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Comparison of Antimicrobial Resistance in Escherichia coli Strains Isolated from Diseased Broiler and Cattle based on Phenotypic and Genotypic Analysis.

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ABSTRACT

Escherichia coli infections in poultry and animals have become a global problem. Currently, little is known regarding the epidemiology of extended-spectrum beta-lactamases (ESBLs) producing *Escherichia coli* in infected poultry and cattle. This study aimed to identify antibiotic resistance patterns and ESBL-producing genes in E. coli strains recovered from diseased poultry and animals in northern Egypt. A total of 100 consecutive, no duplicate clinical samples from diseased broiler poultry and 50 samples from diseased cattle were collected during a one-year period. ESBL was found in 44% (24 out of 54) isolates of E. coli isolated from infected poultry positive for double disc diffusion, and 66% (16 out of 24) isolates of E. coli recovered from cattle positive for double disc diffusion. ESBL genes (*blaCTX-M1, blaCTX-M2, blaOXA10, blaSHV, blaTEM, blaOXA-2, blaVEB, blaPER-2*, and *blaGES*) were detected in strains by multiplex polymerase chain reaction with gene -specific primers. From 40 phenotypically positive ESBL isolates, we discovered that blush, *blaTEM, blactx-m1*, and *blactx-m9* (100%) were the most prevalent genotypes in cattle isolates, whereas *blaGes* and *blactx-m9* (70% of isolates from poultry). This is the first study to look at ESBL resistance patterns and ESBL-producing genes in E. coli isolated from poultry and cattle. Our analysis found varying occurrences among isolates from various hosts.

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INTRODUCTION

Avian pathogenic Escherichia coli (APEC), an extra-intestinal pathogenic E. coli (ExPEC), causes diverse local and systemic infections in poultry. Colibacillosis is one of the leading causes of mortality (up to 20%) and morbidity in poultry, as well as decreased meat (2% decline in live weight, 2.7% deterioration in feed conversion ratio) and egg production (up to 20%), reduced hatching rates, and increased carcass condemnation (up to 43%) at slaughter (**Dho-Moulin and Fairbrother 1999**, **Guabiraba and Schouler 2015**) Furthermore, APEC causes substantial mortality in young chicks for (up to 53.5%) (**Mellata, 2013**).

Cattle are one of the main sources of animal protein, becoming one of the most consumed meat and milk around the world, one of the main constituents of the human food chain (**Alexandratos and Bruinsma, 2012**). It is also one of the main sources of biological fertilizers, due to the high production of fecal mass of these animals (**Smith and Williams, 2016).** All this highlights the importance of cattle production in the context of the food chain and the contaminated environment as reservoirs and transmitting/disseminating vehicles of E-ESBL, thus configuring a threat to the world's public health. This circulation of E-ESBL within our ecosystem creates a consensual concern among the scientific community and the authorities involved in the One Health approach (**Robinson et al. 2016**).

Cattle diarrhea is frequently caused by enterotoxigenic Escherichia coli (ETEC), the most prevalent colibacillosis in newborn calves (**Nagy and Fekete, 2005**). In Egypt, new natal calf diarrhea (NCD) remains the leading cause of calf death, and bacterial infections are still responsible for more than half of episodes of diarrhea in neonatal calves (**Kumar et al. 2012**). Enterotoxigenic Escherichia coli (ETEC) and Salmonellae are the most economically significant pathogens (**Achá et al. 2004**).

Antibiotics are commonly used to treat colibacillosis in poultry and calves (**Agunos et al. 2012**). Resistance to antibiotics, particularly β-lactams, colistin, and carbapenems, poses issues for controlling APEC infections in hens (**Nhung et al. 2017**). Furthermore, there is no effective vaccination available to protect hens from APEC infections, owing to the wide range of APEC serotypes linked with colibacillosis cases in field outbreaks (**Ghunaim et al. 2014**).

Drug resistance in E. coli has risen rapidly during the last decade over the world. The rise is mostly due to an increase in ESBL-producing E. coli (**Hawkey and Jones, 2009**).

Beta-lactamases are bacterial enzymes that confer resistance to beta-lactam antibiotics, such as penicillin derivatives and cephalosporin by hydrolyzing the beta-lactam ring. In recent years, new types of beta-lactamase enzymes including extended-spectrum beta-lactamases (ESBLs) and AmpC beta-lactamases have emerge **(Paterson 2006 and Babic et al. 2006)**.

ESBL genes are found on plasmids, which may be easily transported between and among bacterial species. Some ESBL genes are mutant variants of existing plasmid-mediated β-lactamases (e.g., *blaTEM*/SHV), while others come from environmental bacteria (e.g., *blaCTX-M*). The majority of ESBL gene reports in the 1990s focused on *blaTEM/SHV* forms, which were associated with hospital cross-infections. However, the current global surge has been mostly driven by *blaCTX*-M -type genes. The epidemiology of ESBL genes is quickly evolving, with significant

regional disparities in the distribution of *blaCTX*-M β-lactamase genotypes **(Paterson DL et al. 2005). Comparisons** of Escherichia coli amino acid sequences revealed nine structural and evolutionary families, including TEM, SHV, CTX-M, PER, VEB, GES, BES, TLA, and OXA.5-7 The four main subtypes of ESBL variations are OXA, TEM, SHV, and CTX-M **(Munita J and Arias C, 2016).**

The purpose of this work was to identify the presence of ESBL-encoding genes (*blaCTX-M1, blaCTX-M2, blaOXA10, blaSHV, blaTEM, blaOXA-2, blaVEB, blaPER-2*, and *blaGES*) in Escherichia coli isolated from chicken and cattle, as well as to investigate their antimicrobial resistance profile.

MATERIALS and METHODS:

2.1. Sample Collection

Ruffled feathers, loss of appetite, and respiratory distress were reported on 100 diseased broiler chicken farms (five hens per farm, ages ranging from 14 to 45 days). In parallel, 50 fecal samples were obtained from infected cattle. Samples were gathered from several geographical regions in northern Egypt (Menoufia, Giza, and Qalubia Governorates) between September 2022 and December 2023. samples were transferred to the Reference Laboratory for Veterinary Quality Control in Poultry Production and submitted to postmortem inspection under aseptic circumstances. Samples were collected from the organs (liver, lung, and heart) of birds with coli septicemia, air vasculitis, perihepatitis, and pericarditis and pooled for bacterial screening and isolation. All sample collection procedures were legally approved by the Committee of Ethics at the Animal Health Research Institute, Egypt, under protocol number **(AHRI -42429)**.

2.2. Isolation and Identification of E. coli:

E. coli was isolated and identified using the methods reported (**Nolan et al. 2013**). Briefly, samples were aerobically incubated in buffer peptone water at 37°C for 24 hours. A loopful of each incubated sample was streaked onto MacConkey's agar (Oxoid, Manchester, UK) and Eosin Methylene Blue agar (Lioflichem, Roseto degli Abruzzi, Italy) plates, which were then incubated aerobically at 37°C for 24 hours. The probable colonies were 1-2 mm in diameter, with a hot-pink color on MacConkey and metallic sheen colonies on Eosin Methylene Blue agar. Suspected E. coli colonies underwent further biochemical testing (indole test, methyl red, Voges Proskauer "VP", citrate utilization, oxidase test, and Triple Sugar Iron "TSI"). Serotyping E. coli by agglutination test This was performed by using rapid diagnostic E. coli antisera sets (DENKA SEIKEN CO., Japan) at the serological department in the Animal Health Research Institute, Dokki, Giza, Egypt as described by **Kok, et al. (1996).**

2.3. Antimicrobial Susceptibility Pattern and ESBL Screening of the Isolated E. coli

2.3.1 Screening for Extended-Spectrum β Lactamases Using Disc Diffusion.

Using a traditional disc diffusion method, the isolates were tested with these medications (30 µg of cefotaxime or ceftriaxone). Zones measuring 27mm or less for cefotaxime and 25mm or less for ceftriaxone were identified as potential ESBL producers and verified using double disc diffusion testing. **(Nipa et al 2016)** and **(Sheetal et al 2023).**

2.3.2 Double Disc Diffusion Testing indicates Extended-Spectrum β Lactamase.

Discs containing cefotaxime with and without clavulanic acid (10 µg) were put 20 mm apart on a Muller Hinton agar (Hi-Media) plate previously inoculated with the test organism and incubated at 37°C for 18 hours. Combining an antimicrobial agent and clavulanic acid resulted in a 5mm or greater increase in zone width compared to the antibiotic alone, indicating a positive test. The test was performed by the **Clinical and Laboratory Standards Institute document (CLSI, 2021).**

2.3.3 Antibiotic test susceptibility for ESBL-producing isolates.

The antibiotic resistance pattern of ESBL-producing isolates to a screen of antibiotics was determined using the Kirby Bauer disc diffusion method following CLSI standards. The isolates were tested against the different antibiotics: norfloxacin (10 µg), trimethoprim and sulfamethoxazole (1.25/23.75 µg), ceftriaxone (30 µg), cefotaxime (30 µg), norfloxacin (µg), amoxicillin-clavulanic (µg), and tetracycline (30 µg). Colistin

2.4 DNA extraction and amplification by PCR:

DNA was extracted from 50 *E. coli* isolates using the Wizrep g DNA Mini kit Extraction (cell /Tissue), REF. W71060-100. We used EmeraldAmp® GT PCR Master Mix (Code No. RR310A) for the PCR amplification of Extended-spectrum β-lactamases (ESBLs). We added 10 µl of Master Mix. We added 3 µl of water, 1.5 µl of each primer (ESBLs), at 20 pmol (forward and reverse primers), and 4 µI of the sample's extracted DNA. The reactions involved one cycle of initial denaturation at 96°C for three minutes, then 40 cycles of denaturation at 98°C for thirty seconds, annealing at 58°C to 67°C According to ESBL genes for thirty seconds **table (1)**, and extension at 72°C for one minute, concluding with one cycle at 72°C for five minutes.

Gel electrophoresis:

Products for amplification Agarose gel electrophoresis was used to examine the PCR, and ethidium bromide staining and a gel documentation system were used to visualize the results. The combination of amplicon and gel loading buffer (50% glycerol/0.1M EDTA, pH 8.0/1% SDS/0.1% bromophenol blue/0.0% xylene cyanole) was loaded into 1.5% agarose in 1x TBE (89 mM tris/89 mM boric acid/2 mM EDTA, pH 8.0) The size standard was a 100-bp ladder (Gibco, BRL).

Table 1. Specific primers for ESBL genes detection of poultry and Cattle

ESBL genes	Forward	Reverse	Annealing Temperatures
blaTeM	ATG AGT ATT CAA CAT TTC CGT	TTA CCA ATG CTT AAT CAG TGA	58
blaVeB	GCC AGA ATA GGA GTA GCA AT	TGG ACT CTG CAA CAA ATA CG	58
blages	TAC TGG CAG SGA TCG CTC AC	TTG TCC GTG CTC AGG ATG AG	62
blaPeR	CTC AGC GCA ATC CCC ACT GT	TTG GGC TTA GGG CAG AAA GCT	62
blashV	CGC CTG TGT ATT ATC TCC CTG	TTA GCG TTG CCA GTG CTC GAT	64
blaOXa2	ATG GCA ATC CGA ATC TTC GC	GCA CGA TTG CCT CCC TCT T	60
blaOXa10	ATG AAA ACA TTT GCC GCA TAT G	TTA GCC ACC AAT GAT GCC CT	60
blaCTX-M 1	AGT TCA CGC TGA TGG CGA CG	GAC GAT TTT AGC CGC CGA CG	67
blaCTX-M 9	GCG TGC ATT CCG CTG CT G C	ACA GCC CTT CGG CGA TGA TTC	67

RESULTS:

3.1. E. coli Isolation, Identification, and Serotyping:

Out of 100 tested farms,54 were positive for *E. coli* isolation from poultry, with a rate of 54%. *E. coli* isolates were recovered from internal organs (liver, lung, and heart) of 100 diseased broiler chicks collected from farms in Menoufia, Giza, and Qalubia governorates, as well as 24 positive diseased samples from cattle distributed throughout 50 farms (48%). E. coli isolates were recognized as TSI acidic at the slant and bottom, producing gas, positive for catalase, methyl red, and indole, and negative for VP, oxidase, and citrate.

Serological investigation of chicken isolates revealed the following serotypes: O25.O55, O125, O157, O1, O126, O158, O169, O6, O148, and O18. Cattle isolates, on the other hand, are classified into the following serotypes: O158, O125, O119, O78, O44, O55, O157, and O126.

3.2 The antimicrobial susceptibility pattern of the isolated E. coli.

3.2.1 Extended-Spectrum β Lactamase Screening and Confirmatory Testing.

Phenotypic screening studies revealed that some E. coli isolates were resistant to third generation cephalosporins **(table 2).**

Table 2. culture and ESBL of poultry and cattle *E. coli* isolates

Source of Bacterial isolates	ESBL screening by the disk diffusion method (Resistance)		ESBL confirmation by double disk diffusion testing
	Cefotaxime (30 µg) *N** (%)	Ceftriaxone (30 µg) *N** (%)	Cefotaxime (30 µg) with clavulanic acid (10 µg)
E. coli isolated from poultry	54 (100%)	40 (74%)	24 (53%)
E. coli isolated from cattle	21 (87 %)	24 (100%)	16 (66%)
Total	75(96%)	64 (82%)	(51.2%)

Notes: The number of E. coli isolated was 54 from poultry and 24 from cattle. total E. coli isolates 78. The latter N* as the number of *E. coli* strains resistant for specific antibiotic-Zones measuring 27mm or less for cefotaxime and 25mm or less for ceftriaxone were identified as potential ESBL producers and verified using double disc diffusion testing. Combining an antimicrobial agent and clavulanic acid resulted in a 5mm or greater increase in zone width compared to the antibiotic alone

3.2.2. Antimicrobial Sensitivity Test (AST):

A total of 54 chicken and 24 cow E. coli isolates were examined shown in **Table (3)**. A similar trend was observed among the chicken and cattle isolates, where the majority of the chicken isolates were resistant to cefotaxime 100%, doxycycline 59.2%, and sulfamethoxazole-trimethoprim 57.4%, but isolates from cattle were resistant to doxycycline and sulfamethoxazole-trimethoprim 25%, and, as shown in Table 3, the sensitive antibiotic for E. coli was colistin 100% in poultry and cattle, and norfloxacin 100% in cattle but 74% poultry. Table 3 shows a detailed percentage of resistant, intermediate, and susceptible isolates to each antibiotic based on the CLSI standard.

Table 3. Antibiotic sensitivity trends in Escherichia coli strains isolated from diseased poultry and cattle

Antimicrobial agent	Resistant No (%)		Intermediated No (%)		Sensitive No (%)	
	Poultry	cattle	Poultry	cattle	Poultry	Cattle
Colistin (10µg)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	54(100%)	24(100%)
Ceftriaxone (30 µg),	14 (25.9%)	3 (12.5%)	12 (22.2%)	6 (25%)	28 (51.8%)	15 (62.5%)
Doxycycline's (30 µg),	32 (59.2%)	6 (25%)	0 (0%)	3(12.5%)	22 (40.7%)	15(62.5)
Norfloxacin (10 µg),	12 (22.2%)	0 (0%)	2(3.7)	0 (0%)	40 (74%)	24(100%)
Sulphur trimethoprim (25 µg),	31 (57.4%)	6(25%)	3 (5.5%)	3(12.5%)	20(37%)	15(62.5)
Fosfomycin (200 µg),	9 (16.6%)	3(12.5%)	3 (5.5%)	0 (0%)	42 (77.7%)	21(87.5%)
Cefotaxime (30 µg),	54(100%)	3(12.5%)	0(0%)	15(62.5)	0 (0%)	3(25%)
Amoxicillin clavulanic acid (10 µg)	14 (25.9%)	0 (0%)	13(24%)	15(62.5)	27 (50%)	9(37.5)

Notes: Percentage as calculated according to No of isolates from each species (The number of E. coli isolated 54 from poultry and 24 from cattle).

Table (4): Antibiotic resistance trends in ESBL-producing and nonproducing Escherichia coli isolated from diseased poultry and livestock

Antimicrobial agent	E. coli isolated from poultry		E. coli isolated from cattle		Total ESBL-producing E. coli (N=40)	Total non-ESBL producing E. coli (N=38)
	ESBL-producing E. coli (N=24)	Non-ESBL-producing E. coli (N=30)	ESBL-producing E. coli (N=16)	Non-ESBL-producing E. coli (N=8)		
Colistin (10 µg)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Ceftriaxone (30 µg),	10(41%)	4(13%)	2(12.5%)	1(12.5%)	12 (30%)	5 (13%)
Doxycyclines (30 µg),	22 (91 %)	10(33 %)	5(31 %)	1(12.5 %)	27(67.5 %)	11 (28.9 %)
Norfloxacin (10 µg),	9 (37.5 %)	3(10 %)	0	0	9 (22.5 %)	3 (7.8 %)
Sulpha tri (25 µg),	21(87.5 %)	10(33.3 %)	3(18.7 %)	3 (18.7 %)	24 (60 %)	13 (34 %)
Fosfomycin (200 µg),	6 (25 %)	3 (10 %)	2 (12.5 %)	1 (12.5 %)	8(20 %)	4 (10.5 %)
Cefotaxime (30 µg),	23 (95 %)	21 (70 %)	2 (12.5 %)	1 (12.5 %)	25 (62.5 %)	22 (58 %)
Amoxicillin clavulanic acid (10 µg)	10 (41 %)	4 (13%)	0 (0%)	0 (0 %)	10 (25 %)	(10.5 %)

Notes: The number of ESBL-producing E. coli isolated 24 from poultry and 16 from cattle. The number of non-ESBL-producing E. coli isolated was 30 from poultry and 8 from cattle. Percentage as calculated according to the No of E. coli isolates (ESBL & Non-ESBL).

Molecular Detection:

Drug resistance was confirmed by culture and sensitivity testing on 78 isolates. Genes linked to resistance were found in 40 isolates (24 isolates from poultry and 16 isolates from cattle). **Table 5 and Figure 1** show that the ratio of all screened ESBL genes was different in poultry and cattle. The bla_{TEM} gene was detected in 27 isolates (16 were cattle, a percentage of 64%, and 11 were poultry, a percentage of 44%) that amplified at 861 base pairs. The bla_{VEB} gene was detected in 23 isolates (5 were cattle, a percentage of 20 %, and 18 were poultry, with a percentage of 72 %) that were amplified at 703 base pair. The bla_{GES} gene was detected in 25 isolates (8 were cattle, a percentage of 32 %, and 17 were poultry, a percentage of 68%) that amplified at 838 base pairs. The bla_{PER} gene was detected in 16 isolates (7 were cattle, a percentage of 28 %, and 9 were poultry, a percentage of 36 %) that amplified at 851 base pairs. The bla_{SHV} gene was detected in 23 isolates (16 were cattle, a percentage of 64%, and 7 were poultry, a percentage of 28 %) that amplified at 849 base pairs. The bla_{OXA2} gene was detected in 15 isolates (3 were cattle, a percentage of 12%, and 12 were poultry, a percentage of 48 %) that amplified at 670 base pairs. The bla_{OXA10} gene was detected in 18 isolates (4 were cattle, a percentage of 22 %, and 14 were poultry, a percentage of 78 %) that amplified at 801 bas pair. The bla_{CTXM1} gene was detected in 29 isolates (16 were cattle, a percentage of 64 %, and 13 were poultry, a percentage of 52 %) that amplified at 839 base pairs. The bla_{CTXM9} gene was detected in 33 isolates (16 were cattle, a percentage of 64 %, and 17 were poultry, a percentage of 68 %) that amplified at 832 base pairs.

Table 5. PCR Amplification of Extended-spectrum β-lactamases (ESBLs) in Escherichia coli isolates from both cattle and poultry.

ESBL genes	Escherichia coli isolates from cattle fecal samples	Escherichia coli isolate from poultry organs	Product Size
blaTeM	16	11	861 bp
blaVeB	5	18	703 bp
blages	8	17	838 bp
blaPeR	7	9	851 bp
blashV	16	7	849 bp
blaOXa2	3	12	670 bp
blaOXa10	4	14	801 bp
blaCTX-M1	16	13	839 bp
blaCTX-M9	16	17	832 bp

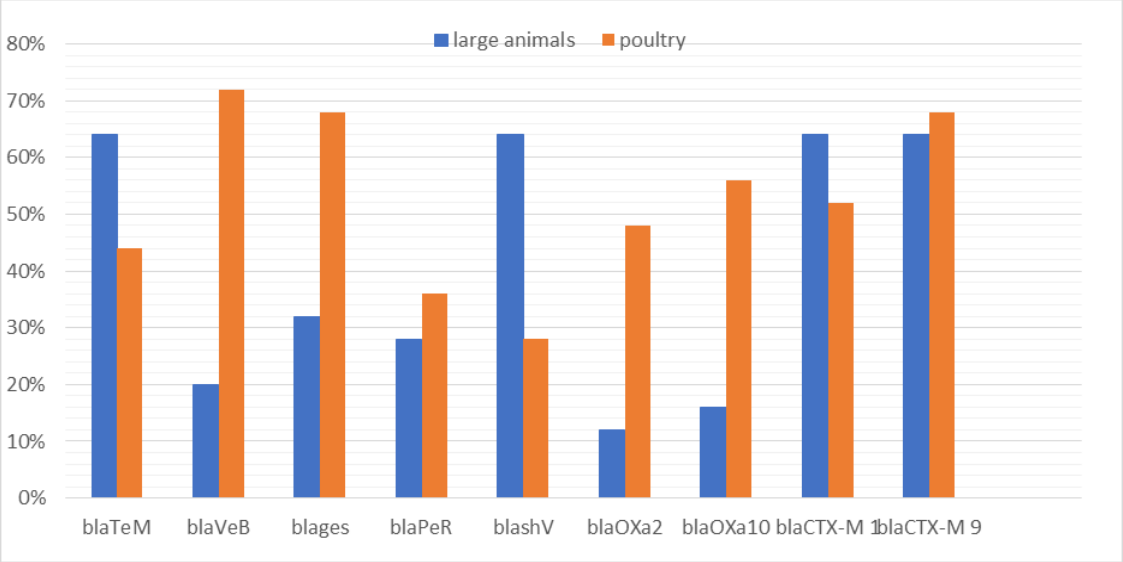

Figure 1: percentage of ESBL genes in cattle and poultry E. coli isolate

DISCUSSION

Extended-spectrum beta-lactamases (ESBLs) resistance bacterial strains present a significant therapeutic problem for veterinarians, as there are few treatment options for illnesses caused by ESBL-producing bacteria. The discovery of genes implicated in ESBL-mediated resistance is often done for monitoring, epidemiological investigations, and control of resistance spread in poultry and cattle farms. (**Prasada et al. 2019)** In our investigation, the total incidence of ESBL production was 51.2%, with 44% E. coli recovered from poultry and 66.6% isolated from cattle. The global incidence of ESBL production among clinical isolates is ranged from less than one to 88%. (Cockerill **et al. 2013; Bora et al. 2014 and Veeraraghavan et al. 2018)**, another study showed that the prevalence of ESBL-producing E. coli is very high, as 100% of tested farms were positive) (**Dierikx et al. 2012)**, and 77 to 94% of all retail chicken meat is contaminated with ESBL-producing E. coli bacteria **(Overdevest et al. 2011 and Leverstein-van Hall et al. 2011),** E. coli. detected that **(**32.8%) were contained ESBL-producing E. coli also this study recorded that the prevalence of ESBL-producing E. coli was much higher in calves (55.7%) than in cows (37.3%) in South Africa **Schmid et al. (2013),** also identified the isolates from cattle feces and beef samples as ESBL producing strains with the percentage of (76.4%).

In our investigation *E. coli* isolates from poultry were most susceptible to colistin, followed by fosfomycin, ceftriaxone, and amoxicillin clavulanic acid, whereas isolates from cattle were most susceptible to colistin and norfloxacin, followed by ceftriaxone **(Tivendal et al. 2010)**. Colistin has been shown in studies to be successful in the treatment of diseased cattle and poultry caused by MDR gram-negative bacteria, although it should be used with caution due to its potential for nephrotoxic effects. **(Karakkattu et al. 2017)**. In our study detected that poultry isolates were 100% resistant to cefotaxime while strains isolated from cattle were resistant to cefotaxime by 12.5%. this agrees with **(Montso, et al. 2019)** the resistance of Cefotaxime ranged from 14.3– 100%. Similar observations had been reported by **(Iweriebor, et al. 2015 and Olowe et al. 2015)** resistance prevalence in cattle isolates (25%) was significantly lower than in swine and poultry, and poultry isolates were 57.4% resistant to sulphamethoxazole trimethoprime, but isolates from diseased cattle were 25% resistant to sulphamethoxazole trimethoprime. this finding agreed with **Guerra et al. (2003),** which showed that most prevalent resistances were to sulfamethoxazole (≥14%) while trimethoprim–sulfamethoxazole (21%) resistance in poultry.

Both E. coli isolates (poultry and cattle) in the research exhibited substantial coresistance to various non-β-lactam antibiotics, which is concerning and poses challenge in managing E. coli infections. Only colistin had 100% action in both isolates. This is extremely troubling and warrants careful consumption of colistin. Few similar investigations have found colistin as the most sensitive antibiotic for both E. coli isolates which is in keeping with our findings, but have reported greater norfloxacin susceptibility, particularly in cattle **(Musicha et al. 2017).**

One of the fastest-growing resistance issues globally is caused by extended-spectrum β-lactamase-producing bacteria **(Petternel, et al., 2014).** According to (**Carattoli 2008),** livestock may play a significant role in the spread of ESBL-producing bacteria throughout the community. The importance of food-producing animals in Egypt has not been enough evaluated, and there is no information on the possibility that large animal and poultry meat contains ESBL-producing Enterobacterales and the genes that encode them **(Kola A et al. 2012)**.

Through this study, the results clarified similarities in the phenotypic and genotypic analysis of ESBL-producing E. coli from chickens and cattle, indicating the possibility of a transmission between the two species **(Abdallah H, et al. 2015).** Amplification of nine antibiotic resistance genes (ESBL) by polymerase chain reaction showed differences in variation between cattle animals and chickens which ranging from 670 bp to 861 bp **(Ryoo, N.H et al. 2005 and Bubpamala, J et al. 2018).** The most common resistance genes in both types are bla_{TEM}, bla_{CTXM1}, and $bla_{\text{CTXM 9}}$, This is consistent with previous studies **(Heba Badr, et al. and Abdallah H, et al. 2022).** Where The first description of an E-ESBL in cattle was in Japan, where a CTX-M-2 *E. coli* producer was detected in cattle feces from an important region close to the Centre of the country (**Shiraki et al. 2004).**

On the other hand, there are five resistance genes *blaVEB, blaGES, blaPER* blaOXA2, and blaOXA10 were mostly found in isolated bacteria from poultry organs than in large animals **(Heba Badr, et al. 2022).** However, the bla_{SHV} gene was mostly found in isolated bacteria from large animals than in poultry organs (**Abdallah H, et al. 2022).** Possibly, the difference in contamination rates between poultry and large animals owes to differences in the production system, which is more intensive in the poultry industry than in the large animal rearing system (**Smet A, et al. 2010).** There may be ecological repercussions from the spread of ESBL-producing bacteria in wild bird populations, including changes to the microbial community's equilibrium and an impact on the well-being of the animals and their environment. Maintaining surveillance for the existence of ESBL-producing bacteria in populations of wild birds and putting preventative measures in place are essential **(World Health Organization, 2021)**.

CONCLUSION

Escherichia coli infections in poultry and animals are a global issue, but the spreading of extended-spectrum beta-lactamases (ESBLs) producing Escherichia coli in infected poultry and cattle is limited. A study aimed to identify antibiotic resistance patterns and ESBL-producing genes in E. coli strains recovered from diseased poultry and cattle. This draws attention to the importance of continued surveillance and the acquisition of more samples, particularly from cattle and poultry, in order to gain a better understanding of how resistance genes were transmitted. Furthermore, wide genetic investigations, including whole genome sequencing, are required to gain a better understanding of the genetic relationship between poultry and cattle isolates.

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Dr.Mohamed I. AbdAllah and **Dr.Mervat E. Hamdy** designed the study and analyzed the PCR data. Also, all authors contributed to the writing and revision**. Dr. Ahmed Shabaan** and **Dr. Mai M. Morsy,** cultured and isolated bacteria from samples. **Dr. Heba Farouk and Dr. Ghada S. Abdelhamed,** data curation of isolate, and sensitivity. **Dr. Nada Th. Momen** collected samples from farms, numbered them, and preserved them. All authors have read and agreed to the published version of the manuscript.

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Conflict of interest:

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.