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PHENOTYPIC AND GENOTYPIC CHARACTERIZATION OF INTRAUTERINE PATHOGENIC *ESCHERICHIA COLI* WITH MULTI-DRUG RESISTANCE ISOLATED FROM COW UTERI WITH ENDOMETRITIS

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ABSTRACT

Endometritis is a common postpartum disorder affecting cows, leading to financial losses. *E. coli* is among the most prominent clinically significant pathogens responsible for severe cases of bovine endometritis. 110 uterine swab samples from cows from several veterinary clinics at Kaliobia Governorate, Egypt (75 cows had subclinical endometritis and 35 cows had clinical endometritis) were used in order to isolate and identify pathogenic *E. coli* and to assess their susceptibility to antibiotics, in addition to their genotypic and phenotypic description of virulence and antimicrobial resistance genes. Our findings provided that endometrial *E. coli* was isolated from 54 examined uterine samples (49.1%). The identified *E. coli* was sensitive to norfloxacin, followed by gentamycin, ciprofloxacin, and co-trimoxazole, whereas remarkably resistant to oxacillin, ampicillin, tetracycline, cefotaxime, cephapirin, and streptomycin. All 54 isolated *E.coli* had Congo Red binding activity, and also all the 45 isolates showed phenotypic evidence of biofilm development. PCR results revealed that *fim*H was present in all five studied *E. coli* strains, and three of them contained *pap*C virulence genes. Antimicrobial resistant genes *bla*TEM , *sul*1, *tet*A(A) were found in all studied strains, *aad*A1 in four strains and three strains exhibited the $bla_{\text{CTX-M}}$ gene. Therefore, according to the results, the recovered *E. coli* was endometrial pathogenic *E. coli* (EnPEC) with multiple antibiotic resistance. Furthermore, the virulence activities and phenotypic resistance to the antibiotics correlated strongly with the presence of the genes *fim*H, *pap*C, bla_{TEM}, $bla_{\text{CTX-M}}$, *aad*A1, *sul*1, *tet*A(A) in these strains.

Keywords: Endometrial pathogenic *Escherichia coli*, endometritis, Congo Red, PCR.

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INTRODUCTION

Postpartum diseases affecting dairy cows can have a significant negative impact on reproduction, as well as economic and

productivity losses (Rocha *et al.*, 2025). *Escherichia coli* is a predominant pathogen associated with subclinical and clinical endometritis in cows and increases the susceptibility of the uterus to subsequent infections by *Trueperella pyogenes* and other bacterial species, leading to enormous economic losses (Sheldon *et al*., 2010; Sheldon and Owens, 2017; Ma *et al*., 2018; Gonzalez *et al*., 2020 and Zhang *et al*., 2024; Chabanenko *et al.,* 2024).

Important pathogenicity traits of Endometrial pathogenic *E. coli* (EnPEC) include adhesion of epithelial cells, invasiveness of endometrial cells, motility mediated by flagella, exotoxins, and lipopolysaccharides, which form the primary biofilm structure and contribute to host immunity and bacterial resistance to antibiotics (Sheldon *et al*., 2010; Carniello *et al*., 2018 and Ostapska *et al.*, 2018). Compared to non-pathogenic *E. coli*, the endometrial epithelial and stromal cells responded to lipopolysaccharide (LPS) extracted from EnPEC by producing more prostaglandin E2 and interleukin-8 (Sheldon *et al*., 2010 and Ostapska *et al.,* 2018). EnPEC possesses numerous virulence factor genes (VFG), such as *fim*H, *pap*C, and *hly*A, which may enable bacteria to attach, colonize cow genitalia, and induce cows' endometritis (Bicalho *et al*., 2012; Ma *et al*., 2018 and Gonzalez Moreno *et al*., 2020), where *fim*H (a type 1 pilus component) is *E. coli* unique gene and substantially related with metritis and endometritis in cows (Gonzalez Moreno *et al*., 2020 and Adiguzel *et al.,* 2021). Antimicrobial therapy is therefore hampered by EnPEC's capacity to form biofilm. Intrauterine antibiotic infusions continue to be the most effective treatment for endometritis and uterine infections in dairy cows. However, prolonged and irregular antibiotic use, coupled with a steadily increasing level of drug resistance, is gradually increasing the prevalence of multidrug resistant (MDR) strains of EnPEC (Senosy and Hussein 2013; Mekibib *et al*., 2024 and Zhang *et al.*, 2024).

The most significant factor in the occurrence of antibiotic resistance in EnPEC was determined as the existence of genes encoding extended-spectrum β-lactamases (ESBLs), including CTX-M, TEM, SHV, that can enzymatically degrade a variety of β-lactamase antibiotics and exhibited MDR, indicating they are resistant to both β-lactam and non-β-lactam antibiotics (Hijazi *et al.,* 2016; Tekiner and Ozpinar, 2016 and Zhang *et al*., 2024).

Furthermore, streptomycin (*aadA*1), sulfonamide (*sul*1), and tetracycline *tet*A(A) are among the chromosomal and plasmid-encoded genes linked to anti-microbial resistance in EnPEC isolated from the uteri of cows suffering from metritis and endometritis (Goldstone *et al*., 2014; Raheel *et al.,* 2020 and Tabaran *et al*., 2022).

As *E. coli* is considered one of the main pathogens linked to endometritis, and in Egypt, little research has been done on the traits of bovine uterine *E. coli* and their association with virulence factors and uterine disorders like endometritis. Therefore, the purpose of this study was to determine the prevalence of pathogenic *E. coli* strains that were isolated from cow uteri that had both clinical and subclinical endometritis, in addition to their phenotypic and genotypic characterization of virulence and antimicrobial resistance genes.

MATERIALS AND METHODS

Ethical approval:

The Bioethics Committee has approved the proposal entitled “Phenotypic and genotypic characterization of intrauterine pathogenic *Escherichia coli* with multi-drug resistance isolated from cow uteri with endometritis” to meet requirements of the Faculty of Veterinary Medicine, Benha

University research ethics, Egypt under approval number (BUFVTM 18-03-25).

Animals and uterine sample collection:
The current investigation was carried out on 110 pluriparous dairy cows from several veterinary clinics within the Kaliobia Governorate, Egypt, between September 2021 and February 2025 (35 cows with clinical endometritis and 75 cows with repeat breeding issues due to subclinical endometritis). Following a clinical diagnosis, uterine swab samples were obtained for each animal using a special catheter of Noakes *et al*. (1989) modified by Maarouf *et al*. (2013). Each swab was collected and directly put in screw-capped tubes with nutrient broth next to the flame, and sent at once to the laboratory for *E. coli* isolation and identification, aside from their phenotypic and genotypic characterization of virulence and antimicrobial resistance genes.

Isolation and Identification of *Escherichia coli* isolates:
One ml of each screw-capped tube was inoculated in 9.0 ml of MacConkey broth and stayed in the incubator for 24 hours at 37 ºC to facilitate primary enrichment for *E. coli* isolation. A loopful of every enrichment culture was streaked onto MacConkey agar plates (CM115, Oxoid, UK) and then incubated at 37 ºC for 24 hours. Suspected lactose fermented *E. coli* colonies (small, round, and bright pink-colored colonies) were obtained and streaked on selective media: Eosin methylene blue (EMB) agar (Oxoid, UK); Tryptone Bile Glucuronide (TBX) agar (3650192, Oxoid, UK, Iso 16649), after that, it was incubated at 37 ºC for an additional 24–48 hours. Purified *E. Coli* colonies were then kept in semi-solid agar for phenotypic and biochemical identification using tests as outlined by Iso (2001), Quinn *et al*. (2011), and Markey *et al*. (2013). These tests included Eijkman, catalase, oxidase, sugar fermentation, nitrate reduction, methyl red, Voges-Proskauer, citrate utilization, urease test, indole, and hydrolysis of gelatin. Additionally, the method outlined by Edward and Ewing (1972) was used to serotype five randomly selected *E. coli* isolates.

Test of *in-vitro* antimicrobial sensitivity:
Standardized antimicrobial discs (Oxoid), ampicillin (AM/10 µg), cefotaxime (CTX/30 µg), gentamicin (GEN/10 µg) Cephapirin (CEPR/30 µg), Ciprofloxacin (CIP/5 µg), Streptomycin (S/10 µg), Doxycycline (DO/30 µg), oxacillin (Ox/10 µg), co-trimoxazole (COT/25 µg), norfloxacin (NOR/10 µg), tetracycline (TE/30 µg), and amoxicillin/clavulanic acid (AMC/30 µg) on Mueller-Hinton agar (Oxoid, UK) plates were used for each *E. coli* isolate for the test of *in-vitro* antimicrobial sensitivity using the disc method of CLSI (2018).

Phenotypic virulence activities for isolates of *E. coli*:
Congo Red (CR) dye binding activity test and biofilm production:
Qualitative biofilm formation phenotypic method was used for detection of biofilm production of all the isolates by Congo red agar method. This test was carried out in accordance with Yadav *et al*. (2014). The medium used for CR dye binding was Trypticase soy agar (TSA) with 0.2% galactose and 0.03% CR dye [addition of an iron source or galactose to CR agar medium enhances absorption of CR dye by some isolates of E. coli (Panigrahy and Yushen 1990)].

An aqueous concentrated solution of Congo red stain was first made, and it was autoclaved at 121ºC for 15 minutes apart from the other medium ingredients. It was then added to the autoclaved medium ingredients at 55 ºC. *E. coli* isolates were streaked onto CR agar plates and incubated at 37 ºC for 24 hours. The plates were further incubated at room temperature for an additional 48 hours. The colonies were

examined at 18, 24, 48 and 72 hours of incubation. The *E. coli* that produced intense orange or brick-red-colored colonies after 24, 48, and 72 hours of incubation showed a positive Congo red reaction (CR+), while pale or white colonies were thought to be Congo red (CR-) negative. CRA is a method used to determine whether isolate has the potential for biofilm production or not. The Congo red dye directly interacts with certain polysaccharides forming colored complexes or more likely some metabolic changes of the dye to form a secondary product could play a more important part in the formation of coloured colonies, Jain & Agarwal (2009) and Arciola *et al*. (2001).

Molecular identification of *E. coli* isolates' genes for virulence and antibiotic resistance

Two virulence genes, *fim*H and *pap*C as well as five antimicrobial resistance genes, tetracycline *tet*A (A), streptomycin (*aad*A1), sulphonamide (*sul*1), β-lactam (*bla*TEM), and extended spectrum β-lactam gene (*bla*CTX-M) were detected genotypically. To do this, five randomly selected isolates of *E. coli* that demonstrated a resistance to antibiotics by the disk diffusion method, as well as strong phenotypic biofilm formation, were examined using the conventional polymerase chain reaction (cPCR). In brief, the DNA of *E. coli* was extracted, following QIA amp® with Catalogue no.51304, DNA Mini Kit instructions (Qiagen, Germany, GmbH), Emerald Amp GT PCR master-mix (Takara, Japan) with Code No. RR310A and 1.5% agarose gel electrophoreses (Sambrook *et al.,* 1989) using the Primers sequences, target genes, amplicons sizes and cycling conditions (Table, 1). A positive control DNA was obtained from confirmed positive *E. coli* field isolate in RLQP (Reference lab for vet. quality control on poultry production, Dokki, Giza, Egypt). On the other hand, the negative control is a PCR mixture without the DNA template.

Table 1: Target genes, oligonucleotide primer sequences and PCR conditions.

Target gene		Primer sequence (5'-3')	Amplified segment (bp.)	Primary Denat	Amplification (35 cycles)			Final extension	References
					Secondary denat	Anneal	Exten		
*fim*H	F	TGCAGAACGGATAAGCCGTGG	508 bp.	94 ºC 5 min	94 ºC 30 sec	50 ºC 40 sec	72 ºC 45 sec	72 ºC 10 min	Ghanbarpour and Salehi, (2010)
	R	GCAGTCACCTGCCCTCCGGTA							
*pap*C	F	TGATATCACGCAGTCAGTAGC	501 bp.	94 ºC 5 min	94 ºC 30 sec	58 ºC 40 sec	72 ºC 45 sec	72 ºC 10 min	Jin *et al*., (2008)
	R	CCGGCCATATTCACATAA							
*bla*TEM	F	ATCAGCAATAAACCAGC	516 bp.	94 ºC 5 min	94 ºC 30 sec	54 ºC 40 sec	72 ºC 45 sec	72 ºC 10 min	Colom *et al*., (2003)
	R	CCCCGAAGAACGTTTTC							
*bla*CTX-M	F	ATG TGC AGY ACC AGT AAR GTK ATG GC	593 bp.	94 ºC 5 min	94 ºC 30 sec	54 ºC 40 sec	72 ºC 45 sec	72 ºC 10 min	Archambault *et al*., (2006)
	R	TGG GTR AAR TAR GTS ACC AGA AYC AGC GG							
*sul*1	F	CGGCGTGGGCTACCTGAACG	433 bp.	94 ºC 5 min	94 ºC 30 sec	60 ºC 40 sec	72 ºC 45 sec	72 ºC 10 min	Ibekwe *et al*., (2011)
	R	GCCGATCGCGTGAAGTTCCG							
*aad*A1	F	TATCAGAGGTAGTTGGCGTCAT	484 bp.	94 ºC 5 min	94 ºC 30 sec	54 ºC 40 sec	72 ºC 45 sec	72 ºC 10 min	Randall *et al.,* (2004)
	R	GTTCCATAGCGTTAAGGTTTCATT							
*tet*A (A)	F	GGTTCACTCGAACGACGTCA	576 bp.	94 ºC 5 min	94 ºC 30 sec	50 ºC 40 sec	72 ºC 45 sec	72 ºC 10 min	
	R	CTGTCCGACAAGTTGCATGA							

RESULTS

After 110 uterine swab samples with endometritis were subjected to bacteriological analysis, 54 *E. coli* isolates (49.1%) were found in {(35/75) repeat breeding cows with subclinical endometritis and (19/35) cows with clinical

endometritis}. Phenotypically, the recovered Gram-negative, medium-sized rods of *E. coli* isolates appeared as single colonies in pairs or colonies in short chain appearance. They had a distinctive greenish metallic sheen appearance on Eosin Methylene Blue media, were typical blue-colored colonies on TBX media (B-glucuronidase positive), and appeared as small, spherical, and brilliant pink-colored colonies (lactose fermenter) on MacConkey agar. Biochemically, all 54 isolates possessed the features of *E. coli*, where in Triple Sugar Iron test (TSI), the isolates showed yellow (acid formation) at slant and butt (lactose and/or sucrose attacked as well as the glucose) with gas formation and without H_2S production. Also, all isolates had positive results for Eijkman, sugar fermentation, methyl red, catalase, and indole tests, but negative results for oxidase, urease, citrate utilization, Voges-Proskauer, and hydrolysis of gelatin tests. Every isolated *E. coli* exhibited 100% motility, spreading like a paintbrush from the site of inoculation into the agar.

Five E. coli isolates were analyzed serologically, two isolates gave positive results with polyvalent antisera group (1), one isolate with group (3), and two isolates with group (5). By using monovalent antisera, only four identified *E. coli* serogroups (O86a, O119, O153, and O158) were discovered serologically, represented as one isolate from serotypes O86a, O119, & O158, and two isolates were O153.

The findings of in-vitro sensitivity testing were shown in Table (2).

Table 2: Antimicrobial sensitivity test (in vitro) for 54 isolates of *Escherichia coli*.

Antimicrobial disc		Disc concentrations	Sensitive		Intermediate		Resistant		
			No.	%	No.	%	No.	%	Aa
Oxacillin	Ox	1 µg	0	0.0	8	14.8	46	85.2	R
Ampicillin	Am10	10 µg	2	3.7	8	14.8	44	81.5	R
Tetracycline	Te/30	30 µg	4	7.4	7	13.0	43	79.6	R
Cefotaxime	Ctx/30	30 µg	4	7.4	10	18.5	40	74.1	R
Cephapirin	Cepr	30 µg	4	7.4	12	22.2	38	70.4	R
Streptomycin	S/10	10 µg	4	7.4	18	33.3	32	59.3	R
Doxycycline	Do/ 30	30 µg	12	22.2	34	63.0	8	14.8	Is
Amoxicillin/clavulanic acid	Amc	30 µg	9	16.7	31	57.4	14	25.9	Is
Norfloxacin	Nor/10	10 µg	42	77.8	8	14.8	4	7.4	S
Gentamicin	Gen/10	10 µg	37	68.5	10	18.5	7	13.0	S
Ciprofloxacin	Cip/5	5 µg	36	66.7	12	22.2	6	11.1	S
Co- trimoxazole	Cot/25	(1.25/23.75) µg	33	61.1	11	20.4	10	18.5	S

Is: Intermediate sensitivity S: sensitive R: Resistant
No.: Number of isolates AA: Antibiogram activity
%: The proportion relative to the total number of *E. coli* isolates (n=54)

Findings from phenotypic virulence activities for 54 isolated *E. coli* revealed that all of them (100%) had CR binding activity and showed development of intense orange to brick red colored colonies on CRA (CR+), so all of them are invasive and pathogenic. Moreover, by CR assay for biofilm formation, 35 *E. coli* isolates (64.8%) were grown as black colonies with a dry, crystalline consistency (strong biofilm formation); 10 (18.5%) as darkened colonies that lacked a dry, crystalline colonial appearance (moderate biofilm formation); and 9 (16.7%) as pink or pale colonies (negative, no-biofilm formation) on CR agar.

Findings of the virulence genes genotyping

detection revealed that three of the *E. coli* studied strains carried *pap*C gene, and all five studied *E. coli* strains harbored the *fim*H gene, giving products of 501 bp. and 508 bp., respectively (Figure 1-a & b). Meanwhile, the antimicrobial resistant genes *bla*TEM, *sul*1, and *tet*A (A) were found in all five studied *E. coli* strains, giving products of 516 bp., 433 bp., and 570 bp., respectively; *aad*A1 was amplified in four strains, giving products of 484bp., and three strains exhibited $bla_{\text{CTX-M}}$ gene, giving products of 593 bp. (Figures, 2- a, b & c and 3- a & b).

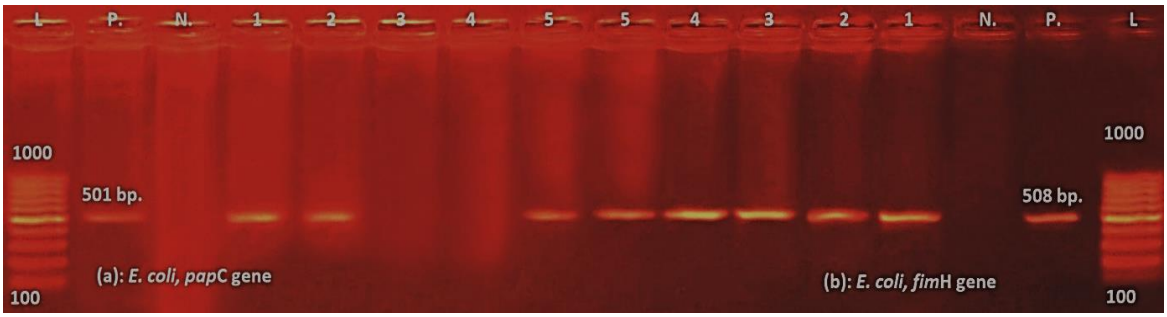

Figure (1-a). PCR screening for P fimbriae (*papC*) gene, (L) 100-1000 bp. Ladder of DNA (P) positive control {*E. coli* from RLQP at 501 bp.}, (N) negative control, lanes (1,2,5) positive amplification of the *pap*C gene in *E. coli* at 501 bp.; Lane (3,4) Negative *E. coli* at 501 bp.

Figure (1-b). PCR screening for Type 1 fimbriae (*fim*H) gene, (P) positive control {*E. coli* from RLQP at 508 bp.}, lanes (1-5) positive *E. coli* at 508 bp.

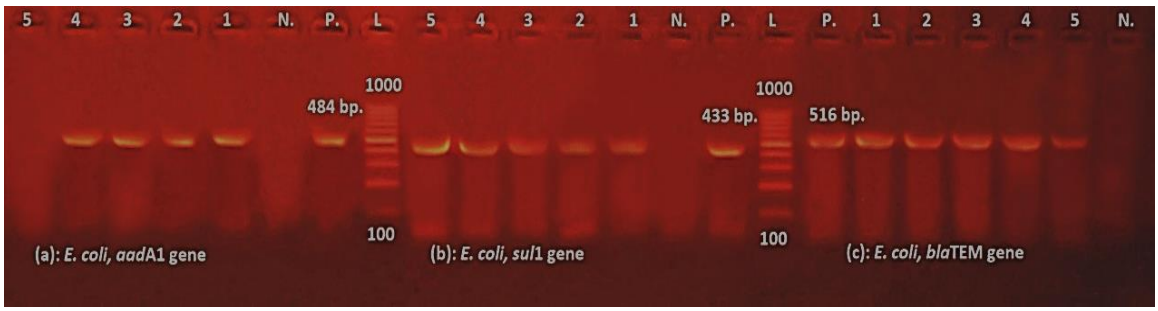

Figure (2-a). PCR screening for streptomycin resistance (*aad*A1) gene, (L) 100-1000 bp. Ladder of DNA, (P) positive control {*E. coli* from RLQP at 484 bp.}, (N) negative control, lanes (1-4) positive amplification of *aad*A1gene in *E. coli* at 484 bp.; Lane (5) Negative *E. coli* at 484 bp.

Figure (2-b). PCR screening for sulphonamide resistance (*sul*1) gene, (P) positive control {*E. coli* from RLQP at 433 bp.}, lanes (1-5) positive amplification of *sul*1 gene in *E. coli* at 433 bp.

Figure (2-c). PCR screening for β-lactam resistance (*bla*TEM) gene, (P) positive control {*E. coli* from RLQP at 516 bp.}, lanes (1-5) positive amplification of the *bla*TEM gene in *E. coli* at 516 bp.

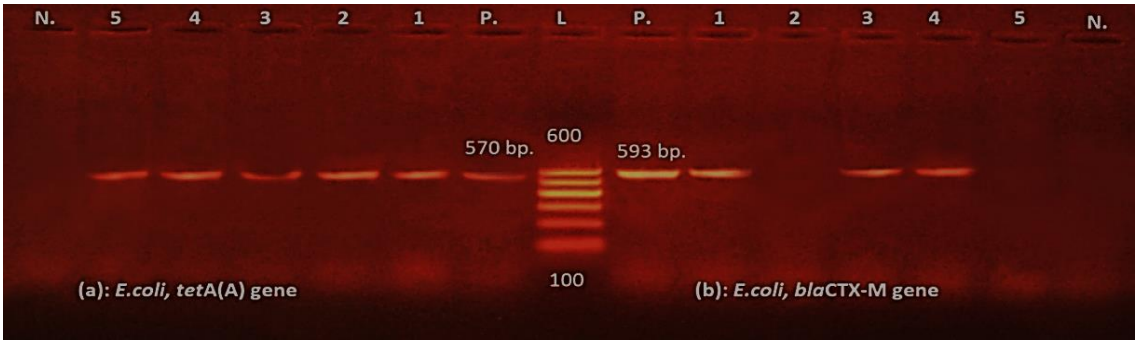

Figure (3-a). PCR screening for tetracycline resistance *tet*A (A) gene, (L) 100-600 bp. Ladder of DNA

(P) positive control {*E. coli* from RLQP at 570 bp.}, (N) negative control, lanes (1-5) positive amplification of the *tet*A (A) gene in *E. coli* at 570 bp.

Figure (3-b). PCR screening for extended spectrum β-lactam resistant (bla_{CTX-M}) gene, (P) positive control {*E. coli* from RLQP at 593 bp.}, lanes (1,3,4) positive amplification of bla_{CTX-M} in *E. coli* at 593 bp.; Lane (2,5) Negative *E. coli* at 593 bp.

DISCUSSION

According to the current study, 54 endometrial *E. coli* isolates (49.1%) were found in 110 uterine swab samples that had both subclinical and clinical endometritis. This higher prevalence of *E. coli* isolation is consistent with earlier research by Kasimanickam *et al*. (2016), Raheel *et al*. (2020), Shafique *et al*. (2021), Mekibib *et al*. (2024), and Zhang *et al*., (2024), that isolated *E. coli* from bovine endometritis cases with incidences of 45%, 48.3%, 36.7%, 45%, and 39.1%, respectively.

Our research pointed to the negligent usage of antibiotics to treat endometritis, whether it is clinical or subclinical, with improper dosage, overuse, and failure to complete the entire course of treatment, as the reason for the increase in the prevalence of certain bacteria that are resistant to antibiotics. These findings congruous with earlier studies reported by Zhao *et al.* (2014), Ma *et al.* (2018), and Zhang *et al.* (2024).

In the context of that, our findings for antimicrobial sensitivity of 54 *E. coli* isolates demonstrated their extreme resistance to oxacillin, followed by ampicillin, then tetracycline, cefotaxime, cephapirin, and streptomycin, almost the same results were attained by Maarouf *et al.* (2013), Yang *et al.* (2016), Ma *et al.* (2018), Raheel *et al.* (2020), Basbas *et al.* (2022), Mekibib *et al.* (2024), and Zhang et *al.* (2024), although they opposed Pohl *et al.* (2018), and Adiguzel *et al.* (2021), as they recorded that tetracycline and β-lactam antibiotics were effective against the isolated endometrial *E. coli*. Furthermore, among the 54 endometrial *E. coli* isolates that were studied, the obtained results showed phenotypic resistance for at least two different antimicrobials and were deemed multi-drug resistant (MDR), which was consistent with earlier research by Zhao *et al*. (2014), Ma *et al*. (2018), Shafique *et al.* (2021), Mekibib *et al.* (2024), and Zhang *et al.* (2024), who stated that *E. coli* isolated from cases of bovine endometritis were MDR. In addition, the studied endometrial *Escherichia coli* isolates were intermediately sensitive to doxycycline and amoxicillin/clavulanic acid; however, they were very sensitive to norfloxacin, followed by gentamicin, ciprofloxacin, and co-trimoxazole. These results came in harmony with those obtained by Raheel *et al.* (2020), Zhao *et al.* (2014), and Maarouf *et al.* (2013).

Bacterial virulence and resistance are mediated by biofilm formation (Cepas *et al*., 2019), where it raises the antimicrobial resistance up to 1,000-fold. To inactivate organisms developing inside biofilm higher antimicrobial concentrations are required (Ahmadi *et al*., 2017 and Schiebel *et al*., 2017).

In this work, recorded results for phenotypic virulence activities seemed to show that all 54 isolated endometrial *E. coli* had CR binding activity, and 45 of them were phenotypically positive for biofilm development (35 isolates produced biofilms strongly, and 10 isolates produced biofilms moderately). So, the identified *E. coli* are Endometrial pathogenic *E. coli* (EnPEC), and this is in accordance with earlier research by Moori Bakhtiari *et al*. (2018), Gonzalez Moreno *et al*. (2020), and Raheel *et al*. (2020).

A PCR technique can be used to identify EnPEC by detecting virulence and antimicrobial resistance genes. In our

research, the virulence-associated *fim*H, Type 1 pili gene, was associated with adherence, invasion, and biofilm development in the epithelial cells of host tissues. This gene was found in each of the five *E. coli* isolates phenotypically positive for biofilm development, and came into alignment with Kassé *et al*. (2016), Yang *et al.* (2016), Ma *et al*. (2018), Bicudo *et al.* (2019), Sheldon *et al*. (2019), Gonzalez Moreno *et al*. (2020), Raheel *et al*. (2020), and Adiguzel *et al*. (2021), who detected the *fim*H gene in EnPEC and showed that this gene is a significant predictor of metritis and endometritis in cows. Additionally, three out of five studied *E. coli* strains possessed the Fimbriae P *pap*C gene, which encodes for bacterial adhesion. The same findings were noted by Kassé *et al*. (2016), Yang *et al.* (2016), and Raheel *et al*. (2020). The existence of *fim*H and *pap*C genes positive *E. coli* in the uterus (EnPEC) increases the risk of uterine infection by other pathogenic bacteria and compromises reproduction (Sheldon *et al.,* 2010 and Yang *et al.,* 2016). Furthermore, the genotyping detection of antimicrobial resistance genes found that *bla*TEM, *sul*1, and *tet*A(A) resistance genes were amplified in all five evaluated *E. coli* strains; the aadA1 gene was present in four strains, and *bla*CTX-M was detected in three strains. Similar finding of these genes in EnPEC strains isolated from the uteri of cows with metritis, clinical and subclinical endometritis was reported by Zhao *et al*. (2014), Ma *et al*. (2018), Raheel *et al*. (2020), Shafique *et al*. (2021), Tabaran *et al*. (2022), and Zhang *et al*. (2024), for *bla*TEM, and *bla*CTX-M genes; Raheel *et al*. (2020), and Tabaran *et al*. (2022), for *sul*1, and *tet*A (A) genes; Tabaran *et al*. (2022), and Zhang *et al*. (2024), for *aad*A1 gene.

Outcomes of our investigation revealed that isolates of *E. coli* from the pluriparous dairy cows' uteri with endometrial inflammation are EnPEC, which had antimicrobial resistance to the majority of antimicrobial medications. Furthermore, virulence activities and phenotypic antibiotic resistance of *E. coli* were positively connected with the existence of *fim*H, and *pap*C virulence genes, as well as *bla*TEM, *bla*CTX-M, *aad*A1, *sul*1, and *tet*A (A) antimicrobial resistance genes in these strains. Therefore, it is advisable to treat bovine endometritis with norfloxacin, gentamicin, ciprofloxacin, and co-trimoxazole.

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التوصيف الظاهري والجيني لبكتيريا الإشريكية القولونية الممرضة داخل الرحم المتعددة المقاومة للأدوية والمعزولة من أرحام الأبقار المصابة بالتهاب بطانة الرحم

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التهاب بطانة الرحم هو اضطراب شائع يصيب الأبقار بعد الولادة، ويؤدي إلى خسائر مالية. تُعد الإشريكية القولونية (*E. coli*) من أبرز المسببات ذات الأهمية الأكلينيكية، والمسؤولة عن الحالات الشديدة من التهاب بطانة الرحم في الأبقار.
تم تجميع ١١٠ عينة مسحة رحمية من أبقار من عدة عيادات بيطرية في محافظة القليوبية بمصر (٧٥ بقرة مصابة بالتهاب بطانة الرحم تحت الأكلينيكي و٣٥ بقرة مصابة بالتهاب بطانة الرحم الأكلينيكي) لعزل وتحديد البكتيريا الإشريكية القولونية المسببة للحالات وتقييم مدى حساسيتها للمضادات الحيوية، بالإضافة إلى التوصيف الظاهري والجيني لجينات الضراوة ومقاومة المضادات الحيوية.
أظهرت النتائج عزل الإشريكية القولونية من ٥٤ عينة رحمية (٤٩,١٪) كانت العترات المعزولة حساسة للنورفلوكساسين، تليها الجنتاميسين، السيبروفلوكساسين، الكوتريموكسازول، بينما أظهرت مقاومة ملحوظة الأوكساسيلين، الأمبيسيلين، التتراسيكلين، السيفوتاكسيم، السيفابيرين والستربتوميسين. جميع العترات المعزولة (٥٤ عترة) أظهرت الارتباط بصبغة الكونغو الحمراء و دليلا ظاهريا على تكون الأغشية الحيوية. أظهرت نتائج الكشف عن جينات الضراوة عن وجود جين *fim*H فى جميع الخمس العترات المختبرة، واحتوت ثلاث منها على جين الضراوة *pap*C. تم الكشف على جينات مقاومة للمضادات الحيوية *bla*TEM، *sul*1 و *tet*A(A) فى جميع الخمس العترات المختبرة، ووجود جين *aad*A1 فى أربع عترات، وظهور جين bla_{CTX-M} فى ثلاث عترات فقط.
وفقًا للنتائج، تعتبر الإشريكية القولونية المعزولة ممرضة لبطانة الرحم (EnPEC) و متعددة المقاومة للعديد من المضادات الحيوية. علاوة على ذلك، عوامل الضراوة والمقامة الظاهرة للمضادات الحيوية مرتبطة ارتباطا وثيقا بوجود الجينات *fim*H، *pap*C، *bla*TEM، bla_{CTX-M}، *aad*A1، *sul*1 و *tet*A(A) فى هذه العترات المعزولة.