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Occurrences, isolation, identification and susceptibility of *Salmonella* sp. in pigeons at Kamrangirchar thana, Dhaka, Bangladesh

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ABSTRACT

Salmonellosis is one of the most common bacterial diseases of poultry which have a great detrimental effect. A very few number of research previously conducted on occurrence, isolation, identification and susceptibility of Salmonella sp. in pigeon at Bangladesh. Therefore present research was carried out to determine the isolation, identification and susceptibility of Salmonella sp. in pigeons at Kamrangirchar, Dhaka, Bangladesh. A total of 1559 diseased pigeons were examined from the first July to last August 2019. They were grouped, into two categories, according to age squabs (0-21 days) and adults (>22 days). Fifty (50) samples were randomly collected from 15 different small-scale farms in the study area. Fecal samples from live pigeons and tissue (liver and lung) samples from the freshly dead birds were examined to detect, isolate, and identify Salmonella sp. by Gram's staining technique and after enrichment of samples in the peptone water followed by growth in S.S. Agar, and X.L.D. Agar simultaneously. The findings of the study revealed that 32 (64%) of samples were positive for Salmonella sp. It also assured that out of 15 farms, 8 farms were infected with salmonellosis consisting of 305 pigeons. The occurrence of Salmonella sp. at the study area within study period in pigeons was 19.56% (305 in 1559). The present study showed that squabs (55.08%) are more susceptible to salmonellosis than adults (44.92%). This study was conducted for the first time in Kamrangirchar, Dhaka, and the time has come to take the necessary steps to save pigeon farming.

***Key Words**: Pigeons, Squabs, Salmonella sp. and Poultry disease*

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I. Introduction

Bangladesh is the world's most densely populated country, with approximately 170 million people crammed into a land area of only 1,47,570 square kilometers. About 21.8 percent of the country's population is poor and struggles to make ends meet on a daily basis (BBS, 2018). The poultry industry is one of Bangladesh's most promising industries, contributing significantly to the country's economy. This industry is also involved in arranging food security and self-employment, creating purchasing power and reducing poverty at a large scale, supplying quality protein to the people of Bangladesh at the lowest price in the world (Mohammad et al., 2014).

Poultry birds refer to a vast group of birds consisting of about 12 species. Chickens, ducks, geese, swan, guinea fowl, quail, pigeon, pheasant, turkeys, emu and ostriches are generally considered as poultry birds (The American Heritage: Dictionary of the English Language, 2009). Because of their short reproduction and incubation periods, the poultry industry is the most efficient and cost-effective source of animal protein in the shortest amount of time. However, it is still unable to close the animal protein supply and demand gap due to rising future demand. Although commercial broiler and layer farms fulfil the present requirement of protein, nowadays, people feel monotonous with chicken meat and desire another safe meat. So many people start pigeon farming to meet public demand and become financially prosperous. Bangladesh had a pigeon population of 13.8 million, with 12 percent kept on what are known as commercial farms (BBS, 2017). Such farms, however, were not identified. Pigeons kept in commercial farm conditions constituted less than 1% of Bangladesh's poultry population in no division (Dolberg, 2008). Domestic pigeons have been raised for meat in Bangladesh for a long time. *Columba livia* (Paul et al., 2015), the scientific name for the common pigeon, is the ancestor of the domestic pigeon. Pigeons fall into three groups, i.e. poultry pigeons, carrier and racing pigeons. Their distribution has made them readily accessible subjects for study. They are accustomed to living close to human beings and are readily bred to provide squabs for the table. Besides, some foreign pigeons like Porter, King, etc., cannot hatch their eggs, but our indigenous pigeons can hatch those pigeon eggs. So, it has a good scope for breeding foreign breeds in the country.

The pigeon is one of the indigenous poultry species, reared conventionally by the poor farmers to maintain their livelihood. In the past, female household members are engaged with pigeon rearing. Pigeon rearing has become a popular second job for many people, including students. Pigeons are plentiful and squab meat is in high demand because of its delicacy and flavour (Asaduzzaman et al., 2012). In order to help the livestock sector's GDP growth, poultry producers are hoping that pigeon and quail meat, a potential future substitute for chicken, will be developed (Basit et al., 2006). Salmonellosis is one of the most common pigeon diseases caused by *S. typhimurium* and *S. enteritidis*. The disease is passed down from parents to their young pigeons and it can also be passed down from sick pigeons through contaminated water or food. Rodents, cockroaches and humans can transmit *salmonella sp.*, but this is a rare occurrence. A high mortality rate in pigeon squabs on their first day of life indicates that they have been infected with *Salmonella* sp. (Ahmed et al., 2015). It affects adult pigeons slowly and causes diarrhoea, anorexia and polydipsia in the gastrointestinal tract. Pigeons begin to lose weight and develop joint inflammation. If left untreated, these inflammations can lead to arthritis or even paralysis. Pigeons avoid flying in some cases and become exhausted quickly. The pigeons' spleen and liver grow in size and nodules in their internal organs can develop (Dumitrache, 2013). Young domestic birds have suffered severe losses as a result of this organism. *S. typhimurium* causes significant squab losses in pigeon lofts; squabs either die soon after hatching or develop swollen wing joints, rendering them unable to fly. In fetal cases, infection with this organism causes enteritis, diarrhoea and septicemia. Another significant symptom is neuromotor defects that result in encephalitis or inner ear infection (Ahmed et al., 2015).

However, *Salmonella* sp. infections occur very frequently in pigeons and are the most common carriers of the bacteria. It is tough to control (Filho et al., 2009) and also involved in zoonosis (Mahmud et al., 2011). *Salmonella* sp. is common in the poultry fecal samples (Assèta et al., 2013) as it can survive low pH of GIT (Foley et al., 2013). *Salmonella enterica* (subsp. Enterica serovar Typhimurium variant Copenhagen) is a host-adapted strain (Rabsch et al., 2002; Pasmans et al., 2003). Host-adapted *Salmonella* sp. isolates are extremely lethal for pigeons (Uzzau et al., 2000; Pasmans et al., 2003). The most striking examples of this apparent adaptation to one particular host reservoir are the serotype Typhimurium phage types DT2 and DT99 cultured from pigeons (Rabsch et al. 2002). The study's

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objectives were to know the occurrence, isolate and identify Salmonella sp. and the susceptibility of *Salmonella* sp. in the study area.

II. Materials and Methods

Study area

The study was carried out in the Kamrangirchar area, Dhaka, Bangladesh, where many small-scale pigeon farms are located. Usually, the farmers rear the pigeon as their pet and as racing birds. Sometimes, the pigeons earned glory to the farmers by winning race.

Study period

The study period was two months (From 1st July to 31st August 2019).

Collection of data

Required data of pigeon diseases on the study period was collected from the bird disease register from the Thana Livestock Office and Veterinary Hospital. In the study period, a total of 1559 diseased pigeons were examined.

Sampling

A total of fifty (50) samples consisting of forty (40) fecal samples from different farms and ten (10) liver and lung samples from the freshly dead birds were collected from the area.

Sample collection and processing

About one (01) gram of fecal swab forty (40) was randomly collected from the Recto-Anal Junction of local pigeons in a sterile plastic container in the morning and shifted immediately to CDIL, Dhaka, for further processing investigation. The remaining ten (10) liver and lung samples were collected from the freshly dead pigeons after post-mortem examination, taken in a sterile plastic container and sent in an icebox to CDIL, Dhaka, for further investigation.

Isolation and identification of *Salmonella* Sp.

For 24 hours, the fecal and swab samples (liver and lung) were kept in Buffered Peptone Water that had been pre-enriched. Then, from each BPW tube, a loop full of enriched culture was put on Xylose-Lysine-Desoxycholate Agar (X.L.D. Agar) and Salmonella-Shigella Agar (S.S. Agar) plates and incubated for 24 hours at 37°. The selective differential agar plates were checked to see if they had typical colonies. *Salmonella* sp. was found in both the X.L.D. Agar and the S.S. Agar.

Microscopic study by Gram's staining method

The purity of salmonella colony from X.L.D. Agar/S.S. Agar was confirmed by Gram's staining method was conducted by following the method described in the manual of veterinary investigation laboratory technique (Smith and Hussey, 2005) to study morphology and staining characters.

III. Results and Discussion

A total of fifty (50) samples were collected, consisting which forty (40) were fecal and ten (10) were tissue (liver and lung) samples.

Post mortem findings on suspected salmonellosis in pigeon

After post mortem examination, the observed gross lesions were as

1. Small, white, necrotic foci in the liver (Figure 01).
2. Liver became swollen and fragile.
3. Distinctive coppery bronze sheen on the surface of the liver.
4. Turbid yellow color fluids in the peritoneal cavity.
5. Congested liver.
6. Hemorrhagic ova with prominent thicken stalks.
7. Peritonitis.
8. Pericarditis.
9. Vent was wet with whitish feces.

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Figure 01. Liver exhibiting congestion and multifocal necrosis

In pigeons, splenomegaly, grey necrotic foci with petechial haemorrhage in lungs, pale and discoloured livers were observed. In young pigeons, yolk sac infection, pneumonia, hepatitis were the most common lesions (Shivaprasad et al. 2008). In chronic causes, an abdominal abscess can be found in the pigeon (Bayram et al., 2016).

Isolation and identification of *Salmonella* Sp.

Based on the staining, cultural, morphological and biochemical properties of *Salmonella* sp. positive samples were identified.

Gram's staining characteristics: The following cultural properties were found under the microscope (Figure 02).

1. Gram negative (-).
2. Small in shape.
3. Rod shaped organisms.
4. Organisms usually arranged singly and sometimes in pair.
5. Motile organisms.

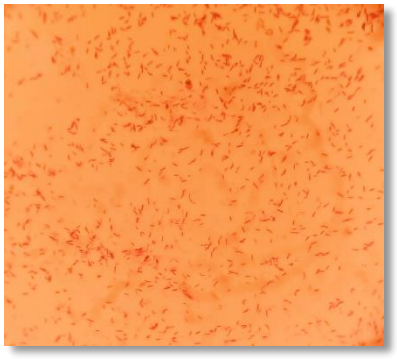

Figure 02. Gram-negative rod-shaped *Salmonella* sp. under microscope (100X).

Morphological characterization revealed that the isolates were Gram negative, short rod shaped organisms and after the motility test they also found that they were motile (Mondal et al., 2008). Kenneth (2004) also reported that *Salmonella* sp. is a motile, rod shaped and gm (-) organism.

Xylose lysine desoxycholate agar (X.L.D agar): Colorless to pale pink colonies, medium red to red violet, colonies mostly with black centered colonies in the X.L.D agar culture plate indicates positive for *Salmonella* sp. (Figure 03). In XLD culture plates, pale pink colonies, medium red to red violet, colonies mostly with black centred were observed has been stated previously by Forbes et al. (2007). Similarly, H. van der Zee (2003) reported that *Salmonella* sp. produces red coloured black centred colonies after 24 hours incubation at 37° temperature in X.L.D. Agar. On the other hand John C. Christenson et al. (2018) revealed X.L.D agar is one of the most used and appropriate selective media for isolating *Salmonella* sp.

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Figure 03. Black centred colonies of *Salmonella* sp. on X.L.D. Agar.

***Salmonella*-Shigella Agar (S.S. Agar):** Colorless, usually with black centred colonies in the S.S. agar culture plate, indicates positive for *Salmonella* sp. (Figure 04). H. van der Zee (2003) stated that *Salmonella* sp. produces colourless black centred colonies after 24 hours incubation at 37° temperature in S.S. agar. Similarly, John C. Christenson et al. (2018) also documented that S.S agar is the most used and best selective media for isolating *Salmonella* sp.

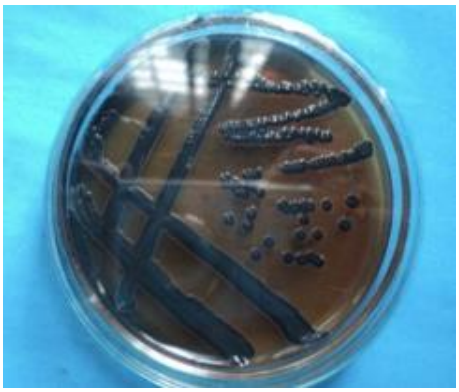

Figure 04. Black centered colonies of *Salmonella* sp. on S.S. Agar

Prevalence of *Salmonella* sp. in study area

A total of 1559 diseased affected pigeons were observed in the study period at TLOVH (Thana Livestock Office and Veterinary Hospital), Kamrangirchar, Dhaka. Fifty (50) samples were randomly collected from 15 different small-scale farms in the study area. Table 01 shows that thirty-two (32) or sixty-four percentage (64%) of samples of fifty (50) pigeons were found positive for *Salmonella* sp. Total fecal samples were forty (40) in which twenty four (24) or sixty percentage (60%) samples were found positive. Total tissue samples were ten (10), of which eight (8) or eighty percentages (80%) of samples were found positive. Our findings are in the same line as Asséta et al. (2013), who reported more than 50% of poultry were positive to the *Salmonella* sp. infection. However, Cedarose et al. (2007) documented a slightly lower percentage of about 47% of poultry were found positive in conventional farms.

Table 01. Positive isolates of *Salmonella* sp. in pigeons of the study area

Total sample	Sample type	No. of sample tested	Cultural examination		Total positive *Salmonella* sp.	
			No. of positive	No. of negative	No.	%
50	Fecal	40	24	16	24	60
	Tissue	10	8	2	8	80
Total	-	50	32	24	32	64

Table 02 shows that the occurrence of *Salmonella* sp. in Kamrangirchar thana, Dhaka was 19.56% within the study period. Rahman et al. (2017) found that the occurrence of *Salmonella* sp. In pigeon is 28.57% in their study.

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Table 02. Occurrence of *Salmonella* sp. in pigeons of the study area.

Total observed diseased population of pigeons	No. of *Salmonella* sp. affected pigeons	Occurrence (%)
1559	305	19.56

Table 03 shows that the age group 0-21 days (squabs) were 55.08% and 22+ days (adults) were 44.92% susceptible to the *Salmonella* sp. infection. The table indicates that the squabs are more susceptible to *Salmonella* sp. infection than the adult pigeons supported by the Ahmed et al. (2015).

Table 03. Susceptibility of *Salmonella* sp. in different age groups of pigeons at the study area

Age group	No. of affected	Susceptibility (%)
0-21 days (Squabs)	168	55.08
22+ days (Adults)	137	44.92

IV. Conclusion

Pigeon farming can emerge as a strong agro-based industry in Bangladesh shortly. One of the significant constraints of pigeon farming is the outbreak of different devastating diseases, including salmonellosis. Study revealed that X.L.D agar and S.S. agar could be excellent culture mediums for *Salmonella* sp. isolation. Out of 50 samples 32 (64%) samples were positive for fecal and tissue sample (lung and liver) *Salmonella* sp. isolates. The occurrence of *Salmonella* sp. at the study area in pigeons were 19.56% (305 in 1559). The present study showed squabs (55.08%) are more susceptible to salmonellosis than adults (44.92%). Due to the inconvenient number of studies on salmonellosis in pigeons in Bangladesh, further study should be conducted to pursue the latest results on the occurrence, isolation, identification and susceptibility of salmonellosis in pigeons in Bangladesh. Since pigeons act as the common carriers of the *Salmonella* sp. and salmonellosis is a zoonotic disease, awareness should be created to control salmonellosis in pigeon. Proper management, feeding and vaccination should be done following the advice of registered veterinarians.

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