Research article

Ciprofloxacin resistant *Escherichia coli*: characterisation of phylogeny, virulence and antimicrobial resistance of isolates from extra intestinal infections of humans and chickens in selected geographical locations of Sri Lanka

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Abstract

Introduction: Ciprofloxacin resistant *Escherichia coli* strains are widely distributed. The phylogenetic backgrounds, antimicrobial resistance profiles, and virulence genes present in ciprofloxacin-resistant *E. coli* strains isolated from chickens and humans were evaluated in the study to identify their connection.

Method: A total of 92 non-duplicate *E. coli* isolates collected between July 2020 to July 2021 from humans with urinary infections (n=57) and chickens with colibacillosis (n=35) were included in the study. Antimicrobial resistance of the isolates was determined using the disk diffusion method following EUCAST guidelines. Phylogroup, plasmid-mediated quinolone resistance genes, and virulent genes were identified using PCR.

Results: Results revealed a high occurrence of ciprofloxacin resistance, with 65% of *E coli* isolates from urinary tract infections (UTI) in humans and 100% of chicken *E. coli* isolates showing resistance. Human strains showed significant co-resistance to third- and fourth-generation cephalosporins ($p<0.001$), whereas chicken isolates were co-resistant to amoxicillin-clavulanate ($p<0.001$) and trimethoprim-sulfamethoxazole ($p<0.001$). Ciprofloxacin resistant human isolates (24%) showed co-resistance to carbapenems. Phylogenetic analysis indicated ciprofloxacin resistant chicken and human *E. coli* isolates belonged to different lineages. Human *E. coli* strains were predominantly in phylogroup B2 of which 58% were O25b-ST131. Chicken isolates were distributed across multiple phylogroups. A significant proportion of ciprofloxacin resistant chicken *E. coli* harbored the *qnrS* gene while human isolates harboured *the qnrB* gene. Variations in the rates of occurrence of virulence genes, *cvaC, iutA, traT, papC, and hlyA* among humans and chickens *E. coli* isolates were also observed.

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Conclusion: There were discernible differences in co-resistant profiles, plasmid-mediated *qnr* genes, and virulent genes between *E. coli* strains derived from humans and chickens.

Key words: E. coli, ciprofloxacin resistant, human, chicken, virulence

Introduction

The emergence of multidrug resistant (MDR) bacterial clones is being increasingly reported throughout the world.[1] These MDR strains have caused severe threats to public health. Hence, antimicrobial resistance has become a serious concern of this century.[2] Previous studies have shown that excessive use and misuse of antimicrobial drugs in livestock and poultry have contributed significantly to the emergence of antimicrobial resistant strains.[3] Many countries have therefore taken measures to minimise the use of antimicrobial drugs in poultry and livestock and have banned the use of antibiotics as growth promoters.[4]

Antibiotics are still used to control bacterial infections in poultry and livestock. Of those antibiotics, quinolones are often used in commercial chicken production due to broad spectrum activity and the availability of poultry specific products. Quinolones are also widely used to treat infections in humans.[5] Clinical bacterial isolates showing resistance to quinolones are increasingly detected throughout the world.[6] The commercial poultry industry is often scrutinised for the emergence and spread of quinolone resistance in *E. coli* strains, assuming that these resistant strains are transmitted to humans through poultry-based food. To test this connection, we determined the occurrence of ciprofloxacin resistance among *E. coli* isolated from commercial broilers with lesions suggestive of colibacillosis and *E. coli* isolated from urinary tract infections (UTI) of humans, and performed a comparative analysis of the phylogenetic backgrounds, antimicrobial resistance profiles, and virulence gene profiles in these isolates.

Material and methods

Sample collection

Isolates identified as coliforms upon culturing urine samples of humans with suspected urinary tract infections were obtained from the microbiology laboratory at the Teaching Hospital, Peradeniya from July 2020 to July 2021. Coliforms isolated from liver, bone marrow and heart blood of diseased chickens with clinical signs and post-mortem lesions suggestive of colibacillosis were obtained from the Veterinary Research Institute, Gannoruwa, Veterinary Investigation Center, Wariyapola and poultry diagnostic laboratory at the Ceylon Grain Elevators, Colombo during the same time period.

Samples were cultured on *E. coli* chromogenic medium (Hi Media, India) and characteristic blue colonies were identified tentatively as *E. coli* and sub-cultured on nutrient agar (Oxoid, UK).

Confirmation of isolates as *E. coli* using PCR

All isolates from humans and chickens were confirmed as *E. coli* using PCR by amplifying the *trpA* gene (Table 1) specific to *E. coli* following the protocol described by Clermont et al. (2009).[7]

Antimicrobial susceptibility testing (AST)

Isolates confirmed as *E. coli* were screened for ciprofloxacin resistance by the disk diffusion method (European Committee on Antimicrobial Susceptibility Testing: EUCAST version 7, January 2019). *E. coli* ATCC 25922 was used as the quality control strain. Ciprofloxacin resistant isolates were subjected to antimicrobial susceptibility testing (AST) for 11 more antibiotics, including ampicillin (AMP:10 µg), amoxicillin-clavulanate (AMC: 20-10 µg), cefuroxime (CFX:30 µg), ceftriaxone (CRO:30 µg), cefotaxime (CTX:5 µg), ceftazidime (CAZ: 30 µg), cefepime (FEP:30 µg), cefpodoxime (CPD:10 µg) ciprofloxacin (CIP: 5 µg), sulphamethoxazole-trimethoprim (SXT: 1.25/23.75 µg), imipenem (IM:10 µg), amikacin (AK: 30 µg), and gentamicin (CN: 10 µg) (Oxoid, UK) following the disk diffusion method and isolates were categorized as susceptible or resistant to particular antibiotics based on EUCAST clinical breakpoints (EUCAST, 2019).

Identification of phylogenetic groups

Genomic DNA of the isolates were extracted by the boiling method. The quadruplex PCR described by Clermont and colleagues (2013)[9] was used to assign *E. coli* strains (n=92) into the phylogroups. PCR mixtures (20 µl) consisted of 4 µl of 5x buffer, 2 µl of 25 mM MgCl2, 20 pmol of each primer (Table 1), each dNTP (Promga, USA) of 5 µM, 2.5 U of Taq polymerase (Himedia HigenoMB®) and 1 µl of genomic DNA.

Identification of *E. coli* O25b-ST131

Isolates belonging to phylogroup B2 were screened for O25b-ST131 using a PCR assay described earlier.[7] Primers O25pabBspe.F (5'-TCCAGCAGGTGCTGGATCGT-3') and O25pabBspe.R (5' -GCGAAATTTTTCGCCGTACTGT-3') were used for the assay. Each reaction was carried out in a 20 µl mixture containing 5x buffer, 1.5 mM $MgCl_2$, 20 pmol of each of the *pabBspe* primers and 12 pmol of each of the *trpA* primers, 2 mM each dNTP (Promega, USA)1 U of Taq DNA polymerase (Promega, USA).

Plasmid mediated quinolone resistance genes

Multiplex PCR assay to detect plasmid-mediated quinolone resistance encoding genes, *qnrA*, *qnrB*, and *qnrS* was performed as described by Cattoir et al.(2007)[10] in a 50 µl reaction mixture containing 10x buffer with 25 mM MgCl2, 0.25 µl dNTP (Promega, USA), 10 pmol of each of the six primers (Table 1), and 1 U of Taq DNA polymerase (Promega, USA).

Table 1: Primer sequences used to identify phylogroup, plasmid associated quinolone resistance genes and virulence associated genes

Gene	Sequence 5'–3'	size (bp)
To confirm isolates as *E. coli*		
trpA[7]	trpA5′-GCTACGAATCTCTGTTTGCC-3′ trpA2. 5′-GCAACGCGGCCTGGCGGAAG-3′	
E. coli Phylogrouping [9]		
chuA	chuA 5′-ATGGTACCGGACGAACCAAC-3′ chuA.2 5′-TGCCGCCAGTACCAAAGACA-3	288
yjaA	yjaA 5′-CAAACGTGAAGTGTCAGGAG-3' yjaA.2b 5′-AATGCGTTCCTCAACCTGTG-3′	211
TspE4C2	TspE4.C2 5′-CACTATTCGTAAGGTCATCC-3′ TspE4C2.2b5′-AGTTTATCGCTGCGGGTCGC-3′	152
AceK.f	arpA 5′-AACGCTATTCGCCAGCTTGC-3′ ArpA1.r 5′-TCTCCCCATACCGTACGCTA-3′	400
Plasmid mediated quinolone resistance genes [10]		
qnrA	F: AGAGGATTTCTCACGCCAGG R: TGCCAGGCACAGATCTTGAC	580
qnrB	F: TTTGCYGYYCGCCAGTCGAA R: TTTGCYGYYCGCCAGTCGAA	264
qnrS	F: GCAAGTTCATTGAACAGGGT R: TCTAAACCGTCGAGTTCGGCG	428
Virulence-associated genes [8]		
papC	F: GTGGCAGTATGAGTAATGACCGTTA R: ATATCCTTTCTGCAGGGATGCAATA	205
hlyA	F: AACAAGGATAAGCACTGTTCTGGCT R: ACCATATAAGCGGTCATTCCCGTCA	1177
cvaC	F: CACACACAAACGGGAGCTGTT R: CTTCCCGCAGCATAGTTCCAT	697
traT	F: GGTGTGGTGCGATGAGCACAG R: CACGGTTCAGCCATCCCTGAG	290
iutA	F: GGCTGGACATCATGGGAACTGG R: CGTCGGGAACGGGTAGAATCG	302

Determination of virulence encoding genes

The presence of five virulence-associated genes, namely *papC*, *iutA*, *hlyA*, *cvaC*, and *traT*, was determined using PCR assays[11].

PCR mixtures (20 µl) consisted of 2 µl of 5x buffer, 1 µl of 25 mM $MgCl_2$, 1.5 µl of each primer (Table 1), 0.2 µl of dNTP (Promega, USA), 0.2 µl of *Taq* polymerase (Promega, USA) and 2 µl of genomic DNA.

All PCR assays were performed in a thermal cycler (Bioer®, China) in 0.2 mL PCR tubes. PCR amplicons were analysed by gel electrophoresis on 2% agarose gel, and the amplicons were visualised by staining with Diamond® nucleic acid dye (Promega, USA).

Statistical analysis

Isolates were regarded as independent samples, since each isolate was derived from a unique host and represented one episode of extra intestinal infection of that host. Statistical associations between resistance identities and categorical variables were performed using the "R" software (version 4.0.3) (http://www.R-project.org/). Categorical variables were compared

using Pearson's $\chi 2$ test with Yates continuity correction or Fisher's exact test. Fisher's exact test was used when comparing groups containing less than 5 isolates)

Results

When the bacterial isolates collected from different laboratories were subjected to the PCR assay, 92 isolates were confirmed as *E. coli*. These isolates comprised 57 isolates obtained from humans and 35 isolates from commercial chickens.

Ciprofloxacin resistance among human and chicken *E. coli* isolates

Among the 57 human UTI isolates, 37 were resistant to ciprofloxacin while all 35 *E. coli* isolates collected from commercial poultry were ciprofloxacin resistant. The variation in antimicrobial resistance profiles of *E. coli* isolates according to the host was assessed and results summarised in Table 2.

Table 2: Antimicrobial resistance of ciprofloxacin resistant *E. coli* isolated from chickens with colibacillosis and humans with urinary tract infections

	Human isolates (n=37)		Chicken isolates (n=35)			Total (n=72)	
	n	%	n	%	*p* value	n	%
Ampicillin (AMP)	35	94.6	33	94.3	1	68	94.4
Amox. clavulanate (AMC)	18	48.6	35	100.0	0.000002	53	73.6
Sulfa. trimethoprim (STX)	26	70.3	33	94.3	0.01	59	81.9
Cefotaxime (CTX)	21	56.8	1	2.9	0.000006	22	30.6
Ceftriaxone (CRO)	24	64.9	2	5.7	0.000001	26	36.1
Cefepime (FEP)	24	64.9	3	8.6	0.000002	27	37.5
Ceftazidime (CAZ)	21	56.8	14	40.0	0.23	35	48.6
Cefpodoxime (CPD)	27	73.0	3	8.6	0.1	30	41.7
Gentamicin (CN)	18	48.6	16	45.7	0.98	34	47.2
Amikacin (AK)	7	18.9	7	20.0	1	14	19.4
Imipenem (IMP)	9	24.3	0	0	-	9	12.5
Multidrug resistance	34	91.9	32	91.4	-	66	91.6.

There was a significant variation in the resistance pattern of *E. coli* isolated from the two hosts as shown in Table 2.

When compared with chicken isolates, ciprofloxacin-resistant human *E. coli* isolates displayed co-resistance to third generation cephalosporins, namely ceftriaxone ($p< 0.001$) and cefotaxime ($p<0.001$), and for fourth generation cephalosporins (cefepime) ($p<0.001$). Except for ceftazidime, chicken *E. coli* displayed minimal resistance frequencies (less than 8.6%) to other third/fourth generation cephalosporins tested. Around 11% of ciprofloxacin isolates were co-resistant to imipenem, all of which were from humas (Table 2).

Phylogenetic distribution of EXPEC

Table 3: Phylogenetic distribution of ciprofloxacin resistant *E. coli* isolated from humans and chickens and ciprofloxacin susceptible *E. coli* isolated from humans

Phylogroup	Ciprofloxacin resistant		Ciprofloxacin susceptible
	urinary (human) isolates	colibacillosis (chicken) isolates	urinary (human) isolates
A	1	4	2
B1	1	8	1
B2	31	8	13
D	1	4	0
E	3	11	4

Distribution of ciprofloxacin resistant human and chicken *E. coli* isolates and ciprofloxacin susceptible isolates in different phylogroups is listed in Table 3. Of the 31 ciprofloxacin-resistant human *E. coli* isolates that belonged to phylogroup B2, 18 were O25b-ST131, while none of the phylogroup B2 isolates from chickens belonged to that clone.

Occurrence of plasmid mediated *qnr* genes

A multiplex PCR assay was used to identify plasmid mediated *qnr* genes and PCR amplicons with 584 bp, 264 bp and 428 bp were identified as *qnrA, qnrB* and *qnrS* respectively. *qnrA*, *qnrB* and *qnrS* were detected in five (colibacillosis =1, UTI = 4), 15 (UTI = 6, colibacillosis = 9) and 29 (UTI =7, colibacillosis = 22) isolates respectively.

When the distribution of plasmid mediated quinolone resistance encoding genes in relation to the host species was assessed, qnrS gene showed significantly high association with chicken *E. coli* isolates ($p < 0.001$). When phylogenetic distribution of these three quinolone resistant genes was evaluated, *qnrA* gene was present in phylogroup B2 (4/5 isolates). The other two genes, *qnrB* and *qnrS,* were mostly present in the isolates of the phylogroups B2 and E respectively. Of the 18 phylogroup E isolates, 11 contained *qnrS* with seven of 52 phylogroup B2 isolates carrying the gene. Fifteen isolates harboring *qnrB* gene were distributed in the phylogroups, A (n=2), B1 (n=3), E (n=4), and B2 (n=6) (Table 4). Of the isolates carrying plasmid mediated *qnr* genes, 13/15 isolates containing *qnrB* gene, 25/27 isolates containing *qnrS* gene and three of the five isolates carrying *qnrA* gene displayed phenotypic resistance to ciprofloxacin.

Table 4: Occurrence of plasmid mediated quinolone resistance encoding genes and virulence encoding genes among ciprofloxacin resistant *E. coli* isolates belonging to different phylogroups.

	Phylogroup				
Gene	**A (n=7)**	**D (n=5)**	**B1 (n=10)**	**E (n=18)**	**B2 (n=52)**
Plasmid mediated *qnr* genes					
qnr A	0.0% (0)	0.0% (5)	10.0% (1)	0.0% (0)	7.7% (4)
qnr B	28.6% (2)	0.0% (5)	30.0% (3)	22.2% (4)	11.5% (6)
qnr S	71.4% (5)	40.0% (2)	40.0% (4)	61.1% (11)	13.5% (7)
Virulence encoding genes					
cvaC	14.3% (1)	40.0% (2)	10.0% (1)	27.8% (5)	11.5% (6)
traT	71.4% (5)	40.0% (2)	70.0% (7)	94.4% (17)	59.6% (31)
iutA	28.6% (2)	80.0% (4)	20.0% (2)	55.6% (10)	63.5% (33)
papC	0.0% (0)	0.0% (0)	10.0% (1)	22.2% (4)	42.3% (22)
hlyA	0.0% (0)	0.0% (0)	0.0% (0)	5.6% (1)	9.6% (5)

Occurrence of virulent genes

Isolates were screened by PCR assays for the presence of *cvaC*, *iutA*, *traT*, *papC* and *hlyA* genes. A total of 38 isolates had more than one virulence encoding gene (i.e: 25 isolates had two genes, seven had three genes, four had four genes and two human isolates had all five genes. Nine isolates did not contain any of the five virulence encoding genes.

Distribution virulence genes particularly, *iutA* and *papC* were similar among ciprofloxacin resistant and ciprofloxacin susceptible *E. coli* isolates. The frequency of occurrence of *cvaC*, *iutA*, *traT*, *papC* and *hlyA* genes in 72 ciprofloxacin resistant *E. coli* isolates were 16.7% (12), 51.4% (37), 66.7% (48), 25.0% (18) and 6.9% (5) respectively.

Around 80% of chicken *E. coli* isolates contain *traT* gene while it was only present in 59% of human *E. coli* isolates. The occurrence of *traT* gene was significantly high ($p = 0.037$) among chicken isolates. and the occurrence of *iutA* ($p = 0.002$) and *papC* genes ($p = 0$) was significantly high in human *E. coli* isolates (Fisher's exact test).

Of the 52 phylogroup B2 isolates, 31 (59.6%), 33 (63.4%) and 22 (42%) isolates contained *traT*, *iutA* and *papC* genes respectively. Of those three genes only *papC* showed significant association with phylogroup B2 ($p = 0.004$).

Discussion

Our results confirm the high occurrence of ciprofloxacin resistance among chicken and human *E. coli* isolates. Around 65% of *E. coli* isolated from urinary tract infections in humans and all the isolates (100%) collected from colibacillosis in chickens were resistant to ciprofloxacin. A study carried out in 2016 in Sri Lanka reported around 46% ciprofloxacin resistance among *E. coli* isolated from humans.[12] In comparison, our results indicate a higher rate of resistance to ciprofloxacin among human urinary *E. coli* isolates. However, due to the limited number of samples collected and differences in the study areas, it is difficult to say whether resistance to ciprofloxacin shows an increasing trend. Other South Asian countries, including India (73%), and Pakistan (84.2%), have also reported high rates of ciprofloxacin resistance among uropathogenic *E. coli* (UPEC).[13,14]

Compared to human isolates, the isolates from chickens displayed higher rates of resistance to ciprofloxacin (100%). A study conducted in 2022 in Sri Lanka also reported a similar rate of ciprofloxacin resistance (around 97%) among *E. coli* isolates obtained from chickens.[15] When compared to the reported ciprofloxacin resistance among avian pathogenic *E. coli* (APEC) isolated from commercial chicken in Bangladesh, Thailand and Australia, ciprofloxacin resistance among avian extra intestinal *E. coli* is extremely high in Sri Lanka.[16,17] The higher resistance rates to ciprofloxacin in chicken *E. coli* isolates in Sri Lanka could be due to the excessive and indiscriminate use of fluoroquinolones (enrofloxacin) to treat diseases and subsequent environmental contamination in poultry farms. This highlights the necessity of legislation to regulate access to antimicrobial agents and restrict their use in commercial chicken production in Sri Lanka.

To investigate whether the ciprofloxacin-resistant isolates of humans and chickens are connected, the phylogenetic background of the ciprofloxacin-resistant isolates was evaluated in this study.

Several studies have demonstrated substantial overlaps in serogroups, phylogenetic groups, and virulence genotypes between APEC and UPEC strains. This genetic similarity has raised concerns about APEC serving as potential reservoirs of virulence and antimicrobial-resistance genes for human extra intestinal pathogenic *E. coli* strains. However, the current study showed that the ciprofloxacin-resistant strains of humans and chickens are phylogenetically different. Around 84% of ciprofloxacin-resistant *E. coli* of human origin belonged to phylogroup B2, and ciprofloxacin-resistant strains isolated from chickens were distributed in all phylogroups, with the highest percentage in phylogroup E followed by B1 and B2. The present study mainly focuses on pathogenic *E. coli* strains isolated from poultry, and it would be important to do further studies including commensal *E. coli* isolates in chickens and humans to gain a better understanding of this relationship.

Ciprofloxacin-resistant human isolates showed a significantly high rate of resistance to 3rd and 4th generation cephalosporins, while ciprofloxacin-resistant chicken *E. coli* isolates showed a low level of resistance to these antibiotics. All ciprofloxacin-resistant chicken *E. coli* isolates showed co-resistance to clavulanate-amoxicillin whereas a smaller proportion of ciprofloxacin-resistant human isolates showed resistance to clavulanate-amoxicillin. The above-mentioned differences in antimicrobial resistance profiles of chicken and human *E. coli* also suggest there is no clonal relationship between ciprofloxacin resistance in human and chicken *E. coli*.

A high proportion of ciprofloxacin-resistant human isolates in this study belonged to O25b-ST131, while none of the chicken isolates belonged to that clone. Previous studies conducted in Sri Lanka reported a high occurrence of ST131 in extraintestinal infections in humans.[18] ST131 belongs to phylogroup B2 and has developed co-resistance to fluoroquinolones (FQs) and extended spectrum cephalosporins (ESCs).[19] The fact that a large proportion of extra intestinal pathogenic *E. coli* isolates belong to a cluster that is exclusive to humans suggests that most of the human pathogenic strains in the current study have no connection to those present in chickens.

Of the three *qnr* genes tested in this study, the *qnrS* gene showed a significant association with ciprofloxacin-resistant chicken *E. coli* isolates. A similar rate of occurrence of the *qnrS* gene in chicken *E. coli* isolates has been reported previously.[20] The *qnrB* gene was also present in 25% of chicken *E. coli* isolates, and the *qnrA* gene was rarely detected. It is important to do further molecular-level analysis to identify whether the isolates carrying *qnr* genes are closely related.

Fluoroquinolone-resistant *E. coli* in poultry is reported to contain mutations in *gyrA* and *parC* genes. These chromosomally encoded fluoroquinolone-resistant genes were not evaluated in our study and analysing those mutations would provide further insight.

Limitations: In the present study, we compared human and chicken *E. coli* isolates that were not epidemiologically linked to each other. Analysis of *E. coli* isolated from humans and chickens with close ecological or occupational association (eg. farm workers regularly exposed to diseased chickens) would provide better insight into the possible transmission of ciprofloxacin-resistant *E. coli* strains from chickens to humans.

Conclusion

The findings of this study suggest that ciprofloxacin-resistant, extra intestinal *E. coli* isolated from humans and chickens in selected geographical locations in Sri Lanka exhibit phylogenetic differences. The well-defined ciprofloxacin-resistant clone, O25b-ST131 was predominant among humans but absent among avian isolates. Additionally, co-resistant profiles, plasmid-mediated *qnr* genes, and virulence genes differed between the *E. coli* isolates from humans and chickens.

Declarations

Conflict of interest: There are no conflicts of interest
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Ethics Statement: Approval for the study was obtained from the ethical clearance committees of the Faculty of Medicine, University of Peradeniya (2019/EC/67) and the Faculty of Veterinary Medicine and Animal Science, University of Peradeniya (VERC-19-9), Sri Lanka.
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References

1. Pitout JD, Peirano G, DeVinney R. The contributions of multidrug resistant clones to the success of pandemic extra-intestinal pathogenic Escherichia coli. *Expert Rev Anti infect Ther.* 2023; 21(4):343-353. *https://doi.org/10.1080/14787210.2023.2184348*
2. Salam MA, Al-Amin MY, Salam MT et al. Antimicrobial resistance: a growing serious threat for global public health. *Healthcare*. 2023; 11(13):946. *https://doi.org/10.3390/healthcare11131946*
3. Ghimpețeanu OM, Pogurschi EN, Popa DC et al. Antibiotic use in livestock and residues in food—A public health threat: *A Rev Foods*. 2022; 16:11(10):1430. *https://doi.org/10.3390/foods11101430*
4. Woodford N, Carattoli A, Karisik E et al. Complete nucleotide sequences of plasmids pEK204, pEK499, and pEK516, encoding CTX-M enzymes in three major Escherichia coli lineages from the United Kingdom, all belonging to the international O25: H4-ST131 clone. *Antimicrob Agents Chemother*.2009; *53*(10):4472-4482. *https://doi.org/10.1128/aac.00688-09*
5. Yoon MY, Kim YB, Ha JS et al. Molecular characteristics of fluoroquinolone-resistant avian pathogenic *Escherichia coli* isolated from broiler chickens. *Poult Sci*. 2020; *99*(7):3628-3636. *https://doi.org/10.1016/j.psj.2020.03.029*
6. Dalhoff A. Global fluoroquinolone resistance epidemiology and implications for clinical use. *Interdiscip Perspect Infect Dis 2012:*(1):976273. *https://doi.org/10.1155/2012/976273*
7. Clermont O, Dhanji H, Upton M *et al*. Rapid detection of the O25b-ST131 clone of Escherichia coli encompassing the CTX-M-15-producing strains. *J Antimicrob Chemother*. 2009; *64*(2):274-277. *https://doi.org/10.1093/jac/dkp194*
8. Johnson JR, & Stell AL. Extended Virulence Genotypes of *Escherichia coli* Strains from Patients with Urosepsis in Relation to Phylogeny and Host Compromise. *J Infect Dis.* 2000; 181(1):261-272. *https://academic.oup.com/jid/article/181/1/261/893888*
9. Clermont O, Christenson JK, Denamur E *et al*. The Clermont *Escherichia coli* phylo-typing method revisited: improvement of specificity and detection of new phylo-groups. *Environ Microbiol Rep*. 2013; *5*(1):58-65. *https://doi.org/10.1111/1758-2229.12019*

10. Cattoir VL, Poirel V, Rotimi CJ et al. Multiplex PCR for detection of plasmid-mediated quinolone resistance qnr genes in ESBL-producing enterobacterial isolates, *J Antimicrob Chemother*. 2007; 1:60(2):394-7. *https://doi.org/10.1093/jac/dkm204*
11. Senadheera GPSG, Ranganathan SS, Patabendige G et al. Resistance and utilisation pattern of antibacterial agents in outpatient settings in two Teaching Hospitals in Colombo. *Ceylon Med J*. 2016; *61*(3): 113-117. *doi: 10.4038/cmj.v61i3.8346*
12. Yamamoto S, Terai A, Yuri K et al. Detection of urovirulence factors in Escherichia coli by multiplex polymerase chain reaction. *FEMS Immunol Med Micro*. 1995; *12*(2):85-90. *https://doi.org/10.1111/j.1574-695X.1995.tb00179.x*
13. Mandal J, Acharya NS, Buddhapriya D et al. common bacterial uropathogens with a special reference to ciprofloxacin resistant *Escherichia coli*. *Indian J. Med. Res*. 2012; *136*(5):842-849.
14. Muhammad I, Uzma M, Yasmin B *et al*. Prevalence of antimicrobial resistance and integrons in *Escherichia coli* from Punjab, Pakistan. *Braz. J. Microbiol*. 2011; *42*:462-466. *https://doi.org/10.1590/S1517-83822011000200008*
15. Priyantha MAR, Pabasara ABS, Fernando PS *et al*. Investigation into the quinolone resistant E. coli isolated from commercial broilers. *Asian J Res Anim Vet Sci*. 2022; *9*:22-27.
16. Saha O, Hoque MN, Islam OK et al. Multidrug-resistant avian pathogenic Escherichia coli strains and association of their virulence genes in Bangladesh. *Microorganisms*. 2020; *8*(8):1135. *https://doi.org/10.3390/microorganisms8081135*
17. Thomrongsuwannakij T, Blackall PJ, Djordjevic SP et al. A comparison of virulence genes, antimicrobial resistance profiles and genetic diversity of avian pathogenic Escherichia coli (APEC) isolates from broilers and broiler breeders in Thailand and Australia. *Avian Pathol*. 2020; *49*(5):457-466. *https://doi.org/10.1080/03079457.2020.1764493*
18. Dewasmika WP, Dissanayake DA, Neelawala ND *et al*. High Occurrence of ST131 Among Extraintestinal Pathogenic Escherichia coli in Humans and Dogs in Sri Lanka. *Zoonoses and Public Health*. 2025; 72(2):200-2006. *https://doi.org/10.1111/zph.13199*
19. Woodford N, Carattoli A, Karisik K et al. Complete nucleotide sequences of plasmids pEK204, pEK499, and pEK516, encoding CTX-M enzymes in three major Escherichia coli lineages from the United Kingdom, all belonging to the international O25: H4-ST131 clone. *Antimicrob. Agents Chemother*. 2009; 53(10):4472-82. *https://doi.org/10.1128/aac.00688-09*
20. Murase T, Phuektes P, Ozaki H *et al*. Prevalence of qnrS-positive Escherichia coli from chicken in Thailand and possible co-selection of isolates with plasmids carrying qnrS and trimethoprim-resistance genes under farm use of trimethoprim. *Poult Sci*. 2022; *101*(1):101538. *https://doi.org/10.1016/j.psj.2021.101*.