Research Article

Epidemiology and Antibiogram Profiles of Salmonella Isolates from Clinical and Nonclinical Sources in Bushenyi District, Uganda

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Abstract: Bacterial food borne contamination by Salmonella species continues to be a challenge to both animal and human health world-wide. This current study was carried out in order to isolate, and determine antibiogram profiles of Salmonella isolates from clinical and non-clinical sources. This was A cross sectional and experimental study carried out. In three selected hospitals, Cattle abattoirs, and water sources in Bushenyi Uganda, between January 2011 and August 2011.Samples were analyzed by used standard microbiological methods. In results out of 74 Salmonella isolates, 47 (63%) were from non-clinical samples, and 27 (37%) were from human patients. Of these isolates, 85% were resistant to 3-5 antimicrobials, and 100% were resistance to Tetracycline. Most of the isolates were relatively sensitive to ciprofloxacin, cotrimoxazole, Ceftriaxone, and Ceftazidime. The predominant servers from cattle, food, and waste water were S. typhimurium, S.arizonae, S.choleraesuis and S.infantis respectively. The only species isolated from humans were Salmonella typhimurium, S. typhi and S. enteritidis. In Conclusion from this study, the prevalence of Salmonella in non-clinical and clinical samples in Bushenyi district was 11.8% and 11.3% respectively. This result was significantly high and could be considered a potential source of food borne salmonellosis.
Keywords: Salmonellosis, Isolates, Resistance, Servers.

INTRODUCTION

Bacterial food borne contamination by Salmonella species continues to be a challenge to both animal and human health world-wide[1,2]. Contamination of foods with Salmonella species occurs during production, processing, and distribution, retail marketing and handling[3,4]. The primary source of contamination is usually animal meat, milk, and infected humans. Alam *et al.;* [5] reported that in the United States, salmonellosis is estimated to affect 1.4 million people each year, and 95% of the cases are food borne. In developing countries, ready to eat foods in particular have been reported to be contaminated with Salmonella and have been implicated in a few outbreaks of food borne diseases[6].

Salmonellosis is caused by two species of Salmonella namely; Salmonella enteric and Salmonella bongori, the later is a zoonotic disease of humans and animals [7]. Salmonella strains which are resistant to antibiotics have emerged worldwide, and is causing great concern to consumers of animal food products. It is particularly serious in low-resource countries where bacterial infections remain among the major causes of death, especially in childhood [8]. Surveillance of antibiotic resistance is a key element for providing updated information on the magnitude and trends in resistance and for planning and monitoring intervention strategies targeted at preserving the therapeutic efficacy of antimicrobial agents[9].

According to Milledge *et al.;* [10], Salmonella serovars such as S. typhi, S.typhimurium, and S.enteritidis which cause extra intestinal infection such as gastroenteritis, enteric fevers, and septicemia exhibit multidrug resistance. The treatment of these infections has become increasingly difficult due to limited choice of antibiotics [11]. This means that more expensive drugs with more potency are needed to treat the Salmonella infection. A study carried out by Agwu *et al.;* in Bushenyi district, among the males surveyed between sentinel centres showed that, age group 10–19 years had the highest typhoid prevalence of 28.6%, while age group 20–29 years had 24.6% highest typhoid prevalence. Despite the large number of antimicrobial agents available, these infections have remained a significant problem because of lack of routine isolation and identification [12]. It's upon this background that

this study on isolation, and identification of Salmonella serovars from clinical and non-clinical sources in Bushenyi district was carried out.

MATERIALS AND METHODS
Study area
The study was done at Kampala International University-Teaching Hospital (KIU-TH), Ishaka Adventist Hospital(IAH), and Kabwohe Health Center IV(KAH-IV) in Sheema district, and at selected government abattoirs in Ishaka-Bushenyi municipality, Kyeizoba parish, and Kyamhunga parish.(See appendix 3).

Sample Size determination
The sample size (n) was calculated using the standard formula below:-

$$n = \frac{Z^2 P (1-P)}{d^2}$$

Where,

d = margin of error of setting a significance level of 0.05 (i.e. 5%).

P= prevalence rate of Salmonella (42%) ₌0.42.

Z=Level of significance (1.96) for confidence interval of 95%.

$$N = \frac{1.96 \times 1.96 \times 0.42(1-0.42)}{0.05 \times 0.05} = 374$$

With the above formula 374 samples were to be collected (each) from selected District abattoirs and Health centres, but due low Salmonella concentration in faecal samples, 640 samples were analysed, 240 from clinical and 400 from non-clinical sources in Bushenyi district.

Study design
The study was a cross-sectional survey. Two types of sample collection were employed; group A clinical samples, and group B non-clinical samples from cattle, Street food, and waste water.

Scope of the study
Phenotypic methods adopted included: cultural and biochemical methods. Confounding variables such as underlying medical condition of patients and health conditions of cattle was not investigated as it was beyond the scope of this study.

Sample collection
The samples were collected every Monday and Wednesdays (slaughter days), from cattle Abattoirs, and clinical samples from Kampala International University-Teaching Hospital, Ishaka Adventist ,and Kabwohe health centers between January 2011 to August 2011. A total of 400 faecal samples from cattle, and 240 stool samples from humans were collected and examined for presence of Salmonella. The faecal specimens (30 gm) of cows were collected in a clean

sterile air tight stool cup directly from the rectum. 30 gm of stool each from cattle and humans were collected in sterile stool cup with an applicator stick. The samples were then transported in an ice box and were examined upon arrival, and those not processed immediately upon arrival were stored at freezer temperature for no longer than 24hrs at Mbarara University of Science and Technology (Microbiology Laboratory).

Isolation and Identification of Salmonella
The isolation of Salmonella was performed, using techniques recommended by International Organizations for Standardization (ISO-6579, 2000).

Pre-enrichment
1 gm of faecal sample and food sample were pre-enriched in buffered peptone water (BPW) (Oxoid CM509, Basingstoke, England) in a ratio 1g of the sample to 9 ml of buffered peptone water and incubated for 24 hrs at 37°C [13].

Selective enrichment
A portion (0.1 ml) of the pre-enriched broth was transferred aseptically into 10 ml of selenite cysteine (SC) (Himedia M025, Mumbi) broth and another 0.1 ml portion was transferred to10 ml of Rappaport and Vassilidis (RV) broth (Merk, Darmstadt, Germany), and incubated at 37°C and 42°C for 24 hrs respectively [13].

Selective plating
Finally, from the selective enrichment media the samples were inoculated on to Xylose Lysine Deoxycholate (XLD) (Oxoid CM0469, Basingstoke, England) agar and incubated at 37°C for 48 hrs. Salmonella colonies, showing a slightly transparent zone of reddish color and a black center, were sub-cultured on nutrient agar (Oxoid CM0003, Basingstoke, England) , and confirmed biochemically using triple sugar iron agar (TSI) (Oxoid CM0277, Basingstoke, England), and Christensen's urea agar (Oxoid CM53, Basingstoke, England) [13].

Identification
Colonies suspected to be Salmonella were picked and sub-cultured into a series of biochemical tests. These included Citrate, Urea and API 20E tests.

Citrate test
This test was performed to determine if the organism was capable of utilizing citrate as a sole carbon source. It mainly aided in the differentiation between genera. Using Simmon's Citrate, a positive test meant growth of the organism with an intense blue color on the slant. This is the usual reaction for Salmonella. A negative test gives no growth and no change in the green color.

Urease test

Christensen's Urea agar was used to determine the ability of the organism to split urea, forming two molecules of ammonia by the action of the enzyme urease with resulting alkalinity. A positive test gives an intense pink-red colour on the slant whilst the negative test gives a yellow-orange colour. Salmonella give a negative test. All urease negative tests, were subjected to Analytical Profile Index (API 20E) test, and the results were read using Mini API instrument.

Analytical Profile Index (API 20E)

API 20 E is a standardized system for the identification of Enterobacteriaceae and other non fastidious Gram-negative rods. The API 20 E strip is made up of 20 test cupules which contain dehydrated substrates. To perform the test, the ampoule is filled with test organism contained in 0.85% Nacl adjusted to 0.5 McFarland Standard. The procedure was carried out according to the instructions laid out by the manufacturer. The inoculated strips were incubated at 37^{oC} for 18 to 24 hours in moist chamber. After incubation the strips were read using the mini API instrument and results subjected to mini API reader.

Antimicrobial susceptibility test

The antimicrobial susceptibility test was performed according to the National Committee for Clinical Laboratory Standards (NCCLS using Kirby-Bauer disk diffusion method. The test was done using Muller-Hinton agar (Oxoid CM0337 Basingstoke, England), [14]. The isolates were grown on blood agar plates at 37^{oC} for 18 hours. Three to four isolated colonies were suspended in 0.5 ml 0.85% saline in polystyrene tubes. The turbidity of the suspension was adjusted to 0.5 McFarland standard (Oxoid) and inoculated onto Muller-Hinton agar plates (Oxoid) using a sterile swab. The inoculated plates were then allowed to dry before a set of antimicrobial disks were dispensed onto the agar surface using a semi-automated dispenser (Difco). Ceftazidime (CAZ, 30ug), erythromycin (E, 15ug), ciprofloxacin (CIP, 5ug), ceftriaxone (CRO, 30ug), cotrimoxazole (SXT, 25ug), tetracycline (TET, 30ug), ampicillin (AM, 10ug) and chloramphenicol (C, 5ug) (BIOLAB Inc., Budapest Hungary) were used. The plates were then incubated for 16-24 hours at 37^{oC}. The zones of inhibition were recorded. The results were interpreted as Sensitive (S) or Resistant (R) according to (CLSI, 2006).

Data analysis

Data was analyzed using computer software SPSS version 13 (SPSS 13.0 Command Syntax Reference. SPSS Inc., Chicago, 2004) and presented in tables and graphs. The Chi-square test was utilized to assess significant differences in antimicrobial resistance of Salmonella isolates from human and cattle and from isolates of food and waste water. A difference was taken as significant at a p-value less than 0.005.

Ethical Approval

The ethical approval of the study was sought from Mbarara University of Science and Technology (MUST), Institutional Research and Ethics Committee (IREC) on Human Research, and Uganda National Council for Science and Technology (UNCST). All experiments were examined and approved by the appropriate ethics committees and performed in accordance with the ethical standards of the committees on human experimentation laid down in the Helsinki declaration of 1975 as revised in 2000.

RESULTS

Human patients, Cattle, Street food and waste Water were considered in this study. Six hundred and forty (640) samples were studied. This included 240(37.5%) samples from clinical sources, and 400(62.5%) samples from non-clinical sources. From the clinical sources, 80 samples were studied from each of the three Health centres. From non-clinical sources, 120(30%) samples were collected from cattle and 140 (35%) were each collected from food and water respectively. The prevalence of Salmonella serovars in the clinical samples were 11.3% (27/240), and 11.8 % (47/400) in non-clinical samples, the difference was not significant (P=.8, OR=1.05 and RR was 1.04).

In the clinical samples Table I below, Ishaka Adventist Health centre IV (IAH) had the highest prevalence of Salmonella serovars 15% (12/80)), followed by Kampala International University Teaching Hospital (KIUTH) 11.3% (9/80), and the least was Kabwohe Health Centre IV 7.5% (6/80). There were no significant differences in the prevalent rates within these Health centers, (p.value=0.32).

In the non-clinical samples Table II below, the highest prevalence of Salmonella serovars was found among the cattle samples 30% (36/120), followed by the water samples 4.3% (6/140). There was a very significant difference in the prevalence of Salmonella serovars within the non-clinical samples (p.value <0.0005). There were 8.33 increased chances of isolating Salmonella serovars in cattle sample than in any other non-clinical sources. In the non-clinical samples, the organisms isolated were S.typhimurium (most abundant-23/48.9%), S.infantis (4/8.5%), S.arizinae (11/23.4%) and S.choleraesius (9/19.1%). In table III below, all the Salmonella spp were resistant to tetracycline. Salmonella serovars resistant to more than four antibiotics were considered multi-drug resistant. Clinical isolates, resistant to five anti-biotics were: S.typhymurium: tetracycline 94.1%, chloramphenicol 88.2%, ampicillin 82.4%, cotrimoxazole 58.8%, and erythromycin 52.9%; S.typhi: tetracycline 100%,

chloramphenicol 100%, ampicillin100%, cotrimoxazole 60%, and erythromycin 60%; S.enteritidis: tetracycline 100%, chloramphenicol 100%, ampicillin100%, cotrimoxazole 75%, and erythromycin 75%; S.infantis: tetracycline 100%, chloramphenicol 100%, ampicillin100%, and erythromycin 100%.

In non-clinical samples studied, most Salmonella serovars showed resistance to only three antibiotics, these were: S.typhimurium: tetracycline 87.0%, chloramphenicol 87.0%, and cotrimoxazole 47.8%; S.arizonae: tetracycline 81.8%, chloramphenicol 63.6%, and cotrimoxazole 54.5%; S.cholaeruisis: tetracycline 100%, chloramphenicol 87.5%, ampicillin 87.5% and S.infantis: tetracycline 100%, chloramphenicol 75%, ampicillin100%, cotrimoxazole 75%, and ceftriaxone 75%. Multi-drug resistance was found to be higher in isolates from clinical samples than in non-clinical samples.

From table V, below, clinical S.infantis was 100% susceptibility to ciprofloxacin, ceftriaxone, ceftazidine, erythromycin and cotrimoxazole, while non clinical S.infantis was sensitive to ciprofloxacin (75%). Non-clinical S.typhimurium was susceptible to ciprofloxacin and ceftazidine (65.2% sensitivity for each), while Clinical S.typhimurium was sensitive to ceftriaxone (76.5%). All isolates identified as S.arizinae and S.typhi were sensitive to ceftazidine (100%) while S.choleraesuis and S.enteritidis were sensitive to ciprofloxacin (87.5% and 100% respectively) and ceftriaxone by (87.5%).

Table VI, below shows p.values and risk estimates of the Salmonella serovars to the antibiotics tested. Ciprofloxacin, erythromycin and cotrimoxazole showed no significant difference in their action against Salmonella isolates from clinical and non-clinical sources (p=.16, .8, .2 respectively), but Salmonella isolates from non-clinical sources were more resistant, (OR=1.6, 1.4 for ciprofloxacin and cotrimoxazole) than the clinical isolates. Clinical isolates were slightly more resistant to Erythromycin (OR=0.9). The rest of the antibiotics tested showed variable action against the non-clinical and clinical Salmonella isolates, (p=.05). Non-clinical isolates were thrice (OR=3.4, 3.2 respectively) more resistant to Ceftriaxone and ceftazidime than the clinical isolates. Clinical Salmonella isolates were more resistant to Ampicillin, chloramphenicol and tetracycline than the non clinical isolates (OR=0.06, 0.1, 0.1)

DISCUSSION

The prevalence rate of Salmonella serovars in non-clinical samples (cattle, food and water) was high, 11.8% (47/400) as compared to studies by Alemayehu *et al.;*) [6], who reported a prevalence of 7.1%, in Ethiopian cattle abattoirs, and ready to eat food items.

Also studies from England by White *et al.;* reported (0.2% and 4%) prevalence, of Salmonella serovars from cattle and ready to eat foods. These reports were much lower than the current investigation (11.8%) prevalence of Salmonella serovars in non-clinical samples from Bushenyi district. These findings were within the range reported earlier by D'Aoust *et al.;* [15], where the rate of Salmonella isolation from Canadian beef carcasses varied from 0.2 to 21.5% with a median of 3.34%. These differences in the prevalence rate of Salmonella serovars may be attributed to the difference in the test method used, the living condition, like housing conditions, feeding habits, and types of feed given to the cattle. Hence non-clinical sources (cattle, food and water), with 11.8% prevalence, could be a potential source of Salmonella infection to individuals working in cattle farms and the community studied in Bushenyi.

In clinical samples analyzed, the prevalence of Salmonella among individuals working in Bushenyi and attending medical care in the health centers was 11.3%. This showed a higher prevalence rate compared to results obtained by Alemayehu *et al.;* [6] who reported prevalence of 6% and 7.6%, among humans working in cattle farms and attending health care at Addis Ababa hospital respectively. This difference in the prevalence of Salmonella serovars from clinical sources may be due to the hygienic status of the people. According to a report by, (MoH,2004), Coverage of public latrines is very low (19%), all located at institutions like primary schools, markets and health units, with as low as 2% and 2.8% in Kotido and Nakapiripirit Districts respectively to only 30% in Bushenyi district in the southwest part of the country. The (11.3%) prevalence of Salmonella serovars in clinical samples (human patients) may be attributed to inadequate hygiene status, improper waste management, and food contamination associated with cattle farmers that supply animal meat, and products to the community. Poor hygienic practices at the cattle slaughter houses could also exacerbate the contamination of carcasses Carli *et al.;*[16] and Arthur *et al.;* [7]). The detection of 47 (76.6%) of the Salmonella isolates in cattle fecal samples, suggests that the process of evisceration could be the main source of animal meat contamination since similar Salmonella serovars (S.typhimurium) were isolated in cattle feaces, food and in water 16 (69.6%), 3 (13.0%) and 4 (17.4%) respectively). The other probable source of contamination is infected abattoir personnel who do the processes of skinning and cleaning of the slaughtered animals as reported in previous studies by Alemayehu *et al.;* [6] in Ethiopian abattoir personnel.

According to Clasen *et al.;*[14] poor waste management in community water sources is the main reason for Salmonella prevalence in drinking water sources. Both cattle waste and human faecal samples

are deposited in rivers and streams that supply the communities around Bushenyi district, is the main reason for prevalence of Salmonella species in water sources. According to the study by Kariuki *et al.;* [1] in Kenya, contamination with human feaces is the major source of spread, and the usual vehicle is contaminated water. The problem is the initial quality of the water source used and the level of treatment provided. Therefore a contaminated water source is a major source of infectious diseases and this highlights the importance of selecting the best possible quality of water source to protect public health (WHO, 2002). A rapid international trade in agriculture, aquaculture and manufactured food products has facilitated the introduction of new Salmonella serovars within the geographic boundaries of importing countries [3]. Of the 47 isolated Salmonella serovars identified, S. Arizona and S. typhimurium were most prevalent. Previous studies in Uganda by Nakavuma *et al.;* in Kampala also reported prevalence of similar Salmonella serovars and other non-host Salmonella serotypes in food animals, meat products and poultry.

A report by Mensah *et al.;* [3] in Accra Ghana, showed that contaminated food usually handled by healthy carriers who harbor Salmonella typhi may be the vehicle of transmission. Analysis of the Salmonella isolates from different sources in Bushenyi suggests that an association exists between the occurrence of certain Salmonella serotypes in food animals, meat products and in man, which could be acquired by man through ingestion of contaminated food and food products. It should be noted that the detection of invasive and pathogenic Salmonella serotypes such as S. typhimurium, S. enteritidis, S. Arizona, and S. infantis is of public health significance since contaminated meat, and meat products may pose health hazards. According to Kabagambe *et al.;* [17] those at risk, are infants, elderly, immunocompromised, and malnourished persons. The risk may further be accentuated if meat is consumed raw or undercooked or if cross-contamination of the meat with Salmonella during meal preparation occurs. This is a common practice among street food venders and food restaurants in Bushenyi district were food (meat) is half roasted or half cooked to avoid profit loss incase customers do not turn up to consume their products .

According to McDowell *et al.;* [18], the control of Salmonella contamination and other food borne pathogens includes the introduction of good manufacturing practices (GMP) and hazard analysis critical control point (HACCP) concepts together with stringent control of all aspects of meat production, preparation, storage, and distribution. Sanitary measures such as protection of source, drainage of the area, animal accessibility, elevation of nearby latrines, surface water collection uphill of the source, presence of a diversion ditch and other pollution sources, have been identified as important factors that influence the water quality. Embankment wall, cattle trough, clothes washing area, water drawing area, diversion channels and fencing is essential. Protection of water sources and treatment of water supplies have greatly reduced the incidence of these diseases in developed countries [19].

Isolation of various serotypes of Salmonella from a wide range of sources indicated the presence and widespread distribution of Salmonella of animal and human origin, which is of significance in the veterinary and public health sectors in Bushenyi district. It also underlines the necessity for a joint and coordinated surveillance and monitoring programs for salmonellosis and other major food borne diseases in the country.

In both clinical and non-clinical samples, Salmonella typhimurium was the most predominant serovars isolated, followed by S.arizonae, S.choleraesuis, S.typhi, S.infantis and S.enteritidis respectively. S. typhimurium represented 54.1% of the six serovars identified. This finding is consistent with the study done by Kariuki *et al.;* [1] where S. typhimurium was isolated from 67% of the positive samples from beef cattle in Ghana dairy cattle. Moreover, 80% of the samples taken from ground beef were heavily contarninated with S. typhimurium. According to Davis *et al.;* [20] S. typhimurium continues to be an important cause of human food borne disease throughout the world.

S. typhimurium was isolated from both clinical and non-clinical samples; but high prevalence of S. typhimurium in cattle (69.9%) compared to (63%) in clinical samples supports the notion that cattle could be a major reservoir of human infection. The findings of this study is in line with Carli *et al.;* [16], whose findings showed that, the most common animal reservoirs are cattle, chickens, turkeys, pigs , as well as other domestic and wild animals.

Most clinical Salmonella isolates were similar to isolates from non-clinical samples (cattle, food and water).In all the samples, S. typhimurium, and S.infantis was isolated. This could be interpreted as evidence of an association between contamination of cattle carcasses, water and people. Cheesbrough *et al.;* [21] and Cummings *et al.;* [22-24] also pointed out that Salmonella has the ability to survive in meats and animal products that are not properly cooked and these could act as the main vehicles of transmission. Furthermore Mensah *et al.;* [3], showed that, the epidemiology of typhoid fever and other enteric fevers primarily involves person-to-person spread because these organisms lack a significant animal reservoir.

Antimicrobial resistance patterns of Salmonella strains isolated from clinical and non-clinical samples indicated that a large proportion of the strains were resistant to a variety of drugs tested. More than half of the Salmonella strains (62.2%) tested exhibited multiple resistance to up to five antimicrobials drugs. According to Fricke *et al.;* [25], and Linsey *et al.;* [26] Salmonella are among those bacteria known to carry plasmids which encode for drug resistance (R plasmid). This implies that wide spread use antimimicrobials in humans and in cattle may cause an increase in the frequency of occurrence of bacteria resistant to other antimicrobials as the R plasmid may encode resistance to additional antimimicrobials. Inappropriate utilization of antibiotics in cattle may favor selection pressure that may lead to increased advantage of maintaining resistance genes in bacteria [27]. Antibiotics are used in the treatment of bacterial infections to avoid contamination, but if wrongly administered, high chances of bacterial drug resistance could be promoted.

Zao *et al.;* [9] reported that the isolates of Salmonella from food items and personnel from Addis Ababa were resistant to antibiotics including streptomycin, ampicillin, and tetracycline. The current research in Bushenyi have also found that Salmonella isolates are resistant to commonly used antimicrobials like ampicillin, tetracycline, and chloramphenicol with resistance rate of 100%, 93%, and 87%%, respectively. All the isolated Salmonella, in this current study, were 100% resistant to ampiciliin. This finding is in line with previous reports[28,29] who reported a similar 100%, over 90% and 100% resistance to ampicillin, respectively. Hughi *et al.;* from Iran reported a resistance rate of 60.3% and72.7% in different study periods among human isolates, which was slightly lower than the current finding.

In the present study, resistance to 4 or more antibiotics was regarded as multidrug resistance. Multidrug resistance was encountered in both clinical and non-clinical isolates among that were isolated during the study. S. typhimurium were resistant (to ampicillin, chloramphenicol, cotrimoxazole, and tetracycline). A study by Poppe *et al.;* [30] also reported that S.typhymuriun DT104 was the top human and animal Salmonella isolate with the high multiple résistance (to ampicilin, chloramphenicol, sulfisoxazole, tetracycline, spectinomycin and kanamycin), and has become aserious health problem in Addis Ababa. The (clinical) human strain of S. typhimurium isolated from hospitalized patients in health centres in Bushenyi district had the characteristic drug resistant pattern seen in non-clinical (cattle) isolates, and showed additional resistance to cotrimoxazole. Also a study by Gebre-Yohanes *et al.;* [19] reported that S.typhymurium strain isolated from hospitalized patients in Addis Ababa showed multiple drug resistance (to ampicilin, chloramphenicol, kanamycin, streptomycin, neomycin and tetracycline). One isolate of S.infantis from cattle had similar resistance patterns to S.infantis from clinical isolates, and was resistant to tetracycline, chlorarnphenicol and tetracycline.

Other Salmonella serovars from human sources that showed multiple drug resistance were S. typhi, and S. infantis. The significance of antimicrobial resistance is most obvious on its impact on limiting therapeutic options which may lead to therapeutic failure and intern result in increased morbidity and even mortality. From the study finding in chapter four, the indiscriminate use of antimicrobial agents in humans and animals may lead to emergence of drug resistant microorganisms because they are cheap, accessible and can be purchased without prescription. According to Alemayehu *et al.;* [6] emergence and spread of antibiotic resistant Salmonella may be attributed to the use of medicated feeds in livestock for growth promotion and prophylaxis, routine treatment with antibiotics and also treatment of various animal species and humans with antibiotics. Multidrug resistant S. typhimurium strain is an example of the increasing emergence of antibiotic resistant strains which are of great concern in many countries [31].

Drug resistance in Salmonella can arise either by acquisition of plasmids and transposons or by chromosome mutation [22]. Chromosomal resistance can only be transferred to the progeny cell whereas; resistance plasmids can be transferred to other species of bacteria sharing the same niche by a process called conjugation[32]. It has been documented by Fricke *et al.;* [33] in USA, that ampicillin resistance gene present in commensal gut E. coli was transferred to Salmonella during the course of antibiotic therapy. Also, enteric flora of food animals can also be a source of antibiotic resistant bacteria. Therefore, this study was conducted to determine the drug resistance pattern of Salmonella isolates from animal and human sources and also to investigate the relatedness of these isolates by comparing their resistance patterns.

The prevalence of Salmonella in non-clinical and clinical samples in Bushenyi district was 11.8% and 11.3% respectively. This result was significantly high compared to studies by Alemayehu *et al.;* [6], in Ethiopian abattoirs. This was therefore considered a potential source of food borne salmonellosis. High proportion (83.3%) of Salmonella isolates were resistant to two or more of the antimicrobials that were commonly used in the veterinary and public health. This was an indication of the difficulty in the treatment of clinical and non clinical Salmonellosis. The study indicated the necessity of a further investigation on the prevalence and antimicrobial susceptibility pattern of

Salmonella, by considering it as a potential food borne pathogen, starting from the cattle farms to table. Molecular characterization of the isolates with emphasis on resistant strains was necessary to identify mechanisms of antibiotic resistance [19].

Table 1: Prevalence Of Salmonella Serovars In Clinical Samples
OR =1.05; RR =1.04, n (%), P = .32

Salmonella Isolates	KIUTH	KBH IV	IAH IV	Total
S.typhimurium	5 (29.4)	4 (23.5)	8 (47.1)	17 (63)
S.typhi	3 (60.0)	0 (00.0)	2 (40.0)	5 (18.5)
S.enteritidis	1 (25.0)	2 (50.0)	1 (25.0)	4 (14.8)
S.infantis	0 (0.0)	0 (00.0)	1 (100)	1 (3.70)
Total positive isolates	9 (33.3)	6 (22.2)	12(44.4)	27 (100)
Prevalence	11.3%	7.50%	15%	11.30%

Foot note: n=number of isolates, %=percentage, OR=Odds ratio, RR=Relative risk

Table 2: Prevalence Of Salmonella Serovars In Non-Clinical Samples
P =.0005; OR=1.05; RR=1.04, n (%)

Salmonella serovars	Cattle	Food	Water	Total
S.typhimurium	16 (69.6)	3 (13.0)	4 (17.4)	23(48.9)
S.infantis	4 (100)	0 (00.0)	0 (00.0)	4 (08.5)
S.arizinae	11 (100)	0 (00.0)	0 (00.0)	11(23.4)
S.choleraesuis	5 (55.6)	2 (22.0)	2 (22.2)	9 (19.2)
Total positive isolates	36 (76.6)	5 (10.6)	6 (12.8)	47 (100)
Prevalence's	30%	3.60%	4.30%	11.80%

Foot note: **n=number of serovars, %=percentage, OR=Odds ratio, RR=Relative risk**

Table 3: Antibiotic Resistant Patterns Of Salmonella Serovars From Clinical Samples

Salmonella Serovars (Number tested)	Antibiograms of resistant Isolates (%)							
S. typhimurium	CIP 5(29.4)	CRO 4(23.5)	CAZ 5(29.4)	E 9(53.0)	AMP 14(82.4)	SXT 10(58.8)	C 15(88.2)	TET 16(94.1)
S. typhi	2(40.0)	1(20.0)	0(0.0)	3(60.0)	5(100)	3(60.0)	5(100)	5(100)
S. enteritidis	0(0.0)	1(25.0)	1(25.0)	3(75.0)	4(100)	3(75.0)	4(100)	4(100)
S. infantis	0(0.0)	0(0.0)	0(0.0)	1(100)	1(100)	0(0.0)	1(100)	1(100)

Foot note: CIP=Ciprofloxacin, CRO=Ceftriaxone, CAZ=Ceftazidime, E=Erythromycin, AMP=Ampicilin, CXT=Cotrimoxazole, C=Chloramphenicol, TET=Tetracycline

Table 4: Antibiotic Resistant Patterns Of Salmonella Serovars From Non- Clinical Samples

Salmonella isolates (Number tested)	Antibiograms of Resistant isolates (%) (From non-clinical sources)							
S.typhimurium	CIP 8(34.8)	CRO 9(39.1)	CAZ 8(34.8)	E 10(43.5)	AMP 10(43.5)	SXT 11(47.8)	C 20(87.0)	TET 20(87.0)
S. infantis	1(25.0)	3(75.0)	2(50.0)	2 (50.0)	4 (100)	3 (75.0)	3 (75.0)	4 (100)
S. arizonae	3(27.3)	4(36.4)	0(00.0)	4 (36.4)	1 (9.1)	6 (54.5)	7 (63.6)	9 (81.8)
S. choleraesuis	1(11.1)	1(11.1)	4(44.4)	4(44.4)	7(77.8)	4(44.4)	7(77.8)	9(100)

Foot note: CIP=Ciprofloxacin, CRO=Ceftriaxone, CAZ=Ceftazidime, E=Erythromycin, AMP=Ampicilin, CXT=Cotrimoxazole, C=Chloramphenicol, TET=Tetracycline

Table 5: Susceptibility Patterns Of Isolates From Cinical And Non-Clinical Sources

Sal Salmonella Isolates		Antibiogram Profiles of Susceptible isolates (%)							
	CLINICAL	CIP	CRO	CAZ	E	AMP	SXT	C	TET
Cli	S.typhimurium	12(70.6)	13(76.5)	12(70.6)	8(47.1)	3(17.7)	7(41.2)	2(11.8)	1(5.9)
	S.typhi	3(60.0)	4(80.0)	5(100)	2(40.0)	0(0.00)	2(40.0)	0(0.00)	0(0.00)
	S.enteritidis	4(100)	3(75.0)	3(75.0)	1(25.0)	0(0.00)	1(25.0)	0(0.00)	0(0.00)
	S.infantis	1(100)	1(100)	1(100)	0(0.0)	0(0.00)	1(100)	0(0.00)	0(0.00)
	NON CLINICAL								
Non	S.typhimurium	15(65)	14(60.9)	15(65.2)	13(56.5)	13(56.5)	12(52.2)	3(13)	3(13.0)
	S.infantis	3(75.0)	1(25.0)	2(50.0)	2(50.0)	0(0.00)	1(25.0)	1(25)	0(0.0)
	S.arizonae	8(72.7)	7(63.6)	11(100)	7(63.6)	10(90.9)	5(45.5)	4(36.4)	2(18.2)
	S.choleraesuis	7(87.5)	7(87.5)	4(50.0)	4(50.0)	1(12.5)	4(50.0)	1(12.5)	0(0.0)

Foot note: CIP=Ciprofloxacin, CRO=Ceftriaxone, CAZ=Ceftazidime, E=Erythromycin, AMP=Ampicilin, CXT=Cotrimoxazole, C=Chloramphenicol, TET=Tetracycline

Table 6: P.Values And Risk Estimates Of Salmonella Serovars To The Antibiotics Tested

Antibiotic	P.values	Odds ratio (95%)	Relative risk (95%)
Ciprofloxacin	0.16	1.6 (0.8-3.4)	1.5 (0.9-2.6)
Ceftriaxone	< 0.005	3.4 (1.7-6.9)	2.4 (1.5-4)
Ceftazidime	< 0.005	3.2 (1.5-6.8)	2.4 (1.4-4)
Erythromycin	0.8	0.9 (0.5-1.7)	0.96 (0.7-1.3)
Ampicillin	< 0.005	0.06 (0.02-0.2)	0.6 (0.5-0.7)
Cotrimoxazole	0.2	1.4 (0.8-2.6)	1.2 (0.9-1.6)
Chloramphenicol	< 0.005	0.1 (0.0-0.4)	0.8 (0.7-0.9)
Tetracycline	0.017	0.1 (0.0-0.9)	0.9 (0.9-1)

Foot note: n=number, %=percentage, OR=Odds ratio, RR=Relative risk

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