RE-EMERGING ZOONOTIC HELMINTH INFECTIONS IN KERALA - AN UPDATE

Lucy Sabu*, Bindu Lakshmanan and K. Devada

Department of Veterinary Parasitology, College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala - 680 651

*Corresponding author: lucysabu@rediffmail.com

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ABSTRACT

Zoonotic cestodes, nematodes and trematodes are widely prevalent in different parts of India. Despite the increasing prevalence of helminth infections among animals, zoonotic infections were not common in Kerala. However, the occurrence of several infections like sparganosis and human dirofilarosis among human subjects in Kerala, being recently diagnosed in the Department of Veterinary Parasitology, reminds of the need to review the situation with special emphasis to the socio economic background of the state. This review is an attempt to present a brief account of the incidence of zoonotic cestode and nematode infections, in the light of our experience of diagnosing such infections in man and animals over three decades. The awareness of the existence of zoonotioc helminths and a sound knowledge of their life cycle would go a long way to curb its spread in the population.

Keywords: Taeniosis, sparganosis, dirofilariosis, Kerala, zoonosis

INTRODUCTION

Zoonoses are diseases transmissible between man and animals. The chances for occurrence of zoonotic diseases in developing countries are high due to the lack of awareness on the etiology and preventive measures, poverty, lack of personal hygiene, open space defecation, scarcity of water, habit of eating raw/ undercooked fish and meat, high population density and abundance of stray animals and vectors. In addition, cultural habits, natural/ man-made calamities, population migration and population explosion also play an important role in the emergence/ re-emergence of such conditions. Though there are many well known zoonotic helminth infections reported from India, majority of them do not find a mention in the reports from Kerala. The high literacy rate and socio economic status of the people have been reported to substantially reduce the incidence of many of these infections in human beings in Kerala. This paper is to describe the increasing occurrence of zoonotic helminthoses among human beings in Kerala over the last three decades from our experience of identifying zoonotic helminth specimens in human beings and animals from different parts of the state. Future investigations are to be directed towards unfolding the molecular epidemiology of zoonotic helminths in Kerala

ZOONOTIC HELMINTH INFECTIONS

Cestode Zoonosis

Taeniosis and Cysticercosis

Taeniosis is an intestinal infection caused by three species of tapeworm: Taenia solium (pork tapeworm), T. saginata (beef tapeworm) and *T. asiatica*. Humans can become infected with T. saginata or T. asiatica when they consume infected beef meat or pig liver tissue, respectively, which has not been adequately cooked. Taeniosis due to T. saginata or T. asiatica has no major impact on human health. Infection with the T. solium tapeworm occurs when humans eat raw or undercooked infected pork. Tapeworm eggs pass with the faeces and are infective for pigs. Infection in humans with the T. solium tapeworm causes few clinical symptoms. The T. solium eggs may also infect humans if they are ingested, causing infection with the larval parasite in the tissues (human cysticercosis). This infection can result in devastating effects on human health. The larvae (cysticerci) may develop in the muscles, skin, eyes and the central nervous system. When cysts develop in the brain, the condition is referred to as neurocysticercosis. Symptoms include severe headache, blindness, convulsions, and epileptic seizures, and can be fatal. Neurocysticercosis is the most frequent preventable cause of epilepsy worldwide, and is estimated to cause 30 per cent of all epilepsy cases in countries where the parasite is endemic (WHO, 2018).

Taenia spp. are identified based on morphologic features of suckers and rostellum on the scolex. It is practically impossible to get the scolex from an infected person and the gravid segments are usually brought for identification. Gravid segments of T. solium are shed in groups and are non motile, while those of T. saginata are motile and excreted individually. The difference in the uterine branching in the gravid proglottids is a method to diagnose the species. In T. saginata the gravid segments contain 15 to 20 lateral uterine branches, while T. solium contains only 7 to 13 lateral branches. The presence (T. saginata) or absence (T.solium) of vaginal spincter muscle is also a differentiating feature in mature and gravid segments (Souslby, 1982). Various *Taenia* species can be identified using PCR amplification of the 5.8S ribosomal RNA gene and subsequent restriction enzyme analysis of the PCR product by multiplex PCR targeting the mitochondrial gene (Mayata et al., 2000).

Though India has still been considered to be endemic for human taeniosis, people of Kerala have been able to successfully break the life cycle at the points of entry and exit. At the same time, the occurrence of taeniosis and cysticercosis is significantly higher in other southern states though they follow a comparatively predominant vegetarian diet. Even when majority of Kerala people eat meat, proper cooking of meat prevents them from being infected. According to IIPS (2007), 96 per cent of households in Kerala own toilets, while in the adjoining states of Tamil Nadu and Karnataka, only 43 and 46 per cent of households, respectively have this facility. It is evident that open defecation is not normal in Kerala unlike in other states, thus contamination of soil with eggs do not happen, which minimizes the chances of animals or man getting cysticercosis from *Taenia* eggs. The habit of eating well cooked food and drinking boiled water,

lesser degree of consumption of raw salads add to the benefit. The overall high socio-economic status aid in seeking medical advice at the face of any infection, thus successfully preventing spread of a disease from its focus. Another fact is the absence of free ranging desi pigs in Kerala.

In the late eighties, a case of adult taeniosis was also identified in our laboratory from a Kashmiri Veterinary graduate student, who had the history of passing motile segments which was confirmed as T. saginata. In 1991 and 1992 another two cases of adult Taenia infection in human beings were diagnosed in our laboratory by identifying the gravid segments. The former was from a young man who had the history of the segments crawling out of the body and hence confirmed as T. saginata. while the latter was a sectioned slide referred from a diagnostic lab in Thrissur where the species could not be identified. An elaborate study on the prevalence of taeniosis in human beings from five districts of Kerala carried out by examining the faecal samples from 1400 people as part of an ICAR project at College of Veterinary and Animal Sciences, Mannuthy could not trace a single case of human taeniosis in Kerala; however screening of 1000 serum samples for qualitative detection of IgG antibodies against T. Solium during the same period using ELISA, revealed the presence in 2.2 per cent cases (Thomas, 2014). Cherian et al. (2014) recorded the seropositivity to neurocysticercosis in Kerala as 15 per cent. It is also worth stating that the detection of cysticerci in beef or pork at the Veterinary University Meat Plant at Mannuthy is very rare. However, the increasing involvement of immigrant labourers, enhanced dependence on other

states for fruits and vegetables and surging habit of consumption of raw salads from outside eateries (where proper washing of salad vegetables are not at all confirmed) are possibly the potential risk factors that Kerala people need to be made aware of.

Sparganosis

Sparganum is the third stage larva (plerocercoid) of a pseudophyllidean tape worm belonging to the genus Spirometra. Dogs and cats are the natural hosts of Spirometra sp. in which the adult worms occur in the small intestine. The eggs passed out along with faeces contaminate water sources and the first stage larva (coracidium) enters into cyclops to form procercoid. The third stage larve (plerocercoid - sparganum) develops in frogs, snakes and other animals on ingestion of infected cyclops along with water. Man is an accidental host for sparganum (Soulsby, 1982). Human infection is widely prevalent in many Asian countries including Japan, Korea and China due to certain peculiar habits and practices such as eating raw meat of cold blooded animals or application of freshly ground frog flesh as poultice on sores especially in the eye (Duggal et al., 2011). Subcutaneous tissues are the most common site for sparganosis. However, worms have also been recovered from other organs including the eyes, brain, urinary tract, pleura, pericardium and spinal canal and also from rare sites such as wall of the intestine, breast, scrotum, epididymis. ureter, urinary bladder, abdominal cavity, heart and lung. A perusal of available literature did not reveal the occurrence of proliferative sparganosis due to Spirometra proliferum in India. The sparganum of S. proliferum is very

invasive and spreads throughout the body by continuous branching and budding; even small pieces of tapeworm left behind after attempted excision may spread and infect distant sites. An unusual case of urinary sparganosis was documented in a 25 year old woman who was reported to have passed 1.5 cm long live worms through urine (Trupti et al., 2018). It is quite unnatural for the sparganum to be shed through excreta and needs further tests for species authentication.

The first case of sparganosis in Kerala was reported from Kunnamkulam, Thrissur (Sabu et al., 2015). It was a seven cm long worm recovered from a painful swelling at the right inguinal region of a seven year old boy. Four cases of sparganosis (0.5-3 cm) were reported from women patients from painful subcutaneous swellings in Kozhikode, Kerala during the period from 2010 to 2015 (Sudarsana et al., 2016). Similar encounters happened in different parts of the state might have gone unreported as the patient recovers soon after removal of the worms and also the lack of interest/ expertise in identifying parasite specimens. Another case of sparganosis (5 cm long) in a woman patient from Thrissur was identified in September, 2017 at Department of Parasitology laboratory, CVAS, Mannuthy. In all these cases, infection has been presumed to be acquired by ingestion of cyclops in pond or river water. Removal of the worm resulted in uneventful recovery. As people of Kerala, do not follow any of the leading causes of infection in endemic regions mentioned above, drinking unfiltered water from open wells or ponds is suspected to have resulted in the infection. At the same time infection due to Diphyllobothrium latum, a closely related Pseudophyllidean tapeworm, is quite common among dogs in Kerala as revealed by postmortem and faecal sample examination (Jacob and Pillai, 1991). As the eggs of both species are similar and does not warrant treatment, specific identification is not routinely made.

To prevent ingestion of cyclops or Spirometra larvae, water in endemic areas should be boiled or treated with disinfectants. Currently effective no treatment for sparganosis is known. The only treatment is surgical excision of localized infection, although limited success has been reported with the use of praziquantel (Torres et al., 1981). The potential threat of misdiagnosis along with the lack of knowledge on the existence of the condition may pose a major challenge in accurate treatment of ailing patients.

Various trematodes such as Clonorchis sp. and pseuduphyllidean tapeworms like Diphyllobothrium sp. are acquired by eating fish. Though these fish borne parasites are quite common in dogs and cats (Jacob and Pillai, 1991), there is hardly any report on the occurrence of these diseases in human beings in Kerala. This may also be attributed to the factors related to the absence of Taenia spp. in people of Kerala.

Nematode zoonoses

Though a variety of nematodes of zoonotic significance are prevalent in India, Kerala is a focus for mosquito borne filarial zoonosis. The most commonly prevalent species in dogs has been identified as Dirofilaria repens, which is found in the subcutaneous tissues in dogs. Species coming under the genus *Dirofilaria* are natural parasites of dogs and other carnivores. These worms have indirect life

cycle and various species of mosquitoes act as intermediate hosts/ vectors in which the first stage larvae develop to infective third stage before being transmitted to their final host. Dirofilaria immitis found in the heart and pulmonary artery is not prevalent in Kerala, while D. repens residing in the subcutaneous tissues of dogs and cats are fairly common. Dirofilaria repens adult female worm measures 25 to 30 cm while the male is up to 13 cm in length with a width of 1 to 2 mm and the microfilariae are unsheathed. However, the sheathed microfilariae found in dogs in and around Thrissur were identified as Brugia malayi by histochemical staining and molecular studies (Ambily et al., 2011).

In the natural hosts, these adult female filarid worms produce thousands of microfilariae (L₁) into the circulation daily, which are ingested by mosquitoes through a blood meal. Larvae develop into infective third stage in the Malpighian tubules and thoracic muscles of the mosquito over the next 10 to 16 days, depending on environmental conditions before being reintroduced into a new host. Various species of moquitoes coming under the genera of *Anopheles, Culex, Aedes, Mansonia* and others are known to act as vectors for *Dirofilaria* spp.

Humans are accidental hosts of *Dirofilaria* spp., because adult worms cannot reach maturity in the heart or in the skin. Most infective larvae introduced into humans are thought to die; therefore, infected individuals usually are not microfilaremic. Infections in humans usually are manifested as a single subcutaneous nodule caused by a worm trapped by the immune cells. Subcutaneous migration of the worm may result in local

swellings with changing localization (creeping eruption). Organ manifestation of the worm in eyes, lung, male genitals or female breast have also been frequently reported. Since only a single worm is present, removal of the parasite is sufficient to treat human infection.

First record of human dirofilariosis in the state dates back to 42 years, when D. conjunctivae was reported from the eye of a 60 year old lady residing in a coastal village (Joseph et al., 1976). Over the last three decades, several worms recovered from human patients from different parts of the state have been identified as D. repens by the Department of Veterinary Parasitology, CVAS, Mannuthy, Thrissur. The first report was in 1999 from a woman patient from Ottappalam, Palakkad (Senthilvel and Pillai, 1999). Twelve specimens of D. repens received from various hospitals in six districts of Kerala during a period of two years from 2002 to 2004 were morphologically identified (Sabu et al., 2005). These included 11 female worms (9.0 - 13.5 cm) and one male worm (6.0 cm). Molecular identification of the human isolates by PCR targeting the mitochondrial and ribosomal RNA genes and further analysis of nucleotide sequence was also successfully attempted in our laboratory to confirm the species identity of D. repens (Nazar et al., 2017). Over and above the published works, worms recovered from human patients in and around Thrissur are routinely identified in our laboratory. In all cases, wherever there is involvement of nematodes, it was confirmed as D. repens. Most of the worms identified were females except a few male worms. All of the worms were from eyes and subcutaneous nodules or abscesses. In almost all cases,

live worms were obtained from adult men or women patients who were over 35 years of age. The youngest case reported so far was in a four month old boy in Srilanka (Schmidt and Robert, 2009).

Recently in June, 2018, a female worm measuring 10.5 cm was recovered from the scrotum of a two year old boy from Kunnamkulam, Thrissur. The child had swelling in the scrotum for about a month before seeking medical advice. In the same month, we further identified an 11 cm long worm removed from the eye of a 45 year old lady from Thrissur as D. repens. Both were live female worms with developed uterus containing immature eggs alone and not larvae proving that the worms have not attained sexual maturity. As human beings are unnatural hosts, the worms fail to attain sexual maturity. The absence of a male worm may also contribute to this. The frequent occurrence of human dirofilarosis in the state along with the high prevalence of microfilarosis among client owned dogs (Sabu, 2005; Abraham et al., 2018) signal the endemic nature of the infection in Kerala. Surprisingly, such a focus is absent in neighboring state of Tamil Nadu, which warrants detailed investigation. Surgical removal of worms results in uneventful recovery and hence no follow up is resorted to. Zoonotic filariosis in Kerala is poorly diagnosed and under reported.

Twenty seven species of mosquitoes coming under seven genera including Anopheles, Culex, Aedes and Mansonia were identified from Thrissur. Kerala which are considered to be potential vectors for Dirofilaria spp. (Sabu, 2005). Preliminary studies to explore the presence of filariid larvae within mosquito species was also undertaken. Sausage shaped second stage larvae and elongate third stage larvae of D. repens could be demonstrated in two out of 1019 mosquitoes collected from human dwellings, these two helminth positive mosquitos belonged to the genera of Armigerus and Culex, respectively (Sabu and Subramanian, 2007). Research should be driven to explore the molecular epidemiology of human and animal isolates of D. repens and to assess the vectorial capacity of the mosquito species.

The occurrence of Loa loa was reported from the subconjunctival space of a woman patient in Calcutta based on a few morphological features like tapering anterior end, mouth without definite lips, anterior opening of vulva and embryo filled uterus, which are typical of all filarid worms (Mandal et al., 2013). The worm was reported to be 10.5 cm long. The authors have not mentioned any specific features suggestive of Loa loa, including its length, which is usually less than seven cm. In this context molecular identification is warranted before confirming the filariid worm as Loa loa. All other reports of filariid worms measuring more than 10 cm identified as L.loa from different parts of India also suffers from the lack of species confirmation.

SUMMARY

The present communication attempted to summarise the scenario of zoonotic helminth infection in the state, in the light of our experience with parasite diagnosis for more than three decades. The prevalence of vector borne helminth zoonosis is on the rise in Kerala. However, cestode zoonosis in the state is rarely reported due to the high standards of personal hygiene and cooking style. The changing food habits and the

immigrant population pose potential risk with regards to human taeniosis. The frequent reports of human sparganosis also points to the need of designing effective public health camps to create awareness.

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