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Prevalence of helminth and protozoan infections in pet birds of Chattogram, Bangladesh

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Abstract

Background: Parasitic diseases such as helminths and protozoa are considered one of the major impediments in the rearing of pet birds. The current study was undertaken to determine the prevalence of helminths and protozoal infections in different captive pet birds in Chattogram metropolitan area, Bangladesh.

Methods: A total of 549 pooled faecal and 311 individual blood samples were collected from different species of pet birds during June 2019 to May 2020. The faecal samples were examined following routine microscopic tests to identify the eggs and oocysts of helminths and protozoan parasites, respectively. Polymerase chain reaction (PCR) was performed to determine the haemoprotozoan parasites.

Results: The prevalence of helminth infestations in pet birds was 8.01% (95% confidence interval [CI]: 5.88–10.61), where infestation caused by nematodes was the highest (7.47%, CI: 5.41–10). *Ascaridia* and *Capillaria* spp. infestations were the commonest helminths recorded in different groups of pet birds. The overall prevalence of intestinal protozoal infections was 11.11% (CI: 8.61–14.04) in pet birds. The most commonly occurring protozoal infections were *Eimeria* spp. (7.83%, CI: 5.73–10.41) followed by *Isospora* spp. (2.91%, CI: 1.67–4.69). The overall prevalence of haemo-protozoan parasites was 2.25% (CI: 0.91–4.58) in different groups of pet birds. The highest prevalence was recorded in *Plasmodium* spp. (1.29%, CI: 0.35–3.26) followed by *Leucocytozoon* and *Haemoproteus* spp.

Conclusions: The data generated in this study are the first of its type, which would be beneficial to the veterinary practice, aviculturists, pet bird owners and breeders in Bangladesh to respond appropriately for the prevention and control of the disease.

KEYWORDS

Chattogram of Bangladesh, haemoprotozoan parasites, helminths prevalence, pet birds, protozoa

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1 | INTRODUCTION

'Pet bird' designates those birds that are housed and bred for an exclusively ornamental purpose. This category includes mainly Passeriformes (e.g., canaries, finches, sparrows, etc.) and Psittaciformes (e.g., parrots, parakeets, budgerigars, lovebirds, etc.) (Mitchell & Mark, 2008). Pet bird rearing is considered an emerging sector in Bangladesh. This trendy pass-time hobby has already become a large commercial endeavour among the youth and contributes to the national economy of the country. Although there are no data available about the number of exotic and pet bird species in the country, the number of species could be more than 30. Many of the species were imported from abroad and bred afterwards to fulfil the demand of pet lovers and aviculturists.

One of the major impediments to this rising sector in Bangladesh is the occurrence of various types of diseases. These birds suffer from different types of diseases like endoparasitic infections (e.g., nematodes, cestodes, trematodes, protozoa) including blood-borne protozoan infections (e.g., Leucocytozoonosis, avian malaria, Haemoproteosis) (Hellgren et al., 2004; Urquhart et al., 1996). In most cases, birds infested with intestinal parasites (e.g., helminths and protozoa) remain asymptomatic other than in severe conditions. Clinical signs varied from anorexia, dullness, decreased feed conversion ratio, reduced body weight, diarrhoea, obstruction of the intestine with a mass of worms, reduced egg production, death of the birds and so forth (Harrison & Lightfoot, 2006). *Eimeria, Isospora, Ascaridia, Capillaria* and *Heterakis* spp. are mostly found in pet birds as GI parasites (Globokar et al., 2017; Hasan et al., 2018).

Other than enteric parasitic infestations, blood parasites are considered another major hindrance in pet bird rearing (Hong et al., 2021; Hellgren et al., 2004). Haemoproteus species are the most common and widespread hemoprotozoan parasites of birds. Some pathogenic species of Haemoproteus cause severe myositis in avian hosts (Tizzani et al., 2020). Leucocytozoon species is another group of vector-borne protozoan parasites of pet birds and many species associated with mortality (Jia et al., 2018). Avian malaria caused by the Plasmodium species is another major mosquito-transmitted disease of pet birds. The haemoparasitic infections are associated with reduced growth and production and reproduction of the birds (Marzal et al., 2005). In severe cases, these diseases may lead to increased mortality of the pet birds (Beadell et al., 2006). However, Plasmodium spp. of the avian species has developmental and morphological characteristics approximately similar to the genera such as Leucocytozoon and Haemoproteus. Therefore, molecular diagnostic tools should be employed to differentiate these blood-borne protozoan infections in pet birds (Hellgren et al., 2004).

Epidemiological investigation of the parasitic infections in birds in a region is very important for the documentation and designing of appropriate prevention and control strategies. To the best of our knowledge, there are few (Hasan et al., 2018) or no specific study has been conducted to investigate the occurrence of endoparasitic infections (e.g., helminths and protozoa) in the country. Therefore, the current study was undertaken to determine the occurrence of GI parasites (e.g., nematodes, cestodes, trematodes, protozoa) and blood-borne protozoan diseases (e.g., *Leucocytozoon, Plasmodium* and *Haemoproteus* spp.

infections) in different groups of captive pet bird species in Chattogram metropolitan areas of Bangladesh.

2 | MATERIALS AND METHODS

2.1 Study design

The current study was undertaken for a period of 12 months starting from June 2019 to May 2020 in Chattogram metropolitan areas of Chattogram district, Bangladesh. A cross-sectional study was designed to collect the pet birds' samples. A standard questionnaire was used to collect demographic data such as the owner's name and address, pet bird species and other relevant information. The selection of the study areas was based on the availability of a higher number of pet bird species in the region.

2.2 Sample collection and preservation

Pooled faeces and individual blood samples were collected from the different species of pet birds. A total of 549 pooled faecal samples (1 pool = 5 random faecal samples) representing 2745 birds of five orders and 23 species were collected to determine the GI parasitic infestation (Supplementary Table S1). Approximately 3–5 gm freshly voided faeces (for each pooled sample) were collected in a plastic container having 10% formalin and refrigerated at 4°C until further analyses.

Further, a total of 311 individual blood samples were collected from six species of Psittaciformes for the determination of blood parasites. Blood samples were collected aseptically by cutting the tips of the nail and a drop of blood from each bird was then immediately put on the Whatman FTA classic cards (Qiagen, Germany) (Stowell et al., 2018). These cards were also kept in refrigerator until further analyses. All the laboratory examinations were performed at Parasitology and Molecular Pathology laboratories of Chattogram Veterinary and Animal Sciences university.

2.3 Examination of samples

2.3.1 Examination of faecal samples

The samples were examined following routine microscopic tests (e.g., direct smear, flotation and sedimentation) to identify the morphological features of eggs and oocysts of helminths and protozoan parasites, respectively (Hendrix & Robinson, 2016; Soulsby, 1982; Urquhart et al., 1996). Briefly, the individual faecal suspension was prepared by homogenising and straining each pooled sample. The direct smear was then carried out by taking a drop of faecal suspension on a glass slide. For the floatation technique, 5 ml of faecal suspension was mixed with 15 ml flotation fluid (sugar-salt solution) and kept in a 20-ml test tube by putting a coverslip on the convex meniscus of the fluid. After 15 min, the coverslip was transferred to a glass side for microscopic

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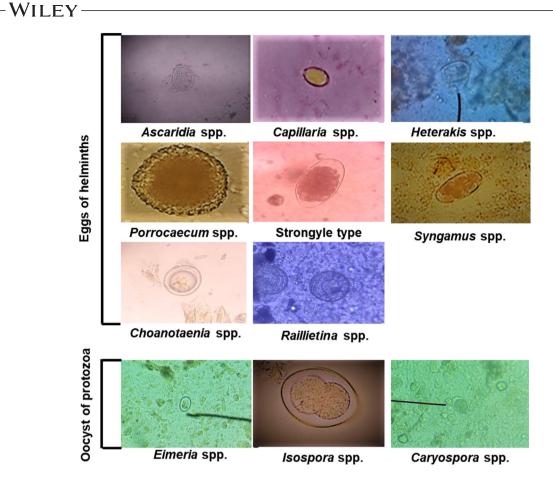


FIGURE 1 Eggs of helminths and oocyst of protozoa identified in pet birds of Chattogram

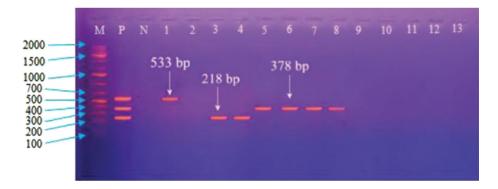


FIGURE 2 Polymerase chain reaction (PCR) confirmation of *Leucocytozoon, Plasmodium and Haemoproteus* at the genus level. Lane M: 2 kb DNA ladder; Lane P: positive control; Lane N: negative control; Lane 1: *Haemoproteus sp*, amplicon size 533bp; Lane 2-4: *Leucocytozoon*, amplicon size 218bp; Lane 5-8: *Plasmodium*, amplicon size 378bp.

examination. For the sedimentation technique, the faecal suspension was kept aside for 15 min, and then a drop of the sediment was examined under the microscope. For each of the sample, duplicate smears were prepared and examined. A sample was considered 'positive' when at least 'one egg' or 'oocyst' was detected in the smears tested. However, the helminths and protozoan species were detected up to the genus level (Atkinson et al., 2009; Hendrix & Robinson, 2016; Soulsby, 1982; Zajac & Conbody, 2012) (Figures 1 and 2).

2.4 | Examination of blood samples

2.4.1 DNA extraction and PCR assay

Total genomic DNA was extracted from the individual blood sample using the commercially available 'FavorPrep Blood Genomic DNA Extraction Mini Kit' (Cat. No: FABGK001, Taiwan) following the manufacturer's instructions with some modifications. Polymerase chain

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TABLE 1 List of multiplex PCR primers for the identification of blood parasites in pet birds

Parasite genera	Primer name	Primer sequence (5' to 3')	Product size (bp)	Reference
Leucocytozoon	LMF	TGGAACAATAATTGSATTATTTACAYT	218	Ciloglu et al. (2019)
	LMR	AACATATCATATTCCATCCATTTAGATTA		
Plasmodium	PMF	CCTCACGAGTCGATCAGG	377-379	
	PMR	GGAAACCGGCGCTAC		
Haemoproteus	HMF	ATTGGATGTCAATTACCACAATC	525-533	
	HMR	GGGAAGTTTATCCAGGAAGTT		

reaction (PCR) was then performed using a multiplex primer list according to the previously published reports (Ciloglu et al., 2019; Table 1). The multiplex PCR reaction was set up in a 25 μ l final volume containing 12.5 μ l master mix, 0.5 μ l forward primer and 0.5 μ l reverse primer for each three species (total 3 μ l), 2.5 μ l DNA template and 7 μ l nuclease free water. The PCR conditions had an initial denaturation step of 95°C for 15 min followed by 35 cycles of denaturation at 94°C for 30 s, annealing at 59°C for 90 s, extension at 72°C for 30 s and a final extension step at 72°C for 10 min. Then, 5 μ l of amplified amplicons was taken and stained using 1% ethidium bromide (Sigma-Aldrich) followed by visualisation of the band after agarose gel (1.5%) electrophoresis (Figure 2).

2.5 | Statistical analysis

The data were manually checked for quality before coding on Microsoft Office Excel 2016. The data were analysed using the statistical tool STATA/IC-13.0 (StataCorp, 4905). We used descriptive statistics for helminths, and blood protozoa of pet birds such as frequencies, percentages and 95% confidence interval (CI).

3 | RESULTS

3.1 | Occurrence of helminth infestations in pet birds

The overall prevalence of endoparasitic (helminths and protozoa) infections was 19.13% (95% CI:15.92–22.67) in different groups of pet birds (Tables 2 and 3). The overall prevalence of helminths was 8.01% (95% CI: 5.88–10.61) in different pet bird species (Table 2). The highest overall prevalence was recorded for nematodes (7.47%, 95% CI: 5.41–10), compared to cestodes (0.55%, 95% CI: 0.11–1.59). Among the nematodes, the overall infestation caused by *Ascaridia* spp. was the highest (3.46%) followed by *Capillaria* spp. (1.28%) and other helminths. In different pet bird groups, *Ascaridia* spp. infestation was the highest in the pigeon (28.57%) of Columbiformes followed by Kadaknath (25%) of Galliformes, finch (15.38%) of Passeriformes, macaw (12.50%), parrots (9.09%) and white cockatoo (9.09%) of Psittaciformes groups of birds. Red-winged parrot (25%), Gouldian finch (14.29%) of Psittaciformes and pigeon (7.14%) of Columbiformes groups of birds were also commonly infested with *Capillaria* spp. The overall prevalence of cestodes (*Choanotaenia* and *Raillietina* spp.) was very low in pet birds, where the infestation with *Raillietina* spp. was only found in yellow-crested cockatoo (11.11%; Table 2).

3.2 Occurrence of enteric protozoal infections in pet birds

The overall enteric protozoal infections were 11.11%, (95% CI: 8.61– 14.04) in the different groups of pet birds. The overall infection caused by the *Eimeria* spp. was the highest (7.83%, 95% CI: 5.73–10.41) followed by *Isospora* and *Caryospora* spp. (Table 3). Further, among different groups of pet birds, the prevalence of *Eimeria* spp. was common in blue-and-yellow macaw (33.33%), eastern rosella (14.29%), horned parakeet (12.50%), lovebird (10%), yellow-crested cockatoo (11.11%), cockatiel (10.53%) of Psittaciformes birds and European goldfinch (10%) of Passeriformes birds. Furthermore, the occurrence of *Eimeria* spp. was also common in Accipitriformes, Columbiformes and Galliformes birds. The occurrence of *Isospora* spp. was recorded in yellowcrested cockatoo, cockatiel and budgerigar. Whereas, *Caryospora* spp. was observed in rose-ringed parakeet and budgerigar of Psittaciformes birds (Table 3).

3.3 | Occurrence of haemoprotozoan infections in pet birds

The overall prevalence of blood parasites was 2.25% (95% CI: 0.91– 4.58) in different groups of pet bird species (Table 4). The highest overall prevalence was recorded for *Plasmodium* spp. (1.29%) followed by *Leucocytozoon* and *Haemoproteus* spp. In different pet birds, the occurrence of *Plasmodium* spp was the highest in macaw (4%) followed by lovebirds (2.38%). The frequency of *Leucocytozoon* spp. was the highest in the parrot (4%) and the cockatiel (2.78%). *Haemoproteus* spp. infection was only recorded in the budgerigar (Table 4).

4 DISCUSSION

The number of pet bird lovers increasing in Bangladesh in recent years due to their ornamental significance. This hobby commences

		Intestations in pet bir	us or Unattogram						
		%, (95% confidence interval), N	erval), N						
		Nematodes						Cestodes	
Order	Common Name (N)	Ascaridia spp.	Capillaria spp.	Heterakis spp.	Porrocaecum spp.	Strongyle type	Syngamus spp.	Choanotaenia spp.	Raillietina spp.
Psittaciformes	Budgerigar (290)	1.03,(0.21–2.99), 3	0.34, (0-1.91), 1	1	1.03, (0.21–2.99), 3	0.34, (0-1.91), 1	0.34, (0–1.91), 1	0.69, (0-2.47), 2	1
	Cockatiel (38)	I	7.89, (1.66–21.38), 3	I	I	I	I	I	I
	White cockatoo (11)	9.09, (0.23-41.28), 1		9.09, (0.23-41.28), 1		9.09; (0.23-41.28), 1	I	I	I
	Yellow-crested	I	I	I	I	I	I	I	11.11,
	cockatoo (9)								(0.28-48.25), 1
	Galah (19)	5.26, (0.13-26.03), 1	I	I	I	I	I	I	I
	Parrots (22)	9.09,(1.12–29.16),2	I	I	I	I	I	I	I
	Red winged Parrot (4)	I	25,(0.63–80.59), 1		ı	ı	I	I	I
	African Gray Parrot (14)	7.14,(0.18–33.87), 1	T	7.14; (0.18–33.87), 1	I	I	I	I	I
	Blue-fronted Parrot(17)	I	I	I	I	I	I	I	1
	Lovebird (20)	5,(0.13-24.87), 1	I	I	I	I	I	I	I
	Lories(5)	ı	I	20,(0.51-71.64), 1	I	I	I	I	I
	Horned Parakeet (8)	I	ı	I	I	I	I	I	ı
	Rose-ringed	I	I	I	1	1	ı	ı	I
	parakeet (o) Eastern Rosella (7)	1		I	I	I	I	1	I
	Macaw (8)	12.50,(0.32-52.65), 1	I	I	I	I	I	I	I
	Blue-and-yellow macaw (3)	I	I	I	I	ı	I	I	1
Passeriformes	Canary (17)	1	1	1	5.88,(0.15–28.69), 1	1	I	I	1
	European Goldfinch (10)	10, (0.25-44.50), 1	I	1	1	1	I	I	1
	Finch (13)	15.38,(1.92-45.45), 2	I	7.69,(0.19-36.03), 1	I	7.69,(0.19–36.03), 1	I	I	I
	Gouldian Finch (7)	14.29,(0.36-57.87), 1	14.29,(0.36–57.87), 1	1	1	1	I	I	1
Accipitriformes	Eagle (1)	Т	ı	I	I	I	I	т	Т
Columbiformes	Pigeon (14)	28.57,(8.39–58.10), 4	7.14,(0.18-33.87), 1	7.14, (0.18–33.87), 1	I	7.14,(0.18-33.87), 1	T	I	I
Galliformes	Kadaknath (4)	25,(0.63-80.59), 1	1	I	I	I	25, (0.63-80.59), 1	T	I
Total (overall)	549	3.46,(2.10–5.35), 19	1.28,(0.51-2.61), 7	0.91,(0.30-2.11), 5	0.73,(0.20-1.85), 4	0.73,(0.20–1.85), 4	0.36, (0–1.31), 2	0.36, (0-1.31), 2	0.18, (0-1.01), 1

 TABLE 2
 Occurrence of helminth infestations in pet birds of Chattogram

TABLE 3 Occurrence of enteric protozoal infections in pet birds of Chattogram

		%, (95% Confidence interv	al), N	
Order	Common name (N)	Eimeria spp.	lsospora spp.	Caryospora spp.
Psittaciformes	Budgerigar (290)	7.93, (5.09–11.66), 23	4.14, (2.16–7.12), 12	0.34, (0-1.91), 1
	Cockatiel (38)	10.53, (2.94–24.80), 4	7.89, (1.66–21.38), 3	-
	White cockatoo (11)	-	-	-
	Yellow-crested cockatoo (9)	11.11, (0.28–48.25), 1	11.11, (0.28–48.25), 1	-
	Galah (19)	-	-	-
	Parrots (22)	-	-	-
	Red-winged parrot (4)	-	-	-
	African grey parrot (14)	7.14, (0.18-33.87), 1	-	-
	Blue-fronted parrot (17)	5.88, (0.15-28.69), 1	-	-
	Lovebird (20)	10, (1.23–31.70), 2	-	-
	Lories(5)	-	-	-
	Horned parakeet (8)	12.50, (0.32–52.65), 1	-	-
	Rose-ringed parakeet (8)	-	-	12.50, (0.32–52.65), 1
	Eastern rosella (7)	14.29, (0.36–57.87), 1	-	-
	Macaw (8)	-	-	-
	Blue-and-yellow macaw (3)	33.33, (0.84–90.57), 1	-	-
Passeriformes	Canary (17)	5.88, (0.15-28.69), 1	-	-
	European goldfinch (10)	10, (0.25-44.50), 1	-	-
	Finch (13)	7.69, (0.19-36.03), 1	-	-
	Gouldian finch (7)	-	-	-
Accipitriformes	Eagle (1)	100, (2.50–100), 1	-	-
Columbiformes	Pigeon (14)	21.43, (4.66–50.80), 3	-	-
Galliformes	Kadaknath (4)	25, (0.63–80.59), 1	-	-
Total (overall)	549	7.83, (5.73-10.41), 43	2.91, (1.67-4.69), 16	0.36 (0-1.31), 2

TABLE 4 Occurrence of haemoprotozoan infections in pet birds of Chattogram

		%, (95% Confidence Interval), N		
Order	Common name (N)	Leucocytozoon spp.	Plasmodium spp.	Haemoproteus spp.
Psittaciformes	Budgerigar (138)	-	0.72, (0-3.97), 1	0.72, (0-3.97), 1
	Cockatiel (36)	2.78, (0-14.53), 1	-	-
	Lovebird (42)	-	2.38, (0-12.57), 1	-
	Lories (20)	-	-	-
	Parrot (25)	4, (0.10-20.35), 1	-	-
	Macaw (50)	-	4, (0.49–13.71), 2	-
Total (overall)	311	0.64, (0-2.30), 2	1.29, (0.35–3.26), 4	0.32, (0-1.78), 1

already to become a large commercial endeavour and contributes to the national economy. However, parasitic infections are remarkable constraints for maintaining the good health of pet birds. It is demonstrated that parasitic infections were common in pet birds. They are suffering from helminth and blood protozoal infections as well. These infections lead to anorexia, decrease body weight, diarrhoea and reduced egg production followed by the death of the birds.

The overall prevalence of endoparasitic infections detected in this study was almost similar to the percentage observed in pet birds of Japan (22.5%; Tsai et al., 1992) and Italy (27%; Papini et al., 2012). However, our findings were lower than the findings of Hasan et al. (2018),

who witnessed 45% in game birds in Dhaka city of Bangladesh. Since the transmission of all GI parasites occurred through the oro-faecal route, contaminated food, water and soil play a significant contribution to the variation of such prevalence. Among helminths, the overall prevalence of nematodes in this investigation was lower than the pet birds of Italy (19%; Papini et al., 2012). Within the nematodes, Ascaridia spp. caused for the highest infection, which was showed consistency with data of pet birds of Japan (1.3%; Tsai et al., 1992); Germany (2.6%; Globokar et al., 2017), Brazil (4.12%; Lima et al., 2017). However, our finding was lower than the prevalence (21.67%) of Ascaridia spp. infestation in game birds in Dhaka city (Hasan et al., 2018) and pigeon (16%) of Chattogram and Sylhet of Bangladesh (Hoque et al., 2014). Geographical distribution, climatic condition and method of rearing could be major reasons for variation in the prevalence of nematodes. However, it is indicated that the prevalence of nematodal infections was found moderately high in this study.

In this study, Ascaridia spp. infestation found the highest in pigeons, which was concordant with different studies conducted in Bangladesh such as 28.33% in Dhaka (Begum & Sehrin, 2012), 35% in Chittagong metropolitan area (Ghosh et al., 2014), 22.81% (Khanum et al., 2018) and 31.74% in Rajshahi (Rahman et al., 2019). Ascaridia spp. infestation in the Galliformes group was also high in this study, which was consistent with the findings of other previous studies that detected 30%-42% in different countries (Abdullah et al., 2021; Alam et al., 2014; Globokar et al., 2017). In addition, the availability of earthworms has a significant role in the occurrence of Ascaridia spp., as it acts as a transport host (Soulsby, 1982). In this study, the occurrence of *Capillaria* spp. was almost similar in pet birds of Germany (2.6%-5.27%; Globokar et al., 2017) and in pet birds of Brazil (3.03%; Lima et al., 2017). Variations in sampling methods, treatment with specific anthelmintics, feeding and managemental practices could be the reasons of the variation of our results. While in the case of chicken and pigeon, the feed is generally offered on the potentially contaminated ground, and feeding dishes are provided for budgerigar, lovebird, macaw and so forth.

Heterakis spp. infestation in this study found mostly similar to the findings of Lima et al. (2017), who detected 1.02% in Northeastern Brazil. The prevalence of strongyle type parasites and *Syngamus* spp. infections was found similar to a previous study in Italy, which observed 1.6% (Papini et al., 2012). In cestodes, the overall prevalence of *Raillietina* spp. infestation was found very low in this study and was mostly similar to the birds in Europe (2.6%; Globokar et al., 2017). The low parasitic infestation might be due to good managemental practice, regular anthelmintics use and less availability of intermediate hosts like ants, beetle or other arthropods.

It was noted that there were no trematode infestations diagnosed in pet birds. Akram et al. (2019) could not detect any trematode eggs in captive birds from Punjab, Pakistan, and Hasan et al. (2018) observed the same in the pet birds of Dhaka city of Bangladesh. For the completion of the life cycle of trematodes, they require one or more intermediate hosts (e.g., snails) and captive rearing of pet birds could be one of the possible reasons for not detecting such infestations in our study (Ebbs et al., 2018). In comparison to helminths, overall enteric protozoal infections (except *Syngamus* spp. infection) were found higher in this study. The infection caused by the *Eimeria* spp. was the highest, and this finding was similar to others who detected 6.3% in pet birds of Italy (Papini et al., 2012) and 7.1% in Iran (Badparva et al., 2015) and 13.13% in Dhaka, Bangladesh (Hasan et al., 2018). However, this finding was much higher than the observation of a prior study in pet birds of temperate countries (0.1%; Globokar et al., 2017). These variations might be due to many environmental factors like different temperatures and humidity in different countries. Mayer and Donnelly (2012) observed that at low temperatures, coccidian parasites cannot survive in the environment. The second most intestinal protozoal infection was isosporiasis. This finding was similar to the findings of Globokar et al. (2017). As the climate of Chattogram is hot and humid, the prevalence of coccidian parasites like *Eimeria* and *Isospora* might be higher.

The occurrence of blood parasitic infections in pet birds was found very low in this study. Some studies found higher rates of blood parasitic infections in different areas of the world; 10.6% in Japanese wild birds (Murata, 2002), 12.4% in birds in Costa Rica (Valkiunas et al., 2004). 21.1% in birds of the Gulf Coast sites in Mexico (Garvin et al., 2006) and 10.7% in social birds from a neotropical savanna in Brazil (Fecchio et al., 2011). These variations might be due to the habitates of birds as these studies were conducted in free-ranging birds. The prevalence of Leococytozoon spp. infection of this study was almost similar to the observation in different birds species of São Paulo State, Brazil (0.06%; Bennett & Lopes, 1980) and Costa Rica (0.3%; Valkiunas et al., 2004). Some studies also observed a higher prevalence of such blood parasites in birds of the west African rainforest (4.6%; Sehgal et al., 2005), Nearctic-neotropical passerine birds of Mexico (1.3%; Garvin et al., 2006) and naturally infected birds of Africa (4%: Valkiunas et al., 2009). These variations might be due to the dissimilitude in the method of study, as our study was conducted through multiplex PCR, while others conducted through microscopic examination. Valkiunas et al. (2009) proved that there are significant variations among the findings of the same study only because of the methods of detection of blood parasites. Our study indicated that the prevalence of Leucocytozoon spp. infection was comparatively lower at Chattogram region.

The prevalence of *Plasmodium* spp. infection was approximately similar to Bennett and Lopes (1980; 1.8%), which was conducted among different types of birds from São Paulo State, Brazil, 1.7% in Japanese wild birds (Murata, 2002) and 1.9% in the birds from Madagascar (Savage et al., 2009). However, the finding was slightly lower than passerine birds from central New Jersey (3%; Kirkpatrick & Suthers, 1988) and 3.6% in birds from a neotropical savanna in Brazil (Fecchio et al., 2011). Moreover, our finding was somewhat higher than the prevalence of *Plasmodium* spp. infection from various bird groups in Costa Rica (0.6%; Valkiunas et al., 2004). These variations might be due to variation in vector availability in different geographical locations such as all these studies were conducted on free-ranging birds, whereas our study focused on captive pet birds.

In this study, the prevalence of *Haemoproteus* spp.infection was mostly similar to Hellgren et al. (2004), who reported 1.2% of a single lineage by PCR method among different avian species in Sweden. This

finding was moderately smaller than the Japanese wild birds (5.1%; Murata, 2002), different birds from Costa Rica (4.8%; Valkiunas et al., 2004) and from West African rainforest birds (7.7%; Sehgal et al., 2005). Sample size, management system, uses of fly repellent and so forth might be influenced by the prevalence rate of this blood parasite.

5 | CONCLUSION

The occurrence of enteric infections caused by nematodes (*Ascaridia*, *Capillaria*, *Heterakis* spp.) and protozoa (*Eimeria* spp.) are more common than cestode (*Raillietina* spp.) in pet birds of Chattogram. Nevertheless, blood-borne protozoan diseases like Leukocytozoonosis, avian malaria and Haemoproteosis are less likely to be common in the studied pet birds. The principal constraint of this study was ununiform sample size for all groups of pet birds due to the lack of availability such. For a greater understanding of helminths and blood parasitic infections in pet birds, further extensive studies are required throughout the country.

AUTHOR CONTRIBUTIONS

Sample collection, all laboratory works (DNA extraction, PCR, microscopic examination), data analysis and writing of the original draft of the manuscript: Mohammad Bayzid. Sample collection, laboratory works (DNA extraction, PCR): Farazi Muhammad Yasir Hasib. Data analysis, writing and review of the manuscript: Tanjila Hasan, Mohammad Mahmudul Hassan. Funding accusation, supervision, resources: Mohammad Masuduzzaman, Mohammad Alamgir Hossain. Research concept, methodology, fund acquisition, study design, supervision, interpretation of the results, writing and reviewing of the manuscript: Mohammad Abdul Alim.

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CONFLICT OF INTEREST

The authors declare no conflicts of interest.

DATA AVAILABILITY STATEMENT

Data files associating this research are available at: https://figshare. com/s/55f1e42a8fe83eed02e6

ETHICS STATEMENT

Ethical approval was obtained from the institutional ethical approval committee of Chattogram Veterinary and Animal Sciences University (CVASU) [CVASU/Dir (R&E) EC/2019/126(13)].

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