

Prevalence and Antimicrobial Resistance of *Salmonella* from Poultry, Poultry Meat, Eggs and Farm Workers

¹Mezene Woyessa, ²Gezahegn Mamo, ³Balako Gumi, ²Bersissa Kumsa, ⁴Boka Tesfaye, ⁴Takele Sori and ⁵Getahun E. Agga

¹School of Veterinary Medicine, Wollega University, P.O. Box: 395, Nekemte, Ethiopia

²College of Veterinary Medicine and Agriculture, Addis Ababa University, P.O. Box: 34, Bishoftu, Oromia, Ethiopia

³Aklilu Lemma Institute of Pathobiology, Addis Ababa University, P.O. Box: 1176 Addis Ababa, Ethiopia

⁴College of Agriculture and Veterinary Medicine, Jimma University, P.O. Box: 307, Jimma, Ethiopia

⁵U.S. Department of Agriculture, Agricultural Research Service, Food Animal Environmental Systems Research Unit, 2413 Nashville Road, B-5 Bowling Green, KY 42101, USA

Abstract: *Salmonella* is the second major cause of foodborne bacterial infections in humans worldwide and poultry is a major source of infection. Drug resistant *Salmonella*, such as quinolone and 3rd and higher generation cephalosporin resistant strains, are regarded by World Health Organization as a critically important highest priority pathogen. Studies from developing countries like Ethiopia are scarce. We conducted a cross sectional study to determine the prevalence and antimicrobial resistance of *Salmonella* in intensive and semi-intensive commercial and backyard poultry farms, poultry meat and eggs and farm workers. The prevalence of *Salmonella* was 18.4% in the fecal samples, 14.8% in the hand swabs of farm workers, 4.5% of eggs and 6% of meat samples. The highest prevalence was observed in intensive production system (16.9%) and the lowest was found in backyard scavenging system (7.4%). Risk factors such as farm type ($P=0.006$), production type ($P=0.001$), breed ($P=0.005$) and sample type ($P=0.001$) were significantly associated with *Salmonella* prevalence. *Salmonella* isolates (n=37) were tested for their resistance against 15 antimicrobials using disc diffusion method. Majority of the isolates (24/37) were resistant or intermediately resistant to at least one antimicrobial. The prevalence of resistance was high to chloramphenicol (62.2%), tetracycline (59.5%), ampicillin (54.1%) and streptomycin (51.4%). More than half of the isolates (56.8%) were multidrug resistant. Our results showed a widespread occurrence of drug resistant *Salmonella* in poultry farms in the study area. Measures to control of *Salmonella* infections in poultry are needed to reduce foodborne infections in humans.

Key words: Antimicrobial resistance • Egg • Poultry • *Salmonella*

INTRODUCTION

Salmonellosis represents a serious threat to public health resulting in considerable economic consequences in many parts of the world [1]. The emergence of antimicrobial resistant *Salmonella* and other enteric pathogens has become a major concern [2]. The use of antimicrobials for therapeutic purposes, disease

prevention and growth promotion in food animal production can potentially lead to a widespread dissemination of antimicrobial resistant bacteria [3, 4]. *Salmonellosis* is an important bacterial disease which affects diverse host species including poultry [5]. Poultry are important reservoirs for many zoonotic bacterial pathogens such as *Salmonella* [6]. The presence of *Salmonella* in healthy poultry is suggested as the main

risk factor for its transmission to humans through table eggs and poultry meat [7, 8]. The prevalence of *Salmonella* associated with poultry and poultry products is high and this high prevalence has both public health and economic implications [9]. The globalization of the food supply and the increased movements of people, animals and goods have increased the burden of *Salmonella* infections in several countries [10-12].

Currently, there is a rapidly growing commercial poultry production in Ethiopia [13]. The government of Ethiopia has prepared a five-year strategic development plan from 2015 to 2020 identifying the poultry production as an important sub-sector. The plan was to increase the total broiler production by 235% and the total egg production by 828% in 2020. The goal of the plan is to reduce poverty, improve household nutrition as well as increased income. Such transformative production systems may however lead to unrecognized risks to human health in terms of antibiotic use and antibiotic resistance as a result of emerging intensified production systems, which often rely on antibiotics as a stopgap for disease control and prevention in low and middle income countries in place of improving hygiene and sanitation in large-scale operations and other management inadequacies leading to increased exposure to bacterial pathogens [14]. Studies from different countries reveal that *Salmonella* serotypes isolated from foods of animal origin have multidrug resistance profiles [15]. The role of poultry products in the dissemination of antimicrobial resistant zoonotic bacterial pathogens also is well documented in developed countries [16]. A review made by Sylvia *et al.* [17] of 40 years (1974 and 2013) of enteric antimicrobial resistance research both in humans and animals in Eastern Africa summarized that despite the high burden of disease in sub-Saharan Africa and the potential health and economic consequences, the research output on antimicrobial resistance in the region was limited. Little data exists to quantify the contribution of different factors to the current levels of antimicrobial resistance. Local knowledge on the prevalence of *Salmonella*, serotype distribution and associated risk factors is important to implement appropriate control strategy to reduce wider dissemination of important zoonotic serovars. The objectives of this study were to investigate the prevalence and antimicrobial resistance of *Salmonella* in backyard and commercial poultry farms, farm workers, poultry meat and eggs in rapidly expanding poultry production areas of central Ethiopia.

MATERIALS AND METHODS

Relevance and Description of the Study Areas: Almost all large-scale commercial poultry farms in Ethiopia are found in East Shewa zone of Oromia regional state. In this zone, there are also an emerging intensive and semi intensive small-scale poultry farms in the urban and peri-urban areas. This activity is being undertaken as a source of income in Adama, Lome, Ada'a and Akaki districts. Most of egg and dressed poultry carcasses supply to supermarkets comes from commercial farms located in the major towns of these districts [18].

Sampling Methods, Sample Collection, Transportation and Storage: A multi-stage sampling procedure was used in which the East Shewa Zone and the four districts were purposively selected because of concentrated poultry production in the area, agro-ecological suitability and accessibility. Within each district for the backyard production system, we selected households. All commercial farms in the major cities of the districts were selected stratified by the size of the farms (small <1000 or large > 1000). For sample collection, ethical clearance certificate (Ref. No: VM/ERC/07/01/12/2020) was obtained from the College of Veterinary Medicine and Agriculture (Addis Ababa University). Written informed consents were obtained from each participant after informing the purpose of the study, the procedure, the risk, benefit and their rights.

Samples were collected according to the method described in ISO-17604 [19]. For chicken fecal samples, 25g pooled fresh feces were collected from the floor by sterile spatula into sterile universal bottle (Oxoid Ltd., London, England). Hand swabs were collected to check for direct transmission of *Salmonella* to farm workers. Swabs were done by rubbing the palm and fingers using pre-moistened swabs (Oxoid Ltd.) in 15 ml conical tubes (Oxoid Ltd.) containing 10ml of BPW. The egg samples were collected from the farms included in the study. Samples were marked with a permanent marker stating the date of collection, the farm and identification number for each egg. For the poultry meat, 25 g of tissues were collected from the commercial farms at slaughterhouses and placed into plastic freezer bags. Precautions were taken at all stages to ensure that the equipment used during sampling, transportation and storage to avoid any contamination. All types of samples were transported to laboratory in cooler containing icepacks and stored refrigerated at 4°C until processed.

Isolation and Identification of *Salmonella*: ISO protocol 6579: 2002 [20] was also used for the isolation and identification of *Salmonella*. Raw meat samples (25 g) were sliced into small pieces on sterile metal trays using sterile scalpel and forceps and placed into flasks containing 225 ml BPW and incubated for 24 hr at 37°C. Fecal samples (25 g) and the contents of individual eggs were placed into a sterile stomacher bag (Oxoid Ltd.) and 225 ml of BPW was added in 1:9 ratio and homogenized using a laboratory blender (Oxoid Ltd.) for 2 minutes and incubated for 24 hrs at 37°C. Hand swabs were transported in 10ml of BPW and incubated for 24 hrs at 37°C. Following incubation, 1 ml and 0.1 ml of the pre enrichment broths were aseptically transferred to 10 ml of tetrathionate broth (Oxoid Ltd.) and 10 ml of Rappaport-Vassiliadis (RV) broth (Oxoid Ltd.), mixed and incubated for 18 to 24 hr at 37°C and 42°C, respectively. Following incubation, a loopful of each broth culture was streaked onto xylose lysine deoxycholate (XLD) agar (Oxoid Ltd.) and brilliant green agar (BGA) (Oxoid Ltd.) and incubated at 37°C for 24 to 48 hrs. The XLD and BGA plates were examined for the presence of *Salmonella* colonies. Red colonies with black centers on XLD and pink colonies with a red zone on BGA plates were considered presumptive positive and further purified by culturing on nutrient agar (Oxoid Ltd.) [21].

For biochemical identification, presumptive *Salmonella* colonies were picked from the nutrient agar and inoculated into triple sugar iron (TSI) agar (Oxoid Ltd.), indol-ornithine agar (Oxoid Ltd) Simon's citrate agar (Oxoid Ltd.), urea broth (Oxoid Ltd.), MR-VP broth (Oxoid Ltd.) and incubated for 24 or 48 hours at 37°C [21]. Colonies producing an alkaline slant with acid (yellow color) butt on TSI with hydrogen sulfide production, positive for lysine (purple color), negative for urea hydrolysis (red color), negative for tryptophan utilization (indole test) (yellow-brown ring), negative for Voges-Proskauer and positive for citrate utilization were considered *Salmonella*-positive [22]. *Salmonella* colonies were tested for sero-grouping by rapid slide agglutination test using *Salmonella* polyvalent O antiserum "I" and "II"(Oxoid Ltd.) according to the manufacturer's instructions [23].

Antimicrobial Susceptibility Testing: Antimicrobial susceptibility was determined by Kirby-Bauer disc diffusion method based on the Clinical and Laboratory Standards Institute (CLSI) [24] with commercially available antibiotic discs. Refreshed pure isolate colonies from the

nutrient agar plates were transferred into tubes containing 5 mL of tryptone soya broth (Oxoid Ltd.). The broth culture was incubated at 37°C for 4 h until it achieved 0.5 McFarland turbidity standards. A sterile cotton swab was used to swab the inoculum uniformly over the surface of Muller Hinton agar plate (Oxoid Ltd.). Plates were held at room temperature for 30 min to dry. The pre-determined antibiotic disks (all from Oxoid Ltd.): norfloxacin (10µg), nalidixic acid (30µg), ciprofloxacin (5µg), amoxicillin-clavulanic acid (20/10µg), tetracycline (30µg), chloramphenicol (30µg), ampicillin (10µg), gentamicin (10µg), streptomycin (10µg), ceftazidime (30µg), cefotaxime (30µg), trimethoprim + sulfamethoxazole (1.25/23.75µg), meropenem (10µg), doxycycline (30µg) and tigecycline (15µg) were then dispensed into the bacterial lawn by means of a sterile pair of forceps and gently pressed to ensure complete contact with the agar. The discs were positioned 15 mm away from the edge of the plate and 25 mm away from each other. The plates were incubated at 35-37°C for 18 to 24 hrs. Zones of inhibition around the discs were measured to the nearest millimeter using a digital caliper, the size of the inhibition zone was compared with the disc manufacturer's standard and classified as sensitive (S), intermediate (I) or resistant (R) to the given drug. *Staphylococcus aureus* (ATCC 25923), *Enterococcus faecalis* (ATCC 29212), *Pseudomonas aeruginosa* (ATCC 27853) and *E. coli* (ATCC 25922) were used as control strains. *Salmonella* colonies that showed resistance to more than or equal to three different classes of antibiotics were considered multidrug resistant (MDR).

Data Analysis: The data was recorded in Microsoft Excel, cleaned, organized and sorted. Descriptive analysis was performed using percentages for each prevalence and resistance to each drug. A Chi-square test was used for initial analysis to determine differences in the proportions of positive between districts, breed, purpose of production, production setting and sample types. Multivariate logistic regression was used depending on the nature of the outcome measures. Significant statistical difference was considered at $P < 0.05$.

RESULTS

Prevalence of *Salmonella*: *Salmonella* was isolated from 92(18.4%) fecal samples, 4 (14.8%) hand swabs sampled from farm workers, 7(4.5%) eggs and 6(6.0%) meat samples. As indicated in Table 1, farm type, production

Table 1: The overall prevalence and risk factors for the occurrence of *salmonella* in poultry, products and farm workers

Risk factor		Sample size	Positive	Prevalence (%)	χ^2	P-value
Study district	Akaki	123	13	10.6	1.785	0.618
	Ada'a	339	44	12.9		
	Lome	116	21	18.1		
	Adama	202	31	15.3		
Farm type	Intensive	467	79	16.9	14.444	0.001
	Semi-intensive	125	16	12.8		
	Backyard	188	14	7.4		
Production type	Layers	313	64	20.4	7.466	0.024
	Broilers	279	31	11.1		
	Dual	188	14	7.4		
Breed	Exotic	606	96	15.8	5.340	0.021
	Local	174	13	7.4		
Sample type	Feces	500	92	18.4	24.696	0.000
	Hand swab	27	4	14.8		
	Egg	153	7	4.5		
	Meat	100	6	6		

Table 2: Associated risk factors for the occurrence of *Salmonella* in poultry by products, products and farm workers based on logistic regression analysis

Variables	Coef.	S.E.	Wald	df	P-value	OR	95% CI for OR	
							Lower	Upper
Farm type			6.134	2	0.047			
Intensive	0.329	0.303	1.18	1	0.277	1.39	0.77	2.52
Semi-intensive	0.809	0.342	5.579	1	0.018	2.25	1.15	4.40
Sample types			17.144	3	0.001			
Fecal sample	1.204	0.447	7.252	1	0.007	3.34	1.39	8.01
Hand swab	0.824	0.696	1.404	1	0.236	2.28	0.58	8.92
Egg sample	-0.154	0.591	0.068	1	0.795	0.86	0.27	2.73
Constant	-3.08	0.519	35.277	1	0	0.05		

Table 3: Fecal prevalence of *Salmonella* by study characteristics

Risk factor		Sample size(n)	Positive(#)	Prevalence (%)	χ^2	P-value
Study district	Akaki	76	11	14.5	2.217	0.529
	Ada'a	199	40	20.1		
	Lome	70	10	14.3		
	Adama	155	31	20		
Farm type	Intensive	288	51	17.7	6.577	0.037
	Semi-intensive	108	28	25.9		
	Backyard	104	13	12.5		
Production type	Layers	242	48	19.8	3.247	0.197
	Broilers	159	32	20.1		
	Dual	99	12	12.1		
Breed	Exotic	403	80	19.6	2.913	0.088
	Local	97	12	12.4		

type, breed and sample type were significantly ($P < 0.05$) associated with prevalence. *Salmonella* prevalence was significantly ($P = 0.001$) higher in the intensive farms (16.9%) than in the back yard farms (7.4%). The prevalence of *Salmonella* was significantly ($P = 0.024$) higher in layer chicken (20.4%) than in broiler chicken (11.1%) and it was twice in the exotic breeds (15.8%) as compared ($P = 0.021$) to local breeds (7.4%). Fecal sample

had significantly ($P = 0.000$) the highest (18.4%) prevalence than poultry products (5.13%) and farm workers (14.8%).

Using multivariable logistic regression analysis, farm type ($P = 0.047$) and sample type ($P = 0.001$) were significantly associated with *Salmonella* prevalence (Table 2). In this regard, semi-intensive farms had higher odds of *Salmonella* prevalence than the backyard

Table 4: Prevalence of *Salmonella* in chicken egg samples by study characteristics

Risk factor		Sample size(n)	Positive(#)	Prevalence (%)	χ^2	P-value
Study district	Akaki	44	1	2.3	3.486	0.357
	Ada'a	34	2	5.9		
	Lome	29	3	10.3		
	Adama	46	1	2.1		
Farm type	Intensive	61	1	1.6	8.855	0.022
	Semi-intensive	8	2	25		
	Backyard	84	4	4.8		
Production type	Layers	52	3	5.8	0.746	0.885
	Broilers	12	0	0		
	Dual	89	4	4.5		
Breed	Exotic	76	4	5.3	0.164	0.493
	Local	77	3	3.9		

Table 5: Antimicrobial resistance among *Salmonella* isolates

Antibiotics	Number (%)		
	Susceptible (%)	Intermediate (%)	Resistance (%)
Norfloxacin (10µg)	37(100)	0(0.0)	0(0.0)
Nalidixic acid (30µg)	32(86.5)	5(13.5)	0(0.0)
Ciprofloxacin (5µg)	34(91.9)	3(8.1)	0(0.0)
Amoxicillin-clavulanic acid (20/10µg)	35(94.6)	2(5.4)	0(0.0)
Tetracycline (30µg)	10(27.0)	5(13.5)	22(59.5)
Chloramphenicol (30µg)	12(32.4)	2(5.4)	23(62.2)
Ampicillin (10 µg)	13(35.1)	4(10.8)	20(54.1)
Gentamicin (10µg)	22(59.5)	5(13.5)	0(0.0)
Streptomycin (10µg)	15(40.5)	3(8.1)	19(51.4)
Meropenem (10µg)	37(100)	0(0.0)	0(0.0)
Doxycycline (30µg)	25(67.6)	5 (13.5)	7(18.9)
Trimethoprim + sulfamethoxazole (1.25/23.75µg)	22(59.5)	8(21.6)	7(18.9)
Tigecycline (15µg)	36(97.3)	1(2.7)	0(0.0)
Cefotaxime (30µg)	36(97.3)	0(0.0)	1(2.7)
Ceftazidime (30µg)	35(94.6)	2(5.4)	0(0.0)

Table 6: Multiple antimicrobial resistance profiles and proportion of *Salmonella* isolates from samples in poultry farms

Number of antibiotics	Pattern of antibiotics	No of resistant isolate	Total no of multi resistant isolate (%)
Three	CHL, AMP, SXT	1	9(42.9)
	TET, CHL, AMP,	2	
	CHL, AMP, STR,	3	
	AMP, STR, SXT	1	
	CHL, AMP, DOX	1	
	TET, CHL, STR	1	
Four	TET, CHL, AMP, STR	4	9(42.9)
	CHL, STR, DOX, SXT	1	
	CHL, AMP, STR, SXT	1	
	CHL, AMP, STR, DOX,	2	
	CHL, AMP, DOX, CTX	1	
Five	TET, CHL, AMP, STR, DOX	1	2(9.5)
	TET, CHL, AMP, STR, SXT	1	
Six	TET, CHL, AMP, STR, DOX, SXT	1	1(4.7)
Total MDR		21	21(100)

† Nalidixic acid (NAL), † Ciprofloxacin (CIP), †Tetracycline (TET), †Chloramphenicol (CHL), †Ampicillin (AMP), †Gentamicin (GMN), †Ceftazidime (CAZ), †Cefotaxime (CTX), †Trimethoprim + sulfamethoxazole (SXT), †Meropenem (MEM), † Doxycycline(DOX), †Tigecycline (TGC), † Streptomycin (STR), †Norfloxacin (NOR)

scavenging system (Odds ratio [OR] = 2.25; 95% confidence interval [CI= 1.15- 4.40; $P= 0.018$]. Fecal samples had three times higher Odds ratio than meat samples [OR = 3.34; 95% CI for OR = 1.39- 8.01; $P= 0.007$] for fecal samples.

Higher fecal prevalence were observed in Ada'a district (20.1%), in semi-intensive farm (25.9%), in broiler chicken (20.1%) and in exotic breed (19.6%) although these differences were not statistically significant (Table 3).

The prevalence of *Salmonella* in the egg samples (Table 4) was relatively higher in Lome district (10.3%) than the other study sites. From the comparison of farm types, production types and breed of chicken, the highest prevalence were 25.0%, 5.8% and 5.3% in semi-intensive farms, layers and exotic breeds respectively as indicated in Table 4. Only farm type was marginally significant ($P=0.022$).

Antimicrobial Resistance Profile: The overall antimicrobial resistance profiles of *Salmonella* isolates from feces (n=20), hand swabs of farm workers (n=4), raw meat (n=6) and raw eggs (n=7) are given in Table 5. Twenty four of the total isolates tested (64.8) were resistant to at least one drug. About fifty-seven percent (21/37) of the isolates tested were resistant to more than or equal to three antibiotic classes indicating multidrug resistance (Table 6). The prevalence of resistance was high for chloramphenicol (62.2%), tetracycline (59.5%), ampicillin (54.1%) and streptomycin (51.4%). All isolates were susceptible to meropenem, norfloxacin, nalidixic acid, ciprofloxacin, amoxicillin-clavulanic acid, gentamicin, ceftazidime and tigecycline.

DISCUSSION

In Ethiopia several factors including under and malnutrition, HIV-AIDS, the unhygienic living circumstances and the close contact between humans and animals can substantially contribute to the occurrence of human salmonellosis [25]. Chicken products and farm workers can acquire *Salmonella* from intestinal contents, fecal matter or from cross-contamination during slaughtering processes, egg collection and other farm management activities [26, 27].

Our finding is nearly similar to 16.7% *Salmonella* isolated in Southern Ethiopia [28], 15.12% recorded in central Ethiopia [29] and nearly 12% in Kenya [30]. However, our result is higher than reported from Addis Ababa (4.7%) in Ethiopia [31] while it is lower than 25.35%

reported in poultry in Bangladesh [32]. The difference in the results may be attributed to differences in sampling procedure, sample type, sample size, locality and other factors.

Based on the univariate analysis using χ^2 test, all the investigated risk factors except district of the study area were associated with the presence of *Salmonella* (Table 1). Finally, multivariable logistic regression analysis was conducted to see the strength of association between risk factors and *Salmonella* isolation. After the effect of other risk factors were adjusted for, only farm type and sample type had a significant effect on *Salmonella* recovery (Table 2). Sample types as a risk factor reveal that, *Salmonella* prevalence was three times higher in fecal sample than in meat sample. This is as a result of *Salmonella* spp. are commonly found in the alimentary tract of animals [33]. The chicken product surface and farm workers can acquire *Salmonella* from intestinal contents, fecal material or from cross-contamination during slaughtering processes, egg collection and other farm management activities [34, 35].

The strength of association indicated that, odds ratio was twice in semi-intensive farms than backyard scavenging system. *Salmonella* prevalence was higher in exotic breeds and in intensive farms than local breed that are usually kept under scavenging backyard system. This may be due to the difference in genetic composition and stress of high physiologic activity in intensive farming systems. The higher prevalence of *Salmonella* in layers (20.4%) than in broilers (11.1%) could be due to physiological stress of egg production and molting, which significantly depresses the immune response and increases the susceptibility to *Salmonella* infection [36].

Antimicrobial-resistant *Salmonella* were observed in 65% of 37 *Salmonella* isolates tested, which is higher than previous studies, 47.6% by Ejo *et al.* [37] in Ethiopia and 19.7% in Spanish by Lamas *et al.* [38]. This observed resistance is lower than what other studies reported, 83% by Zelalem. *et al.*[39] in Ethiopia and 87.2% by Kim *et al.* [40] in Korea. The overall resistance result was relatively in agreement with 60.6% recorded in Ghana by Andoh *et al.* [41]. This resistance variation could be due to indiscriminate use of antimicrobials in animal production without prescription in the animal health sector especially in developing countries, duration of exposure to antibiotics and others which might favor selection pressure that increased the advantage of maintaining resistance genes in bacteria.

Majority (56.8%) of the isolates in the current study was resistant or intermediately resistant to more than two

antimicrobial classes and the prevalence of resistance was high to chloramphenicol (62.2%), tetracycline (59.5%), ampicillin (54.1%) and streptomycin (51.4%). This is consistent with the report of Phagoo & Neetoo [42] in which all isolates were resistant to tetracycline while 60% and 80% of isolates were resistant to chloramphenicol and erythromycin respectively. On the other hand, the current result is in contrary to the report from Brazil by Arboite *et al.*[43], in which the highest sensitivity was demonstrated for ampicillin (94.9%), tetracycline (91.1%) and chloramphenicol (98.7%). The possible reason for high resistance rate to chloramphenicol, tetracycline, ampicillin and streptomycin in Ethiopia could be, these antibiotics are the most commonly used in veterinary medicine in the country [44].

High sensitivity of isolates was recorded for norfloxacin (100%), meropenem (100%), tigecycline (97.3%), cefotaxime (97.3%), ceftazidime (94.6%) and amoxicillin-clavulanic acid (94.6%) which can be associated with no or limited use of these antibiotics in poultry production in Ethiopia. Due to the relatively limited access and high price to get the newly developed cephalosporins and quinolones, the high prevalence of antimicrobial-resistant *Salmonella* to relatively low-priced and commonly available antibiotics is alarming for a low-income society living in most developing countries, like Ethiopia. However, it is important to note that these antibiotics are commonly used in veterinary medicine and infections with these resistant *Salmonella* isolates could lower the efficiency of antibiotic treatments in human infections that require antibiotic therapy [45].

In current study, more than half of the isolates (57%) were MDR. The remarkably high occurrence of MDR in *Salmonella* is probably an indication of their availability and frequent use of the antibiotics in the poultry production system in Ethiopia. Studies conducted elsewhere in Ethiopia [46] also indicated high proportion of drug-resistant *Salmonella* isolates that could be due to the irrational use of antimicrobials and inappropriateness of the prescription and dispensing methods in both the public and private veterinary services.

CONCLUSIONS

This study showed about 20% fecal prevalence of non-typhoidal *Salmonella* in the chicken sampled at poultry farms. This finding warrants improved farm hygienic measures to reduce its dissemination to humans and to the environment through all steps of product collection, processing, transportation and storage.

The overall prevalence of MDR isolates (56.8%; n= 37) indicate the importance of chicken as a source of MDR *Salmonella* infections in humans. There are inappropriate practices and attitudes associated with improper antibiotic handling and management issues in the professionals, awareness problems in the community and easy accessibility of the drugs in the black markets that could potentially affect the drug effectiveness. Personnel training on personal hygiene, biosafety and farm biosecurity for all farm attendants and visitors, education on zoonotic pathogens, major risk factors that affect awareness on safe handling and management of veterinary drugs, restrictions on uncontrolled use of antimicrobials and establishment of a standard pathogen and AMR monitoring system in poultry production are recommended. Overall, we hypothesize that control of *Salmonella* infections in poultry is the important step to control *Salmonella* infections in humans.

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Conflict of Interest: The authors declare that they have no competing interests.

REFERENCES

1. Eng, S.K., P. Pusparajah, N.S. Ab Mutalib, H.L. Ser, K.G. Chan and L.H. Lee, 2015. *Salmonella*: A review on pathogenesis, epidemiology and antibiotic resistance. *Frontiers in Life Science*, 8(3): 284-293. <https://doi.org/10.1080/21553769.2015.1051243>.
2. Doyle, M.P., G.H. Loneragan, H.M. Scott and R.S. Singer, 2013. Antimicrobial resistance: Challenges and perspectives. *Comprehensive Reviews in Food Science and Food Safety*, 12(2): 234-248. <https://doi.org/10.1111/1541-4337.12008>.
3. Abatcha, G., G. Kaur and L. Thong, 2014. Review Article: A trends of *Salmonella* and antibiotic resistance. *Advances in Life Science and Technology*, 17: 9-21. <http://iiste.org/Journals/index.php/ALST/article/view/10821>.

4. Marshall, B.M. and S.B. Levy, 2011. Food Animals and Antimicrobials?: Impacts on Human Health. 24(4):718-733. <https://doi.org/10.1128/CMR.00002-11>
5. Bangera, S.R., S. Umakanth, G. Chowdhury and R.N. Saha, 2019. Poultry?: a receptacle for non-typhoidal *Salmonellae* and Antimicrobial Resistance, 11(1): 31-38.
6. Mir, I.A., S.K. Kashyap and S. Maherchandani, 2015. Isolation, serotype diversity and antibiogram of *Salmonella enterica* isolated from different species of poultry in India. Asian Pacific Journal of Tropical Biomedicine, 5(7): 561-567. <https://doi.org/10.1016/j.apjtb.2015.03.010>.
7. Antunes, P., J. Mourão, J. Campos and L. Peixe, 2016. Salmonellosis: The role of poultry meat. Clinical Microbiology and Infection, 22(2): 110-121. <https://doi.org/10.1016/j.cmi.2015.12.004>.
8. Messens, W., L. Vivas-Alegre, S. Bashir, G. Amore, P. Romero-Barrios and M. Hugas, 2013. Estimating the public health impact of setting targets at the European level for the reduction of zoonotic *Salmonella* in certain poultry populations. International Journal of Environmental Research and Public Health, 10(10): 4836-4850. <https://doi.org/10.3390/ijerph10104836>.
9. Cosby, D.E., N.A. Cox, M.A. Harrison, J.L. Wilson, R. Jeff Buhr and P.J. Fedorka-Cray, 2015. *Salmonella* and antimicrobial resistance in broilers: A review. Journal of Applied Poultry Research, 24(3): 408-426. <https://doi.org/10.3382/japr/pfv038>.
10. Shrestha, K.L., N.D. Pant, R. Bhandari, S. Khatri, B. Shrestha and B. Lekhak, 2016. Re-emergence of the susceptibility of the *Salmonella* spp. isolated from blood samples to conventional first line antibiotics. Antimicrobial Resistance and Infection Control, 5(1): <https://doi.org/10.1186/s13756-016-0121-8>.
11. Tadesse, G. and T.S. Tessema, 2014. A meta-analysis of the prevalence of *Salmonella* in food animals in Ethiopia. BMC Microbiology, 14(1): 1-9. <https://doi.org/10.1186/s12866-014-0270-y>.
12. Tadesse, G. and E.Z. Gebremedhin, 2015. Prevalence of *Salmonella* in raw animal products in Ethiopia: A meta-analysis. BMC Research Notes, 8(1): 1-8. <https://doi.org/10.1186/s13104-015-1127-7>.
13. Shapiro, B., 2015. Livestock Sector Analysis (LSA) & Livestock Master Plan (LMP) Process.
14. Nhung, N.T., N. Chansiripornchai and J.J. Carrique-Mas, 2017. Antimicrobial resistance in bacterial poultry pathogens: A review. Frontiers in Veterinary Science, 4(8): 1-17. <https://doi.org/10.3389/fvets.2017.00126>
15. Benacer, D., K.L. Thong, H. Watanabe and S. Devi Puthuchery, 2010. Characterization of drug-resistant *Salmonella enterica* serotype typhimurium by antibiograms, plasmids, integrons, resistance genes and PFGE. Journal of Microbiology and Biotechnology, 20(6): 1042-1052. <https://doi.org/10.4014/jmb.0910.10028>.
16. Basler, C., T.A. Nguyen, T.C. Anderson, T. Hancock and C.B. Behravesh, 2016. Outbreaks of human *Salmonella* infections associated with live poultry, United States, 1990-2014. Emerging Infectious Diseases, 22(10): 1705-1711. <https://doi.org/10.3201/eid2210.150765>.
17. Sylvia, O., M.M. Samuel, N. Kariuki and R.C. Douglas, 2015. A review of 40 years of enteric antimicrobial resistance research in Eastern Africa: what can be done better? Antimicrobial Resistance and Infection Control 4:1 DOI 10.1186/s13756-014-0041-4.
18. FAO, 2008. Review of the new features of the Ethiopian poultry sector Biosecurity implications.
19. ISO-17604. Microbiology of food and animal feeding stuffs: Carcass sampling for microbiological analysis. ISO, Geneva; 2003. pp: 1-17.
20. ISO-6579. Microbiology of food and animal feeding stuffs: Horizontal method for the detection of *Salmonella* spp. ISO, Geneva; 2002. pp: 511-52.
21. WHO 2010. Laboratory Protocol "Isolation of *Salmonella* spp. From Food and Animal Faeces" 5th Ed
22. Mikoleit, M.L., 2015. A WHO network building capacity to detect, control and prevent foodborne and other enteric infections from farm to table & quot; Biochemical Identification of *Salmonella*, 1-43.
23. Grimont, P. and F.X. Weill, 2007. Antigenic Formulae of the *Salmonella* Serovars. 9th ed. Paris: Institut Pasteur.
24. CLSI, 2012. Performance Standards for Antimicrobial Susceptibility Testing; Twenty Second Informational Supplement. CLSI document M100-S22, Wayne PA.
25. Tadesse, G., 2014. Prevalence of human *Salmonellosis* in Ethiopia: A systematic review and meta-analysis. BMC Infectious Diseases, 14(1): <https://doi.org/10.1186/1471-2334-14-88>.
26. Agyare, C., V.E. Boamah, C.N. Zumbi and F.B. Osei, 2018. Antibiotic Use in Poultry Production and Its Effects on Bacterial Resistance. <https://doi.org/10.5772/intechopen.79371>.
27. Uro, W.T., 2019. A Review: *Salmonellosis*: Int. J. Adv. Res. Biol. Sci, 6(10): 79-88. <http://dx.doi.org/10.22192/ijarbs.2019.06.10.008>.

28. Abdi, R.D., F. Mengstie, A.F. Beyi, T. Beyene, H. Waktole, B. Mammo and F. Abunna, 2017. Determination of the sources and antimicrobial resistance patterns of Salmonella isolated from the poultry industry in Southern Ethiopia. BMC Infectious Diseases, 17(1): 1-12. <https://doi.org/10.1186/s12879-017-2437-2>.
29. Abunna, F., M. Bedasa, T. Beyene, D. Ayana, B. Mamo and R. Duguma, 2016. *Salmonella*: Isolation and antimicrobial susceptibility tests on isolates collected from poultry farms in and around Modjo, Central Oromia and Ethiopia. Journal of Animal and Poultry Sciences, 5(2): 21-35. <http://www.japsc.com>
30. Langata, L.M., J.M. Maingi, H.A. Musonye, J. Kiiru and A.K. Nyamache, 2019. Antimicrobial resistance genes in Salmonella and Escherichia coli isolates from chicken droppings in Nairobi, Kenya. BMC Research Notes, 12(1): 1-6. <https://doi.org/10.1186/s13104-019-4068-8>.
31. Eguale, T., 2018. Non-typhoidal Salmonella serovars in poultry farms in central Ethiopia: Prevalence and antimicrobial resistance. BMC Veterinary Research, 14(1):1-8. <https://doi.org/10.1186/s12917-018-1539-4>.
32. Karim, M.R., G. Md, M. Samad, M.R. Karim, M. Giasuddin, M.A. Samad and M.A. Yousuf, 2017. Prevalence of *Salmonella* spp in Poultry and Poultry Products in Dhaka, Bangladesh Prevalence of Salmonella spp. in Poultry and Poultry Products in Dhaka, Bangladesh. International Journal of Animal Biology, 3(4): 18-22. <http://www.aiscience.org/journal/ijabhttp://creativecommons.org/licenses/by/4.0/>.
33. Garede, L., Z. Hagos, Z. Ddis, R. Tesfaye and B. Zegeye, 2015. Prevalence and antimicrobial susceptibility patterns of Salmonella isolates in association with hygienic status from butcher shops in Gondar town, Ethiopia. Antimicrobial Resistance and Infection Control, 4(1): 1-7. <https://doi.org/10.1186/s13756-015-0062-7>.
34. Kagambèga, A., T. Lienemann, L. Aulu, A.S. Traoré, N. Barro, A. Siitonen and K. Haukka, 2013. Prevalence and characterization of Salmonella enterica from the feces of cattle, poultry, swine and hedgehogs in Burkina Faso and their comparison to human Salmonella isolates. BMC Microbiology, 13(1). <https://doi.org/10.1186/1471-2180-13-253>.
35. Mąka, Ł. and M. Popowska, 2016. Antimicrobial resistance of *Salmonella* spp. isolated from food. Roczniki Panstwowego Zakladu Higieny, 67(4): 343-358.
36. Holt, P.S., 2003. Molting and Salmonella enterica serovar enteritidis infection: The problem and some solutions. Poultry Science, 82(6): 1008-1010. <https://doi.org/10.1093/ps/82.6.1008>.
37. Ejo, M., L. Garede, Z. Alebachew and W. Worku, 2016. Prevalence and Antimicrobial Resistance of *Salmonella* Isolated from Animal-Origin Food Items in Gondar, Ethiopia. Bio. Med. Research International, 2016. <https://doi.org/10.1155/2016/4290506>.
38. Lamas, A., I.C. Fernandez-No, J.M. Miranda, B. Vázquez, A. Cepeda and C.M. Franco, 2016. Prevalence, molecular characterization and antimicrobial resistance of Salmonella serovars isolated from northwestern Spanish broiler flocks (2011-2015). Poultry Science, 95(9): 2097-2105. <https://doi.org/10.3382/ps/pew150>.
39. Zelalem, A., K. Nigatu, S. Zufan, Haile, Y. Alehegne and K. Tesfu, 2011. Prevalence and antimicrobial resistance of *Salmonella* isolated from lactating cows and in contact humans in dairy farms of Addis Ababa: a cross sectional study. BMC Infectious Diseases, 11: 222. <http://ovidsp.ovid.com/ovidweb.cgi>.
40. Kim, M.S., T.H. Lim, J.H. Jang, D.H. Lee, B.Y. Kim, J.H. Kwon and C.S. Song, 2012. Prevalence and antimicrobial resistance of Salmonella species isolated from chicken meats produced by different integrated broiler operations in Korea. Poultry Science, 91(9): 2370-2375. <https://doi.org/10.3382/ps.2012-02357>.
41. Andoh, L.A., A. Dalsgaard, K. Obiri-Danso, M.J. Newman, L. Barco and J.E. Olsen, 2016. Prevalence and antimicrobial resistance of Salmonella serovars isolated from poultry in Ghana. Epidemiology and Infection, 144(15): 3288-3299. <https://doi.org/10.1017/S0950268816001126>.
42. Phagoo, L. and H. Neetoo, 2015. Antibiotic Resistance of Salmonella in Poultry Farms of Mauritius. In J. World's Poult. Res. (Vol. 5).
43. Arboite De Oliveira, F., A. Brandelli and E.C. Tondo, 2006. Antimicrobial resistance in *Salmonella* Enteritidis from foods involved in human salmonellosis outbreaks in southern Brazil. In the New Microbiologica, pp: 29.
44. Tufa, T.B., F. Gurmu, A.F. Beyi, H. Hogeveen, T.J. Beyene, D. Ayana, F.D. Gutema and J.A. Stegeman, 2018. Veterinary medicinal product usage among food animal producers and its health implications in Central Ethiopia. BMC Veterinary Research, 14: 409. <https://doi.org/10.1186/s12917-018-1737-0>.

45. Tadesse, G., 2015. A meta-analysis of the proportion of animal Salmonella isolates resistant to drugs used against human salmonellosis in Ethiopia. *BMC Infectious Diseases*, 15: 84. DOI 10.1186/s12879-015-0835-x.
46. Beyene, T., S. Assefa, D. Ayana, T. Jibat, F. Tadesse, D. Nigussie and A. Feyisa, 2016. Assessment of Rational Veterinary Drugs Use in Livestock at Adama District Veterinary Clinic, Central Ethiopia *J. Veterinar. Sci. Techno.*, 7: 319. doi:10.4172/2157-7579.1000319.