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Short communication

A new hotspot of tick-borne encephalitis virus (TBEV) in the Autonomous Province of Trento, Italy

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ABSTRACT

The tick-borne encephalitis virus (TBEV) has expanded its distributional range in Europe over recent decades. Italy is considered a low tick-borne encephalitis (TBE) incidence country, nonetheless, human cases have recently increased, reaching their maximum so far in 2022. This study reports on a new TBE hotspot in the Autonomous Province of Trento (Italy), along with a description of TBEV genetic variants at the provincial level. Tick sampling was performed where several TBE human cases were reported in 2022. As a result, 458 *Ixodes ricinus* ticks were collected (38 females, 49 males, 371 nymphs) and screened for TBEV through real-time PCR, pooling the nymphs while analyzing the adults individually. Eight samples were found to be positive for TBEV, corresponding to a 4.6 % prevalence in adults and a 1.1 % minimum infection rate in nymphs in the area. The identified TBEV genetic variants were compared with other TBEV European subtype (TBEV-Eu) strains. The relatively high prevalence in ticks and the reported human cases beyond the year of vector collection suggest a sustained virus circulation. Additionally, phylogenetic analysis of the genetic variants in the Trentino Province shows a separation among eastern and western sites. This study highlights the crucial importance of active surveillance for mitigating TBE risk, especially in highly anthropized Alpine areas.

1. Introduction

Tick-borne encephalitis (TBE) is a severe viral zoonosis affecting the human central nervous system. It is caused by *Orthoflavivirus encephalitidis* (previously known as tick-borne encephalitis virus TBEV; genus *Orthoflavivirus*, family *Flaviviridae*; https://ictv.global/taxonomy). Infection with TBEV is usually acquired through the bite of infected ticks belonging to some species of the Ixodidae family (Kahl et al., 2019; see Table 4.1), but also through the consumption of unpasteurized dairy products and other non-vectorial modes of transmission (Martello et al., 2022). The sylvatic enzootic cycle relies on certain tick species and only on certain wild hosts, such as small mammals (reservoir hosts for the virus) and deer (amplifier hosts for tick populations) (Gray et al., 2024). Land use, climate change and modification in wildlife host community composition and animal movement, including migration, can affect vector and host interactions, favoring TBEV spillover in previously non-endemic areas (Dagostin et al., 2023; Rosà et al., 2019).

In the last decades, the number of TBE cases showed a general increasing trend (ECDC, 2024), although this differed among European countries (ECDC, 2024), with an increase in some countries as in Austria, Estonia, Finland, Italy, Norway, and Poland. Morbidity and rate of case fatality differed according to the viral subtypes (European-Eu, Siberian-Sib, Far Eastern-FE, and the newly-discovered Baikalian-Bkl and Himalayan-Him) (Burrell et al., 2016; Ruzek et al., 2019). TBEV distribution is patchy, with foci (hotspots) that might be stable or highly variable in space and time (Kreusch et al., 2019; Stoefs et al., 2021). Italy has a low TBE incidence (0.05 over 100.000 inhabitants and a total of 460 cases in the period 2010–2023) with cases mainly located in the north-eastern regions (Tagliapietra et al., 2024). Since 2017, a national TBE surveillance system of neuro-invasive infections has been

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established, among which Trentino Province reported an increase in the number of cases, reaching its maximum in 2022 (Fig. 1a).

The municipality of Canal San Bovo (Upper Vanoi Valley, Autonomous Province of Trento, Italy), that never reported TBE human cases so far (Fig. 1a and 1b), stood out with an unexpected reported cluster of seven neuro-invasive human cases in 2022 (Del Manso et al., 2022). Afterwards, in the same or neighboring areas, four further cases of TBE were reported and specifically in Predazzo and Ziano di Fiemme (2023), Canal San Bovo (2024) and Mezzano (2024) (data provided by the local Public Health Agency, APSS; see Fig. S1.1, Appendix 1). According to this, we decided to investigate tick infection prevalence in the area of Canal San Bovo and to assess TBEV genetic variants across the Trentino Province by means of phylogenetic analysis (Alfano et al., 2020).

2. Materials and methods

We collected questing ticks from the vegetation in May 2022 to determine TBEV occurrence, although this is generally considered time-consuming and costly due to the large numbers of samples required in view of the low prevalence and the overdispersion of this vector in the environment. Ticks were collected by dragging a standard 1 m^2 white blanket above the vegetation (Salomon et al., 2020) along transects of variable length in ecotones of mixed forests. Two sites located in the municipality of Canal San Bovo (Upper Vanoi Valley, Autonomous Province of Trento, Italy) were selected, namely Pralongo (46.182210 °N 11.693330 °E; 780 m a.s.l.) and Rebrut (46.162350 °N 11.703180 °E; 920 m a.s.l.). Collected ticks were washed in 70 % ethanol, rinsed with deionized water for molecular biology and dried with paper. Tick species, sex and stages were morphologically identified using a stereomicroscope and taxonomic keys (Estrada-Peña et al., 2017, 2004; Manilla, 1998).

The RNA of the collected ticks was extracted from males and females individually, while nymphs were grouped in pools of maximum five specimens (74 pools in total). Tick pools and individuals were homogenized in 350 μL lysis buffer (RLT) (as we estimated they could have the same weight (Mierzejewska et al., 2015; Randolph, 2004)), and we then centrifuged the lysate. Total RNA was then extracted using the RNeasy Mini Kit (Qiagen-GmbH—Hilden, Germany) following the manufacturer's instructions. The extraction protocol was ameliorated by adding for three times the wash buffer (RPE) to the RNeasy spin columns. TBEV-specific real-time PCR was performed, and samples were screened by a real-time Taq-Man based one-step RT–PCR, targeting a fragment of the 3′ non-coding region of the TBEV genome, as described previously (Alfano et al., 2020; Schwaiger and Cassinotti, 2003). Extracted RNA of strain Hypr (a highly neuroinvasive and neurovirulent strain) was used as a positive control, while distilled water served as a negative control.

Positive samples were confirmed by amplifying a fragment of the E gene (∼520 bp long) encoding envelope (E) protein, as described previously (Skarpaas et al., 2006); see also Alfano et al. (2020) for the real-time PCR protocol). We visualized PCR products through capillary electrophoresis on a QIAxcel system (Qiagen), and then they were purified using the MinElute PCR Purification Kit (Qiagen) and sequenced on an ABI 3730xl DNA Analyzer (Applied Biosystems). We calculated infection prevalence in nymphs using the minimum infection rate (MIR), where the number of pools was related to the total number of ticks tested, expressed as a percentage, while in adults, prevalence was estimated. MIR is based on the assumption that only one infected tick is present in each positive pool (Cowling et al., 1999), an index accepted for arboviruses occurring at low prevalence in their vector populations, such as TBEV (Alfano et al., 2020).

We selected 474 TBEV sequences from GenBank, reflecting the 95.7 % of identity, and we aligned them with the eight obtained from Canal San Bovo (GenBank Accession number: PP426229-PP426236) and the four obtained from Alfano et al. (2020) to construct an European phylogenetic tree (Maximum-Likelihood method based on the General Time Reversible Substitution model, Γ distribution and 1000 bootstraps; IQ-TREE web server and FigTree v1.4.4 software). We then selected the clade of the European phylogenetic tree that embedded the Trentino Province sequences (from Canal San Bovo and from Alfano et al. (2020), composed overall by 21 TBEV sequences), aligning them with 41 TBEV-Eu-strains used by Alfano et al. (2020), that represented most European countries for which TBEV-Eu sequences were available. We

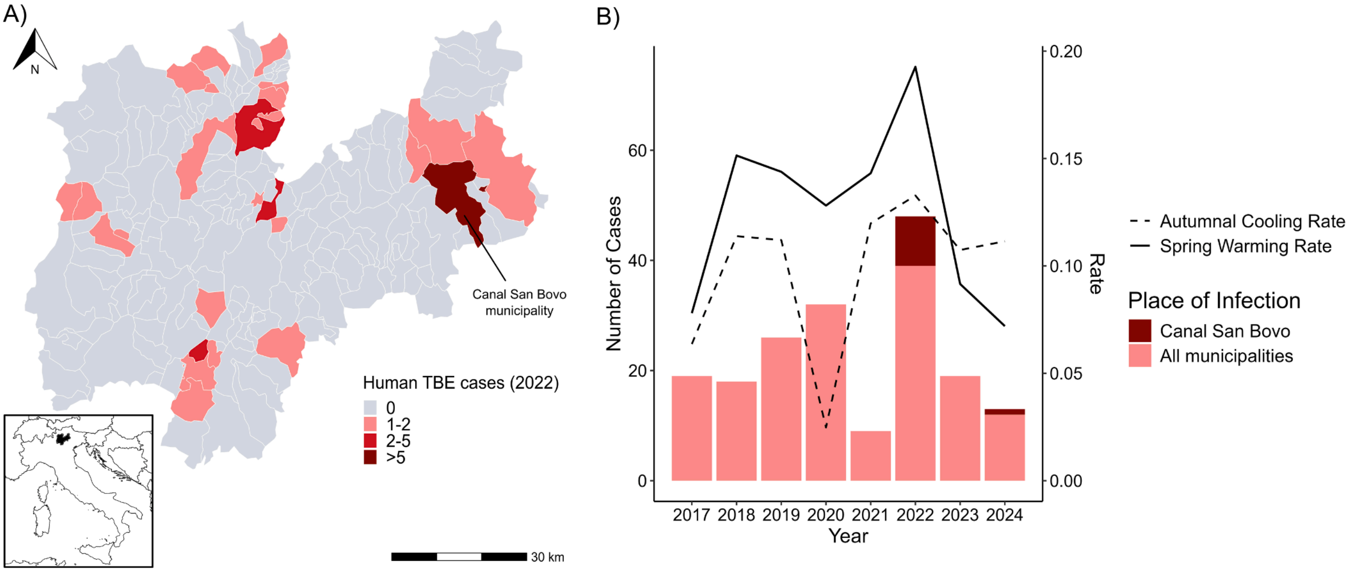

Fig. 1. Integrative framework of the new hotspot of Tick-borne encephalitis virus (TBEV) occurred in eastern Trentino (Canal San Bovo 46°09′23″ N 11°44′01″ E, Autonomous Province of Trento, Italy). *Panel A)* Map of Trentino Province and its municipalities in 2022. The red color gradient highlights the number of human tick-borne encephalitis (TBE) cases recorded in 2022 (data provided by the local Public Health Agency, Azienda Provinciale per i Servizi Sanitari Provincia Autonoma di Trento, APSS). *Panel B)* Number of human TBE cases recorded in Trentino Province (Italy) from 2017 to 2024. Dark red color: human cases from Canal San Bovo; light red color: human cases in other municipalities. The climatic data considered are: (i) the autumnal cooling rate of the previous year (showed as absolute value; dashed line) and (ii) the spring warming rate (solid line), calculated by applying linear regression to the average daily temperatures against the Julian day in the period 1 August to 31 October and 1 February to 30 April, respectively (5) (data obtained from Meteotrentino https://www.meteotrentino.it, Caoria weather station at 803 m a.s.l.).

constructed a second comparative phylogenetic tree (Maximum-Likelihood method based on the General Time Reversible Substitution model, Γ distribution and 1000 bootstraps; MEGAX software) to evaluate the origin of the Trentino Province TBEV sequences compared with those identified and selected in the dendrogram by Alfano et al. (2020). The Omsk hemorrhagic fever virus (OHFV) was used as an outgroup for both phylogenetic trees (GenBank Accession number: AB507800). The Nexus format of the sequences from the second comparative phylogenetic tree (62 TBEV sequences, and OHFV outgroup) that stores information about morphological and molecular characters of the lineages was then used to establish the genetic variants' network using the Minimum Spanning Model of PopArt 1.7, based on the European geographic position of the countries.

3. Results

In the study area of Canal San Bovo, 458 ticks belonging to the species *Ixodes ricinus* were collected (38 females, 49 males, 371 nymphs). Despite the low number of ticks screened, two females, two males, and four pools of nymphs resulted positive for TBEV with a prevalence of 4.6 % (4/87) in adults and a MIR of 1.1 % (4/371) in nymphs.

From the European phylogenetic tree, we identified a separate clade embedding the TBEV isolates from Trentino Province (see red box in Fig. S2.1, Appendix 2) together with other sequences from western and central European countries (e.g., Germany, Slovenia, Switzerland). Considering the second comparative phylogenetic tree and the genetic variants' network, the isolates from Trentino Province belonged to different genetic lineages within the TBEV-Eu subtype (see Fig. 2 and Fig. S3.1, Appendix 3). The sequences of the eight TBEV-positive samples from Canal San Bovo formed a single cluster with one sequence obtained by Alfano et al. (2020), which was collected in eastern Trentino, and one sequence found in the neighboring Belluno Province. These sequences differed from those from Switzerland, Germany, and Slovenia by a couple of mutations. Moreover, the eastern Trentino isolates were distinct from the three sequences retrieved by Alfano et al. (2020) from western Trentino, differing by a single mutation. These latter sequences grouped together with sequences from Germany and Lithuania, differing by one and two mutations, respectively. The Trentino Province isolates demonstrated a close relationship with those from the Friuli-Venezia-Giulia (FVG) region, although they belonged to a distinct clade. In contrast, the remaining sequences from FVG and those obtained from Trentino Province in 2006 clustered apart in the phylogenetic tree, being consistent with the findings reported by Alfano et al. (2020).

4. Discussion

The TBEV prevalence found in ticks and the incidence of human cases recorded in 2022 in the municipality of Canal San Bovo may indicate the occurrence of a TBEV hotspot, which was previously undetected. The municipality of Canal San Bovo is part of a TBE-endemic region which includes the north-eastern alpine and prealpine area of Italy, where the virus is known to circulate for at least 30 years (Tagliapietra et al., 2024).

Based on our previous findings (Marini et al., 2023; Rosà et al., 2019), we hypothesize that TBE incidence of human cases upsurge in Canal San Bovo may reflect the 2-year time lag cascading effect of the abundant production of spruce (*Picea abies*) and beech (*Fagus sylvatica*) seeds (mast) in 2020 (pers. observation). In particular, abundant seed

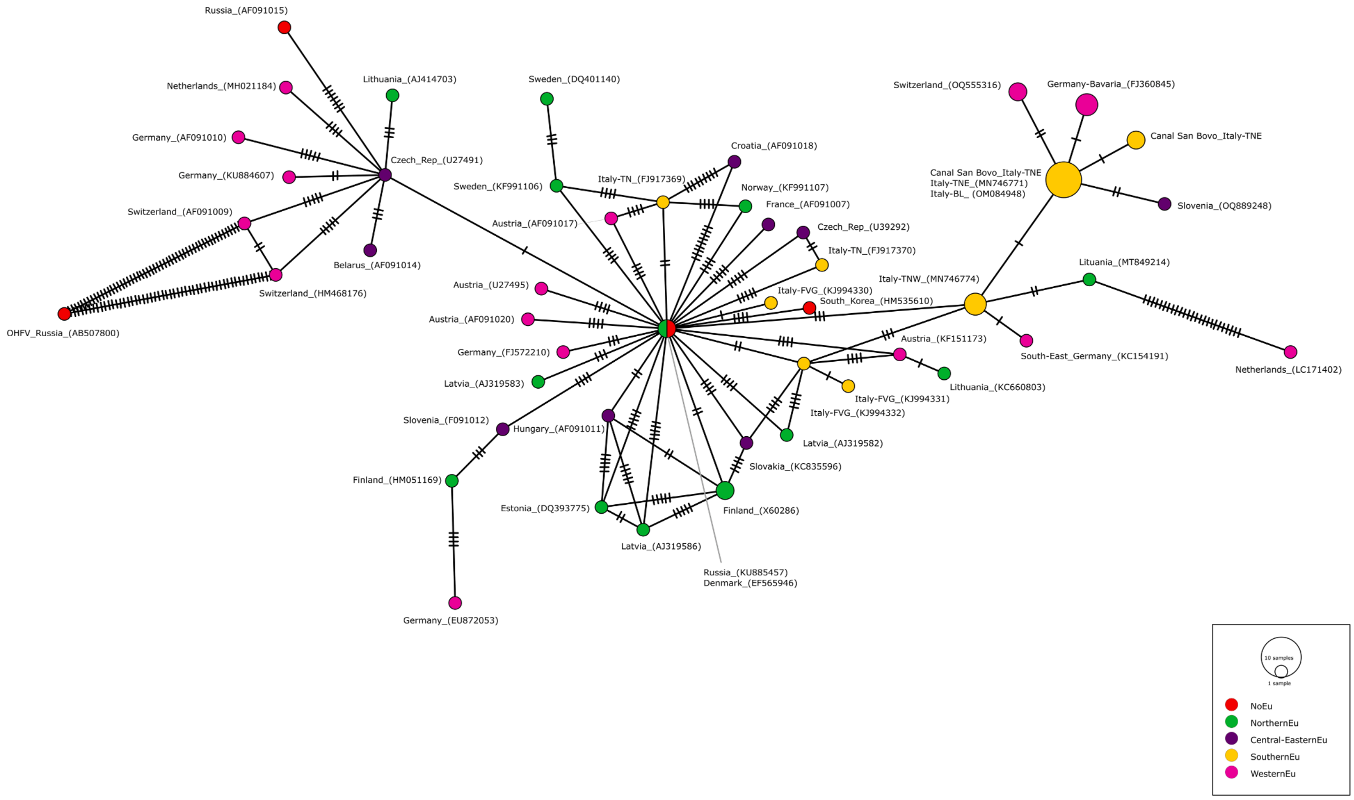

Fig. 2. Minimum spanning tick-borne encephalitis (TBEV) genetic variants' network inferred from 62 tick-borne encephalitis virus (TBEV) sequences from Trentino Province and Europe (4 derived from Alfano et al. (2020), 8 identified in Canal San Bovo, both expressed by yellow nodes) and 50 belonged to European TBEV strain retrieved from GenBank) described by nodes. The Omsk hemorrhagic fever virus (OHFV) was used as an outgroup for both phylogenetic trees. The colors of nodes express the European geographical locations where samples were collected: non-European (red nodes), Northern (green nodes), Eastern (purple nodes), Southern (yellow nodes) and Western Europe (pink nodes). The length of the black lines defines the genetic distance between TBEV genetic variants given by the number of mutations (vertical segments). The size of nodes represents the number of samples with the same TBEV sequence.

production can favour high density of granivorous woodland rodents that, by feeding on tick larval stage, increase the probability of presence of infected nymphs and therefore TBEV risk of transmission two years later. The cascading effects of pulsed mast seed production on the abundance of reservoirs and infected ticks thereafter (Brugger et al., 2017; Imholt et al., 2015) emerged also in other contexts and within other disease systems. For instance, beyond the importance for TBE risk (Rubel et al., 2020), the role of mast seeding has been demonstrated to drive Lyme borreliosis risk (Bogdziewicz and Szymkowiak, 2016; Bregnard et al., 2020; Ostfeld et al., 2006). Beyond masting and its cascading consequence on reservoir abundance, also climatic conditions may govern TBEV transmission risk. In particular, temperature changes play an important role in questing tick behavior, but are not necessarily responsible for the timing of the behavioral diapause, which in turn is mainly regulated by the photoperiod. (Gray et al., 2016). Therefore, rapid autumn temperatures drop (namely autumnal cooling) in 2021 and fast spring temperatures increase (namely spring warming) in 2022 in the area (Fig. 1b) may have positively affected the appearance of questing ticks and synchronized their emergence in spring, ultimately promoting TBEV circulation (Dagostin et al., 2023; Rosà et al., 2019).

Trentino Province is divided north-to-south by the Adige river and homonymous valley, largely occupied by human infrastructures. The differences observed between local eastern and western TBEV sequences suggest that natural or artificial barriers might affect dispersal of infected ticks and hosts from one area to the other, as also observed in our previous findings (Carpi et al., 2016). The host dispersal and movements could also explain the similarity of TBEV found in some European countries, such as Germany, Switzerland and Slovenia, as well as Belluno Province, which are geographically closer to Trentino Province. For example, migratory bird routes may be implied in different introductions of TBEV in Europe, especially from neighboring countries and nesting areas (Sparagano et al., 2015; Waldenström et al., 2007; Wilhelmsson et al., 2020).

The main limitation of the study is that it is temporally restricted to one year of tick surveillance, which may not adequately express the presence or absence of an active focus in the area. To overcome this issue, we suggest that long-term and integrated surveillance studies are needed, together with the evaluation of the variation of other factors influencing the circulation of TBEV. This is paramount to adapt the surveillance-control strategies and forecast new areas with a higher probability of disease emergence. It is of interest that the gross number of vaccine doses administered to people living in the investigated area tripled in 2022, highlighting the awareness of the local health issue (courtesy of the local Public Health Agency, APSS).

Overall, this study highlights the importance of implementing active surveillance and prevention campaigns for mitigating TBE risk, especially in highly-anthropized Alpine areas where the coincidence of a set of critical environmental covariates (Dagostin et al., 2023) predicts a rise in TBEV transmission risk.

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CRediT authorship contribution statement

Giulia Ferrari: Writing – original draft, Visualization, Formal analysis, Data curation, Conceptualization. **Fausta Rosso:** Writing – review & editing, Methodology, Investigation, Data curation, Conceptualization. **Matteo Girardi:** Writing – review & editing, Methodology, Investigation, Formal analysis, Data curation. **Francesca Dagostin:** Writing – review & editing, Visualization, Formal analysis, Data curation. **Daniele Arnoldi:** Writing – review & editing, Investigation. **Maria Grazia Zuccali:** Writing – review & editing. **Chiara Mocellin:** Writing – review & editing. **Silvia Molinaro:** Writing – review & editing. **Valentina Tagliapietra:** Writing – original draft, Project administration, Methodology, Investigation, Conceptualization. **Annapaola Rizzoli:** Writing – review & editing, Project administration, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare that they have no conflict of interest.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.ttbdis.2025.102513.

Data availability

Data will be made available on request.

References

Alfano, N., Tagliapietra, V., Rosso, F., Ziegler, U., Arnoldi, D., Rizzoli, A., 2020. Tick-borne encephalitis foci in northeast Italy revealed by combined virus detection in ticks, serosurvey on goats and human cases. Emerg. Microbes Infect. 9, 474–484. https://doi.org/10.1080/22221751.2020.1730246.

Bogdziewicz, M., Szymkowiak, J., 2016. Oak acorn crop and Google search volume predict Lyme disease risk in temperate. Europe. Basic Appl. Ecol. 17, 300–307. https://doi.org/10.1016/J.BAAE.2016.01.002.

Bregnard, C., Rais, O., Voordouw, M.J., 2020. Climate and tree seed production predict the abundance of the European Lyme disease vector over a 15-year period. Parasites Vectors 13. https://doi.org/10.1186/s13071-020-04291-z.

Brugger, K., Walter, M., Chitimia-Dobler, L., Dobler, G., Rubel, F., 2017. Seasonal cycles of the TBE and Lyme borreliosis vector *ixodes ricinus* modelled with time-lagged and interval-averaged predictors. Exp. Appl. Acarol. 73, 439–450. https://doi.org/10.1007/S10493-017-0197-8/FIGURES/4.

Burrell, C.J., Howard, C.R., Murphy, F.A., 2016. Fenner and White's Medical Virology: Fifth Edition, Fenner and White's Medical Virology: Fifth Edition. Elsevier Inc.

Carpi, G., Kitchen, A., Kim, H.L., Ratan, A., Drautz-Moses, D.I., McGraw, J.J., Kazimirova, M., Rizzoli, A., Schuster, S.C., 2016. Mitogenomes reveal diversity of the European Lyme borreliosis vector *ixodes ricinus* in Italy. Mol. Phylogenet. Evol. 101, 194–202. https://doi.org/10.1016/J.YMPEV.2016.05.009.

Cowling, D.W., Gardner, I.A., Johnson, W.O., 1999. Comparison of methods for estimation of individual-level prevalence based on pooled samples. Prev. Vet. Med. 39, 211–225. https://doi.org/10.1016/S0167-5877(98)00131-7.

Dagostin, F., Tagliapietra, V., Marini, G., Cataldo, C., Bellenghi, M., Pizzarelli, S., Cammarano, R.R., Wint, W., Alexander, N.S., Neteler, M., Haas, J., Dub, T., Busani, L., Rizzoli, A., 2023. Ecological and environmental factors affecting the risk of tick-borne encephalitis in Europe, 2017 to 2021. Eurosurveillance 28, 2300121. https://doi.org/10.2807/1560-7917.ES.2023.28.42.2300121.

Del Manso, M., Di Maggio, E., Perego, G., Petrone, D., Mateo-Urdiales, A. Caporali, M.G., Bella, A., Venturi, G., Di Luca, M., Giannitelli, S., Ferraro, F., Pezzotti, P., Riccardo, F., 2022. Arbovirosi in Italia - 2022.

ECDC, 2024. European Centre for Disease Prevention and Control. Tick-borne encephalitis. In: ECDC. Annual epidemiological Report for 2022. https://doi.org/10.1016/B978-0-323-88305-4.00314-X.

Estrada-Peña, A., Martinez, J.M., Sanchez Acedo, C., Quilez, J., Del Cacho, E., 2004. Phenology of the tick, *ixodes ricinus*, in its southern distribution range (central Spain). Med. Vet. Entomol. 18, 387–397. https://doi.org/10.1111/J.0269-283X.2004.00523.X.

Estrada-Peña, A., Mihalca, A.D., Petney, T.N., 2017. Ticks of Europe and North Africa. A guide to Species identification, 1st ed, Ticks of Europe and North Africa. Springer International Publishing. https://doi.org/10.1007/978-3-319-63760-0.

Gray, J., Kahl, O., Zintl, A., 2024. Pathogens transmitted by *Ixodes ricinus*. Ticks Tick. Borne. Dis. 15, 102402. https://doi.org/10.1016/J.TTBDIS.2024.102402.

Gray, J.S., Kahl, O., Lane, R.S., Levin, M.L., Tsao, J.I., 2016. Diapause in ticks of the medically important *Ixodes ricinus* species complex. Ticks Tick. Borne. Dis. 7, 992. https://doi.org/10.1016/J.TTBDIS.2016.05.006.

Imholt, C., Reil, D., Eccard, J.A., Jacob, D., Hempelmann, N., Jacob, J., 2015. Quantifying the past and future impact of climate on outbreak patterns of bank voles (*Myodes glareolus*). Pest Manag. Sci. 71, 166–172. https://doi.org/10.1002/ps.3838.

Kahl, O., Chitimia-Dobler, L., Mackenstedt, U., Petney, T.N., 2019. Zirkulation des FSME-Virus im Freiland, in: FSME in Deutschland: Stand der Wissenschaft 4, 53–66.

Kreusch, T.M., Holding, M., Hewson, R., Harder, T., Medlock, J.M., Hansford, K.M., Dowall, S., Semper, A., Brooks, T., Walsh, A., Russell, K., Wichmann, O., 2019. A probable case of tick-borne encephalitis (TBE) acquired in England, July 2019. Eurosurveillance 24, 1–5. https://doi.org/10.2807/1560-7917.ES.2019.24.47.1900679/CITE/REFWORKS.

Manilla, G., 1998. Acari: Ixodida 36, Fauna d'Italia.

Marini, G., Tagliapietra, V., Cristofolini, F., Cristofori, A., Dagostin, F., Zuccali, M.G., Molinaro, S., Gottardini, E., Rizzoli, A., 2023. Correlation between airborne pollen data and the risk of tick-borne encephalitis in northern Italy. Sci. Reports 2023 131 (13), 1–8. https://doi.org/10.1038/s41598-023-35478-w.

Martello, E., Gillingham, E.L., Phalkey, R., Vardavas, C., Nikitara, K., Bakonyi, T., Gossner, C.M., Leonardi-Bee, J., 2022. Systematic review on the non-vectorial transmission of Tick-borne encephalitis virus (TBEv). Ticks Tick. Borne. Dis. 13, 102028. https://doi.org/10.1016/J.TTBDIS.2022.102028.

Mierzejewska, E.J., Welc-Faleciak, R., Karbowiak, G., Kowalec, M., Behnke, J.M., Bajer, A., 2015. Dominance of Dermacentor reticulatus over *ixodes ricinus* (Ixodidae) on livestock, companion animals and wild ruminants in eastern and central Poland. Exp. Appl. Acarol. 66, 83. https://doi.org/10.1007/S10493-015-9889-0.

Ostfeld, R.S., Canham, C.D., Oggenfuss, K., Winchcombe, R.J., Keesing, F., 2006. Climate, deer, rodents, and acorns as determinants of variation in Lyme-disease risk. PLoS Biol. 4, 1058–1068. https://doi.org/10.1371/journal.pbio.0040145.

Randolph, S.E., 2004. Tick ecology: processes and patterns behind the epidemiological risk posed by ixodid ticks as vectors. Parasitology 129, 37–65. https://doi.org/10.1017/S0031182004004925.

Rosà, R., Tagliapietra, V., Manica, M., Arnoldi, D., Hauffe, H.C., Rossi, C., Rosso, F., Henttonen, H., Rizzoli, A., 2019. Changes in host densities and co-feeding pattern efficiently predict tick-borne encephalitis hazard in an endemic focus in northern Italy. Int. J. Parasitol. 49, 779–787. https://doi.org/10.1016/j.ijpara.2019.05.006.

Rubel, F., Walter, M., Vogelgesang, J.R., Brugger, K., 2020. Tick-borne encephalitis (TBE) cases are not random: explaining trend, low- and high-frequency oscillations based on the Austrian TBE time series. BMC Infect. Dis. 20, 448. https://doi.org/10.1186/s12879-020-05156-7.

Ruzek, D., Avšič Županc, T., Borde, J., Chrdle, A., Eyer, L., Karganova, G., Kholodilov, I., Knap, N., Kozlovskaya, L., Matveev, A., Miller, A.D., Osolodkin, D.I., Överby, A.K., Tikunova, N., Tkachev, S., Zajkowska, J., 2019. Tick-borne encephalitis in Europe and Russia: review of pathogenesis, clinical features, therapy, and vaccines. Antiviral Res. 164, 23–51. https://doi.org/10.1016/J.ANTIVIRAL.2019.01.014.

Salomon, J., Hamer, S.A., Swei, A., 2020. A beginner's guide to collecting questing hard ticks (Acari: ixodidae): a standardized tick dragging protocol. J. Insect Sci. 20, 11. https://doi.org/10.1093/JISESA/IEAA073.

Schwaiger, M., Cassinotti, P., 2003. Development of a quantitative real-time RT-PCR assay with internal control for the laboratory detection of tick-borne encephalitis virus (TBEV) RNA. J. Clin. Virol. 27, 136–145. https://doi.org/10.1016/S1386-6532(02)00168-3.

Skarpaas, T., Golovljova, I., Vene, S., Ljøstad, U., Sjursen, H., Plyusnin, A., Lundkvist, Å., 2006. Tick-borne encephalitis virus, Norway and Denmark. Emerg. Infect. Dis. 12, 1136. https://doi.org/10.3201/EID1207.051567.

Sparagano, O., George, D., Giangaspero, A., Špitalská, E., 2015. Arthropods and associated arthropod-borne diseases transmitted by migrating birds. The case of ticks and tick-borne pathogens. Vet. Parasitol. 213, 61–66. https://doi.org/10.1016/J.VETPAR.2015.08.028.

Stoefs, A., Heyndrickx, L., de Winter, J., Coeckelbergh, E., Willekens, B., Alonso-Jiménez, A., Tuttino, A.M., Geerts, Y., Ariën, K.K., van Esbroeck, M., 2021. Autochthonous cases of tick-borne encephalitis, Belgium, 2020. Emerg. Infect. Dis. 27, 2179. https://doi.org/10.3201/EID2708.211175.

Tagliapietra, V., Del Manso, M., Riccardo, F., Palamara, A.T., 2024. TBE in Italy, in: The TBE Book. Global Health Press Pte Ltd, Singapore, pp. 251–253.

Waldenström, J., Lundkvist, Å., Falk, K.I., Garpmo, U., Bergström, S., Lindegren, G., Sjöstedt, A., Mejlon, H., Fransson, T., Haemig, P.D., Olsen, B., 2007. Migrating birds and tick-borne encephalitis virus. Emerg. Infect. Dis. 13, 1215. https://doi.org/10.3201/EID1308.061416.

Wilhelmsson, P., Jaenson, T.G.T., Olsen, B., Waldenström, J., Lindgren, P.E., 2020. Migratory birds as disseminators of ticks and the tick-borne pathogens *Borrelia* bacteria and tick-borne encephalitis (TBE) virus: a seasonal study at Ottenby Bird Observatory in south-eastern Sweden. Parasites Vectors 13, 1–17. https://doi.org/10.1186/S13071-020-04493-5/FIGURES/6.