

E. coli O157:H7 and *Salmonella* Species: Public Health Importance and Microbial Safety in Beef at Selected Slaughter Houses and Retail Shops in Eastern Ethiopia

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Abstract

A cross-sectional study was conducted from March 2014 to January 2015 in Dire Dawa city and Haramaya University slaughterhouses and retail shops to identify major foodborne pathogens, assess the beef microbial safety and potential contamination risk factors. 320 beef and environmental pooled samples were examined for the presence of *E. coli*, *E. coli* O157:H7 and *Salmonella* in accordance with international standard guidelines. On each visit of the slaughter house a total of four pooled swab samples were taken each from cleaned, disinfected and dry surfaces, other from hooks, knives and aprons, the third from personnel's hands who works on flaying, evisceration and carcass cutting before the beginning of operation. For isolation and identification of pathogens from meat, 25 gram of sample was weighed, cut in to small piece with different sterile scalpel blade and placed into sterile stomacher bags. The questionnaire survey data and laboratory results of the collected samples were entered into databases using Micro-Soft Excel computer program and analyze using SPSS version-19.0. Of 290 total beef samples collected, *E. coli* was isolated from 36(12.41%) and of which 6(2.06%) were confirmed on Sorbitol MacConkey agar to be *E. coli* O157 H7. On the other hand 8(2.75%) *Salmonella* spp. was identified using culture and biochemical tests. The present study indicated significantly higher fecal coliforms counts in beef samples from HU slaughterhouse (7.50 log₁₀ cfug⁻¹) than in carcass sample from Haramaya retail shop (4.80 log₁₀ cfug⁻¹). Out of 30 environmental pooled samples, *E. coli*, *E. coli* O157:H7 and *Salmonella* were investigated in 7(23.33%), 2(6.66%) and 2(6.66%), respectively. From the visual observations: slaughterhouse, hygienic practice employed, personnel habit were found below the minimum standards. Therefore, good management and hygienic practices should be introduced in order to enhance the overall safety and hygienic quality of beef for the safeguard of consumers.

Keywords: APC; Beef; *E. coli*; *E. coli* O157:H7; FCC; *Salmonella*; Slaughterhouse

Introduction

Foodborne pathogens are one of the leading causes of illness and death in the world. They place heavy burden costing billions of dollars in medical care, social costs and overall economic and infrastructure effects of countries [1]. It mostly affects developing countries, due to major contributing factors such as overcrowding, poverty, changes in eating habits, mass catering, complex and lengthy food supply procedures with increased international movement, inadequate sanitary conditions and poor general hygiene practices [2,3]. In developing countries including Ethiopia up to 2 million people die per year due to disease of foodborne pathogens [4]. Over the last 20 years, the emergence of major foodborne pathogens such as *Salmonella* and *E. coli* have persisted as a major public health concerns and provide clear examples of the persistence of foodborne pathogens despite considerable efforts aimed at prevention and control [5].

Meat processing at retail level is likely to contribute for the higher levels of contamination in minced beef as compared to carcasses. The presence of even small numbers of pathogens in meat and edible offal may lead to heavy contamination of minced meat when it is cut into pieces and the surface area of the meat increases; as more microorganisms are added to the surfaces of exposed tissue [6]. Previous studies conducted in many parts of the country indicated the occurrence of pathogens including *Salmonella* in different food animals, meat and meat products. In addition, outbreaks of infections related with poor hygiene and consumption of contaminated food were reported in Ethiopia where some were by *Salmonella* and *E. coli* [7-9].

In Ethiopia, the widespread habit of raw beef consumption is a potential cause for foodborne illnesses besides the common factors such as overcrowding, poverty, inadequate sanitary conditions and poor general hygiene [7]. Raw meat is available in open-air local retail shops without appropriate temperature control and this is purchased by households and also minced meat (Kitfo) is served as raw, slightly-cooked or well-cooked in Dire Dawa administrative city and Haramaya University. Therefore, the main objectives of this study were to determine the microbial safety of beef through isolation and identification of foodborne bacterial pathogens, assess potential sources of beef contamination, investigate hygienic conditions, practices and hygienic quality of beef in slaughterhouses and retail meat shops.

Materials and Methods

Study area and population

The study was conducted at slaughterhouse and ten retail shops

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in Dire dawa administrative city and in slaughterhouse and one retail meat shop in Haramaya University from March 2014 to January 2015. Cattle, camel, sheep and goats were the main animals slaughtered at Dire dawa municipal abattoir and additionally small ruminants were also slaughtered at different hotels and restaurants in the city. On the other hand, cattle are the only animal slaughtered at Haramaya University slaughterhouse for students' (cafeteria) and staff lounge consumptions.

Study protocol

A cross-sectional study was conducted to determine the microbial safety and hygienic quality of beef samples drawn from municipal slaughterhouse and retail meat shops. In addition checklist and interview were made on food handlers working at food establishment, to determine the hygienic status of the premises and safety practices of meat handlers. In the present study beef samples and environmental pooled samples were collected from slaughterhouses and retail shops in both study areas.

Semi-structured questionnaire was developed and distributed for 50 slaughterhouse workers to assess their understanding and knowledge in handling meat safely, personnel practices regarding prohibited habits and actions, personnel hygiene and educational backgrounds. The abattoir visit and direct assessment was also conducted to support questionnaires in testing the hygienic and sanitation practices employed at the slaughterhouses.

Sample size determination

The sample size required for this study was determined according to [10] by taking 5.6% and 3% previous prevalences for *Salmonella* and *E. coli* O157:H7 at Dire Dawa [11] and HU [12] respectively; assuming that the slaughter animals had come from the same origin in both study areas. Accordingly, 81 beef samples from slaughterhouse and the other 81 from 10 randomly selected retail shops (from 542 total retail shops) in Dire Dawa. However, to increase precision of the study, sample size was increased by 1.23 folds and a total 290 beef sample (200 from Dire Dawa and 90 from HU) were collected. Additionally, 30 environmental pooled samples (from equipment, surface, workers hands, vehicles and etc.) were collected during the study (Table 1).

Sample collection and laboratory techniques procedures

The microbial safety and hygiene quality were then assayed by using the methods recommended by International Commission on Microbiological Specifications for foods [13]. All the samples were investigated with respect to *Salmonella*, *E. coli* and *E. coli* O157:H7 detection and aerobic plate and fecal coliforms counts.

On each visit of the slaughter house a total of four pooled swab samples were taken each from cleaned, disinfected and dry surfaces, other from hooks, knives and aprons, the third from personnel's hands who works on flaying, evisceration and carcass cutting before the beginning of operation. The fourth from the surface of transporting vehicles by rubbing thoroughly with a moistened swab. The pooled environmental sample collections were conducted two times within three months. In each visit of each retail shops a total of three pooled swab samples were taken each from cutting boards and meat grinder, other from hooks, knives and protective cloth. the third sample from personnel's hands (butcher men) before the start of their routine work, by rubbing thoroughly with a moistened swab. The samples were then returned to a test tube containing 9 ml sterile buffered peptone water (BPW). All samples were transported to the Microbiology Laboratory

of College of Veterinary Medicine, HU using ice box on ice packs and then the samples were analyzed upon arrival and sometimes within 24 hours of sampling.

For isolation and identification of pathogens from meat, 25 gram of sample was weighed, cut in to small piece with different sterile scalpel blade and placed into sterile stomacher bags. And then the sample was diluted with 225 ml of sterile BPW and homogenized in a stomacher at 230 R for 2 minutes [14]. In case of environmental samples; pooled swab samples were placed into a test tubes that contained 9 ml sterile BPW. Subsequently, 10-fold serial dilutions were made to 10^{-6} for spread-plating. Samples were analyzed for the presence of *E. coli* O157:H7 and *Salmonella* spp. by employing standard guidelines. The *E. coli* O157:H7 detection was carried out according to the protocol of ISO 16654, 2001 standard. For isolation and identification *Escherichia coli* O157:H7 does not ferment sorbitol and, therefore, produces colorless colonies. In contrast, most other *E. coli* strains ferment sorbitol and form pink colonies and Latex *E. coli* O157:H7 agglutination test was performed to determine strains. The procedures for isolation of *Salmonella* from food were based on protocol of the ISO 6579, 2002 standard. *Salmonella* was isolated from beef sample (25 g) homogenized in 225 mL of 0.1% buffered peptone water (BPW) (HiMedia, India). Aliquot (1 ml) was added to 10 mL of Rappaport Vassiliadis (Oxoid, England) [15]. Following this procedure, eventually two even more colonies from pure isolate were inoculated on urea broth (SRL, India) and incubated at 37°C for 24 hours. All test tubes that were urease negative were treated as suspects of *Salmonella*. In addition isolate that was Gram-negative rod, methyl red and citrate positives but oxidase indole and voges-proskauer negatives which are non-lactose and sucrose fermenting were accepted putatively as *Salmonella* [16,17].

The aerobic plate count (APC) were enumerated using plate count agar (APC); twenty five grams of beef sample was weighed and homogenized in 225 mL of 0.1% sterile peptone water using a sterile homogenizer as described in [15]. In this study fecal coliforms were also employed using violet red bile agar (VRBL); 25 gram of beef sample was weighed and homogenized in 225 mL of 0.1% sterile peptone water using a sterile homogenizer. From the 10-fold dilutions of the homogenates, 0.1 mL of 10^{-4} , 10^{-5} and 10^{-6} dilutions of the homogenates were spread on to agar plates. Finally, on the VRBL medium, pale colonies with greenish zones reflect lactose fermentation by fecal coliforms, which appear slowly [18].

Statistical analysis

The data collected through questionnaire survey and laboratory results of the collected samples were entered into databases using Micro-Soft Excel computer program and analyze using SPSS version-19.0. Descriptive statistics were used to describe the nature and the characteristics of the questionnaire survey result. The aerobic bacterial and fecal coliform counts were expressed as mean using excel and compared by ANOVA. A Chi-Square test was applied to examine whether the differences between the values and the level of contamination between slaughterhouse and retail shops and associated risk factor were significant. A p-value of less or equal to 0.05 and chi-square value were considering indicative of a statistically significant difference.

Results

Isolation of bacteria from beef slaughtered and marketed at Dire Dawa city and Haramaya university

Out of 90 beef samples from HU (45 beef samples from

slaughterhouse and 45 beef samples from retail shop) examined bacteriologically 21(23.3%), 1(1.1%) and 6(6.7%) had *E. coli*, *E. coli* O157:H7 and *Salmonella* spp. respectively. None of the 90 beef samples from HU had mixed bacterial contamination.

Out of 200 beef samples from Dire Dawa (100 beef samples from slaughterhouse and 100 from ten randomly selected retail shops) examined bacteriologically, 15(7.5%), 5(2.5%) and 2(1%) had *E. coli*, *E. coli* O157:H7 and *Salmonella* spp., respectively. One of the 200 samples of beef had yielded both groups of bacteria. The prevalence of *E. coli*, *E. coli* O157:H7 and *Salmonella* in Dire dawa slaughterhouse and retail shop were presented in Table 2.

Hygienic quality of beef from Dire Dawa and HU slaughterhouses and retail shops

The results of total Aerobic bacteria (AB) in beef by using detection methods are summarized in Table 3. The total beef AB was 27/290 (9.31%) from slaughterhouse and retail shops. In this study, fecal coliforms (FC) were detected and enumerated irrespective of pathogenicity of the strain to estimate the level of hygiene. Out of 290 samples, FC were present in 11(3.79%) of which 8 (17.77%) and 2 (2%) were found in HU and Dire dawa slaughterhouses, respectively (Table 3).

Results of mean APCs of beef in this study are presented in Table 4. Fecal coliforms count (FCC) in beef indicates the hygienic qualities of meat.

Major source of microbial contamination for beef from slaughterhouse and retail shops

It is generally accepted that microbial loads on surfaces and equipment vary in different food plants depending on the microbial quality of the food [19].

Average microbial load for APCs and FCCs in beef contact surfaces at slaughterhouse and retail shops are shown in Table 5. In current study Total Aerobic Bacteria in different sample groups in retail shop (knives and hooks, cutting boards and personnel hands), were examined. Out of 30 environmental pooled samples (8, 8, 8 and 6 were from HU slaughterhouse, HU retail shop, Dire dawa slaughterhouse and Dire dawa retail shops respectively; out of 10 randomly selected retail shops due to the fact that 7 of the selected retail shops could not be voluntary to take swab sample), *E. coli*, *E. coli* O157:H7 and *Salmonella* was present in 7(23.33%), 2(6.66%) and 2(6.66%) samples. The occurrence of *E. coli*, *E. coli* O157:H7 and *Salmonella* spp. in beef

contact surfaces from slaughterhouses and retail shops are summarized in Table 6.

Fecal coliforms counts in different sample groups in retail shops and slaughterhouse (knives and hooks, cutting boards, personnel hands and transporting vehicle), were examined. The result varied from 3.11 log₁₀ cfu/cm² - 7.20 log₁₀ cfu/cm² in knives and hooks, cutting boards, balance, personnel hand and transport vehicle at slaughterhouse and retail shops. The overall mean of coliforms count in retail shops environment was 5.40 log₁₀ cfu/cm² and 2.38 log₁₀ cfu/cm² in slaughterhouse. Furthermore, the result of aerobic plate counts and coliform count were compared by ANOVA showed that there is significant (P ≤ 0.01) variation in the means of fecal coliforms count found in different meat contact surfaces in retail shop and slaughterhouse.

Hygienic practices in Dire Dawa and hu slaughterhouses and retail shops

According to Abattoir, Cutting and Packing Plant Standard [20], abattoir wall, floors, ceilings, windows, doors, lighting, air-conditioning/ventilation, services and equipment must be constructed to withstand and facilitate thorough cleaning and minimize contamination of product, either through pests, harboring of dirt or other physical, chemical or microbiological hazards.

In Dire dawa slaughterhouse except in Muslim slaughter premises, it is well organized beef slaughterhouse than HU slaughterhouse, In Dire dawa slaughterhouse for Christian have clear division of slaughtering process into stunning, bleeding, skinning and evisceration, whereas in Muslim slaughter premises and HU slaughterhouse no clear division existed. In both slaughterhouses, horizontal bleeding on killing floor was conducted, however, only vertical dressing process on overhead rail procedure was conducted in Dire dawa slaughterhouse.

The visual observation result in HU slaughterhouses indicated that the animal brought to slaughterhouse without prior ante-mortem inspection was done and without fasting the animal for 12 to 24 hours before slaughter which increases the micro floral load and sometimes the animal brought to slaughterhouse immediately after arrival from market results in shading of microorganisms. But in Dire dawa slaughterhouse the pre-slaughter procedure was done 12 hours before the slaughtering process presided. The animal also encountered stressful handling during riding on foot from the HU farm to HU slaughter house in the night sometimes they even suffered fracture and excitement. Beside these, stunning process was done by kicking using the back of axe and most of the time the workers couldn't made

No	Sample type	Sample collected area	Total sample
1	Raw beef meat	45 from HU slaughter house 45 from HU retail shop 100 from DD slaughter house 100 from DD retail shop	290
2	Environmental sample Equipment Workers hand Contact surface Balance Vehicle	2 from each of the four site 2 from each of the four site 2 from each HU and DD slaughter houses 2 from HU retail shop 2 from each HU and DD slaughter houses 2 from each HU and DD retail shops	30
3	Cutting board and table Respondents	22 respondents from HU 28 respondents from DD	50

HU=Haramaya University; DD=Dire Dawa

Table 1: Summary of the type and total amount of samples collected from the study areas.

Samples source	Number of Samples Processed	Bacterial Isolates (%)		
		<i>E. coli</i>	<i>E.coli</i> O157:H7	<i>Salmonella</i> spp.
HU slaughterhouse	45	16(35.6%)	1(2.2%)	3(6.7%)
HU retail shop	45	5(11.1%)	0(0%)	3(6.7%)
DD slaughterhouse	100	9(9%)	4(4%)	1(1%)
DD retail shops	100	6(6%)	1(1%)	1(1%)
Total	290	36(12.41%)	6(2.06%)	8(2.75%)

P ≤ 0.01, df=3 for *E. coli*, P>0.05, df=3 for *E. coli* O157:H7 and *Salmonella*

Table 2: Frequency of bacterial isolate of beef samples from Dire Dawa and HU slaughterhouse and retail shops.

Sample source	No of sample	Organisms detected	
		No (%) AB	No (%) FCs
HU slaughter house	45	6(13.33)	8(17.77)
HU retail shop	45	8(17.77)	1(2.2)
DD slaughter house	100	10(10)	2(2)
DD retail shops	100	3(3)	0(0)
Total	290	27(9.31)	11(3.79)

FCs=Fecal coliforms, AB=Aerobic bacteria; P>0.05, df=1

Table 3: Indicator organisms detected from beef sampled from HU and Dire Dawa slaughterhouse and retail shops.

Sample Source	No of Sample	Bacterial colonies log ₁₀ cfug ⁻¹					
		APCs			FCCs		
		Mean	Min	Max	Mean	Min	Max
HU slaughter house	45	7.11	4.00	8.80	7.50	3.60	9.20
HU retail shop	45	2.30	4.10	8.80	4.80	0.00	5.70
DD slaughter house	100	5.63	0.30	8.80	1.13	0.33	4.89
DD retail shop	100	3.10	6.72	9.73	0.00	0.00	0.00

FCCs=Fecal coliform counts, APCs=Aerobic plate counts; P ≤ 0.01, df=1

Table 4: Microbial loads of indicator organisms on beef in HU and Dire dawa slaughterhouse and retail shops.

Sources	No of Sample	Enumerated organisms log ₁₀ cfu/m ²					
		APCs			FCCs		
		Mean	Min	Max	Mean	Min	Max
HU slaughterhouse							
Equipments	2	3.05	TFC	6.10	TFC	TFC	TFC
Surfaces	2	TFC	TFC	TFC	TFC	TFC	TFC
Workers hands	2	TFC	TFC	TFC	TFC	TFC	TFC
Vehicle	2	TFC	TFC	TFC	TFC	TFC	TFC
HU Retail shops							
Equipments	2	TFC	TFC	TFC	5.38	5.10	5.67
Cutting boards	2	TFC	TFC	TFC	4.78	4.24	5.33
Workers hands	2	TFC	TFC	TFC	5.06	4.50	5.63
Balance	2	TFC	TFC	TFC	TFC	TFC	TFC
DD slaughterhouse							
Equipment	2	TFC	TFC	TFC	4.43	3.11	5.76
Surface	2	TFC	TFC	TFC	4.42	3.20	5.65
Worker hand	2	TFC	TFC	TFC	4.32	3.32	5.32
Vehicle	2	5.73	4.56	6.91	5.87	4.54	7.20
DD retail shops							
Equipment	2	TFC	TFC	TFC	4.96	4.32	5.61
Cutting board	2	TFC	TFC	TFC	6.26	5.65	6.88
Worker hand	2	TFC	TFC	TFC	5.98	4.87	7.1

TFC=Too Few to Count, P ≤ 0.01, df=1,

Table 5: Microbial loads of indicator organisms on beef contact surfaces from HU and Dire Dawa slaughterhouse and retail shops.

stunning by a single kick rather they kick several times which result the animal to suffering from pain. In general, the pre-slaughtering process in HU slaughterhouse brought the animal to stress which facilitate the rapid multiplication and shading of *E. coli* O157:H7 and *Salmonella* spp. this could be one of the source of contamination of meat. The

hygienic and sanitation practices employed at the study areas are summarized in Table 7.

Hands are rarely free from microorganisms. It is of the utmost importance that soap (preferably in a dispenser) and hot running water

are used for this purpose, thus aiming to reduce the microbiological load on hands [21,22] suggested that soap and hot water, at 45°C, should always be available at the washing-basins. [21] recommend that hand-washing alone has no effect on the reduction of bacteria on hands; it depends on the mechanical action, the duration and the type of soap and sanitizers being used.

It is important to know the educational background, type and terms of employment in the abattoir, and how the meat handler acquired their skills to establish their knowledge in handling meat safely. Accordingly **hygienic quality of beef** had significantly associated ($p=0.001$) with educational status of meat handler's, fecal coliform organisms in meat were more likely prevalent in slaughter house handled by poor knowledge and low level educational status food handler's. The knowledge and educational level of personnel working in both food establishments are summarized in Table 8.

In this study, personnel practices regarding prohibited habits and actions were also assessed. The visual observations indicated that, fraudulent activity and habits like eating, chewing and smoking in the slaughterhouse by the workers were common practices in both slaughterhouses especially prominent in Dire dawa slaughterhouse while they were on duty of meat processing. The overall result regarding habit, personnel cloth and cleanness in both slaughterhouses was summarized in Table 9.

Discussions

The occurrence of *E. coli* in meat samples from HU slaughterhouse in this study was in close agreement with the result of [12] who isolated *E. coli* in 30.97% of the meat samples studied in the same slaughterhouse. The present result is much lower than the finding of [23] who isolated *E. coli* in 91.4% of meat samples from abattoir in Mekelle.

Generally, the high prevalence of *E. coli* in the meat samples from HU slaughterhouse indicated the contamination of meat with intestinal content since evisceration take place in the same place. There was a significant difference in the prevalence of *E. coli* between HU slaughterhouse and Dire Dawa slaughterhouse ($P \leq 0.01$). This difference could be due to difference in hygienic condition and practice in both slaughterhouses.

The prevalence of *E. coli* O157:H7 isolated from beef in HU slaughterhouse (2.2%) and Dire dawa slaughterhouse (4%) in this study was in agreement with the reported prevalence of 2.60% [23] and 2.65% [12] in Ethiopia. There was no statistically significant difference in the prevalence of *E. coli* O157:H7 between HU slaughterhouse and Dire dawa slaughterhouse ($P>0.05$).

In comparison to the present study, a higher prevalence of *E. coli* O157:H7 were reported from different countries; 8% in Debre Zeit and Mojo [24] and 8.1% in Mojo, Ethiopia [25], 9% in India [26]. In the current study, lower prevalence of *E. coli* O157:H7 was also isolated from Dire Dawa retail shop (1%) which is in agreement with the report from America (0.8%) [27] and Kenya (0.2%) [28].

The frequency of isolation of *Salmonella* spp. in meat samples in this study was 6.7% from both HU slaughterhouse and retail shop. This result was in agreement with 5.6% prevalence reported from muscle in Addis Ababa, Debre Zeit, Dire dawa and Jigjiga [11], 8.5% from minced beef in Addis Ababa [29] and 4.8% from beef in Bahir Dar [30].

The detection of 6.7% of *Salmonella* in beef in HU slaughterhouse and retail shop as compared to Dire dawa slaughterhouse and retail shops (1%) suggests that the process of evisceration could be the main source of carcass contamination in addition to carrier state. Cross-contamination can also occur during the skinning process as a result of poor hygienic conditions. The other probable source of contamination is infected abattoir personnel. When comparing with the present study a relatively high prevalence of *Salmonella* (14.4%) was reported by [6] from minced beef in Addis Ababa. It was also lower than the 40% prevalence reported by [31]. Similarly, [32] reported *Salmonella* contamination rate of 42% from minced meat (locally known as «kitfo») samples collected from different hotels, bars and restaurants in Addis Ababa.

Presence of microbes in high numbers ($APC>5 \log \text{cfu/cm}^2$ or g^{-1}) fast tracks the spoilage of the meat. According to the international standard organization [33].

APC of 80% of analyzed samples must not exceed $5 \log \text{cfug}^{-1}$ or cm^2 , whereas 20% of the samples may have counts of up to $5 \log \text{cfug}^{-1}$ or cm^2 [34]. In this study 5.8% of samples had APCs more than 5.00

Sources		No of sample	Bacterial detected		
			No (%) <i>E. coli</i>	No (%) <i>E. coli</i> O157H7	No (%) <i>Salmonella</i>
HU slaughterhouse	Equipments	2	2(100)	1(50)	0(0)
	Surfaces	2	1(50)	0(0)	0(0)
	Workers hands	2	1(50)	0(0)	0(0)
	Vehicle	2	0(0)	0(0)	0(0)
HU Retail shops	Equipments	2	0(0)	0(0)	1(50)
	Cutting boards	2	1(50)	0(0)	0(0)
	Workers hands	2	0(0)	0(0)	0(0)
	Balance	2	0(0)	0(0)	0(0)
DD slaughterhouse	Equipment	2	1(50)	0(0)	1(50)
	Surface	2	0(0)	0(0)	0(0)
	Worker hand	2	0(0)	0(0)	0(0)
	Vehicle	2	1(50)	1(50)	0(0)
DD retail shops	Equipment	2	0(0)	0(0)	0(0)
	Cutting board	2	0(0)	0(0)	0(0)
	Worker hand	2	0(0)	0(0)	0(0)
Total		30	7(23.33)	2(6.66)	2(6.66)

$p>0.05$, $df=3$

Table 6: Bacterial species detected from beef contact surfaces sampled from HU and Dire Dawa slaughterhouse and retail shops.

Practices	Dire Dawa	HU
Cleaning and disinfection of knives and hooks		
Before the commencement of work	28 (100%)	21 (95.5%)
When excessively and visibly soiled		1 (4.5%)
Manner of cleaning and disinfection		
Using detergents and water	22 (78.6%)	21 (95.5%)
Rinsing with water only	6 (21.4%)	1 (4.5%)
Floor Surface cleaning and disinfection		
Before commencement of work	28 (100%)	20 (90.9%)
When excessively and visibly soiled		1 (4.55%)
After commencement of work		1 (4.55%)
Manner of cleaning and disinfection of surface		
Using detergents and water	10 (35.7%)	15 (68.2%)
Rinsing with water only	18 (64.3%)	7 (31.8%)
Hand washing before starting handling raw meat		
Yes	28(100%)	22(100%)
Manner of hand washing		
Using detergents and water	27(96.4%)	17(77.3%)
Rinsing with water only	1(3.6%)	5(22.7%)
Presence of sanitary regulatory system		
Yes	2 (7.1%)	0
No	26 (92.9)	22 (100%)

p ≤ 0.01, df=1

Table 7: Hygienic and sanitation practices employed at Dire dawa and HU slaughterhouses and retail shops.

Skills	D.D slaughterhouse frequency (%)	HU slaughterhouse frequency (%)
Educational status		
None	0	1 (4.55)
Elementary/junior	11 (39.3%)	13 (59.1%)
High school	12 (42.9%)	7 (31.8%)
College	4 (14.3%)	1 (4.55%)
Graduate	1 (3.6%)	
Sources of meat processing skills		
Observation	21 (75%)	19 (86.4%)
Parents	4 (14.3%)	3 (13.6%)
Formal training	3 (10.7%)	0

Table 8: Educational status of meat handler's.

Prohibited habits	Dire Dawa	HU
Jewelry		
Worn	19 (67.9%)	2 (9.1%)
Not worn	9 (32.1%)	20 (90.9%)
Finger nails		
Short and polished	22 (78.6%)	15 (68.2%)
Short/ not polished	6 (21.4%)	5 (22.7%)
Long and polished		2 (9.1%)
Smoking in meat processing plants		
Yes	13 (46.4%)	3 (13.6%)
No	15 (53.6%)	19 (86.4%)
Hair cover		
Used	24 (85.7%)	14 (63.6%)
Not covered	4 (14.3%)	8 (36.4%)
Gum boots		
Used	18 (64.3%)	22 (100%)
Not used	10 (35.7%)	0

p ≤ 0.01, df=1

Table 9: Practices of the meat handlers regarding prohibited habits and actions.

log₁₀ cfu⁻¹ the condition was unacceptable. Lower level of aerobic plate count in this study was much lower than previous studies [2,35-37]. However, the microbial contamination level of slaughterhouse and retail shops were higher as compared to reports from developed countries and our results do not conform to EU specifications [38,39].

The higher aerobic plate count enumerated from HU slaughterhouse (7.11 log₁₀ cfu⁻¹), suggests an unusual high level of contamination and/or growth which was similar with [40] report. Given the hygienic status of the slaughterhouse and meat processing observed in the slaughterhouse.

The result of this study was much lower than presence of fecal coliforms in meat and meat studied by many researchers [41,42]. Other study results have also been reported for retail chicken (50% incidence of *E. coli*) in Australia [43] which was much higher than the present study.

From the data of retail meats it was evident that the highest FCCs (6.26 log₁₀ cfu/cm²) levels were found in the cutting boards at Dire dawa retail shop. Cutting board from HU retail shop got the smallest values of FCCs (4.78 log₁₀ cfu/cm²) and in HU slaughterhouse was too few to count from knives and hooks, surface, vehicle and workers hand. Based on the data, the highest FCCs (5.87 log₁₀ cfu/cm²) and APCs (5.73 log₁₀ cfu/cm²) levels found in the transporting vehicle from Dire dawa slaughterhouse while the smallest values of FCCs (4.32 log₁₀ cfu/cm²) found in workers hand in Dire dawa slaughterhouse and APCs was found too few to count in both slaughterhouses (Table 5).

In both slaughterhouses personnel interviewed to assess the hygienic conditions in the slaughterhouse responded that there was adequate potable water supply in the slaughterhouse. However, there is no hot water supply in all meat processing facilities. In both slaughterhouses, there were no facilities for knife sterilization and no rooms for retention of conditionally approved carcasses. Regarding latrine facility, both slaughterhouses had communal latrine which was properly placed but with poor management. There were no enough water supplies as a result, flies infestation of the facilities were observed. Hand washing is an essential component of infection control [44]. In general, both abattoirs have no mechanism of ensuring sanitation standards, proper waste disposal mechanism and vermin's and scavenger's protection mechanisms. Therefore, there are opportunities of contamination of slaughter facilities which in turn contaminate the exposed tissues of the carcass with microorganisms.

The behavior of worker and hygienic practices of retail shop in HU was relatively good as compared to Dire dawa retail shop and meat handlers do not have close contact with money and they do only cutting and weighing the meat. To get rid of germs and dirt, it is important to wash hands properly and frequently with detergents and warm water. Hands that have long nails are more difficult to clean thoroughly and can collect small pieces of debris and bacteria that do not wash off easily [45].

The clean gum boots are just as important as clean overalls, because they may also be a source of contamination. Gum boots should therefore be washed at the facility provided (washing-basins supplied with hot and cold water, liquid soap and a brush) before entering the processing room [22]. The purpose of hairnets and beard nets is twofold: to prevent loose hairs and dandruff from falling into the food and also to discourage the workers from running their fingers through their hair or scratching their scalps [46,47].

Conclusion

In conclusion the results obtained from this study showed contamination sources of beef are more likely to be associated with insufficient hygienic practices and improper handling of meat in the slaughterhouse and retail shops. Floor surface, cutting boards, hooks and knives, workers hands and transporting vehicle in slaughterhouses as well as, in retail shops are potential sources of beef contamination. The study indicated that slaughterhouses and retail shops in HU and Dire Dawa could be the source of contamination of beef. HU and Dire dawa slaughterhouses and retail shops are not well structured and the working habits in the slaughterhouse are not good enough to satisfy an acceptable hygienic standard practices for slaughtering and processing of beef for human consumption. The study suggested that beef could be a significant source of foodborne pathogens for people in the study areas. Based on the findings of the present study the following recommendations are forwarded in order to guarantee the microbial quality of beef and minimize the risk of *E. coli* O157:H7 and Salmonellosis outbreak in Dire dawa and HU and its surrounding areas.

Competing Interests

There is no conflict of interest regarding the publication of this paper.

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