13

Fascioliasis

AUTHORS DETAIL

Shadan H Abdullah

Department of Microbiology, College of Veterinary Medicine. Sulaimani University. Iraq
*Corresponding author: Shadan H Abdullah

Received: Sept 25, 2022 Accepted: Oct 2, 2022

INTRODUCTION

Fascioliasis is a zoonotic disease caused by a trematode parasite belongs to the genus *Fasciola* (Bargues et al. 2016). It has a wide range of geographical distribution and found across the world (Charlier et al. 2020). *Fasciola* species are commonly known as liver flukes, as they are leaf shaped (David 1990). The flukes are hermaphrodite and are mainly confined to the bile ducts of the liver or gall bladder of infected hosts. They cause liver-rot in ruminant hosts, that may lead to the death (Khoramian et al. 2014).

Fascioliasis has been listed as a neglected zoonotic disease by the World Health Organization (Mas-Coma et al. 2018). It has a predictable impact on livestock production (Kalu 2015). According to an estimate, over 600 million animals were infected with it worldwide, having an annual estimated economic loss to nearly $3 billion (Toet et al. 2014). The public health risk with Fascioliasis among people was estimated to be about 17 million cases worldwide (Mas-Coma et al. 2009a), and up to 180 million at risk of infection (Mas-Coma et al., 2018).

The global increase in human fascioliasis prevalence rates are greatly correlated with a high proportion of infected ruminant hosts (Ashrafi et al. 2014; Diyana et al. 2019).

Flukes of the genus *Fasciola* has complex life cycle. Their larval stages depend on Lymnaeids snails as an intermediate host for their growth and development (Munita et al. 2019). A wide range of mammals including cattle, sheep, goat, horse (Taylor et al. 2013), buffalo, camel, deer and human serve as the definitive host (John et al. 2019). Donkey and mules can also harbor the flukes and become a reservoirs host (Meray Sierra 2020). In addition, the fluke was reported from pigs, alpacas, kangaroos, wallabies, and rabbits (Alemneh 2019). The occurrence of Fasciolaiasis depends on several factors related to the biology of vectors and parasites, and the management of animal herds (Khoramian et al. 2014). The distribution of each *Fasciola* species depends on the availability of intermediate hosts of Lymnaeid snails (Prasad et al. 2008).

Fascioliasis appears in two forms, acute or chronic, depending on the extent of the disease and the required time for its occurrence (Radiostis et al. 2007). The main economic impact of fascioliasis is the condemnation of the infected liver, along with a decrease in productivity and a reduction in the growth rate of infected animals (Usip et al. 2014).

Fascioliasis is an emerging disease in many countries, especially when there is a tradition of eating uncooked vegetables harboring the infective metacercarial stage (Ashrafi et al. 2006a). So, it is regarded as one of the foodborne diseases with greater pathogenic effects mostly in the acute phase of infection during 3-4 months (Chen and Mott 1990). Global changes appear to have a correlation with the emergence of fascioliasis such as importation/exportation and livestock management (Mas-Coma et al. 2009b), environmental anthropogenic modifications, travel (Ashrafi et al. 2014) and alteration in human diet traditions (Ashrafi et al. 2006a).

Etiology

Fascioliasis is considered as food and water borne zoonotic infection caused by digenean trematodes of the genus *Fasciola* (Alemneh 2019). *Fasciola (F.) hepatica* (Linnaeus 1758) and *Fasciola (F.) gigantica* (Cobbold 1856) are the common and more prevalent flukes causing infection in human and animals (Admassu et al. 2015; Amer et al. 2016). *F. hepatica* is nearly distributed throughout all continents, while *F. gigantica* is mostly restricted to the parts of Asia and Africa (Meray Sierra 2020).

Taxonomy

The parasitic tapeworms belong to the Phylum platyhelminths involve two classes: Class Cestoda (the tapeworms) and Class Trematoda (the flukes). The class Trematoda is further divided into two main subclasses, Monogenea (direct life cycle) and Digenia (involving intermediate host). The trematodes belong to the family Fasciolidae comprising of the parasites of major veterinary importance. According to Urquhart et al. (1996) the taxonomic classification of *Fasciola* is as follows;

Kingdom: Animalia
Phylum: Platyhelminthes
Class: Trematoda
Subclass: Digenia
Order: Echinostomida
Family: Fasciolidae
Genus: *Fasciola*
Species: *F. hepatica*, *F. gigantica*

Citation: Abdullah SH, 2023. Fascioliasis. In: Aguilar-Marcelino L, Younus M, Khan A, Saeed NM and Abbas RZ (eds), One Health Triad, Unique Scientific Publishers, Faisalabad, Pakistan, Vol. 3, pp: 78-85. https://doi.org/10.47278/book.oht/2023.80

Morphology

F. hepatica and *F. gigantica* can be distinguished morphologically based on characteristics of their body length and width (Ashrafi et al. 2006b; Itagaki et al. 2009). The adult fluke of *F. hepatica* is large flattened and leaf-like, anteriorly provided with cone shaped projection followed by a pair of prominent shoulder, with wider and rounded posterior end (Hendrix and Robinson 2006). Flukes are grayish brown in color changing to gray when preserved (Wagari 2021). The adults possess two suckers for attachment. The oral sucker at the anterior end surrounds the mouth and the ventral one, situated on fluke´s ventral surface (Urquhart et al. 1996). The flucks´ tegument is absorptive and armed with backward directed spines, together with the suckers, assist to preserve the parasitic position in the bile ducts by an effective mechanism (Smyth 1994). The muscles lie directly under the tegument, and the organs are packed in a parenchyma since they lack the body cavity. The digestive system starts with oral opening leading into a pharynx, esophagus and a pair of blindly branched intestinal ceca. A large number of ciliated flame cells together forms the excretory system, and the waste metabolic products pass through a connected tubular system and exposed externally. The simple nervous system consists of two anterior ganglia and a pair of longitudinal trunks arising from them (Urquhart et al. 1996; Rickard 2001).
Based on geomorphology, *F. hepatica* is short and possess broad shoulders, whilst *F. gigantica* is elongated and with narrower body (Mas-Coma and Burger 1997; Lotfy and Hiller 2003). *F. hepatica* measures "30- 20 mm × 10 mm" and *F. gigantica* measure "27 to 75mm" × 12mm" (Brown 1980). When hybridization of both species occurs within the host's body, subsequent offspring have intermediate phenotypes (Vara-Del Rio et al. 2007; Beesley et al. 2018). Due to the presence of the intermediate form combining of morphological and molecular techniques for distinguishing of *Fasciola* species is critical especially in regions where fluke species overlap (Haridwala et al. 2021). Both species have the ability to reproduce sexually or through self-fertilization (Shoriki et al. 2014).

Egg

The eggs of *Fasciola* spp. are large in size, oval, yellow brown in color, with a thin shell and possess a distinct operculum. The eggs of *F. hepatica* measure up to "130 to 150 µm" by "60 to 90 µm" (Hendrix and Robinson 2006), and in *F. gigantica* measures up to "120 -180 µm" by "80 - 110 µm" (Phalee et al. 2015). Eggs consist of a fertilized ovum with vitelline cells surrounded with proteinous shell (Andrews 1999). The ova contain one cell stage embryo surrounded by a group of oval body yolk cells. Development of eggs to reach maturation in both *Fasciola* species required 12-16 days, and the miracidia hatch within 4 days after maturation (Hussein et al. 2020).

Miracidium

It has an elongated conical body with a broad anterior end and tapering posterior end, and swims at great speed (Malek 1980). The outer surface cover with numerous cilia, except in lateral connection regions of epidermal plats. These cilia appear longer on the apical parts of both anterior and posterior end than the rest parts of the body, and remain viable for about 9-12 hours (Hussein et al. 2010).

Cercaria

It has a large heart shaped body and simple long tail. The body covered with thick wall and is surrounded by tiny spines all over its surface (Hussein et al. 2010).

Metacercaria

It is spherical white color cyst directly infective to the definitive host. With time it becomes yellow and darker in color after 1 or 2 days, the cyst measures up to "0.26 to 0.30 mm" in diameters, and protected by thick wall capsules of double outer and inner layer for protection against environmental impacts (Phalee et al. 2015).

Transmission

The transmitted vectors for *Fasciola* spp. are amphibious freshwater lymnaeid snails (Mas-Coma et al. 2009a). It was estimated that nearly 30 species of lymnaeid snail are recognized as intermediate hosts for *Fasciola* spp. globally (Vázquez et al.2018). *Galba truncatula* is the common lymnaeid act as a transmitter for *F. hepatica* in endemic temperate and subtropical areas (Artigas et al 2011; Bargues et al 2020).
Different *Lymnaea* species including: *L. cousin, L. columella, L. ollula, L. natalensis,* and *L. viridis* act as an intermediate host for *Fasciola* spp. (Hussein and Khalifa 2008). Both *Radix (R.) auricularia* and *R. natalensis* lymnaeid snails that live in the subtropical and tropics area, can transmit *F. gigantica* (Mas-Coma et al. 2009b). *Biomphalaria alexandrina* has also been reported as a transmitter for *F. gaigantica* (Farag and El Sayad 1995).
Other cosmopolitan freshwater lymnaeid snails which are responsible for transmission of *Fasciola* spp. in different areas include: *Radix rubiginosa, Austropeplea tomentosa, Pseudosuccinea columella, Stagnicola corvus,* and *Hinkleyia caperata* (Vázquez et al. 2018). The high transmission capacity of vectors is connected to the duration and persistence of the life span of the infected snails after infection (Mas-Coma et al. 2001).
Humans act as the incidental hosts for liver flukes (Alemneh 2019). Ingestion of freshwater wild plants including watercress is the main source of infection to humans (Mas-

Coma et al. 2018). In spite of watercress, various freshwater plant species might be involved in *Fasciola* transmission and human infection, which depend mainly on geographical distribution of those plants and the dietary traditions of peoples in that region (Mas-Coma et al. 1999). Water had been mentioned as another source for infection in human, either directly by drinking or indirectly by contaminating vegetables, fruits, and kitchen utensils (Chen and Mott 1990). Humans also become infected with fascioliasis after eating raw dishes prepared freshly from an infected liver with immature flukes (Taira et al. 1997).

Epidemiology

Previous studies revealed that fascioliasis has a higher spreading capacity, which is greatly related to the biological ability of intermediate lymnaeid hosts and the fluke adaptation capacity (Mas-Coma et al. 1999). Due to the ability of parasites and snails to develop in diverse adaptation strategies, the transmission rates become higher (Mas-Coma 1996).

F. hepatica is distributed commonly in Europe (Robinson and Dalton 1999), temperate regions of Asia, Africa, Oceania and America, while *F. gigantica* is mainly restricted to Africa and Asia (Lotfy and Hillyer 2003; Mas-Coma et al. 2009a). Both fluke types appear to be present in the same geographical areas especially in some subtropical and warm temperate regions in Africa and Asia (Mas-Coma et al. 2009b; Kalu 2015).

The larval stages of fasciolids species as well as their intermediate host snails, are highly dependent on climate features, so changes in environmental conditions have an impact on liver fluke infection (Fuentes et al. 2001). The dissemination of fascioliasis to a new geographical area is essentially related to the distribution of intermediate lymnaeid hosts, the presence of an infected definitive host, and the presence of appropriate environments for the snail vector. High lands areas with acid soils, poorly drained marshy grazing field and waterlogged are frequently estimated to be appropriate for their propagation and providing high endemic areas for the development of fascioliases (Ayele and Hiko 2016).

Up to 50% of infective overwinter metacercariae might remain viable on pasture and infect grazing livestock and capable to infect livestock hosts following grazing in next spring (John et al. 2019). Their survival is mainly dependent on dampness and diffident temperature, as they can tolerate repeated freeze-thawing action (Boray and Enigk 1964).

Metacercariae of *Fasciola* species might remain viable for more than one year, occasionally for up to two years with infectivity to induce infection in definitive hosts. Additionally, metacercariae from different livestock species origins do not show significant differences in definitive host infectivity (Valero and Mas- Coma 2000; Valero et al. 2002).

The occurrence of fascioliasis in humans has increased in the past 20 years, due to the global increase in the number of infected humans and animals (Alemneh 2019). Previous studies have demonstrated the significant role of human in the spreading of fascioliasis, especially in hyperendemic zones (Esteban et al. 1997), particularly where outdoor defecation is practiced (Mas-Coma et al., 1999), or where the correct services for waste and sewage disposal are absent (Hillyer and Apt 1997).

Life cycle

Fasciola spp. have a complex life cycle requiring the mammalian definitive hosts and a freshwater snail as an intermediate host (Vázquez et al. 2018). In subtropical areas, infection persist during the whole year but significantly slow down during winter (López Lemes et al. 1996). The essential point in trematode life cycle is that, one egg of trematode ultimately develops into hundreds of adults, when it passes through paedogenesis phenomenon in the body of snail intermediate hosts (Alemneh 2019).

The flukes are oviparous: the mature adult in bile ducts of definitive host lay eggs with an operculum. Eggs are transported from the bile medium to the small intestine where they mix up with feces (Nyindo and Lukambagire 2015).

In ruminant, eggs are dropped with feces on to the pasture, and undertake embryonation to the pyriform ciliated larva called a miracidium. Hatching of embryonated eggs can happen in response to the outside stimuli such as light, humidity and temperature (Vázquez et al. 2018). The developing free-swimming ciliated miracidia must find a suitable lymnaeid snail intermediate host for its further development (Urquhart et al. 1996; Graber et al. 2005). It was believed to use chemotactic and phototactic movements for vector finding in less than 24 hours (Vázquez et al. 2018). Upon contact, the miracidia mechanically attack the soft tissues of snail hosts by the effects of proteolytic enzymes and their penetrating styles (Zhang et al. 2019).

The entire penetration process occurs within thirty minutes and later on the miracidium loss its tail and cilia and changes to an elongated saclike structure named sporocyst, that contain a number of germinal cells. These cells undergo a development to the next stage, the redia which migrate to the hepato-pancreatic region of the snail and ultimately leads to the formation of cercaria. The second generation of redia may form during unfavorable environmental conditions. The cercaria is the young flucks with long tail arises actively from the snail in considerable numbers. The majority of infected snails die prematurely due to the disruption in their hepato- pancreas (Urquhart et al. 1996; Rickard 2001; Graber et al. 2005).

The development of flukes inside the snail required about 6 weeks depending on the environmental temperature (Beesley et al. 2018). The asexual development of parasite inside the snail refers as “clonal expansion”; a single miracidium can produce nearly ten to seventy hundred cercaria (Graczyk and Fried 1999).

Finally, the cercaria locates the wet leave of vegetations by negative geotactic movements and attach themselves, shed their tail and metamorphose into metacercariae.

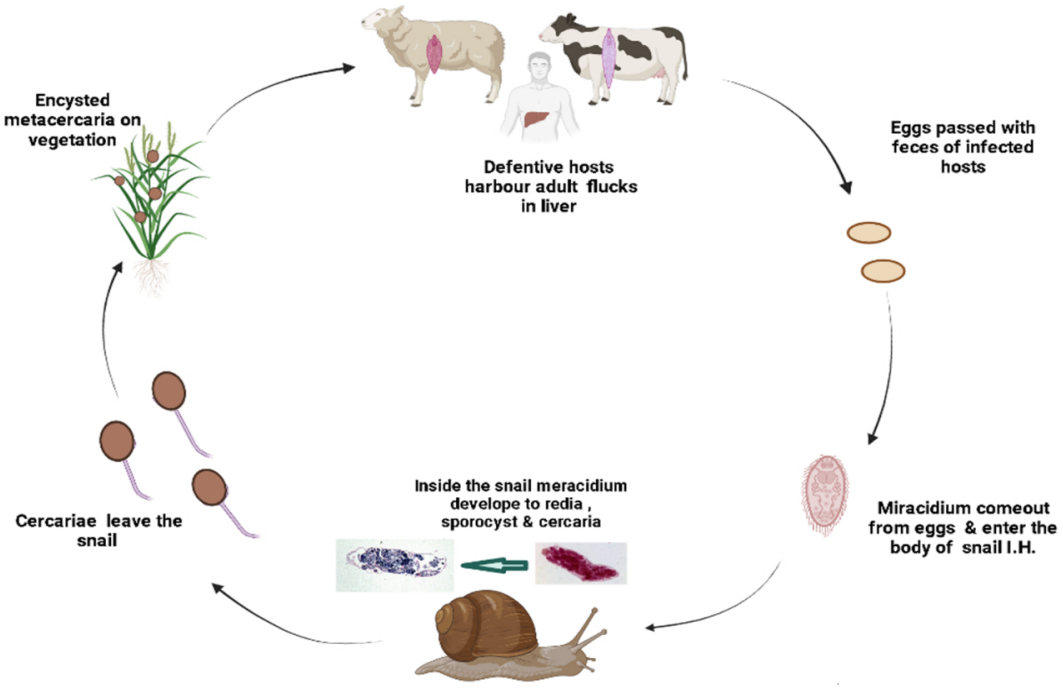

Fig. 1: Life cycle of *Fasciola* spp.

Encysted metacercariae have a great possibility of survival (Nyindo and Lukambagire 2015).

Metacercariae are the infective form of flukes and upon ingestion by suitable definitive hosts, an immature fluke might liberate. During mastication the outer cyst layer is removed, and inner cyst ruptured in the intestine depending on the enzymatic hatching mechanism, which is activated by suitable oxidation reduction and CO_2 system provided by intestinal environment (Urquhart et al. 1996; Rickard 2001; Graber et al. 2005).

The juvenile flukes burrow through the wall of the small intestine and temporarily settle in the peritoneal cavity for several hours (Atalabi and Lawal 2019). Afterward it migrates and penetrate the liver during four to six days, wounder there for another four to seven weeks leading to the entrance in the bile ducts, settle down and lay eggs after sexual reproduction. The life cycle reinitiates, when it lives for several years (Urquhart et al. 1996; Rickard 2001; Graber et al. 2005). The adult worm produces various number of eggs per day in different definitive hosts; reports have shown that in cow, it extruded 25,000 eggs and in sheep 12,000 eggs (Valero et al. 2002). Fig. 1 demonstrate different life cycle stages of *Fasciola* spp.

Pathogenesis

Pathogenesis occurs in two phases: the first phase is acute fascioliasis that occurs after liver penetration by enormous parasitic stages within a short period of time, and migration through the liver parenchyma. Its outcome is the severe liver damage and hemorrhage with subsequent sudden death mainly in sheep. The second phase is chronic fascioliasis, happen when fewer numbers of fluke result in infections over the long period of times even in weeks or months. The adult flukes reach the bile ducts, and result in the damage of the biliary mucosa by their cuticular spines. Sometimes, acute and chronic infections can occur simultaneously. The subclinical form is a common type of fasciolosis, occurring as a result of infection with low numbers of fluke, which accompanying reduction in weight gain and wool quality (Hayward et al. 2021). In both acute and chronic phases, the disease demonstrates high pathogenicity and immunosuppressive capacity (Valero et al. 2003; Girones et al. 2007).

Other pathogenic effects concurrent with fascioliasis include traumatic hepatitis and hemorrhage caused by juvenile flukes and fibrosis of the migratory tracts that eventually calcifies, caused by adult flukes. Moreover, anemia and hypoalbuminemia might occur (Roberts and Suhardono 1996; Javid et al. 2011).

In humans, the complexity of fascioliasis is sometimes related to the capability of the flucks to invade vital organs, leading to the significant outcome and even death of the patient (Mas-Coma et al. 2014).

Furthermore, the metabolites release from the liver flukes into the host circulatory system associated with anemia, increases the concentration of serum enzymes and dysfunction of the adrenal and thyroid glands (Sharma et al. 2011).

The pathogenicity of liver fluke infection can be affected by numerous factors including the breed of host, body

condition, dietary status and the burden of infection (Chauvin et al. 2001).

Clinical Signs

Fascioliasis is associated with significant morbidity and mortality in livestock (Hosseini-Safa et al. 2019). Acute phase often distinguished by sudden death of up to 10% of the flock, due to high levels of blood loss from physical damage to the liver. Typical clinical signs primarily in sheep and goats include reduced appetite, abdominal pain, depression, anemia, weight loss, and sudden death in a few days. Secondary bacterial infection of liver by *Clostridium noyvi*, during the acute phase resulting in clostridial necrotic hepatitis (Lalor et al. 2021).

During the chronic phase, additional clinical signs appear, such as inappetence and lower weight gain, anemia, and ascites (Urquhart et al. 1996; Rickard 2001), decrease in milk yield, diarrhea, and submandibular edema (Fufa 2009). Emaciation during chronic fascioliasis is prominent, especially in more susceptible animals and ewes during the advanced gestation period. The Inflammatory mediators arising from liver damage could have an effect on early pregnancy (Sargison and Scott 2011). Liver fluke infection is also considered as a predisposing risk factor for mastitis (Mavrogianni et al. 2014). The flukes incidentally infect the peritoneal cavity, lungs, subcutaneous tissue, lymph nodes, eye and other locations (Hosseini-Safa et al. 2019). In humans, various complex clinical disorders appear including severe neurological, psychiatric and ophthalmological conditions (Mas-Coma et al. 2014), during the acute phase of infection caused by migration of numerous juvenile parasitic stages (Gonzalez-Miguel et al. 2019).

Diagnosis

In endemic areas, rapid and accurate diagnosis for animal fascioliasis is considered as a successful prevention and treatment measure. Although there is significant progress in the application of new therapeutic agents, little attention has been paid to confirm the diagnosis of fascioliasis in animals (Amiri et al. 2021). Fascioliasis has been diagnosed by parasitological, immunological and molecular methods (Atalabi and Lawal 2020).

Generally, fascioliasis is diagnosed by fecal testing and finding eggs of parasitic flukes in stool, bile or duodenal fluid through wet mount and/or concentration techniques such as formalin- ether.

The expertise of the examiner and the number of parasite eggs in the stool sample are the main disadvantages associated with previous diagnostic techniques. In addition, a number of serologic procedures such as IFA, IHA and ELISA are relevant for diagnosis of fascioliasis during different stages of the disease (Hamoo et al. 2019).

Serological methods give the advantages for early diagnosis of fascioliasis, however circulating antibodies could persist in the blood for several months after effective treatment (Salimi-Bejestani et al. 2005; Arifin et al. 2016).

Moreover, the nucleic acid-based techniques appear to be expectant for diagnosis of recent fascioliasis (Rojas 2014; Davies Calvani et al. 2018). Various molecular procedures are applicable for diagnosis of fascioliasis, i.e., nested PCR provide higher sensitivity than existing diagnostic methods, when fascioliasis could be detected in the feces of infected sheep two weeks post infection (Martınez-Perez 2012; Beesley et al. 2018). Furthermore, sequencing the whole genome, and polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) assay can also be used for diagnosis (Hamoo et al. 2019). Loop mediated isothermal amplification (LAMP) is an alternative technique, because molecular diagnostic techniques using PCR are not available everywhere (Martınez-Valladares and Rojo-Vazquez 2016). It is with low-cost and simple performing test, which permits quick amplification of small amount of DNA with high sensitivity (Amiri et al. 2021) and can be applied for diagnosis of a variety of zoonotic helminths including *Fasciola* species (Ai 2010). It has been recognized to be more sensitive and specific by detecting fascioliasis one-week post infection in experimentally infected sheep (Martınez-Valladares et al. 2016).

Treatment

The recommended treatment depends on the nature of the disease. Some of the existing anti helminthic drugs are not effective against immature flukes, so these are not recommended during acute flukes outbreak. The commonly used flukicides is Triclabendazole, which is effective against both immature and adult flukes (Ahmed et al. 2005). Triclabendazole is also an efficient drug available for human treatment (Gandhi et al. 2019).

Control

The control strategy should be directed at the application of preventive measures rather than a curative basis. The effective control measures include the treatment with appropriate anthelmintics drugs to decrease the number of parasitic flukes in the host body and the number of fluke eggs in the pasture, reduction in the number of snail intermediate host by using mollucicided and improvement of drainage (Ahmed et al. 2005; Fufa 2009). Other control measures include the development of management system (housing, grazing practice and animal watering), reduce snail population by drying the marshy or wet areas or using biological control methods like, introducing the frogs and birds (Alemneh 2019).

Conclusion

Fasciolosis is a common parasitic infection which affects the ruminant productivity by its direct or indirect losses.

Different factors including change in climatic condition and human activities play a role in further spread and distribution of liver flukes. Great concerns should be directed against resistance to flukicides to reduce the number of parasites that led to restrictions in their use. The drug residues in animal products i.e., meat and milk are another issue that restricts anthelmintic usage at any time, due to the long withdrawal period of some products. Moreover, increase in the frequency of liver fluke infection among animals adversely leads to rise the infection rates in human at different regions of the world.

REFERENCES

Admassu B et al., 2015. A review on bovine fasciolosis. European Journal of Biological Sciences 7: 139-146.

Ahmed S et al., 2005. Diversity and Prevalence of Trematodes in liver of Sheep and Goat in Quetta, Pakistan. Pakistan Journal of Zoology 37: 205-210.

Ai L, 2010. Rapid identification and differentiation of *Fasciola hepatica* and *Fasciola gigantica* by a loop-mediated isothermal amplification (LAMP) assay. Veterinary Parasitology 174: 228-233.

Alemneh T, 2019. An Introductory to Fasciolosis. Concept of Dairy and Veterinary Sciences 2(3): 190-194.

Andrews SJ, 1999. The Life Cycle of *Fasciola hepatica*. In: Dalton JP, editors. Fasciolosis. CAB International: Oxon; pp: 1-29.

Amer S et al., 2016. Identity of *Fasciola* spp. in sheep in Egypt. Parasites and Vectors 9: 623.

Amiri S et al., 2021. Accurate and rapid detection of *Fasciola hepatica* copro-DNA in sheep using loop-mediated isothermal amplification (LAMP) technique. Veterinary Medicine and Science 7(4): 1-9.

Arifin MI et al., 2016. Comparison of molecular and conventional methods for the diagnosis of Fasciola hepatica infection in the field. Veterinary Parasitology 232: 8-11.

Artigas P et al., 2011. Characterization of fascioliasis lymnaeid intermediate hosts from Chile by DNA sequencing, with emphasis on Lymnaea viator and Galba truncatula. Acta Tropica 120: 245-257.

Ashrafi K et al., 2006a. Plant-borne human contamination by fascioliasis. American Journal of Tropical Medicine and Hygiene 75: 295-302.

Ashrafi K et al., 2006b. Phenotypic analysis of adults of *Fasciola hepatica*, *Fasciola gigantica* and intermediate forms from the endemic region of Gilan, Iran. Parasitology International 55: 249- 260.

Ashrafi K et al., 2014. Fascioliasis, a worldwide parasitic disease of importance in travel medicine. Travel Medicine and Infectious Disease 12: 636-649.

Atalabi TE and Lawal OT, 2020. Fascioliasis: A Foodborne Disease of Veterinary and Zoonotic Importance. In: Umar B, editors. Rural Health.

Ayele M and Hiko A, 2016. Review on the Biology of *Fasciola* Parasites and the Epidemiology on Small Ruminants. Advances in Life Science and Technology 48: 2224-7181.

Bargues MD et al., 2016. Human fascioliasis endemic areas in Argentina: multigene characterization of the lymnaeid vectors and climatic-environmental assessment of the transmission pattern. Parasites and Vectors 9: 306.

Bargues MD et al., 2020. Genetic uniformity, geographical spread and anthropogenic habitat modifications of lymnaeid vectors found in a One Health initiative in the highest human fascioliasis hyperendemic of the Bolivian Altiplano. Parasites and Vectors 13: 171.

Beesley NJ et al., 2018. *Fasciola* and fasciolosis in ruminants in Europe: Identifying research needs. Transboundary and Emerging Diseases 65(1): 199-216.

Boray J and Enigk K, 1964. Laboratory studies on the survival and infectivity of *Fasciola hepatica* and *F. gigantica* metacercariae. Zeitschrift Für Tropenmedizin Und Parasitologie 15: 324-331.

Brown DS, 1980. Fresh water snails of Africa and their medical importance, Taylor and France Ltd., London.

Charlier J et al., 2020. Initial assessment of the economic burden of major parasitic helminth infections to the ruminant livestock industry in Europe. Preventive Veterinary Medicine 182: 105103.

Chauvin A et al., 2001. Responses of Fasciola hepatica infected sheep to various infection levels. Veterinary Research 32: 87-92.

Chen MG and Mott KE, 1990. Progress in assessment of morbidity due to *Fasciola hepatica* infection: a review of recent literature. Tropical Diseases Bulletin 87: 1-38.

David C, 1990. The veterinary book for sheep farms, 2nd Ed., Butler and Tanner, London, UK.

Davies Calvani NE et al., 2018. Comparison of early detection of *Fasciola hepatica* in experimentally infected Merino sheep by real-time PCR, coproantigen ELISA and sedimentation. Veterinary Parasitology 251: 85- 89.

Diyana JNA et al., 2019. A retrospective study on bovine fascioliasis in veterinary regional laboratories in Peninsular Malaysia. Journal of Parasitology Research: 7903682.

Esteban JG et al., 1997. A population-based coprological study of human fascioliasis in a hyperendemic area of the Bolivian Altiplano. Tropical medicine and international health 2: 695-699.

Farag HF and El Sayad MH, 1995. *Biomphalaria alexandrina* naturally infected with *Fasciola gigantica* in Egypt. Transactions of the Royal Society of Tropical Medicine and Hygiene 89: 36.

Fuentes MV et al., 2001. Validation of a mapping and predicting model for human fasciolosis transmission in Andean very high-altitude endemic areas using remote sensing data. Acta Tropica 79: 87-95.

Fufa A, 2009. Bovine fasciolosis; coprological, abattoir survey and its economic impact due to liver condemnation at Soddo Municipal abattoir, Southern Ethiopia. Tropical Animal Health and Production 12(3): 221-240.

Gandhi P et al., 2019. Triclabendazole in the treatment of human fascioliasis: a review. Trans Royal Society of Tropical Medicine and Hygiene 113: 797-804.

Girones N et al., 2007. Immune suppression in advanced chronic fascioliasis: an experimental study in a rat model. Journal of the Infectious Diseases 195: 1504-1512.

Gonzalez-Miguel J et al., 2019. Numerous *Fasciola* plasminogen-binding proteins may underlie blood-brain barrier leakage and explain neurological disorder complexity and heterogeneity in the acute and chronic phases of human fascioliasis. Parasitology 146: 284-298.

Graber M et al., 2005. Helminthes and Helminthiasis of Domestic and Wild Animal in Ethiopia. Revue D'élevage et de Médecine Vétérinaire des Pays Tropicaux 1: 13-95.

Graczyk TK and Fried B, 1999. Development of *Fasciola hepatica* in the intermediate host. In: Dalton JP, editors. Fasciolosis: CABI Publishing; pp: 31-46.

Hamoo RN et al., 2019. Molecular characterization and phylogenetic analysis of *Fasciola gigantica* in Iraqi sheep using ITS1. Advance Animal Veterinary Science 7: 256-260.

Haridwala S et al., 2021. Morphological and molecular characterization of *Fasciola hepatica* and *Fasciola gigantica* phenotypes from co-endemic localities in Mpumalanga and KwaZulu-Natal provinces of South Africa. Food and Waterborne Parasitology 22: Article # 00114

Hayward AD et al., 2021. The influence of liver fluke infection on production in sheep and cattle: a meta-analysis. International Journal for Parasitology 51(11): 913-924.

Hendrix CM and Robinson E, 2006. Diagnostic parasitology for veterinary technicians. 3rd Ed., pp: 107 -109.

Hillyer GV and Apt W, 1997. Food-borne trematode infections in the Americas. Parasitology today 13: 87-88.

Hosseini-Safa A et al., 2019. High-resolution melting analysis as an appropriate method to differentiate between *Fasciola hepatica* and *F. gigantica*. Iran Journal of Public Health 3: 501-507.

Hussein AN and Khalifa RM, 2008. Experimental infections with *Fasciola* in snails, mice and rabbits. Parasitology Research 102: 1165-1170.

Hussein ANA et al., 2010. Description of Eggs and Larval Stages of *Fasciola*, Light and Scanning Electron Microscopic Studies. Research Journal of Parasitology 5: 1-12.

Hussein AA et al., 2020. Development and hatching mechanism of *Fasciola* eggs, light and scanning electron microscopic studies. Saudi Journal of Biological Sciences 17(3): 247-251.

Itagaki T et al., 2009. Occurrence of spermic diploid and a spermic triploid form of *Fasciola* in Vietnam and their molecular characterization based on nuclear and mitochondrial DNA. Parasitology International 58: 81-85.

Javid A et al., 2011. Some Epidemiological Aspects of Fascioliasis among Cattle of Ladakh. Global Veterinaria 7(4): 342-346.

John BC et al., 2019. A review of our current understanding of parasite survival in silage and stored forages, with a focus on *Fasciola hepatica* Metacercariae. Grass Forage Science 74(2): 211-217

Kalu E, 2015. Bovine fascioliasis: a review. IOSR Journal of Agriculture and Veterinary Science 8: 23-26.

Khoramian H et al., 2014. Prevalence of ruminant's fascioliasis and their economic effects in Kashan, center of Iran. Asian Pacific Journal of Tropical Biomedicine 4(11): 918-922.

Lalor R et al., 2021. Pathogenicity and virulence of the liver flukes *Fasciola hepatica* and *Fasciola gigantica* that cause the zoonosis Fasciolosis. Virulence 1: 2839-2867.

Lotfy WM and Hillyer GV, 2003. *Fasciola* species in Egypt. Experimental pathology and parasitology 6: 9-22.

López Lemes MH et al., 1996. Fascioliasis en la República Oriental del Uruguay. Revista Médica del Uruguay 12(1): 37-43.

Malek EA, 1980. Snail Transmitted Parasitic Diseases, 2nd Vol., CRC Press Inc., Boca Raton, Florida.

Martınez-Perez JM, 2012. Comparison of three different techniques to diagnose *Fasciola hepatica* infection in experimentally and naturally infected sheep. Veterinary Parasitology 190: 80-86.

Martınez-Valladares M and Rojo-Vazquez FA, 2016. Loop-mediated isothermal amplification (LAMP) assay for the diagnosis of fasciolosis in sheep and its application under field conditions. Parasites and Vectors 9: 73-77.

Mas-Coma S, 1996. Human fascioliasis in Latin America. In: MartõÂnez FernaÂ ndez AR editors. Parasitismos y desarrollo. Jornadas Iberoamericanas de Ciencias FarmaceÂ uticas. Real Academia de Farmacia: Madrid; pp: 31-86.

Mas-Coma S and Bargues MD, 1997. Human liver flukes: a review. Research and Reviews in Parasitology 57(3-4): 145-218.

Mas-Coma S et al., 1999. Epidemiology of human fascioliasis: a review and new proposed classification. Bulletin of the World Health Organization 77 (4).

Mas-Coma S et al., 2001. *Fasciola hepatica* and lymnaeid snails occurring at very high altitude in South America. Parasitology 123: 115-127.

Mas-Coma S et al., 2009a. Climate change effects on trematodiases, with emphasis on zoonotic fascioliasis and schistosomiasis. Veterinary Parasitology 163: 264-280.

Mas-Coma S et al., 2009b. *Fasciola*, lymnaeids and human fascioliasis, with a global overview on disease transmission, epidemiology, evolutionary genetics, molecular epidemiology and control. Advances in Parasitology 69: 41-146.

Mas-Coma S et al., 2014. Neurological and ocular fascioliasis in humans. Advances in Parasitology 84: 27-149.

Mas-Coma S et al., 2018. Human fascioliasis infection sources, their diversity, incidence factors, analytical methods and prevention measures. Parasitology 145: 1665-1699.

Mavrogianni VS et al., 2014. Trematode infections in pregnant ewes can predispose to mastitis during the subsequent lactation period. Research in Veterinary Science 96: 171-179.

Meray Sierra R, 2020. Equines as reservoirs of human fascioliasis: transmission capacity, epidemiology and pathogenicity in *Fasciola hepatica*-infected mules. Journal of Helminthology 94: Article # 189.

Munita MP et al., 2019. Liver fluke in Irish sheep: prevalence and associations with management practices and co-infection with rumen fluke. Parasites and Vectors 12: Article # 525.

Nyindo M and Lukambagire AH, 2015. Fascioliasis: An ongoing zoonotic trematode infection. BioMed Research International: Article # 786195.

Phalee A et al., 2015. Experimental Life History and Biological Characteristics of *Fasciola gigantica* (Digenea: Fasciolidae). Korean Journal of Parasitology 53(1): 59-64.

Prasad PK et al., 2008. Molecular identification of the Indian liver fluke, *Fasciola* (Trematoda: Fasciolidae) based on the ribosomal internal transcribed spacer regions. Parasitology Research 103: 1247-1255.

Radiostis OM et al., 2007. A text book of the disease of cattle, horses, sheep, pigs, and goats. Veterinary Medicine, 10th Ed., Saunders Elsevier, London, UK, England.

Rickard BL, 2001. The Practical Veterinarian. Veterinary Parasitology 273-302.

Robinson MW and Dalton JP, 1999. Zoonotic helminth infections with particular emphasis on fasciolosis and other trematodiases. Philosophical Transactions of the Royal Society B: Biological Sciences 364: 2763-2776.

Roberts JA and Suhardono, 1996. Approaches to control fasciolosis in Ruminants. International Journal for Parasitology 26(8-9): 971-981.

Rojas CAA, 2014. Techniques for the diagnosis of Fasciola infections in animals: Room for improvement. Advanced Parasitology 85: 65-107.

Salimi-Bejestani MR et al., 2005. Development of an antibody-detection ELISA for *Fasciola hepatica* and its evaluation against a commercially available test. Research in Veterinary Sciences 78: 177-181.

Sargison ND and Scott PR, 2011. Diagnosis and economic consequences of triclabendazole resistance in *Fasciola hepatica* in a sheep flock in south-east Scotland. Veterinary Record 168: 159.

Sharma RL et al., 2011. Epizootiology, pathogenesis and immunoprophylactic trends to control tropical bubaline fasciolosis: An overview. Journal of Parasitic Diseases 35(1): 1-9.

Shoriki T et al., 2014. Molecular phylogenetic identification of *Fasciola* flukes in Nepal. Parasitology International 63: 758-762.

Smyth JD, 1994. Introduction to animal parasitology, 3rd Ed., Cambridge University Press, England.

Taira N et al., 1997. Zoonotic potential of infection with *Fasciola* spp. by consumption of freshly prepared raw liver containing immature flukes. International Journal for Parasitology 27: 775-779.

Taylor M et al., 2013. Veterinary parasitology, 3rd Ed., Wiley Press, Oxford, England.

Toet H et al., 2014. Liver fluke vaccines in ruminants: strategies, progress and future opportunities. International Journal for Parasitology 44: 915-927.

Urquhart HM et al., 1996. Veterinary Parasitology, 2nd Ed., Black well Science Ltd, London, UK, England.

Usip LP et al., 2014. Prevalence of fascioliasis and the economic loss of condemned liver due to *Fasciola* infection in cattle slaughtered at three abattoirs in Eket Urban, Akwa Ibom State of Nigeria. Global Advanced Research Journal of Food science and Technology 3(2): 54-75.

Valero MA and Mas-Coma S, 2000. Comparative infectivity of *Fasciola hepatica* metacercariae from isolates of the main and secondary reservoir animal host species in the Bolivian Altiplano high human endemic region. Folia Parasitologica 47: 17-22.

Valero MA et al., 2002. Patterns in size and shedding of *Fasciola hepatica* eggs by naturally and experimentally infected murid rodents. The Journal of Parasitology 88(2): 308-313.

Valero MA et al., 2003. Risk of gallstone disease in advanced chronic phase of fascioliasis: an experimental study in a rat model. Journal of Infectious Diseases 188: 787-793.

Vara-Del Río MP et al., 2007. Genetic heterogeneity of *Fasciola hepatica* isolates in the northwest of Spain. Parasitology Research 101(4): 1003-1006.

Vázquez AA et al., 2018. Lymnaeid snails hosts of *Fasciola hepatica* and *Fasciola gigantica* (Trematoda: Digenea): A worldwide review. Center for Agriculture and Bioscience International 13(62): 1-15.

Wagari A, 2021. A Review on cattle Fasciolosis. Journal of Veterinary Medicine and Surgery 5 (4): Article # 6740

Zhang XX et al., 2019. Complex and dynamic transcriptional changes allow the helminth *Fasciola gigantica* to adjust to its intermediate snail and definitive mammalian hosts. BMC Genomics 20: 729.