

## Occurrence of *Escherichia coli* O157: H7 from Sheep and Goats Slaughtered at Elfora Abattoir by Using Latex Agglutination and Biochemical Tests

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**Abstract:** *Background* :*Escherichia coli* (*E. coli*) is an important member of the normal intestinal micro flora of humans and other mammals it has also been widely exploited as a cloning host in recombinant DNA technology. A cross sectional study was conducted from, November, 2018 to April, 2019 at Bishoftu Elfora export Abattoir. For the purpose of study a total of 384 meat swab samples (192 samples from each species) were collected from different body parts were cultured for detection of *E. coli* which was then tested for antimicrobial susceptibility. Results: The overall prevalence of *E. coli* was 10.4% (40/384), Out of which 5.7% (22/192) were from sheep meat and 4.7% (18/192) from goat meat. All of 40 positive samples subjected for sorbitol MacConky agar test and finally for confirmation by agglutination antibody of latex kit formed for O157: H7. From total positive result only 14 (3.67%) of samples shows positive reaction to the test. Accordingly the prevalence of *E. coli* O157: H7 for sheep and goat was 4.6% (9/192) and 2.6% (5/192) respectively, with no statistical significance variation ( $P > 0.05$ ). The distribution of *E. coli* O157: H7 on brisket, flank and thorax among 384 sampled animals (sheep and goat) were 3.3% (3/120), 4.9% (6/122) and 2.8% (4/142) respectively. The prevalence of *E. coli* O157:H7 in young and adult age group was found to be similar (3.8%). Out of the 14 *E. coli* O157:H7 5, 6 and 3 of them identified from animals originated from Borena, Harar and Jinka respectively. The result of antimicrobial resistance tested on *E. coli* isolates showed intermediate susceptibility to two antimicrobials (kanamycin and sulphonamides) which varied from 40% to 60% and 75%- 100% susceptibility was exhibited to oxytetracyclin, nalidixic acid, streptomycin and ceftiraxone. All the isolates showed 100% susceptibility to, ceftriaxone and nalidixic acid. However, *E. coli* isolate exhibited a 100% resistance for amoxicillin and trimethoprim. Conclusion: The detection of *E. coli* O157:H7 in raw meat from apparently healthy sheep and goat shows an important public health concern. The existence to amoxicillin and trimethoprim highlight the potential threat to public health. Hence implementation of *E. coli* O157:H7 prevention and control strategies from farm production to consumption of meat and meat products are crucial.

**Key words:** *E. coli* O157: H7 • Antibiotic Resistance • Small Ruminant • Meat • Prevalence

### INTRODUCTION

*Escherichia coli* (*E. coli*) is an important member of the normal intestinal micro flora of humans and other mammals it has also been widely exploited as a cloning host in recombinant DNA technology. However, *E. coli* is not a laboratory workhorse or harmless intestinal inhabitants it can also be a highly versatile and frequently deadly pathogen [1]. The recognition of

Enterohemorrhagic *Escherichia coli* (EHEC) as an aetiological agent of diarrhoea with life-threatening complications has made this kind of infection a public health problem of serious concern. It has been demonstrated that certain *E. coli* isolates produce a toxin, which was initially called verotoxin because of its distinct effect on vero cells. This family of toxins was subsequently also called Shiga-like toxins (SLT) and more recently Shiga toxins (*Stx*), because of the close relation

to the Stx of *Shigella dysenteriae* type 1. Shiga toxins are categorized into two main groups, *stxI* and *stxII*. The majority of *Stx* genes are bacteriophage-borne, which may be important for the spread of shiga toxin-producing *E. coli* (STEC) [2]. Verocytotoxin-producing *E. coli* (VTEC), also referred to as Shiga toxin-producing *E. coli*, including serotype O157:H7, are one such group, causing severe, chronic and potentially fatal illness such as hemorrhagic colitis, hemolytic uremic syndrome, thrombotic thrombocytopenic purpura and, in severe cases, death, related to their ability to produce one or more toxins known as verotoxin or Shiga-like toxin [3]. EHEC strains are characterized by the ability to form attaching and effacing (A/E) lesions on the surface of epithelial cells in the gastrointestinal tract and the production of shiga toxins. The first gene to be associated with A/E activity was the intimin gene, *eae* and its presence is often used as a marker for the infections caused by EHEC [4]. *E. coli* O157:H7 was the first serotype associated with hemorrhagic colitis, although more than 100 STEC serotypes have been isolated from different sources, such as food and recreational and drinking water. However, not all pathogenic STEC isolates have been shown to produce intimin [5]. EHEC appears to be transmitted primarily through the ingestion of faecal contaminated foods, particularly undercooked beef meat. The most frequent mode of transmission for *E. coli* O157:H7 infection to human is through consumption of contaminated food and water. However, it may also spread directly from person to person and occasionally through occupational exposure [6].

Ruminants, particularly cattle and sheep, are the most important reservoir hosts for EHEC O157:H7 [7]. Cattle have been identified as a major reservoir of *E. coli* O157 [8] and consumption of foods of bovine origin have been associated with some of the largest food poisoning outbreaks in which this organism was identified as the etiologic agent [9]. The microbiological contamination of carcasses can occur during processing and manipulation, such as skinning, evisceration, storage and distribution at slaughter houses. Fecal matter is a major source of contamination and could reach carcasses through direct deposition, as well as by indirect contact through contaminated equipment, workers, installations and air [10]. Animal slaughtering operations, such as bleeding, dressing and evisceration expose sterile muscle to microbiological contaminants that were present on the skin, the digestive tract and in the environment [6, 11]. *E. coli* O157:H7 infections as a cause of disease have shown a marked increase in many countries. Many serotypes other than O157:H7 are capable of producing

Shiga toxins and similar clinical manifestations, but this serotype was the one most commonly isolated in North America and Europe as a cause of illness [12]. Biochemical identification and serotyping are slow and inefficient, especially in outbreak situations and large-scale screening of clinical and food samples. Recently, latex agglutination tests (*Escherichia coli* O157 latex test, Oxoid Ltd., Hampshire, England; SeroBact *E. coli* O157, D.P. Diagnostics, Adelaide, Australia) have become commercially available for rapid presumptive detection of *E. coli* belonging to the serogroup O157 [13]. Antimicrobial resistance is a serious public health problem limiting the therapeutic options available to clinicians treating complicated *E. coli* infections. The emergence of antimicrobial resistance in *E. coli* O157:H7 is complicated because the use of antibiotics for therapeutic purposes in veterinary medicine and as growth promoters in animal feed may promote the emergence of resistance, thus presenting a potential risk to public health from zoonotic infections [14]. Consequently, antibiotic resistance presents a great challenge to the treatment of infections [15]. In case of Ethiopia the majority of the studies on *E. coli* O157:H7 were done on cattle [15, 16].

Therefore the objectives of this research were:

- To detect and determine occurrence of *Escherichia coli* O157: H7 among sheep and goats slaughtered at Elfora abattoir in Bishoftu.
- Assess the antimicrobial resistance profile of *E. coli* isolates.

## MATERIALS AND METHODS

**Study Area:** The study was conducted at Elfora abattoir, Over a period of 5 months (November 2018 to April 2019), in Bishoftu town which is located at 9°N and 4° E with an altitude of 1880 m.a.s.l in the central highlands of Ethiopia, lying 47 km south east of Addis Ababa, the capital city. It has annual rain fall of 1151.6 mm of which 84% falls during the long rainy season that extends from June to September and the remaining during the short rainy season that extends from March to May. The mean annual minimum and maximum temperatures are 12.3°C and 27.7°C, respectively and the mean relative humidity is 61.3%.

**Study Population:** The study population constituted of local breeds of ovine and caprine coming from lowland and midland areas of the country and slaughtered at Elfora abattoir, found in Bishoftu town. Majority of the

slaughter animals come from places such as: Harar, Borana and Jenka areas which represent the lowland and midland regions of the country. The animals slaughtered in Elfora abattoir are mostly male and adult.

**Study Design:** A cross sectional study was conducted from, November, 2018 to April, 2019 by random sample collection on events associated with prevalence and antimicrobial resistance of *E. coli* O157:H7 of ovine and caprine slaughtered in Elfora abattoir.

**Sample Size Determination and Sample Collection:** The sample size was determined based on the formula recommended by Thrustfield [17].

$$n = \frac{1.96^2 \times P_{\text{exp}} (1 - P_{\text{exp}})}{d^2}$$

where,

n = sample size required

P<sub>exp</sub> = expected prevalence,

d = desired absolute precision

Since the prevalence of *E. coli* of caprine and ovine slaughtered in Efora abattoir has not been reported, 50% expected prevalence rate was used. In addition, 95% confidence interval and 5% desired absolute precision was also used. Using desired 95% confidence interval, 5% precision and 50% expected prevalence the number of slaughtered caprine and ovine needed to investigate drug resistance profiles of *E. coli* were 384.

**Sample Collection, Handling and Transport:** A total of 384 swab samples were collected from Elfora export abattoir. The swab samples were from caprine (n=192) and ovine (= 192). All swab samples were taken from carcass ready-to-eat fresh or cooked and taken by random sampling technique from the carcasses of dailyslaughtered animals at the abattoir. Selected carcasses were swabbed using the method described in ISO17604 [18] by placing sterile template (10 x 10 cm) on specific sites of a carcass. A sterile cotton tipped swab, (2X3 cm) fitted with shaft, was first soaked in an approximately 10 ml of buffered peptone water (Oxoid Ltd., Hampshire, England) rubbed first horizontally and then vertically several times on the carcasses. The abdomen (flank), thorax (lateral) and breast (lateral); which are sites with the highest rate of contamination [18]; were chosen for sampling using the same swab over all the sites. On completion of the rubbing process, the shaft was broken by pressing it against the inner wall of the test tube and disposed leaving the cotton swab in the test

tube. All sampling process was done aseptically and sample were placed into sterile containers and transported to the Microbiology Laboratory. Up on arrival, the samples were stored in refrigerator at 4°C until being processed for isolation. Samples were transported in ice boxes and analyzed within 6 to 12 hours.

**Culture and isolation of *Escherichia coli*:** All enriched swab samples were subsequently cultured on to eosin methylene blue (EMB) agar (Difco Laboratories, USA) for primary screening of *E. coli* and incubated aerobically at 37°C for 24 hours [19].

**Morphological Characterization by Gram Staining**

**Method:** Isolated pure colonies from the bacteria inoculated at 37°C for 24 hrs culture media was peaked using sterile wire loop and thin film was made on the clear slide having adrop of distilled water on the center and air dried. After it has been fixed by passing 3 time on Bunsen burner flame, it the subjected to gram stain by flooding with chemical; crstalviolat for 30-60 sec, iodine for 1min acetone for 3-5 sec and safranin for 30-60 sec then air dried and observed under oil immersion microscope and recorded according to the gram section, gram negative and positive. Bacteria having pink color and rod shape under the microscope were gram negative [20].

**Identification of Isolated *Escherichia coli* by Using**

**Specific Biochemical Tests:** For this study, catalase test, MR-VP reaction, indole reaction and sheep blood hemolysis test were selected for identification of *E. coli*. Allsuspected colonies were tested according to the methods given in the Qunin *et al.* [21].

**Methyl Red Test:** The test was performed by inoculating a colony of the test organism in 0.5ml sterile glucose phosphate broth (as used in the V-P test). After overnight incubation at 37°C, a drop of methyl red solution will be added in to test tube and observed for any color change and the resultwas recorded and compared according to the standard recommended. A positive methyl red test was shown by the appearance of a bright red color, indication of acidity. Colony of *E. coli* show bright red, yellow or orange color is treated as negative [21].

**Voges-Proskauer (V-P) Test:** Two ml of sterile glucose phosphate peptone water were inoculated with the test organisms. It is incubated at 37°C for 48 hours. A very small amount Alpha naftol and three (3) ml of sodium hydroxide reagent was added and mixed by shaking and observed closely for the slow development of a pink-red

color for positive reactions after 20-30 min. The result was recorded and compared according to the standard recommended. *E. coli* was negative and there is no development of pink-red color [21].

**Indole Test:** The test organism was inoculated in to the test tube containing tryptone broth prepared using the standard which is a rich source of amino acid tryptophan and incubated at 37°C for 24-28 hours. The indole test was carried out after 24hr of incubation by the addition of half (0.5) ml Kovac’s reagent. Indole positive bacteria produce tryptophanase enzyme, an enzyme that converts tryptophan to indole. The result was recorded and compared according to the standard recommended. A red color layer will indicate indole positive. No red color development indicate negative [21].

**Hemolysis on Sheep Blood:** Hemolysis test of isolated bacteria was performed by inoculating them into blood agar plates (Nutrient agar 2.4 gm% and 7 % Sheep blood). Plates were incubated at 37°C for 24 hr. A clearing zone surround the bacterial colony was observed and recorded. *E. coli* ATCC 23509 and *E. coli* ATCC 25922 were taken as negative and positive control, respectively [22].

**Serological Identification:** After brief, 2-3 biochemically confirmed. *Escherichia coli* were sub cultured on Sorbitol MacConkey Agar (Oxoid), supplemented with 0.05 mg/l cefixime and 2.5 mg/l tellurite (Oxoid) (SMAC-CT) (used to selectively differentiate the non-sorbitol-fermenting *E. coli* O157:H7 strains from other *E. coli* strains) and incubated at 37°C for 18 to 24 hours. Following the end of the incubation period, the SMAC-CT agar plates were examined for the presence of non-sorbitol fermenter colonies and subsequently they were further processed for serological identification. Sorbitol fermenter colonies appears pink while non fermenter (O157:H7)

looks like pale and Blood agar (Oxoid CM55) was used to differentiate hemolytic organisms from non-hemolytic ones [23].

All non-sorbitol fermenting colonies from the SMAC-CT agar were subjected to slide agglutination with the *E. coli* O157 Latex kit test (Oxoid). The latex beads were coated with antibodies which bind to any O157 or H7 antigens on the test organisms, forming a visible antigen antibody precipitate. Colonies giving a precipitation reaction were confirmed as *E. coli* O157:H7 positive [24].

**Antimicrobial Susceptibility Pattern:** The antimicrobial susceptibility test was performed following the standard agar disk diffusion method according to CLSI [13] using commercially available antimicrobial disks Table 1. Indicate antibiotic disks used to test *Escherichia coli* and their respective concentrations. Each isolated bacterial colony from pure fresh culture was transferred into a test tube of 5 ml Tryptone Soya Broth (TSB) (Oxoid, England) and incubated at 37°C for 6 hrs. The turbidity of the culture broth was adjusted using sterile saline solution or added more isolated colonies to obtain turbidity usually comparable with that of 0.5 McFarland standards (approximately  $3 \times 10^8$  CFU per ml).

Mueller-Hinton agar (Bacton Dickinson and Company, Cockeysville USA) plates were prepared according to the manufacturer guidelines. A sterile cotton swab was immersed into the suspension and rotated against the side of the tube to remove the excess fluid and then swabbed in three directions uniformly on the surface of Mueller-Hinton agar plates. After the plates dried, antibiotic disks were placed on the inoculated plates using sterile forceps. The antibiotic disks were gently pressed onto the agar to ensure firm contact with the agar surface and incubated at 37°C for 24 hrs. Following this the diameter of inhibition zone formed around each disk was measured using a black surface, reflected light and

Table 1: Antibiotic disks used to test *E. coli* and their respective concentrations

No Antibiotic disks	Diameter of zone of inhibition in millimeter (mm)				
	Disc code	Concentration	Resistant	Intermediate	Susceptible
Triptoprime	W5	30µg	10	11-15	16
Ceftiaxone	Cro	30	13	14-20	21
Streptomycine	S	25	11	12-14	15
Kanamycin	K	30	13	14-18	19
Amoxycilin	Amc	30	14	15-16	17
Sulphonamides	S3	300	12	13-16	17
Nalidixicacid	Na	30	13	14-18	19
Oxytetracyclin	OT	30	14	15-16	19

transparent ruler by lying it over the plates. The results were classified as sensitive, intermediate and resistant according to the standardized table supplied by the manufacturer [13].

**Data Management and Analysis:** The coded data was entered in MS Excel and then analysed using SPSS version 20 [25]. The overall prevalence of *E. coli* was determined by taking the positive samples and dividing it by the total number of samples examined multiplied by 100. In addition to these, the prevalence in each meat type of the abattoir was determined in the same way by dividing positive value with corresponding total examined samples. Difference among and between proportions of the groups with certain determinant factor was determined by chi-square ( $\chi^2$ ) test. A p-value <0.05 was considered indicative of a statistical significant difference.

## RESULTS

**Prevalence of *Escherichia coli* Based on Emb Test in Sheep and Goat in Elfora Abattoir:** The overall prevalence of *E. coli* in two types of carcass swab samples (sheep and goat's meat) was 10.4% (40/384). Out of which 5.7% (22/192) were from sheep meat and 4.7% (18/192) from goat meat. The test statistics among two types of raw meat samples indicated that there was no statistical significance difference in prevalence rate ( $p > 0.05$ ) Table 2.

**Prevalence of *Escherichia coli* in Elfora Abattoir in Different Age Groups of Sheep and Goat:** From 40 (10.4%) positive result 22 (11.9%) are young sheep and goat while the rest 18 (9%) are adult. This indicates that the prevalence of *E. coli* was similar in the young age group and adult. The test statistics among two age group samples indicated that there was no statistical significance difference in prevalence rate ( $p > 0.05$ ).

**Prevalence of *Escherichia coli* in Sheep and Goat Coming from Different Origin:** From overall positive result, 10 (7.8%) are sheep and goat come from Borena, 13 (11.02%) were from Jinka and the rest 17 (12.31%) positive result were from Harar. However there was no statistically significant relationship difference in prevalence of *E. coli* isolated from the three different origins.

**Prevalence of *Escherichia coli* in Different Site of Sample Collection:** Out of total positive results (40), 10 (8.2%) were sample taken from flank, 12 (10%) were

sample taken from Brisket while the rest positive result 17 (12.7%) taken from thorax. There was no statistically significant relationship difference in prevalence of *E. coli* isolated from the three different site of sample collection.

**Serology Result:** Out of 40 positive result only 14 (3.67%) are make agglutination of antibody of latex kit formed for O157; H7 while the other 26 (6.77%) were negative and not form gel (agglutination).

**Prevalence of *E. coli* O157:H7 Based on Different Specie:** From total serological positive *E. coli* (O157:H7), 9 (4.6%) was sample of sheep while the rest, 5 (2.6%) were from goat. The prevalence of *E. coli* O157:H7 among two different species are similar. The test statistics among two species indicated that there was no statistical significance difference in prevalence rate ( $p > 0.05$ ).

**Prevalence of *E. coli* O157:H7 Based on Different Age Group:** The prevalence of *E. coli* O157; H7 are similar in both young and adult age group and the test statistics among two age group indicated that there was no statistical significance difference in prevalence rate ( $p > 0.05$ ). From total 14 positive result 9 of them are young and the rest 5 are adult.

**Prevalence of *Escherichia coli* O157:H7 among the Origin:** From all serological positive *Escherichia coli* O157:H7 6 (1.6%) was from the sheep and goat originated from Borena, 5 (1.3%) were from Harar while the rest result 3 (0.8%) from Jinka. The test result indicates that there is no statistically significant relationship difference in prevalence of *E. coli* O157:H7 isolated from the three different origins.

**Prevalence of *Escherichia coli* O157:H7 According to Site of Sample Collection:** The distribution of *E. coli* O157; H7 on different body part of sample taken; brisket, flank and thorax among 384 sampled animals (sheep and goat) was as followed: 3.3% (3/120), 4.9% (6/122) and 2.8% (4/142) respectively.

**Antimicrobial Susceptibility Pattern:** The result of antimicrobial susceptibility test of 40 positive *Escherichia coli* isolated from raw meat samples with 8 selected antimicrobial agents is shown in Table 10. The antimicrobial sensitivity test of *E. coli* isolated from different raw meat types revealed a varying degree of susceptibility to antimicrobial agents tested (Table 10). Isolates recovered from sheep meat were found to be 40% to 60% susceptible to three antimicrobial agents tested

Table 2: Prevalence of *Escherichia coli* based on EMB test in sheep and goat in Elfora abattoir

Species	Examined	Positive (%)	95% CI	$\chi^2$	(P-value)
Sheep	192	22(5.7)	7.84-16.74	0.44	(0.504)
Goa	192	18(4.7)	6.01-14.33		
Over all	384	40(10.4)	7.74-13.87		

Table 3: Prevalence of *Escherichia coli* in Elfora abattoir in different age groups of sheep and goat

Age	Examined	Positive (%)	95% CI	$\chi^2$	(P-value)
Adult	200	18(9)	5.77-13.78	0.89	0.343
Young	184	22(11.9)	8.03-17.44		
Over all	384	40(10.4)	7.74-13.87		

Table 4: Prevalence of *Escherichia coli* in sheep and goats coming from different origin

Origin	Examined	Positive (%)	95% CI	$\chi^2$	(P-value)
Borena	128	10(7.8)	4.3-13.78	1.51	(0.47)
Harar	138	17 (12.31)	7.8-18.87		
Jinka	118	13 (11.02)	6.55-17.84		
Overall	384	40(10.42)	7.74-13.87		

Table 5: The prevalence of *E. coli* in different body parts of sampled animals

Site of sample Collected	Examined	Positive (%)	95% CI	$\chi^2$	(P-value)
Brisket	120	12( 10)	5.8-16.67	1.44	(0.48)
Flank	122	10(8.2)	4.51-14.43		
Thoracic	142	18(12.7)	8.17-19.15		

Table 6: Prevalence of *Escherichia coli*O157:H7 based on different specie

Species	Examined	Positive (%)	95% CI	$\chi^2$	(P-value)
Sheep	192	9( 4.6)	2.49-8.67	1.18	(0.276)
Goat	192	5(2.6 )	1.12-5.95		
Over all	384	14 (3.6)	2.18-6.03		

Table 7. Prevalence of *Escherichia coli* O157: H7 in Elfora abattoir in different age groups of Sheep and goat

Age	Examined	Positive (%)	95% CI	$\chi^2$	(P-value)
Adult	200	7 (3.8)	1.71-7.5	0.25	(0.87)
Young	184	7 (3.8)	1.85-7.64		

Table 8: Prevalence of *E.coli* O157:H7 among origin

Origin	Examined	Positive (%)	95% CI	$\chi^2$	(P-value)
Borena	128	6(1.6)	4.3-13.78	1.51	( 0.47)
Harar	138	5(1.3)	7.8-18.87		
Jinka	118	3 (0.8)	6.55-17.84		
Overall	384	14(3.6)	7.74-13.87		

Table 9: Prevalence of *Escherichia coli* O157:H7 according to site of sample collection

Site of sample Collected	Examined	Positive (%)	95% CI	$\chi^2$	(P-value)
Brisket	120	3( 3.3)	5.8-16.67		
Flank	122	6 (4.9)	4.51-14.43	1.44	(0.48)
Thoracic	142	4 (2.8)	8.17-19.15		

Table 10: Antimicrobial susceptibility pattern of *E. coli* isolates (n = 40)

Type of raw meat sample sheep goat	Total									
	R No (%)	I No (%)	S No (%)	R No (%)	I No (%)	S No (%)	R No (%)	I No (%)	S No (%)	
Antibiotic										
Triptoprime	19(4.9)	3(0.8)	(0%)	13(3.2)	5(1.3)	(0%)	32(7)	8(2.1)	0(0)	
Ceftiraxone	0%	4(1)	18(4.7)	0%	2(0.5)	16(4.2)	0%	6(1.5)	34(8.9)	
Amoxycilin	14(3.6)	1(0.3)	7(1.7)	12(3.1)	0%	6(1.6)	26(6.8)	1(0.3)	34(8.9)	
sulphonamides	7(1.8)	10(2.6)	5(1.3)	3(0.8)	8(2.1)	7(1.8)	10(2.6)	18(4.7)	12(3.1)	
Streptomycin	0%	14(3.6)	8(2.1)	0%	8(2.1)	10(2.6)	0%	22(5.7)	18(4.7)	
Nalixicacid	0%	9(2.3)	13(3.4)	0%	4(1)	14(3.6)	0%	13(3.4)	27(7)	
oxytetracyclin	0%	11(2.9)	11(2.9)	0%	7(1.8)	11(2.9)	0%	18(4.7)	22(5.7)	
Kanamycin	3(0.8)	10(2.6)	9(2.3)	1(0.3)	11(2.9)	6(1.6)	4(1)	21(5.5)	22(5.7)	

except ceftriaxone, streptomycin and nalidixic acid antimicrobial agents, which showed 100% susceptibility. Similarly, *E. coli* isolates from goat meat showed 100% susceptibility to, ceftriaxone, streptomycin and tetracycline. However, for the remaining drugs (amoxicillin and trimethoprim) show 100% resistance.

## DISCUSSION

Raw meat and its products are commonly consumed in traditional Ethiopian diets, but *E. coli* is rarely studied in Ethiopia compared to other countries. In the present study, *E. coli* was isolated from sheep meat and goat meat slaughtered in abattoir. The overall prevalence of *E. coli* in the present study (10.4%) was relatively similar with prevalence (15.7%) reported by Ousman Mohammed *et al.* [26] from Jijiga and higher than the prevalence (4.2%) reported in Modjo and Addis Ababa [27] in Ethiopia. On the other hand, prevalence of present study was lower than the 22.2% prevalence report of *E. coli* isolated from meat samples collected from Mekelle Municipality abattoir in northern Ethiopia [16]. In this study, the prevalence of *E. coli* in sheep meat (5.7%) and goat meat (4.7%) were higher than the previous study done by Hiko *et al.* [27] in Ethiopia who report a prevalence of 2.5% in sheep meat and 2% in goat meat and lower than prevalence reported by Bekele, Tizeta *et al.* [15] who report prevalence of sheep meat (9.4%) and goat meat (7.8%) in Addis Ababa. The prevalence of *E. coli* was similar at the abattoir in sheep meat and goat meat. There is no significant variation ( $p > 0.05$ ) among the two types of raw meat samples. The prevalence of *Escherichia coli* are also similar in the age group where there are prevalence of young age group (11.9%) was recorded and in adult prevalence (9%), this prevalence raised from skin contamination during skinning. With regards to the site of sample collection, relatively similar prevalence of *E. coli* was recorded, from brisket (10%), from flank (8.2%) and (12.7%) was from thoracic. Thoracic area is more exposed to contamination than the other parts of the body and relatively similar with the prevalence (15.3%) reported by Ousman Mohammed *et al.* [26]. The presence of *E. coli* in sheep and goat meat might be due to contamination either from gastrointestinal content and/or skin [28]. The splashing of water during floor washing may also contribute to contamination of this area. The lower back area, similarly, is prone to contamination with fecal matter from tail which can come in contact with carcass after skinning. Samuel *et al.* [29] and Maja [30]

reported that feces and soil adhering to the animals are carried into the abattoir on the hair, hide, hooves and tail of the animal and become a major source of carcass contamination.

The prevalence of *E. coli* O157:H7 (3.67%) was relatively similar with prevalence (2.7%) reported by Isibor *et al.* [31] from Nigeria and less than prevalence (10.2%) reported by Bekele, Tizeta *et al.* [15] from Addis Ababa. This study shows that non-O157 STEC were the major cause of carcass contamination in the study area. Non-O157 STEC were also isolated in other countries like Germany, Italy and Denmark with higher frequency than O157:H7 strains [32]. Similar results were found in France and in Switzerland, Non-O157 STEC may also play a more important role in disease compared to STEC O157:H7 as shown in Argentina, Australia, Chile and South Africa [5]. In Canada, Japan, England and Scotland, in contrast, the prevalence of non-O157 is very low. However, the prevalence of *E. coli* O157:H7 in the United States was reported to be 1.3 to 4.0 per cent, with non-O157 strains 0.25 to 0.63 per cent of human isolates [32]. Results of this study support the fact that the occurrence of *E. coli* O157:H7 was very low, but other STEC played the main role in human infection.

**Antimicrobial Susceptibility Pattern of *E. coli*:** The use of antibiotics in the treatment of infection with *E. coli* is controversial, since antimicrobial therapy may increase the risk of development of HUS (Hemolytic uremic syndromes) [33]. Although some studies do not advise antibiotic treatment for infections caused by such bacteria [34], others suggest that disease progression may be prevented by administering antibiotic at early stage of infection [35]. Thus, for better response, antimicrobial susceptibility test is necessary [36]. Hence, on the basis of this necessity, antimicrobial susceptibility test was conducted on the isolates recovered from raw meat. Antimicrobial resistance of *E. coli* isolates from animal raw meat sources have been reported in Ethiopia by Hiko *et al.* [27].

In the present study *E. coli* showed intermediate susceptibility to two antimicrobials (kanamycin and sulphonamides) which varied from 40% to 60% and 75%- 100% susceptibility was noticed to oxytetracyclin, nalidixic acid, streptomycin and ceftriaxone. The 100% susceptibility of all the isolates to, ceftriaxone and nalidixic acid is similar with the findings of Rangel and Marin [37], Rahimi and Nayebpour [38] and 100% resistance to amoxycillin and trimethoprim was recorded.

The high resistance to amoxycilin in this study is in agreement with Hiko *et al.* [27] who reported antimicrobial resistance of *E. coli* isolates from raw meat samples to some of above mentioned antimicrobials especially to amoxicillin and 100% resistance to the trimethoprim were relatively similar with the finding of Bassam *et al.* [39] and contradict with Bekele, Tizeta *et al.* [15] who report 100% susceptibility to trimethoprim. There was susceptibility of 40-60% for kanamycin and sulphonamide. The significantly high level of resistance to these antimicrobials was probably an indication of their extensive usage in the veterinary sector for therapeutic and prophylactic purpose both for *E. coli* and other infections. Antimicrobial resistance emerges from the use of antimicrobials in animals and human and the subsequent transfer of resistance genes and bacteria among animals, humans and animal products and the environment [40]. The shedding of the resistant bacteria into the environment by cattle may lead to a widespread dissemination of antibiotic resistant genes to the resident bacteria in the environment [41]. Evidence has been found which indicates that resistant strains of pathogens can be transmitted to humans through food [42]. Antibiotic resistance among food borne pathogens may create an increased burden to human health through: it's potential to reach humans, increasing the risk of acquiring an infection in human who taking prior antibiotic treatment, limiting illness treatment options and may be by developing increased virulence [43]. Recently, multidrug resistant (MDR) phenotypes have been spread widely among Gram negative bacteria [44]. Most of these antimicrobials are not commonly used in Ethiopia in the treatment of animals that served as a source of meat. More over, the susceptibility might have contributed to the effectiveness of these antimicrobials mostly against Gram negative bacteria like those of the family of Enterobacteriaceae to which *E. coli* belongs.

## CONCLUSIONS

The results of the current study suggest that healthy sheep and goats slaughtered in Eflora abattoir from different areas of the country could be an important reservoir of highly pathogenic *E.coli* O157:H7 and be potential sources of infection for the public.

The prevalence of *E. coli* O157:H7 has shown no statistical association among different age group (young and adult), site of sample collection (Brisket, flank and thorax) in the animal's body and the origin of sampled animals (Borena, Harar and Jinka).

This study demonstrated that *E. coli* isolate used for the test exhibited a 100 % resistance for amoxicillin and trimethoprim.

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**Availability of Data and Materials:** The data sets developed and/or analyzed during the current study are available from the first author or from the corresponding auth or up on request.

**Limitation of the Study:** The isolated *Escherichia coli* O157:H7 species were not molecularly characterized due to unavailability of these resources to the laboratory facility in our country.

**Authors' Contributions:** SA, AY, and FA participated in conception of the research idea, methodology and review of the draft manuscript. SA carried out the laboratory work, sample collection and analysis and write up of the first draft.All authors read and approved the final manuscript.

**Ethics Approval and Consent to Participate:** The study was conducted after obtaining ethical clearance from the Jimma university school of veterinary medicine. Apermission letter was obtained from Addis Ababa university facult of veterinary medicine. Written informed consent was obtained from all study participants.

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