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**Original Article**

# Prevalence of *Linguatula serrata* Infection in Domestic Bovids Slaughtered in Tabriz Abattoir, Iran

***M Hami[1], *SR Naddaf[2], I Mobedi[3], M Zare-Bidaki[4], SS Athari[5], B Hajimohammadi[5], G Anaraki-Mohammadi[3]***

[1] *Dept. of Pathobiology, School of Veterinary Medicine, Islamic Azad University, Malekan Branch, Malekan, Iran*

[2] *Department of Parasitology, Pasteur Institute of Iran, Tehran, Iran,*

[3] *Dept. of Medical Parasitology and Mycology, School of Public Health, Tehran University of Medical Sciences, Iran*

[4] *Dept. of Microbiology, School of Medicine, Rafsanjan University of Medical Sciences, Rafsanjan, Iran*

[5] *Faculty of Veterinary Medicine, University of Urmia, Urmia, Iran*

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## Abstract

**Background:** Linguatulosis is a rare zoonotic parasitic infection, in which human plays the role of both definitive and intermediate host and can be occasionally infected. This study determines the status of infection in livestock and its potential risk to men in the northwestern province of Azarbaijan-e-Sharghi, Iran.

**Methods:** In a cross-sectional study from June 2007 to June 2008, 800 slaughtered animals including 400 cattle and 400 buffaloes from Tabriz abattoir in Azarbaijan-e-Sharghi Province were randomly selected and examined for *L. serrata* nymphs. After primary macroscopical inspection, all liver and lung samples were cut to small pieces, treated with a tissue digestion method and checked macroscopically and microscopically for free or encapsulated nymphs.

**Results:** Out of 800 animals, 3 (0.38%) were found to be infected with *L. serrata* nymphs and the prevalence of infection in cattle and buffaloes was determined to be 0.25% and 0.5%, respectively.

**Conclusion:** *Linguatula* infection occurs as an endemic zoonosis in the study area and has an active transmission life cycle.

**Keywords:** Linguatula serrata, *Linguatulosis, Bovid, Iran*

* **Corresponding Author:** Tel & Fax: +98 21 66968855, E-mail: snaddaf_2001@yahoo.com

## Introduction

Linguatulosis (linguatuliasis) is a rare zoonotic parasitic disease resulted from invasion of the body by wormlike parasites of the genus *Linguatula*, also known as tongue worms (1). The most common reported species involved in linguatulosis is *Linguatula serrata*; (family Linguatulidae, order Porocephalida, phylum Pentastomida), which is commonly classified between annelids and arthropods (2, 3).

The human infection is often contracted through ingestion of contaminated food and may manifest as nasopharyngeal, visceral, or ocular forms (4, 5). *L. serrata* commonly resides within the nasal passages of canines and felines and occasionally humans. The symptoms in humans are nasopharyngitis, violent coughing, asphyxiation, edematous congestion of gums, tonsils and eustachian tube, aural pruritus, deafness, frontal headache, sneezing, lacrimation, coryza, yellow nasal discharge, facial edema, vomiting and breathing blockage (1, 2, 6).

The life cycle of linguatulids involves two hosts. Development of larva occurs in an intermediate host, which begins by ingesting the eggs present in sputum, feces, or body cavity of definitive hosts. Female worms produce several million eggs with the size of 90-70 µm. The eggs, containing fully-grown larvae, are discharged with nasal excreta of definitive hosts including carnivorous, reptiles, birds and mammals into water or on vegetables. Once the eggs are ingested by intermediate hosts (e.g., fish, cattle, sheep, rabbits, rodents, ungulates) they hatch in the intestine and release the larvae, which then burrow through the intestine wall and lodge themselves in liver, lungs and other viscera. Within the viscera, the larvae mature to a pupa-like stage, and then to a nymph (infective larva) stage in which hooks, annular rings, and spines develop. After maturation to the nymph stage, the infective larva migrates to the pleural cavity. Once the intermediate host is eaten by a natural definitive host, the larvae migrate to the nasal cavities, where they mature into adults.

Humans may serve both as intermediate host with encapsulated larvae in inner organs, and definitive hosts with adult worms in nose(7). Human infections can result from ingesting of raw or undercooked visceral tissues of the intermediate hosts such as sheep, goats, cattle, buffaloes or other herbivores harboring the larval stages of the parasite. The infection can also occur through drinking of water or ingestion of fruits and vegetables contaminated with *Linguatula* eggs. Halzoun Syndrome (Middle East) or Marrara syndrom (Sudan) are the human diseases described as pentastomids infection of men (2). Human infection with *L. serrata* has been reported from different parts of the world including tropical regions of North and South America, Europe, Asia, Africa and Australia (8, 9). There are also some reports of the infection from different regions of Iran (4, 10-14).

This study determines the prevalence of *Linguatula* infection in cattle and buffaloes slaughtered in Tabriz abattoir and show the epizootic status of the infection in the area.

## Materials and Methods

From June 2007 to June 2008, 800 slaughtered animals including 400 cattle and 400 buffaloes (100 cattle and 100 buffaloes in each season) were randomly selected from Tabriz abattoir in northwestern province of Azarbaijan-e-Sharghi and examined for *L. serrata* nymphs. On visiting the abattoir in each season of the study year, liver and lung samples of the slaughtered animals were collected and transferred to our laboratory. The samples were cut to small pieces and

were macroscopically examined for free or encapsulated nymphs. A tissue digestion method was also used to check the specimens more precisely (3). The digestion solution was prepared by adding 5g of pepsin enzyme (Merck, 7178) to 25 ml of hydrochloric acid (Merck, 374) and topping it off with water to a total volume of 1 litter.. One hundred gr of minced tissues was submerged in digestion solution and incubated at 37 °C for 24 hours. After digestion process, the suspensions were transferred to Petri dishes and examined under a stereoscope and the suspected ones were checked by optic microscope with 4× to 40× magnifications. The number of nymphs was recorded in positive cases. Morphologic characteristics of recovered nymphs were studied using a light microscope equipped with a Camera Lucida apparatus.

## Results

Out of 800 examined carcasses, three (0.38%) were found to be infected with *L. serrata* nymphs. Out of 400 cattle, only one and out of 400 buffaloes two harbored the nymphs. Thus, the prevalence rate of infection in cattle and buffaloes was 0.25% and 0.5%, respectively. Worm burden in three positive cases was 5, 3 and 1. Seasonal frequency of positive cases and organ involvement are shown in Table 1. All *L. serrata* nymphs were recovered by the digestion method.

There was no considerable morphological difference between recovered *L. serrata* nymphs from cattle and buffaloes (Fig. 1).

**Table 1:** Season frequency and organ involvement of positive cases

| Positive cases | Season of sampling | Stock species | Organ involved | Number of isolated nymphae |
|---|---|---|---|---|
| Case 1 | winter | cattle | liver | 5 |
| Case 2 | summer | buffalo | liver | 1 |
| Case 3 | summer | buffalo | lung | 3 |

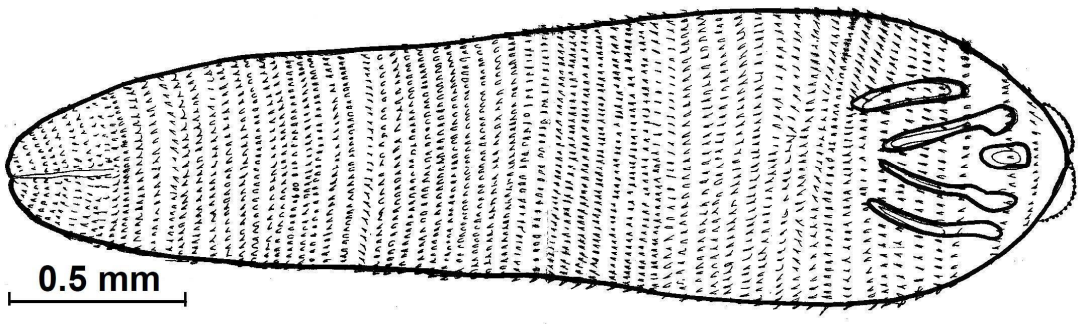


**Fig. 1:** Microscopic drawing of a *L. serrata* nymph recovered fromcattle in this study

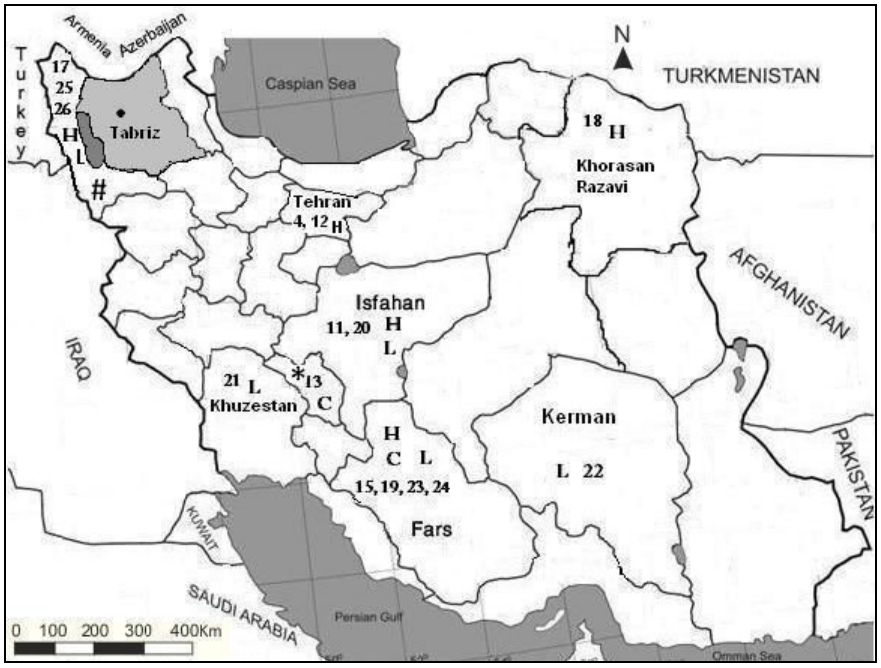


**Fig. 2:** Geographical distribution of *L. serrata* Infection in Iran.
Regions from which *L. serrata* infection has been reported: (H) human Linguatuliasis; **(C**) carnivores infection; (L) livestock infections; ▭ the present Study Area; numbers represents the location of studies cited in this paper according to the allocated number in the reference section; (*) Chaharmahal va Bakhtiari province and **( # )** Azarbaijan-e-Gharbi province.

## Discussion

Up to present, ten cases of human infection with *L. serrata* have been reported from different regions in Iran including Tabriz, Tehran, Mashhad, Kashan, Azarbaijan-e-Gharbi and Shiraz (Fig. 2) (4, 11, 12, 14-18). Regarding animals as definitive hosts, there are merely two reports on infection of stray dogs with *L. serrata* from Iran (Fig. 2). In the first study 143 stray dogs from Shahrekord, the center of Chaharmahal-e-Bakhtiari Province, were examined and 89 (62.2%) showed to harbor pentastomid parasites in their nasal cavities (13). In the second study in Shiraz, the center of the Fars Province, nasopharyngeal area, nasal turbinates, sinuses, eustachian tubes, and brain of 85 stray dogs were examined and 65 (76.5%) showed to harbor *L. serrata worms.* Most worms were found to be in nasal tubinates, the posterior region of nasopharynx area. Only four were recovered from the frontal sinus and no worm was detected in eustachian tubes, maxillary sinuses, and brain cavity. Age, sex, weight, and geographical locations had no significant effects on the prevalence of the parasite. The average worm burden was 4.06 per infected dog. They concluded that the rate of infection in dogs and possibly other carnivores, herbivores, and man was high in Shiraz and suggested adoption of strict control measures to prevent the risk of human infection with this zoonotic disease (19).

There are also some reports on *L. serrata* infection of intermediate hosts in Iran (Fig. 2). Examination of livers and mesenteric

lymph nodes of 400 one-humped camel (*Camelus dromedarius*) from Najaf-Abad in Isfahan province showed that 84 (21.0%) of them harbored *L. serrata* nymphs in lymph nodes and 18 (4.5%) in livers (20). Saiyari et al. examined 3958 goats from Ahwaz in Khuzestan Province and detected nymphs of *L. serrata* in the lungs of 9 (0.23%) cases. The nymphs were embedded in 4-6 mm hemorrhagic cysts below the surface of the pleurae in both caudal lobes. The numbers of cysts varied from 10 to 15 in each lobe of lung (21). One *L. serrata* nymph was also detected in lungs of a native sheep from Kerman Province (22). The rate of infection with *L. serrata* nymphs in livers and mesenteric lymph nodes of 204 goats from Shiraz in Fars Province, was 6.4% and 29.9%, respectively (23). While out of 200 sheep, in the same area, 3.0% and 11.5% had nymphs in livers and mesenteric lymph nodes, respectively (24). Examination of 871 mesenteric lymph nodes from 110 native cattle in Urmia slaughterhouse showed that 44% of the cattle were positive for *L. serrata* infection. The number of parasites isolated from each infected lymph node varied from 1 to 69 with a mean of 5.48 (25).Tavassoli et al studied 1333 mesenteric lymph nodes from 200 native sheep from Urmia area in Azarbaijan-e-Gharbi and showed that 105 (52.5%) of them were infected with the parasite nymphs (26).

The earlier study on *L. serrata* infection in bovids of Urmia showed a higher infection rate than ours (25). The difference is probably due to examination of mesenteric lymph nodes instead of livers or lungs. Many studies have shown that worm burden and infection rate in mesenteric lymph nodes to be much higher than liver or any other visceral organ (20, 23, 24, 26). Therefore, it is necessary to emphasize on mesenteric lymph nodes inspection by veterinary professionals in slaughterhouses. Besides, the fact that all *L. serrata* nymphs were recovered by tissue digestion method indicates that mere macroscopic examination of specimen is not a sensitive and reliable method for the diagnosis of *Linguatula* infection in livestock.

Two reports on nasopharyngeal infection of human with adult *L. serrata* from Azarbaijan-e-Sharghi (16, 17) together with the findings of this study indicate the existence of an active transmission cycle of infection in the area. The present study in accordance with other reports indicates that *Linguatula* infection is an endemic zoonosis in many parts of Iran. Since livestock are a major source of infection for human and other carnivores, adopting proper and reliable diagnostic methods to detect infection in slaughterhouses along with introducing preventative measures are undeniable responsibilities for both veterinary and medical authorities.

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