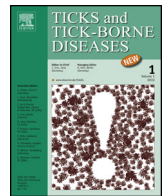




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Original article

Seasonal dynamics of tick species in an urban park of Rome

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ABSTRACT

Regular collections were obtained in the Natural Reserve of the Insugherata of Rome during 2011 in order to obtain the tick species composition and the respective seasonal dynamics of the area. A total of 325 ticks was collected in selected sites by means of drag sampling. Among the identified species, *Rhipicephalus turanicus* was the most abundant (72.3%), followed by *Ixodes ricinus* (19.7%), *Dermacentor marginatus* (6.5%), *Haemaphysalis punctata* (1.2%), and *Rhipicephalus bursa* (0.3%). *R. turanicus* occurred mainly in pastures, showing a mono-modal seasonal activity pattern from spring to early summer. Questing *I. ricinus* were prevalent in woodland from October to May, and the seasonal trend of specimens showed a weak peak in winter. Although adult *D. marginatus* exhibited seasonal dynamics similar to *I. ricinus*, with an activity period from October to April, this species occurred in a different environment (pasture) and with considerably lower densities. *Haemaphysalis punctata* and *R. bursa* were rare, with an apparent autumn and autumn–winter seasonal activity, respectively. While the species diversity recorded appears as an unequivocal consequence of the natural state of the park, the remarkable *R. turanicus* density could be a direct effect of the recent introduction of wild boar, as carriers, from the close Veio Park. The presence of the species, a proven vector of various diseases in humans and domestic animals, is discussed in the light of the possible risk of tick-bite exposure of park workers and visitors.

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Introduction

In recent decades, many urban areas have been created for the conservation of natural heritage. Although suburban territories once supporting wild environments have been surrounded or occupied by human settlements, several members of flora and fauna have managed to survive and to develop inside the changing synanthropic situations (Alekseev et al., 2008). In particular, town parks and suburban green zones, where sometimes a great variety of livestock and wild animals may live together, represent local hot spots of recreational activity in urban areas, but at the same time offer suitable environments for the introduction and/or spread of bloodsucking arthropods, for example ticks. In such areas, ticks can survive, feed on different vertebrate hosts, and develop often becoming infected with several pathogens. The role played by ticks as vectors of diseases in (sub)urban conditions is widely studied especially in northern Europe (Bašta et al., 1999; Gray et al.,

1999; Junntila et al., 1999; Wielinga et al., 2006; Földvári et al., 2011; Schorn et al., 2011). In Italy, although many investigations have been conducted in wild environments, few studies exist on tick occurrence in urban and periurban parks used for recreational activities (Rivosecchi et al., 1980; Cacciapuoti et al., 1985; Corrain et al., 2012).

Since a proper understanding of tick ecology is pivotal in predicting tick-borne pathogen transmission risk in a given area and in order to bridge this gap, a series of surveys carried out in different rural and urban ecosystems of central Italy have been planned. From this extensive research, we report here the findings of an acarological investigation undertaken in an urban park of Rome, during 2011.

Materials and methods

Study area

The Insugherata Natural Reserve (41°57'17" N, 12°25'35" E), so called for the presence of cork oaks (*Quercus suber*), was founded in 1997 and spans 740 ha (Fig. 1). The park is situated in the north-western sector of Rome, which is characterized by woods and bush strips in contrast to the south-eastern sector of the city, where

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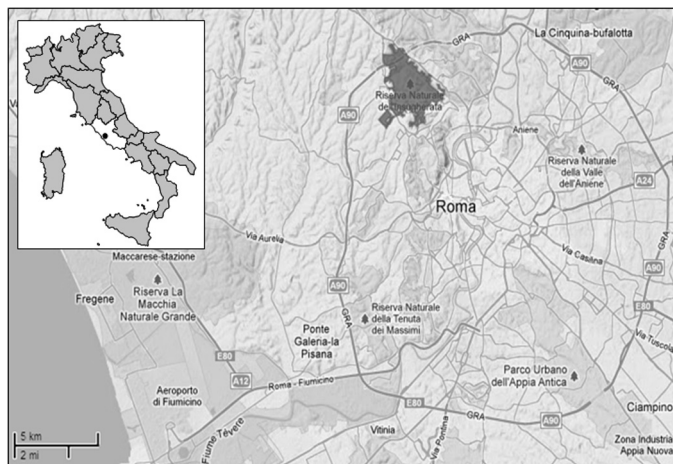


Fig. 1. The Insugherata Natural Reserve is situated in the north-western sector of Rome, Lazio Region, central Italy (map modified from <http://www.parks.it/riserva.insugherata/map.php>).

pastures and uncultivated areas are prevalent. Together with other urban natural areas (Acquatraversa, Monte Mario, Monti della Farnesina, Pineto, and Infernaccio Parks), the Insugherata belongs to a weakly connected woody belt, which is still linked with the green zones outside the urban band (Anzalone, 1953; Celesti-Grappow and Fanelli, 1991). The reserve, with its Mediterranean climate hosts a rich fauna and flora. The sunny slopes of the hills harbor several oaks (*Q. suber*, *Quercus pubescens*, *Quercus cerris*, *Quercus frainetto*, and *Quercus ilex*), while a deciduous wood extends on the shadier slopes, including species such as hornbeams (*Carpinus betulus*), ashes (*Fraxinus ornus*), English oaks (*Quercus robur*), field maples (*Acer campestre*), Spanish chestnuts (*Castanea sativa*), and hazels (*Corylus avellana*). Along the valleys and the streams, willows (*Salix alba*), poplars (*Populus alba*), and several ferns occur. Foxes, weasels, and porcupines are very common, while badgers occur only sporadically. Many small mammals (*Apodemus sylvaticus*, *Microtus savii*, *Suncus etruscus*, *Erinaceus europaeus*, *Talpa europaea*, and *Muscardinus avellanarius*) and a great variety of birds, reptiles, and amphibians complete the wild fauna of the reserve. Only in the past few years, wild boar have spread from northern boundaries of the park. Although characterized by a significant biodiversity with different natural biotopes, also with wheat cultivation and sheep pasture, the park represents an important area for human recreation for many inhabitants of Rome.

Tick collections

After two exploratory surveys carried out in June and July 2010, tick collections were conducted in three selected sites within the park twice a month from January to December 2011, along transects of 100 m each for a total of 12 fixed transects covered per visit. Site 1 was characterized by wheat fields and pasture for sheep flocks and rare horses. Site 2 was a deciduous mixed wood with mainly oaks reflecting the typical vegetation formation of the natural Roman area (Anzalone, 1953; Blasi, 1984). Ecotonal areas with bushy glades and sporadic trees surrounding small lawns, pastures, and uncultivated fields distinguished site 3. Questing ticks were collected in all sites by dragging a 1-m² woolen blanket through the vegetation. Collection sessions were performed by 2 investigators between 8:00 and 12:00 in the morning. Because collection efficiency may vary individually, collectors were systematically changed in each site. The number of ticks on the cloth was counted and picked up every 10 m, and tick abundance was calculated as

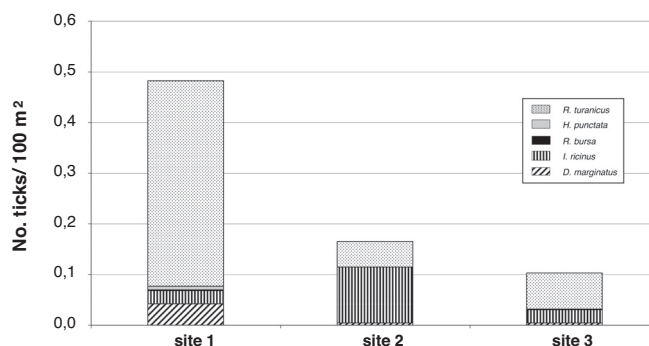


Fig. 2. Species composition by site in the Insugherata Natural Reserve.

the number of ticks collected per 100 m². During each sampling, daylight (h) was assessed and temperature (*T*) and relative humidity (RH) were recorded using a portable thermo-hygrometer. The samples were identified according to morphological characters (Manilla, 1998), and stored at −80 °C.

Statistical analysis

Chi-square test was used to assess the association between species composition and sampling site and between species activity and environmental parameters like temperature and humidity. A *p* value <0.05 was considered statistically significant. Statistical analyses were processed by Stata 11 software.

Results

During 2010–2011 surveys, a total of 325 ticks was collected in selected sites: *Rhipicephalus turanicus* Pomerantzev, 1940, was the most abundant species (72.3%) with 112 males, 106 females, and 17 nymphs, followed by *Ixodes ricinus* (Linnaeus, 1758) (19.7%) with 33 males and 31 females, *Dermacentor marginatus* (Sulzer, 1776) (6.5%) with 10 males and 11 females, *Haemaphysalis punctata* Canestrini and Fanzago, 1877 (1.2%), with one male and 3 females, and *Rhipicephalus bursa* Canestrini and Fanzago, 1877 (0.3%), with one female.

Chi-square test showed a significant association between species composition and collection site and between species dynamics and environmental parameters, *T* and RH. Species composition by site is shown in Fig. 2. Seasonal dynamics of the tick populations from all sites were calculated and are reported in Fig. 3.

During the beginning of spring, ixodid tick abundance showed a dramatic increase which led to 2 remarkable abundance peaks during this season, mainly due to the *R. turanicus* activity. At the end of spring, the decreasing trend was followed by an absence of ticks from our collections during the whole summer period, while the occurrence of specimens was always constant during the autumn and winter months, even if at low densities and fluctuating. About 67% of the whole tick sample was collected in site 1, 20% in site 2, and the remaining specimens in site 3.

Although widespread in all sites, *R. turanicus* significantly occurred in site 1 (*p* < 0.05). All specimens of this species were found from the end of March to the beginning of July, with average temperatures ranging between 14 and 25 °C. *R. turanicus* reached highest densities on 21st of April and 20th of May with 7.42 and 3.33 specimens/100 m², respectively, in correspondence with the lowest relative humidity values during that period (59% RH). Moreover, almost identical numbers of males and females were collected (sex ratio 1.05:1.0), while all nymphs were found in a single survey (20th of May) in site 1. During 2 sporadic surveys carried out

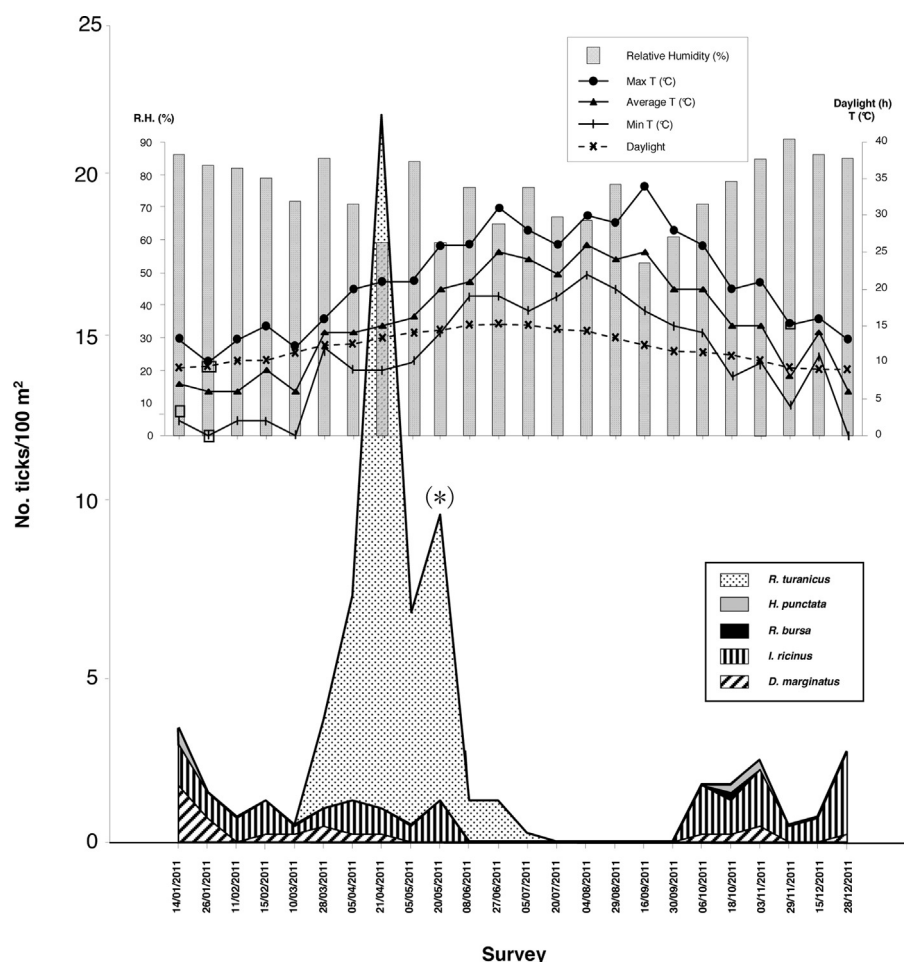


Fig. 3. Seasonal variation in adult activity of the 5 tick species collected in the Insugherata Natural Reserve from blanket dragging and climatic factors recorded for each survey (temperature, relative humidity, and daylight). (*): In this collection survey, 17 *R. turanicus* nymphs were found, but not included in the analysis.

in 2010, only this species was collected (24 specimens on 30th of June and one specimen on 15th of July).

I. ricinus was observed mainly in site 2 ($p < 0.05$). The species was collected from January to May and from October to the end of the year, reaching the maximum density in December (0.83 specimens/100 m²) and almost disappearing in the warmer months when average temperatures were $>20^{\circ}\text{C}$ ($T_{\min}$ was $0\text{--}14^{\circ}\text{C}$, and $T_{\max}$ was $10\text{--}26^{\circ}\text{C}$). It is worth mentioning that the same temperatures were measured (average T was 20°C , $T_{\min}$ 14°C , and $T_{\max}$ 26°C) at the last record of *I. ricinus* in spring and the first record in fall (on 20th of May and 6th of October, respectively). The sex ratio was 1.06:1.0.

Significantly present in site 1 ($p < 0.05$), adult *D. marginatus* were found in low density and not continuously (a total of 21 specimens) from January to April and from October to December, with a peak of abundance recorded on 14th of January with 0.58 specimens per 100 m². This species was active at average temperatures ranging between 6 and 20°C . The tick collections showed a sex ratio of 0.91:1.0.

Only 4 specimens of *H. punctata* were collected in the park, 2 found on 14th of January, with an average temperature of 7°C , the other 2 on 18th of October and 3rd of November, with milder temperatures (average T 15°C). Three of them were found in site 1, but only one specimen in site 3.

Only one specimen of *R. bursa* was reported. It was collected in site 1 on 18th of October.

Discussion

Because in Italy field studies on tick occurrence and population dynamics in urban and suburban areas are lacking or limited, we planned an acarological survey in an old park of Rome. Located in the north-western outskirts of the city, the Insugherata Park is characterized by both natural, unchanged biotopes and others influenced by human activities, like grazing, cultivation, and recreational areas, where many citizens spend their working and leisure time on a daily basis.

During the year, host-seeking ticks were always found on low vegetation in the study area, except for the warmest months.

In the park, the seasonal distribution shows mainly the pattern of *R. turanicus* activity, representing about 70% of the whole tick collection. This species is thermophilic, with a great affinity for Mediterranean habitats (Estrada-Peña et al., 2004). Moreover, Al-Khalifa et al. (2006) showed *R. turanicus* as a species tolerant to low RH levels, also at warm temperatures. Its resistance to desiccation might explain its ability to colonize many arid habitats. In Italy, this species, a proven vector of different animal and human pathogens like *Rickettsia conorii*, *Babesia* spp., *Anaplasma* spp., *Theileria ovis*, *Coxiella burnetii*, and Crimean-Congo hemorrhagic fever virus (Torina et al., 2010; Lalar et al., 2012; Yesilbag et al., 2013), is widespread in central and southern regions, where it was recorded for the first time in the Agro Romano, near Rome by Saratsiotis and Battelli (1975). Unlike *Rhipicephalus sanguineus* (Latreille, 1806)

with which it has often been confused in the past (Manilla, 1998), *R. turanicus* shows an exophilic behavior and a high ecological plasticity, exhibiting a great adaptability to survive in different biotopes and to feed on a broad spectrum of hosts.

At our latitudes, this species was often collected in periurban fields, characterized by clay soils, where grain or legumes are extensively farmed and in pastures where its usual hosts are found (Manilla, 1998). The adults of this species feed particularly on even-toed ungulates, like ovine, goat, and bovine, but also on swine (Gilot et al., 1992). In the Insugherata Park, the species occurred mainly in pastures and showed a typical monomodal seasonal pattern from spring to early summer, confirming the seasonal fluctuations recorded in other areas of the Mediterranean region (Gilot and Pautou, 1981). The samples collected in mid-summer of 2010 are explained by milder temperatures and higher RH values recorded in May and June when compared with the following year (Bianchi et al., 2011).

As expected, the castor bean tick, *I. ricinus*, the main vector of *Borrelia burgdorferi* sensu lato and of the TBE virus in Italy (Cinco et al., 1998; Hudson et al., 2001; Floris et al., 2006), was prevalent in woodland showing a fluctuating seasonal trend with a peak in fall/winter, a weak peak in spring, and completely disappeared from our collections during the summer period.

Adult *D. marginatus* were active from October to April, with a peak in January and occurred mainly in pastures. This species can bite humans and is a vector of some animal and human pathogens (Selmi et al., 2009; Torina et al., 2010; Masala et al., 2012). The other 2 tick species collected in the park, *H. punctata* and *R. bursa* were very scarce in our collections. *H. punctata* is a potential vector of several pathogens (Márquez, 2008; Barandika et al., 2008). The collection of 2 out of four specimens in the winter appears peculiar because the species typically shows a bimodal seasonal activity with peaks in spring and in autumn (Iori et al., 2005). Finally, the rarest species in our collections was *R. bursa* with a single specimen found in October, and no conjecture on its phenology was achieved. This species, which rarely bites humans, is considered the main vector of *Babesia* spp. of small ruminants in Mediterranean countries (Altay et al., 2008).

Although the ixodid fauna recorded is common in central Italy (Iori et al., 2005), the rich variety of tick species seems an explicit consequence of the ecological naturalness of the Insugherata Park, as previously highlighted for other protected natural areas (Stella and Khoury, 1980; Rivoecchi and Khoury, 1986; Frusteri et al., 1994; Iori and Di Paolo, 1999). Furthermore, the park is only partially surrounded by buildings, maintaining an important natural corridor connected to other green zones, outside the urban band. In this sector of the city, the former forest phytocoenosis still prevails, whereas pastures and uncultivated areas characterize the eastern part. These 2 divergent landscapes, with specific flora and fauna and divided by the Tevere River, are determined by microclimatic, morphological and edaphic as well as historic and urban factors (Celesti-Grappow and Nazzaro, 1992). The heavy anthropogenic impact due to an intensive agriculture and urbanization at the eastern and southern periphery of Rome has progressively caused a drastic decline of biodiversity, leading to a massive prevalence of a few ecologically adaptable species, such as *R. sanguineus* (Rivoecchi et al., 1980; Stella et al., 1988). The presence of wild mammals and livestock has a direct influence also on the ixodid fauna. In the park, wide areas are pastures for permanent ovine flocks, and *R. turanicus*, *D. marginatus* as well as *H. punctata* are considered the most frequently found species on small ruminants, especially in central-southern Italy (Genchi and Manfredi, 1999; Deiana and Arru, 1960). The recent invasion of wild boar (*Sus scrofa scrofa*) from the more northern Veio Park (the earliest tracks go back to 2005) could have directly affected the distribution and the abundance of ticks. For example, the high *R. turanicus* density may

be explained by the spread of this invasive wild ungulate, acting as an additional host for this species and enhancing its abundance (Manfredi et al., 1999; Keysary et al., 2011). In addition, it should be taken into account that wild boar may bring still other tick species and some tick-borne pathogens from sylvatic to periurban environments and may act as a reservoir for them (Ortuño et al., 2007). In recent years, several tick-borne diseases have emerged in temperate climates (Piesman and Eisen, 2008) due to pathogen introduction into naïve areas.

Although all tick species recorded in the park are recognized as potential vectors of different pathogens for animal and humans, very little is known about the possible circulation of tick-borne pathogens in Rome. A few studies carried out in wild areas of the province of Rome, investigated the occurrence of pathogens in ticks, identifying 4 *Borrelia burgdorferi* genospecies and *Anaplasma phagocytophilum* in *I. ricinus* (Cinco et al., 1997; Santino et al., 2002) and spotted fever group rickettsiae in *R. sanguineus* (Cacciapuoti et al., 1985).

In Italy, the incidence of tick-borne diseases in humans still seems highly underestimated because of poor surveillance of these zoonoses, misdiagnoses, asymptomatic cases, and an infectious disease-reporting system not widely applied, with an unrealistically low number of notifications.

Although in Rome there has been no recent evidence of the circulation of any pathogens transmitted by ticks, there is a high risk of being bitten by these arthropods for people in the Insugherata park especially in spring and autumn when more people visit the area.

To reduce tick exposure, prevention efforts should focus on 2 objectives, one general and one individual. The reserve management should try to reduce host/tick interactions by preventing the forays of wild boar from the northern natural areas, and by controlling grass and brush particularly along paths and in picnic areas, as well as checking the health of flocks grazing in the park. The individual approach should provide both visitors and professionals frequently exposed to ticks (i.e. forestry rangers, shepherds, farmers) with specific information about the risk of tick bites and recommend appropriate behavioral measures to be taken, including protective clothing, the use of insect repellent, tick checks, and early tick removal (Mead, 2011). In the light of global changes and of consequent concerns raised about the possible introduction and spread of several important tick species (Randolph and Rogers, 2003; Ergonul, 2006; Sumilo et al., 2008; Jaenson and Lindgren, 2011), resources should be made available and addressed to allow continuity of monitoring activities for collection of both questing ticks and ticks parasitizing mammals or birds, at the same time implementing a search for potentially circulating pathogens.

Finally, an evaluation of tick infestation is crucial for understanding the entity of tick bite risk and tick-borne disease transmission; in fact this knowledge is the base to plan aimed control strategies.

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References

- Alekseev, A.N., Dubinina, H.V., Zygutienė, M., Efremova, G.A., 2008. Ticks and other bloodsucking arthropods as urban pests inside and outside dwellings. In: Robinson, W.H., Bajomi, D. (Eds.), Proceedings of the 6th International Conference on Urban Pests. Budapest, Hungary, pp. 361–365.

- Al-Khalifa, M.S., Al-Lahoo, A.A., Hussein, H.S., 2006. The effect of temperature and relative humidity on moulting of engorged larvae and nymphs of *Rhipicephalus turanicus* Pomerantsev, 1936. *Saudi J. Biol. Sci.* 13, 35–43.
- Altay, K., Aktas, M., Dumanli, N., 2008. Detection of *Babesia ovis* by PCR in *Rhipicephalus bursa* collected from naturally infested sheep and goats. *Res. Vet. Sci.* 85, 116–119.
- Anzalone, B., 1953. Residuo di vegetazione spontanea in Roma (Monte Mario e i Monti della Farnesina). *Ann. Bot. (Roma)* 24, 1–29.
- Barandika, J.F., Hurtado, A., García-Sanmartín, J., Juste, R.A., Anda, P., García-Pérez, A.L., 2008. Prevalence of tick-borne zoonotic bacteria in questing adult ticks from northern Spain. *Vector Borne Zoonotic Dis.* 8, 829–835.
- Bašta, J., Plch, J., Hulínská, D., Daniel, M., 1999. Incidence of *Borrelia garinii* and *Borrelia afzelii* in *Ixodes ricinus* ticks in an urban environment, Prague, Czech Republic, between 1995 and 1998. *Eur. J. Clin. Microbiol. Infect. Dis.* 18, 515–517.
- Bianchi, R., Toma, L., Khoury, C., Quarchioni, E., Mancini, F., Ciervo, A., Di Luca, M., 2011. Tick bionomics in a natural park in Rome and detection of pathogens potentially transmitted. In: de la Fuente, J., Estrada-Peña, A. (Eds.), *Proceedings of the 7th International Ticks and Tick-borne Pathogens (ITTP7) Conference*. Zaragoza, Spain, August 28th–September 2nd, 2011, p. 375.
- Blasi, C., 1984. *Quercus cerris* and *Quercus frainetto* woods in Latium (Central Italy). *Ann. Bot. (Roma)* 42, 7–19.
- Cacciapuoti, B., Rivoecchi, L., Stella, E., Ciceroni, L., Khoury, C., 1985. Preliminary studies on the occurrence of rickettsiae of the spotted fever group in *Rhipicephalus sanguineus* captured in suburban areas. *Boll. Ist. Sieroter. Milan.* 64, 77–81.
- Celesti-Grapow, L., Fanelli, G., 1991. A map of vegetation complexes in the urban area of Rome (South-West Sector). *Phytocoenosis* 3, 331–336.
- Celesti-Grapow, L., Nazzaro, G., 1992. Aspetto urbanistico e successione nell'ambiente urbano. In: Marchetti, R., Cotta Ramusino, M. (Eds.), *Atti V Congr. Naz. Soc. Ital. Ecol. (S. It. E.)*, Milano, pp. 249–260.
- Cinco, M., Padovan, D., Murgia, R., Maroli, M., Frusteri, L., Heldtander, M., Johanson, K.E., Engvall, E.O., 1997. Coexistence of *Ehrlichia phagocytophila* and *Borrelia burgdorferi* sensu lato in *Ixodes ricinus* ticks from Italy as determined by 16S rRNA gene sequencing. *J. Clin. Microbiol.* 35, 3365–3366.
- Cinco, M., Padovan, D., Murgia, R., Poldini, L., Frusteri, L., van de Pol, I., Verbeek-De Kruif, N., Rijpkema, S., Maroli, M., 1998. Rate of infection of *Ixodes ricinus* ticks with *Borrelia burgdorferi* sensu stricto, *Borrelia garinii*, *Borrelia afzelii* and group VS116 in an endemic focus of Lyme disease in Italy. *Eur. J. Clin. Microbiol. Infect. Dis.* 17, 90–94.
- Corrain, R., Drigo, M., Fenati, M., Menandro, M.L., Mondin, A., Pasotto, D., Martini, M., 2012. Study on ticks and tick-borne zoonoses in public parks in Italy. *Zoonoses Public Health* 59, 468–476.
- Deiana, S., Arru, E., 1960. Sulla riduzione dell'indice di mortalità negli erbivori della Sardegna per malattie protozoarie ematiche. *Parassitologia* 2, 145–147.
- Ergonul, O., 2006. Crimean-Congo haemorrhagic fever. *Lancet. Infect. Dis.* 6, 203–214.
- Estrada-Peña, A., Quílez, J., Sánchez Acedo, C., 2004. Species composition, distribution, and ecological preferences of the ticks of grazing sheep in north-central Spain. *Med. Vet. Entomol.* 18, 123–133.
- Floris, R., Altobelli, A., Boemo, B., Mignozzi, K., Cinco, M., 2006. First detection of TBE virus sequences in *Ixodes ricinus* from Friuli Venezia Giulia (Italy). *New Microbiol.* 29, 147–150.
- Földvári, G., Rigó, K., Jablonszky, M., Biró, N., Majoros, G., Molnár, V., Tóth, M., 2011. Ticks and the city: ectoparasites of the Northern white-breasted hedgehog (*Eriacus roumanicus*) in an urban park. *Ticks Tick Borne Dis.* 2, 231–234.
- Frusteri, L., Khoury, C., Maroli, M., 1994. Temporal distribution of ticks (Acarina: Ixodidae) in "Macchia Grande" park in Manziana in the province of Rome. *Parassitologia* 36, 295–300.
- Genchi, C., Manfredi, M.T., 1999. Tick species infesting ruminants in Italy: ecological and bio-climatic factors affecting the different regional distribution. *Parassitologia* 41 (Suppl. 1), 41–45.
- Gilot, B., Laforge, M.L., Cabassu, J.P., Romani, M., 1992. Éléments pour la cartographie écologique des populations de *Rhipicephalus* du groupe *sanguineus* dans l'agglomération Marseillaise, en relation avec les diverses formes d'urbanisation. *Acarologia* 33, 17–33.
- Gilot, B., Pautou, G., 1981. Répartition et intérêt épidémiologique de *Rhipicephalus turanicus*. Écologie de cette espèce dans le Midi méditerranéen. *Ann. Parasitol.* 56, 547–558.
- Gray, J.S., Kirstein, F., Robertson, J.N., Stein, J., Kahl, O., 1999. *Borrelia burgdorferi* sensu lato in *Ixodes ricinus* ticks and rodents in a recreational park in south-western Ireland. *Exp. Appl. Acarol.* 23, 717–729.
- Hudson, P.J., Rizzoli, A., Rosa, R., Chemini, C., Jones, L.D., Gould, E.A., 2001. Tick-borne encephalitis virus in Northern Italy: molecular analysis, relationships with density and seasonal dynamics of *Ixodes ricinus*. *Med. Vet. Entomol.* 15, 304–313.
- Iori, A., Di Giulio, A., De Felici, S., 2005. Zecche d'Italia, Parte III. In: Cringoli, G. (Ed.), *Mappe parassitologiche*, Vol. 6 (Zecche), Napoli, Italy, pp. 3–199.
- Iori, A., Di Paolo, M., 1999. Acarological studies in two protected areas of Central Italy. *Parassitologia* 41, 53–55.
- Jaenson, T.G.T., Lindgren, E., 2011. The range of *Ixodes ricinus* and the risk of contracting Lyme borreliosis will increase northwards when the vegetation period becomes longer. *Ticks Tick-Borne Dis.* 2, 44–49.
- Junttila, J., Peltomaa, M., Soini, H., Marjamäki, M., Viljanen, M.K., 1999. Prevalence of *Borrelia burgdorferi* in *Ixodes ricinus* ticks in urban recreational areas of Helsinki. *J. Clin. Microbiol.* 37, 1361–1365.
- Keysary, A., Eremeeva, M.E., Leitner, M., Beth Din, A., Wikswo, M.E., Mumcuoglu, K.Y., Inbar, M., Wallach, A.D., Shanas, U., King, R., Waner, T., 2011. Spotted fever group rickettsiae in ticks collected from wild animals in Israel. *Am. J. Trop. Med. Hyg.* 85, 919–923.
- Lalzar, I., Harrus, S., Mumcuoglu, K.Y., Gottlieb, Y., 2012. Composition and seasonal variation of *Rhipicephalus turanicus* and *Rhipicephalus sanguineus* bacterial communities. *Appl. Environ. Microbiol.* 78, 4110–4116.
- Manfredi, M.T., Dini, V., Piacenza, S., Genchi, C., 1999. Tick species parasitizing people in an area endemic for tick-borne diseases in north-western Italy. *Parassitologia* 41, 555–560.
- Manilla, G., 1998. *Acari Ixodida*. Fauna d'Italia 36. Ed. Calderini, Bologna, Italy.
- Márquez, F.J., 2008. Spotted fever group Rickettsia in ticks from southeastern Spain natural parks. *Exp. Appl. Acarol.* 45, 185–194.
- Masala, G., Chisu, V., Satta, G., Socolovschi, C., Raoult, D., Parola, P., 2012. *Rickettsia slovaca* from *Dermacentor marginatus* ticks in Sardinia, Italy. *Ticks Tick Borne Dis.* 3, 393–395.
- Mead, P., 2011. Education, behaviour change, and other non-pharmaceutical measures against Lyme and other tick-borne diseases. In: Critical needs and gaps in understanding prevention, amelioration, and resolution of Lyme and other tick-borne diseases: the short-term and long-term outcomes. Workshop Report of Committee on Lyme Disease and Other Tick-Borne Disease: The State of the Science; Oct 11–12, 2010; Washington, DC. The National Academies Press, Washington, DC.
- Ortuño, A., Quesada, M., López-Claessens, S., Castellà, J., Sanfeliu, I., Antón, E., Segura-Porta, F., 2007. The role of wild boar (*Sus scrofa*) in the eco-epidemiology of *R. slovaca* in Northeastern Spain. *Vector Borne Zoonotic Dis.* 7, 59–64.
- Piesman, J., Eisen, L., 2008. Prevention of tick-borne diseases. *Annu. Rev. Entomol.* 53, 323–343.
- Randolph, S.E., Rogers, D.J., 2003. Ecology of tick-borne disease and the role of climate. In: Ergonul, O., Whitehouse, C. (Eds.), *Crimean-Congo Hemorrhagic Fever. A Global Perspective*. Springer, The Netherlands.
- Rivoecchi, L., Khoury, C., 1986. Osservazioni su alcuni Artropodi di interesse medico-veterinario in un Parco (Migliarino-S. Rossore-Massaciuccoli) della Regione Toscana, con note su due aree protette (Castel Porziano e Palo Laziale) dei dintorni di Roma. *Frustula Entomol.* 7–8, 283–306.
- Rivoecchi, L., Khoury, C., Lezzneri, C., Dell'Uomo, G., 1980. Osservazioni su *Rhipicephalus sanguineus* (Ixodidae) nella periferia di Roma. *Riv. Parassitol.* 41, 273–276.
- Santino, I., del Piano, M., Sessa, R., Favia, G., Iori, A., 2002. Detection of four *Borrelia burgdorferi* genospecies and first report of human granulocytic ehrlichiosis agent in *Ixodes ricinus* ticks collected in central Italy. *Epidemiol. Infect.* 129, 93–97.
- Saratsiotis, A., Battelli, C., 1975. *Rhipicephalus turanicus* en Italie. Comparaison morphologique avec *Rhipicephalus sanguineus* s. str. *Riv. Parassitol.* 36, 207–214.
- Schorn, S., Pfister, K., Reulen, H., Mahling, M., Silaghi, C., 2011. Occurrence of *Babesia* spp., *Rickettsia* spp. and *Bartonella* spp. in *Ixodes ricinus* in Bavarian public parks, Germany. *Parasit Vectors* 4, 135.
- Selmi, M., Martello, E., Bertolotti, L., Bisanzio, D., Tomassone, L., 2009. *Rickettsia slovaca* and *Rickettsia raoultii* in *Dermacentor marginatus* ticks collected on wild boars in Tuscany, Italy. *J. Med. Entomol.* 46, 1490–1493.
- Stella, E., Khoury, C., 1980. Nuove osservazioni su alcuni Ixodidi dei dintorni di Roma. *Riv. Parassitol.* 41, 145–154.
- Stella, E., Khoury, C., D'Amato, F.R., Rivoecchi, L., 1988. Nuovi dati sulla biologia di *Rhipicephalus sanguineus* (Ixodidae) nella periferia di Roma. *Riv. Parassitol.* 49, 1–11.
- Sumilo, D., Bormane, A., Asokliene, L., Vasilenko, V., Golovljova, I., Avsic-Zupanc, T., Hubalek, Z., Randolph, S.E., 2008. Socio-economic factors in the differential upsurge of tick-borne encephalitis in central and Eastern Europe. *Rev. Med. Virol.* 18, 81–95.
- Torina, A., Alongi, A., Scimeca, S., Vicente, J., Caracappa, S., de la Fuente, J., 2010. Prevalence of tick-borne pathogens in ticks in Sicily. *Transbound. Emerg. Dis.* 57, 46–48.
- Wielinga, P.R., Gaasenbeek, C., Fonville, M., de Boer, A., de Vries, A., Dimmers, W., Jagers, G.A.O., Schouls, L.M., Borgsteede, F., van der Giessen, J.W.B., 2006. Longitudinal analysis of tick densities and *Borrelia*, *Anaplasma*, and *Ehrlichia* infections of *Ixodes ricinus* ticks in different habitat areas in The Netherlands. *Appl. Environ. Microbiol.* 72, 7594–7601.
- Yesilbag, K., Aydin, L., Dincer, E., Alpaly, G., Girgisin, A.O., Tuncer, P., Ozkul, A., 2013. Tick survey and detection of Crimean-Congo hemorrhagic fever virus in tick species from a non-endemic area, South Marmara region, Turkey. *Exp. Appl. Acarol.* 60, 253–261.