GASTROINTESTINAL PARASITES OF TURKEY (*Meleagris gallopavo* Linnaeus, 1758) IN NAGARJUN TURKEY FARM, KATHMANDU, NEPAL

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In partial fulfillment of the requirements for the award of the degree of Master of Science in Zoology with special paper Parasitology

Submitted to

Central Department of Zoology

Institute of Science and Technology

Tribhuvan University

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Nepal

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DECLARATION

I hereby declare that the work presented in this thesis entitled **"Gastrointestinal parasites of turkey (*Meleagris gallopavo* Linnaeus, 1758) in Nagarjun turkey farm, Kathmandu, Nepal"** 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 author(s) or institution(s).

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LETTER OF APPROVAL

On the recommendation of supervisor "**Mr. Janak Raj Subedi**" this thesis submitted by Kabita Kunwar entitled **"Gastrointestinal parasites of turkey (*Meleagris gallopavo* Linnaeus, 1758) in Nagarjun turkey farm, Kathmandu, Nepal"** is approved for the examination and submitted to the Tribhuvan University in partial fulfillment of the requirements for Master's Degree of Science in Zoology with special paper Parasitology.

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CERTIFICATE OF ACCEPTANCE

This thesis work submitted by Kabita Kunwar entitled **"Gastrointestinal parasites of turkey (*Meleagris gallopavo* Linnaeus, 1758) in Nagarjun turkey farm, Kathmandu, Nepal"** has been accepted as partial fulfillment for the requirements of Master's Degree of Science in Zoology with special paper Parasitology.

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LISTS OF ABBREVIATIONS

Abbreviation form	**Details of abbreviation**
BMR	Basal metabolic rate
CI	Confidence interval
E	Eastern
EPG	Egg per gram
et al.,	ET alia (and other)
GI	Gastrointestinal
$K_2Cr_2O_7$	Potassium dichromate
MHC	Major Histocompatibility Complex
NaCl	Sodium chloride
PR	Prevalence Rate
P value	Probability value
rpm	Rotation per minute
T.U	Tribhuvan University

ABSTRACT

Gastrointestinal (GI) parasites can affect poultry productivity by compromising its health. The study was conducted from December to June to determine the prevalence of gastrointestinal parasites in turkey in the Nagarjun turkey farm, Kathmandu, Nepal. A total of 200 fecal samples, male turkeys (100) and female turkeys (100) were collected and preserved in a 2.5% potassium dichromate solution. These samples were examined microscopically by direct wet mount method and concentration methods viz. flotation technique and sedimentation technique, in the laboratory of Central Department of Zoology for detection of GI parasites. A total of 73.5% of turkeys were infected with one or more GI parasites. Six different parasitic genera were identified in turkey. Among the nematodes, *Ascaridia* sp. (26%), *Capillaria* sp. (17.5%) and *Heterakis* sp. (16.5%) were recorded. The cestodes were *Hymenolepis* sp. (10%) and *Raillietina* sp. (6.5%), while only *Eimeria* sp. (22.5%) was recorded as protozoa. The study revealed that female turkey (80%) had higher parasitic infection compared to male (67%). Statistically, the difference in sex-wise prevalence of GI parasite in turkey was found to be significant ($\chi 2=4.338$; $P>0.05$). Finding of this study shows that the prevalence of GI parasites of turkey was the highest during summer season (78%) and lowest in winter season (69%) with no statistical significant difference in between summer and winter season ($\chi 2= 2.079$; $P>0.05$). Single mode of infection was high revealing that maximum number of turkey was infected with single parasites with no significant differences in the prevalence of single and mixed infection ($\chi 2= 0.250$; $P>0.05$). Common parasites were detected from male and female turkeys because of their similar climate, food resources and environment. No any activities on the health care of turkeys regarding the GI parasites were found. Therefore, increasing awareness of regular anti-parasitic treatments for turkeys as well as other preventative and control measures is crucial.

Keywords: Turkey, Gastrointestinal parasites, Prevalence

1. INTRODUCTION

1.1 Background

Poultry farming has grown significantly over the last several decades and emerged as one of the most demanding component of livestock industry (Puttalakshmamma et al., 2008). In Nepal, poultry farming systems were introduced in January 2001 to address food shortages in low-income countries (Adebayo et al., 2013). Although poultry primarily refers to chicken, the term surrounds a variety of birds such as turkey, quail, duck, guinea fowl, and geese (Shreshtha, 2018). Due to its agricultural nature, Nepal's GDP heavily relies on this sector, contributing around 29%. With poultry production composing over 4% of the country's GDP, it emerges as one of the Nepal's proliferating agricultural industries (Nirmal and Pokharel, 2017). The poultry sector in Nepal can be classified into two types: commercial poultry and backyard poultry, accounting for approximately 55% and 45% respectively (Nirmal and Pokharel, 2017). Nepal has the poultry population of about 73 million with chickens contributing 1.63%, ducks 0.01% and turkeys 0.001% to the Agriculture GDP (MOALD, 2022). Turkey farming seems to be the least exploited among them. Global turkey production is increasing at a pace of 3% each year on average (Evans, 2003).

Turkey belongs to the order Galliformes which is native to North America and was named by Carl Linnaeus as "*Meleagris gallopavo*" based on Greek and Latin name (Aldrich, 1967). The ancient Greco-Romans gave the genus name "*Meleagris*" which means "guinea fowl". Latin meaning "peafowl" is used to describe the Asian species called "*gallopavo*" (*gallus* for cock and *pavo* for chicken like) (Al-Mahmodi et al., 2012). Turkey's live in small groups and feed on ground dwelling arthropods, molluscs and amphibians, vegetables nuts, seeds, and leaves (Udoh et al., 2014). Dietary requirements vary by age, sex, and season, but there were no consistent differences in feed use among turkeys older than poults (Laudadio et al., 2009). Turkey lives freely and feeds on food waste or get food from educators as well as various water sources (Yildirim et al., 2016). The majority of turkey population is disease-free or have adapted to naturally occurring diseases (V et al., 2013). Disease can occasionally be harmful but can cause losses due to decreased egg production, decreased hatchability, and increased poult mortality (Radfar et al., 2012). Intestinal complications in turkeys can arise from various factors like bacterial, viral, and parasitic infections, as well as management difficulty and nutritional deficiencies (ShahrokhRanjbarBahadory et al., 2014).

Poultry is the largest and the most popular components of the livestock industry, vertically integrated and intensified of the animal production industries and are also providing jobs and income for small farmers, especially non growing seasons for crops (Udoh et al., 2014). Farm animals are a crucial component of survival for rural poor people as they produce food, fertilizer, and income (Rodrigues Fortes et al., 2020). Turkey rearing is a popular practice worldwide, particularly in Europe and America, as they can thrive with proper care and protection from diseases, predators, and harsh weather (Ghimire et al., 2019). In 2005, the total production of meat Global poultry of 81 million tons/year is about 7%; it is France, Germany, Canada, USA, Netherlands, and Sweden are among most productive of turkey meat countries, which ranks second after chickens in the production of poultry meat (Al-Mayali and Kadhim, 2015). Turkeys are said to thrive more in arid conditions, they tolerate heat as compared to chickens and has higher quality meat with low fat content (Ilori et al., 2010). Their males are bigger than the female turkey and reported that carcass of turkey has higher number of protein than carcass of chicken (Ajayi et al., 2012; Oso et al., 2008). Turkeys are not only valued for their meat but also for their use in sacrificial offerings and the utilization of their bones, feathers, and other byproducts (Thornton et al., 2012).

1.2 Parasites in turkey

Parasitism can lead to the detrimental effects on birds, causing severe consequences such as malnutrition, stunted growth, reduced egg production, increased vulnerability to other infections, and even death in young birds (Radfar et al., 2012). The gastrointestinal tract plays a significant role in digestion and absorption of foods; so, it may lead to improper food absorption and slow growth performance as well as disruption of production if any changes occur in intestinal health and digestion (ShahrokhRanjbarBahadory et al., 2014). Turkey lives a symbiotic relationship with human societies and can be a factor fit parasites especially endoparasites (Gowthaman et al., 2013). The gastrointestinal tract of the turkey is invaded by some parasites such as protozoans, nematodes, acanthocephalans, cestodes and trematode through direct contact with parasite vectors, feces, and soil (Derksen et al., 2018). Poor hygiene, proximity to humans, as well as captivity conditions and environmental factors like rainfall, humidity, and temperature can favor parasite survival in free-range scavenging environments (Mohamed El-Dakhly et al., 2016a). While a few parasites may not pose significant issues, high parasites burden can have profound consequences on growth, egg production and overall health of birds (Tesfaheywet et al., 2012). When domestic turkeys are more crowded, they show an increased number of parasites (Littman, 2014).

In the poultry industry, there are several species of *Eimeria*, which are a type of coccidiosis-causing pathogen belonging to the Apicomplexa phylum (Mesa-Pineda et al., 2021). There are seven recognized species of Eimeria in poultry, each targeting a specific niche within the intestines and exhibiting varying levels of pathogenicity (Vrba et al., 2010). These species include *Eimeria meleagridis*, *Eimeria dispersa*, *Eimeria meleagrimitis*, *Eimeria adenoides*, *Eimeria gallopavonis*, *Eimeria innocua*, and *Eimeria subrotunda* (Chapman, 2008; Williams, 2010). The initial comprehensive examination of coccidia in turkeys were presented by (Hawkins, 1952; Tyzzer, 1929). These parasites are intracellular and rely on specialized organelles within the apical complex for invading the intestinal cells of the host (McDougald et al., 2020). Coccidial parasites are extremely species-specific, and once the coccidia have completed their life cycle, acquired immunity can be attained (Lee et al., 2022). However, since the birds may both carry the illness and become carriers after they become infected, the likelihood of coccidiosis spreading is increased (Ahad et al., 2015). Other protozoan parasite that can infect turkey flocks is '*Histomonas meleagris'* which is the most susceptible species found in turkeys causing the common poultry disease refers as histomoniasis (Daş et al., 2021). It causes blackhead disease in many galliform birds and its infection led to necrosis and ulceration of the cecal mucosa and liver and sulfur-yellow stools (Liu et al., 2018).

Helminth parasites have been identified as a significant factor contributing to poor health and decreased productivity in poultry (Uhuo et al., 2013a). The flocks of turkeys are afflicted by many helminth parasite kinds that can result in symptoms like catarrh, diarrhea, and intestinal blockage, loss of appetite, anemia, weakness, paralysis, and impaired feather quality in birds (Jegede et al., 2015). They can live comfortably in the crop, gizzard, gut, caecum, windpipe, and even the eyelids

(Gauthier and Ludlow, 2013). They can lead to various issues such as reduced feed conversion ratio, weight loss, decreased egg production in layers, and even mortality (Afolabi et al., 2016). The worms that are discovered in the caecum of the large intestine are known as *Heterakis* sp., those that are located in the eye are known as *Oxyspirura mansoni,* and tracheal worms are known as *Syngamus trachea* (Gauthier and Ludlow, 2013). *Heterakis gallinarum* is required for *Histomonas meleagridis* to survive outside of the host and for flock transmission. *H. meleagridis* can continue to cause disease in a flock in the absence of *H. gallinarum* (Hu and McDougald, 2003). Certain insects serve as carriers for helminths which are particularly attracted to higher temperature and to some degree of humidity (Jegede et al., 2015). Nematodes, Cestodes, and trematodes are significant parasites in poultry farming which are commonly found in the intestine or in fresh fecal samples (Fakae and Paul-Abiade, 2003). There are over 1400 cestodes species that can live in the intestines of birds, especially in free-range or backyard flocks (Biu and Haddabi, 2005). Cestodes are commonly found during warmer seasons, when plenty of intermediate hosts are available in birds (Uhuo et al., 2013b). The intermediate hosts for most of the cestodes species are beetles and houseflies that live in poultry farms (Singh and Nama, 2018). There are a variety of cestode species that have been found in turkeys, but none of them have been linked to anything more than mild pathology. In order to infect turkeys, all cestodes required intermediate hosts, typically invertebrates (Davidson, 2008). *Raillietina* sp. usually infects the bird's small intestines and can lead to weight loss, abnormal growth, and blockage in the digestive system (Khan et al., 2022). Though *Raillietina* sp. infection doesn't always cause high morality, it may result in long-lasting and gradual harm to birds (Zhang et al., 2021).

Capillaria is a nematode of small intestines of domestic and wild birds such as chicken, duck, and geese, guinea fowl that causes severe infection of weight loss, diarrhea, and economic losses (Hoque et al., 2014). *Capillaria* can infect and cause inflammation in upper digestive tract, especially in gallinaceous birds (Benisheikh et al., 2020). In domestic and wild birds, a nematode of small intestine *Ascaridia galli* causes severe infection such as diarrhea, decreased egg production, emaciation, and anemia and which are distributed worldwide (Daş et al., 2010). *Ascaridia* eggs have the remarkable ability to withstand the extreme conditions in poultry houses and can remain infectious for over 6 months, leading to reinfection and contamination of the environment with new eggs (Collins et al., 2021). *Echinostoma* trematode of small intestine of birds and are found on the area where there are suitable conditions for growth of intermediate host (Labony et al., 2022). Acanthocephalans are non-segmented, but these infection are rare and are sometimes considered incidental which usually cause subclinical infections in turkeys (Davidson, 2008).

Backyard poultry farming is a vital aspect of the rural population for a significant source of revenue and a means of ensuring food security in unprotected communities (Luka and Ndams, 2007). This is because it is believed that small-scale livestock production is a practical substitute for ensuring food security, and assistance for backyard poultry farming has been frequently employed (Ara et al., 2021). There is a high prevalence of parasitic infestations, which results in poor economic conditions, increased mortality, increased prophylaxis, low production, animal deaths, and low productivity due to low biosecurity (Derksen et al., 2018). These flocks have poor biosecurity and have frequent access to the outdoors, which allows them to meet wild birds and other animals, such as rodents, that can transmit diseases (Whitehead and Roberts, 2014). Inadequate management techniques, including wet litter that encourages oocyst sporulation, contaminated feeders and drinks, inadequate ventilation systems, and excessive stocking densities, might make the clinical infection worse (Khan et al., 2006). Although, the incidence of parasitic infections cannot be decimated completely but their number can be controlled by improving poultry management techniques and educating the small holders about the danger of the diverse gastrointestinal infections (Jaiswal et al., 2020).

1.3 Objectives

1.3.1 General objective

- To determine the prevalence of gastrointestinal (GI) parasites of turkey (*Meleagris gallopavo* Linnaeus, 1758) in Nagarjun Turkey Farm, Kathmandu, Nepal.

1.3.2 Specific objectives

- To determine the prevalence of general and specific gastrointestinal parasites of turkey.

- To determine the prevalence of gastrointestinal parasites of turkey based on sex and season.

1.4 Significance of the study

The industry of raising turkeys is severely hampered by the parasite infections that turkeys are vulnerable to. There have been no studies conducted till date on the prevalence of turkey gastrointestinal parasites in Nepal. Animal protein (meat and eggs) becomes a more crucial nutrient food as the world's population of human rises. Due to their varied breeding and raising practices, turkeys are prone to a wide range of helminthic and protozoan infections in addition to their many other uses. This study therefore has the potential to prevent infection in turkeys by identifying the parasites and incorporating it into poultry management through the provision of a sanitary environment, nutritious feed, and veterinarian suspension. Future researchers will be able to use this study as a reference.

1.5 Hypothesis

The following are the null and alternative hypothesis for the present study:

H_0= the level of gastrointestinal parasites and risk factors did not differ significantly.

H_1= the level of gastrointestinal parasites and risk factors differed significantly.

2. LITERATURE REVIEW

Domestic turkeys show an increased number of parasites when turkeys are more crowded (Markley, 1967). Turkeys are host to many different parasites; these parasites cause the loss of nutrients, damage to the intestines and other organs, loss of blood, secondary infections, and behavioral changes such as a higher likelihood to leave the nest (Oates et al., 2005). Most of these cause subclinical infections and very few cause disease or mortality (Davidson, 2008). Poultry farming is the primary source of commercial meat and egg production. But still, an excess of intestinal parasites can have a disastrous impact on development, egg production, and general health (Markley, 1967). Researchers from around the world have conducted studies on parasites of turkeys and other poultry birds, focusing on Africa and Asia.

2.1 In global context

Globally, the majority of studies were conducted on the African subcontinent. In Nigeria, (Ola-Fadunsin et al., 2019) reported a higher GI prevalence of 95.65% from Ilorin and recorded parasites like *Eimeria* sp., *Ascaridia galli*, *Heterakis gallinarum*, *Subulura brumpti* and *Capillaria annulata* by direct wet mount examination and floatation techniques were performed. A similar study was conducted by (Jegede et al., 2019) reported 95 % parasitic prevalence and found 16 different species of parasites like *Ascaridia* sp., *Strongyloides* sp., *Capillaria* sp., *Heterakis* sp., *Tetrameres* sp., *Spirurid* sp., *Raillientina* sp., *Davainea* sp., *Subulura* sp., *Oxyuris* sp., *Cyathostoma* sp., *Syngamus* sp., *Eimeria* sp., *Cryptosporidium* sp., *Sarcocystis* sp., and *Trichomonas* sp. in Gwagwalada. Likewise, the higher occurrence of GI prevalence was also reported by other Asian and American subcontinent. The higher prevalence of GI parasites (77.3%) were conducted by (Montes-Vergara et al., 2021) in Colombia, which includes one protozoan, four cestodes, and six nematodes species. *Capillaria* sp., *Ascaridia galli*, *Tetrameres* sp., *Heterakis gallinarum*, *Syngamus trachea* and *Strongylus* were the nematodes observed. *Choanotaenia infundibulum*, *Davainea proglottina*, *Raillietina* sp., *Hymenolepis* sp. were the cestodes and *Eimeria* sp. was the observed protozoa. Similar highest prevalence of GI parasites of turkeys in Iran was studied by (ShahrokhRanjbarBahadory et al., 2014) which reported 75% of samples were infected with nematode, cestode and trematode i.e. *Capillaria*, *Ascaridia galli*, *Raillietina tetragona*, *Raillietina echinobothrida* and *Echinostoma*. A study conducted by (Nipu, 2019) reported (74%) prevalence of helminth infections i.e. *Heterakis gallinarum* and *Capillaria philippinensis* in Dhaka, Bangalore. A study carried out in Iran by (Farhang,H. H., 2012) reported highest GI prevalence with six helminth species comprising four nematodes and two cestode species i.e. *Ascaridia galli*, *Heterakis gallinarum*, *Subulura brumpti*, *Raillietina tetragona* and *Raillietina echinobothrida* respectively. A study conducted by (Dauda et al., 2016) found overall prevalence of 68.25% among 400 turkeys in Bukuru – Jos metropolis, Nigeria with some nematodes i.e. *Ascaridia* sp., *Capillaria* sp., and *Cheilospirura spinosa*. Similar prevalence of 60% GI parasites was recorded by (Opara et al., 2014) with *Ascaridia* sp. infected the turkeys. A study conducted by (Martinez-Guerrero et al., 2010) in Sierra Madre Occidental of Durango, Mexico on the gastrointestinal tracts (GIT) of the wild Gould's turkey (*Meleagris gallopavo mexicana*) which showed 100 % parasitic prevalence from the species *Raillietina tetragona* and *Eimeria* spp. and has the highest abundance of 59.4 % for *Heterakis gallinarum*, which is clinically important by being associated with the disease enterohepatitis. According to (Jegede et al., 2019; Ola-Fadunsin et al., 2019; Udoh et al., 2014) a higher prevalence rate of helminth parasites compared to protozoan parasites was recorded in Nigeria. Similarly in Iraq, (Khalaf, 2022) found high helminthic infection consists of five nematode species including *Heterakis gallinarum* (28%), *Capillaria sp*. (24%), *Trichostrongylus sp*. (16%), *Strongyloides avium* (12%), and *Ascaridia galli* accounts for 12% compared to protozoan infection i.e. *Eimeria spp*. (48%). Comparatively high prevalence of *Ascaridia* sp. over *Heterakis* sp. has been reported from Iran, Nigeria, Colombia and Pakistan (Dauda et al., 2016; Farhang,H. H., 2012; Montes-Vergara et al., 2021; Sadaf, T, 2021) respectively. Whereas in Iraq, a study conducted by (Jegede et al., 2019) reported high prevalence of *Heterakis* sp. over *Ascaridia* sp.. According to (Jegede et al., 2019) both male and female turkeys were reported to infected with helminths and protozoan parasites in their local and exotic breeds. A study carried out by (Ola-Fadunsin et al., 2019) reported a higher prevalence of GI parasites in female turkey compared to male turkey in their study on different avian species. Similar study was conducted by (Matur et al. 2010) on exotic and local chicken which reported female chickens were more prone to GI parasites than in male. Likewise, (Atsanda et al. 2015) reported that the female guinea fowls were more infected with GI helminths than male guinea fowls. But the result conflicted with the research done by (Dauda et al., 2016; Udoh et al., 2014) which recorded the higher incidence rate of

GI parasites infection in male than in female turkeys. Similarly, a study performed by (Attah et al. 2013) reported the higher prevalence of GI helminths in male chicken and guinea fowls than in female ones. According to our findings, in Marathwada region of Maharashtra (Naphade & Chaudhari 2013) described the seasonal prevalence of helminth parasites which showed higher prevalence in summer season compared to winter season. Likewise, (Shahin et al., 2011) reported the higher incidence rate of cestodes occurred in summer and autumn season whereas lowest in winter and spring season. In Egypt, (Nagwa et al., 2013)found the highest rate of infection in summer (88%) while the lowest rate was recorded in winter (19%) for turkey. Likewise, (Ahad et al., 2015) reported the higher prevalence of coccidian parasites in broliers during summer and lowest during the winter season in Kashmir valley. Another study conducted in Egypt by (Mohamed El-Dakhly et al., 2016b) over the course of the months only helminthes species found in turkeys where roundworms and tapeworms were especially common in summer and trematodes was discovered during the winter season. But this result conflicted with the study done by (Islam et al. 2009) which reported the higher occurrence of GI parasites was found in winter season followed by rainy and then in summer season. According to the studies carried out by (Mungube et al., 2008; Percy et al., 2012; Soomro et al., 2010) reported higher infection of *Ascaridia galli* in summer season which was observed in Kenya, Pakistan, Zimbabwe respectively. The higher prevalence of single infection was seen than mixed infections (Adang et al., 2008) in Nigeria. This result is supported by (Adang et al., 2009; Bahrami et al., 2013) in Nigeria and Iran which recorded higher rate of single infection compared to mixed type. Whereas, this result conflicted with the research done by (Jegede et al., 2019) conducted a study between the local and exotic breeds of turkeys which detected the majority of the birds having mixed infection. None of the turkeys examined was infected with trematodes. According to the study carried by (Khalaf, 2022), single infection showed the majority of infection, followed by double infection and mixed infection. According to our findings, (Jayentakumar Singh and Mohilal, 2017) showed the higher GI prevalence of 66.7% consists of *Eimeria* with mixed infection of protozoan and helminthes in valley districts of Manipur, which was examined microscopically by direct wet smear with 2.5 % potassium dichromate. A study carried out by (Ola-Fadunsin et al., 2019) describes multiple parasites co-infection found in all the avian species such as broilers, cockerels, layers, indigenous chickens, pigeons, guinea fowls, ducks, and turkeys. Likewise, a study performed by (ElKhawas, 2020) examined 100 fecal samples and buccal swabs from 80 sick and 20 healthy turkeys on the private farms where the infection rates for single and combined infections were, respectively, 68.75%, 31.25%, 45.45%, and 54.55%. The internal parasites discovered in the investigation were *Tritrichomonas eberthi, Ascaridia galli,* and *Eimeria meleagrimitis*.

Likewise, the mid-level occurrence of GI prevalence was studied by (Udoh et al., 2014) reported (57.7%) having parasites like *Ascaridia* sp. that was discovered the most frequently, followed by *Eimeria* sp., *Subulura brumpti, Raillietina cesticillus, Heterakis gallinarum, Capillaria* sp., *Choanotaenia infundibulum, Davainea meleagridis,* and *Methroliasthes lucida*. In Colombia, (Montes-Vergara et al., 2021) found *Eimeria* sp. was the most common intestinal parasite followed by *Heterakis gallinarum*, *Raillietina* sp., *Hymenolepis* sp., *Capillaria* sp., *Syngamus trachea, Tetrameres* sp., *Ascaridia galli*, and *Strongylus* but in Mexico, (Martinez-Guerrero et al., 2010) recorded *Eimeria* spp. and *Raillietina tetragona* has 100 % parasitic prevalence followed by 59.4% for *Heterakis gallinarum.* In the Konya Central villages, Turkey, a study conducted on domestic turkeys by (Sevinç, 2000) reported GI prevalence of 52.5% through the macroscopic inspection of the gastrointestinal tract following parasites were detected such as *Eimeria* sp., *Heterakis* sp., *Subulura* sp., *Echinostoma* sp., and *Ascaridia galli* and in the microscopic inspection, eggs of *Eimeria* sp., *Capillaria* sp., *Ascaridia* sp., *Choanotaenia infundibulum, Trichostrongylus tenuis*, and *Heterakis gallinarum* were found. In Florida, (Hon et al., 1975) reported 34 different helminth species including trematode (10), cestode (six), nematode (17), and one acanthocephalan species. This result supported by (Maxfield et al., 1963) cestode species, two trematode species, one nematode species, and one acanthocephalan species. In Umiam, Meghalaya, a study was carried out by (Das, 2015) in which eggs of *Ascaridia galli* (EPG 50-350) and *Capillaria sp*. (EPG 50-100) by floatation technique using saturated sugar solution through microscopic examination was reported in turkeys.

An investigation studied by (Khalaf, 2022) in Iraq taken from 71 random samples of droppings where 35.21% of overall prevalence of gastrointestinal parasite infection had been found where five nematode species and one protozoan species were observed including *Heterakis gallinarum*, *Capillaria* sp., *Trichostrongylus* sp., *Strongyloides avium*, *Ascaridia galli* and *Eimeria* species. A similar method was used and a low parasitic prevalence (23.5%) was obtained by (Assam et al., 2020) in Kaduna state with various parasites like Coccidia, *Ascaridia,* nematode larvae, *Capillaria*, *Syngamus, Raillietina and Trichuris* for each endoparasites. A study is conducted by (ElKhawas, 2020) on 100 fecal samples and buccal swabs from 80 sick and 20 healthy turkeys (25 from each, 20 sick, and 5 healthy) on the private farms showed the lower infection rate of 40% in the sick birds and 55% in the healthy birds. The internal parasites discovered in the investigation were *Tritrichomonas eberthi, Ascaridia galli,* and *Eimeria meleagrimitis*. Similarly, the lower occurrence of GI parasites (6%) was observed in Egypt which was conducted by (Mohamed El-Dakhly et al., 2016b) where only helminthes species found in turkeys i.e. *Ascaridia dissimilis*. A study was conducted in Pakistan by (Sadaf, T, 2021) reported the lower prevalence rate of GI parasites where 100 fecal samples for each pet birds were analyzed and 6 species of nematode were recorded from fecal samples i.e. *Syngamus trachea*, *Capillaria anatis*, *Capillaria annulata*, *Heterakis gallinarum*, *Ascaridia galli* and *Allodpa suctoria*. Two species of trematodes i.e. *Prosthogonimus ovatus* and *Prosthogonimus macrorchis*, single cestode species *Raillietina echinobothrida* and three protozoan species i.e. *Eimeria maxima, Histomonas meleagridis* and *Giardia lamblia* were recorded from the fecal samples. Whereas, (Sevinç, 2000) described the protozoan (37.5%) infection as dominant GI parasites than helminthic (25.5%) infection in Konya Central villages, Turkey. This result agrees with data of a study done by (Assam et al., 2020) reported in Kaduna State of Nigeria. A study was carried out by (Singh and Nama, 2018) in India which recorded 5% of turkeys tested positive for *Capillaria* sp. (EPG 42-25) in representative feces samples. Likewise, in Malaysia, a study conducted by (Ab Hamid, 2017) examined two species of endoparasite infection in the parasite eggs of nematodes and protozoa with 7300 EPG compared to *Capillaria* sp. with only 1200 EPG.

2.2 In context of Nepal

No article has been carried out in context of gastrointestinal parasites of turkey in Nepal. However, some articles on poultry bird GI parasites have been described.

Similarily, a study conducted in 120 fecal samples of ducks reared in three different locations (Bishnu-Devi, Kanchan-Basti, Balambu) of Chandagiri Municipality shows higher nematode positive rate (74.49%), followed by cestode (52.04%) and protozoans (41.84%) (Shrestha et al. 2020). The study conducted by (Adhikari et al., 2008) discovered five *Eimeria sp*. in layer chickens in Chitwan, with highest prevalence of mixed infection (*Eimeria acervulina*, *Eimeria maxima, Eimeria necatrix, Eimeria brunette*, and *Eimeria tenella*). Similar studies have been conducted in Kathmandu and Lalitpur district poultry farms (Jayswal et al., 2014) reported four species of *Eimeria,* with *Eimeria tenella* having the highest prevalence rate. Two species of *Eimeria* i.e. *Eimeria tenella* (12%), *Eimeria maxima* (7%) in Kadaknath and cross breed from farm was discovered by (Khadka, 2019). The higher prevalence of single infection was seen than mixed infection in pigeon which is described by (Gurung, 2016) in three temples of Pokhara valley. Similarly, (Sukupayo, 2018) reported the same result among GI parasites having higher single infection rate than multiple in pigeons of Bhaktapur.

Over 24 million domestic poultry are kept in Nepal, with 55% kept in backyards using a free-range approach (Khanal et al., 2015). Among the poultry, helminthic infection showed a high prevalence rate than protozoan infection (Khadka, 2019; Resmi, 2021; Shreshtha, 2018). A study conducted by (Mujahid, 2017) showed chicken nematodes in Lalitpur recorded a high prevalence showed *Heterakis gallinarum* (22.4%) followed by *Capillaria* sp. (16%), *Ascaridia galli* (10.4%), unidentified (4.8%) and *Raillietina teragona* (4%). But the result conflicted with the research done by (Resmi 2021) found a high prevalence of *Ascaridia* sp. (48.57%) followed by *Echinostoma* sp. (35.23%), *Heterakis* sp. (19.05%), *Strongyle* sp. (11.43%) and *Trichostrongylus* sp. (9.52%) in barn swallows of Tansen Municipality of Palpa District. This result is supported by (Khadka, 2019; Shreshtha, 2018) showed *Ascaridia* sp. to be the most common helminth parasites of poultry in Nepal.

3. MATERIALS AND METHODS

3.1 Study area

Nagarjun municipality is in Kathmandu district in Bagmati Province of Nepal. At the time of 2011 Nepal census, it had a total population of the municipality is 67,420 people residing in 16,746 households. The total area of Nagarjun Municipality is about 29.8 km^2 and is divided into 10 municipal wards. It lies on the geographical coordinates of $27^0$43'57"N and $85^0$15'24"E at an altitude of 1300 meters to 2500 meters above sea level. The climate of this area evenly distributed precipitation throughout the year. The annual temperature is about 17.6^0C and the annual precipitation is about 65.4 inch per year. Many people in the region are involved in poultry farming on both small and large scales. Summer season is very hot and winter is very cold. Summer lasts from March to mid-June, monsoon lasts from mid- June to September and winter lasts from December to February.

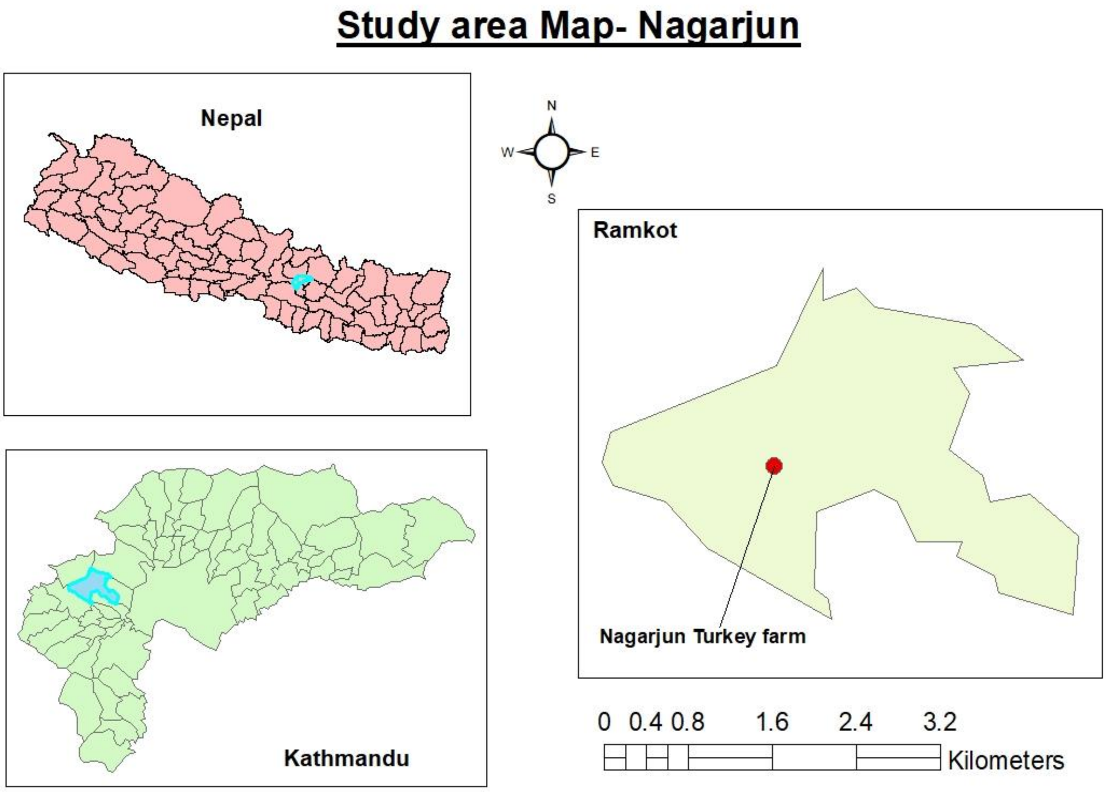

Figure 1: Map of study area

3.2 Material required

3.2.1 Materials

Collecting sterile vials, gloves, masks, stage microscope and ocular micrometer, centrifuge machine, centrifuge tube, measuring cylinder, glass slides, weighing machine, cover slip and compound microscope.

3.2.2 Chemicals

2.5% $K_2CR_2O_7$ (Potassium Dichromate), Normal saline (0.85%), distilled water, Lugol's iodine solution, 10% formalin, Ethyl acetate, sodium monophosphate and sodium bi-phosphate, Immersion oil, hand wash, NaCl solution and Ziehl-Neelsen (ZN) Acid-fast stain.

3.3 Methods

3.3.1 Preparation of Normal saline

8.5gm of sodium chloride is dissolved in 1000ml of distilled water for preparation of normal saline and used in unstained preparation (Zajac et al., 2021).

3.3.2 Preparation of 2.5% Potassium dichromate

2.5gm of $K_2Cr_2O_7$ was dissolved in 100 ml in distilled water after measured accurately. This solution is essential for the preservation of parasites found in the collected stool samples and ensuring the preservation of integrity of cysts and eggs within the samples (Zajac et al., 2021).

3.3.3 Preparation of Lugol's Iodine solution

A solution was created by dissolving 10gm potassium iodine with100 ml of distilled water. Additional 5gm of iodine was added to the solution which later was filtered and stored in a bottle. To examine the internal characteristics and identify the different species of protozoans, iodine solution was utilized (Zajac et al., 2021).

3.3.4 Preparation of buffered 10% formalin solution

In 90 ml distilled water, 10 ml of concentrated formaldehyde (35-40%) was added. And after that 0.08gm of phosphate buffer was added to the solution. 6.1gm of disodium phosphate and 0.15gm of monosodium phosphate was weighted first for the preparation of phosphate buffer, then it was mixed together to form a phosphate powder. Finally, 0.08gm of buffered phosphate was weighted and added to the 10% formalin and mixed well (Garcia, 2021).

3.4 Study design

The purpose of the present study was to assess the severity of gastrointestinal parasitic infection of turkey in turkey farm. The study consists of the following steps:

a) Collection of fresh fecal samples in the sterile glass vials.
b) Preserving fecal samples in a solution containing 2.5% potassium dichromate.
c) Conduct a key informant interview.
d) Examining feces samples through the use of direct smear, sedimentation, flotation, and acid-fast stain procedures.
e) Measurement and identification of parasite eggs and cysts.
f) Data analysis through MS-Excel and SPSS (Statistical Package for Social Science).

3.4.1 Sample collection method

The fresh fecal samples of turkeys were gathered in early hours of morning. It took a total of 10 days for sample collection on the site. A total of 20–25 g of feces was gathered in the morning with the help of farm's caretaker. To avoid the potential of contamination from the ground, plastic sheets were placed on the floor of poultry areas where turkeys were kept. It creates a physical barrier and hinders the transmission of parasites from the ground to the fecal samples. Each turkey's sex is identified by using leg bands. Disposable gloves were used during the collection of samples, to prevent any kind of contamination.

3.4.2 Preservation of fecal samples

The collected fecal sample was placed in a sterile bottle and fully covered with 2.5% potassium dichromate (2.5gm potassium dichromate powder dissolved in one liter of distilled water). It aid in preserving the morphology of protozoan parasites and halting the ongoing growth of helminthes egg and larva. In semisolid or solid state, the fecal samples were collected. The sex of turkeys was identified during fecal collection (Table 1).

Table 1: Sex categories used in the study

S.N	Category	Description
1.	Male turkey	Larger individuals with longer tails and legs, a long and droopy snood on their bills.
2.	Female turkey	Smaller individuals with shorter tails and legs, have dull coloring.

All of the samples that were gathered had accurate labels. Vials were then kept in an airtight cool box, following which the sample was taken to the Central Department of Zoology.

3.4.3 Sample size

There are around 300 turkey birds in the whole study area. To study the gastro-intestinal parasites of turkey, a total of 200 samples were collected comprising 100 of male and 100 of female turkey from the farm in Nagarjun, Kathmandu. These samples were collected from the Nagarjun municipality of Kathmandu valley from December 2021 to June 2022 A.D. The sample collected from the study area was brought to the laboratory for examination of GI parasites. The sample size occupies about 68% of whole population.

3.4.4 Interview

Verbal surveys were conducted with the workers and the owner of turkey farm concerning gastrointestinal parasites in turkey.

3.5 Laboratory examination

Fecal samples were preserved and transported, and then all samples were examined at the Central Department of Zoology laboratory at T.U. Kirtipur in Kathmandu. By using stained smear preparation and concentration methods, such as floatation and sedimentation procedures, the fecal samples were analyzed under a microscope for trophozoites, cysts, oocysts, eggs, and larvae of gastro-intestinal parasite. The Stoll's count technique was used to determine mix infection of parasites (Arora and Arora, 2015).

3.5.1 Direct smear method

For many laboratory procedures, smear preparation is necessary. Making a smear is done to stick the parasitic cysts, ova, and eggs to the slide.

3.5.1.1 Saline wet mount examination

1-2 drops of sample were placed on a clean slide using a clean bamboo toothpick after mixed thoroughly, and then cover with a cover slip. Excess fluid was discarded with the help of filter paper which is followed by observation of sample under the microscope at magnification of 100 × and 400× power (Pradhan et al., 2014).

3.5.1.2 Stained preparation of stool smear

2-3 drops of feces was collected and emulsified with Lugol's iodine solution on a clean glass slide, and then covered with a cover slip. The smear was examined at a total magnification of 100 × and 400× power. It is helpful for studying the nuclear character and recognizing protozoan cysts and trophozoites (Malla et al., 2004).

3.5.2 Concentration methods

Techniques for the detecting parasites of eggs, cysts, trophozoites, and larva are included in the concentration procedures, including floatation and sedimentation methods (Arora and Arora, 2015). Parasites can be easily detected in smears in case of heavy infection, but it can be challenging to find the parasitic form in mounts or smears in cases of light infection. As a result, the study used the concentration methods of floatation and sedimentation.

3.5.2.1 Floatation Technique

This approach relied on the use of a saturated salt solution. The parasite with a density lower than saturated salt is floated using the flotation method (Arora and Arora, 2015). A 15 ml centrifuge tube containing 2 g of the material was filtered, combined with regular saline, and centrifuged for 5 minutes at 1200 rpm. Only around 4-5 ml of the floatation solution remained in the tube after the supernatant had been disposed of. The particles were re-suspended after a thorough mixing. After adding more concentrated NaCl solution to fill the tube to the full 14 ml, the tube was centrifuged once more for five minutes at 1200 rpm. Concentrated NaCl was added to the tube drop by drop until the top created a convex surface. After covering the tube with a clean coverslip to prevent bubbles, it was left alone for at least ten minutes. After carefully removing the coverslip to prevent the sample from falling out, the coverslip was put over a spotless glass slide. The slide was examined using a compound microscope with and without Lugol's iodine at total magnifications of 100 × and 400 × power.

3.5.2.2 Sedimentation Technique

The parasites with densities greater than the solution's density are revealed using the sedimentation method. It mostly finds trematode eggs, however because they float on the concentration solution, certain nematode eggs and larvae and some cestode eggs are also found with this method (Arora and Arora, 2015).

After a thorough filtering, about 2 g of the sample were combined with regular saline in a 15 ml centrifuge tube. The suspension then centrifuged at 1000 revolutions per minute for 5 minutes (rpm). After discarding the supernatant, the sediment was thoroughly mixed. After that, 3 ml of ethyl acetate and 10 ml of 10% buffered formalin were added to the tube, and it was centrifuged once more. There were four layers: 10% formalin, a plug of debris, ethyl acetate, and sediment. Using a wooden applicator stick, the debris plug was removed, and all of the supernatant fluid was extracted and disposed of. Under a microscope, ethyl acetate produces large bubbles; therefore it was important to remove it before turning the tube upright. If the sediments were excessively dry, add one or two drops of 10% formalin and well mix. A clean slide was used to hold a drop of sediment, which was then covered with a cover slip and examined under a microscope with and without Lugol's iodine (Garcia and Procop, 2016).

3.6 Acid-Fast Staining Technique

A thin smear of 1 to 2 drops of specimen was prepared on the clear and dry glass slide and was dried with gentle heat in room temperature. Then the slide was flooded with Carbolfuchsin stain and heated to steaming for 5 minutes, avoid boiling. Without the additional heat for 5 minute standing period, the slide was washed in the running tap water. Acid- Fast Decolorizer was utilize for 2 minute or until no more stain came off. Washing thoroughly was crucial for false positive results. The slide was washed again and counterstained with Methylene blue for 30 minutes. After the final wash, the slide was air dried and examined under an oil immersion objective (Henriksen and Pohlenz, 1981).

3.7 Identification of the eggs, cysts and larva

Prepared slides were examined under microscope under 10X and 40X respectively. The eggs, cysts and larva were identified by comparing the structure, color and size of eggs, cysts and larva of published articles, journals and books (Soulsby 1982, Zajac & Conboy 2012).

3.8 Measurement of eggs, cysts and larva

The size of the eggs and cysts was determined using calibrated ocular and stage micrometers. The length, breadth and diameter of the parasite egg, cyst and larva were measured by calibrated ocular and stage micrometer. They were measure with the calibration factors (C.F).

C.F= (No. of S.D/ No. of O.D) × 10 µm

C.F for 10x= 10 µm

C.F for 40x= 2.6 µm

3.9 Data analysis

The data was recorded based on a laboratory experiment. The purpose of the study was to identify various intestinal parasites. MS-Excel 2007 was used to analyze the data, and the Chi-square test from Statistical Package for Social Sciences (SPSS) version 20 was used for statistical analysis. In each case, the 95% confidence interval (CI) and $P > 0.05$ were used to determine whether a difference was statistically insignificant. Prevalence was calculated using a percentage. The prevalence rate (PR) was computed using the following formula:

PR of parasite= number of stool sample found positive with the parasite /total number of stool samples analyzed *100

4. RESULTS

During the study period, a total of 200 fecal samples of turkey (100 males and 100 females) were taken from farm in Nagarjun municipality of Kathmandu district and examined by using saline wet mount method, floatation method and sedimentation method.

4.1 General prevalence of GI parasites

Out of 200 fecal samples examined, 147 fecal samples were positive for one or more specific GI parasites, showing 73.5% prevalence of parasitic infection whereas 26.5% fecal samples were negative.

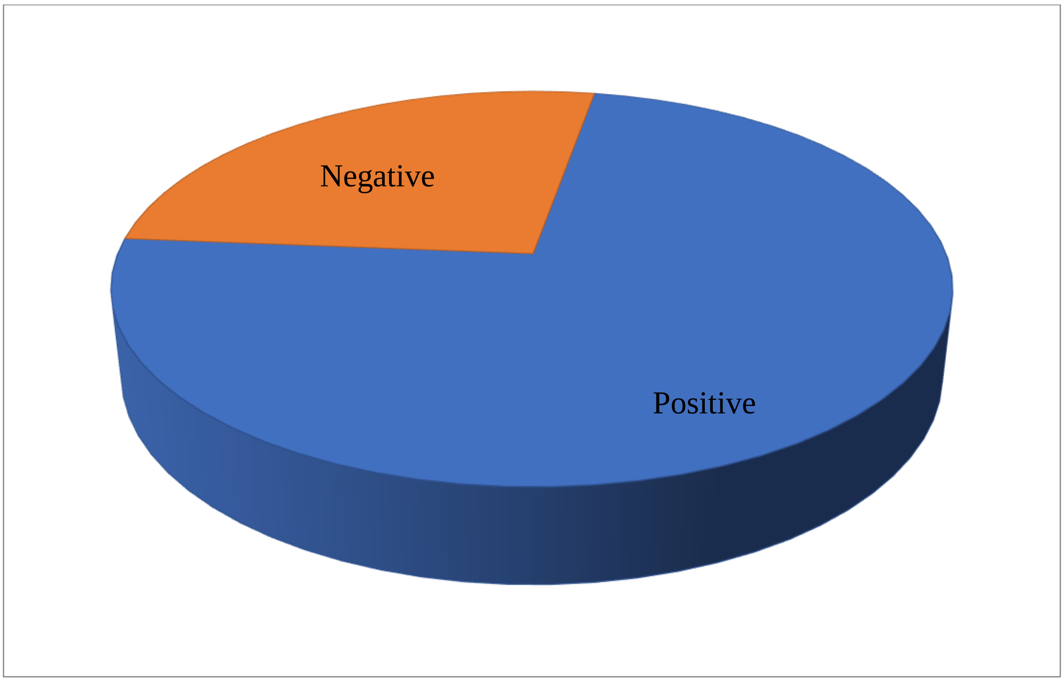

Figure 2: General prevalence of GI parasites

4.2 Prevalence of specific GI parasites

Out of 200 total samples, a total of three nematodes (60%), two cestodes (16.5%) and one protozoan (22.5%) parasitic eggs were isolated and identified. The prevalence rate of *Eimeria* sp. was 45 (22.5%) followed by five helminths: *Ascaridia* sp. 52 (26%), *Capillaria* sp. 35 (17.5%), *Heterakis* sp. 33 (16.5%), *Raillietina* sp. 13 (6.5%), *and Hymenolepis* sp. 20 (10%).

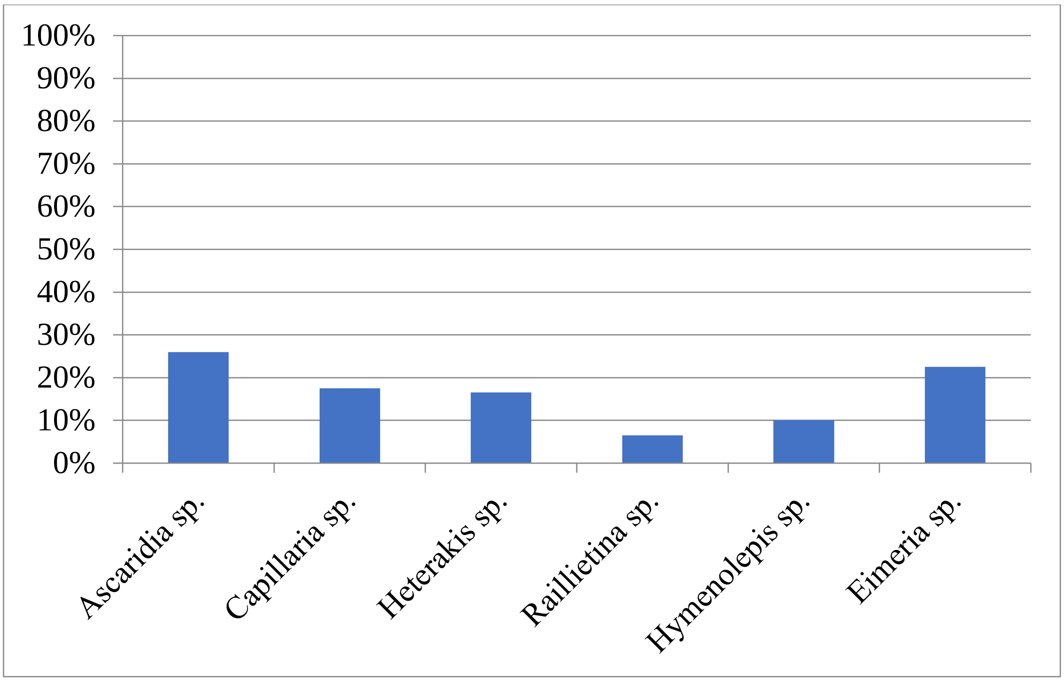

Figure 3: Prevalence of specific of GI parasites

4.3 Sex-wise prevalence

The sex-wise prevalence of GI parasite in turkey was categorized into male and female. Out of 200 samples, 100 were males and 100 were females. Sex-wise 67% males and 80% females were found to be infected with one or more parasite. Statistically, the difference in sex-wise prevalence of GI parasites in turkey was found to be significant ($\chi 2=4.338$; $P>0.05$).

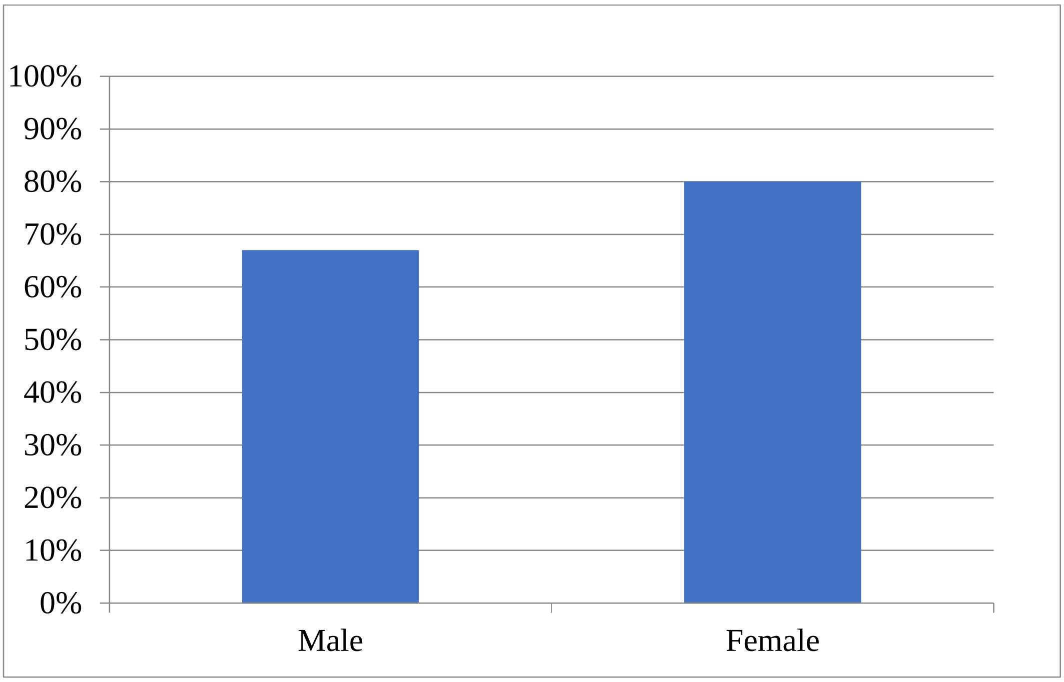

Figure 4: Sex-wise prevalence of GI parasites

4.3.1 Sex-wise comparative prevalence of specific GI parasites in turkey

The study showed the effects of sex-wise comparative prevalence of specific gastrointestinal parasites in turkey and there was no statistical difference of the prevalence of intestinal parasites among sexes ($\chi 2= 9.698$; $P>0.05$) (Figure 5). *Ascaridia* sp. was found to be more prevalent in female (38%) than in male (14%). The highest prevalence of *Capillaria* sp. was found in male (20%) than in female (15%). Incidence of *Heterakis* sp. was found more in female (21%) than in male (12%). Likewise, the prevalence of two cestodes i.e. *Raillietina* sp. and *Hymenolepis* sp. was found more in female (8% and 13%) than in male (5% and 7%) respectively. The prevalence of protozoan parasite i.e. *Eimeria* sp. was found more in female (32%) than in male (13%).

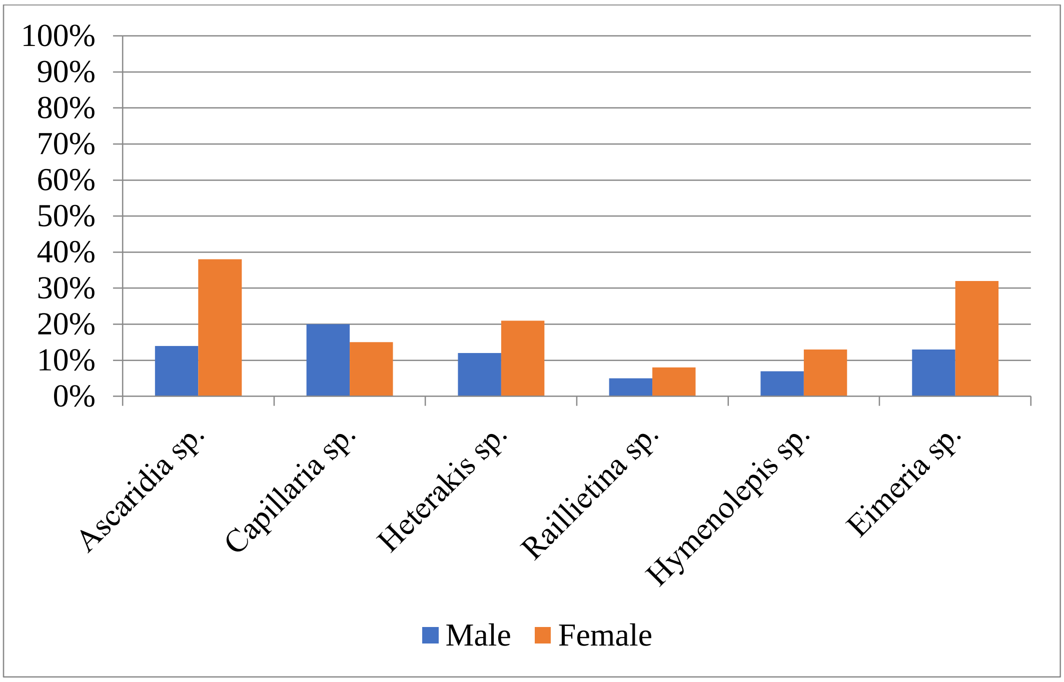

Figure 5: Sex-wise prevalence of specific GI parasites

4.4 Season-wise prevalence of GI parasites

The season-wise prevalence of GI parasite in turkey was categorized into summer and winter. Out of 200 samples, 100 were collected in summer and 100 were in winter. Seasonal differences in prevalence of GI parasite in turkeys have shown in the figure 6. The prevalence of GI parasites was the highest during summer (78%) and lowest in winter (69%). The chi-square test indicated that there was no statistical significant difference in season-wise prevalence of GI parasite in turkey ($\chi 2= 2.079$; $P>0.05$).

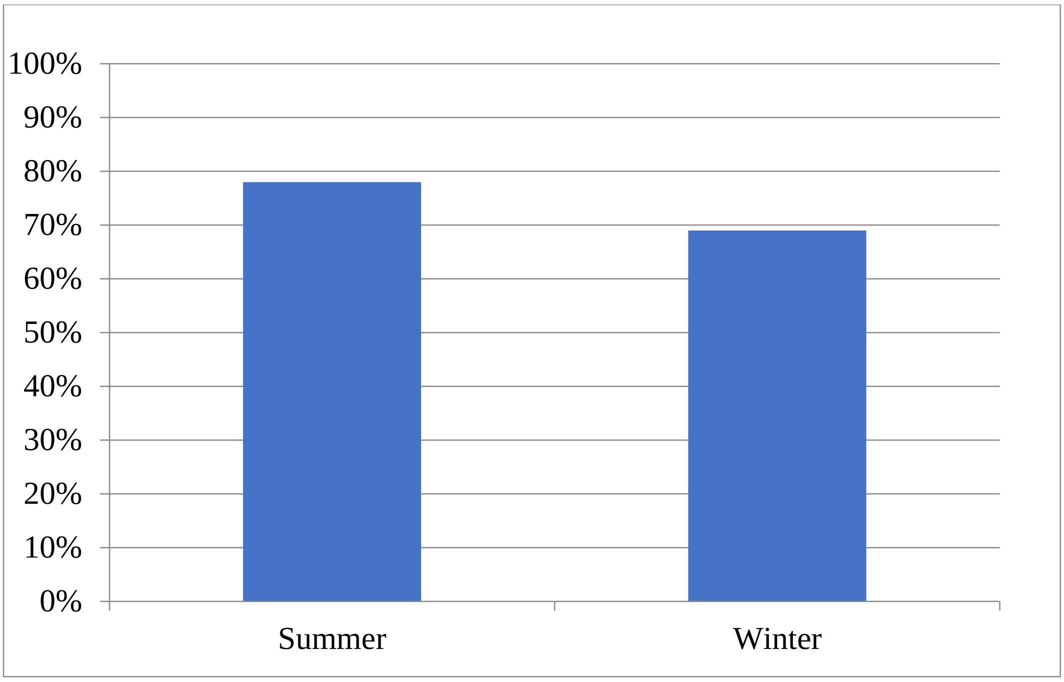

Figure 6: Season-wise prevalence of GI parasites

4.4.1 Season-wise comparative prevalence of specific GI parasites in turkey

A total of 200 samples were collected from the study area (Nagarjun turkey farm) for fecal examination. In summer season, the higher incidence of *Eimeria* sp. (26%) was detected than in winter season (19%). Similarly, *Ascaridia* sp. was found to be more prevalent in summer (32%) than in winter (20%), *Capillaria* sp. was also found more in summer (20%) than in winter (15%). Likewise, *Heterakis* sp. was found to be more prevalent in summer (21%) compare to winter (12%). Additionally, two cestodes i.e. *Raillietina* sp. and *Hymenolepis* sp. were also shown to be more prevalent in summer (8% and 11%) than winter (5% and 9%) respectively. Statistically, the difference in season-wise prevalence of specific GI parasites in turkey was found to be insignificant ($\chi2= 0.650$; $P>0.05$).

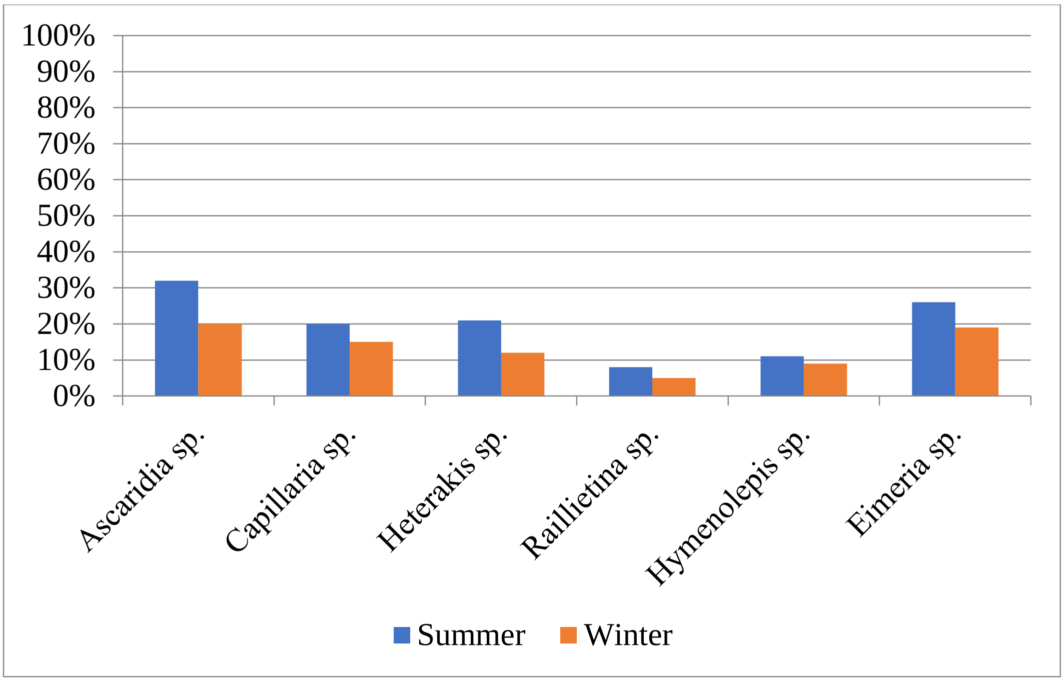

Figure 7: Season-wise prevalence of specific GI parasites

4.5 Types of infection

Out of 200 samples, the higher prevalence was of single infection 89 (44.5%) than mixed infections (double or triple infections) 58 (29%). Statistically, the differences in the prevalence of single and mixed infection were found to be insignificant ($\chi 2= 0.250$; $P>0.05$).

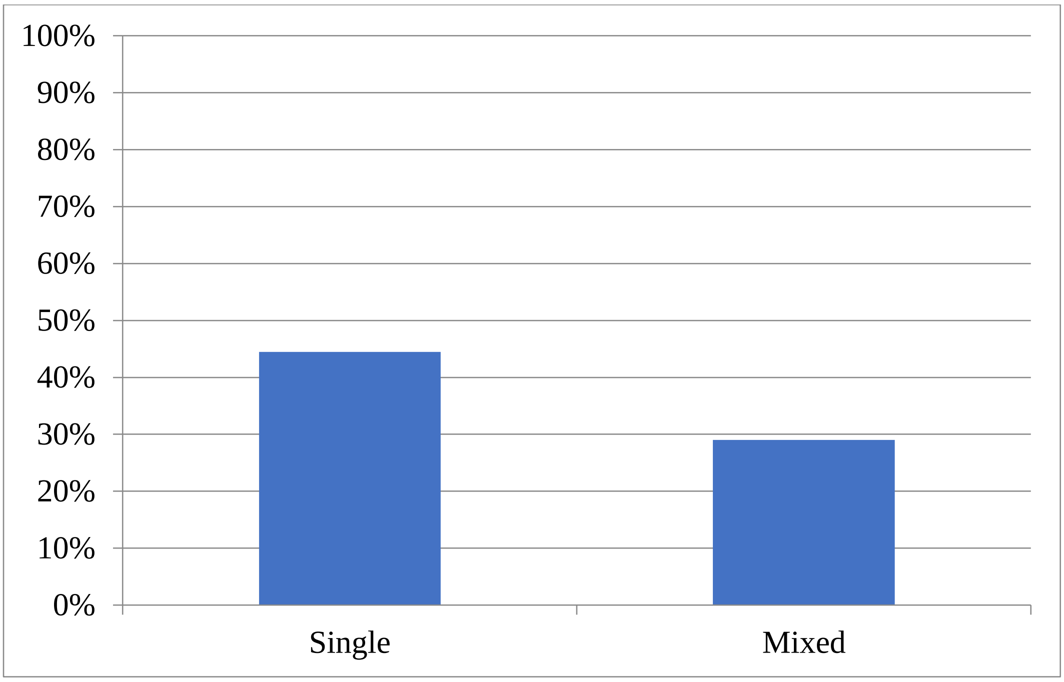

Figure 8: Prevalence of single and mixed infections

4.5 Health care

Health care related to GI parasites for the turkeys such as fecal examination, routine deworming, or other treatment was not found from the Nagarjun turkey farm and District Livestock Services Office, Lalitpur.

4.6 Eggs and cysts of GI parasites in turkey under (10X*40X) electronic microscope

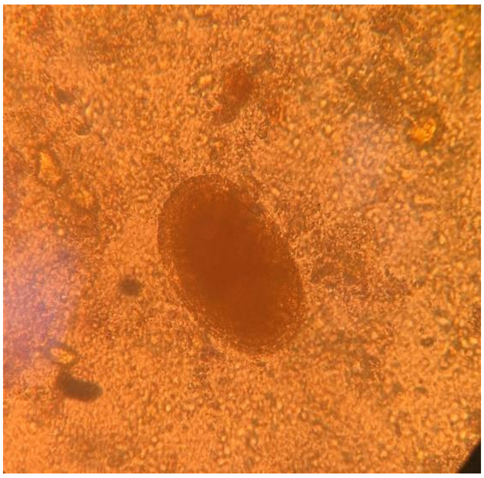

Photo 1: Egg of *Eimeria* sp. (20.60 µm) **Photo 2:** Egg of *Ascaridia* sp. (72.52 X 35 µm)

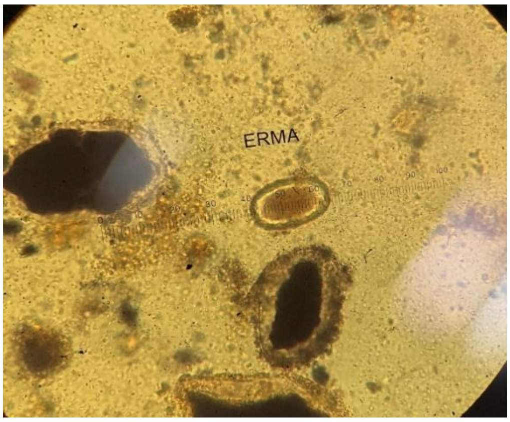

Photo 3: Egg of *Capillaria* sp. (55.3 X 15µm) **Photo 4:** Egg of *Heterakis* sp. (62.22 X40µm)

Photo 5: Egg of *Raillietina* sp. (68 X 25µm) **Photo 6:** Egg of *Hymenolepis* sp. (66.8 X 42 µm)

5. DISCUSSION

Poultry has become one of the important sources of national income in many countries in the world as it provides high nutritional value and other economic benefits (Udoh et al. 2014). The amount of parasites in domestic fowl is greatly influenced by the climate (Husby et al. 2011). In the poultry sector, parasite diseases are frequently neglected in addition to viral and bacterial diseases. Most protozoan parasites, including coccidian parasites, can result in significant loss because of their high mortality and morbidity rates (Fatoba & Adeleke 2018). Similarly, because they slow down the fowl' growth, helminth infections can also result in significant losses in poultry production, both directly and indirectly (Ashenafi & Eshetu 2004). The likelihood of the turkeys carrying more gastrointestinal parasites has grown due to the extensive backyard farming of these birds. Also, a significant amount of human and animal waste is dumped into the soil every day, contaminating it with harmful organisms in their infective stages, which the birds readily ingest when grazing (Audu et al. 2004).

The overall prevalence of infection with gastrointestinal parasites recorded in this study was 73.5%. This is in relation to the 74% reported by (NIPU 2019) in Dhaka, Bangalore and slightly above 75% reported by (ShahrokhRanjbarBahadory et al. 2014) in Iran and 77.3% by (Montes-Vergara et al. 2021) in Colombia. Comparatively, higher occurrence was also reported in Iran (90.8%) and Nigeria (95% and 95.65%) (Jayentakumar Singh & Mohilal 2017, Jegede et al. 2019, Ola-Fadunsin et al. 2019). However, the lower occurrence than this study was reported in Egypt (6%) and Florida (14.0%); (Mohamed El-Dakhly et al. 2016) and (Hon et al. 1975). In this study, turkey farm were practicing a free ranging system and did not have veterinary care and which may have contributed to a higher incidence of gastrointestinal parasites (Permin & Hansen 1998).

The study showed 76.5% among all samples examined were infected by one or more species of helminth parasites which found higher than the protozoan parasitic infection (22.5%), agrees with the work of (Udoh et al. 2014) who found 35.20% infection of domestic turkey in in Kaduna metropolis, northern Nigeria. The present study is also more or less similar to the report of other worker who reported the prevalence rate of 71% (Jegede et al. 2019), 60.00% (Opara et al. 2014) and 74% (Nipu 2019). In the present study five species of helminth were identified comprising three nematodes and two cestodes compared to eight species of helminth identified by (Udoh et al. 2014) comprising of four nematodes and four cestodes, seven species by (Sadaf et al. 2021) comprising six nematodes and one cestode, six species by (Farhang 2012) comprising of four nematodes and two cestodes and four species by (ShahrokhRanjbarBahadory et al. 2014) comprising two nematodes and two cestodes. The study found that many helminth parasites, particularly nematodes, had infected the turkeys. This might be because parasites are thought to be more common in tropical nations, considering the local environment and climate tend to promote the growth of parasites. Parasites the turkey can acquire depend on both its management system and the lifespan of the parasites. Because of their increased exposure to larger areas of land and different, intermediate hosts of parasites, turkeys raised extensively on free range tend to become infected with a wide variety of parasites with direct life cycles (Fabiyi 1972). Among the 147 positive cases, three nematode species (*Ascaridia* spp., *Capillaria* spp., Heterakis spp.), two cestode species (*Raillietina* spp., *Hymenolepis* spp.), and one protozoan species (*Eimeria* spp.), accounting for 60%, 16.5%, and 22.5% of the total, were identified. Similar findings of more nematodes over cestodes were reported in Nigeria (Jegede et al. 2019, Assam et al. 2020), Iran (Farhang 2012, Shahrokh Ranjbar Bahadory et al. 2014) and Colombia (Montes-Vergara et al. 2021). A high incidence of the infective stage and intermediate hosts of the parasites, such as beetles, ants, earthworms, and snails, which are part of the diet of turkey and aid in the indirect lifecycle of nematodes, may be the cause of the high prevalence of nematode infections observed in the farm. Most of the nematode spp. does not require intermediate hosts for infestation within environment. Moreover, adult nematodes produce a large number of eggs per day, some of which can remain viable for up to a year. As a result, domestic poultry regularly consumes fertile eggs from the contaminated environment's bird droppings (Permin & Hansen 1998). Thus, excessive fertility and their lack of sanitation may be key sources of nematodes infection.

Similar, comparatively high prevalence rate of *Ascaridia* sp. over *Heterakis* sp. has been reported from turkeys of Nigeria (Udoh et al. 2014, Dauda et al. 2016, Ola-Fadunsin et al. 2019), Iran (Farhang 2012), Pakistan (Sadaf et al. 2021) and Colombia (Montes-Vergara et al. 2021). This finding strongly showed that the most prevalent and significant helminth infection of poultry is *Ascaridia* sp. The increased prevalence of *Ascaridia* sp. is consistent with other studies that identify this species as the most prevalent and significant helminth infection in poultry (Cervantes-Rivera et al. 2016). *Ascaridia galli* eggs have thick shells that protect them from desiccation and may increase their chances of encountering a new host while they are still living in the environment. By sharing the host's nutrition, *Ascaridia galli* has a severe negative impact on the health of turkeys. This results in stunted growth and decreased egg and meat output (Ashenafi & Eshetu 2004). Where proper managerial practices are not in place, feed and water sources of birds can easily be contaminated because farm handlers can bring the eggs of these parasites from other sources to the farm site. In the deep litter system, the eggs can likely remain infectious for years depending on the temperature, humidity, pH, and ammonium concentration. Although *Heterakis gallinarum* was found in turkeys and chickens with a significantly lesser incidence than *Ascaridia galli*, its pathology and its function as a carrier of the serious pathogen *Histomonas meleagridis* should be strongly regarded (Ashenafi & Eshetu 2004). Moreover, *H. gallinarum* has the ability to transmit the protozoan *Histomonas meleagridis* to birds (Dimitrov et al. 2015). This study reported *Capillaria* spp. (17.5%) which found to be commonly infecting poultry birds. Different species of *Capillaria* in several poultry were recorded by (Castle & Christensen 1984, Rabbi et al. 2006, Muhairwa et al. 2007, Kaufmann et al. 2011, Udoh et al. 2014, Dauda et al. 2016, Jegede et al. 2019, Montes-Vergara et al. 2021, Sadaf et al. 2021). Domestic birds may become infected by *Capillaria* species in areas where there are many eggs in the soil or the litter (Permin & Hansen 1998).

In the present study two species of cestode i.e. *Raillietina* sp., *Hymenolepis* sp. was recorded with the prevalence of 16.5%. The prevalence rate was somewhat similar to the finding of (ShahrokhRanjbarBahadory et al. 2014) who reported *Raillietina tetragona (*8%)*, Raillietina echinobothrida* (8%) among the infected turkeys. Infection with *Raillietina* sp. in turkey with lower prevalence has also been reported from United States (Maxfield et al. 1963), from Nigeria (Udoh et al. 2014, Jegede et al. 2019). *Choanotaenia infundibulum* (22.6%) and *Hymenolepis* spp. (76.1%) were the two species of cestodes identified by (Montes-Vergara et al. 2021) from sick turkeys in the Savanna region, Department of Sucre, Colombia. The availability of intermediate hosts, the degree of host resistance, ecological traits and the different seasons these experiments were done in could all be contributing factors.

Meanwhile *Eimeria* spp. infection in birds can lead to coccidiosis, the most common disease caused by a protozoan that is becoming a problem for poultry globally (El-Shahawy 2010). The overall occurrence of *Eimeria* spp. infestation in this study was 22.5%. This occurrence was in-line with the occurrence in Nigeria (Udoh et al. 2014), this occurrence was lower than that in Bukurus-Jos metropolis, Nigeria (Dauda et al. 2016), Iran (Jayentakumar Singh & Mohilal 2017), Colombia (Montes-Vergara et al. 2021). These differences could be the result of various management strategies and coccidiostats usage as preventative measures in the study areas. The high prevalence found in this study may be related to the large range of diet that domestic turkeys naturally consume, which puts them at risk for parasite diseases. Food like seeds, forages, and kitchen trash expose them to the intermediate hosts of several pathogens, such as cockroaches, beetles, grasshoppers, earthworms, etc. (Naem & Eskandari 2005). Moreover, oocyst walls are resistant to external condition and *Eimeria* spp. have a shorter life cycle than helminths. The gastrointestinal fecal samples of turkey had no trace of trematodes, which was a unique aspect of this investigation. This might be as a result of the complicated trematode life cycle, which necessitates at least one intermediate host, which might share the same habitat as the turkeys. The lack of these environments contributes to the trematode lifecycle being disrupted, which slows the spread of the worms (Adang et al. 2008).

According to (Lawal et al. 2016, Jegede et al. 2019) both male and female were infected with helminth parasites and *Eimeria* species. We found that females were more susceptible to helminth infections and *Eimeria* infections than males, despite the fact that these parasite diseases are not sex biased. In the present study, the overall prevalence of the parasites between females (79%) and males (68%), which showed some degree of preference for female birds, as higher infection rate was observed in female than male turkey. Statistically, difference in prevalence of GI parasitic infection between male and female turkey was found to be insignificant ($\chi2=3.106$; $P>0.05$). It might be because of similar climate, food resources and environment. According to our findings, (Ola-Fadunsin et al. 2019) reported a higher prevalence of GI parasites in female than male in their study on different avian species. (Jegede et al. 2019) reported higher prevalence of female than male studied in local and exotic breeds of turkeys. (Matur et al. 2010) reported female chickens were more prone to GI helminths than male in their study on exotic and local chickens. A higher prevalence of *Eimeria* species in exotic female chickens than its male studied by (Pam et al. 2015). (Atsanda et al. 2015) described that female guinea fowls were more infected with GI helminths than male. The investigation showed that female birds recorded more gastrointestinal helminths and protozoa than the males. This might be a chance or associated to their eating habits, as females are known to be more voracious eaters than males, who are mostly selective, especially while producing eggs (Sonaiya 1990, Matur et al. 2010). Despite the fact that some zoonotic parasites can be detected in turkeys, the ones found in this study were not zoonotic and may not put handlers at risk for infection. In contrast to the study, higher prevalence of GI helminths in male chicken and guinea fowls than female ones was reported by (Attah et al. 2013). (Dauda et al. 2016) documented higher prevalence of gastrointestinal nematodes in male than female turkeys. Female avian species are more likely to be infected with helminth parasites due to frequent scratching of the ground as they find food for their chicks and in the process pick up helminth eggs, sporulated *Eimeria* oocysts. Some helminth parasites use earthworms, beetles, flies, grasshoppers, and cockroaches as intermediate hosts (Taylor et al. 2007, Radfar et al. 2012). Because the development of parasites in the host is largely dependent on stress factors and the immune system of the host, it is possible that the higher prevalence recorded in females was also caused by the physiological stress of brooding (Taylor et al. 2007).

In the present study, the prevalence of gastrointestinal parasites is found in turkeys but their findings are correlated with different types of poultry birds. Overall prevalence of gastrointestinal parasitic infection in summer and winter seasons was 78% and 69% respectively. Insignificant ($p= 0.149$) relationship between the seasonality and prevalence of gastrointestinal parasites was observed ($\chi2= 2.079$), summer season being more favorable for the prevalence of parasites. The results are in accordance with the findings of (Naphade & Chaudhari 2013) who reported the seasonal prevalence of helminth parasites in Marathwada region of Maharashtra during summer season was higher than winter season. According to (Magwisha et al. 2002), climatic factors (temperature and humidity) may change the parasite population dynamics, leading to differences in the frequency and severity of helminth infections. (Shahin et al. 2011) reported the highest incidence of cestodes occurred during summer, autumn and lowest in winter and spring season. Several insects that may serve as helminth infection vectors are, to some extent, favored by high temperatures, which is the most likely cause of the seasonal shifts. These elements could account for the high variety and spread of cestode and nematode species found in poultry (Permin et al. 1997, Hørning et al. 2003). According to this study, a rise in the population of its intermediate hosts may account for its higher intensity in the summer. Higher infection of *A. galli* in summer season was observed in a semi-arid area of Kenya in local chickens (Mungube et al. 2008), in local and exotic chickens in district of Hyderabad, Pakistan (Soomro et al. 2010) and in free range chicken in rural district of Zimbabwe (Percy et al. 2012). For the development of eggs into infective stages, moist and humid conditions are required. As earthworms, paratenic hosts of *A. galli* and their numbers rise in the summer, the parasite's activity may also increase. However, a research by (Magwisha et al. 2002) discovered that the intensity of *A. galli* was constant throughout the year. Similarly, the prevalence of coccidian parasites in broilers in Kashmir valley was found to be highest during summer and lowest during winter season (Ahad et al. 2015). In the present study, the prevalence of coccidian parasites was reported to be significantly higher in summer than in winter, which might be attributed to unfavorable temperatures that are unsuitable for sporulation of parasitic eggs (Ahad et al. 2015). Since the turkeys were free-ranging and had access to environmental infective stages and intermediate hosts like beetles, earthworms, ants, and other creatures that are intermediate hosts for helminth parasites, the high prevalence rate of gastrointestinal parasitism in turkey in the current study may be explained by this.

In the present study, the single parasitic infection was found to be more common in turkey. This finding is kind of similar to the finding reported by (Dauda et al. 2016, Ola-Fadunsin et al. 2019) i.e. 47.50% had single infection followed by mixed infection (26%). In this study, mixed infections of two or more parasite species per turkey were common, while their prevalence was lower than that of single infections. This result may be explained by the food preferences at a specific period, which has in great extent determined the establishment of a mixed or single infection. The prevalence of mixed infections has increased due to the ability of two or more parasites to coexist in the same host, but as the number of parasites per host increases, the prevalence decreases because the parasites cannot tolerate one another (Smyth 1976).

6. CONCLUSIONS AND RECOMMENDATIONS

6.1 CONCLUSIONS

From this study, it is concluded that turkey of Nagarjun turkey farm, Kathmandu, Nepal were infected with different GI protozoan and helminth parasites. The most prevalent parasites identified were *Ascaridia* sp., *Eimeria* sp., *Capillaria* sp., *Heterakis* sp., *Hymenolepis* sp. and *Raillietina* sp. interestingly; no trematodes were detected in this particular study. The prevalence of gastrointestinal parasites among the turkeys was found to be high, with significant differences observed on sex and with no significance difference observed on season. It was noted that there were single parasite infections in most of the turkeys. The study highlighted the ineffective management of litter on the farm and the lack of awareness about poultry among the owner. Therefore effective medication for controlling the parasitic infection and further studies need to be designed for the health and conservation of turkeys.

6.2 RECOMMENDATIONS

- In order to effectively control GI parasites, turkey health care programs like routine fecal examination and deworming should be carried out.
- On a species-level, parasites could be further identifying.
- Age-wise study can be studied.

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ANNEX

Photo Plates

Photo 7: Grazing of turkey in the farm

Photo 8: Collection of stool samples

Photo 9: Stool sample preservation

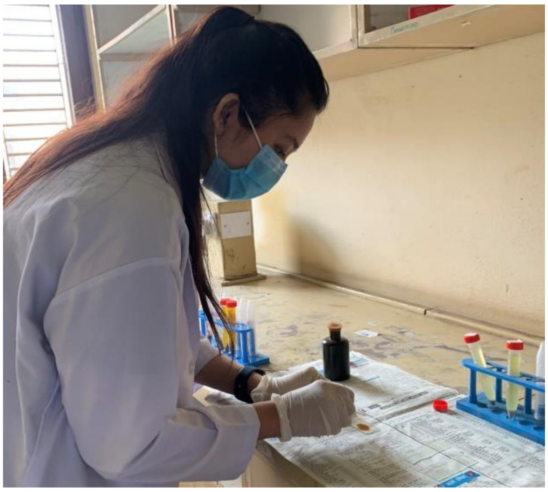

Photo 10: Slide preparation of stool sample

Photo 11: Microscopic fecal observation