

Occurrence of ticks in the subcutaneous tissue of red foxes, *Vulpes vulpes* in Czech Republic and Romania



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ABSTRACT

An extensive survey of parasites in red foxes (*Vulpes vulpes*) was independently conducted in Romania and the Czech Republic. Carcasses were examined by necropsy, and small, dark nodules apparently containing ticks were noticed in the subcutaneous tissue of several foxes. Histopathological examination was performed using hematoxylin and eosin (HE) staining. Of the 91 foxes examined from the Czech Republic, 14 (15.4%) were harboring ticks in the subcutaneous tissue. In the majority of these cases, 1–3 nodules/fox were found, with a maximum of 31 nodules/fox. In Romania a single examined fox had subcutaneous ticks. All ticks collected from subcutaneous tissue were partially engorged adults. Based on morphological features, *Ixodes ricinus*, *I. hexagonus*, *I. crenulatus* and *Dermacentor reticulatus* were identified. The histopathological examination revealed chronic granulomatous panniculitis with peripheral fibrosis and intralesional presence of the ticks. Only few data are available regarding ticks localized in the subcutaneous tissue of any host. All the ticks were dead or already decomposed and it is evident that subcutaneous location does not represent an evolutionary advantage, as the detachment and finishing the life cycle is impossible.

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1. Introduction

Ticks are blood-feeding arthropods, parasitizing on a wide range of vertebrate species, typically firmly attached on the host skin surface (Sonenshine, 1991). Nevertheless, a report published more than a century ago in United Kingdom (Nuttall, 1914) and few later reports in Poland (Drozd, 1958), Austria (Hinaidy, 1976), Slovakia (Lebeda, 1962; Tovornik, 1984; Pet'ko and Stanko, 1991) and United States of America (Smith et al., 1986) mention the presence of ticks in the subcutaneous tissues of red foxes (*Vulpes vulpes*). Moreover, one case of subcutaneous ticks was recently recorded in a domestic dog from Sweden (Zakrisson, 2010) and in a child from South Korea

(Chang et al., 2006). All these studies represent only isolated reports and do not provide any data on the frequency of this phenomenon or associated pathology. The aim of our study was to evaluate how commonly the ticks do occur in the subcutaneous tissues of red foxes in Czech Republic, to identify the diversity of tick species being able to reach this location and, in one fox from Romania, to describe the lesions in order to provide data for understanding the underlying mechanism of subcutaneous localization.

2. Materials and method

During an extensive survey on the parasites of red foxes performed in the Czech Republic, 91 carcasses were collected between June 2014 and February 2015 all over the country, and examined by full necropsy. In several cases, after removing the skin of foxes (as a routine method for the detection of subcutaneous filariae), small,

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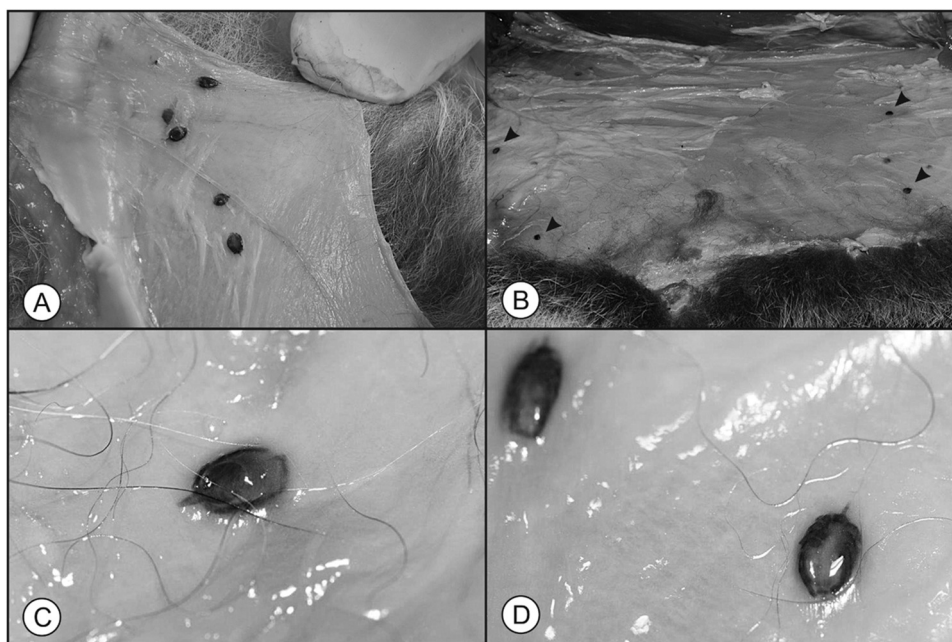


Fig. 1. The macroscopic aspect of the nodules containing partially engorged adult ticks in various stages of decompositions. A. General aspect in a red fox from Czech Republic. B. Multiple nodules (arrowheads) in a red fox from Romania. C. Partially engorged *Ixodes ricinus* female in a red fox from Romania in ventral position. D. Partially engorged female ticks (unidentified species) in a red fox from Romania in dorsal position.

dark nodules apparently containing ticks were noticed. The prevalence (foxes with subcutaneous ticks) and the intensity (number of subcutaneous ticks per fox) were calculated. The nodules were dissected and the ticks were removed. Identification of the subcutaneous ticks was possible only in four foxes due to the advanced state of degradation of the arthropods; ticks were identified using morphological criteria (Nosek and Sixl, 1972). Following the findings in Czech Republic, for the only purpose of describing the lesions, the same procedure was used for a single road killed fox from Romania (Corușu, Cluj County, Romania) delivered to the Department of Parasitology and Parasitic Diseases of the Faculty of Veterinary Medicine in Cluj-Napoca. No prevalence studies were performed in this country due to lack of sufficient number of samples.

In order to evaluate the microscopical characteristics of the lesions, skin pieces along with surrounding subcutaneous tissue containing ticks were collected and immediately fixed in 10% neutral buffered formalin and routinely processed for paraffin wax embedding. From the paraffin block, multiple 4 μ m thick sections were cut with a rotary microtome (Leica RM2125 RT) and stained with hematoxylin and eosin (HE). Histological slides were examined under an Olympus BX41 optical microscope equipped with an Olympus UC30 digital camera. Images were collected and processed using Olympus Stream Basic image analysis software.

3. Results

The prevalence of the subcutaneous nodules in foxes collected in the Czech Republic was 15.4% (14/91, 95% confidence interval: 8.7–24.5). As in Romania only a single fox was examined, no prevalence data is provided. Most of the nodules were located in the subcutaneous tissue on the dorsal part of the body (Fig. 1A–B) and contained dead ticks in various stage of decomposition (Fig. 1C–D). The number of nodules per fox varied between 1 and 31 (mean intensity: 4.6). The gross appearance of the skin revealed the absence of any external epidermal lesions at the site of the subcutaneous nodules. Nine nodules containing ticks were collected from the Czech foxes; eight ticks were partially engorged adults of the

following species: *Ixodes ricinus* (1 female and 1 male), *I. hexagonus* (2 females and 1 male), *I. crenulatus* (1 female and 1 male) and *Dermacentor reticulatus* (1 female); one tick was not identifiable due to extensive degradation. In the Romanian fox, of the 3 nodules found, only one contained a morphologically identifiable tick (female of *I. ricinus*).

Histological examination (Fig. 2) revealed granulomatous panniculitis with peripheral fibrosis and the intralesional presence of ticks, with certain structures still distinguishable (Fig. 2A–D). The inner part of the tick contained a mixture of cell debris, cholesterol crystal clefts, macrophages and neutrophils. The cuticle of the tick was surrounded by inflammatory reaction consisting of macrophages, epithelioid, multinucleated, giant cells neutrophils and sparse eosinophils (Fig. 2E–F). No blood meal traces were observed during the histopathological examination.

4. Discussion

Red foxes are important hosts of ticks all over Europe, with at least 16 tick species known to occur on foxes, with regional differences among tick fauna (Ross and Fairley, 1969; Sréter et al., 2003; Dominguez, 2004; Martínez-Carrasco et al., 2007; Lorusso et al., 2011; Sobrino et al., 2012). There are three species of ticks (*I. canisuga*, *I. hexagonus*, *I. ricinus*) which are commonly found on foxes in most studied regions (Dominguez 2004; Martínez-Carrasco et al., 2007). Of these, we reported two species (*I. hexagonus* and *I. ricinus*) occurring in subcutaneous tissue.

Most of the previously published data in carnivores are isolated case reports of subcutaneous *I. ricinus* in red foxes in various parts of Europe (Nuttall, 1914; Drozd, 1958; Lebeda, 1962; Hinaidy, 1976; Pet'ko and Stanko, 1991). Additionally, a fox harboring a subcutaneous *Amblyomma americanum* was reported in USA (Smith et al., 1986). None of these reports provide information on the extent of the occurrence of ticks under the skin of their hosts. The prevalence of subcutaneous ticks was 15.4% of examined foxes in our study, suggesting the frequent occurrence of this phenomenon in red foxes from this part of Europe. In addition to the records of *I. ricinus* reported previously, our study identified a variety of tick

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