UNIVERSITÀ DEGLI STUDI DI BOLOGNA
SCUOLA DI AGRARIA E MEDICINA VETERINARIA

DIPARTIMENTO DI SCIENZE MEDICHE VETERINARIE

DOTTORATO DI RICERCA IN SCIENZE VETERINARIE (XXVIII CICLO)

Coordinatore: Prof. Arcangelo Gentile

Settore scientifico-disciplinare: VET/06

Settore concorsuale: 07/H3

HELMINTH PARASITES OF WILDLIFE IN EMILIA-ROMAGNA REGION

Relatore:

Prof. GIOVANNI POGLAYEN

Dottorando:

Dott. ALFREDO FIOCCHI

ESAME FINALE ANNO 2017

INDEX

PREMESSA

Il percorso di Dottorato svolto presso il Servizio di Malattie Trasmissibili e Sanità Pubblica Veterinaria (MTSPV) si è snodato approfondendo prevalentemente argomenti di parassitologia inerenti la fauna selvatica. Cinque dei relativi capitoli corrispondono ad altrettante indagini pubblicate, in corso di pubblicazione o di futura proposizione, e riguardano la volpe ed uno gli uccelli rapaci. I parassiti intestinali del carnivoro sono stati indagati con due successive ricerche, una nelle province di Modena e Bologna ed un'altra in quelle di Forlì e Rimini. In entrambi i casi si è potuta apprezzare una variegata fauna parassitaria che, sotto il profilo eco-patologico, appare in equilibrio con l'ospite. Degno di nota il fatto che la situazione non appaia molto differente da quella di lustri or sono e che, al suo interno vi siano rappresentati parassiti comuni al cane nonché alcuni agenti di zoonosi. Fra questi ultimi, abbiamo però potuto confermare, ancora una volta, l'assenza del temuto cestode *Echinococcus*. Se i parassiti dell'apparato digerente della volpe sono stati ampiamente studiati nel nostro Paese, non altrettanta attenzione hanno ricevuto quelli dell'apparato respiratorio, che presentano al loro attivo solo un numero esiguo di indagini. Le nostre ricerche hanno individuato una fauna elmintica non molto diversificata ma in sintonia con l'ambiente che supporta la presenza degli ospiti intermedi. In questo caso, trattandosi di elementi di ridotte dimensioni, quando non di larve, alla classica classificazione morfologica ha fatto seguito la conferma molecolare. Sempre nell'ambito della volpe, il riscontro di un parassita intestinale particolarmente raro come *Alaria alata*, sospettato come agente di zoonosi, ha meritato un'accurata descrizione morfologica cui è seguita una comunicazione al congresso della Società Italiana di Parassitologia e soprattutto un proficuo contatto con i colleghi dell'Università di Zagabria, esperti di questo trematode per una futura pubblicazione in comune.

Un altro aspetto originale è stato quello relativo ai parassiti dei rapaci, argomento che per la sua complessità ha avuto ben pochi altri seguaci in Italia. Molti sono stati i problemi incontrati nel corso del lavoro, innanzitutto di tipo burocratico (si tratta di specie particolarmente protette le cui carcasse erano conservate presso un CRAS). Si tratta inoltre di specie molto diverse sia sotto il profilo zoologico (diurni /notturni) ma anche come comportamento alimentare. Vi si aggiunga inoltre che le chiavi di identificazione degli elminti di questi uccelli sono di difficile reperibilità ed interpretazione ed il quadro sarà completo. Se oltre l'80% dei soggetti è risultato parassitato possiamo solo affermare che si tratta, per usare una terminologia oggi in voga, di un argomento particolarmente negletto.

Con questo termine sarebbe giusto appellare anche un'altra categoria di parassiti più vicina al medico veterinario tradizionale, quelli del bovino. In particolare gli elminti dell'apparato digerente da decenni non sono oggetto di interesse nel nostro Paese nonostante le nostre eccellenze allevatoriali e produttive. Non ci siamo fatti sfuggire l'opportunità di sondare l'argomento sfruttando la disponibilità di un mattatoio in provincia di Bologna. Questo ha dimostrato, ancora una volta, di rappresentare un osservatorio epidemiologico privilegiato anche per le malattie parassitarie, in particolare oggi che l'anagrafe bovina funziona a pieno regime e permette di risalire all'intera storia dell'animale, della zona e dell'allevamento di provenienza. Poche, ma ancora rappresentate le principali specie di elminti gastrici.

Abbiamo cercato di editare questa tesi, come ormai in uso in ambito internazionale, suddividendola in capitoli corrispondenti ad altrettanti contributi scientifici pubblicati, in corso di pubblicazione o di futura proposizione.

GENERAL OVERVIEW OF THE THESIS

The **first chapter** of this thesis is a survey on the helminthic fauna of 60 wild canids, 57 red foxes (*Vulpes vulpes*) and three wolves (*Canis lupus italicus*), collected in Modena and Bologna provinces, Emilia-Romagna region, Italy. The study focused mainly on the gastrointestinal and hepatic helminths. In total, 15 helminth species were identified, two trematodes, seven cestodes and six nematodes. Our study allowed us to evidence that foxes are reservoir hosts of potential zoonotic parasites, such as *Taenia crassiceps*, *Uncinaria stenocephala* and *Toxocara canis*. Moreover we demonstrated the presence of *Alaria alata* in a few animals, a very rare digenean trematode in the Italian paeninsula. In the **second chapter** we present a research on *Echinococcus granulosus* in 313 red foxes from Eastern Emilia-Romagna. Despite not succeeding in isolating the parasite from the intestines analysed, we identified 16 helminth species, among which the trematodes *Alaria alata* and *Phagicola* sp., the cestode *Joyeuxiella* sp. and the nematode *Molineus legerae*, all representing rare helminth species in the Emilia-Romagna region. The **third chapter** consists of a deeper analysis of the discovery of the trematode *Alaria alata* in our region. In the **fourth chapter** we present a research on the respiratory helminths of 90 red foxes from Eastern Emilia-Romagna. Three nematode species were described, namely *Eucoleus aerophilus, Filaroides hirthi* and *Crenosoma vulpis*. This study represents one of the few descriptions of *F. hirthi* in European red foxes. The **fifth chapter** is a parasitological survey on the helminthic fauna of 67 diurnal and nocturnal birds of prey, belonging to eight species. The helminth community was richer in falconiform and accipitriform than in strigiform birds. In particular *Accipiter nisus* and *Buteo buteo* showed the highest number of parasite species. This survey allowed us to recover parasitological findings which are consistent with recent surveys on raptors' helminthic fauna in other European countries and it could represent a further contribute to better understand the health and biology of birds of prey. The **sixth chapter** is a study on gastro-intestinal nematode parasitic infection in adult cattle (dairy and brood cows) bred in Italy. The survey was performed collecting 427 fecal samples from a bovine slaughterhouse in the province of Bologna. One hundred abomasa were also examined to assess the presence of worm burdens. Gastro-intestinal nematode eggs were detected in 31% of individual fecal samples examined. The fecal output of nematode eggs was significantly related with the livestock category and the stocking density.

1. HELMINTH PARASITES OF THE RED FOX (*VULPES VULPES* L., 1758) AND THE WOLF (*CANIS LUPUS ITALICUS* ALTOBELLO, 1921) IN EMILIA-ROMAGNA, ITALY.

ALFREDO FIOCCHI[1], ANDREA GUSTINELLI[1], LUCA GELMINI[2], GIANLUCA RUGNA[2], MARIA RENZI[3], MARIA CRISTINA FONTANA[3], GIOVANNI POGLAYEN[1]

[1]*Department of Veterinary Medical Sciences, Alma Mater Studiorum University of Bologna, Ozzano Emilia (BO), Italy*

[2]*Lombardy and Emilia Romagna Experimental Zooprophylactic Institute, Modena, Italy*

[3]*Lombardy and Emilia Romagna Experimental Zooprophylactic Institute, Bologna, Italy*

ABSTRACT

In the period 2013-2014 a survey was carried out on the helminthic fauna of 60 wild canids, 57 red foxes (*Vulpes vulpes*) and three wolves (*Canis lupus italicus*), collected in the Emilia-Romagna region, Italy. The study focused mainly on the gastrointestinal and hepatic helminths. Parasites were recovered in 91.2% of the red foxes and in all the wolves examined. Multiple infections were found in the majority of the animals (71.9% of the foxes and 100% of the wolves). In total, 14 intestinal helminth species were identified, two trematodes (*Alaria alata*, *Brachylaima* spp.), seven cestodes (*Mesocestoides* spp., *Taenia crassiceps*, *Taenia pisiformis*, *Taenia polyacantha*, *Dipylidium caninum*, *Taenia ovis*, *Taenia hydatigena*) and five nematodes (*Uncinaria stenocephala*, *Toxocara canis*, *Trichuris vulpis*, *Pterigodermatites affinis*, *Ancylostoma caninum*). The heartworm *Dirofilaria immitis* was also recovered in two foxes. No *Echinococcus* spp. were found. Our study shows that foxes are reservoir hosts of zoonotic parasites, including *A. alata*, a rare digenean trematode in the Italian paeninsula. Results are compared with those of other surveys on helminths of wild canids carried out in Italy and other European countries.

Keywords: Red fox, wolf, helminths, Emilia-Romagna, Italy.

INTRODUCTION

The red fox (*Vulpes vulpes* L., 1758) belongs to the order Carnivora, family Canidae. It is a common species in Italy and also in the Emilia-Romagna region, occupying a wide range of habitats from urbanised lowlands to mountainous territories (Spagnesi & De Marinis 2002). It is an opportunistic predator, whose varied diet includes lagomorphs, small- and medium-sized rodents, domestic and wild birds, reptiles, invertebrates, and also fruit and vegetables especially in summer and autumn. If available, they feed on rubbish, carrion of domestic and wild ungulates, and acquatic animals such as fish and amphibians. Prey availability and geographical setting may influence the parasitic fauna of these animals. Moreover, red foxes are potential reservoir of zoonotic helminth species as several authors have demonstrated in Europe (Loos-Frank & Zyhle 1982; Richards et al. 1995; Papadopoulos et al. 1997; Segovia et al. 2004; Vervaeke et al. 2005; Letková et al. 2006; Saeed et al. 2006; Sikó Barabási et al. 2010; Bružinskaité-Schmidhalter et al. 2012), including Italy (Guberti & Poglayen 1991; Capelli et al. 2003; Di Cerbo et al. 2008; Macchioni et al. 2012). The aim of this study was to analyse the gastro-intestinal and hepatic helminthic fauna of red foxes from Emilia-Romagna, northern Italy. Three wolves (*Canis lupus italicus*), found dead in Modena Apennines, were also included in the survey in order to highlight any differences from foxes' helminthic fauna.

MATERIALS AND METHODS

The study areas were the provinces of Modena and Bologna, in Emilia Romagna, northern Italy. The northern part of the two provinces, which is the more populated and industrialised, is characterised by lowlands (Padan Plain) whereas the southern part is rich in hills and mountains reaching the top altitude of the whole northern Apennines (Mount Cimone, 2165 m). These territories have a subcontinental temperate climate which turns into fresh temperate in the highest valleys of the Apennines. Average annual precipitation ranges between 600 and 800 mm in the lowlands, but tends to increase with altitude, reaching as much as 2000 mm in some mountainous areas.

Between January 2013 and December 2014 we analysed a total of 60 wild canids, 57 red foxes (*Vulpes vulpes*) and three wolves (*Canis lupus italicus*), collected in official culling campaigns or found dead in the Emilia-Romagna region. Twenty-eight foxes and all the wolves came from Modena province, and 29 foxes from Bologna province. Thirty-three foxes (58%) came from

lowland territories, and 24 foxes (42%) and the three wolves came from hilly or mountainous territories. The stomach and the intestine were removed during necropsy and parasites were collected using the sedimentation and counting technique (SCT). The organs were placed on a plastic sheet and opened up along their entire length. Visible helminths were removed immediately from the mucosa and placed in Petri dishes with water. The mucosa was deeply scraped with a sharp spoon and the collected material was allowed to precipitate in tap water inside conical jars for nearly 30 min. Repeated washings and decantations were performed until the sediment in the jars was sufficiently clear to be examined by the stereomicroscope. Livers and gallbladders were also minutely checked for helminths. Several incisions were done in all the hepatic lobes in order to assess the presence of flukes. The bile was also inspected for parasites using both the stereomicroscope and the optical microscope. Only the hearts of foxes from Modena province (n=28) were opened and inspected for the presence of *Dirofilaria immitis* adults. Recovered helminths were counted, stored in 70% alcohol, clarified in lactophenol and then identified on the basis of taxonomic literature and previous descriptions (Pétavy & Deblock 1980; Euzeby 1982; Khalil et al. 1994). Prevalence (P; infected animals/examined hosts), mean intensity (MI; total number of parasites/number of infected hosts) and mean abundance (MA; total number of parasites/number of examined hosts), with their confidence intervals (95%), and an aggregation index (variance-to-mean ratio) have been calculated according to Bush et al. (1997) and Poulin (2006) using the software QP web (Reiczigel et al. 2013)**.** The importance value (I), which allows us to classify helminth parasites in dominant, codominant and subordinate species, has been calculated according to Thul et al. (1985). Comparison of prevalence values of different helminth groups and species has been evaluated with Chi-square test using the software EpiInfo 3.5.1.

RESULTS AND DISCUSSION

The helminthological findings in foxes and wolves are presented in Tables I and II with data regarding prevalence (P), mean intensity (MI), mean abundance (MA) of infection, aggregation index and Thul index. Only five foxes out of 57 (8.7%) were negative for gastro-intestinal helminths. All the wolves examined proved positive for gastro-intestinal helminths. Overall, 15 helminth species were identified: two trematodes (*Alaria alata*, *Brachylaima* spp.), seven cestodes (*Mesocestoides* spp., *Taenia crassiceps*, *Taenia pisiformis*, *Taenia polyacantha*, *Dipylidium caninum*, *Taenia ovis*, *Taenia hydatigena*) and six nematodes (*Uncinaria*

stenocephala, *Toxocara canis*, *Trichuris vulpis*, *Pterigodermatites affinis*, *Ancylostoma caninum* and *Dirofilaria immitis*). The most prevalent parasites in foxes were *Mesocestoides* spp. (17.5%), *Taenia crassiceps* (17.5%), *Uncinaria stenocephala* (75.4%) and *Toxocara canis* (52.6%), whereas *Taenia ovis* (100%), *Taenia hydatigena* (66.6%) and *Uncinaria stenocephala* (66.6%) had higher prevalences in the wolves examined. Differences between prevalence values of helminth groups and species in foxes, which were found to be statistically significant, are presented in Table III. Mixed infections were more common than single ones both in foxes (71.9%) and in wolves (100%). Intestinal parasitism involving only one species was found in 19.2% of the cases (11 foxes), two species in 29.8% (17 foxes), three species in 22.8% (13 foxes), four species in 19.2% (11 foxes). One wolf had two, the others three and four, parasite species, respectively. Most parasites in foxes presented an aggregated distribution (variance-to-mean ratio > 1; Table I). Some cestodes could not be identified to the species level either because they lacked scolices or because these were too degraded to allow a precise identification. No gastric helminths such as spirurid nematodes or *Aonchotheca* (*Capillaria*) *putorii* were recovered. Only one fox (1.75%) had hepatic parasites, which were identified as *Mesocestoides* tetrathyridia.

Alaria alata (Goeze, 1782) is a common intestinal trematode of red foxes in European countries (Möhl et al. 2009). Conversely, the presence of *A. alata* in Italy has been rarely reported. Gestaldi (1854) first described the larval stage of the trematode in frogs ("*Distoma tetracystis*"). A few years later, Molin (1861) found the parasite in a fox near Padua (Veneto region). More recently, Ferroglio et al. (2012) described the infection in a dog from Aosta Valley. We did not find any other reports of this trematode in several parasitological surveys on Italian foxes (Soldati et al. 1976; Rossi et al. 1983; Poglayen et al. 1985; Leoni et al. 1986; Iori et al. 1990; Guberti & Poglayen 1991; Capelli et al. 2003; Manfredi et al. 2003; Di Cerbo et al. 2008; Magi et al. 2009, 2016). In the present study, the infection was observed in only three foxes (prevalence 5.3%; Table I), all living in lowlands rich in humid areas and channels, a suitable environment for the life cycle development of the parasite. A similar prevalence was found in Zagreb county, Croatia (Rajković-Janje et al. 2002: 4.7% of 85 foxes) (Table IV). Regarding mean intensity (3.3), Schöffel et al. (1991) and Murphy et al. (2012) also noted that the majority of infected foxes had a small parasitic burden. Several authors (Loos-Frank & Zeyhle 1982; Criado-Fornelio et al. 2000; Eira et al. 2006; Murphy et al. 2012) confirm that the prevalence of

A. alata is higher in territories rich in wet habitats. The complex life cycle of *A. alata* requires a freshwater snail as the first intermediate host and an amphibian as the second one. Reptiles, rodents, wild boars and other vertebrates can act as paratenic hosts after feeding on infected amphibians (Möhl et al. 2009). Definitive hosts, usually members of the family Canidae, become infected after ingesting mesocercariae in amphibians or paratenic hosts. *A. alata* is regarded as a potential zoonotic agent, especially considering the contamination of wild boar meat with viable mesocercariae (Möhl et al. 2009; Széll et al. 2013). Interestingly, we found another digenean trematode, ***Brachylaima*** spp., in three foxes (Table I) living in the same territories as those infected by *A. alata*. Different species of *Brachylaima* were reported in foxes in Spain (Segovia et al. 2004), Denmark (Al-Sabi et al. 2013: *B. tokudai*), the United Kingdom (UK; Richards et al. 1995: *B. recurva*) and Australia (Dybing et al. 2013: *B. cribbi*) (Table IV).

Mesocestoides spp. is probably the most common tapeworm of red foxes in Europe (Loos-Frank & Zeyhle 1982; Sikó Barabási et al. 2010; Al-Sabi et al. 2014). Prevalences of infection higher than 70% have been reported from Spain (Gortázar et al. 1998), Germany (Schöffel et al. 1991), Denmark (Al-Sabi et al. 2014), Lithuania (Bružinskaité-Schmidhalter et al. 2012), Hungary (Széll et al. 2004), Ukraine (Zvegintsova et al. 2007), Greece (Papadopoulos et al. 1997) and Iran (Dalimi et al. 2006) (Table IV). Its life cycle involves a coprophagous arthropod (a mite) as first intermediate host and different kinds of vertebrates, including amphibians, reptiles, birds and mammals (e.g. rodents) as second intermediate hosts (Loos-Frank & Zeyhle 1982; Papadopoulos et al. 1997). Therefore, foxes and other carnivora can get infected after the ingestion of a wide range of prey. Tetrathyridia commonly develop inside the coelomatic cavity of vertebrate second intermediate hosts, but can also live in the abdominal cavity of their definitive hosts (Urquhart et al. 1996; Lahmar et al. 2014). A few infections have been described in humans as a result of the ingestion of raw viscera of birds (e.g. chickens), reptiles or small mammals (Eom et al. 1992; Fuentes et al. 2003). In the present research, we found *Mesocestoides* spp. in the small intestine of 10 red foxes (prevalence 17.5%; Table I). The mean intensity recovered was the highest among tapeworms (91.7; Table I), as already reported by other authors (e.g. Segovia et al. 2004; Saeed et al. 2006; Magi et al. 2009, 2016). Curiously another fox had several tetrathyridia encysted in the hepatic parenchyma, mainly under the gallbladder; no mature tapeworms were recovered in the small intestine. ***Taenia crassiceps*** (Zeder, 1800) has been found in red foxes from several European countries (Loos-Frank & Zeyhle 1982; Sikó Barabási et al. 2010; Al-Sabi et al. 2014). Li et al. (2013) described the

parasite also in foxes from Qinghai, China. In Italy it was reported mainly from northern regions (Rossi et al. 1983; Iori et al. 1990; Guberti & Poglayen 1991; Capelli et al. 2003; Di Cerbo et al. 2008) (Table IV). The larval stage of *T. crassiceps* (*Cysticercus longicollis*) usually lodges in the subcutis of different species of rodents, mainly microtines such as *Microtus arvalis* (Pétavy & Deblock 1980; Loos-Frank & Zeyhle 1982; Sikó Barabási et al. 2010). Humans can get infected with the parasite after the accidental ingestion of viable ova shed in canid faeces. Cysticerci are usually located in the subcutis, muscles, eyes and, rarely, the central nervous system (Lescano & Zunt 2013; Ntoukas et al. 2013). Underlying immunosuppression can play a role in facilitating the infection, at least in cases involving the skin and muscles (Ntoukas et al. 2013). In the present study we identified *T. crassiceps* in 10 out of 57 foxes (prevalence 17.5%; Table I), all of which were living in lowlands. A similar prevalence of infection in foxes was found in Auvergne, France (Pétavy & Deblock 1980: 17.3% of 69 foxes), and, despite a much higher number of samples, in Germany (Schöffel et al. 1991: 15% of 100 foxes; Wessbecher et al. 1994a: 19.9% of 801 foxes; Pfeiffer et al. 1997a: 17.7% of 1300 foxes) and Austria (Suchentrunk & Sattmann 1994: 14.6% of 307 foxes; Lassnig et al. 1998: 14.6% of 500 foxes) (Table IV). Lower prevalences were reported in previous studies on foxes from Emilia-Romagna (Guberti & Poglayen 1991: 2% of 153 foxes; Capelli et al. 2003: 9% of 109 foxes) (Table IV).

Three foxes were found positive for ***Taenia pisiformis*** (Bloch, 1780) (prevalence 5.3%; Table I). This value is comparable with those evidenced in Portugal (Eira et al. 2006: 3.2% of 62 foxes), Spain (Gortázar et al. 1998: 4.9% of 81 foxes; Martínez-Carrasco et al. 2007: 7.3% of 55 foxes), France (Pétavy & Deblock 1980: 2.9% of 69 foxes), Germany (Schöffel et al. 1991: 7% of 100 foxes) and Hungary (Széll et al. 2004: 4% of 100 foxes) (Table IV). The mean intensity we found (2.6) is similar to the values reported by Eira et al. (2006: mean intensity 4) and Gortázar et al. (1998: mean intensity 3.3). Lagomorphs (rabbits, hares) host the larval forms of the tapeworm (*Cysticercus pisiformis*) mainly in the liver and the peritoneal cavity. As noted by Loos-Frank and Zeyhle (1982), the remarkably different levels of prevalence among European surveys depend on the availability of lagomorphs in the fox diet. High frequencies of infection were recorded, for example, in several studies made in the UK (Hackett & Walters 1980: 13.9% of 280 foxes; Richards et al. 1995: 13.7% of 843 foxes) and in Piedmont, Italy (Rossi et al. 1983: 45.4% of 33 foxes) (Table IV). A high level of infection in lagomorphs might also be explained by a corresponding elevated prevalence of *T. pisiformis* in domestic dogs (especially those used in hunting activities) which, according to some authors (Beveridge & Rickard 1975; Suchentrunk

& Sattmann 1994; Eira et al. 2006), are more appropriate hosts for this taenid than foxes are. ***Taenia polyacantha*** (Leuckart, 1856) was reported mainly from mountainous territories in Italy (Iori et al. 1990; Manfredi et al. 2003; Capelli et al. 2003). In the present study we found the parasite in two out of 57 foxes (prevalence 3.5%; Table I), both of them living in mountainous areas. In the Emilia-Romagna region, the Apennines are regarded a favourable place for *Myodes glareolus* (Bertusi & Tosetti 1986; Spagnesi & De Marinis 2002), which has often been found infected by *T. polyacantha* metacestodes in other European countries (Loos-Frank & Zeyhle 1982; Sikó Barabási et al. 2010). Differently from *T. crassiceps*, the larval stage is located mainly in the pleural or peritoneal cavities of rodents (Pétavy and Deblock 1980; Fujita et al. 1991). The prevalence in foxes from our study is similar to those reported by Gortázar et al. (1998) in Ebro Valley, Spain (3.7% of 81 foxes), Suchentrunk and Sattmann (1994) in Austria (2.1% of 307 foxes), Széll et al. (2004) in Hungary (3% of 100 foxes) and Iori et al. (1990) in the Piedmont (3% of 33 foxes) and Veneto (3.6% of 28 foxes) regions, Italy (Table IV). Higher values were reported by Manfredi et al. (2003: 24.4% of 42 foxes) and Capelli et al. (2003: 20% of 89 foxes) in Trentino Alto-Adige and Veneto, respectively (Table IV). ***Dipylidium caninum*** (Linnaeus, 1758), a flea-transmitted tapeworm, has often shown low levels of prevalence in European countries (Loos-Frank & Zeyhle 1982; Richards et al. 1995; Vervaeke et al. 2005; Al-Sabi et al. 2014). High prevalence values were found in Romania (Sikó Barabási et al. 2010: 14.7% of 561 foxes), Central Italy (Magi et al. 2009: 57.3% of 129 foxes), Tunisia (Lahmar et al. 2014: 55.6% of 9 foxes), Kyrgyzstan (Ziadinov et al. 2010: 33.1% of 151 foxes) and Eastern Australia (Dybing et al. 2013: 27.7% of 147 foxes) (Table IV). This parasite seems to be also common in some Middle-East countries such as Jordan (El-Shehabi et al. 1999), Iraq (Mohammad et al. 2003) and Iran (Dalimi et al. 2006; Meshgi et al. 2009) (Table IV). In our research we found a prevalence of 3.5% (Table I), which is consistent with previous studies in Emilia-Romagna (Guberti & Poglayen 1991: 2.6% of 153 foxes; Capelli et al. 2003: 3% of 109 foxes). ***Taenia ovis*** is an uncommon species among red fox's helminths. Reports exist from Wales (Williams 1976: 1.3% of 149 foxes), Auvergne, France (Pétavy & Deblock 1980: 1.4% of 69 foxes), Germany (Lucius et al. 1988: 0.99% of 101 foxes), Austria (Suchentrunk & Sattmann 1994: 0.4% of 307 foxes) and Romania (Sikó Barabási et al. 2010: 3.7% of 561 foxes) (Table IV). Cysticerci live inside sheep skeletal muscles in the case of *T. ovis ovis* (Cobbold, 1869), and cervid skeletal muscles in the case of *T. ovis krabbei* (Moniez, 1879). However, it is not possible to differentiate between them on a morphological basis (Flueck & Jones 2006). We found the

parasite in only one red fox (prevalence 1.8%; Table I) and in the three wolves necropsied (Table II). They all came from mountainous territories of the province of Modena. Soldati et al. (1976) reported the presence of *T. multiceps* in foxes inhabiting this area of the Apennines. Hence, animals living here might be more prompted to feed on carrions of sheep or wild cervids. *T. ovis* was also described in Italian wolves (n=89) as a subordinate species by Guberti et al. (1993) with a prevalence of 3%. ***Taenia hydatigena*** (Pallas, 1766) is another tapeworm transmitted to canids through the consumption of domestic or wild ungulates viscera. Larvae (*Cysticercus tenuicollis*) are usually found inside the peritoneal cavity of intermediate hosts. Its presence in foxes has been reported by various authors (e.g. Ballek et al. 1992a; Richards et al. 1995; Segovia et al. 2004; Dalimi et al. 2006; Sikó Barabási et al. 2010) (Table IV). Differently from Guberti and Poglayen (1991), we were not able to demostrate *T. hydatigena* in foxes, but found it in two wolves in a mixed infection with *T. ovis* (Table II). In their review on helminthic parasites of wolves, Craig and Craig (2005) consider *T. hydatigena* the second most prevalent parasite (after *U. stenocephala*) in the montane-temperate biome, and the most prevalent one in the boreal biome. ***Echinococcus granulosus*** (Batsch, 1786) is an extremely rare helminth of red foxes in Europe. Low prevalences have been found in southern England (Richards et al. 1995: 0.12% of 843 foxes), Spain (Sànchez et al. 1977; Segovia et al. 2004: 0.25% of 399 foxes) and Sardinia, Italy (Leoni et al. 1986: 1.18% of 85 foxes; Arru et al. 1988: 1.43% of 629 foxes). According to some authors (Arru et al. 1988; Guberti & Poglayen 1991; Papadopoulos et al. 1997; Segovia et al. 2004) this canid does not play an important role in the maintainance of the life cycle of *E. granulosus*. Arru et al. (1988) in particular stated that no gravid proglottids were found in any *Echinococcus* specimen isolated in foxes from Sardinia, also in experimental studies. According to Garippa (2006), cystic echinococcosis (CE) has a sporadic diffusion in production animals in Emilia-Romagna. However, Battelli et al. (2004) evidenced a CE risk area for humans in the mountains (northern Apennines) between Reggio Emilia and Modena. In our survey, *E. granulosus* was not found in the 60 canids examined. Guberti et al. (2004) reported a prevalence of 15% for this parasite in 119 wolves from the Italian Apennines and suggested that echinococcosis in this species is still related to the classic dog-sheep cycle, mainly diffused in Southern and Central Italy (Guberti et al. 1993, 2004). More recently, Gori et al. (2015) recorded a lower occurence of *E. granulosus* (5.6%), analysing by polymerase chain reaction (PCR) and sequencing faecal samples of wolves from Liguria region.

Uncinaria stenocephala (Railliet, 1884) is one of the most prevalent intestinal nematodes of red foxes in Europe (Loos-Frank & Zeyhle 1982; Richards et al. 1995; Vervaeke et al. 2005; Al-Sabi et al. 2014) (Table V). They are mainly infected by ingestion of third-stage larvae, but active cutaneous penetration is also possible (Gibbs 1961; Urquhart et al. 1996). According to several authors (Suchentrunk & Sattmann 1994; Richards et al. 1995; Gortázar et al. 1998; Criado-Fornelio et al. 2000; Eira et al. 2006; Martínez-Carrasco et al. 2007; Dybing et al. 2013), moist soil conditions and warm temperatures are important factors determining the viability of free-living larvae and hence the diffusion of the parasite in wild Carnivora. Both *U. stenocephala* and *A. caninum* free-living third-stage larvae can cause cutaneous *larva migrans* in people (Feldmeier & Schuster 2012). In high-income countries, the disease occurs sporadically and is usually associated with untypical climatic conditions, such as prolonged periods of rainfall and warm weather (Feldmeier & Scuster 2012). We found a prevalence of 75.4% and a mean intensity of 14.9 in foxes (Table I). Similar values were reported by Eira et al. (2006) in Portugal (P = 77.4%, MI = 16.2 of 62 foxes), Pétavy and Deblock (1980) in France (P = 68.1%, MI = 13.4 of 69 foxes) and Capelli et al. (2003) in Veneto region, Italy (P = 74%, MI = 14 of 89 foxes). The high prevalence of infection might be due to the high humidity levels of the climate, which fit the humidity requirements of preparasitic stages of *U. stenocephala*. Two of the wolves examined in our study (66%) were infected with *U. stenocephala* (Table II). According to the review by Craig and Craig (2005), this nematode is the most prevalent helminth in wolves living in the temperate/montane biome. Guberti et al. (1993) regarded it as a dominant species in Italian wolf populations. Only one fox was found infected with ***Ancylostoma caninum*** (Ercolani, 1859) (P = 1.8%) (Table I). This is in agreement with several other European studies, where a low level of prevalence for this parasite was observed (Pétavy & Deblock 1980: 1.44% of 69 foxes; Loos-Frank & Zeyhle 1982: 0.03% of 3573 foxes; Carvalho-Varela & Marcos 1993: 2% of 306 foxes; Wessbecher et al. 1994b: 1.1% of 801 foxes; Capelli et al. 2003: 2% of 109 foxes; Segovia et al. 2004: 0.5% of 399 foxes; Széll et al. 2004: 1% of 100 foxes; Saeed et al. 2006: 0.6% of 1040 foxes) (Table V). Regarding other strongylid nematodes, we recovered unidentified trichostrongylids in two foxes from Bologna province (Table I). ***Toxocara canis*** (Werner, 1782) is another common nematode of red foxes in Europe (Loos-Frank & Zeyhle 1982; Richards et al. 1995; Vervaeke et al. 2005; Al-Sabi et al. 2014) (Table V). Canids usually become infected by ingesting embryonated eggs or paratenic hosts (e.g. rodents) containing second-stage larvae. A transplacental and transmammary infection route is also possible (Urquhart et al. 1996). The

presence of red foxes in urban and suburban areas could increase the risk of *Toxocara* infection in humans (Richards et al. 1993; Reperant et al. 2007). In our survey we found a prevalence of 52.6% in foxes with a mean intensity of 6 (Table I). Even though a lower number of samples were analysed, our results are similar to those reported by Deblock et al. (1988) in France (P = 51.3% of 154 foxes), Richards et al. (1995) in the UK (P = 55.8%, MI = 7.1 of 843 foxes), Saeed et al. (2006) in Denmark (P = 59.4%, MI = 6.9 of 1040 foxes) and other authors in Italy (e.g. Guberti & Poglayen 1991: P = 46.4%, MI = 5.7 of 153 foxes; Manfredi et al. 2003: P = 56.1% of 42 foxes). Differently from Soldati et al. (1976), Magi et al. (2009, 2016) and Capelli et al. (2003), we couldn't find any *Toxascaris leonina*. This actually agrees with most studies on helminths of foxes, in which a higher prevalence of *T. canis* was usually reported (Loos-Frank & Zeyhle 1982; Richards et al. 1995; Vervaeke et al. 2005; Al-Sabi et al. 2014). However *T. leonina* was observed at a higher prevalence than *T. canis* in Portugal (Carvalho-Varela and Marcos 1993), Spain (Gortázar et al. 1998; Criado-Fornelio et al. 2000), France (Pétavy and Deblock 1980), Hungary (Széll et al. 2004), Ukraine (Zvegintsova et al. 2007), Turkey (Gicik et al. 2009), Iran (Dalimi et al. 2006) and China (Li et al. 2013) (Table V). Only one wolf was infected with *T. canis* and at an extremely low intensity (i.e. 1; Table II). Guberti et al. (1993) reported a prevalence of 17% in their survey on helminths of Italian wolves (n = 89) and considered *T. canis* a codominant species. We recovered ***Trichuris vulpis*** (Frölich, 1789) in the caecum of seven foxes at the same prevalence (12.3%; Table I) as indicated by Manfredi et al. (2003), but higher than that reported in other surveys from our region (Soldati et al. 1976: 8.6% of 23 foxes; Guberti & Poglayen 1991: 3.3% of 153 foxes; Capelli et al. 2003: 4% of 109 foxes) (Table V). ***Pterigodermatites affinis*** (syn. *Rictularia affinis*) (Jägerskiöld, 1904) was found in three foxes (prevalence 5.3%; Table I) and in one wolf (Table II). Similar prevalences in foxes were reported in Portugal (Eira et al. 2006: 3.2% of 62 foxes) and, despite a higher number of animals, in Haute-Savoie and Cantal, France (Pétavy et al. 1990: 5% of 150 foxes; Deblock et al. 1988: 4% of 154 foxes), Liguria, Italy (Magi et al. 2016: 5.6% of 180 foxes) and Slovenia (Vergles Rataj et al. 2013: 4.2% of 428 foxes) (Table V). In Italy we have reports from several northern regions (Iori et al. 1990; Manfredi et al. 2003; Capelli et al. 2003; Di Cerbo et al. 2008; Magi et al. 2016) and Sardinia (Leoni et al. 1986: *Rictularia* sp.), but not from Emilia-Romagna (Table V). The presence of *P. affinis* in Carnivora depends on the frequency of ingestion of suitable intermediate hosts (insects) and paratenic hosts, including reptiles (Deblock et al. 1988; Papadopoulos et al. 1997; Martínez-Carrasco et al. 2007). Out of 60 gastro-intestinal tracts

examined, we were able to identify remnants of reptiles (lizards) in the stomach of only one fox. Most of the other stomachs contained parts of small mammals, birds, insects and fruit. According to Craig and Craig (2005), *Rictularia* spp. were found in wolves in Russia (Mech 1970) and Greece (Papadopoulos et al. 1997). Guberti et al. (1993) did not find any *Rictularia* spp. in a large survey on Italian wolves. ***Dirofilaria immitis*** (Leidy, 1856) is the only cardiopulmonary helminth that was checked for in our survey. It has been found also in red foxes from Portugal (Carvalho-Varela & Marcos 1993: P = 11.8% of 306 foxes; Eira et al. 2006: P = 3.2% of 62 foxes), Spain (Gortázar et al. 1998: P = 12.7% of 415 foxes; Segovia et al. 2004: P = 0.2% of 399 foxes), Italy (Leoni et al. 1986: P = 1.1% of 85 foxes; Magi et al. 2009: P = 6.2% of 129 foxes), Iran (Meshgi et al. 2009) (Table V), and in the Iberian wolf (Segovia et al. 2001: P = 2.1% of 47 wolves). Since mosquitoes are biological vectors of the parasite, humid temperate habitats are most suitable for its diffusion (Gortázar et al. 1998; Eira et al. 2006; Martínez-Carrasco et al. 2007). We found a prevalence of 7.1% in foxes from the province of Modena (Table I). They were all shot in lowlands during the spring. This territory, rich in moist habitats, characterised also by high temperatures and humidity in the warm season, has ideal environmental conditions for the trasmission cycle of *D. immitis*.

In conclusion, the present survey shows that red foxes are definitive hosts for several helminth species in Emilia-Romagna, Italy. Only a few trematodes were recovered, namely *A. alata* and *Brachylaima* spp. Our study provides, notably, one of the few recent description of *A. alata* in the Italian paeninsula. Among cestodes, *Mesocestoides* spp. and *T. crassiceps* were the most common parasites, whereas *T. pisiformis*, *T. polyacantha*, *D. caninum* and *T. ovis* had lower prevalences. Regarding nematodes, we report high prevalence values of *U. stenocephala* and *T. canis*, whereas lower prevalences of *T. vulpis*, *P. affinis* and particularly *A. caninum* were found. These differences were statistically significant, as indicated in Table III. We also found a few animals positive for *D. immitis*. Therefore, the role of the red fox as a reservoir host for some potential zoonotic parasites such as *T. crassiceps*, *U. stenocephala* and *T. canis* must be taken into consideration. Most parasites in foxes presented an aggregated distribution (variance-to-mean ratio > 1; Table I). From an ecological perspective these results confirm the stability of the host-parasite relationship in a wild population with a particular and well-structured helminthofauna, also hierarchically. The present parasitological findings also confirm also the fact that a significant part of red foxes' diet is based on small- or medium-sized vertebrates (e.g. rodents, lagomorphs, birds, reptiles and amphibians). Conversely, the extremely low prevalence

of tapeworms transmitted by ungulates, such as *T. ovis*, indicates that they probably represent a less important part of foxes' diet in these territories. Considering wolves, only a few animals were available for analysis. They show a helminthic fauna similar to that of foxes, with the exception of taenids. Indeed, the high prevalence of *T. ovis* and *T. hydatigena* and the absence of other species of tapeworms allow us to speculate that domestic and wild ungulates play an important part in wolves' diet.

Acknowledgements

We are grateful to Mr Bruno Negrini, technician at Lombardy and Emilia Romagna Experimental Zooprophylactic Institute, Bologna, Dr. Mauro Ferri, former DVM at AUSL (Azienda Unità Sanitaria Locale) of Modena, Prof. Maria Letizia Fioravanti, Prof. Fabio Ostanello and Dr. Benedetto Morandi, DVM, for their kind collaboration in this research.

Table I. Helminth species recovered in 57 foxes from Modena and Bologna provinces, Emilia-Romagna region.

Helminth species	N° positive	Prevalence % (IC 95%)	Mean Intensity (IC 95%)	Intensity range	Mean Abundance (IC 95%)	Aggregation index (s^2/m)	Importance value[a]
SUBCLASS DIGENEA							
Alaria alata	3	5.3 (1.1-14.6)	3.3 (3.0-3.7)	3 - 4	0.2 (0.0-0.4)	3.28	0.06 (C)
***Brachylaima* spp.**	3	5.3 (1.1-14.6)	1.3 (1.0-1.7)	1 - 2	0.1 (0.0-0.2)	1.46	0.02 (C)
CLASS CESTODA							
***Mesocestoides* spp.**	10	17.5 (8.7-29.9)	91.7 (38.8-187)	2 - 347	16.1 (5.7-40.1)	216.82	20.13 (D)
Taenia crassiceps	10	17.5 (8.7-29.9)	28.5 (14.5-49.2)	1 – 85	5 (2.0-11.5)	51.22	6.25 (D)
Taenia pisiformis	3	5.3 (1.1-14.6)	2.7 (2.0-3.0)	2 - 3	0.1 (0.0-0.4)	2.66	0.05 (C)
Taenia polyacantha	2	3.5 (0.4-12.1)	22.5 (1.0-22.5)	1 - 44	0.8 (0.0-4.2)	43.01	0.19 (C)
Dipylidium caninum	2	3.5 (0.4-12.1)	3 (1.0-3.0)	1 - 5	0.1 (0.0-0.4)	4.30	0.02 (C)
Taenia ovis	1	1.8 (0.0-9.4)	1 n.c.	1	0.01 (0.0-0.1)	1.00	0.002 (S)
Unidentified cestodes	7	12.2	–	–	–	–	–
PHYLUM NEMATODA							
Uncinaria stenocephala	43	75.4 (62.2-85.9)	14.9 (10.0-23.4)	1 - 112	11.3 (7.2-18.0)	36.34	60.61 (D)
Toxocara canis	30	52.6 (39.0-66.0)	6 (3.9-10.1)	1 - 30	3.2 (1.9-5.3)	13.10	11.92 (D)
Trichuris vulpis	7	12.3 (5.1-23.7)	3.4 (1.6-8.1)	1 - 13	0.4 (0.1-1.4)	7.71	0.36 (C)
Pterigodermatites affinis	3	5.3 (1.1-14.6)	15.7 (1.0-30.3)	1 – 45	0.8 (0.0-4.0)	43.06	0.30 (C)
Trichostrongylidae [b]	2	3.5 (0.4-12.1)	2.5 (1.0-2.5)	1 - 4	0.1 (0.0-0.4)	3.37	0.02 (C)
Ancylostoma caninum	1	1.8 (0.0-9.4)	1 n.c.	1	0.01 (0.0-0.1)	1.00	0.002 (S)
Dirofilaria immitis [c]	2/28	7.1	–	–	–	–	–

[a] The letter under the importance value (Thul index) refers to the following categories: (D) dominant species; (C) codominant species; (S) subordinate species.
[b] Unidentified *Trichostrongylid* nematodes.
[c] Prevalence data refer only to foxes from Modena.
CI = confidence interval.

Table II. Helminth species recovered in three wolves from Modena and Bologna provinces, Emilia-Romagna region.

Helminth species	N° positive (prevalence)	Mean Intensity	Intensity range	Mean Abundance
CESTODA				
***Taenia* spp.**	3 (100 %)	50	37 (*T.ovis*) 45 (*T. ovis, T. hydatigena*) 68 (*T. ovis, T. hydatigena*)	50
NEMATODA				
Uncinaria stenocephala	2 (66.6 %)	76.5	8 - 145	51
Toxocara canis	1 (33.3%)	1	1	0.3
Pterigodermatites affinis	1 (33.3 %)	1	1	0.3

Table III. Significant differences in prevalence values between helminth groups and species recovered in 57 foxes from Modena and Bologna provinces, Emilia-Romagna region.

Helminth groups/species compared		Chi-square value	p
Trematodes	Cestodes	24.14	< 0.01
Trematodes	Nematodes	68.12	< 0.01
Cestodes	Nematodes	16.24	< 0.01
Mesocestoides /T. crassiceps	*T.pisiformis*	4.25	0.03
Mesocestoides /T. crassiceps	*T. polyacantha*	5.96	0.01
Mesocestoides /T. crassiceps	*D. caninum*	5.96	0.01
Mesocestoides /T. crassiceps	*T. ovis*	8.15	< 0.01
U. stenocephala	*T. canis*	6.44	0.01
U. stenocephala	*T. vulpis*	46.17	< 0.01
U. stenocephala	*P. affinis*	58.31	< 0.01
U. stenocephala	Trichostrongilids	61.72	< 0.01
U. stenocephala	*A. caninum*	65.29	< 0.01
T. canis	*T. vulpis*	21.17	< 0.01
T. canis	*P. affinis*	31.09	< 0.01
T. canis	Trichostrongilids	34.06	< 0.01
T. canis	*A. caninum*	37.26	< 0.01

Table IV- Prevalence of some cestode and trematode infections in red foxes (*Vulpes vulpes*) from European and extra-european countries, according to recent surveys.

Country	N° foxes	*T. crassiceps*	*T.polyacantha*	*T.pisiformis*	*T. ovis*	*T. hydatigena*	*Taenia spp.**	*D. caninum* *Joyeuxiella*	*Mesocestoides spp.*	*A. alata*** *Brachylaima*	Reference
Portugal	306	1.3	2.0	3.3	_	_	_	_ 9.8**	50.0	_	Carvalho-Varela and Marcos 1993
Portugal (Coimbra)	62	_	_	3.23	_	_	_	_	30.65	27.42	Eira et al. 2006
Spain (Galicia)	201	23	_	_	_	_	_	0.5	2.5	_	Alvarez et al. 1995
Spain (Ebro Valley)	81	_	3.7	4.9	_	_	_	1.2 1.2	71.6	_	Gortázar et al. 1998
Iberian Peninsula	399	4.3	10.3	6.3	_	2.0	_	_ 2.8, 6.0	28.8	2.0 1.5	Segovia et al. 2004
Spain (Murcia)	55	_	_	7.3	_	_	_	1.8 34.6	61.8	_	Martínez-Carrasco et al. 2007
France (Auvergne)	69	17.39	27.53	2.89	1.44	_	_	_	27.5	_	Pétavy and Deblock 1980
France (Auvergne)	154	23.4	11.03	1.29	_	_	_	_	26	_	Deblock et al. 1988
France (Haute-Savoie)	150	29	14	_	_	_	_	_	4.6	_	Pétavy et al. 1990
Belgium (Flanders)	219	_	_	_	_	_	2.7	0.9	_	_	Vervaeke et al. 2005
Netherlands	137	P****	_	P	_	_	53.3	_	_	10.9	Borgsteede 1984
Netherlands	136	22.1		_	_	_	_	_	5.9	16.9	Franssen et al., 2014
Germany (South-West)	3573	24	7.7	0.03	_	0.03	_	0.06	19.8	0.08	Loos-Frank and Zeyhle 1982
Germany (Schleswig-H.)	101	21.78	1.98	2.97	0.99 (*T.cervi*)	_	_	_	_	29.7	Lucius et al. 1988
Germany (Berlin)	100	15	7	7	_	_	_	_	72	11	Schöffel et al. 1991
Germany (Hessen,Westfalen)	397	28.5	14.4	2.0	_	0.8	_	_	4.3	_	Ballek et al. 1992a
Germany (Karlsruhe)	801	19.9	7	_	_	_	_	0.5	16.6	_	Wessbecher et al. 1994a
Germany (Sachsen-Anhalt)	1300	17.7	11.9	0.15	_	_	_	0.2	54.1	_	Pfeiffer et al. 1997a
Germany (Schleswig-H.)	470	_	_	_	_	_	26.2	_	1.9	13	Manke and Stoye 1998
Denmark	1040	0.2	_	0.3	_	0.4	21.5	_	35.6	15.4	Saeed et al. 2006

Country	N° foxes	*T. crassiceps*	*T.polyacantha*	*T.pisiformis*	*T. ovis*	*T. hydatigena*	*Taenia spp.**	*D. caninum* *Joyeuxiella*	*Mesocestoides*	*A. alata*** *Brachylaima*	Reference
Denmark	384	_	_	_	_	_	30.7	1	42.7	34.4 1.3	Al-Sabi et al. 2013
Denmark (Copenaghen)	70	_	P	_	_	_	22.9	1.4	78.6	20	Al-Sabi et al. 2014
Denmark (Southern Jutland)	48	P	P	_	_	_	39.6	_	8.3	2.1	Al-Sabi et al. 2014
UK (Wales)	149	_	_	10.7	1.3	2.7	_	_	_	_	Williams 1976
UK (Wales)	280	_	_	13.9	_	6.4	_	0.7	_	_	Hackett and Walters 1980
UK (England)	843	_	_	13.76	_	2.49	_	3.8	_	_ 2.85	Richards et al. 1995
UK	588	_	_	2	_	_	20.7	0.7	_	_	Smith et al. 2003
Ireland	77	_	_	_	_	_	P	_	_	27.3	Wolfe et al. 2001
Poland	380	_	_	_	_	_	39.5	_	63.7	2.1	Ramisz et al. 2004
Belarus	94	27.7	17	12.8	_	5.3	_	4.3	13.8	42.6	Shimalov and Shimalov 2003
Lithuania	269	26.4	61.7	_	_	_	_	_	78.4	94.8	Bružinskaitė-Schmidhalter et al. 2012
Slovakia	302	_	_	_	_	_	20.86	1.99	61.2	_	Letková et al. 2006
Austria	307	14.6	2.1	_	0.4	_	_	_	24.9	_	Suchentrunk and Sattmann 1994
Austria (Steiermark)	500	14.6	6.8	0.2	_	_	_	_	15.6	_	Lassnig et al. 1998
Switzerland (Geneva)	228	_	_	_	_	_	41.9	2.2	5.7	_	Reperant et al. 2007
Italy (Trentino)	29	37.9	27.6	_	_	_	_	_	10.3	_	Iori et al. 1990
Italy (Trentino)	42	_	24.4	_	_	_	_	_	4.9	_	Manfredi et al. 2003
Italy (Veneto)	28	_	3.6	3.6	_	_	_	_	7.1	_	Iori et al. 1990
Italy (Veneto)	89	9	20	_	_	_	_	_	_	_	Capelli et al. 2003
Italy (Lumbardy)	93	3.2	11.8	8.6	_	_	_	_	21.5	_	Iori et al. 1990
Italy (Piedmont)	33	30.3	3	45.4	_	_	_	_	_	_	Iori et al. 1990 Rossi et al. 1983
Italy (Liguria, Piedmont)	180	_	1.1	1.1	_	_	6.1	29.4	81.7	_	Magi et al. 2016

Country	N° foxes	*T. crassiceps*	*T.polyacantha*	*T.pisiformis*	*T. ovis*	*T. hydatigena*	*Taenia spp.**	*D. caninum* *Joyeuxiella*	*Mesocestoides*	*A. alata**** *Brachylaima*	Reference
Italy (Alps)	645	28.99	_	_	_	0.32	_	_	27.44	_	Di Cerbo et al. 2008
Italy (E.-Romagna, MO)	23	_	_	8.6	_	_	_	_	13	_	Soldati et al. 1976
Italy (E.-Romagna, FC)	153	2	_	_	_	3.3	17	2.6	11.1	_	Guberti and Poglayen 1991
Italy (E.-Romagna, FE)	109	9	_	_	_	_	_	3	1	_	Capelli et al. 2003
Italy (Tuscany)	100	_	_	_	_	_	_	4	37	_	Capelli et al. 2003
Italy (Tuscany)	129	_	_	_	_	_	_	57.3	45.4	_	Magi et al. 2009
Italy (Lazio)	182	_	1.6	_	_	_	_	_ 15.9	14.3	_	Iori et al. 1990
Italy (Sardinia)	85	_	_	_	_	_	_	_ 48.2	60	_	Leoni et al. 1986
Slovenia	428	22.2	6.5	2.1	_	_	_	1.4	27.6	_	Vergles Rataj et al. 2013
Croatia	85	_	_	P	_	_	24.7	_	4.7	4.7	Rajković-Janje et al. 2002
Former Jugoslavia	532	51	16	22	_	2	_	_	62	65	Pavlović et al. 1997
Hungary	100	7	3	4	_	_	_	_	73	48	Széll et al. 2004
Romania	561	5	5.3	12	3.7	8.2	_	14.7	28.7	15	Sikó Barabási et al. 2010
Ukraine	25	20	_	_	_	_	_	_	72	_	Zvegintsova et al. 2007
Greece	314	0.3	_	_	_	0.9	_	3.2 24.5	73.2	1.6	Papadopoulos et al. 1997
Tunisia	9	_	_	_	_	_	_	55.6	22, 33, 55.6 (3 species)	_	Lahmar et al. 2014
Turkey (Kars)	20	_	_	10	_	_	_	_	60	30	Gicik et al. 2009
Jordan	9	_	_	_	_	_	_	11.1 44.4	44.4	_	El-Shehabi et al. 1999
Iraq	54	_	_	_	_	_	_	14.8 7.4	_	_	Mohammad et al. 2003
Iran (Western)	22	_	_	_	_	9.09	_	9.1 27.3	81.8	_	Dalimi et al. 2006
Iran (3 Climatic Zones)	37	-	-	-	-	- 5.4 -	-	10.8 8.1 -	43.2 59.5 67.5	-	Meshgi et al. 2009
Iran (Western)	52	_	1.9	_	_	13.4	38.4	3.8	71.9	1	Khanmohammadi et al. 2011

Country	N° foxes	*T. crassiceps*	*T.polyacantha*	*T.pisiformis*	*T. ovis*	*T. hydatigena*	*Taenia spp.**	*D. caninum* *Joyeuxiella*	*Mesocestoides*	*A. alata****	Reference
Kyrgyzstan	151	_	P	_	_	P	31.7	33.1	65.6	_	Ziadinov et al. 2010
China (Qinghai)	27	7.4	_	7.4	_	_	_	_	59	11	Li et al. 2013
Japan	7	_	29	29	_	_	_	_	_	29	Sato et al. 1999
Australia (Western)	147	_	_	_	_	0.7	4.1	27.7	_	0.1	Dybing et al. 2013
Argentina (Patagonia)*****	81	_	_	_	_	4.9	_	_	1.2	_	Zanini et al. 2006

(*) Prevalence values have been inserted when they are the only or main information provided by the authors about *Taenia* species.

(**) Prevalence values in orange refer to *Joyeuxiella* species.

(***) Prevalence values in orange refer to *Brachylaima* species.

(****) P= present.

(*****) Foxes examined belong to the species *Pseudalopex griseus.*

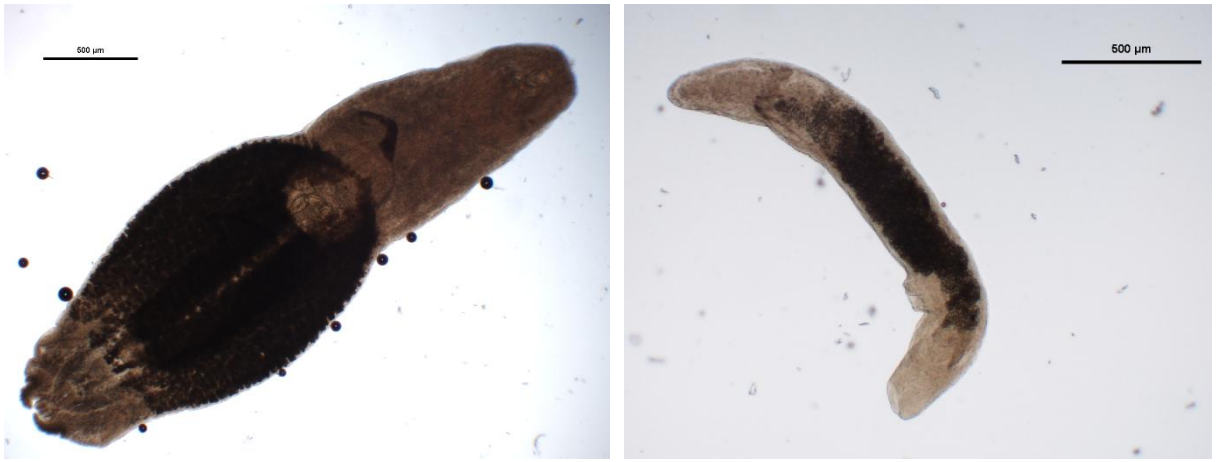
Table V- Prevalence of some nematode infections in red foxes (*Vulpes vulpes*) from European and extra-european countries, according to recent surveys.

Country	N° foxes	*U. stenocephala*	*A. caninum*	*Toxocara canis*	*T. leonina*	*P. affinis*	*T. vulpis*	*Molineus spp.*	*D. immitis*	Reference
Portugal	306	57.2	2.0	11.1	11.4	8.5	2.0	_	11.8	Carvalho-Varela and Marcos 1993
Portugal (Coimbra)	62	77.42	_	37.10	_	3.23	8.06	4.84	3.23	Eira et al. 2006
Spain (Galicia)	201	28	_	23	1	_	_	_	_	Alvarez et al. 1995
Spain (Ebro Valley)	81	30.9	_	6.2	66.7	23.4	12.3	_	12.7	Gortázar et al. 1998
Iberian Peninsula	399	71.2	0.5	29.3	19.3	22.3	8.3	4.3	0.2	Segovia et al. 2004
Spain (Murcia)	55	1.8	_	45.5	5.4	54.5	9.1	_	_	Martínez- Carrasco et al. 2007
France (Auvergne)	69	68.11	1.44	27.53	33.3	_	_	1.44	_	Pétavy and Deblock 1980
France (Auvergne)	154	58.4	_	51.3	25.3	3.9	16.2	_	_	Deblock et al. 1988
France (Haute-Savoie)	150	52	_	44	10	5	8	_	_	Pétavy et al. 1990
Belgium (Flanders)	219	31.5	_	47.9		_	_	_	_	Vervaeke et al. 2005
Netherlands	137	59.9	_	73.7	_	_	_	5.1	_	Borgsteede 1984
Netherlands	136	54.4	_	61	2.2	_	16.9	_	_	Franssen et al. 2014
Germany (South-West)	3573	25.8	0.03	31.3	3.4	_	_	_	_	Loos-Frank and Zeyhle 1982
Germany (Schleswig-H.)	101	57.42	_	74.25	_	_	_	_	_	Lucius et al. 1988
Germany (West Berlin)	100	89	_	69	12	_	1	22	_	Schöffel et al. 1991
Germany (Hessen,Westfalen)	397	3.5	3.8	32.7	11.1	_	_	_	_	Ballek et al. 1992b
Germany (Karlsruhe)	801	24.3	1.1	30.2	2.0	_	_	_	_	Wessbecher et al. 1994b
Germany (Sachsen-Anhalt)	1300	15.9	1.7	26.5	10.5	_	_	_	_	Pfeiffer et al. 1997b
Germany (Schleswig-H.)	470	32.7	_	66	0.9	_	_	_	_	Manke and Stoye 1998
Denmark	1040	68.6	0.6	59.4	0.6	_	0.5	_	_	Saeed et al. 2006

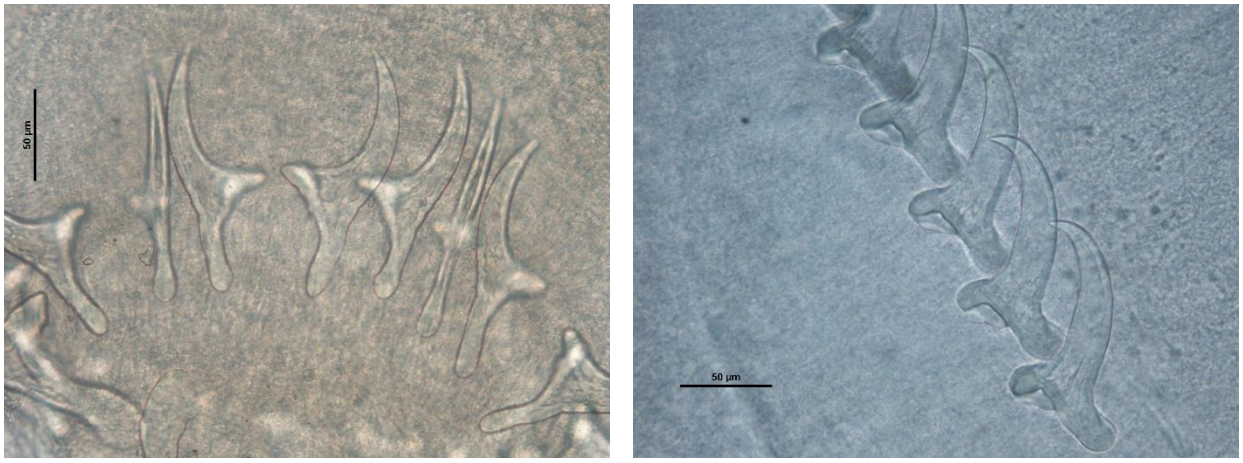
Country	N° foxes	*U. stenocephala*	*A. caninum*	*Toxocara canis*	*T. leonina*	*P. affinis*	*T. vulpis*	*Molineus spp.*	*D. immitis*	Reference
Denmark	384	84.1	_	60.9	_	_	_	_	_	Al-Sabi et al. 2013
Denmark (Copenaghen)	70	84.3	_	48.6	_	_	_	_	_	Al-Sabi et al. 2014
Denmark (South. Jutland)	48	60.4	_	64.6	6.3	_	_	_	_	Al-Sabi et al. 2014
UK (Wales)	149	57	0.7	52	11.5	_	_	_	_	Williams et al. 1976
UK (Wales)	280	87.1	_	63	2.9	_	_	_	_	Hackett and Walters 1980
UK (Southern England)	843	67.97	_	55.87	1.54	_	0.47	_	_	Richards et al. 1995
UK	588	41.3	_	61.6	0.3	_	0.3	_	_	Smith et al. 2003
Ireland	77	92.2	_	37.7	_	_	_	_	_	Wolfe et al. 2001
Poland	380	13.4	6.8	25.8	1.1	_	3.4	_	_	Ramisz et al. 2004
Belarus	94	40.4	3.2	25.5	18.1	_	3.2	3.2	_	Shimalov and Shimalov 2003
Lithuania	269	76.9	_	40.5	_	_	_	_	_	Bružinskaitė-Schmidhalter et al. 2012
Slovakia	302	1.98	5.63	25.82	17.55	_	6.9	_	_	Letková et al. 2006
Austria	307	27.5	_	42.9	0.4	_	_	1.3	_	Suchentrunk and Sattmann 1994
Austria (Steiermark)	500	43	_	46.8	0.6	_	0.2	_	_	Lassnig et al. 1998
Switzerland (Geneva)	228	78.2	_	44.3	37.3	_	8.3	_	_	Reperant et al. 2007
Italy (Trentino)	29	48.3	_	48.3	_	6.9	48.3	_	_	Iori et al. 1990
Italy (Trentino)	42	51.2	_	56.1	_	17.1	12.2	9.8	_	Manfredi et al. 2003
Italy (Veneto)	28	28.6	_	46.4	_	17.8	28.6	_	_	Iori et al. 1990
Italy (Veneto)	89	74	_	66	_	8	9	_	_	Capelli et al. 2003
Italy (Lumbardy)	93	63.4	_	54.8	_	21.5	_	_	_	Iori et al. 1990
Italy (Piedmont)	33	81.8	12.1	45.4	_	_	18.1	_	_	Iori et al. 1990 Rossi et al. 1983
Italy (Liguria, Piedmont)	180	70	_	26.7	25.6	5.6	21.1	27.2	_	Magi et al. 2016

Country	N° foxes	*U. stenocephala*	*A. caninum*	*Toxocara canis*	*T. leonina*	*P. affinis*	*T. vulpis*	*Molineus spp.*	*D. immitis*	Reference
Italy (Alps)	645	51.32	_	54.42	_	19.38	0.16	2.95	_	Di Cerbo et al. 2008
Italy (E.-Romagna, MO)	23	4.34	_	69.5	13	_	8.6	_	_	Soldati et al. 1976
Italy (E.-Romagna, FC)	153	11.8	3.9	46.4	_	_	3.3	_	_	Guberti and Poglayen 1991
Italy (E.-Romagna, FE)	109	11	2	70	_	_	4	_	_	Capelli et al. 2003
Italy (Tuscany)	100	41	_	32	1	_	1	_	_	Capelli et al. 2003
Italy (Tuscany)	129	39.1	_	9.1	5.4	_	_	_	6.2	Magi et al. 2009
Italy (Lazio)	182	43.4	1.1	31.9	1.6	_	_	_	_	Iori et al. 1990
Italy (Sardinia)	85	91.76	_	3.53	_	4.71	5.88	_	1.18	Leoni et al. 1986
Slovenia	428	58.9	_	38.3	2.6	4.2	0.7	30.6	_	Vergles Rataj et al. 2013
Croatia	85	25.88	_	28.23	_	_	_	_	_	Rajković-Janje et al. 2002
Former Jugoslavia	532	44	15	44	11	_	20	_	_	Pavlović et al. 1997
Hungary	100	72	1	12	48	_	_	1	_	Széll et al. 2004
Romania	561	15	18.2	29.4	4.6	_	27.2	_	_	Sikó Barabási et al. 2010
Ukraine	25	36	4	40	48	60	24	_	_	Zvegintsova et al. 2007
Greece	314	43.9	5.1	28.6	2.5	17.5	8	_	_	Papadopoulos et al. 1997
Tunisia	9	44	11	_	_	67	33	_	_	Lahmar et al. 2014
Turkey (Kars)	20	_	_	20	65	_	_	_	_	Gicik et al. 2009
Jordan	9	33.3	_	_	_	_	_	_	_	El-Shehabi et al. 1999
Iraq	54	_	_	_	_	_	_	_	_	Mohammad et al. 2003
Iran (Western)	22	13.64	4.54	4.54	31.82	54.54	_	_	_	Dalimi et al. 2006
Iran (3 Climatic Zones)	37	- 8.1 -	- 5.4 -	- 10.8 32.4	-	-	-	-	- 8.1 -	Meshgi et al. 2009
Iran (Western)	52	16.3	9.1	35.6	30.1	47	3.8	_	_	Khanmohammadi et al. 2011

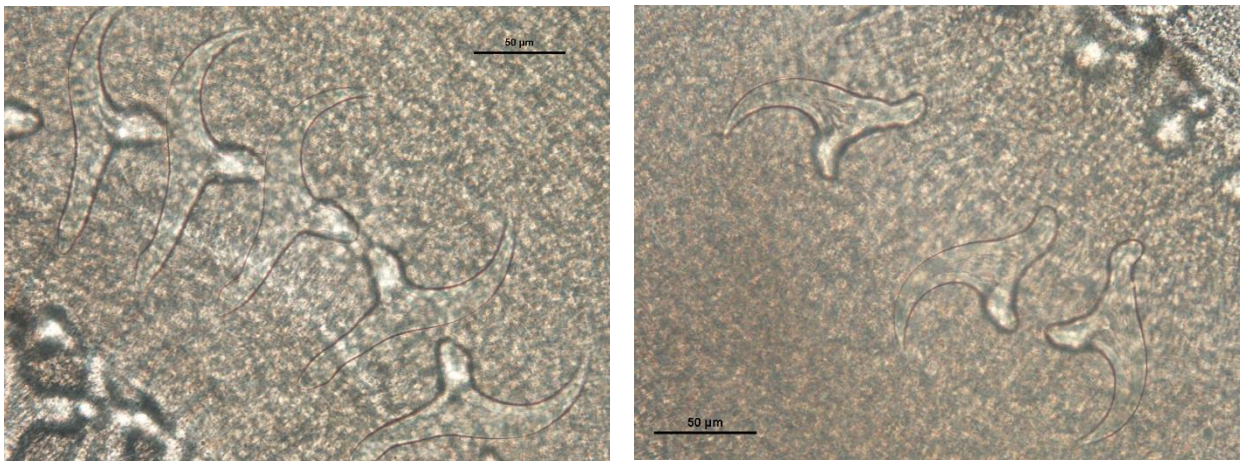
Country	N° foxes	*U. stenocephala*	*A. caninum*	*Toxocara canis*	*T. leonina*	*P. affinis*	*T. vulpis*	*Molineus spp.*	*D. immitis*	Reference
Kyrgyzstan	151	_	_	30.4	5.9	_	_	_	_	Ziadinov et al. 2010
China (Qinghai)	27	_	_	_	44	_	_	_	_	Li et al. 2013
Japan	7	_	_	71	_	_	_	29	_	Sato et al. 1999
Australia (Western)	147	18.2	_	14.9	4.7	_	_	_	_	Dybing et al. 2013
Argentina (Patagonia)	81	2.5 *Uncinaria* spp.	3.7 *Ancylostoma* spp.	_	24.7	_	_	_	_	Zanini et al. 2006



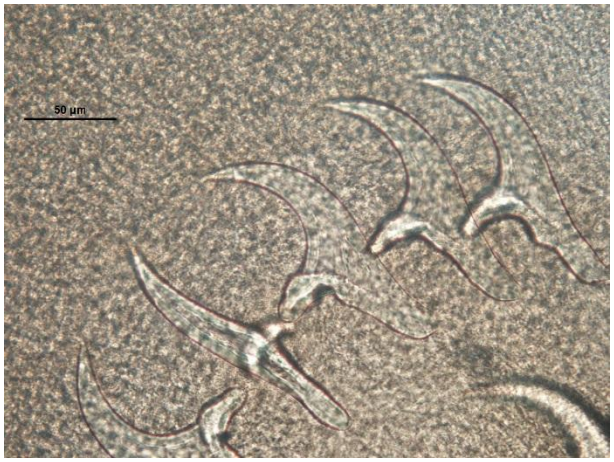
Alaria alata (on the left) and *Brachylaima* sp. (on the right) from red fox

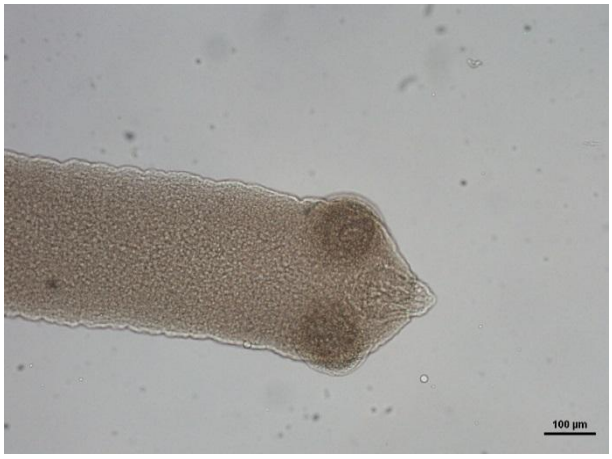


Small rostellar hooks of *Taenia crassiceps* (on the left) and small rostellar hooks of *Taenia polyacantha* (on the right) from red fox

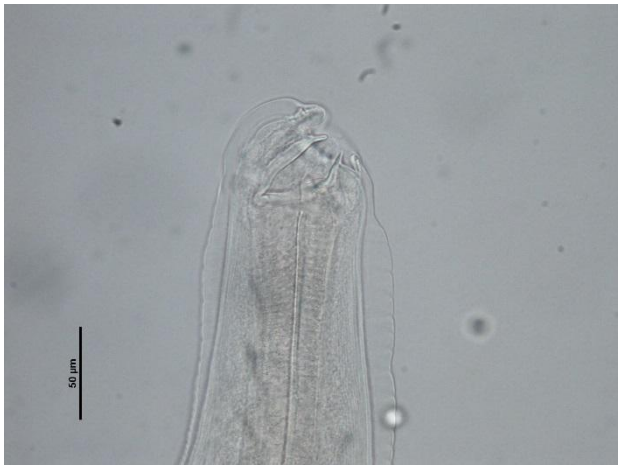


Small rostellar hooks of *Taenia hydatigena* (on the left) and small rostellar hooks of *Taenia ovis* (on the right) from a wolf





Small rostellar hooks of *Taenia pisiformis* (on the left) and scolex of *Dipylidium caninum* (on the right) from red fox



Cephalic end of two specimens of the nematode *Pterigodermatites affinis* from a red fox

Cephalic end of *Uncinaria stenocephala* from a red fox

REFERENCES

Al-Sabi MN, Chriél M, Jensen TH, Enemark HL. 2013. Endoparasites of the raccoon dog (*Nyctereutes procyonoides*) and the red fox (*Vulpes vulpes*) in Denmark 2009-2012 - A comparative study. International Journal for Parasitology: Parasites and Wildlife 2:144–151.

Al-Sabi MN, Halasa T, Kapel CM. 2014. Infections with cardiopulmonary and intestinal helminths and sarcoptic mange in red foxes from two different localities in Denmark. Acta Parasitologica 59:98-107.

Alvarez MF, Iglesias R, García J, Paniagua E, Sanmartin ML. 1995. Intestinal helminths of the red fox (*Vulpes vulpes* L.) in Galicia (Northwest Spain). Wiadomości Parazytologiczne 41: 429-442.

Arru E, Garippa G, Fico R. 1988. Sulla presenza di *Echinococcus granulosus* nella volpe (*Vulpes vulpes*) e nel lupo (*Canis lupus*). Atti della Società Italiana di Scienze Veterinarie 42:927–929.

Ballek D, Takla M, Ising-Volmer S, Stoye M. 1992a. The helminth fauna of red foxes (*Vulpes vulpes* Linnaeus 1758) in North Hesse and East Westphalia. 1. Cestodes. Deutsche Tierärztliche Wochenschrift 99:362–365.

Ballek D, Takla M, Ising-Volmer S, Stoye M. 1992b. The helminth fauna of red foxes (*Vulpes vulpes* Linnaeus 1758) in North Hesse and East Westphalia. 2. Nematodes. Deutsche Tierärztliche Wochenschrift 99:435-437.

Battelli G, Ostanello F, Baldelli R, Di Francesco A, Grilli R, Vizioli M. 2004. Human echinococcosis in the Emilia-Romagna region (northern Italy) in the years 1997 to 2002: an updating. Parassitologia 46:415–416.

Bertusi MG, Tosetti T. 1986. I Mammiferi dell'Emilia-Romagna Bologna: I.B.C. Regione Emilia-Romagna, Edizioni Grafis, Bologna. 134 pp.

Beveridge I, Rickard MD. 1975. The development of *Taenia pisiformis* in various definitive host species. International Journal for Parasitology 5:633–639.

Borgsteede FH. 1984. Helminth parasites of wild foxes (*Vulpes vulpes* L.) in the Netherlands. Zeitschrift fur Parasitenkunde 70: 281-285.

Bružinskaitė-Schmidhalter R, Šarkūnas M, Malakauskas A, Mathis A, Torgerson PR, Deplazes P. 2012. Helminths of red foxes (*Vulpes vulpes*) and raccoon dogs (*Nyctereutes procyonoides*) in Lithuania. Parasitology 139:120–127.

Bush AO, Lafferty KD, Lotz JM, Shostak AW. 1997. Parasitology meets ecology on its own terms: Margolis et al. revisited. The Journal of Parasitology 83:575–583.

Capelli G, Stancampiano L, Magi M, Poglayen G, Guberti V. 2003. Diversità delle comunità parassitarie intestinali in tre popolazioni di volpi. Journal of Mountain Ecology 7:199–205.

Carvalho-Varela M, Marcos MVM. 1993. A helmintofauna da raposa (*Vulpes vulpes silacea* Miller, 1907) em Portugal. Acta Parasitologica Portuguesa 1:73–79.

Craig HL, Craig PS. 2005. Helminth parasites of wolves (*Canis lupus*): a species list and an analysis of published prevalence studies in Nearctic and Palaearctic populations. Journal of Helminthology 79:95–103.

Criado-Fornelio A, Gutierrez-Garcia L, Rodriguez-Caabeiro F, Reus-Garcia E, Roldan-Soriano MA, Diaz-Sanchez MA. 2000. A parasitological survey of wild red foxes (*Vulpes vulpes*) from the province of Guadalajara, Spain. Veterinary Parasitology 92:245–251.

Dalimi A, Sattari A, Motamedi G. 2006. A study on intestinal helminthes of dogs, foxes and jackals in the western part of Iran. Veterinary Parasitology 142:129–133.

Deblock S, Pétavy AF, Gilot B. 1988. Helminthes intestinaux du Renard commun (*Vulpes vulpes* L.) dans le Massif central (France). Canadian Journal of Zoology 66:1562–1569.

Di Cerbo AR, Manfredi MT, Trevisiol K, Bregoli M, Ferrari N, Pirinesi F, Bazzoli S. 2008. Intestinal helminth communities of the red fox (*Vulpes vulpes* L.) in the Italian Alps. Acta Parasitologica 53:302–311.

Dybing NA, Fleming PA, Adams PJ. 2013. Environmental conditions predict helminth prevalence in red foxes in Western Australia. International Journal for Parasitology: Parasites and Wildlife 2:165–172.

Eira C, Vingada J, Torres J, Miquel J. 2006. The helminth community of the red fox, *Vulpes vulpes*, in Dunas de Mira (Portugal) and its effect on host condition. Wildlife Biology in Practice 2:26–36.

El-Shehabi FS, Abdel-Hafez SK, Kamhawi SA. 1999. Prevalence of intestinal helminths of dogs and foxes from Jordan. Parasitology Research 85:928-934.

Eom KS, Kim SH, Rim HJ. 1992. Second case of human infection with *Mesocestoides lineatus* in Korea. Korean Journal of Parasitology 30:147-150.

Euzeby J. 1982. Diagnostic expérimental des helminthoses animales (animaux domestiques-animaux de laboratoire-primates). Travaux pratiques d'Helminthologie Vétérinaire. Livre 2. Paris: Informations Techniques des Services Vétérinaires, Ministére de l'Agriculture. pp. 99–117.

Feldmeier H, Schuster A. 2012. Mini review: hookworm-related cutaneous larva migrans. European Journal of Clinical Microbiology and Infectious Diseases 31:915–918.

Ferroglio E, Trisciuoglio A, Lazzaron S, Filippetto E, Zanet S. 2012. Clinical and molecular findings of *Alaria alata* infection in dog from Aosta Valley, Italy. Mappe Parassitologiche, XXVII Congresso Nazionale Società Italiana di Parassitologia 18:160.

Flueck WT, Jones A. 2006. Potential existence of a sylvatic cycle of *Taenia ovis krabbei* in Patagonia, Argentina. Veterinary Parasitology 135:381–383.

Franssen F, Nijsse R, Mulder J, Cremers H, Dam C, Takumi K, van der Giessen J. 2014. Increase in number of helminth species from Dutch red foxes over a 35-year period. Parasites and Vectors. 7:166.

Fuentes MV, Galán-Puchades MT, Malone JB. 2003. Short report: a new case report of human *Mesocestoides* infection in the United States. The American Journal of Tropical Medicine and Hygiene 68:566-567.

Fujita O, Oku Y, Okamoto M, Sato H, Ooi HK, Kamiya M, Rausch RL. 1991. Early development of larval *Taenia polyacantha* in experimental intermediate hosts. Journal of the Helminthological Society of Washington 58:100-109.

Garippa G. 2006. Updates on cystic echinococcosis (CE) in Italy. Parassitologia 48:57–59.

Gestaldi B. 1854. Cenni sopra alcuni nuovi Elminti della *Rana esculenta* con nuove osservazioni sul *Codonocephalus mutabilis* (Diesing). Tesi per aggregazione al Collegio della Facoltà delle Scienze Fisiche e Mathematiche nella R. Università di Torino. pp. 25–36.

Gibbs HC. 1961. Studies on the life cycle and developmental morphology of *Dochmoides stenocephala* (Railliet, 1884) (Ancylostomatidae: Nematoda). Canadian Journal of Zoology 39:325–348.

Gicik Y, Kara M, Sari B, Kiliç K, Arslan MÖ. 2009. Intestinal parasites of red foxes (*Vulpes vulpes*) and their zoonotic importance for humans in Kars Province. Kafkas Üniversitesi Veteriner Fakultesi Dergisi 15:135-140.

Gori F, Armua-Fernandez MT, Milanesi P, Serafini M, Magi M, Deplazes P, Macchioni F. 2015. The occurrence of taeniids of wolves in Liguria (northern Italy). International Journal for Parasitology: Parasites and Wildlife 4:252–255.

Gortázar C, Villafuerte R, Lucientes J, Fernández-de-Luco D. 1998. Habitat related differences in helminth parasites of red foxes in the Ebro valley. Veterinary Parasitology 80:75–81.

Guberti V, Bolognini M, Lanfranchi P, Battelli G. 2004. *Echinococcus granulosus* in the wolf in Italy. Parassitologia 46:425–427.

Guberti V, Poglayen G. 1991. Parasitic zoonoses: survey in foxes (*Vulpes vulpes*) in the Northern Apennines. Hystrix 3:167–173.

Guberti V, Stancampiano L, Francisci F. 1993. Intestinal helminth parasite community in wolves (*Canis lupus*) in Italy. Parassitologia 55:59–65.

Hackett F, Walters TMH. 1980. Helminths of the red fox in mid-Wales. Veterinary Parasitology 7:181–184.

Iori A, Costantini R, Cancrini G. 1990. Parassiti di volpi (*Vulpes vulpes*) provenienti da alcune regioni italiane. Parassitologia 32:153–154.

Khalil LF, Jones A, Bray RA. 1994. Keys to the cestode parasites of vertebrates. Wallingford, Oxon: Cab international. 768 pp.

Khanmohammadi M, Fallah E, Reyhani rad S. 2011. Epidemiological studies on fauna and prevalence of parasite helminthes on red fox (*Vulpes vulpes*) in Sarab District, East Azerbaijan Province, Iran. Annals of Biological Research 2:246-251.

Lahmar S, Boufana B, Ben Boubaker S, Landolsi F. 2014. Intestinal helminths of golden jackals and red foxes from Tunisia. Veterinary Parasitology 204:297–303.

Lassnig H, Prosl H, Hinterdorfer F. 1998. Zur Parasitenfauna des Rotfuchses (*Vulpes vulpes*) in der Steiermark. Wiener Tierärztliche Monatsschrift 85:116–122.

Leoni A, Garippa G, Masala S. 1986. Osservazioni sull'elmintofauna della volpe (*Vulpes vulpes*) in Sardegna. Parassitologia 28:268–269.

Lescano AG, Zunt J. 2013. Other cestodes: sparganosis, coenurosis and *Taenia crassiceps* cysticercosis. Handbook of Clinical Neurology 114:335–345.

Letková V, Lazar P, Čurlík J, Goldová M, Kočišová A, Košuthová L, Mojžišová J. 2006. The red fox (*Vulpes vulpes* L.) as a source of zoonoses. Veterinarski Arhiv 76:73–81.

Li W, Guo Z, Duo H, Fu Y, Peng M, Shen X, Tsukada H, Irie T, Nasu T, Horii Y, Nonaka N. 2013. Survey on helminths in the small intestine of wild foxes in Qinghai, China. The Journal of Veterinary Medical Science 75: 1329-1333.

Loos-Frank B, Zeyhle E. 1982. The intestinal helminths of the red fox and some other carnivores in southwest Germany. Zeitschrift fur Parasitenkunde 67:99–113.

Lucius R, Böckeler W, Pfeiffer AS. 1988. Parasiten der haus-, nutz- und wildtiere Schleswig-Holsteins: parasiten der inneren organe des rotfuchses (*Vulpes vulpes*). Zeitschrift fur Jagdwissenschaft 34:242–255.

Macchioni F, Gustinelli A, Caffara M, Chelucci L, Magi M, Fioravanti ML. 2012. *Red fox (Vulpes vulpes) as wild reservoir of Opisthorchis felineus (Digenea: Opisthorchiidae) in Italy.* Mappe Parassitologiche XXVII Congresso Nazionale Società Italiana di Parassitologia 18:186.

Magi M, Guardone L, Mignone W, Prati MC, Macchioni F. 2016. Intestinal helminths of red foxes (*Vulpes vulpes*) in north-west Italy. Helminthologia 53:31–38.

Magi M, Macchioni F, Dell'Omodarme M, Prati MC, Calderini P, Gabrielli S, Iori A, Cancrini G. 2009. Endoparasites of red fox (*Vulpes vulpes*) in Central Italy. Journal of Wildlife Diseases 45:881–885.

Manfredi MT, Giacometti A, Fraquelli C, Piccolo G. 2003. Studio della popolazione elmintica in volpi (*Vulpes vulpes*) del Trentino Alto-Adige. Journal of Mountain Ecology 7:261–263.

Manke K, Stoye M. 1998. Parasitological studies of red foxes (*Vulpes vulpes*) in the northern districts of Schleswig-Holstein. Tierärztliche Umschau 53:207-214.

Martínez-Carrasco C, Ruiz de Ybánez MR, Sagarminaga JL, Garijo MM, Moreno F, Acosta I, Hernández S, Alonso FD. 2007. Parasites of the red fox (*Vulpes vulpes* Linnaeus, 1758) in Murcia, southeast Spain. Revue de Médicine Vétérinaire 158:331–335.

Mech LD. 1970. The wolf. The ecology and behavior of an endangered species. Natural History Press. N.Y.: Doubleday Publishing Co. 389 pp.

Meshgi B, Eslami A, Bahonar AR, Kharrazian-Moghadam M, Gerami-Sadeghian A. 2009. Prevalence of parasitic infections in the red fox (*Vulpes vulpes*) and golden Jackal (*Canis aureus*) in Iran. Iranian Journal of Veterinary Research 10:387-391.

Mohammad MK, Jasim MK, Al-Moussawi AA. 2003. The parasitic fauna of the red fox *Vulpes vulpes* (L., 1758) in Iraq with some notes on its biology and ecology. Bulletin of the Iraq Natural History Museum 10:59-76.

Möhl K, Grosse K, Hamedy A, Wüste T, Kabelitz P, Lücker E. 2009. Biology of *Alaria* spp. and human exposition risk to *Alaria mesocercariae*-a review. Parasitology Research 105:1–15.

Molin R. 1861. Prodromus Faunae Helminthologicae Venetae. Wien: Kaiserlich-Königlichen hof- und staatsdruckerei. 193 pp.

Murphy TM, O'Connell J, Berzano M, Dold C, Keegan JD, McCann A, Murphy D, Holden NM. 2012. The prevalence and distribution of *Alaria alata*, a potential zoonotic parasite, in foxes in Ireland. Parasitology Research 111:283–290.

Ntoukas V, Tappe D, Pfütze D, Simon M, Holzmann T. 2013. Cerebellar cysticercosis caused by larval *Taenia crassiceps* tapeworm in immunocompetent woman, Germany. Emerging Infectious Diseases 19:2008–2011.

Papadopoulos H, Himonas C, Papazahariadou M, Antoniadou-Sotiriadou K. 1997. Helminths of foxes and other wild carnivores from rural areas in Greece. Journal of Helminthology 71:227–231.

Pétavy AF, Deblock S. 1980. Helminths of the common fox (*Vulpes vulpes* L.) from the Massif Central (France). Annales de Parasitologie Humaine et Comparee 55:379–391.

Pétavy AF, Deblock S, Prost C. 1990. Epidémiologie de l'échinococcose alvéolare en France I. Helminthes intestinaux du renard commun (*Vulpes vulpes* L.) en Haute-Savoie. Annales de Parasitologie Humaine et Comparee 65:22–27.

Pfeiffer F, Kuschfeldt S, Stoye M. 1997a. Helminth fauna of the red fox (*Vulpes vulpes* Linne 1758) in south Sachsen-Anhalt. 1. Cestodes. Deutsche Tierärztliche Wochenschrift 104:445-448.

Pfeiffer F, Kuschfeldt S, Stoye M. 1997b. Helminth fauna of the red fox (*Vulpes vulpes* Linne 1758) in south Sachsen-Anhalt. 2. Nematodes. Deutsche Tierärztliche Wochenschrift 104: 461-450.

Poglayen G, Guberti V, Leoni B. 1985. Parasites present in foxes (*Vulpes vulpes*) of the province of Forli. Parassitologia 27:303–311.

Poulin R. 2006. Evolutionary ecology of parasites. 2nd ed. Princeton: Princeton University Press. 360 pp.

Rajković-Janje R, Marinculić A, Bosnić S, Benić M, Vinković B, Mihaljević Ž. 2002. Prevalence and seasonal distribution of helminth parasites in red foxes (*Vulpes vulpes*) from the Zagreb County (Croatia). Zeitschrift fur Jagdwissenschaft 48:151–160.

Ramisz A, Nicpoń J, Balicka-Ramisz A, Pilarczyk B, Pacoń J, Piekarska J. Die prävalenz von darmhelminthen bei rotfüchsen (*Vulpes vulpes* L.) aus dem südwestlichen teil Polens. Tierärztliche Umschau 59:601-604.

Reiczigel J, Rozsa L, Reiczigel A, Fabian I. 2013. Quantitative Parasitology (QPweb). Available: http://www2.univet.hu/qpweb**.** [Accessed 8 October 2016].

Reperant LA, Hegglin D, Fischer C, Kohler L, Weber JM, Deplazes P. 2007. Influence of urbanization on the epidemiology of intestinal helminths of the red fox (*Vulpes vulpes*) in Geneva, Switzerland. Parasitology Research 101:605–611.

Richards DT, Harris S, Lewis JW. 1993. Epidemiology of *Toxocara canis* in red foxes (*Vulpes vulpes*) from urban areas of Bristol. Parasitology 107:167–173.

Richards DT, Harris S, Lewis JW. 1995. Epidemiological studies on intestinal helminth parasites of rural and urban red foxes (*Vulpes vulpes*) in the United Kingdom. Veterinary Parasitology 59:39–51.

Rossi L, Iori A, Cancrini G. 1983. Osservazioni sulla fauna parassitaria della popolazione di volpi presente nel Parco Regionale “La Mandria”. Parassitologia 25:340–343.

Saeed I, Maddox-Hyttel C, Monrad J, Kapel CM. 2006. Helminths of red foxes (*Vulpes vulpes*) in Denmark. Veterinary Parasitology 139:168–179.

Sánchez C, Lucientes J, Albala F. 1977. Contribución al conocimiento de la helmintofauna del zorro (*Vulpes vulpes*) en la provincia de Zaragoza. Anales de la Facultad de Veterinaria, Universidad de Zaragoza 11-12:335–339.

Sato H, Inaba T, Ihama Y, Kamiya H. 1999. Parasitological survey on wild carnivora in north-western Tohoku, Japan. The Journal of Veterinary Medical Science 61:1023-1026.

Schöffel I, Schein E, Wittstadt U, Hentsche J. 1991. Parasite fauna of red foxes in Berlin (West). Berliner und Münchener Tierärztliche Wochenschrift 104:153–157.

Segovia JM, Torres J, Miquel J. 2004. Helminth parasites of the red fox (*Vulpes vulpes* L., 1758) in the Iberian Peninsula: an ecological study. Acta Parasitologica 49:67–79.

Segovia JM, Torres J, Miquel J, Llaneza L, Feliu C. 2001. Helminths in the wolf, *Canis lupus*, from north-western Spain. Journal of Helminthology 75:183–192.

Shimalov VV, Shimalov VT. 2003. Helminth fauna of the red fox (*Vulpes vulpes* Linnaeus, 1758) in southern Belarus. Parasitology Research 89:77-78.

Sikó Barabási S, Fok E, Gubányi A, Mészáros F, Cozma V. 2010. Helminth fauna of the small intestine in the European red fox, *Vulpes vulpes*, with notes on the morphological identification of *Echinococcus multilocularis.* Scientia Parasitologica 11:141–151.

Smith GC, Gangadharan B, Taylor Z, Laurenson MK, Bradshaw H, Hide G, Hughes JM, Dinkel A, Romig T, Craig PS. 2003. Prevalence of zoonotic important parasites in the red fox (*Vulpes vulpes*) in Great Britain. Veterinary Parasitology 118:133-142.

Soldati G, Pavesi M, Canestri-Trotti G, Cocchi MG, Gaiardi S, Morganti L, Prosperi S, Sanguinetti V, Stanzani F. 1976. Research on infections and parasitic agents in foxes of the Modenese Apennines. Rivista di Parassitologia 37:329–332.

Spagnesi M, De Marinis AM. 2002. Mammiferi d'Italia. Quaderni di Conservazione della Natura, 14. Ministero dell'Ambiente e della Tutela del Territorio e del Mare - Istituto Nazionale Per la Fauna Selvatica "A. Ghigi". 309 pp.

Suchentrunk F, Sattmann H. 1994. Prevalence of intestinal helminths in Austrian red foxes (*Vulpes vulpes* L.) (Cestoda, Nematoda) Annalen des Naturhistorischen Museums in Wien 96B:29–38.

Széll Z, Sréter T, Varga I. 2004. Prevalence, veterinary and public health aspects of gastrointestinal parasites in red foxes (*Vulpes vulpes*) in Hungary. Magyar Allatorvosok Lapja 126:293–299.

Széll Z, Tolnai Z, Sréter T. 2013. Environmental determinants of the spatial distribution of *Alaria alata* in Hungary. Veterinary Parasitology 198:116–121.

Thul JE, Forrester DJ, Abercrombie CL. 1985. Ecology of parasitic helminths of Wood Ducks, *Aix sponsa,* in the Atlantic flyway. Proceedings of the Helminthological Society of Washington 52:297–310.

Urquhart GM, Armour J, Duncan JL, Dunn AM, Jennings FW. 1996. Veterinary Parasitology. 2nd ed. Glasgow: Blackwell Science Ltd. 307 pp.

Vergles Rataj A, Posedi J, Zele D, Vengušt G. 2013. Intestinal parasites of the red fox (*Vulpes vulpes*) in Slovenia. Acta Veterinaria Hungarica 61:454–462.

Vervaeke M, Dorny P, De Bruyn L, Vercammen F, Jordaens K, Van Den Berge K, Verhagen R. 2005. A survey of intestinal helminthes of red foxes (*Vulpes vulpes*) in northern Belgium. Acta parasitologica 50:221–227.

Wessbecher H, Dalchow W, Stoye M. 1994a. The helminth fauna of the red fox (*Vulpes vulpes* Linne 1758) in the administrative district of Karlsruhe. 1. Cestodes. Deutsche Tierärztliche Wochenschrift 101:322–326.

Wessbecher H, Dalchow W, Stoye M. 1994b. The helminth fauna of the red fox (*Vulpes vulpes* Linne 1758) in the administrative district of Karlsruhe. 2. Nematodes. Deutsche Tierärztliche Wochenschrift 101:362–364.

Williams BM. 1976. The intestinal parasites of the red fox in South West Wales. British Veterinary Journal 132:309–312.

Wolfe A, Hogan S, Maguire D, Fitzpatrick C, Vaughan L, Wall D, Hayden TJ, Mulcahy G. 2001. Red foxes (*Vulpes vulpes*) in Ireland as hosts for parasites of potential zoonotic and veterinary significance. The Veterinary Record 149:759-763.

Zanini F, Laferrara M, Bitsch M, Pérez H, Elissondo MC. 2006. Epidemiological studies on intestinal helminth parasites of the patagonian grey fox (Pseudalopex griseus) in Tierra del Fuego, Patagonia Argentina. Veterinary Parasitology 136:329-334.

Ziadinov I, Deplazes P, Mathis A, Mutunova B, Abdykerimov K, Nurgaziev R, Torgerson PR. 2010. Frequency distribution of *Echinococcus multilocularis* and other helminths of foxes in Kyrgyzstan. Veterinary Parasitology 171:286-292.

Zvegintsova NS, Dumenko VP, Varodi EI. 2007. Helminthes of the Red Fox (*Vulpes vulpes*) in the Askania Nova Biosphere Reserve (Ukraine). Vestnik Zoologii 41:153-157.

The results of this research have been published in the Italian Journal of Zoology, 83(4):503-513.

Taylor & Francis
Taylor & Francis Group

Italian Journal of Zoology

ISSN: 1125-0003 (Print) 1748-5851 (Online) Journal homepage: http://www.tandfonline.com/loi/tizo20

Helminth parasites of the red fox Vulpes vulpes (L., 1758) and the wolf Canis lupus italicus Altobello, 1921 in Emilia-Romagna, Italy

A. Fiocchi, A. Gustinelli, L. Gelmini, G. Rugna, M. Renzi, M. C. Fontana & G. Poglayen

To cite this article: A. Fiocchi, A. Gustinelli, L. Gelmini, G. Rugna, M. Renzi, M. C. Fontana & G. Poglayen (2016) Helminth parasites of the red fox Vulpes vulpes (L., 1758) and the wolf Canis lupus italicus Altobello, 1921 in Emilia-Romagna, Italy, Italian Journal of Zoology, 83:4, 503-513, DOI: 10.1080/11250003.2016.1249966

To link to this article: http://dx.doi.org/10.1080/11250003.2016.1249966

2. RESEARCH ON *ECHINOCOCCUS GRANULOSUS* IN RED FOXES (*VULPES VULPES*) FROM EASTERN EMILIA-ROMAGNA REGION.

ALFREDO FIOCCHI[1], ANDREA GUSTINELLI[1], BENEDETTO MORANDI[1], GIOVANNI TOSI[2], GIOVANNI POGLAYEN[1]

[1]*Department of Veterinary Medical Sciences, Alma Mater Studiorum University of Bologna, Ozzano Emilia (BO), Italy*

[2]*Lombardy and Emilia-Romagna Experimental Zooprophylactic Institute (IZSLER), Forlì, Italy*

TO BE PUBLISHED IN A PEER-REVIEWED JOURNAL

ABSTRACT

Between January 2013 and June 2015 we analysed a total of 313 wild red foxes (*Vulpes vulpes*) collected in the provinces of Forlì-Cesena and Rimini, Emilia-Romagna region. The first intestinal tract (approximately the first quarter of the small intestine) was removed during necropsy and helminths were collected using the sedimentation and counting technique (SCT). The main aim of the research was *Echinococcus granulosus*, which commonly inhabits the first tract of the small intestine. Overall 16 helminth species were identified, three trematodes (*Alaria alata, Phagicola* sp., *Brachylaima* sp.), seven cestodes (*Mesocestoides* spp., *Taenia polyacantha, Taenia crassiceps, Dipylidium caninum, Joyeuxiella* sp., *Taenia pisiformis, Taenia martis*) and six nematodes (*Toxocara canis*, *Uncinaria stenocephala*, *Molineus legerae, Pterigodermatites affinis, Toxascaris leonina* and *Toxocara cati*). The most prevalent parasites were *Mesocestoides* spp. (13.7%), *Taenia polyacantha* (5.4%), *Toxocara canis* (24.6%) and *Uncinaria stenocephala* (13.7%). Despite not succeeding in recovering any specimen of *E. granulosus*, a cestode that has been recently detected in faeces of wolves from Emilia-Romagna eastern Apennines, the present survey confirms that red foxes are definitive hosts for several helminth species in these territories. Moreover *A. alata, Phagicola* sp., *Joyeuxiella* sp. and *M. legerae* all represent rare parasitological findings for the Emilia-Romagna region.

Keywords: Red fox, helminths, *Echinococcus granulosus*, Emilia-Romagna, Italy.

INTRODUCTION

Echinococcus granulosus is a zoonotic cestode whose life cycle is primarily domestic. However *E. granulosus*-complex sylvatic cycles are well established in North America (*E. canadensis*, cervid strain G8-G10), Northern Eurasia (*E. canadensis*, cervid strain G6-G8-G10) and Australia (*E. granulosus*, sheep strain G1), where especially wolves and australian wild dogs are the most important definitive hosts (Carmena and Cardona 2014). Significant sylvatic cycles are probably present also in cystic echinococcosis (CE) endemic northern african countries (e.g. Tunisia) and Middle-East countries (e.g. Western Iran), where wild canids such as golden jackals and foxes have been found infected (Dalimi et al. 2002; Carmena and Cardona 2014). Regarding Europe, the occurrence of a noteworthy *E. granulosus* prevalence rate in wild carnivores has been reported in wolves in mediterranean countries, such as Spain and Italy. However assessing the existence of a true sylvatic cycle of the parasite is difficult (Guberti et al. 2004; Carmena and Cardona 2014; Poglayen et al. 2017). Infection of wild red foxes has been documented in the United Kingdom, Spain and Sardinia (Italy), but most of the authors agreed that these mammals are not important in the epidemiology of *E. granulosus* and should be regarded as mere spill-over events from the domestic cycle (Arru et al. 1988; Richards et al. 1995; Segovia et al. 2004). Higher prevalences of infection in red foxes were found in certain areas of Great Britain, like Wales (Cook 1965: 22%; Williams 1976: 2.7%; Hackett and Walters 1980: 7.1%), in Eastern Turkey (Gicik et al. 2009: 5%), Western Iran (Dalimi et al. 2002: 5%; Dalimi et al. 2006: 4.5%), Tunisia (Lahmar et al. 2009: 13.3%) and South-eastern Australia (Jenkins and Morris 2003: 18.6%). In these countries, all endemic for cystic echinococcosis in men and domestic animals, the mentioned authors reported higher rates and intensities of infection with *E. granulosus* in dogs (Hackett and Walters 1980; Gicik et al. 2009; Dalimi et al. 2002,2006), jackals (Lahmar et al. 2009, 2014) and dingoes (Jenkins and Morris 2003; Jenkins 2006) living in the same territories as the red foxes examined. Therefore this species may not be such a favourable host for *E. granulosus* as wild or domestic dogs. Zanini et al. (2006) drew the same conclusion studying the diffusion of the parasite in grey foxes (*Pseudalopex griseus*) in Tierra del Fuego, Argentina**.** The aim of the present study was to analyse the possible presence of *E. granulosus* in the intestine of red foxes from South-Eastern Emilia-Romagna region. Despite the diffusion of CE in this region being considered sporadic in men and domestic animals (Grosso et al. 2012), these territories are adjacent to Central Apennines (Toscana and Marche regions), in the recent past an endemic area for cystic echinococcosis in sheep. An epidemiological survey showed that

CE was found in 47% of sheep slaughtered in the province of Arezzo (Bio and Fagiolo 2004), a territory bordering with Emilia-Romagna eastern Apennines. Moreover *E. granulosus* (G1-G3) has been recently detected in feces of wolves living in mountainous districts of eastern Emilia-Romagna (Poglayen et al. 2017: prevalence 5.5%), leading us to check if the parasite has eventually spread to other wildlife definitive hosts like the red fox.

MATERIALS AND METHODS

Between January 2013 and June 2015 we analysed a total of 313 wild red foxes (*Vulpes vulpes*), collected in official culling campaigns or found dead in the provinces of Forlì-Cesena and Rimini, Emilia-Romagna region . Fifty-one animals (16.3%) came from lowland territories, 262 (83.7%) from hilly or mountainous territories. The first intestinal tract (approximately the first quarter of the small intestine) was removed during necropsy at the Veterinary Diagnostic Laboratory (IZSLER, Lombardy and Emilia-Romagna Experimental Zooprophylactic Institute, Forlì), frozen at – 20 °C and subsequently sent to the Parasitology Laboratory (DIMEVET, Department of Veterinary Medical Sciences, University of Bologna). Once defrosted, the intestinal tract was placed inside a plastic tray and opened up along its entire length. The mucosa and the intestinal content were minutely inspected for the presence of visible helminths, which were subsequently removed and placed in Petri dishes with deionizied water. Afterwards, the mucosal layer was repeatedly washed with deionizied water and scraped with a spoon. The collected material was transferred to a glass container, closable with a lid in which a 1 mm wire mesh had been firmly inserted. Repeated washings with running tap water through the mesh were carefully performed until the content was sufficiently clear to be examined by the stereomicroscope. Next, the material was allowed to precipitate inside a conical jar for nearly 30 minutes and then observed by the stereomicroscope. Recovered helminths were counted, stored in 70% alcohol, clarified in lactophenol and subsequently identified according to standard protocols and previous descriptions (Pétavy and Deblock 1980; Euzeby 1982; Khalil et al. 1994).

Table 1. Intestinal helminths recovered in the first intestinal tract of 313 red foxes (*Vulpes vulpes*) from Forlì-Cesena and Rimini provinces, Emilia-Romagna, Italy.

Helminth species	N°positive (Prevalence)	Mean Intensity	Intensity range
***Mesocestoides* spp.**	43 (13.7%)	15.8	1-340
Taenia polyacantha	17 (5.4%)	7.2	1-23
Taenia crassiceps	4 (1.3%)	13.7	1-48
Dipylidium caninum	3 (0.9%)	1	1
***Joyeuxiella* sp.**	2 (0.6%)	9	9
Taenia martis	1 (0.3%)	1	1
Taenia pisiformis	1 (0.3%)	1	1
***Taenia* spp.**	16 (5.1%)	-	-
Toxocara canis	77 (24.6%)	3.5	1-41
Uncinaria stenocephala	43 (13.7%)	3.6	1-17
Molineus legerae	13 (4.1%)	1.5	1-3
Pterigodermatites affinis	2 (0.6%)	1.5	1-2
Toxascaris leonina	1 (0.3%)	2	2
Toxocara cati	1 (0.3%)	1	1
Alaria alata	3 (0.9%)	2	1-4
***Phagicola* sp.**	3 (0.9%)	158	2-460
***Brachylaima* sp.**	1 (0.3%)	1	1

RESULTS

The helminthological findings in red foxes are presented in Table 1 with data regarding prevalence, mean intensity of infection and intensity range. Overall 16 helminth species were identified, three trematodes (*Alaria alata*, *Phagicola* sp., *Brachylaima* sp.), seven cestodes (*Mesocestoides* spp., *Taenia polyacantha*, *Taenia crassiceps*, *Dipylidium caninum*, *Joyeuxiella* sp., *Taenia pisiformis*, *Taenia martis*) and six nematodes (*Toxocara canis*, *Uncinaria stenocephala*, *Molineus legerae*, *Pterigodermatites affinis*, *Toxascaris leonina* and *Toxocara cati*). The most prevalent parasites were *Mesocestoides* spp. (13.7%), *Taenia polyacantha* (5.4%), *Toxocara canis* (24.6%) and *Uncinaria stenocephala* (13.7%). Some cestodes could not be identified to the species level either because they lacked scolices or these were too degraded to allow a precise identification. In four animals unidentified ascarids were found.

DISCUSSION

The results shown in Table 1 are partially representative of the intestinal helminthic fauna of red foxes from Forlì-Cesena and Rimini provinces, since only the first part of the small intestine was examined in our survey. This choice was motivated by the fact that the main aim of the research was *Echinococcus granulosus*, which commonly inhabits the first tract of the intestine, especially in animals harboring low intensity rates of infection (El-Shehabi et al. 1999). Consequently, prevalence and mean intensity rates of all the helminths described, in particular those of *U. stenocephala* and cestodes, must be regarded as partial data, because these parasites can be also recovered in the intermediate and posterior parts of the small intestine (Sikó Barabási et al. 2011; Al-Sabi et al. 2013). Nevertheless, we could find a high number of helminth species and several considerations can undoubtedly be done. Unfortunately we were not able to isolate any ***E. granulosus*** from the 313 samples examined. This confirms the fact that this cestode is extremely rare in the red fox, a wild canid regarded by several authors as an unfavorable host for this parasite and not so important in the maintenance of its life cycle (Hackett and Walters, 1980; Arru et al. 1988; Guberti and Poglayen 1991; Papadopoulos et al. 1997; Segovia et al. 2004). We chose the sedimentation and counting technique (SCT) because, despite being a time-consuming method, it has a very high level of sensitivity and is a cheaper technique compared to copro-antigen (Cp-Ag) ELISA and PCR-based tests (Carmena and Cardona 2014). In addition, it allows us to directly demonstrate the presence of adult forms of the parasite. ***Mesocestoides* spp.** resulted the most common cestode (prevalence 13.7%) (Table 1), in agreement with several other

european surveys (Loos-Frank and Zeyhle 1982; Sikó Barabási et al. 2010; Al-Sabi et al. 2014). The high prevalence of this parasite can be explained by the fact that different kinds of vertebrates, including amphibians, reptiles, birds and mammals (e.g. rodents) can act as second intermediate hosts (Loos-Frank and Zeyhle 1982; Papadopoulos et al. 1997). Therefore foxes and other carnivores can get infected after the ingestion of a wide range of prey. Similar prevalence values were reported by Soldati et al. (1976) (13% of 23 foxes) and Guberti and Poglayen (1991) (11% of 153 foxes) in Emilia-Romagna Apennines. Despite only the first intestinal tract being analysed, interestingly ***Taenia polyacantha*** showed a slightly higher prevalence rate (5.4%) (Table 1) than in foxes from central Emilia-Romagna (Fiocchi et al. 2016: 3.5% of 57 foxes). This might be due to the fact that most part of the animals in the present survey came from hilly or mountainous territories, a typology of habitat that favors intermediate host of the cestode (e.g. *Myodes glareolus*, *Sciurus vulgaris*, *Apodemus flavicollis*) (Loos-Frank and Zeyhle 1982). Poglayen et al. (2017) also recovered *T. polyacantha* (prevalence 1.8%) in feces of wolves from the same geographical area. Conversely, an extremely low prevalence rate of ***T. crassiceps***, another rodent-transmitted cestode, was found in foxes (1.3%) (Table 1). In our previous survey, 17.5% of the red foxes were found infected with *T. crassiceps* (Fiocchi et al. 2016), however all these animals came from lowlands territories, not much represented in the present study. Regarding *Dipylidiidae*, three animals harbored ***Dipylidium caninum*** and two a ***Joyeuxiella*** species (Table 1). This uncommon genus of cestodes has been previously reported in red foxes from Liguria and Piedmont (Magi et al. 2016), Lazio (Iori et al. 1990) and Sardinia (Leoni et al. 1986) in Italy. It is typical of other southern European countries such as Portugal and Spain (Carvalho-Varela and Marcos 1993; Gortázar et al. 1998; Segovia et al. 2004; Martínez-Carrasco et al. 2007), and Greece (Papadopoulos et al. 1997). It was also described in several Middle East countries such as Turkey (Mimioğlu et al. 1965), Jordan (El-Shehabi et al. 1999), Iraq (Mohammad et al. 2003) and Iran (Dalimi et al. 2006; Meshgi et al. 2009; Nabavi et al. 2014). According to some authors (Papadopoulos et al. 1997; Mohammad et al. 2003), reptiles are important intermediate hosts in the life cycle of this tapeworm. Only one fox was positive for ***T. pisiformis***, another one for ***T. martis***, a cestode typical of mustelids (Loos-Frank and Zeyhle 1982; Brunet et al. 2014). ***Toxocara canis*** (prevalence 24.6%) and ***Uncinaria stenocephala*** (prevalence 13.7%) (Table 1) were the most common nematodes recovered and this in agreement with several other european surveys (Loos-Frank and Zeyhle 1982; Richards et al. 1995; Vervaeke et al. 2005; Al-Sabi et al. 2014). Two foxes were infected

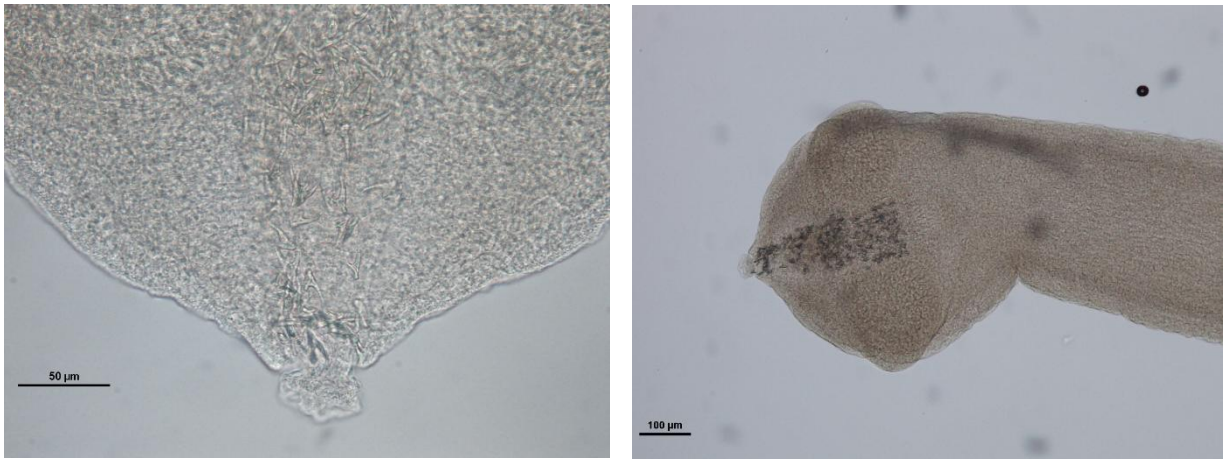
with ***Toxascaris leonina*** and ***Toxocara cati*** respectively. Interestingly, the prevalence rate of *U. stenocephala*, despite being higher than previously reported by Guberti and Poglayen (1991) (11.8%) in the same geographical area, showed a notably lower value in comparison with foxes from Modena and Bologna provinces (75.4%) (Fiocchi et al. 2016). A possible reason might be the fact that the latter territory is characterized by a more humid climate, especially in hilly and mountainous areas. This factor probably fits better the humidity requirements of preparasitic stages of *U. stenocephala.* Several animals were infected with ***Molineus legerae*** (prevalence 4.1%) (Table 1), this representing the first report of the parasite in red foxes from the Emilia-Romagna region. Nematodes of this genus (fam. *Trichostrongylidae*) have been described in foxes from Portugal (Eira et al. 2006: *M. patens*), Spain (Segovia et al. 2004: *M. patens* and *M. legerae*), France (Petavy and Deblock 1980: *M. patens*), the Netherlands (Borgsteede 1984: *M. patens*), Germany (Schöffel et al. 1991: *M. patens*), Belarus (Shimalov and Shimalov 2003: *M. patens*), Hungary (Széll et al. 2004: *M. patens*), Austria (Suchentrunk and Sattmann 1994: *M. patens*), Slovenia (Vergles Rataj et al. 2013: *M. patens*), Northern Italy (Manfredi et al. 2003; Di Cerbo et al. 2008; Magi et al. 2016: *M. legerae*) and Japan (Sato et al. 1999). According to some authors (Suchentrunk and Sattmann 1994; Vergles Rataj et al. 2013), *M. patens* may also occur accidentally in red foxes after predation on small carnivores like mustelids. Data from our survey demonstrate that this nematode is not only present in the Alpine region (Magi et al. 2016), but also on the northern Apennines range. Only two foxes were found infected with ***Pterigodermatites affinis*** (prevalence 0.6%) (Table 1), a spirurid nematode whose intermediate and paratenic hosts are insects and reptiles respectively (Deblock et al. 1988; Papadopoulos et al. 1997). Regarding digenean trematodes, three foxes were positive for ***Alaria alata*** (Table 1), an amphibian-transmitted parasite which has been rarely reported in Italy (Fiocchi et al. 2016). The animals were killed near the city of Forlì and in nearby low hills. *A. alata*'s complex life cycle requires a freshwater snail as the first intermediate host and an amphibian as the second one. Reptiles, rodents, wild boars and other vertebrates can act as paratenic hosts after feeding on infected amphibians (Möhl et al. 2009). Definitive hosts, usually members of the family *Canidae*, become infected after ingesting mesocercariae contained in amphibians or paratenic hosts. The young flukes migrate through the gut and other viscera, change into the metacercarial stage in the lungs and, after being swallowed, reach the small intestine (Möhl et al. 2009). Three other foxes were infected with ***Ascocotyle* (*Phagicola*)**, a genus of trematodes rarely reported in

other parasitological surveys on European foxes (Eira et al., 2006), and only one with a ***Brachylaima*** species (Table 1).

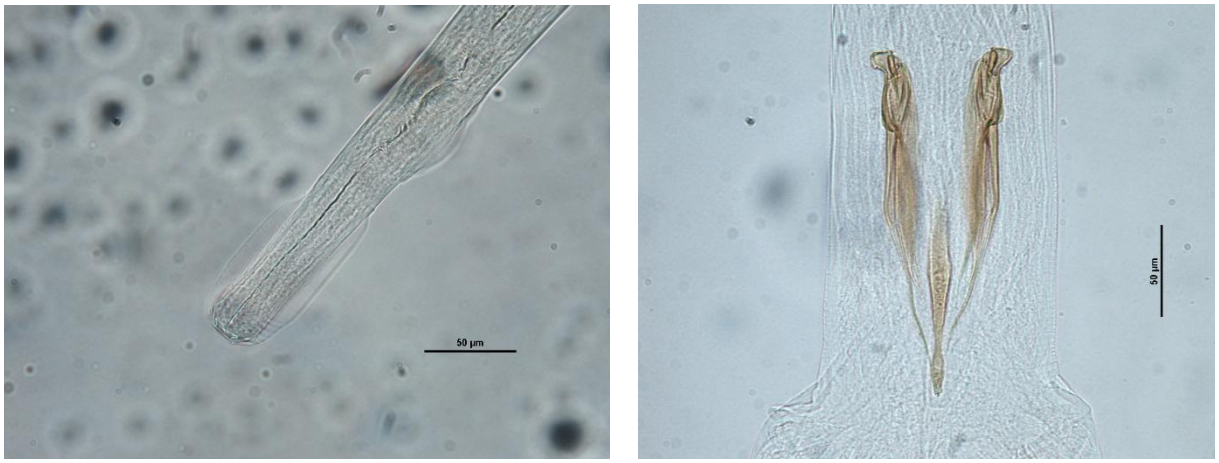
The present survey confirms that red foxes are definitive hosts for numerous helminth species in the Emilia-Romagna region, Italy. Only a few digenean trematodes were recovered, namely *A. alata*, *Brachylaima* sp. and *Phagicola* sp., all representing rare parasites for the Italian paeninsula. Among cestodes, *Mesocestoides* spp. and *T. polyacantha* were the most common species. Regarding nematodes, we report a high prevalence of *T. canis*, whereas lower prevalence values of *U. stenocephala*, *M. legerae* and particularly *P. affinis*, *T. leonina* and *T. cati* were found. Therefore, the role of the red fox as a reservoir host for some potential zoonotic parasites such as *T. canis* and *U. stenocephala* must be taken into consideration. Despite a considerable effort, we were not able to find any specimen of *E. granulosus*, a parasite that has been recently detected in scats of wolves from Emilia-Romagna eastern Apennines (Poglayen et al. 2017). This most likely confirms the fact that the red fox is not such a favourable definitive host for this tapeworm. In addition, despite the territory examined being an significant area for sheep rearing in the Emilia-Romagna region, we suppose, as already observed in a previous study (Fiocchi et al. 2016), that animals living here probably rely much more on small or medium-sized vertebrates (e.g. rodents, lagomorphs, birds, reptiles and amphibians) as sources of food rather that on carrions of domestic or wild ungulates.

Acknowledgements

We are grateful to Miriam Marrone for her kind collaboration in this research.



Cephalic end of *Joyeuxiella* sp. from a red fox



Cephalic end (on the left) and posterior part with spicules in detail (on the right) of the nematode *Molineus legerae* in a red fox

Rostellar hooks of *Taenia martis* from a red fox

REFERENCES

Al-Sabi MN, Chriél M, Jensen TH, Enemark HL. 2013. Endoparasites of the raccoon dog *(Nyctereutes procyonoides*) and the red fox (*Vulpes vulpes*) in Denmark 2009-2012 - A comparative study. International Journal for Parasitology. Parasites and Wildlife 2:144-151.

Al-Sabi MN, Halasa T, Kapel CM. 2014. Infections with cardiopulmonary and intestinal helminths and sarcoptic mange in red foxes from two different localities in Denmark. Acta Parasitologica 59:98-107.

Arru E, Garippa G, Fico R. 1988. Sulla presenza di *Echinococcus granulosus* nella volpe (*Vulpes vulpes*) e nel lupo (*Canis lupus*). Atti della Società Italiana di Scienze Veterinarie 42:927–929.

Bio C, Fagiolo A. 2004. Incidence of Hydatidosis in a sheep's slaughterhouse in the province of Arezzo, Italy. Parassitologia 46 (Suppl 1):28.

Borgsteede FH. 1984. Helminth parasites of wild foxes (*Vulpes vulpes* L.) in the Netherlands. Zeitschrift fur Parasitenkunde 70:281-285.

Brunet J, Pesson B, Chermette R, Regnard P, Grimm F, Deplazes P, Ferreira X, Sabou M, Pfaff AW, Abou-Bacar A, Candolfi E. 2014. First case of peritoneal cysticercosis in a non-human primate host (Macaca tonkeana) due to Taenia martis. Parasites and Vectors 7:422.

Carmena D, Cardona GA. 2014. Echinococcosis in wild carnivorous species: epidemiology, genotypic diversity, and implications for veterinary public health. Veterinary Parasitology 202:69-94.

Carvalho-Varela M, Marcos MVM. 1993. A helmintofauna da raposa (*Vulpes vulpes silacea* MILLER, 1907) em Portugal. Acta Parasitologica Portuguesa 1:73-79.

Cook BR. 1965. The epidemiology of *Echinococcus granulosus* in Great Britain. PhD Thesis, University of Liverpool, UK.

Dalimi A, Motamedi G, Hosseini M, Mohammadian B, Malaki H, Ghamari Z, Ghaffari Far F. 2002. Echinococcosis/hydatidosis in western Iran. Veterinary Parasitology 105:161-171.

Dalimi A, Sattari A, Motamedi G. 2006. A study on intestinal helminthes of dogs, foxes and jackals in the western part of Iran. Veterinary Parasitology 142:129-133.

Deblock S, Pétavy AF, Gilot B. 1988. Helminthes intestinaux du Renard commun (*Vulpes vulpes L.*) dans le Massif central (France). Canadian Journal of Zoology 66:1562-1569.

Di Cerbo AR, Manfredi MT, Trevisiol K, Bregoli M, Ferrari N, Pirinesi F, Bazzoli S. 2008. Intestinal helminth communities of the red fox (*Vulpes vulpes* L.) in the Italian Alps. Acta Parasitologica 53:302–311.

Eira C, Vingada J, Torres J, Miquel J. 2006. The helminth community of the red fox, *Vulpes vulpes*, in Dunas de Mira (Portugal) and its effect on host condition. Wildlife Biology in Practice 2:26–36.

El-Shehabi FS, Abdel-Hafez SK, Kamhawi SA. 1999. Prevalence of intestinal helminths of dogs and foxes from Jordan. Parasitology Research 85:928-934.

Euzeby J. 1982. Diagnostic expérimental des helminthoses animales (animaux domestiques-animaux de laboratoire-primates). Travaux pratiques d'Helminthologie Vétérinaire. Livre 2. Paris: Informations Techniques des Services Vétérinaires, Ministére de l'Agriculture, pp.99-117.

Fiocchi A, Gustinelli A, Gelmini L, Rugna G, Renzi M, Fontana MC, Poglayen G. 2016. Helminth parasites of the red fox (*Vulpes vulpes* L., 1758) and the wolf (*Canis lupus italicus altobello*, 1921) in Emilia-Romagna, Italy. Italian Journal of Zoology 83:503-513.

Gicik Y, Kara M, Sari B, Kiliç K, Arslan MÖ. 2009. Intestinal parasites of red foxes (*Vulpes vulpes*) and their zoonotic importance for humans in Kars Province. Kafkas Üniversitesi Veteriner Fakultesi Dergisi 15:135-140.

Gortázar C, Villafuerte R, Lucientes J, Fernández-de-Luco D. 1998. Habitat related differences in helminth parasites of red foxes in the Ebro valley. Veterinary Parasitology 80:75-81.

Grosso G, Gruttadauria S, Biondi A, Marventano S, Mistretta A. 2012. Worldwide epidemiology of liver hydatidosis including the Mediterranean area. World Journal of Gastroenterology 18: 1425-1437.

Guberti V, Poglayen G. 1991. Parasitic zoonoses: survey in foxes (*Vulpes vulpes*) in the Northern Apennines. Hystrix 3:167–173.

Guberti V, Bolognini M, Lanfranchi P, Battelli G. 2004. *Echinococcus granulosus* in the wolf in Italy. Parassitologia 46:425-427.

Hackett F, Walters TMH. 1980. Helminths of the red fox in mid-Wales. Veterinary Parasitology 7:181-184.

Iori A, Costantini R, Cancrini G. 1990. Parassiti di volpi (*Vulpe vulpes*) provenienti da alcune regioni italiane. Parassitologia 32:153-154.

Jenkins DJ, Morris B. 2003. Echinococcus granulosus in wildlife in and around the Kosciuszko National Park, south-eastern Australia. Australian Veterinary Journal 81:81-85.

Jenkins DJ. 2006. Echinococcus granulosus in Australia, widespread and doing well! Parasitology International 55 Suppl:S203-206.

Khalil LF, Jones A, Bray RA. 1994. Keys to the cestode parasites of vertebrates. Wallingford, Oxon: Cab international, 768 pp.

Lahmar S, Boufana BS, Lahmar S, Inoubli S, Guadraoui M, Dhibi M, Bradshaw H, Craig PS. 2009. Echinococcus in the wild carnivores and stray dogs of northern Tunisia: the results of a pilot survey. Annals of Tropical Medicine and Parasitology 103:323-331.

Lahmar S, Boufana B, Ben Boubaker S, Landolsi F. 2014. Intestinal helminths of golden jackals and red foxes from Tunisia. Veterinary Parasitology 204:297-303. .

Leoni A, Garippa G, Masala S. 1986. Osservazioni sull'elmintofauna della volpe (*Vulpes vulpes*) in Sardegna. Parassitologia 28:268–269.

Loos-Frank B, Zeyhle E. 1982. The intestinal helminths of the red fox and some other carnivores in southwest Germany. Zeitschrift für Parasitenkunde 67:99-113.

Magi M, Guardone L, Mignone W, Prati MC, Macchioni F. 2016. Intestinal helminths of red foxes (*Vulpes vulpes*) in north-west Italy. Helminthologia 53:31-38.

Manfredi MT, Giacometti A, Fraquelli C, Piccolo G. 2003. Studio della popolazione elmintica in volpi (*Vulpes vulpes*) del Trentino Alto-Adige. Journal of Mountain Ecology 7 (Suppl):261-263.

Martínez-Carrasco C, Ruiz de Ybánez MR, Sagarminaga JL, Garijo MM, Moreno F, Acosta I, Hernández S, Alonso FD. 2007. Parasites of the red fox (*Vulpes vulpes* Linnaeus, 1758) in Murcia, southeast Spain. Revue de Médicine Vétérinaire 158:331–335.

Meshgi B, Eslami A, Bahonar AR, Kharrazian-Moghadam M, Gerami-Sadeghian A. 2009. Prevalence of parasitic infections in the red fox (Vulpes vulpes) and golden Jackal (Canis aureus) in Iran. Iranian Journal of Veterinary Research 10:387-391.

Mimioğlu MM, Güralp N, Tolgay N, Sayin F. 1965. Ankara civarinda tilkilerde (*Vulpes vulpes*) bulduğumuz helmintler. Ankara Üniversitesi Veteriner Fakültesi Dergisi 12:164-190.

Mohammad MK, Jasim MK, Al-Moussawi AA. 2003. The parasitic fauna of the red fox *Vulpes vulpes* (L., 1758) in Iraq with some notes on its biology and ecology. Bulletin of the Iraq Natural History Museum 10:59-76.

Möhl K, Grosse K, Hamedy A, Wüste T, Kabelitz P, Lücker E. 2009. Biology of *Alaria* spp. and human exposition risk to *Alaria mesocercariae*-a review. Parasitology Research 105:1–15.

.

Nabavi R, Manouchehri Naeini K, Zebardast N, Hashemi H. 2014. Epidemiological study of gastrointestinal helminthes of canids in Chaharmahal and Bakhtiari Province of Iran. Iranian Journal of Parasitology 9:276-281.

Papadopoulos H, Himonas C, Papazahariadou M, Antoniadou-Sotiriadou K. 1997. Helminths of foxes and other wild carnivores from rural areas in Greece. Journal of Helminthology 71:227–231.

Pétavy AF, Deblock S. 1980. Helminths of the common fox (*Vulpes vulpes* L.) from the Massif Central (France). Annales de Parasitologie Humaine et Comparee 55:379-391.

Poglayen G, Gori F, Morandi B, Galuppi R, Fabbri E, Caniglia R, Milanesi P, Galaverni M, Randi E, Marchesi B, Deplazes P**.** 2017. Italian wolves (Canis lupus italicus Altobello, 1921) and molecular detection of taeniids in the Foreste Casentinesi National Park, Northern Italian Apennines. International Journal of Parasitology Parasites and Wildlife 6:1-7.

Richards DT, Harris S, Lewis JW. 1995. Epidemiological studies on intestinal helminth parasites of rural and urban red foxes (*Vulpes vulpes*) in the United Kingdom. Veterinary Parasitology 59:39–51.

Sato H, Inaba T, Ihama Y, Kamiya H. 1999. Parasitological survey on wild carnivora in north-western Tohoku, Japan. Journal of Veterinary Medical Science 61:1023-1026.

Schöffel I, Schein E, Wittstadt U, Hentsche J. 1991. Parasite fauna of red foxes in Berlin (West). Berliner und Münchener Tierärztliche Wochenschrift 104:153–157.

Segovia JM, Torres J, Miquel J. 2004. Helminth parasites of the red fox (*Vulpes vulpes* L., 1758) in the Iberian Peninsula: an ecological study. Acta Parasitologica 49:67–79.

Shimalov VV, Shimalov VT. 2003. Helminth fauna of the red fox (*Vulpes vulpes* Linnaeus, 1758) in southern Belarus. Parasitology Research 89:77-78.

Sikó Barabási S, Fok E, Gubányi A, Mészáros F, Cozma V. 2010. Helminth fauna of the small intestine in the European red fox, *Vulpes vulpes*, with notes on the morphological identification of *Echinococcus multilocularis.* Scientia Parasitologica 11:141–151.

Soldati G, Pavesi M, Canestri-Trotti G, Cocchi MG, Gaiardi S, Morganti L, Prosperi S, Sanguinetti V, Stanzani F. 1976. Research on infections and parasitic agents in foxes of the Modenese Apennines. Rivista di Parassitologia 37:329–332.

Suchentrunk F, Sattmann H. 1994. Prevalence of intestinal helminths in Austrian red foxes (*Vulpes vulpes* L.) (Cestoda, Nematoda) Annalen des Naturhistorischen Museums in Wien 96B:29–38.

Széll Z, Sréter T, Varga I. 2004. Prevalence, veterinary and public health aspects of gastrointestinal parasites in red foxes (*Vulpes vulpes*) in Hungary. Magyar Allatorvosok Lapja 126:293–299.

Vergles Rataj A, Posedi J, Zele D, Vengušt G. 2013. Intestinal parasites of the red fox (*Vulpes vulpes*) in Slovenia. Acta Veterinaria Hungarica 61:454–462.

Vervaeke M, Dorny P, De Bruyn L, Vercammen F, Jordaens K, Van Den Berge K, Verhagen R. 2005. A survey of intestinal helminthes of red foxes (*Vulpes vulpes*) in northern Belgium. Acta parasitologica 50:221–227

Williams BM. 1976. The intestinal parasites of the red fox in South West Wales. British Veterinary Journal 132:309–312.

Zanini F, Laferrara M, Bitsch M, Pérez H, Elissondo MC. 2006. Epidemiological studies on intestinal helminth parasites of the patagonian grey fox (Pseudalopex griseus) in Tierra del Fuego, Patagonia Argentina. Veterinary Parasitology 136:329-334.

3. *ALARIA ALATA*, A REDISCOVERED PARASITE IN NORTHERN ITALY.

ALFREDO FIOCCHI[1], ANDREA GUSTINELLI[1], LUCA GELMINI[2], GIANLUCA RUGNA[2], MARIA RENZI[3], MARIA CRISTINA FONTANA[3], GIOVANNI TOSI[4], MARIA LETIZIA FIORAVANTI[1], GIOVANNI POGLAYEN[1]

[1]*Department of Veterinary Medical Sciences, Alma Mater Studiorum University of Bologna, Ozzano Emilia (BO), Italy*

[2]*Lombardy and Emilia Romagna Experimental Zooprophylactic Institute, Modena, Italy*

[3]*Lombardy and Emilia Romagna Experimental Zooprophylactic Institute, Bologna, Italy*

[4]*Lombardy and Emilia Romagna Experimental Zooprophylactic Institute, Forlì, Italy*

TO BE PUBLISHED IN A PEER-REVIEWED JOURNAL

ABSTRACT *Alaria alata* is an intestinal helminth of carnivores widespread in Eurasia and potentially zoonotic. This trematode has been found rarely in Italian wildlife and most of the reports date back to the XIX century. Six infected red foxes (*Vulpes vulpes*) were found in the Emilia-Romagna region, Northern Italy. Most of them lived in lowlands rich in wet areas, a suitable environment for the life cycle of the parasite. The prevalence in the fox population examined was low and low were the parasitic burdens found. Further studies are still needed to better determine the prevalence and distribution of this trematode in wild carnivores of our country.

Keywords: Red fox, wolf, *Alaria alata*, Emilia-Romagna, Italy.

INTRODUCTION

Alaria alata (Goeze, 1782) (Figure 1) is a digenean trematode of the family *Diplostomatidae*, which has been reported in wild red foxes (*Vulpes vulpes*) from several European and extra-European countries (Loos-Frank et al. 1982; Sato et al. 1999; Möhl et al. 2009; Sikó Barabási et al. 2010; Al-Sabi et al. 2014). According to literature, its prevalence values in the fox populations examined range from 0.1% (Loos-Frank et al. 1982) to 94.8% (Bružinskaité-Schmidhalter et al. 2012). The presence of *A. alata* in Italy has been reported rarely. Gestaldi (1854) first described the larval stage of the trematode in frogs ("*Distoma tetracystis*"). A few years later Molin (1861) found the parasite in a fox near Padua (Veneto region). More recently Ferroglio (2012) described the infection in a dog from Aosta Valley. We did not find any other reports of this trematode in several parasitological surveys on Italian foxes (Soldati et al. 1976; Rossi et al. 1983; Poglayen et al. 1985; Leoni et al. 1986; Iori et al. 1990; Guberti and Poglayen 1991; Capelli et al. 2003; Manfredi et al. 2003; Di Cerbo et al. 2008; Magi et al. 2009, 2016). The complex life cycle of *A. alata* requires a freshwater snail as the first intermediate host and an amphibian as the second one (Figure 2). Reptiles, rodents, wild boars and other vertebrates can act as paratenic hosts after feeding on infected amphibians (Wolfe et al. 2001; Möhl et al. 2009; Riehn et al. 2014). The spectrum of paratenic hosts is potentially broad, they may accumulate the mesocercariae with each transition and they may also serve to pass over the infection from the aquatic to the terrestrial environment (Möhl et al. 2009). Definitive hosts, usually members of the family Canidae, become infected after ingesting mesocercariae contained in amphibians or paratenic hosts (Figure 2). The young flukes migrate through the gut and other viscera, change into the metacercarial stage in the lungs and, after being swallowed, reach the small intestine (Möhl et al. 2009). Moreover mesocercariae might undergo hypobiosis in the definitive carnivore host, then reactivate around parturition and be transmitted to the offspring by the milk (Shoop 1994). *A. alata* is also a potential zoonotic agent (Odening 1961). Humans could acquire the infection after eating undercooked frog legs or raw game meat containing mesocercariae (Murphy et al. 2012). Clinical signs of human larval alariosis, so far reported only in North America and caused by american *Alaria* species, include low-grade respiratory and cutaneous symptoms, diffuse unilateral sub-acute neuroretinitis and anaphylactic shock (Möhl et al. 2009).

Figure 1. Adult specimen of *Alaria alata*.

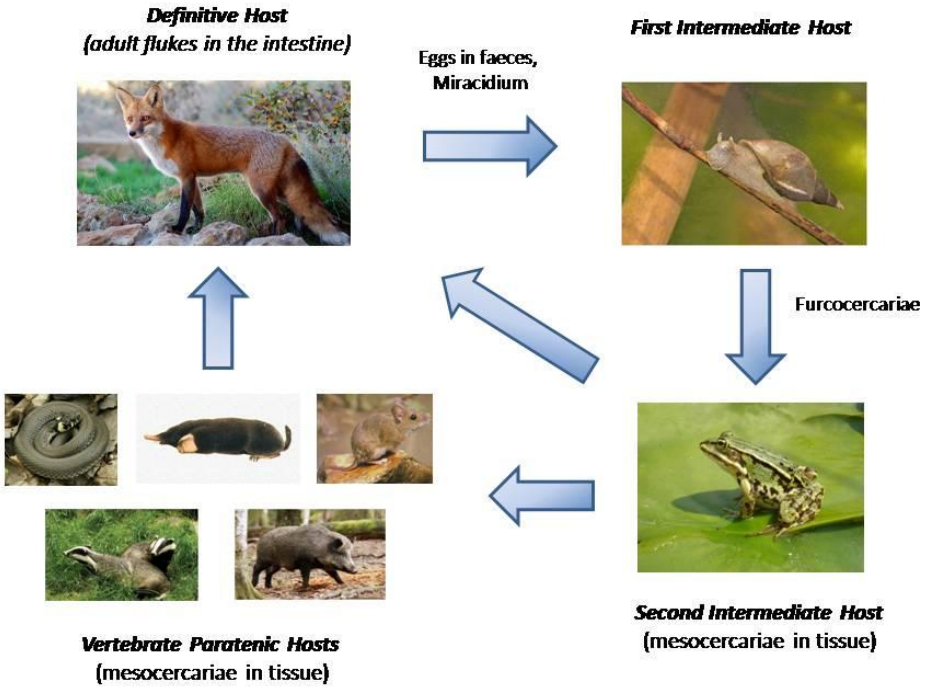


Figure 2. Life cicle of *Alaria alata*.

MATERIALS AND METHODS

Between January 2013 and June 2015 we analysed 373 wild canids (370 red foxes and three wolves) collected in official culling campaigns or found dead in the provinces of Modena, Bologna, Forlì-Cesena and Rimini, Emilia-Romagna region (Figure 3). Eighty-four animals (22.5%) came from lowland territories, 289 (77.5%) from hilly or mountainous territories (Table 1). The stomach and the intestine were removed during necropsy and intestinal parasites were collected using the sedimentation and counting technique (SCT). Recovered trematodes were counted and stored in 70% alcohol for morphological examination. Eight parasites were clarified by Amman's lactophenol and glycerol, then two stained with boracic carmine (Pritchard and Kruse 1982) and observed at light microscope. Morphometric analysis has been carried out by a Leica DMLS light microscope, digital camera Nikon DS-Fi1 and imaging analysis softwtare NIS-elements D 3.22©. All the measurements are expressed in µm.

RESULTS AND DISCUSSION

Morphological analysis of the trematodes from the six positive foxes confirmed our presumptive identification of *A. alata* showing slight differences with the description reported by Mohl et al. (2009) (Table 2). The prevalence of infection with *A. alata* was low in foxes (1.62%), with a mean intensity of 2.6 (Table 1). Considering foxes from Modena and Bologna, three out of 57 were found positive for the parasite (prevalence 5.26%), one of them harboring 4 trematodes and the other two 3 trematodes each (mean intensity 3.3) (Table 1). These animals lived in a lowland territory, known as "Bassa Pianura" (Figure 3, blue stars), rich in humid habitats and channels, a suitable environment for the life cycle development of *A. alata.* In the group of foxes from Forlì-Cesena and Rimini, only three animals out of 313 were infected (prevalence 0.95%), harboring respectively 4 and the other two 1 trematode each (mean intensity 2) (Table 1). They were killed near the city of Forlì and in nearby low hills (Figure 3, red stars). All the wolves examined were negative.

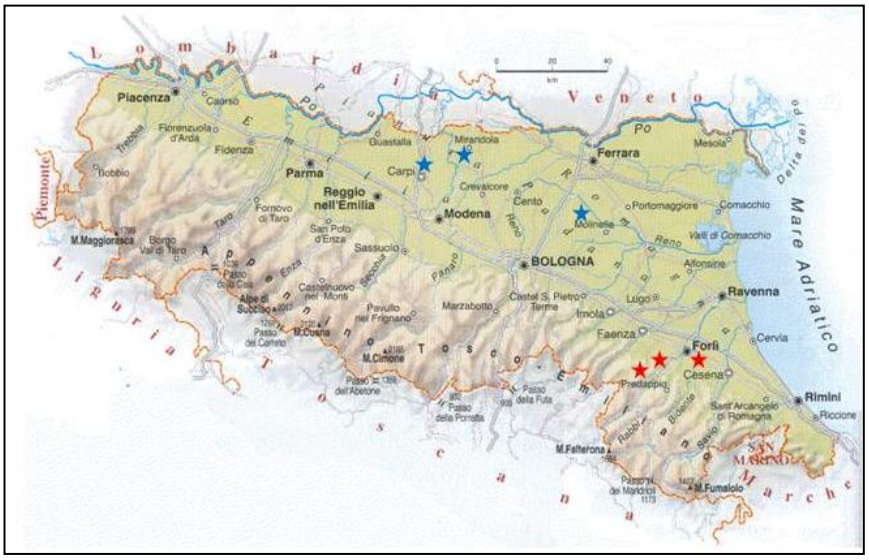


Figure 3. Emilia-Romagna region. The blue and the red stars indicate where the *Alaria*-infected foxes lived.

Table 1. Territory of origin of wild canids examined in this study, prevalence (P%) and mean intensity (MI) of *A. alata*.

Provinces of origin	**N° Animals**	**Lowlands**	**Hill, Mountain**	***Alaria* positive foxes (P%)**	***Alaria* MI**
Modena and Bologna	60 (57 foxes, 3 wolves)	33	27 (24 foxes, 3 wolves)	3/57 (5.26)	3.3
Forlì-Cesena and Rimini	313 (foxes)	51	262	3/313 (0.95)	2
Total	373	84	289	6/370 (1.62)	2.6

Table 2 Measurements (expressed in µm) of *Alaria alata*.

	MIN	**MAX**	**MEAN**
Body Lenght	2049,33	3723,21	3198,02
Body Width	619,93	1630,44	1110,77
Oral Sucker Lenght	62,52	119,62	95,26
Oral Sucker Width	61,04	135,49	98,64
Ventral Sucker Lenght	71,21	108,40	89,52
Ventral Sucker Width	57,66	142,64	117,12
Pharynx Lenght	72,73	147,32	123,04
Pharynx Width	72,19	124,90	94,06
Eggs Lenght	98,59	115,14	109,215
Eggs Width	71,57	86,61	78,095

Alaria alata is a digenean trematode that has been reported rarely in Italy. Our research demonstrates for the first time the presence of this potential zoonotic helminth in the Emilia-Romagna region and stresses the importance of water-rich territories in its life-cycle. Other studies such as those of Loos-Frank et al. (1982) in Germany, Criado-Fernelio et al. (2000) in Spain, Eira et al. (2006) in Portugal, Murphy et al. (2012) in Ireland confirm that the prevalence of this trematode is higher among wildlife hosts in wetland habitats. Even though we examined animals living at different altitudes, infected foxes were found mainly in lowland territories. In Emilia-Romagna these plain lands are rich in channels and wetlands. The difference in the *Alaria* prevalence between the two groups of animals may be attributed to the fact that a larger number of foxes in the first group (Modena, Bologna provinces) were killed or found dead in plains. The parasite's mean intensity was very low. Other authors in Europe (Schöffel et al., 1991; Murphy et al., 2012) noted that the majority of infected foxes had a small parasitic burden. We don't know if *A. alata* is endemic to Padan Plain or should be considered an exotic parasite. Historical data (Gestaldi, 1854; Molin, 1861) and the wide diffusion of potential second intermediate hosts (e.g. *Rana esculenta, Bufo bufo*) and paratenic hosts (e.g. *Natrix natrix, Talpa europaea*) in Emilia-Romagna lowlands suggest the first hypothesis. *A. alata's* presence could be alternatively

the consequence of a past importation of game animals (e.g. wild boars) from Eastern Europe. However no historical reports exist regarding the finding of mesocercariae in wild boar meat, differently from other European countries (Portier et al., 2011; Riehn et al., 2012, 2014; Paulsen et al., 2013; Szell et al., 2014). Further studies on a higher number of specimens are necessary both to better determine the prevalence and the distribution of the parasite in our country and to exclude the presence of other *Alaria* species. The importation of bullfrogs (*Lithobates catasbeianus*), suitable host for *Alaria mustelae* and *Alaria spp*. (Möhl et al., 2009), from extra-European countries and their present diffusion into the wild, could actually lead to the diffusion of exotic parasites.

REFERENCES

Al-Sabi MN, Halasa T, Kapel CM. 2014. Infections with cardiopulmonary and intestinal helminths and sarcoptic mange in red foxes from two different localities in Denmark. Acta Parasitologica 59:98-107.

Bružinskaitė-Schmidhalter R, Šarkūnas M, Malakauskas A, Mathis A, Torgerson PR, Deplazes P. 2012. Helminths of red foxes (*Vulpes vulpes*) and raccoon dogs (*Nyctereutes procyonoides*) in Lithuania. Parasitology 139:120–127.

Capelli G, Stancampiano L, Magi M, Poglayen G, Guberti V. 2003. Diversità delle comunità parassitarie intestinali in tre popolazioni di volpi. Journal of Mountain Ecology 7:199–205.

Criado-Fornelio A, Gutierrez-Garcia L, Rodriguez-Caabeiro F, Reus-Garcia E, Roldan-Soriano MA, Diaz-Sanchez MA. 2000. A parasitological survey of wild red foxes (*Vulpes vulpes*) from the province of Guadalajara, Spain. Veterinary Parasitology 92:245–251.

Di Cerbo AR, Manfredi MT, Trevisiol K, Bregoli M, Ferrari N, Pirinesi F, Bazzoli S. 2008. Intestinal helminth communities of the red fox (*Vulpes vulpes* L.) in the Italian Alps. Acta Parasitologica 53:302–311.

Eira C, Vingada J, Torres J, Miquel J. 2006. The helminth community of the red fox, *Vulpes vulpes*, in Dunas de Mira (Portugal) and its effect on host condition. Wildlife Biology in Practice 2:26–36.

Ferroglio E, Trisciuoglio A, Lazzaron S, Filippetto E, Zanet S. 2012. Clinical and molecular findings of *Alaria alata* infection in dog from Aosta Valley, Italy. Mappe Parassitologiche, XXVII Congresso Nazionale Società Italiana di Parassitologia 18:160.

Gestaldi B. 1854. Cenni sopra alcuni nuovi Elminti della *Rana esculenta* con nuove osservazioni sul *Codonocephalus mutabilis* (Diesing). Tesi per aggregazione al Collegio della Facoltà delle Scienze Fisiche e Mathematiche nella R. Università di Torino. pp. 25–36.

Guberti V, Poglayen G. 1991. Parasitic zoonoses: survey in foxes (*Vulpes vulpes*) in the Northern Apennines. Hystrix 3:167–173.

Iori A, Costantini R, Cancrini G. 1990. Parassiti di volpi (*Vulpes vulpes*) provenienti da alcune regioni italiane. Parassitologia 32:153–154.

Leoni A, Garippa G, Masala S. 1986. Osservazioni sull'elmintofauna della volpe (*Vulpes vulpes*) in Sardegna. Parassitologia 28:268–269.

Loos-Frank B, Zeyhle E. 1982. The intestinal helminths of the red fox and some other carnivores in southwest Germany. Zeitschrift fur Parasitenkunde 67:99–113.

Magi M, Guardone L, Mignone W, Prati MC, Macchioni F. 2016. Intestinal helminths of red foxes (*Vulpes vulpes*) in north-west Italy. Helminthologia 53:31–38.

Magi M, Macchioni F, Dell'Omodarme M, Prati MC, Calderini P, Gabrielli S, Iori A, Cancrini G. 2009. Endoparasites of red fox (*Vulpes vulpes*) in Central Italy. Journal of Wildlife Diseases 45:881–885.

Manfredi MT, Giacometti A, Fraquelli C, Piccolo G. 2003. Studio della popolazione elmintica in volpi (*Vulpes vulpes*) del Trentino Alto-Adige. Journal of Mountain Ecology 7:261–263.

Möhl K, Grosse K, Hamedy A, Wüste T, Kabelitz P, Lücker E. 2009. Biology of *Alaria* spp. and human exposition risk to *Alaria mesocercariae*-a review. Parasitology Research 105:1–15.

Molin R. 1861. Prodromus Faunae Helminthologicae Venetae. Wien: Kaiserlich-Königlichen hof- und staatsdruckerei. 193 pp.

Murphy TM, O'Connell J, Berzano M, Dold C, Keegan JD, McCann A, Murphy D, Holden NM. 2012. The prevalence and distribution of *Alaria alata*, a potential zoonotic parasite, in foxes in Ireland. Parasitology Research 111:283–290.

Odening K. 1961 Der “Dunckersche Muskelegel” kann experimentell auf den Affen übertragen werden. Monatshefte für Veterinärmedizin 16:395-399.

Paulsen P, Forejtek P, Hutarova Z, Vodnansky M. 2013. *Alaria alata* mesocercariae in wild boar (*Sus scrofa*, Linnaeus, 1758) in south regions of the Czech Republic. Veterinary Parasitology 197:384-387.

Poglayen G, Guberti V, Leoni B. 1985. Parasites present in foxes (*Vulpes vulpes*) of the province of Forli. Parassitologia 27:303–311.

Portier J, Jouet D, Ferté H, Gibout O, Heckmann A, Boireau P, Vallée I. 2011. New data in France on the trematode *Alaria alata* (Goeze, 1792) obtained during *Trichinella* inspections. Parasite 18:271-275.

Pritchard MH, Kruse GO. 1982. The collection and preservation of animal parasites. Technical Bulletin No. 1. The Harold W. Manter Laboratory, University of Nebraska Press. 141 pp.

Riehn K, Hamedy A, Grosse K, Wüste T, Lücker E. 2012. *Alaria alata* in wild boars (*Sus scrofa*, Linnaeus, 1758) in the eastern parts of Germany. Parasitology Research 111:1857-1861.

Riehn K, Lalkovski N, Hamedy A, Lücker E. 2014. First detection of *Alaria alata* mesocercariae in wild boars (*Sus scrofa* Linnaeus, 1758) from Bulgaria. Journal of Helminthology 88:247-249.

Rossi L, Iori A, Cancrini G. 1983. Osservazioni sulla fauna parassitaria della popolazione di volpi presente nel Parco Regionale “La Mandria”. Parassitologia 25:340–343.

Sato H, Inaba T, Ihama Y, Kamiya H. 1999. Parasitological survey on wild carnivora in north-western Tohoku, Japan. The Journal of Veterinary Medical Science 61:1023-1026.

Schöffel I, Schein E, Wittstadt U, Hentsche J. 1991. Parasite fauna of red foxes in Berlin (West). Berliner und Münchener Tierärztliche Wochenschrift 104:153–157.

Shoop W. 1994. Vertical transmission in the trematoda. Journal of the Helminthological Society of Washington 61:153-161.

Sikó Barabási S, Fok E, Gubányi A, Mészáros F, Cozma V. 2010. Helminth fauna of the small intestine in the European red fox, *Vulpes vulpes*, with notes on the morphological identification of *Echinococcus multilocularis.* Scientia Parasitologica 11:141–151.

Soldati G, Pavesi M, Canestri-Trotti G, Cocchi MG, Gaiardi S, Morganti L, Prosperi S, Sanguinetti V, Stanzani F. 1976. Research on infections and parasitic agents in foxes of the Modenese Apennines. Rivista di Parassitologia 37:329–332.

Széll Z, Tolnai Z, Sréter T. 2013. Environmental determinants of the spatial distribution of *Alaria alata* in Hungary. Veterinary Parasitology 198:116–121.

Wolfe A, Hogan S, Maguire D, Fitzpatrick C, Vaughan L, Wall D, Hayden TJ, Mulcahy G. 2001. Red foxes (*Vulpes vulpes*) in Ireland as hosts for parasites of potential zoonotic and veterinary significance. The Veterinary Record 149:759-763.

Partial results of this research were presented at the XXVIII SOIPA (Italian Society of Parasitology) national congress, held in Rome in June 2014.

FIRST REPORT OF *ALARIA ALATA* IN WILD RED FOXES (*VULPES VULPES*) FROM EMILIA ROMAGNA REGION , ITALY

Fiocchi A., Gustinelli A., Poglayen G.

Department of Veterinary Medical Sciences Alma Mater Studiorum University of Bologna, Bologna, Italy

INTRODUCTION:

Alaria alata is a digenean trematode of the family *Diplostomatidae*, which has been reported in wild red foxes (*Vulpes vulpes*) from several European countries (Loos-Frank et al., Z Parasitenkd. 1982, 67(1):99-113.; Möhl et al., Parasitol Res. 2009, 105(1): 1-15; Barabási et al., Sci Parasitol. 2010, 11(3):141-151). According to literature, its prevalence values in the fox populations examined ranges from 0.1 % (Loos-Frank et al., 1982, l.c.) to 94.8 % (Bružinskaité-Schmidhalter et al., Parasitology 2012, 139(1):120-7). The presence of *A. alata* in Italy has been reported rarely. Molin (Prodromus Faunae Helminthologicae Venetae, 1854) found the parasite in a fox near Padua (Veneto Region) in the XIX century. More recently Ferroglio (Mappe Parassitol., 2012, 18:160) described an infection in a dog from Valle d'Aosta. We didn't find any reports of this trematode from Italy in several parasitological surveys (Soldati et al., Riv Parassitol 1976, 37: 329-332; Poglayen et al., 1985, Parassitologia, 27(3): 303-11.; Capelli et al., J. Mt. Ecol. 2003, 7 (Suppl.): 199-205; Di Cerbo et al., Acta Parasitologica 2008, 53 (3): 302-311; Magi et al., J. Wildl. Dis. 2009, 45(3): 881-885) with the exception of some photos on an atlas of wildlife pathology (Alborali et al., Collana Fond. In. Zooprof. Brescia, 2012, 91: 566).

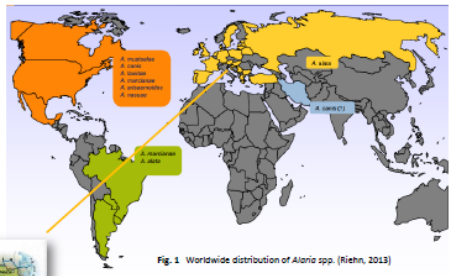


Fig. 1 Worldwide distribution of *Alaria* spp. (Riehn, 2013)

Fig. 2 Emilia-Romagna Region. The red circle and the blue circle indicate where the *Alaria* infected foxes lived.

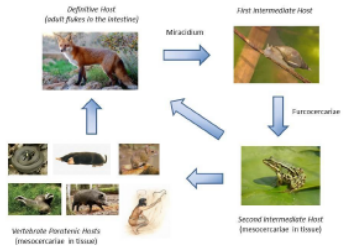


Fig. 3 Life cycle of *Alaria alata*

LYFE CYCLE: *A. alata*'s complex life cycle requires a freshwater snail as first intermediate host and an amphibian as second intermediate host. Reptiles, rodents, wild boars and other vertebrates can act as paratenic hosts after feeding on infected amphibians (Möhl et al., 2009, l.c.). Definitive hosts, usually members of the family *Canidae*, become infected after ingesting mesocercariae contained in amphibians or paratenic hosts (Fig. 3). The young flukes migrate through the gut and other viscera, change into the metacercarial stage in the lungs and, after being swallowed, reach the small intestine (Möhl et al., 2009, l.c.). *A. alata* is also a potential zoonotic agent. Humans can acquire infection after eating undercooked frog legs or raw game meat containing mesocercariae (Murphy et al., Parasitol Res. 2012, 111(1):283-290). Clinical signs of human larval alariosis include low-grade respiratory and cutaneous symptoms, diffuse unilateral sub-acute neuroretinitis and anaphylactic shock (Möhl et al., 2009, l.c.).

MATERIALS AND METHODS:

Between February 2013 and March 2014, we analyzed 29 carnivores (28 red foxes and one wolf) collected from hunters or found dead in the Province of Modena and Bologna (group A). The stomach and the gut were removed during necropsy and parasites were collected using SCT ("Sedimentation and Counting Technique") according to standard protocols. In the same period we analyzed the duodenal tract of 80 red foxes from the Province of Forlì-Cesena and Rimini (group B) during a survey for *Echinococcus granulosus*. Recovered trematodes were counted, stored in 70 % alcohol and stained with carmine.

RESULTS:

In group A only two foxes out of 28 were found positive for *Alaria alata* (prevalence *7.1 %*), harboring respectively 3 and 4 trematodes. These two animals lived in a lowland territory, known as "Bassa Pianura"(Fig. 2, red circle), rich in wet areas, channels and rice fields, a suitable environment for the life cycle development of *A. alata* (Fig. 5). In group B, only two foxes out of 80 were infected (prevalence *2.5 %*), harboring respectively 4 and 1 trematodes. They were killed near the city of Forlì (Fig. 2, blue circle). Morphological analysis of the trematodes from the four foxes confirmed our presumptive identification showing measures agreeing with the description reported by Mohl et al. (2009) (Fig. 4).

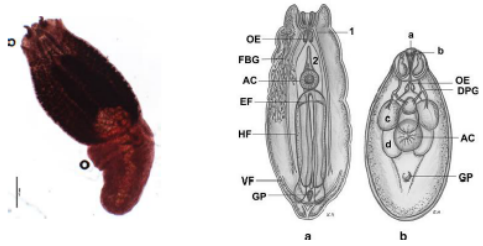


Fig. 4 Adult specimen of *A. alata* compared to illustrations by Mohl et al. (2009)

Fully developed adult (a) and mesocercarial (b) stage of *Alaria* spp. 1 - remnant of penetration gland duct, 2 - caecum (elonged), OE oesophagus, FBG forebody glands and their ducts, AC acetabulum, EF edge of fold over anterior and of holdfast organ, HF holdfast organ, VF ventral lip of spathiform forebody, GP genital primordium, a oral opening and oral sucker, b gland cells, c penetration glands, d caecum, DPG duct of penetration gland

Fig. 5. Wetlands in the lowlands in the Province of Modena

Fig. 6 Lithobates catasbeianus

CONCLUSIONS: *Alaria alata* is a digenean trematode that has been reported rarely in Italy. Despite a low number of carnivores were analyzed, our report demonstrate for the first time the presence of this zoonotic helminth in our Region and stress the importance of water-rich territories in its life-cycle. We don't know if *A. alata* is endemic to Padan Plain or should be considered an exotic parasite. An hypothesis is that its presence could be a consequence of the importation of game animals (e.g. wild boars) from Eastern Europe. Further studies on a higher number of specimens are also necessary to exclude the presence of other *Alaria* species since the importation of bullfrogs (*Lithobates catasbeianus*)(Fig. 6), suitable host for *Alaria mustelae* and *Alaria spp*. (Möhl et al., 2009, l.c.), from extra-European countries, and their diffusion into the wild, could lead to the diffusion of exotic parasites.

ACKNOWLEDGEMENTS:
The Authors wish to thank the Veterinary Personnel of the Sections of Modena, Bologna and Forlì of I.Z.S.L.E.R. (Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia-Romagna) for kindly providing the biological material for this parasitological survey.

4. SURVEY OF PULMONARY HELMINTH PARASITES OF THE RED FOX (*VULPES VULPES* L., 1758) IN EMILIA-ROMAGNA, ITALY.

BENEDETTO MORANDI[1], SOFIA BERTASO[1], ALFREDO FIOCCHI[1], ANDREA GUSTINELLI[1], ROBERTA GALUPPI[1], GIOVANNI TOSI[2], ANTONIO VARCASIA[3], ANNA PAOLA PIPIA[3], GIOVANNI POGLAYEN[1]

[1]*Department of Veterinary Medical Sciences, Alma Mater Studiorum University of Bologna, Ozzano Emilia (BO), Italy*

[2]*Lombardy and Emilia-Romagna Experimental Zooprophylactic Institute (IZSLER), Forlì, Italy*

[3]*Department of Veterinary Medicine, University of Sassari, Sassari, Italy*

TO BE PUBLISHED IN A PEER-REVIEWED JOURNAL

ABSTRACT In the period 2014-2015, 90 respiratory tracts of red foxes (*Vulpes vulpes*) from the Emilia-Romagna region, Italy, were analyzed. The Baermann method was used to recover helminth parasites which were found in 30% of the animals. Multiple infections were found in 5.5% of the animals. Three helminth species were described, *Eucoleus aerophilus* (1.1%), *Filaroides hirthi* (6.6%) and *Crenosoma vulpis* (27.7%). Results are compared with those of other surveys on pulmonary helminths of red foxes carried out in Italy and other European countries.

Key words: red fox, *Vulpes vulpes*, pulmonary helminths, Baermann method, Emilia-Romagna, Italy.

INTRODUCTION

The red fox (*Vulpes vulpes,* Linnaeus 1758) belongs to the Order *Carnivora*, Family *Canidae*. It is a common species in Italy and also in the Emilia-Romagna region, occupying a wide range of habitats from urbanized lowlands to hilly and mountainous territories (Spagnesi and De Marinis 2002). It's an opportunistic predator, whose varied diet includes lagomorphs, small and medium-sized rodents, domestic and wild birds, reptiles, invertebrates but also fruit and vegetables especially in summer and autumn. If available they feed on rubbish, carrion of domestic and wild ungulates, acquatic animals such as fish and amphibians. Prey availability and geographical setting may influence the parasitic fauna of these animals, including the helminthofauna of the respiratory tract (Segovia et al. 2004; Martínez-Carrasco et al. 2007). The aim of the present study was to analyse the pulmonary helminthic fauna of red foxes from Emilia-Romagna, northern Italy. Results are compared with those of other surveys carried out in other italian regions and european countries.

MATERIALS AND METHODS

The study areas were the provinces of Forlì-Cesena, Ravenna and Rimini in Emilia Romagna, northern Italy. The northern part of these provinces, which is the more populated and industrialized, is characterized by lowlands (Padan Plain) whereas the southern part is rich in hills and mountains reaching the top altitude of 1654 m (near Mount Falterona). These territories have a subcontinental temperate climate which turns into fresh temperate in some valleys of the Apennines. Average annual precipitation ranges between 600 and 800 mm in the lowlands, but tends to increase with altitude reaching as much as 1500 mm or more in some mountainous areas. Spring and autumn are usually the seasons with the highest level of rainfall. Temperatures vary greatly during the year. They can easily reach 35 °C in the summer, particularly in the lowlands, and high levels of humidity are common. On the contrary winter months are cold with temperatures ranging generally between – 5 °C and 10 °C, but occasionally dropping until – 20 °C particularly in the Apennines.

A total of 90 respiratory tracts from wild red foxes were analyzed. The animals were collected in official culling campaigns or found dead in the provinces of Forlì-Cesena, Rimini and Ravenna, Emilia-Romagna region. Forty-eight red foxes came from Forlì-Cesena province, twenty-nine from Rimini province and 2 from Ravenna. No information was available about the other eleven animals. The trachea and lungs were removed during necropsy at the Veterinary Diagnostic

Laboratory (IZSLER, Lombardy and Emilia-Romagna Experimental Zooprophylactic Institute, Forlì), frozen at – 20 °C and subsequently sent to the Parasitology Laboratory (DIMEVET, Department of Veterinary Medical Sciences, University of Bologna). Each specimen was labeled with information regarding the geographic origin and sex. Before analysis, the organs were defrosted at room temperature. The trachea, bronchi and bronchioles were cut open and carefully inspected for the presence of parasites. The lungs were then chopped into pieces and nematode larvae (L1) were collected using the Baermann method according to standard protocols. Measurements and pictures were taken using the software NIS-Elements D 4.10.01 64-bH. Helminths were identified according to the morphologic keys of McGarry and Morgan (2009) and Conboy (2009). The identification of nematode larvae was confirmed by molecular tools thanks to the colleagues of University of Sassari. Comparison of prevalence values of different helminth species was evaluated with Chi-square test using the software EpiInfo3.5.1.

RESULTS AND DISCUSSION

In total 271 larval forms of lung nematodes were recovered, belonging to the following species, *Crenosoma vulpis* and *Filaroides hirti*. Only one animal was positive for *E. aerophilus* eggs. Thirty per cent of the lungs were infected by at least one nematode species. Pulmonary parasitism involving one species was found in 24.4% of the cases (22 foxes), two species in 5.5% (5 foxes). *Crenosoma vulpis* showed a prevalence of 27.7% (25 positive foxes), *F. hirti* 6.6% (6 positive foxes) and *E. aerophilus* 1.1% (1 infected fox) respectively, as indicated in Table 1. Differences between the prevalence values of *C. vulpis* and other parasites were found to be statistically significant ($p < 0.001$). No adult parasites were found. Medium length of *C. vulpis* and *F. hirthi* larvae are indicated in Table 2.

Eucoleus aerophilus (Fam. Capillaridae) is a thin nematode living attached to the trachea, bronchi and bronchioles of domestic and wild carnivores, in which it can cause from moderate to severe inflammation and even foci of broncopneumonia. Definitive hosts get infected ingesting either embyonated eggs or intermediate hosts like earthworms. This nematode is probably the most common pulmonary helminth of European red foxes. High prevalence values have been detected in several countries such as Spain (Gortázar et al. 1998: 34.8%; Segovia et al. 2004: 48.1%), Great Britain (Morgan et al. 2008: 39%), the Netherlands (Borgsteede 1984: 46.8%; Franssen et al. 2014: 67.7%), Germany (Schöffel et al. 1991: 49%; Steinbach et al. 1994: 77.8%; Manke and Stoye 1998: 51.9%), Austria (Lassnig et al. 1998: 43.9%), Danmark (Saeed et al.

2006: 74.1%), Norway (Davidson et al. 2006: 88%), Lithuania (Bružinskaité-Schmidhalter et al. 2012: 97.1%) and Hungary (Sréter et al. 2003: 64%) (Table 3). In Italy the highest prevalence was found in Piedmont and Liguria (Rossi et al. 1983: 51.5%; Magi et al. 2015: 41.8%), whereas lower prevalence values were reported in Trentino Alto-Adige (Manfredi et al. 2003: 14.6%), Veneto (Iori et al. 1990: 3.6%), Tuscany (Poli et al. 1985: 29%; Magi et al. 2009: 7%), Lazio (Iori et al. 1990: 10.7%) and Sardinia (Leoni et al. 1986: 1.2%) (Table 3). In the present survey we found a prevalence of 1.1%, with only one animal infected. No adult helminths were macroscopically recovered in the trachea, bronchi and lower airways, as already reported by Traversa et al. (2009) in a study on *E. aerophilus* infection in domestic carnivores. Surprisingly we found a higher prevalence (6.6%) of ***Filaroides hirthi*** (Fam. Metastrongylidae) (Figure 1). Nematodes of this genus, transmitted through ingestion of first-stage (L1) larvae present in the saliva or in feces, are located mainly in the lung parenchyma and have been unfrequently described in European and Italian red foxes (Carvalho-Varela and Marcos 1993; Magi et al. 2015) (Table 3). ***Crenosoma vulpis*** (Figure 2) is another metastrongylid nematode living in the trachea, bronchi and lower airways of canids. In some cases bronchitis and broncopneumonia can develop. Intermediate hosts of the parasite are snails where it develops in third-stage larvae. This helminth is rather common in European red foxes and prevalence values range from 1.3% (Carvalho-Varela and Marcos 1993) to 58% (Davidson et al. 2006) (Table 3). In most European studies *C. vulpis* had lower prevalence values compared to *E. aerophilus* (e.g. Borgsteede 1984: 4.5%; Schöffel et al. 1991: 35%; Steinbach et al. 1994: 4.2%; Lassnig et al. 1998: 24.9%; Manke and Stoye 1998: 0%; Gortázar et al. 1998: 2.5%; Sréter et al. 2003: 24%; Segovia et al. 2004: 20.3%; Davidson et al. 2006: 58%; Saeed et al., 2006: 17.4%; Morgan et al. 2008: 2%; Bružinskaité-Schmidhalter et al. 2012: 53.8%; Franssen et al. 2014: 16.7%) (Table 3). Conversely in this research we found a prevalence of 27.7%, much higher than that of *E. aerophilus*. In our country lower prevalence values were reported in Piedmont and Liguria (Rossi et al. 1983: 9%; Magi et al. 2015: 15.8%), Trentino Alto-Adige (Iori et al. 1990: 20.7%; Manfredi et al. 2003: 17.3%), Veneto (Iori et al. 1990: 0%), Tuscany (Poli et al. 1985: 3%; Magi et al. 2009: 14.7%), Lazio (Iori et al. 1990: 13,1%) and Sardinia (Leoni et al. 1986, 0%).

The results of the present survey differ from many European studies on pulmonary helminths of red foxes. We gave one of the few descriptions of *F.hirthi*, a sporadic metastrongylid nematode of wild canids. Moreover we found an extremely low prevalence of *E. aerophilus*, similar to that described by Leoni et al. (1986) in Sardinia, Italy (1.2%) and Carvalho-Varela and Marcos

(1993) in Portugal (1%). It is difficult to explain why the prevalence of this helminth, transmitted both directly and by intermediate hosts like earthworms, was so low at least compared to that of *C. vulpis* (27.7%), a metastrongylid nematode transmitted only by snails or slugs. Indeed in most European surveys *E. aerophilus* showed higher prevalence values than *C. vulpis*. However opposite results were described in Portugal (Carvalho-Varela and Marcos 1993), Croatia (Rajković-Janje et al. 2002) and in other Italian regions (Iori et al. 1990; Manfredi et al. 2003; Magi et al. 2009) (Table 3). Regarding *Angyostrongylus vasorum*, a nematode described in Italy by several authors (e.g. Poli et al. 1985; Leoni et al. 1986; Iori et al. 1990; Magi et al. 2009,2015), its presence in these territories can't be ruled out since hearts and pulmonary arteries were not inspected in this study. However one possible reason for its absence or low prevalence in foxes might be the decreasing number of amphibian paratenic hosts, such as *Rana temporaria*, in the Emilia-Romagna region. Further parasitological analyses are necessary to better understand the ecological factors determining the distribution and diffusion of these helminths in our region.

Table 1. Prevalence of *C. vulpis*, *F. hirthi* and *E. aerophilus* in the present research.

Helminths	Total Prevalence (%)	Prevalence (%) per helminth species	Prevalence (%) on positive foxes
Crenosoma vulpis	30	27.7	92.6
Filaroides hirthi		6.6	22.2
Eucoleus aerophilus		1.1	3.7

Table 2. Medium length and standard deviation of *C. vulpis* and *F. hirthi* larvae.

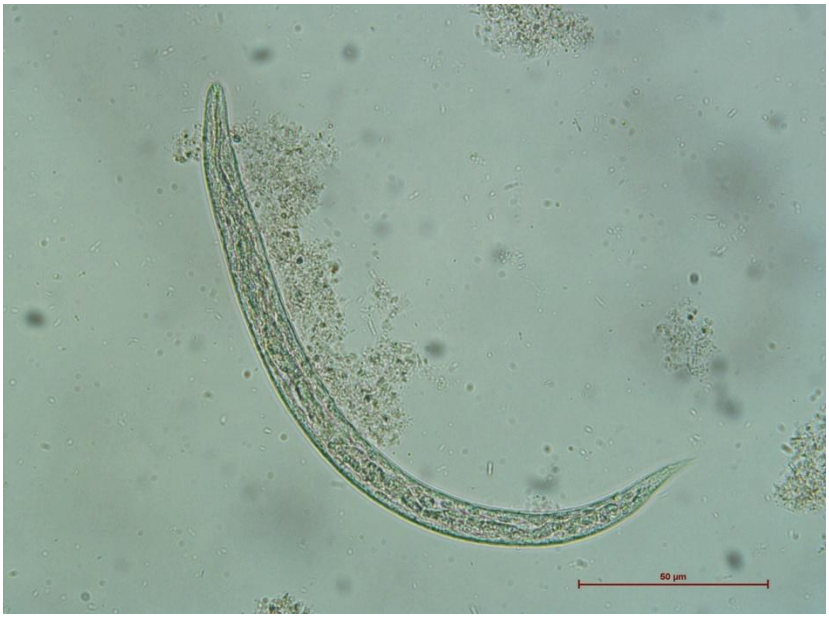
Helminths	Medium length	Standard deviation
Crenosoma vulpis	292.860 μm	16.76743
Filaroides hirthi	276.04 μm	8.289693

Table 3. Prevalence of *E. aerophilus*, *C. vulpis*, *Filaroides* spp. and *A. vasorum* in red foxes according to recent surveys.

Country	N° foxes	*Eucoleus aerophilus*	*Crenosoma vulpis*	*Filaroides spp.*	*Angiostrongylus vasorum*	Reference
Portugal	306	1	1.3	1 (*F. marti*)	0.3	Carvalho-Varela and Marcos 1993
Portugal (Coimbra)	62	4.8	3.2	-	16.1	Eira et al. 2006
Spain (Ebro Valley)	161	34.8	2.5	-	20.7 (of 29 foxes)	Gortázar et al. 1998
Iberian Peninsula	399	48.1	20.3	-	17.5	Segovia et al. 2004
Spain (Catalonia)	251	59	33.9	-	22.7	Mañas et al. 2005
Spain (Murcia)	55	5.4	-	-	1.8	Martínez-Carrasco et al. 2007
Netherlands	111	46.8	4.5	-	-	Borgsteede 1984
Netherlands	96	67.7	16.7	-	4.2	Franssen et al. 2014
Germany (Schleswig-H.)	101	50.5	-	-	-	Lucius et al. 1988
Germany (Berlin)	100	49	35	-	-	Schöffel et al. 1991
Germany (Niedersachsen)	72	77.8	4.2	-	-	Steinbach et al. 1994
Germany (Schleswig-H.)	470	51.9	-	-	-	Manke and Stoye 1998
Austria (Steiermark)	474	43.9	24.9	-	-	Lassnig et al. 1998
Denmark	748	74.1	17.4	-	48.6 (of 463 foxes)	Saeed et al. 2006
Denmark (Copenaghen)	70	87.1	22.9	-	80	Al-Sabi et al. 2014
Denmark (South. Jutland)	48	93.8	22.9	-	-	Al-Sabi et al. 2014
Norway	393	88	58	-	-	Davidson et al. 2006
United Kingdom	546	39	2	-	7.3	Morgan et al. 2008
Lithuania	104	97.1	53.8	-	-	Bružinskaité-Schmidhalter et al. 2012
Belarus	94	9.6	5.3	-	-	Shimalov and Shimalov 2003
Croatia	85	4.7	28.2	-	1.2	Rajković-Janje et al. 2002
Hungary	100	64	24		5	Sréter et al. 2003
Italy (Piedmont)	33	51.5	9	-	-	Rossi et al. 1983
Italy (Piedmont, Liguria)	165	41.8	15.8	4.8	78	Magi et al. 2015
Italy (Trentino)	29	-	20.7	-	-	Iori et al. 1990
Italy (Trentino)	42	14.6	17.3	-	-	Manfredi et al. 2003
Italy (Veneto)	28	3.6	-	-	-	Iori et al. 1990
Italy (Tuscany)	355	29	3	_	35	Poli et al. 1985
Italy (Tuscany)	129	7	14.7	-	7	Magi et al. 2009
Italy (Lazio)	205	10.7	13.1	-	6.3	Iori et al. 1990
Italy (Sardinia)	85	1.2	-	-	15.3	Leoni et al. 1986

Figure 1. *Filaroides hirthi* larva (scale bar = 50 µm).

Figure 2. *Crenosoma vulpis* larva (scale bar = 50 µm).



REFERENCES

Al-Sabi MN, Halasa T, Kapel CM. 2014. Infections with cardiopulmonary and intestinal helminths and sarcoptic mange in red foxes from two different localities in Denmark. Acta Parasitologica 59:98-107.

Borgsteede FH. 1984. Helminth parasites of wild foxes (*Vulpes vulpes* L.) in the Netherlands. Zeitschrift fur Parasitenkunde 70:281-285.

Bružinskaitė-Schmidhalter R, Šarkūnas M, Malakauskas A, Mathis A, Torgerson PR, Deplazes P. 2012. Helminths of red foxes (*Vulpes vulpes*) and raccoon dogs (*Nyctereutes procyonoides*) in Lithuania. Parasitology 139:120–127.

Carvalho-Varela M, Marcos MVM. 1993. A helmintofauna da raposa (*Vulpes vulpes silacea* Miller, 1907) em Portugal. Acta Parasitologica Portuguesa 1:73–79.

Conboy G. 2009. Helminth parasites of the canine and feline respiratory tract. Veterinary Clinics of North America: Small Animal Practice 39:1109-1126.

Davidson RK, Gjerde B, Vikøren T, Lillehaug A, Handeland K. 2006. Prevalence of Trichinella larvae and extra-intestinal nematodes in Norwegian red foxes (*Vulpes vulpes*). Veterinary Parasitology 136:307-316.

Eira C, Vingada J, Torres J, Miquel J. 2006. The helminth community of the red fox, *Vulpes vulpes*, in Dunas de Mira (Portugal) and its effect on host condition. Wildlife Biology in Practice 2:26–36.

Franssen F, Nijsse R, Mulder J, Cremers H, Dam C, Takumi K, van der Giessen J. 2014. Increase in number of helminth species from Dutch red foxes over a 35-year period. Parasites and Vectors 7:166.

Gortázar C, Villafuerte R, Lucientes J, Fernández-de-Luco D. 1998. Habitat related differences in helminth parasites of red foxes in the Ebro valley. Veterinary Parasitology 80:75–81.

Iori A, Costantini R, Cancrini G. 1990. Parassiti di volpi (*Vulpes vulpes*) provenienti da alcune regioni italiane. Parassitologia 32:153–154.

Lassnig H, Prosl H, Hinterdorfer F. 1998. Zur Parasitenfauna des Rotfuchses (*Vulpes vulpes*) in der Steiermark. Wiener Tierärztliche Monatsschrift 85:116–122.

Leoni A, Garippa G, Masala S. 1986. Osservazioni sull'elmintofauna della volpe (*Vulpes vulpes*) in Sardegna. Parassitologia 28:268–269.

Lucius R, Böckeler W, Pfeiffer AS. 1988. Parasiten der haus-, nutz- und wildtiere Schleswig-Holsteins: parasiten der inneren organe des rotfuchses (*Vulpes vulpes*). Zeitschrift fur Jagdwissenschaft 34:242–255.

Magi M, Guardone L, Prati MC, Mignone W, Macchioni F. 2015. Extraintestinal nematodes of the red fox (*Vulpes vulpes*) in north-west Italy. Journal of Helminthology 89:506-511.

Magi M, Macchioni F, Dell'Omodarme M, Prati MC, Calderini P, Gabrielli S, Iori A, Cancrini G. 2009. Endoparasites of red fox (*Vulpes vulpes*) in Central Italy. Journal of Wildlife Diseases 45:881–885.

Manfredi MT, Giacometti A, Fraquelli C, Piccolo G. 2003. Studio della popolazione elmintica in volpi (*Vulpes vulpes*) del Trentino Alto-Adige. Journal of Mountain Ecology 7:261–263.

Manke K, Stoye M. 1998. Parasitological studies of red foxes (*Vulpes vulpes*) in the northern districts of Schleswig-Holstein. Tierärztliche Umschau 53:207-214.

Mañas S, Ferrer D, Castellà J, Loppez-Martin JM. 2005. Cardiopulmonary helminth parasites of red foxes (*Vulpes vulpes*) in Catalonia, norteastern Spain. Veterinary Journal 169:118-120.

Martínez-Carrasco C, Ruiz de Ybánez MR, Sagarminaga JL, Garijo MM, Moreno F, Acosta I, Hernández S, Alonso FD. 2007. Parasites of the red fox (*Vulpes vulpes* Linnaeus, 1758) in Murcia, southeast Spain. Revue de Médicine Vétérinaire 158:331–335.

McGarry JW, Morgan ER. 2009. Identification of first-stage larvae of metastrongyles from dogs. The Veterinary Record 165:258-261.

Morgan ER, Tomlinson A, Hunter S, Nichols T, Roberts E, Fox MT, Taylor MA. 2008. Angiostrongylus vasorum and Eucoleus aerophilus in foxes (*Vulpes vulpes*) in Great Britain. Veterinary Parasitology 154:48-57.

Poli A, Arispici M, Marconcini A, Mancianti F, Corsi C. 1985. Lungworms in red foxes (*Vulpes vulpes*) from the maritime provinces of Tuscany. Erkrankungen der Zootiere. Akademie Verlag, Berlin, pp. 507-512.

Rajković-Janje R, Marinculić A, Bosnić S, Benić M, Vinković B, Mihaljević Ž. 2002. Prevalence and seasonal distribution of helminth parasites in red foxes (*Vulpes vulpes*) from the Zagreb County (Croatia). Zeitschrift fur Jagdwissenschaft 48:151–160.

Rossi L, Iori A, Cancrini G. 1983. Osservazioni sulla fauna parassitaria della popolazione di volpi presente nel Parco Regionale "La Mandria". Parassitologia 25:340–343.

Saeed I, Maddox-Hyttel C, Monrad J, Kapel CM. 2006. Helminths of red foxes (*Vulpes vulpes*) in Denmark. Veterinary Parasitology 139:168–179.

Schöffel I, Schein E, Wittstadt U, Hentsche J. 1991. Parasite fauna of red foxes in Berlin (West). Berliner und Münchener Tierärztliche Wochenschrift 104:153–157.

Segovia JM, Torres J, Miquel J. 2004. Helminth parasites of the red fox (*Vulpes vulpes* L., 1758) in the Iberian Peninsula: an ecological study. Acta Parasitologica 49:67–79.

Shimalov VV, Shimalov VT. 2003. Helminth fauna of the red fox (*Vulpes vulpes* Linnaeus, 1758) in southern Belarus. Parasitology Research 89:77-78.

Spagnesi M, De Marinis AM. 2002. Mammiferi d'Italia. Quaderni di Conservazione della Natura, 14. Ministero dell'Ambiente e della Tutela del Territorio e del Mare - Istituto Nazionale Per la Fauna Selvatica "A. Ghigi". 309 pp.

Sréter T, Széll Z, Marucci G, Pozio E, Varga I. 2003. Extraintestinal nematode infections of red foxes (*Vulpes vulpes*) in Hungary. Veterinary Parasitology 115:329-334.

Steinbach G, Welzel A, M. Von Keyserlingk M, Stoye M. 1994. On the helminthic fauna of the red fox (*Vulpes vulpes* L.) in southern lower Saxony. 1. Nematodes and trematodes. Zeitschrift fur Jagdwissenschaft 40:30–39.

Traversa D, Di Cesare A, Milillo P, Iorio R, Otranto D. 2009. Infection by *Eucoleus aerophilus* in dogs and cats: is another extra-intestinal parasitic nematode of pets emerging in Italy ? Research in Veterinary Science 87:270-272.

5. HELMINTH PARASITES OF RAPTORS IN NORTHERN ITALY

ALFREDO FIOCCHI [1], ANDREA GUSTINELLI [1], LUCA GELMINI [2], STEFANO BASSI [2], GIOVANNI POGLAYEN [1], MARIA LETIZIA FIORAVANTI [1]

[1] *Department of Veterinary Medical Sciences, Alma Mater Studiorum University of Bologna, Ozzano Emilia (BO), Italy*

[2] *Lombardy and Emilia Romagna Experimental Zooprophylactic Institute, Modena, Italy*

TO BE PUBLISHED IN A PEER-REVIEWED JOURNAL

ABSTRACT

A parasitological survey on the helminthic fauna of 67 birds of prey from Emilia-Romagna region, Italy was carried out during 2009-2011. Eight species of raptors were analyzed, namely 15 *Accipiter nisus*, 9 *Falcus tinnunculus*, 8 *Buteo buteo*, 1 *Falcus peregrinus*, 12 *Asio otus*, 9 *Athene noctua*, 8 *Strix aluco* and 5 *Otus scops*.They all represent protected avian species and a few data are available on their helminthic fauna in our country. Helminth infestation was found in 55 (82.1%) out of 67 animals. The following taxa were recovered: trematodes (*Strigea falconis*, *Neodiplostomum* spp.), nematodes (*Eucoleus dispar*, *Capillaria tenuissima*, *Baruscapillaria falconis*, *Physaloptera alata*, *Microtetrameres cloacitectus*, *Microtetrameres* sp., *Synhimantus laticeps*, *Synhimantus robertdollfusi*, *Dyspharinx* spp., *Procyrnea leptoptera*, *Procyrnea spinosa*, *Porrocaecum* spp., *Subulura* sp., *Serratospiculum* sp.) and acanthocephalans (*Echinorhynchus* sp., *Centrorhynchus aluconis*, *Centrorhynchus globocaudatus*). The helminth community was richer in falconiform and accipitriform than in strigiform birds. In particular *A.nisus* and *B. buteo* showed the highest number of parasite species, *A. noctua* and *O. scops* the lowest.

Keywords: helminths; birds of prey; Emilia-Romagna; Italy.

INTRODUCTION

Diurnal and nocturnal birds of prey are protected or even endangered species. A few recent reports focused on Southern regions are available in literature on the parasitofauna of these avian species in Italy (Santoro et al. 2010, 2012a, 2012b). Being located at the top of the food chain and involved as definitive hosts of the indirect life cycles of several parasite species, the possibility to examine a high number of dead specimens represents an infrequent and important opportunity to study the parasite occurrence in raptors and to better understand both biological and health aspects concerning these animals. At this regard, a parasitological survey on the helminthic fauna of 67 birds of prey from Emilia-Romagna region, Italy was carried out in the past years. Our survey focused on the prevalence of helminth parasites of the digestive and respiratory tracts.

MATERIALS AND METHODS

Between 2009 and 2011, we analysed 67 diurnal and nocturnal birds of prey, 15 sparrowhawks (*Accipiter nisus*), 9 common kestrels (*Falcus tinnunculus*), 8 common buzzards (*Buteo buteo*) and 1 peregrine falcon (*Falcus peregrinus*), 12 long-eared owls (*Asio otus*), 9 little owls (*Athene noctua*), 8 tawny owls (*Strix aluco*) and 5 scops owls (*Otus scops*). All the birds were collected in a wildlife recovery center of Emilia-Romagna region (Northern Italy), frozen after death and subsequently sent to the diagnostic laboratory. During necropsy, the esophagus, the stomachs and the whole intestine were carefully inspected for helminths. The mucosa was gently scraped with a sharp spoon and the collected material was allowed to precipitate in tap water inside conical jars for nearly 30 minutes. Repeated washings and decantations were performed until the sediment in the jars was sufficiently clear to be examined by the stereomicroscope. Helminths found were washed and fixed in 70% ethanol for identification based on morphological features. Parasite examinations were not quantitative. The trachea, bronchi, lungs and the air sacs were also inspected for helminths. In the case of the peregrine falcon, it was possible to collect only the nematodes present in the respiratory tract, since the gastro-intestinal tract was not available for parasitological analysis.

RESULTS

All the birds analysed had been kept frozen for several months so that most of them were not in a good state for macroscopic and particularly histopathological analysis. Twenty-five animals

(37.3%) had various kind of traumatic injuries. We also evidenced one *A. otus* with an extended phlegmon, another one with severe cutaneous acariasis and one *A. nisus* with avian tuberculosis. Esophageal and gastro-intestinal helminths were recovered in 54 raptors (81.8%). Twelve birds (4 *F. tinnunculus*, 4 *O. scops*, 3 *A. noctua*, and 1 *A. otus*) were negative. No parasites (except the nematode *Serratospiculum* sp. in *F. peregrinus*) were recovered in the trachea, lungs and air sacs of the examined animals. The helminth species detected and infestation rates per host species are indicated in Table 1. *Capillaria tenuissima* and *Baruscapillaria falconis* were found infecting five avian species each; *Synhimantus laticeps*, *Dispharynx* spp., *Procyrnea leptoptera* and *Porrocaecum* spp. four avian species each; *Strigea falconis*, *Neodiplostomum* spp. and *Eucoleus dispar* three avian species each; *Physaloptera alata*, *Microtetrameres cloacitectus*, *Microtetrameres* sp., *Synhimantus robertdollfusi*, *Procyrnea spinosa*, *Subulura* sp., *Echinorhyncus* sp., *Centrorhyncus aluconis* and *C. globocaudatus* one avian species each. Genera *Physaloptera* and *Microtetrameres* were recovered only in diurnal raptors, whereas *Subulura* and *Echinorhyncus* only in nocturnal raptors. Specimens indicated in Table 1 as *Capillaria* spp., *Physaloptera* sp., *Synhimantus* spp., *Procyrnea* spp. could not be identified at species level either because they were too damaged or because only female parasites were present. Specimens of *Microtetrameres* sp. recovered in five *A. nisus* belonged to a different species from *Microtetrameres cloacitectus*.

DISCUSSION

Concerning digenean trematodes, ***Strigea falconis*** (Szidat, 1928) (order *Strigeida*) is a common intestinal fluke of birds of prey. Its complex life-cycle requires a water snail as the first intermediate host (sporocysts stage), an amphibian as the second one (mesocercaria stage) and another vertebrate, which can be a mammal, bird, reptile or amphibian, as the third intermediate host (metacercaria stage) (Krone and Streich 2000; Krone and Cooper 2002). Strigeid and diplostomatid trematodes are usually considered as low-pathogenic in raptors (Smith 1996; Krone and Cooper 2002). We recovered the parasite in *A. nisus*, *B. buteo* and *A. otus* (Table 1). Prevalence of infection was higher in *B. buteo* than in *A. nisus* also in the Czech Republic (Sitko 1994), Germany (Krone 2000) and Catalonia (Ferrer et al. 2004b). *A. otus* was the only strigiform found infected also in Galicia (Sanmartín et al. 2004). ***Neodiplostomum*** species were found in the intestinal lumen of *A. nisus*, *B. buteo* and *S. aluco* with the highest prevalence in *B. buteo* (37.5%) (Table 1). This avian species was the most frequently affected also in Germany

(Krone 2000), Galicia (Sanmartín et al. 2004) and Southern Italy (Santoro et al. 2010, 2012a). Infestations in tawny owls were also described by Kutzer et al. (1982) in Austria, Borgsteede et al. (2003) in the Netherlands, Ferrer et al. (2004a) and Sanmartín et al. (2004) in Spain and Santoro et al. (2012b) in Southern Italy.

Eucoleus dispar (Duj., 1845) (order *Enoplida*) has a direct life cycle, but earthworms may act as paratenic hosts. It can cause from mild to severe inflammation of the upper alimentary tract. Diphtheritic membranes can develop in the buccal cavity, pharynx, esophagus and crop (Krone and Cooper 2002). Krone (2000) described an extremely high prevalence of infection in *A. nisus*. We also found *E. dispar* in *A. nisus* (prevalence 33.3%) and less frequently in *B. buteo* and *S. aluco* (Table 1). Conversely Ferrer et al. (2004b), Sanmartín et al. (2004) and Santoro et al. (2010, 2012a) all reported higher prevalence values in *B. buteo* than in *A. nisus*. Similarly to Ferrer et al. (2004a, 2004b), Sanmartín et al. (2004) and Santoro et al. (2012a, 2012b), who reported the parasite only in falconiforms, no strigiform bird except one tawny owl was found infected. Okulewicz (1988) also described *E. dispar* in *S. aluco* from Poland.

Capillaria tenuissima (Rud., 1809) and ***Baruscapillaria falconis*** (Goeze, 1782) are both intestinal threadworms of birds of prey. At low densities, affected animals do not demonstrate clinical signs. However, in heavy infections they can show diarrhea, anorexia and weight loss (Krone and Cooper 2002). In our survey *C. tenuissima* was the most common species in birds with intestinal capillariasis, with the exception of *S. aluco* (Table 1). Similarly to Illescas Gomez et al. (1993) and Ferrer et al. (2004b) neither esophageal nor intestinal capillarid nematodes were found in *F. tinnunculus*. Krone (2000) and Sanmartín et al. (2004) similarly reported high prevalence values of *C. tenuissima* in *B. buteo* and *A. nisus*, but did not find any *B. falconis*. Among strigiforms, the latter author described the highest prevalence of *C. tenuissima* in *A. noctua*. Conversely, we found the highest frequency of infection in *A. otus* (75%). A high prevalence in this owl was also reported by Borgsteede et al. (2003). Coinfections were found in three *A. otus*, one *S. aluco* and one *A. nisus*. In one *O. scops* and one *S. aluco* we recovered unidentified *Capillaria* specimens (Table 1).

Physaloptera species (order *Spirurida*) are normally found in the esophagus, proventriculus and gizzard of raptors. In heavy infections they may cause irritation and inflammation of the mucous membrane. Insects are believed to act as intermediate hosts (Krone and Cooper 2002). We found *P. alata* (Rud., 1819) only in *A. nisus* (prevalence 46.6%) and *Physaloptera* sp. in one *Falcus*

tinnunculus (Table 1). Similarly Ferrer et al. (2004b), Sanmartín et al. (2004) and Santoro et al. (2010, 2012a) reported high prevalence values of *Physaloptera* species in *A. nisus*.

Microtetrameres species (order *Spirurida*) inhabit mainly the proventriculus, where females live in the glandular tissue and the much smaller males in the lumen of the organ. After ingesting the eggs released in faeces, insects and isopods act as intermediate hosts (Krone and Cooper 2002). We found *Microtetrameres* sp. in *A. nisus* (prevalence 33.3%) and *Microtetrameres cloacitectus* (Oschmarin, 1956) in *B. buteo* (prevalence 25%) (Table 1). In other birds examined (two buzzards, one sparrowhawk and one kestrel) we recovered unidentified small tetramerid larvae. Ferrer et al. (2004b) and Sanmartín et al. (2004) similarly reported high prevalence values of *Microtetrameres* sp. in *A. nisus* in Spain. Baruš (1966), Kutzer et al. (1980), Sitko (1994) and Krone (2000) also described *M. cloacitectus* in common buzzards. Both Ferrer et al. (2004a) and Sanmartín et al. (2004) did not find this genus in strigiform birds. On the contrary, Illescas Gomez et al. (1993) described *Microtetrameres* sp. in *A. noctua*.

Synhimantus species (order *Spirurida*) are nematodes living both in the stomachs of hawks and owls and in their upper alimentary tract (pharynx, esophagus). The irritation caused by these parasites can affect the feeding behavior of the parasitized birds (Krone and Cooper 2002). Insects and isopods are supposed to be intermediate hosts (Sanmartín et al. 2004). In the present study we found *Synhimantus laticeps* (Rud., 1819) in *A. nisus*, *B. buteo*, *F. tinnunculus* and *A. otus* with the highest prevalence values in *F. tinnunculus* (44.4%) and *A. otus* (41.6%) (Table 1). Krone (2000), Sanmartín et al. (2004) and Santoro et al. (2010, 2012a) all reported the parasite in the above-mentioned diurnal raptors. Similarly Borgsteede et al. (2003) and Sanmartín et al. (2004) reported the highest prevalence in *A. otus* among strigiforms. Conversely Ferrer et al. (2004a) described *Synhimantus* spp. in several strigiform species except *A. otus* in Catalonia, Spain. *Synhimantus robertdollfusi* was found in one *B. buteo*. Unidentified *Synhimantus* specimens were also recovered in *A. nisus* and *A. otus* (Table 1). Moreover ***Dispharynx*** species were found in the stomachs of *A. nisus*, *F. tinnunculus* and *A. noctua*. Six *A. otus* (prevalence 50%) had helminths of this genus in the esophagus (Table 1).

Procyrnea leptoptera (Rud., 1819) is another spirurid nematode inhabiting the proventriculus and gizzard of birds of prey. Insects (orthopterans, coleopterans) are probable intermediate hosts (Sanmartín et al. 2004). In the present research it was found in *A. nisus*, *B. buteo*, *F. tinnunculus* and *S. aluco* with the highest prevalence in *A. nisus* (40%) (Table 1). Unidentified *Procyrnea* specimens were recovered in *B. buteo*, *F. tinnunculus* and *S. aluco*. Similarly to Baruš (1966),

Kutzer et al. (1980), Sitko (1994) and Krone (2000) *Procyrnea spinosa* (Gendre, 1922) was found in *F. tinnunculus* (Table 1).

Porrocaecum species (order *Ascaridida*) were found in *A. nisus, B. buteo, F. tinnunculus* and *A. otus* with the highest prevalence values in *B. buteo* (62.5%) and *A. nisus* (33.3%) (Table 1). Similar results in the latter hosts were described by Krone (2000) and Ferrer et al. (2004b). These nematodes live mainly in the small intestine of birds of prey, but can be recovered also in their stomachs. In large number they can lead to obstruction of the lumen of the gut or even perforate its wall (Smith 1996; Santoro et al. 2010). Sanmartín et al. (2004) reported high prevalence values for *Porrocaecum angusticolle* in *B. buteo* and *A. nisus*, whereas Santoro et al. (2010, 2012a) mainly in *B. buteo*. The life cycle of *P. angusticolle* is indirect with Insectivora as intermediate hosts (Krone and Cooper 2002). ***Subulura*** sp. was recovered only in one *O. scops* (Table 1). Similarly Ferrer et al. (2004a) and Santoro et al. (2012b) reported this species in scops owls.

The spirurid ***Serratospiculum*** sp., recovered in the air sacs of the one peregrine falcon examined, was the only helminth found in the respiratory tract of the birds analysed. Santoro et al. (2010, 2012a, 2016) described a high prevalence of infection in this host with consequent cachexia, pneumonia and air sacculitis.

Acanthocephalans are intestinal parasites of birds of prey. Their life cycle involves an arthropod as intermediate host and one or more vertebrates (small mammals, reptiles, amphibians) as paratenic hosts (cystacanth stage). In heavy infections thorny-headed worms can cause severe enteritis and even generalized peritonitis (Smith 1996; Santoro et al. 2010). We found *Echinorhynchus* sp. in one *A. otus*, *Centrorhynchus globocaudatus* in one *B. buteo*, *Centrorhynchus* sp. in one *F. tinnunculus* and *Centrorhynchus aluconis* in one *S. aluco* (Table 1). One unidentified acanthocephalan was also found in one *S. aluco*. *Centrorhynchus* species were reported by several authors (e.g. Ewald and Crompton 1993; Borgsteede et al. 2003; Ferrer et al. 2004a, 2004b; Sanmartín et al. 2004; Papazahariadou et al. 2008; Santoro et al. 2012a, 2012b) and according to Krone and Cooper (2002) they represent the most frequently diagnosed genus of acanthocephalans in European raptors. Unidentified tapeworms were found only in one *B. buteo* and one *O. scops.* Since all the animals were deeply frozen after death and kept in this status for several months we suspect, agreeing with Ferrer et al. (2004a), that the freezing and thawing process probably did not allow a good conservation status of this group of parasites.

Despite the sample size of different avian species being not fully homogeneous, we can point out that diurnal birds of prey (Accipitriforms and Falconiforms) showed a richer helminthic fauna of the digestive tract (15 taxa) than strigiforms (12 taxa). Two species (plus an unidentified tapeworm) were found in *Otus scops*, three species in *A. noctua*, six in *S. aluco*, seven in *A. otus*, whereas seven in *F. tinnunculus*, eleven in *A. nisus* and eleven (plus an unidentified tapeworm) in *B. buteo*. The richest helminth component community was found in common buzzards and sparrowhawks. This is not surprising in the case of *B. buteo*. Its diet is known to be extremely varied including small mammals, birds, reptiles, amphibians, insects and other arthropods. Consequently it should be exposed to a high number of potential intermediate hosts of parasites (Santoro et al. 2012a; Tezel et al. 2015). On the contrary sparrowhawk's diet is usually restricted to birds and small mammals (Brichetti and Dicapi 1991; Spagnesi and Serra 2004; Sanmartín et al. 2004). Therefore, it is quite surprising that helminths known to be transmitted mainly through invertebrate intermediate hosts were recovered at high prevalences in this host. This is the case of *Eucoleus dispar*, *Microtetrameres* sp., *Physaloptera alata* and *Procyrnea leptoptera*. Ferrer et al. (2004b) and Sanmartín et al. (2004) also reported similar data. We agree with the latter author on the hypothesis that *A. nisus* may therefore ingest a higher quantity of invertebrates than usually thought whether directly or indirectly in prey stomach contents. The helminthic fauna recovered in *F. tinnunculus*, with high prevalences of genera *Synhimantus*, *Dispharynx* and *Procyrnea*, is actually in accordance with its typology of diet, in which insects and other arthropods are known to play an important part (Brichetti and Dicapi 1991; Spagnesi and Serra 2004; Ferrer et al. 2004b). Regarding nocturnal raptors, similarly to Sanmartín et al. (2004), little owls showed a lower species richness compared with tawny owls and long-eared owls. Moreover we also noticed the absence of genera *Microtetrameres* and *Physaloptera* and the extremely low prevalence of *E. dispar* in strigiforms. The scops owl, characterized mostly by an insectivorous diet (Spagnesi and Serra 2003), is another species that showed a restricted helminthic fauna. The difference between *A. otus*, *S. aluco* and the smaller strigiforms (*A. noctua* and *O. scops*) might be related to the more complex diet of the former species. However, if scops owls rely mainly on arthropods, little owls are reported to have a much varied diet which can include rodents, small birds, reptiles and invertebrates (Brichetti and Dicapi 1991). Interestingly in other surveys (Ferrer et al. 2004a; Santoro et al. 2012b) both *A. noctua* and *O. scops* showed a richer helminth community, much wider than *A. otus*. This divergence among surveys is difficult to justify. It is most likely explained by the different sample size of avian species among different surveys, but

might be also related to the diversity and adaptability of owls' diet in different geographical areas and to the diversity of local helminthic fauna.

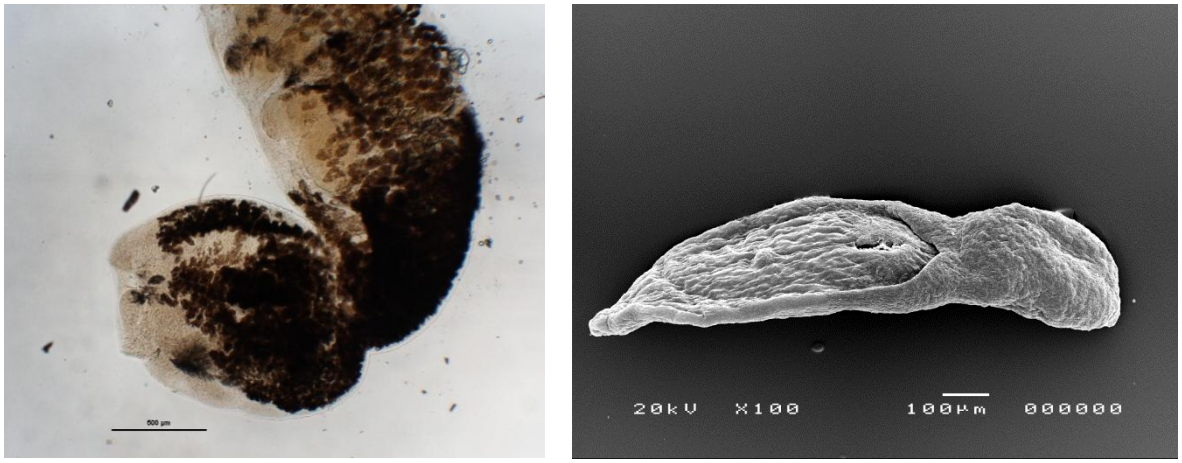
CONCLUSIONS

The present survey focuses on the helminthic fauna of birds of prey from Northern Italy. Although the results of researches dealing with animals coming from wildlife rehabilitation centers cannot be considered fully representative of the normal parasite fauna of free-living bird populations, it is often the only opportunity to study these protected animals. Our survey allowed to recover parasitological findings which are consistent with recent surveys on raptors' helminthic fauna in other European countries and it could represent a further contribute to better understand the health and biology of birds of prey.

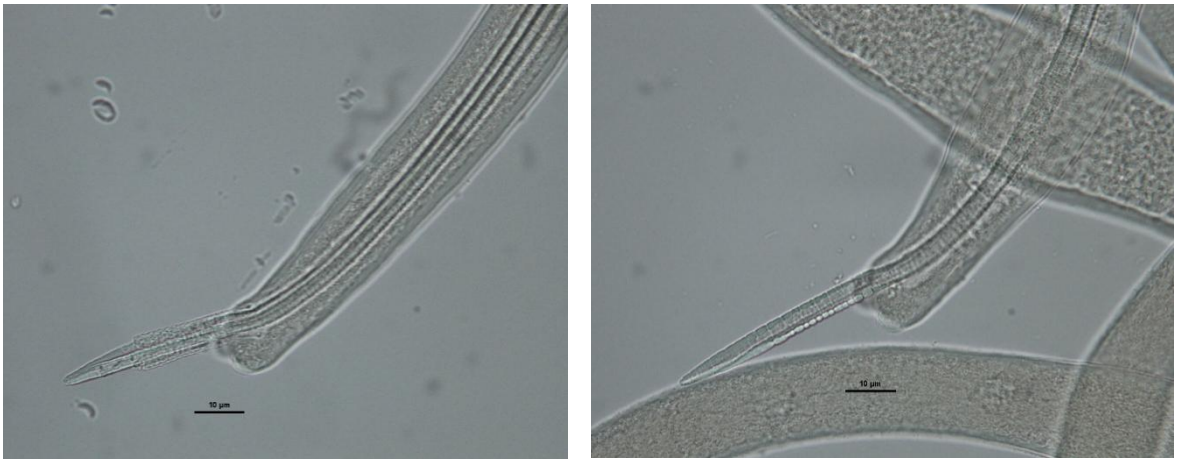
Acknowledgements: the authors are grateful to the wildlife recovery centre "Il Pettirosso", Modena, Italy, for providing samples and to the AUSL of Mirandola, Modena for logistic support during the survey.

Table 1. Helminth prevalences in raptors. Percentages in relation to total animals examined.

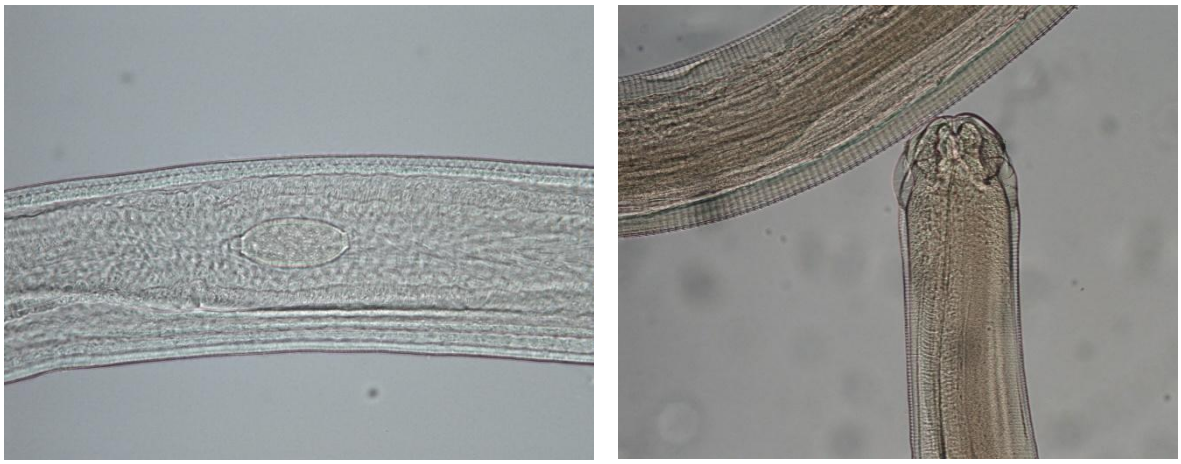
	A. nisus (n=15)	*Buteo buteo* (n=8)	*F. tinnunculus* (n=9)	*Asio otus* (n=12)	*A. noctua* (n=9)	*Strix aluco* (n=8)	*Otus scops* (n=5)
Strigea falconis	**13.3**	**25**	_	**16.6**	_	_	_
Neodiplostomum spp.	**6.6**	**37.5**	_	_	_	**25**	_
Eucoleus dispar	**33.3**	**12.5**	_	_	_	**12.5**	_
Capillaria tenuissima	**40**	**50**	_	**75**	**44.4**	**25**	_
Baruscapillaria falconis	**6.6**	**25**	_	**33.3**	**11.1**	**75**	_
Capillaria spp.						**12.5**	**20**
Physaloptera alata	**46.6**	_	_	_	_	_	_
Physaloptera sp.	_	_	**11.1**	_	_	_	_
Microtetrameres cloacitectus	_	**25**	_	_	_	_	_
Microtetrameres sp.	**33.3**	_	_	_	_	_	_
Synhimantus laticeps	**13.3**	**25**	**44.4**	**41.6**	_	_	_
Synhimantus robertdollfusi	_	**12.5**	_	_	_	_	_
Synhimantus spp.	**6.6**	_	_	**25**	_	_	_
Dispharynx spp.	**13.3**	_	**22.2**	**50**	**11.1**	_	_
Procyrnea leptoptera	**40**	**37.5**	**22.2**	_	_	**12.5**	_
Procyrnea spinosa	_	_	**11.1**	_	_	_	_
Procyrnea spp.	_	**12.5**	**33.3**	_	_	**12.5**	_
Porrocaecum spp.	**33.3**	**62.5**	**11.1**	**16.6**	_	_	_
Subulura sp.	_	_	_	_	_	_	**20**
Echinorhyncus sp.	_	_	_	**8.3**	_	_	_
Centrorhyncus spp.	_	**12.5** (*C.globocaudatus*)	**11.1**	_	_	**12.5** (*C. aluconis*)	_



Strigea falconis in *A. nisus* at the light microscope (on the left) and *Neodiplostomum* sp.in *S. aluco* at the Scanning Electron Microscope (on the right).

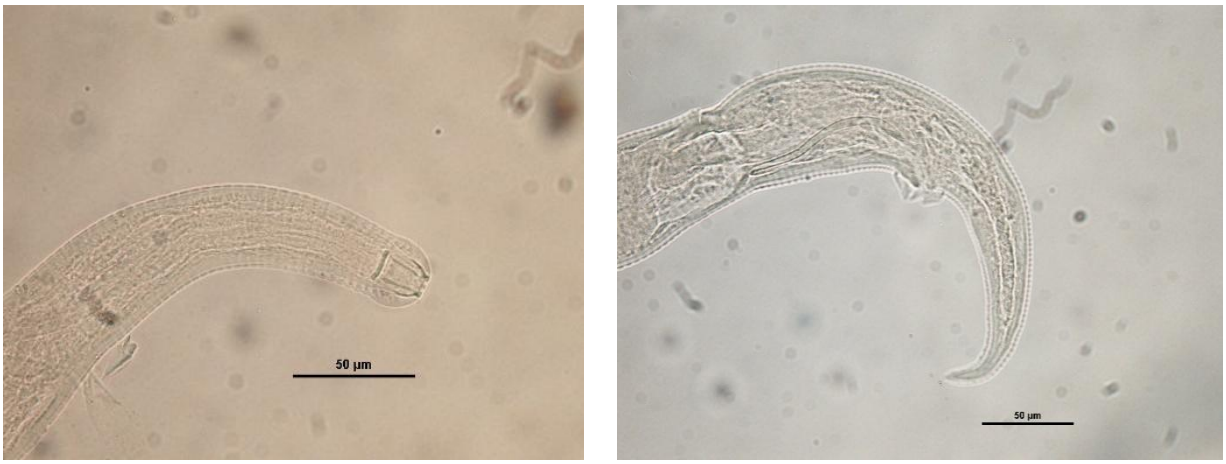


Spicules of *Capillaria tenuissima* (on the left) and *B. falconis* (on the right) in *A. otus*.

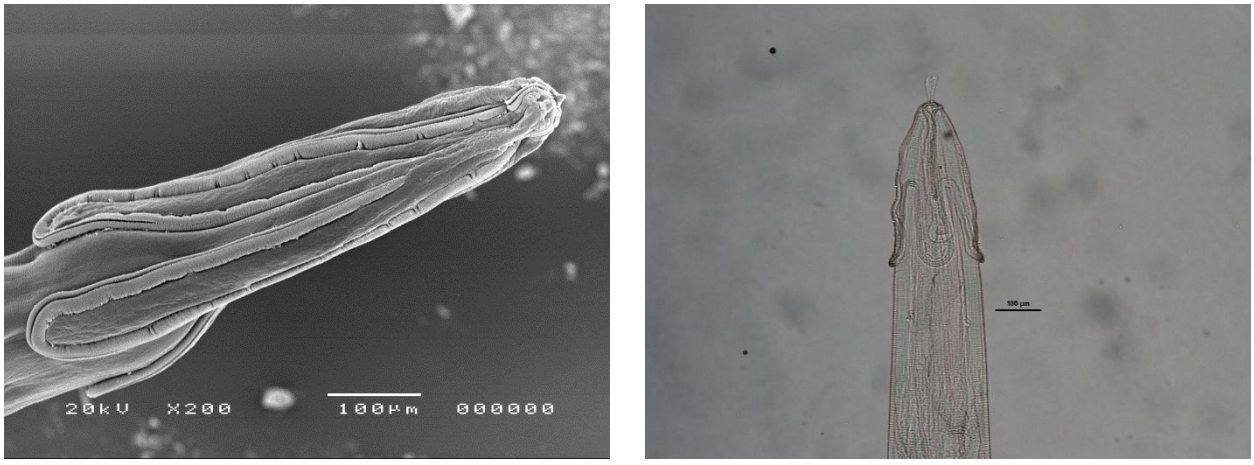


Egg of *Eucoleus dispar* in *B. buteo* (on the left) and cephalic part of *Porrocaecum* sp. in *A. otus* (on the right).

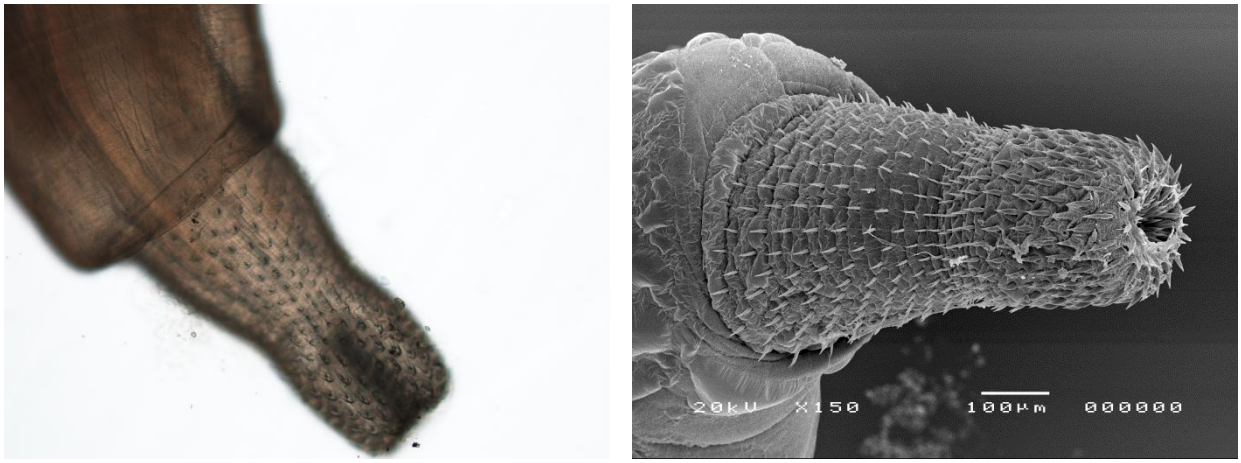
Physaloptera alata in *A. nisus*: anterior part (on the left) and posterior part (on the right).



Microtetrameres sp. in *A. nisus*: anterior part (on the left) and posterior part (on the right).



Cephalic part of *Dispharynx* sp. at the Scanning Electron Microscope (on the left) and of *Synimanthus laticeps* at the light microscope (on the right) in *A. otus*.



Specimens of the acanthocephalan *Centrorhynchus globocaudatus*: everted proboscis at the light microscope (on the left) and at Scanning Electron Microscope (on the right).

REFERENCES

Baruš V. 1966. Parasitic nematodes of birds in Czechoslovakia I. Hosts: *Columbiformes*, *Piciformes*, *Falconiformes* and *Strigiformes*. Folia Parasitologica (Praha) 13:7-27.

Borgsteede FHM, Okulewicz A, Zoun PEF, Okulewicz J. 2003. The helminth fauna of birds of prey (Accipitriformes, Falconiformes and Strigiformes) in the Netherlands. Acta Parasitologica 48:200-207.

Brichetti P, Dicapi C. 1991. Il grande libro degli uccelli d'Italia e d'Europa. De Vecchi Editore, Milano, Italy, pp. 122-124.

Ewald JA, Crompton DWT. 1993. *Centrorhynchus aluconis* (Acanthocephala) and other helminth species in tawny owls (*Strix aluco*) in Great Britain. The Journal of Parasitology 79:952-955.

Ferrer D, Molina R, Castellà J, Kinsella JM. 2004a. Parasitic helminths in the digestive tract of six species of owls (Strigiformes) in Spain. The Veterinary Journal 167:181-185.

Ferrer D, Molina R, Adelantado C, Kinsella JM. 2004b. Helminths isolated from the digestive tract of diurnal raptors in Catalonia, Spain. The Veterinary Record 154:17-20.

Illescas Gomez MP, Rodríguez Osorio M, Aranda Maza F. 1993. Parasitation of falconiform, strigiform, and passeriform (Corvidae) birds by helminths in Spain. Research and Reviews in Parasitology 53:129-135.

Krone O. 2000. Endoparasites in free-ranging birds of prey in Germany. In: Lumeij JT, Remple JD, Redig PT, Lierz M and Cooper JE, Editors. Raptor Biomedicine III. Zoological Education Network, Inc., Lake Worth, Florida, pp. 101-116.

Krone O, Streich WJ. 2000. *Strigea falconispalumbi* in Eurasian buzzards from Germany. Journal of Wildlife Diseases 36:559-561.

Krone O, Cooper JE. 2002. Parasitic disease. In: Cooper JE, Editor. Birds of Prey. Health and Diseases, Third ed. Iowa State Press, pp. 105-120.

Kutzer E, Frey H, Kotremba J. 1980. Zur parasitenfauna österreichischer Greifvögel (Falconiformes). Angewandte Parasitologie 21:183-205.

Kutzer E, Frey H, Nöbauer H. 1982. Zur parasitenfauna österreichischer eulenvögel (Strigiformes). Angewandte Parasitologie 23:190-197.

Okulewicz A. 1988. Nematodes in Falconiformes and Strigiformes of Lower Silesia. Wiadomości Parazytologiczne 34:137-149.

Papazahariadou M, Diakou A, Papadopoulos E, Georgopoulou I, Komnenou A, Antoniadou-Sotiriadou K. 2008. Parasites of the digestive tract in free-ranging birds in Greece. Journal of Natural History 42:381-398.

Sanmartín ML, Álvarez A, Barreiro G, Leiro J. 2004. Helminth fauna of falconiform and strigiform birds of prey in Galicia, Northwest Spain. Parasitology Research 92:255-263.

Santoro M, Tripepi M, Kinsella JM, Panebianco A, Mattiucci S. 2010. Helminth infestation in birds of prey (*Accipitriformes* and *Falconiformes*) in Southern Italy. The Veterinary Journal 186:119-122.

Santoro M, Kinsella JM, Galiero, G, degli Uberti B, Aznar FJ. 2012a. Helminth community structure in birds of prey (*Accipitriformes* and *Falconiformes*) in Southern Italy. Journal of Parasitology 98:22-29.

Santoro M, Mattiucci S, Nascetti G, Kinsella JM, Di Prisco F, Troisi S, D'Alessio N, Veneziano V, Aznar FJ. 2012b. Helminth communities of owls (Strigiformes) indicate strong biological and ecological differences from birds of prey (Accipitriformes and Falconiformes) in Southern Italy. Plos One 7(12):e53375.

Santoro M, D'Alessio N, Di Prisco F, Kinsella JM, Barca L, Degli Uberti B, Restucci B, Martano M, Troisi S, Galiero G, Veneziano V. 2016 The occurrence and pathogenicity of *Serratospiculum tendo* (Nematoda: Diplotriaenoidea) in birds of prey from southern Italy. Journal of Helminthology 90:294-297.

Sitko J. 1994. Helminths of birds of prey (*Falconiformes*) and owls (*Strigiformes*) in the Czech Republic and their influence on health condition of caged birds. Zprávy MOS 52:53-84.

Smith SA. 1996. Parasites of Birds of Prey: their diagnosis and treatment. Seminars in Avian and Exotic Pet Medicine 5:97-105.

Spagnesi M, Serra L. 2003. Uccelli d'Italia. Quad. Cons. Natura, 16, Ministero Ambiente- Ist. Naz. Fauna Selvatica, pp. 204-205.

Spagnesi M, Serra L. 2004. Uccelli d'Italia. Quad. Cons. Natura, 21, Ministero Ambiente- Ist. Naz. Fauna Selvatica, pp. 38-39, 60-61.

Tezel M, Girişgin AO, Birlik S, Yildirimhan HS, Şenlik B. 2015. Helminths of the digestive tract in *Buteo buteo* (Falconiformes: Falconidae) in Bursa Province of Northwest Turkey. Turkish Journal of Zoology 39:323-327.

6. IL MATTATOIO COME OSSERVATORIO EPIDEMIOLOGICO, UN ASPETTO NEGLETTO. AGGIORNAMENTO SUGLI ENDOPARASSITI BOVINI IN ITALIA.

THE SLAUGHTERHOUSE AS AN EPIDEMIOLOGICAL OBSERVATORY, A NEGLECTED TASK. UPDATING OF BOVINE ENDOPARASITES IN ITALY.

COSTANZA ROMANELLI[1], GIANLUCA PIO ZAFFARANO[1], BENEDETTO MORANDI[1], ALFREDO FIOCCHI[1], VANNES BENFENATI[2], GIOVANNI POGLAYEN[1]

[1] *Dipartimento di Scienze Mediche Veterinarie, Alma Mater Studiorum-Università di Bologna*

[2] *A.S.L. Bologna, Via Altura 3, 40139 Bologna*

RIASSUNTO Scopo dell'indagine è stato quello di fornire dati aggiornati sulla prevalenza e la diffusione di infestazioni da nematodi gastrointestinali in bovini adulti allevati in Italia. Lo studio ha avuto inizio con la raccolta di 427 campioni di feci da bovini processati in un macello della provincia di Bologna. Questi, prelevati da singoli animali, sono stati analizzati in laboratorio mediante esame coprologico qualitativo. Dagli stessi animali sono stati scelti a caso 100 abomasi, esaminati poi al tavolo necroscopico per valutare la presenza di forme adulte di nematodi. Uova di nematodi gastrointestinali sono state individuate nel 31% dei campioni di feci esaminati. Negli abomasi è stata rilevata una positività del 13% per elminti gastro-intestinali. Il loro isolamento ha permesso di identificarli come appartenenti ai generi *Ostertagia*, *Trichostrongylus* e *Cooperia*. Sulla base di indagini statistiche l'eliminazione di uova di elminti gastro-intestinali è risultata associata in maniera significativa sia alla categoria produttiva di appartenenza sia alle dimensioni in numero di capi dell'allevamento. Il presente studio dimostra che il parassitismo gastro-intestinale da nematodi è un problema che deve essere considerato diffuso nei bovini allevati in Italia, con livelli di prevalenza non trascurabili. Tuttavia sembra essere ancora sottostimato da parte dei tecnici del settore.

Parole chiave: bovini, parassiti, strongili gastro-intestinali, mattatoio.

ABSTRACT Introduction Gastro-intestinal nematodes are important helminth parasites in all animal species. However, they must regarded particularly dangerous in domestic ruminant species, also in relation to consequent economic losses. **Aim** The present study focused on providing current data, missing since several decades, on gastro-intestinal nematode parasitic infection, prevalence and epidemiology in adult cattle (dairy and brood cows) bred in Italy. **Materials and methods** The survey was performed collecting 427 fecal samples from a bovine slaughterhouse in the province of Bologna (Italy). Samples, obtained from single animals processed, were analyzed by qualitative coprological examinations. From the same animals 100 abomasa were randomly selected and examined by necropsy technique to assess the presence of worm burdens. **Results** Gastro-intestinal nematode eggs were detected in 31% of individual fecal samples examined. Evaluation of abomasa exhibited a prevalence of 13% of helminthes. *Ostertagia*, *Trichostrongylus* and *Cooperia* were the isolated genera. The fecal output of nematode eggs was significantly related with the livestock category and the stocking density. **Discussion** The influence of livestock category on the occurrence of positive coprological results

can be attributed to the condition of animal husbandry: brood cows are often pasture raised. The correlation observed between positive samples and herd size, with intermediate class (50-99 animals) associated with higher prevalence, may be explained by a different effectiveness of hygiene management among classes of stocking density. **Conclusions** The study results show that endoparasitic infection by nematodes in adult cattle is a problem that must be considered ubiquitous in Italy, with a relatively high prevalence rate. Nevertheless, it seems to be still underestimated by technicians in the field.

Key words: cattle, parasites, gastro-intestinal strongyles, slaughterhouse.

INTRODUZIONE

Il patrimonio bovino nazionale ammonta a 5535696 animali (www.statistiche.izs.it). I parassiti che colpiscono questa specie sono numerosi, anche se l'evoluzione di un certo tipo di allevamento e lo sviluppo di nuove molecole tende a ridurne l'impatto. Tra tutti gli elminti in grado di parassitare i ruminanti, i nematodi appartenenti al sottordine degli *Strongyloidea*, volgarmente designati sotto il nome generico di "strongili", sono il gruppo più numeroso, vario e maggiormente patogeno. Le strongilosi gastro-intestinali in particolare sono ben conosciute nei piccoli ruminanti, ma si tende generalmente a trascurarle nei Bovini (Euzeby 1969). Sebbene alcune forme di strongilosi possano dare origine a disturbi gravi, anche nel momento in cui non determinano manifestazioni cliniche imponenti, esse sono sempre causa di disturbi latenti (Euzeby 1969). In particolare questi nematodi possono interferire con il complesso processo digestivo del ruminante andando ad inficiare in maniera subdola le performances produttive. Nonostante queste ben note considerazioni, da molti anni sono particolarmente carenti le informazioni relative al parassitismo bovino nel nostro Paese. Esse sono così carenti da indurre nell'immaginario collettivo dei tecnici del settore il pensiero dell'inutilità di intervento diagnostico-terapeutico, se non limitato ai broutard. Da molti anni manca quindi nel panorama nazionale un tentativo di analisi a largo raggio che, sfruttando la disponibilità di un piccolo macello (sarebbe stato impossibile lavorare in una struttura ad elevata produttività), la puntuale identificazione dell'animale e della sua storia attraverso le informazioni dell'anagrafe bovina, fornisse una visione di insieme, la cui utilità potesse riverberarsi su tecnici ed allevatori. Dopo un'attenta rivisitazione della bibliografia internazionale sul parassitismo bovino, con questa indagine abbiamo cercato di raggiungere tale obiettivo aggiungendo al dato coprologico, per sua natura carente in sensibilità, quello necroscopico che ci ha permesso di isolare i più comuni parassiti abomasali. Per praticità abbiamo limitato l'indagine necroscopica a questo viscere, considerando che rappresenta il primo habitat che incontrano i nematodi dell'apparato digerente e rappresenta anche una delle prime stazioni in cui il processo digestivo ha inizio. L'abomaso può essere considerato inoltre un buon indicatore delle condizioni di parassitismo degli animali, sufficiente per indicare il livello di infestazione generale (Giannetto et al. 1998).

MATERIALI E METODI

La nostra indagine si è articolata in tre fasi successive, la raccolta dei campioni, la loro processazione e la successiva elaborazione dei dati. **Raccolta dei campioni**. La raccolta dei

campioni è stata possibile presso la tripperia di un piccolo macello della provincia di Bologna, la cui attività è principalmente finalizzata alla processazione di bovini (circa 400 capi al mese) a fine carriera. Ci è stato consentito di operare in questa struttura nel corso dell'anno 2015 raccogliendo complessivamente 100 abomasi e 427 campioni di feci, prelevate direttamente dall'ampolla rettale. La possibilità di consultare i registri del macello ci ha permesso di associare il codice identificativo di ciascun animale e risalire, attraverso la Banca Dati Nazionale (BDN), ad ulteriori informazioni relative a segnalamento e anamnesi del singolo bovino macellato. Tra queste: data di nascita, categoria produttiva di appartenenza, provincia e allevamento di provenienza, numero dei capi ivi presenti, data di inizio attività, numero di lattazioni e figli durante la carriera produttiva. I campioni, immediatamente dopo la raccolta, sono stati portati al laboratorio di Parassitologia e Malattie Parassitarie del Dipartimento di Scienze Mediche Veterinarie (DIMEVET) dell'Università di Bologna e qui conservati a ± 5 °C. **Processazione dei campioni.** Per l'analisi degli abomasi si è utilizzata la tecnica del "Total worm count" (Euzeby 1982) che ha permesso di isolare i nematodi presenti. Questi ultimi sono stati conservati in alcol 70°, glicerinato al 5%, e successivamente montati su vetrino, chiarificati e identificati utilizzando le chiavi tassonomiche del manuale MAFF-ADAS (1986). Per l'esame coprologico qualitativo ci siamo avvalsi della tecnica di sedimentazione e successiva flottazione, descritta da Euzeby (1982), completata dal riconoscimento delle uova attraverso le chiavi di Sloss e Kemp (1978). **Elaborazione statistica dei dati.** I risultati parassitologici sono stati inseriti in un foglio di calcolo assieme alle informazioni ricavate dalla BDN, la cui elaborazione si è ottenuta applicando la funzione software Microsoft Excel per la generazione di tabelle e grafici. Il test del chi quadrato (Epiinfo 3.5.1) è stato utilizzato per la validazione delle correlazioni eventualmente riscontrate.

RISULTATI

Complessivamente abbiamo sottoposto ad analisi 427 bovini provenienti da 31 province. Il 93% (397/427) degli animali erano femmine, di cui 63% da latte ed il 37% da carne, il 7% (30/427) erano maschi. Il range di età dei nostri soggetti variava da 1 a 26 anni. La distribuzione geografica del nostro campione è risultata indipendente dalla nostra volontà in quanto legata alle politiche commerciali del macello. L'abbiamo riassunta nella Figura 1 dove si evidenzia come la maggior parte dei campioni origini dal Centro Italia, con alcuni bovini provenienti dalla Puglia, dal Trentino Alto-Adige e dalla Sardegna. Le province "più generose" sono state Pesaro-Urbino

(66), Ferrara (54), Bologna (48), Perugia (40), Sassari (35) e Grosseto (29). La ricerca dei nematodi nei 100 abomasi ha permesso di individuarne 13 positivi per strongili gastro-intestinali. I generi identificati sono stati *Ostertagia sp*, *Trichostrongylus spp*. e *Cooperia spp*. L'esame coprologico ha consentito di identificare come positivi 31% degli animali. Nel confronto tra la positività coprologica e le diverse caratteristiche estrapolate dalla BDN, gli unici fattori che hanno beneficiato della significatività statistica ($p<0.01$) sono stati la categoria zootecnica a favore dei bovini da carne (22%) rispetto a quelli da latte (13%) (Figura 2), e le dimensioni dell'allevamento, che vede le classi intermedie (2 e 3) significativamente più parassitate (Figura 3).

Figura 1. Distribuzione geografica dei bovini sottoposti ad analisi.

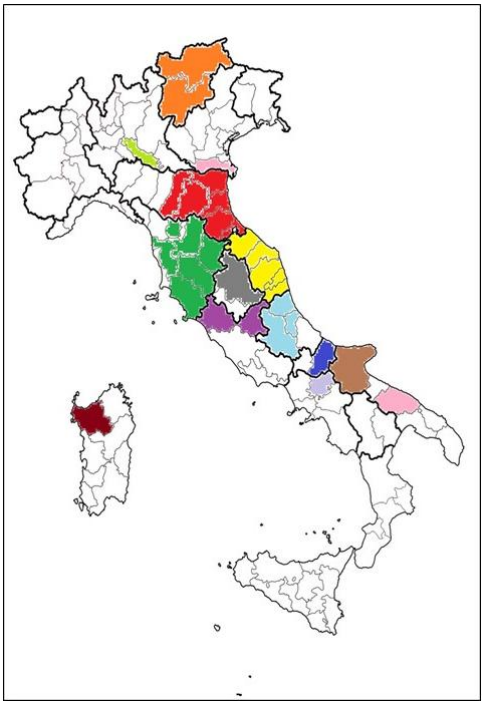


Figura 2. Distribuzione dei bovini positivi per categoria produttiva

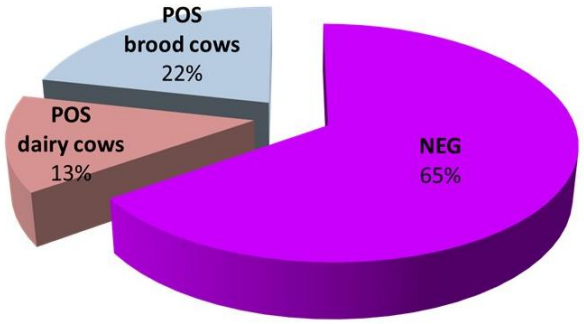
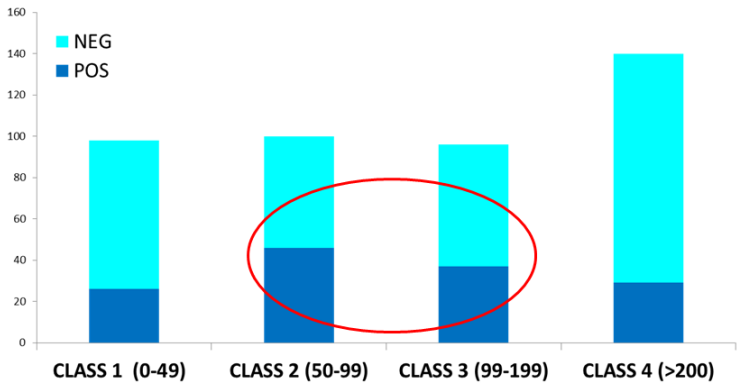


Figura 3. Distribuzione dei bovini postivi per dimensione in numero di capi dell'allevamento



DISCUSSIONE

I nostri risultati ci inducono a ritenere che il parassitismo da strongili dell'apparato digerente sia ancora ben rappresentato nei bovini allevati nel nostro Paese. Trattandosi di nematodi le cui uova non sono morfologicamente distinguibili all'esame coprologico, i risultati di quest'ultimo non possono essere correlati direttamente ai reperti anatomo-patologici limitati al solo abomaso; il loro habitat si estende lungo tutto l'apparato digerente e pertanto le uova da noi osservate non sono state necessariamente emesse a livello dello stomaco. La presenza di questi parassiti anche solo in quest'organo è risultata rilevante (13%) e vi sono rappresentati tre generi di riconosciuta patogenicità (*Ostertagia* sp., *Trichostrongylus* spp., *Cooperia* spp.). Anche la coprologia fornisce risultati oltremodo interessanti, quasi un terzo degli animali (31%) è risultato emettere uova e quindi presumibilmente albergare strongili gastro-intestinali. La significativa maggiore positività negli animali da carne è probabilmente da attribuire al fatto che questi con maggior frequenza usufruiscono di periodi di pascolo, riconosciuto fattore di rischio per le strongilosi che beneficiano di una fase subaerea a loro particolarmente favorevole. Più difficile da spiegare la significativa maggior positività riferita agli allevamenti di medie dimensioni; potremmo azzardare l'ipotesi che in questa tipologia la cura dell'igiene sia meno accurata in quanto subordinata ad altre attività che sostentano economicamente l'azienda agricola di cui l'allevamento bovino è solo una parte. Negli allevamenti di ridotte dimensioni il management igienico è sicuramente più semplice da realizzare, mentre in quelli di grandi dimensioni (>200) l'attività allevatoriale è quella preponderante nell'azienda e vi si dedica maggiore attenzione.

CONCLUSIONE

Prescindendo dal dato puramente parassitologico, l'avere lavorato in una struttura che processa animali adulti di diverse provenienze, usufruendo della BDN, ci ha fornito un quadro della situazione zootecnica che appare ancora molto tradizionale con allevamenti anche di ridotte dimensioni e un'età degli animali che agli occhi di un attento buiatra potrebbe apparire anti-economica. In questa realtà gli strongili gastro-intestinali sono presenti, sicuramente producono danni e altrettanto sicuramente necessiterebbero di maggiore attenzione da parte di tecnici e allevatori. Il mattatoio, oltre a garantire salubrità alle carni, se correttamente utilizzato, dimostra ancora una volta di possedere un'importante valenza come osservatorio epidemiologico.

BIBLIOGRAFIA E SITOGRAFIA

Euzeby J. 1969. “Caratteri morfologici, biologici e fisiopatologici degli strongili gastrointestinali dei bovini” in Atti del convegno “Le strongilosi gastro-intestinali dei bovini in Piemonte” Cinzano 27 Settembre 1969, pp.15-62.

Euzeby J. 1982. “Helminthes gastro-intestinaux des ruminants” In “Diagnostic expérimental des helminthoses animales (animaux domestiques-animaux de laboratoire-primates). Travaux pratiques d’Helminthologie Vétérinaire. Livre 2. Informations Techniques des Services Vétérinaires, Ministére de l’Agriculture, Paris: 51-157.

Giannetto S, Virga A, Poglayen G, Ferlazzo M. 1998. Fauna elmintica gastro-intestinale in ovini della Sicilia Orientale. Atti XIII Congresso Nazionale S.I.P.A.O.C. Palermo 16-19 aprile 1998, pp. 264-266.

MAFF-ADAS Manual of Veterinary Parasitological Laboratory Techniques. 1986. HMSO Publication Centre, London.

Sloss MW, Kemp RL. 1978. Veterinary Clinical Parasitology. Fifth Edition. The Iowa State University Press, Ames, USA.

http:// www.statistiche.izs

This research has been published in the Journal "Large Animal Review" 22: 245-248.

Il mattatoio come osservatorio epidemiologico, un aspetto negletto. Aggiornamento sugli endoparassiti bovini in Italia

C. ROMANELLI[1], G.P. ZAFFARANO[1], B. MORANDI[1], A. FIOCCHI[1], V. BENFENATI[2], G. POGLAYEN[1]

[1] Dipartimento di Scienze Mediche Veterinarie, Alma Mater Studiorum-Università di Bologna
[2] A.S.L. Bologna, Via Altura 3, 40139 Bologna

RIASSUNTO

Scopo dell'indagine è stato quello di fornire dati aggiornati sulla prevalenza e la diffusione di infestazioni da nematodi gastrointestinali in bovini adulti allevati in Italia. Lo studio ha avuto inizio con la raccolta di 427 campioni di feci da bovini processati in un macello della provincia di Bologna. Questi, prelevati da singoli animali, sono stati analizzati in laboratorio mediante esame coprologico qualitativo. Dagli stessi animali sono stati scelti a caso 100 abomasi, esaminati poi al tavolo necroscopico per valutare la presenza di forme adulte di nematodi. Uova di nematodi gastrointestinali sono state individuate nel 31% dei campioni di feci esaminati. Negli abomasi è stata rilevata una positività del 13% per elminti gastro-intestinali. Il loro isolamento ha permesso di identificarli come appartenenti ai generi *Ostertagia*, *Trichostrongylus* e *Cooperia*. Sulla base di indagini statistiche l'eliminazione di uova di elminti gastro-intestinali è risultata associata in maniera significativa sia alla categoria produttiva di appartenenza sia alle dimensioni in numero di capi dell'allevamento. Il presente studio dimostra che il parassitismo gastro-intestinale da nematodi è un problema che deve essere considerato diffuso nei bovini allevati in Italia, con livelli di prevalenza non trascurabili. Tuttavia sembra essere ancora sottostimato da parte dei tecnici del settore.

PAROLE CHIAVE

Bovini, parassiti, strongili gastro-intestinali, mattatoio.

INTRODUZIONE

Il patrimonio bovino nazionale ammonta a 5535696 animali[1]. I parassiti che colpiscono questa specie sono numerosi, anche se l'evoluzione di un certo tipo di allevamento e lo sviluppo di nuove molecole tende a ridurne l'impatto. Tra tutti gli elminti in grado di parassitare i ruminanti, i nematodi appartenenti al sottordine degli *Strongyloidea*, volgarmente designati sotto il nome generico di "strongili", sono il gruppo più numeroso, vario e maggiormente patogeno. Le strongilosi gastro-intestinali in particolare sono ben conosciute nei piccoli ruminanti, ma si tende generalmente a trascurarle nei Bovini[2]. Sebbene alcune forme di strongilosi possano dare origine a disturbi gravi, anche nel momento in cui non determinano manifestazioni cliniche imponenti, esse sono sempre causa di disturbi latenti[2]. In particolare, questi nematodi possono interferire con il complesso processo digestivo del ruminante andando ad inficiare in maniera subdola le performances produttive. Nonostante queste ben note considerazioni, da molti anni sono particolarmente carenti le informazioni relative al parassitismo bovino nel nostro Paese. Esse sono così carenti da indurre nell'immaginario collettivo dei tecnici del settore il pensiero dell'inutilità di intervento diagnostico-terapeutico, se non limitato ai broutard. Da molti anni manca quindi nel panorama nazionale un tentativo di analisi a largo raggio che, sfruttando la disponibilità di un piccolo macello (sarebbe stato impossibile lavorare in una struttura ad elevata produttività), la puntuale identificazione dell'animale e della sua storia attraverso le informazioni dell'anagrafe bovina, fornisse una visione di insieme, la cui utilità potesse riverberarsi su tecnici ed allevatori. Dopo un'attenta rivisitazione della bibliografia internazionale sul parassitismo bovino, con questa indagine abbiamo cercato di raggiungere tale obiettivo aggiungendo al dato coprologico, per sua natura carente in sensibilità, quello necroscopico che ci ha permesso di isolare i più comuni parassiti abomasali. Per praticità abbiamo limitato l'indagine necroscopica a questo viscere, considerando che rappresenta il primo habitat che incontrano i nematodi dell'apparato digerente e rappresenta anche una delle prime stazioni in cui il processo digestivo ha inizio. L'abomaso può essere considerato inoltre un buon indicatore delle condizioni di parassitismo degli animali, sufficiente per indicare il livello di infestazione generale[3].

Autore per la corrispondenza:
Giovanni Poglayen (giovanni.poglayen@unibo.it).

MATERIALI E METODI

La nostra indagine si è articolata in tre fasi successive, la raccolta dei campioni, la loro processazione e la successiva elaborazione dei dati.

Raccolta dei campioni

La raccolta dei campioni è stata possibile presso la tripperia di un piccolo macello della provincia di Bologna, la cui attività è principalmente finalizzata alla processazione di bovini