

# Questing *Ixodes ricinus* ticks and *Borrelia* spp. in urban green space across Europe: A review

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## Abstract

For more than three decades, it has been recognized that *Ixodes ricinus* ticks occur in urban green space in Europe and that they harbour multiple pathogens linked to both human and animal diseases. Urban green space use for health and well-being, climate mitigation or biodiversity goals is promoted, often without consideration for the potential impact on tick encounters or tick-borne disease outcomes. This review synthesizes the results of over 100 publications on questing *I. ricinus* and *Borrelia* spp. infections in ticks in urban green space in 24 European countries. It presents data on several risk indicators for Lyme borreliosis and highlights key research gaps and recommendations for future studies. Across Europe, mean density of *I. ricinus* in urban green space was 6.9 (range; 0.1–28.8) per 100 m<sup>2</sup> and mean *Borrelia* prevalence was 17.3% (range; 3.1%–38.1%). Similar density estimates were obtained for nymphs, which had a *Borrelia* prevalence of 14.2% (range; 0.5%–86.7%). Few studies provided data on both questing nymph density and *Borrelia* prevalence, but those that did found an average of 1.7 (range; 0–5.6) *Borrelia*-infected nymphs per 100 m<sup>2</sup> of urban green space. Although a wide range of genospecies were reported, *Borrelia afzelii* was the most common in most parts of Europe, except for England where *B. garinii* was more common. The emerging pathogen *Borrelia miyamotoi* was also found in several countries, but with a much lower prevalence (1.5%). Our review highlights that *I. ricinus* and tick-borne *Borrelia* pathogens are found in a wide range of urban green space habitats and across several seasons. The impact of human exposure to *I. ricinus* and subsequent Lyme borreliosis incidence in urban green space has not been quantified. There is also a need to standardize sampling protocols to generate better baseline data for the density of ticks and *Borrelia* prevalence in urban areas.

## KEYWORDS

*Borrelia miyamotoi*, global change, Lyme disease, public health, ticks, urban ecology, urbanization

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## 1 | INTRODUCTION

Lyme borreliosis remains the most important tick-borne disease in Europe and North America (Killilea et al., 2008; Sykes & Makiello, 2016). Incidence has been increasing in many European countries, with 5%–15% average annual increases reported (Vandekerckhove et al., 2019). In some areas, similar increases in both rural and urban locations have been observed (Brzozowska et al., 2021). Transmitted via the bite of infected ticks, *Borrelia burgdorferi* sensu lato can infect humans, resulting in over 650,000 cases in the United States and Western Europe annually (Marques et al., 2021). The distribution of Lyme borreliosis is directly linked to a range of wildlife hosts that feed, infect and transport ticks, and there is risk of transmission in areas where members of the public may encounter infected ticks (Pfäffle et al., 2013). *Ixodes ricinus* is the main vector of Lyme borreliosis in most of Europe, although *Ixodes persulcatus* also plays a key role in North-Eastern Europe (Mencke, 2013). *Ixodes ricinus* can be found in a range of habitats including woodland, moorland, scrub and rough pasture, heathland, and also urban parks and gardens, where local establishment is dependent on suitable microclimate and host availability (Anderson & Magnarelli, 2008; Estrada-Peña, 2015; Randolph, 2000). Host composition also plays a key role in the local prevalence of *B. burgdorferi* s.l. (Braks et al., 2016), with different *Borrelia* genospecies linked to different host species and associated with different clinical presentations (Mannelli et al., 2012; Radolf et al., 2012; Sprong et al., 2018).

The acquisition of Lyme borreliosis in humans is associated with outdoor activities, and urban green space has been identified as an area for potential human/tick exposure (Rizzoli et al., 2014). During the last 32 years, numerous studies have been conducted to investigate tick presence, density and *Borrelia* prevalence across Europe (Appendix S1). Understanding the distribution and abundance of ticks and prevalence of tick-borne pathogens is of paramount importance, because urban green spaces are key targets for habitat change, to fulfil policy goals relating to climate adaptation, biodiversity and health and well-being (Defra, 2011; Her Majesty's Government, 2018; Lovell et al., 2018; Natural England, 2003; Twohig-Bennett & Jones, 2018; Wheeler et al., 2015). The dramatic changes already occurring and planned for urban expansion and greening could be setting a course for the emergence of urban Lyme borreliosis hot spots, which is of increasing concern if members of the public and clinicians are unaware of the risk (Bayles et al., 2013; Piesman & Gern, 2004).

Here, we conduct a qualitative review of this previous work on questing *I. ricinus* density and infection with *B. burgdorferi* s.l. in peri/urban green space across Europe. Previously, Rizzoli et al. (2014) provided an extensive narrative review of urban tick species, host ecology and reported prevalence rates of associated tick-borne pathogens. Furthermore, Grochowska et al., (2020) quantified pathogen prevalence in urban ticks in relation to climate and geographical location. Our current review updates these previous studies by incorporating data from an additional 68 publications not previously captured. The density of ticks, prevalence of *Borrelia*

### Impacts

- Urban green space provides habitat for *Borrelia*-infected *Ixodes ricinus*, and exposure of humans to infected ticks could potentially occur within several habitat types and across seasons.
- The density of ticks and prevalence of *Borrelia* is variable, but high in some urban locations, and on average, up to 2 nymphs per 100 m<sup>2</sup> are infected with a range of *Borrelia* spp.
- Clearer descriptions of urban green space sampling locations, tick density assessments and prevalence estimates are needed. Linking human tick bites or disease incidence with ecological data sets could help further assess the impact of *Ixodes ricinus* in urban green space.

within the tick population and the density of infected nymphs can be useful indicators of Lyme borreliosis risk (Estrada-Peña & De La Fuente, 2014; Killilea et al., 2008; Kilpatrick et al., 2017). Data on these risk indicators were captured and presented at the study, country and European level to bring together the latest data and current knowledge on urban *I. ricinus* and *B. burgdorferi* s.l. in Europe. An additional aim of this review was to identify and discuss key research gaps and to make recommendations for further developing our understanding of urban tick risk.

## 2 | METHODS

### 2.1 | Literature review

We performed an extensive literature review on field-based research that quantified questing *I. ricinus* density and/or *B. burgdorferi* s.l. or *Borrelia miyamotoi* prevalence in *I. ricinus* in peri/urban areas across Europe, using EMBASE (343 publications), MEDLINE (309 publications) and Scopus (538 publications). Literature published up until the 31 August 2021 was considered, and articles were restricted to those written in English. The following search terms were used; tick\* OR "*Ixodes ricinus*" OR *Borrelia* OR Lyme disease OR Borreliosis AND urban\* OR park OR city OR recreation with each country in Europe also included. *Ixodes ricinus* was targeted, as this species has the most significant impact on public health and is the most widespread across Europe (Medlock et al., 2013).

Reference management was performed in EndNote X9. Of 1190 publications identified, duplicate titles were removed, and 601 publications remained. Titles and abstracts were screened to identify studies estimating one or more of the following: tick presence, density, *Borrelia* spp. detection/prevalence or the density of infected nymphs. A total of 115 publications were identified for further review (Appendix S1). Some studies also contained data on rural survey locations. Data from rural locations were removed from study totals

for density or prevalence if study authors described them as rural or “natural” habitat and when searching Google maps, they appeared to be away from peri/urban environment. Some studies included data on non-*Borrelia* pathogens. Tick presence and density data were extracted from such non-*Borrelia* studies, but pathogen data were not included, as this was outside the scope of this review. Notes are provided in Appendix S1 to indicate studies that captured non-*Borrelia* pathogens. This review did not include data on *Borrelia* spp. detected in ticks removed from animals (following Estrada-Peña et al., 2018).

## 2.2 | Data extraction and analysis

Data captured from each study included the following: country, publication year, city, total ticks and total individual ticks per life stages, the distance/time unit sampled, the method used for *Borrelia* spp. detection (if applicable), the total ticks/life stages tested for *Borrelia*, the number of ticks testing positive and genospecies composition. Location information was also captured (if available), along with site descriptions. Data were also captured or generated to estimate five Lyme borreliosis risk indicators; mean density of questing ticks (DOT) and density of questing nymphs (DON); *Borrelia burgdorferi* s.l. tick infection prevalence (TIP; adults and nymphs only) and questing nymph infection prevalence (NIP); and the density of infected nymphs (DIN). Additionally, *B. burgdorferi* s.l. infection prevalence in adult ticks was also captured (AIP).

Study means for DOT or DON per 100 m<sup>2</sup> were captured directly from published studies or calculated using raw data on the total ticks (adults and nymphs only) and nymphs collected, and total distance sampled. Only raw data were used to estimate overall mean DOT and DON per 100 m<sup>2</sup> for urban green space in Europe. Only studies using a standardized method of blanket dragging/flagging to collect ticks (Estrada-Peña et al., 2013) were included in DOT and DON. DOT and DON were not available for all studies. Details of other studies estimating density per unit of time are captured in Appendix S1.

For TIP, AIP and NIP, total adults and nymphs, total adults and total nymphs, respectively, tested and testing positive for *B. burgdorferi* s.l. were used to estimate prevalence at the country and European levels. Study means and ranges of prevalence are presented in Appendix S1, and 95% confidence intervals for country- and European-level estimates were calculated using the Rmisc function in R (Hope, 2013). Country and European-level *B. burgdorferi* s.l. estimates were generated from studies using PCR for detection in individually tested ticks only. Studies using dark-light microscopy or direct immunofluorescence or studies with pooled samples are captured in Appendix S1. Studies which included *Borrelia miyamotoi* in overall *Borrelia* spp. estimates, without a clear breakdown of numbers, were excluded from TIP, AIP, NIP and DIN, but details of these are provided in Appendix S1.

Mean DIN was calculated per study by multiplying DON by NIP (from figures presented in publications or from calculations described above). Mean DIN was also calculated at the European level,

but only included studies with total nymphs and distance presented, and nymphs tested individually/positive, using PCR as the method of detection. DIN based on nymphs collected per hour, alternative detection methods or pooled samples are included in Appendix S1.

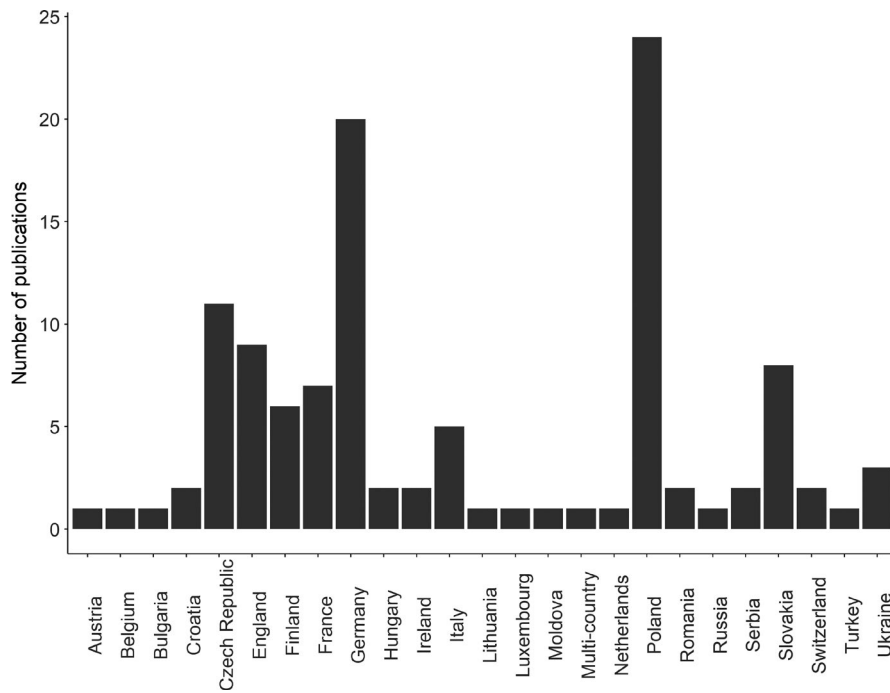
The proportion of genospecies was estimated for each country and overall for Europe using the total number of successfully genotyped, individually tested ticks and the total number of each genospecies. Studies using pooled data were excluded, along with studies that did not provide a breakdown of the number of ticks testing positive for each genospecies. Studies not able to differentiate genospecies were also excluded. For country-level proportions, studies with fewer than 30 ticks successfully genotyped were excluded, but these studies were included for overall estimates for Europe. Finally, publications reporting the detection of *Borrelia miyamotoi* were used to estimate an overall mean prevalence for Europe, with 95% confidence intervals. These studies included all life stages, as *B. miyamotoi* is transovarially transmitted (Richter et al., 2012). Further details of genospecies reported in each study, including co-infections and *B. miyamotoi* detection, can be found in Appendix S2. Data were visualized using the ggplot2 package (Wickham, 2016) in R version 4.1.1 (R Core Development Team, 2019).

## 3 | RESULTS

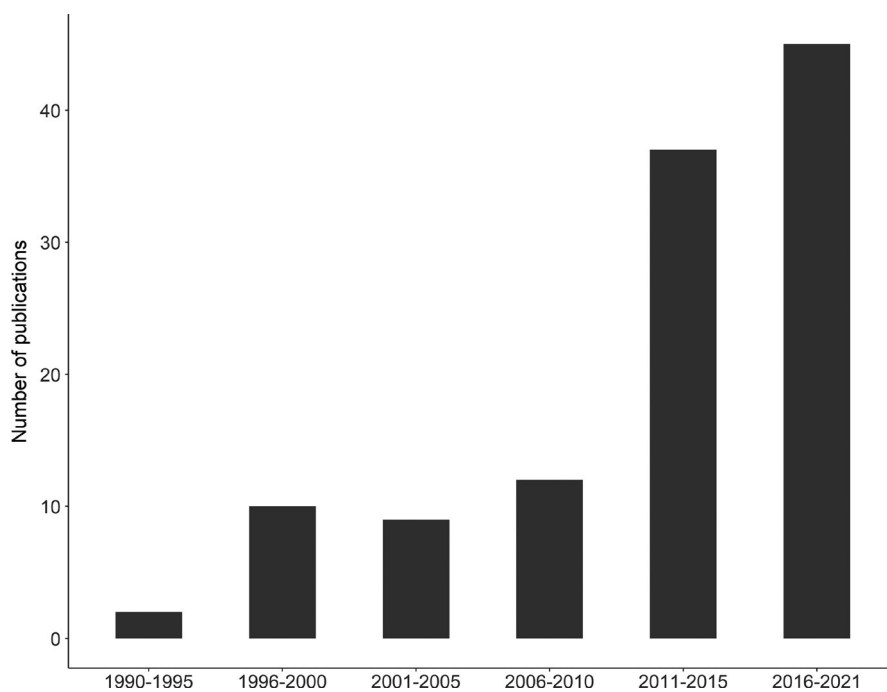
*Ixodes ricinus* ticks have been reported in the literature from urban green space in 24 countries across Europe (Figure 1) based on an analysis of 115 studies published during the period 1990–2021. Most studies were conducted in central and western Europe, with Poland, Germany, Czech Republic, England, Slovakia and France contributing the most data (68.7%;  $n = 79$ ). All other countries had six or fewer studies, with the majority having just one study (Figure 1). The number of publications investigating questing *I. ricinus* in urban green space has increased over time, with almost 50 published in the past 6 years (Figure 2).

The simultaneous presence of all life stages of *I. ricinus* in urban green space was reported from 40.0% ( $n = 46$ ; Appendix S1) of studies conducted in 15 countries covering all European regions. Where total ticks and total nymphs were available (85 studies; 73.3%), nymphs made up the largest proportion of life stages collected (64.4%). Where adults were collected and sex reported (62 studies; 53.4%), the proportion of males (49.6%) and females (50.4%) was similar. The presence of larvae was reported in over a third ( $n = 46$ ; 40.0%) of all studies. Whilst tick presence was reported from all studies, apparent absence of ticks was reported from several locations in 10 studies located in Belgium, Croatia, England, Finland, Germany, Ireland, Italy and Switzerland (Gray et al., 1999; Hansford et al., 2021; Heylen et al., 2019; Klemola et al., 2019; Krčmar et al., 2014; Maetzel et al., 2005; Mäkinen et al., 2003; Nelson et al., 2015; Oechslin et al., 2017; Olivieri et al., 2017).

A range of habitat types were sampled and found to have ticks, including large city parks, urban or suburban forests (including deciduous, mixed, coniferous), cemeteries, meadows and botanical



**FIGURE 1** Total number of publications per country contributing to the assessment of risk posed by questing *Ixodes ricinus* in peri/urban areas of Europe, during the period 1990–2021



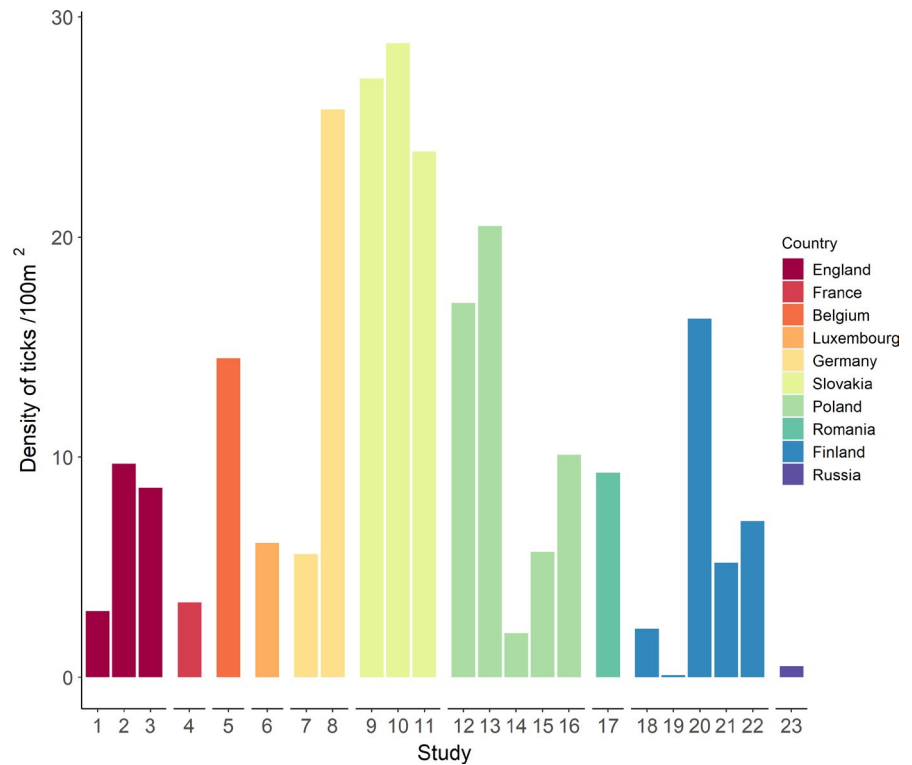
**FIGURE 2** Total number of publications per year contributing to the assessment of risk posed by questing *Ixodes ricinus* in peri/urban areas of Europe, during the period 1990–2021

gardens. The location of study sites within cities varied from centrally isolated patches, to larger spaces with greater connectivity to surrounding rural habitat. Some studies focused on single locations with known tick populations (e.g. Richmond Park, London and Senart Forest, Paris), whereas others assessed a broader range of urban green space habitats across cities. Different approaches were also taken regarding the timing of data collection, with some studies targeting peak questing periods only, and others capturing all seasons, or multiple years. In addition to tick presence, density and prevalence assessments, some studies investigated the effects of microclimate or weather, habitat or connectivity factors, with a handful of studies incorporating less common aspects such as the

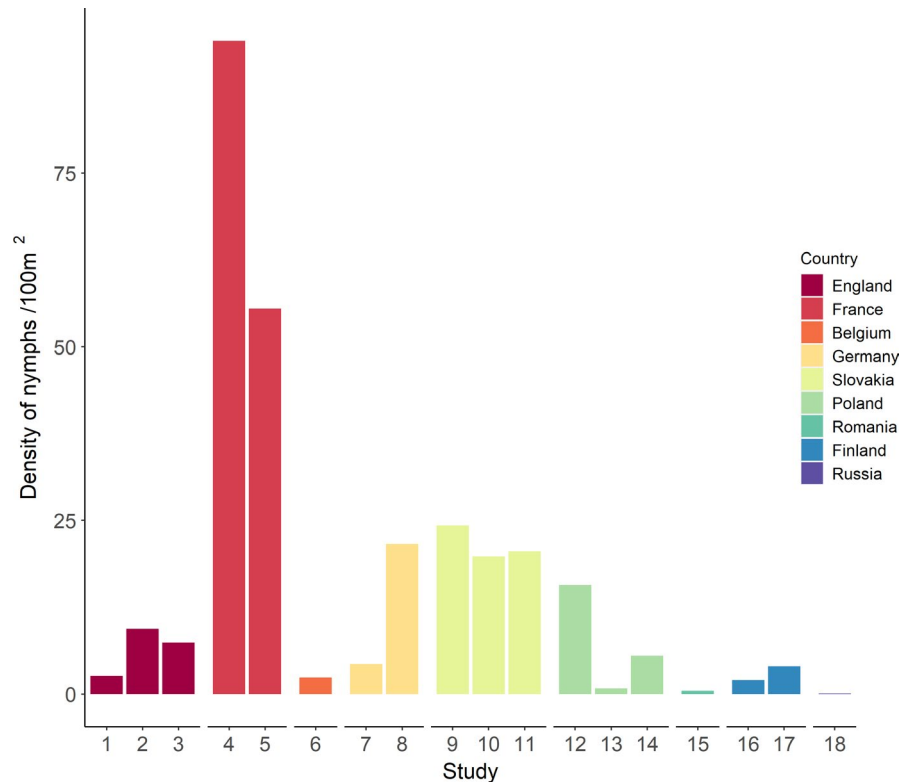
effect of tick/*Borrelia* spp. hosts (small mammals, birds), the impact on Lyme borreliosis incidence locally, the efficacy of repellents and the effect of tick exoskeletal anomalies (Appendix S1).

Tick density assessments were available in 38 (33.0%) publications, and most studies used distance as the unit of measure ( $n = 30$ ) rather than time ( $n = 8$ ). Tick density (all tick life stages) ranged from 0 to 152.3 per 100 m<sup>2</sup> and 2–138/h (Appendix S1) for all locations included per study. Study means, using adults and nymphs only, were available from 23 publications (Figure 3) and sixteen provided data (183 individual datapoints) for calculating DOT per 100 m<sup>2</sup> for Europe overall (6.9 ticks per 100 m<sup>2</sup>). Nymph density was reported in 29 publications (25.2%, 26 using

**FIGURE 3** Mean tick density of *Ixodes ricinus* per 100 m<sup>2</sup> from studies in peri/urban green space across Europe (ordered West to East) during the period 1990–2021; 1 (Hansford et al., 2017); 2 (Greenfield, 2011); 3 (Hansford et al., 2021); 4 (Mathews-Martin et al., 2020); 5 (Heylen et al., 2019); 6 (Reye et al., 2010; Junntila et al., 1999); 7 (Maetzel et al., 2005); 8 (Hauck et al., 2020); 9 (Chvostáč et al., 2018); 10 (Minichová et al., 2017); 11 (Svitáľková et al., 2015); 12 (Bukowska et al., 2003); 13 (Kubiak & Dziekońska-Rynko, 2006); 14 (Kubiak et al., 2019); 15 (Pet'ko et al., 1997); 16 (Kowalec et al., 2017); 17 (Borşan et al., 2020); 18 (Klemola et al., 2019); 19 (Mäkinen et al., 2003); 20 (Junntila et al., 1999); 21 (Sormunen et al., 2020); 22 (Cayol et al., 2017); 23 (Makenov et al., 2019)



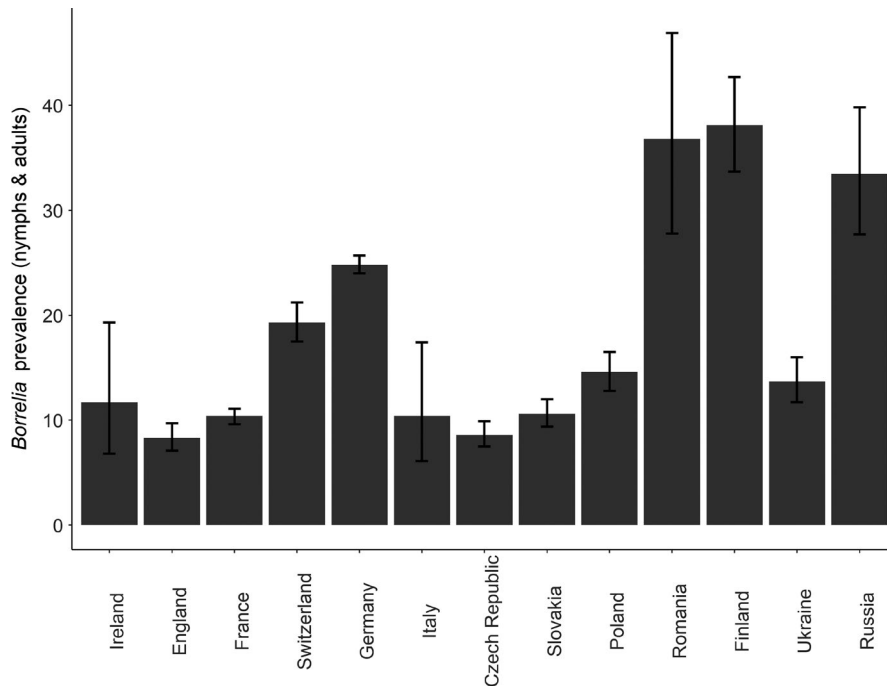
**FIGURE 4** Mean nymph density of *Ixodes ricinus* per 100 m<sup>2</sup> from studies in peri/urban green space across Europe (ordered West to East) during the period 1990–2021; 1 (Hansford et al., 2017); 2 (Greenfield, 2011); 3 (Hansford et al., 2021); 4 (Marchant et al., 2017); 5 (Vourc'h et al., 2016); 6 (Heylen et al., 2019); 7 (Maetzel et al., 2005); 8 (Hauck et al., 2020); 9 (Chvostáč et al., 2018); 10 (Svitáľková et al., 2015); 11 (Minichová et al., 2017); 12 (Bukowska et al., 2003); 13 (Pet'ko et al., 1997); 14 (Kowalec et al., 2017); 15 (Borşan et al., 2020); 16 (Klemola et al., 2019); 17 (Sormunen et al., 2020); 18 (Makenov et al., 2019)



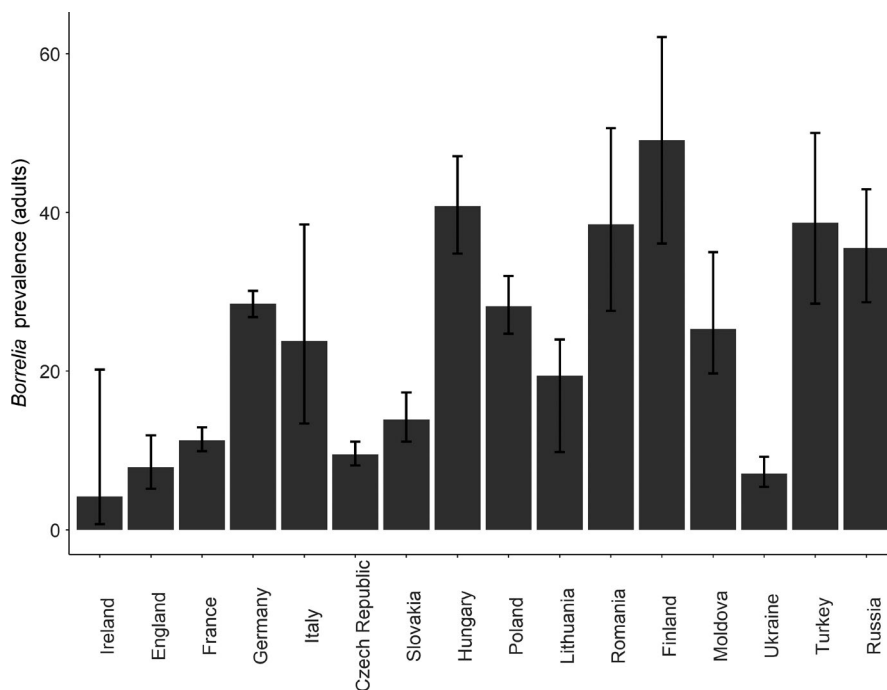
distance, three using time) and ranged from 0 to 159.5 per 100 m<sup>2</sup> and 4.7–75.0/h (Appendix S1). Study means were available in 18 publications (Figure 4), and information on distance sampled and total nymphs collected to calculate DON was available in 15 publications (12.9%; 178 individual datapoints). Overall mean density of nymphs in urban green space in Europe was 12.2 nymphs per 100 m<sup>2</sup>. When removing data from studies in France, which did not

include data to calculate mean DOT, the overall mean density of nymphs for Europe was 6.1 per 100 m<sup>2</sup>.

Ticks were tested for *B. burgdorferi* s.l. via several methods in 57 studies (49.6%), the most popular being PCR (real-time or conventional;  $n = 46$ ) on individual tick DNA extractions as opposed to pooled samples ( $n = 11$ ). Overall mean *B. burgdorferi* s.l. infection prevalence of individually tested ticks reached 38.1% (168/441



**FIGURE 5** Mean *Borrelia burgdorferi* sensu lato prevalence in *Ixodes ricinus* adults and nymphs collected from peri/urban green space across Europe (ordered West to East), during the period 1990–2021. Error bars show 95% confidence intervals. Number of analysed ticks per country; Czech Republic ( $n = 2083$ ); England ( $n = 1739$ ); Finland ( $n = 441$ ); France ( $n = 6199$ ); Germany ( $n = 9755$ ); Ireland ( $n = 103$ ); Italy ( $n = 115$ ); Poland ( $n = 1434$ ); Romania ( $n = 95$ ); Russia ( $n = 227$ ); Slovakia ( $n = 2145$ ); Switzerland ( $n = 1798$ ); Ukraine ( $n = 976$ )



**FIGURE 6** Mean *Borrelia burgdorferi* sensu lato prevalence in *Ixodes ricinus* adult ticks collected from peri/urban green space across Europe (ordered West to East), during the period 1990–2021. Error bars show 95% confidence intervals. Number of analysed adults per country; Czech Republic ( $n = 1485$ ); England ( $n = 252$ ); Finland ( $n = 53$ ); France ( $n = 1676$ ); Germany ( $n = 2854$ ); Hungary ( $n = 240$ ); Ireland ( $n = 24$ ); Italy ( $n = 42$ ); Lithuania ( $n = 36$ ); Moldova ( $n = 198$ ); Poland ( $n = 571$ ); Romania ( $n = 65$ ); Russia ( $n = 172$ ); Slovakia ( $n = 475$ ); Turkey ( $n = 75$ ); Ukraine ( $n = 679$ )

tested) in one study in Finland (Figure 5), but most studies reported a prevalence of less than 17.0% (Appendix S1). Total ticks (adults and nymphs) tested and total ticks positive were available from 32 (27.8%) publications. Overall, mean TIP was 17.3% (4568/26,374; 95% CI 16.9%–17.8%). Forty publications (34.8%) provided data to calculate prevalence in adult ticks (Figure 6), with a mean AIP of 21.1% (1648/7797; 95% CI: 20.2%–22.1%). Finally, 33 publications (28.7%) provided data to calculate NIP (Figure 7), with 14.2% of nymphs testing positive (3154/22,179; 95% CI: 13.8%–14.7%).

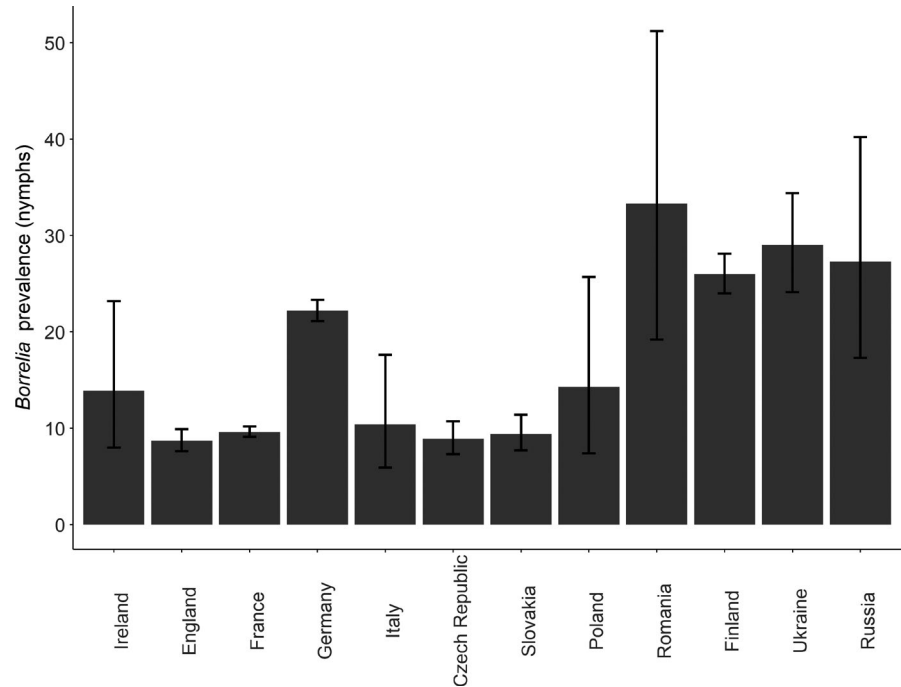
The density of infected nymphs (DIN) per 100 m<sup>2</sup> was available in 10 (8.7%) publications and ranged from 0.03 to 9.8 (Figure 8; Appendix S1). Only eight studies provided data on total nymphs

collected, tested and positive for *B. burgdorferi* s.l., as well as distance sampled. Between these studies, 24,947 nymphs were collected from 207,425 m<sup>2</sup> giving a DON of 12.0 per 100 m<sup>2</sup>. In total, 8090 nymphs were tested via PCR, and 1167 (14.4%; 95% CI: 13.7%–15.2%) were positive for *B. burgdorferi* s.l. Mean DIN for all available data in studies conducted in urban green space in Europe is therefore estimated at 1.7 per 100 m<sup>2</sup>, but as shown in Figure 8, variation across locations is very large.

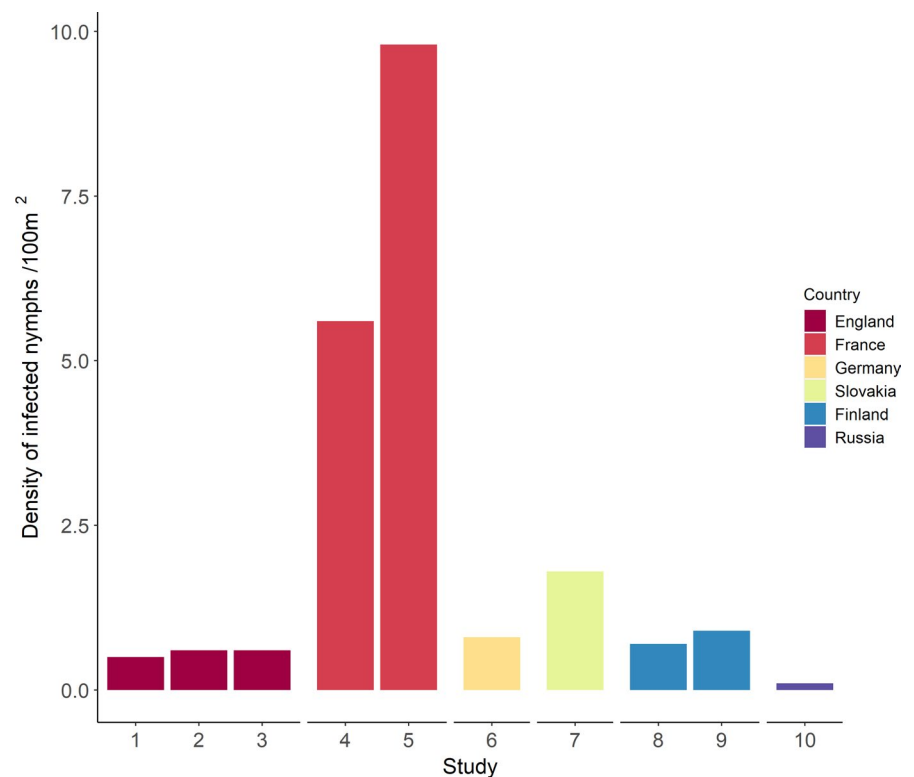
*Borrelia burgdorferi* s.l. genospecies were investigated in 48 (41.7%) studies across 21 countries. The dominant genospecies reported was *Borrelia afzelii*, which was also the dominant genospecies (48.1%) based on a closer review of 29 of these publications



**FIGURE 7** Mean *Borrelia burgdorferi* sensu lato prevalence in *Ixodes ricinus* nymphs collected from peri/urban green space across Europe (ordered West to East), during the period 1990–2021. Error bars show 95% confidence intervals. Number of analysed nymphs per country; Czech Republic ( $n = 1079$ ); England ( $n = 2400$ ); Finland ( $n = 1774$ ); France ( $n = 9874$ ); Germany ( $n = 5429$ ); Ireland ( $n = 79$ ); Italy ( $n = 106$ ); Poland ( $n = 56$ ); Romania ( $n = 30$ ); Russia ( $n = 55$ ); Slovakia ( $n = 1000$ ); Ukraine ( $n = 297$ )

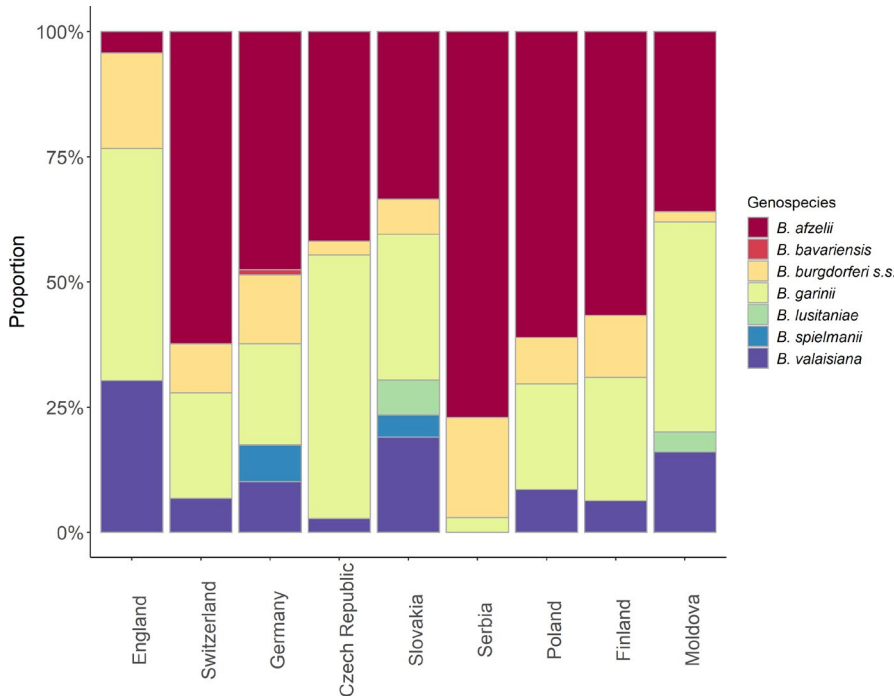


**FIGURE 8** Mean density of *Borrelia burgdorferi* sensu lato infected *Ixodes ricinus* nymphs per 100 m<sup>2</sup> from studies in peri/urban green space across Europe (ordered West to East), during the period 1990–2021; 1 (Hansford et al., 2017); 2 (Hansford et al., 2021); 3 (Nelson et al., 2015); 4 (Marchant et al., 2017); 5 (Vourc'h et al., 2016); 6 (Maetzel et al., 2005); 7 (Chvostáč et al., 2018); 8 (Klemola et al., 2019); 9 (Sormunen et al., 2020); 10 (Makenov et al., 2019)



that provided information on the number of ticks testing positive for each genospecies (Figure 9). *Borrelia garinii* was the second most detected (24.6%), followed by *Borrelia burgdorferi* sensu stricto (11.9%) and *Borrelia valaisiana* (10.8%). Other genospecies detected included *Borrelia spielmanii* (3.0%), *Borrelia lusitaniae* (1.2%), *Borrelia bavariensis* (0.4%) and *Borrelia bissettii* (no prevalence data available) (Figure 9; Appendix S2), which made up the minority. Many different genospecies co-infections were reported, but overall sample sizes were often small making associations hard to ascertain

(Appendix S2). *Borrelia miyamotoi* was detected in questing ticks collected from peri/urban areas from ten countries (Belgium, England, Finland, France, Germany, Poland, Romania, Slovakia, Switzerland and Ukraine; Appendix S2). Mean prevalence per study was always lower compared with *Borrelia burgdorferi* s.l., ranging from 0.4% to 8.9% at individual study locations, with the overall study mean only exceeding 2.5% in one study (Blazejak et al., 2018). Overall mean prevalence in all life stages for all studies was low (1.5%; 95% CI: 1.2%–1.8%).



**FIGURE 9** Proportion of *Borrelia burgdorferi* sensu lato genospecies per country in *Ixodes ricinus* ticks collected from peri/urban green space across Europe (ordered West to East), during the period 1990–2021

## 4 | DISCUSSION

This review aimed to summarize available data on questing ticks in urban green space in Europe and to identify research gaps. Focusing on several Lyme disease risk indicators, estimates for tick density (DOT), nymph density (DON), *B. burgdorferi* s.l. prevalence (TIP, NIP) and the density of infected nymphs (DIN) have been obtained, along with the diversity and proportion of urban *Borrelia* genospecies in questing *I. ricinus*. In some study locations, tick or nymph density exceeded 100 per 100 m<sup>2</sup> (Brugger et al., 2016; Dobson et al., 2011; Hauck et al., 2020; Marchant et al., 2017; Svitálková et al., 2015), *B. burgdorferi* s.l. prevalence was higher than 30% (Fingerle et al., 1999; Junttila et al., 1999; Klemola et al., 2019; Makenov et al., 2019; May et al., 2015; Rogovskyy et al., 2018) and more than 10 infected nymphs could be found in every 100 m<sup>2</sup> of urban green space (Marchant et al., 2017). A wide range of genospecies were also detected, along with the emerging tick-borne pathogen, *B. miyamotoi*. Furthermore, an understanding of the current state of research has highlighted key gaps in our knowledge. Such gaps reduce the impact of local interventions and enable urban green space planning and management to continue without consideration of the impact of tick-borne diseases.

A small proportion of publications provided a standardized measure of tick/nymph density per unit area, making it difficult to compare studies and eliminating the opportunity to calculate DIN in most cases. When available, overall density of nymphs and adults combined, or nymphs alone, was 6–7 per 100 m<sup>2</sup>. Overall densities were low, and studies reported variation between habitat types, urban gradient and season. High densities (50– > 100 ticks per 100 m<sup>2</sup>) were typically reported in woodland habitat, including both centrally located parks and fringe habitat in and around the cities of Bratislava, Hanover, London and Paris (Dobson et al., 2011; Hauck et al., 2020; Marchant et al., 2017; Svitálková et al., 2015). Some of these locations had established tick populations supported by deer

(which may not be typical of urban green space) and a suitable microclimate due to the wilder nature of the areas and increased leaf litter (Schorn et al., 2011). Woodland habitats were often reported to have significantly higher tick/nymph densities (Di Luca et al., 2013; Hansford et al., 2017; Krstić et al., 2016) and activity periods started sooner compared with other urban habitat types (Dobson et al., 2011). A number of studies also suggested that the type of peri/urban woodland can influence tick density, with deciduous and mixed woodland having higher densities compared with coniferous (Michalik et al., 2003; Vourc'h et al., 2016).

Although peri/urban woodland may be important for *I. ricinus* and could be a key area for Lyme borreliosis acquisition, this habitat was not always associated with ticks (Mäkinen et al., 2003) or the highest densities across an urban landscape (Borşan et al., 2020) and other urban green spaces (cemeteries, parks and gardens) were found to be important (Hornok et al., 2014; Pet'ko et al., 1997; Rogovskyy et al., 2018). Urban gradient studies also found mixed results, with some suggesting higher levels of urbanization were associated with lower tick densities in urban green space (Buczek et al., 2014; Hauck et al., 2020; Stańczak et al., 2015), whereas others suggested the opposite (Kowalec et al., 2017; Kubiak et al., 2019; Svitálková et al., 2015). This discrepancy is likely explained by local host availability for feeding and infecting ticks, the types of habitat surveyed and connectivity with other suitable tick habitats (Daniel & Cerny, 1990; Heylen et al., 2019; Maetzel et al., 2005; Rollins et al., 2021; Vaculová et al., 2019). Based on study site descriptions, lower risk urban green spaces associated with apparent absences of ticks appear to be centrally located urban parks of varying size, surrounded by the built environment and lacking connectivity to surrounding rural habitat. In these locations, it is likely that the lack of wildlife (perhaps due to poor connectivity), habitat suitability and management practices such as grass cutting are having a negative impact on tick presence and survival (Hansford et al., 2021;



Heylen et al., 2019; Krčmar et al., 2014; Maetzel et al., 2005; Nelson et al., 2015; Oechslin et al., 2017).

Variation in density was observed depending on the season, with some evidence for unimodal activity and spring peaks (Buczek et al., 2014; Bukowska et al., 2003; Drelich et al., 2014; Hansford et al., 2017; Kazimirová et al., 2016; Lejal et al., 2019; Mehlhorn et al., 2016; Oechslin et al., 2017; Olivieri et al., 2017; Pangráčová et al., 2013; Vogelgesang et al., 2020; Vollmer et al., 2011; Vucelja et al., 2019; Zákovská et al., 2008) and others with bimodal patterns and second peaks later in the year (Boršan et al., 2020; Cayol et al., 2017; Hauck et al., 2020; Michalik et al., 2003; Sormunen et al., 2020; Szekeres et al., 2017; Tappe et al., 2014; Vollack et al., 2017). This suggests a wide range of urban green spaces can support ticks, and tick exposure could occur at multiple times throughout the year.

Almost half of studies provided some assessment of *B. burgdorferi* s.l. prevalence. Overall prevalence in questing *I. ricinus* collected in peri/urban green space (adults 21.1%, nymphs 14.2%) appears to be slightly elevated compared with other estimates for *I. ricinus* across Europe (including both urban and rural locations; adults 14.9%–18.6%, nymphs 10.1%–11.8%), which also found higher prevalence in adults compared with nymphs (Hubálek & Halouzka, 1998; Rauter & Hartung, 2005; Strnad et al., 2017). This highlights that prevalence may be significant in and around urban areas. In fact, some individual survey locations had a prevalence over 30% (with sample sizes over 100), including popular central urban parks in Bern, Hamburg, Hanover, Helsinki, Moscow, Munich and Turku (Blazejak et al., 2018; Fingerle et al., 1999; Junttila et al., 1999; Klemola et al., 2019; May et al., 2015; Oechslin et al., 2017; Tappe et al., 2014).

Much like density, prevalence in some areas appeared to be influenced by habitat, level of urbanization and season. Forest habitat in Budapest, Hungary (Hornok et al., 2014), and peri-urban woodland in Hanover, Germany (Tappe et al., 2014), had higher prevalence compared with other study areas without forest/woodland. Furthermore, a study in Luxembourg found a positive association between prevalence and an increasing level of urbanization (Reye et al., 2010) and similar results were reported in southern Germany in a study that found higher prevalence in urban compared with rural sites (Răileanu et al., 2021). In some studies, however, *B. burgdorferi* s.l. prevalence did not appear to differ significantly across peri/urban green space, including areas with and without woodland habitat (Cekanac et al., 2010; Hansford et al., 2017; Krstić et al., 2016; Kubiak et al., 2019; Maetzel et al., 2005; Michalik et al., 2003; Oechslin et al., 2017; Pet'ko et al., 1997; Plch & Basta, 1999). The effects of the level of urbanization on prevalence should be further investigated.

Differences in *B. burgdorferi* s.l. prevalence across locations are likely due to local host composition and perhaps the natural variation in *Borrelia* transmission cycles that can occur. For example, *B. burgdorferi* s.l. was not detected in over 100 ticks collected from a large deer park in central London (Hansford et al., 2015), despite other studies in the same park finding up to 8% of ticks infected (Hansford et al., 2021; Nelson et al., 2015; Sorouri et al., 2015). Other studies have also found significant annual fluctuations in prevalence (Marchant et al., 2017; Michalik et al., 2003) and seasons,

with peaks identified in autumn in peri-urban woodland in France (Lejal et al., 2019), in August in a suburban park in Brno, Czech Republic (Žáková et al., 2013) and in April in urban forest in South-Western Slovakia (Chvostáč et al., 2018). Other studies have found a lack of variation across season (Cekanac et al., 2010), with some even reporting stability in prevalence over several years (Blazejak et al., 2018). This highlights that seasonal fluctuations could occur and in some instances may coincide with peak use of such spaces by members of the public.

The density of ticks and prevalence can be good indicators of risk, but the density of infected nymphs is perhaps most relatable to Lyme borreliosis incidence (Braks et al., 2016). Although most studies did not report data to calculate DIN, those that did provided evidence of up to 10 infected nymphs per 100 m<sup>2</sup> in some instances (Marchant et al., 2017). This appeared to be primarily driven by the density of nymphs (rather than high prevalence), which has been highlighted as a key driver for increased DIN in other studies (Braks et al., 2016; Randolph, 2001). If DON is a significant contributor to DIN, density assessments alone could be underestimated in their potential to provide assessments of risk locally. The study in France with high DIN and all other study locations with a DIN of 2 to 10 per 100 m<sup>2</sup> were reported in peri/urban woodland habitat. Once taking into consideration all habitat types from all study locations, DIN was found to be less than two per 100 m<sup>2</sup>. This reduction in DIN is likely due to the inclusion of less suitable urban green space habitat that may have fewer reservoir hosts, may be located in parts of a city that is less accessible to wildlife or may not have suitable habitat/microclimate to support the tick life cycle. This has already been demonstrated in Belgium where DIN was negatively associated with increased urbanization or lack of connectivity to rural space (Heylen et al., 2019). How peri/urban DIN translates into Lyme borreliosis incidence is not yet clear, and most studies did not incorporate clinical or tick-bite data.

*Borrelia afzelii* was the dominant genospecies or made up a large proportion of genospecies in most countries. This finding is supported by other reviews of genospecies data across Europe (Hubálek & Halouzka, 1998; Rauter & Hartung, 2005; Strnad et al., 2017). Genospecies composition in England appears to be very different to all other countries, being dominated by bird-associated genospecies (Mannelli et al., 2012; Mencke, 2013; Piesman & Gern, 2004; Tilly et al., 2008). The reason for such higher prevalence of *B. garinii* in England is not fully understood, but this follows the same trend as a country-wide study in England where it was suggested there could be a link to increased distribution of game birds (Cull et al., 2021) which can be key hosts for *B. garinii* (Hoodless et al., 1998; Kurtenbach et al., 1998). The reporting of seven different *B. burgdorferi* s.l. genospecies, *Borrelia* spp. co-infections, as well as *B. miyamotoi* shows that urban green space can harbour a wide range of Borrelial pathogens. *Borrelia miyamotoi* was less likely to be found in infected ticks compared with *B. burgdorferi* s.l., but despite the lower prevalence, the detection of this pathogen, as well as *Anaplasma phagocytophilum*, *Babesia* spp, *Rickettsia* spp and Tick-borne encephalitis virus in several cities across Europe (Appendix S1) highlights these areas as risk zones for tick-borne pathogen exposure. The transient or established nature of these pathogens will vary locally, but it is likely

that exposure to at least *B. afzelii* and *B. garinii* is common in urban green space.

The vast difference in study locations (including lack of clarity on urban/peri-urban classification of survey sites), timings and methods to collect and test ticks could have influenced density and prevalence estimates generated in this review. Mean densities reported would have been higher if conducted in known tick areas during peak questing seasons, compared to broader urban green spaces sampled throughout or over multiple years. It is also known that blanket dragging to estimate density can vary dramatically between surveyors (Nyrhilä et al., 2020), and this too could have influenced density and ultimately, DIN. To minimize the impact of different pathogen detection methods, only PCR results were included in prevalence assessments (following Estrada-Peña et al., 2018) and only papers differentiating genospecies were included. Although this may have biased genospecies proportion estimates (particularly for the closely related *B. garinii* and *B. bavariensis*), most publications reported *B. afzelii* as the dominant genospecies (regardless of the methodology used). Although an extensive and systematic search for relevant literature was undertaken, some peri/urban study locations may have been missed.

From a public health perspective, it is of paramount importance to bring together the current data on ticks in urban green space, and this review will enable comparisons with future studies. In addition, it also highlights key gaps in our knowledge. Although over 100 studies have started to contribute to our understanding of urban tick-borne disease risk and some make suggestions on how to reduce risk, many of them lacked baseline data on tick densities or *Borrelia* spp. prevalence. None of the studies incorporated the testing of control measures to reduce tick density or human exposure to ticks in urban areas. Only a few studies were able to collate data on Lyme borreliosis incidence at the time of tick surveys (Cisak et al., 2008; Pangrácová et al., 2013), and there is currently a gap in knowledge of how people interact with peri/urban green space in a way that leads to ticks bites or subsequent Lyme borreliosis diagnosis. Further assessment of the true public health impact of tick bites in peri/urban green space is required. Better evidence of public health impact would provide further justification for studies on interventions in high-risk urban green space and could rally change at the planning stages of existing or new urban developments.

Ecological studies on *I. ricinus* in urban green space should be incorporating a density assessment of DOT or DON, preferably DON as nymphs are the most common life stage found biting humans (Kilpatrick et al., 2017). If funding is available, prevalence assessments using sensitive PCR analysis should be used and should focus on nymphs (NIP). If both sets of data are available, they can be combined to generate DIN. Repeating studies over several years, and from multiple locations classified as truly urban, would also help to improve current density and prevalence estimates. To provide further context on tick-borne disease risk in urban areas, studies incorporating urban to rural gradients or different types of urban green spaces would be beneficial. Additionally, study methods should specify if site survey selection was random, or targeted to known tick areas, as this has implications when interpreting results.

Such efforts will lack wider impact if there is lack of clarity on survey effort and numbers of ticks tested, as well as lack of information on survey locations and habitat descriptions. This lack of transparency makes global comparisons across all urban green space challenging and limited to a subset of studies. More time should be spent to fully present data and capture habitat descriptions, to enable better impact of studies through contribution to reviews like this, or projects like the European Centre for Disease Prevention and Control's VectorNet project which is currently mapping and monitoring the distribution of key tick species across Europe, North Africa and the Middle East. Where possible, additional information should be incorporated into future research studies assessing *I. ricinus* in urban areas, including the monitoring of tick bites or acquisition of Lyme borreliosis during ecological surveys (Cisak et al., 2008; Krstić et al., 2016; Pangrácová et al., 2013). Such studies suggest that higher tick density and *Borrelia* prevalence may not always result in higher rates of tick bites or Lyme borreliosis incidence (either diagnosed cases or serological response to antigens) and that human behaviour also needs to be considered. Similar data would provide additional risk assessment and might help to explain why some locations report relatively high DIN and visitor access, with few anecdotal reports for Lyme borreliosis (Hansford et al., 2021). Finally, experimental studies within cities that target development areas such as meadow introduction or greater connectivity through wildlife corridors, and also changes in management practices such as reduced mowing, are needed to better assess the potential impacts of future climate mitigation and habitat management strategies on urban tick populations.

Although some themes appear to be emerging from questing urban tick data in Europe, including the importance of woodland habitat, the potential impacts of connectivity, seasonal fluctuations in prevalence and dominance of *B. afzelii*, risk appears to be present in a range of urban green spaces and across multiple seasons. Some areas may present a significant risk to public health, with high tick densities, *B. burgdorferi* s.l. prevalence and densities of infected ticks that may not be expected within such settings. Further data on DON, NIP and DIN are needed to provide further support of these emerging themes, and data on human behaviour in these settings, including the acquisition of tick bites and subsequent Lyme borreliosis incidence, would help to further identify and pinpoint control and interventions in high-risk areas.

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## CONFLICT OF INTEREST

The authors declare no conflicts of interest.

## DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available in Appendices S1 and S2.

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