

Carriage, Risk Factors, and Antimicrobial Resistance Patterns of Salmonella Isolates from Raw Beef in Jimma, Southwestern Ethiopia

Minda Asfaw Geresu ¹
Wondimu Zeleke Desta²

¹Department of Veterinary Science,
College of Agriculture and Environmental
Science, Arsi University, Asella, Ethiopia;

²School of Veterinary Medicine, College
of Agriculture and Veterinary Medicine,
Jimma University, Jimma, Ethiopia

Purpose: Consumption of raw beef infected with multidrug-resistant Salmonella is pertinent to the world public health risk of antimicrobial resistance. Henceforth, this study aimed to investigate the carriage, antimicrobial resistance (AR) patterns, and the revealing risk factors of Salmonella-contaminating beef in abattoirs and butcher shops in Jimma town.

Methods: A cross-sectional study was conducted to investigate the carriage and AR patterns and to reveal the risk factors of beef contaminated by Salmonella spp. Three hundred and forty-eight swab samples were collected from abattoirs (n=210) and butcher shops (n=138) and the conventional cultural methods were employed for identification of Salmonella. Isolates were subjected to 12 antimicrobials using the Kirby–Bauer disk diffusion method for AR patterns.

Results: The occurrence of Salmonella isolates from the abattoir samples was 11.4%, whereas about 6.52% of isolates were recovered from butcher shops. Educational status (abattoir: odds ratio (OR)=8.40, confidence interval (CI)=1.186–59.493; butcher shops: OR=9.17, CI=1.15–73.239), job related training (abattoir: OR=5.50, CI=1.065–28.416), contamination risk perception (abattoir: OR=5.31, CI=1.256–22.489), neatness of knives (abattoir: OR=7.6, CI=0.892–65.376), source of contamination (abattoir: OR=8.44, CI=1.682–42.39), wearing of protective cloth (butcher shops: OR=8.44, CI=1.682–42.39), manner of hand washing (butcher shops: OR=7.25, CI=1.210–43.442), and money handling (butcher shops: OR= 9.69, CI=1.578–59.474) were among the potential risk factors significantly associated with Salmonella carcass contamination in the abattoir and butcher shops. Of the 33 Salmonella isolates, 14 (58.3%) and six (66.7%) of the abattoir and butcher shops isolates, correspondingly, were resistant to two or more antibiotics.

Conclusion: The finding of this investigation exhibited extensive multidrug-resistant Salmonella isolates in the study setting. Hence, establishing standard meat safety requirements and provision of training for meat handlers and prudent use of antimicrobials are recommended.

Keywords: abattoir, antimicrobial resistance, beef, butcher shop, Jimma, Salmonella

Correspondence: Minda Asfaw Geresu
Department of Veterinary Science,
College of Agriculture and Environmental
Science, Arsi University, PO Box 193,
Asella, Ethiopia
Tel +251910431505
Email minda.asfaw@gmail.com

Introduction

Salmonella is one of the foremost causes of foodborne disease epidemics worldwide.^{1,2} Salmonella spp. are the prominent bacterial pathogens amid other food-borne pathogens and are responsible for instigating gastroenteritis in humans.³ Infections caused by Salmonella spp. in farm animals has been documented as the leading cause of considerable economic losses worldwide.^{4,5} Globally, about 93.8 million cases and 155,000 deaths are associated with gastroenteritis due to

Salmonella species every year.⁶ Reports revealed that about 85.6% were estimated to be food borne, and infection was associated with many different food types, including beef and beef products.⁷

Studies revealed that contaminated feeds, transportation of animals to the abattoir, the slaughtering operation, and edible organs being contaminated by fecal storage, distribution, and preparation for consumption may contribute to Salmonella contamination of raw meat.⁸ Uncleanliness of equipment, utensils, and personal hygiene of food handlers aid in the spread of Salmonella.⁹

Due to the habit of consuming raw animal origin food, the infection rate of food-borne Salmonella has been augmented intensely in Ethiopia during the past few years. Though the extent of food-borne illness and its severity has been known much more, effort has not been made to overcome the problem.⁸ Investigations carried out in different corners of the country have demonstrated the occurrence of Salmonella in diverse food animals and food products.^{6,8,10,11}

Though the above studies revealed that Salmonella spp. are the leading food-borne pathogens that extremely overwhelm the economic growth of the country, there are only a few small-scale reports on the bacteriological quality, sanitary conditions, and practices in abattoirs and retailer (butcher) shops of beef meat in Jimma Zone.

Therefore, the absence of a comprehensive recent study on the carriage and antimicrobial resistance patterns of Salmonella isolates from raw beef in Jimma municipal abattoir and meat retailer (butcher) shops multifaceted with increased consumption of raw/minced meat (locally known as “kitfo” and “Kurt”) by the community at large entails isolation, identification, antimicrobial profile characterization, hygienic practices of the abattoir and butcher shop workers/consumers, and concomitant putative risk factors exposing to Salmonella food borne infections in the study setting.

Materials and Methods

Study Site

This study was carried out in Jimma municipal abattoir and meat retailers (butcher) shops from September 2016 to July 2017. One municipal abattoir and 74 meat retailer (butcher) shops were located in Jimma town, 352 km southwest of Addis Ababa, which directly receive a slaughter service from the abattoir.⁶ Yet, almost all of the residents practiced illegitimate backyard slaughtering

of animals in the zone. Daily about 30–50 cattle, 10–25 sheep, and 5–10 goats which originated from different areas of the zone were slaughtered in the abattoir. In the town municipal abattoir, ante-mortem and post-mortem inspection was carried out by veterinarians, whereas the rest of the activities were performed by technicians.

Population

All cattle in the abattoir ready for slaughtering were the source of the population. The target study population were comprised of all ages and sexes of beef cattle slaughtered in Jimma town municipal abattoir and those meat cutters in the butcher houses of the town, and beef meat consumers.

Study Design

A cross-sectional study supported by questionnaire survey was carried out to investigate the carriage of Salmonella isolates and their antimicrobial resistance patterns and to assess the knowledge gaps of abattoir workers and meat handlers. Carcass samples from municipal abattoir and retailer (butcher) shops were selected by simple random sampling. Microbiological sample analysis was undertaken to isolate and characterize Salmonella spp. as per standards set in ISO-17604.¹²

Information pertaining to demographic data (educational status and sex), predisposing factors for salmonellosis such as hygiene (hand washing, knives, and equipment), knowledge and attitude of abattoir workers, retailers and consumer of raw beef were collected by using a pretested, structured, and self-administered questionnaire.

Sample Size Determination

A single population proportion formula developed by Thrusfield¹³ was used to determine the sample size to be collected from the abattoir and retailer (butcher) shops.

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

where 1.96² is the *z* statistic for level of confidence, *n* is the required sample size, *P_{exp}* is the expected prevalence, and *d* is the desired absolute precision.

Sample size calculation was based on the pooled expected prevalence of 4.53% beef carcass from the abattoir in different parts of the country.¹⁴ Hence, by using 4.53% as expected prevalence, the minimum calculated sample size was 67.

However, in order to increase the precision of the estimate, the sample size was inflated by 5% so 70 samples (carcass, workers hand, and knife samples) were collected from the abattoir.

The sample size to be collected from the retailer (butcher) shops was 118 using pooled expected prevalence of 8.34%.¹⁴ Accordingly, the sample size was inflated by 5% so as to increase the precision of the estimate, the sample size for retailer (butcher) shops was 124. Nevertheless, there are only 74 retailer (butcher) shops in Jimma town which were lower than the calculated sample size for the study. In order to solve this inconsistency, finite population correction factor (FPC factor) formula was used to adjust the sample size to the population size. Since the original required sample size was 124, adjustment of the sample size to the population size was made as per the correction formula and the sample size was calculated as follows.

$$n_a = \frac{n^r}{1 + \frac{n^r - 1}{N}}$$

$$n_a = \frac{124}{1 + \frac{124-1}{74}} = \frac{124}{1 + \frac{123}{74}} = \frac{124}{1 + 1.7} = \frac{124}{2.7} = 46$$

Adjusted sample size for retailer (butcher) shops was 46, where n_a is the adjusted sample size, n^r is the original required sample size, and N is the population size.¹⁵

Sampling Strategy and Salmonella Isolation

A total of 210 municipal abattoir swab samples (carcass, $n=70$; workers hand, $n=70$; and knife, $n=70$) and 138 retailer (butcher) shops swab samples (carcass (meat for sale), $n=46$; workers hand, $n=46$; and chopping board, $n=46$) were collected for conventional bacteriological culturing.

About 100 cm² of beef carcass surface around the neck, brisket, fore rib, flank, and rump was swabbed by wiping the cotton swabs on each sampling site, over the delineated area horizontally and then vertically several times, whereas meat cutting equipment (knives) and workers hand in contact with the meat in the abattoir were swabbed from a 15–20 m² surface of the knives and workers hand using a sterile surgical glove. Then, the samples were kept in separate sterile plastic bags (Seward, England) to prevent spilling and cross-contamination.

Butcher (retailer) shops swab samples were collected from the surface of the carcass, meat chopping boards, and

hands of the meat cutter. The swab cotton was moistened with sterile buffered peptone water (BPW) (Oxoid) and rubbed firmly across the exposed area of the sample source several times in all directions and introduced into a test tube containing 10 mL diluents of BPW and shaken vigorously.¹⁶ Finally, all samples were labeled for identification, kept in an ice-box at 4°C and 1 hour after collection, transported to the Veterinary Microbiology Laboratory of School of Veterinary Medicine of Jimma University. Detection and phenotypic characterization of Salmonella isolates from raw beef sample was performed according to the standard culture technique (ISO-6579).¹² And then, briefly, the following isolation activities were carried out.

Pre-Enrichment and Selective Enrichment

For isolation and identification of Salmonella spp. the swab samples were pre-enriched in an appropriate amount of BPW and incubated at 37°C for 24 hours. One hundred microliters of the pre-enriched culture was transferred to 10 mL of Rappaport-Vassiliadis Enrichment Broth (RVB), (Oxoid) and incubated for 24 hours at 42°C. Another 1 mL of the suspension was also transferred to 10 mL of Selenite F broth (SFB) (Oxoid) and incubated for 24 hours at 37°C.

Plating Out and Identification

A loop full of inoculums from each RVB and SFB cultures were plated onto Xylose lysine desoxycholate (XLD) (Oxoid) and Brilliant green (BGA) (Oxoid) agar plates and incubated at 37°C for 24 hours for identification. Characteristic red colonies with black centers on XLD and pink colonies on BGA plates were examined and considered as presumptive Salmonella colonies. Five typical colonies from both XLD and BGA were streaked onto the surface of pre-dried nutrient agar plates (Oxoid) and incubated at 37°C for 24 hours.

Then, typical colonies from nutrient agar were inoculated into the following biochemical tests for further identification as per ISO-6579 guidelines¹² and incubated for 24 hours at 37°C. The biochemical tests conducted include triple sugar iron (TSI) agar (Hi Media), lysine iron agar (Hi Media), Simmon's citrate agar (Hi Media), urea broth (Hi Media), and indole reaction (motility indole ornithine, MIO) medium (Pronadisa)

Antimicrobial Resistance Pattern Testing

Salmonella isolates susceptibility to a panel of antimicrobials were performed according to the Clinical and Laboratory Standards Institute (CLSI) guidelines¹⁷ using

Kirby–Bauer disk diffusion method on Muller–Hinton agar plates (Oxoid). Antimicrobial disks used in this study were all from Sensi-Discs, Becton, Dickinson and Company and they were the following, with their disc potencies (μg): ampicillin (10), cefoxitin (30), chloramphenicol (30), ciprofloxacin (5), gentamicin (10), kanamycin (30), nalidixic acid (30), neomycin (30), norfloxacin (10), streptomycin (10), trimethoprim (5), and tetracycline (30). The quality control organism used to carry out the antimicrobial susceptibility test was *Escherichia coli* ATCC 25922 and the interpretation of the susceptibility test result was based on the CLSI guidelines.¹⁸

Data Analysis

Microbiological and questionnaire survey data were analyzed using SPSS version 20 software. The prevalence of *Salmonella* isolates in raw beef of abattoir and retailer (butcher) shops was calculated by dividing the frequency of positive samples by the total number of samples examined. A Chi-square Fisher exact test was used to compare the occurrence of *Salmonella* in both abattoir and retailer (butcher) shops of the study setting. Data from the questionnaire were used to assess the association of the risk factors with *Salmonella* identification. A univariate analysis (Chi-square Fisher exact test) was conducted using the *Salmonella* status of the abattoir and butcher (retailer) shops as the outcome variable. A multivariable analysis was performed to relate the likely risk factors to *Salmonella* outcomes (presence or absence of the isolate) in samples and abattoir or butcher shops. Finally, associations were reported as being statistically significant whenever the *p*-value was <0.05 .

Results

Identification of *Salmonella* by Conventional Culture

In the current study, out of 210 and 138 swab samples from the abattoir and retailer (butcher) shops, correspondingly, the overall prevalence of *Salmonella* positive swab samples from the abattoir was 11.43% (24/210), whilst it was 6.52% (9/138) from butcher shops. Of the abattoir samples, the highest were obtained from carcasses compared to the isolates obtained from knives (11.43%, 8/70) and workers hands (8.57%, 6/70), whereas out of the nine isolates from retailer (butcher) shops, 8.7% (4/46), 6.52% (3/46), and 4.35% (2/46) were recovered from the carcass, chopping boards, and workers hands, respectively, as depicted in Table 1.

Risk Factors Revealing Beef Contamination by *Salmonella* in Municipal Abattoir

A binary logistic regression analysis revealed that educational status (OR=8.4, 95% CI=1.186–59.493), job related training (OR=5.5, 95% CI=1.065–28.416), contamination risk perception (OR=5.31, 95% CI=1.256–22.489), neatness of knives (OR=7.6, 95% CI=0.892–65.376), and source of contamination (OR=8.44, 95% CI=1.682–42.39) were among the potential risk factors associated with *Salmonella* occurrence in the abattoir, as illustrated in Table 2.

Putative Risk Factors Related with Beef Contamination by *Salmonella* in Butcher Shops

Of the risk factors considered for the butcher shops, educational status (OR=9.17; 95% CI=1.15–73.239),

Table 1 The Overall Prevalence of *Salmonella* Isolated from Abattoir and Butcher Shops in the Study Setting

Sample Source	Sample Location	Sample, N	Positive Sample, N (%)	95% CI
Abattoir	Carcass	70	10 (14.29)	7.07–24.71
	Worker's hand	70	6 (8.57)	3.21–17.73
	Knife	70	8 (11.43)	5.02–21.28
	Total	210	24 (11.43)	7.46–16.53
Butcher shop	Carcass	46	4 (8.70)	2.42–20.79
	Worker's hand	46	2 (4.35)	0.53–14.84
	Chopping board	46	3 (6.52)	1.37–17.90
	Total	138	9 (6.52)	3.03–12.02

Abbreviation: CI, confidence interval.

Table 2 Putative Exposing Risk Factors to Beef Contamination by Salmonella in Jimma Abattoir Workers

Factors	Category	Sample, N	Positive Sample, N (%)	OR (95% CI)	p-value*
Educational status	Uneducated	8	5 (62.5)	8.40 (1.186–59.493)	0.049
	Primary school	28	6 (21.43)	-	
	Secondary school	13	2 (15.38)	R	
Job related training	No	29	11 (37.93)	5.50 (1.065–28.416)	0.047
	Yes	20	2 (10.00)	R	
Contamination risk perception	No	11	6 (54.55)	5.31 (1.256–22.489)	0.047
	Yes	38	7 (18.42)	R	
Clean clothing	No	39	11 (28.21)	1.57 (0.287–8.595)	0.709
	Yes	10	2 (20.00)	R	
Neatness of knives	No	34	12 (35.29)	7.6 (0.892–65.376)	0.043
	Yes	15	1 (6.67)	R	
Source of contamination	Fecal	14	8 (57.14)	8.44 (1.682–42.39)	0.014
	Handling with dirty equipment and hand	13	2 (15.38)	1.15 (0.166–7.990)	
	Water and floor	22	3 (13.64)	R	
Proper placing of equipment	No	8	1 (12.50)	2.90 (0.321–26.158)	0.663
	Yes	41	12 (29.27)	R	

Notes: *Pearson Chi² and Fisher exact test.

Abbreviations: OR, odds ratio; R, reference.

wearing of protective cloth (OR=11.42; 95% CI=1.829–71.419), manner of hand washing (OR=7.25; 95% CI=1.210–43.442), and money handling (OR=9.69; 95% CI=1.578–59.474) were among the putative risk factors significantly associated with the occurrence of Salmonella in a contaminated carcass, as designated in Table 3.

Antimicrobial Resistance Patterns Mono Antimicrobial Resistance Patterns of Salmonella Isolates

In the present study, antimicrobial resistance patterns of 33 Salmonella isolates recovered from raw beef were tested against 12 antimicrobials so as to evaluate their resistance level (Table 4). Out of 24 isolates from abattoir raw beef,

Table 3 Putative Risk Factors Associated with the Incidence of Salmonella in Butcher Shop Workers

Factors	Category	Sample, N	Positive Sample, N (%)	OR (95% CI)	p-value
Educational Status	Uneducated	9	4 (44.44)	9.17(1.15–73.239)	0.037
	Primary	14	1 (7.14)	-	
	Secondary	23	2 (8.70)	R	
Wearing protective cloth	Not used	12	5 (41.67)	11.42(1.829–71.419)	0.009
	Used	34	2 (5.88)	R	
Manner of hand washing	Rinsing with water only	15	5 (33.3)	7.25(1.210–43.442)	0.029
	Using detergent and water	31	2 (6.45)	R	
Handling of money	Butcher with hand	14	5 (35.71)	9.69(1.578–59.474)	0.014
	Cashier	32	2 (6.25)	R	

Table 4 Mono-Antimicrobial Resistant Patterns of Salmonella Isolates from Raw Beef in Jimma

Antibiotics	Resistant (%)		Intermediate (%)		Susceptible (%)	
	Abattoir	Butcher	Abattoir	Butcher	Abattoir	Butcher
Tetracycline	14 (58.3)	5 (55.6)	3 (12.5)	1 (11.1)	7 (29.2)	3 (33.3)
Streptomycin	16 (66.7)	6 (66.7)	3 (12.5)	1 (11.1)	5 (20.8)	2 (22.3)
Trimethoprim	2 (8.33)	2 (22.2)	0	0	22 (91.67)	7 (77.8)
Chloramphenicol	4 (16.7)	2 (22.2)	7 (29.2)	2 (22.2)	13 (54.2)	5 (55.6)
Kanamycin	4 (16.67)	3 (33.3)	7 (29.2)	0	13 (54.6)	6 (66.7)
Norfloxacin	0	0	0	0	24 (100)	9 (100)
Gentamicin	0	0	0	0	24 (100)	9 (100)
Ampicillin	14 (58.3)	5 (55.6)	4 (16.67)	1 (11.1)	6 (25)	3 (33.3)
Ciprofloxacin	0	0	0	0	24 (100)	9 (100)
Cefoxitin	2 (8.33)	1 (11.1)	3 (12.5)	1 (11.1)	19 (79.2)	7 (66.7)
Neomycin	5 (20.8)	3 (33.3)	7 (29.2)	1 (11.1)	12 (50)	5 (55.6)
Nalidixic acid	2 (8.33)	2 (22.2)	7 (29.2)	2 (22.2)	15 (62.5)	5 (55.6)

14 (58.3%), 14 (58.3%), and 16 (66.7%) of them were resilient to tetracycline, ampicillin, and streptomycin, correspondingly, whereas no isolate was resistant to norfloxacin, gentamicin, and ciprofloxacin. Besides, of the nine Salmonella isolates from retailer (butcher) shops, 5 (55.6%), 5 (55.6%), and 6 (66.7%) of them were resistant to tetracycline, ampicillin, and streptomycin, respectively, while no isolate was resistant to norfloxacin, gentamicin, and ciprofloxacin. A higher intermediate resistance of the isolates to streptomycin (66.7%) was observed compared to the rest of the antibiotics, as designated in Table 4.

Multi-Drug Resistance Patterns of Salmonella Isolates

Out of 24 isolates obtained from the abattoir, 14 (58.3%) were resistant to two or more antibiotics. Among the 14 multi-drug resistant isolates, five (35.7%) were resistant to tetracycline, ampicillin, and streptomycin, whereas two were resistant to tetracycline, streptomycin, chloramphenicol, kanamycin, ampicillin, and neomycin. Moreover, only two isolates from retailer (butcher) shops were resistant to five different antibiotics, as depicted in Table 5.

Discussion

In the current study, the carriage and antimicrobial resistance pattern of Salmonella isolates from raw beef in Jimma town abattoir and retailer (butcher) shops were estimated. Consequently, Salmonella isolates were recovered from 24 (20.2%) and nine (6.52%) swab samples of abattoir and retailer (butcher) shops, respectively. The frequency of Salmonella isolated from the abattoir is consistent with the report of Sibhat et al,¹⁹ who reported 10.9% from a commercial Slaughterhouse in Debre Zeit,

Table 5 Multiple Antimicrobial Resistance Profile of Salmonella Isolates from Raw Beef in Jimma

No. of Antibiotics	Antimicrobials Shown Resistance (No. of Isolates)	No. of Isolates (%)
	Resistance Antimicrobials to Abattoir Isolates	14 (58.3%)
Two	TE, AMP	1
Three	TE, S, AMP	5
Four	TE, S, AMP, CXT	1
Five	TE, S, AMP, CXT, NA	1
	TE, S, C, K, AMP	1
Six	TE, S, C, K, AMP, Neo	2
	TE, S, AMP, Neo, NA	1
	TE, S, AMP, CXT, Neo, NA	1
Seven	TE, S, W, C, K, AMP, Neo	1
	Resistance antimicrobials to butcher shop isolates	6 (66.7%)
Two	TE, S	1
Four	TE, K, AMP, Neo	1
Five	C, AMP, CXT, Neo, NA	1
	TE, S, W, K, AMP	2
Six	TE, S, C, AMP, Neo, NA	1
Total		20 (60.61%)

Abbreviations: TE, tetracycline; AMP, ampicillin; S, streptomycin; CXT, cefoxitin; NA, nalidixic acid; K, kanamycin; Neo, neomycin; W, trimethoprim; C, chloramphenicol.

whereas Beyene et al²⁰ reported a slightly comparable result from Asella abattoir (8.5%). The current finding is higher than the report of Shilangale et al,²¹ who reported

a 0.85% incidence of *Salmonella* from the three beef export abattoirs in Namibia and the 1.5% isolation rate reported by Ibrahim²² from cattle carcasses at Kano abattoir in Nigeria. Contrarily, this finding is lower than the reports of Wassie et al,²³ Hiko et al,²⁴ and Amenu²⁵ from Gonder (20.8%), Addis Ababa (26.6%), and Arbaminch (31.5%) municipal abattoirs, respectively.

In our observation, the abattoir was extremely poor in sanitation due to the absence of water and blood drainage, and the accumulation of waste materials which were disposed of near to the slaughtering house. Therefore, the difference in the frequency of *Salmonella* isolation rate in the present study from different authors work could be attributed to variation in sampling strategy, detection procedures, target populations, topographical origins of the animals, numbers of animals sampled, study design, season, hygienic status of the abattoir and retailer shops, and antimicrobial treatment warranted during the process. This might be due to the poor hygiene to the abattoir, which is consistent with the report of Kusumaningrum et al,²⁶ who stated that poor hygienic conditions are the source of carcass contamination in the abattoir.

The current study finding depicted that about 6.52% of *Salmonella* spp. was isolated from butcher shops in the study setting. This finding is consistent with the report of Mengistu et al²⁷ from Haramaya University, eastern Ethiopia. Contrarily, a relatively higher prevalence of *Salmonella* isolates recorded in the current investigation was higher than the report of Mengistu et al²⁷ from Dire Dawa retailer shops (1%), revealing that the course of evisceration might be the basis for carcass uncleanness besides carrier state. However, this result was lower than the incidence of 12% from raw meat²⁸ and 17.3% from minced meat²⁹ in Gondar town. This difference might be due to poor personnel hygiene in the butcher houses and equipment (viz. knives, table, and balance) (Personal observation).

The proportion of *Salmonella* isolates, in the current study, recovered from swabs of carcasses of abattoir origin was 14.29%. This finding corroborates with the findings of Takele et al⁶ and Dabassa and Bacha³⁰ who reported 11.3% and 13.3% from beef carcasses in Jimma municipal abattoir, respectively, and Hiko et al²⁴ who reported 11.8% from Addis Ababa Abattoirs Enterprise whereas this prevalence is lower than other reports from Tigray region (16.4%),³¹ Dire Dawa abattoir (17.7%),³² and Senegal (42.8%).³³ Counter to this, the current finding is higher than the report of El-Gamal and EL-Bahi,³⁴

Renatus et al,³⁵ Thongsay et al,³⁶ Kalambhe et al,³⁷ Bahnass et al,³⁸ Sefinew and Bayleyegn,³⁹ and Gizachew and Mulugeta⁴⁰ from Egypt (0.0%), Namibia (0.50%), Thailand (4.5%), Central India (6%), Saudi Arabia (8.5%), and Bahir Dar (4.8, 7.6%), respectively. This might be due to variation in the nature of samples, sampling strategy, and procedures origin of animals, contamination from intestinal tract breakage and fecal leakage during evisceration, and from lairage due to lack of care in the study setting.

Salmonella isolates detected from abattoir workers hand swab (8.57%) of the present study setting is higher than the reports of Bahnass et al³⁸ and Nyeleti et al⁴¹ in Najran region (2.4%) and Addis Ababa (6.0%), respectively. Personal sanitation disparity of the food handlers could help to elucidate this inconsistency since the majority of workers in the present study setting handled rumen content and gastrointestinal tracts without washing their hands.

In the current investigation, the prevalence of *Salmonella* isolates detected on the cutting knife (11.4%) was slightly higher than the reports of Teklu and Nigusie,⁴² and Garedeew et al²⁹ from Mojo (7.4%) and Gondar (9.1%) towns, respectively. This variation is attributed to a high frequency of *Salmonella* in raw beef samples which might act as a cause of contamination for the knives, and due to the unhygienic condition of knives, viz. abattoir workers in the study setting put knives on the floor and then use them without washing or disinfecting.

The occurrence of *Salmonella* on the butcher shops cutting (chopping) boards was 6.5%. This finding is almost comparable to the 5.6% and 5.7% prevalence rates which were reported by Garedeew et al²⁹ and Wassie et al²³ from chopping boards of butcher shops and an abattoir in Gonder town, respectively. A comparatively lower incidence rate of *Salmonella* isolates from cutting boards than that of carcasses could be ascribed to the hygienic condition of the chopping board. Reports revealed that bacteria such as *Salmonella* were not isolated from wooden surfaces soon after they were applied, unless huge numbers were used. Studies revealed that wood is intrinsically porous, which permits food juices and bacteria to enter the body of the wood unless a highly hydrophobic filtrate covers the surface.⁴³

Of the risk factors considered in the current study, the educational level of both abattoir and butcher shop workers was positively associated with the occurrence of *Salmonella*. The probability of carcass contamination by

uneducated workers was 8- and 9-times more likely than educated workers in the abattoir and retailer shops, respectively. This finding is consistent with the report of Ntanga et al⁴⁴ in Tanzania which revealing that the educational level of workers was the risk factor for positive results. The educational level and training of food handlers are important for basic perception and necessities of personal hygiene and its environment play important roles in preserving the quality of food products to consumers.⁴⁵ Best commencement of hygiene practices has been attributed to those employees with a basic level (at least primary) of education, while bad practices are attributed to those who were illiterate.⁴⁶

Salmonella infection was more prevalent in untrained abattoir workers (37.93%) compared to the trained ones (10%). This study revealed that job related training was significantly associated with Salmonella carriage ($p<0.05$). Untrained personnel working in the abattoir were 6-times more likely to contaminate the carcasses with Salmonella than trained personnel.

The current finding corroborates the report of Niyonzima et al,⁴⁷ who reported that job related training is found to be positively associated ($p<0.05$) with a reduced risk of Salmonella incidence in retailed meat in Rwanda. Job-related training of food handlers regarding basic concepts and requirements of personal hygiene play an integral part in ensuring safe products to the consumers.⁴⁸ Our result indicated there was a poor knowledge on meat hygiene practice in both abattoir and retailer shop workers of the town. Hence, there is a need for more operative training in both individual and general hygiene practices for these workers. Pertaining to contamination risk perception, a higher frequency rate of Salmonella was isolated from personnel (54.55%) who had a lack of perception on contamination of carcasses in the slaughtering process as a risk. This result revealed that personnel who had a lack of knowledge on contamination in the slaughtering process were significantly associated ($p<0.05$) with Salmonella isolation. This in turn corroborates the report of Haileselassie et al⁴⁸ from north-central Nigeria.

Lack of wearing personal protective cloth during the handling of meat was insignificantly associated with Salmonella occurrence in the abattoir workers of the current study. Though insignificantly associated, a higher prevalence of Salmonella isolates was obtained from personnel who did not use clean personal protective clothing.

This finding agrees with that of Alhaji and Baiwa,⁴⁹ who reported that a substantial proportion of the workers did not follow the recommended personal protective practices in Niger state of north-central Nigeria. Though many studies identified bare hands, dirty clothes, and workers hair could be the sources of microbial contaminating the meat,^{50–52} in our current work, abattoir workers had no awareness about the importance of wearing clean protective cloths which may prevent the transmission of microorganisms from handlers to meat.

Using an uncleaned knife for splitting of carcasses was also significant with the infection of meat by Salmonella in the abattoir ($p<0.05$). The chance of contamination of the carcass by Salmonella was 8-times more likely when using dirty knives than clean knives. Our finding corroborates with the report of Muluneh and Kibret⁴⁰ in Bahir Dar town, who reported washing the knife before beginning slaughtering reduces the risk of contamination by Salmonella. A higher bacterial load detected on raw beef from the slaughterhouses could be the possibility of cross-contamination associated to the use of uncleaned knives.⁴⁷

Feces as a source of contamination was positively associated with the Salmonella isolation rate from the carcass ($p<0.05$) in the current study. Fecal meat contamination of Salmonella was 8-times more likely to contribute to the contamination than floor and water contamination to the carcass. On top of this, handling of carcasses with dirty equipment and hand had one more chance of contaminating of carcasses than water and floor at Jimma municipal abattoir. This result is inconsistent with the previous reports from Sudan by Abdalla et al.⁵³

Microbial contaminations of meat carcasses by hides and intestinal contents are occurred mainly during hide removal and the evisceration process.¹⁹ In the current study setting, the esophagus and rectum were not ligated during the evisceration process, which could be attributed to high microbial contamination of raw beef.

Lack of wearing personal protective cloth during handling of meat in the current investigation was also significantly correlated to Salmonella incidence ($p<0.05$) in the butcher shop workers. This outcome agrees with the work of Chepkemioi et al,⁵⁴ which revealed that butcher shop workers who did not wear protective clothes had a risk factor for carcass contamination. Besides this, the current study observed that only about 5.88% of butcher shop workers wore clean protective clothes, which corroborates with the report of Garedew et al.²⁹ This could be attributed to a lack of knowledge about good sanitary practices as

none of the butchers had taken formal training in food safety in the study site.

In this study, hand washing before handling meat by the butcher men respondents was statistically associated with *Salmonella* occurrence ($p < 0.05$). Workers washing their hands with water only were contaminating the carcass and equipment 7-times more than those washing their hands with detergent and water. This result agrees with the report of Ntanga et al⁴⁴ from Tanzania. In the current study, we observed that butcher shop workers did not wash their hands mostly due to lack of hand-washing facilities, washing basin, soap, etc., as reported by Todd et al.⁵⁵

Meat cutters in butcher shops handling money with their hands while selling meat was significantly contributing to the contamination of *Salmonella* ($p < 0.05$). The isolation rate of *Salmonella* in the current work revealed that butcher men collecting money with their hands during selling were 10-times more likely contaminating carcasses than those collecting money using a cashier. This finding closely corroborates with the report of Garedew et al,²⁹ who reported almost all shops workers collecting money with bare hands while serving meat. The unhygienic conditions and habits of handling money in circulation usually subjects the money to contamination with a variety of microorganisms. The money can thereafter act as a vehicle for contaminating the hands of the food seller/handler, and thus cross-contamination of food.⁵⁶

Our current study finding revealed that all *Salmonella* isolates recovered from both abattoir and butcher shops were sensitive to norfloxacin, gentamicin, and ciprofloxacin, which is comparable to a study conducted in Jimma town.⁵⁷ This might be because they are not widely used in Ethiopia for animal treatment, and those resistance drugs can easily available for both treatment of animal.

The resistance level of the abattoir and butcher shop *Salmonella* isolates to streptomycin in the current study was higher than the resistance reports of Wolde and Bacha⁵⁷ in Jimma town (50.8%). Contrarily, a highest resistance (100%) level of *Salmonella* isolates to streptomycin was reported by Kebede et al⁵⁸ in slaughtered bovines and ovines at Addis Ababa abattoir Enterprise. Detection of AR *Salmonella* could be attributed to recurrent usage of antibiotics in both livestock and public health sectors as these antimicrobials are comparatively cheaper and frequently available.

The higher number of tetracycline resistant *Salmonella* isolates (58.3%) recorded in the present work is closely comparable with the finding of Madoroba et al,⁵⁹ who

reported 51.9% resistance in a South African abattoir, whereas about 55.6% tetracycline resistant isolates were obtained from butcher shops in this finding, which agrees with the finding of Eguale et al⁶⁰ in Addis Ababa (58.3%). Contrary to this, a higher resistance (75%) to tetracycline was reported by Worku et al⁶¹ in food handlers in Jimma town. This higher resistance profile of *Salmonella* isolates to tetracycline might be attributed to the high level of utilization of this drug in veterinary medicines by the local community in the current study setting.

Salmonella isolates resistance to ampicillin (58.3%), from the abattoir, recorded in the current study is similar with the report of Beyene et al²⁰ in Assella Abattoir (58.3%), whereas the prevalence of resistant isolates from the butcher shops (55.6%) in our work is higher than the report of Garedew et al²⁹ in retailer shops in Gondar town (88.7%). The differences could be due to frequent use and easy accessibility of the drug everywhere in the country including the current research site.

The occurrence of multidrug-resistant *Salmonella* isolates in the abattoir and butcher shops sample was observed in this study and a higher frequency of the isolates were recovered from both abattoir (58.3%) and butcher shops (66.7%), of which were resistant to two or more of the antimicrobials. This result was lower than studies carried out in Addis Ababa and Gondar towns where 83% and 75.5%, respectively, of isolates were resistant to two or more antimicrobials.^{29,62}

Nevertheless, the current finding is relatively higher than the report of Beyene et al²⁰ from dairy farms, abattoir, and in contact humans of Asella town, in which about 50% of *Salmonella* isolates were resistant to two or more antimicrobials. A higher level of resistance could be attributed to irrational use of antibiotics in our study setting, as previously reported.^{63–65} In general, these findings call for constant monitoring and evaluation of antimicrobials stewardship.

Conclusion

The isolation proportion of *Salmonella* from Jimma town abattoir and butcher shops was 11.43% and 6.52%, respectively. Educational status, job related training, perception of contamination risk, neatness of knife, wearing protective cloth, manner of hand washing, handling of money, and fecal contamination were amid the risk factors contributing to *Salmonella* raw beef contamination in the current study. A higher mono antimicrobial resistance rate of *Salmonella* isolates was observed against

tetracycline, ampicillin, and streptomycin amid the antibiotics considered in the study. Besides, multi-drug resistance (two or more antibiotics) was registered for 14 (58.3%) and six (66.7%) isolates of the abattoir and retailer (butcher) shops, respectively. These drugs are popular and usually used in the veterinary and human medicines in Ethiopia. This could attribute to the limit of therapeutic choice to prevent and control salmonellosis and other bacterial diseases in livestock and human health. Lack of public awareness about *Salmonella* contamination of beef meat in abattoir and butcher shops, and its transmission to humans, was found to be high in the study area.

Moreover, it is mandatory for the concerned authority to strictly and consistently provide training on keeping personal, meat, working equipment, and environmental hygiene requirements so as to decrease the level of contamination of raw beef by abattoir and butcher shop workers. More comprehensive training has to be given on proper antimicrobial usage for the concerned stakeholders and further studies are needed to describe all the virulence gene and serotype of pathogenic *Salmonella* strain for the emergence of drug resistance isolates in order to develop the best prevention and control measures.

Data Sharing Statement

The data accustomed to support the findings of this research work are offered from the first and/or corresponding author upon request.

Ethics Approval and Consent to Participate

Abattoir workers and owners of beef butcher shops, which were involved in the business of beef meat, were informed about the purpose of the study and the consent of each abattoir worker, and owners and workers of each beef butcher shops were obtained before physical examination of raw beef and collection of the sample. Only volunteer abattoir workers and owners' and workers of the butcher shops have participated in the study. The questionnaire and methodology for this study was approved by the research and ethical review committee of Jimma University and the study was conducted in accordance with the Declaration of Helsinki.

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Author Contributions

Both authors made a significant contribution to the work reported, either in the conception of the study, study design, execution, acquisition of data, analysis, and interpretation, or in all these areas. Moreover, both authors took part in revising or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

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Disclosure

The authors declare that they have no competing interests.

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