KUFA JOURNAL FOR VETERINARY MEDICAL SCIENCES

https://journal.uokufa.edu.iq/index.php/kjvs

Phenotypic Identification and Antibiotic Resistance Profile of *Escherichia coli* Isolated from Lambs with Colibacillosis in Mosul, Iraq

Khalid Waleed Muhammad and Mohammad A. Hamad*

Department of Microbiology, College of Veterinary Medicine, University of Mosul. Mosul, Iraq.

ABSTRACT

Neonatal diarrhea and colibacillosis generally lead to critical morbidity and mortality rates in affected sheep flocks caused by Escherichia coli (E. coli). The aim of this study was to detect and isolate E. coli from lambs with dysentery and their antibiotic resistance profile. 67 Rectal swabs from affected lambs in different regions of Mosul city were collected and cultured in MacConkey broth and incubated aerobically for 24 hours at 37ºC to cultivate and decrease the growth of undesired bacteria. Then, each sample was cultured on MacConkey, EMB, and HicromTM E. coli agar at the same incubation conditions for isolation of E. coli. Growing and purified isolates were identified based on Gram's staining reaction and biochemical IMViC tests (Indole, Methyl Red, Voges-Proskauer, and Citrate). E. coli was isolated from 65 samples (97.01%). Then, an antibiotic resistance profile was performed on all 65 isolates by using the disk diffusion method on 12 common antibiotics. The results of the research showed a high distribution of E. coli in affected cases, with varying resistance patterns, especially all isolates showed absolute resistance to spiramycin, lincomycin, and ceftriaxone. These findings point to the immediate requirement to reform antibiotic administration policies in veterinary medicine and ensure the importance of antibiotic resistance in E. coli isolates to find the most effective treatment strategies for colibacillosis in lambs. Future studies are needed to find alternative treatment options and the role of antibiotic management in veterinary lines.

ARTICLE INFO
Article history:
Received: 23 March 2025
Accepted: 19 June 2025
Published: 31 December 2025
DOI:
https://doi.org/10.36326/kjvs/2025/v16i219148

E-mail addresses:
khalid.23vmp27@student.uomosul.edu.iq.
ORCID:https://orcid.org/0009-0009-4016-8514

mahmah1073@uomosul.edu.iq.
ORCID: https://orcid.org/0000-0001-9278-5391

* Corresponding author

P- ISSN: 2077-9798
E- ISSN: 2959-8478

KEYWORD: *E. coli*, Colibacillosis, lambs, Hicrom^{TM} *E. coli* agar.

INTRODUCTION

Neonatal diarrhea is a significant challenge in the early stages of sheep rearing and the primary cause of mortality in ruminants, including lambs under one month old. It often causes serious problems, including a decrease in growth and high risks of mortality, which lead to high economic losses. Diarrhea in neonates may compromise the efficiency of feed and overall productivity of

the flock. Various viruses, bacteria, and protozoa are the causative agents of neonatal diarrhea [1].

Among all diseases and cases of diarrhea in lambs, Colibacillosis is the most common and important one. Colibacillary diarrhea is a bacterial disease caused by pathogenic strains of *E. coli* in small age ruminants, particularly within 1-3 days after birth. Diarrhea may also occur in older ages and around the time of weaning [2].

Colibacillary diarrhea occur in four distinct clinical form based on the causes and changes in physiological conditions which are hemorrhagic diarrhea also known as dysentery, this type of diarrhea characterized by the presence of blood or mucus in feces, watery diarrhea persist for a couple of hours or days, the risk associated with these cases is dehydration, Continuous diarrhea which persist for for 14 days or longer, Diarrhea related to severe malnutrition, danger associated with this condition include dehydration, vitamin and minerals deficiencies and heart failure [3].

The relevance of studying *colibacillosis* in lambs, especially small farm animals in general, is important because *colibacillosis* is considered a predominant challenge facing sheep breeding and a significant health threat [4].

Colibacillosis occurrence is facilitated by overcrowding, lack of skilled workforce, bad rearing systems, poor hygienic housing, improper passive immunity, and less colostrum uptake [5]. Although the economic importance of lambs, especially in regions such as Mosul city, is notable, little data presents a knowledge gap due to the rarely performed studies about Colibacillosis in Iraq in general and Mosul city in particular, especially considering the increasing mortality rates among neonatal lambs this study was done to investigate the prevalence and effects of the disease in lambs within Mosul city.

Colibacillosis in ruminants, including lambs, is of public health concern due to zoonotic and pathogenic *E. coli* strains, especially those strains that harbor Shiga toxin genes. Therefore, comprehensive and intensive molecular studies are required to develop prevention strategies and control different measures, which include perfect vaccination programs and controlling random antibiotic use. Zoonotic

pathotypes of *E. coli* related to colibacillosis in lambs have significant concerns in public health because they are a potential cause of many diseases in humans. Among these pathotypes, Shiga toxin-producing *E. coli* (STEC), especially the O157:H7 serotype, is the main cause of hemolytic uremic syndrome (HUS) and hemorrhagic colitis in humans. Goats, sheep, and some other ruminants are considered reservoirs for *E. coli* O157:H7, and they facilitate transmission of zoonotic disease by direct contact and/or ingestion of contaminated and uncooked products [6].

MATERIALS AND METHODS

ETHICAL APPROVAL

The research processes were done with the agreement of the Institutional Animal Care and Use Committee (UM.VET.2024.041), which licensed this study on 9/7/2024.

Study design:

The diagram below diagram highlight on the main methods used in the current study.

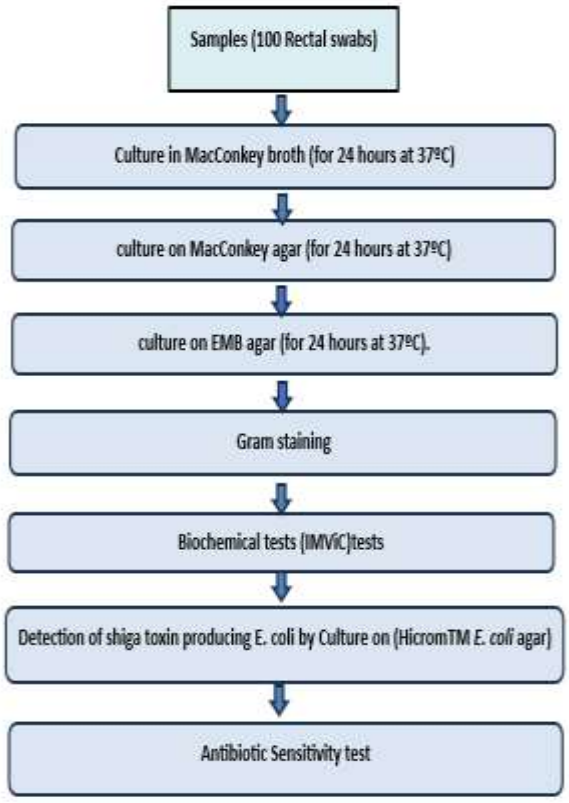

Fig. 1. The experimental design of the study

Specimens Collection

Sixty-seven rectal swabs were collected from lambs aged between one and four weeks of age suffering from dysentery or diarrhea. The samples were collected from different fields and veterinary clinics located in Mosul city during the period between October and December 2024, and the lambs

The samples were obtained by using a swab container containing 5 ml of MacConkey broth to ensure the bacteria reached alive to the lab alive and to support bacterial growth.

Laboratory Diagnosis (Isolation and identification of *E. coli*)

A loopful of the MacConkey broth was cultured onto MacConkey agar to distinguish between lactose and non-lactose-fermenting bacteria. A single pure pink colony was selected and subcultured onto EMB agar to observe the colonies with the metallic sheen phenomenon. Then subcultured on chromogenic agar (HicromTM *E. coli* agar-Himedia, India) to differentiate Shiga toxin-producing *E. coli* from non-producing colonies by making greenish-blue colonies (according to manufacturing instructions) [7]. All plates were incubated aerobically for 24 hours at 37ºC.

Identification by conventional methods

Pure colonies from EMB agar were selected and used for different conventional diagnoses, including the indole test, methyl red (MR), Voges-Proskauer (VP), and citrate test [8]. Gram staining for isolates was done between each culturing step on selective media (MA, EMB, and HicromTM *E. coli* agar).

Antimicrobial susceptibility test:

The antimicrobial susceptibility testing of *E. coli* was done using the disk diffusion method on Muller-Hinton agar. A total of twelve antibiotic disks were supplied by Bioanalyse Company (Turkey) (Table 1). Bacterial colonies were suspended in sterile normal saline, and the turbidity was adjusted to match the 0.5 McFarland standard. Then, the antimicrobial disks were placed on the agar by sterile forceps, and the Plates were incubated for 24 hours at 37 °C. After incubation, the diameter of inhibition zones was measured and classified as resistant (R), intermediate (I), or sensitive (S). The results were interpreted depending on the guidelines of the Clinical and Laboratory Standards Institute [9].

Table 1. Antimicrobial agents, abbreviations, and their concentrations

No.	Antibiotics	Abbreviations	Concentrations(µg)
1	Ciprofloxacin	CIP 10	10 µg
2	Spiramycin	SP 30	30 µg
3	Amoxicillin	AX 10	10 µg
4	Chloramphenicol	C 1O	10 µg
5	Lincomycin	L 10	10 µg
6	Oxytetracycline	T 30	30 µg
7	Metronidazole	MET 30	30 µg
8	Norfloxacin	NOR 30	30 µg
9	Ofloxacin	0FX 5	5 µg
10	Neomycin	N 10	10 µg
11	Ceftriaxone	CRO 10	10 µg
12	Trimethoprim and Sulphamethoxazole	SXT 25	1.25/23.5 µg

1. Abbreviations represent antimicrobial agents and their respective disk diffusion concentrations used for susceptibility testing, expressed in micrograms (µg).
2. Concentrations refer to the amount of antimicrobial agent per disk used in standard susceptibility testing protocols.

RESULTS

Isolation rates: The results revealed that almost all diarrheic cases (97.01%) were positive for isolation of pathogenic *E. coli*.

Culture characterization

1. Colonies on MacConkey agar

The colonies of isolated bacteria on MacConkey agar (MA) appeared smooth, shiny, with a moist texture and bright to thick pink color due to the ability of the isolates to ferment lactose, which results in pH reduction of the media. Out of Sixty-seven specimens, sixty-five (97.01%) showed pure, perfect pink color colonies on MacConkey agar (Fig. 2), indicating perfect isolation. while the two specimens (2.98%) appeared very contaminated and could not be purified.

Fig 2. Lactose-fermenting colonies of *E. coli* on MacConkey agar.

2. Colonies on EMB agar

All 65 cultured isolates on MA produced a metallic green sheen phenomenon on EMB (Figure 3), which is a perfect index for *E. coli* colonies.

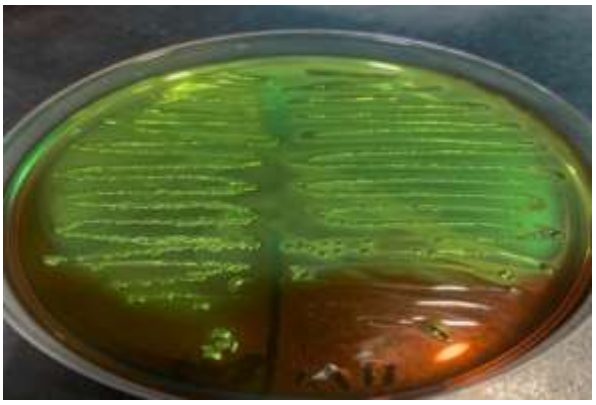

Fig. 3. Metallic sheen phenomenon of *E. coli* colonies on EMB agar.

Staining reaction:

All sixty-five isolates appeared as Gram-negative coccobacilli- short bacilli in all performed smears.

Biochemical tests

Single pure colonies that produced lactose fermentation on MacConkey agar and metallic sheen phenomenon on EMB agar (65 isolates) were positive for indole and methyl red tests, and negative Voges-Proskauer and Citrate tests (Fig. 4).

Fig. 4. (IMViC) tests

Left to Right (Indole +, M.R +, V.P: –, Citrate –)

Detection of Shiga toxin-producing *E. coli*

The purified colonies of isolated bacteria on EMB and MA that were cultured on HiCrome™ *E. coli* agar revealed that out of Sixty-five isolates, fifty-two (80%) isolates produced a greenish-blue color on Chromogenic agar, indicating Shiga toxin-producing *E. coli* (STEC). In comparison, thirteen isolates (20%) produce a white-colored pigmentation, indicating non-Shiga toxin-producing *E. coli* (Fig. 5).

Fig. 5. Greenish-blue and white Pigmentation of *E. coli* on Chromogenic agar

Greenish-blue color on Chromogenic agar, indicating Shiga toxin-producing *E. coli* (STEC). White-colored pigmentation, indicating non-Shiga toxin-producing *E. coli*.

Antimicrobial susceptibility test:

All *E. coli* isolates were faced against twelve antimicrobial agents, and the zone of inhibition were measured depending on guidelines of Clinical and Laboratory Standards Institute[9]., and the results revealed complete resistance of all isolates to four usually used antibiotics (spiramycin, lincomycin, metronidazole, and ceftriaxone), relatively high resistance of (83.1%) of isolates to antibiotics which *E. coli* isolates are commonly resistant to them (amoxicillin, oxytetracycline), 50.8% to neomycin, 32.3% to (Chloramphenicol and Trimethoprim and Sulphamethoxazole). In contrast, the majority of isolates (83.1%) were clearly sensitive to the fluoroquinolone antibiotic group (ofloxacin, norfloxacin, and ciprofloxacin) (Table 2, Fig. 6). In statistical analysis, antibiotic resistance patterns are analyzed using Fisher's Exact Test and the Chi-Square Test. The comparison between Spiramycin and Amoxicillin demonstrated extreme statistical significance ($p = 0.0001$), with Spiramycin having complete resistance in all 84 isolates compared to Amoxicillin's 70 resistant and 14 intermediate cases. Further analysis of Amoxicillin and Neomycin showed highly divergent resistance profiles ($\chi^2 = 19.7059$, $p < 0.00001$), while Neomycin versus Chloramphenicol also indicated significant differences in resistance patterns ($\chi^2 = 6.2694$, $p = 0.012$). Ofloxacin emerged as particularly effective, showing significantly greater sensitivity compared to Chloramphenicol ($\chi^2 = 5.4527$, $p = 0.019$), with 83.3% of isolates being non-resistant. According to international standards, the isolates were classified as multidrug resistant (MRD) because the isolates are resistant to many antibiotics; moreover, the isolates were not classified as extensively drug resistant or pan drug resistant because fluoroquinolones sensitivity were observed.

Fig. 6. Different inhibition zones depending on the isolate sensitivity.

Table 2. Susceptibility of the isolates to antimicrobial agents

Antibiotics	R	%	I	%	S	%
Spiramycin	65	100	0	0	0	0
Lincomycin	65	100	0	0	0	0
Metronidazole	65	100	0	0	0	0
Ceftriaxone	65	100	0	0	0	0
Amoxicillin	54	83.1	11	16.9	0	0
Oxytetracycline	54	83.1	11	16.9	0	0
Neomycin	33	50.8	32	49.2	0	0
Chloramphenicol	21	32.3	0	0	4	67.7
Trimethoprim and Sulphamethoxazole	21	32.3	11	16.9	33	50.8
Ofloxacin	11	16.9	0	0	54	83.1
Norfloxacin	11	16.9	0	0	54	83.1
Ciprofloxacin	11	16.9	0	0	54	83.1

Resistance to different antibiotics: 12 antimicrobial agents, all isolates demonstrated 100% resistance to Spiramycin, Lincomycin, Metronidazole, Ceftriaxone antibiotics, underscoring a serious multidrug resistance profile.

DISCUSSION

Colibacillosis in lambs is one of the most commonly reported diseases of lambs less than three months old because the sheep are the lifeline of the agro-economy in many different tropical regions, so characterization, identification, and treatment of the causative agents have significant economic importance. The results of this study showed high detection of *E. coli* in diarrheic lambs, ensuring the importance of these bacteria as a significant causative agent at the first stages of their life, which leads to great economic losses. The high isolation could be influenced by any factors, including age, geographical location, weak immunity, and stress, especially the cold weather, indicating a high rate of bacterial recovery under applied laboratory conditions and could be from mixed infections [10]. The existing study revealed that all isolated bacteria from dysentery or diarrheic cases were Gram-negative lactose-fermenting bacteria on MacConkey agar and metallic sheen producers on EMB. These phenomena are typical for *E. coli* isolates [11]. The results of this study were similar to the findings of several recent studies in the neighboring regions, including one conducted in Basrah governorate, Iraq [12], which showed that the majority of diarrhea cases were caused by *E. coli* pathogenic strains, and this is a close result achieved in the current study. According to this, *E. coli* is one of the most common causes of diarrhea and enteritis.

All *E. coli* isolates tested showed perfect reactions for the principal biochemical tests since they appeared positive for indole production and the methyl red test and negative for the Voges-Proskauer and citrate tests, and these results are identical for *E. coli* bacteria [12].

The isolation rate in the existing study was very extraordinary (97.01%) and much consider than the recorded observations in similar studies like the local study. such results suggest a high association of *E. coli* in diarrheic lambs; these high results raise the possibility of environmental contamination as

feed, water, and surfaces. It gives valuable insights to transmission pathways on sheep farms as mentioned by [13], which recorded an isolation rate of up to 47%, and even higher than the two Egyptian studies [14, 15], that revealed an isolation rate of *E. coli* up to 32.00% and 65%, respectively. Also, higher than the percentage recorded by an Iranian study [16]. (40.34%), and the same thing with the outcomes of past research in the Izmir governorate [17], which recorded a lower rate (36.4%) of enterovirulent *E. coli* strains, and most of their isolates were Shiga toxin-producing *E. coli*. Also, the current recording rate was higher than the Indian study, which registered 31.6% [18]. The differences in the isolation rates in all the mentioned studies and the existing study may be attributed to the specificity of the mode of collection of samples, since the specimens in the current study were collected from exactly diarrheic young lambs, and the culture techniques used were quality for E. coli isolation. In addition, the climate and environment may play a role in the isolation rate variety.

The majority of isolated bacteria (80%) produced a greenish-blue color on Chromogenic agar, indicating Shiga toxin-producing *E. coli* (STEC). In comparison, thirteen isolates (20%) produce a white-colored pigmentation, indicating non-Shiga toxin-producing *E. coli* [19].

The findings of the current study were near to the outcomes of several recent studies in nearby countries, including a study done by [15], in Egypt, where their results showed that most *E. coli* isolates indicated as pathogenic by pigment production (Bluish-green pigmented).

The isolated bacteria showed variety in their resistance or sensitivity toward many antibiotics that are used in the veterinary line for the treatment of bacterial diseases in general and diarrhea in particular, where the isolated *E. coli* revealed complete resistance to four main antibiotics (spiramycin, lincomycin, metronidazole, and ceftriaxone) and relatively high resistance (83.1-50%) to several antibiotics like amoxicillin, oxytetracycline, and neomycin. In contrast, the majority of isolates (83.1%) were evidently sensitive to the fluoroquinolones antibiotic group. This phenomenon of variety in sensitivity and resistance made these bacteria in terms of multi-drug resistance (MDR) and is well-known in all *E. coli* infections in sheep and other farmed animals like cattle, goats, poultry, and other animals, and the most common explanation for these varieties in resistance and susceptibility is related to owning the antibiotic resistance genes [15].

The existing results are similar to the findings of a recent study in Turkey performed by Kandir and Öztürk, who recorded absolute resistance (100%) to lincomycin in *E. coli* isolates from calves. Similar results to our study were performed by Haque *et al*., who found all *E. coli* isolates from calves suffering from diarrhea in India also showed complete resistance to tetracycline and amoxicillin compared to the 83.3% resistant isolates identified in our results [20]. Highly resistant *E. coli* isolates to these antibiotics in different studies and different countries reflect an international trend of resistance development to many antibiotics used in the veterinary line.

In another study performed by Asadi *et al*., in Tabriz City in Iran, they found that only 22.2% of *E. coli* isolates from calves suffering from diarrhea were resistant to ceftriaxone, while this study showed 100% of *E. coli* isolates were resistant to it. The highly resistant difference may be due to local variations in the usage of antibiotics, selection pressures, and/or sample sources [20].

The results of this study showed that the majority of *E. coli* isolates (83.3%) were evidently sensitive to the antibiotics belonging to the fluoroquinolones group, including

Ciprofloxacin, Ofloxacin, and Norfloxacin. But a study performed by Malekzadegan *et al*. from a tertiary care hospital in Iran showed relatively higher resistance of E. coli strains to Ciprofloxacin, Ofloxacin and Norfloxacin, where they showed relatively higher resistance of *E. coli* to these antibiotics than the results of our study as follows: norfloxacin, (44.6%), ofloxacin (46.3%) and ciprofloxacin (48.4%). The difference between the results of the two studies reflects the risk of antimicrobial resistance because *E. coli* can develop antimicrobial resistance genes in different environments and increase the risk of zoonotic diseases through the consumption of animal products. The difference between the results might be due to host variation and the degree of antibiotic exposure. These results highlight the need for regulation of antibiotic use to prevent cross-species transmission of resistance genes [21].

In the current study, Neomycin showed resistance in 51.2% of *E. coli* isolates, and 48.8% of them showed intermediate sensitivity. The results of another study were found by Jerzsele *et al*. in Hungary, where 74.3% of isolates were resistant to neomycin in turkeys. This resistance indicates that *E. coli* isolates to neomycin might no longer be effective in many cases, and it should be used [22].

The results of trimethoprim-sulphamethoxazole resistance in this study were 32.1%, which was higher compared to the results found by Morris *et al*, who showed that only 4.9% of the isolates were resistant to Trimethoprim-sulphamethoxazole in cow-calf. The relatively higher sensitivity of the isolates than other antibiotics in our study and the low resistance of the isolates in other studies reflect that it is a more effective antibiotic [23]. Also, moderate resistance to chloramphenicol was observed in this study, particularly in 32.1% of isolates. But in a study performed by Rasheed *et al*. reported absolute resistance to chloramphenicol from *E. coli* strains isolated from chronic respiratory disease cases in poultry. The differences in results between the two studies might be due to historical and excessive use of the antibiotic among host species. Also, it reflects the ability of *E. coli* isolates to develop resistance genes against these bacteria [24].

The results of the current study showed that the resistance to fluoroquinolone antibiotics is lower than other antibiotics in *E. coli* isolates, which causes colibacillosis, possibly because of their proper use till now in sheep farming and the difficulty in growing resistance against these antibiotics. But the usage of these antibiotics should be to prevent the distribution of resistance genes in both humans and animals.

The high antibiotic resistance rate observed in this study is similar to the results of a study performed by Subhi *et al*., where they linked the high resistance of *E. coli* isolates to the mobile genetic elements, including plasmids and integrons, which exist in the cytoplasm of the bacterial cell [25]. A study performed by Blanco Crivelli *et al.* about the relationship between the presence of virulence genes in *E. coli* isolates and antimicrobial resistance in lambs indicated that pathogenic strains of *E. coli* were more likely to carry resistance genes. These results showed the idea to integrate the phenotypic antimicrobial assay and genotypic detection of virulence genes [26].

Our results suggest that *E. coli* strains with diarrhea and enteritis have different virulence factors, which make them pathogenic. This emphasizes the requirement for targeted specific treatments and also different preventive measures to control *E. coli* infections in lambs, mainly on farms with frequently occurring diarrhea.

The current study is a step in serial studies that will confirm the isolates

molecularly and detect their main virulence genes (Shiga toxin 1 *'stx1'* gene, Shiga toxin 2 '*stx2*' gene, and Intimin gene '*eaeA*') that play the principal role in the pathogenicity of *E. coli* colibacillosis-causing strains.

CONCLOSION

In conclusion, colibacillosis is a principal disease and the main cause of death in young lambs, with a high prevalence observed in the studied population. The antibiotic resistance of these strains is highly prevalent in thriving lambs. This result highlights the need to find different antimicrobial strategies.

CONFLICTS OF INTEREST

Regarding the research data and instruments utilized in this work, the authors declare that there is no conflict of interest.

|REFERENCES

1. Grünberg W. Diarrhea in Neonatal Ruminants (Scours). Veterinary Content Pet Health Resource Quizzes, Faculty of Veterinary Medicine, Justus-Liebig-Universität Giessen, Giessen, Germany. 2021. https://www.msdvetmanual.com/digestive-system/intestinal-diseases-in-ruminants/diarrhea-in-neonatal-ruminants
2. Sun J, Chen W, Yuan Z. Characterization of intestinal microbiota in lambs with different susceptibility to *Escherichia coli* F17. Vet Sci 2022;9(12):670. https://doi.org/10.3390/vetsci9120670
3. Arasaradnam RP, Brown S, Forbes A, Fox M R, Hungin P, Kelman L, Thomas P. Guidelines for the investigation of chronic diarrhea in adults: British Society of Gastroenterology, 3rd edition. 2018;67(8):1380-1399. https://doi.org/10.1136/gutjnl-2017-315909
4. Muktar Y, Mamo G, Tesfaye B, Belina D. A review on major bacterial causes of calf diarrhea and its diagnostic method. J Vet Med Anim Health [Internet]. 2015;7(5):173–85. https://doi.org/doi:10.5897/JVMAH2014.0351
5. Robi DT, Mossie T, Temteme S. A comprehensive review of the common bacterial infections in dairy calvesand advanced strategies for health management. Vet Med Res Rep 2024;31:1-4. https://doi.org/10.2147/VMRR.S452925
6. Ramatla T, Tutubala M, Motlhaping T, de Wet L, Mokgokong P, Thekisoe O, Lekota K. Molecular detection of Shiga toxin and extended-spectrum beta-lactamase (ESBL)-producing *Escherichia coli* isolates from sheep and goats. Mol Biol Rep 2024;51(1):57. https://doi.org/10.1007/s11033-023-08987-0
7. Hi-Media. Hi-Crome™ *E. coli* Agar. Available from: https://www.himedialabs.com.
8. Aryal S. Biochemical test of *Escherichia coli* (*E. coli*). Microbe Notes [Internet]. 2022. https://microbenotes.com/biochemical-test-of-escherichia-coli-e-coli/
9. Wayne PA. Clinical and Laboratory Standards Institute. Performance standards for antimicrobial susceptibility testing. 31st ed. 2020.https://www.nih.org.pk/wp-content/uploads/2021/02/CLSI-2020.pdf
10. Talah RS, Jafar NA. Animal as potential reservoir of diarrheagenic *Escherichia coli*. Egypt. J. Vet. Sci. 2024; 10:1-8. DOI: 10.21608/EJVS.2024.285118.2037.
11. Mueller M, Tainter CR. *Escherichia coli* infection. StatPearls. National Library of

Medicine; 2023. https://www.ncbi.nlm.nih.gov/sites/books/NBK564298

12. Farhan ZA, Al-Iedani AA. Molecular detection of shiga toxin (stx1 and stx2) and intimin (eae A) genes in *Escherichia coli* isolated from fecal samples of cattle, sheep, and human in Basrah governorate. Basrah J Vet Res. 2019;18(2):288-305. https://iasj.rdd.edu.iq/journals/uploads/2024/12/29/9ffb9eef6689ed94bc1a94ab4a4c0a38.pdf

13. Al-Sabawi AH, Jwher DM. A comparative study of *Escherichia coli* isolates from open and closed sheep breeding systems in Nineveh, Iraq. Basrah J Vet Res. 2022; 21(3):12-22. https://www.researchgate.net/publication/365565237_A_comparative_study_of_Escherichia_coli_isolates_from_open_and_closed_sheep_breeding_systems_in_Nineveh_Iraq

14. Mokhbatly AA, Elsheikh N, Ghazy EW, Elgamal AM, Hegazy YM, Assar DH. Prevalence of Shiga toxin-producing *Escherichia coli* and Salmonellae and some associated hematologic and biochemical profile alterations in lambs. Vet. Res. Forum. 2022;13(2):155. https://doi.org/10.30466/vrf.2020.124018.2907

15. El-Nady HH, Eissa MI, Abou-Zeid NZ, Abd-Elfatah EB, Shehata AA, Fawzi EM. Colibacillosis in lambs and kids in Egypt: Prevalence, serogroups, antibiogram profile, virulence genes distribution and antimicrobial resistance genes. Open Vet J 2023;16;13(9):1106-15. https://www.ajol.info/index.php/ovj/article/view/259336

16. Ghanbarpour R, Askari N, Ghorbanpour M, Tahamtan Y, Mashayekhi K, Afsharipour N, Darijani N. Genotypic analysis of virulence genes and antimicrobial profile of diarrheagenic *Escherichia coli* isolated from diseased lambs in Iran. Trop Anim Health Prod. 2017;49:591-7. https://link.springer.com/article/10.1007/s11250-017-1234-7

17. Türkyılmaz S, Eskiizmirliler S, Tunaligil S, Bozdogan B. Identification, characterization and molecular epidemiology of *Escherichia coli* isolated from lamb and goat kids with diarrhoea. Acta Vet Brno. 2014;13;82(4):357-62. https://doi.org/10.2754/avb201382040357

18. Bandyopadhyay S, Mahanti A, Samanta I, Dutta TK, Ghosh MK, Bera AK, Bandyopadhyay S, Bhattacharya D. Virulence repertoire of Shiga toxin-producing *Escherichia coli* (STEC) and enterotoxigenic *Escherichia coli* (ETEC) from diarrhoeic lambs of Arunachal Pradesh, India. Trop Anim Health Prod. 2011;43:705-10.1 https://link.springer.com/article/10.1007/s11250-010-9757-1

19. Kandir H, Öztürk D. Antimicrobial resistance of *E. coli* and Salmonella isolated from wild birds in a Rehabilitation Center in Turkey. Arch. Razi Inst. 2022;77(1):257. https://doi.org/10.22092/ARI.2021.356322.1823.

20. Asadi H, Nadari M, Onagh SA, Abdolghaffar N. Study on antibiotic resistance pattern of *E. coli* in diarrheic calves of Tabriz dairy farms. Arch. Razi Inst.2024; 22;34(2):88–99. https://doi.org/10.22034/as.2023.56083.1703

21. Malekzadegan Y, Rastegar E, Moradi M, Heidari H, Sedigh Ebrahim-Saraie H. Prevalence of quinolone-resistant uropathogenic Escherichia coli in a tertiary

care hospital in south Iran. Infect. Drug Resist. 2019;19:1683. https://www.tandfonline.com/doi/full/10.2147/IDR.S206966

22. Jerzsele Á, Szabó Á, Barnácz F, Csirmaz B, Kovács L, Kerek Á. Antimicrobial Susceptibility Profiles of *Escherichia coli* Isolates from Clinical Cases of Turkeys in Hungary (2022–2023). Antibiotics. 2025; 14(4): 338. https://doi.org/10.3390/antibiotics14040338

23. Morris C, Wickramasingha D, Abdelfattah EM, Pereira RV, Okello E, Maier G. Prevalence of antimicrobial resistance in fecal *Escherichia coli* and Enterococcus spp. isolates from beef cow-calf operations in northern California and associations with farm practices. Front. Microbiol. 2023; 14:1086203. https://doi.org/10.3389/fmicb.2023.1086203

24. Rasheed BY, Hamad MA, Isihak FA. Molecular study of resistance genes in *Escherichia coli* isolated from chronic respiratory disease cases in broilers. Iraq J Vet Sci. 2024;38(1):119-24. https://doi.org/10.33899/ijvs.2023.139069.2873

25. Subhi A, Saad AS, Osman K, Hashad ME, Deif HN. Prevalence and Antibiogram of *Escherichia coli* isolates recovered from bovine milk. J Appl Vet Sci. 2023;8(3):82-90. https://doi.org/10.21608/javs.2023.215720.1238

26. Blanco Crivelli X, Bonino MP, Sanin MS, Petrina JF, Disalvo VN, Massa R, Miliwebsky E, Navarro A, Chinen I, Bentancor A. Potential zoonotic pathovars of diarrheagenic Escherichia coli detected in lambs for human consumption from Tierra del Fuego, Argentina Microorganisms. 2021;9(8):1710. https://doi.org/10.3390/microorganisms9081710.