Salmonella transmission in poultry farms: The roles of rodents, lizards and formites


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ABSTRACT

Introduction: Salmonellosis constitutes a major threat to poultry industry in Nigeria. The study investigated the roles of rodents, lizards and fomites in the transmission of Salmonella in poultry farms.

Methods: A total of 320 samples consisting of litter (n = 70), feed (n = 50), water from drinkers (n = 50), cloacal swabs from chicken (n = 50), faeces from rodents (n = 50) and faeces from lizards (n = 50), were collected from selected poultry farms in Maiduguri, Borno State between March and August, 2011. The samples were subjected to standard bacteriological analyses and presumptive Salmonella isolates were serotyped using slide agglutination technique.

Results: A total of 31 (9.7%) Salmonella isolates comprising 7 serovars were obtained. Salmonella serovars Kentucky and Enteritidis were the major serovars recovered from all sample sources and these represent 3.4 % (n= 11) and 3.1 % (n= 10) respectively. These serovars were the predominant in rodents and lizards and were equally isolated from chicken and other samples, however, Salmonella Hadar was only isolated from litters, but not from chicken or other environmental samples (feed or water).

Significance: It was concluded that rodents and lizards play significant roles in poultry Salmonella infection via environmental contamination. Farmers should focus more attention on these agents of Salmonella carriage in the poultry environment.

Keywords:
Salmonella
Rodents
Lizards
Poultry
Environment

Introduction
Salmonellosis is among the major diseases of public health importance affecting both humans and animals. It is caused by Salmonella and characterised by gastrointestinal disorders (Ao et al., 2015). Salmonella has been reported to cause invasive disease conditions, such as bacteraemia, skin and bone infections, urinary tract infections, meningitis, and splenic abscess (Berendes et al., 2007; Ahmed et al., 2016). Globally, human salmonellosis has been incriminated in approximately 93.8 million cases annually (Majowicz et al., 2010). Salmonella serotypes can be grouped into host specific, host restricted, and generalist serotypes, with important epidemiological and public health implications (Uzzau et al., 2000). Host specific serotypes cause disease in only one host type. Serotypes Paratyphi A, and Typhi for instance, are specific for human while Gallinarum biovars Gallinarum and Pullorum are specific for avian (Karín et al., 2011). Host restricted serotypes majorly affect one host species but can cause disease in other species of animals as well (Uzzau et al., 2000). The generalist serotypes commonly cause disease in a large number of hosts. For instance, Salmonella Typhimurium has been documented from various host species (Gyles, 2004; Ahmed et al., 2017).

Worldwide, avian species are raised for food, companionship, scientific and educational purposes. There has been rapid expansion in poultry industry in Nigeria in spite of challenges such as disease outbreak, the global financial crisis, inadequate credit facilities, and poor infrastructures (FAO, 2008). Salmonellosis is common among galliform birds and has been documented at high rates from commercially reared chicken, turkeys, and other poultry (Ahmed et al., 2017; Raufu et al., 2013; Fasheh et al., 2010). Wild birds and mammals are generally regarded as the main reservoir for Salmonella in the environment. These warm-blooded animals can harbour the bacterium in their gastrointestinal tracts, mostly without showing clinical disease (Robert and Mark, 2004). Infected wild animals can transmit this bacterium from the farm environment via faeces to food animals, and subsequently to man. Although, studies have shown that rodents can be infected with Salmonella (Bastiaan and Aize, 2007), the actual roles of rodents and reptiles in the transmission of salmonellosis is difficult to be related for practical reasons. This study was undertaken to determine the roles of rodent and lizard (Agama) in poultry salmonellosis via environmental contamination.
Materials and methods

Sample collection

Five commercial poultry farms that consented and agreed to participate in the study within Maiduguri metropolitan centre were visited between November, 2010 and February, 2011. A total of 320 samples (64 samples/farm) were collected from litters (n = 70), feed (n = 50), water from drinkers (n = 50), cloacal swabs from layers chicken (n = 50), rodent faeces from poultry pen (n = 50) and lizard faeces from poultry pen (n = 50). Samples of faeces and litters were collected in sterile polythene bags, swabbing was done using sterile swab stick and approximately 5 g of feed was collected in sterile polythene bags while 5 ml of non-medicated water from drinkers were collected into sterile universal bottles. All samples were labelled properly and stored in a box containing ice packs and transported to the Veterinary Microbiology laboratory, University of Maiduguri for analyses.

Isolation and identification of Salmonella species

One gram each of faecal, litter, feed sample and 1 ml of water sample were pre-enriched in 9 ml of buffered peptone water while each cloacal swab with sample was inoculated into 10 ml of buffered peptone water (Fluka Biochemika, Steinheim, Germany). These were incubated at 37°C for 24 h. Thereafter, 1 ml of pre-enriched culture of each sample was inoculated into 9 ml of Selenite-F broth (Fluka Biochemika, Steinheim, Germany) and incubated at 42°C for 24 h. The enriched cultures were inoculated onto deoxycholate citrate agar (DCA) (Laboratorioes Britania, Buenos Aires, Argentina) and incubated at 37°C for 24 h. Non-lactose fermenting colonies with dark centre were sub-cultured onto xylose lysine deoxycholate (XLD) agar (Oxoid, Hampshire, UK) and incubated at 37°C for 18–24 h. All colonies suggestive of Salmonella species on the selective agars were purified on Nutrient agar plates (Lab M, Lancashire, UK) at 37°C for 24 h and subjected to further biochemical typing which included reaction on triple sugar agar, sugar fermentation tests, urease, IMViC (Indole, Methyl red, Voges-Proskauer, Citrate), ONPG (ortho-nitrophenyl galactosidase), amino acid (lysine and arginine) decarboxylase, catalase, and motility (Oxoid Ltd, Hampshire, UK) (Cheesbrough, 2002; Perilla, 2003).

Serotyping of Salmonella isolates

All isolates that showed biochemical characteristics typical of Salmonella were serotyped on the basis of somatic (O), flagellar (H), and capsular (Vi) hyperimmune sera (S and A Reagents Laboratory, Ltd., Bangkok, Thailand). Salmonella O and Vi antigens were identified by slide agglutination procedure. The serotypes were assigned according to the Kauffmann–White scheme (Popoff and Minor, 2007). Serotyping was performed at the WHO National Salmonella and Shigella Centre, Bangkok, Thailand.

Statistical analysis

Prevalence was determined and the significance (p < 0.05) of differences between isolation rates of Salmonella from various sample sources were determined using Chi-square test for independent proportion.

Results

Of the 320 samples collected, 31 (9.7 %) were positive for Salmonella. Salmonellae were isolated from 9 (2.8 %) of the rodents and 8 (2.5 %) each of the lizards and the litter samples, respectively. The association between rates of isolation and sample type was found to be significant (p < 0.05) (Table 1). Seven (7) serovars were obtained in this study, with serovar Kentucky accounting for 3.4 % (n =11), followed by serovar Enteritidis, with isolation rate of 3.1 % (n =10). Salmonella Kentucky which was isolated at the rates of 0.6 % (n=2) and 1.6 % (n=5) from lizards and rodents, respectively, was isolated from all sample sources. Similarly, Salmonella Enteritidis isolated at 0.9 % (n=3) and 0.6 % (n=2) also from lizards and rodents respectively, was equally isolated from all other sample sources, however, Salmonella Hadar, and Salmonella 47:mt- were recovered only from litter, Salmonella Elisabethville and Salmonella Mbadaka were both recovered from lizard only, while Salmonella Bonarensis was obtained only from Rodent (Table 2).

Table 1. Occurrence of Salmonella species from different sources in poultry environment in Maiduguri metropolitan centre

<table>
<thead>
<tr>
<th>Sample</th>
<th>n</th>
<th>n +ve (%)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Formites</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Litter</td>
<td>70</td>
<td>8(2.5)</td>
<td>0.0345</td>
</tr>
<tr>
<td>Feed</td>
<td>50</td>
<td>2(0.6)</td>
<td></td>
</tr>
<tr>
<td>Water</td>
<td>50</td>
<td>2(0.6)</td>
<td></td>
</tr>
<tr>
<td>Faeces</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lizard</td>
<td>50</td>
<td>8(2.5)</td>
<td></td>
</tr>
<tr>
<td>Rodent</td>
<td>50</td>
<td>9(2.8)</td>
<td></td>
</tr>
<tr>
<td>Chicken</td>
<td>50</td>
<td>2(0.6)</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>320</td>
<td>31(9.7)</td>
<td></td>
</tr>
</tbody>
</table>

*p < 0.05; χ² = 12.01, df = 5

Discussion

The isolation rate of Salmonella in this study closely correlated with earlier study on poultry farm from Ibadan, Nigeria in which the isolation rate was 11.0 % (Fashae et al., 2010). However, higher rates of 12.5% in Awka, Nigeria and 16.0 % in Maiduguri, Nigeria were reported by Mike et al. (2004) and Raufu et al. (2013), respectively. This variation might be due to differences in climatic conditions, variation in the farm management practices or differences in the sample sizes in each study, since large sample size can affect the probabilities of obtaining more isolates on culture as previously reported (Raufu et al., 2009; Ahmed et al., 2016).

In this study, the predominant serovars were Salmonella Kentucky and Salmonella Enteritidis. These serovars have also been previously reported to be predominant in poultry and poultry environment (Fashae et al., 2010; Fasure et al., 2013; Raufu et al., 2013). Salmonella Enteritidis and Salmonella Kentucky have been reported to be important serovars causing human salmonellosis in African countries (Galanis et al., 2006; Gordon et al., 2008; Morpeth et al., 2009; Fashae et al., 2010; Raufu et al., 2013), and their prevalence in poultry settings constitute major hazard to public health as poultry products have been suggested...
as the major reservoir of human non-typhoidal Salmonella (Hugas and Beloeil, 2014). Salmonella Kentucky and Salmonella Enteritidis were the major serovars recovered from rodents and lizards in the present study and were equally isolated from all the sources tested. The detection of serovars Enteritidis and Kentucky from all the environmental samples (feed, water and litters) examined in this study may be attributed to environmental contamination by faeces of rodents and lizards, which subsequently served as source of infection to chicken, although there is a need to undertake further studies in future, to confirm this hypothesis by using molecular techniques to study the epidemiological relationship between the serovars isolated from all the sources. Contamination of animal feeds with Salmonella pose a risk of infection to livestock and humans (Davies and Wales, 2010) and studies have shown strong correlations between contamination of feedstuffs and infections of chickens with Salmonella of the same serovars (Davies and Wales, 2010; Raufu et al., 2013; Idowu et al., 2017; Ahmed et al., 2017). It has also been reported that Salmonella remains infective in the faeces of rodents for up to 3 months in the environment (Davies and Wray, 1995; 1996). Rodents and reptiles were reported to be attracted to poultry settings probably because of spilled feedstuffs, availability of water, and the presence of shelter (Bastiaan and Aize, 2007) and the possibility of Salmonella persistence on poultry farms are about two times higher when rodents are present the farm (Rose et al., 2000). Salmonella Enteritidis and Salmonella Kentucky have also been reported from rodents and lizards inhabiting poultry environment (Henzler and Opitz, 1992; Garber et al., 2003). In contrast, serovar Hadar which was predominant in litters was not isolated from any other source. This may be due to the rearing method employed on all the farms sampled (intensive cage management system) in which birds have little or no contact with litters. However, contaminated litters from farms may pose a public health hazard, even though high biosecurity standards in most commercial poultry productions probably minimize the risk (Karin et al., 2011).

**Conclusion**

The present investigation provided insight to the indispensable roles of rodents and lizards in poultry salmonellosis. In addition, the role of environment as a route via which diseases agents could be transmitted to farm animals was also illustrated and therefore, there is a need for farmers to ensure strict biosecurity measures on the farms to minimize rodent/lizard population in and around the farms and reduce the threat of salmonellosis on public health. Furthermore, farmers should practice effective vermin control programmes on their farms when rodent or lizard populations are high.

**Acknowledgments**

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**Conflict of interest**

The authors declare no conflicting interest.

**References**


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**Table 2. Distribution of Salmonella serovars from different sources in poultry environment in Maiduguri metropolitan centre**

<table>
<thead>
<tr>
<th>Sample</th>
<th>No of Sample</th>
<th>47:mt</th>
<th>Kentucky</th>
<th>Hadar</th>
<th>Enteritidis</th>
<th>Bonarensis</th>
<th>Elishabettile</th>
<th>Mbadaka</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Litter</td>
<td>70</td>
<td>1(0.3)</td>
<td>1(0.3)</td>
<td>5(1.6)</td>
<td>1(0.3)</td>
<td>0(0)</td>
<td>0(0)</td>
<td>0(0)</td>
<td>8(2.5)</td>
</tr>
<tr>
<td>Feed</td>
<td>50</td>
<td>0(0.0)</td>
<td>1(0.3)</td>
<td>0(0.0)</td>
<td>1(0.3)</td>
<td>0(0.0)</td>
<td>0(0.0)</td>
<td>0(0.0)</td>
<td>2(0.6)</td>
</tr>
<tr>
<td>Water</td>
<td>50</td>
<td>0(0.0)</td>
<td>1(0.3)</td>
<td>0(0.0)</td>
<td>1(0.3)</td>
<td>0(0.0)</td>
<td>0(0.0)</td>
<td>0(0.0)</td>
<td>2(0.6)</td>
</tr>
<tr>
<td>Lizard</td>
<td>50</td>
<td>0(0.0)</td>
<td>2(0.6)</td>
<td>0(0.0)</td>
<td>3(0.9)</td>
<td>0(0.0)</td>
<td>1(0.3)</td>
<td>1(2.0)</td>
<td>8(2.5)</td>
</tr>
<tr>
<td>Rodent</td>
<td>50</td>
<td>0(0.0)</td>
<td>5(1.6)</td>
<td>0(0.0)</td>
<td>2(0.6)</td>
<td>2(0.6)</td>
<td>0(0.0)</td>
<td>0(0.0)</td>
<td>9(2.8)</td>
</tr>
<tr>
<td>Chicken</td>
<td>50</td>
<td>0(0.0)</td>
<td>1(0.3)</td>
<td>0(0.0)</td>
<td>1(0.3)</td>
<td>0(0.0)</td>
<td>0(0.0)</td>
<td>0(0.0)</td>
<td>2(0.6)</td>
</tr>
<tr>
<td>Total</td>
<td>320</td>
<td>1(0.3)</td>
<td>11(3.4)</td>
<td>5(1.6)</td>
<td>10(3.1)</td>
<td>2(0.6)</td>
<td>1(0.3)</td>
<td>1(0.3)</td>
<td>31(9.7)</td>
</tr>
</tbody>
</table>


Robert, D. and Mark, B. (2004). Observations on Salmonella contamination of eggs from infected commercial laying flocks where vaccination for Salmonella enterica serovar Enteritidis had been used. Avian Pathology, 33(2): 133-144.
