

Prevalence of *Linguatula* sp., a food-borne zoonotic aberrant arthropod, in river buffaloes slaughtered at Tabriz slaughterhouse, Iran

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Background: Canids and herbivores are the definitive and intermediate hosts of *Linguatula* sp., respectively.

Methods: Mesenteric lymph nodes (n=32 525) were randomly collected from 7585 buffaloes from July 2016 to July 2019 and examined macroscopically.

Results: Results showed that 388 (5.11%) buffaloes were infected. The intensity of infection was determined to be 3.07 ± 0.07 . Significant statistical association was identified between infection rate and age and sex. Although there were no significant differences in the infection rate over different seasons, the highest infection rate was observed in autumn.

Conclusions: These data highlight the importance of inspection at slaughter.

Keywords: Buffalo, food-borne zoonosis, Iran, *Linguatula*

Introduction

Belonging to the class Pentastomida, *Linguatula* sp. is a zoonotic abnormal arthropod,¹ the adult form of which usually invades the respiratory system of carnivores, the final host, while the nymph form invades the mesenteric lymph nodes (MLNs), liver, lungs and spleen of herbivores, the intermediate host. In most cases, the final hosts show no clinical signs; however, massive infections can cause rhinitis and nasopharyngitis with chronic sneezing and/or coughing, purulent nasal discharge and/or nose bleeding. Human infection with *Linguatula* occurs following consumption of raw or undercooked infected internal organs of the intermediate host and can manifest as a nasopharyngeal infection resulting in nasal discharge, coughing and headaches.

Most research on the epidemiology of *Linguatula* and sources of human infection have focused on small ruminants and cattle, with *Linguatula serrata* being identified in these hosts. However, recent molecular studies indicate that parasites identified as *L. serrata* morphologically might be a group of closely related species, with those from Iran forming a distinct clade phylogenetically.^{1,2} This increases the importance of understanding prevalence and factors contributing to prevalence in different

hosts. Therefore the current study was undertaken to evaluate the prevalence of the nymph stage of *Linguatula* in MLNs of Iranian river buffaloes, an intermediate host for which there are limited data, and to assess the effect of age, sex and season on prevalence.

Materials and methods

MLNs were collected from 7585 buffaloes categorized in three age groups (<2, 2–3 and >3 y) from July 2016 to July 2019 during 3–4 d/week after slaughter at the Tabriz abattoir. The samples were processed following previously published protocols.³ After recovering nymphs, they were flattened, dehydrated in ascending grades of ethyl alcohol and examined under a stereomicroscope (10× and 40× magnification).

The effects of host factors (age and sex) and season on the prevalence was assessed using the χ^2 test with SPSS version 21 (IBM, Armonk, NY, USA). A p-value <0.01 was considered to be statistically significant.

Table 1. The prevalence of *Linguatula* sp. nymphs in MLNs of buffaloes slaughtered at Tabriz slaughterhouse, Iran, based on sex, age and season

Year	Infested buffaloes, n/N (%)	Infested MLNs, n/N (%)	Nymphs (mean± standard error)	Sex ^a , n/N (%)		Age ^a (years), n/N (%)			Season ^b , n/N (%)			
				Male	Female	<2	2-3	>3	Spring	Summer	Autumn	Winter
2016	155/2585 (5.99)	400/12925 (3.09)	2.06 ± 0.01	45/1125 (4)	110/1460 (7.53)	43/925 (4.64)	52/893 (5.82)	55/767 (9.49)	30/579 (5.18)	25/689 (3.62)	70/841 (8.32)	30/476 (6.30)
2017	65/1200 (5.41)	350/5980 (5.85)	3.50 ± 0.11	20/500 (4)	45/700 (5.42)	10/350 (2.85)	17/450 (3.77)	38/400 (9.50)	8/300 (2.66)	10/250 (4)	32/350 (9.14)	15/300 (5)
2018	98/2300 (4.26)	490/11420 (4.29)	3.27 ± 0.09	33/1100 (3)	65/1200 (5.41)	22/782 (2.81)	28/800 (3.50)	48/718 (6.68)	15/580 (2.58)	17/600 (2.83)	35/500 (7)	31/620 (5)
2019	70/1500 (4.66)	300/7500 (4)	2.93 ± 0.08	20/600 (3.33)	50/900 (5.55)	15/500 (3)	20/500 (4)	35/500 (7)	15/350 (4.28)	10/350 (2.85)	28/500 (5.60)	17/300 (5.66)
Total	388/7585 (5.11)	1540/32525 (4.73)	3.07 ± 0.07	118/3325 (3.54)	270/4260 (6.33)	90/2557 (3.51)	117/2643 (4.42)	140/2385 (5.87)	68/1809 (3.75)	62/1889 (3.28)	165/2191 (7.5)	93/1696 (5.48)

^aLinguatula sp. prevalence was significantly higher in female buffaloes and in buffaloes > 3 y of age (p<0.0001).
^bLinguatula sp. prevalence was higher in autumn, but there were no significant seasonal differences. Spring (April–June); summer (July–September); autumn (October–December); winter (January–March).

Results and discussion

A total of 32 525 MLNs were examined from 7585 buffaloes slaughtered at the Tabriz abattoir.

A prevalence of 5.11% (Table 1) was found, which is similar to the 5.73% reported by Tajik and Sabet Jalali.⁴ However, it is lower than the prevalence reported in buffaloes in India and South Africa and small ruminants in Iran.^{2,3,5} The observed differences in the prevalence can potentially be related to the heterogeneous forage habitats of buffaloes, climate, and annual variation in or low exposure to dogs, the definitive hosts.³ It also could be related to differences in the *Linguatula* strain or species, with only those in buffaloes from South Africa being analysed genetically and determined to be *Linguatula nuttalli*; those found in other studies in Iran have not yet been genetically analysed.^{1,2} The *Linguatula* recovered in the study presented here resemble those found in small ruminants in Iran (unpublished morphological data), which potentially belong to an as yet unnamed species that could be related to what has been found in South Africa.^{1,2}

The finding of this study regarding the significantly higher prevalence in female buffaloes than male buffaloes (p<0.05) is in agreement with other studies of *Linguatula* in Iran, in contrast to what has been seen in buffalo in India.³⁻⁵ The observed differences in various studies can be attributed to the age of female buffaloes, which are older than male animals at slaughter, and a true parasite preference for older animals.³ Our findings revealed that the parasitic infestation rate in MLNs of buffaloes increased with the age of the animal (p<0.001), which was also reported by Tajik and Sabet Jalali.⁴ The lower prevalence in younger animals is probably due to the fact that it takes 5–6 months for nymphs to develop. The infection rate was not significantly different by season (p>0.01). However, in the autumn the prevalence trended towards being higher than in the other seasons, which was similar to the results of studies of sheep and goats from other regions of Iran.³ This discrepancy may be related to grazing and exposure periods. If intermediate hosts grazing in spring swallow infected eggs, *Linguatula* sp. nymphs will emerge in the autumn, as 6 months is required for the egg to develop into a larva. In northwest Iran, where this study was carried out, suitable weather conditions for egg development occur in the late spring, coinciding with the time of grazing sheep, therefore nymphs emerge in MLNs in the autumn.

Conclusions

The high prevalence of infestation observed in river buffalo is of concern owing to the zoonotic nature of the parasite. Genetic analysis is needed of *Linguatula* in buffaloes and people to better understand the zoonotic risk.

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