

MEAT SURFACE CONTAMINATION IN JIGJIGA: A MICROBIOLOGICAL STUDY OF ABATTOIRS AND RETAIL HOUSES

¹Abebe G. Kebede and ²James L. Anderson

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Abstract

Slaughter facilities involved in various meat processing stages, such as skinning, evisceration, storage, and distribution, can serve as significant sources of contamination for meat and meat products. In developing countries, traditional methods of handling, processing, and marketing meat often undermine quality due to poor sanitation, leading to substantial product loss and increased risk of food-borne diseases. External contamination of meat, particularly in abattoirs, is a major concern, with microbial surface contamination of carcasses significantly impacting meat shelf life. Fecal matter is a primary source of microbial contamination, which can reach carcasses through direct deposition or indirect contact via contaminated equipment, surfaces, and workers. This study explores the extent of contamination in meat processing facilities in developing countries and highlights the critical need for improved sanitation practices to enhance meat quality and safety.

INTRODUCTION

Slaughter facilities used at different meat processing stages such as skinning, evisceration, storage and distribution could serve as a source of contamination to meat and meat products (Abdalla et al., 2009a). In most developing countries, traditional methods of handling, processing and marketing of meat undermine quality where poor sanitation leads to considerable loss of product as well as the risk of food-borne diseases (Garcia, 2007).

The external contamination of meat comprises a major problem in most developing countries' abattoirs and microbial surface contamination of carcasses has been repeatedly reported to have a significant effect on the meat shelf life (Yen, 2003). Fecal matter is a major source of microbial contamination and could reach carcasses through direct deposition as well as by indirect contact through contaminated and unclean equipment, surfaces, workers and so on (Abdalla et al., 2009b).

In Ethiopia, the current intervention strategies used in the abattoir and retail houses are insufficient in maintaining hygienic level of meat processing establishments (Molla et al., 2003). Presently, operations of abattoir and retail houses are hardly inspected by veterinary and public health officers as per the regulations. There is high

¹ College of Veterinary Medicine, Haramaya University, P.O. box 138, Dire Dawa, Ethiopia.

² Alabama A & M University, Normal, AL 35811, Alabama, United States.

possibility of detecting pathogens in meat processing establishments designed for production of meat intended for human consumption (Balcha et al., 2014).).

Since there is an increasing demand for meat and meat products, it is of paramount importance to assess the contamination levels of meat contact surfaces in the municipal abattoir and retail houses with pathogens of public health significance (*Staphylococcus aureus*, *Escherichia coli* O157:H7, *Listeria monocytogenes*, and *Campylobacter* spp.). The information on meat production area hygiene status will facilitate designing microbial contamination preventive strategies in abattoir and retail houses and provide baseline data for related studies. The study determined the microbiological quality and hygiene levels of meat contact surfaces at municipal abattoir and retail houses in Jigjiga town, Somali National Regional State of Ethiopia.

MATERIALS AND METHODS

Sample collection and preparation

A total of ninety pooled environmental samples were collected from the municipal abattoir and four retail houses. On each visit to the abattoir, a total of three pooled swab samples were taken from: cleaned, disinfected and dry floor surface, hooks and knives and the third from butchers' hands before beginning of work. During each visit to the retail house, a total of three pooled swab samples were taken from: cutting boards, hooks and knives and the third from butchers' hands before the beginning of work by rubbing thoroughly with a moistened swab.

The microbiological quality of meat contact surfaces and environment were assayed by using the methods described in the Compendium of Methods for the Microbiological Examination of Foods (Doores et al., 2013). Sterile swabs were used to sample 1 cm² areas of meat contact surfaces. Before sampling, sterile swabs were kept in 9 ml sterile peptone water. After sampling, the swabs were placed again aseptically back into peptone water. All samples were transported to the Veterinary Microbiology Laboratory of Haramaya University in an ice box with ice packs and analyzed upon arrival or within 24 h of sampling.

Microbiological quality assessment

In the laboratory, each tube containing the swab was vortexed 10 s to ensure mixture of the sample and 10-fold serial dilutions were made with 0.1% sterile buffered peptone water (BPW) (Oxoid, CM0509) to 10⁻⁶. All the samples were analyzed for the presence and/or concentration of *S. aureus*, *E. coli* O157:H7, *L. monocytogenes*, aerobic plate counts (APCs); fecal coliforms (FC); yeast and molds (Y&M) counts and *Campylobacter* spp.

Aerobic plate count (APC)

For counting aerobic bacteria, 0.1 ml of homogenate was plated onto the surface of plate count agar (Oxoid, CM0325). Plates were incubated at 35°C for 48 h and plates containing between 30 and 300 colonies were counted (ISO, 2009).

Fecal coliforms count (FCC)

Fecal coliforms were enumerated using violet red bile (lactose) agar (VRBL) (Oxoid, CM0107); 0.1 ml of the homogenate were spread on to agar plates and incubated at 44 ± 1°C for 24 h, typical and atypical colonies were enumerated (American Public Health Association, 2012).

Yeast and mold counts

Enumeration of yeasts and molds was done using potato dextrose agar (PDA) (Oxoid, CM0139). The inoculums (0.1 ml of the homogenate) were spread onto PDA and incubated at 30-32°C for 2-7 days or longer. Yeasts were growing as creamy to white colonies whereas molds were growing as filamentous colonies of various colors (United States Pharmacopeia Convention, 2007).

Detection and enumeration of *S. aureus*

Appropriate portion of dilution (0.1 ml) was transferred to Baird Parker Agar (Oxoid, CM0275) plates and distributed over the surface using a sterile, bent glass rod. After allowing inoculums to be absorbed by the medium

before inverting the plates, they were incubated at 37°C for 48 h. Plates having 30-300 colonies were examined, counting colonies typical of *S. aureus* using Stuart sc6plus colony counter (Bibby scientific Limited, UK). Coagulasepositive staphylococci produce black, shiny, convex colonies with entire margins and clear zones, with or without an opaque zone (United States Pharmacopeia Convention, 2007).

Detection and enumeration of *E. coli* O157:H7

Appropriate portion of dilution (0.1 ml) was transferred onto Sorbitol-MacConkey agar (SMA) (Oxoid, CM0813, SR0172) and incubated at 35°C for 20 to 22 h. Presumptive *E. coli* O157:H7 colonies were counted using colony counter (Timothy and Smith, 2012). *E. coli* O157:H7 does not ferment sorbitol and produces colorless colonies. In contrast, most other *E. coli* strains ferment sorbitol and form pink colonies.

Detection and enumeration of *L. monocytogenes*

For the detection and enumeration of *L. monocytogenes*, 0.1 ml of the dilution was spread onto *Listeria* selective agar (LSA) (Oxford, CM0856, SR0140). After incubation for 48 h at 37°C, presumptive *L. monocytogenes* colonies were counted. *L. monocytogenes* produce special brown color producing black zones around the colonies due to the formation of black iron phenolic compounds derived from the aglucon, so can be easily identified (Tavakoli et al., 2008).

Test for *Campylobacter* spp.

One milliliter of each of the collected samples was suspended in 9 ml of Bolton selective enrichment broth (Oxoid, CM0983, SR0183) and incubation at 37°C for 4 h, followed by further incubation at 41.5°C for 44 h and sub cultured to *Campylobacter* selective agar (CCDA; Oxoid, CM739, SR0155) at 41.5°C for 48 h. A typical *Campylobacter* on CCD-agar had a gray, moistening and effuse appearance. *Campylobacter jejuni* has a green or gray appearance that can be very dry. At the same time, the appearance can be with or without a shine of metal. A creamy grey, moistening and raised colony is a typical *Campylobacter coli* (Salihu et al., 2009).

Table 1. Bacterial species detected from meat contact surfaces sampled from Jigjiga town municipal abattoir and retail houses.

Sources	No. of sample	Bacterial species detected			
		<u>S. aureus</u>	<u>E. coli</u>	<u>L. monocytogenes</u>	<u>Camplyobacter</u>
		No. (%)	O157H7 No. (%)	No. (%)	No. (%)
Abattoir					
Knives and hooks	15	4(26.67)	1(6.67)	0(0)	0(0)
Surfaces	15	5(33.33)	2(13.33)	0(0)	3(20)
Butchers’ hands	15	4(26.67)	2(13.33)	0(0)	0(0)
Retail houses					
Knives and hooks	15	7(46.67)	0(0)	0(0)	0(0)
Cutting boards	15	3(20)	0(0)	0(0)	0(0)
Butchers’ hands	15	6(40)	1(6.67)	0(0)	0(0)
Total	90	29(32.22)	6(6.67)	0(0)	3(3.33)

Table 2. Pathogenic bacteria load of meat contact surfaces from Jigjiga municipal abattoir and retail houses.

Sources	No. of sample	Bacterial colonies log10 cfug/cm ²					
		<i>S. aureus</i>			<i>E. coli</i> O157H7		
		Mean±SE	Min	Max	Mean±SE	Min	Max
Abattoir							

Knives and hooks	15	6.39±0.07	6.19	6.47	5.78±0.00	5.78	5.78
Surfaces	15	5.98±0.07	5.75	6.18	5.94±0.02	5.91	5.96
Butchers' hands	15	5.82±0.11	5.71	5.93	6.03±0.03	6.00	6.06

Retail houses

Knives and hooks	15	6.14±0.11	5.84	6.44	-	-	-
Cutting boards	15	5.61±0.10	5.50	5.71	-	-	-
<u>Butchers' hands</u>	<u>15</u>	<u>6.43±0.34</u>	<u>5.77</u>	<u>6.85</u>	<u>5.96±0.00</u>	<u>5.96</u>	<u>5.96</u>

Statistical analysis

The results of microbial counts (CFU/cm²) were converted into log₁₀ and descriptive statistics were used to calculate mean, standard error, minimum and maximum values considering the type of sample and origin. Percentages were calculated to express the frequency of contamination. Microbial counts were compared by ANOVA. P-value <0.05 was considered statistically significant at the 95% confidence level. All data were analyzed using SPSS (Statistical Package for Social Science) software version 20.

RESULTS AND DISCUSSION

The study determined the microbiological quality and hygiene levels of meat contact surfaces at municipal abattoir and retail houses in Jigjiga town, Somali National Regional State of Ethiopia.

Bacterial profile and load of meat contact surfaces at abattoir and retail houses

The occurrence and average microbial concentrations of *S. aureus* in abattoir and retail house contact surface samples are summarized in Tables 1 and 2, respectively. In the present study, 29 (32.22%) samples out of 90 pooled contact surfaces samples revealed typical colonies of *S. aureus* on BPA. *S. aureus* positive samples ranged from 4/15 (26.67%) for hooks and knives, and butchers' hands to 5/15 (33.33%) for floor surface of abattoir whereas for retail houses, *S. aureus* positive samples ranged from 3/15 (20%) for cutting boards and 7/15 (46.67%) for hooks and knives. In general, the occurrences of *S. aureus* from meat contact surfaces were high [7/15(46.67%)] in hooks and knives, and low [3/15 (20%)] in cutting boards samples of retail houses.

The mean count for *S. aureus* from abattoir samples ranged from 5.82±0.11 log₁₀ cfu/cm² for butchers' hands to 6.39±0.07 log₁₀ cfu/cm² for the hooks and knives. Whereas, the mean *S. aureus* count for retail houses ranged from 5.61±0.10 log₁₀ cfu/cm² for cutting boards to

$6.43 \pm 0.34 \log_{10} \text{ cfu/cm}^2$ for butchers' hands. The mean *S. aureus* count from meat contact surfaces were high ($6.43 \pm 0.34 \log_{10} \text{ cfu/cm}^2$) in butchers' hands and low ($5.61 \pm 0.10 \log_{10} \text{ cfu/cm}^2$) in cutting boards samples of retail houses. High *S. aureus* counts in meat contact surfaces samples indicate high levels of contamination due to use of unhygienic processes both at abattoir and retail houses (Timm et al., 2013) or failure of the abattoir workers to observe proper hand washing regularly.

The occurrence and load of *E. coli* O157:H7 in meat contact surface samples from Jigjiga municipal abattoir and retail houses are summarized in Tables 1 and 2, respectively. Out of 90 pooled contact surfaces samples, *E. coli* O157:H7 was present in 6 (6.67%) samples from hooks and knives, floor surfaces and butchers' hands at the abattoir. *E. coli* O157:H7 positive samples ranged from 1/15 (6.67%) for abattoir hooks and knives, and retail houses butchers' hands to 2/15 (13.33%) for floor surface and butchers' hands at the abattoir. *E. coli* O157:H7 was not detected on hooks and knives, and from cutting boards of retail houses.

The highest average *E. coli* O157:H7 ($6.03 \pm 0.03 \log_{10} \text{ cfu/cm}^2$) counts were detected from butchers' hands samples from the abattoir. Whereas the lowest

($5.78 \pm 0.00 \log_{10} \text{ cfu/cm}^2$) count were detected in abattoir hooks and knives samples. Considering the very low infective dose (10-100 cfu(g/l) for *E. coli* O157:H7, its detection at such concentrations in contact surfaces of the abattoir and retail houses represents high contamination levels and poses significant public health risks to meat consumers. Moreover, presence of *E. coli* showed the presence of recent fecal contamination (Timm et al., 2013). In this study *L. monocytogenes* were not detected both in the abattoir and retail houses meat contact surfaces. However, Kornacki and Gulner (2007) reported that *L. monocytogenes* are highly prevalent in meat processing facilities within North America and Europe, especially beef processing plants (28-92%), poultry processing plants (13.3%) and fish processing plants (12.8%). Variation in the occurrence of *L. monocytogenes* from meat contact surface samples reported in other studies may be due to differences in sampling techniques employed, seasonal effects and/or laboratory methodologies employed in different studies. Therefore, determination of the ultimate contamination level would require an intensive on-site investigation of microbiological quality which is essential, especially on the surface of processing hooks and knives and facilities that have direct contact with the meat, where biofilm may exist (Pui et al., 2011).

Campylobacter spp. were only detected in abattoir floor surfaces, other contact surface samples were negative for the organism. Overall, 3 (3.33%) of meat contact surface samples were contaminated with *Campylobacter* spp. Among the commonly encountered *Campylobacter* spp. in meat processing environment, *C. jejuni* and *C. coli* were identified. Occurrence of *Campylobacter* spp. may be due to cross-contamination during manual skinning, evisceration and processing in the slaughter house or insufficient hygiene during processing in the retail houses. *Campylobacters* naturally present in the intestinal tract of animals represent a potential risk for contamination of meat contact surfaces and subsequently the carcasses depending on shedding patterns and hygienic manufacturing practices adopted by the abattoirs or slaughterhouses (Ghafir et al., 2007).

Hygienic quality of meat contact surfaces at abattoirs and retail houses

The occurrence and average microbial concentrations of aerobic bacteria (AB), fecal coliforms (FCs) and yeast and molds (Y&Ms) in abattoir and retail houses contact surface samples are summarized in Tables 3 and 4, respectively. Out of 90 pooled contact surfaces samples, FCs, AB and Y&Ms were present in 54/90 (60%), 40/90 (44.44%) and 31/90 (34.44%) samples, respectively. The occurrences of FCs, AB and Y&Ms in meat contact surfaces samples were high in floor surface [13/15(86.67)], hooks and knives [9/15(69.23)] and butchers' hands samples [8/15(53.33)] of abattoir and low in butchers' hands at abattoir [5(33.33)], hooks and knives, and butchers' hands at retail houses [4(26.67)], and butchers' hands at abattoir [2(13.33)], respectively.

Based on the data from abattoir, the highest FCCs ($6.25 \pm 0.075 \log_{10} \text{ cfu/cm}^2$) and Y&MCs ($5.19 \pm 0.513 \log_{10} \text{ cfu/cm}^2$) levels found in the floor surfaces while the highest APCs ($6.08 \pm 0.126 \log_{10} \text{ cfu/cm}^2$) found in butchers' hands. The smallest values of FCs ($5.72 \pm 0.094 \log_{10} \text{ cfu/cm}^2$), APCs ($5.99 \pm 0.135 \log_{10} \text{ cfu/cm}^2$) and Y&MCs ($4.55 \pm 0.045 \log_{10} \text{ cfu/cm}^2$) were seen in butchers' hands, floor surface and hooks and knives, respectively.

In the retail houses, it was evident that the highest FCCs ($5.83 \pm 0.076 \log_{10} \text{ cfu/cm}^2$), APCs ($6.03 \pm 0.153 \log_{10} \text{ cfu/cm}^2$) and Y&MCs ($4.81 \pm 0.045 \log_{10} \text{ cfu/cm}^2$) levels were found in the hooks and knives, cutting boards and butchers' hands, respectively. Meat handlers had the smallest values of FCCs ($5.72 \pm 0.090 \log_{10} \text{ cfu/cm}^2$) and 2 APCs ($5.90 \pm 0.074 \log_{10} \text{ cfu/cm}^2$), while hooks and knives had the smallest values of Y&MCs ($4.62 \pm 0.049 \log_{10} \text{ cfu/cm}^2$).

In general, it was evident that the highest FCCs ($6.25 \pm 0.075 \log_{10} \text{ cfu/cm}^2$) and Y&MCs ($5.19 \pm 0.513 \log_{10} \text{ cfu/cm}^2$) were detected in the floor surfaces while the highest APCs ($6.08 \pm 0.126 \log_{10} \text{ cfu/cm}^2$) found in butchers' hands at abattoir. The smallest values of FCCs ($5.72 \pm 0.094 \log_{10} \text{ cfu/cm}^2$), APCs ($5.90 \pm 0.07 \log_{10} \text{ cfu/cm}^2$) and Y&MCs ($4.55 \pm 0.045 \log_{10} \text{ cfu/cm}^2$) got abattoir and retail house butchers' hands, retail houses butchers' hands and abattoir hooks and knives, respectively.

The findings of this study indicated that meat contact surfaces harbor important foodborne pathogens such as *S. aureus*, *E. coli* O157H7 and *Campylobacter* hence posing a risk of meat contamination and subsequently foodborne intoxication or infection for the consumers. Similar findings were reported by Temelli et al. (2006), Gill and McGinnis (2004). In the present study, all types of contact surfaces displayed average levels of contamination exceeding $4.00 \log_{10} \text{ cfu/cm}^2$, which is sufficient to initiate the formation of biofilms (Timm et al., 2013). This level of contamination was observed in all contact surfaces including hooks and knives, cutting board, butchers' hands and floor surface, which can accumulate large amounts of organic matter, which favors microbial growth. An unacceptable levels for both pathogenic and indicator bacteria were observed in meat contact surface samples. Therefore, these organisms can be decreased by applying good manufacturing practices and HACCP procedures (Brady and Morris, 2005). Because the identified pathogenic bacteria can cause serious public health problems, it is necessary to eliminate or minimize their presence. This in turn can only be achievable if attention is given to training and supervision of meat handlers to ensure proper hand washing and appropriate cleaning and sanitation procedures in meat processing plants to reduce or eliminate cross-contamination (Kusumaningrum et al., 2003). Based on the standards used in the food processing industry; a standard of less than $1.3 \log_{10} \text{ cfu}$ was used for aerobic plate count and less than $1.0 \log_{10} \text{ cfu}$ for Enterobacteriaceae such as *E. coli* count. According to this standard, the results for food contact surfaces in our study were unacceptable (Sneed et al., 2004).

Table 3. Indicator organisms detected from meat contact surfaces sampled from Jigjiga municipal abattoir and retail houses.

Organisms detected				
Sources	No. of sample	FCs	AB	Y&Ms
		No. (%)	No. (%)	No. (%)
Abattoir				
Knives	and15	11(73.33)	9(69.23)	5(33.33)
hooks				
Surfaces	15	13(86.67)	9(60)	8(53.33)
Butchers' hands	15	5(33.33)	6(40)	2(13.33)
Retail houses				

Knives and hooks	15	9(60)	4(26.67)	7(46.67)
Cutting boards	15	8(53.33)	8(53.33)	3(20)
Butchers' hands	15	8(53.33)	4(26.67)	7(46.67)
<u>Total</u>	<u>90</u>	<u>54(60)</u>	<u>40(44.44)</u>	<u>31(34.44)</u>

Table 4. Microbial loads of indicator organisms on meat contact surfaces from Jigjiga abattoir and retail houses.**Enumerated organisms \log_{10} cfug/cm²**

Sources	No. of sample	FCCs			APCs			Y&Ms	
		Mean±SE	Min	Max	Mean±SE	Min	Max	Mean±SE	MinMax
Abattoir									
Knives and hooks	15	6.00±0.00	6.00	6.00	6.05±0.07	5.89 4.63	6.31	4.55±0.05	4.47
Surfaces	15	6.25±0.08	5.89	6.47	5.99±0.14	5.65 5.70	6.45	5.19±0.51	4.68
Butchers' hands	15	5.72±0.09	5.49	5.91	6.08±0.13	5.95 4.93	6.33	4.79±0.05	4.67
Retail Houses									
Knives and hooks	15	5.83±0.08	5.57	5.99	6.01±0.07	5.95 4.76	6.07	4.62±0.05	4.53
Cutting boards	15	5.80±0.12	5.50	6.00	6.03±0.15	5.70 4.76	6.44	4.67±0.09	4.57
<u>Butchers' hands</u>	15	<u>5. 72±0.09</u>	<u>5.54</u>	<u>5.90</u>	<u>5.90±0.07</u>	<u>5.69</u>	<u>6.02</u>	<u>4.81±0.05</u>	<u>4.69</u>
						4.94			

Conclusion

This study showed that all the meat contact surfaces had pathogenic and indicator bacteria. The findings have highlighted high contamination levels of abattoir and retail houses meat contact surfaces. This may contribute towards a high incidence of food associated illnesses through cross contamination of different food products. Maintenance of slaughter hygiene and regular monitoring of meat establishments is essential to minimize the risk of direct and cross-contamination of the meat thereby ensuring meat quality and public health protection.

Conflict of interests

The author(s) did not declare any conflict of interest.

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