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MSC THESIS

INVESTIGATION ON TREMATODES OF RUMINANTS: CURRENT STATUS, SNAIL INTERMEDIATE HOSTS, AND COMMUNITY KNOWLEDGE IN AND AROUND BISHOFTU, ETHIOPIA

DEPARTMENT OF PATHOLOGY AND PARASITOLOGY

BY

ABRAHAM DIRIBA KENEA

JUNE, 2024

BISHOFTU, ETHIOPI

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INVESTIGATION ON TREMATODES OF RUMINANTS: CURRENT STATUS, SNAIL INTERMEDIATE HOSTS, AND COMMUNITY KNOWLEDGE IN AND AROUND BISHOFTU, ETHIOPIA

A Thesis submitted to the College of Veterinary Medicine and Agriculture of Addis Ababa University in partial fulfilment of the requirements for the degree of Master of Science in Veterinary Parasitology

BY

ABRAHAM DIRIBA KENEA

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DR. GEREMEW HAILE (DVM, MVSC, ASSI. PROF)

JUNE, 2024

BISHOFTU, ETHIOPIA

Approval Sheet

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As MSc research advisor, I certify that I have read and evaluated the thesis prepared under my guidance by: **Abraham Diriba Kenea**: Entitled "**Investigation on Trematodes of Ruminants: Current status, Snail intermediate hosts, and Community knowledge in and around Bishoftu, Ethiopia"** and recommended that it be accepted as fulfilling the thesis requirement for the degree of: **Masters of Science in Veterinary Parasitology**.

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DEDICATION

I dedicate this MSc paper to my beloved wife, Yadate Mengasha. Throughout this journey, your unwavering support, love, and encouragement have been my greatest sources of strength. Your patience, understanding, and belief in me have inspired me to persevere through the challenges and obstacles encountered along the way. Your presence has brought joy and meaning to every moment of this academic pursuit. This achievement is as much yours as it is mine, and I am deeply grateful for your constant presence by my side. Thank you for being my rock, my confidante, and my greatest cheerleader. This paper is dedicated to you with all my love and gratitude.

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LIST OF ABBREVIATIONS

CGS	College of Graduate Studies
CSA	Central Statistical Agency
DAFWA	Department of Agriculture and Food, Western Australia
DNA	Deoxyribo Nucleic Acid
ELISA	Enzyme-Linked Immunosorbent Assay
FGC	Faculty Graduate Council
KAP	Knowledge Attitude and Practice
NMA	National Meteorological Agency
OR	Odds Ratios
PCR	Polymerase Chain Reaction
QGIS	Quantum Geographic Information System
SPSS	Statistical Package for the Social Sciences
StataSE	Stata Special Edition

ABSTRACT

The occurrence of trematode infections in ruminants, facilitated by their snail intermediate hosts, poses a major concern across the diverse agro ecological regions of Ethiopia. A cross-sectional study was conducted in and around Bishoftu town from November 2023 to April 2024 to determine the current prevalence of trematode infections and associated risk factors in ruminants, identify snail intermediate hosts of trematodes and determine their infectivity through the emergence of cercariae, and assess the knowledge, attitudes, and practices (KAP) of farmers regarding the prevention and traditional management of trematode infections. A total of 336 fecal samples were collected from cattle, sheep, and goats and subjected to copromicroscopic examination. 400 snails were collected, identified and examined for cercarial shedding. The copromicroscopic examination revealed an overall prevalence of 11.61% trematode infections in the study animals. Logistic regression analysis showed that risk factors such as species, age, body condition score (BCS), and season had significant associations with overall trematode infections. The identified genera were *Fasciola* (8.93%) and *Paramphistomum* (4.46%), with coinfection of 1.79%. Sheep had higher infection of *Fasciola* compared to goats and cattle, which was statistically significant (OR = 7.713, 95% CI: 2.197-27.076, p = 0.001). Cattle had higher *Paramphistomum* infection than sheep and goat which was statistically significant (OR = 18.403, 95% CI: 2.111-160.375, p = 0.008). Young animals had higher *Paramphistomum* infection compared to old animals (OR = 14.786, 95% CI: 2.295-95.276, p = 0.005). Poor body condition animals were more affected than those with medium and good body condition for both Fasciola (OR = 12.27, 95% CI: 3.097-48.118, p = 0.000) and Paramphistomum (OR = 6.277, 95% CI: 1.147-34.363, p = 0.034). The semi-dry season was associated with higher *Fasciola* infection compared to dry season (OR = 3.265, 95% CI: 1.307-8.155, p = 0.011). The malacology study involved the collection of 400 snails, and revealed *Lymnaea natalensis* (37%), *Physa* spp (21.25%), *Bulinus truncatus* (15.75%), *Lymnaea truncatula* (10%), *Biomphalaria pfeifferi* (9.5%), and *Bulinus africanus* (6.5%). The overall cercarial rate of infection of snails was 7.75%, with the highest rate of infection observed in *Lymnaea natalensis* (13.51%), and followed by *Bulinus truncatus* (11.11%). Rate of infections varied significantly by species ($\chi^2 = 18.76$, p = 0.002), habitat type, with ponds having the highest rate (13.48%) ($\chi^2 = 15.6$, p = 0.004), and season, with higher rates during the semi-dry season (12%) compared to the dry season (3.5%) ($\chi^2 = 10.11$, p = 0.001). The quiestionnaire survey revealed that there was significant associations ($p < 0.05$) between education level and KAP of farmers; gender and age with knowledge of farmers about trematode infections. In conclusion, the study provided important insights into the epidemiology of trematode infections in the study area and strategic deworming to reduce pasture contamination with flukes and snail control is needed.

Key words: *Bishoftu, Epidemiology, Fasciola, Paramphistomum, Ruminant, Snail*

1. INTRODUCTION

Livestock is one of the sub-sectors of agriculture in Ethiopia with the most potential, playing an indispensable role in promoting the national economy of the country (Shapiro *et al*., 2015). Ethiopia has a large number of livestock population, including an estimated 65 million cattle, 39 million sheep, 54 million goats, 49 million poultry (CSA, 2020). However, the profits gained from this vast livestock resource do not match the substantial size, as various diseases continue to undermine productivity and profitability (Alemu *et al.*, 2018). Among the major challenges facing the livestock sector, parasitic diseases, particularly trematode infections, are significant impediment to the sector's growth and development (Abera *et al*., 2021). These parasitic infections not only cause direct production losses but also treatment and control costs, hindering the realization of the full economic potential of Ethiopia's livestock industry (Regassa *et al*., 2019).

Fasciola, *Paramphistomum*, *Schistosoma*, and other less common trematode parasites, such as *Echinostoma*, *Heterophyes*, *Clonorchis*, *Opisthorchis*, and *Paragonimus*, are affecting both animals and humans, causing substantial economic losses globally and within Ethiopia's livestock sector (Colley *et al*., 2022). The global prevalence of *Fasciola* infection in livestock ranges from 10% to 80%, as reported by Charrlier *et al*. (2020), while in Ethiopia; the prevalence report ranged from 8.1% to 56.4% which can also reach 80% recorded in Bahir Dar (Japaro, 2023). Similarly, *Paramphistomum* infections in livestock have a global prevalence of up to 80%, and in Ethiopia, the prevalence can reach 45.83% in cerctain areas (Netsa *et al*., 2022). *Schistosoma* infections in livestock have a global prevalence ranging from 10% to 60% (Lund *et al.*, 2022), and in Ethiopia, the prevalence can be as high as 40% in some regions (Erko *et al*., 2019).

Trematode infections significantly affect livestock health and productivity, leading to substantial economic losses. Infected animals often experience reduced body weight and decreased milk production due to the parasites' consumption of vital nutrients and blood, which results in anemia and reduced feed efficiency (Charlier *et al*., 2020). Chronic infections can cause liver damage, leading to poor growth rates, decreased fertility, and overall weakened immune systems, making animals more susceptible to secondary infections (Rizwan *et al*., 2021). These effects cumulatively result in profit gained for infected animals and increased costs for treatment and management (Mwabonimana *et al*., 2019; Alemu *et al*., 2021). This underscores the importance of investigating the epidemiology of trematodes, their hosts, and the environmental conditions (Xie *et al*., 2023).

Despite trematode infections causing considerable challenges to livestock sectros, the epidemiology of these infections, particularly in wetland areas like Bishoftu, Ethiopia, where suitable habitats for snail intermediate hosts are abundant, remains poorly understood. As the previous studies focus on abattoir based study, there is a lack of comprehensive field-based investigations on the prevalence of trematode infections and their associated risk factors in ruminants in these regions. Moreover, the identification of snail intermediate hosts, their occurrence, and infectivity levels in transmitting these parasites are crucial but had not been thoroughly studied in this context. Additionally, the knowledge, attitudes, and practices of farmers regarding the prevention and management of trematode infections in these areas were not well documented. Addressing these gaps is crucial for developing targeted interventions and control strategies to mitigate the impact of trematode infections on both animal and human health.

General objective

- The general objective was to investigate prevalence of trematode infections in ruminants and distribution of snail intermediate hosts in and around Bishoftu

Specific objectives

- To estimate the current prevalence of trematode infections and associated risk factors in ruminants
- To identify snail intermediate hosts of trematodes and access their infectivity through the emergence of cercariae
- To assess the knowledge, attitudes and practices of the farmers regarding the prevention and traditional management of trematode infection

2. TREMATODE INFECTIONS IN RUMINANTS

2.1. Definition and Taxonomy

The definition of trematodes, also known as flukes, encompasses a group of parasitic flatworms belonging to the phylum Platyhelminthes. These organisms are characterized by their flattened, leaf-shaped bodies and specialized suckers that allow them to attach to and feed on their host's tissues. Trematodes have a complex life cycle that typically involves multiple hosts, including intermediate hosts where larval stages develop, and definitive hosts where adult worms reside (Chai and Jung, 2022). They are known for their ability to cause a variety of diseases in humans, livestock, and wildlife, making them a noteworthy concern in the fields of public health and veterinary medicine. Understanding the definition of trematodes is essential for recognizing their unique characteristics and life cycle strategies, which play a crucial role in their pathogenicity and transmission dynamics (Widdicombe *et al.*, 2024).

The taxonomy of trematodes is a complex and intricate field that involves the classification and categorization of these parasitic flatworms based on their morphological, anatomical, and genetic characteristics. Trematodes are classified within the phylum Platyhelminthes, along with other flatworms, and are further subdivided into the class Trematoda. Within the class Trematoda, trematodes are organized into various orders, families, genera, and species based on their physical features, life cycle patterns, and evolutionary relationships (Pitaksakulrat *et al.*, 2022).

Taxonomic identification of trematodes traditionally relied on morphological traits such as body shape, size, and reproductive structures, as well as host specificity and geographical distribution. However, with the advancement of molecular techniques, including DNA sequencing and phylogenetic analysis, researchers have been able to refine and clarify the taxonomy of trematodes at a molecular level. This has led to the discovery of new species, revisions of existing classifications, and a deeper understanding of the evolutionary history and diversity of trematodes (Robinson and Sotillo, 2022).

Understanding the taxonomy of trematodes is essential for studying their biology, epidemiology, and control strategies. By accurately identifying and classifying trematode species, researchers and public health officials can better assess the risks associated with these parasites and develop targeted interventions to prevent and manage infections in both animals and humans (Pitaksakulrat *et al*., 2022).

2.2. Major Trematodes of Domestic Ruminants

2.2.1. Fasciola

Fasciola species (*Fasciola* spp.), particularly *Fasciola hepatica (F. hepatica)* and *F. gigantica*, are noteworthy parasites of domestic ruminants, causing hepatic damage and economic losses worldwide. The adult *Fasciola*, particularly *F. hepatica* and *F. gigantica*, exhibits a broad, leaf-shaped, dorsoventrally flattened body, with *F. hepatica* typically measuring 20-30 mm in length and 8-15 mm in width, and *F. gigantica* being larger at 25-75 mm in length and around 12 mm in width (Mas-Coma *et al*., 2022). The body is grayish-brown, covered with a tegument that has spines for anchorage to host tissues. Adults have two suckers: an oral sucker located at the anterior end for feeding, and a ventral sucker near the middle of the body for attachment. They are hermaphroditic, containing both male and female reproductive organs, with branched testes in the posterior region and an ovary situated anteriorly. Their digestive system includes a mouth, pharynx, esophagus, and a branched intestine extending laterally (Hodova *et al*., 2018).

The eggs of *Fasciola* are oval-shaped and measure approximately 130-150 micrometers in length and 63-90 micrometers in width. They are yellowish-brown with a distinct operculum (lid) at one end. The shell is smooth with a thin but tough outer layer, and inside, a developing embryo (miracidium) becomes visible as it matures. The cercaria of *Fasciola* is small, around 200-400 micrometers in length, with an oval or pear-shaped body and a long, simple, unbranched tail used for swimming (Jia, 2023).

F. hepatica is prevalent in temperate regions, while *F. gigantica* is more common in tropical and subtropical areas. The pathogenesis of fascioliosis progresses in two distinct phases, influenced by the host species and the developmental stage of the parasite within the liver (Jia, 2023). The initial phase occurs during the migration of the parasite through the liver parenchyma, resulting in liver damage and haemorrhaging. Subsequently, the second phase ensues as the adult flukes engage in hematophagic activities within the bile ducts, causing damage to the mucosa through their cuticular spines (Mas-Coma, *et al.,* 2022).

It features an oral sucker at the anterior end for initial attachment and a ventral sucker on the ventral side for movement and further attachment. The cercaria contains penetration glands that secrete enzymes to facilitate penetration into the snail host. Upon leaving the snail, the cercaria encysts on aquatic vegetation or other surfaces, forming the metacercaria, which is the infective stage for the definitive host (Jia, 2023).

2.2.2. Paramphistomum

There are more than 70 recognized species of paramphistomes that parasitize various animals. The adult *Paramphistomum* flukes are generally conical or pear-shaped and measure between 5-13 mm in length and 2-5 mm in width. They have a distinct anterior oral sucker and a large posterior acetabulum or ventral sucker, which is located at the posterior end of the body. The tegument is smooth, lacking the spines seen in other trematodes like *Fasciola*. They possess both male and female reproductive organs, with the testes usually lobed and located in the posterior half of the body, and the ovary positioned anteriorly to the testes (Rafiq *et al*., 2023). The digestive system comprises an oral sucker, pharynx, esophagus, and a simple, unbranched intestine (Che-Kamaruddin and Isa, 2023).

Paramphistomum eggs are ovoid and measure about 120-180 micrometers in length and 60-80 micrometers in width. They are transparent and have a smooth shell with an operculum at one end (Rafiq *et al*., 2023). The cercaria of *Paramphistomum* is typically oval and measures around 200-300 micrometers in length, with a tail approximately 400-500 micrometers long. It has an oral sucker at the anterior end and a ventral sucker located further down the body. The cercaria uses its tail for swimming and encysts on aquatic vegetation or other substrates to form metacercariae, which are then ingested by the definitive host (Che-Kamaruddin and Isa, 2023).

While adult *Paramphistomum* flukes are generally considered nonpathogenic to their hosts, the invasion of immature worms through the duodenal mucosa can lead to severe enteritis, including necrosis and haemorrhage (Rafiq *et al.,* 2023). This invasion can result in symptoms such as anorexia, polydipsia, unthriftiness, profuse diarrhea, weight loss, and mortality in the definitive host (Buddhachat *et al.,* 2023). Paramphistomosis can lead to substantial economic losses in affected herds, especially in areas with high prevalence (Che-Kamaruddin and Isa, 2023).

2.2.3. Schistosoma

The adult *Schistosoma* species, also known as blood flukes, are elongated and cylindrical, unlike the flat, leaf-shaped bodies of other trematodes. Males are generally shorter and stouter, measuring around 10-20 mm in length, while females are longer and more slender, measuring 20-30 mm (Jain and Rana, 2024). They possess two suckers: an oral sucker at the anterior end and a ventral sucker located just below it. The tegument is covered with tiny spines, which help in attachment to the host's blood vessels (Paul, 2024).

Schistosoma eggs are characteristically oval and have a size range of 100-170 micrometers in length, depending on the species. They possess a distinct spine, which can be terminal or lateral, aiding in tissue penetration and migration (Jain and Rana, 2024). The cercariae of *Schistosoma* are fork-tailed and measure about 200-300 micrometers in length. They have an anterior oral sucker and a ventral sucker, which are used for attachment to the host skin. Cercariae are highly motile and are adapted for penetrating the skin of the definitive host directly, where they transform into schistosomula and migrate to the blood vessels to mature (Paul, 2024).

Schistosoma, a genus of parasitic trematodes commonly known as blood flukes, infects both humans and animals. While less prevalent in ruminants compared to humans, *Schistosoma* infections in animals can still occur and may lead to clinical manifestations such as anemia, weight loss, and reduced productivity. They primarily inhabit blood vessels in various parts of the body, including the alimentary canal, nasal veins, and bladder (Lund *et al.,* 2022). Their clinical impact is associated with the passage of spined eggs through gut tissues, causing damage during migration and egg-induced irritation in the intestine and other organs (Jain and Rana, 2024). Additionally, the worms' blood-feeding behavior and access to vesical veins can lead to haematuria, further complicating the infection's clinical presentation.

Several other trematodes besides *Fasciola, Paramphistomum,* and *Schistosoma* can infect domestic ruminants, including *Dicrocoelium dendriticum* (lancet liver fluke), *Calicophoron daubneyi* (rumen fluke), *Eurytrema pancreaticum* (pancreatic fluke), and *Gastrothylax spp*. These less prevalent species can still cause clinical disease, affecting animal health and productivity. *D. dendriticum* can lead to liver damage, affecting production performance, with a unique life cycle involving terrestrial snails and ants as intermediate hosts. *C. daubneyi*, a rumen fluke, has gained attention due to increasing prevalence and economic losses in affected herds (Busin *et al.,* 2023). *E. pancreaticum* infects the pancreas and bile ducts, causing pancreatic damage and potential digestive disturbances. *Gastrothylax* spp. can infect the gastrointestinal tract, with varying clinical significance based on species and geographical location (Chai and Jung, 2024).

2.3. Zoonotic Importance of Trematodes

Trematodes, commonly referred to as flukes, are a diverse group of parasitic worms that hold substential zoonotic importance, presenting risks to both animal and human health. In ruminants, various trematode infections raise public health concerns due to their potential for zoonotic transmission (Mohamed *et al.,* 2024). Notably, *F. hepatica* and *F. gigantica*, the causative agents of fascioliasis, exemplify this concern, as they can induce considerable disease in both humans and ruminants. Infection occurs through the consumption of metacercariae-contaminated vegetation or water, leading to the migration of flukes to the host's liver and subsequent hepatobiliary diseases.

Another concerning group is the genus *Schistosoma*, whose species are responsible for schistosomiasis. Of particular zoonotic relevance are *Schistosoma japonicum* and *Schistosoma mekongi* (Chai and Jung, 2022).

In addition to these, other trematodes also pose zoonotic risks. For instance, *Paragonimus* species, which cause paragonimiasis, can infect both humans and animals, particularly through the consumption of undercooked or raw crustaceans (Robinson and Sotillo, 2022). Likewise, *Echinostoma* species, known to cause echinostomiasis, have zoonotic potential, with human infections linked to the consumption of raw or undercooked fish, amphibians, or vegetables contaminated with parasite larvae. *Dicrocoelium dendriticum* is recognized for its indirect transmission, which involves ant intermediate hosts that may come into contact with human food, leading to a condition known as dicrocoeliasis. *Eurytrema pancreaticum*, the pancreatic fluke, is known to infect ruminants and has been reported in human cases, indicating its zoonotic significance (Sharma *et al.*, 2020).

2.4. Economic Importance of Trematodes

Trematode infections impose substantial economic losses for domestic ruminant's worldwide. Global economic losses due to trematode infections in domestic ruminants are to range from 1.5 billion to 3 billion annually, considering direct and indirect costs. These losses encompass direct impacts on animal health and productivity, as well as indirect costs related to treatment, control measures, and reduced market value of infected animals (Tidman *et al.*, 2023).

In specific regions such as Europe and North America, the economic impact of trematode diseases on livestock production is estimated to be around 200 million to 500 million per year. The cost of treatment and control measures for trematode infections in livestock operations can account for up to 10-20% of total production costs, further adding to the economic impact (Tidman *et al.*, 2023). Indirect losses, such as reduced market value of infected animals and decreased reproductive performance, can contribute to the overall economic impact of trematode diseases, with estimates ranging from 50 to 100 per infected animal (Hammoud *et al.*, 2022).

Furthermore, the zoonotic potential of some trematode species, such as *F. hepatica*, underscores the interconnected nature of the economic impact of these parasites. Human infections can result in health issues and additional healthcare costs, adding to the overall economic impacts of trematode diseases (Tidman *et al*., 2023).

2.5. Epidemiology of Trematodes

2.5.1. Geographical distribution

Trematodes, or flukes, are parasitic flatworms that exhibit a wide geographical distribution, with different species being prevalent in specific regions around the world (Kathleen and Sanzo, 2022; Tidman *et al.,* 2023). The distribution of trematodes is influenced by various factors, including environmental conditions, host availability, and human activities. Understanding the global distribution of trematodes is essential for assessing the burden of disease, implementing control measures, and predicting potential spread to new areas (Chai and Jung, 2019).

Tropical and subtropical regions are prime areas for trematode infections due to the presence of suitable intermediate hosts (snails) and warm, humid climates that promote parasite development. In regions such as sub-Saharan Africa, Southeast Asia, and parts of South America, trematode infections are endemic and pose considerable health risks to humans and animals. Trematode such as *Schistosoma* and *Fasciola* are particularly prevalent in these regions, causing diseases such as schistosomiasis and fascioliasis (Kathleen and Sanzo, 2022).

In temperate regions, trematode infections are also common, though with different species and transmission dynamics. In areas of Europe, North America, and parts of Asia, trematodes such as *F. hepatica* and *D. dendriticum* are prevalent in livestock and wildlife populations. The seasonal variation in temperature and rainfall in temperate regions can influence the transmission of trematodes, with peak rate of infections occurring during specific times of the year (Paterson *et al.,* 2024).

Trematodes that have aquatic intermediate hosts are often found in freshwater bodies such as lakes, rivers, and wetlands. These environments provide a conducive habitat for both the parasite and its intermediate hosts, facilitating the completion of the trematode life cycle. Species such as *Schistosoma mansoni* (*S. mansoni*) and *S. haematobium),* which cause schistosomiasis, are commonly found in areas with freshwater snails as intermediate hosts (Hailegebriel *et al.,* 2020).

Urban and peri-urban areas can harbour trematode infections, especially in areas with poor sanitation and water management practices. Contaminated water sources, inadequate hygiene, and close proximity to livestock can increase the risk of trematode transmission to humans and animals in urban environments. In recent years, emerging and re-emerging trematode infections have become a global concern. Factors like climate change, human migration, and changes in land use play significant roles in the spread of trematodes to new areas or the resurgence of infections in previously controlled regions (Jia and Peng, 2022).

2.5.2. Host Range

Trematodes are known for their remarkable ability to infect a wide range of hosts, including mammals, birds, reptiles, and humans. These parasitic flatworms have evolved complex life cycles that often involve multiple host species, each playing a crucial role in the parasite's development and transmission (Mark *et al.,* 2022). The host range of trematodes can vary considerably depending on the species, with some parasites exhibiting a more restricted host specificity, while others can infect a diverse array of hosts (Kathleen and Sanzo, 2022).

In addition to domestic and wild mammals like ruminants, trematodes can also parasitize various aquatic and terrestrial snails, which serve as intermediate hosts in their life cycle (Svinin *et al*., 2023). These intermediate hosts play a critical role in the transmission of trematodes, serving as a site for larval development and multiplication before infecting the definitive host. Certain trematode species have adapted to infect specific host groups, while others demonstrate a broader host range, infecting a wide variety of vertebrates. This versatility in host selection is a testament to the evolutionary success of trematodes as parasitic organisms (Kirill *et al.,* 2010).

Understanding the host range of trematodes is essential for elucidating the dynamics of parasite transmission, identifying potential reservoir hosts, and assessing the risk of transmission to humans and other susceptible species. By studying the interactions between trematodes and their diverse hosts, researchers can gain valuable insights into the ecological and evolutionary factors that shape parasite-host relationships and contribute to the persistence of these parasitic infections in natural ecosystems (Mark *et al.,* 2022).

2.5.3. Life cycle

The life cycle of digenetic trematodes, as illustrated in (Figure 1), begins with the production of eggs by adult trematodes residing in the definitive host's (often a vertebrate) gastrointestinal tract or other organs. These eggs are usually excreted into the environment through the host's feces. Once the eggs are released into water, they hatch and release free-swimming larvae called miracidia. Miracidia are equipped with ciliated epidermis and are capable of actively seeking out and infecting intermediate hosts, such as snails, through penetration of the host's soft tissues (Akramova *et al.,* 2022). Upon infecting the intermediate host, the miracidium transforms into a sporocyst or redia stage within the host's tissues. These stages undergo asexual reproduction, giving rise to multiple larvae known as cercariae. Cercariae are the larval stage that emerges from the intermediate host and actively seeks out the definitive host for infection (Reqgi *et al.,* 2022). They often encyst on vegetation or other surfaces, waiting to be ingested by the definitive host. In some trematode species, the cercariae encyst and transform into metacercariae, a dormant stage that remains infective until consumed by the definitive host. Upon ingestion by the definitive host, metacercariae undergo excystation in the host's digestive tract, maturing into adult trematodes. The adults then reproduce sexually, completing the life cycle by producing eggs that are excreted into the environment to start the cycle a new (Nesterenko *et al.,* 2020).

This detailed review outlines the intricate life cycle of trematodes, highlighting the various stages involved from egg production to the completion of the life cycle in the definitive host. The complexity of life cycles in parasitic worms like trematodes is attributed to benefits such as transmission efficiency and increased reproductive success in definitive hosts (Gayo *et al.*, 2020).

2.5.4. Transmission dynamics and influencing factors

Transmission of trematodes to animals and humans can occur through various routes. In the case of animals, including livestock and wildlife, ingestion or penetration of infective stages of the parasite is the primary mode of transmission. Animals grazing on contaminated vegetation or drinking contaminated water can get infection by trematodes, leading to disease distribute within populations. Similarly, humans can acquire infections by consuming raw or undercooked seafood containing infective larvae or through direct contact with contaminated water sources. Understanding these transmission routes is crucial for safeguarding both animal and human health (Robinson and Sotillo, 2022).

Temperature, humidity, and water quality play crucial roles in the survival and distribution of trematode stages and their intermediate hosts. Suitable environmental conditions can promote the proliferation of intermediate hosts, increasing the risk of transmission (Mark *et al.*, 2022). The grazing behavior of animals and dietary habits of humans can influence their exposure to contaminated sources, affecting the likelihood of infection. The compatibility between trematode species and their hosts can influence transmission dynamics. Some hosts may exhibit resistance to infection, while others may serve as efficient reservoirs for the parasite (Stepanenko *et al.*, 2022).

2.5.5. Risk factors for animals

Ruminant animals, such as cattle, sheep, and goats, may become infected with trematode parasites by grazing on contaminated pastures or drinking from contaminated water sources. Improper pasture management and lack of access to clean water can increase the risk of ruminants acquiring trematode infections, which can then be transmitted to humans through consumption of contaminated animal products (Tidman *et al.,* 2023). Failure to implement regular deworming programs for ruminant livestock can lead to high parasite burdens in animals, increasing the likelihood of trematode transmission within herds. Infected animals shed trematode eggs in their feces, contaminating the environment and posing a risk of human infection through indirect exposure to contaminated soil or water. Inadequate veterinary care and monitoring of ruminant health can result in undiagnosed trematode infections in livestock, allowing the parasites to persist and spread within animal populations (Gayo *et al.,* 2020).

2.5.6. Prevalence

Trematode infections are prevalent in tropical and subtropical regions where environmental conditions support the life cycles of these parasites (Perina *et al.,* 2023). The presence of suitable intermediate hosts and environmental factors conducive to parasite development contribute to the high prevalence of trematode infections in these regions. The lack of adequate hygiene practices and poor sanitation infrastructure in these areas contribute to the increased transmission of trematodes. Additionally, populations engaged in occupations that involve close contact with contaminated water sources, such as farmers and fishermen, are also at a heightened risk of infection (Onohuean and Nwodo, 2023).

The prevalence of trematode infections may exhibit dynamic changes over time due to various factors, including environmental modifications, climate change, and shifts in human behaviour. These factors can influence the distribution and transmission of trematodes, potentially altering the epidemiology of these infections. Understanding the evolving patterns of trematode infections is essential for adapting control strategies to address emerging challenges and mitigate the impact of these parasites on human and animal health (Tidman *et al.,* 2023) (Prevalence of *Fasciola* and *Paramphistomum* in some region of Ethiopia are shown in Table 1 and Table 2 respectively).

Table 1: Prevalence of Fasciola in ruminant across different Ethiopia

Place	**Prevalence**	**Ruminant species**	**Technique**	**Reference**
Ada’a district of Central Ethiopia	13.48%	Cattle and sheep	Coprology	Gebeyehu *et al*. (2024)
Asella	26%	Ruminant	Coprology	Megersa *et al*. (2024)
Tarcha, Southern Ethiopia	20.3%	Cattle	Coprology	Mathewos *et al (2023)*
Hosanna Town, Southern Ethiopia	8.25%	Cattle	Coprology	Tiele (2023)
Nekemte	19%	Cattle	Post mortem examination	Turuna and Adugna (2019)
Bahir Dar	14%	Cattle	Coprology	Yihdego (2017)
Ambo	10.2%	Small ruminant	Coprology	Ayana and Ifa (2015)
Bishoftu	5.4%	Ovine	Coprplogy	Dinka and Hika (2013)
Dire Dawa	24.44%	Cattle	Post mortem examination	Mebrahtu and Beka (2013)
Adwa municipal abattoir	32.3%	Cattle	Post mortem examination	Bekele *et al*. (2010)

Table 2: Prevalence of *Paramphistomum* in ruminant across different Ethiopia

Place	**Prevalence**	**Ruminant species**	**Technique**	**Reference**
Ada’a District	4.19%	Cattle and sheep	Coprology	Gebeyehu *et al.* (2024)
Hosanna	9.5%	Cattle	Coprology	Tiele *at al*. (2023)
Kutaber Woreda, South Wollo	39.1%	Sheep and goat	Coprology	Wondmnew *et al*. (2019)
Bahir Dar	48.5%	Cattle	Coprology	Aragaw and Tilahun (2019)
Nekemte	41.5%	Cattle	Post mortem	Turuna and Adugna (2019).
Bahir Dar	35.5%	Cattle	Coprology	Yihdego (2017)
Gondar	51.82%	Cattle	Post mortem	Ayalew *et al.* (2016)
Ashenge	23.7%	Sheep	Coprology	Kifleyohannes *et al*. (2015)
Bishoftu	40.1%	Cattle	Post mortem	Sintayehu and Mekonnen (2012)

2.6. Diagnostic Methods of Trematodes

2.6.1. Coprological techniques

Sedimentation methods are complementary to flotation techniques and are especially useful for detecting heavier trematode eggs that may not float during flotation procedures (Perina *et al.*, 2023). The formalin-ethyl acetate sedimentation technique is a common approach for concentrating trematode eggs in fecal samples, enhancing the sensitivity of diagnosis, especially in cases of low-level infections or certain trematode species with denser eggs. While coprological techniques are valuable tools for diagnosing trematode infections, they have certain limitations. Factors such as intermittent egg shedding, variations in egg production, and the presence of immature or prepatent infections can affect the sensitivity and specificity of these methods. Additionally, coprological techniques may not be able to differentiate between different species of trematodes or detect early-stage infections (Dravigney, 2022).

2.6.2. Serological techniques

ELISA is a highly sensitive and specific serological test for detecting antibodies against trematode antigens in ruminant serum samples. By immobilizing trematode antigens on a solid phase and detecting specific antibodies in the sample, ELISA can provide quantitative information on the host's immune response to infection. ELISA is valuable for diagnosing subclinical infections, monitoring treatment efficacy, and assessing the prevalence of trematode infections in ruminant populations (Markoulatos *et al*., 2002).

2.6.3. Molecular techniques

Molecular techniques have revolutionized the diagnosis of trematode infections in animals due to their high sensitivity, specificity, and ability to detect low levels of infection. Key methods include Polymerase Chain Reaction (PCR), which amplifies specific DNA sequences and allows for the quantification of parasite load, and Loop-Mediated Isothermal Amplification, which is rapid, cost-effective, and suitable for field diagnostics (Tomita *et al*., 2008). DNA sequencing, including next-generation sequencing and Sanger sequencing, enables the identification and characterization of trematode species. Restriction Fragment Length Polymorphism differentiates between closely related species by analyzing DNA patterns, while multiplex PCR allows for the simultaneous detection of multiple trematode species in a single reaction (Markoulatos *et al*., 2002). Real-time PCR (qPCR) provides quantitative data on parasite load. These molecular techniques facilitate early diagnosis, effective treatment, and improved management of trematode infections, ultimately enhancing the health and productivity of livestock. Continued advancements in molecular diagnostics hold promise for even more efficient and accessible tools in the future (e (Kreitmann *et al.,* 2023).

2.6.4. Post-mortem examination

Post-mortem examination, including necropsy and histopathological analysis, remains a valuable method for detecting trematode parasites in ruminants. Necropsy allows for the direct visualization and identification of trematode stages in various tissues, while histopathology provides detailed information on tissue pathology and the host's immune response to infection. Post-mortem examination is essential for confirming the presence of trematodes, assessing the severity of infection, and investigating the impact of parasites on ruminant health (Hadjiev *et al.,* 2022).

2.7. Control and Prevention Methods of Trematode Infections in Ruminants

2.7.1. Deworming approach

Regular deworming is a cornerstone of managing trematode infections in ruminants. Administering anthelmintic drugs at strategic intervals can significantly reduce the parasite burden. Timing the administration of these drugs is critical and should be based on the local epidemiology of the parasites to coincide with periods of peak transmission (Kaplan and Vidyashankar, 2012). Triclabendazole is commonly used for treating infections caused by *Fasciola* species due to its high efficacy against both immature and adult flukes. Praziquantel, on the other hand, is the drug of choice for *Schistosoma* species and other trematodes. Its broad-spectrum activity and safety profile make it a valuable tool in controlling these parasitic infections. However, the effectiveness of these treatments relies on their proper use; incorrect timing or dosages can lead to suboptimal outcomes and contribute to the development of drug resistance (Coles, 2020).

The repeated use of the same anthelmintic can lead to the development of drug-resistant trematode populations. To prevent this, it is crucial to rotate anthelmintics from different chemical classes. This practice helps to reduce the selection pressure for any one drug, thereby slowing the development of resistance. Monitoring the efficacy of anthelmintic treatments is also essential. This can be achieved through fecal egg count reduction tests, which involve comparing the number of parasite eggs in the feces before and after treatment. A significant reduction in egg counts indicates effective treatment, while a lack of reduction may signal emerging resistance. Regular monitoring allows for timely adjustments in treatment protocols and the selection of alternative drugs if resistance is detected (Sangster and Gill, 2017).

2.7.2. Pasture management

Implementing rotational grazing is an effective pasture management strategy to control trematode infections. This practice involves dividing the pasture into smaller sections and rotating the ruminants through these sections. By doing so, each section is given time to rest and recover, which helps to break the life cycle of the trematodes. During the rest periods, the environmental conditions become less favorable for the survival of trematode larvae, reducing the risk of re-infection when the animals return. Additionally, rotational grazing can improve pasture health and productivity, providing better nutrition for the ruminants, which can enhance their overall resistance to infections (Abbott *et al.*, 2012).

Ensuring proper drainage of wetland areas is another critical aspect of pasture management. Wet, marshy areas provide ideal habitats for snails, which are intermediate hosts for many trematodes. By improving drainage, the habitat for these snails is reduced, which in turn disrupts the life cycle of the trematodes. Practical measures include the construction of drainage ditches, the use of pumps to remove excess water, and the strategic planting of vegetation that can absorb water. These efforts not only reduce the snail population but also improve the overall quality of the pastureland, making it less conducive to trematode transmission (Stothard and Rollinson, 2021).

2.7.3. Biological control

Biological control of snail populations is a crucial strategy for reducing trematode infections, as snails serve as intermediate hosts for many trematode species. Introducing biological agents that prey on or compete with snails can significantly decrease their numbers. Certain fish species, such as black carp (*Mylopharyngodon piceus*) and redear sunfish (*Lepomis microlophus*), are known to feed on snails and can be introduced into water bodies to naturally reduce snail populations (Brown *et al.*, 2021). Additionally, plant molluscicides derived from natural sources, such as the saponins from soapberry (*Sapindus* spp.) and the alkaloids from endod (*Phytolacca dodecandra*), have been shown to be effective in controlling snail populations without causing significant harm to the environment (Adewumi *et al.*, 2020). These biological methods provide a sustainable and eco-friendly alternative to chemical molluscicides.

Another innovative approach involves the use of predatory nematodes that specifically target snail hosts. For example, the nematode *Phasmarhabditis hermaphrodita* has been studied for its potential to infect and kill various snail species. These nematodes can be applied to areas with high snail populations, where they seek out and parasitize the snails, ultimately leading to their death. This method of biological control is advantageous because it specifically targets the snails without affecting other non-target organisms, making it a precise and environmentally friendly option for managing intermediate host populations (Wilson and Denham, 2019).

2.7.4. Environmental and cultural practices

Effective management of aquatic vegetation is vital in reducing habitats where snails thrive. Aquatic plants such as water hyacinth, reeds, and other emergent vegetation provide ideal environments for snail breeding and development. By systematically clearing or managing these aquatic plants, the habitats for snails can be significantly reduced. Mechanical removal, controlled burning, or the use of herbicides (in a controlled and environmentally sensitive manner) can help achieve this goal.

Additionally, introducing competitive plant species that do not support snail populations can alter the habitat to be less conducive to snails (Denny and Tun, 2022).

Proper water management practices are essential to limit the breeding sites for snails. Stagnant water bodies are prime locations for snail proliferation. By improving water flow and reducing standing water through the construction of drainage ditches, installing pumps, and creating channels to increase water movement, the environments suitable for snail breeding can be minimized. Additionally, promoting land use practices that reduce waterlogging, such as contour plowing and the use of raised beds in agriculture, can further decrease the likelihood of snail habitats forming (Kumar *et al.*, 2021).

2.7.5. Vaccination and immunoprophylaxis

Research into vaccines for trematodes holds great promise for long-term control of these infections in ruminants. Developing vaccines that induce immunity in animals could provide a sustainable solution by reducing the reliance on chemical treatments and other control measures. Current research focuses on identifying and characterizing key antigens from trematodes that can stimulate a protective immune response in ruminants. Experimental vaccines targeting *Fasciola hepatica*, for example, have shown some success in inducing partial immunity, which reduces the burden of infection and transmission potential (Dalton *et al.*, 2022). Continued research and development efforts aim to enhance the efficacy and practicality of these vaccines, potentially leading to widespread adoption and significant reductions in trematode infections.

3. MATERIALS AND METHODS

3.1. Description of the Study Area

A study was conducted in and around Bishoftu, East Shoa zone, Oromia, Ethiopia. Bishoftu town is located 45 km southeast of Addis Ababa, the capital city of Ethiopia. It is located at a latitude of 8.7599°' N and a longitude of 39.002°'E, at an altitude of around 1,920 meters above sea level. The town experiences a subtropical highland climate. Bishoftu receives an average annual rainfall of 866 mm, with a bimodal rainfall pattern - the main rainy season is from June to September, and the short rainy season is from March to May. The average annual temperature in Bishoftu ranges from a minimum of 10°C (50°F) to a maximum of 25°C (77°F), with an average of around 18°C (64°F). The relative humidity in Bishoftu is typically between 60-70% (NMA, 2007). According to the latest census data from 2023, the population of Bishoftu is approximately 217,964 (CSA, 2023). The Bishoftu area is also home to a notable livestock population, including approximately 160,697 cattle; 22,181 sheep; 37,510 goats; 1,660 equines, and 191,380 poultry. The livestock production system in the area is both intensive and extensive type (CSA, 2023).

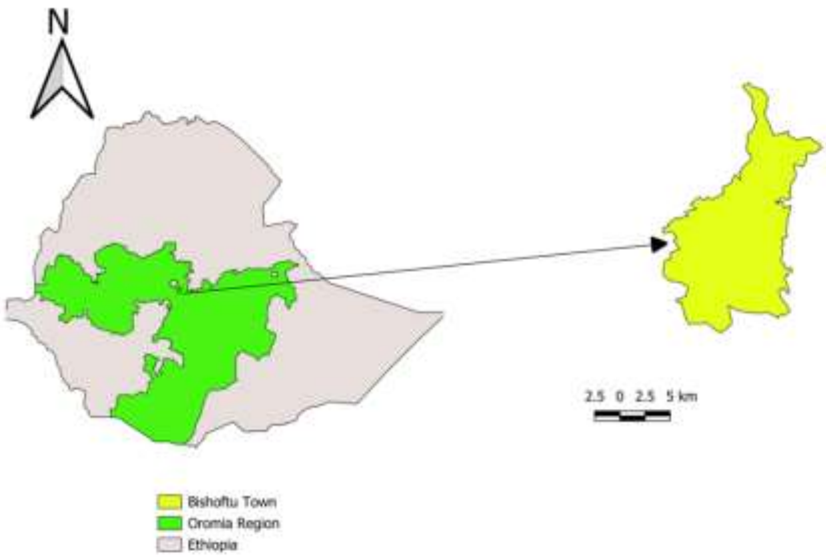

Figure 1: Map of the study area

(QGIS version 3.30 Ethiopia Admin shape file 2021).

3.2. Study Population

The coprological investigation was conducted on indigenous breeds of cattle, sheep, and goats, representing all age groups and both sexes, which were kept under traditional extensive husbandry systems with communal grazing practices. The questionnaire was administered to livestock owners aged 21 and above, including both genders. The malacological study was conducted on snails collected from various water bodies within the wetland areas of Bishoftu, with the sampling sites selected based on their potential to harbor snail intermediate hosts of trematode parasites, considering factors such as water availability, vegetation, and proximity to grazing areas.

3.3. Study Design and Sampling strategy

A cross-sectional study was conducted from November 2023 to April 2024 to investigate the prevalence of trematode infections in ruminants, the distribution and infectivity of snail intermediate hosts, and the knowledge, attitudes, and practices of farmers. The study location, or locations, were purposively selected based on their agro-ecological characteristics. Selection for the second stage (household level) was done based on their willingness (volunteer). A simple random sampling technique was then employed to select the ruminants (cattle, sheep, and goats) for coprological investigation. For the questionnaire survey, from participating household, individuals aged above 21 years were randomly selected. For the malacological study, snails were collected from various water bodies using hand collection technique.

3.4. Sample Size Determination

The sample size for this study was determined using different approaches for the various components of the study: ruminant sampling, farmer surveys, and snail collection. For the coprological investigation, the sample size calculation was based on a formula recommended by Thrusfield (2005). The expected prevalence of trematode infections was estimated at 5.4%, based on a previous copromicroscopic study conducted on the ovine fasciolosis of Bishoftu area (Dinka and Hika, 2013). To achieve a 95% confidence interval and a desired level of absolute precision set at 5%, the minimum required sample size was calculated to be 80 animals per ruminant species (cattle, sheep, and goats).

Substituting the values:

$$n = \frac{1.96^2(0.054)(1-0.054)}{(.05)^2} = \frac{3.84 \times 0.054 \times 0.946}{0.0025} = \frac{0.20}{0.0025} = 80$$

To increase the level of precision, 40% of the estimated value was added, resulting in 112 animals for each ruminant species. Therefore, a total of 336 ruminants were selected and examined (112 cattle, 112 sheep, and 112 goats).

For the questionnaire survey, the sample size was calculated based on the formula recommended by Arsham (2002): $N = 0.25/SE^2 = 100$. Where: N= sample size, SE = standard error, assuming a standard error of 5%. Accordingly, the respondents required for interviewe was 100 livestock owners.

For the malacological study, snail collection was conducted in various water bodies within the selected locations, aiming to collect as many snails as possible to ensure a representative sample. The total snails collected were 400 snails from water bodies.

3.5. Study Methodes

3.5.1. Data Collection and laboratory examination

A prescribed questionnaire with the information of the host, deworming history, and management system and owners was used by the participants. Detailed primary data on each sampled animal's sex, age, and Body Condition Score (BCS), were recorded (Annex 8). The age of cattle was determined using tooth eruption and wear techniques as described by DeLahunta and Habel (1986), dividing the animals into young (<2 years), adult (2-5 years), and old (>5 years) categories. For sheep and goats, age classification followed Samad's (2008) method, dividing them into kids (1-6 months), young (>6–18 months), and old (>18 months). Body condition scoring was conducted based on established matrices; for sheep and goats, the scoring system adopted from Suiter (1994) was used, while for cattle, the system from Nicholson and Butterworth

(1986) was employed. These matrices consider various physical attributes such as fat cover and muscle tone. Each sampled animal was associated with specific environmental data, including the sampling site (location) and the exact sampling date. The study involved two distinct seasons: the semi-dry season from November to January and the dry season from February to April, crucial for understanding the seasonal variations in trematode prevalence and snail intermediate hosts activity.

Fecal samples were collected directly from the rectum of the animals using disposable gloves to ensure hygienic and uncontaminated samples. In cases where direct collection was not feasible, samples were obtained from freshly voided fecal masses on the ground. Each sample was immediately placed in a clean, labeled universal bottle, with labels including detailed information about the animal's origin (location), species, age, sex, and body condition score. Collected fecal samples were promptly transported to the parasitology laboratory at the Addis Ababa University College of Veterinary Medicine and Agriculture. Samples that could not be examined immediately upon arrival were stored in a temperature-controlled refrigerator at 4°C.

Snail collection was conducted in various water body such as wetlands, ponds, rivers, irrigation canals, and lakes known for their potential to harbor snail populations that serve as intermediate hosts for trematodes. Sites were selected based on frequent grazing and human activity, which are factors that could influence snail populations and trematode transmission (Annex 4 and 9). Snail sampling was performed using hand collection, where snails visually searched and manually collected from aquatic vegetation and rocks in the water. Collections were made during early morning and late afternoon, periods of high snail activity. Each collection site was recorded with detailed information about the location, type of water body, and date of collection. These dates were later used to classify the sampling season into semi-dry (November to January) and dry (February to April) periods. The collected snails were placed in labeled plastic containers with adequate vegetation and a small amount of water from the collection site to maintain their natural habitat conditions. These containers were then transported to the parasitology laboratory at Addis Ababa University College of Veterinary Medicine and Agriculture for further analysis.

3.5.2. Copromicroscopic analysis

A standardized sedimentation technique recommended by DAFWA (2013) was employed for the detection of trematode eggs in the fecal samples. Briefly, 3 grams of feces were placed in a mortar, and 50 ml of tap water was added and thoroughly mixed to create a homogeneous suspension. The suspension was then filtered through a fine tea strainer to remove any large particles and debris. The filtered suspension was allowed to stand for 5 minutes to facilitate the settling of the eggs at the bottom of the test tube. The supernatant was carefully decanted, and the sediment was re-suspended in tap water. This sedimentation process was repeated two more times to ensure the removal of fecal debris, resulting in a clear supernatant. After the final sedimentation, the supernatant was carefully removed, and the sediment was transferred into a test tube and re-suspended in approximately 5 ml of tap water. The supernatant was then discarded. A drop of methylene blue was added to the sediment, and the mixture was allowed to stand for 5 minutes to facilitate the effective identification of trematode eggs.

The stained sediment was taken with pipet and transferred onto a microscopic slide and examined under a compound microscope with 10x objectives. The characteristic yellow color of *Fasciola* eggs were used to differentiate them from those of *Paramphistomum*, as described by Urquhart *et al*. (1996). Additionally, the presence of specific structures, such as operculum, spines, and knobs on the eggshell, was considered for the identification of different trematode species, following the guidelines provided by Esteban *et al*. (2014).

3.5.3. Snail identification and examination

Snail identification was carried out using morphological characteristics, focusing on shell morphology as described in identification keys and guidelines by Brown (1994). Key features such as shell shape, size, color, presence of specific markings, and the structure of the aperture and operculum were used to identify the snail species. Detailed examination of these morphological traits was performed using a stereomicroscope, and each snail was classified according to its species. The identified snails were recorded with details of their species, location of collection, and any other relevant information such as the type of water body and at the time of collection.

In the laboratory, snails were further examined for cercarial shedding following established protocols. Each snail was individually placed in a clean petri dish containing a small amount of dechlorinated water and exposed to artificial light for 2-3 hours to stimulate the shedding of cercariae, based on the protocol by Frandsen and Christensen (1984). This procedure was conducted daily for a period of two weeks to ensure the detection of any latent infections. During this time, snails were maintained in aquaria with fresh lettuce provided as food to keep them alive and active. The water in each petri dish was examined under a stereomicroscope daily to check for the presence of cercariae (Annex 7).

The characteristic morphology of the cercariae was carefully observed under setereo microscopy. Specifically, the key morphological traits of trematode cercariae include tail, motility and an elongated body typically equipped with an oral sucker at the anterior end (Urquhart *et al.*, 1996). Snails shedding cercariae were recorded as infected.

3.5.4. Questionery survey

A semi-structured questionnaire survey was conducted to assess the knowledge, attitudes, and preventive measures of farmers regarding trematode infections. The questionnaire included both closed-ended and open-ended questions and was divided into four main sections (Annex 1). Initially, a pilot test was conducted with some farmers included under target populations. This approach was taken to identify any ambiguities and ensure the clarity and interpretability of the questions. The feedback received indicated that the questionnaire was clear and understandable. This feedback was used to refine the questionnaire, resulting in the final version.

The data for this survey were collected using the Kobo Collect application on a smartphone. The first section of the questionnaire collected sociodemographic data, including age, sex, and educational status. The second section evaluated the farmers' knowledge about trematode infections in ruminants, focusing on aspects such as signs and symptoms, transmission, sources of infection, timing of occurrence, zoonotic awareness, treatment, and prevention. Responses were scored with "Yes," "No," or "I don’t know," where correct answers received 1 point and incorrect or unknown responses received 0 points. With 18 questions in this section, the total possible score ranged from 0 to 18. Knowledge scores were calculated as percentages and categorized as poor (<60%), moderate (60-79.9%), or good (80-100%) based on Bloom's cutoff points (1968).

The third section assessed attitudes towards trematode infections through 8 questions, with responses scored to reflect positive or negative attitudes. Scores were categorized as poor (<60%), medium (60-79.9%), and good (80-100%). The fourth section focused on preventive practices, evaluating how farmers prevent the disease using both modern techniques (such as anthelmintics) and traditional practices. This section consisted of 17 questions, with responses again recorded as "Yes," "No," or "I don’t know." Correct answers indicating acceptable practices received 1 point, while incorrect or unknown responses received 0 points. Practice scores were computed as percentages and categorized as acceptable (practice score percentage > 80%) or unacceptable (practice score percentage < 80%).

3.6. Data Analysis

The raw data from different studies were systematically collected and organized using Microsoft Excel databases. The data were then analyzed using StataSE 14 (64-bit) software. For the coprology study, logistic regression analysis was employed to examine the associations between parasite taxa and potential risk factors, with the results expressed using Odds Ratios (OR), P-values, and 95% confidence intervals. The malacology study data were analyzed using Pearson's chi-square tests to examine significant associations between different risk factors and the infection status of snails, with results interpreted using P-values. In the questionnaire survey analysis, Pearson's chi-square test was used to explore the relationships between knowledge, attitudes, and practices with sociodemographic characteristics, identifying significant associations between these variables.

3.7. Ethical Consideration

The study was carried out in accordance with the Animal Research Ethics Review Committee of the College of Veterinary Medicine and Agriculture of Addis Ababa University and the approval certificate ensured ethical approval for the study with reference number VM/ERC/02/16/024, 26/03/2024 indicated in (Annex 10). The owners were informed about the purpose of the study and consent was sought. The samples for this investigation were carefully gathered without causing any harm to the animals and in accordance with all ethical standards for sample collection and sampling methods.

4. RESULTS

4.1. Coprology Study Results

The copromicroscopic study indicated the overall prevalence of 11.61% (95% CI: 8.58-15.52) trematode infections among ruminants in the study area. *Fasciola* and *Paramphistomum* egg idenfitied in this study (Annex 2). Among the identified trematodes, *Fasciola* was the most prevalent, followed by *Paramphistomum* with a prevalence of 8.93% (95% CI: 6.3-12.5) and 4.46% (95% CI: 2.71-7.29), respectively. Additionally, co-infections of *Fasciola* and *Paramphistomum* were detected in 6 samples, indicating a co-infection prevalence of 1.79% (95% CI: 0.8-3.93) (Table 3).

Table 3: Prevalence of trematodes infections

Name of parasite	Positive sample	Prevalence	95% CI
Fasciola	30	8.93	6.3-12.5
Paramphistomum	15	4.46	2.71-7.29
Fasciola + *Paramphistomum*	6	1.79	0.8-3.93
Overall	**39**	**11.61**	**8.58-15.52**

The logistic regression analysis of the prevalence of trematodes in ruminants around Bishoftu town identified several significant associations with various risk factors. Specifically, the site of Sardo (OR=6.97, 95% CI=1.10-44.05, P=0.039) and Yatu (OR=6.75, 95% CI=1.028-44.36, P=0.047) showed a significantly higher prevalence of trematode infections compared to Kata. Among species, both cattle (OR=4.86, 95% CI=1.55-15.21, P=0.007) and sheep (OR=6.51, 95% CI=2-19.9, P=0.001) had a significantly higher prevalence of trematodes compared to goats. Additionally, ruminants with poor body condition scores (BCS) had a much higher likelihood of trematode infection (OR=11.20, 95% CI=3.40-36.91, P=0.000) compared to those with good BCS. Seasonally, the semi-dry season was associated with a higher prevalence of infections (OR=3.79, 95% CI=1.63-8.84, P=0.002) compared to the dry season. Other factors, such as sex and age group, showed no significant associations (Table 4).

Table 4: Logistic regression analysis of the association between trematode infections and risk factors in ruminants around Bishoftu

Variable	**Species**	**OR**	**(95%CI)**	**P-value**
Site	Cheleleka	4.76	0.70-32.41	0.110
	Dhankaka	4.75	0.73-30.71	0.102
	Dire	2.30	.33-16.09	0.402
	Godino	4.734	.71-31.43	0.107
	Kality	1.39	.20-9.70	0.737
	Sardo	6.97	1.10- 44.05	0.039
	Yatu	6.75	1.028- 44.36	0.047
	Kata	Reference		
Species	Cattle	4.86	1.55-15.21	0.007
	Sheep	6.51	2-19.9	0.001
	Goat	Reference		
Sex	Female	1.89	.84-4.22	0.122
	Male	Reference		
Age group	Young	3.30	.99-10.96	0.052
	Adult	2.06	.83-5.14	0.121
	Old	Reference		
BCS	Poor	11.20	3.40-36.91	0.000
	Medium	1.44	.37-5.64	0.601
	Good	Reference		
Season	Semi-dry	3.79	1.63- 8.84	0.002
	Dry	Reference		

N.B: * Significant difference. Description of age group: Cattle: Young: < 1.5 yr, Adult: 1.5-5 year, Old: >5 yr. Sheep and goat: Young: 1-6 months, Adult: 6–18 months, Old: >18 months.

The prevalence of *Fasciola* and *Paramphistomum* infections was assessed across different factors. By location, the prevalence of *Fasciola* was highest in Sardo at 14.29%, followed by Dhankaka at 11.9%, and Cheleleka, Godino, and Yatu each at 9.52%. In terms of *Paramphistomum*, Yatu had the highest prevalence at 7.14%, while several locations including Cheleleka, Dhankaka, Dire, Godino, and Sardo had a prevalence of 4.76%. Among species of ruminants, sheep exhibited the highest prevalence of *Fasciola* at 14.29%, while cattle had the highest prevalence of *Paramphistomum* at 10.71%. For sex, females showed a higher prevalence of both *Fasciola* and *Paramphistomum* at 10.47% and 6.4%, respectively, compared to males with 7.32% and 2.44%. Age-wise, young animals had a *Fasciola* prevalence of 11.54% and a notably higher *Paramphistomum* prevalence of 19.23%. Adults showed a *Fasciola* prevalence of 15.38% and *Paramphistomum* prevalence of 6.15%, whereas old animals had lower prevalences at 6.94% for *Fasciola* and 2.45% for *Paramphistomum*. Body condition score (BCS) data indicated that animals with poor BCS had the highest prevalence of *Fasciola* at 16.79% and *Paramphistomum* at 9.16%. Animals with medium BCS had prevalences of 4.55% for *Fasciola* and 0.91% for *Paramphistomum*, while those with good BCS had the lowest prevalences at 3.16% and 2.11%, respectively. Seasonally, the semi-dry season saw a higher prevalence of *Fasciola* at 13.10% and *Paramphistomum* at 5.95% compared to the dry season, which had prevalences of 4.76% and 2.98%, respectively. These findings highlight notable variability in the prevalence of these infections based on location, species, sex, age, body condition, and season.

The study assessed the prevalence of trematode infections (*Fasciola* and *Paramphistomum*) in ruminants, considering factors such as location, species, sex, age, body condition score (BCS), and seasonality. Logistic regression analysis identified several significant associations between risk factors and trematode infections. With fasciolosis, sheep were over seven times more likely to be infected with *Fasciola* compared to goats (OR = 7.713, 95% CI: 2.197-27.076, $p = 0.001$). Cattle were also more likely to be infected compared to goats (OR = 3.550, 95% CI: 0.993-12.696, $p = 0.051$), though this was not statistically significant. Additionally, ruminants with poor BCS were over twelve times more likely to be infected compared to those with good BCS (OR = 12.27, 95% CI: 3.097-48.118, $p < 0.001$). Those in medium condition had a higher likelihood of infection compared to those in good condition (OR = 1.959, p = 0.391), but this was not significant. Seasonality played a role; the semi-dry season was associated with a more than threefold increase in the likelihood of infection compared to the dry season (OR = 3.265, 95% CI: 1.307-8.155, p = 0.011) (Table 5).

Regarding *Paramphistomum* infections, cattle were over eighteen times more likely to be infected compared to goats (OR = 18.403, 95% CI: 2.111-160.375, p = 0.008). Sheep had a higher likelihood compared to goats (OR = 1.367, p = 0.817), but this was not significant. Age was another critical factor; young ruminants were nearly fifteen times more likely to be infected with *Paramphistomum* compared to older ones (OR = 14.786, 95% CI: 2.295-95.276, p = 0.005). Adults had a higher likelihood of infection compared to older ruminants (OR = 2.246, p = 0.311), though this was not significant. Poor BCS was significant again, with ruminants in poor condition being more likely to be infected (OR = 6.277, 95% CI: 1.147-34.363, p = 0.034). Those in medium condition had a lower likelihood compared to those in good condition (OR = 0.201, p = 0.237), but this was not significant (Table 5).

Table 5: Logistic regression analysis for factors potentially associated with *Fasciola* and *Paramphistomum* infections in ruminants

Factor	Variable	*Fasciola*				*Paramphistomum*			
		OR	95% CI		*p-value*	OR	95% CI		*p-value*
			Lower	Upper			Lower	Upper	
Location	Cheleleka	6.511	0.603	70.264	0.123	7.522	211.60	211.604	0.2360
	Dhankaka	7.154	0.702	72.900	0.097	4.836	0.186	125.984	0.343
	Dire	3.06	0.265	35.482	0.370	3.635	0.137	96.263	0.440
	Godino	7.106	0.800	74.279	0.101	9.738	0.359	264.315	0.177
	Kality	1.785	0.160	19.965	0.638	1.411	0.0427	46.680	0.847
	Sardo	9.921	0.992	99.252	0.051	7.850	0.291	211.473	0.220
	Yatu	7.299	0.696	76.568	0.097	15.40	0.529	449.420	0.112
	Kata	Reference				Reference			
Ruminant species	Cattle	3.550	0.993	12.696	0.051	18.403	2.111	160.375	0.008*
	Sheep	7.713	2.197	27.076	0.001*	1.367	0.097	19.306	0.817
	Goat	Reference				Reference			
Sex	Female	1.997	0.822	4.849	0.127	4.015	0.925	17.435	0.064
	Male								
Age	Young	1.036	0.233	4.595	0.963	14.786	2.295	95.276	0.005*
	Adult	2.445	0.921	6.493	0.073	2.246	0.469	10.751	0.311
	Old	Reference				Reference			
BCS	Poor	12.27	3.097	48.118	0.000*	6.277	1.147	34.363	0.034*
	Medium	1.959	1.421	9.120	0.391	0.201	0.014	2.879	0.237
	Good	Reference				Reference			
Season	Semi-dry	3.265	1.307	8.155	0.011*	2.603	0.667	10.157	0.168
	Dry	Reference				Reference			

N.B: * Significant difference. Description of age group: Cattle: Young: < 1.5 yr, Adult: 1.5-5 year, Old: >5 yr. Sheep and goat: Young: 1-6 months, Adult: 6–18 months, Old: >18 months.

4.2. Malacology Study Results

4.2.1. Frequency of snails

A total of 400 snails were collected from various water bodies, including lakes, rivers, ponds, irrigation canals, and wetland areas in Bishoftu. The collected snails belonged to different species (Figure 2). Among these, *Lymnaea natalensis* was the most widely distributed species, making up 37% of the total collection. *Physa* spp was the second most prevalent, comprising 21.25% of the snails collected. *Bulinus (B.) truncatus* accounted for 15.75%, while *Lymnaea truncatula* made up 10%. *Biomphalaria pfeifferi* constituted 9.5%, and *Bulinus (P.) africanus* was the least collected species at 6.5%. All collection sites had regular animal contact for drinking and grazing purposes. The snails were predominantly found attached to aquatic vegetation, stones, and other aquatic objects (Annex 4).

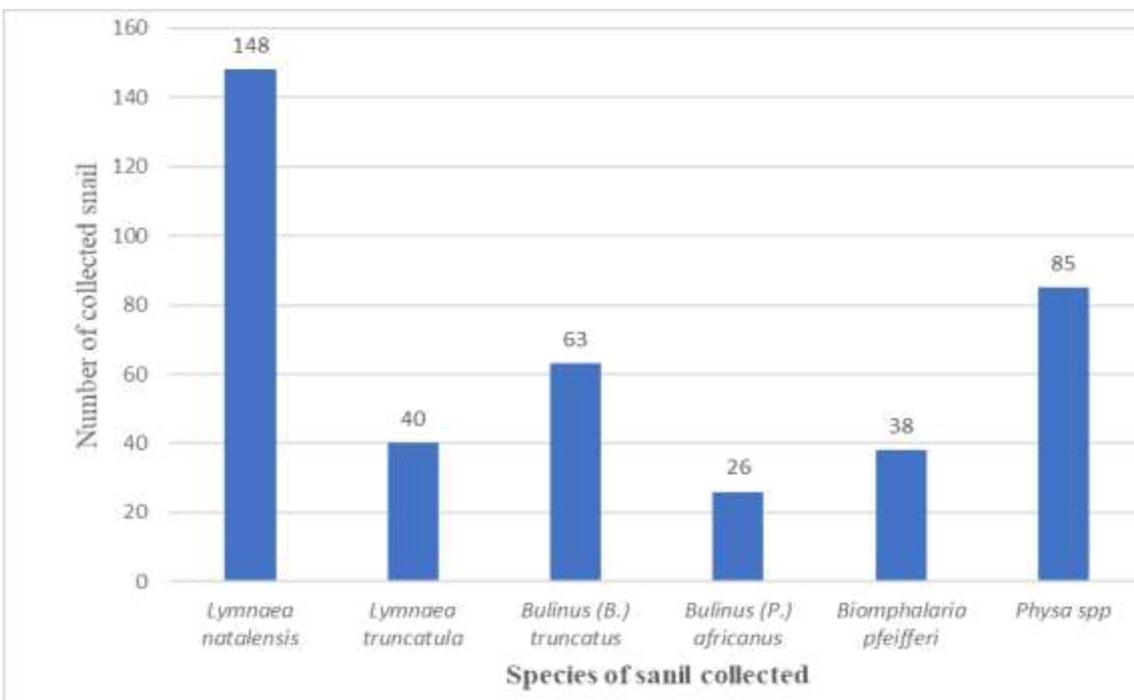

Figure 2: Distributions of different snail species in the study area

4.2.2. Cercarial shedding by snails

Out of the 400 snails that were collected, 31 of them (7.75%) were found to be infected with larval trematodes (Annex 3). *Lymnaea natalensis* exhibited the highest rate of infection, with 20 out of 148 snails (13.51%) tested positive. *Lymnae truncatula* had an rate of infection of 7.50% (3 out of 40 snails). *Bulinus (B.) truncatus* had an rate of infection of 11.11% (7 out of 63 snails), and *Bulinus (P.) africanus* had an rate of infection of 3.85% (1 out of 26 snails). No infections were found in *Biomphalaria pfeifferi* (38 snails) and *Physa spp* (85 snails). The chi-square value for species of snail was 18.76 with a p-value of 0.002, indicating a significant difference in rate of infections among the species (Table 6).

Table 6: Snail species identified and their rate of infection in the study area

Species of snail identified	**Number of snail collected**	**№ of infected snail**	**Chi2**	**p-value**
Lymnaea natalensis	148	20 (13.51%)	18.76	0.002
Lymnaea truncatula	40	3 (7.50%)		
Bulinus(B.) truncatus	63	7(11.11%)		
Bulinus(P.) africanus	26	1(3.85%)		
Biomphalaria pfeifferi	38	-		
Physa spp	85	-		
Total	**400**	**31(7.75%)**		

Snails collected from Sardo had the highest rate of infection at 16%, followed by Dhankaka at 12%, Yatu and Godino both at 8%, Cheleleka and Kality each at 6%, and Kata with the lowest rate of infection at 2%. However, no significant difference in rate of infections among the different locations. The rate of infections of cercaria varied significantly across different habitats, and seasons. Ponds had the highest rate of infection at 13.48%, followed by wetlands at 10.67%, rivers at 2.94%, and irrigation canals at

1.56%, with no infections found in lakes. The chi-square value for habitat type was 15.6 with a p-value of 0.004, indicating a significant difference in rate of infections among habitat types. The rate of infections also varied significantly by season. During the semi-dry season, 12% of snails were infected, compared to 3.50% during the dry season. The chi-square value for season was 10.11 with a p-value of 0.001, indicating a significant difference in rate of infections between the semi-dry and dry seasons (Table 7).

Table 7*:* Occurrence of cercaria infection in the snail vectos based on different risk factor

Factors	**Variables**	**Number of Snail collected**	**Percentage of infected snail**	**Chi2**	**P- value**
Location	Dire	50	2(4%)	9.76	0.203
	Cheleleka	50	3(6%)		
	Dhankaka	50	6(12%)		
	Godino	50	4(8%)		
	Kality	50	3(6%)		
	Kata	50	1(2%)		
	Sardo	50	8(16%)		
	Yatu	50	4(8%)		
Habitat type	Ponds	400	19 (13.48%)	15.6	0.004
	Wetlands	75	8 (10.67%)		
	River	102	3 (2.94%)		
	Irrigation canal	64	1 (1.56%)		
	Lakes	18	-		
Season	Semi-dry	200	24(12%)	10.11	0.001
	Dry	200	7 (3.50%)		

4.3. Questionnaires Survey Results

4.3.1. Demography of respondents

The largest group of participants was males, comprising 78% of the population. The age group that participated the most was 31-40, representing 46% of the respondents. The respondents' educational background included elementary school, high school or above, and leman (Table 8).

Table 8*:* Respondent's demographic characteristics

Sociodemographic	**Variables**	**Percent (N=100)**
Sex	Male	78%
	Female	22%
Age	21-30	14%
	31-40	46%
	41 years and above	40%
Educational status	Leman	35%
	Elementary	57%
	High school/above	8%
Total		**100%**

4.3.2. Knowledge level on trematode infections

The results indicated majority of respondents (62%) heard about trematode infections, yet awareness specifically about these infections in ruminants is somewhat lower (57%). Symptoms such as swelling under the jaw (73%) and diarrhea (67%) are well recognized, whereas weight loss (42%) is less commonly identified. Knowledge about transmission sources is varied; while there is good awareness of the movement of infected animals (74%) and contaminated grazing areas (63%), awareness of aquatic snails (0%) and aquatic plants (12%) as sources is strikingly low. The majority of respondents correctly identify the rainy season as the peak time for infections (73%). Awareness of the impacts of trematode infections, particularly in terms of production loss (83%) and body weight loss (84%), is strong. However, awareness of the zoonotic potential of trematode infections is very low (7%) (Table 9).

Table 9: Knowledge of participants about trematode infection

No.		Knowledge Questions	Correct (%)	Incorrect (%)
1		Heard about trematode infections	62	38
2		Awareness of trematode infections in ruminants	57	43
3		**Symptoms of trematode infections include**		
	I	Reduced appetite	61	39
	II	Weight loss	42	58
	III	Diarrhea	67	33
	IV	Swelling under the jaw (bottle jaw)	73	17
4		**Possible sources of trematode infections**		
	I	Contaminated grazing area a	63	37
	II	Contaminated water	40	60
	III	Movement of infected animals	74	26
	IV	Aquatic snails	0	100
	VI	Acquatic plants	12	88
5		**Trematode infections most frequent at**		
	I	During dry season	64	36
	II	During the rainy season	73	17
	III	Similar throughout the year	65	35
6		**Impacts of trematode infections**		
	I	Loss due to mortality of animals	73	27
	II	Sick animals cause production loss	83	17
	III	Loss of body weight of animal	84	16
7		Awareness of trematode zoonosis	7	93

Out of the 100 participants, 49% had a poor knowledge score (<60%), 34% had a moderate knowledge score (60-79%), and 17% had a good knowledge score (80-100%). The mean score for knowledge among the study respondents was 47.53% (Figure 3).

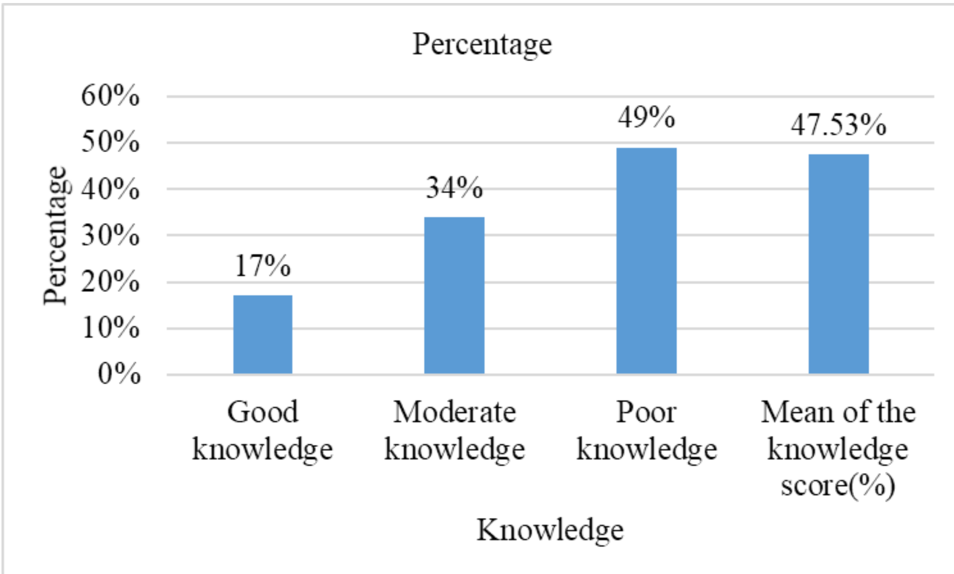

Figure 3: Percentage distribution of respondents' knowledge categories on trematode infections

4.3.3. Attitude toward trematode infections

The attitudes of respondents towards trematode infections reveal a spectrum of perceptions. While a considerable portion expressed concern about the potential impact of trematode infections on livestock health and productivity (67%), others demonstrated uncertainty or indifference. Statements regarding the seriousness of the threat posed by trematode infections to animal health received high levels of agreement (71%), indicating a widespread recognition of the severity of these infections. However, beliefs regarding the effectiveness of traditional methods in controlling trematode infections were more varied, with a minority (22%) agreeing that traditional methods are effective. Similarly, while there was widespread concern about the potential public health risks associated with trematode infections in ruminants (70%), a notable portion expressed neutral or disagreeing attitudes (Table 10).

Table 10: Distribution of participants' attitudes toward trematode infections

№	Statement	Attitude Scale		
		Agree	**Neutral**	**Disagree**
1	Drainage is important for the reduction of snail-born parasitic diseases.	31	42	27
2	I am concerned about the potential impact of trematode infections on the health and productivity of my ruminant livestock.	67	24	9
3	Trematode infection can bring mortality of animals.	34	43	23
4	Animal health is seriously threatened by trematode infections.	71	22	7
5	Concerns regarding trematode infections should be increasing.	61	34	5
6	Traditional methods of prevention and management are effective in controlling trematode infections in my herd/flock.	22	63	15
7	The animals infected with trematode diseases can be treated and cured.	71	20	9
8	I am concerned about the potential public health risks associated with trematode infections in ruminants.	70	22	8

Out of 100 respondents, the majority of them have a moderate attitude (47%), while a considerable portion exhibit a poor attitude (37%) and only a small percentage show a good attitude (16%). The average attitude score across all respondents is 55.16% (Figure 4).

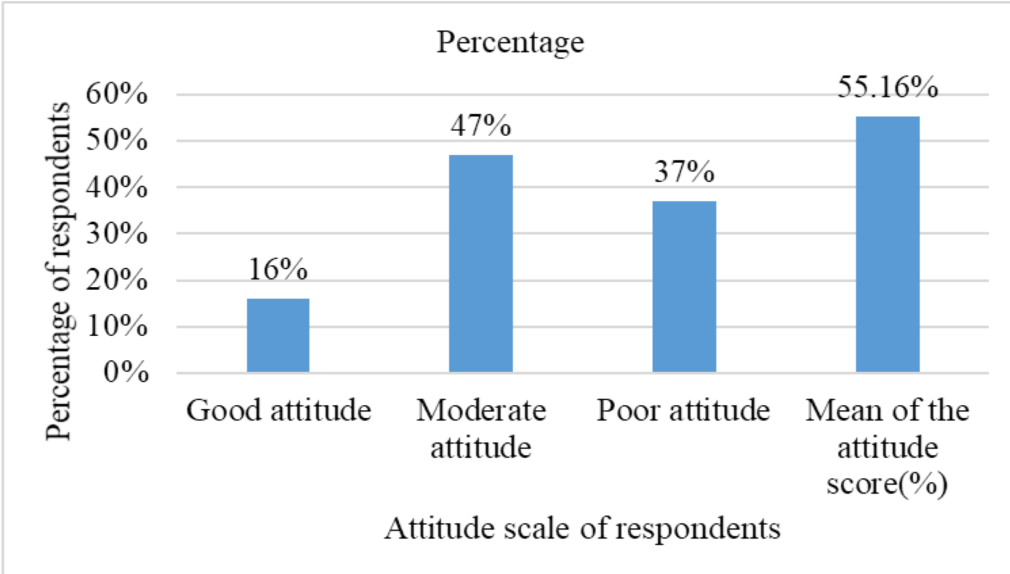

Figure 4: Respondents percentage categories based upon attitude

4.3.4. Preventive practices against trematode infections

The findings illustrate the diverse methods employed by respondents to prevent and manage trematode infections, reflecting a blend of traditional and modern practices within their communities. While a minority (17%) report using traditional herbal or plant-based remedies, the majority (76%) adhere to regular deworming with anthelmintic drugs, aligning with conventional veterinary recommendations. Interestingly, only a small fraction (12%) engage in pasture or grazing management to reduce snail populations, indicating potential gaps in environmental management practices. Similarly, few respondents (7%) focus on improving drainage and water sources, suggesting limited awareness or resources for environmental modifications (Table 11).

Regarding traditional trematode management practices, a notable proportion of respondents (27%) acknowledge the use of specific plant parts with claimed anthelmintic properties, while fewer engage in practices involving ritual or spiritual elements (8%) or homemade concoctions (14%). Interestingly, a few participants (9%) identified specific traditional remedies like *Baala Haadha Bocoo*/*Indiriffaa*/*Insilii*, *Waalleessii*, and *Hoolotoo* originated from Oromic language for treating trematode infections (Annex 5). However, despite the recognition of traditional anthelmintic plants in the study area by some respondents (9%), there is a prevailing uncertainty or lack of knowledge regarding their effectiveness, as evidenced by the high percentage of "Incorrect/I don't know" responses (Table 11).

Table 11: Participants' responses to practice questions on trematode infections

№		Practice Question/Statements	Correct (%)	N	Incorrect/I don't know (%)
1		**Describe the methods used to prevent and manage trematode infections**			
	I	Use of traditional herbal/plant-based remedies	17		83
	II	Regular deworming with anthelmintic drugs	76		24
	III	Pasture/grazing management to reduce snails	12		88
	IV	Improving drainage and water sources	7		93
2.		**Describe any traditional trematode management practices used in your area.**			
	I.	Use of specific plant parts (leaves, roots, seeds) with claimed anthelmintic properties	27		73
	II.	Traditional practices involving ritual or spiritual elements (e.g., chants, prayers)	8		92
	III.	Home-made concoctions using herbs, spices, other ingredients	14		86
3		**Mentions of traditional anthelmintic plants in the study area**	9		91
4.		**How effective have the traditional treatments been in your experience?**			
	I.	Observed reduction in clinical signs and improved health	24		76
	II.	Reduction in frequency or severity of infections	18		82

	III. Limitations in quantifying effectiveness compared to conventional treatments	43	57
5.	**Why do you use traditional remedies instead of/in addition to modern treatments?**		
	I. Lower cost and better accessibility of traditional remedies	22	78
	II. Preference for "natural" treatments and mistrust of modern pharmaceuticals	6	94
	III. Cultural beliefs and traditional knowledge passed down	19	81
6.	**Where did you learn about the traditional trematode control methods?**		
	I. Knowledge acquired from elders or experienced farmers	43	57
	II. Sharing of traditional practices among fellow livestock keepers	40	60
	III. Self-experimentation and observation of outcomes	7	93

Around 40% of respondents demonstrated scores ranging from 80% to 100%, indicating a high level of acceptance towards recommended practices. In contrast, 50% of respondents fell into the category of unacceptable practices, with scores ranging from 60% to 79%. The mean practice score across all respondents was 58.33% (Figure 5).

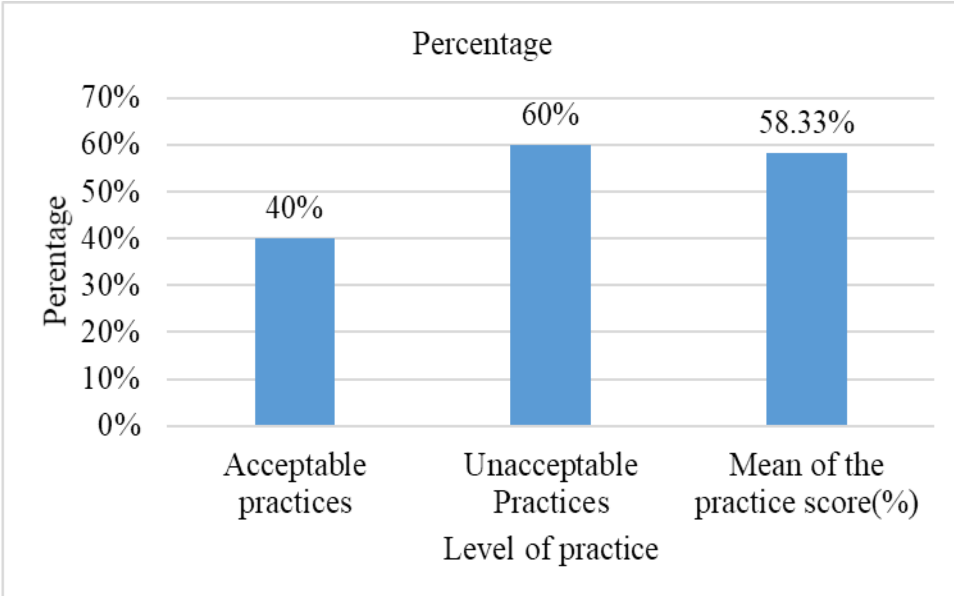

Figure 5: Percentage categories of participants on preventive practices

4.3.5. Sociodemographic associations between knowledge, attitude, and preventive practices

The table presents the association between knowledge, attitude, and practice (KAP) regarding trematode infections of ruminants and demographic factors, namely gender, age, and education level. Notably, significant associations were observed between education level and all three aspects of KAP ($p = 0.045$ for knowledge, $p = 0.000$ for attitude, and $p = 0.004$ for practice). Specifically, individuals with higher education levels tended to have better knowledge, more positive attitudes, and exhibit more accepted practices concerning trematode infections. Additionally, gender and age were found to influence knowledge scores significantly ($p = 0.042$ for gender and $p = 0.011$ for age), highlighting potential disparities in understanding the risks associated with trematode infections among different demographic groups. However, gender and age did not show significant associations with attitude or practice scores, suggesting that factors beyond demographic characteristics may play a role in shaping individuals' attitudes and behaviors towards trematode infections (Table 12).

Table 12: Association between KAP to sociodemographic factor towards trematode infection

	Knowledge				**Attitude**				**Practice**		
	Poor (49)	**Moderate (34)**	**Good (17)**	**P value**	**Poor (37)**	**Moderate (47)**	**Good (16)**	**P value**	**Accepted (40)**	**Unaccepted (60)**	**P value**
Gender											
Male	33 (67.3)	30 (88.2)	15 (88.2)	0.042*	30 (81.1)	35 (74.5)	13 (81.2)	0.724	32 (80.0)	46 (76.7)	0.693
Female	16 (32.7)	4 (11.8)	2 (11.8)		7 (18.9)	12 (25.5)	3 (18.8)		8 (20.0)	14 (23.3)	
Age											
20-30yr	9 (18.4)	4 (11.8)	1 (5.9)	0.011*	5 (13.5)	7 (15.0)	2 (12.5)	0.648	7 (17.5)	7 (11.7)	0.239
31-40yr	29 (59.2)	12 (35.3)	5 (29.4)		17 (46.0)	19 (40.4)	10 (62.5)		21 (52.5)	25 (41.6)	
Over 41yr	11 (22.4)	18 (52.9)	11 (64.7)		15 (40.5)	21 (44.6)	4 (25.0)		12 (30.0)	28 (46.7)	
Education											
Leman	21 (42.9)	9 (26.5)	5 (29.4)	0.045*	18 (48.6)	15 (31.9)	2 (12.6)	0.000*	7 (17.5)	28 (46.7)	0.004*
Elementary	27 (55.1)	22 (64.7)	8 (47.1)		19 (51.4)	31 (66.0)	7 (43.7)		27 (67.5)	30 (50.0)	
High school or above	1 (2.0)	3 (8.8)	4 (23.5)		-	1 (2.1)	7 (43.7)		6 (15.0)	2 (3.3)	

5. DISCUSSION

5.1. Coprology Study

The overall prevalence of trematode infections was found to be 11.61%, with specific prevalence of 8.93% for *Fasciola* and 4.46% for *Paramphistomum*, as well as a co-infection prevalence of 1.79%. The study area, known for its extensive wetlands, provides an ideal environment for the intermediate hosts of these trematodes, underscoring the importance of understanding infection dynamics in such habitats.

This overall prevalence of 11.61% is significant for understanding the presence of trematode infections in ruminants within the Bishoftu area. However, comparisons with other studies are challenging due to the limited availability of data on the overall prevalence of trematode infections in ruminants. One notable study by Megersa *et al*. (2024) reported a significantly higher prevalence of 35.5% in ruminants across various agro-ecological zones in Ethiopia, including Asela, Hawassa, and Batu. This discrepancy could be attributed to regional differences in environmental conditions, intermediate host density, differences in ruminant management practices**,** veterinary extension services and community knowledge (Cringoli *et al.*, 2010). Despite the scarcity of studies reporting overall trematode prevalence, the available data on specific trematode genera, such as *Fasciola* and *Paramphistomum*, provide valuable insights and allow for more detailed comparisons.

In this study, higher prevalence of *Fasciola* was observed in sheep (14.29%), followed by cattle (8.93%) and goats (3.57%). In sheep, the prevalence compared with several other studies. For instance, Dinka and Hika (2013) reported a significantly lower prevalence of 5.4% in sheep in Bishoftu, the same study area, using coprology. Similarly, Ayana and Ifa (2015) found a prevalence of 10.2% in sheep in Ambo, Central Oromia, Ethiopia, also lower than the current study. On the other hand, Regassa *et al*. (2022) reported a much higher prevalence of 42.81% in sheep in Shirka District, Arsi Zone, Eastern Ethiopia. Additionally, Oljira *et al*. (2022) observed a prevalence of 20.7% in sheep in Central

Ethiopia through post-mortem examination. Furthermore, Isah (2019) reported a prevalence of 39.1% in sheep in Nigeria through post-mortem examination, which is considerably higher than the current study. These comparisons demonstrate that the prevalence of *Fasciola* in sheep can vary widely based on the diagnostic technique and geographical location (Groning *et al*., 2023).

In cattle, the current prevalence of *Fasciola* compared to other studies such as Tiele *et al*. (2023) reported a prevalence of 8.25% in cattle in Hosanna Town, Southern Ethiopia, which is slightly lower than the current study. Martindah *et al.* (2023) found a prevalence of 4.5% in large ruminants in Indonesia, significantly lower than the current study. In contrast, Yitayal *et al*. (2015) reported a higher prevalence of 32.3% in cattle in Bahir Dar, North Western Ethiopia. Tumusiime *et al*. (2023) found a prevalence of 20% in cattle in Nyagatare District, Rwanda, significantly higher than the current study. Additionally, Isah (2019) reported very high prevalences of 45.7% in cattle in Nigeria through post-mortem examination, which is much higher than the prevalence reported in the current study. Aragaw and Tilahun (2019) found a prevalence of 20.1% in cattle in Bahir Dar, northwest Ethiopia, which is more than double the prevalence found in the current study. These comparisons highlight the variability in *Fasciola* prevalence in cattle across different regions and diagnostic methods.

In goats, the current prevalence of *Fasciola* was lower than some other reports, such as the 8.06%, prevalence found by Ayana and Ifa (2015) in Ambo, Central Oromia, Ethiopia using coprology. Additionally, Isah (2019) reported a much higher prevalence of 35% in goats in Nigeria through post-mortem examination. These comparisons indicate that the prevalence of *Fasciola* in goats varies significantly across different regions and diagnostic methods, with the current study showing a relatively low prevalence compared to these reports.

The prevalence of *Paramphistomum* infections in this study was found highest in cattle (10.71%), followed by sheep (1.79%) and goats (0.89%). This indicates significant variations in infection prevalence among different ruminant species. Compared to other regions in Ethiopia, the observed prevalence in cattle is relatively higher. For example, a study by Tiele *et al.* (2023) in and around Hosanna town, Southern Ethiopia, reported a prevalence of 9.5%. In contrast, several studies have reported higher prevalence. Yitayal *et al.* (2015) and Yihdego (2017) found prevalences of 32.3% and 35.5%, respectively, in cattle in Bahir Dar, North Western Ethiopia. Additionally, Tumusiime *et al.* (2023) observed a 69% prevalence in cattle in Nyagatare District, Rwanda. Differences in prevalence can be influenced by environmental factors (Mas-Coma *et al.*, 2009), diagnostic methods, and veterinary practices. Regions with favorable conditions for intermediate hosts tend to have higher prevalence. Furthermore, post-mortem examinations often report higher prevalence compared to coprological methods due to their better detection sensitivity (Cringoli *et al.*, 2010).

In sheep, the current prevalence observed lower compared to other reports from Ethiopia and beyond. A study by Kifleyohannes *et al.* (2015) in the Tigrai region reported a prevalence of 23.7% in sheep, which is higher than this finding. Similarly, a study by Kebede *et al.* (2023) found a prevalence of 5.4% in small ruminants in Central Ethiopia. These differences might be attributed to variations in study design, diagnostic techniques, or local environmental conditions.

In goats, the current prevalence of *Paramphistomum* was lower compared to other studies. For instance, Alemu *et al.* (2021) in the Amhara region reported a prevalence of 4.5% in goats. Additionally, a study by Chiejina *et al.* (2020) in Nigeria reported a prevalence of 9.3% in goats, indicating higher rate of infections. The lower prevalence in goats observed in this study could be due to differences in grazing patterns, management practices, or environmental factors that reduce exposure to the snail intermediate hosts essential for the *Paramphistomum* life cycle. The variations in prevalence observed in different studies and regions can be attributed to several factors, including differences in ecological conditions, livestock management practices, and the diagnostic methods employed (Mwabonimana *et al.*, 2019).

The prevalence of mixed infections with *Fasciola* and *Paramphistomum* observed in this study was 1.79%, and these were exclusively found in cattle. This prevalence is lower compared to several previous studies. For instance, Tumusiime *et al*. (2023) reported a mixed infection prevalence of 15.9% in cattle in Nyagatare District, Rwanda using coprology. Dey *et al*. (2022) found a prevalence of 4.8% in small ruminants in Bangladesh, while Aragaw and Tilahun (2019) observed a prevalence of 12.2% in cattle in Bahir Dar, northwest Ethiopia. Kebede *et al*. (2023) reported an 8.33% prevalence in cattle in Wolaita, Ethiopia using coprology. These differences in prevalence can be explained by ecological conditions, veterinary and management practices, and diagnostic sensitivity. Regions with favorable conditions for both *Fasciola* and *Paramphistomum* intermediate hosts tend to have higher mixed rate of infections (Kaplan and Vidyashankar, 2012; Taylor *et al*., 2015).

In this study, no significant difference in the prevalence of trematode infections was observed among different locations in and around Bishoftu. This consistency can be attributed to the similar ecological conditions favorable for snail and parasite transmission across the locations. Keyyu *et al*. (2005) found no significant regional differences in *Fasciola* prevalence in Tanzania, likely due to similar environmental conditions and livestock management practices. Mage *et al*. (2002) similarly reported comparable prevalences of *Fasciola* in cattle across different regions in Rwanda due to similar ecological conditions. In contrast, Dinka and Hika (2013) found that location had a significant effect on the prevalence of *Fasciola* in Bishoftu, possibly due to climate changes and increased human intervention that facilitate the distribution of snail intermediate hosts. Walker *et al*. (2008) in Kenya and Phiri *et al*. (2005) in Zambia also found significant regional differences, linked to varying environmental conditions and human interventions.

This finding showed no significant difference in the prevalence of *Fasciola* and *Paramphistomum* between sexes, likely because both sexes have equal exposure. Turuna and Adugna (2019) who found no significant association between sex and *Fasciola* and *Paramphistomum* infections in Nekemte, Ethiopia support this. Khan *et al*. (2009)

similarly reported no significant difference in fasciolosis prevalence between male and female animals in Pakistan. Bhutto *et al*. (2012) also found no significant association between sex and paramphistomosis in buffaloes in Pakistan. In contrast, Rizwan *et al*. (2021) reported a significantly higher prevalence of fasciolosis in female animals, possibly due to hormonal and physiological factors affecting immunity. Mbaya *et al*. (2010) in Nigeria and Ahmed *et al*. (2007) in Egypt also reported higher prevalence in female cattle and sheep, respectively, potentially due to increased stress and hormonal fluctuations.

Sheep showed the highest prevalence of *Fasciola* infection (14.29%), followed by cattle (8.93%) and goats (3.57%). Conversely, *Paramphistomum* prevalence was highest in cattle (10.71%), with (1.79%) and goats (0.89%) showing much lower prevalence. This finding aligns with Kipyegen *et al*. (2022) in Kenya and Megersa *et al*. (2024), who reported higher prevalence of *Fasciola* in sheep and *Paramphistomum* in cattle, respectively. In contrast, Isah (2019) from Nigeria found no significant difference in prevalence among different ruminant species, indicating regional management practices and environmental factors might influence these results. The significant effect of ruminant species on both *Fasciola* and *Paramphistomum* infections can be attributed to differences in physiological susceptibilities. Host specificity in *Fasciola* spp. infections is influenced by various factors, including genetic diversity, immunological differences, and environmental conditions (Rekani and Mero, 2023).

Age had no significant effect on the prevalence of *Fasciola* in this study. Tumusiime *et al*. (2023) from Rwanda similarly reported no significant association between age and *Fasciola* prevalence. However, younger ruminants had significantly associated with *Paramphistomum* infection compared to older ones, possibly due to weaker immune systems and higher likelihood of consuming contaminated feed or water. Dorny *et al*. (2011) also reported higher prevalence of *Paramphistomum* in younger ruminants in Vietnam. In contrast, Khedri *et al*. (2015) found no significant correlation between age and *Paramphistomum* infection in ruminants in South-Eastern Iran, suggesting that regional differences in parasite exposure and management practices might play a role.

Poor BCS had significantly associated with both *Fasciola* and *Paramphistomum* infection compared to those with medium and good BCS. Poor nutrition weakens the immune system, making animals more susceptible to infections (Chandra and Newberne, 2012). Additionally, animals in poor condition may be more exposed to contaminated environments due to lower grazing selectivity. Mwabonimana *et al*. (2019) found a significant association between poor BCS and higher prevalence of *Fasciola* and *Paramphistomum* in Tanzania. Alemu *et al*. (2021) similarly reported higher prevalence of fasciolosis in animals with poor BCS in Ethiopia. Conversely, Nwosu *et al*. (2006) found no significant correlation between BCS and fasciolosis in Nigeria, suggesting other factors like breed and management practices might play a more crucial role. Terefe *et al*. (2012) also did not find a significant association between BCS and *Paramphistomum* infection, indicating that even well nourished animals were susceptible, possibly due to high environmental contamination (Haenni, 2020).

This study showed a significant association between season and *Fasciola* prevalence, with higher susceptibility of infection in the semi-dry season. The semi-dry season may provide optimal conditions for snail populations and parasite transmission, with increased moisture facilitating the life cycle of *Fasciola* (Purwaningsih *et al*., 2022). This finding is supported by Mwabonimana *et al*. (2019) in Tanzania, who found a significant association between the rainy season and higher prevalence of *Fasciola* infections, and Alemu *et al*. (2021) in Ethiopia, who observed higher prevalence of fasciolosis during the rainy season due to favorable conditions for snail intermediate hosts. In contrast, Tumusiime *et al*. (2023) reported no significant association between season and the prevalence of *Fasciola* infections in cattle in Rwanda, suggesting year-round transmission, and Ahmed *et al*. (2007) found no significant seasonal variation in fasciolosis prevalence in sheep in Egypt, indicating continuous transmission. No significant effect of season on *Paramphistomum* infection was observed in this study, whereas Khedri *et al*. (2015) reported a significant association between season and *Paramphistomum* prevalence in South-Eastern Iran, could be influenced by various factors such as specific environ mental conditions, parasite life cycle dynamics, host population dynamics, and agricultural practice (Pokharel and Sharma, 2023).

5.2. Malacology Study

The findings from this malacology study in and around Bishoftu highlight the significant role of various snail species in the transmission of trematode infections among ruminants in this area. *Lymnaea natalensis* the most dominant species, accounting for 37% collected snails, aligning with its established role as the primary intermediate host for *Fasciola* species, reflected in the 8.93% prevalence of *Fasciola* infections found among ruminants (Ngcamphalala *et al.*, 2022). The distribution of other snail species, such as *Physa* spp. (21.25%) and *Bulinus (B.) truncatus* (15.75%) collected were also noteworthy. Although *Physa* spp are not well-documented snail intermediate hosts for *Paramphistomum* (Nguhiu-Mwangi *et al.*, 2019), the presence of snails such as *Lymnae spp*. and *Bulinus* spp. (Sibula *et al.*, 2024) indicates potential roles in the transmission of *Paramphistomum*, observed at a 4.46% prevalence. Additionally, the presence of *Biomphalaria pfeifferi* (9.5%) and *Bulinus (P.) africanus* (6.5%), further underscores the diversity of snail intermediate hosts in the area.

These findings are consistent with other reports from Ethiopia. For instance, recent surveys conducted by Yihenew *et al*. (2020), Girma *et al*. (2021), and Mekonnen *et al*. (2021) also reported *Lymnaea natalensis* as the predominant snail species in various parts of the country. However, the lower prevalence of *Biomphalaria pfeifferi* in this study contrasts with findings from neighboring countries such as Kenya and Uganda, where *B. pfeifferi* is more prevalent (Kariuki *et al*., 2004; Stothard *et al*., 2002). These differences may be attributed to variations in environmental conditions and snail habitat preferences across regions.

The observed overall rate of infection of 7.75% aligns with the range reported in several recent malacological surveys conducted across different parts of Ethiopia and other parts of the world. For instance, a study conducted in the Gilgel Gibe and Meskan districts reported an rate of infection of 33% among snails (Mereta *et al*., 2023). Additionally, research in the Awash River Basin documented an rate of infection of 3.6% (Mereta *et al*., 2019). These findings suggest that the level of trematode infection in freshwater snail populations in Ethiopia is within the typical range observed in other East African countries. For example, a study in the Iringa and Arumeru districts of Tanzania reported an infection prevalence of 6.6% among collected snails (Nzalawahe, 2021), while a study in Kenya reported a much higher cercarial shedding rate of 62.4% (Joof *et al*., 2020). The notable differences in rate of infections between these studies could be attributed to variations in environmental conditions, snail species, and sampling methods.

However, rate of infections with cercaria varied significantly among snail species. *Lymnaea natalensis* exhibited the highest rate of infection at 13.51%, followed by *Bulinus (B.) truncatus* at 11.11%. These results highlight the importance of these species as intermediate hosts for trematodes. The high rate of infection in *L. natalensis* is particularly noteworthy as it is a primary host for *Fasciola* species, responsible for fascioliasis in both humans and livestock. Similar high rate of infections in *L. natalensis* have been reported in other studies in Ethiopia, emphasizing its role in the epidemiology of fascioliasis (Solomon *et al.*, 2017).

The study also found significant differences in rate of infections based on habitat type and season. Snails collected from ponds and wetlands had the highest rate of infections, at 13.48% and 10.67%, respectively. These environments provide stable conditions and abundant vegetation, which are conducive to the proliferation of snails and the development of trematode larvae. These findings are consistent with those from tropical regions where similar habitats are associated with higher snail rate of infections (Yilma and Malone, 1998; Phiri *et al.*, 2005).

Seasonal variation in rate of infections was also observed, with higher rates during the semi-dry season (12%) compared to the dry season (3.5%). This seasonal pattern may be due to changes in snail population density and environmental conditions that favor cercarial release during the wetter semi-dry season. These observations align with those reported by Nwoko *et al*. (2023) in South Africa, where seasonal changes significantly influenced snail rate of infections.

Globally, the study's findings underscore the importance of local ecological conditions, snail population dynamics, and water contact patterns in determining the distribution and rate of infections of snail hosts. For example, lower rate of infections in lakes and rivers in Bishoftu contrast with higher rates reported in lake-dominated regions in South America (Mas-Coma *et al.*, 2009), suggesting that targeted local interventions are crucial for effective control.

Recent studies have further highlighted the impact of climate change on snail-borne diseases. Changes in temperature and precipitation patterns can influence snail habitats and their population dynamics, potentially altering rate of infections. For instance, studies in Tanzania have shown an increase in snail populations and rate of infections following periods of heavy rainfall (Nzalawahe, 2021). Similarly, research in China has demonstrated that warmer temperatures can extend the breeding season of snails, leading to higher cercarial shedding rates (Zhou *et al.*, 2019). These findings suggest that climate change could exacerbate the spread of trematode infections, making it essential to incorporate climate data into future monitoring and control strategies.

5.3. Questionnaires Survey

The questionnaire survey revealed varying levels of knowledge about trematode infections among livestock farmers in Bishoftu area. While a majority of respondents had heard about trematode infections, specific knowledge about snail intermediate hosts and symptoms was limited. Attitudes towards trematode infections showed a high level of concern for livestock health, but beliefs about the effectiveness of traditional treatments were mixed. Preventive practices were mainly focused on regular deworming, with less emphasis on environmental management strategies.

The mean knowledge score of 47.53% observed in this study is lower compared to findings from regions with more extensive veterinary extension services. For instance, Singh *et al.* (2018) reported that over 80% of respondents in the Punjab region of India were aware of trematode infections and their transmission pathways. This higher awareness is attributed to ongoing educational programs and better access to veterinary information. This contrast underscores the importance of targeted educational programs and veterinary intervention program in enhancing farmers' knowledge. A notable finding in this study was the extremely low awareness of aquatic snails as intermediate hosts (0%), which is consistent with a study by Mwabonimana *et al*. (2015) in rural Kenya, where similar gaps were observed. In contrast, a study in Vietnam by Phan *et al.* (2011) reported that 45% of farmers identified snails as intermediate hosts, highlighting regional differences in awareness likely influenced by local education efforts.

The high level of concern about the impact of trematode infections on livestock health (71% of respondents agreed) aligns with findings from Mungube *et al*. (2012) in Kenya, where farmers similarly recognized the impacts of these infections. This dynamic concern reflects a general awareness of the threat posed by trematode infections to livestock productivity. However, beliefs about the effectiveness of traditional methods varied, with only 22% of respondents agreeing that traditional methods are effective. This is lower compared to findings by Swai *et al*. (2005) in rural Tanzania, where 48% of ruminant farmers relied on traditional practices due to limited access to veterinary care. Similarly, Ravindran *et al*. (2014) in rural India found a 55% belief in the effectiveness of traditional methods, indicating a stronger cultural reliance on traditional medicine.

Preventive practices by livestock owners around the Bishoftu area, particularly the high adherence to regular deworming (76%), align with findings from Ibrahim *et al*. (2017) in central Ethiopia, where regular deworming was also a common practice among livestock owners. However, the low engagement in pasture management (12%) and drainage improvement (7%) contrasts with practices reported in South Africa by Van Wyk *et al*. (2000), where more comprehensive environmental management practices are employed to control helminth infections. The use of traditional remedies by 17% of respondents in this study is similar to findings in rural areas of Kenya by Njoroge *et al*. (2004), where traditional practices are still in use, though often supplemented by modern veterinary treatments. Cultural beliefs and practices significantly influence attitudes and preventive measures. Regions with broader veterinary extension services often report higher knowledge and better preventive practices. The lower reliance on traditional methods in Bishoftu compared to Kenya may be due to better access to modern veterinary care. Additionally, the preference for natural treatments and cultural traditions, as described by Ravindran *et al*. (2014), can greatly influence the reliance on traditional versus modern methods.

Furthermore, the findings from this study highlight significant associations between sociodemographic factors and the knowledge, attitude, and practice (KAP) regarding trematode infections among farmers in and around Bishoftu, Ethiopia. The substantial impact of education on all three aspects of KAP suggests that educational interventions could be crucial in enhancing the understanding and management of trematode infections. Specifically, individuals with higher education levels demonstrated better knowledge ($p = 0.045$), more positive attitudes ($p = 0.000$), and more accepted preventive practices ($p = 0.004$). This pattern underscores the pivotal role of education in equipping farmers with the necessary information and skills to prevent and manage trematode infections effectively.

These findings are consistent with other studies conducted in Ethiopia and other parts of the world. For instance, a study by Mekonnen *et al*. (2012) in the Amhara region of Ethiopia reported that education level was significantly associated with farmers' knowledge and practices regarding livestock diseases. Furthermore, in a study conducted in Nigeria, higher education levels were linked to better awareness and practices concerning livestock health among farmers (Afolabi *et al*., 2010). These parallels indicate that educational attainment universally influences farmers' KAP (knowledge, attitudes, and practices) concerning animal health.

The significant association between gender and knowledge scores (p = 0.042) suggests that male farmers in the study area had better knowledge about trematode infections compared to female farmers. This finding aligns with studies from rural parts of Ethiopia, where men typically have more access to agricultural training and extension services (Lemma *et al*., 2017). However, the lack of significant associations between gender and both attitude and practice scores suggests that while men may be more knowledgeable, both men and women hold similar attitudes and engage in similar practices regarding trematode infections. This indicates a potential area for targeted educational programs that can bridge the knowledge gap between genders without altering their generally positive attitudes and practices.

Age was also found to significantly influence knowledge scores (p = 0.011), with older individuals likely having more experience and thus better knowledge about trematode infections. This is in line with findings from Tanzania, where older farmers were reported to have better knowledge about livestock diseases due to their extensive experience in farming (Swai *et al*., 2010). However, similar to gender, age did not significantly influence attitude or practice scores. This suggests that while experience enhances knowledge, it does not necessarily translate to more positive attitudes or better practices, highlighting the need for continuous education across all age groups.

These findings indicate that factors beyond demographic characteristics, such as access to veterinary services, community health programs, and cultural beliefs, may play a significant role in shaping farmers' attitudes and behaviors towards trematode infections. For instance, a study in Kenya found that cultural beliefs and the perceived severity of disease symptoms strongly influenced farmers' preventive practices (Kimeli *et al*., 2013).

6. CONCLUSION AND RECOMMENDATIONS

In conclusion, this study highlights the notable prevalence of trematode infections in ruminants in the wetland areas of Bishoftu, revealing that 11.61%, with *Fasciola* and *Paramphistomum* contributing majorly to the disease prevalence. The study highlighted several key associations, such as higher prevalence of *Fasciola* in sheep and *Paramphistomum* in cattle, as well as the increased susceptibility of young animals and those with poor body condition, and seasonal variations in increasing susceptibility to these infections. The malacology study further underscored the critical role of snail intermediate hosts, particularly *Lymnaea natalensis* and *Bulinus* spp., in the transmission of trematode infections. High cercaria shedding in these snails were linked to specific habitats such as ponds and wetlands, with seasonal fluctuations influencing infection dynamics. The questionnaire survey revealed gaps in knowledge among farmers regarding the transmission and prevention of trematode infections. Despite high levels of concern for livestock health, there was limited awareness about the role of snail intermediate hosts and effective management practices beyond regular deworming. Based on the above conclusion, the following recommendations were forwarded:

- Strengthen veterinary extension services to provide regular deworming programs and promote the use of effective anti-trematodal drugs.
- Implement seasonal monitoring and control programs to address the higher prevalence observed during the semi-dry season, focusing on both livestock and snail populations.
- Conduct targeted educational programs for livestock owner to increase awareness of role of snail intermediate hosts in trematode transmission and avoiding grazing in areas with high snail populations, to reduce the impacts of trematode infections.
- Support ongoing research to explore more sensitive diagnostic methods for detecting trematode infections in both livestock and snail intermediate hosts.
- Studying the impact of climate change on intermediate hosts and trematodes can provide valuable information for the management of the disease.
- Further research on traditional plants used by farmers and explore their potential integration into modern veterinary medicine.

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8. ANNEXES

Annex 1: Interview format to assess the knowledge, attitudes and practices of the farmers regarding the prevention and traditional management of trematode infection

Part I: Basic Information of Socio-demographic characteristic

1. Respondent’s name: ____
2. Address: Zone________District___________Location________Phone No________
3. Gender: [A] Male [B] Female
4. Age: [A] 21-30 years [B] 31-40 years [C] 41 years and above
4. Educational background: [A] Leman [B] Elementary [D] High school or above

Part II: Knowledge about Trematode infections

1. Have you heard about trematode infections?

   [A] Yes [B] No [C] I don't know

2. Are trematode infections caused by:

   I. Contagious bacterial diseases?

   [A] Yes [B] No [C] I don't know

   II. Airborne microorganisms that cause respiratory problems?

   [A] Yes [B] No [C] I don't know

   III. Worms/parasites that live within the internal organs of animals?

   [A] Yes [B] No [C] I don't know

3. Are you aware of trematode infections in ruminants?

   [A] Yes [B] No [C] I don't know

4. Do symptoms of trematode infections include:

   I. Reduced appetite?

   [A] Yes [B] No [C] I don't know

II. Weight loss?

[A] Yes [B] No [C] I don't know

III. Diarrhoea?

[A] Yes [B] No [C] I don't know

IV. Swelling under the jaw (bottle jaw)?

[A] Yes [B] No [C] I don't know

5. Possible sources of trematode infections and transmission include:

I. Contaminated grazing area?

[A] Yes [B] No [C] I don't know

II. Contaminated water?

[A] Yes [B] No [C] I don't know

III. New introduction of animals?

[A] Yes [B] No [C] I don't know

IV. Infected snail?

[A] Yes [B] No [C] I don't know

V. Marketing?

[A] Yes [B] No [C] I don't know

VI. Contaminated air?

[A] Yes [B] No [C] I don't know

6. Are trematode infections most frequent at:

I. During dry season?

[A] Yes [B] No [C] I don't know

II. Shortly after rainy season?

[A] Yes [B] No [C] I don't know

III. During the rainy season?

[A] Yes [B] No [C] I don't know

IV. Similar throughout the year?

[A] Yes [B] No [C] I don't know

7. Can trematode infections be treated with:

I. Antibiotic?

[A] Yes [B] No [C] I don't know

II. Anthelmintic?

[A] Yes [B] No [C] I don't know

III. Multivitamins?

[A] Yes [B] No [C] I don't know

8. What are the impacts of trematode infections? (Select all that apply):

I. Loss due to mortality of animals?

[A] Yes [B] No [C] I don't know

II. Sick animals cause production loss?

[A] Yes [B] No [C] I don't know

III. Lack of draft power?

[A] Yes [B] No [C] I don't know

IV. Loss of body weight of animal?

[A] Yes [B] No [C] I don't know

9. Are you aware of trematode-related zoonotic risks?

[A] Yes [B] No [C] I don't know

Part III: Attitude toward trematode infections

1. Drainage is important for the reduction of snail-born parasitic diseases.

[A] Agree [B] Neutral [C] Disagree

2. I am concerned about the potential impact of trematode infections on the health and productivity of my ruminant livestock.

[A] Agree [B] Neutral [C] Disagree

3. Trematode infection can bring mortality of animals.

[A] Agree [B] Neutral [C] Disagree

4. Animal health is seriously threatened by trematode infections.

[A] Agree [B] Neutral [C] Disagree

5. Concerns regarding trematode infections should be increasing.

[A] Agree [B] Neutral [C] Disagree

6. Traditional methods of prevention and management are effective in controlling trematode infections in my herd/flock.

[A] Agree [B] Neutral [C] Disagree

7. I would be receptive to adopting new practices or technologies recommended by veterinary authorities to prevent and manage trematode infections in my livestock.

[A] Agree [B] Neutral [C] Disagree

8. The animals infected with trematode diseases can be treated and cured.

[A] Agree [B] Neutral [C] Disagree

9. Addressing trematode infections in livestock is a high priority for ensuring food security and sustainable animal production in my community.

[A] Agree [B] Neutral [C] Disagree

10. I am concerned about the potential public health risks associated with trematode infections in ruminants.

[A] Agree [B] Neutral [C] Disagree

Part IV: Preventive practices against trematode infections

1**.** Describe the methods used to prevent and manage trematode infections:

I. Use of traditional herbal/plant-based remedies?

[A] Yes [B] No [C] I don't know

II. Regular deworming with anthelmintic drugs?

[A] Yes [B] No [C] I don't know

III. Pasture/grazing management to reduce snails?

[A] Yes [B] No [C] I don't know

IV. Improving drainage and water sources?

[A] Yes [B] No [C] I don't know

2. Describe any traditional trematode management practices used in your area:

I. Use of specific plant parts (leaves, roots, seeds) with claimed anthelmintic properties? [A] Yes [B] No [C] I don't know

II. Traditional practices involving ritual or spiritual elements (e.g., chants, prayers)?

[A] Yes [B] No [C] I don't know

III. Home-made concoctions using herbs, spices, other ingredients?

[A] Yes [B] No [C] I don't know

3. If (**Q 2.I**), is **Yes**, could you list the medicinal plants that are now used in traditional medicine in your area?

4. How effective have the traditional treatments been in your experience?

I. Observed reduction in clinical signs and improved health?

[A] Yes [B] No [C] I don't know

II. Reduction in frequency or severity of infections?

[A] Yes [B] No [C] I don't know

III. Limitations in quantifying effectiveness compared to conventional treatments?

[A] Yes [B] No [C] I don't know

5. Why do you use traditional remedies instead of/in addition to modern treatments?

I. Lower cost and better accessibility of traditional remedies?

[A] Yes [B] No [C] I don't know

II. Preference for "natural" treatments and mistrust of modern pharmaceuticals?

[A] Yes [B] No [C] I don't know

III. Cultural beliefs and traditional knowledge passed down?

[A] Yes [B] No [C] I don't know

6. Where did you learn about the traditional trematode control methods?

I. Knowledge acquired from elders or experienced farmers?

[A] Yes [B] No [C] I don't know

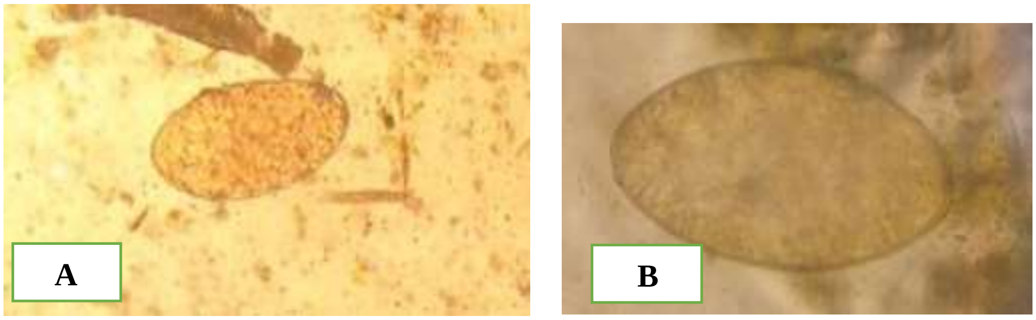
II. Sharing of traditional practices among fellow livestock keepers?

[A] Yes [B] No [C] I don't know

III. Self-experimentation and observation of outcomes?

[A] Yes [B] No [C] I don't know

THANK YOU FOR INTEREST!



Annex 2: Egg of *Fasciola* and *Paramphistomum*

(A: *Fasciola*, B: *Paramphistomum*)

Annex 3: Cercaria of trematode








Annex 4: Pictures taken at sampling sites in the different Water body around Bishoftu

A: *Baala Haadha Bocoo (Indiriffaa) (Insilii) B:Hoolotoo*

Annex 5: Some of traditional medicinal plants claimed anthelmintic properties used by farmes in the study area.

Annex 6*:* Shell of snail collected from study area

Annex 7: Daily follow up of snail for cercaria shedding

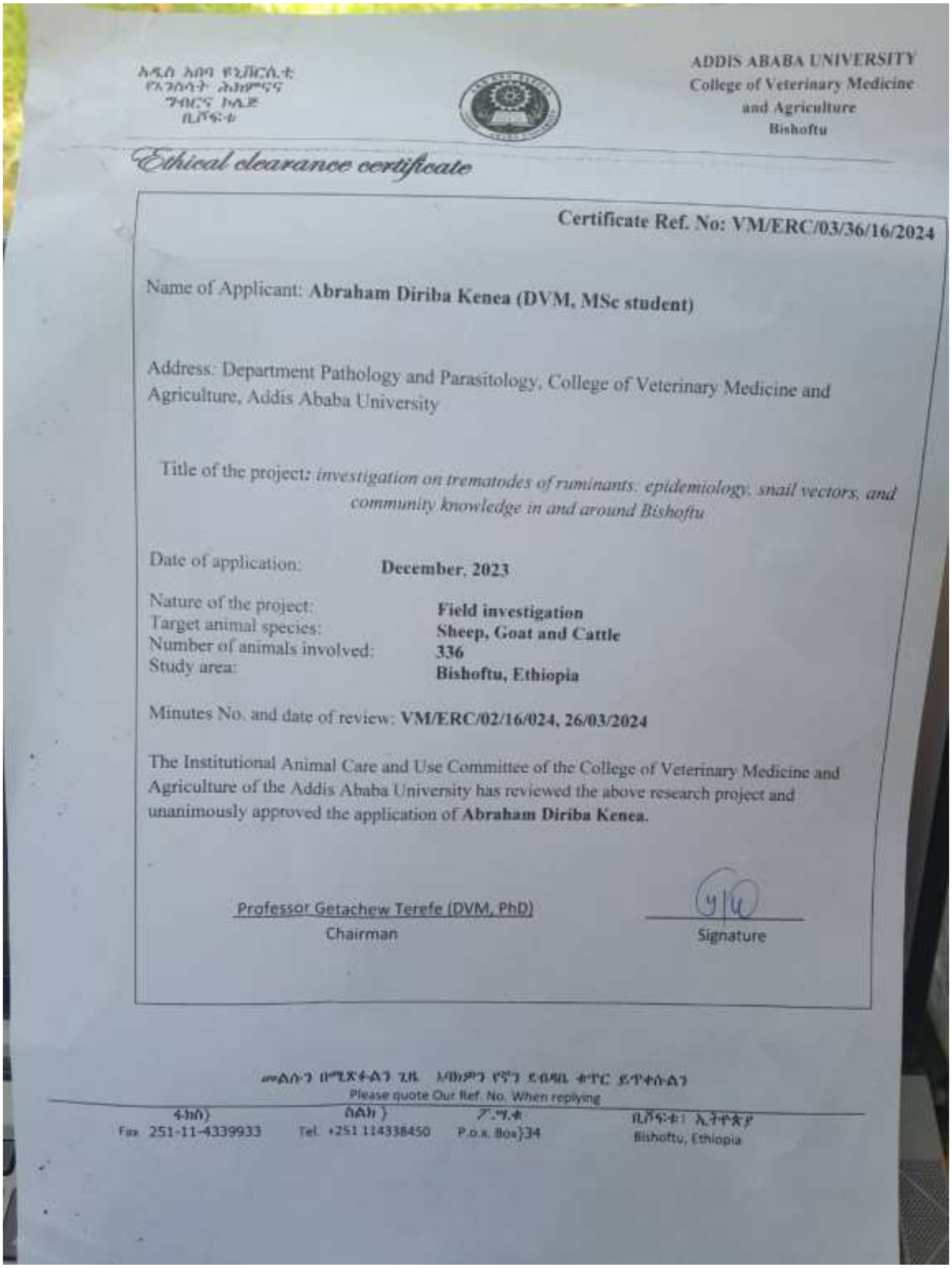
Annex 8: Format of faecal sample collection

L/N	Owner Name	Address	Spp	Breed	Sex	Age	B C	Result	Date
1									
2									
3									
4									
5									
6									
7									
8									
9									
10									

Annex 9: Format of snail sample collection

L/N	Location	Water body type	Season	Spp of snail identied	Infection status
1					
2					
3					
4					
5					
6					
7					
8					
9					
10					

Annex 10: Ethical clearance certificate



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ADDIS ABABA UNIVERSITY
College of Veterinary Medicine
and Agriculture
Bishoftu

Ethical clearance certificate

Certificate Ref. No: VM/ERC/03/36/16/2024

Name of Applicant: **Abraham Diriba Kenea (DVM, MSc student)**

Address: Department Pathology and Parasitology, College of Veterinary Medicine and Agriculture, Addis Ababa University

Title of the project: *investigation on trematodes of ruminants: epidemiology, snail vectors, and community knowledge in and around Bishoftu*

Date of application: **December, 2023**

Nature of the project: **Field investigation**
Target animal species: **Sheep, Goat and Cattle**
Number of animals involved: **336**
Study area: **Bishoftu, Ethiopia**

Minutes No. and date of review: **VM/ERC/02/16/024, 26/03/2024**

The Institutional Animal Care and Use Committee of the College of Veterinary Medicine and Agriculture of the Addis Ababa University has reviewed the above research project and unanimously approved the application of **Abraham Diriba Kenea**.

Professor Getachew Terefe (DVM, PhD)
Chairman

Signature

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Please quote Our Ref. No. When replying

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