HYGIENE HANDLING PRACTICES, PREVALENCE AND ANTIMICROBIAL SENSITIVITY TESTING OF S. AURES ISOLATED FROM DIFFERENT CRITICAL POINT OF BEEF SUPPLY CHAIN ON SLAUGHTER PREMISE AND BUTCHER SHOP AT MENGE DISTRICT, BENISHANGUL GUMUZ REGIONAL STATE, WESTERN ETHIOPIA

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ABSTRACT: Across-sectional study was conducted on Assessment of hygienic handling practice, prevalence and antimicrobial sensitivity testing of *S. aureus* isolated from different critical point of beef supply chain in Abattoir and Butcher shop retail meat at Menge district, western Ethiopia from October 2024 to April 2025**,** with the objectives to isolate and identify *S.aureus* from critical control points of Abattoir and Butcher shops supply chain, to assess the public health significance associated with risk factors and to estimate antimicrobial susceptibility patterns of S.aureus isolates. A total of 384 samples were collected from Abattoir (N=240) and Butcher shop (N=144) processed bacteriologically and the isolate were tested with a number of biochemical tests for confirmation and identification of *S. aureus.* The study revealed that 34.11 % of the collected swab samples were predominant contaminants of *S. aureus*. Higher (50%) *S.aureus* contaminates were reported in Butcher swab followed by (24.58%) abattoir samples, with butcher contaminants 1.06 times more likely harbor the *S.aureus* infection (P<0.05). In this study, there was in-significant association (p>0.05) among the age, body conditions, hygiene and breed of the cattle with the isolates of S.aureus. In other way, meat handling practices, waste disposal practices, sample types, and *S.aureus* risk factors was significant (P<0.05). Majority (100%) of drug resistance prevalence was reported in penicillin G, followed by (84.61%) Amoxicillin; 64.1% cefoxitin and 56.41% sulphonamides. Whereas higher (100%) of drug susceptible was recorded in (ciprofloxacin), followed by 92.30% Gentamycin, 89.74% in (chloramphenicol); 50% kanamycin, 45% streptomycin, 40% vancomycin and 32.5% chloramphenicol. The presence and consumption of raw meat may constitute a public health hazard and reduced meat quality due to *S. aureus*. Thus health professionals should create awareness about meat handling practice, storage and loading and unloading process to Abattoir owners, butchers' and to its value chain. Therefore, regular resistance follow-up, using antimicrobials sensitivity tests helps to select effective antibiotics and reduce the problems of drug resistance developments towards commonly used antimicrobials. Besides this, there should be hygienic practices at each critical control points of beef supply chain, and sanitation during slaughtering operation, marketing and transportation of animals in Menge District.

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1. INTRODUCTION

Background

In developing countries, food-borne infections constitute the major cause of sickness and death. Food-related illnesses are caused by changes in eating patterns, mass catering, and improper food true in developing countries like Ethiopia, where the production of meat and meat products often occurs under unsanitary conditions and the consumption of raw meat is common. Meat is the most valuable livestock product and for many people serves as their first choice from animal protein which provides all the essential amino acids and various micronutrients in proper proportion to the human beings. This includes all processed or manufactured products prepared from animal tissues (Sebsibe *et al*., 2007).

However, apart from its nutritional and health benefits, meat can be a source of both chronic (Wyness, 2016) and infectious diseases (Battaglia *et al*., 2015). Foodborne diseases occur in developing countries because of the poor food handling and sanitation problems. Although animal's tissue is sterile, during slaughtering, microorganisms could contaminate the tissue primarily from the exterior or the interior environments (A. M. Goja et al., 2013); (T. Beyene et al., 2017). Even though modern technologies are advanced for safe meat production, the safety of meat processing in developing countries including Ethiopia is still a concern. Except in the big cities of Ethiopia, the animals are slaughtered locally in open areas without any hygienic prerequisites (Gutema et al., 2021). As a result, the chance of the meat being contaminated with pathogenic microbes is exceedingly high. The contamination of meat occurs during the removal of hides, evisceration, processing, packaging and storage, and distribution at slaughterhouses and retail outlets (Lonergan et al., 2019).

Staphylococcus aureus is a Gram-positive, facultative anaerobic bacterium with size 0.5- 1.0μm in diameter which grows individually, in pairs, short chains or grape-like clusters (Schleifer KH et al., 2009). The bacterium is catalase and coagulase positive, oxidase-negative, non-motile microorganism that does not form spores. On agar medium it creates smooth, convex, glistening, circular colonies with entire margins reaching a size of 6-8mm in diameter (Le Loir Y et al., 2003); (Medveďová, 2012). They are resistant to desiccation and can survive on different surfaces, and resist sanitation, forming biofilms (Moretro T, 2017).

S. aureus is able to tolerate pH ranges from 4.5 to 9.0 and NaCl concentrations up to 9% and can grow and express virulence in a wide range of environmental conditions (Le Loir Y et al., 2003). *Staphylococcus aureus* is one of the most common foodborne pathogens causing intoxication. Staphylococci can be introduced in the environment of food processing installations through various routes, such as raw materials, food handlers, or poor hygiene in food processing equipment. It is one of the food borne diseases transmitted from the contaminated animal source foodstaffs. (S. Nouichi and T. M. Hamdi, 2009). It produces heat stable and proteolytic enzyme resistant enterotoxins that cause food poisoning in humans leading to vomiting, abdominal pain and diarrhea (L. Busani et al., 2006). It is found in 30% nonclinical nasal carrier population (W. Levinson, 2008).This could be the sole source of contamination in abattoir and butcher workers for those who do not have enough awareness on the nature of the disease. Billions of people are at risk every year and thousands die as a result of consuming unsafe food (Sibanyoni JJ et al., 2017).

Staphylococcus *aureus* is a foodborne pathogen which is responsible for contamination of different food products and results food spoilage, reduction of food safety and shelf life and cause foodborne poisoning via production of deadly enterotoxins. It is one of the most contaminates and prevalent causes of clinical infections globally and has garnered substantial public attention due to increasing mortality associated with multidrug resistance (Kwon NH et al., 2006).

In the last few decades, staphylococcal food poisoning has been reported as the third cause of foodborne illnesses in the world (Zadoks R, et al., 2000). The prevalence and burden of S. *aureus* is growing worldwide and it is a major concern of public health programs (Kadariya et al., 2014). However, the true incidence of staphylococcal food poisoning is underestimated due to misdiagnosis, under-reporting, improper samplecollection and laboratory examination (Argudín *et al*., 2010). The global prevalence of S. *aureus* ranges from 23.3% to 73% (Reygaert, 2013).

In Africa, there is scarce data regarding to the public health impact and burden of S. *aureus*. Considering the poor hygienic conditions during food production and processing, coupled with poor cooling facilities (Tarekgne *et al*.,2015), there is a likely of high impact of staphylococcal food poisoning in African countries like Ethiopia. The prevalence varies from 16.0% in Tunisia to 52.0% in Egypt (Falagas et al., 2013) and up to 57.8% in Ethiopia (Alebachew *et al*., 2012). In addition, the prevalence in Ethiopia from meat reaches up to 40% reported form butcher shops of Mekelle city (Endale, 2013). Ethiopian raw beef consumption habit is the potential cause of foodborne illnesses (Beyene *et al*., 2017); (G. Girmay *et al*., 2015). Raw meat is available in open-air local butchers without the cold-chain process and purchased by consumers. Meat processing at butchers is likely to contribute for the contamination of minced beef meat as compared to the carcasses (Haimanot *et a*l., 2010).

1.1. Statement of the problem

Billions of people are at risk and millions fall ill every year; many die as a result of consuming unsafe food throughout the world (WHO, 2004). In Ethiopia slaughtering of beef cattle is carried out either poorly sanitized abattoir or open field and raw meat is available in open-air local retail shops without appropriate food safety control. Despite of this eating of raw meat is common habit in Ethiopia (Tesfay Kebede et al., 2014). S. aureus which is found as normal flora on the skin and upper respiratory tract of human and animal is major contaminant of meat along value chain and causes of foodborne intoxication (Diep et al., 2006; Kadariya et al., 2014).

In addition to these, meat contaminating S. aureus had developed antimicrobial resistance against regularly used antimicrobial agents for treatment of S. aureus infection especially methicillin resistant S. aureus (MRSA) and

possibly transmitted to public by eating raw beef (Tesfaye K *et al*., 2014). However, there is dearth of information on identification and testing antimicrobial susceptibility pattern of S. aureus isolates from beef at the municipal abattoir and butcher shops in Menge District..

In spite of the fact that it is difficult to prove the role of drug resistance in bacterial contaminating food with increased clinical cases, the presence of such bacteria in food items could play a great role in the spread of antimicrobial resistance. Therefore, information should be gathered to develop an effective strategy to reduce the foodborne illness and drug resistance (J. Lin *et al*., 2009).

1.3. Basic Research Questions

- What is the prevalence of *S. aureus* in beef at butcher shops of Menge District?
- What are the potential risk factors of diseases and occurrence of *S. aureus*?
- What is status of drug susceptibility pattern of *S. aureus?*

1.4. Objectives of the study

1.4.1. General Objective

To study the Prevalence, Associated Risk Factors and Drug Resistance Pattern of *Staphylococcus aureus* in beef in butcher shops and Slaughter Premises in Menge District.

1.4.2. Specific Objectives

- To determine the prevalence of *S. aureus* in beef at butcher shops and Slaughter Premises of Menge District
- To assess associated risk factors for occurrence of *S. aureus*.
- To determine the antibiotic drug susceptibility pattern of *S.aureus*.

2. MATERIAL AND METHODS

2.1. Study Area

The study was conducted from October 2024 to May 2025 in Menge District. Menge District is one of the Woredas of Benishangul Gumuz Regional State. It is located at 10°23′and 10°37′ N latitude and 34°21′ and 34°46′ E longitude, 739 km western of Addis Ababa according to the CSA (CSA, 2015). The altitude of Menge District ranges from 1100m to over 1200 m above sea level. The area is characterized by low land plane agro-ecology which has 'kola' micro climate with land coverage of 62 km^2 areas. According to national meteorological service agency/ NMSA/ the average annual rainfall of the town is 850–1126 mm with unimodal type of rainfall that occurs between April and October (NMSA, 2014). Its mean annual temperature ranges between 22.75°C and 39°C. The estimated total human population of the town was 5500 of whom 3,017 are men and 2483 are women (CSA, 2021). Cattle population of the area was 1,300 and about 65 cattle will be ready for Slaughter every week (Source: Menge District Agricultural Office, 2022).

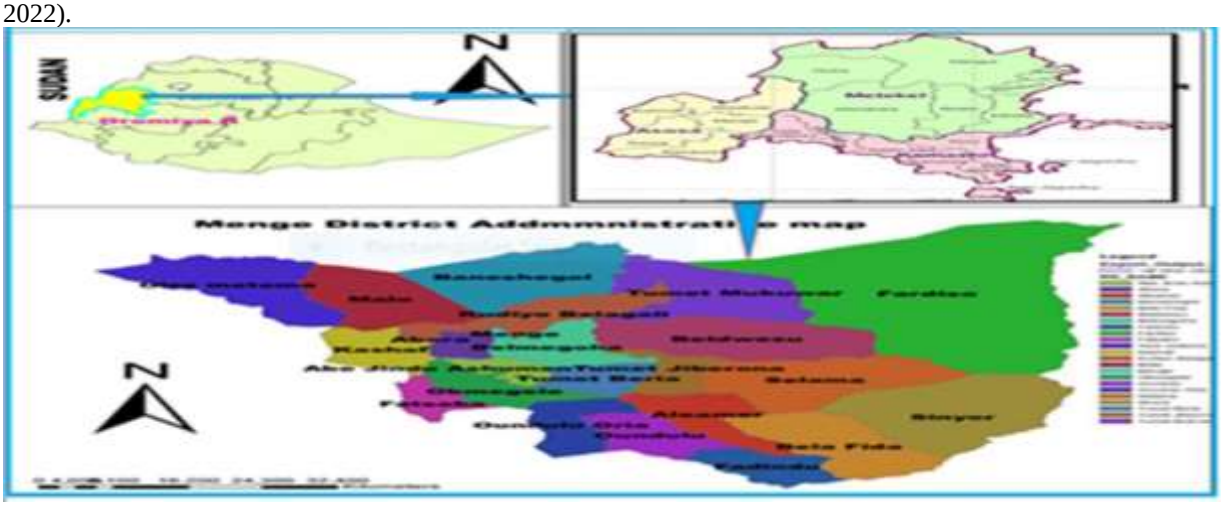


Figure 1. Menge District Map

2.2. Study design

A cross - sectional study design was conducted from November 2024 to May 2025 to determine prevalence, risk factors and drug susceptibility pattern of *S. aureus*. The sanitary conditions of the butcher shops, Slaughter Premises, the meat handler's food safety knowledge, attitudes and practices will be assessed by using personal observation and structured questionnaire survey.

2.3. Study population

The study population was beef carcasses at butcher shops, Slaughter premises, butchers, butchers' equipment and the water used for working and processing.

2.4. Sample Size Determination and Sampling Procedures

Using the Thrusfield formula (Gallina *et al*., 2015), the approximate sample size required for the investigation was determined based on the expected prevalence of *S.aureus* and desired absolute precision. The sample size for this study is determined at 95% confidence interval, 5% precision, and 50% predicted prevalence. n = (1.96)2 x P (1-P) d 2 Where: $n = the\ required\ sample\ size\ Pexp = expected\ prevalence\ d = desired\ absolute\ precision.$ Based on the above given formula, the total sample size is expected to be 384. Accordingly, the sample sizes are distributed to swab samples from slaughter house and retail shops as given in the table below.

Table 2. The amount and type of samples collected from abattoir, butcher houses and retailers.

Sampling sites	Sample type	Sample No.
At slaughter house	Nasal swab during antemortem examination	36
	Hand swab after skinning or before evisceration	36
	Knife and axe swab after skinning or before evisceration	36
	Carcass swab after skinning or flying or before evisceration	36
	Carcass swab after washing the carcass	36
	Carcass swab after evisceration	36
	Swab from transporting material like packs, vehicles.	24
At butcher shops	Carcass swab from retail shops	36
	Hand swab from workers in in retail shops	36
	Material swab from knife, axe, cutting table and carcass	36
	board and money from retail shops	36
	Total sample	384

Therefore, a total of 384 microbiological samples of swabs samples were collected and analyzed from Menge slaughter houses and meat retail/butcher shops.

2.5. Sample and Sampling technique

Simple random sampling was employed to select swabs from the butcher shops. Hence, as described above, **144 carcass swabs** were collected from butcher shops and 240 Abattoir Swab samples were collected from slaughter house. Samples were also taken from **knives, splitting axe, personal hands, working cloths, cutting boards and table, and hooks, rectal swab,** carcass swab after evisceration, washing, and skinning. For convenience, the swabs acquired from the equipment and the working environment of butcher shops were considered as samples from the immediate working environment. Convenience sampling was used to take 144 swab samples from different types of butchers' equipment. Personal observation and structured questionnaire survey was used to assess knowledge, attitudes and practices of beef handlers /butchers/ and sanitary conditions of butcher shops at Menge District.

2.6. Sample collection procedure and Transportation

The samples was collected twice per month from the shops, Slaughter Premises and the equipment. The selected carcasses was swabbed using the method described in ISO 6888-2 (NCCLS, 2012) by placing sterile template (10 × 10 cm) on specific sites of a carcass. Using a sterile cotton tipped swab (2 × 3 cm) fitted with shaft and first soaked in an approximately 10 ml of buffered peptone water (Oxoid Ltd., Hampshire, England) was rubbed horizontally and then vertically several times on the carcasses. **The abdomen (flank), thorax (lateral), crutch and breast (lateral)**

sites were sampled according to ISO 6888-2 (NCCLS, 2012). On completion of the rubbing process, the shaft was broken by pressing it against the inner wall of the test tube and disposed leaving the cotton swab in the test tube.
Swab samples was also be taken from **knives, splitting axe, personal hands, working cloths, cutting boards, display cabinets, digital scales and hooks** while a certain volume of cleaning water was taken directly from the pipe line. For convenience, the swabs acquired from the equipment and the working environment is considered as samples from the immediate butcher shops' environment. Finally, all samples was kept in the cold chain (+4°C) using icebox and shipped to Assosa Regional Veterinary Laboratory for analysis.

2.6.1. Questionnaire survey and observation

Personal observation and structured questionnaire survey was used to assess the food safety knowledge, attitudes and practices of beef handlers and sanitary conditions of butcher shops and Slaughter Premises of Mange District. The questionnaire was also be prepared to include issues addressing demographic characteristics of respondents, health status and personal hygiene of beef handlers or butchers. The questionnaire was pre-tested and modified as needed before the formal study begins. The questionnaire was filled by face to face interview with 100 beef handlers or butchers (Appendix I).

2.7. Laboratory work

2.7.1. Isolation and identification of *Staphylococcus aureus*

The swab samples which were dipped into 10 ml BPW during collection was directly be incubated for 24 h at 37°C. The water samples was also be enriched at proportion of 1 ml water to 9 ml of BPW and incubated for 24 h at 37°C. A loop full of enrichment was streaked on 7% heparinized sheep blood based agar plate (BAP) enriched and was incubated at 37°C for 24 hrs under aerobic condition. The presumptive colonies of *S. aureus* with beta hemolysis was selected and sub cultured on to Mannitol salt agar. Bacteria colonies having small sized, pale, pink and yellowish color was observed accordingly colonies with golden yellow color was taken as suspected *S. aureus* colony. Further isolation and identification was done by conducting biochemical tests such as gram staining, hemolysis, catalase test, oxidase test, coagulase, motility test, citrate utilization test, O-F test, urease test, carbohydrate utilization test (glucose, mannitol, lactose and maltose) (Quinn *et al*., 2002).

2.7.2. Drug susceptibility testing of *Staphylococcus aureus*

Antibacterial susceptibility testing of *S. aureus* isolates was evaluated against ten different antibiotics (cefoxitin, gentamycin, vancomycin, penicillin, tetracycline, streptomycin, chloramphenicol, nalidixin acid, amoxicillin, Sulphamethoxazole-trimethoprim) by using Kibry-Bauer disk diffusion method following Clinical and Laboratory Standards Institute guidelines (CLSI, 2018). From each isolate, four biochemically confirmed well-isolated colonies grown on nutrient agar was transferred into tubes containing 5 ml of normal saline or Tryptone soya broth. The broth culture was incubated at 37ºC for 8 hrs until it achieved the 0.5 McFarland turbidity standards. Sterile cotton swab was dipped into the suspension and the bacteria was swabbed uniformly over the surface of Muller-Hinton agar plate with in a sterile safety cabinet. The plates was kept at room temperature for 15 minutes to allow drying. Antibiotic discs with known concentration of antimicrobials will be placed and the plates will be incubated for 24 hrs at 37ºC. Following incubation, the **diameters of zone of inhibition** was recorded to nearest millimeters for each disc used and then will be classified as **resistant, intermediate, and susceptible** according to published interpretive chart of CLSI (2018).

2.8. Data Management and Statistical Analysis

Processing of data will be done by computer software. Data was coded and entered to MS Excel spreadsheet and checked for accuracy. After validation, it was transferred and processed using computer software STATA version 13 for analysis. Percentages, proportions, and frequency distributions will be applied to compute some of the data. Logistic regression tests was used when appropriate to analyze the proportions of categorical data. Odd ratio and 95% CI was computed, the 95% confidence level was used, and results was considered significant at ($P < 0.05$).

2.9. Data Quality Control

Sample was collected following strictly aseptic conditions of sample collection procedure for microbial analysis to avoid cross contamination. Laboratory result was evaluated for their consistency with standard using manuals or positive/negative control isolates. Data validation was performed during data entry process for its consistency, accuracy and missing value before analysis. Each stapes of the research was conducted strictly following the data collection, analysis and presentation procedures. Communication to butcher shop owners before the commencement

of sample collection and involving voluntary butchers, confidentiality on the information and disseminating the result was considered concerning the ethical issues.

2.10. Expected Output

The expected output of the current study will be the figurative amount of prevalence of *S.aureus*, drug susceptible pattern and associated risk factors of the disease in the study butcher shops and Slaughter Premises. Therefore, the overall status of *S.aureus* in the Menge District of butcher shops and Slaughter Premises will be determined and quantified, which in turn help to advocate the intervention strategies.

3.RESULTS

3.1. Occurrence of Staph. Aureus in the Abattoir and Butcher shops

In the present study, (n=240 abattoir sample and (n=144) butcher samples were processed, and identified by culturing and biochemical methods. Overall, (N=384, 131/384 (34.11%) of the staph .aureus isolates were able to produce golden yellow colonies on Mannitol salt agar, and blue or violet colored, clustered, Staphy, cocci gram- positive on Gram's staining. The results of hemolysis, coagulase, manitol, catalase, MR and VP test of the staphylococci isolates were positive but it was oxidase test negative. The isolates were able to ferment manitol and maltose and O-F test fermentative.

Of the total abattoir and butcher samples examined, 131/384 (34.11%) staphylococcus aureus prevalence was found to be affected with infection. They were found to be statistically significant (P<0.05). Higher Staphylococcus (50%) contaminates were recorded in Butchers' swab samples and it was 1.06 times higher than abattoir positive samples (N=59/240=24.58%, OR=0.85, p=0.04) as Table 3.

Table3. Prevalence of sample source by binary outcomes- logistic regression

No.	**Sample source**	N=384	Prevalence (%)	OR	95%CI	CHI2	P- value
1	Abattoir	240	59(24.58)	0.85	0.72-0.99	4.10	0.04*
2	Butcher shop	144	72(50)	1.06*	0.79-1.43	0.20	0.65
Total		384	131(31.51%)	1.56	1.0-2.43		

*Abattoir (N=240, LRCHI2=4.10, Prob>chi2=0.0428; SE=0.068; pseudo R2=0.0153); * Butchers' samples (N=144; SE=0.15; LRCHI2=0.20; prob>R2=0.001)

Table 4. Prevalence *of* *Staph aureus* with sample type in Menge Slaughter House (n=240)

Sample Type	**N=240**	**Prevalence (%)**	**OR**	**95%CI**	**Z**	**P –value**
Rectal swab (AM)	36	11 (30.55)	-	0.36-1.38	-1.00	0.32
Hand swab	36	15(41.66)*	0.71	0.36-1.38	-1.00	0.32
knife and axe swab	36	9(25)	0.57	0.39-0.84	-2.85	0.004*
Carcass swab after skinning	36	7(19.44)	0.62	0.47-0.81	-3.38	0.47
Carcass swab after washing	36	3(8.33)	0.54	0.40-0.73	-0.3.98	0.000*
Carcass swab after evisceration	36	8(22.22)	0.77	0.66-0.91	-3.12	0.002*
Swab from transporting material, (pack, vehicle)	24	6(25)	0.83	0.71-0.97	-2.33	0.02*
Total	**240**	**59 (24.58%)**			**12.35**	**0.05***

As Table 4 indicated, the highest Staphylococcus prevalence was identified in workers' hand swab(41.66%) followed by rectal swab(30.55%); knife and axe swab(25%), transporting material swab(25%); carcass swab after evisceration(22.22%); carcass swab after skinning (19.44%); and carcass swab after washing(8.33%), which was statistically significant(P<0.05). The overall prevalence of the Staphylococcus aurues in the present study was 24.58% (N=240, LRCHI2=4.10, Prob>CHI2=0.0428; SE=0.068; pseudo R2=0.0153).

Table 5. Prevalence *of Staph. Aureus* with sample type in Menge Butcher shop (n=144)

Sample Type	N=144	Prevalence %	OR	95%CI	SE	Z	P-value
Carcass swab =retail shop	36	13(36.11)	0.92	0.83-1.02	0.046	-1.64	0.10
Butchers' hand swab	36	23(63.88)*	1.07	0.98-1.16	0.046	1.64	0.10
Knife and axe=Cutting Table=material swab	36	20(55.55)**	1.02	0.95-1.102	0.036	0.67	0.50
Cutting board swab	36	16(44.44)**	1.16	-	1.89	0.01	0.99
Total	**144**	**72(50)**				6.44	0.09

As described in Table 5, the major Staphylococcus aureus prevalence was recorded in Butchers' hand swab (63.88%), followed by knife and axe (55.6%), cutting board swab(44.44%), and carcass retail swab (36.1%), which was significantly associated (P<0.05) (Butchers' samples (N=144; SE=0.15; LRCHI2=0.20; Prob>R2=0.001).

3.2 Staphylococcus Associated Risk factors

Prevalence of *Staphylococcus aureus* infection related to the specific risk factors were determined as the proportion of affected abattoir and butcher swab sample types of examined total samples. As indicated in (Table 6), the questionnaire survey and observation data result shows association of sample type, and sources, origin, beef carcass handling practices, waste management, are amongst the potential risk factors, which are associated with staphylococcus of disease in beef slaughtered and butcher retail shops. Accordingly, Staphylococcus prevalence showed significant variation among different origin groups (p = 0.00), waste management (0.000), and beef handling practices (p=0.000. However, breed, age, and body condition have no significant difference with staph p>0.05).

With regards to Staphylococcus aureus associated risk factors, majority of staphylococcus prevalence (50%) was recorded in Butchers' retail as compared to local slaughtered house (24.58%), which was significantly associated(P<0.05). Higher Staphylococcus prevalence was recorded with advancing age in beef industry, and hence, (39.44%) in greater than 7 years age; followed by (33.47%) in 4-7 years; 21.2% in 3years age, which hadn't significant association (P>0.05). 43.22% of staphylococcus contaminants were identified in cross breed while 30.07% of infection was isolated in local zebu breed, which was no significant (P>0.05). Higher(43.2%) staphylococcus prevalence was identified in poor body conditions whereas26.97% of contaminants were isolated in good body conditions of abattoir and butcher retail samples, which was in -significant(P>0.05).

Table 6: Result of multivariate binary logistic regression of attribute risk factors with Staph

Risk factors	Categories	N =384	Prevalence (N=131, %)	OR	CHI2	P-value
Sample source	Abattoir	240	59(24.58%)	3.09*	25.85	0.000*
	Butcher	144	72 (50%)			
Age(years)	≤3 (year)	33	7 (21.2%)	1.33	1.43	0.2
	4-7 years	242	81(33.47%)			
	> 7 years	109	43 (39.44%)			
Breed	Cross	118	51(43.22%)	1.52	1.62	1.00
	Zebu	266	80(30.07%)			
BCS	Good	215	58 (26.97%	0.91	0.54	0.70
	Poor	169	73(43.19%)			
Drainage(waste) management	*poor*	*172*	*77 (44.76)*	*2.59*	4.04	0.00*
	good	*212*	*54(25.47)*			
General hygiene	*poor*	*245*	*91(37.14)*	*0.84*		0.67
	good	*139*	*40(28.77)*			

Meat handling practice	*good*	*230*	*62(26.95)*	*3.6*	5.39	0.000*
	poor	*154*	*69(44.80)*			
Origin	*Assosa area*	*119*	*31(26.05)*	*1.28*	2.14	0.03*
	wollega	*149*	*62(41.61)*			
	Other	*116*	*38(32.75)*			

NB: * significant, OR= odd ratio; CHI2- chisquar

3.3. Antimicrobial Susceptibility Test

Out of 121 *S.aureus*, 39 isolates were subjected to antimicrobial susceptibility tests. Amoxicillin and Penicillin were drugs to which a large proportion of *S aureu*s isolates' resistance. As it is indicated in Table 10, most Isolates(84.61% and 100%) were resistance to these two drugs respectively. *S. aureus* isolates were 56.41% resistance to Sulphonamide and 64.10% of Cefoxitin. All 39 testes species of *S. aureus* were highly susceptible to Ciprofloxacilin (100%) and Chloramphenicol (94%) followed by Gentamicin (92.30%).

Table 8: Resistance and susceptible of *S. aureus* isolates to different antimicrobials (n = 39)

Antimicrobial agents	Disc content (µg)	No. of Isolates	Resistance	Intermediate	Susceptible
			No (%)	No (%)	No (%)
Cefoxitin	CK-30	39	25(64.10)	0(0)	14(35.89)
Amoxicillin	Amx-10	39	33(84.61)	0(0)	6(15.38)
Sulphonamide	S-300	39	22(56.41)	12(30.76)	5(12.82)
Gentamycin	Gen-10	39	3(7.69)	0(0)	36(92.30)
Penicillin G	P-10	39	39(100)	0(0)	0(0)
Chloramphenicol	C-30	39	3(7.69)	1(2.56)	35(89.74)
Ciprofloxacin	Cip-5	39	0(0)	0(0)	39(100)

Key:%=percent, S=susceptible; I=intermidiate; R=resistance

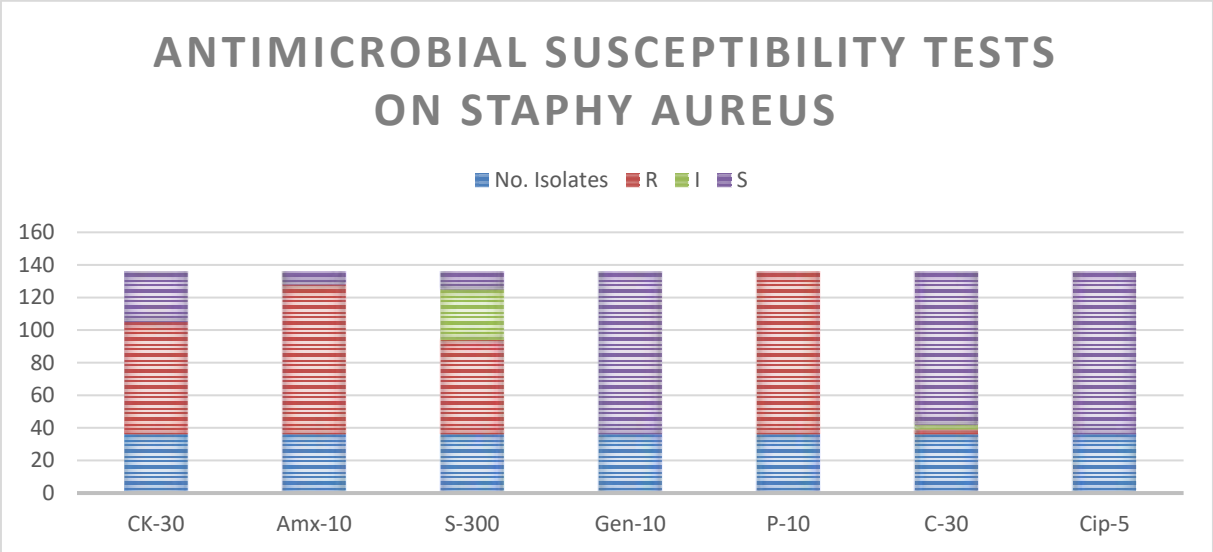


Figure 3: Antimicrobial drugs resistance and susceptibility profile of *S.aureus* isolated from milk; **Key:** Fax30, CN10, Pen-10, AMX-10; S-300; Gen-10; C-30; CIP-5

3.3.1 Multi drug resistance of S. aureus

In this study, 12 isolates of *S. aureus* showed resistance to two classes of antimicrobial drugs. From this isolates, (8) eight for amoxicillin and penicillin, three (3) for amoxicillin and cefoxitin and one for penicillin and sulfonamide.

Of the 17 isolates of S aureus, 14 isolates were resistance to three classes of antimicrobial (amoxicillin, penicillin and sulfonamides); 1 isolates were each for (chloramphenicol, penicillin and sulfonamides) and 2 isolates for (amoxicillin,

penicillin and cefotitin). The maximum multiple drug resistance registered for 3 isolates were resistance to four classes of antimicrobials as indicated in Table 9.

Table 9. Multi drug resistance /MDR/ pattern among S. auerus isolates

No. AMR	AMR patterns	No. isolates	No.of isolates (%)
Two	S3, PG AMX, PG AMX, FOX	1 8 3	1 (2.56) 8 (20.51) 3(7.69)
THREE	CHL, PG, S3 AMX, PG, S3 AX, PG, FOX	1 14 2	1(2.56) 19(35.89) 2(5.12)
FOUR	AMX, PG, S3, CHL AMX, PG, S3, FOX	1 2	1(2.56) 2(5.12)

bKey; no= number; %=percent; AMX= amoxicillin; PG=penicillin, S3= sulfonamides; CHL= chloramphenicol; FOX= Cefoxitin

3.5. Questioner and Observational Survey

3.5.1. Demographic information of abattoir and butcher shops workers

With regards to demographic information of Abattoir and butcher workers', 65% of abattoir workers were 18- 40 years age groups and 35% workers were greater than 40 years age while 75% of butcher workers were in 18- 40 years age and 25% were greater than 40 years. The dominant abattoir and butcher workers were male domain as compared to female. Concerning the workers' experience, higher (50%) abattoir workers had 4- 5 years of experience, followed by 30% in greater than 5 years and 20% had less than 3 year experience whereas, 55% of butchers' work experience was 4- 5 years; 25% in less than 3 years and 20% in greater than 5 years**.** In line with the education level of abattoir and butcher workers, majority of abattoir workers (40%) were primary education level , followed by 30% in secondary level; (20%) illiterate; (10%) college level whereas in butcher retail workers, 45% of education categories of workers were primary; (25%) secondary; (15%) illiterate; (15%) college level.

With regards to capacity building, majority (75%) of abattoir workers had not attained food safety training, but 25% had got training; 70% of butcher workers did not obtained training while the rest obtained training. 80% of abattoir workers hadn't taken health certificate but 20% got health certificate; whereas in case of butcher workers 85% did not got health certificate however, 15% got health certificate (Table 10).

Table 10. Demographic information of Meat handlers working in Abattoir and retail Beef Shop

Variables	**Category**	**Abattoir workers (N=20, %)**	**Butchers Shop workers (N=20, %)**
Age	18-40years	13 (65%)	15(75%)
	>40 years	7(35%)	5(25%)
Sex	Female	2(10%)	1(5%)
	Male	18(90%)	19(95)
Experience	Less than 3 years	4(20%)	5(25%)
	4-5 years	10 (50%))	11(55%)
	> 5 years	6(30%)	4(20%)
Education	Illiterate	4(20%)	3(15%)
	Primary	8(40%)	9(45%)
	secondary	6(30%)	5(25%)
	college	2 (10%)	3(15%)
Food safety training	Yes	5(25%)	6(30%)
	No	15(75%)	14(70%)
Health certificate	Yes	4(20%)	3(15%)
	No	16(80%)	17(85%)

3.5.2. Hygienic meat handling Practice of abattoir workers

Results from this study revealed that from 30 of the abattoir workers interviewed, 80% of the workers use water only for washing and 36.66% of them have habit of disinfect knife before commencing slaughtering. About, 66.66% of workers did not wear protective clothes, and all of them handled meat with bare hands. About 88.2% of the respondents have no health certificate there was no habit of reporting illness. Majority of abattoir workers (76.47%) respond that there was no sanitary control system (Table 6). 70% of the abattoir workers did not aware of zoonotic / foodborne pathogen while 30% of workers aware of foodborne pathogen. 73.33% of abattoir workers had awareness on source of meat contamination while 26.66% did not. 56.66% of abattoir workers wash equipment once in a day while 43.30% of workers wash equipment's twice a day; 73.33% of workers clean slaughter house once a day but 26.66% cleaning slab house twice a day. Majority (63.33%) of abattoir workers did not clean cutting boards while 36.66% cleaning cutting boards. 16.66 % of abattoir workers use Jewelry while 83.33% did not used(Table 11).

Table 11: Survey on hygienic meat handling practices of Menge municipal abattoir workers

Factors	Categories	Frequency n=30	Percentage (%)
Hand washing	water only	24	80
	water and soap	6	20
Sterilizing knife	Yes	11	36.66
	No	19	63.33
Protective clothing (apron, hair coat)	Yes	10	33.33
	No	20	66.66
Awareness on source of meat contamination?	Yes	22	73.33
	No	8	26.66
Knowledge of zoonotic/ foodborne pathogen	Yes	9	30
	No	21	70
Cutting board condition	clean	11	36.66
	No	19	63.33
Jewellery(watch, ring,bracelet)	Yes	5	16.66
	No	25	83.33
House cleaning frequency	Once	22	73.33
	Twice	8	26.66
Freq. of washing equipment's'	Once	17	56.66
	Twice	13	43.33

3.5.3. Hygienic meat handling Practice of butcher shops workers

The study showed that from interviewed (N=30) butcher shops workers, 70% of the butcher shop workers used only water for cleaning and all of them never have habit of sterilizing knife every consecutive working days. About 86.66% of the workers at the butcher shops did not wear aprons and 70% of them did not cover their hair. However, 83.33% of the butcher shop workers wore dirty white gown. Most of butcher shops didn't have casher and therefore, on this study 76.66% of butcher shops workers handle money while serving in meat processing (Table 12).

Table 12: Survey on hygienic meat handling practice of butcher shops workers of Menge

Observations	Categories	Frequency	Percentage (%)
Cleaning and hand washing	water only	21	70
	water and soap	9	30
Sterilizing knife	Yes	0	0
	No	30	100

Hair cover	Yes	9	30
	No	21	70
White gown	Yes	25	83.33
	No	5	16.66
Apron use	Yes	4	13.33
	No	26	86.66
Handling money	Butcher	23	76.66
	casher	7	23.33
Cutting board condition	Clean	7	23.33
	non	23	76.66
Jewelery	Yes	18	60
	No	12	40
House cleaning freq.	once	21	70
	twice	9	30
Freq. of washing equipment's	once	22	73.33
	twice	8	26.66

4. DISCUSSION

Abattoir is one of the food industries that contribute to the problem of possible food-borne diseases and health hazards associated with food unless the principles of food-borne hygiene practices are implemented (H. Roberts, L *et al*., 2009).

Staphylococcus aureus originating from meat is a leading causes of foodborne illness and the presence and spreading of antimicrobial resistant *S. aureus* is one of the great global public health issue especially in developing countries like Ethiopia. Detection of S. aureus from meat indicates that poor hygienic condition of value chain process from abattoir to retail houses. S. aureus which is natural flora of skin and mucous membranes of animals and human can cause beef contamination during processing (Norrung *et al.*, 2009; Komba *et al*., 2012).

In present study, from a total of 384 swab sample taken from municipal abattoir and butcher shops, 34.11 % were S. aureus positive. Of these, 24.58% (N=240, 59/240=24.58%, OR=0.85; CI=0.72-0.99) were from municipal abattoir and 50 % (N=72/144=50%, OR=1.06; CI=0.79-1.43) were from butcher shops swab samples (P<0.05), which is concordant with study reported by Bizuneh Tsehayneh, (2019) 54.45% S. aureus prevalence from ready to eat meat in Bahir Dar city. Besides this, 24.6% S.aureus prevalence reports of Lema *et a*l., (2021) in Addis Ababa and 21.46% of Tibebu *et a*l., (2021) from Bishoftu, Ethiopia. This similarity might be due to presence of common poor food safety knowledge in abattoir and butcher shops workers in the study areas.

In other study, 19.8% prevalence of S. aureus was reported by Feben Adugna *et al*., (2018) in Addis Ababa abattoir and butcher shops which is lower than present finding and 78.5% S. aureus isolates have been reported by Das *et al.*, (2019) in Bangladesh from carcasses at slaughter house, which was higher than present finding. This difference might be due to variation in the level of knowledge and practice of hygienic meat handling at abattoir and butcher shops workers.

This finding was consistent with the previous findings of Dessalew T, (2021) in Dangila town, Amhara Region, in Municipal Abattoir and Butcher, which was 48.43% Staph. aureus in Municipal abattoir and 54.68% butcher shop; overall, 51.56% S. aureus prevalence. Besides this, Dessalew T (2021) reported that, 5.5, 8.2, 7.5, 5.7 and 5.7, 7.2, 6.7, and 5.9 log10cfu/cm2 staphylococcus bacterial count in carcass, knives, cutting table and hooks swab in abattoir and butcher shop respectively. Comparably, Dessalew T, (2021) reported that, 48.44% of staph. aureus prevalence in Abattoir with 43.75% in carcass, 62.5% in knife swab, 50% in cutting board, and 37% in hooks; whereas, 51.56%

staph. aureus prevalence was reported in butcher shops with 68.75% in carcass; 56.25% in knife, 75% in cutting board; and 31.25% in hooks, which were significantly associated (P<0.05).

In this investigation, *S. aureus* isolated from butcher shops were more than abattoir which is in line with study reported by Henok A *et al.,* (2019). This similarity might be due to contamination of meat during unhygienic transportation from abattoir to retail shops. In addition to this meat could get further contamination in butcher shops during handling and preparation from personal and equipment contacts.

Present study revealed that, S. aureus isolated from rectal swab, abattoir workers' hand swab, knife and axe swab, carcass swab after skinning, washing, evisceration and transporting material swab of abattoir were 30.55%, 41.66%, 25%, 19.44%, 8.33%, 22.22%, and 25% and S.aures isolates from carcass swab, butcher workers' hand swab, knife and axe swab (material swab); and cutting board swab of butcher shop were 36.11%, 63.88%, 55.55%, and 44.44%, respectively, which were highly contaminated and this result was consistent with the earlier findings of Dessalew T, (2021) , who reported, S. aureus isolated from Knife, Cutting boards and Hooks of abattoir of 62.5% , 50%, 37% and 54.69 % and from butcher shops 68.75%, 56.25%, 75% and 31.25 % respectively which is highly contaminated. Besides this, this result is found to be concomitant with study reported by Feben Adugna *et al.*, (2018). This similarity could be due to butchers in abattoir and retail shops didn't have practice of cleaning and disinfection of equipments involved in meat processing before and after each slaughtering activity at both study areas.

The present study showed that, 34.1 % overall prevalence of ***S. aureus*** in both abattoir and Butcher shop in the study area, which is higher than 15.6% prevalence reported from Ambo and Bako towns in the west Shoa zone of the Oromia regional state, Ethiopia (Bizunesh *et al*., 2023). Besides this, this findings were higher as compared to the previous findings of Feben A *et al*., (2018) in Addis Ababa, who reported 9.4% Staph. Aureus prevalence in Municipal abattoir; 19.8% in Butcher shops; 15% in cutting table, 15% in hooks, and 22.5% in knife at each critical control points. The variable prevalence of *S. aureus* in Abattoir and Butcher shop at each critical points may be attributed to differences in hygienic practice of slaughtering house, sanitary, Equipment(Utensils and cutting boards, washing practices, Abattoir management practices, environmental conditions, and awareness of disease transmissions. *S. aureus* is a contagious pathogen that spreads from infected cows to healthy ones during unhygienic milking practices and contact with animals

Staphylococci, which are natural flora of skin and mucous membranes of animals and human can cause meat contamination (Nørrung *et al*., 2009). As compared to present findings, Ahmed et al., (2013) in Lahore University, indicated that, mean S. aureus counts for the beef meat from abattoirs and retail outlet were 2.76 and 3.07 log10 CFU/cm2 respectively. *S. aureus* was isolated from the 72 (51%) samples, which indicate poor sanitary quality of abattoirs and retail outlets. S. aureus positive samples were significantly higher for beef outlets as compared to beef abattoirs (70% vs 55%), sheep outlets as compared to sheep abattoirs (45% vs 25%), and goat outlets as compared to goat abattoirs (70% vs 40%)

In the present study the higher S. aureus prevalence was reported in abattoir workers hand(41.66%) followed by 30.55% in rectal swab in abattoir categories whereas in Butcher shop categories, 63.88% S.aureus contaminants were reported in buchers' hand swab; followed by 55.53% in knife and axe swab; and 44.44% in cutting board which was significant(P<0.05). Similarly, Butchers' hand swab were 1.07 times more harbor S. aureus; knife swab 1.02 times harbor S.aureus; and cutting board swab 1.16 times more likely harbor S. aureus contaminants. This implies that there was poor sanitary practice of abattoir and butcher shops knives and axe, Cutting boards, and workers hand swabs at each critical control points; which is convenient with study reported by Ali *et al*., (2010) from Pakistan ; Dessalew T (2021) from Dagila town, and Feben A *et al*., (2018) from Addis Ababa. This similarity could be due to poor cleaning and disinfecting practice of cutting boards before and after each activity of meat processing by the workers at study areas. In this study relatively small bacterial load was identified from hooks of abattoir and butcher shops this could be due to small surface area and less frequently contact with personnel hands.

With regards to Staph aureus associated risk factors, 34.1% of the abattoir and butcher shop samples were contaminated with Staph. aureus. In this findings, the prevalence of S.aureus infection was significantly influenced by age categories. Significant *S.aureus* infection associated with age categories which was higher in (39.44%) in greater than 7 years age followed by (33.47%) of 4-7 years age and in 3 years (21.2%) which was non - significant (P>0.05). Similar result was reported by Shimelis (2014) in Selale /Fitche,). Old followed by adult cattle in this study were more susceptible to S.aureus infection than young cows. The increasing occurrence of infection with increasing age were agreed with the findings by Kerro and Tareke, (2003) who found that, the risk of infection with the advancing age of the cow. This might be due to the increased opportunity of infection with time and the prolonged duration of infection, especially in a herd without mastitis control program (Radostits *et al*., 2007).

Significant (43.22%) S.aures infection was recorded in cross breeds followed by 30.07% in local zebu breeds (p>0.05). Comparable findings were reported by with Bitew *et al*. (2010) who reported in Bahir Dar, between Cross and Fogera breed, Lakew *et al*. (2009) in cross and local Arsi breed.

This study revealed that beef house with poor drainage was 44.76 times more likely to be harbor S.aureus than well drainage housing systems. The association can be attributed to poor sanitation practices and poor waste disposal practices in the slaughter house, that promote the survival and transmission of pathogens (Bizunesh *et al*., 2022**).**

Occurrence of *S.aureus* infection was significantly associated with hygienic practice (p>0.05). Beef farms with poor hygiene standard are severely affected (37.14%) than those with good hygiene practices (28.77%). Similarly, in abattoir and butcher shop, 44.80% of S.aureus was contaminated with poor meat handling practices and 26.95% contaminates recorded in good handling practices, which was significant (P<0.05). This findings were comparable with the previous findings of (Mulugeta and Wassie, 2013; Lakew *et al*., 2009; Sori *et al*., 2005). This might be due to absence of abattoir hygiene, handling of meat with common abattoir and butcher shop workers' and using of common abattoir, and butcher protective cloths, which could be vectors of spread especially for zoonotic diseases (Radostitis *et al*., 2007).

The hygienic conditions of abattoirs and butcher shops as well as equipment are major factors contributing in bacterial contamination of meat (Gill *et al*., 2000). However, in this study overall sanitation of the abattoir and butcher shops were poor. There was no proper disposing system as a result solid wastes such as faces, skin, bones and horns were found near to the abattoir. Most of the butcher shops were found on the road side, exposed to dust due to wind or vehicle, and so there is high probability of contamination with microbes. According to WHO (2004) food handlers should be educated.

With regards to Abattoir and butcher shop workers' demographic information, in the present study, 40 abattoirs and butcher shop workers were interviewed to collect information which can directly or indirectly contribute to the contamination of meat. Out of the total (n= 40) respondents interviewed, 2(10%) were female and 18 (90%) were male in Abattoir workers, whereas from butcher workers, 5% were female and 95% were males workers'. The present findings were agreed with earlier report of Wassie B et al., (2017) in Godar Town, Ethiopia with 17% female and 83% male domains.

The level of education and training of food handlers about the basic concept and requirements of personal hygiene and its environment plays an important part in safeguarding the safety of products to consumers. During the study it was revealed that, the abattoir workers had low level of education and this could make difficult in acceptability of modern slaughtering practices as well as adherence to strict hygienic and standard slaughtering practices that contribute to microbial contamination. Based on their educational status of respondents 4 (20%) were illiterate, 8(40%) primary, 6(30 %) secondary, 2 (10%) colleges of Abattoir workers'; while in butchers' workers 15% illiterate ; 45% primary; 25% secondary; and 15% in college. It was comparable with wassie B. et al.(2017) with report of 3.8% illiterate; 20.8% primary; 43.4% secondary and 31.7% college level workers were interviewed. Consistently, Abebe B et al.(2019) indicated 66.67%; 16.67%, 8.33%, and 8.33% of Butcher workers' education categorized as grade1-4; grade 5-8; 9-12 and greater than 12 respectively.

Comparably, the present result was inline with earlier works of Dessalew T (2021) from Dangila town, who reported , 41.2% abattoir and 43.75% butcher shops workers were illiterate and 82.4% abattoir and butcher shops of 81.25% workers didn't have training regarding meat hygiene. Besides this, This result was in line by Feben A et al., (2018) and Haileselassie M *et al*., (2013) where they reported 58.3% illiteracy level and 61.5% untrained personnel, respectively

Food safety training (N=40), 25% had attained food safety training and 75% didn't attained training in abattoir; whereas, 30% had food safety training; and 70% hadn't attained food safety training. Majority (20%) of the respondents had got health certificate whereas 80% of the respondents hadn't got health certificate in Abattoir while 15% of the Butcher workers' got health certificate and 85% butcher workers' didn't have health certificate . This result was comparable with the previous report of Wassie B *et al*., (2017) with 33.96% of Butcher workers had access of training and 66.04% hadn't have access of training. Abebe B *et al*., (2019) in Bishoftu, Central Ethiopia, revealed, 41.67% of Butcher workers receive training and 58.33% had not received training. In contrary with the present findings, Abebe B *et al*., (2019) in Bishoftu, Central Ethiopia reported, 58% and 42% of workers had health certificate and hadn't have health certificate respectively. In line with the present findings, Fanta D. G *et al*.,(2021) in Bishoftu, Ethiopia reported that, 60.7% of abattoir workers received on the job training on meat hygiene practices and 39.3% of them didn't received on job training.

Besides this, Muhammed N., and Mulu D.(2021) at Slaughterhouses and Butcher's Shops in West Hararghe Zone Ethiopia stated that 15% attained meat hygiene training and 85% of abattoir workers didn't attained training. Comparable result was reported by Birhanu S and Menda S., (2017) in Gojjam Area, Ethiopia indicated that, 1.85% of worker has health certificate and 98.15% of abattoir worker didn't given health certificate by checking health status of workers. Muhammed N. and Mulu D.(2021) at Slaughterhouses and Butcher's Shops in West Hararghe Zone Ethiopia stated that 7.5% f abattoir workers got health certificate and 92.5% of abattoir workers had not got certificate. Consistently, Fanta D.G *et al*.,(2021) in Bishoftu, Ethiopia indicated that, 50 % of workers had got frequency of medical checkup was every 3mth and 50%% of them stated as it was every 6 mth of years in abattoir.

The hygienic condition of the abattoir workers has potential to contribute for contamination in meat processing. Author (L. Adetunde, R et al., 2011) reports unclean slaughter men's hands, clothing, and equipment used in carcass dressing process accounted for the microbial contamination. (PPE) Personal protective clothes such as overalls or hair cover should be worn at all times when handling meat and the overalls should be light in color so that contamination can be easily identified and cleaned easily (MoA, 2010). This study revealed that 66.66% of abattoir workers didn't use protective clothes, and all of them handled meat with bare hands and 86.66% of workers at the butcher shops didn't wear aprons and 70% of them didn't cover their hair, This findings were compared with the previous findings of Dessalew T (2021), who revealed that, 88.2 % of abattoir workers did not use protective clothes, and all of them handled meat with bare hands and 81.25% of the workers at the butcher shops did not wear aprons and 68.75 % of them did not cover their hair. In addition, the present finding is lower than survey reported by Haileselassie M *et al*., (2013) 11.3% butcher shop workers did not use protective clothes and 50.7% did not cover their hair. This difference could be due to variation in the level of awareness of workers in meat handling. Paper money used as medium of exchange handled under poor hygienic conditions could possibly contaminated by microorganism and could be impose a major health hazard (Gosa Girma *et al*., 2014)..

However, this study revealed that 76.66 % butcher shops workers handle money during serving in meat processing. The present study is concordant with Endale Balcha *et al*. (2013) and Abebe B *et al*., (2019) reported that 68.75% and 66.67% respectively. In other study Muinde and Kuria (2005) from Kenya have reported 91.7% of butchers handled money while serving food which is much higher than present survey. This difference might be due to difference in awareness and knowledge of butcher shop owners and workers in meat handling in different areas.

About 34% of abattoir and butcher workers are carriers of *S. aureus* as normal flora. Food which has been handled by humans has a high probability of contamination with bacteria (Kluytmans & Wertheim, 2005).

With regards to cleaning and hand washing, 70% of butcher workers used water only for cleaning, and 30% used water and soap ,whereas 80 % of abattoir workers used water only for hand washing and equipment's and 20% of workers used water and soap for cleaning and washing hand, and slab house. Similarly, 63.33% of abattoirs' cutting boards were not clean properly while 36.66% only cleaned properly whereas 76.66% of butchers, shop cutting boards were not clean properly but only 23.33% of cutting boards clean properly. 56.66% of abattoirs' the slab house equipment's were cleaned once a day and 43.33% of equipment's' clean twice a day and 73.33% of slab house was cleaned once a day and 26.66% cleaned twice a day; whereas; 70% butchers house clean once aday and also 73.33% of butchers' equipments' washing once aday ; which is in line with 41.1% investigated by Feben Adugna *et al*.,(2018). Consistently, Abebe B *et al*. (2019) in Bishoftu, Central Ethiopia stated that 75% of abattoir workers apply water only and 25% apply (water, soap) and detergent as cleaning strategy in the area. And also 41.67% of workers cleaning abattoir daily, and 58.33% of workers clean every other days once every 48-72 hrs. Besides this, Fanta DG *et al*.,(2021) in Bishoftu, Ethiopia revealed that, 46.0% of abattoirs workers hand washing between activities during the work time and 53.6% of workers didn't wash hands between activities.

Consistently, Adeolu, A.T *et al*., (2019) in Karu Abattoir of Nigeria indicated that 98.3% of workers indicated as there was washing of hand after each operation while 1.7% of worker noted as there was no washing of hand after each operation. Comparably, Adaoha Pearl Agu *et al*., 2021 in Abakaliki, Ebonyi State South-East Nigeria stated that 80.3%,71.3%,69.7%,60.1%, and 43.6% of abattoir workers food and environment safety were proper waste disposable, proper storage of leftover meat and regular hand washing , wear PPE, and washing hand after each operation respectively. Muhammed N., and Mulu D. (2021) at Slaughterhouses and Butcher's Shops in West Hararghe Zone Ethiopia stated that 62.5% and 37.55 of abattoir workers hygienic condition of coat were good and poor.

Muhammed N., and Mulu D., (2021) at Slaughterhouses and Butcher's Shops in West Hararghe Zone Ethiopia, stated that 66.7% of abattoir workers indicated as there was washing and sterilizing of knives after skinning and evisceration and 33.3% of workers did not washed and sterilized of knives after skinning and evisceration. Similarly, 100%, 0.0%, 0.0% of frequency of cleaning and disinfection was done daily, twice week and trice week respectively.

In the present study, all butcher shop workers didn't use sterilizing knife while 36.66% of abattoir workers' use sterilized knife for cutting meats and majority(63.33%) didn't sterilize knife. This finding was agreed with results, B. Endale and G. Hailay, (2013) revealed, knives were washed only with water and there is poor sterilization and continuous use of a single knife despite contact with dirty or contaminated surfaces. The presence of bacterial pathogens in meat contact surfaces may contribute to the contamination of meat. In a similar way, the present study revealed the total mean of APC from butcher house equipment (cutting knife and cutting table). The variations of bacterial load observed in different studies might be due to lack of good processing and handling practices and sanitary standard operating procedures of meat along the meat production chain (D. Pius 2013).

Moreover, performing skinning and evisceration on the ground without separating the dirty and clean areas increases the risk of cross contamination during meat processing, putting meat consumers at risk of foodborne illness. Comparably, the present result was similar with the previous report of Birmaduma G *et al*, (2019) in Eastern Oromia who noted that, 57.89%, 15.79%, 26.3% of city Vans truck, private ways and Hucar of carcass transport. Besides this, study was comparable with Birmaduma G *et* al, (2019) of 12.28%, 10.52%, and 57.89% of abattoir workers noted as there was loading and unloading, meat inspectors and carcass split respectively. Comparably, Adaoha Pearl Agu *et al*. (2021) in Abakaliki, Ebonyi State South-East Nigeria stated that 70.2% and 29.85 of Abattoir workers were actively involved by government and health offices ensuring good hygienic practices in areas. Adeolu, A.T *et al*., 2019 in Karu Abattoir of Nigeria indicated that current waste disposable method were 73 .6% , 13.2%, and 13.2% of abattoir of open dumping, land filling and composting way of waste disposable. Comparably, Adaoha Pearl Agu et al., 2021 in Abakaliki, Ebonyi State South-East Nigeria stated that 76.1%, 6.9%, 11.2%, and 5.9% of abattoir workers method of waste disposable were open dumping, burning, land filling and don't know response rates respectively.

The sanitation and hygiene status of the butcheries was generally poor as most of them did not meet the sanitary requirements for operation as stipulated in (Adaoha Pearl Agu *et al*., 2021). Meat hygiene was not properly observed as factors that directly affect it such as discarding wastes; sufficient drainage system and hand washing practices were not adhered to. Additionally, practices such as use of protective wear were low and other requirements such as medical examination of meat handlers and hand washing after toilet was neither consistent nor verifiable. Observation showed that domestic flies were abundant in most parts of the abattoir. It is important to control flies since they feed on meat and other wastes, where they pick up and transport various diseases causing agents with potential to causes such as enteric and eye infections. Based on findings from this study, the sources of meat contamination originated from the slaughter process in the abattoir and poor hygienic environmental conditions, unhygienic handling of meat by the abattoir workers and crosscontamination from the floor and other equipments were used in the abattoir.

The present study showed that the resistance of *S. aureus* to Penicillin G (100%), amoxicillin (84.61%), sulphonamides (56.41%), and (64.10%) cefoxitin, and 7.69% in chloramphenicol observed in milk samples. Comparable research works were reported in various parts of Ethiopia by Biniam T (2014) revealed resistance of *S.aureus* to Penicillin G (100%) in and around Wolaita Sodo, southern, Ethiopia**.** Besides this, Alemayehu (2015) indicated resistance of *S.aureus* to Penicillin G (95.8%), Cefoxitin (75.7%), and Tetracyline (72.2%), from Bovine mastitic milk in Dairy farms of Bahir Dar.

In addition, this research is in accordance with the findings of Abebe *et al.* (2013) who reported resistant of *S.aureus* to penicillin G 96.7% and tetracycline 73.8% around Addis Ababa, and Abera *et al.* (2010) 94.4% resistance to penicillin G in Adama; in addition to this study has demonstrated the existence of alarming level of resistance of *S. aureus* to commonly used antimicrobials (penicillin G, and tetracycline) in dairy farms. This results were in consistent with reports from earlier studies in the other countries (Edward *et al*., 2002; Gentilini *et al*., 2002 and Jakee *et al*., 2008) suggesting a possible development of resistance from prolonged and indiscriminate usage of some antimicrobials. Hence, penicillin and tetracycline are the only most commonly used antimicrobials for the treatment of other infections as well as mastitis in veterinary practice in Ethiopia, as the result, there was spread of drug resistance reported by many researchers which was in line with the recent findings.

The resistance of *S.aureus* isolates to beta-lactam antibiotic was evident. High percentage of *S.aureus* was resistant to the most frequent drugs. In agreement with the finding of by Derese *et al*. (2012), the study showed cefoxitin resistant isolates were obtained from the milk. All cefoxitin resistant *S. aureus* were also resistant to penicillin G. (64.10%) cefoxitin was resistant to *S. aureus* isolates, (84.61%) and (100%) were also resistant to amoxicillin and Penicillin G respectivelly. This is an indicator of MRSA (Daka *et al*., 2012). This is due to the fact that resistance of *S. aureus* to these drugs may be attributed to the production of β-lactamase, an enzyme that inactivates penicillin and closely related antimicrobials (Wubishet *et al*., 2012; Sharma *et al*., 2011; Green and Bradely, 2004).

In addition, the current investigation was agreed with the reports of Tsige 2018 who reported the resistance of s. aureus to penicillin 100%. Moreover, the present report was compared with the results of Markos *et al*., 2023 in Shishicho town recording 100% for both penicillin and amoxicillin. This is supported by the findings of Endries *et al*., 2022 from Holeta who reported S.aureus resistance to Amoxicillin 95% and Oxacillin 87.50%. Similarly, S aureus resistance to penicillin 94%, were reported by bishoftu Tibebu *et al*. (2021).

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Antimicrobial susceptibility tests of S.aureus isolates revealed that the highest rate of susceptibility among the isolates was recorded against Ciprofloxacin, Gentamycin, chloramphenicol. The results agree with the findings of Tibebu *et al*. (2021) who reported susceptibility to S aureus to Ciprofloxacin and Gentamycin. The variability in susceptibility results could partly arise from how frequently a drug was in use for dairy cows treatment in the study area. The resistance of S. aureus isolates to penicillin G may be attributted to the production of B- lactamase enzyme that inactivates penicillin and closely related antibiotics. Resistance to pencillin G is used as a marker to assess the susceptibility of S. aureus isolates against other beta lactam antibiotics (Markos *et a*l., 2023).

5. CONCLUSION AND RECOMMENDATIONS

This study showed that S. aureus was a major contaminant of beef from abattoir to butcher shops in menge district. Most of beef at abattoir and butcher shops were highly contaminated with S. aureus and below acceptable standard limit level for human consumption. The overall higher loads of the S. aureus occurs on the beef at the butcher shops relative to abattoir due to further contaminations during transportation and handling at the retail houses. This study confirmed that all of isolated S. aureus were resistant to single to five antimicrobial agents. The highest resistance was observed to penicillin G followed by Amoxicillin, cefoxitin, and Sulphonamides. However, most of S. aureus isolates were susceptible to Chloramphenicol, Ciprofloxacin, and Gentamycin. Therefore beef could be the major sources of Staphylococcal food borne diseases and spreading of antimicrobial resistance to the consumer unless good hygienic practices are implemented in meat value chain process.

Based on the above conclusion the following recommendations are forwarded:

- Appropriate strategies should be designed and implemented in meat processing units to reduce S. aureus contamination.
- Abattoir and butcher shops hygienic practice should monitored regularly.
- Abattoir and butcher shops workers should have training on hygienic meat handling.
- Further research should be conducted on antimicrobial resistance and enterotoxin producing strains of S. aureus

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