Isolation, Identification and Drug Resistance Profile of salmonella from Apparently Healthy Cattle Slaughtered at Wolaita Sodo Municipality Abattoir, Ethiopia

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Abstract
The muscle (meat) of a healthy animal is sterile. Contamination may be due to infection within the animal or external contamination during slaughter and handling processes. High bacterial load on carcasses may pose potential risk of meat contamination with Foodborne pathogens. A cross-sectional observational study was undertaken in Wolaita Sodo municipal abattoir from November, 2015 to April, 2016 for isolation, identification and drug susceptibility of salmonella from cattle. Methods followed in the Study were ISO 6579: 2002 procedures for Salmonella isolation which included pre-enrichment, Selective enrichment, plating out and confirmatory tests. Accordingly, the overall 16/300 (5.3%) prevalence of Salmonella was revealed at abattoir. A total of 100 healthy slaughtered cattle’s were systematically collected and examined for the presence of Salmonella following the standard techniques and procedures. Out of the total of 300 samples, 16 (5.3%) were positive to Salmonella. Of all isolates, 13 (81.25%) were multiple antimicrobial resistant and highest level of resistance was observed for ampicillin (56.25%), cefoxitin (43.75%), chlorophenicol (37.5), kanamycin (35.5%).

Keywords: Abattoir, Antimicrobial resistance, Cattle, identification, Isolation, Salmonella

1. INTRODUCTION
Food-borne diseases are common in developing countries including Ethiopia because of the prevailing poor food handling and sanitation practices, inadequate food safety laws, weak regulatory systems, lack of financial resources to invest in safer equipment, lack of education for food-handlers [1]. Most of them are zoonotic and have reservoirs in healthy food animals from which they spread to variety of foods. Therefore food of animal origin is considered as major vehicles of food borne infections [2].

Salmonellosis is considered as one of the most widespread food borne zoonoses associated with food of animal origin in industrialized as well as developing countries even though the incidence seems to vary between countries [3]; [4] and [5] reports that, Salmonella covers 88% of the food borne infections. In many registers non-typhoidal Salmonella species are documented as one of the leading causes of bacterial diseases. Food borne Salmonella typically causes acute gastroenteritis and may cause a more Septicemic disease usually in very young, the elderly and immune compromised subject [6].

The ubiquitous distribution of Salmonella in the natural environment and its prevalence in the global food chain, the physiological adaptability and its potentially serious economic impact on the food chain industry predicate the need for continued attention and stringent control at all levels of food production [7]. The continued prominence of raw meats, eggs, dairy products, vegetable sprouts, fresh fruits, and fruit juices as the principal vehicle of human food borne salmonellosis arises from major difficulties to coordinate sectorial control efforts within each industry. The problem of salmonellosis is further compounded by the massive and unrestricted movement of food in international trade, the national disparities in the hygienic agricultural and aqua cultural production of foods and the non-uniform government and industry food safety controls during production of foods and the non-uniform government and industry food safety controls during the processing, distribution and marketing of fresh and processed food products [8][6].

Moreover antimicrobial resistant Salmonella are increasing due to the use of antimicrobial agents in food animals at sub-therapeutic level or prophylactic doses which may promote on-farm selection of antimicrobial resistant strains and markedly increase the human health risk associated with consumption of contaminated meat products [9]. Routine assessment of patterns of emerging antibiotic resistant Salmonella strains is of principal importance because such information channeled to physicians and veterinarians help to timely redirect drug use so as to diminish the development and spread of resistance.

Food animals harbour a wide range of Salmonella serotypes and so act as source of contamination, which is of paramount epidemiological importance in non-typhoid human salmonellosis. Salmonella infection in meat animals, including cattle, swine and sheep, arises from intensive rearing practices and the use of contaminated feeds [10]. Contamination of meat by Salmonella may occur at abattoirs from the excretion of symptomless animals, contaminated abattoir equipment, floors and personnel and the pathogen can gain access to meat at any stage during butchering. Cross-contamination of carcasses and meat products could continue during subsequent handling, processing, preparation and distribution [11]. Contamination of equipment, utensils and hands of workers can spread Salmonella to uncontaminated carcasses and parts, which can occur in
subsequent handling, processing, transport, storage, distribution and preparation for consumption [10]. Despite the presence of many studies on the prevalence and antimicrobial susceptibility pattern of *Salmonella* in different parts of Ethiopia, so far nothing has been done in Wolaita where cattle meat is commonly consumed raw. The objectives these studies are;

- To isolate *Salmonella* and determine prevalence in slaughtered cattle
- To determine antimicrobial pattern of *Salmonella* isolated from slaughtered cattle at Wolaita Sodo municipal abattoir.

**MATERIALS AND METHODS**

**Materials and method**

**Study Area**

The study was conducted in Wolaita Sodo Municipal abattoir, Wolaita zone was found in Southwest part SNNPR of Ethiopia, and 390km from Addis Ababa following the tarmac road that passes through Shashamane to Arbaminch. Alternatively, it is located 330km Southwest of Addis Ababa following the tarmac road that passes through Hosanna to Arbaminch. Wolaita Sodo is town of the zone. It has total area of 4,541 km² and is composed of 12 Woredas and 3 registered towns. It is approximately 2000 meters above sea level and its altitude ranges from 700-2900 meters. The area is divided into three ecological zones: Kola (lowland <1500m), Woina Dega (mid-altitude 1500-2300m) and Dega (highland > 2300m). Most of the area lies within the mid altitude zone. Rainfall is bimodal, with an average amount of about 1000mm (lower in the lowlands and higher in the highlands). Mean monthly temperature vary from 26°C in January to 11°C in August. Soils (mainly Vertisols and Nitosols) vary in pH from 5-6. Primary occupation of the zone is farming. Mixed crop-livestock production predominates, but there are some pastoralists in the lowlands. Generally, the climatic condition is conducive to livestock production. The livestock population in the area is estimated to be 68,900 cattle, 1992 sheep, 382 goats, 121 horses, 131 mules, 488 donkeys and 55,191 [12].

**Study animals**

The study was conducted on 100 apparently healthy slaughtered cattle at Wolaita Sodo municipality abattoir from November, 2015 to April, 2016. The animals were originated from different agro-ecological zones which have different management system. Animals were both local and cross breed cattle kept under extensive and semi intensive farming systems. Even though, the study animals were kept under broad range of management, animals in most of the rural areas were kept to graze pasture on grassland and supplementary feedings of crop residue when pasture is scarce especially during long dry season. In Sodo town, semi intensive management system is practiced and animals were feed with concentrate and hay. The means of transport of animals to abattoir is by using trek and by their foot. After they were presented to the abattoir they were kept for an average 6hrs without feed and water in holding pens. Almost all the slaughtered animals were adult male cattle and majority of them were those which have finished traction life.

**Study design**

A cross-sectional observational study was undertaken in Wolaita Sodo municipal abattoir from November, 2015 to April, 2016 for isolation and identification of *Salmonella* from cattle. On each sampling day, usually once a week from a site, apparently healthy animals were randomly selected based on the number of slaughtered cattle in the abattoir and sampled. Carcass *Salmonella* status was considered as an output variable of interest at slaughter houses. The explanatory variables considered in slaughtered houses were *Salmonella* status of liver and intestinal content.

**Sampling**

The sample size was determined based on sample size determination in random sampling for infinite population using expected prevalence of *Salmonella* and desired absolute precision according to[13], as follows.

\[
N = \frac{1.96^2 \times \text{pexp} \times (1-\text{pexp})}{d^2}
\]

Where;

- \( N \) = required sample size
- \( \text{pexp} \) = expected prevalence
- \( d \) = desired absolute precision

The previous study on *Salmonella* in abattoirs at Debre Zeit which is almost agro ecologically similar area revealed an average prevalence of 7% [14]. Therefore using 7% expected prevalence and 5% absolute precision at 95% confidence level, the number of animals needed to estimate the prevalence of *Salmonella* in municipal abattoir of selected town of Wolaita Sodo was calculated 100.
Sampling procedure
Sample animals from abattoirs where selected randomly and systematically using the identification number given for the animals both for ante mortem and postmortem examination, depending on the number of animals slaughtered on each day, a number was drawn to fix the beginning of the sample and following slaughter order. Liver tissue, intestinal content and carcass swab from each selected slaughtered cattle were collected in separate containers.

Liver tissue (more than 25gm) was collected from each of the selected animals after evisceration. Intestine containing approximately 25gm of its content was collected in sterile universal bottle by cutting at both ends with scalpel blade. Separate sterile scalpel blades and sterile forceps were used for each and every cutting during sample collection.

Sterile cotton swabs moistened in 10ml bpw were rubbed on the both sides of the carcass from hindquarter to for quarters uniformly. Swabbing of carcass was conducted holding the sterile stick on opposite end of its tips. The swab samples were inserted in to the universal bottles containing bpw after cutting off the part the stick which was in contact with the hand, by binding out on the mouth of bottle. Carcass swab samples were collected at the end of slaughtering process before it is prepare for loading. At the end of each sample collection, every sampling bottle was labeled including date of sampling and the type of sample collected corresponding to animal identification number.

Sample processing
Twenty-five grams of liver was weighed, minced and added in to 225ml of BPW and was agitated. Twenty five grams of the intestinal content was weighed on sterile aluminum foil and put in sterile test tubes. About 225 ml BPW was added and agitated manually to disperse the content. Each carcass swab was agitated manually while in the original sterile universal bottle containing 10mlBPW.

Isolation and Identification
International organization for standardization (ISO) specifies food and animal feeding horizontal method for detection of salmonella through different successive stages as shown in figure 1 [15]. Therefore the isolation and identification was made based on this standard [16] had been used to complement media preparation and in identifying colonies. The bacteriological media used in different stages were prepared according to the manufactures recommendation.

Pre-enrichment in non-selective liquid media
Processed samples in appropriate amount of BPW (1:9) were incubated for 18±2h at 37\(^0\)c±1\(^0\)c.

3.5.2 Enrichment in selective liquid media
Enrichment in selective liquid media was done by transferring 0.1ml of culture obtained from non-selective pre-enrichment media to a tube containing 10ml of Rappaport Vassiliadis (RV) salmonella enrichment broth and 1ml to a tube containing 10ml of Selenite Cystine broth (SC). The incubated RV salmonella enrichment broth was incubated at 42\(^0\)c for 24± 3h, whilst SC broth was incubated at 37\(^0\)c for 24± 3h [15].

Plating out and identification
Xylose- Lysine Deoxycholate agar (XLD) (Hemedia laboratories pvt.ltd, Mumbai, India) and Brilliant green agar (BG) plates were used for plating out and identification purpose. A loop full of inoculums from SC broth and RV broth cultures was streaked on to XLD and BG agar plates and the inoculated plates were incubated at 37\(^0\)c for 24 ±3h. After proper incubation, plates were examined for the presence of typical salmonella colonies. Typical colonies of salmonella grow on XLD medium produce hydrogen sulphide (H2S) and have black H2S center and lightly transparent zone of reddish color due to the color change of the indicator [15] while on BG media plate appear red and impart a red/pink color to the surrounding agar. Other enteric typically appear green or yellow. Note the presence of typical Salmonella-like colines on BGA with a + in the record sheets.

Confirmation
For confirmation, at least five presumptive colonies were selected from every selective plating media. Whenever the suspected colonies on each plate were fewer than five, all the colonies were selected. The selected colonies were streaked on to the surface of pre-dried nutrient agar (Hemedia laboratories pvt.ltd, Mumbai, India) plates, in manner that allow well isolated to develop and incubated at 37\(^0\)c±1\(^0\)c for 24± 3h. Then the pure culture on nutrient agar was used for biochemical confirmation.

Biochemical test; pure culture obtained from nutrient agar were used for biochemical confirmation. Triple sugar iron agar (TSI) (Oxoid LTD, Basingstoke Hampshire, England) slants were inoculated from pure culture by streaking the slant and stubbing the butt and without flaming the wire lysine iron agar (LSI) (Difco, Becton Dickinson) was inoculated just below the surface and both tubes were incubated at 37\(^0\)c±1\(^0\)c for 24±3h.
loosely capped to maintain aerobic condition and to prevent excessive production of H2S production. Typical salmonella culture in TSI agar show alkaline (red) slants, and acid (yellow) butts with gas (bubbles) formation and (in about 90% of the cases) formation of H2S blackening of the agar. Alkaline reaction (purple color) both in the slant and butt superimposed with H2S after incubation indicates a typical positive reaction for salmonella in lysine iron agar.

Pure isolates were inoculated on Symons’s citrate agar (Hemedia laboratories pvt.ltd, Mumbai, India) by streaking the slant. Both of the inoculated tubes were incubated at 37°c±1°c for 24 ±3h [15] and change in the incubated media were interpreted for salmonella after the end of incubation following the guide line. Salmonella colonies on Simmons’s agar produce alkaline product using the medium as the sole Carbon source hence deep blue color indicates positive reactions.

The pure isolates were also inoculated in to tryptose soya broth TSI to determine the ability of an organism to spilt amino acid tryptophan to form the compound indole. For this an inoculated broth culture was incubated for 37°c 1°c for 24 h and kovac’s reagent was added. In positive reaction pink colored ring is formed on the top of broth due to reaction of indole with kovac’s reagent. However typical salmonella colonies due not hydrolyze amino acid tryptophan; therefore the medium remain unchanged after addition of appropriate reagent.

**Antimicrobial resistances test**

Isolates biochemically confirmed as salmonella were tested for their resistance to individual antimicrobial drugs by disc diffusion technique [17]. Four to five well isolated colonies grown on nutrient agar were transferred on to tubes containing 5ml of topic soy broth. The broth culture inoculated at 350c for 4h until it achieves or exceeds the 0.5 McFarland turbidity standard. For these tubes which exceeded the turbidity standard, adjustment were made by adding the sterile saline solution to obtain turbidity usually comparable to the standard.

With in 15 minute after adjusting turbidity of inoculums suspension, sterile swab immersed in each of dilution suspension and swabbed uniformly over surface of two plates of Muller Hinton agar (Oxoid LTD, Basingstoke Hampshire, England) for each inoculums. The plates were held at room temperature for 30 minutes to allow drying. Using sterile forceps, disc impregnated with known concentration of antimicrobials were dispensed on to the surface of Muller Hinton agar plates. Plates were incubated at 370c for 20 hrs and examine for zone of inhibition. The diameter of zone inhibition were recorded to the nearest millimeter, and classified as resistance intermediate, or susceptible according to published interpretive chart. The type of tested antimicrobials, their concentration in the discs and the zone of inhibition in deciding susceptibility are given.

<table>
<thead>
<tr>
<th>Antimicrobial agents and symbols</th>
<th>Disc contents(Zone diameter Nearest whole mm)</th>
<th>Resistance</th>
<th>Intermediate</th>
<th>Susceptible</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ampicillin (AMP)</td>
<td>10</td>
<td>≤ 13</td>
<td>14-16</td>
<td>≥ 17</td>
</tr>
<tr>
<td>Erythromycin (E)</td>
<td>5</td>
<td>≤ 13</td>
<td>14-22</td>
<td>≥ 18</td>
</tr>
<tr>
<td>Gentamycin (GCN)</td>
<td>10</td>
<td>≤ 12</td>
<td>13-14</td>
<td>≥ 15</td>
</tr>
<tr>
<td>Kanamycin(K)</td>
<td>30</td>
<td>≤ 13</td>
<td>14-17</td>
<td>≥ 18</td>
</tr>
<tr>
<td>Streptomycin(S)</td>
<td>10</td>
<td>≤ 11</td>
<td>12-14</td>
<td>≥ 15</td>
</tr>
<tr>
<td>Trimethoprim-sulfamethoxazole(SXT)</td>
<td>1.25/23.75</td>
<td>≤ 10</td>
<td>11-15</td>
<td>≥ 16</td>
</tr>
<tr>
<td>Amoxacillin(AML)</td>
<td>20</td>
<td>≤ 13</td>
<td>14-16</td>
<td>≥ 17</td>
</tr>
<tr>
<td>Tetracyclin(TE)</td>
<td>30</td>
<td>≤ 11</td>
<td>12-14</td>
<td>≥ 15</td>
</tr>
<tr>
<td>Chloramphenicol(C)</td>
<td>30</td>
<td>≤ 12</td>
<td>13-17</td>
<td>≥ 18</td>
</tr>
<tr>
<td>Cefoxitine(FOX)</td>
<td>30</td>
<td>≤ 14</td>
<td>15-17</td>
<td>≥ 18</td>
</tr>
<tr>
<td>Ciprofloxacine(CIP)</td>
<td>5</td>
<td>≤ 14</td>
<td>15-17</td>
<td>≥ 21</td>
</tr>
<tr>
<td>Nalixic acid</td>
<td>30</td>
<td>≤ 13</td>
<td>14-18</td>
<td>≥ 19</td>
</tr>
</tbody>
</table>

3.6. Data Analysis

All data were entered in to a Microsoft Excel spread sheet. After validation, the data were imported to STATA version 11 for windows (stata corp. College station, TX, USA) for descriptive analysis and association tests.

An animal in abattoir was considered positive when intestinal content or liver or both samples were culture positive for Salmonella.

4 RESULTS

The present study was conducted on 100 apparently healthy slaughtered cattle at Wolaita Sodo municipal abattoir from November, 2015 to April, 2016 to estimate prevalence and antimicrobial profile of Salmonella. Bacteriological examination was conducted on liver tissue, intestinal contents and carcass swab samples (each n=100).
4.1 Prevalence of Salmonella

In studying prevalence at animal level, the animal was considered Salmonella positive. When, it was bacteriologically positive either for one, two or liver tissue and intestinal content. The status of Salmonella on the carcass has been considered as indicator of contamination. However a sample was considered Salmonella positive when it was bacteriologically positive for Salmonella in determining sample prevalence.

Out of the total 100 animals examined for Salmonella 8% (8 of 100) were bacteriologically positive for Salmonella. Different level of detection of Salmonella was observed for different Salmonella positive samples. From 8 of Salmonella positive animals the six were positive from the intestinal content and the two were positive from liver tissue. Generally the animals were positive either from the liver or from the intestinal content not from both.

Of the total 300 samples taken from 100 animals 5.3% (16 of 300) were culture positive for Salmonella. Salmonella was detected from all sample types and there was considerable variation in frequency of isolation among sample types (Table 2.). However there was no significant difference between the prevalence in different samples (P>0.05)

<table>
<thead>
<tr>
<th>Sample types</th>
<th>Number of samples</th>
<th>Positive (%)</th>
<th>95%confidence interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Liver</td>
<td>100</td>
<td>2(12.5%)</td>
<td>0.8 -3.1</td>
</tr>
<tr>
<td>Intestinal content</td>
<td>100</td>
<td>6(37.5%)</td>
<td>3.5 - 9.0</td>
</tr>
<tr>
<td>Carcass swab</td>
<td>100</td>
<td>8(50%)</td>
<td>4.3 - 9.8</td>
</tr>
<tr>
<td>Total</td>
<td>300</td>
<td>16(5.3%)</td>
<td>2.3 – 6.7</td>
</tr>
</tbody>
</table>

The level of carcass contamination was considered as dependent variable taking the liver and intestinal content Salmonella status as a risk factor for carcass contamination. Therefore the association was assessed using logistic regression analysis but no association was found between the carcass contamination and any of other variables.

The highest sample prevalence 8% (8 of 100) was found on carcass swab which contributed 50% (8 of 16) of total isolates while the lowest prevalence 2% (2 of 100) was found on liver tissue samples contributed 12.5% (2 of 16) of total isolates.

4.2. Antibiotics susceptibility profile of Salmonella isolates

All of the isolates obtained from the study (n=16) were tested for eight different antimicrobials that were commonly used in human as well as animal treatment, and available in the market. 13 of the total 16 isolates (81.25%) were resistant to one or more of the tested antimicrobials.

All the isolates were susceptible to the antimicrobial effect of Ciprofloxacin, 93.75% to gentamycin, 87.5 % to sulphamethoxazinetrimethoprim and 62.5% to nalidixic acid. The isolates showed high resistance to ampicillin (56.25%), cefoxitin (43.75%), chloramphenicol (37.5%) and kanamycin (37.5%).

<table>
<thead>
<tr>
<th>Drug type</th>
<th>No of isolates</th>
<th>No of resistant isolates (%) &amp;95%CI</th>
<th>No of intermediate isolates (%)</th>
<th>No of susceptible isolates (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kanamycin (30µg)</td>
<td>16</td>
<td>6(37.5; 95%CI: 25.9-49.9)</td>
<td>2(12.5 ;95%CI:0.8-24.5)</td>
<td>8(50; 95%CI:37.6-61.3)</td>
</tr>
<tr>
<td>Nalidixic acid (30µg)</td>
<td>16</td>
<td>4(25; 95%CI:16.2-38.3)</td>
<td>2(12.5; 95%CI:0.8-24.5)</td>
<td>10(62.5; 95%CI: 48.5-72.7)</td>
</tr>
<tr>
<td>Gentamycin (10 µg)</td>
<td>16</td>
<td>1(6.25; 95%CI: 0.4-9.7)</td>
<td>16(100; 95%CI: 100-100)</td>
<td>1(6.25; 95%CI: 0.4-9.7)</td>
</tr>
<tr>
<td>Cefoxitin (30 µg)</td>
<td>16</td>
<td>7(43.75; 95%CI: 45.3-69.8)</td>
<td>3(18.75; 95%CI: 37.5-49.8)</td>
<td>6(37.5; 95%CI:30.1-54.6)</td>
</tr>
<tr>
<td>Chloramphenicol (30 µg)</td>
<td>16</td>
<td>6(37.5; 95%CI: 25.9-49.9)</td>
<td>1(6.25; 95%CI:0.8-24.5)</td>
<td>9(56.25; 95%CI: 43.8-68.3)</td>
</tr>
<tr>
<td>Sulphamethoxazine</td>
<td>16</td>
<td>1(6.25; 95%CI: 9.8-29.5)</td>
<td>16(100; 95%CI: 100-100)</td>
<td>14(87.5; 95%CI: 65.1-86.4)</td>
</tr>
<tr>
<td>Trimethoprim (25 µg)</td>
<td>16</td>
<td>9(56.25; 97%CI: 43.8-68.4)</td>
<td>2(12.5; 95%CI: 5.1-22.1)</td>
<td>5(32.25; 95%CI: 18.9-41.7)</td>
</tr>
</tbody>
</table>

5. DISCUSSION

The overall Salmonella prevalence estimated in this study (5.3%) was compared with Previous reported prevalence from Ethiopia and other countries. The overall prevalence of Salmonella in beef samples determined in this study was higher than the prevalence in Australia, 0.1% [18], Ethiopia, 4.2% [8], Wolaita Sodo abattoir was higher than Australia and Ethiopia, due to differences in the hygienic and sanitary practiced in those two abattoirs. The current study concerns about the municipal abattoir, that has poor sanitation and hygienic standard in comparison to the others abattoirs. In addition to this workers in the current abattoir were found to be with poor general and personal hygiene and lack of knowledge in hygienic processing of meat, due to lack of training regarding hygienic and sanitation of slaughtering and working environment generally and there was no disinfectants, hot water and separate room for final carcass and live animals in the abattoir. The overall high level of carcass contamination with Salmonella is of special public health significance for a country like Ethiopia, where raw and under cooked meat is the favorite
in most areas [19].

Wolaita Sodo abattoir lower prevalence than Ethiopia, 7.1%[14], the finding was in consonance with earlier studies made by [20] who reported 6% (18/300) prevalence of Salmonella in Addis Ababa abattoir worker, Nigeria, 25% [21], South Africa, 60%[22], Senegal, 43 and 87% at abattoir respectively [23]. The difference in the prevalence reported could be due to differences in study sites (abattoirs) and animal. In these studies, three sample types were examined and a higher percentage prevalence would probably have been obtained if other organs had been examined of the sample types taken from each animal during the study period, the mesenteric lymph nodes, carcass swab, and liver proved to be useful indicators of infection, as most of the bovine Salmonella-positive. Usually, healthy carriers intermittently excrete only a few Salmonellae, unless they undergo some kind of stress, for example during transport or holding in the lairage prior to slaughter.

The frequency of isolation of Salmonella varied between organs ranged from 12.5% to 50%. The specific prevalence of Salmonella from liver, intestinal content, and carcass swab collected from Wolaita Sodo abattoir were found to be 2 (12.5%), 6 (37.5%), and 8 (50%) respectively. The prevalence of Salmonella varied among the sampling organs. The hypothesis behind that the prevalence of Salmonella would be higher from carcass swab, medium of intestinal content and small on liver as more personnel and meat contribute for cross-contamination. In the present study, the overall prevalence of Salmonella in bovine carcass slaughtered at Wolaita Sodo municipal abattoir was found to be 5.3%. This study shows that 100 cattle carcasses were sampled, out of which 16 (5.3%) were contaminated by bacteria. The finding of this study revealed that cattle carcasses in Wolaita Sodo abattoir are contaminated with pathogenic Gram – negative bacteria of salmonellosis. Salmonella is among the most common agents of Foodborne illness in humans and have been isolated from beef and dairy cattle at all stages of production, but fecal shedding may be intermittent and difficult to detect, however the organism appears to be fairly spread throughout bovine population [24]. The low prevalence of salmonella in liver of bovine in this study supports other findings, indicating that localization of the organisms in this organ is most likely minimal. The liver is usually free of Salmonella at slaughter, but the surfaces can become contaminated during processing. The ultimate source of this contamination is likely to be the Salmonella present in the carcass swab and intestinal contents either of the same animal or of other animals slaughtered on the same day [25].

This in turn implies that the abattoir used for slaughtering process, abattoir personnel and butchers could have been contaminated to serve a source of contamination. The corrective action required include evaluation of cattle cleanliness, improving working procedure/instructions, retraining review of cleaning/disinfection materials and maintenance/cleaning equipment, improve supervision. The possible source of contaminates may be due to the unhygienic manner of handling meat in abattoirs, the environment upon which the meat is slaughtered as well the water [26].

Resistance to multiple antimicrobials (81.25%), which was observed in current study was Higher than other studies was conducted in Ethiopia. For instance [14] [27] reported 52%, 23.5% and 44.8% respectively. The multidrug resistance of Salmonella isolated from food of animal sources, animals and humans, as well higher than reports from elsewhere in the [28][29][30][31] and [32] reported multidrug resistance of Salmonella isolates respectively as follow: 16%, 50% (from raw meats), (1.2%, 14.1% and 23.7%) Salmonella isolated from different type of samples, 51.7% and 37.82%. This difference could be because of that, antimicrobial-resistant Salmonella are increasing due to the use of antimicrobial agents in food animals at sub-therapeutic level or prophylactic doses which may promote on-farm selection of antimicrobial resistant strains and markedly increase the human health risks associated with consumption of contaminated meat products [8] [33]and [34].

[34] reported that the isolates of Salmonella from food items and Workers from Addis Ababa were resistant to the commonly used antibiotics including Ampicillin. Furthermore, [9] also indicated resistance of Salmonella isolates to commonly used antimicrobials including ampicillin, and kanamycin, with resistance rate of 100%, and 33.3%, respectively. Similarly previous reports from South India [35] from Nigeria [36] and from Cameroon [37] indicated a similar 100%, over 90% and 100% respectively resistance to ampicillin. The result of the current research also indicated resistance of Salmonella isolates to commonly used antimicrobials including ampicillin, cefoxitin, chlorphenicol, and kanamycin with resistance rate of 56.25%, 43.75%, 37.5%, and 37.5% respectively. However, kanamycin was higher resistance rate than previous reports while ampicillin was lower resistance and resistance to further drugs as well as to Sulphamethoxazole trimethoprim and nalidixic acid with resistance rate of 6.25 % and 25% respectively was observed in this result. This difference could be due to the increasing rate of inappropriate utilization of antibiotics which favors selection pressure that increased the advantage of maintaining resistance genes in bacteria [38] [39] . It is as well recognized that recent resistance additions include resistance to Sulphamethoxazole trimethoprim. The continuing development of antibiotic resistance may lead to sufficient pressure ultimately to restrict the antibiotics available to the veterinary profession for animal treatment [40]. Moreover, this increase antibiotic resistance, in addition to public health problems, may lead to economic loss in the countries due to loss of exporting meat and animal products and cost of drug of choice to treat human and animals due to resistance development.
Ciprofloxacin showed a good antimicrobial activity against these Salmonella isolates. It was found that all of 16 (100%) isolates were susceptible to ciprofloxacin. This result was comparable with previous reports by [33] from central part of Ethiopia among isolates of sheep and goat meat, [36] from Nigeria, from human isolates and [9], isolates of Salmonella from dairy farms in Addis Ababa. The effectiveness of such drugs like ciprofloxacin could be because of that they are not widely used in countries like Ethiopia and other African countries [9]. In addition to this, effectiveness of this drug could be because of this drug is not well distributed to all societies and not simply prescribed rather than it is used as drug of choice in antibiotic resistant person. In addition to this, ciprofloxacin is not commonly used to treat animals in Ethiopia. Other good antimicrobial activity against salmonella isolates susceptible to gentamycin, Sulphamethohazole and nalidixic drugs were found 93%, 87% and 62% respectively.

6. CONCLUSION AND RECOMMENDATIONS

In present study 8% animal level prevalence of Salmonella in apparently healthy slaughtered cattle in wolaita sodo was observed. The study also revealed 5.3% over all prevalence of Salmonella in different sample types. Salmonellae were detected in liver tissue, intestinal content and carcass swab samples. The high prevalence of Salmonella in beef implies that consumption of undercooked and raw meat and meat products may lead the consumers to a risk of infection with the pathogen in areas like Wolaita where meat is consumed raw.

The majority of the Salmonellae isolates (81.25%) from the study were resistant to one or more of the tested antimicrobials. Based on the results and conclusions of this study, following recommendations are forwarded.

- Further detailed studies involving different sources and different types of food animals on the Contamination levels, diversity of serovars and antimicrobial resistance of Salmonella should be done.
- Personal hygiene and food sanitation should be practiced to prevent infection from different sources.
- Awareness should be created among public about the risks associated with consumption of raw and under cook meat for prevention of human salmonellosis.
- New legislations or public awareness campaigns should be maintained to decrease Salmonella incidence.

7. REFERENCES

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