Prevalence of *Linguatula serrata* nymphs in slaughtered sheep in Isfahan province, southwest of Iran

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Abstract *Linguatula serrata*, well known as tongue worm; is an aberrant cosmopolitan parasite, which inhabits the carnivorous mammals (especially *Canidae*) respiratory system. The discharged eggs infect many plant feeder animals including human that produces visceral and nasopharyngeal linguatulosus which is known as Marrara syndrome in man. In current study, the prevalence rate of infection with *L. serrata* nymphs in mesenteric and mediastinal lymph nodes (MLNs) of slaughtered sheep was investigated in Esfahan Province, Iran. The MLNs of 506 slaughtered sheep, including 236 females and 270 males, were examined for *L. serrata* nymphs by cutting the MLNs longitudinally and then microscopic studies for *L. serrata* nymphs. Sheep were categorized into four age groups, including <1 year, 1–2 years, 2–3 years and >3 years. Results showed that 11.66 % of examined sheep were infected with *L. serrata*. Age had significant effect on the prevalence rate of this parasite in sheep (infection in sheep with >3 years old was more than other groups significantly) and sex had no significant effect on the prevalence rate of this parasite in sheep. Infection rate in winter was significantly lower than infection rate in spring; but there were no significant differences between the other

seasons. As high prevalence rate of infection in sheep, suggesting possibly similar high rate of infection in other animals and man in the investigated area, which this emphasizes undertaking strict control measures to reduce risk of zoonotic outbreaks. This study was demonstrated infection rate of *L. serrata* in sheep in central parts of Iran.

Keywords Human · Iran · Isfahan province ·
Linguatula serrata · Sheep

Introduction

Linguatula serrata Frohlic 1789, cosmopolitan parasite, is a member of small group of parasites from phylum Pentastomida (Gosling 2005; Muller 2002). Adult infects the nasal sinuses and nasopharynx of carnivorous mammals, especially *Canidae* and probably *Hyaenidae* and *Felidae* (Khalil 1970; Riley 1986). A wide range of mammals are intermediate hosts for *L. serrata*, but herbivores such as camels, cattle, goats, sheep and other ruminants are the best hosts for development of parasite's nymphal stages (Alcala-Canto et al. 2007). Eggs containing fully developed larvae are discharged into the environment by nasal secretion and ingested by intermediate hosts in which they develops to nymphal stage in various organs, particularly in mesenteric lymph nodes (Eslami et al. 1976; Berger and Marr 2006; Khalil and Schacher 1965; Soulsby 1982). Man can be infected with both adult and nymphal stages of *L. serrata* (Tabibian et al. 2012). Linguatulosus in humans has been reported from various parts of the world, particularly in countries of the Middle East, Africa, America and in Southeast Asia (Gardiner et al. 1984; Acha and Szyfres 2003; Baird et al. 1988; El-Hassan et al. 1991; Yagi et al. 1996; Lazo et al. 1999). Nasopharyngeal linguatulosus,

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which is known as Halzoun or Marrara syndrome, is the common form of infection in man and is often produced following consumption of raw or undercooked infected viscera (liver, lung and lymph nodes) of infected animals (El-Hassan et al. 1991; Beaver et al. 1984; Drabick 1987). This parasite has been reported in humans in Iran (Hodjati and naghili 1987; Fata et al. 1994; Sadjjadi et al. 1998; Maleky 2001). Several studies have shown the prevalence of *L. serrata* infection in animals including, dogs (Oryan et al. 1997, 2008; Khalil 1970; Tavassoli et al. 2000; Meshgi and Asgarian 2003), camels (Mobedi et al. 1998; Oryan et al. 1993; Tajik et al. 2007; Pourjafar et al. 2007; Shakerian et al. 2008), buffaloes (Sisakumar et al. 2005), sheep (Esmail-Nia et al. 2000; Shekarforoush et al. 2004; Tavassoli et al. 2007; Ravindran et al. 2008; Nourollahi-Fard et al. 2011), cattle (Nourollahi-Fard et al. 2010a) and goats (Saiyari et al. 1996; Razavi et al. 2004; Nourollahi-Fard et al. 2010b).

This study aimed to determine the prevalence of *L. serrata* nymphs in MLNs of slaughtered sheep in Esfahan Province, Central Iran.

Materials and methods

Lymph nodes of 506 slaughtered sheep at slaughterhouses of Esfahan province located in central part of Iran were examined for *L. serrata* nymphs from January 2011 to December 2012. After determining the sex, sheep were divided into four age groups (including <1 year, 1–2 years, 2–3 years and >3 years) using the eruption of permanent incisor teeth criteria as already described. At least, four

mesenteric and mediastinal lymph nodes (MLNs) form each animal; and totally 2,024 lymph nodes were collected in phosphate buffer saline (PBS) and transferred to the Parasitology Laboratory of Veterinary Faculty of Shahrekord University. Then each lymph node was cut longitudinally and tested using a dissecting microscope for *L. serrata* nymphs. Nymphs identified based on the key of Soulsby (1982). The number of collected nymphs from each node was recorded and then nymphs stored in PBS at 4 °C for further studies. Digestive method was carried out for detection of more nymphs which were not recognized in first method. For this purpose, 6 g pepsin was solved in 10 ml hydrochloric acid, and then 600 ml of distilled water was added. After 24 h in 37 °C samples were examined.

Statistical analysis

Chi square test (SPSS version 17.0) was used to compare the relative frequency of infection among different ages, sexes and seasons. A value of $P < 0.05$ was considered significant.

Results

The results showed that MLNs in 59 sheep (11.66 %) were infected with *L. serrata* nymphs. The number of collected nymphs from each infected lymph node varied from 1 to 17 (approximately mean = 10). The infection rate in mesenteric lymph nodes was significantly higher than the infection rate in mediastinal lymph nodes (Table 1). Infection rate had no significant difference

Table 1 Infection rate by *Linguatula serrata* nymphs in MLNs in slaughtered sheep in different sex, age and season

	Lymph node					
	Mesentric		Mediastinal		Mesentric + mediastinal	
	Animals no.	Infected no. (%)	Animals no.	Infected no. (%)	Animals no.	Infected no. (%)
Sex						
Male	270	27 (10.0)	270	7 (2.59)	270	6 (2.22)
Female	236	30 (12.7)	236	7 (2.96)	236	6 (2.54)
Total	506	57 (11.3)	506	14 (2.76)	506	12 (2.37)
Age (year)						
<1	350	17 (4.85)	460	0 (0)	460	0 (0)
1–2	71	15 (21.120)	91	3 (3.29)	91	3 (3.29)
2–3	33	8 (24.24)	40	4 (10)	40	4 (10)
>3	52	17 (32.69)	29	7 (24.13)	29	5 (17.24)
Season						
Spring	100	12 (12)	100	3 (3)	100	2 (2)
Summer and autumn	306	36 (11.76)	306	9 (2.94)	306	9 (2.94)
Winter	100	9 (9)	100	2 (2)	100	1 (1)

between male and female sheep (Table 1). There was significant difference between the groups with different ages (Table 1). Infection rate in winter was significantly lower than infection rate in spring; but there were no significant differences between the other seasons (Table 1).

Discussion

The prevalence of linguatulososis in dogs have been determined in different part of Iran, which found to be 76.2 % in Shiraz (Oryan et al. 2008), 65.5 % in Shahre-Kord (Meshgi and Asgarian 2003) and 76.47 % in Fars province (Oryan et al. 1997). In Bursa, Turkey, 20 % of dogs have been found infected (Akyol et al. 1995). Many studies were done on prevalence of *L. serrata* in various domestic ruminants in Iran and other parts of world (Youssefi et al. 2012; Mirzaei et al. 2011; Yakhchali et al. 2009; Tavassoli et al. 2007; Tajik et al. 2006; Razavi et al. 2004). The infected nymphs were obtained from different visceral organs and in most studies MLNs were evaluated because the nodes are first place of infection with *L. serrata* nymph. Therefore, possibility of infection in MLNs is higher than other visceral organs (Shakerian et al. 2008; Yakhchali and Tehrani 2011). Only, few studies on the prevalence of *L. serrata* nymphs in sheep were conducted in some area of Iran, in Mashhad (Tajik et al. 2007) and in Najaf-Abad (Pourjafar et al. 2007; Shakerian et al. 2008). In Egypt 4.9 % of MLNs from examined camels were infected (Razavi et al. 2004). In recent study, from 232 examined goats, the parasite was reported in MLNs of goats, that was higher than the reports of Oryan et al. (1993) and Wahba et al. (1997) with infection rate of 7.5 and 4.9 %, respectively. Above mentioned data were less than that reported by Tajik et al. (2007) and Pourjafar et al. (2007) with the infection rate of 75 and 35 %, respectively. Therefore, our results showed that Esfahan province is an endemic area for linguatulososis in sheep. There was significant difference among different age groups and among different seasons, but, there was no significant difference in the rate of infection of the male and female sheep. Due to the fact that sheep meat is one of the common food in Iran, particularly in central part of Iran such as Esfahan Province, the high rate of infection with the *L. serrata* nymphs in sheep in this area clearly indicates the high risk of transmission of the disease from consumption of raw or undercooked sheep viscera to human beings. Also, these results show that infection rate in final hosts such as dogs must be high, and this is the other risk factor for human and ruminant linguatulososis. Furthermore, sheep offal's can be transmit the infection to final hosts and cause the maintenance of life cycle of parasite in this area. Based on high prevalence of infection in sheep we suppose that the rate

of problems associated with linguatulososis in humans is higher in this province.

Results obtained from this research, suggests that presence of such gross changes in the color and consistency of the MLNs could be considered as an indication of infection with nymphs of *L. serrata* and elimination of such lymph nodes is necessary to interrupt the life cycle of parasite.

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