SHORT COMMUNICATION

Survey on the role of brown hares (*Lepus europaeus*, Pallas 1778) as carriers of zoonotic dermatophytes

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Abstract

The occurrence of dermatophytes and keratinophilic fungi was investigated by hair-brush technique on the coat of 986 apparently healthy brown hares (*Lepus europaeus*, Pallas 1778) caught in 9 restocking and capture zones in Central Italy. Overall, 7.5% hair samples gave positive results. *Trichophyton terrestre* (2.1%), *Chrysosporium* sp, *Chrysosporium keratinophilum*, *Microsporum gypseum*, *Trichophyton gloriae* and *Trichophyton mentagrophytes* (0.6% each), *Trichophyton erinacei* and *Scopulariopsis brevicaulis* (0.4% each), *Chrysosporium asperatum* (0.3%), *Arthroderma* sp and *Microsporum canis* (0.1% each) were identified in cultures with single isolates, whereas *Chrysosporium* sp/*T. mentagrophytes* (0.3%), *Chrysosporium* sp/*T. terrestre* and *M. gypseum*/*T. terrestre* (0.2% each), *Chrysosporium tropicum*/*T. terrestre*, *M. canis*/*T. terrestre* and *T. ajelloi*/*T. terrestre* (0.1% each) were identified in cultures with mixed isolates. *T. erinacei* and *M. canis* have not previously been isolated from hares. *M. canis*, T. *erinacei* and *T. mentagrophytes* were the most clinically important dermatophytes found. Altogether, they were isolated only from 1.5% hair samples. Thus, it is concluded that brown hares may play a limited epidemiological role as carriers of zoonotic dermatophytes.

Nevertheless, this should be taken into consideration as many people may be exposed to zoonotic agents from brown hares during hunting and trapping activities.

Introduction

The Brown Hare or European Hare (*Lepus europaeus*, Pallas 1778), also known as Common Hare, is an indigenous species to Italy. European hares have naturally expanded throughout most of Europe as well as through the Middle East to Central Asia. In addition, they have been introduced extensively by humans to several other countries and have become one of the most important game species throughout the world due to their highly appreciated meat. Keratinophilic fungi are a group of molds that colonize various keratinous substrates and degrade them to components of low molecular weight. Many keratinophilic fungi are common saprophytes in soil such as *Chrysosporium* species and *Scopulariopsis brevicaulis*, though they can be found as contaminants on keratinous tissues, including skin, nails, feathers and hair. Some species share certain characteristic features, constituting a special group called dermatophytes. Most species of dermatophytes have strict parasitic aptitude towards animals (zoophilic), man (anthropophilic) or both (zoonotic) and are considered as true dermatophytes such as *Microsporum canis*, *Trichophyton erinacei* and *Trichophyton mentagrophytes*. Others are soil saprophytes with different parasitic aptitude and are indicated as geophilic dermatophytes such as *Microsporum gypseum*, *Trichophyton ajelloi* and *Trichophyton terrestre*. Over the last years, an increasing number of zoophilic dermatophytes have been recognized as agents of skin infections in humans causing clinical lesions with greater frequency than anthropophilic dermatophytes (Tampieri *et al*., 2006).

In recent years, wildlife is emerged as a large and often overlooked source of zoonotic diseases (Deutz *et al*., 2003; Lévesque *et al.*, 2007). Many studies gathered a wealth of scientific information on biology (Paci *et al*., 2007), management (Pelorosso et al., 2008), and health status of European hare populations (Wibbelt and Frolich, 2005), including attention to agents which may have an impact on human health (Treml *et al*., 2007). However, data concerning the carriage of zoonotic dermatophytes are very few. In Italy the studies on the epidemiology of dermatophytes in domestic animals are scarce (Cafarchia *et al*., 2004) and only two reports have been published on hares (Mantovani *et al.*, 1982; Zanni *et al*., 1995). As the epidemiology of dermatophytes may be changing overtime, it is important to review periodically their prevalence and distribution.

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Therefore, the aim of this study was to report about the role of brown hares as carriers of dermatophytes in Italy during a 5-year period.

Materials and methods

Study area and sampling design

According to the Italian legislation, the regional administration of Tuscany (Central Italy) has established the creation of restocking and capture zones in each province to manage the activities of game hunting in the territory. These special protected areas, where hunting is prohibited, aim to allow the population growth of wild game species. In our reality, restocking and capture zones are mainly used for the growth of brown hare populations. Once a year (usually between December and January), many hares are captured alive by trammel nets and then relocated in hunting territories.

A total of 986 apparently healthy European hares (*Lepus europaeus*, Pallas 1778) were examined for the presence of dermatophytes and related keratinophilic fungi on their coats. Data collected from each animal included age and sex. Animals were classified as juveniles (≤9-month-old) or adults (>9-month-old) according to the presence of the Stroh's tubercle (Broekhuizen and Maaskamp, 1979). There were 719 adults and 267 juveniles as well as

499 females and 487 males. Between January 2003 and January 2008, they were caught from 9 restocking and capture zones with different habitat characteristics in the province of Pisa, expanding from 429 to 1,152 hectares (mean ± standard deviation=862±267). As soon as possible after trapping, animals were selected randomly and sampled by brushing each over the face and neck as well as the dorsal, lateral and ventral surfaces of the body using a small sterilized plastic hair brush (Mancianti *et al*., 1993, 2002). Then they were placed in darkened, wooden capture-boxes, transferred and released in hunting areas.

Identification of isolates

Samples were seeded on Petri dishes with Mycobiotic agar (Difco Laboratories, Detroit, USA), incubated at 25°C, and daily inspected for fungal growth up to 4 weeks. Genera and species of dermatophytes and keratinophilic fungi were identified as previously described (Mancianti *et al*., 1993, 2002).

Analysis of results

Prevalence of dermatophytes and keratinophilic fungi was determined as the number of positive samples/total number of samples x 100. Differences in prevalence rates were compared according to sex and age using the χ^2-test. P values ≤0.05 were considered significant.

Results and discussion

Overall, 74 out of 986 (7.5%) brown hares were carrying dermatophytes and/or keratinophilic fungi on their coats. Altogether, 13 fungal species were identified. In order of frequency, the number of single isolates obtained in cultures was the following: 21 isolates of *Trichophyton terrestre* (2.1%), 6 each of *Chrysosporium* sp, *Chrysosporium keratinophilum*, *M. gypseum*, *Trichophyton gloriae* and *T. mentagrophytes* (0.6%), 4 each of *T. erinacei* and *S. brevicaulis* (0.4%), 3 of *Chrysosporium asperatum* (0.3%), 1 each of *Arthroderma* sp and *M. canis* (0.1%). In some cases, two isolates were detected in cultures from single hair samples. These included 3 (0.3%) cases with *Chrysosporium* sp/*T. mentagrophytes*, 2 (0.2%) with *Chrysosporium* sp/*T. terrestre* or *M. gypseum*/*T. terrestre* and 1 (0.1%) with *Chrysospo-rium tropicum*/*T. terrestre*, *M. canis*/*T. terrestre* or *Trichophyton ajelloi*/*T. terrestre*.

When statistical analysis of results by the χ^2-test was carried out, it was evident that males were more likely (P=0.0115) to harbour dermatophytes and keratinophilic fungi on their coat in comparison to females (47/487, 9.6% *vs* 27/499, 5.4%). With respect to age, the prevalence rate was higher in juveniles (26/267, 9.7%) than in adults (48/719, 6.7%) but this difference did not reach a statistically significant value (P=0.1049). Probably, males may be associated with a significantly higher prevalence rate of dermatophytes and keratinophilic fungi because of their habits. Indeed, male hares generally live in a wider territory covering the territories of several females and fight each other often during the breeding season. Thus have increased possibility to find sources of fungal infection and to transmit mycotic agents to each other by direct contact.

From a public health perspective, *M. canis*, *T. erinacei* and *T. mentagrophytes* were the most clinically important dermatophytes found, as they are commonly reported to cause *Tinea corporis*, *Tinea barbae*, *Tinea cruris* or *Tinea capitis* in man, whereas the other keratinophilic species isolated are less frequently associated with disease in humans, mostly in cases of immunodepression or other underlying pathologies. Altogether, *M. canis*, *T. erinacei* and *T. mentagrophytes* were isolated from 15 out of 986 (1.5%) hair samples. The present range and prevalence of medically relevant dermatophytes are slightly greater than previously reported. Indeed, *T. mentagrophytes* was the only clinically important dermatophyte species isolated from 1 out of 415 (0.2%) and 3 out of 270 (1%) brown hares, respectively (Mantovani *et al*., 1982; Zanni *et al*., 1995). This difference in range and prevalence is probably due to the larger number of animals sampled in our survey.

T. mentagrophytes is known to parasite almost permanently the coat of rodents, small mammals and lagomorphs. The finding of *M. gypseum* was probably due to contamination from soil, its basic substrate. *T. mentagrophytes* and *M. gypseum* have previously been reported in brown hares (Mantovani *et al*., 1982). Thus, their isolation was to be expected, whereas the occurrence of *T. erinacei* and *M. canis* was rather unexpected.

To the best of authors' knowledge, these two zoophilic dermatophytes with high zoonotic potential have not previously been isolated from hares. *T. erinacei* is commonly associated with hedgehogs. Its isolation was probably due to coat contamination with soil infected by hedgehogs shedding skin scales and sharing the same environment with hares. *M. canis* has been rarely found in wildlife, though it has been reported in asymptomatic foxes (Mancianti *et al*., 1993), marmots (Gallo *et al.*, 2005a) and eastern cottontails (Gallo *et al.*, 2005b). It is mostly associated with urban environment as cats are its natural reservoir and can be easily transmitted to dogs, rabbits, fur animals and mostly humans. Foxes, dogs and cats are amongst the main predators on brown hares (Haerer *et al*., 2001; Schmidt *et al*., 2004). M. canis has been reported in foxes, dogs and cats from Central Italy (Mancianti *et al.*, 1993, 2002). Thus, it is likely that infected predators' attacks may have favoured the spread of *M. canis* to brown hares.

Conclusions

As only 1.5% of animals examined in this study were carrying dermatophytes of medical importance on their coat, it is concluded that brown hares play a limited epidemiological role as carriers of zoonotic dermatophytes.

Nevertheless, this should be taken into consideration as many hunters, wildlife operators and researchers may be especially exposed to zoonotic pathogens touching and handling brown hares, dead or alive, during hunting and trapping activities (Deutz *et al*., 2003; Lévesque *et al*., 2007).

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