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# Isolation, identification and antimicrobial susceptibility profile of *Salmonella* isolated from poultry farms in Addis Ababa, Ethiopia

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## Abstract

**Background:** *Salmonella* has been found to be the major cause of foodborne diseases and a serious public health problem in the world, with an increasing concern for the emergence and spread of antimicrobial-resistant strains.

**Method:** A cross-sectional study was conducted on poultry and poultry farms in Addis Ababa from February 2016 to April 2016 to determine the occurrence and evaluate the antimicrobial susceptibility profile of *Salmonella* isolates. A total of 200 samples consisting of the cloacal swab ($n = 168$), pooled litter ($n = 12$), hand swab ($n = 8$), pooled feed and water ($n = 12$) were collected separately from six poultry farms. The samples were examined for the presence of *Salmonella* through culturing on bacteriological media. Descriptive statistics, Pearson's Chi-square ($X^2$) and bivariate logistic regression were used in the analysis of the data. Accordingly, out of 200 samples, 23 (11.50%) were *Salmonella* positive, of which 21(12.50%) were in cloacal swab and 2(16.67%) in the pooled litter. There was no statistical association between the bacteriological status of sample type and *Salmonella* positivity ($X^2 = 3.07$, $P = 0.545$). However, there was a statistical association between farms and the rate of *Salmonella* isolation ($X^2 = 22.21$, $P \leq 0.00$). The antimicrobial susceptibility testing for *Salmonella* isolates was conducted following the Kirby–Bauer disc diffusion method (1961).

**Results:** Out of 23 samples tested, 95.65% of them were resistant to at least one or more antimicrobials. Multiple drug resistances were observed for 69.56% of *Salmonella* isolates. The highest resistance (73.9%) was observed in kanamycin followed by tetracycline (65.2%) and streptomycin (56.3%). gentamycin was the most effective antibiotic (95.7%; sensitivity) followed by ciprofloxacin (78.3% sensitivity) and ampicillin (69.6% sensitivity.

**Conclusion:** This current study finding indicated that further detailed epidemiological and molecular studies are essential on the frequency and sources of acquisition of resistant genes.

KEYWORDS
Addis Ababa, antimicrobial susceptibility, isolation, poultry, *Salmonella*

## 1 | INTRODUCTION

Foodborne pathogens are the most common problems of recent times and distributed across the world that had ultimately become a public health concern. Nowadays, individuals have been suffering from various foodborne diseases worldwide due to contaminated food and water consumption (Hendriksen et al., 2007; Majowicze et al., 2010). Among the pathogens, the genus *Salmonella* is considered the most prevalent foodborne pathogen globally and has been found to be the major zoonotic organism with a serious public health implication (Carrasco et al., 2012; Sánchez-Vargas et al., 2011). Foodborne 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 and lack of education for food handlers (WHO, 2004). Salmonellosis is an infectious and economically important disease of humans and animals caused by different *Salmonella* species (OIE, 2000). The ubiquity of *Salmonella* isolates makes a constant impurity hazard in all raw food items and is associated with occurrences of human salmonellosis (Carrasco et al., 2012; Tadesse, 2014). Foodborne *Salmonella* infection spread is recognised as the key reason for *Salmonella* contaminations, with many food sources and supplies implicated in these infections (Majowicz et al., 2010). *Salmonella* consists of mainly two species, namely, *Salmonella enterica* (*S. enterica*) and *S. bongori*. Furthermore, *S. enterica* was previously categorised into six subspecies upon biochemical tests and molecular characterisation. *Salmonella enterica* subspecies *enterica* is the major subspecies that is highly a distress as a result of virulent strains that could cause salmonellosis in animals and human beings. Food items that are derived from animal products, particularly poultry and poultry products, are frequently implicated in erratic circumstances and outbreaks of *Salmonella* infection in humans (Sánchez-Vargas et al., 2011). Previously FAO (2002), indicated that poultry and poultry products are a major foodborne infection vector and regularly among the prominent animal sources of human salmonellosis during food supply. The author has also indicated that human beings could acquire *Salmonella* infection through ingestion of raw or undercooked animal-derived food items, mainly poultry products. Isolation of *Salmonella* species in poultry foodstuffs is a worldwide public health distress including our country, Ethiopia. Infection with various species of *Salmonella* could result in diverse clinical manifestations such as inflammation of intestinal epithelia, diarrhoea, vomiting and typhoid fever (Crump & Mintz, 2010).

Various contamination levels have been reported by numerous studies worldwide. A study finding in Addis Ababa revealed a prevalence of 5.21% *Salmonella* isolates from eggs purchased from local markets (Bayu et al., 2013). A prevalence of 5.3% has been found from eggs shells from the open market purchased eggs in and around Haramaya Town (Kemal et al., 2016) and a prevalence of 18% from eggs in Gondar. The common exercise of using antimicrobial agents to human and animals to mitigate disease-causing agents play a crucial role in the emergence and antimicrobial-resistant bacteria that are successively transmitted to humans through the food chain. Moreover, antibiotics have a surprising ability to accelerate animal growth or growth promoter. Currently, more antibiotics are used in poultry, swine and cattle to promote growth and prevent disease than are used by the entire human population (FDA, 2015). Human salmonellosis is considered as a main bacterial root cause of foodborne diarrhoeal problem across the globe, and food items derived from animal origin act as a vehicle in the transmission of *Salmonella* species. Feed items such as water, faeces, poultry wastes, cages and litter contaminated with *Salmonella* are central sources of infection. Several research results showed the distribution of *Salmonella* in various sampling points in poultry surroundings or on antibiotic resistance, virulence and control strategies.

Discontinuation treatment and continuous undiscriminating uses of antimicrobials against diarrhoeal pathogens including *Salmonella* have presumptively assumed the possible causes for the development of antibiotic resistance and antimicrobial resistance is, therefore, a multi-sectorial problem encompassing the interface between humans, animals and the environment (Marshall & Levy, 2011). There is inadequate information available on the *Salmonella* isolated from poultry products in the country though the disease is nationally as well globally a concern and a leading zoonotic pathogen (Carrasco et al., 2012). Despite being a major zoonotic, foodborne and pathogen with high drug resistance along with diversity in its strains and host ranges, there is limited study on *Salmonella* in Ethiopia. Therefore, the purpose of this research work was to determine the occurrence of *Salmonella* from poultry and poultry farms and to evaluate the antimicrobial susceptibility profile of *Salmonella* isolates in Addis Ababa, Ethiopia.

## 2 | MATERIALS AND METHODS

### 2.1 | Study area

The study was conducted in Addis Ababa, which is located 2408 m.a.s.l and receives an annual mean rainfall of 1200 mm, with average minimum and maximum annual temperature of 9.4 and 23.2°C, respectively (National Metrological Service Agency, 2002). Based on the preliminary 2007 census results, Addis Ababa has a total population of 2,738,248, consisting of 1,304,518 men and 1,433,730 women. The city is fully urban, with no rural dwellers within the city's administrative boundaries. Addis Ababa contains 22.9% of all urban dwellers in Ethiopia. With an estimated area of 530.14 square km, this chartered city has an estimated density of 5165.1 inhabitants per square km (Central Statistical Agency of Ethiopia, 2008).

### 2.2 | Study population

The study populations were healthy chickens, farm attendants, feed and drinking water for chickens.

## 2.3 | Study design and sample collection

A cross-sectional study was conducted in this research. The samples were collected from February 2016 to April 2016. A total of 200 samples consisting of cloacal swab ($n = 168$), personnel hand swab from farm attendants ($n = 8$), pooled house litter ($n = 12$), pooled feed ($n = 6$) and pooled water ($n = 6$) were collected from chicken farms in Addis Ababa. Out of six poultry farms studied in this research, five farms consisted of layers, and one farm consisted of broilers. All farms were small-scaled and the farms comprised 257–400 layers and broilers: Farm A (257), Farm B (318), Farm C (400), Farm D (367), Farm E (339) and Farm F (349) and all chickens aged above 45 days old. The cloacal swab was collected from the cloaca of broiler ($n = 103$) and layer ($n = 65$). Pooled litter samples were taken directly from five areas (four from the corner and one from the middle) of each chicken house. Personnel hand swab samples were taken after the personnel were working and before washing their hand. Pooled water and feed samples were taken directly from drinking water and chicken feed available on each farm. Each sample was taken aseptically and put into a test tube containing pre-enrichment media (buffered peptone water [BPW]). The samples were transported after being collected in a portable container with ice packs (at 4°C) to the Microbiology Laboratory and were processed upon arrival or kept at +4°C overnight.

## 2.4 | Isolation procedure of *Salmonella*

The methods used in the isolation of *Salmonella* were according to the techniques recommended by the International Organisation for Standardisation (ISO 6579, 2002). The isolation involves three steps: pre-enrichment in pre-enrichment broth media, enrichment in selective media, plating on selective media and biochemical confirmation of suspected colonies from selective agar media.

### 2.4.1 | Pre-enrichment in non-selective broth medium

All samples were pre-enriched separately with an appropriate amount of BPW (CONDA) (1:9) and were incubated for 18–24 h at 37 ± 1°C.

### 2.4.2 | Enrichment in selective broth media

Tetrathionate broth base (Titan Biotech Ltd.) and Rappaport Vassiliadis *Salmonella* enrichment broth (Himedia MH1491) were used for selective enrichment of all samples. A portion (1 ml) of the pre-enriched culture was aseptically transferred to 10 ml of tetrathionate broth base containing test tube, and another 0.1-ml pre-enriched culture was aseptically transferred to test tubes containing 10 ml of Rappaport Vassiliadis *Salmonella* enrichment broth and incubated at 37 ± 1°C and 41.5 ± 0.5°C for 24 h, respectively.

### 2.4.3 | Plating out and isolation

Xylose Lysine Deoxycholate (XLD) agar (Oxoid CM0469) and brilliant green agar (BGA) base modified (Himedia M016) plates were used for plating out and isolation purpose. A loop full of inoculums from RV broth and tetrathionate broth was transferred and streaked onto the surface of XLD agar and BGA base modified separately. The plates were incubated at 37 ± 1°C for 18–24 h. After incubation, the plates were examined for typical and atypical colonies of *Salmonella*. On XLD agar, typical colonies can be colourless, very light, slightly shiny and transparent (colour of the medium) with a dark tinted centre, surrounded by a light red area and yellow edge, or of pink to red colour, with a black centre or without a black centre. Hydrogen sulphide ($H_2S$) (+) colonies are colourless or light pink with darker centres, and lactose (+) colonies are yellow or without the characteristic blackening; whereas on BGA, typical colonies are transparent, colourless or light pink, and the colour around colonies changes from pink to light red. For confirmation, presumptive *Salmonella* colonies were selected from every selective plating media and subcultured on nutrient agar (Oxoid CM0003) in a manner that allow isolated colonies to develop and incubate at 37°C for 18–24 h for further confirmation by biochemical tests.

## 2.5 | Biochemical confirmations

The biochemical identification of the organism was done by performing the biochemical tests. The biochemical tests were done as stated on Bergey's *Manual of Determinative Bacteriology*. Each identified colony with typical *Salmonella* morphology was confirmed biochemically by triple sugar iron (TSI) agar (Oxoid CM0277), Urease (Himedia M111A), Simmons' citrate agar (Himedia M099, India), Indole (Oxoid CM0129), lysine iron agar (LIA; Oxoid CM0579), methyl red (MR) and Voges–Proskauer (VP) (Himedia M070) tests. Colonies producing red slant (alkaline), yellow butt (acidic) on TSI agar with $H_2S$ production and bubbles formation/cracking at the butt (gas production), negative urea utilisation (yellow), positive citrate utilisation (deep blue slant), negative for indole production from tryptophan, positive LIA agar (alkaline slant/alkaline butt), positive for MR test and negative for VP test were considered *Salmonella* positive (ISO 6579, 2002). LIA (Oxoid CM0579) was used to demonstrate hydrogen sulfide production and the decarboxylation or deamination olysine. These *Salmonella* positive samples showed alkaline slant/alkaline butt. Isolates presumptive of *Salmonella* for all tests were cultured on nutrient agar (Oxoid CM0003) for antimicrobial susceptibility testing.

## 2.6 | Antimicrobial susceptibility test of *Salmonella* isolates

The antimicrobial susceptibility testing for *Salmonella* isolates was carried out following the Kirby–Bauer disc diffusion method on Mueller–Hinton agar (Oxoid CM0337) as described in the Clinical and

Laboratory Standards Institute (CLSI) guidelines (CLSI, 2013). From each isolate, biochemically confirmed well-isolated colonies grown on nutrient agar (Oxoid) were transferred into sterilised tubes containing 5 ml of tryptone soya broth (Oxoid). The broth culture was incubated at 37°C for 4 h until it achieved the 0.5 McFarland turbidity standard. A sterilised cotton swab was dipped into the suspension, and the bacteria were swabbed uniformly on the entire surface of Muller–Hinton agar plate. The plates were held at room temperature for 30 min to allow drying. Antibiotic discs with known concentrations of antimicrobial were applied aseptically onto the surface of the plates at an appropriate special arrangement with the help of a sterile pair of forceps on Mueller–Hinton agar plates. The plates were then inverted and incubated at 37°C for 24 h. The antibiotic discs (Oxoid) uses: ampicillin (AMP,10 $\mu$g), chloramphenicol (C,30 $\mu$g), tetracycline (TE,30 $\mu$g), streptomycin (S,10 $\mu$g), gentamycin (CN,10 $\mu$g), nalidixic acid (NA,30 g), ciprofloxacin (CIP,5 $\mu$g), cefoxitin (FOX,30 $\mu$g), kanamycin (K,30 $\mu$g) and sulphamethoxazole/trimethoprim (SXT,25 $\mu$g). After incubation, the diameter of clear zones produced by antimicrobial inhibition of bacterial growth was measured to the nearest mm using a transparent straight line ruler and classified as susceptible, intermediate and resistant categories according to CLSI guidelines (CLSI, 2013).

### 2.7 | Data management and analysis

The data were entered into Microsoft excel 2010 and analysed using the SPSS statistical software package version 20 (IBM SPSS statistics). Descriptive statistics were used to compute proportions and frequency distributions of the rate of *Salmonella* isolation among various sampling point. Pearson's Chi-square ($X^2$) test was used to assess the association in the positivity of *Salmonella* isolates from samples originating from poultry, poultry house litter and other sources. *Salmonella* isolates were further screened for susceptibility to 10 different drugs and classified as susceptible, intermediate and resistant using frequency and proportions. Multi-drug resistance (MDR) was considered if and only if one *Salmonella* isolate was resistant in three or more antimicrobial categories. A difference was taken as significant at $P < 0.05$ at 95% confidence interval for the variables analysed by $X^2$.

## 3 | RESULTS

### 3.1 | Frequency of isolation of *Salmonella*

Out of the total 200 samples collected for *Salmonella* bacteriological isolation, 23 (11.50%) were positive. Out of 23 *Salmonella* isolates, the distribution of the isolates with respect to sample type are described as follows: 21(12.50%) from cloacal swab and 2(16.67%) from pooled litter, there was no *Salmonella* isolated from personnel's hand swabs, pooled feed and pooled drinking water. This is to mean that *Salmonella* isolates were obtained from some sampling points such as cloacal swab and pooled litter. On the other hand, we could not get *Salmonella* isolates from the other sampling points like personnel's hand swabs, pooled feed and pooled drinking water. However, this *Salmonella* positivity among the various sampling points has no statistical association between these different sample types (sampling points) and *Salmonella* positivity ($X^2 = 3.79$, $P = 0.545$).

Conversely, among the poultry farms that were assessed for the occurrence of *Salmonella* bacterium, we have found that there was a statistical association between farms and the rate of *Salmonella* isolation ($X^2 = 22.21$, $P \leq 0.00$) as depicted in Table 1. In detail, more *Salmonella* isolates 29.79% ($n = 14$) and 9.30% ($n = 4$) were found in Farm C and Farm F, respectively, than the remaining farms. The possible reason for the variation of *Salmonella* occurrence among the poultry farms could be the hygienic status difference between farms and close vicinity to the main roads and parking. Moreover, relatively the farm's size was slightly high as compared to the others, and the number of visitors has increased to the farms. So, this could be one contributing factor for the occurrence of *Salmonella* bacterium. Some study findings also showed that chickens might be contaminated by the mechanical carriage of *Salmonella* on the wheels of vehicles or human footwear from areas of access to inside the premises.

*Note*: Among the sample types (sampling points) we have determined, cloacal swab samples were more positive 12.5% ($n = 12$) followed by pooled litter samples though no statistical association between sample type and *Salmonella* positivity was found ($X^2 = 3.07$, $P = 0.545$). As we mentioned, Farm C and F were positive even from pooled litter. Sample type: CS, cloacal swab; F, feed; PHS, personnel hand swab; L, litter and w, water. Poultry type: 0 = sample source other than poultry, b = Broiler and ly = layer.

### 3.2 | Antimicrobial susceptibility pattern of *Salmonella* isolated from different sample types

Antimicrobial susceptibility results of 23 *Salmonella* isolates were shown in Table 2. Gentamycin was the most effective antibiotic (95.7% sensitivity) followed by ciprofloxacin (78.3% sensitivity) and ampicillin (69.6% sensitivity). The highest resistance (73.9%) was observed to kanamycin followed by tetracycline (65.2%) and Streptomycin (56.3%). No intermediate resistance was seen in all isolates to gentamycin, ampicillin and kanamycin as indicated in (Table 3).

### 3.3 | Multiple drug resistance patterns of *Salmonella* isolated from different sample sources

Of the 23 isolates, 69.56% showed resistance for three or more of the antimicrobials tested, and all 23(100%) were resistant to one or more antimicrobials. These MDRS are summarised in Table 3.

## 4 | DISCUSSION

The main rationale of taking samples from different sampling points (poultry house elements) of the poultry farm is to look at the

**TABLE 1** The rate of *Salmonella* occurrence from different sampling points

| | | Positive | Negative | Total | Proportion (%) | Chi-square ($X^2$) | *P*-value | 95% CI | |
|---|---|---|---|---|---|---|---|---|---|
| Farm name | A | 2 | 18 | 20 | 10.00 | 22.21 | 0.00 | 8.61 | 11.39 |
| | B | 1 | 23 | 24 | 4.17 | | | 3.35 | 4.98 |
| | C | 14 | 33 | 47 | 29.79 | | | 28.23 | 31.35 |
| | D | 0 | 33 | 33 | 0.00 | | | – | – |
| | E | 2 | 31 | 33 | 6.06 | | | 5.22 | 6.90 |
| | F | 4 | 39 | 43 | 9.30 | | | 8.39 | 10.21 |
| Total | | 23 | 177 | 200 | 11.50 | | | 11.03 | 11.97 |
| Sample type | CS | 21 | 147 | 168 | 12.50 | 3.79 | 0.545 | 11.97 | 13.03 |
| | F | 0 | 6 | 6 | 0.00 | | | – | – |
| | L | 2 | 10 | 12 | 16.67 | | | 14.36 | 18.98 |
| | PHS | 0 | 8 | 8 | 0.00 | | | – | – |
| | W | 0 | 6 | 6 | 0.00 | | | – | – |
| Total | | 23 | 177 | 200 | 11.50 | | | 11.03 | 11.907 |
| Poultry type | 0 | 2 | 30 | 32 | 6.25 | 2.273 | 0.321 | 5.38 | 7.12 |
| | B | 15 | 87 | 102 | 14.71 | | | 13.96 | 15.45 |
| | Ly | 6 | 60 | 66 | 9.09 | | | 8.36 | 9.82 |
| Total | | 23 | 177 | 200 | 11.50 | | | 11.03 | 11.97 |

distribution of *Salmonella* bacterium among the sampling points in particular and poultry farms in general. As a result, we found variations in the distribution of *Salmonella* isolates, but the variations were not statistically significant. Moreover, having detailed information about the distribution of this zoonotic bacterium on the various sampling points will ultimately help us to know the remarkable health problems to these people who engage in poultry farms that have not followed strict biosecurity protocols and also aid in taking measures targeting in the mitigation of the problem based of the magnitude of the bacterium in these different sampling points. In our current study finding, from the total of 200 different samples from poultry farms examined for *Salmonlla*, 11.50% (23/200) were positive, of which 12.50% (21/168) were from cloacal swab, and 16.67% (2/12) were from pooled house litter. This result was in agreement with the findings of Marianne et al. (2007) who reported 11.8% *Salmonella* from apparently healthy poultry imported to Denmark for slaughter. In contrast, a report from Pakistan by Aijaz et al. (2010) indicated that 38% of *Salmonella* was isolated from poultry meat, which was higher than the results of our current study. This may be due to the difference in geographical and poultry management conditions, like housing, feeding, and so forth. *Salmonella* was not isolated from pooled feed samples. This finding disagrees with Maung (2004) who reported 0.6% of *Salmonella* isolated from poultry feed sources. This may be due to the low number of our sample size, the absence of rodents in the farms, which are main contaminants of feed, or heat treatment of the feed. In the present study, the rate of isolation of *Salmonella* from different farms showed a statistically significant difference ($X^2 = 22.21$, $P \leq 0.00$). This may be related to the difference in hygienic status among farms and close vicinity to roads and parking. Rose et al. (2000) reported an increased risk of *Salmonella* contamination associated with trucks running and parking in close vicinity to the poultry houses. It is thus possible that chickens might be contaminated by mechanical carriage of *Salmonella* on the wheels of vehicles or human footwear from areas of access to inside the premises.

The occurrence of *Salmonella* in the broiler (17.71%) in this study was slightly higher than the findings of Ibrahim et al. (2013) who reported 16.67% isolates of *Salmonella* from commercial broiler farms in Egypt. However, our findings are lower than those detected by Caldwell et al. (1995) (18.89%). Regarding the occurrence of *Salmonella* in layers, the isolation rate (9.09%) was to some extent in agreement with the data obtained by Ibrahim et al. (2013) who demonstrated that *Salmonella* was isolated with a percentage of 9.01% from layer farms. The high level of *Salmonella* isolation in broilers evaluated in this study may be attributed to ingestion of contaminated feeds, water or litter or using contaminated equipment (Gast, 2003).

All the *Salmonella* isolates were tested against a panel of 10 antimicrobials available at the local market. Out of 23 isolates, all (100%) of them were resistant to one or more antimicrobials tested. Accordingly, higher antimicrobials resistance of 73.9%, 65.2% and 47% was observed against kanamycin, streptomycin and nalidixic acid. Resistance to tetracycline was observed in 65.2% of the isolates, which is higher than that reported in different studies: 46.6% in Senegal (Bada-Alambedji et al., 2006) and 36% in Portugal (Antunes et al., 2003). Tetracycline has been one of the most commonly used antibiotics for production animals; from day-old chicks to broiler chickens, they are exposed to antimicrobial drugs during their growth phase. Therefore, resistance to drugs such as tetracycline could be expected since the members of this class (chlortetracycline and oxytetracycline) are approved for use in broiler feeds for the purpose of growth

**TABLE 2** Multiple drug resistance patterns of *Salmonella* isolated from different sample sources

| No. antimicrobial resistance | Antimicrobials | No. of *Salmonella* isolates (%) |
|---|---|---|
| One | TE | 3 (13.04) |
| | S | |
| | K | |
| Two | TE-NA | 3 (13.04) |
| | TE-K | |
| | NA-K | |
| Three | TE-SXT-K | 3 (13.04) |
| | AMP-K-SXT | |
| | AMP-SXT-S | |
| Four | TE-AMP-S-K | 4 (17.39) |
| | TE-AMP-K-SXT | |
| | TE-FOX-S-K | |
| | NA-S-K-C | |
| Five | TE-FOX-S-K-C | 5 (21.74) |
| | TE-NA-FOX-S-K (2) | |
| | TE-AMP-NA-K-SXT | |
| | TE-NA-S-K-C | |
| Six | NA-FOX-S-K-SXT-C | 2 (8.70) |
| | CN-TE-AMP-S-K-SXT | |
| Seven | TE-NA-CIP-S-K-SXT-C | 1 (4.35) |
| Eight | TE-AMP-NA-FOX-S-K-SXT-C | 1 (4.35) |
| Nine | TE- CIP- AMP- FOX-S-K-SXT | 1 (4.35) |
| Overall | | 23 (100) |

*Note*: We say multi-drug resistance, if and only if *Salmonella* isolates are resistant ≥ 3 antimicrobials.
Abbreviations: AMP, ampicillin; C, chloramphenicol; CIP, ciprofloxacin; CN, gentamycin; FOX, cefoxitin; NA, nalidixic acid; No, number;
S, streptomycin; K, kanamycin; SXT, sulphamethoxazole/trimethoprim; TE, tetracycline.

**TABLE 3** Antimicrobial sensitivity test results of *Salmonella* isolates

| Type of antimicrobial | Status of antimicrobial sensitivity | | |
|---|---|---|---|
| | Resistant (%) | Intermediate (%) | Sensitive (%) |
| CN | 1 (4.3%) | 0 (0.0%) | 22 (95.7%) |
| TE | 15 (65.2%) | 3 (13.0%) | 5 (21.7%) |
| AMP | 7 (30.4%) | 0 (0.0%) | 16 (69.6%) |
| NA | 11 (47.8%) | 2 (8.7%) | 10 (43.8% |
| FOX | 6 (26.1%) | 5 (21.7%) | 12 (52.2%) |
| CIP | 1 (4.3%) | 4 (17.4%) | 18 (78.3%) |
| S | 13 (56.3%) | 5 (21.7%) | 5 (21.7%) |
| K | 17(73.9%) | 0 (0.0%) | 6 (26.1%) |
| SXT | 9 (39.1%) | 3 (13.0%) | 11 (47.8%) |
| C | 7 (30.4%) | 4 (17.4%) | 12 (52.2%) |

Abbreviations: AMP, ampicillin; C, chloramphenicol; CIP, ciprofloxacin; CN, gentamycin; FOX, cefoxitin; NA, nalidixic acid; S, streptomycin; K, kanamycin; SXT, sulphamethoxazole/trimethoprim; TE, tetracycline.

promotion according to reports by Jones and Ricke (2003). Resistance to streptomycin (56.3%) was also higher and is in conformity with other findings (Cardoso et al., 2006). This resistance to tetracycline and streptomycin commonly observed among the *Salmonella* isolates has been frequently reported; this elevated resistance may be explained by the possible diffusion of the *tet*(A) resistance gene observed in an epidemiological study with *Salmonella* strains isolated from animals (Pezzella et al., 2004).

Of 23 isolates, all 100% (23/23) and 78.3% (18/23) showed high susceptibility to gentamycin and ciprofloxacin, respectively. This was lower than with the findings of Begum et al. (2010), who reported (100%) of *Salmonella* strains isolated from chicken eggs, intestines, and environmental samples were susceptible to gentamycin and ciprofloxacin. This investigation indicates that *Salmonella* isolates becoming resistant to these antimicrobials. Our findings regarding resistance to kanamycin (73.9%) were completely contrasted with the reports of Carraminana et al. (2004) who reported (2.8%) found in *Salmonella* isolated from a poultry slaughterhouse in Spain.

A total of 15 different multiple drug resistance patterns were observed: one isolate resistance to eight antimicrobials, one isolate resistance to seven antimicrobials, two isolates resistant to six antimicrobials with two different resistance patterns, five isolates resistance to four antimicrobials with four different resistance patterns, four isolates resistance to four antimicrobials with four different resistance patterns and three isolates resistant to three antimicrobials with three different resistance patterns. Moreover, the result of the current study revealed intermediate resistance of commonly used antibiotics including streptomycin, tetracycline and sulphamethoxazole/trimethoprim. This implies *Salmonella* is not inhibited by the usual achievable concentration of the antimicrobials with a normal dosage schedule as reported by Abebe et al. (2013).

## 5 | CONCLUSION

In general, from this cross-sectional study, it can be concluded that the occurrence of *Salmonella* from poultry farms in Addis Ababa was 11.50%. This result is significantly high to be a potential source of foodborne salmonellosis putting human health at risk via the food chain. The results of the present study indicate single or multiple resistance of *Salmonella* isolated from poultry, which constitutes a potential source of transmission of these resistant strains to man and poses a problem in public health. Resistance was mainly observed to kanamycin followed by tetracycline and streptomycin, whereas gentamycin and ciprofloxacin seem still effective to be used. Therefore, a detailed epidemiological survey has to be made to determine the sources of contamination and associated risk factors in poultry and poultry farms. Moreover, molecular studies are essential on the frequency, sources

of acquisition of resistant genes and distribution of antimicrobial resistant *Salmonella* among food animals, food products and humans in Ethiopia to apply appropriate measures to minimize *Salmonella* infection.

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## ETHICAL STATEMENT

Written ethical approval and consent for this study was obtained from Addis Ababa University, College of Veterinary Medicine of Animal Research Ethics and Review committee. Oral consent was also obtained from the owners to take samples from their chicken and for further research use of the samples.

## CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest regarding the publication of this paper.

## AUTHOR CONTRIBUTION

*Conceptualisation, methodology, writing–review and editing*: Yasin Mohammed.

Teshager Dubie: Contributed to data collection, data analysis, interpretation of data, writing and editing of the manuscript.

## DATA AVAILABILITY STATEMENT

The data sets used and/or analysed during the current study could be available from the corresponding author on reasonable request. The method has been described in detail and is reproducible. All relevant results obtained were presented and discussed.

## PEER REVIEW

The peer review history for this article is available at https://publons.com/publon/10.1002/vms3.762

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## REFERENCES

Abebe, A. Rakshit, K., & Anal, K. (2013). Genotypic and phenotypic characterization of antimicrobial resistance patterns of *Salmonella* strains isolated from raw milk in Sebeta, Ethiopa. *International Journal of Advance Life Science*, *6*, 192–199.

Aijaz, H. S., Muhammad, K., Muhammad, B. B., Ghiasuddin, S., Azizullah, M., & Parkash, D. (2010). Prevalence and antimicrobial resistance of *Salmonella* serovars isolated from poultry meat in Hyderabad, Pakistan. *Turkish Journal of Veterinary and Animal Sciences*, *34*, 455–460.

Antunes, P., Reu, C., Souza, J. C., Peixe, L., & Pestana, N. (2003). Incidence of *Salmonella* from poultry products and their susceptibility to antimicrobial agents. *International Journal of Food Microbiology*, *82*, 97–103.

Bada-Alambedji, R., Fofana, A., Sedi, M., & Akakpo, A. J. (2006). Antimicrobial resistance of *Salmonella* isolated from poultry carcasses in Dakar (Senegal). *Brazilian Journal of Microbiology*, *37*, 510–515.

Bayu, Z., Asrade, A., Kebede, N., Sisay, Z., & Bayu, Y., (2013). Identification and characterization of *Salmonella* species in whole egg purchased from local markets in Addis Ababa, Ethiopia. *Journal of Veterinary Medicine and Animal Health*, *5*(5), 133–137.

Begum, K., Reza, T. A., Haque, M., Hossain, A., Hassan, F. M. K., Hassan, S. N., Akhter, N., Ahmed, A., & Barua, U. (2010). Isolation, identification and antibiotic resistance pattern of *Salmonella* spp. from chicken eggs, intestines and environmental samples. *Bangladesh Pharmaceutical Journal*, *13*, 23–27.

Caldwell, D. J., Hargis, B. M., Corrier, D. E., Vidal, L., & Deloach, J. R. (1995). Evaluation of persis-tence and distribution of *Salmonella* serotype isolation from poultry farms using drag–swab sampling. *Avian Diseases*, *39*, 617–621.

Cardoso, M. O., Ribeiro, A. R., Santos, L. R., Pilotto, F., Moraes, H. L. S., Salle, C. T. P., Rocha, S. L. S., & Nascimento, V. P. (2006). Antibiotic resistance in *Salmonella enteritidis* isolated from broiler carcasses. *Brazilian Journal of Microbiology*, *37*, 368–371.

Carraminana, J. J., Rota, C., Agustin, I., & Herrera, A. (2004). High prevalence of multiple resistance to antibiotics in *Salmonella* serovars isolated from a poultry slaughterhouse in Spain. *Veterinary Microbiology*, *104*, 133—139.

Carrasco, E., Morales-Rueda, A., & Garcťıa-Gimeno, R. M., (2012). Cross-contamination and recontamination by *Salmonella* in foods: A review. *Food Research International*, *45*(2), 545–556.

Clinical and Laboratory Standards Institute (CLSI). (2013). *Performance standards for antimicrobial susceptibility testing; twenty third informational supplement, CLSI document M100-S23*. Clinical and Laboratory Standards Institute.

Crump, J. A., Mintz, E D. (2010). Global trends in typhoid and paratyphoid fever. *Clinical Infectious Diseases*, *50*(2), 241–246. https://doi.org/10.1086/649541

CSA (Central Statistical Agency of Ethiopia) (2008). *Report on livestock and livestock characteristics (II: 583)*. Statistical Bulletin.

FAO (2002). Microbiological risk assessment series no. 1 risk assessment of Salmonella in eggs and broiler chickens. Interpretative Summary. *World Health Organization*.

Food and Drug Administration (FDA). (2015). *Drugs@FDA data files*. www.fda.gov/Drugs/InformationOnDrugs/ucm079750

Gast, R. K. (2003). Paratyphoid infections: Diseases of poultry, 11th ed. Iowa State University Press, Ames., 583–613.

Hendriksen, R., Vieira, A. R., Karlsmose, S., Lo Fo Wong, D. M. A., Jensen, A. B., Wegener, H. C., & Aarestrup, F. M. (2007). Global monitoring of *Salmonella* serovar distribution from the World Health Organization global foodborne infections network country data bank: Results of quality assured laboratories from 2001 to 2007. *Foodborne Pathogens and Disease*, *8*(8), 887–900.

Ibrahim, M. A., Emeash, H. H., Ghoneim, N. H., & Abdel Halim, M. A. (2013). Seroepidemiological studies on poultry salmonellosis and its public health importance. *Journal of World's Poultry Research*, *3*, 18–23.

Jones, F. T., & Ricke, S. C. (2003). Observations on the history of the development of antimicrobials and their use in poultry feeds. *Poultry Science*, *82*, 613–617.

Kemal, J., Berhanu, S., Sissay, M., Desta, B., (2016). Prevalence, assessment, andantimicrobial resistance patterns of Salmonella from raw chicken eggs in Haramaya, Ethiopia. *The Journal of Infection in Developing Countries*, *10*(11), 1230–1235.

Majowicz, S. E., Musto, J., Scalene, E., Angulo, F. J., Kirk, M., O'Brien, S. J., Jones, T. F., Fazil, A., Hoekstra, R. M., & International Collaboration on

Enteric Disease 'Burden of Illness' Studies (2010). The global burden of non-typhoidal *Salmonella* gastroenteritis. *Clinical Infectious Diseases*, *50*(6), 882–889.

Majowicze, E., Musto, J., Scallan, E., Angulo, F. J., Kirk, M., O'Brien, S. J., Jones, T. F., Fazil, A., Hoekstra, R. M., & International Collaboration on Enteric Disease 'Burden of Illness' Studies. (2010). The global burden of non-typhoidal *Salmonella* gastroenteritis. *Clinical Infectious Diseases*, *50*(6), 882–889.

Marianne, N. S., Andersen, J. S., Aabo, S., Ethelberg, S., Aarestrup, F. M., Sørensen, A. H., Sørensen, G., Pedersen, K., Nordentoft, S., Olsen, K. E. P., Gerner-Smidt, P., & Baggesen, D. L., (2007). Antimicrobial drug resistance of *Salmonella* isolates from meat and humans, Denmark. *Emerging Infectious Diseases*, *13*(4), 638–641.

Marshall, B. M., & Levy, S. B., (2011). Food animals and antimicrobials: impacts on human health. *Clinical Microbiology Review*, *24*, 718–733.

Maung, S. M. (2004). Epidemiology of *Salmonella* contamination of poultry meat products. Knowledge gaps in the farm to store products.

National Metrological Service Agency (2002). Year book bulletin.

Office International des Épizooties (OIÉ), (2000). Salmonellosis. In OIE Review committee, *Manual of standards for diagnostic tests and vaccines* (pp. 832–842). OIÉ.

Pezzella, C., Ricci, A., Di Giannatale, E., Luzzi, I., & Carattoli, A. (2004). Tetracycline and streptomycin resistance genes, transposons, and plasmids in *Salmonella enterica* isolated from animals in Italy. *Antimicrobial Agents Chemotherapy*, *48*, 903–908.

Rose et al. (2000). Risk factors for Salmonella persistence after cleansing and disinfection in French broiler-chicken houses, Prev. *Vet. Med.*

Sánchez-Vargas, F. M., Abu-El-Haija, M. A., & Gt'omez-Duarte, O. G., (2011). *Salmonella* infections: An update on epidemiology, management, and prevention. *Travel Medicine and Infectious Disease*, *9*(6), 263–277.

Sánchez-Vargas, F. M. Abu-El-Haija, M., & Gómez-Duarte, O., (2011). *Salmonella* infections: An update on epidemiology, management, and prevention. *Travel Medicine and Infectious Disease*, *9*(6), 263–277.

Tadesse, G., (2014). Prevalence of human Salmonellosis in Ethiopia: A systematic review and meta-analysis. *BMC Infectious Diseases*, *14*, 88.

World Health Organization (WHO), (2004). *Regional office for Africa "developing and maintaining food safety control systems for Africa current status and prospects for change"*.. Second FAO/WHO Global Forum of Food Safety Regulators, Bangkok, Thailand (pp. 12–14).

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