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Gastrointestinal Parasites of Common Teal (Anas Crecca) in the Wetlands of Punjab, Pakistan

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Abstract – During the present study total 40 samples (20♂ 20♀) were collected from different six wetlands of Punjab Pakistan. Among these samples total 17 samples (08% ♂, 09% ♀) were found infected with different gastro- intestinal parasites. The parasitic load was almost equally shared by the bird sampled from different sites (Chashma Barrage 37.50%; Taunsa Barrage and Head Marala 40% both; waterlogged area of Bahawalnagar 44.4% and Head Sulaimanki and waterlogged areas of Faisalabad 50% both. The frequency of distribution in two sexes vary at different sampling sites, parasitic load was almost equally shared by both sexes (40% ♂, 45% ♀), and difference of both sexes were statistically non-significant (P>0.05). The total seven species of Gastrointestinal parasites were identified which belongs to four classes/phylum (Nematode, Trematodes, Cestodes and protozoan) and five families (*Dioctophymidae, Echinostomatidae, Hymenolepidiae, Hexamitidae*and *Cryptosporidiidae*), in which Nematodes 15%, Trematodes 32%, Cestodes 23% and Protozones 30 %. The total seven species of parasites which were recorded as *Hystrichis tricolor*, *Echinostoma revolutum, Hypoderaeum conoideum, Hymenolepis longicirrosa, Diorchis stefanski, Giardia sp.* and *Cryptosporidium sp*.
There is inadequate information on the parasites of wild migratory birds and this study will help to understand host-parasite interaction and provides valuable base line information for the evaluation of strength and health status of this species.

Keywords – ***Parasitic fauna, Common Teal, Wetlands, Punjab, Pakistan.***

I. INTRODUCTION

Ducks are hardy and fairly disease resistant birds, also they are known for being good scavengers outside of their nests. So, more risk factors are affected by these rural birds than industrial species. However, this scavenger bird can be considered as final or intermediate host of many helminths or protozoan parasites, the eggs of gastrointestinal parasites are mostly passed through the feces and shed into the environment of birds and can distribute infection to human and other animals [1].

Because of the food and feeding habit Common teal acquires parasitic infection from their habitats. Jones *et al.* (2005) reported mammals reported that the genus *Paramonostomum* Lühe, 1909 was found in the intestinal tract of birds. Parasites are markers of bird migration pathways and feeding habits, and therefore need to be researched. Helminthes of *Anatidae* living on Lake Ladoga have been partially studied [2].

The common teal is highly sensitive to *Sarcocystis* cysts that were discovered in the bodies of common Teal hunted in Lithuania and Finland [3]. Moreover, *Toxoplasma gondii* was found in 12 species of Teal found at the wetlands of Italy, when inspected with advanced agglutination tests for *Toxoplasma gondii.* In some studies it was also found that

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these Teals are also responsible for transmitting the virus specifically, Avian Influenza Viruses in Europ[4].

II. Methodology

2.1 Study area

There are six wetlands selected for the sample collection including, Head Marala, Chashma Barrage, Taunas Barrage, Head Sulaimanki, Water logged areas of District Faisalabad and District Bahawalnagar. These wetlands had complex terrestrial and aquatic habitat of common teal (*Anas crecca)* as well as for some other migratory birds.

2.2 Gastrointestinal parasites

Immediately after the arrival of a sample in the laboratory two cm was cut from each end of intestine with a sterile scalpel to eliminate contamination during processing. The fecal material was then forced into a sterile 50 ml centrifuge tube containing 10 ml of 2.5 % wt/vol $K_2Cr_2O_7$ to maintain the cysts during storage. Fecal samples were then kept at 4° C less than 4 weeks before processing [5]. Fecal specimens were tested for *Giardia* spp. by examining trichrome-stained direct smear of fecal pellets[6]. Slides were screened at × 400 magnification and cysts of species were confirmed at 1000 × magnification. Internal characteristics that were used to identify the cysts were included to two to four bodies, and aloneness. The modified zeinlnelson staining technique was used to identify *Cryptosporidium,* oocysts [7]. Fecal samples were analyzed by both fecal smear and concentration methods (centrifugal floatation technique) at the Parasitological Laboratory, Punjab Wildlife Research Centre; Faisalabad (Pakistan). Fecal smears were prepared on a microscope slide air dried and fixed with the methanol for 5 minutes. Fixed smears were stained with dilute carbol (1:10) for 3 to 5 min and washed with tap water. Smears were decolorized using acid alcohol, and then counterstained with 0.5 % Malachite Green solution for 1 min. Smears slides were dried in air and examined under the microscope at 400 × magnification. Fecal samples found negative with direct smear method was examined either by centrifugal floatation method [8].For centrifugal floatation method, samples were mixed with 30-50 ml of water. The solution was sedimented for 10-15 minutes until the supernatant was clear. The sediment was mixed with the saturated saline solution in a centrifuge tube and centrifuged at 1500 revolutions per minute (rmp) for one or two minutes. The floating oocysts were removed by touching with cover glass and transferred to clean glass slide and examined under microscope (400X). The parasites were identified up to the lowest possible taxonomic level based upon oocyst and egg morphology[9].

2.3 Statistical Analysis

On completion of sample collection and laboratory analysis, data was tabulated and statistically analyzed by using relevant statistical techniques. T-test and chi-square test for fecal samples were applied, using SPSS-22 statistical softer package.

III. Results

3.1 Gastrointestinal parasites

During the present study total 40 samples (20♂, 20♀) were collected from different six wetlands of Punjab Pakistan. Among these samples total 17 samples (08% ♂, 09% ♀) were found infected with different gastro- intestinal parasites. The parasitic load was almost equally shared by the bird sampled from different sites (Chashma Barrage 37.50%; Taunsa Barrage and Head Marala 40% both; waterlogged area of Bahawalnagar 44.4% and Head Sulaimanki and waterlogged areas of Faisalabad 50% both (Tab-3.1; Fig-3.1). The frequency of distribution in two sexes vary at different sampling sites, parasitic load was almost equally shared by both sexes (40% ♂, 45% ♀), and difference of both sexes were statistically non-significant (P>0.05).

Table-3.1 Gut parasite load (%) in common teal sampled from deferent wetlands of Punjab, Pakistan. Values in parenthesis indicate frequencies.

Wetland	Male n=20	Female n=20	Total n=40	Chi-square value	P-value
Chashma	66.67(2/3)	20.00 (1/5)	37.50 (3/8)	1.742^{NS}	0.187
Taunsa	20.00 (1/5)	60.00 (3/5)	40.00 (4/10)	1.667^{NS}	0.197
Marala	33.33 (1/3)	50.00 (1/2)	40.00 (2/5)	0.139^{NS}	0.709
Sulemanki	50.00 (1/2)	50.00 (1/2)	50.00 (2/4)	-	-
Bahawalnagar	40.00 (2/5)	50.00 (2/4)	44.44 (4/9)	0.090^{NS}	0.764
Faisalabad	50.00 (1/2)	50.00 (1/2)	50.00 (2/4)	-	-
Total	40.00(8/20)	45.00(9/20)	42.50(17/40)	0.102^{NS}	0.749

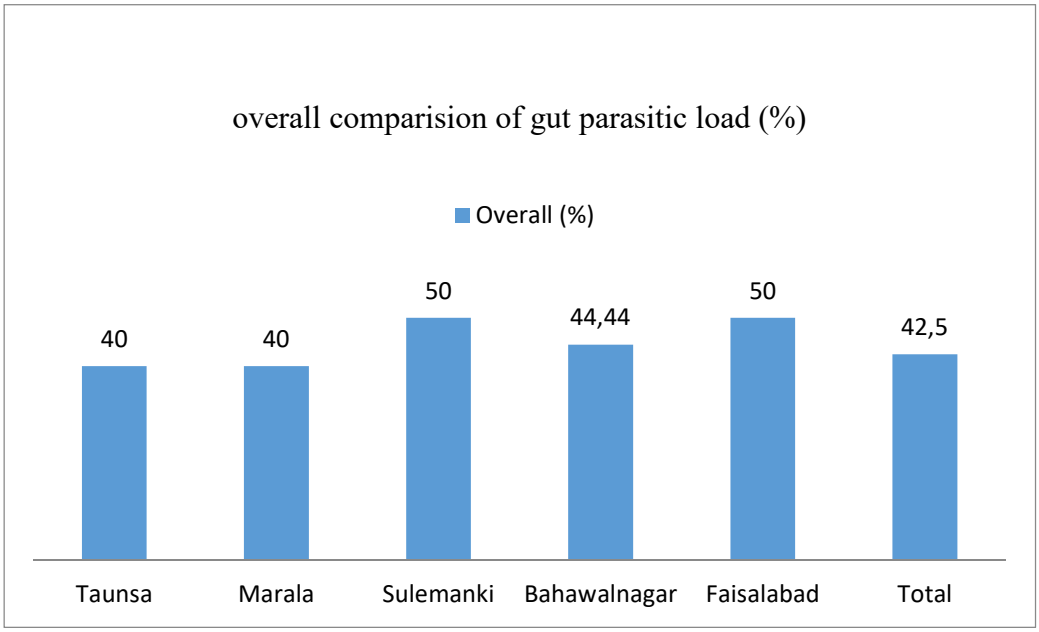

Fig-3.1 The overall comparison of gut parasitic load (%) in common teal samples collected from wetlands of Punjab

The total seven species of Gastro-intestinal parasites were identified which belongs to four classes/phylum (Nematode, Trematodes, Cestodes and protozoan) and five families (*Dioctophymidae, Echinostomatidae, Hymenolepidiae, Hexamitidae* and *Cryptosporidiidae*), in which Nematodes 15%, Trematodes 32%, Cestodes 23% and Protozones 30 % (Fig-3.2). Among the total five families, two families (*Echinostomatidae and Hymenolepidiae*) were most prevalent (28.57 %) and other three families (*Dioctophymidae, Cryptosporidiidaeand Hexamitidae*) were equally prevalent (14.28%) among the birds. The total seven species of parasites which were recorded as *Hystrichis tricolor*, *Echinostoma revolutum, Hypoderaeum conoideum, Hymenolepis longicirrosa, Diorchis stefanski, Giardia sp.* and *Cryptosporidium sp*.

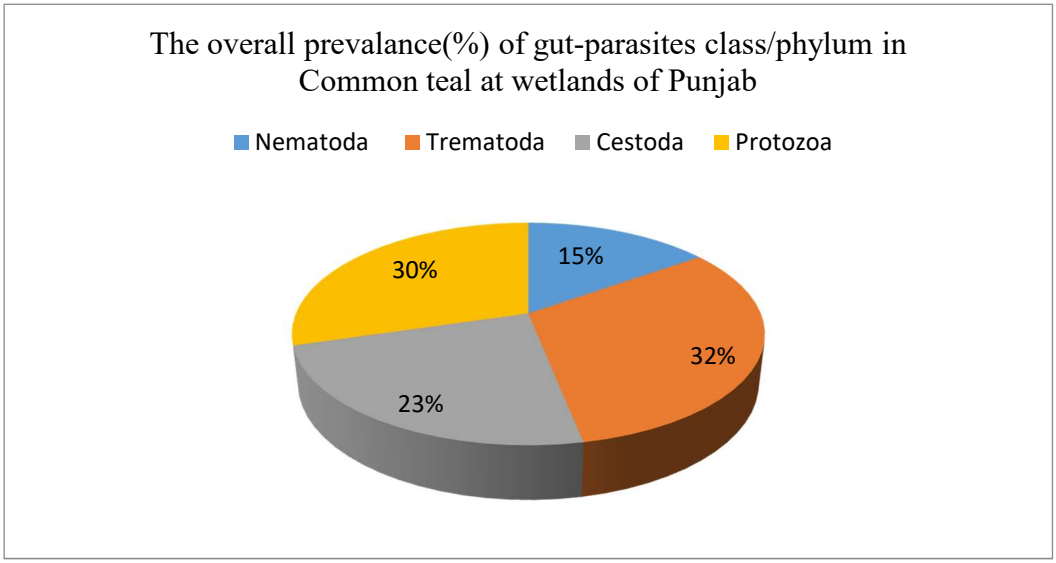

Fig-3.2 The overall prevalence (%) of gut-parasites class/phylum in Common teal at wetlands of Punjab

The overall prevalence of these endoparasites was 17.58% *H. tricolor*, 14.81% *E. revolutum*, 22.58% *H.conoideum* , 16.12 % *H. longicirrosa*, 11. 62% *D. stefanski*. The overall mixed infection was 37.5% among all the infected Birds (Table- 3.2; Fig-3.3). Five species were recorded at Taunsa Barrage (*Hystrichis tricolor*, *Echinostoma revolutum, Hypoderaeum conoideum, Hymenolepis longicirrosa, Cryptosporidium sp.)* and at Water-logged areas of Bahawalnagar (*Hystrichis tricolor*, *Hypoderaeumconoideum*, *Hymenolepislongicirrosa,*,*Giardia sp*., *Cryptosporidium sp.*). Four species were recorded at Chashma Barrage (*Echinostomarevolutum, Hypoderaeumconoideumm, Hymenolepislongicirrosa, Giardia sp*) and water-logged areas of Faisalabad (*Echinostomarevolutum, Hymenolepislongicirrosa, Diorchisstefanski,Giardiasp*) and three species were recorded at Marala(*Hystrichis tricolor*, *Echinostomarevolutum,Diorchisstefanski*) and at Head Sulaimanki (*Hystrichis tricolor*, *Hypoderaeumconoideum, Diorchisstefanski*).

Table- 3.2 Prevalence (%) of different species of gut endoparasites common teal sampled from the wetlands of Punjab. Values in parenthesis present frequencies.

			Wetlands						Overall
Family	**Order**	**Species**	**Chashma**	**Taunsa**	**marala**	**Sulaimanki**	**Bahawalnagar**	**Faisalabad**	
Nematode									
Dioctophymidae	Rhabditida	*Hystrichis tricolor*	-------	10(1/10)	50 (1/5)	50(2/4)	11.11 (1/9)	-----	17.85 (5/28)
Trematodes									
Echinostomatidae	Plagiorchiida	*Echinostoma revolutum*	12.5 (1/8)	10(1/10)	20 (1/5)	--------	------	25 (1/4)	14.81 (4/27)
		Hypoderaeum conoideum	25 (2/8)	20 (2/10)	--------	25 (1/4)	22.22(2/9)	------	22.58 (7/31)
Cestodes									
Hymenolepidiae	Cyclophyllidea	*Hymenolepis longicirrosa*	12,5 (1/8)	20 (2/10)	------	-------	11.11 (1/9)	25 (1/4)	16.129(5/3)
		Diorchis stefanski	------	-------	40 (2/5)	25 (1/4)	-------	50(2.4)	11.62 (5/13)
Protozoans									
Hexamitidae	Diplomonadida	Giardia sp.	12.5(1/8)	----------	---------	----------	22.22(2/9)	25(1/4)	19.04 (4/21)
Cryptosporidiidae	Eucocciorida	Cryptosporidium sp.	---------	10(1/10)	-------	-------	22.22(2/9)	----------	15.78(3/19)
Mixed infection			25(2/8)	30(3/10)	40(2/5)	50(2/4)	44.44(4/9)	50(2/4)	37.5 (15/40)

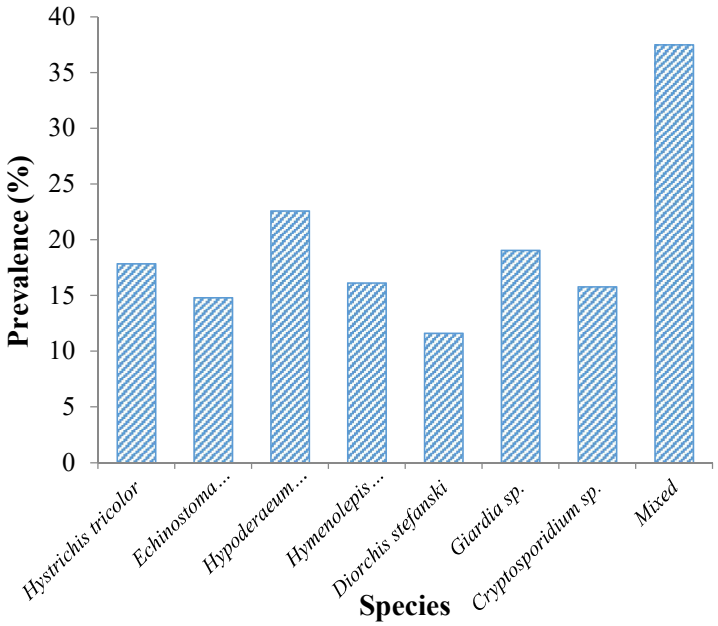

Fig-3.3 The prevalence (%) of different species of gut endoparasites common teal sampled from wetlands of Punjab.

Tricolour (Nematodes) was recorded among the three wetlands (Taunsa Barrage, Marala and waterlogged area of Bahawalnagar) and was most prevalent at Marala (50%), which leads Bahawalnagar (11.11 %) and least at Taunsa Barrage (10.8%). The parasitic species *E. revolutum*(Trematodes) was recorded among the four wetlands (Chashma, Taunsa, Marala and water-logged areas of Faisalabad) and was most prevalent at Chashma Barrage and Head Sulaimanki (25%) which leads to Water-logged areas of Bahawalnagar (22.22 %) and least present at Taunsa Barrage (20%). *E. conoideum*(Trematodes) was recorded among the four wetlands (Chashma, Taunsa, Head Sulaimanki and water-logged areas of Faisalabad) and was most prevalent at Water-logged areas of Faisalabad (25%), which leads to the Marala (20%), Chashma Barrage (12.5%) and least present at Taunsa Barrage (10%). *H. longicirrosa*(Cestodes)was recorded among the four wetlands (Chashma, Taunsa, water-logged areas of Faisalabad and Bahawalnagar) and was most prevalent at water-logged area of Faisalabad (25%) which leads Taunsa Barrage (20%), Chashma Barrage (12.5%) and least present at Bahawalnagar (11.11 %). *D. stefanski*(Cestodes) *was* recorded among the three wetlands (Head Marala, Sulaimanki and water-logged areas of Faisalabad) and was most prevalent at water-logged areas of Faisalabad (50%), which leads to Marala (40%) and least present at Head Sulaimanki (25%). *Giardia sp*. (protozoan) was recorded among the three wetlands (Chashma barrage, Water-logged areas of Bahawalnagar and Faisalabad) and was most abundant at Water-logged areas of Faisalabad (25 %) which leads to Water-logged areas of Bahawalnagar (22.22 %) and least prevalence at Chashma Barrage (12.5%). *Cryptosporidium sp*.(protozoan) was recorded among the two wetlands (Taunsa barrage, Water-logged areas of Bahawalnagar) and was most abundance at Water-logged areas of Bahawalpur (22.22 m%) which leads to Taunsa Barrage (10%). The overall mixed infection was 37.5% among of the infected Birds. The mixed infection was most prevalence at Head Sulaimanki and Water-logged areas of Faisalabad (50%), which leads Bahawalnagar (44.44%), Head Marala (40%), Taunsa Barrage (30%) and least present at Chashma Barrage (25%) (Table.-3.2;Fig-3.3, Fig-3.4).

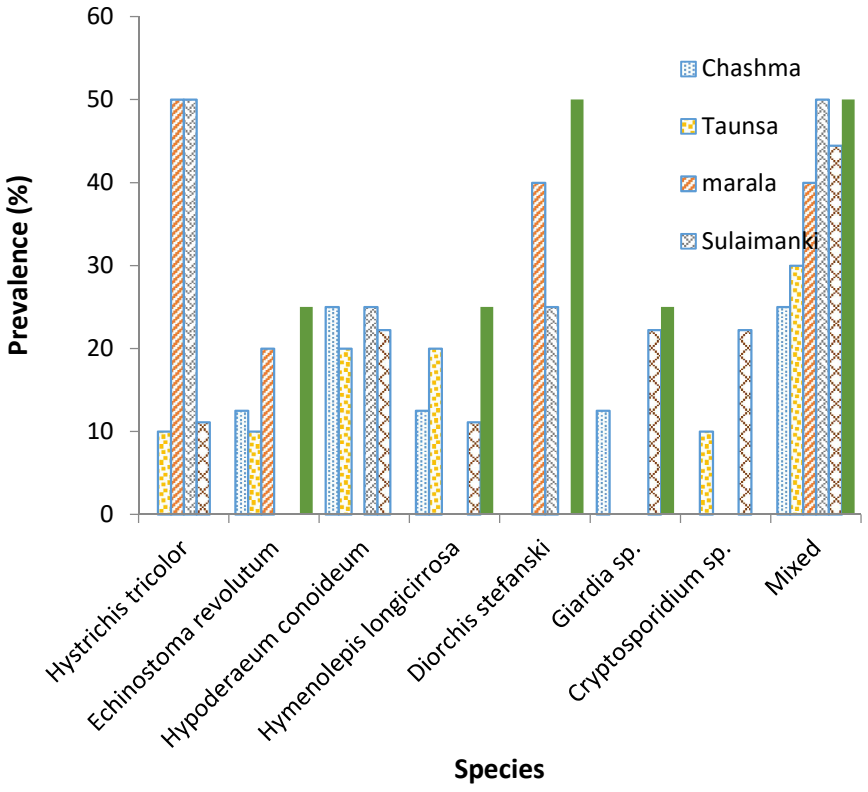

Figure-3.4 The comparison of different endo-parasite species in common tea (Anas crecca) at different wetlands of Punjab

IV. Discussion

The total 17 samples (08 ♂, 09 ♀) were found infected with different gastro- intestinal parasites. The total seven species of Gastro-intestinal parasites were identified which belongs five families were recorded as *Hystrichis tricolor*, *Echinostomarevolutum, Hypoderaeumconoideum, Hymenolepislongicirrosa, Diorchisstefanski, Giardia sp.* and *Cryptosporidium sp*.

Waterfowl can act as a main source of different types of parasites; they can pick up infection from their habitat, carry and spread them in the environment, including drinking water supplies and domestic animals [10]. Previous re-searchers have suggested a positive relationship between migration and parasite richness could stem from a weakening of the immune system during migration [11] a greater aggregation of hosts [12] or exposure to a wider range of habitats and parasite types.

E. revolutum produced haemorrhagic enteritis and parasites were firmly attached with the mucosa. This finding confirmed the observations of Forester et al. (1994) who observed haemorrhagic enteritis and thickened intestinal mucosa. They also detected extensive destruction and degeneration of the villi caused by the attachment and pressure of the flukes. Bhowmik et al. (1987) observed severe enteritis in turkeys due to *E. recurvatum*. Both the *E. revolutum* and *E. recurvatum* have spiny head collar and scaly cuticle.They remain firmly attached to the site of attachment and give irritation. Parasites were firmly attached with the mucosa of small intestine. Cestode fauna in avian hosts is directly related to the type of food ingested [13].The prevalence of helminths is often greater in adult female waterfowl and in first-year birds because of a greater amount of invertebrates in their diet [14]. Moreover, it is estimated that 80 to 96% of surface waters in the United States are contaminated with *Cryptosporidium* and *Giardia* [15].The contributions of *Giardia* cysts and *Cryptosporidium* oocysts from avian species to the concentrations of cysts and oocysts in water samples are largely unknown, as are the extent of transmission of bird-vectored organisms to mammalian hosts and the importance of these parasites in avian species [16].Migration of various species of waterfowl toward the northern parts of Iran has important impact on parasitic diseases in birds and human in Northern part of Iran. *Giardia* spp. commonly was reported from various birds including budgerigars (*Melopsittacusundulatus*), cockatiels (*Nymphicushollandicus*), love birds (*Agapornis* spp.), grey-cheeked parakeets (*Brotogerispyrrhopterus*) and other psittacines [17].*Giardia*cysts are commonly found in sewage and surface waters and occasionally in drinking water. In Canada, a cross sectional survey in 72 municipalities performed between 1991 and 1995, Wallis et al. (1996) found that 72.6%, 21% and 18.2% of raw sewage, raw water, and treated water samples, respectively, contained *Giardia* cysts. In a similar study, fecal samples were taken from wild ducks on the lower Rio Grande River around Las Cruces, from 2000 to 2001. The results of this study indicated that 49% of the ducks were carriers of *Cryptosporidium*, also 28% of the ducks were positive for *Giardia*[18].In our study, prevalence of giardiosis is very close to the result of Kuhn et al.'s (2002). Contaminated drinking water, recreational water, and food are very important and are a major pathway for transmission. *Cryptosporidium* oocysts are commonly found in sewage and surface waters and occasionally in treated water [19]. To date, only two valid species of *Cryptosporidium* (*Cryptosporidium baileyi*and *Cryptosporidium meleagridis*) have been proven to cause infections in birds and *Cryptosporidium* has been detected in more than 30 species of birds including geese (*Anseranser*), tundra swans (*Cygnus* sp.), black-headed gulls (Larusridibundus), chickens (Gallus gallus)[20], turkeys (Meleagris gallopavo), mallards (*Anas platyrhynchos*), and (*Cairinamoschata*) Muscovy ducks.

V. Conclusion

The further studies may be conducted an account for various factors that could influence the ecological health of wet lands and habitat quality for migratory birds, nutrients from catchment area, initiation of vegetation control program, regular wildlife research and monitoring programs and formulation of species recovery plan for threaten species. The further studies should be taken to screen birds with molecular tools to better understand the biological and medical patterns of host-parasite in these areas.

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