Ticks and Tick-borne Diseases xxx (xxxx) xxx–xxx

Contents lists available at ScienceDirect

Ticks and Tick-borne Diseases

journal homepage: www.elsevier.com/locate/ttbdis

Original article

Distribution of *Ixodes ricinus* ticks and prevalence of tick-borne encephalitis virus among questing ticks in the Arctic Circle region of northern Norway

A. Soleng[a,*], K.S. Edgar[a,1], K.M. Paulsen[b,c,1], B.N. Pedersen[b], Y.B. Okbaldet[b], I.E.B. Skjetne[a], D. Gurung[b], R. Vikse[b], Å.K. Andreassen[b]

[a] Norwegian Institute of Public Health, Division for Infection Control and Environmental Health, Department of Pest Control, PO-Box 4404 Nydalen, NO-0403 Oslo, Norway

[b] Norwegian Institute of Public Health, Division for Infection Control and Environmental Health, Department of Virology, PO-Box 4404 Nydalen, NO-0403 Oslo, Norway

[c] Norwegian University of Life Sciences, Faculty of Veterinary Medicine, Department of Production Animal Clinical Sciences, Ullevålsveien 72, NO-0454 Oslo, Norway

ARTICLE INFO

Keywords:
Ixodes ricinus
Hard ticks
TBE
TBEV
Geographical distribution
Tick-borne diseases
Real-time PCR

ABSTRACT

This study investigated the geographical distribution of *Ixodes ricinus* and prevalence of the tick-borne encephalitis virus (TBEV) in northern Norway. Flagging for questing *I. ricinus* ticks was performed in areas ranging from Vikna in Nord-Trøndelag County, located 190 km south of the Arctic Circle (66.3°N), to Steigen in Nordland County, located 155 km north of the Arctic Circle. We found that ticks were abundant in both Vikna (64.5°N) and Brønnøy (65.1°N). Only a few ticks were found at locations ~66°N, and no ticks were found at several locations up to 67.5°N. Real-time PCR (RT-PCR) analyses of the collected ticks (nymphs and adults) for the presence of TBEV revealed a low prevalence (0.1%) of TBEV among the nymphs collected in Vikna, while a prevalence of 0% to 3% was found among nymphs collected at five locations in Brønnøy. Adult ticks collected in Vikna and Brønnøy had higher rates of TBEV infection (8.6% and 0%–9.0%, respectively) than the nymphs. No evidence of TBEV was found in the few ticks collected further north of Brønnøy. This is the first report of TBEV being detected at locations up to 65.1°N. It remains to be verified whether viable populations of *I. ricinus* exist at locations north of 66°N. Future studies are warranted to increase our knowledge concerning tick distribution, tick abundance, and tick-borne pathogens in northern Norway.

1. Introduction

The geographical distribution of castor bean ticks (*Ixodes ricinus*) in Norway has been previously examined in several studies. An extended survey by Tambs-Lyche (1943) found *I. ricinus* to be distributed in the coastal areas of Norway, from southeastern Østfold County, along the southern and western coastline, up to Nordland County at ~66°N. Tambs-Lyche (1943) reported that the number of ticks varied among different locations. In the late 20th century, Mehl (1983) reported finding no major changes in the geographical distribution of ticks when compared with data reported by Tambs-Lyche (1943). Nevertheless, he observed an increased density of tick populations in certain parts of the distribution range. Although Mehl (1983) argued that *I. ricinus* was mainly distributed along the coast, ticks were also found distant from the coast and at elevations as high as 800 m above sea level. Furthermore, Mehl (1983) reported that *I. ricinus* were found in scattered foci in the periphery of their range, and that ticks found outside of their normal range were probably transported to those locations by migratory birds or larger mammals. Jore et al. (2011) suggests that tick populations in Norway have undergone recent shifts in latitudinal and altitudinal range. That study was based on confirmed cases of borreliosis, veterinary surveys, and tick registration maps published in public newspapers that showed tick populations extending to ~69°N. However, Jenkins et al. (2012) found only few ticks attached to dogs and cats in a region north of Brønnøy at 66°. Hvidsten et al. (2014) studied the distribution of ticks in northern Norway, and concluded that further studies are needed to clarify if tick populations are established north of the Arctic Circle.

Hard ticks similar to *I. ricinus* are considered important vectors for both human and animal diseases (Granström, 1997; Parola and Raoult, 2001; Charrel et al., 2004). Questing ticks from Brønnøy in northern Norway have been examined for the presence of *Borrelia burgdorferi* sensu lato and *Anaplasma phagocytophilum* (Soleng and Kjelland, 2013). Jenkins et al. (2012) and Hvidsten et al. (2014) examined fully engorged female ticks collected from dogs and cats in northern Norway, and found the same prevalence of *Borrelia* as in ticks collected in

* Corresponding author.
E-mail address: arnulf.soleng@fhi.no (A. Soleng).
[1] These authors equally contributed to the study.

http://dx.doi.org/10.1016/j.ttbdis.2017.10.002
Received 11 May 2017; Received in revised form 29 August 2017; Accepted 2 October 2017

southern Norway. Information concerning the prevalence of other tick-borne pathogens in northern Norway is very limited.

Tick-borne encephalitis (TBE) is a disease caused by the tick-borne encephalitis virus (TBEV) which belongs to *flavirus* genus, and has been a growing public health concern in Europe and other parts of the world for the past 20 years (Kunze, 2016). TBE is the single most important viral tick-borne disease in Europe, although only 33% of individuals infected with TBEV exhibit symptomatic disease (Kaiser, 2012). Three different subtypes of TBEV (the European, Siberian, and Far Eastern subtypes) exist in Europe (Ecker et al., 1999), and the European subtype is found in Norway (Ecker et al., 1999; Andreassen et al., 2012). While ticks are considered both vectors and reservoirs for TBEV (Lindquist and Vapalahti, 2008), humans can also acquire TBE by consuming unpasteurized milk and cheese (Mansfield et al., 2009). Various species of mammals and migratory birds also play an important role in the transmission and distribution of TBEV (Labuda and Nuttall, 2004; Jaenson et al., 2012). Human cases of TBE are highly uncommon in Norway. According to the Norwegian Surveillance System for Communicable Diseases (MSIS), only 106 cases of TBE were reported during the years 1994–2016, giving an incidence of 0.2 per 100,000 inhabitants. To date, human TBE cases have only been reported in the coastline regions of southern Norway. However, the disease might be underdiagnosed, as TBEV has been found in questing ticks gathered from areas of Norway with no known human TBE cases (Larsen et al., 2014; Paulsen et al., 2015).

The present study was conducted for two purposes: (1) to identify the northern geographical distribution of *I. ricinus* in Norway and (2) to examine the prevalence of TBEV found in questing ticks in this area.

2. Materials and methods

Questing *Ixodes ricinus* were collected by flagging vegetative undergrowth with a white terry towel (90 × 60 cm) attached to a wooden stick. Only nymphs and adult ticks were collected, and tick larvae were excluded from the study. Flagging was performed at 17 different locations situated south and north of the Arctic Circle at 66.3°N (Fig. 1, Tables 1 and 2). All sampling was done during daytime hours (10 am–4 pm) except in 2017 when flagging/dragging in Geitvågen and Saltstraumen (see Table 2) were performed as late as 6 pm and 8–9 pm, respectively. The habitat vegetation was dry unless otherwise stated (Tables 1 and 2). In 2017 both flagging (described above) and dragging with a flannel cloth (105 × 115 cm) was performed. The vegetation consisted of small deciduous trees and various types of undergrowth such as grass, herbs, ferns, and heather – a typical habitat for ticks. Numerous rodent burrows, rodent runways, bedding sites, and cervid tracks were observed in many of the locations. The total flagging times in areas where no ticks were found ranged from 45 min to 2 h, but sampling continued for several hours at locations where ticks were present.

Nymphs and adults attached to the sampling cloth were gently removed with tweezers and placed into small plastic microtubes (Axygen; Union City, CA, USA). Tubes with ticks were placed into double plastic zip-lock bags and kept on crushed ice in a cooler during transportation to the laboratory; after which, they were stored in an ultra-freezer at −80 °C until analysis. The ticks from each sampling site were counted and sorted by life stage on blocks of frozen CO_2 prior to analysis. Adult ticks were separated individually by sex, and nymphs were randomly sorted in groups of ten. A randomly selected subpopulation consisting of 429 adult ticks and 1830 nymphs was analysed for presence of TBEV by RT-PCR, and the results were confirmed by pyrosequencing, as previously described by Andreassen et al. (2012) and Paulsen et al. (2015).

The estimated prevalence calculations were based on confirmed positive results as verified by pyrosequencing. Numerical values for the estimated prevalence, minimum infection rate (MIR), and estimated pooled prevalence (EPP), were calculated as previously described (Andreassen et al., 2012).

3. Results

High tick densities, comparable to the southern parts of Norway, was found in the southernmost locations at Vikna (64.5°N) and Brønnøy (65.1–65.2°N) where 4–5 people collected more than 750 nymphs in two to three hours. However, only a few ticks were found at locations just north of Brønnøy at Alstahaug (65.5°N) and Dønna (66.1°N) after several hours of flagging, and no ticks were found further north up to 67.6°N. All locations north of the Arctic Circle was examined a second time by flagging and dragging in 2017, and no ticks were found. At Torghatten 1 in Brønnøy, we found a remarkably high number of adult ticks when compared with nymphs (Table 1).

TBEV was detected in questing tick nymphs collected in both Vikna and Brønnøy. A low prevalence of TBEV (0.1%) was found in Vikna, while the prevalences at five locations in Brønnøy ranged from 0% to 3% (Table 3). The prevalence of TBEV among adult ticks was higher than that among nymphs (i.e., 8.6% among adult ticks in Vikna and 0%–9.0% among adult ticks in Brønnøy) (Table 4). Note that the number of sampled ticks varies and are low and not representative at some locations. No TBEV was detected in the few ticks collected further north in Alstahaug and Dønna (Tables 3 and 4).

4. Discussion

In this study, we found a geographic distribution of *I. ricinus* that was similar to those reported by Tambs-Lyche (1943) and Mehl (1983). There is generally a high density of ticks in suitable habitats along the Norwegian coastline extending from the border with Sweden in the southeast and northwards up to 65.3°N (pers. obs.). In the present study, only a few ticks were found in areas located at 66.1°N, and no ticks were found

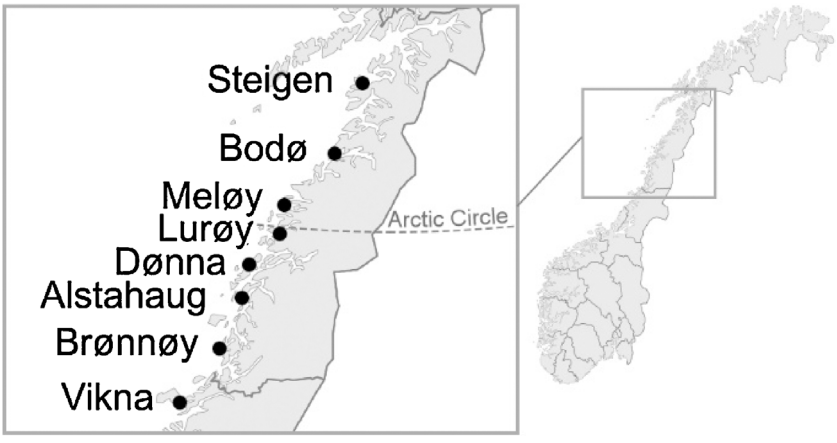

Fig. 1. Geographical locations of the sampling sites in northern Norway, as shown on a map created by Creative Commons Attribution ShareAlike 3.0 from Kartverket.