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Other

# An Emerging Role for Ticks as Vectors of Tularaemia in Sweden

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**Keywords:** cat | climate change | *Francisella tularensis* | hare | northern hemisphere | tularaemia

## Abstract

**Background:** The zoonotic bacterium *Francisella tularensis*, the causative agent of tularaemia, can be transmitted to humans via multiple routes, including through contact with infected animals, contaminated water or arthropod vectors. Ticks have not previously been described as transmitting the disease in Sweden. Recently, Ixodid tick species have expanded their latitudinal and altitudinal range in Sweden to areas where the disease is endemic. Tularaemia is a cause of growing concern, spreading to new areas in Sweden and infecting hares and humans.

**Objectives:** To establish whether ticks could be a potential arthropod vector in the transmission of *F. tularensis* subsp. *holarctica* in Sweden.

**Methods:** Ticks were collected from northern Sweden and screened for *F. tularensis*. A follow-up study with ticks collected from *F. tularensis*-positive hares was performed. Ticks were analysed using real-time PCR and a pathological examination was performed on the hares.

**Results:** *F. tularensis* subsp. *holarctica* was identified in ticks from one cat and three *F. tularensis*-infected hares. Two hares had skin lesions associated with tick bites with intralesional *F. tularensis* bacteria.

**Conclusions:** *F. tularensis* subsp. *holarctica* was isolated from ticks collected from the hares and cat, the first such reports in ticks in Sweden. Identification of the bacteria at the tick bite site and the more chronic character of the skin lesions compared to those of inner organs suggest that the ticks infected the hares. The cat showed no clinical signs of disease, suggesting that its tick was indeed the vector. These new findings suggest that ticks play a role in the transmission of *F. tularensis* to human and animal hosts in Sweden.

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## 1 | Introduction

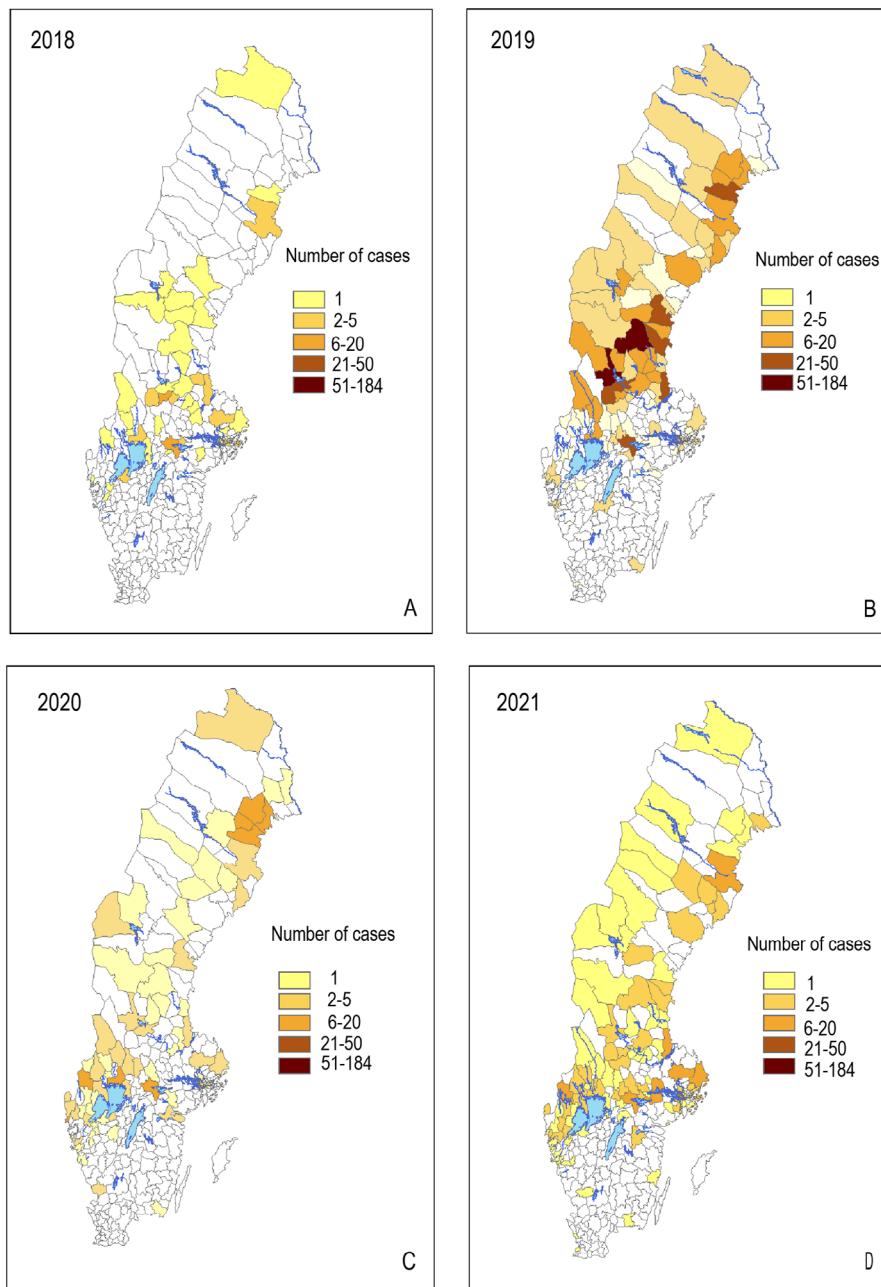
Tularaemia is a vector-borne zoonosis caused by *Francisella tularensis*, a bacterium with a broad host range that includes invertebrates, mammals and birds. *F. tularensis* is categorised into the subspecies *F. tularensis* subsp. *tularensis* (Type A) and *F. tularensis* subsp. *holarctica* (Type B), which differ with respect to the severity of illness in humans and animals and its geographical distribution (Michelet et al. 2014; Sjöstedt 2007). In the northern hemisphere, Type B is the most common subspecies. It is less virulent than Type A, which is found almost exclusively in North America (Svensson et al. 2005). *F. tularensis* subsp. *holarctica* can be transmitted to humans via multiple routes, including through contact with infected animals, contaminated water or arthropod vectors (Hestvik et al. 2015), which are reflected in the different clinical presentations of the disease (Plymoth et al. 2024; Sjöstedt 2007). The disease presents itself in different forms; ulceroglandular, oropharyngeal, oculoglandular, pneumonic and typhoid/septic form. The clinical presentation depends on the subspecies of the bacteria and the route of the infection. In Sweden, the ulceroglandular and pneumonic forms dominate, and the ulceroglandular form is most prevalent with around 65% of the cases. It is characterized by a typical ulcer with enlarged regional lymph nodes. Usually, patients also exhibit nonspecific symptoms such as prolonged fever, severe fatigue and myalgia (Plymoth et al. 2024). Two principal life cycles of tularaemia are described in the literature: a terrestrial life cycle and an aquatic life cycle (Maurin and Gyuranecz 2016). Both life cycles are suspected to be present in association with *F. tularensis* subsp. *holarctica* in Europe (Maurin and Gyuranecz 2016). In Sweden and Finland, the aquatic life cycle is the most predominant environment (Maurin and Gyuranecz 2016), and mosquitos are considered to play a major role in transmission of the disease (Dryselius et al. 2019; Sormunen et al. 2021). To the best of our knowledge, outbreaks among humans and hares have not been reported to be linked to tick-bites in the Nordic countries. However, Sharma et al. (2023) have confirmed that the transmission cycle of *F. tularensis* varies in different ecosystems. Both Types A and B are associated with diverse life cycles in different animal hosts and arthropod vectors and can also coexist. Type B tularaemia is more frequently associated with the aquatic cycle, although outbreaks of tick-borne tularaemia involving subspecies *holarctica* have been reported (Maurin and Gyuranecz 2016). The simultaneous presentation of the terrestrial and aquatic life cycles has been shown in Ukraine where *F. tularensis* was found in water, mammals and ticks (Hightower et al. 2014).

The clinical presentation varies depending on the route of infection. Among animals, the disease is frequent in hares and rodents. In Sweden, hares have previously been described as primarily showing acute fatal infections (Hestvik et al. 2017); thus, it is considered that they die promptly after infection and therefore are not able to maintain the infection for prolonged periods. In Sweden, the number of infected humans and hares fluctuates greatly from year to year. Figure 1A–D shows the geographical distribution of human cases of *F. tularensis* subsp. *holarctica* in Sweden between 2018 and 2021 (Public Health Agency of Sweden 2023). In the same period, 118 necropsied hares were diagnosed with *F. tularensis* subsp. *holarctica* via the national general wildlife disease surveil-

lance programme at the Swedish Veterinary Agency (SVA) (Figure 2).

Ticks transmit several pathogens of medical and veterinary importance (De la Fuente et al. 2017; Heyman et al. 2010). In Europe's temperate and cold regions, the hard tick *Ixodes ricinus* is the most important vector of tick-borne diseases in terms of both animal and human health, for example, tick-borne encephalitis virus, *Borrelia* spp., *Anaplasma phagocytophilum*, *Rickettsia* spp., *Neorhlichia mikurensis* and *Babesia* spp. (Springer et al. 2020). *F. tularensis* has been detected in different tick species in several European countries (Guryčová et al. 1995; Hestvik et al. 2015), but to the authors' knowledge, it has not been reported in *I. ricinus* in Sweden. However, several European studies have shown evidence that the *I. ricinus* tick species is a biological vector of *F. tularensis* subsp. *holarctica* (Hestvik et al. 2015). Detection of *F. tularensis* DNA in questing *I. ricinus* ticks has been confirmed in the Czech Republic (Hubalek and Halouzka 1997), France (Reis et al. 2011), Serbia (Milutinović et al. 2008) and Slovakia (Guryčová et al. 1995), in nymph stages of *I. ricinus* in Switzerland (Wicki et al. 2000) and in *I. ricinus* ticks collected from vertebrate hosts in Germany (Franke et al. 2010). However, the detected prevalence rate of *F. tularensis* in *I. ricinus* ticks is low, at between 0% and 3%, as summarized by Hestvik et al. (2015). It is known that the bacterium can be present and replicate in the ticks' midgut and salivary glands, and transstadial transmission has also been demonstrated experimentally (Petersen, Mead, and Schriefer 2009; Reif et al. 2011). Furthermore, ticks can transmit the infection during a blood meal (Reif et al. 2011). An adult female tick may feed on their host for up to seven days, which exceeds the normal incubation period of tularaemia. *I. ricinus* occurs in a wide variety of habitats throughout Europe as far north as 66°N and is known to be the most common tick species in Fennoscandia (Hvidsten et al. 2020; Omazic et al. 2023). Global environmental changes, such as shifts towards a warmer and wetter climate with milder winters and less snow cover, are likely to improve tick survival and affect their distribution and abundance (Jore et al. 2014). Furthermore, scenario models show that climate change this century will probably increase the geographic range of *I. ricinus* as vegetation communities and mammals associated with high tick densities expand their geographic ranges due to a prolonged vegetation period (Hvidsten et al. 2020; Jaenson and Lindgren 2011). Compared to mosquitoes (that have a shorter lifespan), ixodid ticks like *I. ricinus* could be important for the long-term persistence of endemic foci of tularaemia in northern Europe since they have longer developmental cycles and can act as reservoir of tick-borne pathogens for years (Sonenshine and Roe 2014). All these factors together have contributed to the ticks' expanded distribution in Fennoscandia and to a shift in their latitudinal and altitudinal distribution (Hvidsten et al. 2020; Jaenson and Lindgren 2011; Jore et al. 2014). This northward expansion of ticks and tick-borne pathogens in Sweden is a considerable public health concern.

The focus of the present study was to investigate whether ticks might be involved in the transmission of *F. tularensis* subsp. *holarctica* in Sweden. Molecular analyses of *F. tularensis* subsp. *holarctica* were performed in ticks collected from (i) a citizen science study in Sweden and (ii) *F. tularensis* subsp. *holarctica*-positive hares.



**FIGURE 1** | Geographical distribution of human cases of *Francisella tularensis* subsp. *holarctica* in Sweden in: (A) 2018, (B) 2019, (C) 2020 and (D) 2021.

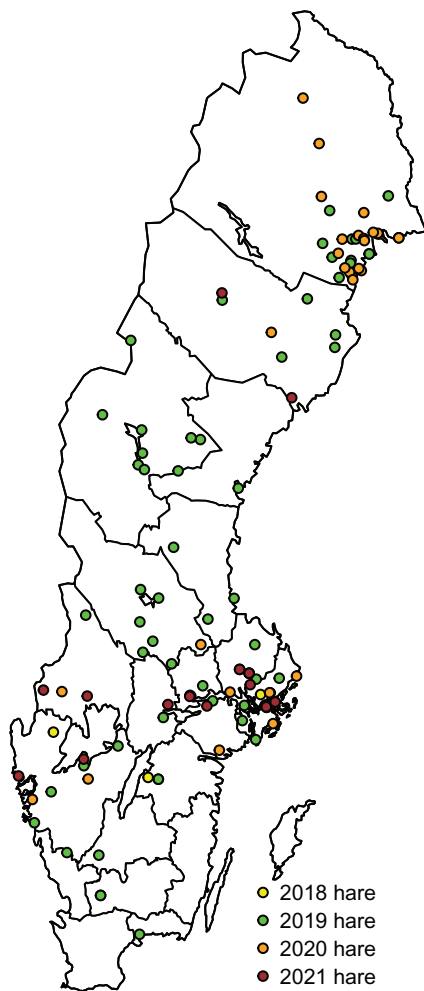
## 2 | Materials and Methods

Ticks ( $n = 3131$ ) were collected from municipalities in the provinces located north of the river Dalälven in Sweden (above latitude  $60^{\circ}\text{N}$ ) in a citizen science study conducted between June and October 2018 (Omazic et al. 2023; Grandi et al. 2024) (manuscript in progress). SVA (Uppsala, Sweden) issued a press release in early June 2018 to inform the public in the study area about the study. Information about the collection was also disseminated with the help of national media channels and social media. Citizens living in or visiting the study area could send in ticks found either on themselves or on domestic and wild animals between June and October in 2018. The tick samples were sent by post to SVA, where all the ticks were registered with information

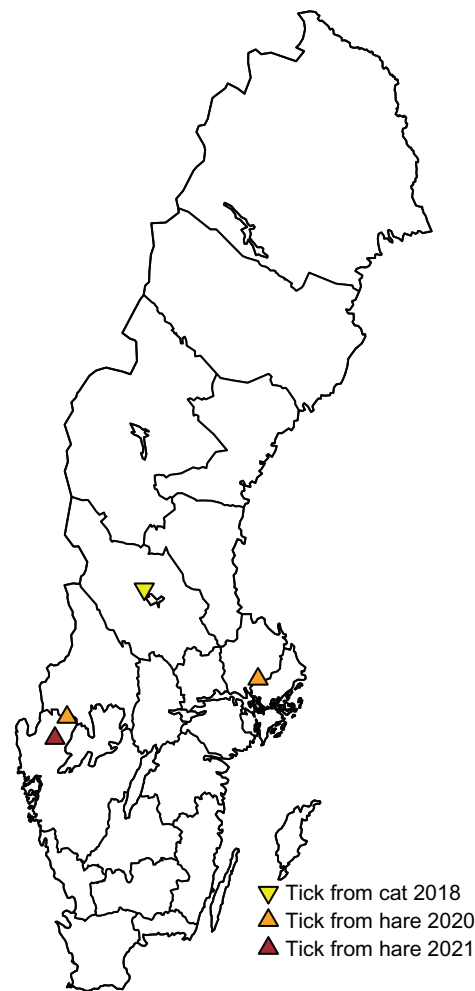
about the municipalities from where they had been submitted and the host species from which they were collected. Individual ticks were stored at  $-80^{\circ}\text{C}$  pending molecular analysis of tick-borne pathogens. Morphological and molecular identification of *I. ricinus* and *Ixodes persulcatus* in the collected ticks was performed at SVA and is described in Omazic et al. (2023).

### 2.1 | High-Throughput Microfluidic Real-Time PCR

A subset of collected ticks ( $n = 1087$ ) was screened for microorganisms using chips with 48.48 dynamic arrays in a Bio-Mark real-time PCR system (Fluidigm, San Francisco, California,



**FIGURE 2** | Geographical distribution of reported *Francisella tularensis* subsp. *holarctica*-positive hares ( $n = 118$ ) between 2018 and 2021 in Sweden.



**FIGURE 3** | Locations of the tick positive for *Francisella tularensis* subsp. *holarctica* found attached to a cat and of the *F. tularensis* subsp. *holarctica*-positive hares that were found to have carried ticks positive for *F. tularensis* subsp. *holarctica*.

USA), enabling 48 samples to be analysed in 48 PCR reactions simultaneously on each chip (Gondard et al. 2018; Michelet et al. 2014). Primers and probes targeting the two gene regions *tul4* and *fopA* were applied for detection of *F. tularensis* subsp. *holarctica*. Microfluidic real-time PCR results were confirmed by a real-time PCR assay for the detection of *F. tularensis* subsp. *holarctica*, targeting the junction between ISFtu2 and the flanking 3'-region (Larsson et al. 2009). Samples with a Ct-value  $\leq 38$  were considered positive. Samples with a clear curve and a Ct-value above 38 but below 45 were considered suspected positive.

## 2.2 | Follow-Up Study

Two to five ticks were collected from the head region of three randomly selected *F. tularensis* subsp. *holarctica*-positive European brown hares (*Lepus europaeus*) from the national general wildlife disease surveillance programme at SVA in Sweden between 2020 and 2021. Ticks from each animal were pooled and then analysed using real-time PCR as described above. A hare was defined as *F. tularensis* subsp. *holarctica*-positive when testing positive

either using PCR or the immunohistochemistry of inner organs. A necropsy of each hare was followed by a histopathologic examination of the collected tissue samples fixed in 4% formalin. Fixed tissues were prepared and stained with Mayer's haematoxylin and eosin, in accordance with standard procedures. To visualise *F. tularensis*, immunohistochemistry was performed, following in-house standard protocols, on prepared tissue samples of inner organs and skin at the site of the tick bite. A monoclonal primary antibody clone BM2369M, formerly clone FB11 (Meridian Life Science Inc., Nordic Biosite AB, Täby, Sweden) directed against *F. tularensis* sp. lipopolysaccharide antigen, diluted 1:6000, was used.

## 3 | Results

### 3.1 | Real-Time PCR of Ticks

Of the 1087 ticks (collected from the citizen science sampling), one *I. ricinus* female tick tested positive for *F. tularensis* subsp. *holarctica* (Figure 3). This tick was collected in Mora municipality in 2018 and had been found on a cat. According to the owner, the

**TABLE 1** | Ticks collected from *Francisella tularensis* subsp. *holarctica*-positive hares via the national general wildlife disease surveillance programme at the Swedish Veterinary Agency in Sweden from 2020 to 2021.

ID	Year of collection	Age	Origin (municipality)	<i>Francisella tularensis</i> in hares <sup>a</sup>	<i>Francisella tularensis</i> in ticks <sup>b</sup> (Ct value)
A	2020	Adult	Säffle	Positive	24.0
B	2020	Adult	Knivsta	Positive	28.4
C	2021	Adult	Bengtsfors	Positive	40.1

<sup>a</sup>A hare was defined as *Francisella tularensis* subsp. *holarctica*-positive when it either tested positive using PCR or the immunohistochemistry of inner organs.

<sup>b</sup>Conventional real-time PCR assay: Samples are considered positive below a cut-off (Ct) value of 38.

cat showed no symptoms of disease at the time the tick was found and removed. Two of the three hares diagnosed with tularemia carried ticks that were positive for *F. tularensis* subsp. *holarctica* (ID A and ID B in Table 1; Figure 3) and one of the hares carried ticks that were suspected positive (ID C in Table 1; Figure 3). The ticks collected from the hares were identified as *I. ricinus* (engorged) females.

### 3.2 | Pathology in Hares

All three hares were infected with *F. tularensis* subsp. *holarctica* at the time of death. Two of the hares (ID A and ID B) died of an acute systemic infection caused by *F. tularensis*, whereas the third hare (ID C) died from blunt trauma.

ID A was an adult female European hare (*L. europaeus*) found dead, with mild autolytic changes, in October 2020 in Säffle municipality (Table 1; Figure 3). The hare was emaciated and heavily infested with ticks in the head and neck region. A focal skin reaction associated with regional lymphadenopathy was seen at the site of the tick bites. The hare presented with splenomegaly. Microscopically, multifocal severe necrosis was observed in multiple internal organs (bone marrow, liver, spleen, lung and parotid lymph node) with limited inflammatory reaction and a multifocal presence of intralesional bacteria suggestive of *F. tularensis*. The most pronounced lesions were detected in the lymph node. At the site of the tick bites, the skin presented with focal necrosis surrounded by a granulomatous inflammatory reaction. *F. tularensis* antigen was identified by immunohistochemistry in inner organs and skin, mainly associated with areas of necrosis (Figure 4).

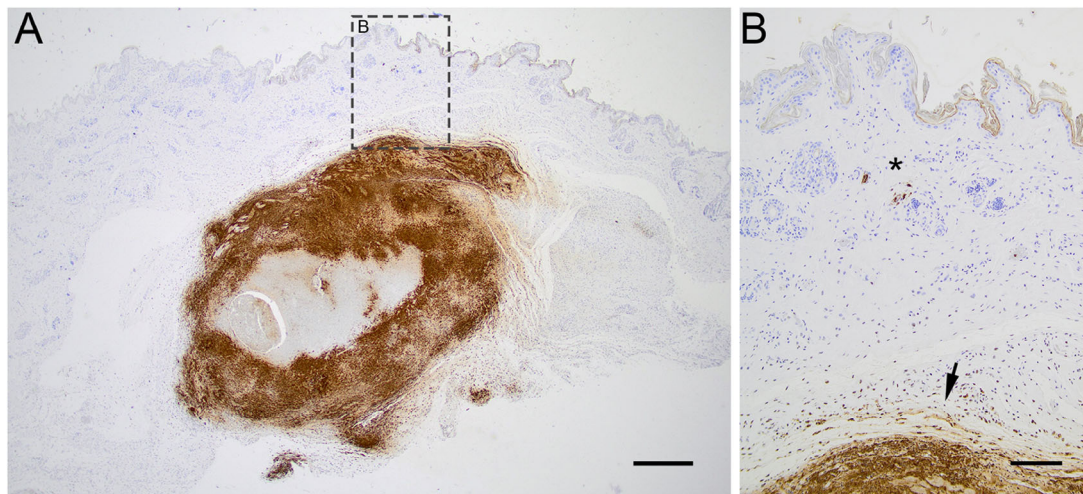
ID B was an adult male European hare found dead, with rigor mortis, in May 2020 in Knivsta municipality (Table 1; Figure 3). The hare was in good body condition, with a few ticks attached to its neck. The hare presented with splenomegaly. Microscopically, multifocal severe necrosis was observed in multiple internal organs (bone marrow, liver and spleen) with limited inflammatory reaction and a multifocal presence of intralesional bacteria suggestive of *F. tularensis*. In the skin of the neck, multiple focal areas of necrosis were observed in association with the tick bites. *F. tularensis* antigen was identified by immunohistochemistry in the inner organs and skin, mainly associated with areas of necrosis (Figure 4).

ID C was an adult male European hare found dead, with mild autolytic changes, in October 2021 in Bengtsfors municipality (Table 1; Figure 3). The hare was emaciated and had general muscle atrophy. Ticks were present in the head region. The diaphragm was acutely ruptured, with displacement of the liver and intestines to the thoracic cavity. Microscopically, purulent meningitis and acute hepatitis were observed. *F. tularensis* was identified by immunohistochemistry in the meninges and choroid plexus. Molecular analysis (PCR) of the inner organs (pooled sample of lung, liver, bone marrow and spleen) tested positive for *F. tularensis* subsp. *holarctica*.

## 4 | Discussion

To the best of the authors' knowledge, this is the first time that *F. tularensis* subsp. *holarctica* has been reported in *I. ricinus* ticks in Sweden. Generally, the transmission of *F. tularensis* subsp. *holarctica* in Sweden is considered to be through the aquatic life cycle (Maurin and Gyuranecz 2016), with mosquitoes a major biological vector of the disease (Dryselius et al. 2019; Sormunen et al. 2021). This is reflected in the predominance of the ulceroglandular form of the disease, as highlighted in a recent study from Sweden (Plymoth et al. 2024). Both ticks and hares are part of the terrestrial life cycle of *F. tularensis*. However, it is likely that the terrestrial and aquatic life cycles can be present simultaneously in different regions in one country and/or on different occasions (Sharma et al. 2023). Previously, hares have not been considered to be part of the reservoir of *F. tularensis* subsp. *holarctica* in Sweden, as the acute fatal infection and death promptly after infection may prevent them being able to maintain the infection for prolonged periods. At present, the pattern for the disease among hares is changing in Sweden, with infected hares now observed in large parts of the country and more frequent reports of cases with chronic lesions (Hestvik et al. 2017). Moreover, antibodies against *F. tularensis* subsp. *holarctica* have been reported in European hares and mountain hares (*Lepus timidus*) found dead with lesions corresponding to tularemia in Sweden (Averhed et al. 2022), which may suggest a chronic manifestation of the disease since seroconversion generally appears after 3 weeks (Kreizinger et al. 2016).

Altogether, the findings of the present study suggest that *I. ricinus* ticks can be part of the disease transmission of *F. tularensis* subsp.



**FIGURE 4** | Microphotograph of the immunohistochemistry visualizing the bacteria *Francisella tularensis* (brown stain) in the skin at the location of a tick bite in a *F. tularensis* subsp.-*holarctica* positive hare (ID A). (A) Section of skin at the site of the tick bite. *F. tularensis* visualized in lytic lesion in the subcutis, bar = 500  $\mu$ m. (B) Close-up of (A), bacteria visualized in the cytoplasm of vascular endothelial cell (asterisk) and lytic lesion in the subcutis (arrow), bar = 200  $\mu$ m.

*holarctica* in Sweden. Results from the citizen science study in 2018 revealed one tick infected with *F. tularensis* attached to a cat. The cat showed no clinical signs of tularaemia, which indicates that the cat did not have septicaemia, and it is therefore likely that the tick acted as a carrying vector of tularaemia, regardless of its host infection status. Although both Types A and B subspecies have been isolated from cats, Type A is responsible for most tularaemia cases in cats (Larson et al. 2014). *F. tularensis* subsp. *holarctica* has previously been reported in clinically healthy cats (Kittl et al. 2020). Case reports from both Norway and Sweden describe humans infected by *F. tularensis* subsp. *holarctica* after cat bites (Petersson and Athlin 2017; Yaqub, Bjornholt, and Enger 2004). Transmission of tularaemia from cats to humans caused by both *F. tularensis* subsp. *holarctica* and *F. tularensis* subsp. *tularensis* has also been described in North America from both sick and healthy cats (Capellan and Fong 1993; Woods et al. 1998). Furthermore, studies performed in Sweden by Eliasson et al. (2002) showed independent associations between humans acquiring tularaemia and owning a cat. Cats are at high risk of exposure to tularaemia predominantly due to their hunting behaviour (Larson et al. 2014), they may eat infected prey, and they are highly exposed to vectors when moving through vegetation associated with high tick densities. Additionally, in 2019, 1 year after the tick was collected from the cat, Sweden experienced its largest outbreak of tularaemia in over 50 years, with Dalarna county (which includes the municipality of Mora) worst affected by the tularaemia outbreak, according to the Public Health Agency of Sweden (Dryselius et al. 2019). Previous studies together with the present findings strongly indicate that cats can increase the risk of human infection both by bringing ticks into the household and through bites and scratches.

Two of the three tularaemic hares (ID A and ID B) carried ticks positive for *F. tularensis* subsp. *holarctica*, and one of the hares (ID C) carried a tick that was suspected positive. The hares carrying positive ticks were diagnosed with acute tularaemia as the cause of death. Interestingly, hare ID A and hare ID B had a concurrent skin reaction, with visualized intralesional tularaemia bacteria

at the site of the tick bite(s). Hestvik et al. (2017) previously described similar skin lesions at the site of tick bites with several foci of necrosis in the dermis and subcutis in two European hares, and the authors discussed the possibility of transmission of the bacteria via tick bites but did not analyse the ticks. In hare ID A, a subacute inflammatory reaction was observed to be linked to foci of necrosis and bacteria in the skin compared with more acute lesions in the inner organs (Figure 4A,B). This observation, together with most severe necrotic lesions seen in the regional lymph node (parotid), suggests that the tick bite is the portal of entry. The hare exhibited a disease pattern consistent with the most common form of tularaemia, the ulceroglandular form, affecting skin at the portal of entry and regional lymph nodes, potentially progressing into systemic disease (Maurin and Gyuranecz 2016; Sjöstedt 2007). This information indicates that the bacteria were transmitted from the tick to the hare, causing a local skin inflammation with lymphogenous spread to the regional lymph node, and further causing a fatal septicaemia. A similar case has previously been described by Park et al. (2009), where immunohistochemistry was performed on both the skin of a Japanese hare (*Lepus brachyurus*) and the tick attached to the hare. A bacterial antigen was detected not only in the tick intestines, but in the haemocoel of the tick as well. The hare displayed skin granuloma on its external ear, with regional lymphadenopathy and acute necrosis in the inner organs. The authors concluded that the hare acquired the tularaemia infection from the tick and not vice versa (Park et al. 2009), as in the present study.

The ticks that tested positive for *F. tularensis* subsp. *holarctica* in the present study were *I. ricinus* adult females. *F. tularensis* subsp. *holarctica* has been found in *I. ricinus* in several European countries (Reis et al. 2011), but biological studies demonstrating that the bacteria can be transmitted by ixodid ticks are scarce. In particular, the vectorial capacity of *I. ricinus* can currently only be hypothesized, and it is not clear if the present findings are due to transmission of the infection from the tick to the animal host or vice versa. In the present study, the pathology of the hare was

consistent with the tick bite as the port of bacterial entry, which suggests that *F. tularensis* subsp. *holarctica* can be transmitted by *I. ricinus* ticks. Additionally, the presence of *F. tularensis* subsp. *holarctica* was observed in a tick on a clinically healthy cat, which further supports the role of *I. ricinus* as a potential vector of the bacteria.

In Sweden, human cases (and outbreaks) of tularaemia have mainly been reported in northern Sweden, but cases in the central and southern parts of the country have been increasing in recent years (Figure 1). The same pattern can be seen in infected hares: In the last two decades, bacterial infection of *F. tularensis* in hares has been shown to be more common in areas south of the previous endemic region in northern Sweden (Andersson et al. 2022). Mosquitos are considered the main vector for tularaemia, and outbreaks are suggested to be dependent on mosquito prevalence in boreal forest regions (Rydén et al. 2012). The species of mosquitos (*Aedes cinereus*) that are often considered to spread tularaemia prefer forests habitat. This habitat is present in a larger degree (13 times as common) in one of the studied northern locations compared to the collection spot in southern Sweden (Schäfer and Lundström 2001). Forest and wetland habitats are present in whole Sweden, that is, forests cover 68% and wetlands cover 22% of the total area, and forests are predominant in northern Sweden (Sweden 2023). In southern and central Sweden, ticks are more abundant and might play a role as a potential vector of transmission of the tularaemia bacteria. Simultaneously, ongoing climatic and environmental changes are favouring the spread of ticks towards the northern parts of Sweden. A recent study by Omazic et al. (2023) confirmed that *I. ricinus* and *I. persulcatus* have enlarged their distributional area in northern Sweden compared with previous reports. The increasing number of ticks might become important in the future transmission of *F. tularensis* subsp. *holarctica* in Sweden. The prevalence of tularaemia in ticks in Europe is low, and in several countries, including Sweden, largely unknown. Mosquitos are considered the major vector for *F. tularensis* subsp. *holarctica* in Sweden with a relatively high presence of the bacterium in mosquito species sampled in endemic areas (Thelau et al. 2014). In the present study, tularaemia has only been demonstrated in a few ticks indicating a low prevalence. This suggests that tularaemia transmission in animals and humans through tick bites might be quite rare in Sweden. However, our study revealed for the first time in Sweden that we have tularaemia positive ticks and that they can infect hares which indicates that ticks may play a role in the transmission of *F. tularensis* subsp. *holarctica* in Sweden. This knowledge is important as an information base for future *F. tularensis* subsp. *holarctica* surveillance in the country. Further studies are needed in order to investigate the role of ticks in the transmission of *F. tularensis* subsp. *holarctica* between animals and humans, and between infected animals, during an outbreak.

## 5 | Conclusions

*F. tularensis* subsp. *holarctica* has been isolated from ticks in Sweden for the first time. The bacterium was detected in ticks collected from three hares and a cat. Immunohistochemical identification of the bacteria at the site of the tick bite, together with associated subacute skin lesions and subsequent acute dis-

seminated disease, suggests that the hares acquired the infection from the tick. The cat showed no clinical signs of disease, which suggests a possible transmission of the bacterium from the tick to the cat. Hares have not previously been considered to be part of the transmission of *F. tularensis* subsp. *holarctica* in Sweden, mainly due to the acute nature of the infection and rapid death. However, the disease pattern has recently changed in Sweden, and hares may now also be diagnosed with chronic infections. Furthermore, this is confirmed by the presence of antibodies, which enables them to be an important part of the transmission cycle of the bacteria. Finally, the findings of this study suggest that ticks may play a role in the transmission of *F. tularensis* to human and animal hosts in Sweden, which merits further investigation.

### Author Contributions

**Ellinor Spörndly-Nees:** conceptualization, investigation, resources, visualization, writing—original draft. **Tomas N. Gustafsson:** conceptualization, funding acquisition, writing—review and editing. **Anna Omazic:** conceptualization, data curation, investigation, funding acquisition, project administration, validation, writing—original draft. **Elina Thorsson:** investigation, resources, visualization, writing—review and editing. **Giulio Grandi:** conceptualization, funding acquisition, writing—review and editing.

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### Ethics Statement

An ethics statement is not applicable as this study was not classified as a human or animal experiment. Ticks were sent in voluntarily by people living in or visiting the study area. Their personal data were only used to locate the geographical area and host information. SVA processed the data in accordance with Data Protection Regulation (EU) 2016/679. The senders were informed that SVA reserves the right to use the submitted ticks and the accompanying metadata for research purposes as part of its activities. The hares were found dead and were sent to the SVA through the national general wildlife disease surveillance programme, therefore no ethical approval was needed.

### Conflicts of Interest

The authors declare no conflicts of interest.

## Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

## Peer Review

The peer review history for this article is available at <https://publons.com/publon/10.1002/vms3.70094>.

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