

GASTROINTESTINAL PARASITES OF FERAL PIGEON (*Columba livia* GMELIN, 1789) AT TWO TEMPLES OF KATHMANDU VALLEY



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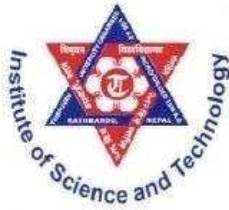
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DECLARATION

I hereby declare that the work presented in this thesis has been done by myself, and has not been submitted elsewhere for the award of any degree. All sources of information have been specifically acknowledged by reference to the authors or institutions.

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RECOMMENDATION AND LETTER OF APPROVAL

This is recommended that the thesis entitled “GASTROINTESTINAL PARASITES OF FERAL PIGEON (*Columba livia* GMELIN, 1789) AT TWO TEMPLES OF KATHMANDU VALLEY” has been carried out by Bina Jha for the partial fulfillment of Master’s Degree of Science in Zoology with special paper Parasitology. This is her original work and has been carried out under my supervision. To the best of my knowledge, this thesis work has not been submitted for any other degree in any institutions.

Subsequently, the aforementioned thesis is approved for the examination and submitted to the Tribhuvan University in the partial fulfillment of Master’s Degree of Science in Zoology with special paper Parasitology.

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CERTIFICATE OF ACCEPTANCE

This thesis work submitted by Bina Jha entitled “GASTROINTESTINAL PARASITES OF FERAL PIGEON (*Columba livia* GMELIN, 1789) AT TWO TEMPLES OF KATHMANDU VALLEY” has been accepted as a partial fulfillment of Master’s Degree of Science in Zoology with special paper Parasitology.

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LIST OF ABBREVIATIONS

| Abbreviated form | Details of abbreviations |
|------------------|---|
| ADPCD | - Animal Disease Protection and Control Division |
| BCN | - Bird Conservation Nepal |
| CI | - Confidence Interval |
| DNPWC | - Department of National Parks and Wildlife Conservation |
| GI | - Gastro-Intestinal |
| IUCN | - International Union for Conservation of Nature |
| LC | - Least Concerned |
| NaCl | - Sodium Chloride |
| PCRC | - Pigeon Control and Resource Centre |
| RH | - Relative Humidity |
| UNESCO | - United Nations Educational Scientific and Cultural Organization |
| Viz: | - Namely |

ABSTRACT

Pigeons (Order Columbiformes) are found worldwide and are known to be originated from Europe, North Africa and Asia. Feral pigeons are found almost everywhere except the Sahara desert, Antarctica and high Arctic. The present study was conducted to evaluate the general prevalence, specific prevalence, compare area-wise prevalence, infection-wise prevalence and identification of gastrointestinal parasites of feral pigeons in two temples viz. Pashupatinath temple and Krishna temple of Kathmandu valley, Nepal. Total 120 fecal samples were collected by opportunistic random sampling method on 22nd and 23rd April, 2016 A.D. to determine the gastrointestinal parasites of feral temple pigeons. The qualitative examination of fecal samples was done by direct microscopic examination, flotation technique and sedimentation technique whereas Microsoft Excel 2007 and “R” version 3.3.1 software package was used for data analysis. Out of total 120 fecal samples examined, 109 fecal samples were positive with overall prevalence of 90.83%. Total of six GI parasites were identified that includes one genera of protozoa: *Eimeria* sp. 52 (43.34%) and five genera of helminthes: *Capillaria* sp. 62 (51.67%) followed by *Ascaridia* sp. 33 (27.50%), *Heterakis* sp. 23 (19.17%), *Syngamus* sp. five (4.17%) and *Tetrameres* sp. two (1.70%). The prevalence rate of helminthes 100 (83.34%) was higher than prevalence rate of protozoan parasites 52 (43.34%). Statistically, the difference in prevalence of helminthes and protozoan parasites were found to be significant ($\chi^2=15.14$, $P<0.05$). The infection rate in two study areas, Pashupatinath temple and Krishna temple was found 57 (95%) and 52 (86.67%) respectively. Statistically, the difference in prevalence of GI parasitic infection among study area was found to be insignificant ($\chi^2=0.24$, $P>0.05$). Mixed infection was more common 61 (50.84%) than single infection 48 (40%). Statistically, the differences in the prevalence of single and mixed infections were found to be insignificant ($\chi^2 =1.56$, $P>0.05$). Among multiple infections, double infection showed the highest rate 53 (86.88%) than the multiple infection 8 (13.12%) in this study. The current study revealed heavy infection in feral pigeons at two temples of Kathmandu valley. The study indicated that feral pigeons are highly susceptible to GI parasites. Therefore, sustainable action should be designed and implemented to control the parasitic infection and reduce the health hazards of feral temple pigeons.

1. INTRODUCTION

1.1 Background

Pigeons (Order Columbiformes) are worldwide free living species which are found since ancient time (BC. 3000-5000) and are known to be originated from Europe, North Africa and Asia (Sari *et al.*, 2008). Feral pigeons are found almost everywhere except the Sahara desert, Antarctica and high Arctic. They have adapted to life in urban, suburban and rural environment and have close communication with humans and are most widely distributed among hoppy in the world, in some countries pigeon are used for human food, hobby, experimental purpose, cultural and religious symbol as well as ornamental purposes, also feral pigeons are used as a bio-indicator of chemical pollution (klein *et al.*, 2008, Nam *et al.*, 2004). *Columba livia* are descended from wild rock Pigeons that live in Mediterranean Europe (Adang, 1999). Common names of *Columba livia* are pigeon, dove, blue rock pigeon, rock dove, wild rock pigeon, rock pigeon, feral pigeon. About 100 different breeds and varieties are described in more or less detail by Levi in his comprehensive reference work on the Pigeon (Levi, 1969). In Nepal, six species of *Columba* are recorded: (*Columba livia* Gmelin, 1789), (*Columba rupestris* Pallas, 1811), (*Columba leuconata* Vigors, 1831), (*Columba palumbus* Linnaeus, 1758), (*Columba hodgsonii* Vigors, 1832) and (*Columba pulchricollis* Blyth, 1846) (BCN and DNPWC, 2016).

1.2 Characteristics of pigeon

Adult of the nominate species of the feral pigeon weight about 369 gm, 32-37 cm long and have 64-72 cm wingspan. Pigeons have dark bluish-grey head, neck and chest with glossy greenish and reddish-purple iridescence around the neck and wing feathers, Orange or red iris with pale inner ring in adult or brown or grayish brown in juveniles. These pigeons consist of black bill with off-white cere, red feet and legs, distinctive twin black wing bars and white lower back feathers (PCRC, 2009).

1.3 Population Status

The global population is estimated to number c.260,000,000 individuals (BirdLife International 2016). This species has an extremely large range, and hence does not approach the thresholds for Vulnerable under the range size criterion (Extent of Occurrence <20,000 km² combined with a declining or fluctuating range size, habitat extent, or population size and a small number of locations or severe fragmentation). Despite the fact that the population trend appears to be decreasing, the decline is not believed to be sufficiently rapid to approach the thresholds for Vulnerable under the population trend criterion. The population size is extremely large, and hence does not approach the thresholds for Vulnerable under the population size criterion (<10,000 mature individuals with a continuing decline estimated to be >10% in ten years or three generations, or with a specified population structure). For these reasons the species is evaluated as LC. The population size is suspected to be decreasing owing to interbreeding with domestic form; declines have been recorded in Israel (Baptista *et al.* 1997).

1.4 Ecology and behavior

Pigeons breed all year round with peak breeding periods in spring and summer (Williams and Corrigan, 1994). They breed well for at least five to six years and will continue to reproduce, but less regularly into an old age of 10 or more years. All columbiformes are monogamous. Wild birds breed on coastal cliffs and some inland cliffs. Feral birds breed on or in buildings, usually in urban areas. Flimsy nest are built on rocky shelf or accessible ledge on a building or in the roof void of a building. Female lays one or two white eggs that are incubated by both parents for 17-19 days. The squab has yellow down and a pink bill. Squabs are fed on 'crop milk' by both parents. Fledging period is approximately 30 days depending on time of year. The young leave the nest at four to six weeks of age. More eggs are laid before the first clutch leaves the nest. Young pigeons reach sexual maturity by six to seven months (Hollander, 1954; Levi, 1969; Sturtevant and Hollander, 1978; Kendall and Scanlon, 1981). A population of pigeons usually consists of equal numbers of males and females. Life expectancy of pigeon varies greatly from 3-5 years through to 15 years dependent on many factors, including natural predation and human interference. In captivity, pigeons commonly live up to 15 years and sometimes longer (Williams and Corrigan, 1994).

Seeds and grains are the major component of diet for pigeons. Some ground feeding species (granivorous species) eat fruit and take insects and worms. One species, the Atoll Fruit Dove, has adapted to taking insects and small reptiles. The feral pigeon found in urban areas exists exclusively on a diet of seed (normally from human sources) and human refuse, such as fast food waste provided for them intentionally or unintentionally by people (Williams and Corrigan, 1994). Wood pigeons have a varied diet which includes vegetables and berries. Food consumption is about one-tenth of the pigeon's body weight and will range from 20-100 gm daily, depending on the strain (Sturtevant and Hollander, 1978). Pigeon consumes 36-60 ml of water daily (Clarkson *et al.*, 1963).

Pigeons can fly at altitudes of 6000 feet or more at average speeds of up to 77.6 mph but have been recorded flying at 92.5 mph. Pigeons can fly between 600 and 700 miles in a single day, with the longest recorded flight in the 19th century taking 55 days between Africa and England and covering 7000 miles. Pigeons are thought to navigate by sensing the earth's magnetic field and using the sun for direction. Other theories include the use of roads and even low frequency seismic waves to find their way home. Pigeons (and all the columbidae family) drink by sucking water and using their beaks like straws. Most birds sip water and then throw their head back to swallow (PCRC, 2009).

Pigeons may be raised under wide seasonal ranges of temperature, humidity, light, and barometric pressure, if kept in flypens. For cage housed birds, temperature should be held between 10°-24°C (50°-57°F) with RH of 30%. A 12-hour light, 12 hour dark diurnal cycle is commonly provided, although 14 hours light will enhance breeding activity (Sturtevant and Hollander, 1978). Pigeons lay successive clutches of two eggs at five

week intervals and raise 10-22 young per annum where 15 to 16 is considered a good commercial production (Levi, 1969; Kendall and Scanlon, 1981; Hollander, 1954; Sturtevant and Hollander, 1978).

1.5 Pigeon, human and parasites in nature

Pigeons have widely colonized most of the world including five continents and extremely isolated islands, facilitated by their high dispersal ability (Pereira *et al.* 2007), most important difference between urban and less modified or wild ecosystems is the major role humans play. Beyond being the dominant urban species, people influence species composition and abundance by killing pests, and feeding and protecting popular species, particularly birds (Tangley, 1986). Indeed, studies have indicated that up to 57% of people may feed wildlife (Rollinson *et al.*, 2003), which can influence population size. Feral pigeons have a long history of human-wildlife conflict, which includes unwanted noise to urban residents (Jerolmack, 2008), the damage feral pigeons cause to buildings and other surfaces from nesting and feces (Sacchi *et al.*, 2002), and the potential of feral pigeons to spread human diseases (Haag-Wackernagel and Moch, 2004). An individual pigeon produces around 12 kg of excrement per year that fouls breeding areas, building faces, monuments, pavements and other public areas (Haag-Wackernagel and Geigenfeind, 2008). Feral pigeons harbor at least 110 different human pathogens, but there has been limited evidence of actual human infection with only 230 recorded human infections worldwide (Haag-Wackernagel and Moch, 2004). For example, pigeons can carry or transmit encephalitis, histoplasmosis, newcastle disease, pigeon ornithosis, cryptococcosis, pigeon coccidiosis, toxoplasmosis, pseudo-tuberculosis and salmonella food poisoning (Rehman, 1993; Opara *et al.*, 2012). And also, pigeons can carry fleas, ticks, mites and other parasites (Balicka-Ramisz *et al.*, 2007; Rehman, 1993). While others view pigeons as complementary to city life and enjoy feeding and interacting pigeons (Johnston and Janiga, 1995). Those who enjoy pigeons in cities are likely to hold strong opinions against control leading to human-wildlife conflict.

1.6 Parasitic infections of feral pigeons

There are well documented data on parasites occurrence of wild and domestic pigeons from different part of the world that indicate wide range of helminthes, protozoa and arthropods infection (Olsen and Braun, 1980; Begum and Sheikh, 1987; Bernard and Blesemans, 1987). The gastro-intestinal tracts of pigeons harbor a wide variety of helminthes, of which nematodes are the most deleterious parasites and are responsible for clinical and sub-clinical parasitism. Earlier study on feral pigeons shows that large roundworms *Ascaridia* sp., cecal worms *Heterakis gallinarum*, hair worms *Capillaria obsignata*, crop worms *Capillaria contorta* and *C. annulata*, tape worms *Raillietina cesticillus*, gapeworms *Syngamus trachea* and coccidians such as *Eimeria* sp. *Isospora* sp. are recorded from their fecal examinations (Bahrami *et al.*, 2013). *Columba livia* is associated with human habitation, often occupying and soiling premises where people work and live (Adang, 1999). Pigeons may be infected with many parasites, some of which are pathogenic to humans (Zwart, 1986). Free-range scavenging

birds are in direct contact with parasite vectors, soil and feces. On the other hand, lack of hygiene, direct contact with humans, and the physical environment (rainfall, humidity and ambient temperature) provides optimum conditions to maintain parasite populations. The probability of disease transmission is influenced by many factors, such as time of infection, latent period, stability of the agent when exposed to the environment, population density, animal handling, virulence and route of infection (Alves *et al.*, 2008). Intermediate hosts of parasites easily infect the *Columba livia* via their diet. Parasites are the etiological agents of most diseases of cage and aviary birds. The high parasitic infection means an indication of poor management and control efforts in either the animal or in the immediate environment where infection or re-infection (directly or indirectly) may emanate. Both sexes of pigeons are equally at risk of being infected by the parasites and as carriers of pathogenic organisms, some of which might be zoonotic (Opara *et al.*, 2012). The effects of parasitism on birds are often severe, including retarded growth, low egg production and susceptibility to other infections (Dranzoa *et al.*, 1999). Parasites often have short life-cycles. This gives them a capacity to change in genetic composition between generations and hosts need ability to respond these changes (Lindstrom, 2000). Feral pigeons have been identified with mycotic, bacterial, protozoal, chlamydial, rickettsial and parasitic diseases as well as dermatosis have been identified from the transmission of pigeons to humans (Weber, 1979).

1.7 Research questions

- ❖ Are the feral pigeons (*Columba livia*) at two different temples of Kathmandu valley infected with gastrointestinal parasites?
- ❖ If yes, what is the prevalence of gastrointestinal parasites of feral pigeons (*Columba livia*) at two different temples of Kathmandu valley?
- ❖ What is the prevalence of helminth and protozoan parasites within gastrointestinal parasites?
- ❖ What is the prevalence of single and mixed infections of different gastrointestinal parasites?

1.8 Objectives

1.8.1 General objective

- ❖ To identify the gastrointestinal parasites found in feral pigeons (*Columba livia* Gmelin, 1789) at two different temples of Kathmandu valley.

1.8.2 Specific objectives

- ❖ To determine the general prevalence of gastrointestinal parasites of feral pigeons (*Columba livia*) at two different temples of Kathmandu valley.
- ❖ To determine the prevalence of helminth and protozoan parasites.
- ❖ To determine the prevalence of single and mixed infections of different gastrointestinal parasites.

1.9 Justification of the study

Feral pigeons are not harmless birds (Weber 1979). Many potential infections of human silently exist in pigeons which are not apparent. They have the potential for transmission of over 30 diseases to humans plus another ten to domestic animals (Weber 1979). Information about pigeon borne diseases and zoonotic parasites is limited and insufficient to the public and pigeon breeders. In global context there is much information written about the problem, but it remains primarily in the professional journals and technical references, neatly stacked away on library shelves. The objective of this paper is to provide a brief account of the major parasites prevalent in feral pigeons. Few studies have been carried regarding the parasitic infections of pigeon in Nepal, if compared to other nations. Fecal examinations were conducted to identify the prevalence of intestinal parasites (helminthes and protozoans) of *Columba livia* at two temples of Kathmandu valley as well as single and multiple infections of parasites. This study will reveal the prevalence of GI parasites in feral pigeons at two temples of Kathmandu valley as well as update the previous findings on GI parasites of pigeons. This study will also create opportunity for the further study.

2. LITERATURE REVIEW

The presence of parasites in birds and other animals is the rule, rather than the exception (Cole *et al.*, 1999). Hundreds of parasite species have been identified from free ranging wild birds; however, the presence of parasites does not necessarily equate with disease. Most of the parasites identified from wild birds cause no clinical disease. Others cause varying levels of disease, including death in the most severe cases. The pathogenicity or the ability to cause disease, of different species of parasites varies with the species of host invaded (infected or infested), the number or burden of parasites in or on the host, and internal factors impacting host response. For example, when birds are in poor nutritional condition, have concurrent infections from other disease agents (including other species of parasites), or are subject to other types of stress, some parasites that do not normally cause disease do cause disease. Lethal infections may result from parasites that generally only cause mild disease (Cole *et al.*, 1999). Pigeons may be infected with many organisms some of which are pathogenic to humans (Mushi *et al.*, 2000). Especially wild pigeons could spread the zoonoses and parasites to people or other birds because they can fly long distances (Piasecki, 2006). They are often the cause of discomfort due to the accumulation of their droppings. Considering this, lots of research work has been carried out on endoparasites of domestic as well as wild and feral pigeons. In case of feral temple pigeons, a very little research work has been carried out regarding parasitic infection. Feral temple pigeons can be infected by different parasites including protozoan, trematodes, cestodes and nematodes. Here, some of the important published work related with the present work has been reviewed.

2.1 In global context

The cestode species present in feral pigeons in Thessaloniki; Northern Greece were recorded (Diakou, 2013). A total of 136 adult feral pigeons were necropsied and the cestodes recovered were preserved for identification using morphological keys. 96 (70.58%) of them were found to be infected with cestode parasites; which were identified to be *Raillietina* spp.: 84 (61.76%) were infected with *R. echinobothrida*; 20 (14.7%) with *R. cesticillus* and 8 (5.88 %) with *R. tetragona*. Single infection was recorded in 80 birds (83.3% of the infected population); while a significantly lower number; i.e. 16 birds (16.7 %) had a mixed infection with *R. echinobothrida* and *R. cesticillus*.

Parasani and Momin (2010). Qualitative fecal examination of 30 samples of pigeons revealed 27 samples (90%) with parasitic infection. Among them nematode, cestode and coccidian infection was 88.88%, 26.92% and 74.07% respectively in Gujarat State of India.

Routine examination within three to four months after death of 609 band tailed pigeons (*Columba fasciata fasciata* Say, 1822) in Colorado was performed. They revealed that 12.50% harbored helminthes that includes two species of cestodes and four species of

nematodes. *Hymenolepis armata*, *Raillietina* sp., *Ascaridia columbae*, *Splendido filaria colubensis*, *S. hibli* and *Chandlerella robinsoni* were helminthes found in pigeons above nine months of age whereas absent in pigeons younger than nine months of age (Olsen and Braun, 1980). Similar prevalence rate (23.50%) of helminthes that includes cestodes only were found while studying the ecto-, gastrointestinal and hemoparasites of live pigeons (*Columba livia*) in Kampala, Uganda by using direct smear, floatation and sedimentation tests. The identification of cestodes was not possible (Dranzoa *et al.*, 1999).

Adang *et al.* (2008) humanely killed dissected and necropsied 240 *Columba livia domestica* in Zaria area, Nigeria. Among which 48.30% were infected by nine species of helminthes, comprising six species of cestodes and three species of nematodes. The higher prevalence of cestodes (*Raillietina tetragona*) was recorded whereas single infection (37.50%) was more common than double (10%) and triple infections (0.83%).

Adang *et al.* (2009) conveyed speckled pigeons as a probable definitive host of some ectoparasites and helminthes. A total of 30 (20 males and 10 females) speckled pigeons were trapped from the wild in Zaria, Nigeria and GI tract of each bird were tested. The prevalence rate of helminth infections was found to be 56.70% represented by four species of cestodes comprising *Raillietina tetragona*, (3.30%), *Raillietina cesticillus* (26.70%), *Amoebotaenia cuneata* (13.30%) and *Hymenolepis carioca* (13.3%). Single infection was the only infection type observed.

Balicka-Ramisz and Bogumila (2014) examined a total of 330 fecal samples with two methods: Willis-Schlaaf's (qualitative) and McMaster's (quantitative). Three species of protozoa were isolated: *Eimeria labbeana*, *E. columbarum*, *E. columbae* and the infections were mixed. The occurrence of *E. labbeana* was most commonly reported, which was shown, depending on the pigeon loft and the age of the birds, in 89–93% of young pigeons and in 63–55% of adults. The species *E. columbarum* and *E. columbae* were found less frequently.

Msoffe *et al.* (2010) found prevalence rate (79.50%) of GI helminthes in domestic pigeons of Morogoro Municipality, Tanzania by the postmortem examination. The three subfamilies represented two cestodes and one nematode, whereas no trematodes were found. Three species of helminthes *Raillietina tetragona* (6%), *Raillietina echinobothrida* (63%) and *Ascaridia galli* (15.50%) were identified. Nestlings appeared to be less susceptible to GI cestodes but more susceptible to nematodes compared with adults.

GI nematodes were highly prevalent among pigeons (*Columbia livia domestica*) in Albania (Bizhga *et al.*, 2013). The average parasitic burden was 76 eggs per gram feces, with significant variations at the values of 48-120 eggs per gram feces. *Capillaria* spp. was ranked as the second nematode in terms of the distribution among pigeons. The burden for capillarids was 26 eggs per gram feces, with variations of 12-36. The genus found according to the prevalence and parasitic burden included *Ascaridia columbae*,

Capillaria spp. *Ascaridia galli* and *Heterakis gallinarum* in a small number and in restricted values and areas.

Radfar *et al.* (2012) performed same examination method for the first survey to determine the prevalence and intensity of parasites among free-range backyard chicken and domestic pigeon (*Columba livia domestica*) in Sistan region. Out of 46 domestic pigeon, 39 (84.78%) were infected with parasites. They found seven species of parasites including two species of nematodes, two species of cestodes and three species of ectoparasites. The parasites were *Ascaridia colombae* (15.21 %), *Hadjelia truncata* (17.39 %), *Raillietina tetragona* (26.08%), *Raillietina echinobothrida* (28.26%), *Argas reflexus* (13.04%), *Menopen gallinae* (32.60%) and *Columbicola Columba* (41.30%).

Omer *et al.* (2015) detected the external and internal parasites and its association with intestinal pathological changes in wild pigeons at Sumel region-Duhok Governorate. Samples of skin (plumage), intestinal contents and intestinal tissue were taken from 100 adult pigeons from March to August 2012 for detection of parasites and for histopathological examination. Results showed that 6 (6 %) pigeons feather samples were found positive for external parasites (*Columbicola columbae*), 22 (22%) of fecal samples were found positive for internal parasites (*Raillietina tetragona*), whereas no trematode and nematode were found. A total of 19 (19%) infected pigeons were appeared pathological changes in intestine, which were manifested by mild catarrhal inflammation and excessive mucoid masses.

Bahrami *et al.* (2013) indicated that young pigeons could be more susceptible to parasitic infection as compared to above two years old birds but these parasites did not cause any visible deleterious effects in the blood parameters. This could be due to immune response of the pigeons to parasitic infections. Out of 250 samples, 79.20% were positive and 19.19% were carrying multiple infections when examined through direct smear method. The parasites that have been identified in this study consist *Raillietina* sp. (24.24%), *Capillaria* (14.14%), *Tetrameres* (8.08%), *Ascaridia* (4.04%), *Syngamus* (9.09%), *Oocyst* protozoa (7.07%), *Phthiraptera* (8.08%), *Ceratophyllus columbae* (6.06%).

Radfar *et al.* (2011) surveyed parasites by postmortem examination of 102 domestic pigeons (*Columba livia domestica*) in a selected semiarid zone of South Khorasan, Iran. They found 42.15% prevalence of helminthes including two species of nematodes, *Ascaridia colombae* (16.66%) and *Hadjelia truncate* (1.96%), while 3 species of cestodes, *Cotugnia digonopora* (13.79%), *Raillietina magninumida* (18.62%) and *Raillietina achinobothridia* (32.35%).

Borji *et al.* (2012) examined 300 domestic pigeons (*Columba livia*) by same method in Mashhad, Iran region. Among which 21.60% and 15.30% were infected with nematodes and cestodes respectively whereas no trematodes were found. The overall prevalence of helminthes recorded was 36.90%. Nematodes found were *Ascaridia columbae* (20.37%), *Capillaria bursata* (3.70%), *Capillaria caudinflata* (1.85%), *Eulimdana clava* (2.70%), *Gongylonema* spp. (0.90) and cestodes were *Choanotaenia infundibulum* (11.11%) and *Raillietina echinobothrida* (6.48%).

Begum and Sehrin (2012) examined the separated parts of the alimentary canal of the pigeons taken in 0.85% normal saline solution to collect helminthes parasites in pigeon (*Columba livia*). They found all the birds infected by eleven species of helminthes parasites. Among which, four species of trematoda found were *Echinostoma revolutum* (15%), *E. trivolvus* (5%), *Patagifer bilobus* (5%), *Ehinoparyphium recurvatum* (8.33%) whereas six species of cestoda found were *Hymenolepis columbae* (63.33%), *Raillietina echinobothrida* (100%), *R. bonini* (43.33%), *R. cesticillus* (100%), *Cotugnia celebesensis* (68.33%), *C. cuneata* (100%) and one species of nematode found was *Ascaridia columbae* (28.33%). In autumn season highest intensity of infection was found.

Radfar *et al.* (2011) found 43.13% of protozoan parasites by postmortem examination of 102 domestic pigeons (*Columba livia domestica*) in a selected semiarid zone of South Khorasan, Iran. *Eimeria* sp. (40.19%) and *Cryptosporidium* oocyst (2.94%) were identified.

Nagwa *et al.* (2013) used direct microscopic examination and concentration floatation technique. The seasonal dynamic revealed that the highest incidence of *Eimeria* sp. and *Cryptosporidium* in pigeons from Gharbia, governorate was in winter (42% and 3.6%) respectively whereas the lowest rate of protozoa in pigeons was in spring (20.4 and 2.6%) respectively.

Patel *et al.* (2000) found 53.57% positive for parasitic infection, out of 106 fecal samples of pigeon by using sedimentation technique in the laboratory. *Ascaridia* sp. and *Capillaria* sp. were helminth found with 20.75% and 13.20% respectively whereas 17.92% of *Eimeria* sp. was only protozoan found. The samples were collected from a zoo of Gujarat, India.

Sari *et al.* (2008) found similar prevalence rate of parasites with 46.12% when examined 251 fecal sample of pigeons (136 domestic pigeons and 115 wild ones) in Turkey, through the centrifugal floatation and modified acid-fast staining methods. Coccidia oocysts were detected 59.60% in domestic pigeons and 30.40% in wild pigeons. Helminth eggs detected were 23.50% in domestic pigeons and 4.30% in wild pigeons. The helminth species identified were *Capillaria* sp. (19.90%), *Ascaridia columbae* (5.10%) and *Heterakis* sp. (3.70%) in domestic Pigeons whereas *Capillaria* sp. (4.30%) and *Syngamus* sp. (1.70%) in wild pigeons.

Opara *et al.* (2012) examined fecal samples of 150 street pigeons (*Columba livia*) in Owerri, Imo State, Nigeria and also found similar 70 (46.70%) prevalence rate of GI parasites. Four GI parasites were identified with *Trichomonas* sp. giving the highest prevalence rate (42%), followed by *Eimeria* sp. (28%) whereas Coccidians and *Ascaridia* sp. returned the least with each having the prevalence rate of 14%.

Marques *et al.* (2007) found 74.14% prevalence rate of GI parasites in urban areas of Lages, Southern Brazil. Protozoa (100% for *Eimeria* sp.) were detected and nematodes

(*Ascaridia* sp. and *Capillaria* sp.) 32.56% were detected among positive case. The multiple parasite infection was 20.93%.

Ghosh *et al.* (2014) also revealed similar prevalence rate (72%) of GI parasitic infection in Chittagong metropolitan area, Bangladesh using direct smear, floatation and sedimentation techniques. They found six different species of parasites, among which highest prevalence was recorded for *Ascaridia galli* (35%) followed by *Capillaria* sp. (22%), *Heterakis gallinarum* (13%), *Eimeria* sp. (11%), *Raillietina* sp. (6%) and *Syngamus trachea* (3%).

Similarly, same methods were used in Y S R Kadapa district of Andhra Pradesh in India (Sivajothi and Sudhakara, 2015). They found 72.70% of the birds harbored parasites including *Ascaridia colombae* (33.30%), *Eimeria* sp. (31%), *Capillaria colombae* (17.40%) and *Raillietina* (9%). *Eimeria* sp. was found to be higher in squabs than compare with adults.

Al-Barwari and Saeed (2012) found 100% parasitic infection in pigeons of Erbil Iraq when examined blood samples, ectoparasites and alimentary canal. Examination of the alimentary canal of *Columba livia* was done for protozoans and helminthes parasite. They found four protozoan species, *Eimeria labbeana*, *Trichomonas gallinae*, *Haemoproteus columbae* and *Plasmodium* sp. whereas eight cestoda species, four of each of the genera *Cotugnia* and *Raillietina* and four nematoda species, *Ascaridia columbae*, *A. galli*, *Capillaria obsignata* and *Synhimantus spiralis*.

Mushi *et al.* (2000) observed *Ascaridia columbae* (30%) and *Dispharynx spiralis* (10%), *Raillietina* sp. (80%) and coccidian oocysts (40%) by isolating digestive tract of pigeon from Sebele Gaborone, Botswana.

Tanveer *et al.* (2011) studied the prevalence of gastrointestinal nematodes in 143 (80 male and 63 female) domestic pigeons in Lahore, Pakistan. Fecal samples were collected to determine the gastrointestinal nematodes of domestic pigeons through qualitative and quantitative fecal examinations. The overall prevalence of gastrointestinal nematodes was 40.5% (58/143) in domestic pigeons. Likewise, the prevalence of gastrointestinal nematodes in males and females was found 41.3% (33/58) and 39.7% (25/58) respectively. The overall prevalence of *Capillaria obsignata* and *Ascaridia columbae* was found to be 67.2% and 32.8%, respectively. The prevalence of *C. obsignata* and *A. columbae* in males was 72.7% (24/33) and 27.8% (9/33) and in females was 60% (15/25) and 40% (10/25), respectively.

Natala *et al.* (2009) also used similar method for examination of samples and observed *Eimeria* sp. (49.20%), *Haemoproteus columbae* (15.60%), *Leucocytooson* sp. (6.40%) and *Plasmodium relicum* (0.80%) protozoan parasites whereas *Raillietina tetragona* (4.90%), *Raillietina cesticillus* (3%), *Raillietina echinobothrida* (7.60%), *Ascaridia columbae* (1.20%), *Ascaridia galli* (1.20%) and *Capillaria anatis* (0.80%) were the

helminthes seen. The presence of these parasites was considered to pose a danger to achieving the full potentials of the up-coming pigeon barbecue business in Zaria.

Similarly, *Echinostoma revolutum* (25%), *Raillietina echinobothrida* (50%) and *Cotugnia cuneata* (25%) were observed by same method in the Parasitology Laboratory, Department of Zoology, University of Dhaka (Musa *et al.*, 2011).

Ledwon *et al.* (2016) diagnosed only two known cases of fluke invasions in racing pigeons (*Columba livia*) originating from different regions of Poland over 4 years. In both cases, the invasion was characterized by a very high mortality (approximately 70%), and the source of the infestation was snails of the Lymnaeidae family eaten by pigeons. Fluke invasions in pigeons are extremely rare. Using molecular biology techniques, infestation with the fluke *Echinostoma revolutum* was determined in the second case.

Eljadar *et al.* (2012) determined endo-parasites in green mountain region of Libya from free range pigeons. Fecal and blood samples were collected from different pigeons species and evaluated for the presence of gastrointestinal parasites and heamoparasites. Protozoa (90% for *Eimeria* spp. and 1% for *Haemoproteus* spp.) and nematodes (20% for *Capillaria* spp. and 10% for *Heterakis* spp.) were detected in number of the cases, whereas 5% of the fecal samples were infected by multiple parasites. The presence of coccidian oocysts was revealed in the most of fecal samples.

2.2. In context of Nepal

Scanty study has been carried out in context of Nepal. Using dissection method, Meggitt, 1924 recorded *Raillietina torquata* from Kathmandu whereas (Sharma, 1943) recorded *Raillietina kantipura*, *Raillietina nagpurensis* and *Raillietina nripendra* from Kathmandu.

Similarly, *Hymenosphenacanthus macracanthus* was reported from Kathmandu (Sharma, 1943).

Helminth parasites *Capillaria* sp. (31.67%), *Ascaridia* sp. (21.66%), *Echinostoma* sp. (7.50%), *Syngamus* sp. (5.83%), *Hymenolepis* sp. (3.33%), *Heterakis* sp. (2.50%) and (19.16%) of coccidians were observed by fecal examination of temple pigeons at Pokhara Valley (Gurung, 2016).

3. MATERIALS AND METHODS

3.1 Study area

The Kathmandu Valley is a region of 230 sq. miles (600 sq. km.) in the Bagmati zone in central Nepal. The valley consists of three fabulous cities of great historic and cultural interest. These legendary cities go by the names of: Kathmandu, Lalitpur or Patan and Bhaktapur as well as 100 of smaller towns and villages. The valley, roughly oval bowl measuring 24 km east-west and 19 km north-south, is encircled by a range of green terraced hills. It's central lower part stands at 1,425 m (4,675 ft) above sea level. It lies on the geographical coordinates of 27°42' 14" north latitude and 85°18'32" east longitude.

Kathmandu valley is not only surrounded by the natural beauty but also rich from the historical view. Here we can find many historical oriented temples reflecting the ancient periods of Nepal which has become the habitat of pigeons too. Sample collection areas of the present study are:

A. Pashupatinath Temple B. Krishna Temple

A. Pashupatinath Temple

The Pashupatinath temple is a famous, sacred Hindu temple dedicated to Pashupatinath and is located on the banks of the Bagmati river, five km north-east of Kathmandu Valley in the eastern outskirts of Kathmandu, the capital of Nepal. It is at the elevation of 4,324 ft. from the sea level and lies at the geographical coordinates of 27.71076 north latitude and 85.34851 east longitude. Pashupatinath temple's existence dates back to 400 B.C. Pashupatinath temple is the oldest Hindu temple in Kathmandu. It is not known for certain when Pashupatinath Temple was built. The temple was erected anew in the 15th century by Lichhavi King Shupuspa after the previous building was consumed by termites. The UNESCO enlisted Pashupatinath temple as world heritage site in 1979. The Pashupatinath temple is regarded as a very special and popular temple. The forest Mrigasthali, Kailash and Aryaghat around Pashupatinath temple have made the surrounding environment attractive, beautiful and worth seeing it.

Pigeons are one of densely populated birds observed in Pashupatinath temple. Besides, Cows (*Bos taurus*), Dogs (*Canis lupus familiaris*) and Monkeys (*Macaca mulatta*) are other animals majorly seen within the premises of Pashupatinath temple. Large numbers of pigeons are found feeding upon the grains scattered by the visitors and right at the main entrance of temple area. Street vendors occupy the outer periphery of temple area selling garlands, flowers, puja items and devotional offerings. Besides, many sweet shops and hotels are also aligned at the outer margins of Pashupati area. Pigeons are often seen feeding upon the residual litters accumulated by these street vendors, sweet shops and hotels. Likewise, lots of people do worships and ritual deify in the huts made outside the main temple and their offertory remainings also makes a major food for the pigeons out there. Bagmati river is the main source of water for pigeons at Pashupatinath temple.

Krishna temple is situated in Patan Durbar Square. It is elevated at 4327 ft. above the sea level and lies at the geographic coordinates of 27.67360 north latitude and 85.32493 east longitude. Krishna temple is perhaps Kathmandu's finest stone wrought monument. Krishna temple is made in Sikhara architectural style, a style that is commonly found in what is now known as the Indian Subcontinent. It was established by Siddhi Narsimha Malla of Patan in 1637 AD. The Patan Durbar Square today is one of the World Heritage Sites listed by UNESCO. It is a city of arts and artists and craftsmen with undisputable skills. The majority of the population follows Buddhism, but we also find exquisite Hindu temples in addition to the bronze gateways, marvelous statues, guardian deities and beautiful carvings in metal, wood and stone.

Unlike Pashupatinath temple, Krishna temple is standing at intersection of busy and conjusted crossroads of Patan Durbar Square. It lies across the river Bagmati having the nearest distance of 2.8 km. Krishna temple is eight km. far away from Pashupatinath temple. Dozens of street vendors sell grains in plastic tupperwares and bags at the side of temple. People buy and feed these grains to the temple pigeons as daily rituals, hobby and matter of interest. Along with those grains, pigeons also feed on the residue of fast food thrown by the public passing through the crossroads. Pigeons are also often seen scratching the rooftops of local residential houses. Food and water offered by the local people at their balconies also makes a major diet of pigeons. Water is also kept in small bowls by the Buddhist monks near by the temple which the pigeons drink.



3.2 Materials used

3.2.1 Equipments

- | | |
|------------------------|-----------------------|
| I. Electric microscope | II. Ocular micrometer |
| III. Stage micrometer | IV. Volumetric flask |
| V. Centrifuge machine | VI. Centrifuge tubes |
| VII. Gloves | VIII. Beakers |
| IX. Cover slips | X. Slides |
| XI. Cotton | XII. Tea strainer |
| XIII. Glass rod | XIV. Cavity slide |
| XV. Watch glass | XVI. Rack |
| XVII. Dropper | XVIII. Tooth picks |
| XIX. Mask | XX. Glass vials |
| XXI. Spatula | XXII. Camera |

3.2.2 Chemicals

- | | | |
|--------------------------------|---------------------|--------------------|
| I. Potassium dichromate (2.5%) | II. Iodine solution | III. NaCl solution |
| IV. Methylene blue | V. Distilled water | |

3.3 Study design

The present study was designed to assess the gastrointestinal parasitic infection in *Columba livia* at two temples of Kathmandu valley. The study comprises:

- a) Selection of temples with abundant *Columba livia*.
- b) Collection of fresh fecal samples in sterile vials by opportunistic random sampling.
- c) Preservation of fecal samples in 2.5% of Potassium dichromate solution.
- d) Examination of fecal samples by using iodine wet mount, floatation and sedimentation techniques.
- e) Identification and measurement of eggs and cysts of parasites.

3.3.1 Sample collection method

All fresh fecal samples of *Columba livia* was collected randomly in early hour of morning to avoid the crowding of human population so that each of pigeons were watched very carefully. One hour of time was only spent at the study area while collecting the samples to prevent the duplication of sample from the same individual host. It took each morning for sample collection of each site. About five gm of fecal sample was collected in clean, sterile vial with the help of a spatula wearing gloves and mask. It was then preserved with 2.5% potassium dichromate. All the samples collected were labeled properly. The same collection process was repeated for all collected fecal samples.

3.3.2 Preservation of fecal samples

After sample collections, it was preserved in 2.5% potassium dichromate solution (2.5 gm potassium dichromate powder dissolved in one liter of distilled water). It helps in upholding morphology of protozoan parasites and prevents further development of helminth eggs and larva.

3.3.3 Sample size

Total 120 fecal samples of *Columba livia* were collected from present study areas. Out of 120, 60 samples of pigeon droppings were collected from each study area. The samples were collected on 22 and 23rd of April, 2016 A.D.

3.4 Laboratory examination

The collected fecal samples in vials were then brought to laboratory of Central Department of Zoology, Kirtipur, Kathmandu for test. The fecal samples were subjected to coprological examination by different concentration technique (floatation and sedimentation) and iodine wet mount method.

3.4.1 Iodine wet mount

About two gm of fecal samples were emulsified in a drop of Lugol's Iodine solution on a clean glass slide and then covered with a clean cover-slip. The smear was examined under electric microscope at 10X and 40X (Soulsby, 1965). This technique is generally used for the recovery of oocysts and motile trophozoites of protozoan parasites such as *Eimeria* sp. and *Giardia* sp. respectively.

3.4.2 Concentration techniques

Eggs, cysts and trophozoites are often in such low number in feces, that they are difficult to be detected in direct smears or mounts. Therefore, these procedures were performed which includes floatation and sedimentation techniques (Soulsby, 1982; Zajac and Conboy, 2012).

3.4.2.1 Floatation technique

This technique ensures the eggs float in the floatation liquid, which helps to identify the nematode and cestode eggs present in *Columba livia* feces.

Approximately two grams of fecal sample was put in a beaker and 28 ml of water was added. The samples were grinded lightly with the help of rod or pistle and the solution was filtered by tea strainer. The filtrate solution was poured into a centrifuge tube of 15 ml and centrifuged at 1000 rpm for five minutes. The tube's water was replaced with super saturated NaCl solution and again centrifuged.

After centrifuged, more saturated NaCl solution was added to develop convex meniscus at the top of the tube and one drop of Methylene blue (to stain) was also added. A cover-slip was placed for a five minutes. It was then removed from tube, placed on glass slide and examined microscopically at 10X and 40X. The photographs of eggs and cysts of parasites were taken and identified on the base of shape, shell and size.

3.4.2.2 Sedimentation technique

Saturated NaCl solution was removed gently from the centrifuge tube after examination of the floatation portion and the sediment content was poured into the watch glass and the content was stirred gently to mix it. One drop from the mixture was taken to prepare a second slide. The specimen was stained with Iodine wet mount's solution and examined microscopically at 10X and 40X. This technique is primarily used to identify eggs of

internal parasites that do not float well due to high specific gravity or presence of an operculum (eggs of flukes and false tapeworms). Such as eggs of trematodes. In this way, two slides were prepared from one sample (one from floatation and one from sedimentation).

3.4.3 Eggs and cysts size measurement

Eggs and cysts size were measured by using micrometry. The calibration factor was found to be 10.37 μ m for 10X and 2.588 μ m for 40X.

3.4.4 Eggs and cysts identification

On the basis of size, shape and shell characters given in the published literature journals and books (Assafa *et al.*, 2006; Cuomo *et al.*, 2009; Davis *et al.*, 1971; Gibbons *et al.*, 2005; Carney, 1991; Ritchie Schantz, 1997: 2006; Soulsby, 1982), eggs and cysts were identified.

3.5 Data analysis

On the basis of laboratory experiment, the data was recorded. The recorded data were coded and entered into Microsoft Excel 2007. Statistical analysis was performed using “R”, version 3.3.1 software packages. Chi-square test was used for statistical analysis of data to measure area wise prevalence, prevalence of protozoan and helminth parasites and prevalence of single and mixed infections. In all cases 95% confidence interval (CI) and $P < 0.05$ was considered for statistically significant difference. Percentage was used to calculate prevalence.

Sample collection, preservation and laboratory activities.



Photo 2: Flocks of pigeon at Krishna Temple.



Photo 3: Sample collection at Krishna Temple.



Photo 4: Sample collection at Pashupatinath.



Photo 5: Flocks of pigeon at Pashupatinath.



Photo 6: Microscopic examination.



Photo 7: Sample examination (Centrifugation).



Photo 8: Sample examination (Flotation technique).

4. RESULTS

4.1 General prevalence of GI parasites

Out of 120 fecal samples examined, 109 fecal samples were positive for one or more specific GI parasites, showing 90.83% prevalence of parasitic infection whereas 11 (9.17%) fecal samples were negative.

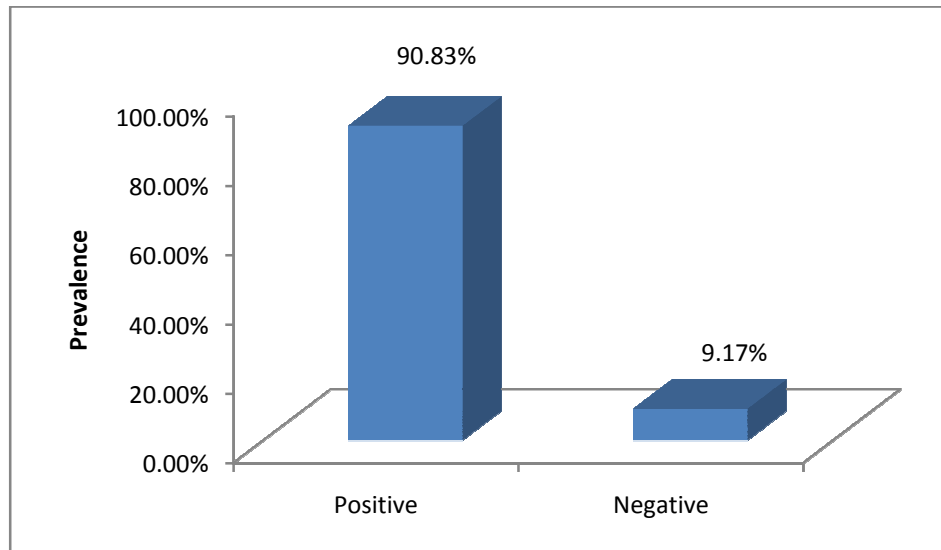


Figure 1: General prevalence of GI parasites.

4.2 Area wise prevalence

Out of two study area, 60 samples from each area (Pashupatinath temple and Krishna temple) were taken for examination. The area with higher prevalence of GI parasites was in Pashupatinath temple 57 (95%) and prevalence in Krishna temple was 52 (86.67%). Statistically, the difference in prevalence of GI parasitic infection among study area was found to be insignificant ($\chi^2=0.24$, $P>0.05$).

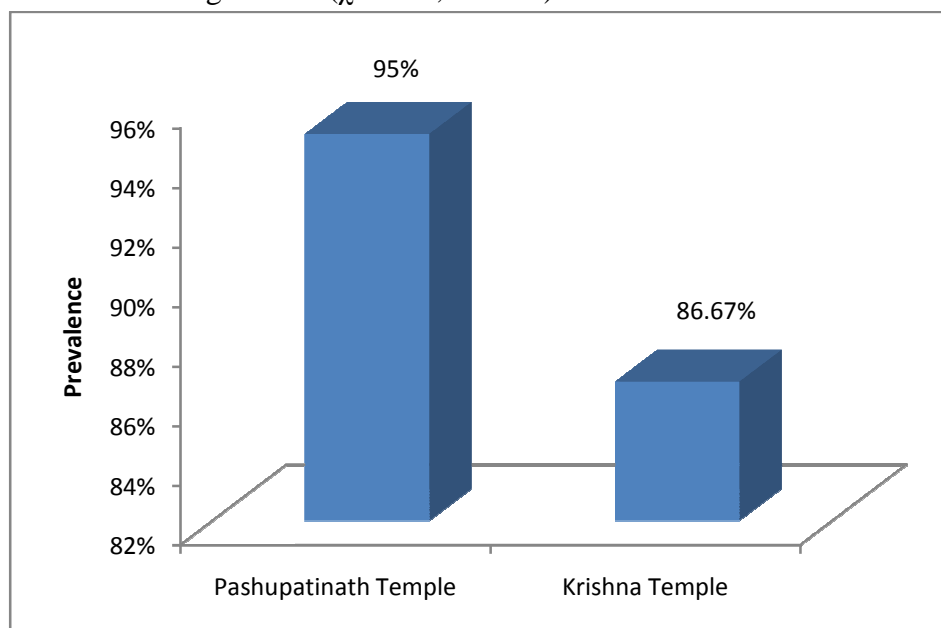


Figure 2: Prevalence of GI parasitic infection among study area.

4.3. Prevalence of protozoa and helminth parasites

Out of 120 total samples, 100 (83.34%) were positive with helminthes whereas 52 (43.34%) were seen positive with protozoan parasites.

Statistically, the difference in prevalence of helminthes and protozoan parasites were found to be significant ($\chi^2=15.14$, $P<0.05$).

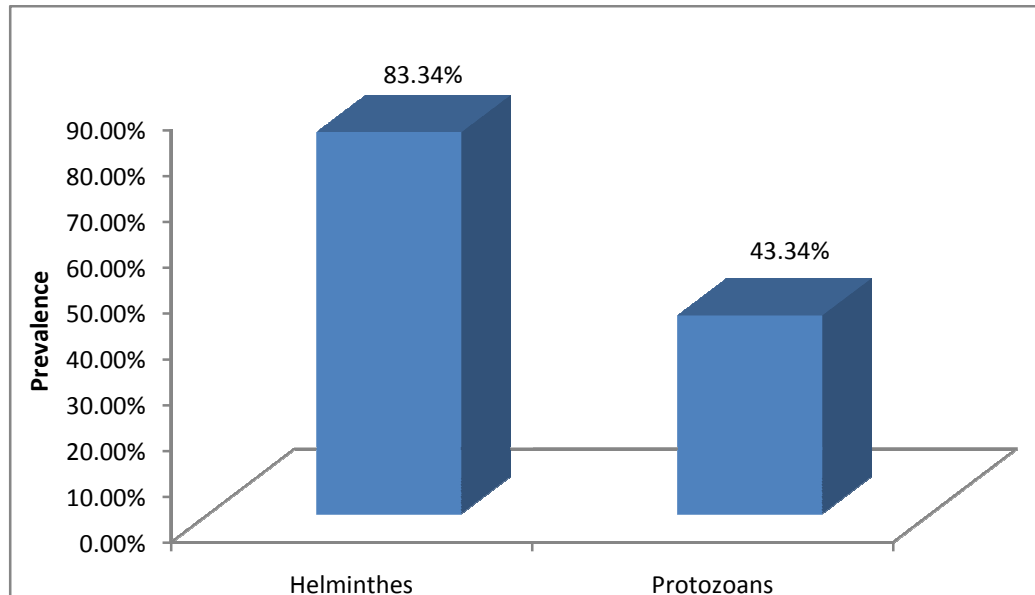


Figure 3: Prevalence of different helminth and protozoan parasites.

4.4. Prevalence of specific GI parasites

Out of 120 total samples, the prevalence rate 52 (43.34%) of *Eimeria* sp. were recorded. *Eimeria* sp. was only one species of protozoan parasite and among five helminth parasites *Capillaria* sp. 62 (51.67%) showed highest prevalence followed by *Ascaridia* sp. 33 (27.5%), *Syngamus* sp. five (4.17%), *Tetrameres* sp. two (1.7%) and *Heterakis* sp. 23 (19.17%). All these helminth parasites belong to the phylum nematoda.

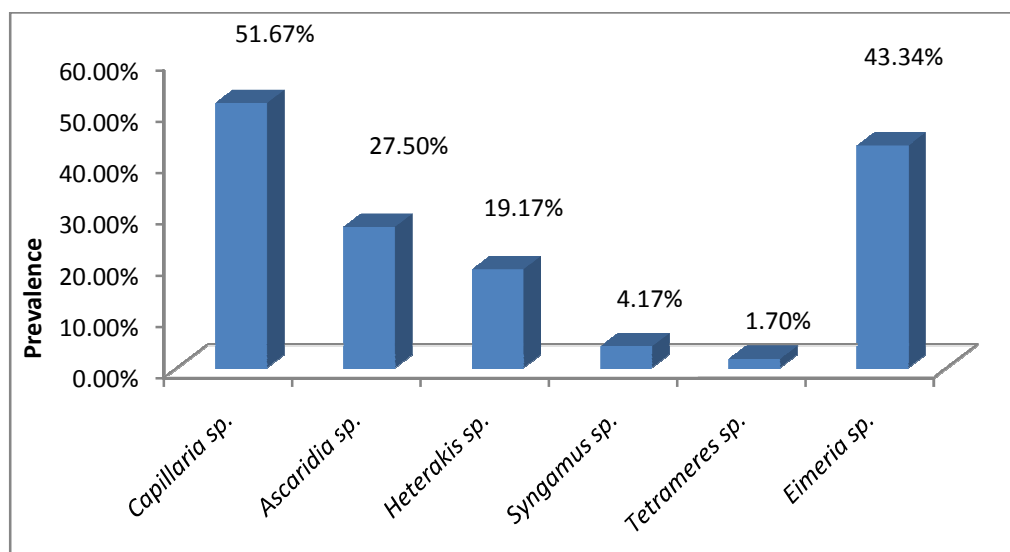


Figure 4: Prevalence of specific GI parasites.

Table 1: Morphometric characters of egg and cyst of specific GI parasites.

| S.N. | Parasites | Photo No. | Size (µm) | Size (µm) Soulsby, 1982 | Shape | Shell | Other features |
|-------------------|-----------------------|-----------|---------------|----------------------------|---------------------------------|-------------------------------|--------------------------------|
| Helminthes | | | | | | | |
| 1. | <i>Ascaridia</i> sp. | 9 | 75.05x54.3 | (68-90x40-60) | Ovoidal | thick and smooth three layers | Contents unsegmented |
| 2. | <i>Capillaria</i> sp. | 11 | 59.53x33.65 | (43-65x20-35) | Barrel shape with bipolar plugs | thick and smooth | Contents granular unsegmented |
| 3. | <i>Heterakis</i> sp. | 10 | 75.05x46.59 | (59-77x31-48) | Ellipsoidal | thick and smooth | Contents unsegmented |
| 4. | <i>Tetrameres</i> sp. | 13 | 30x47 | (25x50) | Ovoidal | thinner and hyaline | Embryonated when shed |
| 5. | <i>Syngamus</i> sp. | 12 | 82.9x45 | (78-100x43-60) | Ellipsoidal | thick | Morula present |
| Protozoa | | | | | | | |
| 6. | <i>Eimeria</i> sp. | 16 | 6-42 diameter | (10-40x10-30) | Round or ellipsoidal or ovoid | smooth | 4 sporocyst with 2 sporozoites |

4.5. Prevalence of single and mixed infections

Out of 120 total samples, the higher prevalence was of mixed infections 61 (50.84%) than the single infection 48 (40%). Statistically, the differences in the prevalence of single and mixed infections were found to be insignificant ($\chi^2 = 1.56$, $P > 0.05$).

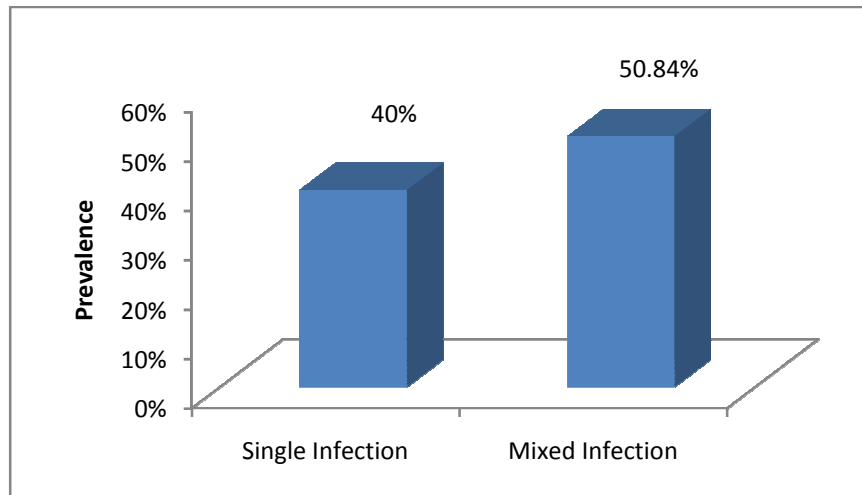


Figure 5: Prevalence of single and mixed infections.

4.6 Type of mixed infection

Out of 120 total samples, mixed infection was encountered more prevalent 61 (50.84%) than the single infection 48 (40%). Among mixed infections, double infection showed the highest rate 53 (86.88%) than the multiple infection 8 (13.12%) in this study.

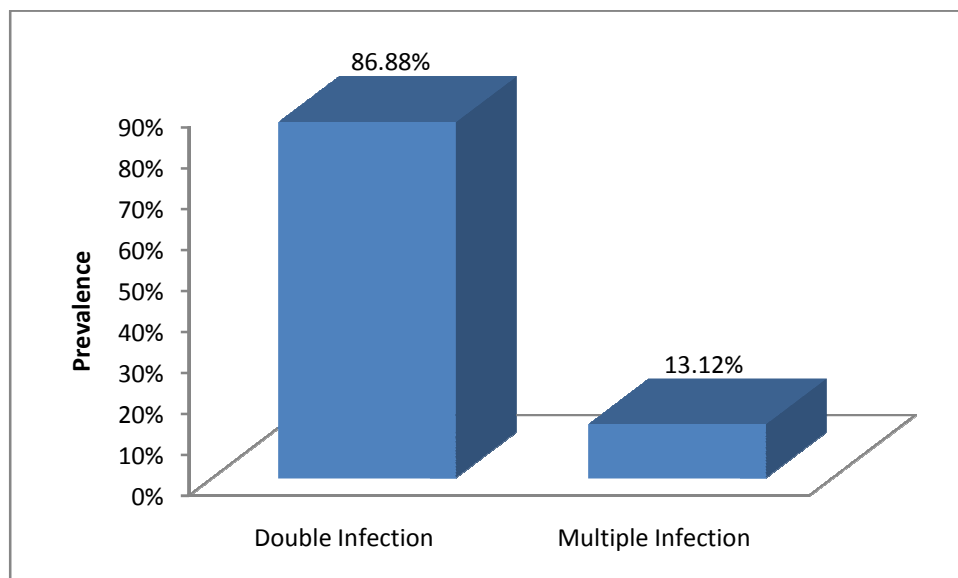


Figure 6: Prevalence of double and multiple infections.

Eggs of Nematode parasites in Pigeon under 10X*40X electron microscope.



Photo 9: *Ascaridia* sp. egg

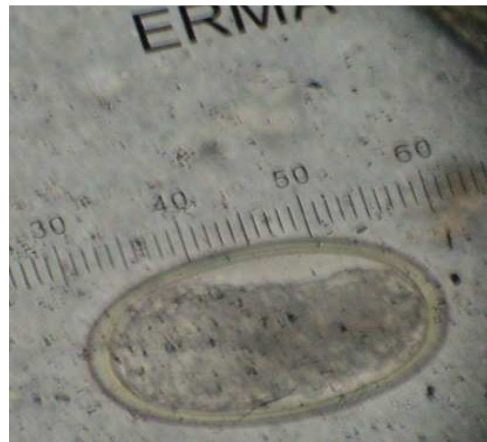


Photo 10: *Heterakis* sp. egg



Photo 11: *Capillaria* sp. Egg



Photo 12: *Syngamus* sp.egg



Photo 13: *Tetrameres* sp. Egg

Oocyst of Protozoan parasites in Pigeon under 10X*40X electron microscope.

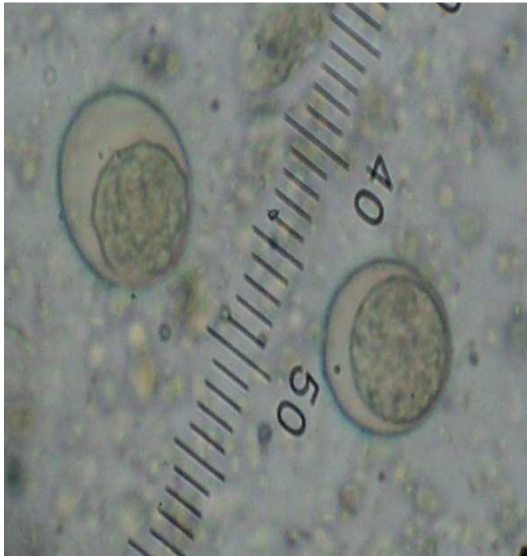


Photo 14: Unsporulated coccidian oocysts.



Photo 15: Sporulated oocyst of *Eimeria* sp.

5. DISCUSSIONS

Feral pigeons (*Columba livia*), also called city doves, city pigeons, or street pigeons, are pigeons that are derived from the domestic pigeons that have returned to the wild. There are about 100 different breeds and varieties described. They are primarily grain eater and seed eater but also feed on garbage, insects, livestock manure and other food materials. Feral pigeons find the ledges of buildings to be a substitute for sea cliffs, have become adapted to urban life, and are abundant in towns and cities throughout much of the world. Generally birds can be affected with both ectoparasites and endoparasites. The major endoparasites reported in pigeons, according to available literatures are *Ascaridia* sp., *Capillaria* sp., *Raillietina* sp., *Eimeria* sp., *Hymenolepsis* sp., *Cotugnia* sp. *Echinostoma* sp. *Tetrameres* sp. and *Heterakis* sp.

The present research was done to study the GI parasites of feral pigeon at two temples (Pashupatinath temple and Krishna temple) of Kathmandu valley. The overall prevalence rate of GI parasites in the present study was found to be 90.84%. The difference in prevalence of gastrointestinal parasitic infection among study area was found to be insignificant ($\chi^2=0.24$, $P>0.05$). It might be because of similar climate, food habits and environment. The prevalence rates of helminthes and protozoan parasites were significantly different ($\chi^2=15.14$, $P<0.05$). It may be because of pigeons have high antibody titres, to the protozoan parasite (Mushi *et al.*, 2000). The high prevalence of helminth infections recorded in this study could be an indication of a high incidence of the infective stages and intermediate hosts of the parasites in places where these pigeons are reared. The intermediate hosts of these parasites; beetles, pill bugs, ants, earthworms cockroaches and snails which form part of the diet of pigeons (Adang, 1999).

The overall prevalence rate (90.84%) of GI parasites in the present study was similar to the prevalence rate 90% recorded by the previous study (Parasani and Momin, 2010). The study was conducted in the Gujarat State of India. Similarly, the overall prevalence rate (90.84%) of GI parasites in the present study was higher than prevalence rates 74.14%, 72% and 72.70% obtained by the previous studies (Marques *et al.*, 2007; Sivajothi and Sudhakara, 2015; Ghosh *et al.*, 2014) respectively. Three different types of fecal qualitative tests; namely direct smear, floatation and sedimentation techniques that were used in present study was also used by them. Prevalence rate 84.78% obtained by (Radfar *et al.*, 2012) was nearly similar to the result determined in the present study. The variation in results may be due to different number of sample species, different methodology used, different climatic conditions or different time period and seasons of sample collection.

Moreover, the prevalence rate of present study was lower as compared to 100% prevalence rate of others finding (Al-Barwari and Saeed, 2012). The 100% prevalence rate of previous study (Al-Barwari and Saeed, 2012) was because of combine prevalence rate of fungi, protozoa, cestodes, nematodes and arthropods. Specific prevalence rate of

GI parasites was not given. 100% parasitic infections may be due to infection or re-infection (directly or indirectly) of parasites which indicates poor management and control efforts in the birds or in the immediate environment (Opara *et al.*, 2012).

Other previous reports showed comparatively lower prevalence rate of GI parasites 53.57%, 46.70% and 46.12% (Patel *et al.*, 2000; Sari *et al.*, 2008; Opara *et al.*, 2012) respectively than obtained by present study, though this rate of prevalence also indicates, health of pigeon is in risk. Patel *et al.* (2000) has studied upon captive birds of Gujarat zoos. Since there is more care in zoos as compared to outside, so this might be the main reason of less prevalence than present study and only sedimentation technique was used by them. Opara *et al.* (2012) examined only adult birds of street pigeons (*Columba livia*) and accepted certain level of host immunological response. They have considered moderate prevalence may be attributed to the food searching habits of the pigeons of not scratching below the surface soil where most infective stages of these nematodes are hidden. Sari *et al.* (2008) has studied parasites of domestic (*Columba livia domestica*) and wild (*Columba livia livia*) pigeons together in Nigde, Turkey. Combination study of wild and domestic pigeons as well as different seasonal and geographical variation might be cause of low prevalence rate of parasites than present study.

Helminth parasites cause watery diarrhea, weakness, weight loss, decreased milk production, reduced product quality, mortality and other secondary infections (Soulsby, 1982). The prevalence rate (83.34%) of helminthes in present study was higher than prevalence rates of 56.10%, 48.30% and 42.15% obtained by previous studies (Adang *et al.*, 2009; Adang *et al.*, 2008; Radfar *et al.*, 2011) respectively. The prevalence rates of helminth 36.90%, 33.33%, 23.50%, 18.70% and 12.50% shown by previous studies (Borji *et al.*, 2012; Musa *et al.*, 2011; Dranzoa *et al.*, 1999; Natala *et al.*, 2009; Olsen and Braun, 1980) respectively were lower than present study whereas prevalence rates of 84.78%, 79.50% and 79.20% obtained by previous studies (Radfar *et al.*, 2012; Msoffe *et al.*, 2010; Bahrami *et al.*, 2013) respectively were similar to present study. Among protozoan, 43.34% prevalence rates was recorded in present study which was lower than 72%, 46.27% and 43.13% of previous studies (Natala *et al.*, 2009; Nagwa *et al.*, 2013; Radfar *et al.*, 2011) respectively. The overall prevalence of various parasites differs greatly among the previous reports as well as when compared with present observation. This might be due to variance in sample collection methods, sample size and sample examination methods. Diversity of bird endoparasite assemblages may be related with many factors, which may include home range, behavior, size and roosting habit of the host. This may also be attributed to difference in the geographical areas and period of study (Begum and Sehrin, 2012).

Among six different GI parasites identified in present study, the prevalence rates of *Capillaria* sp. (51.67%) were higher than others. The prevalence of *Capillaria* sp. (51.67%) were higher than 24.20% and 22% of previous studies (Sari *et al.*, 2008; Ghosh *et al.*, 2014) respectively whereas it was also higher than 17.40%, 14.14%, 13.20% and 5.55% as compared to others findings (Sivajothi and Sudhakara, 2015; Bahrami *et al.*,

2013; Patel *et al.*, 2000; Borji *et al.*, 2012) respectively. Some *Capillaria* sp. (*Capillaria anatis*, *Capillaria obsignata*) have a direct life cycle. Domestic and wild birds ingest these infective eggs with contaminated food or water whereas other species (*Capillaria annulata*, *Capillaria bursata*, *Capillaria caudinflata*) have various earthworm species as obligate intermediate hosts. The larva develops inside the earthworms which become infective in two to four weeks and can survive for years inside it (Davis *et al.*, 1971). The high prevalence of *Capillaria* sp. in present study than other species is may be due to both direct and indirect life cycles of different species of *Capillaria*. The lining of the crop and the esophagus becomes inflamed and swollen, which can make swallowing impossible for affected birds. Fatalities are frequent in cases of heavy infections. White rumped vulture and slender billed vulture were reported infected with species of *Capillaria* (Gupta and Pandey, 2007). Similarly, literatures are also available for buffalo (Mukhia *et al.* 2007) and cat (Khanal and Gupta, 2004) for being infected by *Capillaria* sp. *Capillaria* sp. was also found prevalent in the poultry animals of Kathmandu (ADPCD, 1982).

In present study, prevalence rate (27.50%) of *Ascaridia* sp. were consistence with 30%, 28.33%, 20.75%, 20.37%, 16.66%, 15.50% and 15.21% obtained by previous studies (Mushi *et al.*, 2000; Begum and Sehrin, 2012; Patel *et al.*, 2000; Borji *et al.*, 2012; Radfar *et al.*, 2011; Msoffe *et al.*, 2010; Radfar *et al.*, 2012) respectively. The prevalence of present study was higher as compared to 5.10%, 4.04% and 1.20% of previous studies (Sari *et al.*, 2008; Bahrami *et al.*, 2013; Natala *et al.*, 2009) respectively. This is probably due to different climatic factors in the study areas (Chege *et al.*, 2015). The possible migration of *Ascaridia* sp. to liver, trachea and lung for development also suggest low prevalence (Michel, 1974). The prevalence rate of present study was lower as compared to 35% and 33.33% reported in previous studies (Ghosh *et al.* 2014; Sivajothi and Sudhakara, 2015) respectively. It might be due to difference in sample collection method. As they have collected samples from adult, squab and nestling pigeons. Gupta (1996) studied helminth parasites in domestic fowl of Kathmandu and determined the presence of *Ascaridia galli*. Infection with *Ascaridia galli* in white rumped vultures and slender billed vultures at Chitwan district was also reported (Gupta and Pandey, 2007).

In Nepal, Singh (1970) detected the presence of *Heterakis gallinae* and *Heterakis spumosa* from domestic fowl and rat respectively. Earlier, *Heterakis vesicularis* was also reported from domestic fowl (Sharma, 1942). In the present study, the prevalence rate (19.17%) of *Heterakis* sp. was higher than 9.02%, 3.7% and 3.3% obtained by previous studies (Ghosh *et al.*, 2014; Sari *et al.*, 2008; Adang *et al.*, 2008) respectively. This could be due to the difference in habitat and physiological condition of pigeons. These species are seen less in winter season in temperate region (Permin and Jorgen, 1998). *Heterakis gallinarum* is non-pathogenic, but a vector for *Histomonas meleagridis* which is highly pathogenic etiologic agent of “Black-head” disease lethal to chickens, turkeys, pheasants and other fowls (Cheng, 1973).

Syngamus sp. infection in the present study encountered the prevalence rate (4.17%) which is quite similar to 3% prevalence obtained by (Marques *et al.*, 2007; Ghosh *et al.*, 2014). In present study, determined prevalence was lower than 9.09%, and higher than

1.70% of previous studies (Bahrami *et al.*, 2013; Ghosh *et al.*, 2014; Sari *et al.*, 2008) respectively. From Nepal, (Gurung, 2016) reported 5.83% prevalence rate of *Syngamus* sp. from temple pigeons of Pokhara.

Bahrami *et al.* (2013) obtained 8.08% of *Tetrameres* sp. which is higher than 1.7% prevalence rate obtained in the present study. Poultry animals were observed with the infection of *Tetrameres fassisipina* (Singh, 1970). ADPCD (1982) shows the record of poultry animals of Kathmandu being infected with *Tetrameres* sp.

The prevalence rate (43.34%) of *Eimeria* sp. in present study showed higher result than 17.92%, 11% and 7.07% of previous studies (Patel *et al.*, 2000; Ghosh *et al.*, 2014; Bahrami *et al.*, 2013) respectively. This rate was similar to 49.20%, 46.27%, 46.20%, 43.15% and 31% of previous studies (Natala *et al.*, 2009; Nagwa *et al.*, 2013; Sari *et al.*, 2008; Radfar *et al.*, 2011; Sivajothi and Sudhakara, 2015) respectively. Result obtained from the present study is lower in comparison to 90% prevalence determined by (Eljader *et al.*, 2012). Coccidiosis infected birds generally exhibit loss of appetite, weakness, ruffled feathers, bloody diarrhea and can only be diagnosed by post-mortem examination (Dingle and Shanawany, 1999). The difference in prevalence rates of coccidian might be due to difference in practice of management area, hygiene of pens, flock structure, samples collected and laboratory techniques. In the temperate regions, the eggs of *Eimeria* cannot embryonate and develop to infectivity during the winter season of temperature below 10-15°C (Permin and Jorgen, 1998) might also cause low prevalence of coccidia.

The presence of *Raillietina* sp. has been recorded in different literature during literature review. In present study, there is absence of *Raillietina* sp. which is also absent in study of (Bahrami *et al.*, 2013). In Nepal, *Raillietina torquata* was recorded by (Meggett, 1924) from Kathmandu whereas (Sharma, 1943) recorded *Raillietina kantipura*, *Raillietina nagpurensis* and *Raillietina nripendra* from Kathmandu. *Raillietina* sp. requires two different host for complete life cycle. The definitive hosts are mostly wild and domestic birds, and sometimes human and several insects such as ants and beetles as intermediate hosts. Diagnosis of *Raillietina* sp. is usually done by post-mortem upon autopsy, since proglottids are seen in feces rather than eggs (Ritchie *et al.*, 1997) might be cause of absence of *Raillietina* sp. or might be because of plenty availability of grains and seeds in study area that prevents the intermediate host of *Raillietina* sp. as food.

There was no significant differences ($\chi^2=1.56$, $P>0.05$) in the prevalence of single and mixed infections in present study. The lower prevalence of single infection (40.00%) was seen than mixed infections (50.34%) which do not show similarity with some of previous study. The prevalence of single infection was seen higher in the previous studies (Adang *et al.*, 2008; Adang *et al.*, 2009; Bahrami *et al.*, 2013). Among the mixed infections, prevalence of double infection was 48.63% followed by multiple infection 7.34%. The lower prevalence of multiple infections in the pigeons may suggest a form of competition

that kept the other species away (Kennedy, 1975). It may be also because of crowding effect in pens and nest. Higher prevalence of mixed infection in present study suggests that health of feral pigeons in these temple areas are in high risk of morbidity and mortality.

This study indicated that pigeons at two different temples of Kathmandu valley are highly susceptible to GI parasites. Helminth infections is highly prevalent than protozoan parasites in this area and prevalence of mixed infection were higher than the prevalence of single infection. GI parasites can be controlled through effective management practices, daily cleaning of surrounding, washing dishes, quarantine new birds before entrance to the flock, clean water resources, eradication of intermediate host, regular treatments of anthelmintic, anticoccidials drugs and dusting of birds with pesticides as well as educating the breeders of birds, visitors in temples. Therefore, sustainable ways for controlling the parasitic infection and further studies need to be designed for the health and conservation of pigeons.

6. CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

The aim of the study was to investigate the general prevalence of GI parasites in feral temple pigeons. The overall prevalence of GI parasites of pigeons (*Columba livia*) was found to be 90.84% with higher prevalence rates of *Capillaria* sp. (51.67%). Out of six parasites, one protozoa: *Eimeria* sp. 52 (43.34%) and five helminthes: *Capillaria* sp. 62 (51.67%), *Ascaridia* sp. 33 (27.50%), *Syngamus* sp. 5 (4.17%), *Tetrameres* sp. 2 (1.70%) and *Heterakis* sp. 23 (19.17%) were identified. All these five helminthes are nematodes, trematodes as well as cestodes were not recorded from this study. Helminthiasis has been emerged as an important parasitic condition of feral pigeons in this study. Nematodes recorded from this study are well known to harm the host upto some degree but these nematodes from pigeons are not contagious to humans because these are not reported zoonotic with regards to human. The higher prevalence of GI parasites was in Pashupatinath temple 57 (95.00%) than the 52 (86.67%) of Krishna temple. Statistically, the difference in prevalence of GI parasitic infection among study area was found to be insignificant ($\chi^2=0.24$, $P>0.05$). The prevalence of helminthes 100 (83.34%) and protozoan parasites 52 (43.34%) were significantly different ($\chi^2=15.14$, $P<0.05$) whereas the difference in prevalence of single infection 48 (40.00%) and mixed infections 61 (50.84%) were insignificant ($\chi^2=1.56$, $P>0.05$). Among mixed infections, double infection showed the highest rate 53 (86.88%) than the multiple infection 8 (13.12%) in this study. The current study revealed heavy infection in feral pigeons at two temples of Kathmandu valley. The study indicated that feral pigeons are highly susceptible to GI parasites. Therefore, sustainable action should be designed and implemented to control the parasitic infection and reduce the health hazards of these feral pigeons.

6.2 Recommendations

- ❖ Routinely de-worming and fecal examination should be done for effective control of GI parasites.
- ❖ Vaccination programs against coccidiosis, anticoccidials drugs and dusting of birds with pesticides should be done.
- ❖ Droppings should be removed at frequent interval and surroundings as well should be kept clean.

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