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Trichinellosis: An under recognised zoonosis in India

H B CHETHAN KUMAR[1], HIMANI DHANZE[2], K N BHILEGAONKAR[3] and ASHOK KUMAR[4]

ICAR – Central Coastal Agricultural Research Institute, Old Goa, Goa 403 402 India

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ABSTRACT

Trichinellosis also known as trichinosis is a meatborne zoonotic disease caused by *Trichinella* larvae present in the infected meat. The disease has worldwide distribution. Although this nematode has wide host range, pigs are the major source of infection to humans. The symptoms of trichinellosis in humans may vary from self-limiting disease to a severe disease terminating in death. On the other hand, animals rarely suffer from the disease. In India ,*Trichinella* infection has been reported both from humans and animals. The disease is still largely under recognised in medical domain masking the true picture of the disease status in the country. The responsibility of preventing the human trichinellosis lies with both medical and veterinary fraternity. Hence, medical and veterinary professionals need to be updated to enable early diagnosis and treatment of trichinellosis cases and to identify and condemn the infected meat. Since the major route of transmission of trichinellosis to humans is by consumption of undercooked meat, strong and well-structured public health awareness program is also the need of the hour.

Key words: Animal, Human, India, Pigs, Prevalence, *Trichinella*

Trichinellosis is a helminth zoonosis caused by the consumption of raw or undercooked meat of domestic or wild animals, infected with larvae of the genus *Trichinella* (OIE 2008). The major source of human infection is inadequately cooked meat from the members of the Family Suidae. *Trichinella* has a wide host range affecting mammals, reptiles and birds. The disease is known to infect ten thousand humans every year with a mortality rate of about 0.2% (Pozio 2007). Trichinellosis is an important disease not only due to its zoonotic potential but also its profound impact on swine production, international trade and food safety (Gottstein *et al*. 2009). Although few human and animal cases of trichinellosis have been reported from India, the disease remains largely under recognised in India (Joshi *et al*. 2014, Singh *et al.* 2015). Since, there is no comprehensive review available on the current status of trichinellosis in the country; in this paper we have discussed the present scenario of *Trichinella* infection in animals and humans in India. Finally, strategies for prevention and control of this disease are also discussed.

Historically presence of *Trichinella* larvae in human diaphragm was first recognized in the year 1835 by James Paget and Richard Owen (Owen 1835). In 1846, the nematode was identified in pigs by Joseph Leidy (Gould 1970). The genus *Trichinella* has been placed under the Phylum: Nemathelminthes, Class: Nematoda, SubClass: Adenophorea, Order: Enoplida, Superfamily: Trichuroidea and Family: Trichinellidae (Soulsby 2005). Hitherto the genus *Trichinella* consists of twelve taxa, of which eight have been assigned with the status of species and four genotypes. The taxa are grouped into two clades, encapsulated (*T. spiralis*, *T. nativa*, *T. britovi*, *T. murrelli*, *Trichinella T6*, *T. nelsoni*, *Trichinella* T8, *Trichinella* T9 and *Trichinella* T12) and non-encapsulated (*T. pseudospiralis*, *T. papuae*, *T. zimbabwensis*) based on presence or absence of collagen capsule around the muscle stage larvae (Pozio *et al.* 2009).

The life cycle of *Trichinella* begins with consumption of infected muscle containing encysted L1 stage larvae by susceptible host. Larvae are released in the gastrointestinal tract and undergo four moults to become adults. Adult male and female worms copulate and the ovo-viviparous female release larvae five to seven days post infection. The larvae enter systemic circulation by penetrating the intestinal mucosa and finally get lodged in the skeletal muscles where they subsequently get encapsulated with a host derived collagen capsule (encapsulated species only). Within the muscle cells the larvae develops into an infective L1 stage in about fifteen days.

Epidemiology of trichinellosis in India

Predisposing factors for Trichinella infection in pigs

According to the 19th livestock census (2012), India has

Present address: [1]Scientist (chethuhb@gmail.com), Animal Sciences Section. [2]Scientist (hdhanze@yahoo.co.in), Division of Veterinary Public Health, Indian Veterinary Research Institute, Izatnagar, Bareilly. [3]Principal Scientist (kiranvph @rediffmail.com), Training and Education Centre, Indian Veterinary Research Institute, Agriculture College Campus, Shivajinagar, Pune. [4]Assistant Director General - Animal Health (ashokakt @rediffmail.com), Division of Animal Science, Krishi Bhavan, ICAR, New Delhi.

10.29 million pigs (DAHD 2015). Major segment of the pig population is concentrated in eastern and north-eastern part of the country. The pig farming is mostly practiced by rural poor or marginal farmers as a source of additional income and employment. The majority of country's pig farming is still under traditional backyard rearing with small holding size. Nevertheless medium to large scale piggeries with scientific farm practices do exist. In addition to scavenging by pigs, a part of the daily ration is provided in the form of fodder, grains and kitchen waste collected from restaurants. The housing system in backyard piggery include either roofed or unroofed pigsties, or sometimes pigs may be completely left loose without any housing. In case of housed pigs, the sty may or may not provide proper rodent control and on the other hand free ranging pigs will come in contact with stray animals such as dogs, cats, birds, wild animals and dead carcasses which may harbour *Trichinella* larvae. There are minimal veterinary care activities including prophylactic vaccination and deworming of backyard piggery (Bujarbaruah 2006).

Once the pig reaches the slaughter weight, the pigs are either sold to butcher, who will slaughter and distribute carcass cuts for retail outlets depending on the demand. In some places pigs are slaughtered by the farmer itself at the farm premises and pork is sold to the consumers (Njuki *et al*. 2010). The slaughter in the farm premises or butcher shops takes place without any veterinary supervision. In pigs the aforementioned husbandry practices are known to increase the risk of acquiring *Trichinella* infection either by feeding of infected meat scraps from restaurant waste, or scavenging on dead rodent and wild animal carcasses. In the recent past, several sporadic cases and outbreak of human trichinellosis have been reported in the country. Not surprisingly most of the infections were contracted through consumption of inadequately cooked pork and pork products derived from either domestic pigs or wild boar.

Trichinella infection in animals

Trichinella has been detected both in domestic and wild animal species from different parts of the country (Table 1). However, the overall prevalence of *Trichinella* spp. recorded was rather low. Considerable data is available regarding the prevalence of other parasitic zoonoses *viz*. hydatidosis, cysticercosis, and cryptosporidiosis in veterinary and medical literature from different states. However, with regards to trichinellosis, in depth information on epidemiology, host range, prevalence, molecular characterization and disease burden is still lacking.

In India, *Trichinella* larvae have been detected in cat (Maplestone and Bhaduri 1942, Kalapesi and Rao 1954, Parmeter *et al*. 1967), domestic pig (Niphadkar *et al*. 1979, Pethe 1991, Chethan Kumar *et al*. 2015), synanthropic animals like rodents (Niphadkar 1973, Niphadkar 1975) and wild animals (Schad and Chowdhury 1965, Singh 2000) *viz.*, civet cat and tiger/panther. Although *Trichinella* larva was not isolated from wild boar meat, in an outbreak of human trichinellosis in Uttarakhand, wild boar meat was epidemiologically implicated as source of infection. In this outbreak, demonstration of *Trichinella* larvae in muscle biopsy of the infected patients and their food consumption history suggested the existence of this nematode in wild boar (Sethi *et al*. 2010). Table 1 summarises the previous surveys of animal trichinellosis in the country.

Table 1. Previous surveys of *Trichinella* infection in animals from India

Species	Infection status	Prevalence (%)	Reference
Cat	Positive	NA	Maplestone and Bhaduri (1942)
Cat	Positive	NA	Kalapesi and Rao (1954)
Civet cat	Positive	NA	Schad and Chowdhury (1965)
Cat	Positive	NA	Parmeter *et al* (1967)
Domestic pig	Negative	0%	Ramamurthi and Ranganathan (1968)
Domestic pig	Negative	0%	Deodhar *et al*. (1968)
Rodents	Positive	25%	Niphadkar (1973)
Rodents	Positive	1.37%	Niphadkar (1975)
Domestic pig	Negative	0%	Upadhyay *et al.* (1977)
Domestic pig	Positive	0.6%	Niphadkar *et al.* (1979)
Domestic pig	Negative	0%	Damodar and Rao (1982)
Domestic pig	Positive	0.4%	Pethe (1991)
Tigers and Panthers	Positive	15.79%	Singh (2000)
Domestic pig	Negative	0%	Gaurat (2002)
Wild boar	Epidemio-logical evidence	Epidemio-logical evidence	Sethi *et al.* (2010)
Domestic pig	Positive	0.69%	Chethan Kumar *et al*. (2015)
Tigers and bears	Negative Bears	0%	Chethan Kumar *et al*. (2015)

NA, Not available.

It is interesting to note that, *Trichinella* infection in domestic pigs was recorded in three different studies conducted in the same municipal abattoir located in Mumbai city (Niphadkar *et al*. 1979, Pethe 1991, Chethan Kumar *et al*. 2015). This municipal abattoir receives pigs for slaughter from different districts of Maharashtra and neighbouring Gujarat state, majority of them being raised under free ranging system. In India, the overall prevalence of trichinellosis is between 0.4–0.69% in domestic pigs (Chethan Kumar *et al*. 2015). Further studies are required to explore the presence of this parasite in other parts of the country in different host species.

Detection of *Trichinella* in wild animals is an indication that the nematode is circulating in sylvatic cycle in Indian wildlife. In this context, the spill over of infection from wild animals to domestic animals or even to humans cannot be overlooked. To further complicate the situation, the practice of bush meat consumption exists among tribal communities of north-eastern and other parts of India

(Hilaluddin *et al*. 2005). Consuming bush meat poses threat of contracting zoonotic infection in general and trichinellosis in particular to the consumers. The most glaring example of this fact is that most of the trichinellosis patients from India contracted the disease after eating wild boar meat. Further, the source of large scale outbreak of human trichinellosis which took place in district Pauri and Tehri of Uttarakhand was undercooked/roasted wild boar meat (Sethi *et al*. 2010).

Human trichinellosis in India

Sporadic and isolated cases of human trichinellosis have been recorded mostly from northern part of India. After reviewing the published literature, at least nine case reports and one outbreak of trichinellosis could be retrieved. The most common source of human infection was either undercooked pork or pork product of domestic pig or wild boar. The first case of human trichinellosis was reported from a woman who was presented with symptoms of fever, muscle weakness, joint pain and facial oedema. The patient had consumed pork fifteen days prior to the onset of the clinical symptoms. Muscle biopsy examination of the patient confirmed the presence of *Trichinella* larvae (Alipuria *et al*. 1996). Handa and co-workers diagnosed a case of trichinellosis in a female patient suffering from muscle weakness. Muscle biopsy examination showed *Trichinella* larvae and further enquiry revealed the fact that patient had also consumed wild boar meat (Handa *et al*. 2000). Bhatoe and co-workers detected *Trichinella* larvae by histopathology from a patient suffering from echinoccosis of thoracic spine (Bhatoe 2000). In another case report, a female patient was presented with symptoms of diarrhoea, nausea, fever, muscle pain and periorbital oedema. History of raw pork consumption and demonstration of *Trichinella* larva in muscle biopsy confirmed the diagnosis (Chatterjee *et al*. 2000). Mohan and others recorded an unusual clinical presentation in which *Trichinella* larvae were isolated from a patient while draining a psoas abscess (Mohan *et al*. 2002).

The first serious outbreak of human trichinellosis took place in district Pauri and Tehri of Uttarakhand state after consuming undercooked/roasted wild boar meat (Sethi *et al*. 2010). The outbreak caused high mortality (26.2%) and left several people hospitalized (Sethi *et al*. 2012). Dubey and associates detected *Trichinella* infection in a 12 year old girl suffering from osteomyelitis using ELISA followed by acid pepsin digestion assay of muscle for larval isolation. The source of infection in this case was suspected to be salami (Dubey *et al*. 2011). In a similar mode, 31 years old woman experienced fever, joint pain and oedema after consumption of under cooked wild boar meat (6 weeks ago) and in this patient also *Trichinella* larva was demonstrated in muscle biopsy (Abrol 2012). One more patient hailing from Uttarakhand was diagnosed with trichinellosis, who was also suffering from gluteal abscess. The food history of patient revealed that he consumed wild boar meat 4 weeks ago (Joshi *et al*. 2014). A sero-prevalence study of major parasitic zoonoses, conducted in Punjab, revealed 5.73% of human subjects as seropositive for trichinellosis (Singh *et al*. 2015). Until recently there was no report of human trichinellosis from southern India, when Alva and co-workers detected *Trichinella* larva in a hisptopathological section from a woman suffering from parotid gland neoplasm (Alva *et al*. 2015).

We believe that the low prevalence of human trichinellosis observed in the country could be just a tip of ice-berg. The gross under reporting of the disease may be attributed to nonspecific symptoms of the disease and lack of awareness in physicians and laboratory technicians about this disease (Handa *et al*. 2000, Dubey *et al*. 2011).

Clinical symptoms in humans

The clinical symptoms of human trichinellosis are not typical of the disease and vary from asymptomatic infection to severe form with life threatening complications (Kociecka 2000). Clinical manifestation begins with nonspecific symptoms such as fever, headache, uneasiness and gastrointestinal disturbances. Fever, facial or peri-orbital oedema, myalgia, are the important features of the human trichinellosis (Gottstein *et al*. 2009, Dupoy Camet and Bruschi 2007). The other symptoms include haemorrhages of conjunctiva, splintered haemorrhages in the finger nail beds and loss of strength etc. Acute trichinellosis may prove fatal because of cardiovascular, neurological, ocular and respiratory complications. Predominant clinical symptoms recorded in trichinellosis patients from India were muscle weakness, myalgia, fever, headache, facial swelling, peri-orbital oedema, difficulty in breathing and gastrointestinal disturbances (Sethi *et al*. 2010, Alipuria *et al*. 1996).

Diagnosis

The diagnosis of human trichinellosis is based on correlation of clinical symptoms with history of consumption of pork or wild animal meat, haematological analysis (eosinophilia), muscle biopsy examination (for larvae detection), serological assays (anti-*Trichinella* antibodies) and wherever possible demonstration of larvae in leftover meat sample. On the other hand, the diagnosis of *Trichinella* infection in animals relies broadly on two aspects, detection of larvae in muscle samples or detection of antibodies to *Trichinella*.

Detection of larvae in muscle

The larvae can be detected in muscle samples collected from predilection sites by using either trichinoscopy or artificial digestion technique. At present artificial digestion assay using acid pepsin is the only recommended technique for detection of *Trichinella* larvae in domestic and wild animal meat at slaughter houses for the purpose of food safety and trade (OIE 2008). Unfortunately, in most part of the country, the pig slaughter and sale of pork takes place in unorganised manner with no room to test the carcass for trichinellosis.

In pig and horse, for the purpose of artificial digestion assay, diaphragm, tongue and masseter are the most preferred muscles. Depending on the desired sensitivity of detection, a quantity of 1 gram to 10 gram muscle can be tested. In case of wild animals tongue, diaphragm, forearm, masseter, intercostals, flippers and pterygoid muscles are the sample of choice and quantity should be preferably not less than ten gram (Gamble *et al*. 2000, Kapel 2000, Kapel *et al*. 2005, Nockler and Kapel 2007).

Although, previous scientific literatures have designated the Indian isolates of *Trichinella* as '*Trichinella spiralis*' there are no molecular studies available to support this claim. And only morphological characters are not sufficient to confirm the species of larva (Krivokapich *et al*. 2008). Therefore, polymerase chain reaction is a valuable technique particularly for exact identification of species of larvae and in establishing species diversity of this parasite in different geographical regions of the world (Krivokapich *et al*. 2008, Zarlenga *et al*. 1999).

Detection of antibodies against Trichinella

The presence of specific anti-*Trichinella* antibodies can be demonstrated either in blood, serum, plasma and tissue fluids. Among several serological techniques available, enzyme linked immunosorbent assay is the most commonly used test for sero-surveillance of trichinellosis in domestic and wild animals (OIE 2008). However, ELISA is not recommended for testing individual animals for food safety purpose, since there is possibility of false negative results during initial stage of infection (Gamble *et al*. 2004).

Medical treatment

Early diagnosis and treatment of infected human subjects is essential to eliminate the adult worms in the intestine and to prevent the subsequent progression of the new born larvae into migratory phase. The therapeutic regimen consists of administration of anthelmintics, corticosteroids and supportive therapy. Mebendaole, albendazole and pyrantel are the anthelmintics of choice. Glucocorticoids alleviate the symptoms of hypersensitivity and prevent further complications (Dupouy Camet and Bruschi 2007).

Way forward in combating trichinellosis in India

Trichinellosis is obviously a preventable disease. Human trichinellosis affects several people worldwide and involve huge financial loss which demands suitable preventive and control measures in animals thereby preventing human infection (Kapel 2005). Established guidelines are already given by Office International Des Epizooties (OIE) and International Commission on Trichinellosis (ICT) which can be adapted for prevention and control of *Trichinella* infection in animals and humans.

In India swine rearing is practiced mostly by small and marginal farmers as a source of supplementary income for livelihood. It is estimated that during the year 2013–14 India produced 0.48 million tonnes of pork (India stat 2015). Further, it is believed that the current pork production is less than the existing demand. Considering the acute shortage and increasing demand for meat including pork the threat of meat borne zoonotic diseases in general and trichinellosis in particular cannot be overlooked.

Pre-harvest infection control strategies should include education of pig farmers to follow good farm practices to prevent the pigs acquiring *Trichinella*. This task can be achieved by preventing indiscriminate scavenging, adapting stall feeding, provision of rodent proof pigsties, eliminating the access of pigs to dead carcasses, thorough cooking of kitchen waste before feeding to pigs and preventive health interventions like regular deworming (Gamble *et al*. 2000).

According to law, the animals intended for meat purpose must be slaughtered in a designated slaughterhouse/abattoir and ante-mortem (AM) inspection; post-mortem (PM) inspection should be performed by a competent authority to provide wholesome meat to the consumer. Further, it is also recommended that the meat inspection procedure should include a means to detect *Trchinella viz*., artificial digestion assay in abattoirs. However, it is a common practice to slaughter the pigs in the butcher's shops without proper AM or PM inspection. It is also not uncommon to find the pigs being slaughtered for ceremonial purposes in certain tribal communities. Nevertheless, this missing link of carcass testing can be addressed by educating the butchers regarding the Indian laws and regulations relating to the slaughter of animals and encouraging them to slaughter the animals at the municipal slaughterhouses only.

The place where establishing a slaughterhouse is not economically feasible and meat cannot be tested for trichinellosis, rigorous consumer education should be undertaken. At the foremost general public should be made aware of importance of meat quality and safety and advice them not to consume meat from pigs/wild animals which have not undergone carcass inspection. If consumption of such meat is inevitable then public should be educated to adopt appropriate cooking practices. *Trichinella* larvae present in the infected meat can be inactivated by cooking to an internal temperature of not less than 71°C for at least 1 min, which is indicated by change in colour of meat to gray (Gamble *et al*. 2000). Freezing and irradiation are other methods which inactivate muscle larvae and can be practiced wherever facilities are available. However, curing, smoking, drying, and microwave cooking may not render the meat safe for human consumption (Gamble *et al*. 2000). The hunting and subsequent consumption of game meat which is not tested for the presence of *Trichinella* should be discouraged with strict enforcement of existing laws like wildlife protection act, 1972.

It is also important to establish species diversity of *Trichinella* isolated from different host species by systematic molecular studies. Since, in the recent past several reports of *Trichinellosis* are available from veterinary and medical field, efforts should be made to characterise the larvae till species level using molecular

methods. Species identification is key to explore the epidemiology of *Trichinella* and also to design suitable control measures (Pozio and La Rosa 2013).

CONCLUSION

Trichinellosis is a worldwide zoonosis caused by larvae of *Trichinella* ingested through raw or semi-cooked meat. The disease has cosmopolitan distribution infecting wide range of animals and birds. Trichinellosis is not only important from zoonotic perspective but also has profound impact on international trade and economy. Human trichinellosis outbreaks are reported frequently worldwide with considerable morbidity and mortality. In India trichinellosis has been reported both in human and animal population. A large scale outbreak of human trchinellosis due to consumption of wild boar meat has also been reported from Uttarakhand state which depicts the importance of mandatory meat inspection and warrants creation of awareness among general public.

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