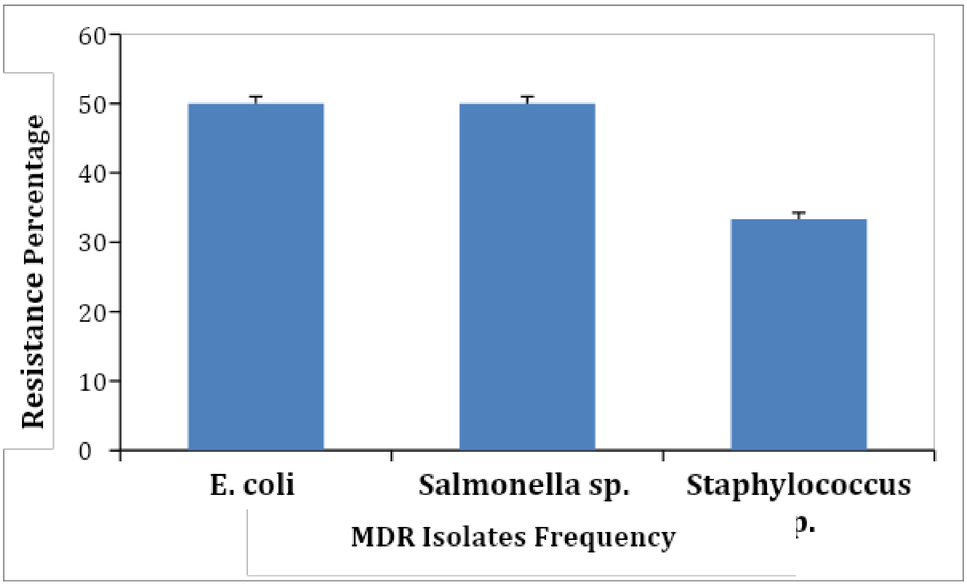

Fig 1: Frequency of MDR Isolates

Table 3: Shown are the Multidrug resistant isolates resistance frequencies against three classes of antibiotic group.

MDR Isolates	Antibiotic Classes	Antibiotics
Staphylococcus spp.	Tetracycline	Tetracycline
	Nitrofurantoin	Nitrofurantoin
	Macrolides	Erythromycin Clindamycin
E.coli	Quinolones	Levofloxacin Ciprofloxacin
	Aminoglycoside	Gentamicin
	Sulfonamides	Trimethoprim/Sulfamethaxazole
Salmonella spp.	Aminoglycoside	Amikacin Gentamicin
	Sulfonamides	Trimethoprim/Sulfamethaxazole
	Cephalosporins	Cefuroxime Cefuroxime Axetil

Note: Antibiotics subjected as per CLSI guidelines.

It illustrates that the three frequent MDR isolates are *Staphylococcus* sp., *E. coli* and *Salmonella* sp. and their resistant characteristics against various antibiotic classes.

4.1 National Database of Antibiotic Resistant Organisms (NDARO) AST data Submission

Among the observed total 12 MDR isolates, 3 isolates were identified with extreme resistance and their phenotypic AST data were submitted to NDARO, a designated biosample accession number was obtained for further genomic studies which includes Biosample - *Staphylococcus aureus*, SAMN14381906: SAU(TaxID: 2717510), Biosample - *E.coli*, SAMN14381070: ESCH (TaxID: 562) and Biosample- *Salmonella* spp., SAMN14363953; SAL(TaxID: 2717816).

5. DISCUSSION

In the present study, out of 50 raw chicken samples from retail shops, all had bacterial growth in different culture medium whereas there was no growth in frozen samples. It is reported that elevated levels of bacterial growth in the fresh chicken owed to unhygienic practices in the slaughterhouse, meat storage on inappropriate temperatures for a longer time. The medium of poultry and the overall livestock are becoming the pool of MDR microorganisms and they are highly disseminating the resistant pathogens as well. A similar study of preliminary data on AMR bacteria from raw buffalo and chicken meat of Bhaktapur, Nepal was conducted and documented the status of resistant isolates dissemination among the buffalo and chicken meat. Also the study stated about the similar report from Kathmandu with 80% resistant coliform and Pakistan with 84.0% among different bacterial species. The reason for this study was the cross contamination that occurred during the slaughter room process along with long time storage in the inappropriate temperature before delivering to the retail markets.[1] Globally the *E. coli* has been evidenced as a predominant resistant bacteria in the poultry food in various studies.[9,10] The common intestinal pathogen *E. coli* and the most concerned poultry pathogen *Salmonella* spp. were observed as the most frequent isolates and 50% of them were MDR in this study. Like Bantawa and *et al* (2019) report, the current study also confirmed *Staphylococcus* spp. as the next frequent resistant bacteria with prevalence rate of 33.3% compared to other isolates.[1] A study from Nigeria conducted by Matthew *et al*., 2017 has confirmed highest percentage of 35.6% of *Staphylococcus* spp. present in poultry feed.[11] Interestingly, *Klebsiella* spp. and *Enterobacter* spp. were the least common with no resistance to all the studied antimicrobials. In contrast many studies have proved the resistant *Klebsiella* spp. presence by recurrent gene transfer mechanism from poultry to human through consumption amid resistance characters from a decade back.[12,13] Out of 50 isolates, three MDR isolates include sample no023 *Staphylococcus* spp., sample no. 33 *E.coli* and sample no. 41 *Salmonella* spp. were confirmed as the most extreme multidrug resistant bacteria based on their antibiotic attributes shown against major classes of antibiotics. As reported earlier, in the current study it is proved that *Staphylococcus* spp. has been continuously gaining resistance to Tetracycline (100%) and Benzylpenicillin (33.3%). It is noted that *Staphylococcus* spp. has frequently attained resistance to the level of third generation of beta lactam antibiotics. A review report by Marshal BM, 2011 has stated about this wide range of frequent tetracycline resistance in animals.[14] From all the observed *Staphylococcus* spp. were found to be with resistant to tetracycline antibiotic whereas antibiotic resistant pattern study conducted by Bantawa *et al* in 2019 has shown 63% resistance to tetracycline. This study also indicated the *Staphylococcus* spp. resistant to Clindamycin (33.3%) and Erythromycin (33.3%). Interestingly no MRSA was found in this study, instead all the observed *Staphylococcus* spp. was found to be resistant with tetracycline antibiotics. There are many literature evidences about MDR *E.coli* resistant to both Aminoglycosides and Sulfonamides classes of antibiotics.[15,16] The current study observed that *E. coli* exhibited resistance to Gentamicin (50%), Trimethoprim Sulfamethoxazole(100%) which is similar to Hussain and *et al*., 2017 and Rahman and *et al*., 2017 studies.[8,17] In addition it has shown resistance to Quinolones class antibiotics, Levofloxacin (50%) and Ciprofloxacin (50%). The distribution of resistant *E.coli* with the living organisms and the environment is completely evolved with resistant pathogenic strains. It is clearly understandable that the raw meats and the slaughter office handling employees and meat selling people, utilities and processing area were completely contaminated with resistant pathogens. This study also highlighted the possible cross contamination in the selling and meat processing areas. Since MDR strains of various bacteria were observed, a keen and continuous monitoring system is an urgent need to eradicate the spread of resistant genes in the food cycle, animals, human and environment. The least prevalent *Pseudomonas* spp. has shown the steady status of resistance against Tigecycline (100%) and Trimethoprim Sulfamethoxazole (100%)[18]. Interestingly, it has been reported in a literature in 1996, a study conducted in a Multidrug Efflux -Intrinsic Resistance to Trimethoprim and Sulfamethoxazole conducted by Kohler and *et al*. The study also reveals that the *Pseudomonas aeruginosa* mutant species were effectively addressed with the following antibiotics quinolones, tetracycline, erythromycin, and the iron chelator dipyridyl using the overexpression of efflux operons mechanisms. Thus, the current hypothesis of efflux pump mediated resistance has a significant role in the spread of MDR phenotypic pathogens. It is also shown that the OprM- and OprJ- over expression strains exhibited higher level of resistance towards the sulfamethoxazole (SMX) and the mexABoprM efflux system has the key role on intrinsic resistance of *Pseudomonas aeruginosa* against the mentioned antibiotics.[19] Although frequent studies confirmed about resistant *Pseudomonas spp.* from poultry worldwide, fortunately the MDR frequency of *Pseudomonas* spp.(18%) in this study is lesser compared to *E. coli(28%)*. As similar to scientific findings, the result was similar for resistance to sulfonamides and Tetracycline antibiotics.[18] The poultry's most alarmed pathogen *Salmonella* spp. was also confirmed as MDR, and the frequency is also higher in this study, it has been proved as MDR in a recent literature with different resistance attributes.[6] In relation to many reports, in this study the resistance pattern of the *Salmonella* spp. observed against the following antibiotics was Aminoglycosides (Amikacin 50%, Gentamicin (50%); *Cephalosporins (Cefuroxime Axetil 50%, Ceftriaxone 50%) and Trimethoprim Sulfamethoxazole (50%)* and the predominant frequency was highly observed with the *E.coli* followed by *Proteus* sp., *Klebsiella* sp., *and Pseudomonas* sp..[20-23] *A* report from Nepal a decade back had shown the 14.5% prevalence of S*almonella* spp. in chicken , in this study the prevalence has been found to have increased to 5.5% (total 20%).[2] Saud and *et al* study in

2019 from Nepal reported about the MDR isolates from chicken and buffalo meat, it was similar with MDR *E.coli* and *Staphylococcus* spp. in our study except *Salmonella* spp. in this study.[1] Consecutively in the same year MDR *Salmonella* spp. exclusively has been studied and reported from an Indian report by Bandyopadhyay *et al*, 2019 against various drugs.[6] A study from Korea by Lee HJ and *et al*, 2018 has revealed the relationship between antibiotic residues and resistant isolates antibiotic compounds in chicken samples.[24] It is noteworthy that both gram positive and gram-negative bacteria are developing a diverse pattern of resistant characteristics worldwide through horizontal gene transfer and drug pressure.[14] In 2008, antibiotic resistance study conducted by Kilonzo *et al*. has noticeably explained about the impending antibiotic-resistant bacteria contamination to humans via poultry.[25] Globally, the overall report on self-prescription of oral antibiotics were noted for about 66% in most of the viral infections such as flu, Pneumonia as well as protozoal infection such as Malaria.[26]

6. CONCLUSION

In conclusion, this is probably first report from South India about resistant bacteria surveillance in raw chicken meat samples collected from retail shops. The confirmed MDRs were submitted to NDARO. By this poultry food investigational study, it is clear that meat products from retail shops are highly contaminated with the MDR resistant bacteria. Rural and Urban awareness campaigns should be initiated along with the national level surveillance programs to limit the antibiotic usage in poultry. The strategies proposed in the national level antibiotic resistance surveillance forum should reach ground level actions to reduce the antibiotic resistance in the environment. This growing resistance issue worldwide will become another panic condition like the deadly virus evolution and emerge as a challenge to the scientific community.

7. AUTHORS CONTRIBUTION STATEMENT

All authors listed in this study have made an extensive and intellectual paper contribution to the work and approved it for publication. SharmilaDevi Natarajan and Balaji Nagarajan conceptualized the work and executed the experiments. R DineshKumar, Duraimurugan M, Mythili R, M Prathaban and Varadharaju Chandrasekar, analyzed these data and provided necessary support to finish this Manuscript successfully.

8. ACKNOWLEDGEMENTS

The authors thank the laboratory personnel of the Department of Microbiology, Karpagam Academy of Higher Education and Bioline Laboratory and Research Institute for excellent technical assistance.

9. CONFLICTS OF INTEREST

Conflict of interest declared none.

10. ABBREVIATIONS

MDR: Multi Drug Resistant
Spp.: Species
CLSI: Clinical and Laboratory Standards Institute
MRSA: Methicillin-Resistant *Staphylococcus aureus*.
VRSA: Vancomycin –Resistant *Staphylococcus aureus*
NDARO- National Database of Antibiotic Resistant Organisms
AMR – Antimicrobial Resistance

11. REFERENCES

1. Saud Bhuvan, Paudel Govinda, Khichaju Sharmila, Bajracharya Dipendra, Dhungana Gunaraj, Awasthi Mamata Sherpa, Shrestha Vikram. Multidrug-resistant bacteria from raw meat of buffalo and chicken Nepal. Vet Med Int. 2019;2019:Article ID 7960268. doi: 10.1155/2019/7960268, PMID 31186828.
2. World Health Organization (WHO). 2001. Global strategy for containment of antimicrobial resistance, summary of recommendations for intervention. World Health Organization [publication]. (WHO, CDS/CSR/DRS. 2001.2a.).
3. Adu-Gyamfi A, Torgby-Tetteh W, Appiah V. Microbiological quality of chicken sold in Accra and determination of D10-value of *E. coli*. Food Nutr Sci. 2012;3:693-8. doi: 10.4236/fns.2012.35094.
4. World Health Organization (WHO). High levels of antibiotic resistance found worldwide; new data shows antibiotic resistance. Available from: http://www.who.int/mediacentre/news/releases/2018/antibiotic-resistance-found/en/. Vol. 2018 (January). BANGKOK; 2018.
5. Yildirim YZ, Gonulalan Z, Pamuk S, Ertas N. Incidence and antibiotic resistance of *Salmonella* spp. on raw chicken carcasses. Food Res Int. 2011;44(3):725-8. doi: 10.1016/j.foodres.2010.12.040.
6. Bandyopadhyay M, Jha V, Ajithkumar BS et al. Prevalence study and resistance profiles of multi-drug resistant Salmonella obtained from poultry across Mumbai region. Acta Sci Microbiol. *2019*;2(5):36-41.
7. Gomez TM, Motarjemi Y, Miyagawa S, Käferstein FK, Stöhr K. Foodborne salmonellosis. World Health Stat Q. 1997;50(1-2):81-9. PMID 9282390.
8. Hussain Arif, Shaik Sabiha, Ranjan Amit, Nandanwar Nishant, Tiwari Sumeet K, Majid Mohammad, Baddam Ramani, Qureshi Insaf A, Semmler Torsten, Wieler Lothar H, Islam Mohammad A, Chakravortty Dipshikha, Ahmed Niyaz. Risk of transmission of antimicrobial resistant Escherichia coli from commercial broiler and free-range retail chicken in India. Front Microbiol. 2017;8:2120. doi: 10.3389/fmicb.2017.02120, PMID 29180984.
9. Mahalmani Vidya M, Sarma Phulen, Prakash Ajay, Medhi Bikash. Positive list of antibiotics and food products: current perspective in India and across the globe. Indian J Pharmacol. 2019;51(4):231-5. doi: 10.4103/ijp.IJP_548_19, PMID 31571708.
10. Rahman MA, Rahman AKMA, Islam MA, Alam MM. Antimicrobial resistance of Escherichia coli isolated from milk, beef and chicken meat in Bangladesh. Bangl J Vet Med. 2017;15(2):141-6. doi: 10.3329/bjvm.v15i2.35525.
11. Matthew O, Chiamaka R, Chidinma O. Microbial analysis of poultry feeds produced in Songhai farms,

Rivers State, Nigeria. J Microbiol Exp. 2017;4(2). doi: 10.15406/jmen.2017.04.00110.
12. Fielding Burtram C, Mnabisa Amanda, Gouws Pieter A, Morris Thureyah. Antimicrobial-resistant Klebsiella species isolated from free-range chicken samples in an informal settlement. Arch Med Sci. 2012;8(1):39-42. doi: 10.5114/aoms.2012.27278, PMID 22457672.
13. Marchaim Dror, Navon-Venezia Shiri, Schwaber Mitchell J, Carmeli Yehuda. Imipenem-resistant Enterobacter species isolation: the emergence of KPC-2 carbapenemase, molecular characterization, epidemiology, and outcomes. Antimicrob Agents Chemother. 2008;52(4):1413-8. doi: 10.1128/AAC.01103-07, PMID 18227191.
14. Marshall Bonnie M, Levy Stuart B. Food animals and antimicrobials: impacts on human health. Clin Microbiol Rev. 2011;24(4):718-33. doi: 10.1128/CMR.00002-11, PMID 21976606.
15. Taneja Neelam, Sharma Megha. Antimicrobial resistance in the environment: the Indian scenario. Indian J Med Res. 2019;149(2):119-28. doi: 10.4103/ijmr.IJMR_331_18, PMID 31219076.
16. Laxminarayan Ramanan, Chaudhury Ranjit Roy. Antibiotic resistance in India: drivers and opportunities for action. PLOS Med. 2016;13(3):e1001974. doi: 10.1371/journal.pmed.1001974, PMID 26934098.
17. Nhung Nguyen Thi, Chansiripornchai Niwat, Carrique-Mas Juan J. Antimicrobial resistance in bacterial poultry pathogens: a review. Front Vet Sci. 2017;4(126):126. doi: 10.3389/fvets.2017.00126, PMID 28848739.
18. Agyare Christian, Boamah Vivian Etsiapa, Zumbi Crystal Ngofi et al. Antibiotic use in poultry production and its effects on bacterial resistance. Antimicrob Resist Glob Threat. 2018. doi: 10.5772/intechopen.79371.
19. KöHler Thilo, Kok Menno, Michea-Hamzehpour Mehri, Plesiat P, Gotoh N, Nishino T, Curty LK, Pechere JC. Multidrug Efflux in intrinsic Resistance to trimethoprim and sulfamethoxazole in *Pseudomonas aeruginosa*. Antimicrob Agents Chemother. 1996;40(10):2288-90. doi: 10.1128/AAC.40.10.2288, PMID 9036831.
20. Bantawa Kamana, Sah Shiv Nandan, Subba Limbu DhirenS, Subba Prince, Ghimire Arjun. Antibiotic resistance patterns of Staphylococcus aureus, Escherichia coli, Salmonella, Shigella and Vibrio isolated from chicken, pork, bufalo and goat meat in eastern Nepal. BMC Res Notes. 2019;12(1):766. doi: 10.1186/s13104-019-4798-7, PMID 31752992.
21. Muhammad Nazir Uddin MN, Farooq Muhammad. Antibiotic assays of Salmonella isolated from poultry chicken of various locations in districts Swat. Pure Appl Biol. 2018;7(1):78-84. doi: 10.19045/bspab.2018.70010.
22. Zhang Lina, Fu Ying, Xiong Zhiying, Ma Yeben, Wei Yihuan, Qu Xiaoyun, Zhang Hongxia, Zhang Jianmin, Liao Ming. Highly prevalent multidrug-resistant *Salmonella* from chicken and pork meat at retail markets in Guangdong, China. Front Microbiol. 2018;9:2104. doi: 10.3389/fmicb.2018.02104, PMID 30258419.
23. Mamber SW, Katz SE. Effects of antimicrobial agents Fed to Chickens on some Gram-negative enteric Bacilli. Appl Environ Microbiol. 1985;50(3):638-48. doi: 10.1128/aem.50.3.638-648.1985, PMID 3852666.
24. Lee Hyo-Ju, Cho Seung-Hak, Shin Dasom, Kang Hui-Seung. Prevalence of antibiotic residues and antibiotic resistance in isolates of chicken meat in Korea. Korean J Food Sci Anim Resour. 2018;38(5):1055-63. doi: 10.5851/kosfa.2018.e39, PMID 30479511.
25. Kilonzo-Nthenge A, Nahashon SN, Chen F, Adefope N. Prevalence and antimicrobial resistance of pathogenic bacteria in chicken and guinea fowl. Poult Sci. 2008;87(9):1841-8. doi: 10.3382/ps.2007-00156, PMID 18753453.
26. Otaibi NN, Almutairi M, Al-Namshan YZ, Al Dhohayan MA, Alotibi RD. Factors influencing misuse of antibiotic therapy in Al-Qassim region, Saudi Arabia. Int J Life Sci Pharm Res. 2019;9(1): (P) 1-6.