

## HAEMOPROTOZOAN INFECTION IN DUCKS: PREVALENCE AND PATHOLOGY

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### ABSTRACT

The prevalence of haemoprotozoa in duck and pathological changes produced by these parasites were studied in different areas of Mymensingh district during July, 2007 to December, 2007. In this study 60% ducks were found to be infected with haemoprotozoa. Two species of haemoprotozoa were identified such as *Leucocytozoon caulleryi* and *Leucocytozoon simondi*. Prevalence of *L. caulleryi* (54.67%) was significantly ( $P<0.01$ ) higher than that of *L. simondi* (5.33%) in ducks. The calculated odds ratio implied that ducks were 19.67 times more susceptible to *L. caulleryi* infection than *L. simondi* infection. Prevalence of haemoprotozoa was significantly ( $P<0.01$ ) higher in male (78.94%) than female ducks (53.57%). Pathological lesions were observed in case of *Leucocytozoon* sp. infections. Grossly lungs were congested but other suspected organs such as liver, lungs, kidney and heart were apparently normal. Microscopically, comma shaped organisms were found in the hepatocyte of liver and alveolar epithelial cell of lungs accompanied with eosinophilic infiltration. Schizonts were also observed in the hepatocyte of liver and alveolar epithelial cell of lungs. The lungs alveoli were distended and ruptured in advanced stages. According to the result, it may be concluded that haemoprotozoan infection in duck is highly prevalent in Bangladesh and produces clinicopathological lesions in duck. So proper control measures should taken to reduce the prevalence of haemoprotozoa of duck in Bangladesh.

**Key words:** Haemoprotozoa, ducks, prevalence, pathology

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### INTRODUCTION

Bangladesh is a developing country where poultry industry is a rising sector. The duck population in Bangladesh is 39.08 millions (Anon, 2007). It plays an important role in rural economy. Local ducks are ubiquitous in the country and smallholder farmers keep them under a subsistent level of management (Islam *et al.*, 2003). But duck rearing is hindered by various problems, of which parasitic infections is one of a major problem.

Many recent studies have focused on avian blood parasites as a model system for host-parasite interactions in an evolutionary and ecological context (Bensch *et al.*, 2004; Hellgren *et al.*, 2004 and Ricklefs *et al.*, 2005). The prevalence of *Leucocytozoon simondi* is 9.2% in ring-necked ducks (*Aythya collaris*) in Florida (Forrester *et al.*, 2001) and 91.2% in Canada (Bennett *et al.*, 1991). The pathogenicity of *Leucocytozoon simondi* has been studied in ducklings. This parasites produces inappetence, weakness listlessness, dyspnea, anaemia, leukocytosis and sometimes death within 24 hr. Splenomegaly and liver degeneration have encountered in *Leucocytozoon simondi* infection in duckling. Extensive tissue damage was noted in spleen and heart of ducks carrying megaloschizont of *Leucocytozoon simondi* (Springer, 1997). In North America, *L. simondi* were associated with heavy mortality of duck especially domestic duck (Shutler *et al.*, 1999). By considering the above mentioned points, this research work was conducted, to detect different haemoprotozoa in duck along with their prevalence and pathological lesion produced by this haemoprotozoa.

### MATERIALS AND METHODS

To study the prevalence of haemoprotozoa of ducks and the pathological changes produced by them, ducks were selected randomly and collected from different local market and from farmer's house of different villages of Mymensingh district.

**Ante-mortem examination**

After collection of birds, age, sex, breed and body weight were recorded. Birds were examined carefully by parting of feathers against their natural direction and simultaneous close inspection to detect clinical signs if any and also to collect ectoparasites.

**Collection and staining of blood and identification of protozoa**

Blood samples were collected from the wing vein of ducks with the help of syringe and needle. The thin smear was made immediately after the collection of blood for identification of blood protozoa. The smear was then air dried, fixed with absolute acetone free methanol, stained with Giemsa's stain and then air dried (Cable, 1957). The slides were then examined under microscope in higher magnification (40X, 100X) for the detection of blood protozoa. Protozoa were identified according to the keys and descriptions given by Levine (1985), Springer (1997) and Soulsby (1982).

**Post-mortem examination**

After ante-mortem examination, birds were slaughtered and allowed to bleed completely and then post-mortem examination was performed as described by Fowler (1990). The skin over-lying the breast and fluff were carefully removed. Then the abdominal wall was incised along the midline, starting from cloaca up to the posterior margin of the keel bone by a scissors. The ribs were then cut just lateral to the keel bone and keel bone was separated. After the opening of thoracic and abdominal cavities, the internal organs such as lung, heart, liver, spleen, kidney and ovary was examined carefully to detect pathological changes.

**Histopathological examination**

During necropsy suspected tissues were collected and fixed in 10% buffered neutral formalin for histopathological studies. Formalin fixed tissue samples were processed, embedded, blocked and sectioned and stained with Haematoxylin and Eosin stain following standard method (Luna, 1968).

**RESULTS AND DISCUSSION****Prevalence of parasites in duck**

In this study, 75 ducks were necropsied whereas 45 (60%) ducks were affected with haemoprotozoa. Ducks were found to be infected with *Leucocytozoon simondi* and *Leucocytozoon caulleryi*. Prevalence of *L. caulleryi* (54.67%) was significantly ( $P < 0.01$ ) higher than that of *L. simondi* (5.33%) in ducks. The calculated odds ratio implied that ducks were 19.67 times more susceptible to *L. caulleryi* infection than *L. simondi* infection (Table 1). Similar studies were also conducted by other scientists in different countries. Benenett *et al.* (1975) reported 30% haemoprotozoan infection in ducks in Nova Scotia and Prince Edward Island. Thul *et al.* (1980) found *L. simondi* infection in 20% ducks in Atlantic Flyway. Williams *et al.* (1977) recorded higher percentage (86%) of *L. simondi* in ducks in Alberta and the Mackenzia Delta. This variation among the present and previous studies may be due to the differences in geographic niches, breeds of ducks, management factors and method of study. In this study only backyard ducks were included and their management is relatively poor than any other system.

Table 1. Haemoprotozoa of duck in Bangladesh

Name of protozoa	Location	Prevalence (%)	Parasitemia	OR
<i>L. simondi</i>	Lymphocyte	4 (5.33)	1-2%	19.67
<i>L. caulleryi</i>	Lymphocyte	41 (54.67)	6-22%	
Total (n = 75)	–	45 (60)	–	–

OR = Odds ratio.

*Haemoprotozoan infection in ducks*

Prevalence of haemoprotozoa was significantly ( $P < 0.05$ ) higher in male (78.94%) than in female (53.47%). The calculated odds ratio showed that male ducks were 3.25 times more susceptible than the female ducks (Table 2). This result could not be compared due to paucity of relevant literatures. The exact cause of higher parasitic infection in male can not be explained. However, the female sex hormones are associated with lower mean parasitic burden (Ackert and Dewhirst, 1950; Todd and Hollingsworth, 1951). This phenomenon may have role in the lower prevalence of haemoprotozoan infection in female.

Table 2. Sex-wise prevalence of haemoprotozoa in duck

Parameter	Name of protozoa	Prevalence (%)	P-value	OR
Male (n = 19)	<i>L. simondi</i>	4 (21.05)	0.685	Male vs female 3.25
	<i>L. caulleryi</i>	11 (57.89)	0.021	
	Total	15 (78.94)	0.0002	
Female (n = 56)	<i>L. simondi</i>	0 (0)	—	
	<i>L. caulleryi</i>	30 (53.57)	0.001	
	Total	30 (53.57)	0.0002	

OR = Odds ratio.

In this study, both *L. simondi* and *L. caulleryi* were found in lymphocytes. In case of *L. simondi* infection, the gametocytes were found in leucocyte and in Giemsa's stain these were more or less elongated measuring upto 14 to 22  $\mu$  long in their host cells. In some cases this size extended upto 45 to 55  $\mu$ . The nucleus of the host cell was seen forming a very long, thin, dark band in one side of the cell. Pale cytoplasmic horns were seen in either sides, extending out beyond the parasite and the nucleus (Fig. 1). The gametocyte found in leucocyte specially lymphocyte. These descriptions were in conformity to the keys and description given by Levine (1985).

In *L. caulleryi* infection, the gametocyte, which was identified from the leucocyte were round and measuring 15.5 by 15.0  $\mu$ . The nucleus of the host cell formed a narrow dark violet coloured band extending about a third of the way around the parasite. Infected cells were somewhat larger (Fig. 2). This description was almost similar to the descriptions of Levine (1985). In *L. caulleryi* infection about 6-22% cells were found to be infected whereas in case of *L. simondi* infection only 1-2% cells were infected. This result could not be compared due to lack of relevant literatures.

***Pathology produced by haemoprotozoa in duck***

Grossly, the lungs were congested (Fig. 3) but there were no other gross lesions in any organs. Microscopically, in lungs, there were lots of eosinophilic infiltration and blood vessels were congested. The congested area contains aberrant structure suggesting the presence of haemoprotozoa (Fig. 4). Few RBC displayed multiple parasitic inclusions. Comma shaped gamete of haemoprotozoa were seen in between the cord of hepatocytes in liver (Fig. 5). Schizont like structures were seen in the alveolar epithelium of the affected lung accompanied with neutrophilic infiltration. The lungs alveoli were distended and ruptured in advance stage (Fig. 6). In kidneys, the capillaries were congested and hemozoinic pigments were present in congested blood. Renal tubules were necrosed with the disintegration of necrotic cellular mass accompanied with focal accumulation of reactive cells (Fig. 7). These findings are somewhat similar to the result described by Yin *et al.* (2002), who studied the pathology of experimentally infected *L. caulleryi* infection in 300 chickens. This change might be due to the liberation of gametocytes from the affected cells.

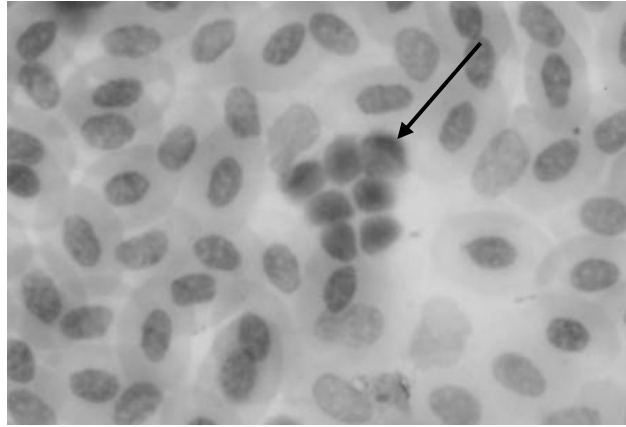


Fig. 1. Developmental stage of *Leucocytozoon simondi* (arrow) in lymphocyte in the blood film of duck, stained with Giemsa's stain (825X).



Fig. 2. *Leucocytozoon caulleryi* (arrow) infected lymphocyte in the blood film of chicken stained with Giemsa's stain (825X).



Fig. 3. Congested lungs affected with *Leucocytozoon* sp. in duck.

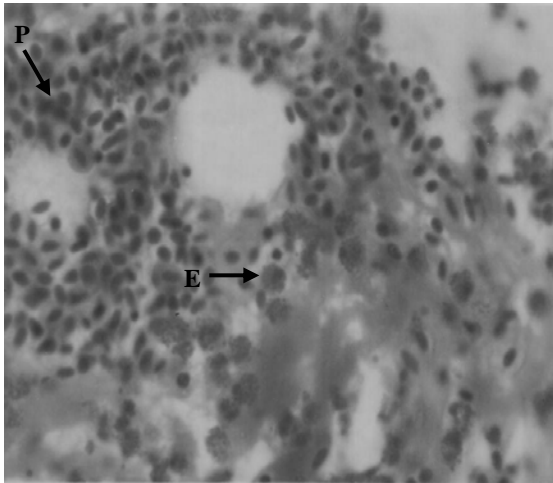


Fig. 4. Lungs infected with hemoprotozoa (P). Characterized by the infiltration of eosinophils (E). Few RBC contain duplicate nuclei seems to be the gamete of protozoa (P) (825X).

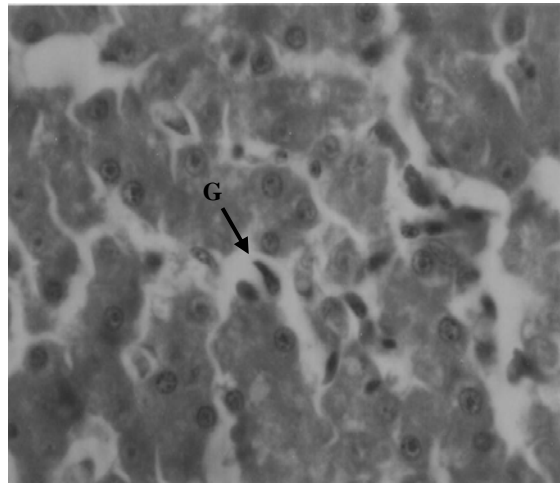


Fig. 5. Liver containing comma shaped gamete (G) of hemoprotozoan in between the cord of hepatocytes (825X).

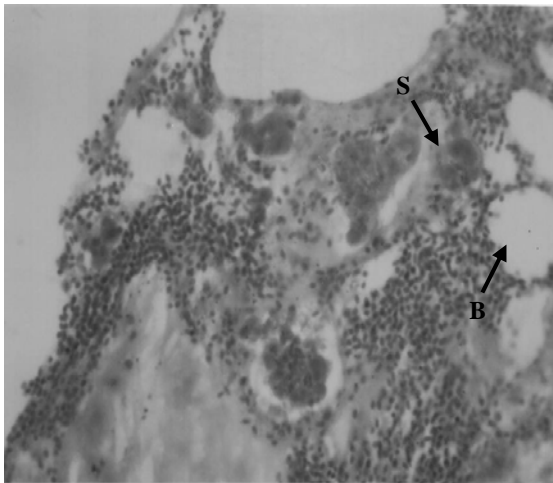


Fig. 6. Lung showing schizonts (S), neutrophilic infiltration and distended ruptured lung alveoli (B) (825X).

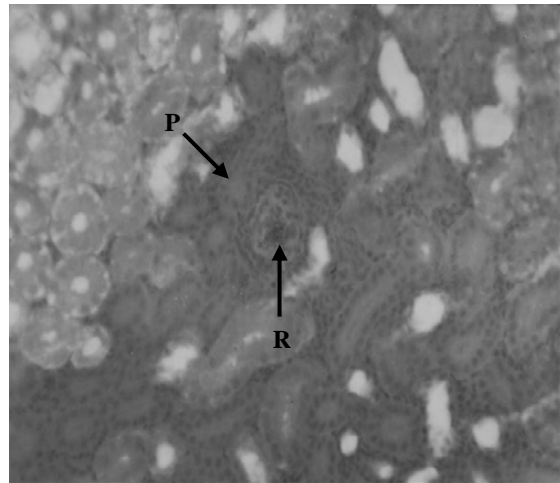


Fig. 7. Necrosis of renal tubular epithelium and blocking of the tubular lumen with proteinaceous mass (P) along with the infiltration of reactive cells (R) in the affected glomeruli (825X).

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