زانكۆی سەلاحەدین - هەولێر
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Pathological Changes in Sheep and Goats Infected with *Haemonchus contortus(R.).*

Research Project

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Abstract

Haemonchus contortus is a parasitic nematode that infects the abomasum (fourth stomach) of sheep and goats, causing significant damage to the host's health and productivity. The present abstract summarizes the pathological changes that occur in sheep and goats infected with H. *contortus*. The pathological changes in H. *contortus*-infected animals are primarily due to the feeding activity of the parasite, which results in significant damage to the abomasal mucosa. The initial phase of infection is characterized by the presence of adult worms in the abomasum, which feed on the host's blood, leading to anemia and hypoproteinemia. In addition, the presence of the worms causes an inflammatory response, leading to the secretion of cytokines and chemokines that attract inflammatory cells to the site of infection. As the infection progresses, the abomasal mucosa becomes hyperemic and edematous, with thickening of the folds and accumulation of exudate. The inflammatory response becomes more pronounced, leading to the infiltration of the abomasal mucosa by lymphocytes, plasma cells, and eosinophils. In severe cases, the abomasal mucosa may undergo erosion and ulceration. H. *contortus* infection in sheep and goats leads to significant pathological changes in the abomasum, including anemia, hypoproteinemia, inflammation, hyperemia, edema, hyperplasia, and ulceration. These changes can result in significant morbidity and mortality in infected animals, leading to significant economic losses for farmers and the livestock industry.

Keywords: H. *contortus*, abomasum, pathological changes, anemia, hypoproteinemia, parasitic nematode.

Table of contents:

List of figures:

1. Introduction

Small ruminants are economically significant because they provide us meat, milk skin ,and wool. Small ruminant health issues are the most serious problem in sheep and goat production. Gastrointestinal nematodes of farmed livestock cause severe economic losses in pasture-based grazing systems. *Haemonchus contortus* (H. *contortus*)is a nematode parasite of ruminants that causes production losses and illness in sheep and goats. It is one of most dangerous pathogenic nematodes, also known as the barber pole worm, red stomach worms, or wire worms of small ruminants, that lives in abomasum (Alam *et al*., 2020). H. *contortus* is adapted to a wide range of climatic zones but survives best in warm, high rainfall environments. The prevalence of gastrointestinal nematodes (GIN) in tropical and subtropical areas has adversely affected the production potential of sheep and goats(Chaudary *et al*.,2007). Therefore, it appears that ambient relative humidity modifies the impact of rainfall on larval migration through fecal moisture content. Sheep feces may act as a larval reservoir in dry conditions, with infection peaks occurring after rainfall. Larvae survived well in dry feces for a number of days but did not migrate in the absence of rainfall. As a result, rates of fecal desiccation and rehydration on pasture may be very important to understanding the temporal patterns of larval availability(Wang *et al*., 2014). H. *contortus* sucks about 0.05 ml of blood per day by ingestion (Qamar and Maqbool., 2012) and additional blood is lost into the gastric lumen due to mucosal irritation and oozing out from when the parasite is completely detached from the feeding site.

Ascites, weight loss, anemia, submandibular edema, and death are typical clinical indica tors. The pathological consequences of helminth parasitism, as well as the host's ability to develop immunity and express resistance to them, are determined by a variety of factors, including host traits (species, breed, age, physiological status, feeding behavior) and worm characteristics (burden, feeding behavior, lifecycle) (Abosse *et al*., 2022). Young animals frequently exhibit the acute and emaciated form of the illness, whereas adults are more tolerant of infestation. Infection with H. *contortus* caused a significant reduction in the erythrocyte counts, hemoglobin concentrations and PCV and significant increased WBC and eosinophil, also produce significant reduction in total protein, calcium and iron(Alam *et al*., 2020). Clinical signs and fecal examinations are usually used to make a diagnosis of Haemonchus. When the damage is done, eggs are discovered in feces. As a result, ELISA can detect subclinical infection (Tehrani *et al*.,2012). some reports described that goat appeared to be more susceptible to Helminthes than sheep as they appear to develop less immunity but sheep picked more parasites because they predominantly grazed on grass which harbor infective larvae while goat mostly consume browse which is uncontaminated with parasite larvae (Kelemework *et al*., 2016). (Trefe *et al*., 2005) have observed a significantly lower H. *contortus* female worm length, fecal egg count (FEC). This was significantly associated with higher blood eosinophilia, higher packed cell volume (PCV) and increased number of tissue eosinophils and globule leucocytes. The aim of studying the pathological changes in sheep and goats infected with *Haemonchus contortus* is to better understand the mechanisms by which this parasitic nematode causes disease in small ruminants. This knowledge can inform the development of more effective prevention and control strategies to minimize the impact of H. *contortus* infections on animal health and welfare, as well as on the productivity and profitability of the livestock industry. Additionally, a deeper understanding of the pathological changes associated with H. *contortus* infection can inform the development of therapies to treat or prevent the disease in small ruminants. Overall, the aim of this review is to contribute to the improvement of small ruminant health and welfare, as well as to the sustainability and profitability of the livestock industry.

2. The taxonomic position of the nematode is as follows:

Kingdom: Animalia, Phylum: Nematoda, Class: Secernentea, Subclass: Rhabditia, Order: Strongylida, Family: Trichostrongylidae, Genus: *Haemonchus*, Species: H. *contortus* (Shakya, 2007)

3. Morphology of H. *contortus*

The parasites are of yellow color. Body is filiform (slender) tapering towards the anterior end in male and towards both ends in female. Anterior end is relatively wide and blunt. Buccal cavity is small with a conspicuous tooth extending from dorsal wall. Buccal capsules absent. In addition to transverse striations longitudinal lines are also present on the body.

3.1 Male

It has a length of 9.55-11.85 mm and a width of 0.15- 0.29 mm. A bursa can be found at the tail end. The bursa is made up of three lobes: two large lateral lobes and a small dorsal lobe. Dorsal ray is bifurcated and asymmetrical. The external dorsal ray is long and thin. Lateral rays emerge from a single trunk, while ventral rays are fused proximally and separated dorsally. Spicules number two and measure 0.25-0.52 mm in length(Casey, 2014) (Kuchai *et al*., 2012).

3.2 Female

It measures 18.38-24.50 mm in length and 0.32-0.64 mm in width. Vulva is situated in the posterior third of the body at a distance of about 2.11-4.55 mm from the posterior end. The valvular lips are inconspicuous but a linguiform process is invariably present. Vulva is covered with valves. Tail is without a spine. Anus is situated at a distance of 0.35-0.69 mm from the posterior end. Eggs measure 0.45-0.09 mm X 0.03-0.06 mm in dimensions (Kuchai *et al*., 2012).

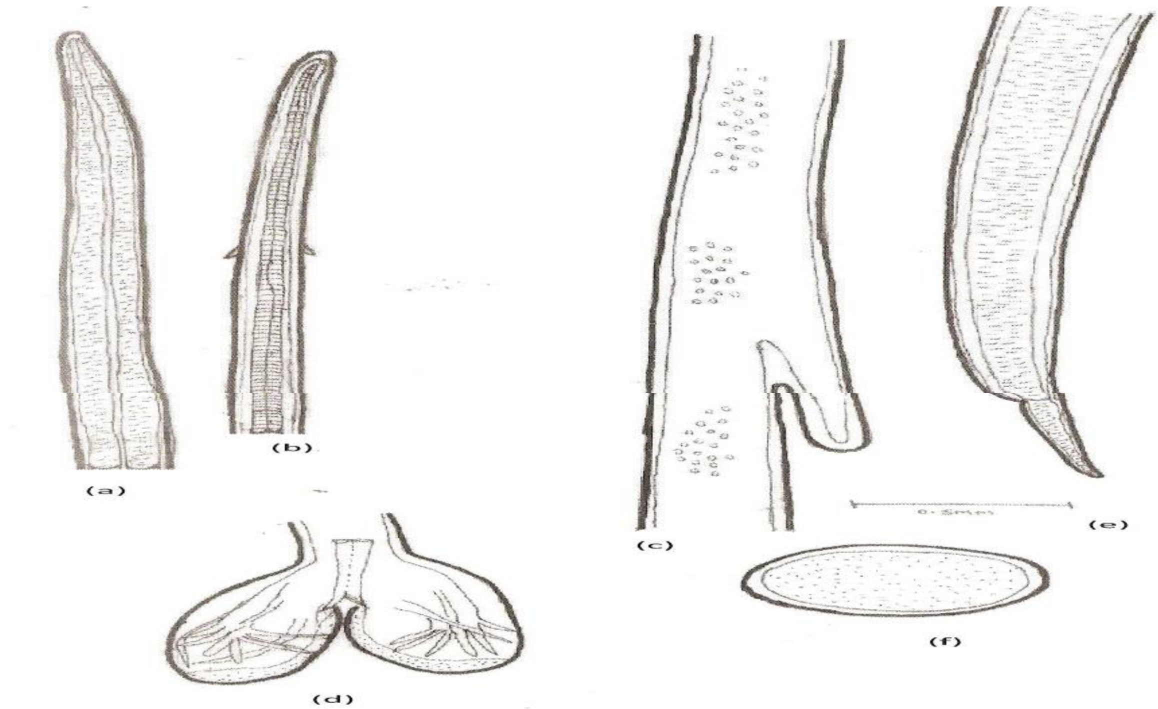

Figure 1: . *Haemonchus contortus* ; A, anterior end of female; B, anterior end of male; C, vulva of female; D, bursa of male showing spicules; E, posterior end of female; F, egg.

4. Life cycle and infectivity of H. contortus

The life cycle of H. *contortus* starts when adult male and female worms reproduce sexually in the host abomasum (Brummett 2019,), with an impressive output per female worm estimated from 5000 to 15,000 eggs/day. The eggs embryonate into first-stage larvae (L1) and second-juvenile stage larvae (L2) within four to six days, and start feeding on bacteria in the dung. Under optimal conditions of 24–29 °C, the L2 larvae molt into the third stage (L3), but they retain the cuticle from the previous molt. During grazing, the infective stage L3 larvae reach the abomasum, shed their cuticle, and burrow into the internal layer of the abomasum and ex-sheath into L4 (pre-adult) stage, usually within 48 h, and finally develop to early (L5) and then late adult worms, which start feeding on blood During the infective L3 or early L4 larval stages, parasites do not proceed directly to the next stage; instead, they remain in the gastric glands of the abomasum. This phenomenon is called developmental arrest (hypobiosis/diapause), and it is described as a "developmental stage in which the parasite becomes dormant, does not cause disease, and metabolically remains inactive". If the conditions outside the host are unfavorable for parasite development, the portion of hypo biotic worms is usually higher so that any egg shed into the environment would be unlikely to develop and survive(Ehsan *et al*., 2020). infection is highest in late summer (mid-July to August) and early fall because *Haemonchus* prefers warm and humid conditions (>25°C). The two biggest sources of pasture contamination with *Haemonchus* eggs are: 1) lambs and kids followed by 2) ewes and does in late gestation and lactation (usually spring). Larvae go into an inactive state inside the animal during winter to survive . In this state, the worms do not lay eggs or cause damage to their hosts. In late April – early May as the worms resume activity, ewes or does develop severe infestations at the same time as late pregnancy or lactation when the animal's immune system is stressed . Ewes/does will contaminate the spring pasture with eggs and immediately expose vulnerable lambs or kids. susceptible hosts, reducing overall levels of pasture contamination, minimizing the effects of parasite burdens, and encouraging the development of immunity in animals. Prompt recognition of factors that favor parasitic infection is essential (e.g., weather, grazing behavior)14. It is also important to distinguish the difference between sheep and goats that are “resistant” and those that are “resilient”. Resistance refers to the ability of the host to actively reduce the number of incoming larvae or worms. Resilience refers to the hosts ability to maintain productivity while tolerating a challenging parasite infection(Simpson).

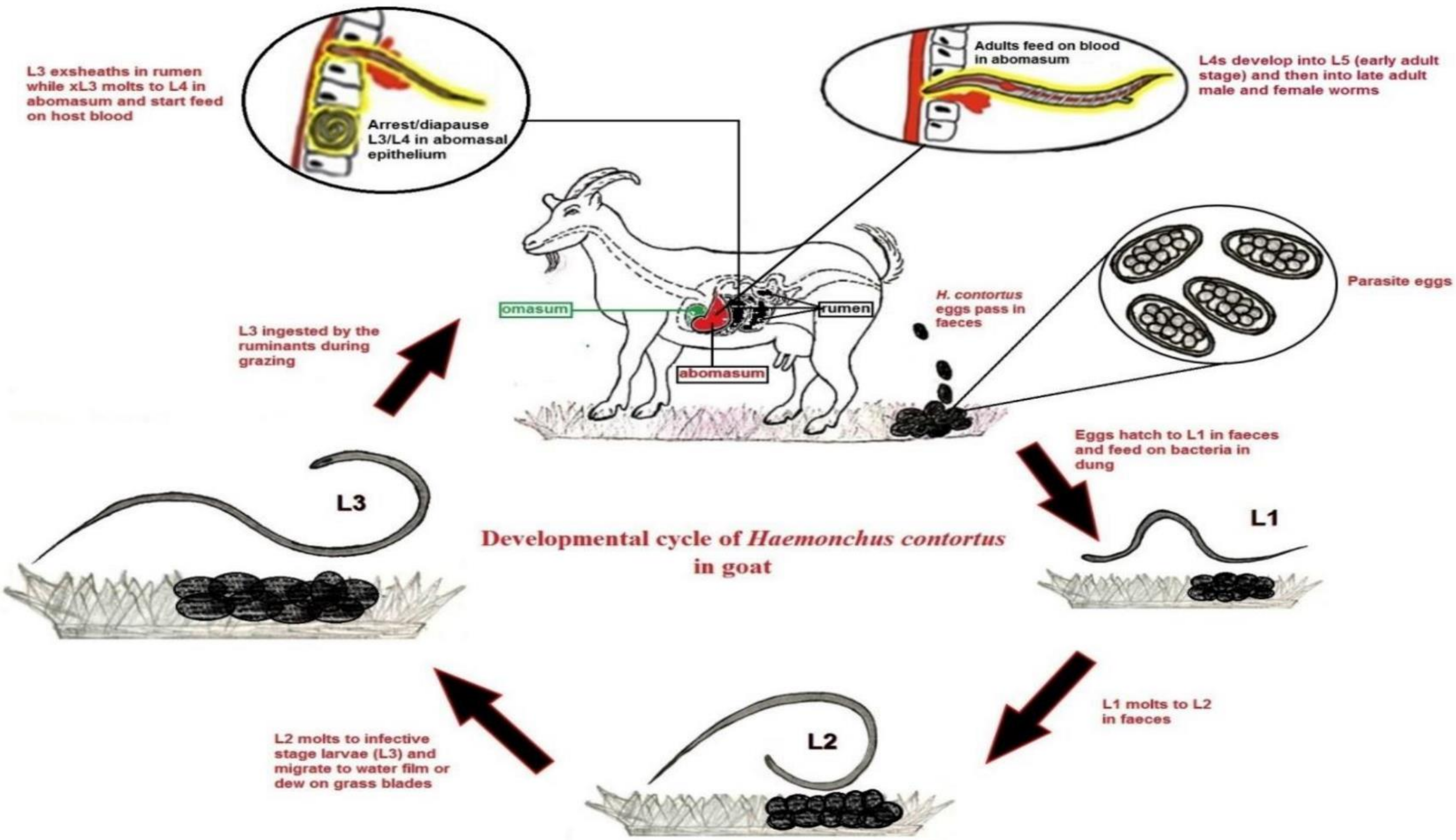

Figure 2: Developmental life cycle of *Haemonchus contortus* parasite in goat.

5. Epidemiology

The free-living stages of H. *contortus* require a warm, humid environment in order to survive and develop outside the host. Haemonchosis can develop in any season if the right circumstances are in place. H. *contortus* thrives and disease prevalence is highest in the tropical climate zones between 23.5° North and South, including South-East Asia, Southern India, Central Africa, and America and Northern South America. H. *contortus* can survive in subtropical areas that are immediately north and south of 23.5° N and 23.5° S, including parts of Australia, Southern Africa, Southern North America, South America, and South-eastern China. Also, Basrah province , Iraq infection with barber pole worm is 12.76%(Abbas and Al-azziz,2018). These areas also have warm, humid summers with heavy rainfall. Temperate zones between 45° and 65° latitude in Sweden, France, Denmark, and the Netherlands are too cool most of the year to permit larval survival and development, whereas warm temperate zones beyond 40° latitude in New Zealand, Northern Europe, Scandinavia, North Asia, and North America have suitable conditions in wet summers and autumns for larval survival. Due to a lack of moisture for H. *contortus* free-living larval stages, Haemonchosis is uncommon in arid areas of the world. However, more rain or irrigation may be able to help larvae survive in hotter arid regions(Flay *et al*., 2022). The fourth larval stage of H. *contortus* ability to experience hypobiosis (Hypobiosis is a characteristic of worm strains or populations that have evolved in particular environments, where the ability to survive)(Swarnkar and Singh, 2015), which helps them endure in cold or dry environments, is also significant. Temperatures between 22 and 26 °C and humidity levels close to 100% are ideal for H. *contortus* egg hatching and larval development. Larvae may stay in desiccated feces during dry weather and emerge after rain, which can lead to an increase in infection. The time it takes for a larva to develop from an egg to an infective third stage larva (L3) depends on the environmental conditions; in an ideal environment, this can happen in four days. Desiccation and low humidity quickly kill both eggs and larvae(Getachew *et al*., 2007). The most hardy, free-living form, the unsheathed L3 larvae retain the cuticle of the second-stage larvae and can survive for extended periods of time under ideal conditions of temperature and humidity. Independent of free water, the L3 migrate randomly up the grass sward in both a horizontal and vertical direction after emerging from the feces. The larvae don't eat and rely on their reserves of energy to survive. Larvae can survive the winter in some climates because they are inactive, use little energy, and can survive in cold temperatures (above freezing). Because the larvae are more active and use up energy stores, the survival of L3 is typically shorter than five weeks in tropical climates. This gives some geographical areas the chance to use spelling of pastures (i.e., times without grazing) as a control method(Flay *et al*., 2022).

6. Pathogenesis and clinical signs

A major factor in the pathogenicity of a *Haemonchus infection* is blood loss. Both the adult worms and the fourth stage larvae are bloodsuckers. They lose every blood component, including red blood cells and plasma proteins, by passing copious amounts of blood from affected sheep through their digestive tract. The calculated average blood loss is 0.05 ml per parasite per day. As a result, infection causes anemia and hypoproteinemia. leakage of proteins to the gastric lumen occurs as a result of the disruption of intercellular unions and increased permeability , epithelial cells loss, tissue reparation, increase in mucus production, and increased requirements for protein synthesis by the immune system . Moreover, many factors prevent such protein losses from being replaced through feeding. Infected animals have lower food intake(Angulo *et al*., 2007). Numerous factors that regulate the dynamic and density of the pathological changes associated with the disease play a role in the pathogenesis of hemochromatosis. These elements include the level of infection, the total number of worms present, the host's nutritional status, host immunity, and the age of the animal(Gidey,2017). Three categories can be used to classify Haemonchosis: acute, hyperacute, and long-lasting.

6.1. Hyperacute Haemonchosis

Although uncommon, the hyperacute form of Haemonchosis is distinguished by a very high infection burden (roughly 30,000 adult parasites), which causes severe anemia and hemorrhagic abomasitis. The affected animals may die suddenly and without warning, while those that do survive experience severe anemia. The most noticeable symptom in these animals is a distinct pallor of the mucous membranes, particularly of the conjunctiva. Due to the severe blood loss, infected animals gradually lose strength and eventually become unable to move. While grazing, they might pass out and pass away. Antiparasitic medication must be taken in order to prevent hypoproteinemia, which is characterized by generalized subcutaneous oedema and severe blood loss. Although not specific to this infection, jaw swelling is a frequent clinical finding, though it is occasionally absent. Haemonchosis does not typically cause diarrhea, and the melaena causes feces to become less frequent, dry, and occasionally dark. Diarrhea may happen when other nematode parasites are also present(Arsenopoulos *et al*., 2021).

6.2. Acute Haemonchosis

Acute Haemonchosis is characterized by lower burdens of H. *contortus* (approximately 2000–20,000 larvae per animal), resulting in a less severe loss of blood and anemia that develops over a longer period of time, compared to hyperacute Haemonchosis. In acute Haemonchosis, Sheep display symptoms of the acute form such as lethargy, lack of appetite, weight loss, thirst, pale mucous membranes, increased heart rate and breathing, and occasionally diarrhea(Iliev *et al*., 2017). A reduction in packed cell volume (PCV) is initially observed, but within the first 14 days of infection, compensatory erythropoiesis kicks in, and over the course of the next six weeks, full compensation and apparent recovery are observed. Three to four days after hemorrhage or hemolysis starts, compensatory erythropoiesis can be seen in most mammals, including ruminants. Until the animal's baseline PCV is reached, compensatory erythropoiesis continues. In sheep infected with H. *contortus*, erythropoiesis and iron utilization have been observed to increase threefold. Therefore, it can be assumed that this compensatory process may lead to the described initial steady normalization of the PCV in the infected sheep. However, the prolonged loss of blood over a period of weeks outpaces the absorption of iron in the intestine, and the gradual depletion of iron reserves results in an iron deficiency condition (Flay *et al*., 2022).

6.3. Long-standing(Chronic) Haemonchosis

Long-lasting Haemonchosis is defined as the infection of ruminants with significantly fewer *Haemonchus* spp. for an extended period of time. Long-lasting Haemonchosis occurs in regions with less ideal conditions for the growth of *Haemonchus* spp. larvae. Clinical recognition of this type of hemochromatosis is frequently missed. Due to the host's poor physical condition, the onset of symptoms is linked to a sudden rise in the parasitic burden or a diminished immune response. Because of the high morbidity, it results in substantial production losses. This form creeps up on the animal and causes cachexia, hyporexia, misshapenness, and even asthenia in some cases. The infested animal no longer follows the herd and presents diarrhea with stools colored black (presence of digested blood). long-standing Haemonchosis is also often associated with blood symptoms with hypochromic microcytic anemia leading to subglossian oedema. (Arsenopoulos *et al*., 2021; DANSOU *et al*., 2021).

6.4. Gross pathological changes in the abomasum

The abomasal mucosa becomes oedematous and congested and exhibits petechial hemorrhages in all *Haemonchus contortus* infected goats. Gross lesions, such as petechial hemorrhages, are frequently observed as a result of the *Haemonchus* parasite attachment and feeding behavior. Compared to the cardiac and pyloric areas, these changes are more pronounced in the fundus. Gastric lymph nodes are also oedematous and markedly enlarged in all infected goats . The abomasal, the content was watery, seldom with foul smelling. In these abomasal, the mucosa had different patterns as multifocal to diffuse thickening and corrugations, focal to multifocal thickening and presence of scattered pale to whitish small nodules in the mucosa (Mesfin,2020). All of the visceral organs were pale after H. *contortus* infection, and the abomasal contents were fluidal and occasionally mixed with free blood with a significant number of adult *Haemonchus contortus* parasites, according to (Biswajit *et al*., 2017). In a few instances, the abomasal mucosa displayed ulcerative hemorrhagic spots where the parasites were found adhered with hyperaemia of the abomasal folds.

7. Diagnosis

The clinical diagnosis of Haemonchosis is based mostly on the detection of anemia in association with a characteristic epidemiological picture, and confirmed at postmortem by the finding of large numbers of *H. contortus* in the abomasum. The detection of impending Haemonchosis relies chiefly on periodic monitoring for anemia, including through the 'FAMACHA' conjunctival-color index, or through fecal worm egg counts and other laboratory procedures (Besier *et al*., 2016) .

7.1. **Direct examination**

Laboratory tests for parasites can confirm the diagnosis of Haemonchosis. The detection and counting of parasitic eggs in fecal samples can be done using qualitative (like the Teleman method) and quantitative (like the modified McMaster method) parasitological tests for the infection. The number of eggs excreted in the feces during experimental infections has been linked to the host's parasite load. Levine claims that 3,000 eggs in fecal samples signify a mild parasite infection while 30,000 eggs signify a severe infection . However, under clinical circumstances, isolated infections with this parasite are uncommon, so this diagnostic strategy is useless. Although genus identification of Haemonchus spp. eggs is challenging, coprocultures make it simple to identify the parasite's L3 larvae .A necessary precondition for the successful diagnosis of Haemonchosis with this approach is the knowledge of L3 morphological features. First, L3 larvae can be distinguished by the filaroid oesophagus (L1 and L2 larvae have a bulbous oesophagus; Figure 3). Further, Haemonchus L3 larvae are recognized by the medium length of their tail (65–82 μm) (Figure 4) and the presence of 16 intestinal cells, whilst no refractile bodies are present (Arsenopoulos *et al.*, 2021).

Figure 3: Haemonchus L2, as seen during microscopic during microscopic observation.

Figure 4: Haemonchus L3 (infective stage), as seen observation.

7.2. **Hematology , and immunological methods**

A complete blood count (CBC) may reveal a variety of changes. From mild to severe, regenerative to non-regenerative anemia can exist. The anemia may be macrocytic and hypochromic if compensatory erythropoiesis is present (as in acute hemochromatosis). Due to iron deficiency brought on by chronic blood loss, anemia may develop that is microcytic, hypochromic, and non-regenerative Sheep with high H. *contortus* worm burdens may have decreased serum immunoglobulins IgG and IgM levels. In addition to commonly used coprodiagnostic methods, a number of bio-chemical and immunological approaches have been developed that aim at the specific diagnosis of infection. These methods are mainly based on the detection and measurement of parameters (e.g., detection of elevated serum pepsinogen and gastrin levels or circulating antibodies) that might be indicative of parasitic infections. immunological methods, including those that are based on the detection of an immune response in an infected animal and those for the detection of parasite antigens, have been developed for the specific diagnosis of parasitic infections Based on the target molecule (antigen or antibody)(Roeber et al., 2013), Experimentally infected sheep with heavy worm burdens show significantly lower IgG (9.36 ± 0.16 g/L) and IgM (0.59 ± 0.01 g/L) concentration compared to controls (IgG 10.35 ± 0.09 g/L, IgM 0.83 ± 0.01 g/L) and sheep with light (IgG 10.22 ± 0.05 g/L; IgM 0.74 ± 0.01 g/L) and moderate (IgG 10.12 ± 0.03 g/L; IgM 0.67 ± 0.01 g/L) burdens of H. *contortus* . This was attributed to immunoglobulins moving into the abomasum's lumen to protect against H. contortus IgG lower motility of its larvae (Flay *et al*., 2022). elevation in the serum IgE level in infected sheep and goats, that may be due to natural infection with Haemonchosis that elevate the production of specific antibodies IgE that associated with worm fertility control, which is characteristic of helminthic Infection (Alam *et al*., 2020).

7.3. FAMACHA system

The name FAMACHA was coined to describe the system evolved for treating only those animals unable to cope with current *Haemonchus contortus* challenge on pasture, by using clinical anemia as the determinant. It is an acronym derived from the name of the originator of the idea, Dr Faffa Malan (FAffa MAlan CHArt) . It is common knowledge that during the course of fatal Haemonchosis the color of the conjunctivae of sheep changes from the deep red of healthy sheep, through shades of pink to practically white, as a result of a progressively worsening anemia. The extent to which these changes relate to a range of hematocrit values (chosen as the Managing ovine Haemonchosis by selective treatment 511 “gold standard” of anemia) was, until recently, still undetermined. The feasibility of grading the degree of anemia clinically in conjunctivalmucous membraneswas investigated by both photographing the mucous membranes and determining the hematocrit of sheep which ranged from very healthy to extremely anemic(Van Wyk and Bath, 2002) .

7.4. Molecular tools

The extraction, culture, and microscopic examination of eggs or larvae from fecal samples in a laboratory are time-consuming traditional methods for determining the type and severity of infection in a sheep flock. The potential for more effective and dependable procedures is provided by developments in molecular technology. Since sequence information from other studies was available, primer/probe sets centered on the ribosomal internal transcribed spacer 2 (ITS2) region were created. As a result, tests utilized both previously published primer/probe sets as well as those created by the Food and Environment Research Agency . The specificity of various primer/probe combinations was examined against DNA isolated from or from H. *contortus* DNA. Using real-time PCR technology, all sets were examined for cross reactivity against four other closely related species. The best primer/probe combination for each species was used to optimize reactions, which were then tested for sensitivity using samples with high concentrations of H. *contortus* DNA. Fecal samples from sheep with H. *contortus* infection were taken, and the eggs were harvested, counted, and DNA was extracted. Real-time PCR reactions were conducted for each using a range of concentrations between roughly 3000 and 50 eggs per sample, and mean cycle time (Ct) values were calculated (Learmoun *et al*., 2009).

8. Control of Haemonchus contortus

different principles of action for controlling gastrointestinal nematodes (GIN) in ruminants, including *Haemonchus contortus*. Here are some strategies mentioned in the paragraph for controlling H. *contortus*:

1- Pharmaceutical Control :There are several classes of anthelmintic drugs effective against Haemonchus; these include benzimidazoles (e.g., albendazole), imidazothiazoles (e.g., levamisole), macrocyclic lactones (e.g., ivermectin), salicylanides (e.g., closantel), amino-acetonitrile derivatives (e.g., monepantel), sporoindoles (e.g., derquantel) . Commercially available preparations may include the above as single active ingredients or in combinations(Arsenopoulos *et al*., 2021).

2- Use of non-chemical anthelmintic materials: using various allopathic, homeopathic, herbal and biological products to control H. contortus. Studies have shown that these materials can eliminate worms and/or negatively affect the parasite's biology (Jaheed, 2021)(Qamar and Maqbool., 2012).

3-Nutritional management: Providing a balanced diet with adequate levels of protein, energy, and minerals can help improve the immune response of the host and reduce the level of H. *contortus* infection.

4- Genetic selection: Selecting animals with genetic resistance to H. *contortus* can reduce the level of infection in the herd or flock over time. This can be achieved through selective breeding programs or by selecting animals with a history of low worm egg counts.

5- Grazing management: Limiting the contact between the hosts and the infective larvae in the field through grazing management methods can reduce the risk of H. *contortus* infection. This can be achieved through techniques such as rotational grazing, mixed grazing with other livestock species, and resting pastures.

It is important to note that the integration of different control tools is essential for the sustainable management of H. *contortus* infections and other GIN infections in ruminants. By combining these different strategies, farmers and veterinarians can reduce the reliance on chemical dewormers, which can help prevent the development of resistance and promote more sustainable control practices(Hoste and Torres-Acosta, 2011).

9. Conclusion

effective control measures such as regular deworming and good management practices are necessary to minimize the impact of *Haemonchus contortus* on the livestock industry. The development of new and effective vaccines and treatment strategies could be critical in reducing the severity of pathological changes in infected animals, leading to improved animal welfare and productivity.

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