academicJournals

Vol. 9(8), pp. 213-227, August 2017
DOI: 10.5897/JVMAH2015.0381
Article Number: BDF731E65487
ISSN 2141-2529

http://www.academicjournals.org/JVMAH

Journal of Veterinary Medicine and Animal Health

Full Length Research Paper

Microbial safety and its public health concern of *E. coli* O157:H7 and *Salmonella* spp. in beef at Dire Dawa administrative city and Haramaya University, Ethiopia

Abayneh Edget*, Daniel Shiferaw and Shimelis Mengistu

College of Veterinary Medicine, Haramaya University, Haramaya, Dire Dawa, Ethiopia.

Received 14 March 2015; Accepted 31 August 2015

A cross-sectional study was conducted in Dire Dawa administrative city and Haramaya University (HU) slaughterhouses and retail shops, with the aim to identify *E.coli* O157:H7and *Salmonella,* to assess the microbial safety of beef and identify potential contamination risk factors. A total of 320 samples consisting of beef samples and environmental pooled samples examined for the presence of *E. coli*, *E. coli* O157:H7and *Salmonella* following standard bacteriological techniques and procedures outlined by the International Organization for Standardization. From a total of 290 beef samples collected, *E. coli* was isolated from 36 (12.41%) and out of these, 6 (2.06%) were confirmed on Sorbitol MacConkey Agar to be *E. coli* O157 H7. 8(2.75%) *Salmonella* spp. was identified by means of culture and biochemical test. The difference in prevalence was statistically significant (P≤ 0.01) between slaughterhouses and retail shops in both study areas. There was significant difference in mean Aerobic Plate Counts between Haramaya University slaughterhouse (7.11 log10 cfug^{-1}) and retail shop (2.3 log10 cfug^{-1}). Fecal coliforms counts (FCC) were significantly higher for beef samples from Haramaya University slaughterhouse (7.50 log10 cfug^{-1}) as compared to 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* was present in 7(23.33%), 2(6.66%) and 2(6.66%), respectively in both study areas. A significant difference (P≤ 0.01) in the prevalence of *E. coli* between Haramaya University slaughterhouse (35.6%) and Haramaya University retail shop (11.1%) and Dire Dawa slaughterhouse (9%). Visual observations of slaughterhouse design, layout, slaughtering process, hygienic practice employed, sanitary regulatory system and personnel habit were below the minimum standards. Slaughterhouse and all meat contact surfaces might have served as sources of contamination for the product. Therefore, good management practice and good hygienic practice should be introduced in order to enhance the overall safety and hygienic quality of beef and safeguard the consumer from foodborne pathogens.

Key words: Aerobic plate counts (APC), beef, Dire Dawa, *E. coli, E. coli* O157:H7, fecal coliforms counts (FCC), Haramaya University (HU), *Salmonella.*

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 on countries (Fratamico et al., 2005). Centers for Disease Control and Prevention (CDC) reported that of 19,056 people who get sick, more than 4,200 are hospitalized and 80 deaths recorded States of America (USA) (CDC, 2013). It mostly affects developing countries, due to major contributing factors such as from foodborne illness among 48 million (15%) population in United 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 (Bhandare et al., 2007; Podpecan et al., 2007; Chhabra and Singla, 2009). In developing countries, including Ethiopia, up to 2 million people die per year due to disease of foodborne pathogens (World Health Organization (WHO), 2007).

Over the last 20 years, the emergence of major foodborne pathogens such as *Salmonella* and *Escherichia 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 (Diane et al., 2010). For this reason, the basic steps in the control of safety and quality of food include analysis of food products for presence of pathogenic microorganisms that cause the majority of alimentary human diseases. Among them are, *Salmonella* and *E. coli* O157:H7. These foodborne pathogens have frequently been linked to a number of cases of human illness (Brown et al., 2000).

Trends in foodborne illness in the industrialized and developing countries indicate that the incidence of foodborne illness is increasing (WHO, 2005). It has resulted in significant social and economic impact and that it is likely to remain a threat to public health well into the next century. There are however, substantial gaps in our understanding of this problem. In 2005, the World Health Organization (WHO) reported that 1.8 million people died from diarrheal diseases, largely attributable to contaminated food and drinking water (WHO, 2005). This is not just only an underdeveloped world problem. Meat processing at retail level is likely to contribute to the higher levels of contamination in minced beef as compared to carcasses (Tegegne and Ashenafi, 1998). 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 (Ejeta et al., 2004). Previous studies conducted in many parts of the country indicated the occurrence of pathogens including *Salmonella* in different food animals, meat and meat products (Haimanot et al., 2010). In addition, outbreaks of infections related with poor hygiene and consumption of contaminated food were reported in Ethiopia (Mache et al., 1997) and some were caused by *Salmonella* and *E. coli* (Alemseged et al., 2009).

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 (Haymanot et al., 2010). 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 in beef, to identify potential sources of contamination of beef in slaughterhouse and retail meat shops, to determine the hygiene conditions and practices of slaughterhouse and retail meat shops and to determine the hygienic quality of beef from slaughterhouse and retail meat shops.

MATERIALS AND METHODS

Study area and population

The study was conducted at slaughterhouse and ten retails shop in Dire Dawa administrative city and slaughterhouse and one retail meat shop in Haramaya University from May to November, 2014. Dire Dawa lies in the eastern part of the Ethiopia 515 km away from Addis Ababa with latitude 9° 27′ to 9° 49′ North and longitude 41° 38′ East (Center of Stastical Agency (CSA), 2007). The city has a total area of 1,213 km^2 coverage's and elevation of 226 to 950 m above sea level (Center of Stastical Agency, 2007), and Haramaya University (HU) is located in the Eastern Hararghe Zone of the Oromia Region of Ethiopia, which is about 17 km from the city of Harar and 40 km from Dire Dawa and 5 km from Haramaya town at an altitude of 1980 m above sea level between latitude 9° 26" N and longitude 42° 3" E (Asrat, 2008). The mean annual rainfall is 870 mm with a range of 560 to 1260 mm, and the mean maximum and minimum temperatures are 23.4 and 8.25°C, respectively (Asrat, 2008). The study population represents apparently healthy cattle slaughtered in Dire Dawa and HU slaughterhouse, cattle subjected to slaughter brought from Water, Kersa, Hirna, Chalanko and Kulibi in both study areas and in addition from Issa (Somali) in Dire Dawa. Both local and cross breeds cattle are reared in and around the study areas for meat production mostly. There is one municipal

*Corresponding author. E-mail: edgetabayneh@gmail.com.

slaughterhouse in which different species of animal such as cattle, goat, sheep and camel is slaughtered and both over head rail and horizontal slaughtering system is practiced. Over an average of 70 cattle, 20 camel and 55 goat and sheep are slaughtered daily in Dire Dawa slaughterhouse but one slaughterhouse and one beef retail shop is available inside HU and only horizontal slaughtering system is practiced. Over 17 workers in HU slaughterhouse and 5 workers in HU retail shop are working daily on temporary basis and in range of 5 to 20 cattle are slaughtered every day depending on the needs of student cafeteria (main campus and Harer), staff lounge and the days of the week in HU slaughterhouse. The sample included raw beef and environmental pooled samples from slaughterhouses and retail shops in both study areas. The slaughterhouse structure in Dire Dawa was relatively good like there is a clear division of work, vertical line system and separate room for evisceration as compared with HU slaughterhouse.

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 interviews 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 retails shops in both study areas. The sample size required for this study to identify the microbial safety of foodborne pathogen from beef was determined according to Thrusfield (2007) by taking expected prevalence of 5.6% for *Salmonella* in Dire Dawa (Bayleyegn et al., 2003) and 3% for *E. coli* O157:H7 in Haramaya University (Taye et al., 2013) with the consideration of slaughter animal coming from the same origin in both study area and confidence interval of 95% and 5% absolute precision.

The sample size of the present study were calculated and 81 beef samples from Dire Dawa slaughterhouse and 81 from 10 randomly selected retails shop and the selection of retail shop were done based on lottery method from 542 retails shop in Dire Dawa, and to increase the precision 19 beef samples for each sample collection centers were added and a total of 200 beef samples were collected from Dire Dawa administrative city and exactly the calculated number of 45 beef samples from HU slaughterhouse and 45 beef samples from retail shop were also collected due to resource limitation. Beside that, to assess the source of contamination level, only 30 environmental pooled samples were taken from equipment, surface, workers hand, vehicles in both study areas due to the vastness of the work and the availability of resource. Therefore, a total of 290 beef samples and 30 environmental pooled samples were collected from both study areas. The microbial safety and hygiene quality were then assayed by using the methods recommended by International Commission on Microbiological Specifications for Foods (ICMSF) (1986). All the samples were investigated with respect to *Salmonella*, *E. coli* and *E. coli* O157:H7 detection and aerobic plate and fecal coliforms counts.

The slaughterhouse and each retail shops were visited once in a week for consecutive weeks, and in each visit, ten beef samples were taken from the slaughterhouse and ten beef samples from ten retails shops in Dire Dawa town and five beef samples from HU slaughterhouse and five beef samples from HU retail shop every week for nine consecutive weeks. Each carcass is represented by meat pieces collected from different locations such as leg, flank, inter costal and neck, and pooled together weighing 200 g. Retail meat samples (200 g) were taken simply from different location under aseptic conditions using sterile blades and sterile containers as described by Gill (2007).

For convenience, before the commencement of the sample collection all the respective samples (meat and environmental swab) were labeled with necessary information including date of sampling, code of sample source (beef) and identification of the shop from which the samples were obtained. The live animals were coded with owners name and the same code were followed for the carcass then the meat and samples were taken from the same carcass from those owner retails shops in Dire Dawa and the same procedure was followed in HU. After completion of sampling, all collected samples from Dire Dawa were placed in nutrient broth or Carry Blair Transport medium (Oxoid Ltd, Basingstoke, Hampshire, England) and immediately transported in cold chain using ice box containing ice pack to Veterinary Microbiology Laboratory of Haramaya University, within an hour and samples from Haramaya University were processed immediately upon arrival. The samples were processed up on arrival or stored overnight in a refrigerator at +4°C and the samples were processed in the next day for identification of pathogenic species, according to the standard set by the International Commission on Microbiological Specification for Food (ICMSF, 1986).

A total of thirty pooled environmental samples were collected from slaughterhouse, retail shop and transport vehicles. The pooled environmental sample collections were conducted two times within three months. On each visit to the slaughter house, a total of four pooled swab samples were taken each from cleaned, disinfected and dry surfaces, others from hooks, knives and aprons, the third from personnel's hands who work flaying, evisceration and carcass cutting before the beginning of the work and the fourth from the surface of transporting vehicles by rubbing thoroughly with a moistened swab. In each visit of each retail shops, a total of three pooled swab samples were taken each from cutting boards and meat grinder, others from hooks, knives and protective cloths and the third from personnel's hands (butcher man) before the beginning of 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 Veterinary Microbiology Laboratory of Haramaya University in an ice box on ice packs and analyzed upon arrival or within 24 h of sampling. The type and the number of samples processed were presented in Table 1.

For isolation and identification of pathogens from meat, 25 g of sample was weighed, cut into small piece with different sterile scalpel blade and placed into sterile stomacher bags, diluted with 225 ml of sterile BPW and homogenized in a stomacher at 230 R for 2 min (ISO TS 11133-1, 2009). For isolation and identification of pathogens from environmental samples, pooled swab samples were placed into a test tube 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. according to published methods as follows:

The *E. coli* O157:H7 detection was carried out according to the protocol of ISO 16654 (2001) standard. The pre-enriched beef samples were subsequently subcultured onto MacConkey agar (Oxoid, England) for primary screening of *E. coli* and incubated at 37°C aerobically for 24 h. Suspected colonies of *E. coli* (pinkish color appearance) were then subcultured onto nutrient agar (HiMedia, India) (non-selective media) and confirmed by triple sugar iron (TSI) (Oxoid, England) and indole, methyl red, Voges-Proskauer and citrate (IMViC) tests on tryptone broth (Oxoid, England), MRVP medium (Oxoid, England) and Simon citrate agar

Table 1. Summary of the type and total amount of sample collected

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	2 from each of the four site	30
	Equipment	2 from each of the four site	
	Workers hand	2 from HU slaughter house and 2 from DD slaughter house	
	Contact surface balance	2 from HU retail shop	
	Vehicle	2 from HU and 2 from DD slaughter house	
	Cutting board and table	2 from HU and 2 from DD retail	
3	Respondents	22 respondents from HU 28 respondents from DD	50

HU= Haramaya University; DD= Dire Dawa

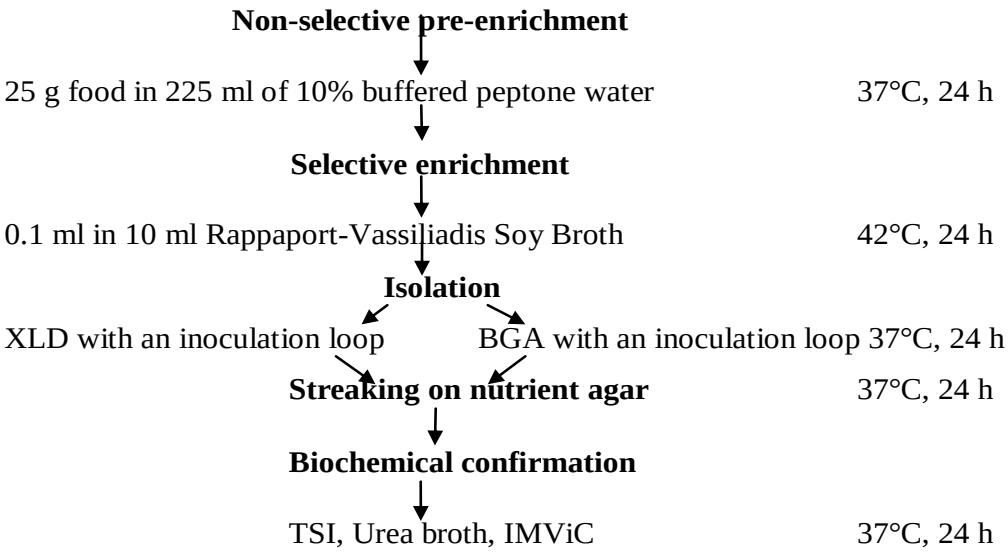

Figure 1. Flow diagram for detection of *Salmonella* (Source, ISO 6579, 2002).

(Oxoid, England), respectively. Then the bacterium that was confirmed as *E. coli* was subcultured onto Sorbitol MacConkey agar (SMA) (Oxoid, England) from nutrient agar (HiMedia, India). SMA (Oxoid, England) plates were incubated at 35°C for 20 to 22 h (Timothy and Smith, 2012). *E. 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 (Figure 1). To diminish the risk of obtaining false negative results, a non-selective pre-enrichment of large food sample, followed by two selective enrichments, but due to availability of the resource, used one selective enrichment media which is mandatory and plating on two selective media was performed.

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). This was incubated at 41 ± 0.5°C over night. After gentle mixing, a loopful of culture from the enrichment broth was streaked parallely onto Xylose lysine desocholate (XLD) agar (Oxoid,

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

Samples source	Number of samples processed	Bacterial isolates (%)		
		E. coli	*E. coli* O157:H7	*Salmonella* 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*

England) and Brilliant green agar (BGA) (Oxoid, England) and incubated at 37°C for 24 to 48 h. Typical *Salmonella* colonies which are pink with or without black centers were isolated from XLD and *Salmonella* colonies grow as red-pink, white opaque colonies surrounded by brilliant red zones in the agar are taken from BGA. The colonies were purified on fresh nutrient agar (HiMedia, India) and then streaked and stabbed into the butt of triple sugar iron (TSI) (Oxoid, England) slants and inoculated into tryptone broth (Oxoid, England), MRVP medium (Oxoid, England) and Simon citrate agar (Oxoid, England) for IMViC test from both selective media. These were incubated at 37°C for 24 h. The test tubes that had alkaline (red) slants and acidic (yellow) butts, with the production of H_2S (blackening) were presumed to be *Salmonella* isolates. Moreover, two or more colonies from pure isolates were inoculated on urea broth (SRL, India) and incubated at 37°C for 24 h. All test tubes that were urease negative were treated as suspects of *Salmonella* (FDA, 1992). In addition, isolate that was Gram-negative rod, oxidase negative, methyl red positive, citrate positive, indole negative, Voges-Proskauer negative and lactose and sucrose non-fermenter were accepted putatively as *Salmonella* (Fawole and Oso, 2001).

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. From the 10-fold dilutions of the homogenates, 0.1 ml of 10^{-4}, 10^{-5} and 10^{-6} dilutions of the homogenates were plated by the spread plate method onto the surface of plate count agar (PCA). Plates were incubated at 35°C for 48 h and plates containing between 30 and 300 colonies were counted (ISO/TS 11133-1, 2009). Fecal coliforms were enumerated using violet red bile agar (VRBL); 25 g 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 onto agar plates. The VRBL was inoculated with 0.1 ml inoculums after the final incubation at 44 ± 1°C for 24 h, typical and atypical colonies were enumerated. Typical colonies on VRBL medium have fuchsia color with a diameter of approximately 0.5 mm and sometimes surrounded by a reddish-fuchsia zone (1 to 2 mm in diameter) of precipitated bile salts, which reveals lactose degradation in acid. On the VRBL medium, pale colonies with greenish zones reflect lactose fermentation by fecal coliforms, which appear slowly (Marshall, 1993).

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 analyzed using SPSS version-19.0 (SPSS, 2012). 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 analysis of variance (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 considered 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. The prevalence of *E. coli*, *E. coli* O157:H7 and *Salmonella* in HU slaughterhouse and retail shop were presented in Table 2. 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 in beef by using detection methods are summarized in Table 3. This study detected total aerobic bacteria in 27/290 (9.31%) of beef

Table 3. Indicator organisms detected from beef sampled from HU and Dire Dawa 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 4. Microbial loads of indicator organisms on beef in HU and Dire Dawa slaughterhouse and retail shops.

Sample source	No of sample	Bacterial colonies log10 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.

samples from slaughter house and retail shops. Results of mean APCs of beef in this study are presented in Table 4. Fecal coliforms count in beef presented in Table 4 indicates the hygienic qualities of meat. In this study, fecal coliforms were detected and enumerated irrespective of pathogencity of the strain to estimate the level of hygiene (Table 3). Out of 290 samples, fecal coliforms were present in 11 (3.79%) samples including HU and Dire Dawa slaughterhouse 8 (17.77%) and 2 (2%), respectively and HU retail shop 1 (2.2%) and Dire Dawa retail shops 0 (0%) (Table 3).

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 (Evans et al., 2004). One of the specific objectives of this study was to examine the sources of microbial contamination of beef intended for human consumption and to determine the acceptability of hygienic levels of slaughterhouse and retail shops. Swab samples were taken from cleaned, disinfected and dry surfaces of slaughterhouse, retail shops facilities and equipment surfaces, and also personnel before the beginning of work by rubbing meat contact surfaces and the hand of meat handler thoroughly with a moistened swab from both sites. All contact surfaces were analyzed for *E. coli* O157:H7 and *Salmonella* spp*.,* detection and enumeration of fecal coliform and aerobic bacteria. Out of 30 environmental pooled samples (8 from HU slaughter house, 8 from HU retail shop, 8 from Dire Dawa slaughterhouse and 6 from Dire Dawa retail shops 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 HU and Dire Dawa slaughterhouse and retail shops are summarized in Table 6. Average microbial load for APCs and FCCs in beef contact surfaces at slaughterhouse and retail shops are shown in Table 5. Total aerobic bacteria in different sample groups in retail shop (knives and hooks, cutting boards and personnel hands) were examined.

Fecal coliforms counts in different sample groups in retail shops and slaughterhouse (knives and hooks, cutting boards, personnel hands and transporting vehicle)

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

Sources	No of sample	Enumerated organisms log10 cfu /cm^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.

were examined. The result varied from 3.11 $\log_{10}$ cfu/cm^2 to 7.20 $\log_{10}$ cfu/cm^2 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 $\log_{10}$ cfu/cm^2 and 2.38 $\log_{10}$ cfu/cm^2 in slaughter house. Furthermore, the result of aerobic plate counts and coliform count were compared by ANOVA and 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

"Abattoir" in terms of the Republic of South African Meat Safety Act, 2000 (Act 40 of 2000) means a slaughter facility in respect of which a registration certificate has been issued in terms of section 8 (1) and in respect of which a grading has been determined in terms of section 8 (2): (i) A well-designed and constructed structure is needed to systematically process the animal that is slaughtered. According to abattoir, cutting and packing plant standard (ABM, 2008), 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 a well organized beef slaughter house than Haramaya University slaughter house. 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

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

Sources	No of sample	Bacterial detected		
		No (%) *E. coli*	No (%) *E.coli* O157H7	No (%) *Salmonella*
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

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 h 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 h 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 could not make 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 sources of contamination of meat.

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 (Desmarchelier et al., 1999). Van Zyl (1995) suggested that soap and hot water, at 45°C, should always be available at the washing-basins. Desmarchelier et al. (1999) 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 was 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. 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.

DISCUSSION

Food borne illnesses caused by *Salmonella* spp., and *E. coli* O157:H7 represents a major public health problem worldwide. These pathogens are transmitted mainly through consumption of contaminated food and the presence of these organisms in meat animals and in raw meat products has relevant public health implications (Sousa, 2008). The occurrence of *E. coli* in meat samples from HU slaughterhouse in this study was in close agreement with the result of Taye et al. (2013) 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 Mekonnen et al. (2012) 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% (Mekonnen et al., 2012) and 2.65% (Taye et al., 2013) 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 (Hiko et al., 2008) and 8.1% in Mojo, Ethiopia (Mersha et al., 2009), 9% in India (Luga et al., 2007). 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%) (Desenclos et al., 1988) and Kenya (0.2%) (Chapman et al., 2000). The frequency of isolation of *Salmonella* spp. in meat samples in this study was 6.7% from both HU slaughter house and retail shop. This result was in agreement with 5.6% prevalence reported from muscle in Addis Ababa, Debre Zeit, Dire Dawa and Jigjiga (Bayleyegn et al., 2003), 8.5% from minced beef in Addis Ababa (Zewdu and Cornelius, 2009) and 4.8% from beef in Bahir Dar (Sefinew and Bayleyegn, 2012).

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 Ejeta et al. (2004) from minced beef in Addis Ababa. It was also lower than the 40% prevalence reported by Molla et al. (2000). Similarly, Tegegne and Ashenafi (1998) reported *Salmonella* contamination rate of 42% from minced meat (locally known as «kitfo») samples collected from different hotels, bars and restaurants in Addis Ababa.

The lower prevalence was also revealed in this study from Dire Dawa slaughterhouse (1%) and Dire Dawa retail shops (1%). This result was in agreement with prevalence report of Sibhat et al. (2011) who reported 2% from carcass in Debre Zeit, Ethiopia and Fegan et al. (2004) also reported carcass contamination of 2% from slaughterhouse in Australia. There was no statistically significant difference in the prevalence of *Salmonella* spp. between Haramaya University and Dire Dawa administrative city ($P > 0.05$). This could be due to unhygienic slaughtering practice in HU slaughterhouse and Dire Dawa halal slaughter house.

Presence of microbes in high numbers (APC > 5 log cfu/cm^2 or g^{-1}) fast tracks the spoilage of the meat. According to the international standard organization (ISO 4833, 2003), APC of 80% of analyzed samples must not exceed 5 log $cfug^{-1}$or cm^2, whereas 20% of the samples may have counts of up to 5 log $cfug^{-1}$ or cm^2 (Mukhopadhyay et al., 2009). In this study 5.8% of samples had APCs more than 5.00 $\log_{10}$ $cfug^{-1}$, the condition was unacceptable. Lower level of aerobic plate count in this study was much lower than previous studies (Alvarez-Astorga et al., 2002; Bhandare et al., 2007; Haque et al., 2008; Hassan et al., 2010). 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 (Gill et al., 2000; Duffy et al., 2001).

The higher aerobic plate count enumerated from HU

slaughterhouse (7.11 $\log_{10}$ $cfug^{-1}$) suggests an unusual high level of contamination and/or growth which was similar with Gill (2007) report, given the hygienic status of the slaughterhouse and meat processing observed in the slaughterhouse. The result of this study was much lower than the presence of fecal coliforms in meat and meat studied by many researchers (Doyle, 2007; Adu-Gyamfi et al., 2012). Other study results have also been reported for retail chicken (50% incidence of *E. coli*) in Australia (Pointon et al., 2008) which was much higher than the present study. Mean fecal coliforms counts were higher for beef samples from HU slaughterhouse (7.50 $\log_{10}$ $cfug^{-1}$) as compared to carcass sample from HU retail shop (4.80 $\log_{10}$ $cfug^{-1}$) and also higher for beef samples from Dire Dawa slaughterhouse (1.13 $\log_{10}$ $cfug^{-1}$) but there was no fecal coliforms in Dire Dawa retail shops. This difference was statistically significant between both slaughterhouses and retail shops ($p \leq 0.01$).

The prevalence of coliforms was much lower than that of any other microorganism studied. Of the 290 beef samples tested, 11 (3.79%) were positive for FCCs and the microorganism were detected at both processing stage. The concentration of fecal coliforms enumerated from beef (3.57 $\log_{10}$ $cfug^{-1}$) was higher than established limits (10 to 100 $cfug^{-1}$) in guidelines (Alvarez-Astorga et al., 2002) which is assumed to be an indicator of fecal contamination. The result showed that only one sample from HU retail shop had count of 6.10 $\log_{10}$ cfu/cm^2 in knives and hooks, while too few to count in cutting boards, balance and personnel hands and 6.91 $\log_{10}$ cfu/cm^2 in vehicle from Dire Dawa slaughterhouse. The overall mean of total aerobic bacteria count in retail shops environment was 3.05 $\log_{10}$ cfu/cm^2 and 5.73 $\log_{10}$ cfu/cm^2 in Dire Dawa slaughterhouse. Furthermore, the result showed that there was significant ($P \leq 0.01$) variation in the means of total aerobic bacteria found in knives and hooks and different meat contact surfaces in retail shops and slaughterhouse.

From the data of retail meats it was evident that the highest FCCs (6.26 $\log_{10}$ cfu/cm^2) 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 $\log_{10}$ cfu/cm^2) and in HU slaughterhouse was too few to count from knives and hooks, surface, vehicle and workers' hands (Table 5). Based on the data, the highest FCCs (5.87 $\log_{10}$ cfu/cm^2) and APCs (5.73 $\log_{10}$ cfu/cm^2) levels found in the transporting vehicle from Dire Dawa slaughterhouse while the smallest values of FCCs (4.32 $\log_{10}$ cfu/cm^2) found in workers hand in Dire Dawa slaughterhouse and APCs was found too few to count in both slaughterhouses (Table 5). The findings of this study indicated that meat contact surfaces might be real risks associated with the persistence of hazardous organisms. Similar findings were reported by Gill and McGinnis (2004) and Temelli et al. (2006). Based on European commission standards used in the food processing industry; a standard of less than 1.3 $\log_{10}$ cfu was used for aerobic plate count, less than 1.0 $\log_{10}$ cfu for Enterobacteriaceae count. According to this standard, the results of average mean of APCs and FCCs in our study for food contact surfaces were 4.39 and 5.29 $\log_{10}$ cfu, respectively, which was unacceptable (Sneed et al., 2004).

In the present study, it was found that all of the meat establishments had pathogenic and indicator bacteria. The findings showed the magnitude of contamination at slaughterhouses and retail shops was high. This may contribute to the incidence of food associated illnesses. In this study, the identification of thermo-tolerant *E. coli* showed the presence of recent fecal contamination (Collee and Mackie, 1999). Hence, basic failures occur in the sanitization procedures applied to these utensils, since the establishments were found not to apply the cleaning process on a daily basis. The knives used for filleting and cutting were not sanitized at any of the retail houses visited. Neither the slaughterhouse, nor any of the platforms (bleeding, evisceration and inspection line) adopted the practice of immersing knives in hot water.

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 (Larson et al., 2003). 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 adequacy of a cleaning program is judged on the basis of the adherence to specified standard operating procedures during the cleaning and disinfection process and the inspection of cleaned facilities and equipment (Gill et al., 1999). Gill et al. (1999) further reports that improperly cleaned equipment have been implicated in outbreaks of foodborne diseases and it is therefore apparent that cleaning and disinfecting processes should fully comply with regulations. Gill and McGinnis (2000) reported that a primary source of *E. coli* deposited on meat during processing appears to be the detritus in equipment which was not removed during daily cleaning.

Assessment on the procedures and frequency of cleaning and disinfection of the equipment in both slaughterhouses are important and the result indicated that, the procedures of cleaning and disinfection of the surface, a notably low percentage (35.7%) in Dire Dawa slaughterhouse and high percentage in HU slaughterhouse (68.2%) of respondents indicated that running water and detergent was used to clean the surfaces. whereas majority of them cleaned their knives whenever they were excessively and visibly soiled with fat or blood before the commencement of work each day. About seventy eight percent of the respondents in Dire Dawa slaughterhouse and ninety five percent in HU slaughterhouse practiced washing their knife with soap and water. The respondents were also questioned on the frequency of cleaning and disinfection of the working surfaces. All (100%) respondents in Dire Dawa and 90.9% in HU slaughterhouse reported that the surfaces were cleaned before the commencement of work each day.

Upon visual observation, the knives used for filleting and cutting were not sanitized at any of the meat retail establishments visited in both study areas. In the slaughterhouse, any of the platforms (bleeding, evisceration and inspection line) did not adopt the practice of immersing knives in hot water. As for the meat hooks for hanging carcasses, most of them were splashing with water and some were not cleaned prior to use and most importantly the floor and surface of transporting vehicles were regularly cleaned with detergent and water but re-contaminated with workers gum boot and in contact with the meat during loading. In both slaughterhouses interview showed that washing of the hand before starting slaughter is not common but after the end of processing hand washing were conducted without the use of hot water and soap. In addition to the frequency, the procedure of hand-washing is also considered important. The proportion of individuals that indicated following the correct procedure of using detergents and water for lathering and rinsing was 96.4% in Dire Dawa slaughterhouse and 77.3% in HU slaughterhouse. Regarding the availability of soap, all of the respondents indicated that soap was not always available.

In the current study, 92.9% of the interviewees in Dire Dawa slaughterhouse and all (100%) of the interviewees in HU slaughterhouse responded that no sanitary regulation system was in place in the slaughterhouse in Table 7. Therefore, effective food safety and quality control programs are essential. The behavior of worker and hygienic practices of retail shop in HU was relatively good as compared to Dire Dawa retails shop, and meat handlers do not have close contact with money and they do so only when 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 (Trickett, 1997).

All the respondents in both slaughterhouses were employed on a temporary basis which makes it difficult to train the staffs. When assessment on the literacy level, the personnel working on food establishment in both areas, 98% of butcher men attend school and 14% of respondent are obtained their skills from their parents, while 80% of the respondents taught themselves through visual observation and 10.7% of respondent in Dire Dawa gained skill through formal training. Training and education of food handlers regarding the basic concepts and requirements of personal hygiene plays an integral part in ensuring a safe product to the consumer (Adams and Moss, 1997). To ensure this, there should be some form of induction training with regular updating and refresher courses for the food handlers. Meat handlers should furthermore understand the risks associated with contamination of food by microbial agents, and should be trained to avoid the contamination of the meat. A formal employee training and assistance program that describes all the training activities should be made attractive to the meat handlers (CFIA, 1990). Ryser and Marth (1991) conclude that the training and education should be directed towards a thorough understanding of food hygiene, which includes aspects of sanitation.

The result from meat handler indicated that 46.4% in Dire Dawa and 13.6% in HU smoke cigarette when they carry out their task. Smoking inside the slaughterhouse or whenever meat is handled should be prohibited, because whenever a cigarette is handled the fingers come into contact with the lips and saliva, together with microorganisms, may consequently be transferred from the hands to the food (Burton, 1996). Smoking may furthermore cause coughing, thus transferring aerosols containing microorganisms to the food (Gordon-Davis, 1998). Moreover 42% of the respondent in both slaughterhouses had worn jewelry materials. Jewelry is a potential source of microorganisms, because the skin under the jewelry provides a favorable habitat for contaminating microorganisms to proliferate (Trickett, 1997).

Regarding protective cloth, the personnel observation and assessment result in both slaughterhouses indicated that, almost all of the food handlers had a uniform protective cloth. However; minimal personal hygiene was practiced during food preparation. Van Zyl (1995) gave emphasis to protective clothing which should not only be on protection, but also on cleanliness, thus he proposed that the overalls, hairnets (beard nets if applicable), hard hats, gum boots and aprons should at all times be worn by meat handlers. Because the purpose of wearing overalls is to protect both the food product and the meat

Table 7. Hygienic and sanitation practices employed at Dire Dawa and HU slaughterhouses 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 8. Educational status of meat handler's

Skills	DD 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

P ≤ 0.001, df= 4

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

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

$P \leq 0.01$, df= 1

handler from cross-contamination, overalls should be suitable to wear over other clothing (CFIA, 1990).

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 (Van Zyl, 1995). 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 (Educational Foundation, 1992; Pelczar et al., 1993).

CONCLUSION AND RECOMMENDATIONS

The results obtained from this study showed that 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 overall prevalence of *E. coli*, *E. coli* O157:H7 and *Salmonella* were 36 (12.4%), 6 (2.06%) and 8 (2.75%) which 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 practice 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 Haramaya University and its surrounding areas.

1. Haramaya University should open the newly constructed slaughterhouses that can improve slaughtering and processing of beef for human consumption and Dire Dawa administrative city authorities should improve their supervision of slaughterhouse workers.
2. Periodic sanitary-hygienic evaluation and inspection of

abattoirs and beef meat retail establishments should be implemented and Health authorities need to enforce legislative requirements and periodic monitoring aimed at insuring the proper slaughtering process and sanitary-hygienic standards. Failure to meet these requirements should result in enforcement action against premises, and this should ultimately lead to prosecution and suspension and/or revocation of their license to operate.

3. Good manufacturing practice and good hygienic practice, together with stringent control of all aspects of meat production, preparation, storage and distribution should be put in place in food establishment in order to reduce contamination of *Salmonella* and other foodborne pathogens to acceptable limit.

4. Training to meat handlers regarding stunning process, food safety and good hygienic practices should be given especially in Haramaya University slaughter house as all workers who had no formal trainings.

CONFLICT OF INTERESTS

The author has not declared any conflict of interest.

ACKNOWLEDGEMENT

We would like to extend our thanks to College of Veterinary Medicine, Haramaya University for all round support.

REFERENCES

Activity Based Management (ABM) (2008). Standards for Abattoir, Cutting and Packing Plant. Version 4.0 January, 2008 Northern Ireland.

Adams MR, Moss MO (1997). Food Microbiol. Cambridge: The R Soc Chem. pp. 200-213.

Adu-Gyamfi A, Torgby-Tetteh W and Appiah V (2012). Microbiological quality of chicken sold in Accra and determination of D10-Value of *E. coli*. Food Nutr. Sci. 3(5):693-698.

Alemseged F, Yami A, Birke W, S/Mariam Z and Worku K (2009). Investigation of Dysentery outbreak and Its causes, Jimma City South west, Ethiopia. Ethiop. J. Health Sci. 19(3):147-154.

Alvarez-Astorga M, Capita R, Alonso-Calleja C, Moreno B, Del M and Garcia-Fernandez C (2002). Microbiological quality of retail chicken by-products in Spain. Meat Sci. 62(1):45-50.

Asrat GT, Berhan T, Solomon M (2008). Inclusion of Different Proportions of Poultry Litter in the Rations of Yearling Hararghe Highland goats. Livest. Res. Rural Dev. 20:48.

Bayleyegn M, Daniel A, Woubit S (2003). Sources and distribution of *Salmonella* serotypes isolated from food animals, slaughterhouse personnel and retail meat products in Ethiopia. Ethiop J. Health Dev. 17:63-70.

Bhandare S, Sherikarv A, Paturkar A, Waskar V, Zende R (2007). A comparison of microbial contamination on sheep/goat carcasses in a modern Indian abattoir and traditional meat shops. Food Control 18: 854-868.

Brown MH, Gill CO, Hollingsworth J, Nickelson R, Seward S, Sheridan JJ (2000). The role of microbiological testing in systems for assuring the safety of beef. Int. J. Food Microbiol. 62:7-16.

Burton GR (1996). Microbiology for the health sciences (4th ed.). New York: Lippencott. pp. 234-239.

Canadian Food Inspection Agency (CFIA) (1990). Meat hygiene manual of procedures, Canada. Available at: http://www.inspection.gc.ca/food/meat-and-poultry-products/manual-of-procedures/eng/1300125426052/1300125482318.

Centers for Disease Control and Prevention (CDC) (2013). FoodNet: Incidence and Trends in Foodborne Illness.

Central of Statistics Agency (CSA) (2007). Agricultural sample survey 2006/07(1999 EC). Report on livestock and livestock characteristics (private peasant holdings), CSA, Addis Ababa, Ethiopia.

Chapman PA, Siddons CA, Cerdan Malo AT, Harkin MA (2000). A one year study of *E. coli* O157 in raw beef and lamb products. Epidemiol. Infect. 124:207-213.

Chhabra MB, Singla LD (2009). Food-borne parasitic zoonoses in India: Review of recent reports of human infections. J. Vet. Parasitol. 23(2):103-110.

Collee JG, Mackie TJ (1999). *Practical Medical Microbiology*. 14th ed Churchill living stone. pp 883-884, 898-907.

Desenclos JC, Zergabachew A, Desmoulins B, Chouteau L, Desve G (1988). Clinical, microbiological and antibiotic susceptibility patterns of diarrhoea in Korem, Ethiopia. J. Trop. Med. Hyg. 91:296-301.

Desmarchelier PM, Higgs GM, Mills L, Sullivan AM, Vanderlinde PB (1999). Incidence of coagulase positive *Staphylococcus* on beef carcasses in three Australian abattoirs. Int. J. Food Microbiol. 47:221-229.

Diane GN, Marion K, Linda V, Erwin D, Awa AK, Hein S, Marieke O, Merel L, John T, Flemming S, Koke V-DG, Hild K (2010). Foodborne diseases. Int. J. Food Microbiol. 139:s3-s15.

Doyle ME (2007). Microbial food spoilage – Losses and control strategies, (A brief review of the Literature), FRI Briefings. Available at: www.wisc.edu/fri/

Duffy EA, Belk KE, Sofos JN, LeValley SB, Kain ML, Tatum JD, Smith GC, Kimberling CV (2001). Microbial contamination occurring on lamb carcasses processed in the United States. J. Food Prot. 64 (4):503-508.

Educational Foundation of the National Restaurant Association (1992). Applied food service sanitation (4th ed.). Canada: John Wiley and Sons. pp: 44-45.

Ejeta G, Molla B, Alemayehu D, Muckle A (2004). *Salmonella* serotypes isolated from minced meat beef, mutton and pork in Addis Ababa, Ethiopia. Revue Méd. Vét. 11:547-551.

Evans JA, Russell SL, James C, Corry JE (2004). Microbiological contamination of food refrigeration equipment. J. Food Eng. 62:225-232.

Fawole MO, Oso BA (2001). *Laboratory manual of microbiology*: Revised edition. Ibadan: Spectrum books Ltd. P 127.

FDA (Food and Drug Authority) (1992). Bacteriological Analytical Manual. 7th Ed. AOAC international 2200 Wilson Blvd, Suite 400, Arlington, VA. Available at: ttp://www.fda.gov/Food/FoodScienceResearch/LaboratoryMethods/ucm2006949.htm.

Fegan N, Vanderlinde P, Higgs G, Desmarchelier P (2004). Quantification and prevalence of *Salmonella* in beef cattle presenting at slaughter, J. Appl. Microbiol. 97:892-898.

Fratamico PA, Bhunia AK, Smith JL (2005). Foodborne Pathogens: Microbiology and Molecular Biology, Caister Academic Press, Wymondham, Norfolk, UK. P 273.

Gill CO (2007). Sampling of red meat. In Microbiological Analysis of red meat, poultry and egg, Mead, G.C. (editor). Cambridge, England; Woodhead Publishing Limited. pp. 132-135.

Gill CO, Mcginnis JC (2004). Microbiological conditions of air knives before and after maintenance at a beef packing plant. Meat Sci. 68:333-337.

Gill CO, Badoni M, McGinnis JC (1999). Assessment of the adequacy of cleaning of equipment used for breaking beef carcasses. Int. J. Food

Microbiol. 46:18.
Gill CO, Bryant J, Brereton DA (2000). Microbiological conditions of sheep carcasses from conventional or inverted dressing processes. J. Food Prot. 63(9):1291-1294.
Gordon-Davis L (1998). The hospitality industry handbook on hygiene and safety: for South African students and practitioners. Kenwyn: Juta publication. pp. 49-65
Haque MA, Siddique MP, Habib MA, Sarkar V, Chou KA (2008). Evaluation of sanitary quality of goat meat obtained from slaughter yards and meat stalls at late market hours. Bangl. J. Vet. Med. 6(1):87-92.
Hassan AN, Farooqui A, Khan A, Khan Y, Kazmi SU (2010). Microbial contamination of raw meat and its environment in retail shops in Karachi, Pakistan. J. Infect. Dev. Ctries. 4(6):382-388.
Haymanot T, Alemseged A, Getenet B, Solomon GS (2010). Microbial flora and foodborne pathogens on minced meat and their susceptibility to antimicrobial agents Jimma city, Ethiopia. Ethiop. J. Health Sci. 20(3):137-143.
Hiko A, Asrat D, Zewde G (2008). Occurrence of *E. coli* O157:H7 in retail raw meat products in Ethiopia. J. Infect Dev. Ctries. 2:389-393.
ICMSF (International Commission on Microbiological Specifications for Foods) (1986). Microorganisms in Foods 2. Sampling for microbiological analysis: Principle and specific application, (2nd ed.) Blackwell Scientific Publications. UK. pp. 132-136.
ISO (International Organization for Standardization)/TS 11133-1 (2009). Microbiology of food and animal feeding stuffs. *Guidelines on preparation and production of culture media*. Part 1: General guidelines on quality assurance for the preparation of culture media in the laboratory.
ISO (International Organization for Standardization)-16654 (2001). Microbiology of food and animal feeding stuffs. Horizontal method for the detection of *E. coli* O157:H7.
ISO (International Organization for Standardization)-4333 (2003). A horizontal method for the enumeration of microorganisms by counting the colonies growing in solid medium after aerobic culturing of aerobic bacteria.
ISO (International Organization for Standardization)-6579 (2002). Microbiology-General guidance on methods for the detection of *Salmonella,* 4rd ed International Organization for Standardization, Geneva.
Larson E, Aiello A, Lee LV, Della-Latta P, Gomez-Duarte C, Lin S (2003). Short and long term effects of hand washing with antimicrobial or plain soap in the community. J. Commun. Health 28(2):139-150.
Luga I, Akombo PM, Kwaga JKP, Umoh VJ and Ajogi I (2007). Sero-prevalence of Faecal Shedding of *E. coli* O157:H7 from Exotic Dairy Cattle in North-Western Nigeria. Niger. Vet. J. 28:6-11.
Mache A, Mengistu Y, Cowley S (1997). *Salmonella* sero groups identified from adult diarrheal out-patients in Addis Ababa, Ethiopia: Antibiotic resistance and plasmid profile analysis. East Afr. Med. J. 74:183-187.
Mekonnen H, Habtamu T, Kelali A, Shewit K (2012). Food safety knowledge and practices of abattoir and butchery shops and the microbial profile of meat in Mekelle City, Ethiopia. Asian Pac. J. Trop. Biomed. 12:952-957.
Mersha G, Asrat D, Zewde BM, Kyule M (2009). Occurrence of *E. coli* O157:H7 in faeces, skin and carcasses from sheep and goats in Ethiopia. Lett. Appl. Microbiol. 50:71-76.
Molla B, Kleer J, Sinell HJ (2000). Occurrence, distribution and level of *Salmonella* in selected food items in Addis Ababa, Ethiopia. Fleischwirt Int. 4:37-39.
Mukhopadhyay HK, Pillai RM, Pal UK, Kumar VJA (2009). Microbial quality of fresh chevon and beef in retail Outlets of pondicherry. Tamilnadu J. Vet. Anim. Sci. 5 (1):33-36.
Pelczar MJ, Chan ECS, Krieg NR (1993). *Microbiology, concepts and applications*. New York: McGraw-Hill. pp. 80-100, 158-161, 370.
Podpecan B, Pengov A, Vadnjal S (2007). The source of contamination of ground meat for production of meat products with bacteria *Staphylococcus aureus*. Slov. Vet. Res. 44:25-30.
Pointon A, Sexton M, Dowsett P, Saputra T, Kiermeier A, Lorimer M, Holds G, Arnold G, Davos D, Combs B, Fabiansson S, Raven G, McKenzie H, Chapman A, Sumner J (2008). A baseline survey of the microbiological quality of chicken portions and carcasses at retail in two Australian states (2005 to 2006). J. Food Prot. 71(6):1123-1134.
Ryser ET, Marth EH (1991). Listeria, Listeriosis, and food safety. New York: Marcel Dekker. pp. 183-189.
Sefinew A, Bayleyegn M (2012). Prevalence and antimicrobial resistance profiles of *Salmonella* enteric serovars isolated from slaughtered cattle in Bahir Dar, Ethiopia. Trop. Anim. Health Prod. 44:595-600.
Sibhat B, Molla B, Zerihun A, Muckle A, Cole L, Boerlin P, Wilkie E, Perets A, Mistry K, Gebreyes WA (2011). *Salmonella* serovars and antimicrobial resistance profiles in beef cattle, slaughterhouse personnel and slaughterhouse environment in Ethiopia. Zoonoses Public Health 58:102-109.
Sneed J, Strohbehn C, Gilmore AS, Mendonca A (2004). Microbiological evaluation of food service contact surfaces in Iowa assisted living facilities. J. Am. Diet. Assoc. 104:1722-1724.
Sousa CP (2008). The Impact of Food Manufacturing Practices on Foodborne Diseases. Braz. Arch. Biol. Technol. 51(4):815-823.
Taye M, Berhanu T, Berhanu Y, Tamiru F, Terefe D (2013). Study on carcass contaminating *E. coli* in apparently healthy slaughtered cattle in Haramaya University slaughter house with special emphasis on *E. coli* O157:H7, Ethiopia. J. Vet. Sci. Technol. 4:132.
Tegegne M, Ashenafi M (1998). Microbial load and incidence of *Salmonella* species in kitfo", traditional Ethiopian spiced, minced meat dish. Ethiop. J. Health Dev. 12:135-140.
Temelli S, Dokuzlu C, Cemsen MK (2006). Determination of microbiological contamination sources during frozen snail meat processing stages. Food Control 17:22-29.
Thrusfield M (2007). Veterinary Epidemiology, 3rd Edition. Blackwell Science, UK. P 332.
Timothy M, Smith JR (2012). Isolation, Identification, and Enumeration of Pathogenic *Salmonella* Serovars from Environmental Waters. J. Food Prot. 76(7):2-251.
Trickett J (1997). Food hygiene for food handlers. London: Macmillan Press. pp. 19-27.
Van Zyl AP (1995). Manual for the abattoir industry (1st ed.). Pretoria: Red Meat Abattoir Association. pp. 123-156.
WHO (World Health Organization) (2007). Food safety and foodborne diseases and value chain management for food safety. "Forging links between Agriculture and Health" CGIAR on Agriculture and Health Meeting in WHO/HQ.
Wilson I (2002). *Salmonella* and *Campylobacter* contamination of raw retail chickens from different producers: a six year survey. Epidemiol. Infect. 129:635-645.
Zewdu E, Cornelius P (2009). Antimicrobial resistance pattern of *Salmonella* serotypes isolated from food items and personnel in Addis Ababa, Ethiopia. Trop. Anim. Health Prod 41:241-249.