

EPIDEMIOLOGICAL STUDY ON TRICHINELLOSIS IN THE FOX (*VULPES VULPES*) IN TUSCANY (CENTRAL ITALY)

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ABSTRACT - During the years 2004-2005, 112 foxes (*Vulpes vulpes*) and 4 badgers (*Meles meles*) were caught in different areas of Tuscany (Central Italy) and examined for *Trichinella* infection, using the diagnostic technique of artificial digestion through Stomacher. No animal was positive for *Trichinella* larvae. According to our results, Tuscany can be considered a low-risk area for trichinellosis in the fox. In this region the presence of the parasite cannot be ruled out, two cases of infection being reported in 1993.

Key words: Red fox, badger, *Trichinella britovi*, wild cycle, prevalence, central Italy

RIASSUNTO – *Epidemiologia della trichinellosi della volpe (Vulpes vulpes) in Toscana (Italia centrale)*. Nel corso degli anni 2004-2005, 112 volpi (*Vulpes vulpes*) e 4 tassi (*Meles meles*) sono stati catturati ed esaminati per la presenza di infestazione da *Trichinella* in differenti aree della Toscana (Italia centrale). L'indagine di laboratorio è stata condotta mediante digestione artificiale tramite Stomacher. Nessun animale è risultato positivo. Da questi risultati si può ritenere la Toscana una regione a basso rischio di infezione. La presenza del parassita non può però essere esclusa totalmente. Infatti, nel 1993 sono stati riportati due casi di infestazione.

Parole chiave: Volpe, tasso, *Trichinella britovi*, ciclo selvatico, prevalenza, Italia centrale

INTRODUCTION

Trichinellosis is a typical zoonosis widespread all over the world, with the exception of the Antarctic continent. In domestic and wild animals trichinellosis is caused by at least eight species

and three unnamed genotypes of nematodes of the genus *Trichinella*. Recent taxonomic studies have identified two clades, one including *Trichinella* species that induce the formation of a capsule in the muscular tissue of the host, and one which does not

produce a capsule. The species and genotypes of the first clade - *T. spiralis*, *T. nelsoni*, *T. britovi*, *T. murrelli*, *T. T9*, *T. T8*, *T. nativa*, *T. T6* - parasitize only mammals. In the non-encapsulated clade, *T. pseudospiralis* infects mammals and birds (Ainsworth *et al.*, 1993), whilst *T. papuae* and *T. zimbawensis* infect mammals and reptiles (Pozio and Zarlenga, 2005).

The parasitosis is diffused through two cycles: 1) a wild cycle, main hosts being scavengers such as the fox (*Vulpes vulpes*), wolf (*Canis lupus*), bear (*Ursus arctos marsicanus*), raccoon (*Procyon lotor*), hyena (*Hyena hyena*) and jackal (*Canis aureus*), but also in omnivores such as wild boars (*Sus scrofa*) and rodents, 2) a domestic cycle, involving in particular swine and sinanthropic animals (mainly rats), but also other animals such as horses (Murrell *et al.*, 2004). It has been recently pointed out (Guberti *et al.*, 2004) that, wherever biocenoses are structured, the parasite can easily survive because of the number of available suitable host species and their ecological relationships.

In Italy *T. spiralis* has been reported only in imported horses in Apulia and Emilia-Romagna regions and in wild boars in Piedmont (Pozio *et al.*, 2001), whilst in autochthonous animals *T. britovi* has been frequently detected (La Rosa *et al.*, 1989). In Italy, *T. pseudospiralis* has been found only in two owls (*Strix aluco* and *Athene noctua*; Pozio *et al.*, 1999, 2001), whilst in France it has been recently identified as responsible for trichinellosis in four people who had eaten the meat of a wild boar hunted in Camargue (Ranque *et al.*, 2000).

Trichinellosis in Italy is now caused only by *T. britovi* through the wild cycle (Pozio *et al.*, 1992). The fox represents the main reservoir of this parasite so the monitoring of *T. britovi* presence in this host provides a reliable indication of the epidemiological situation (Pozio, 1991). The wolf can also sustain the biological cycle of the parasite, but its epidemiological role is less important due to the small size of its population. In research conducted by Stancampiano *et al.* (1993), 19% of wolves were positive for *T. britovi*, with the highest prevalence in the district of Rieti (central Italy). The parasite has been found also in the badger (*Meles meles*), beech marten (*Martes foina*), brown rat (*Rattus rattus*), porcupine (*Hystrix cristata*) and brown bear (*Ursus arctos marsicanus*) (Fico *et al.*, 1988; Pozio *et al.*, 2001; Orusa *et al.*, 2003).

Foxes can indirectly infect man through other animals such as wild boars, pigs and horses (Pozio *et al.*, 2000). The direct transmission from fox to man is also possible through the consumption of undercooked fox meat, as occurred in Trentino in 1961 (Pampi-gliione and Doglioni, 1971) and in Basilicata in 1985 (Pozio, 1987b). The direct transmission among wild animals may be facilitated when the carcasses of wild carnivores are abandoned after hunting and fur taking (Pozio, 1991). Several studies on trichinellosis in wild carnivores and in particular in the fox are available for Italy, particularly in the second half of the 20th century: Leinati and Marazza (1959) (Italian Alps, 32.8%); Nardi (1959) (Apulia, 8.5%); Cagnolati and Merighi (1959) (Rome, 16.6%); Marazza (1960)

(Lombardy, 58.3%); Morozzi (1960) (Umbria, 19.4%); Mantovani (personal communication) (1960-62); Baldelli and Frescura (1963) (Umbria, 33%); Goffredo (1971) (Apulia); Soldati *et al.* (1976) (Emilia-Romagna, 2.7%); Cancrini *et al.* (1982) (Rome, 0.85%); Corselli *et al.* (1983) (Rome, 1.1%); Piergili Fioretti *et al.* (1986) (Umbria, 31.5%); Panebianco and Marra (1987) (Calabria, 5.88%); Pozio *et al.* (1987a) (Northern Italy); De Carneri and Di Matteo (1988) (Northern Italy, 3.9%); Fico *et al.* (1988) (Abruzzo, 3.3%); Boni *et al.* (1988) (Lombardy, 3.2%); Fico *et al.* (1988) and Scaramozzino *et al.* (1993) (4.4% in foxes, 0.4% in wild boars, and 28% in wolves; zones surrounding the National Park of Abruzzi); Iori *et al.* (1990) (Lazio, 1.1%); Rossi and Dini (1990) and Rossi *et al.* (1992) (Turin, 2.3%); Balbo and Rossi (1992) (Piedmont and Liguria, 6.6%); Stancampiano *et al.* (1998) (Vicenza province, 1-1.9%); Remonti *et al.* (2005) (Aosta Valley, 3.5%). Di Matteo *et al.* (1990, 1991) suggested that the marked decrease of prevalence of the infection, which occurred between the 1950s and 1986 (34% - 4.2%) in foxes from Lombardy was due to the strong hunting pressure exerted for the control of rabies. In Sardinia, epidemiological surveys (Arru, 1962; Pintore *et al.*, 1996) have always given negative results, but in 2004-05 two outbreaks of human trichinellosis were detected in Orgosolo, where 11 people were hospitalised after eating sausages from a house raised pig (Cossu *et al.*, 2006). In Sicily, from 1958 to 1987 (Tu-mino *et al.*, 1987) all the examined foxes were negative. In Tuscany trichinellosis has been repor-

ted by Pozio *et al.* (1987b), who assumed presence of the parasite in wild boars of the Grosseto province on the basis of indirect immunofluorescence tests. In 1993 a case of trichinellosis was reported in a pig living in a natural reserve of Arezzo province (Pagni *et al.*, 1993). Moreover, in 1993 a positive fox was found in Lucca province (Magi, pers. com.). Searches for trichinellosis in the fox in Tuscany, conducted by Marconcini *et al.* (1970) and by Magi and Sassetti (1990), gave negative results. Tuscany is considered a low-risk area for trichinellosis as indirectly confirmed by the absence of the infection in wild boars from various zones of the province of Grosseto in 1989 (Magi *et al.*, 1989) and by research carried out by Martini *et al.* (2005) on wild boars from the whole of Tuscany.

METHODS

In the period 2004-2005 carcasses of foxes and badgers, killed during a control campaign (about 95% of the sample) or in road accidents, have been examined. The animals were sent to the Department of Animal Pathology of Pisa University from four different zones: Monte Amiata (Grosseto), in areas localized between 630 and 900 m a.s.l., the hills around Cecina (Livorno), in areas localized between 250 and 550 m a.s.l., the municipalities of Cascina and Bientina (Pisa province), situated at sea level, and the hills around Siena, where foxes were collected in areas ranging between 250 and 400 m a.s.l. (Fig. 1).

For each animal the area of origin, sex, age (inferred by the status of the teeth, according to Toschi, 1965), and weight of the animal were noted. Foxes were grouped according to the altitude of collection

(above or below 500 m a.s.l.) and age (older or younger than 2 yr). These cut-off values were chosen since in Western Europe the prevalence of the parasite is higher in foxes of more than 2-3 years and from areas above 500-600 m a.s.l. (Rossi *et al.*, 1992; Guberti *et al.*, 2004, Balestrieri *et al.*, 2007). In Italy, however, this pattern does not seem to be found all over the country (Balestrieri *et al.*, 2007).

Muscle samples were collected from the diaphragm, the tongue and the anterior tibial muscles. In order to increase the chance of detecting *Trichinella* larvae more

than 10 grams of muscle sample underwent *in vitro* digestion by Stomacher (Pepsin 1:10.000 U.I; 17.5% HCl; digestion for 25' at 41°C and filtration with sieve) (Thomsen, 1978, 1997), according to the Commission Regulation (EC) N. 2075/2005. Artificial digestion was preferred to serological tests because the use of this last method to detect *Trichinella* infection in wild and domestic animals has still not been standardised (Gamble *et al.*, 2004).

The prevalence of *Trichinella* infection and its 95% exact binomial confidence interval were evaluated (Armitage *et al.*, 2002).

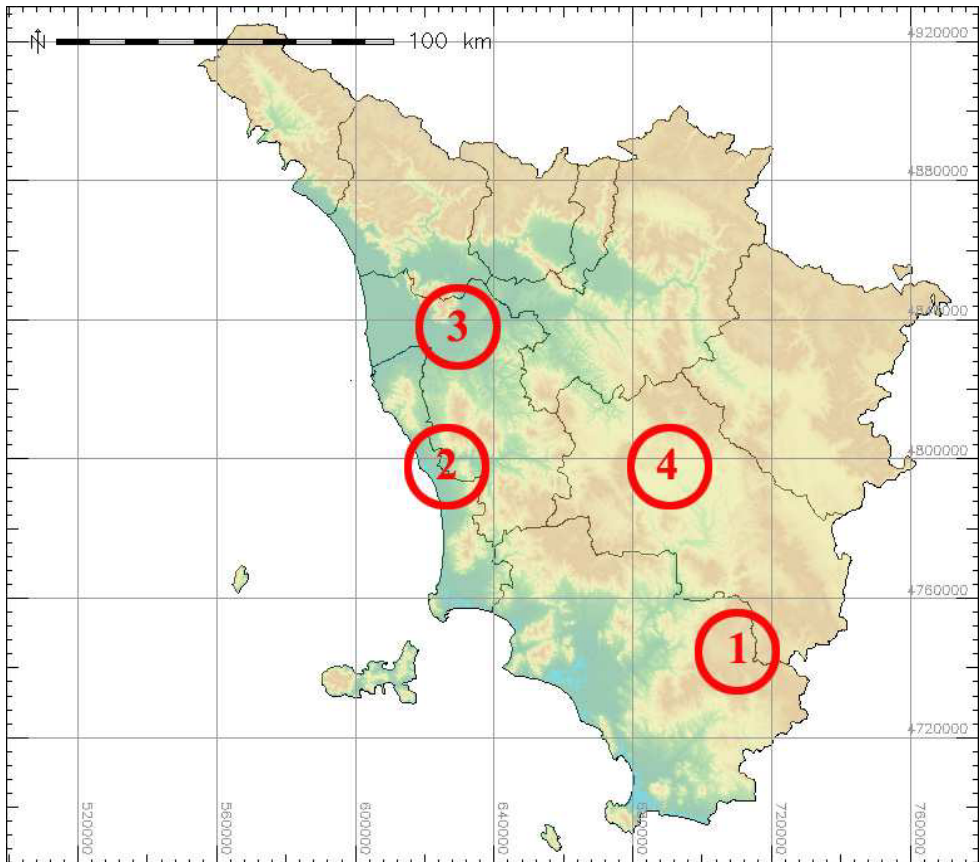


Figure 1 - Map of Tuscany showing the sampling zones. 1 = M. Amiata, 2 = Cecina, 3 = Cascina and Bientina, 4 = Siena.

RESULTS AND DISCUSSION

One hundred and twelve foxes and four badgers were examined. Fox distribution according to age and altitude of sampling area is shown in Table 1. All the animals collected above 500 m a.s.l. came from Monte Amiata, with the exception of two foxes (older than 2 yr) which came from the hills around Cecina.

No animal proved positive for the presence of larvae of *Trichinella*. The 95% confidence interval for the prevalence of *Trichinella* infection in the fox is 0.0% - 2.1%. This result agrees with that reported by Magi and Sasseti (1990), who, in the period 1988-1990, found no positive fox out of 350 analysed animals.

In the analysis of the results it should be considered that in the present research only 28 foxes (25% of the total) were adults coming from areas above 500 m a.s.l.. Moreover, in the hunting season, fox control is carried out, which, shortening the life span of foxes, could contribute to a decrease in the prevalence of *Trichinella* infection (Guberti *et al.*, 2004).

The results of our study together with those of Magi and Sasseti (1990) and Marconcini *et al.* (1970), carried out over a time span of nearly 20 years, em-

phasize that Tuscany can be considered a low-risk area for *Trichinella* infection in the fox. Unfortunately the presence of the parasite cannot be ruled out in this region. Accordingly, in 1993 two cases of infection were reported, of which one caused also infection in humans (Pagni *et al.*, 1993). Therefore diagnostic-epidemiological surveys on foxes older than 3 years are needed, in order to determine the prevalence of the infection in wildlife.

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Table 1 - Cross-classification of 112 red foxes according to their age and the altitude of their recovery zones.

	age \geq 2 yr	age < 2 yr	total
altitude \geq 500m	28	30	58
altitude < 500m	48	26	74
total	76	56	112

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