Lyme disease is the most common arthropod-borne disease of humans in Europe and North America. In Europe, disease has been reported in a variety of animal species closely associated with human (Kasbohrer and Schonberg, 1990; Parker and White, 1992; Blowey et al., 1994; Ciceroni et al., 1997; Magnarelli et al., 1997; Stefancikova et al., 2000). Approximately 40 species of mammals and birds have been recognized as a reservoir for *Borrelia burgdorferi* (Bb) (Gern et al., 1998). Among the pet animals dog has been identified as the competent reservoir for *B. burgdorferi* sensu stricto (Mather et al., 1994). Moreover, various researchers have proposed the dog as ‘sentinel animal’ for the detection of emerging risk areas of Lyme disease (Lindenmayer et al., 1991; Falco et al., 1993; Merino et al., 2000; Bhide et al., 2002). In Europe considerable study on canine Lyme borreliosis has been done with respect to symptoms and seroprevalence. The most common symptom in dog is migratory arthritis without divergent radiographic findings (Magnarelli et al., 1987). Intermittent lameness can also be seen with several episodes. Other clinical signs consist of anorexia and general malaise. There are some reports of heart block (Levy and Dury, 1988), neurological sign like seizures (Azuma et al., 1993), and fatal kidney failure (Dambach et al., 1997). Although the various symptoms have been reported so far, the diagnosis of Lyme borreliosis in dogs is much more difficult. Recent serological techniques have made the diagnosis easier and more confirmative.

Antibody profile in Lyme disease is an interesting criterion that can be used for risk assessment of Lyme disease in humans. Purpose of this review is to focus on the possible use of dog as a marker for identification of new developing Lyme disease foci and to elaborate their eco-epidemiological importance.

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Canine Lyme disease serodiagnosis and antibody profile

Till today sensitive serological techniques like ELISA, western blot, immuno fluorescent assay (IFA) etc. are developed to detect and confirm the B. burgdorferi infection in dogs. With the help of standard ELISA or IFA, antibody titers can be detected between 4–6 weeks after exposure to infected ticks. Serological tests are also developed to distinguish between early and late stages of Borrelia infection. Though the sensitive ELISAs are used widely in humans and pet animals, their specificity is still doubtful. False positive results can be obtained because of vaccination especially in the dogs. In efforts to improve the specificity of serological tests as well as to distinguish between antibodies after Borrelia infection and vaccination several strategies have been attempted, for example use of flagellin-enriched (Coleman and Benach, 1987) and purified (Hansen et al., 1988) antigen preparations or specifically VlsE antigen (Liang et al., 2000), and the use of recombinant antigens of B. burgdorferi expressed in Escherichia coli (Zumstein et al., 1992). Recently, the more specific and confirmative borreliacidal antibody test has been used in dogs (Callister et al., 2000). B. burgdorferi infection in humans and other animals results in production of killing (borreliacidal) antibodies. These antibodies are directed against several B. burgdorferi proteins including outer surface protein A (OspA), OspB, OspC, decorin binding protein A (DbpA) and outer membrane protein p66 (Scriba et al., 1993; Probert and Lefebvre, 1994; Rousselle et al., 1998; Exner et al., 2000). Borreliacidal antibodies can be detected in dogs one week after attachment of infected tick (Callister et al., 2000). These borreliacidal antibodies not only increase the specificity but also effectively distinguish between early and late Lyme disease.

In untreated infected animal, antibody level increases (IgG), reaching maximum at approximately 90–120 days after tick exposure, and then remain at its level up to one and a half year in the absence of re-exposure (Straubinger, 2000). On the other hand, shorter span of anti-Borrelia antibodies has been reported in dogs. Moreover, Hovius et al. (1999) and Goossens et al. (2001) have reported obligatory yearly reinfection to maintain seropositivity in dogs. On the contrary the period of seropositivity in humans after an infection with B. burgdorferi is much longer.

Advantages of canine seroprevalence over serosurvey in other animals

Prediction of potential area for Lyme disease is a difficult task. An epidemiologist may experience more complexities in declaring any new geographical area as emerging risk zone for Lyme borreliosis. The complexity heightens particularly when human case prevalence is low. To identify the endemic area it is also very necessary to study vector-host relationship, vector population and anti-Borrelia antibody prevalence in the reservoir hosts. Some researchers have suggested a close association between population/distribution of Ixodid ticks and Lyme disease prevalence in humans and dogs (Lissman et al., 1984; Magnarelli et al., 1987). Canine anti-Borrelia serosurvey offers a promising tool for targeting areas presenting potential human risk (Rand et al., 1991, 1996). Advantages of canine serosurvey over other animals are: simplicity in sample collection, effective follow up and/or feed back, known history of treatment and vaccination, and greater correlation for Lyme disease risk assessment to human being. On the other hand, tick vector distribution surveys, flagging, small mammal trapping or examination of deer and other wild free-living animals are laborious and time consuming (Eng et al., 1988). Moreover, the seroprevalence in wild animals can not be applied directly to assess the Lyme disease risk to common people who have rare or no contact to forested areas. Measuring the tick density and prevalence of infected ticks in and around cities especially parks, playgrounds and recreational places near human habitat is one of the imperative approach to assess a Lyme risk for common people. As dogs have free and frequent access to such areas, combination of tick density and tick infectivity study with canine seroprevalence can be effective tool to judge the actual Lyme disease risk in the area under study.

Facts of canine seroprevalence

Till to date considerable work has been done in the field of canine seroprevalence. Anti-Borrelia antibodies in dogs have been reported in most of the major European countries (Table 1). Particularly in Slovakia, seropositivity in hunting dogs was 40% whereas in service and pet dogs positivity observed was 11.80% and 29.40%, respectively (Stefancikova et al., 1996). Difference in seropositivity according to
use and nature of dogs is also reported by Cohen et al. (1990), Stefancikova et al. (1998) and Merino et al. (2000) whereas, antibody prevalence was not associated with sex and season (Delgado and Carmenes, 1995). Outdoor activity is the prime factor, which governs percent seropositivity against Lyme borreliosis in any given species of animal. In short, the difference in seroprevalence may due to differences in tasks performed by dogs and therefore the different tick exposition (Daniels et al., 1993). Age de-

<table>
<thead>
<tr>
<th>Country</th>
<th>County</th>
<th>Method of detection</th>
<th>Prevalence (%)</th>
<th>No. of sample (n)</th>
<th>Reference</th>
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pendent variation in seroprevalence of Lyme disease in dogs is reported in Slovakia (Stefancikova et al., 1996), Spain (Merino et al., 2000) and North America (Cohen et al., 1990; Lindenmayer et al., 1991). Some researchers have tried to correlate the seropositivity and geno-phenotypic characteristics of dogs. In dogs with hard type of hairs greater seropositivity against B. burgdorferi was reported in comparison to others (Merino et al., 2000). No correlation between size of dog and positivity was reported. Similarly gender is a factor, which does not affect the seropositivity (Magnarelli et al., 1987; Delgado and Carmenes, 1995; Merino et al., 2000). Apart from above explained factors, environment can also play an important role. Dogs living at higher altitude expressed minor seroprevalence in comparison with dogs living in lower region (Lindenmayer et al., 1991). Study in Soria province in Spain by Merino et al. (2000) confirmed this hypothesis by comparing the seroprevalences in dogs from other altitudes. All environmental factors ultimately control the tick population in specific area. Tick population governs vector-host relationship as well as tick attachment rate and thus affect the seropositivity in dogs. In the canine surveillance system for Lyme borreliosis in Wisconsin and Illinois (Guerra et al., 2001), seroprevalence pattern by county (0–40%) was significantly correlated with human incidence of Lyme disease and with abundance of tick vector, Ixodes scapularis. In the same study a geographic information system (GIS) was used to integrate environmental data with the location of the residences of the dogs to determine environmental risk factors. In Europe environmental risk factors for Lyme disease have been determined using satellite, climatological, and ecological data (Estrada-Pena, 1997; Daniel et al., 1998; Randolph, 2000). Thus, seropositivity in dogs is positively associated with increased tick exposure, time spent outdoor, living in deciduous forested areas etc. Because of close similarity between Lyme disease risk factors of dogs and humans, canine surveillance system is useful method for assessing the risk as well as geographic distribution of Lyme disease.

Complement resistance of Borrelia burgdorferi and reservoir competence of dog

Complement-mediated killing of B. burgdorferi in hosts have ecological implications as it can determine the reservoir competence (Kurtenbach et al., 1998b; Hovius et al., 2000). The pattern of serum complement sensitivity of different Borrelia genospecies matches the known reservoir status of many vertebrate species (Kurtenbach et al., 1998b). Studies indicate that B. garinii and B. valaisiana are mainly transmitted to ticks by avian hosts whereas, B. afzelii is transmitted to tick by rodents (Humair et al., 1995; Kurtenbach, 1998a). In vitro canine complement sensitivity test (Hovius et al., 2000) against three different Borrelia strains (B31, B. burgdorferi sensu stricto; pKo, B. afzelii; and A87S, B. garinii) showed B31 and pKo as resistant species to dog complement than A87S. It was observed that Borrelia isolates differ in their ability to activate complement and resist killing by serum bactericidal activity (Brade et al., 1992). Though there is no extensive study available to compare species specific complement sensitivity of Borrelia and reservoir competence of dog, one can extrapolate the available complement sensitivity results to propose reservoir status of dog for particular Borrelia species (Hovius et al., 2000). Such a correlation was made previously in rodents and squirrels by Kurtenbach et al. (1998b). Rodent complement resistance of B. afzelii parallels the prime transmission competence of rodent species (Humair et al., 1995) and squirrels (Craine et al., 1997). Furthermore, complement mediated lyses of B. garinii explains why the European rodents are insufficient reservoir for European B. garinii strains, while its resistance to pheasant complement makes clear the concept of reservoir competence ability of pheasant for the same Borrelia genospecies. In case of an incompetent reservoir sika deer active killing of Borrelia by complement takes place (Nelson et al., 2000). Similarly lysis of Borrelia regardless of genospecies correlates the incompetent reservoir nature of deer explained by Jaenson and Talleklint (1992).

Lyme disease risk assessment for pet owners and hunters

Overall Lyme disease risk assessment data compiled in various reviews and reports (Fliisiak and Zabicka, 1995; Arteaga and Garcia-Monco, 1999; Werner et al., 2001), indicate the morbidity exceeds 100 cases per 100 000 inhabitants per year in central Europe. Comparatively higher prevalence observed in outdoor workers than indoor workers in southwest Sweden (Werner et al., 2001) and Spain (Arteaga and Garcia-Monco, 1999) indicates posi-
tive correlation between human contact with tick vector and Lyme disease risk. Only measurement of human contact with a tick is not sufficient criteria to assess the risk of Lyme disease. Apart from this criterion, the population of reservoir competent domestic, wild as well as pet animals in particular area is necessary to study.

There are many controversies about zoonotic importance of pet animals as far as Lyme disease is concern. Even if some authors have had put forth hypothesis about greater risk of Lyme disease to pet owners (Mather et al., 1994), there is no concrete evidence of direct infection from pet animals or dogs to human. Survey in the Netherlands by Goossens et al. (2001), showed no correlation between sole ownership of dogs and seropositivity against Lyme disease. However, recently a case of one and a half year old girl suffering form gonarthitis has been reported by Zajadacz and Juszkiewicz (2002) with high antibody titre. The girl was never in the forest and had no contact with animals except a pet dog. Authors suggested the most possible transmission of Lyme disease from pets to the girl.

Hunting dogs usually carry infected ticks from the forest. Loosely attached ticks as well as infected females from dog drop near human habitat. Female ticks lay eggs in the spring, which hatches to larvae. These Ixodid larvae preferentially feed on small mammals and rodents. Presence of rodent population in and around human habitat facilitates feeding of larvae and nymphs and consequently helps in establishment of the tick population. Rodents are known competent reservoir from which Ixodid larvae acquire Borrelia infection. In the following spring larvae moult into nymphs, with an acquired infection from rodents. Ixodid nymphs have wide host range including dogs and humans. Nymphs moult to adult in fall and act as the most important source of infection for dogs. Transmission of Borrelia from Ixodid ticks to dogs, cats and human has been reported (Smith et al., 1993). Higher seroprevalence (33%) in the domestic cats does not exclude the importance of this pet animal in Lyme disease epidemiology (Magnarelli et al., 1990).

To conclude, screenings of dog for seropositivity is good indicator of actual and present risk of Lyme disease in particular area due to shorter span of anti-Borrelia IgG antibodies. Dogs stay seropositive for a much shorter period after an infection with Borrelia. On the other hand the seropositivity in other animals as well as in humans persists for several years. Similar seroprevalence in hunting dogs and humans particularly in hunters (Goossens et al., 2001) illuminates close relation and linked epidemiological aspects of Lyme disease. Evolution and establishment of Lyme disease focus may occur quickly due to favourable climatic conditions and geocoecological suitability of central Europe for tick vectors. Hunting dogs can serve as seroindicators and/or sentinel for identifying new focuses as well as assessing the changes in endemicity of well known focuses of Lyme disease.

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