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Research Article

Food Safety, Isolation and Antibiogram *Escherichia Coli* Along Beef Value Chain in Chelenko Town, Eastern Ethiopia

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Abstract

Escherichia coli (E. coli) is a common bacterium that can cause significant diseases in both humans and animals. The growing threat of antimicrobial resistance (AMR) poses serious risks to public health and food safety, contributing to treatment failures, increased morbidity, and rising healthcare costs. This study, conducted in Chelenko town, Ethiopia, aimed to isolate E. coli and assess its antimicrobial resistance along the beef value chain. The cross-sectional study, carried out from March to September 2022, sought to isolate and identify E. coli in beef samples and evaluate the hygienic practices in abattoirs and butcher shops within Chelenko town, East Hararghe zone, Oromia State, Ethiopia. A total of 384 samples were collected, including 78 beef meat samples, 36 feces samples, 36 water samples, and 234 swab samples from abattoir and butcher staff. Additionally, semi-structured interviews and site observations were used to assess hygienic practices. E. coli was detected in 33 (8.6%) of the total samples, with 16 (7.41%) positive samples originating from abattoirs and 17 (10.12%) from butcher shops. In abattoirs, E. coli was isolated from 2.8% of meat, 2.8% of hand swabs, 2.8% of knife swabs, 19.44% of feces, and 2.8% of water samples. In butcher shops, it was detected in 21.4% of meat, 2.4% of hand swabs, 2.4% of knife swabs, and 11.9% of feces samples. Antimicrobial susceptibility testing revealed high resistance levels, with 97% of isolates resistant to Enrofloxacin, 78.8% to Oxytetracycline and Streptomycin, 72.8% to Tetracycline, and 63.6% to Gentamycin. All E. coli isolates showed complete resistance to Amoxicillin. The study also highlighted poor hygiene practices in both abattoirs and butcher shops, emphasizing the urgent need for improved food safety training and infrastructure to ensure better hygiene in the beef supply chain.

Keywords

Abattoir, Beef Meat, Butcheries, Chelenko, *Escherichia Coli*

1. Introduction

Despite outstanding advances in science and technology in developed countries in recent decades, microbial food-borne illness remains a global concern [1]. Food-borne disease is also common in developing countries, particularly in Africa, due to poor food handling and sanitation practices, insufficient food safety laws, a weak regulatory system, a lack of financial resources to invest in safer equipment, and a lack of education for food handlers [2]. Food-borne diseases are the leading cause of disease and mortality in underdeveloped countries, with related medical and societal costs in the bil-

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lions of dollars. Most notably, foodborne infections have a large impact on developing countries [3]. Contaminated food is a major source of food-borne illnesses. The majority of food-borne illnesses are caused by biological food contamination, specifically bacteria such as *E. coli*, salmonella, and S. aureus [2].

Escherichia coli is a major pathogenic agent that is commonly connected with foodborne diseases. Reports of *E. coli* as one of the deadliest foodborne bacteria producing serious sickness and high mortality rates in people have increased in the near past, particularly from throughout the world [5]. EHEC (entero hemorrhagic *Escherichia coli*), EIEC (entero invasive *Escherichia coli*), EPEC (entero pathogenic *Escherichia coli*), and ETEC (entero toxigenic *Escherichia coli*) are the five virulent classifications. The powerful strain of Escherichia coli O157:H7 from the genus *Escherichia coli* causes hemorrhagic colitis, hemolytic uremic syndrome (HUS), and thrombotic thrombocytopenic purpura. Human hemorrhagic colitis, hemolytic uremic syndrome (HUS), and thrombotic thrombocytopenic purpura (TTP) are frequently caused by *Escherichia coli* O157:H7, a virulent strain of the species [6].

A significant portion of *Escherichia coli* zoonotic transmission to humans comes through the ingestion of contaminated bovine consumables such as beef and dairy products [7-9]. *E. coli* are bacteria that live commensally in the gastrointestinal tracts of mammals, often without causing harm to the animals. Infection in humans can also occur as a result of direct contact with diseased animals or fecal contamination of other food products. Even though the majority of *E. coli* strains are not dangerous to humans, the presence of *E. coli* in foods intended for human consumption suggests poor hygiene during production, processing, or preparation [10]. *Escherichia coli* has emerged as a significant threat to public health worldwide, particularly in developing countries. The majority of outbreaks involving a large number of illnesses have been linked to eating undercooked or cross-contaminated prepared foods. Foods mostly of bovine origin, such as beef and beef burgers, as well as unpasteurized milk, are the most frequently associated to *Escherichia coli* outbreaks [11, 12].

Meat from animals may contain microorganisms that are dangerous to humans [13]. Zoonotic bacteria, especially pathogenic *E. coli* serotypes such O157:H7, are the most prominent [14, 15]. The main sources of carcass contamination are contact with the skin during removing the hide and contamination from stomach contents pouring out as the carcass is eviscerated. Furthermore, as part of the process of removing an animal's hide, some bacteria from the hide are released into the abattoir's air [14, 1]. There is a critical requirement to investigate this organism and its characteristics in order to lessen the harmful effect caused by this growing virus in poor countries where there is still an alarming prevalence of filthy living conditions, hunger, and a lack of health care facilities [16].

Previously, investigations on the danger of *E. coli* were conducted in Ethiopia, documenting the level of incidence in foods of animal origin, especially meat and milk [17-19]. Almost all of the studies were conducted in central Ethiopia, where there is a large animal husbandry business. Furthermore, studies that assess the organism's presence in foods of animal origin are being done across the country. The measurement and quantification of *Escherichia coli* prevalence at the national level can assist authorities in preventing and managing its occurrence in foods before it reaches end consumers, hence reducing its impact [20].

At the national level, Ethiopian meat and meat products continue to provide a problem for risk-based approaches to food safety since it is difficult to establish the extent of contamination with this virus. Furthermore, there is a food shortage and insufficient procedures in place to ensure food safety, both of which have an impact on the nation's economic development and public health safety [21, 22]. *E. coli's* intestinal home in animals allows simple access to animal meat at slaughter and downstream in the food production chain [23]. The greatest food safety challenge is potential contamination of edible carcass tissue, and the degree and form of such contamination are related to the *E. coli* state of the animal prior to slaughter and any activities impacting the organism within or between carcasses disseminated during dressing [24].

Foodborne illnesses are frequently underreported, and Ethiopia is no exception. Because of the lack of strict surveillance of foodborne pathogens in meat and meat products in Ethiopia, risk-based approaches to improving food safety have difficulties in demonstrating the amount of contamination with this disease [25]. The meat industry, including slaughterhouses and butcher shops, is highly susceptible to *E. coli* contamination, leading to spoilage and food-borne infections. The environment surrounding these establishments is a significant source of contamination, causing economic and health losses. Hence, more data is needed to understand the prevalence of *E. coli* in the meat industry [26].

Despite a growing demand for food safety standards in the Ethiopian meat industry, the government has yet to establish effective policies to enhance meat hygiene. This challenge is compounded by inadequate sanitary practices in abattoirs and butcheries, as well as issues like dark cutting, improper handling, careless packing, and poor management during transport. Beef is often processed in unsanitary conditions and distributed through informal channels, raising the risk of microbial contamination. Contributing factors include insufficient sanitation and sterilization of equipment, a lack of training on hygiene and foodborne diseases, widespread illiteracy, and poor personal hygiene among workers. These conditions pose significant health risks to consumers, lead to meat losses and quality deterioration, and result in economic setbacks. This study aimed to isolate and identify Escherichia coli along the cattle value chain while assessing sanitary practices in Chelenko to address these pressing issues.

2. Materials Methods

2.1. Description of Study Area

The research was carried out in two towns of Meta district, Chelenko town in Eastern Hararghe, Oromia, Ethiopia. The area is located 445 kilometers east of Ethiopia's capital city, Addis Ababa, and 80 kilometers west of Harar. It is geographically positioned between 9'55" and 9'28'45"N latitude and 41'31" to 41'52'30"E longitude (Figure 1). The Woreda is bounded to the south by Goro Muti, to the west by Deder, to the northwest by Goro Gutu, to the southeast by Bedeno, to the northeast by Kersa, and to the north by the Somali Region. Metta Word's elevation ranges from 1311 to 2830 meters above sea level. It has an annual rainfall of 600-900 mm and temperatures ranging from 15 to 37 degrees Celsius [27].

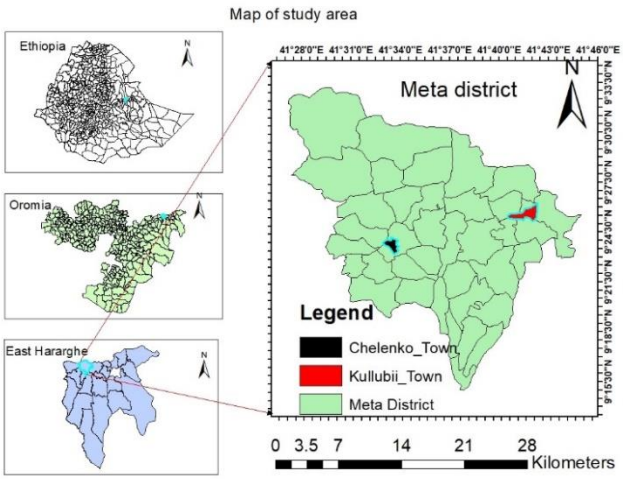


Figure 1. *Map of the study area.*

Source: ESRI. "World Topographic Map." ArcGIS Online, 2022. Figure caption.

2.2. Study Design

From March to September 2022, a cross-sectional investigation was done to identify *E. coli* in beef meat and ambient samples. In addition, the check list and questionnaire were used to examine the hygiene practices at the slaughterhouse and butchery shop.

2.3. Study Samples

At slaughterhouse and butchery shops, the swab samples have been collected from the carcass, knife, saw, hook, and workers' hands after slaughtering and skinning the animals. Water samples were also collected from the water tank and the tap used for washing the carcasses. At butcher shops, the swab samples were collected from the, cutting board, and from the displayed meat and the refrigerated meat (Table 1).

Table 1. *The amount and type of samples collected from abattoir, butcher houses and retailers.*

NO	Column2	Unit	N
1	Meat	25 g	78
2	Faecal sample	3 g	36
3	Water	9 ml	36
4	Swab sample		
	Workers hand	2 hand	78
	Knife swab	2 side	78
	Cutting tables	40 cm^2	78
	Total		384

*Table Footer.

2.4. Sample Size Determination

Using the Thrusfield formula [28], the approximate sample size required for the investigation was determined based on the expected prevalence of *E. coli* and desired absolute precision. The sample size for this study is determined at 95% confidence interval, 5% precision, and 50% predicted prevalence.

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

Where:

n = the required sample size

$Pexp$ = expected prevalence

d = desired absolute precision.

2.5. Sample Collection Procedure and Transportation

The slaughtered animal was chosen for sampling using systematic random sampling. Samples were taken aseptically from cattle slaughtered at the Chelenko abattoir, with six samples collected from the total number of slain cattle, including fecal and meat samples, and recorded in the appropriate manner. The hand, knife, and cutting board had been swabbed using the procedure for sampling beef carcasses and surface swab. To summarize, a sterile gauze pad of 7.6 cm x 7.6 cm was applied on certain areas. A sterile gauze pad was soaked in approximately 10 ml of buffered peptone water (Oxoid Ltd., Hampshire, England) and rubbed horizontally and then vertically numerous times [29].

All samples were labeled with the essential information, namely the date of sampling, sample code and sample type, sample weight, and aseptically inserted.

2.6. Laboratory Analysis

2.6.1. Isolation and Identification of *E. coli*

The detection of *E. coli* was carried out in accordance with the ISO 17604, (2005) standard protocol. The samples were then subcultured onto MacConkey agar for primary *E. coli* screening and incubated at 37 ℃ for 24 hours. The probable lactose positive *E. coli* colonies (red color) were then subcultured onto EMB. EMB agar was subcultured into nutrient agar and validated by Triple Sugar Iron and IMViC (Indole, Methyl red, Voges-Proskauer, Citrate utilization) assays on tryptone broth (Oxoid, England), MRVP medium (Oxoid, England), and Simon citrate agar (Oxoid, England), respectively [30].

2.6.2. Biochemical Conformational Test

The bacteria in the indole test can metabolize tryptophan for their capacity to create indole using the enzyme tryptophanase. Pyruvic acid, indole, and amino acids are formed as a result of enzyme breakdown. Kovac's reagent contains indole, which interacts with the aldehyde to form a crimson or pink ring at the top of the tube. Bacterial isolates were inoculated in a tube with tryptophan-containing peptone water, and the mixture was incubated at 37 ℃ for 24 hours. After a few drops of Kovac's reagent were added to the mixture, the ring formed. A red ring at the top progressed in a positive direction. The *E. coli* result is then indole-positive [5, 31].

The Methyl-red test detects microorganisms that can produce acid from glucose fermentation. The bacteria were evaluated before being placed in glucose phosphate (MRVP) broth, which contains glucose and a phosphate buffer, and cultured for 48 hours at 37 ℃. Then, in the tube, add three to five drops of MR reagent. When the bacteria produce enough acid to counteract the phosphate buffer, a positive response known as red color creation occurs. Yellow is the color of MR-negative bacteria. *E. coli* is a bacterium that has MR [5, 31].

The test bacteria have been placed in a tube containing glucose phosphate (MRVP) broth and grown for 72 hours for the VP test. The test was then shaken with 15 drops of alpha-naphthol. Shake the soup after adding five drops of 40% potassium hydroxide (KOH). Allow the tube to sit for 15 minutes before looking for a bright red color. If the color of the isolate does not change after an hour, it is called VP negative. The Voges-Proskauer test can be used to detect the presence of acetone in bacterial medium. When acetone, sodium hydroxide, and oxygen are present, diacetyl is formed. The interaction of guanidine with diacetyl creates a red color in the presence of alpha-naphthol [5, 31].

The citrate utilization test assesses the bacteria's ability to use citrate as its only carbon and energy source. Bromothymol blue is a PH indicator found in citrate agar media. At an alkaline PH, the agar media turns blue rather than green. A loopful of bacteria should be placed over a citrate agar slant without sticking the bottom of the body, and it should be incubated at 37 ℃ with a loose cap for 24 hours. Citrate in the medium is broken down into oxaloacetate and acetate by the citrate enzyme. Oxaloacetate is then oxidized further, producing pyruvate and CO_2. When sodium citrate is converted into Na_2CO_3, the medium's pH shifts to an alkaline state, causing the medium to turn blue [5, 31].

In general, the biochemical properties of the *E. coli* isolate were positive for catalase, Methyl red, and Indole tests but negative for Voges-Proskauer, urease, and citrate. In addition, reactions in TSI agar slant indicated yellow but with gas and the generation of hydrogen sulfide. Almost all *E. coli* isolates fermented lactose, sucrose, and glucose, producing acid and gas in the process. Lactose fermentation was found to be positive, Simmons' citrate was found to be negative, Indole production was found to be positive, and Nitrate reduction was found to be positive. Methyl Red is positive, and Voges-Proskauer is negative [5, 31].

2.6.3. Antimicrobial Susceptibility Testing

The Clinical and Laboratory Standards Institute's agar disc diffusion technique was used to isolate *E. coli* and perform antimicrobial susceptibility tests [32]. Amoxicillin, Gentamicin, Streptomycin, Tetracycline, Oxytetracycline, and Enrofloxacin were among the antibiotics tested for resistance. A sterile cotton swab was dipped into the standardized bacteria suspension and then streaked consistently across the whole surface of the Mueller-Hinton agar. Each isolated bacterial colony was put into a 5 ml test tube of Tryptone Soya Broth (TSB) (Oxoid, England) and cultured at 37 ℃ for 16-24 hours. The turbidity of the culture broth was adjusted using sterile saline solution or by adding more isolated colonies to achieve turbidity similar to 0.5 McFarland standards (about 3 x 108 CFU per ml). After 24 hours, clear zones of inhibition created by bacterial growth and antibiotic diffusion, which were measured in millimeters with a caliper or ruler and interpreted as susceptible, intermediate, and resistant [33].

The study used antibiotic discs made of agar to test E. coli drug sensitivity. The discs were impregnated with specific antibiotics, such as Amoxicillin, Gentamycin, Streptomycin, Tetracycline, Oxytetracycline, and Enrofloxacin. The antibiotics inhibited E. coli growth, creating a clear zone. The concentration of the antibiotic in the discs, expressed in micrograms per milliliter (μg/ml), affected the size of the zone, determining the antibiotic's susceptibility.

2.6.4. Questionnaire Survey

A descriptive survey design was made to obtain information about the hygienic condition of the abattoir and butchery shops in which workers in relation to meat processing and handling. And also, observational checklist was prepared to assess environmental hygiene, cleanliness of food, and food handling practices during each visit. The questionnaires were prepared in English, but during the interview, the questionnaire was translated according to [34] into the pre-

ferred language of the respondents, particularly, Afaan Oromo.

2.7. Data Management and Analysis

Data from the research area and laboratory studies were entered into Microsoft Excel, modified, coded, and analyzed with SPSS version 26. The frequency and proportion of the results were described using descriptive statistics. *E. coli* prevalence was calculated by dividing the number of positive samples by the total number of samples examined. The data was then subjected to chi-square analysis to determine the relationship between similar variable connections of possible risk variables and *E. coli* contamination. With 95% confidence intervals, relationships were considered significant when the p-value was less than 0.05.

3. Results

3.1. Isolation Prevalence of *E. coli*

The study included 384 samples from beef meat and environmental samples (slaughter house workers' hands, knives, and cutting boards), 216 from abattoirs, and 168 from butchers and retailers. 33 (8.6%) of 384 distinct samples tested were positive for *E. coli* at the Chelenko slaughterhouse and butchery shop. Among the positive samples, 17 (10.12%) were from meat butcher shops and 16 (7.41%) from the abattoir (Table 2).

Table 2. *The Occurrence of E. coli isolated from different sample source.*

Sample Source	Examined sample	Positive (%)	X2	P value
Abattoir	216	16 (7.41%)		
Butchery	168	17 (10.12%)		
Total	384	33 (8.6%)		

*Table Footer.

The overall occurrence of *E. coli* among the 216 samples evaluated was 16 (7.41%) at the Chelenko abattoir and 168 samples at the butchery shop (17 (10.12%), as indicated in Table 6. The abattoir had 11.11% positive meat samples, 2.8% positive hand swab samples, 2.8% positive knife swab samples, 19.44% positive fecal samples, 2.8% positive water samples, and 5.56% positive cutting board swab samples, while the butchery had 21.4% positive meat samples, 4.8% positive hand swab samples, 2.4% positive knife swab samples, and 11.9% positive cutting board swab samples (Table 3).

Table 3. *Isolation frequency of E. coli from different sample type.*

Sample Source	Sample type	Examined	Positive (%)	X2	P value
Abattoir	Meat	36	4 (11.11)		
	Hand swab	36	1 (2.8)		
	Knife swab	36	1 (2.8)		
	Feces	36	7 (19.44)		
	Water	36	1 (2.8)		
	Cutting board	36	2 (5.56)		
	Total	216	16 (7.41)	21.130	0.001
Butchery	Meat	42	9 (21.4)		
	Hand swab	42	2 (4.8)		
	Knife swab	42	1 (2.4)		
	Cutting board	42	5 (11.9)		
	Total	168	17 (10.12)	19.280	0.002

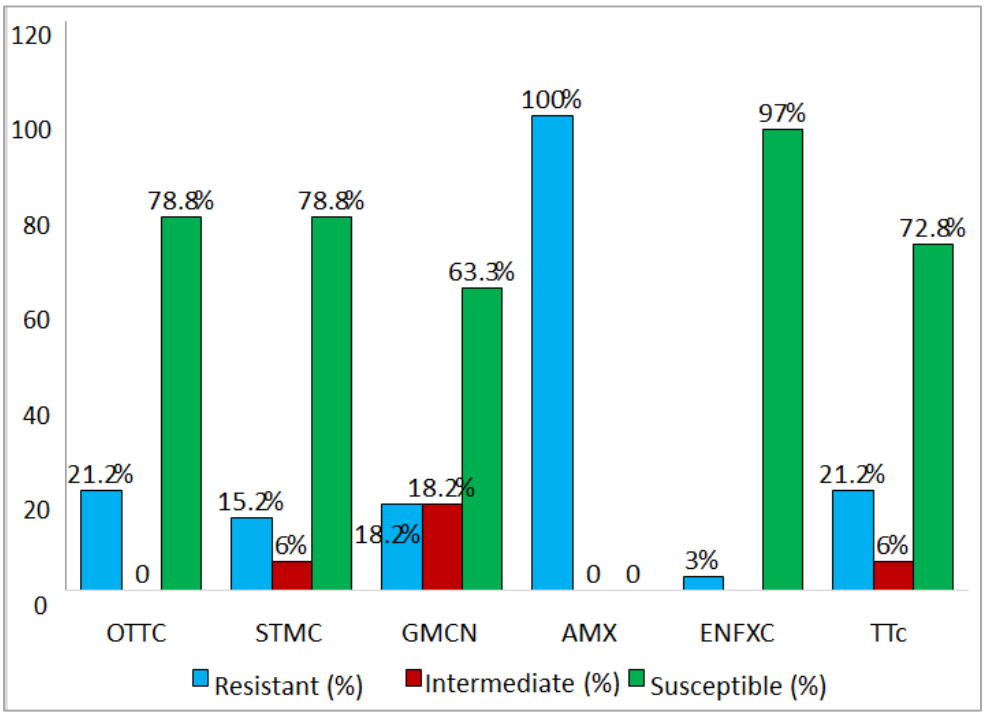
3.2. Antimicrobial Susceptibility of *E. coli*

Following CLSI recommendations, isolates from 33 E. coli were screened against six antimicrobials. These E. coli isolates were shown to be 97% susceptible to Enrofloxacin, 78.8% susceptible to Oxy-tetracycline and Streptomycin, 72.8% susceptible to Tetracycline, and 63.6% susceptible to Gentamicin. All E. coli isolates, on the other hand, were found to be completely resistant to amoxicillin. As a result, the most effective medications against E. coli were Enrofloxacin, Oxytetracycline, streptomycin, tetracycline, and gentamicin (Table 4).

Table 4. *E. coli isolates and their susceptibility to antimicrobial drugs.*

No	Drug	Concentration	Frequency of E. coli isolation		
			Resistant	Intermediate	Susceptible
1	Amoxicillin	20 µg/ml	33(100%)	-	-
2	Gentamicin	10 µg/ml	6(18.2%)	6(18.2%)	21(63.6%)
3	Streptomycin	10 µg/ml	5(15.2%)	2(6%)	26(78.8%)
4	Tetracycline	30 µg/ml	7(21.2%)	2(6%)	24(72.8%)
5	Oxytetracycline	30 µg	7(21.2%)	-	26(78.8%)
6	Enrofloxacin	10 µg	1(3%)	-	32(97%)

All of the isolates were tested positive for several drugs. A multi-drug test revealed that about 98% of the *E. coli* isolates were sensitive. The test result showed that *E. coli* was highly resistant with 100% and susceptible with 97%, to Amoxicillin and Enrofloxacin respectively (Figure 2).



Hint: - OTTC=Oxytetracycline, STMC=Streptomycin, GMCN=Gentamycin

Figure 2. *Antimicrobial susceptibility pattern of E. coli.*

3.3. Findings of Questionnaire Survey

3.3.1. Status of Slaughterhouse Employees

The two sets of respondents included both permanent and temporary workers. Permanent employees are referred to as "permanent personnel," and they work in abattoirs permanently, whereas contract employees are referred to as "temporary staff," and they work in abattoirs by expanding their job with the approval of a cooperative group. In the abattoir, 18% are permanent employers and 82% are contract workers. The abattoir was surveyed with a total of 30 respondents. About 33.3% of abattoir workers have completed elementary education, 40% have not attended school, and 26.7% have completed high school. Although none of the interviewees chewed or smoked, some admitted to drinking alcohol while working (Table 5).

Table 5. Demographic information of both abattoirs and butchery shops workers.

Variables	Respondent	Butchery (n=40)		Abattoir (n=30)	
		N	%	N	%
Age	20-28	8	20	3	10
	29-35	20	5	20	66.6
	36-45	6	15	7	23.4
	>46	6	15	-	-
Educational status	Non school	10	25	12	40
	Elementary	21	52.5	10	33.3
	High School	6	15	8	26.7
	College	3	7.5	-	-
Employment Status	Permanent	-	-	5	16.7
	Temporary	40	100	25	83.3
Work experiences	>5 Yrs	6	15	18	60
	6-10 Yrs	26	65	9	30
	>10 Yrs	8	20	3	10

3.3.2. Assessment of Hygienic Practices

A large number of employees have never been taught any of their responsibilities. Those who had not received training were much more aware of the risks associated with food safety and slaughtering cleanliness. The majority of the monitoring was not centered on hygiene, and not all of the personnel had taken the needed medical checks to work in the slaughterhouse. Few abattoir workers learned their trade from their parents, while the majority of respondents learned through visual observation. The majority of responders stated that they had no training in sanitary techniques.

The protective apparel was filthy, covered with blood, and frequently in touch with carcasses, and none of them worked while wearing an apron, a face mask, gloves, or a hair cover. There are no aprons, white coats, boots, or hair coverings available, nor are sinks for hand washing available in the slaughterhouse. Slaughtering implements were put on dirty surfaces during our visit. Knives were strewn around the floor and on the skins of slaughtered animals, and a single knife was used to carve meat and offal. However, all of the workers washed their instruments in buckets rather than with running water, and they did so while working on dirty surfaces. Every day before commencing work, many of them cleaned their blades, which had become excessively and visibly dirty with fat or blood.

A total of 30 abattoir employees were interviewed. Approximately 40% of workers are uneducated, 33.3% are elementary school graduates, and 26% are high school graduates. Personal hygiene lessons were not taken by 83.3% of respondents, whereas they were taken by 16.7%. During working hours, approximately 56.7% and 43.3% of respondents wash their hands twice and once each day, respectively. In addition, 20% of respondents reported using a detergent to wash their hands after using the restroom (Table 6).

Table 6. Summary results of hygiene practices at slaughter house and butchery shops.

		Abattoir		Butchery	
Variables	**Respondent**	**Frequency**	**%**	**Frequency**	**%**
Lesson on personal hygiene	Yes	5	16.7	3	7.5
	No	25	83.3	37	92.5
Hand washing interval	Once	17	56.7	14	35
	Twice	13	43.3	26	65
Washing hands after toilet	Yes	20	66.7	36	90
	No	10	33.3	4	10
Washing of hands	Water only	24	80	34	85
	Use Soap	6	20	16	15
Used Protective cloth	Yes	16	53.3	35	87.5
	No	14	46.7	5	12.5
Wash cloth	Yes	8	26.7	14	35
	No	22	73.3	26	65
Hair cover	Always	0	0	14	35
	Sometimes	2	6.7	25	62.5
	Never	28	93.3	1	2.5
Washing of knives	Before	2	6.7	15	37.5
	After	21	70	17	42.5
	Between	7	23.3	8	20
Working surface cleaning interval	Once	23	76.7	18	45
	Twice	7	23.3	22	65
Training about foodborne disease	Yes	3	10	0	0
	No	27	90	40	100
Medical tests	Yes	2	6.7	0	0
	No	28	93.3	40	100
Wear jewelry	Yes	7	23.3	9	22.5
	No	23	76.7	31	77.5
Money taking	Butchery	-	-	11	27.5
	Cashier	-	-	29	72.5
Status closet	Clean	-	-	25	62.7
	Dirty	-	-	15	38.3
Availability of toilet	Yes	18	60	38	95
	No	12	40	2	5
Waste removal and storage	Yes	4	13.3	8	20
	No	14	46.7	32	80
Medical tests	Yes	3	10	18	45
Mouth/nose pipetting	Yes	22	73.3	37	92.5

4. Discussion

Hygienic approaches are important routes for producing safe and high-quality products for consumers, reducing microbial contamination [35]. Because contaminated hands of food workers spread many foodborne infections, hand cleanliness is regarded as one of the most efficient techniques of preventing foodborne disease [36]. The source and kind of

water used for washing the hands of slaughter crew and utensils have an important impact on microbial contamination of the meat and are a good source of the causes for the mode of transmission of the food borne disease. Approximately 85% of those interviewed at abattoirs and butcher shops used simply water to wash their hands, whereas 15% used both water and soap [37].

According to the current study, the interviewed respondents just washed their hands before handling meat and had no sanitary regulating mechanism in place. Employees in the food industry may be carriers of foodborne diseases. As a result, inappropriate personal hygiene practices such as not washing hands after using the restroom or engaging in non-food related activities, a lack of periodic medical health examinations, and careless sneezing and coughing can contaminate meat and represent a public health risk [38]. As a result, sanitation and hygiene training should improve personal behavior and attitudes while also imparting knowledge [39].

The microbial contamination was caused by unclean slaughtermen's hands, clothing, and equipment used in the carcass dressing process [40]. The majority of abattoir and butcher shop employees understand that personal protective equipment (PPE) is employed for personal protection rather than meat contamination prevention. This revealed that the workers were aware that viruses from meat could affect them but were unaware that meat could be infected by them, which also occurred due to a lack of ability and training. The examinations revealed that the butcher shop workers' bodies, notably their hands and the tools they use, were heavily polluted by harmful bacteria that might potentially transmit to meat. Butcher shop personnel are unable to prevent the spread of bacteria from workers to meat because they only comprehend the one-way benefits of PPE.

About 70.4% of those surveyed did not use protective clothing when working in butcheries and abattoirs, and they learned how to butcher from observation and their parents. The present result was agreed with the study results that reported in Ethiopia by Bersisa *et al.* [41] and eastern Nepal by Bhattarai *et al.* [42], in Aden University by Sallami [43]. In contrast, 46% of 70 interviewed workers wash their work coat once a week. According to the study's findings, the majority of abattoir and butcher personnel lacked information about hygienic meat handling and the impact of personal hygiene on meat safety.

In the current study, 70 abattoir and butcher shop workers were interviewed regarding their meat hygiene knowledge, butchering skills, sanitary control system, and meat related disease awareness. According to the workers interviewed, their butchery lacked a hand sanitary regulatory guideline and a sanitary regulation system. This could be due to a lack of training in meat handling, personal and environmental hygiene for their employees, butchery owners, and businesses who hire on a contract and daily basis and this agreed with report of Hogan *et al.* [44].

In this survey, all respondents were asked if they were aware of any disease that may be passed from animal to human. The study's findings revealed that only a small number of respondents were aware of zoonotic diseases in Chiro. The diversity in the provision of information about zoonotic diseases and food habits could explain the general awareness in the research sites for zoonotic diseases. Because of their lack of knowledge, butcher workers are more susceptible to get infections caused by zoonotic agents. In general, the interviewed workers' response result show in the present study, there was low awareness on the foodborne diseases. The present result was in contrary with that of Zelalem *et al*. [45] and Seifu and Sintayehu [46], who indicated that all the respondents in Addis Ababa were mentioned about zoonotic diseases.

The lack of understanding of zoonotic illnesses in the current investigation region could be attributed to weak or missing awareness creation actions by government medical and veterinary health care personnel. Due to a lack of water, awareness, and competent management, the abattoir and butchery, which is part of the research site, was unable to complete its usual cleaning and disinfection activities. The butchers were cleaned twice a day, before and after sales. A deeper check of the tables and other furniture revealed that no one had cleaned them in a number of days, contrary to the butcher's claim. The location and design of the abattoirs were not correctly chosen, since they were built in the town center, and the environment was heavily polluted as a result of waste product released from the abattoir being down to the water that the town used for various purposes.

The overall prevalence of E Coli was 33 (8.6%) in both abattoir and butchery houses, with 16 (7.41%) and 17 (10.12%) discovered from a total of 384 analyzed samples. The current investigation found that samples from the Wolaita Sodo abattoir and butchery shops were statistically significant (p 0.05). Cross-contamination of meat and meat-associated contact surfaces along the supply chain in abattoirs and butcher shops may have led to this. Following slaughter and dressing, animal carcasses can be contaminated with primarily enteric bacteria, including *E. coli*, originating from the skin, hair, gastrointestinal system, and the atmosphere at slaughtering facilities. Another aspect is that due to a lack of lariage, the animals were slaughtered as soon as they arrived at the abattoir from the owner's residence, and they stayed in one area before to slaughter. This drives the animals to fight, resulting in stress and the release of pathogens via feces [47].

The current finding is lower than different finding in Ethiopia, 17.29% in Dire Dawa ELFORA abattoir, 24.48% Dire Dawa Municipality abattoir [48], 29.55% in Addis Ababa [49], 27% in Mekelle [2], 35.5% in Bishoftu and 40% in Mekelle [50], 22.2% in Mekelle [2], 20.2% in Jimma [51], 35% of *E. coli* from cow hide in Nigeria [52], and 37.86% in Bangladesh [53], 62.26% in the Khon Kaen [54], 46.5% in Nigeria [23], 32.5% in Indonesia [55], 35.21% in India [56], 49% and 73% from formal meat sector informal meat sectors

respectively in South Africa [57] and higher than 0.44% in Algeria [58].

This variation in the current study findings could be attributed to differences in study design, methods of detecting isolates, isolation technique, differences in sample type, size, and sampling techniques, and differences in the quality of work, storage, and processing conditions, slaughterhouse hygienic measures, and different rates of carcass contamination during the slaughtering procedure, as well as cross-contamination in the abattoir environment and butchery shop [46].

In the current investigation, *E. coli* was found in 16 (7.41%) of abattoirs and 17 (10.12%) of butchery shops, respectively. Butchery shops had a higher prevalence (10.12%) than Sodo abattoirs (7.41%). This variation could be attributed to variances in the butcher shop's hygienic and sanitary practices, the environment in which the meat was sold, the water used in the meat's processing, the mode of meat transportation to the butcher house, and the state of the butchery house. Furthermore, due to the butchery house worker experience, hygiene and sanitation status of the workers, because all of the workers did not receive training, and workers acted carelessly in retail houses.

E. coli prevalence at the meat level was 11.11% in slaughterhouse meat and 21.4% in butchery shop meat. The incidence of *E. coli* in butchery shop meat was greater (21.4%) than in slaughterhouse meat (11.11%). This variance could be attributed to the time of meat sample collection, the state of the meat lay out, the hygiene of the hook, the hygiene of the knife used to cut the meat, and the sanitary condition of the butchery shop.

The prevalence of *Escherichia coli* at the Chiro abattoir was 7.41%, which was lower than a prior study rate reported in Ethiopia, which was 19.3% in Jimma [51], 24.48% in Dire Dawa Municipality abattoir [48], 28% in Bishoftu [41], and 22.2% in Mekelle [2].

And also, the current result was a higher prevalence than that of reported 2.65%, by [20], 7.29% in Dire Dawa ELFORA abattoir [48], and from other countries, 1.8% in United State, and 3.2% in United Kingdom [24]. The version of the result was attributable to abattoir hygiene, sample methodologies, isolation procedure, personal performance, and worker quality.

The Current result of *E. coli* obtained in meat sample at the Chiro abattoir were 11.11% low compared with the previous study result, 28% in Bishoftu [41], 15% in Adigrat, 20% in Jimma [51], 27.3% in Mekelle municipal abattoir [2], and 46.5% reported in Nigeria [23].

When compared to prior results from various regions throughout the world, the new study indicated a lower prevalence rate, 21.1% in Jimma [51], and higher than 4.7% in Haweresa [18]. This could be due to the season in which the sample was collected, animal diet, or geographical location.

The prevalence of *E. coli* was 9 (21.4%) from meat samples, 2 (4.8%) from hands, 1(2.4%) from a knife swab sample, and 5 (11.9%) from a cutting board at the Sodo butchery business. The current prevalence value for *E. coli* obtained at the butcher house in Sodo on meat sample (21.4%) was slightly higher than the 12.5% at Jimma town abattoir [51], and 0.44% in Algerian frozen meat [58], 0.4% in France [59], 1.7% in goat meat and 1.3% in camel meat in Iran [60], 0.29 % [61] from Nigeria in beef meat, and 1.6% in Eldoret in Kenya reported by [62], 3.6% of the prevalence of *E. coli* in Dagoretti in Kenya [62], 8.3% from Iran [63] and low compared with 53.6% in beef meat in Northwest Turkey [64], 48.2% in ground beef meat and 25% in beef meat sample [64], 30% of ground beef [65], 24.8% in Dire Dawa [48], 28.9% reported in Jimma town butchers shop [51], 22.2% in Mekelle [50]. The current variation in the meat samples from the previous study could be attributed to the hygienic and sanitary condition of the butchery house, the building condition, meat handling practices of workers, materials used in butchery, and meat transporting condition.

E. coli was found in 11.9% of Chiro butcher businesses via cutting board swab samples. This finding (11.9%) from butcher shop cutting boards was greater when compared to prior research in other locations; 4% in Addis Abeba, 3.3% in Bishoftu, and 6.67% in Holeta [66], 10% from cutting boards in Jimma, and 3.6% in central Ethiopia [66] and low compared with 36% in Bishoftu [41], 23.3% in Jimma town butcher's shop [51]. This could be attributed to poor cleanliness of equipment and workers, as well as the sanitation of the butcher house. The current prevalence result for *E. coli* obtained at the butchery shops on hand was 4.8%, which was lower than the 12.5% found in Jimma town butchery shop [51], 87.5% in Bishoftu and Mojo [67]. The current achieved result of 2.4% of the knife at butchery houses was low in comparison to the prior study findings of 16.7% of the knife in Jimma Town butchery shops and 20.0% in Jimma municipal abattoir [51], 28% in Bishoftu [41], 31.25% in Bishoftu and Mojo [67].

In general, there was a statistically significant difference in *E. coli* prevalence between Chiro abattoir and meat retail outlets. The level of significance was discovered to be (p-value 0.05) which was 0.002. This variation could be related to hygiene differences, a lack of training, and meat handling techniques in the abattoir and butchery house. Antimicrobial resistance has been identified as a growing global issue in both human and veterinary medicine in both developed and developing countries. Antimicrobial resistance occurs as a result of antimicrobial use in humans and animals, as well as the subsequent transmission of resistance genes and bacteria across animals, people, and the environment [68].

The release of antibiotic-resistant bacteria by cattle into the environment has the potential to widely distribute such genes among local microorganisms [69, 70]. The resistant pathogen strains can be transmitted to people through food [71]. Antibiotic resistance among foodborne pathogens may increase the burden on human health by increasing the risk of infection in humans who have previously received antibiotic treatment,

limiting illness treatment options, and possibly increasing virulence [72].

In this investigation, 33 *E. coli* isolates were screened against six antimicrobials using CLSI criteria. Among those *E. coli* isolates, 97% were sensitive to Enrofloxacin, 78.8% to Oxy-tetracycline and Streptomycin, 72.8% to Tetracycline, and 63.6% to Gentamicin, which was consistent with the findings of Hiko *et al.* [73]; Mude *et al.*, [74] and disagree with that of Negesse Welde *et al.* [75].

The current study revealed that all isolates were highly resistant to Amoxicillin and this result is in line with similar findings were reported by Mora *et al.* [76]; Srinivasan *et al.* [77]; Taye *et al.* [20]. The resistant level of Gentamycin was agreed with Negesse Welde *et al.* [75] and also the level of streptomycin resistant was agreed with that of reported in Bishoftu Elfora export Abattoir and Addis Ababa [78].

In the present study, the *E. coli* isolates show two or more drug resistance. The results of this study are comparable to those of earlier studies Guerra *et al.* [79]; Zhao *et al.* [80]; Akond *et al.* [81]. This might be due to inappropriate use of antibiotics for treatment of diseases [82] and also excessive use of antimicrobials for therapeutic and prophylactic treatment [83]. This variance is most likely due to the pathogen's development of resistant gene code, which is related with emerging and re-emerging characteristics of the isolates in terms of varied agro ecology [84].

5. Conclusion and Recommendations

Butcheries are crucial in the meat production and processing chain, but they also pose health risks due to substandard facilities and unhygienic practices. Escherichia coli, a microbial contamination, is a significant contributor to health issues. The prevalence of *E. coli* isolates was 8.6% in both abattoirs and butcheries, with higher occurrences in feces, water, meat, and utensils. A preventative approach is needed to control *E. coli* in beef meat production. This study reveals poor personal and general hygiene practices among slaughter staff, including non-food activities, lack of medical health examinations, and careless sneezing and coughing, which can contaminate meat and pose public health concerns.

Based on the above conclusion, the following recommendations are forwarded:

1) The government should enforce strict regulations and inspections on the butcheries and abattoirs to ensure compliance with hygiene standards and prevent microbial contamination of meat products.
2) The slaughter staff should undergo regular medical examinations and training on personal and general hygiene practices, such as washing hands, wearing gloves, and avoiding non-food activities in the slaughter area.
3) The consumers should be educated on the health risks of *E. coli* and the proper handling and cooking of beef meat to avoid infection and illness.
4) The researchers should conduct further studies on the sources and transmission of *E. coli* in the meat production and processing chain, as well as the potential interventions and treatments to control and eliminate the bacteria.

Abbreviations

EHEC	Entero Hemorrhagic *Escherichia coli*
EIEC	Entero Invasive *Escherichia coli*
EPEC	Entero Pathogenic *Escherichia coli*
ETEC	Entero Toxigenic *Escherichia coli*

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Author Contributions

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The data is available from the corresponding author upon reasonable request.

Conflicts of Interest

The authors declare no conflicts of interest.

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Biography

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