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Multi-drug resistant phenotypes of extended-spectrum β-lactamase (ESBL)-producing *E. coli* from layer chickens

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Abstract

Antimicrobial resistance (AMR) is a growing and emerging public health problem worldwide. This research determines the occurrence of ESBL *E. coli* and antimicrobial resistance profiles of *E. coli* on eggshells from selected layer chickens. The shells of 270 egg samples were swabbed to detect the presence of *E. coli*. *E. coli* isolates were recovered from 73(23%) of the 270 samples collected. The isolates were subjected to antimicrobial susceptibility testing using six panel antibiotics (ampicillin, tetracycline, sulphamethoxazole-trimethoprim, gentamicin, imipenem, and ciprofloxacin) using the disk diffusion method. The isolates showed the highest resistance to Ampicillin 95.9%, closely followed by tetracycline 89%, sulphamethoxazole-trimethoprim 72%, gentamicin 41.1%, and imipenem 1.4%. Also, 78% of the isolates were multi-drug resistant. A 56/73 (76.7%) out of seventy-three isolates were screened as presumptive ESBL-*E. coli* by culture on ESBL CHROM agar and 42/56 (75%) of the isolates yielded ESBL-producing *E. coli* based on the production of ESBL by double disc diffusion method. The questionnaire survey results showed that all farms used antimicrobial agents for therapeutic or prophylactic purposes. Also, not all the farms had suitable biosecurity measures. The findings of this study indicated that eggshells are potential reservoirs for multi-drug resistant *E. coli* and ESBL-Producing *E. coli.*

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Introduction

Antimicrobial resistance (AMR) in food-producing animals has negatively impacted veterinary and human medicine during the last decade. Bacteria from food animals carrying resistance determinants are being investigated for their potential to transfer AMR to humans via the environment, food chain, and direct contact (1). Antimicrobial resistance is not only a problem for pathogenic bacteria but also normal gut microbiota (2). *E. coli* is the most common and essential indicator of Gram-negative gut microbiota, and it is also regarded as a significant reservoir of acquired resistance genes (3). Phenotypes of *E. coli* are used as a proxy of AMR in the gut of healthy animals, including resistance genes mediated by conjugative plasmids (4).

Extended Spectrum β-lactamases confers resistance to many β-lactam antimicrobials, particularly the third generation cephalosporins, including ceftriaxone, cefotaxime, and ceftazidime, and aztreonam, but not to carbapenem and cephamycin. Many of these enzymes are $bla_{CTX\text{-}M}$, bla_{TEM}, and bla_{SHV} types, and *E. coli* isolates producing CTX-M types have been recognized as significant causes of community and hospital-acquired infections. ESBL *E. coli* have gained the notoriety of being an indicator for the spread of resistance determinants and the transfer of resistance genes between bacteria of the same and varied species through transmissible plasmids. These resistance plasmids have a global distribution and have been reported to confer resistance to 1st, 2nd, and 3rd generation cephalosporins and monobactams. Studies have indicated that transmissible plasmids carrying resistance genes to cephalosporins are widely disseminated in humans and animals. Thus, suggesting the likelihood of food animals serving as significant transmission sources of resistance determinants bacteria to humans via horizontal gene transfer (5-15). To investigate the occurrence of MDR-phenotypes in ESBL *E. coli*, egg samples were collected from nine different farms within Jos and environs, first to isolate and identify *E. coli* and investigate their resistance profiles and ESBL production.

Materials and methods

Ethical approval

This study was approved by research ethics committee of the Faculty of Veterinary Medicine University of Jos on the 11th of November 2020: ID UJ/VM/0044.

Study area

This study was conducted in Jos North Local Government Area and environs. Jos, the capital of Plateau State, is located at an altitude of 1217 m) above sea level. Jos climate is closer to temperate than that of the clear majority of Nigeria. Average monthly temperatures range from 21-25°C, and from mid-November to late January, night-time temperatures drop to as low as 7°C. Jos receives about 1400 millimeters of rainfall annually, with a unimodal pattern of rainfall and a downward trend, and rainfall variability has never gone above or below 400 mm with a mean of 1326.253 mm, the precipitation arising from both conventional and orographic sources, owing to the location of the city on the Jos Plateau. Jos North Local Government area has fourteen wards, covering 291 square kilometers and a population projection of 429,400 people as of the 2006 census (National Population Commission of Nigeria, National Bureau of Statistics web).

Sample size

A convenient random sampling technique was used in this study. Samples were collected from nine different layer farms located within Jos North Local Area and environs. A total of 270 eggshell swab sample was collected. Utmost care was taken to manage the eggs with sterile gloves aseptically. For examination, samples were adequately packaged and transported to the Veterinary Microbiology Laboratory, Faculty of Veterinary Medicine, University of Jos.

Antibiotic susceptibility testing and detection of ESBL production

The antimicrobial susceptibility test was performed using the disc diffusion method as described in the guidelines of the Clinical and Laboratory Standard Institute CLSI (16). The sensitivity of all confirmed *E. coli* isolates was evaluated against six-panels of antibiotics, each representing classes of antimicrobials commonly used in poultry on the Plateau. The tested antibiotics includes Ampicillin 10 µg, Ciprofloxacin 5 µg, Gentamicin 10 µg, Imipenem 10 µg, Sulphamethoxazole-Trimethoprim 1.25/23.75 µg, and Tetracycline 30 µg. *E. coli* ATCC 25922 and *Pseudomonas aeruginosa* ATCC 27853 were used as quality control. For confirmed *E. coli* isolates, production of ESBL enzyme was determined using a combination of disc diffusion method with ceftazidime (CAZ) and cefotaxime (CTX) alone and in combination with clavulanic acid (CAZ/CLA and CTX/CLA) according to the guidelines of CLSI (16). A ≥5 mm increase in zone diameter for either antimicrobial agent evaluated in combination with clavulanic acid versus the zone diameter of the antimicrobial agent when evaluated alone was considered for ESBL Production (17).

Determination of multiple antibiotic resistance index

The multiple antibiotic resistance (MAR) index for each recovered *E. coli* isolates was determined by dividing the number of antibiotics to which the isolate was resistant, divided by the total number of antibiotics evaluated (18). MAR Index = No. of antibiotics to which the isolate is resistant / Total No. of antibiotics evaluated.

Data analysis

Data were entered into and analyzed in Microsoft office excel 2016. The data were analyzed using descriptive statistics to show the percentage of Farms with *E. coli*, ESBL *E. coli*, and resistance phenotypes.

Results

Occurrence of *E. coli* from selected layer chicken farms

Of the 270 egg samples collected, 23% (73/270) were phenotypically confirmed as *E. coli*. The recovery rate across farms showed that farms two 60%, one 40%, and four 37% had the highest occurrence of *E. coli* isolates, while farms five and nine had an occurrence of 3.3% and 10%. Farms seven and eight had 30%, respectively, while the percentage of *E. coli* isolates recovered in farms three and six were 16% (Table 1).

Antimicrobial Susceptibility of *E. coli*

The overall occurrence of AMR in *E. coli* is high for most of the six evaluated antibiotics (Figure 1). The highest rates of resistance were found for Ampicillin 95.9% (70/73), tetracycline 89% (65/73), sulphamethoxazole-trimethoprim 72.6% (53/73), and ciprofloxacin 50.7% (37/73) followed by gentamicin 41.1% (30/73) and imipenem 1.4% (1/73) (Table 2 and Figure 2). *E. coli* isolates recovered from farm two were resistant to all tested antimicrobials. Farms 1, 3, 4, 6, 7, and 8 were resistant to all antimicrobials tested but imipenem. In contrast, farms 5 and 9 were resistant to two (Ampicillin and Ciprofloxacin) and three (Ampicillin, Tetracycline, and Sulphamethoxazole) tested antimicrobial agents, respectively (Figure 1). For most *E. coli* isolates, 78% (57/73) presented multiple drug resistance profiles (≥3 antimicrobial class) and with a MAR index of ≥ 0.2 (Figure 3). Additionally, 56% (41/73) and 20.5% (15/73) of the *E. coli* isolates showed resistance to four and five different classes of antibiotics, respectively (Table 3).

Table 1: Occurrence of *E. coli* isolates recovered from freshly laid eggs from selected Chicken farms

Farm ID	+ve farm (n)	% +ve isolates
Farm 1	12	40%
Farm 2	18	60%
Farm 3	5	16%
Farm 4	11	37%
Farm 5	1	3.3%
Farm 6	5	16%
Farm 7	9	30%
Farm 8	9	30%
Farm 9	3	10%
Total	73	23%

A total of 30 eggs were collected from each farm.

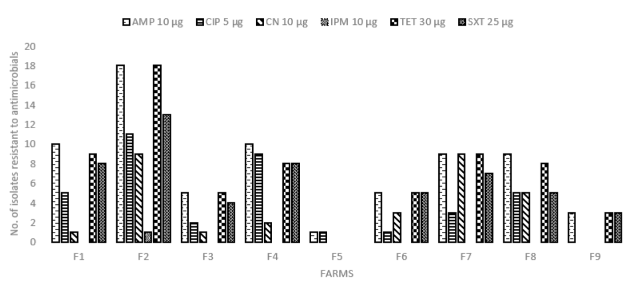

Figure 1: Farm-level antimicrobial resistance profiles of *E. coli* isolates recovered from the surfaces of freshly laid eggs from selected layer chicken farms in Jos and Environs; AMP 10µg- Ampicillin, CIP 5µg -Ciprofloxacin, CN 10µg -Gentamicin, IPM 10µg -Imipenem, TET 30µg -Tetracycline, SXT 25 µg - Sulphamethoxazole, F1 to F9= Farm 1 to Farm 9.

Phenotypic detection of ESBL *E. coli*

Nine small-scale layer chicken farms were used in this study. A total of 73 *E. coli* isolates were recovered, and out of this, 56/73(76.7%) were initially screened as presumptive ESBL-*E. coli* by culture on ESBL CHROMagar. Confirmation of ESBL production by the double-disc diffusion method showed that 42/56 (75%) yielded ESBL-producing *E. coli*. All farms sampled in this study yielded ESBL- producing *E. coli*. Among all the farms, two farms (farm two and farm one) had the highest ESBL *E. coli*, while one had the least ESBL-producing *E. coli*.

Farm details and management practices

A well-structured questionnaire was administered to collate information on farm details, farm management practice, history of antimicrobial use, and biosecurity measures in all the farms visited. The farms visited in this study were all located within Jos north local government and environs. The stock size ranged from one hundred birds to 3700 within 42 to 70 weeks. The common breeds of layer chickens encountered were Isa brown and Leghorn, and 55.6% of the farmers had other birds on their farms other than layer chickens. All farms practiced a deep litter system; 88.9% used commercially compounded feeds, while 11.1% compounded their feeds.

Additionally, 66.7% of the farmers used well water as a water source, and 33.3% used boreholes. The egg production of the farms ranged from three and a half to one hundred crates per day. None of the farmers decontaminate their eggs before selling.

Farm vaccination history showed that all farms have fully vaccinated their flocks and six farms (66.7%) identified New Castle disease, coccidiosis, and fowl typhoid as the most encountered diseases. Out of this, only five farms utilized the services of a veterinarian to confirm the diagnosis of disease in their farms, while the other diagnosed diseases in their farms based on previous experience.

All farms used antimicrobials in their farms; 66.7% of the farmers' used antimicrobials for therapeutic purposes only, and 33.3% used antimicrobials for prophylaxis. The commonly used antimicrobial class used in the farms were quinolones, tetracyclines, aminoglycosides, and macrolides. In all the farms visited, 77.8% utilized the services of a veterinarian as their source of prescription, and 22.2% prescribed antimicrobials based on previous experience. The administration of antimicrobials in five farms was less than seven days, and four farms administered antimicrobials for seven days. Similarly, five farms used other supplements as growth promoters.

In terms of biosecurity, only 3 (33.3%) of the farms had foot dips at the entrance of the farms and each pen. Eight farms 88.9% provided personal protective attires to their workers, and out of this, only seven farms 77.8% agreed that their workers always use personal protective clothing on the farms, one farm responded that their workers sometimes used personal protective clothing, while the other said their workers never used personal protective equipment. In terms of workers' hygiene, seven farms agreed that their workers' hygiene was poor and that flies, flying birds, and other insects had access to the poultry farms. All farms agreed to change their litter, but the frequency varied. For instance, 66.7% of the farmers changed their litter once in two weeks, 22.2% once a month, and 11.1% only after harvest.

Table 2: Antimicrobial susceptibility profiles of *E. coli* recovered from the surfaces of freshly laid eggs from layer chickens

Antimicrobial agents	Disc content	+ve isolates	Susceptibility profiles of *E. coli* isolates		
			Resistant (%)	Intermediate (%)	Susceptible (%)
Ampicillin	10 µg	73	70 (95.9)	2 (2.7)	1 (1.4)
Gentamicin	10 µg	73	30 (41.1)	0 (0)	43 (58.9)
Ciprofloxacin	5 µg	73	37 (50.7)	23 (31.5)	13(17.8)
Imipenem	10 µg	73	1(1.4)	1 (1.4)	71(97.2)
Tetracycline	30 µg	73	65 (89.0)	0 (0)	8 (10.9)
Sulphamethoxazole	25 µg	73	53(72.6)	2 (2.7)	18(24.7)

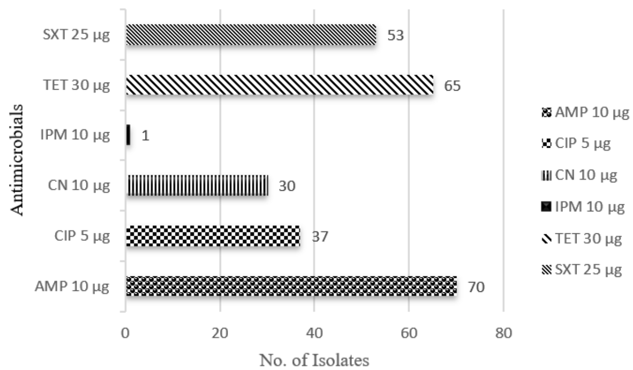

Figure 2: Antimicrobial resistance of *E. coli* recovered from the surface of freshly laid eggs from selected layer chicken farm in Jos and Environs (n = 73); AMP 10µg- Ampicillin, CIP 5µg -Ciprofloxacin, CN 10µg - Gentamicin, IPM 10µg -Imipenem, TET 30µg - Tetracycline, SXT 25 µg – Sulphamethoxazole.

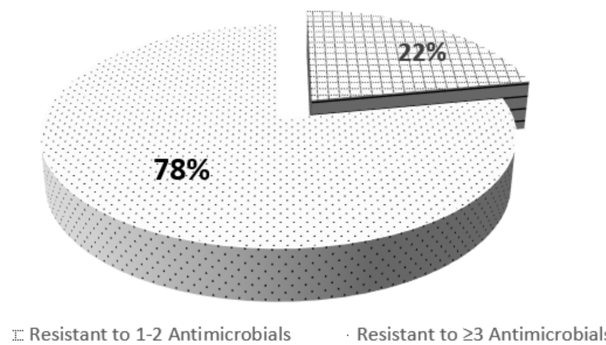

Figure 3: Percentage of MDR *E. coli* isolates recovered from eggshells.

Table 3: *E. coli* isolates with Multiple Antimicrobial (MAR) Index of ≥ 0.2 or more

Resistance profiles	Resistant (n)	MAR Index
Single resistance		
AMP	2	≤0.2
SXT	2	≤0.2
Double resistance		
AMPCIP	3	≥0.2
TETCIP	1	≥0.2
AMPSXT	2	≥0.2
AMPTET	6	≥0.2
Multi-drug resistance		
AMPSXTTET	13	≥0.2
AMPTETCN	1	≥0.2
AMPTETCIP	2	≥0.2
AMPSXTTETCN	9	≥0.2
AMPSXTTETCIP	11	≥0.2
IPMAMPSXTTET	1	≥0.2
AMPTETCIPCN	5	≥0.2
AMPSXTTETCIPCN	15	≥0.2
Total	73	

Discussion

Bacteria from animal sources constituted a considerable proportion of opportunistic pathogens and are known to carry clinically relevant resistance determinants, owing partly to the frequent and indiscriminate use of antimicrobials in human and veterinary medicine (17). This study investigated the antimicrobial resistance profiles of commensal ESBL *E. coli* isolated on eggshells from selected layer chicken farms in Jos and environs. Our results showed that 73 out of the 270 egg samples collected were identified as *E. coli*. The *E. coli* recovery rate across farms showed that three farms had the highest recovery rate, and two farms had the least identified *E. coli* isolates. The high occurrence of potentially clinically relevant *E. coli* obtained in this study is not surprising, as *E. coli* is one of the notable members of the family *Enterobacteriaceae* known to colonize poultry's gastrointestinal tract.

Additionally, *E. coli* is a known indicator organism for studying the spread of antimicrobial resistance genes (3,19). These have a significant public health impact since these bacteria, and their resistance genes can be transmitted to humans via the food chain. This can also lead to contamination of the environment when fecal content from these birds is used either as organic manure to grow vegetables or feed pigs and fish. The variable *E. coli* recovery rate observed in this study could be related to the farm biosecurity measures and workers' hygiene. Farm hygiene plays a critical role in reducing contamination by pathogens. This finding agrees with Musa *et al*. (5) report, where the authors reported that animal handlers and equipment contributed to a persistent contamination of the farm environment.

The highest number *of E. coli* resistant isolates recovered from eggshells were found to be for Ampicillin, tetracycline, and sulphamethoxazole. This was followed by resistance to ciprofloxacin, gentamicin, and imipenem. All these antimicrobial agents are commonly used either singly or in combination with others in poultry production for prophylactic or therapeutic purposes, thus favoring the selection of resistant bacteria that can potentially be transmitted to humans via the food chain, direct animal contact, or environmental contamination (19-23).

E. coli isolates were resistant to Ampicillin, ciprofloxacin, gentamicin, tetracycline, and sulphamethoxazole, and three farms were resistant to two and three antimicrobials, respectively. These resistance profiles directly reflect antimicrobial use on the farms and support the theory that indiscriminate or sustained use of antimicrobials in conventional poultry farms exerts selective pressure on the microbial community, aiding the persistence and dissemination of resistance genes among bacterial bacteria species. The findings also agree with Davis *et al*. (24) report, where empirical research evidence revealed a higher prevalence of antimicrobials resistance among *E. coli* isolates from traditional turkey farms than organic turkey farms with no history of antimicrobial use. It is unclear why the *E. coli* isolates recovered and used had variable resistance levels in this study. However, this phenomenon existed because of the magnitude of use of antimicrobials in each of the farms visited. Additionally, Mathew *et al*. (25) demonstrated a relationship between the level of resistance and antimicrobial use. Another explanation could be the horizontal transfer of resistance determinants to susceptible *E. coli* strains or via intrinsic resistance mechanisms.

In this study, a considerable number of the *E. coli* isolates were MDR phenotypes and with a multiple antimicrobial resistance (MAR) index of ≥ 0.02 representing seven out of the nine farms studied. Studies have demonstrated a higher prevalence of MDR *E. coli* in chickens and the environment in Nigeria 91.8% (26,27), Bangladesh 75.06% (28), India 60%, and Nepal 80% (29). These high occurrences of MDR phenotypes could be associated with indiscriminate use of antimicrobials, which may replace susceptible strains of bacteria in an environment already overwhelmed by antimicrobials.

To investigate the occurrence of ESBL-producing *E. coli* on eggshells from selected layer chicken farms. The results showed that ESBL *E. coli* is circulating in poultry farms, which presents a significant health risk to humans. The occurrence of ESBL-producing *E. coli* reported in this study was 75%, higher than those reported in a study by Aworh *et al*. (26). This high occurrence of ESBL-producing *E. coli* observed in this study could be attributed to easy access to antimicrobial agents for veterinary use. Contamination of Egg by ESBL *E. coli* phenotypes is likely to occur during laying. This can be due to contaminated hens through horizontal transmission from environmental dust or feces (30,31). This is evident because the questionnaire survey results showed that all farms used antimicrobial agents for therapeutic or prophylactic purposes. Additionally, not all farms have reasonable biosecurity measures.

Conclusion

The findings of this study affirmed the presence of antimicrobial resistance *in E. coli* isolates and ESBL *E. coli* phenotypes on eggshell from selected layer chicken farms in Jos and environs. Antimicrobial resistance phenotypes in poultry farms are a critical concern as they can enter the food chain and directly affect public health. There is a need to ensure comprehensive surveillance and more sensible use of antimicrobial agents in layer chick farms to stem the tides against the spread of resistant phenotypes.

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Conflict of Interest

The authors declare that there are no conflicts of interest regarding the publication of this manuscript.

References

1. Ceccarelli D, Hesp A, Van Der Goot J, Joosten P, Sarrazin S, Wagenaar JA, Dewulf J, Mevius DJ. Antimicrobial resistance prevalence in commensal *E. coli* from broilers, fattening turkeys, fattening pigs, and veal calves in European countries and associated with antimicrobial usage at the country level. J Med Microbiol.2020;69:537-47. DOI: 10.1099/jmm.0.001176
2. Hoelzer K, Wong N, Thomas J, Talkington K, Jungman E, Coukell A. Antimicrobial drug use in food-producing animals and associated human health risks:what, and how strong, is the evidence? BMC Vet Res.2017;13:1-38. DOI: 10.1186/s12917-017-1131-3
3. Thu WP, Sinwat N, Bitrus AA, Angkittitrakul S, Prathan R, Chuanchuen R. Prevalence, antimicrobial resistance, virulence gene,

and class 1 integrons of Enterococcus faecium and Enterococcus faecalis from pigs, pork, and humans in Thai-Laos border provinces. J Glob Antimicrob Resist. 2019;18:130-8. DOI: 10.1016/j.jgar.2019.05.032
4. Scott AM, Beller E, Glasziou P, Clark J, Ranakusuma RW, Byambasuren O, Bakhit M, Page SW, Trott D, Del Mar C. Is antimicrobial administration to food animals a direct threat to human health? A rapid systematic review. Int J Antimicrob Agents.2018;52:316-23. DOI: 10.1016/j.ijantimicag.2018.04.005
5. Musa L, Casagrande Proietti P, Branciari R, Menchetti L, Bellucci S, Ranucci D, Marenzoni ML, Franciosini MP. Antimicrobial susceptibility of *E. coli* and ESBL-producing *E. coli* diffusion in conventional, organic, and antibiotic-free meat chickens at slaughter. Animals.2020;10:1215. DOI: 10.3390/ani10071215
6. Proietti PC, Stefanetti V, Musa L, Zicavo A, Dionisi AM, Bellucci S, Mensa AL, Menchetti L, Branciari R, Ortenzi R, Franciosini MP. Genetic Profiles and Antimicrobial Resistance Patterns of Salmonella Infantis Strains Isolated in Italy in the Food Chain of Broiler Meat Production. Antibiotics.2020;9:814. DOI: 10.3390/antibiotics9110814
7. Brower CH, Mandal S, Hayer S, Sran M, Zehra A, Patel SJ, Kaur R, Chatterjee L, Mishra S, Das BR, Singh P. The prevalence of extended-spectrum beta-lactamase-producing multidrug-resistant *E. coli* in poultry chickens and variation according to farming practices in Punjab, India. Environ Health Perspect. 2017;125:077015. DOI: 10.1289/EHP292
8. Schmid A, Hörmansdorfer S, Messelhäusser U, Käsbohrer A, Sauter-Louis C, Mansfeld R. Prevalence of extended-spectrum β-lactamase-producing *E. coli* on Bavarian dairy and beef cattle farms. Applied Environ Microbiol. 2013;79:3027-32. DOI: 10.1128/AEM.00204-13
9. Bitrus AA, Chuanchuen R, Luangtongkum T. Emergence of colistin resistance in extended-spectrum beta-lactamase-producing Enterobacteriaceae isolated from food animals and its public health implication:A review. J Adv Vet Anim Res.2018;5:1-1. DOI: 10.5455/javar.2018.e246
10. Watson E, Jeckel S, Snow L, Stubbs R, Teale C, Wearing H, Horton R, Toszeghy M, Tearne O, Ellis-Iversen J, Coldham N. Epidemiology of extended-spectrum beta-lactamase *E. coli* (CTX-M-15) on a commercial dairy farm. Vet Microbiol. 2012;154:339-46. DOI: 10.1016/j.vetmic.2011.07.020
11. Yang H, Rehman MU, Zhang S, Yang J, Li Y, Gao J, Gu L, Wang M, Jia R, Chen S, Liu M. High prevalence of CTX-M belonging to ST410 and ST889 among ESBL producing *E. coli* isolates from waterfowl birds in China’s tropical island, Hainan. Acta Trop.2019;194:30-5. DOI: 10.1016/j.actatropica.2019.03.008
12. Yu ZN, Wang J, Ho H, Wang YT, Huang SN, Han RW. Prevalence and antimicrobial-resistance phenotypes and genotypes of *E. coli* isolated from raw milk samples from mastitis cases in four regions of China. J Glob Antimicrob resist. 2020;22:94-101. DOI: 10.1016/j.jgar.2019.12.016
13. Hartmann A, Amoureux L, Locatelli A, Depret G, Jolivet C, Gueneau E, Neuwirth C Occurrence of CTX-M producing *E. coli* in soils, cattle, and farm environment in France (Burgundy region). Front Microbiol. 2012;3:83. DOI: 10.3389/fmicb.2012.00083
14. Hijazi SM, Fawzi MA, Ali FM, Abd El Galil KH. Multidrug-resistant ESBL-producing Enterobacteriaceae and associated risk factors in community infants in Lebanon. The J Infect Develop Countries.2016;10:947-55. DOI: 10.3855/jidc.7593
15. Tamang MD, Nam HM, Kim SR, Chae MH, Jang GC, Jung SC, Lim SK. Prevalence, and molecular characterization of CTX-M β-lactamase-producing *E. coli* isolated from healthy swine and cattle. Foodborne Pathog Dis.2013;10:13-20. DOI: 10.1089/fpd.2012.1245
16. Clinical and Laboratory Standards Institute. Performance Standards for Antimicrobial Susceptibility Testing. USA: Twentieth Informational Supplement; 2013. 30 p.
17. Lay KK, Torio HE, Bitrus AA, Chuanchuen WM. Multidrug-resistant *E. coli* Harboring Extended-spectrum β-Lactamase-encoding genes isolated from clinically healthy pigs. Thai J Vet Med. 2021;51(2):303-10. DOI: 10.14456/tjvm.2021.38
18. Krumperman PH. Multiple antibiotic resistance indexing of *E. coli* to identify high-risk sources of fecal contamination of foods. Applied Environ Microbiol. 1983;46(1):165-170. DOI: 10.1128/aem.46.1.165-170.1983
19. Savin M, Bierbaum G, Hammerl JA, Heinemann C, Parcina M, Sib E, Voigt A, Kreyenschmidt J. Isolation and characterization of ESKAPE-bacteria and ESBL-producing *E. coli* from waste-and process water of German poultry slaughterhouses. Appl Environ Microbiol. 2020;86:e02748-19. DOI: 10.1128/AEM .02748-19
20. Wassenaar, TM. Use of antimicrobial agents in veterinary medicine and implications for human health. Crit Rev Microbiol. 2005;31:155-169. DOI: 10.1080/10408410591005110
21. Diarra MS, Silversides FG, Diarrassouba F, Pritchard J, Masson L, Brousseau R, Bonnet C, Delaquis P, Bach S, Skura BJ, Topp E. Impact of feed supplementation with antimicrobial agents on growth performance of broiler chickens, Clostridium perfringens and Enterococcus counts, and antibiotic resistance phenotypes and distribution of antimicrobial resistance determinants in *E. coli* isolates. Appl Environ Microbiol. 2017;73:6566-76. DOI: 10.1128/AEM.01086-07
22. Ljubojević D, Radosavljević V, Milanov D. The role of gulls (Laridae) in the emergence and spreading of antibiotic resistance in the environment. World's Poult Scie J.2016;72:853-64. DOI: 10.1017/S0043933916000659
23. Ljubojević D, Pelić M, Puvača N, Milanov D. Resistance to tetracycline in *E. coli* isolates from poultry meat:Epidemiology, policy, and perspective. World's Poult Scie J.2017;73:409-17. DOI: 10.1017/S0043933917000216
24. Davis GS, Waits K, Nordstrom L, Grande H, Weaver B, Papp K, Horwinski J, Koch B, Hungate BA, Liu CM, Price LB. Antibiotic-resistant *E. coli* from retail poultry meat with different antibiotic use claims. BMC Microbiol. 2018;18:1-7. DOI: 10.1186/s12866-018-1322-5
25. Mathew AG, Upchurch WG, Chattin SE. Incidence of antibiotic resistance in fecal *E. coli* isolated from commercial swine farms. J Anim Scie. 1998;76:429-34. DOI: 10.2527/1998.762429x
26. Aworh MK, Kwaga J, Okolocha E, Mba N, Thakur S. Prevalence, and risk factors for multi-drug resistant *E. coli* among poultry workers in the Federal Capital Territory, Abuja, Nigeria. PloS one. 2019;14:e0225379. DOI: 10.1371/journal.pone.0225379
27. Aworh MK, Kwaga JK, Hendriksen RS, Okolocha EC, Thakur S. Genetic relatedness of multidrug-resistant *E. coli* isolated from humans, chickens, and poultry environments. Antimicrob Resist Infect Contr. 2021;10:1-3. DOI: 10.1186/s13756-021-00930-x
28. Rahman MM, Husna A, Elshabrawy HA, Alam J, Runa NY, Badruzzaman AT, Banu NA, Al Mamun M, Paul B, Das S, Rahman MM. Isolation, and molecular characterization of multidrug-resistant *E. coli* from chicken meat. Scie Reports.2020;10:1-1. DOI: 10.1038/s41598-020-78367-2
29. Shrestha A, Bajracharya AM, Subedi H, Turha RS, Kafle S, Sharma S, Neupane S, Chaudhary DK. Multi-drug resistance and extended-spectrum beta-lactamase-producing Gram-negative bacteria from chicken meat in Bharatpur Metropolitan, Nepal. BMC Res Notes.2017;10:1-5. DOI: 10.1186/s13104-017-2917-x
30. Szmolka A, Nagy B. Multidrug resistant commensal *E. coli* in animals and its impact on public health. Front Microbiol.2013;4:258. DOI: 10.3389/fmicb.2013.00258
31. Shim JB, Seo KW, Kim YB, Jeon HY, Lim SK, Lee YJ. Molecular characteristics of extended-spectrum and plasmid-mediated AmpC β-lactamase-producing *E. coli* isolated from commercial layer in Korea. Poultry Scie. 2019;98:949-56. DOI: 10.3382/ps/pey41

الأنماط الظاهرية المقاومة للأدوية المتعددة من الإشريكية القولنية المنتجة للبيتا لاكتاميز الواسعة الطيف (ESBL) من الدجاج البياض

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الخلاصة

مقاومة مضادات الجراثيم هي مشكلة صحية عامة متنامية وناشئة في جميع أنحاء العالم. يحدد هذا البحث ظهور جراثيم الإشريكية القولونية المنتجة لأنزيمات البيتا لاكتاميز واسعة الطيف (Extended-spectrum beta-lactamases ESBL E. coli) وملامح مقاومة المضادات الجرثومية للإشريكية القولونية على قشر البيض من الدجاج البياض المختار. أخذت مسحات من قشور ٢٧٠ عينة بيض للكشف عن وجود الإشريكية القولونية. تم الحصول على ٧٣ (٢٣٪) عزلة من الإشريكية القولونية من مجموع العينات (٢٧٠ عينة). إخضعت العزلات لاختبار الحساسية للمضادات الحيوية باستخدام ستة أنواع من المضادات الحيوية (الأمبيسيلين، التتراسيكلين، السلفاميثوكسازول - تريميثوبريم، الجنتاميسين، الإيميبينيم، والسيبروفلوكساسين) باستخدام طريقة الانتشار للأقراص. أظهرت العزلات أعلى مقاومة للأمبيسيلين (٩٥,٩٪)، تليها التتراسيكلين (٨٩٪)، سلفاميثوكسازول-تريميثوبريم (٧٢٪) ، الجنتاميسين (٤١,١٪)، والإيميبينيم (١,٤٪). كما أن ٧٨٪ من العزلات كانت متعددة المقاومة للمضادات الجرثومية. إعتماداً على الزرع على وسط ESBL CHROM agar كانت ٥٦ عزلة (٧٦,٧٪) من المجموع الكلي (٧٣ عزلة) تبدو من نوع ESBL-E. coli وبإستخدام طريقة انتشار القرص المزدوج ثبت ان ٥٦/٤٢ (٧٥٪) من العزلات كانت من نوع E. coli المنتجة لـ .ESBL أظهرت نتائج الاستبيان أن جميع حقول الدجاج البياض تستخدم مضادات الجراثيم للأغراض العلاجية أو الوقائية. أيضًا ، لم يكن لدى جميع الحقول إجراءات أمن حيوي مناسبة. أشارت نتائج هذه الدراسة إلى أن قشور البيض هي مصادر وخزائن محتملة لجراثيم الإشريكية القولونية المقاومة للأدوية المتعددة و المنتجة لـESBL.