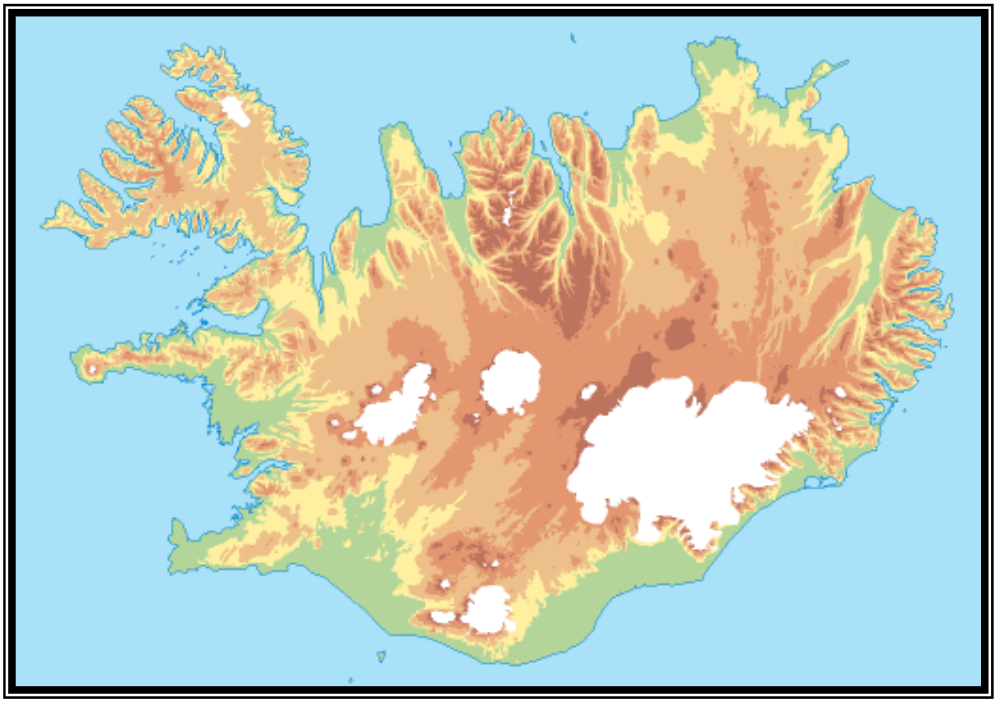

A seroepidemiological survey for antibodies to *Borrelia burgdorferi* sensu lato in dogs in Iceland

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Abstract

A random epidemiological study was undertaken to estimate the prevalence of antibodies to *Borrelia burgdorferi* sensu lato, the causative agent of Lyme Borreliosis, in apparent healthy dogs in Iceland. The presence of antibodies was determined by whole-cell Enzyme-linked immunosorbent assay (ELISA). Due to possible cross reactions, all positive results were to be confirmed by Western blot. Sera from 86 dogs of various ages, breed and both sexes from different regions were tested. Of all the dogs tested, 94,2% (81/86) were seronegative, 5,8% (5/86) were considered borderline. No dog was found seropositive (0/86). This study concludes that Lyme Borreliosis is not an endemic disease in Iceland with an estimated prevalence in the Icelandic dog population below 2%.

Key words: Epidemiology, antibodies, *Borrelia burgdorferi* s.l., ELISA, dogs, Iceland, Lyme Borreliosis

1. Introduction

Lyme Borreliosis (also termed Lyme disease) is a complex multiorgan disorder and represents the most important tick-borne zoonosis in Europe and in the United States (1, 2, 3, 4). It is caused by a spirochete of the genus *Borrelia*, collectively termed *Borrelia burgdorferi* sensu lato (s.l.) but actually represented by a very large and somewhat diverse group of isolates (5). The organism is transmitted by ticks of the genus *Ixodes* (6, 7). The species *I. ricinus* is considered the most important vector in relation to the epidemiology in Europe (7, 8). Other *Ixodes* ticks, *I. hexagonus* and *I. uriae* being nidicolous species, also contribute to the circulation of *B. burgdorferi* s.l. in Europe (7, 8, 9). Transmission by other vectors such as flees and mosquitoes has also been reported (5, 10).

Lyme Borreliosis (LB) is categorized as a zoonotic disease, because the infection is maintained in nature by humans and domestic animals as incidental hosts, but the reservoirs hosts are mainly wild small mammals, deer and birds (9, 11).

Most data on Lyme Borreliosis in domestic animals concern dogs and horses, with isolated reports of infection in cattle (12), sheep (13) and cats (14). Because of its association with a tick vector and the ideal climate for ticks being high humidity and dense vegetation, the prevalence of LB varies geographically (6, 7). In Europe relatively few reports exists, compared to USA, where hundreds of studies on the subject have been published. Many studies reveal that antibody profiles in dogs are an interesting criterion that can be used for risk assessment for LB in humans, e.g. using dogs as sentinels for human Lyme Borreliosis (7, 15, 18).

Several serologic methods have been established for detecting IgM and IgG antibodies in dogs. These include immunofluorescent antibody test (IFA), Enzyme-linked immunosorbent assay (ELISA) and Western Blotting (WB). Cultivation and Polymerase chain reaction (PCR) are frequently used for the identification and classification of the bacteria in ticks.

Seroepidemiological surveys for antibodies to *B. burgdorferi* s.l. in healthy, asymptomatic dogs have been performed in Sweden (16), Denmark (17), The Netherlands (18), France (19), Slovakia (20), Norway (21, 22), Spain (23), Czech Republic (24) and in the United Kingdom (25), categorizing these countries as being endemic, at least in some regions. Other countries, for example Belgium (26) and Germany (27) have reported seroprevalence in symptomatic dogs.

Although *I. ricinus* the most important vector of LB has hitherto not been believed to be endemic in Iceland (28), cases have increased through the years where it has infested domestic animals and humans. *I. uriae*, the seabird tick, on the other hand is endemic in some regions in Iceland, e.g. Vestmannaeyjar and Breiðafjarðareyjar. A study by Bunikis et al. 1996 on seabird colonies in Flatey Breiðafjörður, reported the presence of *B. garinii* in *I. uriae* ticks. Since no territorial mammals are present on Flatey the authors suggest that birds play an important part in the maintenance of *B. burgdorferi* and that *I. uriae* is a potential carrier of human pathogenic borrelia strain (29). A study from Frandsen et al. 1999 on the prevalence of *B.burgdorferi* s.l. in *I. uriae* ticks, collected from puffins (*Fratercula arctica*) in Vestmannaeyjar and Breiðafjarðareyjar indicated the prevalence as approximately 50% (48/93) by the IFA and 40% by the PCR method. The serotype was not confirmed (30).

LB has not been described in dogs in Iceland. One case of human LB has been reported. It was a 14 year old boy with arthritis, in the third (chronic) stage of the disease. He probably did not get infected in Iceland (31).

Two seroepdemiological surveys of antibodies to *Borrelia burgdorferi* s.l. in puffin hunters in Vestmannaeyjar have been performed in 1988 and 1995. None of the samples where seropositive, indicating that there is no seroconversion among the hunters, although frequently exposed to the *I. uriae* ticks (unpublished data 32).

The name Lyme Borreliosis (LB) refers to an outbreak of oligoarthritis in children in Old Lyme, Connecticut in 1975, who all had a history of tick-bites. In 1982 Burgdorfer et al. and a year later Barbour et al. isolated the spirochete *Borrelia burgdoferi* sensu lato (s.l.) from the hard bodied ticks *I. dammini* in the USA and *I. ricinus* in Europe (33, 34). After analyses of the agent, it became clear that the manifold of the symptoms in humans, all were a part of the same disease-complex, but caused by different species of *B. burgdorferi* s.l. The symptoms of human LB can be divided into 3 stages. In its early stage it is characterized by influenza-like symptoms, followed in 60-80% of the cases by erythema migrans, a skin lesion that spreads outward from around the site of a tick bite. If untreated, the disease may proceed to a second or a third stage in which neurological disorders and arthritis are common symptoms (18).

Lyme Borreliosis was first suggested and described in dogs 1984 by Lissmann et al. in a Doberman Pinscher suffering from fever, lethargy and swollen joints (35). In 1992 Wasmoen et al. fulfilled the Koch's postulates for *B. burgdorferi* as the causative agent of LB in dogs (36). Numerous reports of canine LB subsequently followed, describing a variety of clinical manifestations.

These include fever, inappetence, lethargy, lymphadenomegaly and acute onset of stiffness or lameness (often intermittent and shifting from one leg to another), swelling or pain in the affected joints are variably observed in acute infections. In chronic LB recurrent, intermittent, non-erosive arthritis is considered the primary, clinical manifestation and does not appear until 2 to 5 months after exposure to infected ticks (1, 7, 37, 38, 39). Heart block and renal disease as well as neurological dysfunctions have also been described (7, 11, 15).

In some previous studies, the diagnoses were based on clinical signs similar to those observed in humans and positive serologic test results. Green 1990 believes that these studies may not have been adequate since serologic surveys of dogs living in endemic areas have shown that up to 50% of dogs can be seropositive yet asymptomatic (40). Skotarczak 2002 demonstrates this problem partly as a result of cross reactions that occur between the antigens of *B. burgdorferi* and related bacteria such as *Treponema* spp, *Bradyspira* sp, and *Leptospira* spp (15). A study by Hovius et al. 1999 showed that prevalence of *B. burgdorferi* s.l. antibodies is usually higher in symptomatic dogs compared with healthy ones (41). Levy et al. 1992 and Goossens et al. 2003 revealed that only clinical signs, exclusions of other diseases in the differential diagnosis of the symptoms, possible exposure to infected ticks and response to treatment are reliable indicators for diagnosis of canine borreliosis (42, 43). Most studies consider a titer of 1:128 or greater as positive, a titer below 1:64 as negative. A titer between 1:64 and 1:128 as borderline (37, 39).

The aim of this seroepidemiological survey is to find out, if Icelandic dogs are exposed to *Borrelia burgdorferi* s.l. the causative agent of LB, using the enzyme-linked immunoassay (ELISA) method. If sera is tested positive, it will be confirmed with Western Immunoblot method.

3. Laboratory diagnostic methods

IFA, with *B. burgdorferi* cell preparations on a glass slides, is a first-generation test. Since IFA test does not allow any differentiation between infected and vaccinated dogs, and cross-reaction to other spirochetes is possible, many false positive results may occur (44). Thus Chambers et al. 1996 developed a novel IFA test where the antigens are adhered to a monolayer of cultured endothelial cells. This procedure made the test easier to evaluate and reduces the variability of test results (45).

ELISA, the second-generation test, with whole-cell preparations or single recombinant antigens is useful for the detection and precise measurement of antibody responses. Cross-reactive antibodies can influence the specificity of the test (44). Schillhorn van Veen et al. 1993 came to the conclusion that periodontal diseases, frequently caused by *Treponema spp.*, may cause false positive results in the antibody tests for LB (46). Most commercial available ELISA tests do not differentiate between infected and vaccinated animals (44).

WB with whole–cell preparations or recombinant antigen is useful for the detection and precise identification of antibody responses. It can differentiate between specific and non-specific cross-reactive antibody reactions and helps to make the distinction between infected and vaccinated animal. It is often used as confirmatory assay for IFA or ELISA (44). A study by Lindenmayer et al. 1990 showed that WB can give false positive results in cases of dogs with immune-mediated diseases and leptospirosis (47). Many authors demonstrate that serological tests may be deceptive in diagnosis, as a high proportion of dogs are seropositive without showing clinical symptoms. Some authors even reveal that serological screening of healthy dogs is controversial because it can lead to overdiagnosis or overtreatment of normal dogs, most of which never develop LB (6).

The direct *B. burgdorferi* detection includes cultivation of the agents in modified Barbour–Stoenner-Kelley (BSK) medium and Polymerase chain reaction (PCR). Cultivation is cumbersome and time-consuming since the organisms are grown in BSK medium over several weeks and are then detected by dark-field microscopy (44). PCR detects the specific microbial DNA and is a more sensitive and specific than bacteriological culture (44).

However PCR lacks the necessary sensitivity for diagnostic purposes because of sample bias resulting from the uneven distribution of *Borrelia* throughout biopsy specimens. Furthermore PCR not only detects living infecting organisms, but also DNA remnants of the causative spirochetes (43).

At least five serotypes are categorized to the group of *B. burgdorferi* s.l. and are believed to play an important role in borrelia infections in Europe (4, 48), see table 1.

Table 1: *Borrelia burgdorferi* s.l. serotypes in Europe. (4, 48).

- *Borrelia burgdorferi sensu stricto*
- *Borrelia afzelii*
- *Borrelia garinii*
- *Borrelia lusitaniae*
- *Borrelia valaisiana*

At least three species of the *B. burgdorferi* s.l. complex, *B. afzelii*, *B. garinii* and *B. burgdorferi* sensu stricto are known to be pathogenic for humans and dogs (4). All serotypes are reported present in all *I. ricinus* populations examined so far in Europe (10).

4. Materials and methods

In the year 2006 July to October (in the period of the highest tick activity), a total of 86 serum samples where randomly obtained (every 3. dog visiting the clinics) from healthy, asymptomatic dogs, from different regions in Iceland. See figure 1.

The samples were collected from the cephalic vein of the dogs, by means of 4,7 ml serum-gel coated tube and a 22G needle. After centrifugation the serum was stored in a freezer until it was sent to VetMedLabor in Ludwigsburg Germany for a whole-cell Enzyme-linked immunosorbent assay (ELISA), and if sera is tested positive it should be confirmed by Western blot (WB).

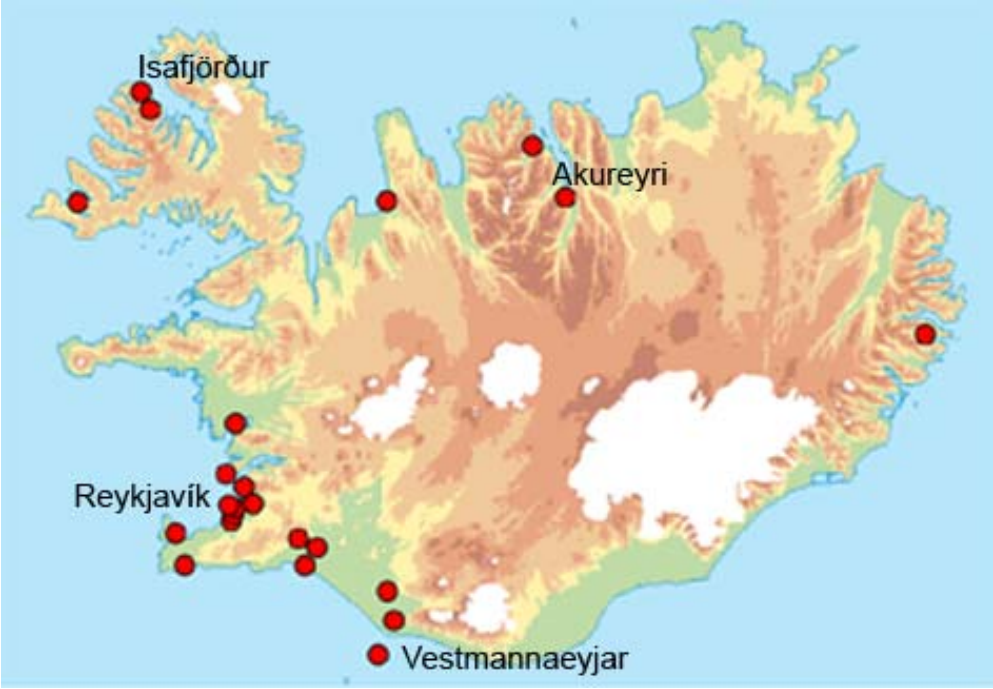

Figure 1: Sampling locations in Iceland.

Sample size was calculated from the program FreeCalc on EpiVetNet (49). The estimated population size was 12000, estimated prevalence below 10% with 95% confidence interval. Excluded were imported dogs, since they might have been vaccinated in their country of origin.

The sera where partly provided by local veterinary surgeons, and the information accompanying the sera included age, sex, breed, history and the dates when the sera were submitted.

The sera were examined by a modified commercial whole-cell ELISA test from the company Genzyme Virothech GmbH, Rüsselsheim Germany. The test is used for detection of specific antibodies against *Borrelia burgdorferi* in dogs and horses, in the IgG and IgM class using the strain *B. burgdorferi* sensu stricto (DC122.00) as antigen (50).

4.1 Test principle

The ELISA is intended for the semiquantiative and qualitative detection of IgG- and IgM-antibodies in dog or horse serum. The antibody searched for in the serum forms an immune complex with the antigen coated on the microtitre-plate. Unbound immunoglobulins are removed by washing processes. The enzyme conjugate attaches to this complex. Unbound conjugate is again removed by washing processes. After adding the substrate solution (tetramethylbenzidin-TMB), a blue dye is produced by the bound enzyme (peroxidase). The color changes to yellow, when the stopping solution is added. The specificity is 98% and sensitivity 100% for IgG and IgM (50).

The concentration of the IgG or IgM antibody titers is given in Virotech Units (VE), see table 2 below.

Table 2: VE units (50).

VE	**IgG**	**IgM**
< 8,0	negative	negative
8,0-12,0	borderline	borderline
> 12,0	positive	positive

VE unit below 8 is considered negative. VE unit between 8 and 12 is considered borderline and should be tested again after 2-3 weeks if the dog is symptomatic. VE units above 12 are considered positive.

5. Results

The dogs were of various breeds, 30 females (34,9%) and 56 males (65,1%) and the age ranged from 6 months to 16 years. See figure 2.

At least 3 of the dogs did have a history of tick bites within previous year.

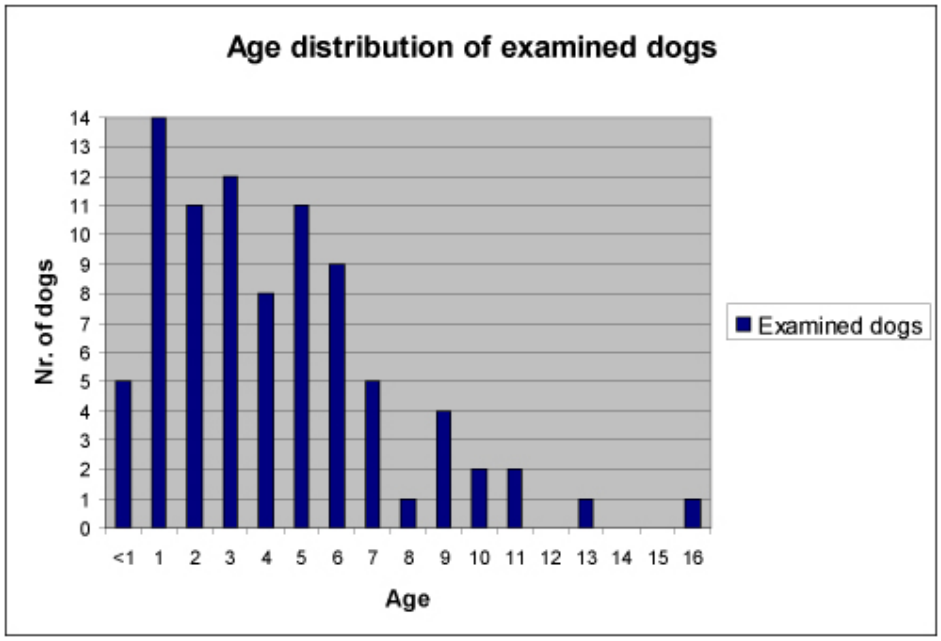

Figure 2: Age distribution of examined dogs

Of the 86 sera examined for the IgG and IgM antibodies to *B.burgdorferi* s.l. 94,2% (81/86) were found negative, inclusive the 3 dogs with previous history of tick bites. Approximately 5,8% (5/86) were considered borderline, e.g. ranging from 8-12 VE units for both IgG and IgM. The mean age of those 5 dogs was 4,6 years (1-9 years) and they were of various breeds. 3 came from Reykjavík, 1 from Ísafjörður and 1 from Vestmannaeyjar. See Appendix 1. No sera (0/86) were found positive. See figure 3.

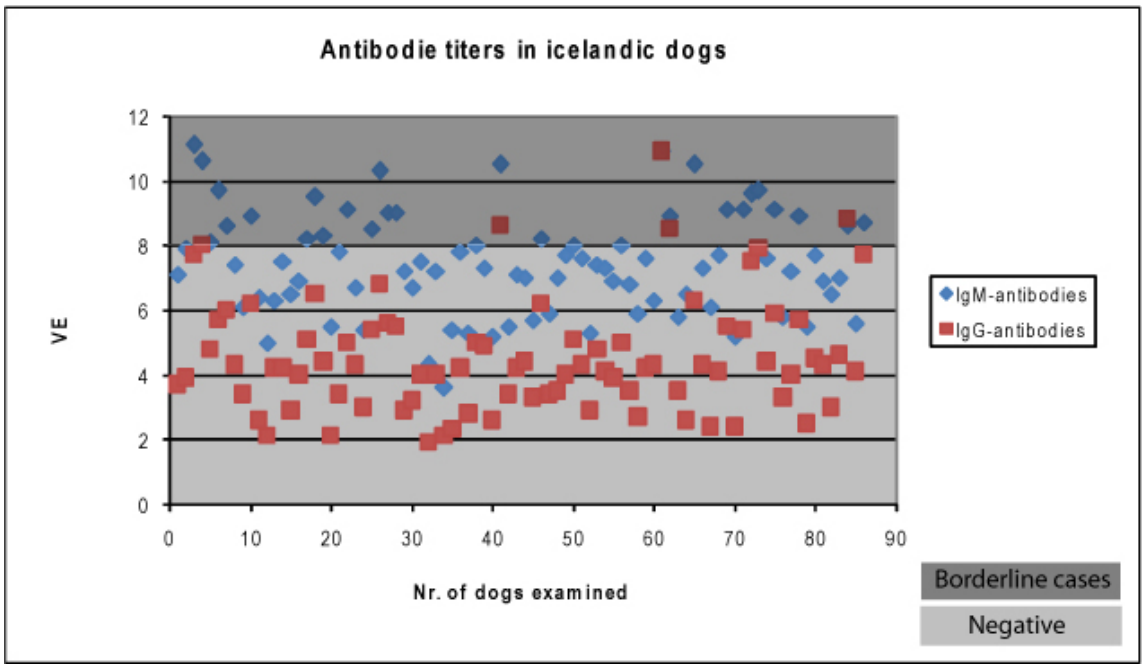

Figure 3: Antibody titers in Icelandic dogs.

6. Discussion

Since none of the 86 dogs tested positive for antibodies against *Borrelia burgdorferi* s.l., the relative risk for dogs in Iceland to acquire LB has to be estimated as low. In order to explain it, the fact has to be considered that the main vector *I. ricinus* has not been considered endemic in Iceland although cases, where they have infested humans and domestic animals, have increased through the years (32). Their ideal climate is high humidity >80% and temperature between 14-23ºC. These conditions are mainly found in woods and wooded areas (51). Due to Iceland´s cold climate, rough vegetation and weather fluctuations the vector probably is not able to settle in Iceland. But due to global warming these conditions can change to the benefit of the vectors.

This has been demonstrated well in Sweden. *I. ricinus* is endemic in South- and Mid-Sweden but North-Sweden has hitherto been considered a non-endemic area. Now the northward spread of ticks and a gradual rise in human LB cases in the region have been related to milder winters, springs and autumns (52). However the effect of climate has been disputed by other authors (53).

A study by Egenvall et al. 2000 on 588 dogs from three regions in Sweden revealed that none of the 96 dogs originating from Norrland (North-Sweden) were positive for *Borrelia burgdorferi* s.l., while the remaining 492 dogs from Götaland (South-Sweden) and Swealand (Mid-Sweden) showed 3,9% seroprevalence (16).

Other Scandinavian studies have given differing results. In Denmark a study by Hansen and Dietz 1997 on 205 healthy dogs showed 16,1% seroprevalence (17). In Norway Åkerstedt et al. 1996 found 13,8% (12/87) of samples from dogs visiting one animal clinic in Aust-Agder were positive (21) and Csango and Stamberg 1996 found antibodies to *Borrelia burgdorferi* s.l. in 27% of 149 dogs (22).

Further studies of unsuspected randomly sampled dogs in Europe have compared hunting dogs with other dogs, since use of the dog is seen as a potential risk factor. In Slovakia Stefancikova et al. 1996 found that the seroprevalance among military service dogs was 11,8%, while it was 40% among hunting dogs (20). In Spain Merino et al. 2000 found 84% seroprevalence among hunting dogs and 35% among watchdogs (23). In the Netherlands Goossens et al. 2001 found no significant differences between hunting dogs 18% and non-hunting dogs 17% (18). In this study both hunting dogs, rescue dogs and also pet dogs were included.

Other potential risk factors like age, breed, sex, habitat, season and presence of ticks on the animal were also considered in current study. The 86 dogs were at different ages (see figure 2), both sexes (65,1% males and 34,9% females), various breeds and from different regions in Iceland (see figure 1). The samples were collected from July to October 2006, in the period of the highest tick activity and 3 dogs had history of tick-bite within the previous year. A study by Pejchalová et al. 2006 in the Czech republic found 6,5% overall seroprevalance in military dogs and a significantly higher seroprevalence among older dogs than younger dogs (24). That corresponds to results of other authors (16, 23).

Baatz et al. 2000 reveal that due to genetic differences, breeds like Golden retrievers and Labrador retrievers are more susceptible and therefore more likely to be infected than other breeds (27). Some authors have suggested that the dog poses a risk for its owner of acquiring LB since it, through outdoor activities, easily comes in contact with infected ticks (20).

According to Goossens et al. 2001 no positive correlation was observed between seropositivity of hunters and their dogs, thus direct transfer of ticks between dog and owner is probably insignificant (18).

The vector competence of *I. uriae* for *B. burgdorferi* s.l. has never been demonstrated under laboratory conditions, but its involvement as a vector of borrelial spirochaetes in transmission cycles in seabird colonies has been shown (29). Two seroepidemiological surveys in Vestmannaeyjar in Iceland in puffin-hunters showed no seroconversion among them, even though 10% of them recalled being bitten by *I. uriae* within the previous year (unpublished 32). On Faeroe Islands a similar survey was performed. Of 81 serum samples from puffin hunters, 3 were found to be positive. The findings of seropositive Faeroe Islanders who are regularly exposed to *I. uriae* indicate that there may be a transfer of *B. garinii* by this tick species to humans (54).

A study by Bunikis et al. 1995 concluded that the reliability of a serological investigation of LB increases when antigens are prepared from local isolated strains (55). In this study all serum samples where sent to Germany for evaluation, using the strain *Borrelia burgdorferi* sensu stricto as antigen. As mentioned before, only the strain *B. garinii* has been isolated from seabird ticks in Iceland (29). This could possibly give false negative results but since a whole-cell ELISA test is used, the cross-reactions among *Borrelia spp.* is over 99%. But the use of whole-cell ELISA can also give cross-reactions to other related spirochetes such as *Treponema spp.* and *Leptospira spp.* (50).

Levy et al. 1993 demonstrate that in a whole-cell ELISA the closer the fit of antibody to antigen, the stronger the reaction. High antibody titer is therefore most likely the result of reactivity to *B. burgdorferi.* Low antibody titer can however represent reactivity to cross-reactive antigens (5). The 5,8% (5/86) of borderline cases in this study could be due to cross-reactions.

7. Conclusions

The conclusion is that Lyme Borreliosis is not an endemic disease in Iceland and the risk assessment of dogs in Iceland acquiring the disease is low.

Yet it has to be considered that the density and geographical ranges of the main vector *I. ricinus*, probably because of global warming, has increased. Therefore Icelandic veterinary surgeons should be on guard for this disease and other infections that ticks may carry.

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Appendix 1

Sampled dogs in Iceland:

Name:	Breed:	Age:	Region:	IgG	IgM	Ticks Y/N	Pos/Neg/Bo
Egó	mixed br	6 mán	Akranes	2,1	5,5	No	Neg
Hómer	Am.cocker	1 árs	Akranes	4	7,5	No	Neg
Garpur	mixed br	1 árs	Akureyri	4,3	6,7	No	Neg
Ringo	R.Collie	2 ára	Akureyri	5,7	8,9	No	Neg
Pjakkur	mixed br	4 ára	Akureyri	4,5	7,7	No	Neg
Ási	mixed br	3 ára	Akureyri	4,3	6,9	No	Neg
Týra	mixed br	4 ára	A-Landeyjar	4,3	7,4	No	Neg
Perla	Labrador	2 ára	Blönduós	5,1	8,2	No	Neg
Lady	E.Setter	4 ára	Bolungarvík	3	6,5	No	Neg
Stjarna	mixed br	5 ára	Borgarnes	6,5	9,5	No	Neg
Bassi	Labrador	1 árs	Borgarnes	4	7,2	No	Neg
Skuggi	Labrador	6 mán	Dalvík	2,5	5,5	No	Neg
Dúlla	I.sheepd	16 ára	Eyrarbakki	2,1	3,6	No	Neg
Kátur	Poodle	6 ára	Fáskrúðsfj	3,9	6,9	No	Neg
Tinna	mixed br	5 ára	Grindavík	3,3	5,8	No	Neg
Prímó	Schaeffer	1 árs	Hafnarfjörður	4	7,7	No	Neg
Kolur	mixed br	3 ára	Hafnarfjörður	5,1	8	No	Neg
Spice	Weimeran	6 ára	Hafnarfjörður	4,1	7,3	No	Neg
Herkúles	Schaeffer	1 árs	Hafnarfjörður	6,3	10,5	No	Neg
Lýsa	Golden ret	1 árs	Hverageröi	3,7	7,1	No	Neg
Lubba	mixed br	6 ára	Hvolsvöllur	2,9	5,3	No	Neg
Skuggi	mixed br	3 ára	Ísafjörður	4	6,9	No	Neg
Rex	Border co	7 ára	Ísafjörður	2,8	5,3	No	Neg
Stjarna	mixed br	8 ára	Ísafjörður	5	8	No	Neg
Dofri	Labrador	5 ára	Ísafjörður	4,9	7,3	No	Neg
Púki	Schaeffer	5 ára	Ísafjörður	2,6	5,2	No	Neg
Tryggur	Labrador	5 ára	Ísafjörður	8,6	10,5	No	Borderline
Prince	Boxer	6 ára	Keflavík	3,3	5,7	No	Neg
Tása	I.sheepd	3 ára	Kjalarnes	2,9	6,5	No	Neg
Prins	Border co	4 ára	Kjalarnes	4,2	7,6	No	Neg
Leo	Schaeffer	6 ára	Kjalarnes	7,5	9,6	No	Neg
Pjakkur	Golden ret	7 ára	Kópavogur	4,8	8,1	No	Neg
Tindri	Labrador	1 árs	Kópavogur	2,9	7,2	No	Neg
Máni	Beagle	5 ára	Kópavogur	6,2	8,2	No	Neg
Tinna	Labrador	3 ára	Kópavogur	3,5	5,8	No	Neg
Tító	Labrador	1 árs	Kópavogur	2,6	6,5	No	Neg
Máni	mixed br	5 ára	Kópavogur	5,9	9,1	No	Neg
Skoti	Border co	6 ára	Mosfellsbær	2,6	6,4	No	Neg
Zorro	Silki terrier	3 ára	Mosfellsbær	4,2	7,5	No	Neg
Kolur	mixed br	2 ára	Mosfellsbær	3,5	6,8	No	Neg
Máni	Golden ret	5 ára	Mosfellsbær	4,3	6,3	No	Neg
Loppa	mixed br	3 ára	Mosfellsbær	4,1	7,7	No	Neg

Name:	Breed:	Age:	Region:	IgG	IgM	Ticks Y/N	Pos/Neg/Bo
Finnur	mixed br	10 ára	Mosfellsbær	2,4	5,2	No	Neg
Baldur	Schaeffer	2 ára	Mosfellsbær	4,4	7,6	No	Neg
Skutla	Border co	2 ára	Patreksfj	3,4	5,9	Yes	Neg
R.Teitur	Vorsteh	4 ára	Reykjavík	3,9	7,9	No	Neg
Check	Am.cocker	2 ára	Reykjavík	7,7	11,1	Yes	Neg
Neró	Schaeffer	1 árs	Reykjavík	8	10,6	No	Borderline
Breki	Golden ret	1 árs	Reykjavík	6	8,6	No	Neg
Bubbi	mixed br	11 ára	Reykjavík	3,4	6,1	No	Neg
Týri	mixed br	1 árs	Reykjavík	6,2	8,9	No	Neg
Kolbeinn	mixed br	4 mán	Reykjavík	2,1	5	No	Neg
Tobbi	West h.t.	7 ára	Reykjavík	4,2	6,3	Yes	Neg
Trilla	mixed br	4 ára	Reykjavík	4,4	8,3	No	Neg
Katla	Rottweiler	3 ára	Reykjavík	3,4	7,8	No	Neg
Fígó	Labrador	2 ára	Reykjavík	5	9,1	No	Neg
Kata	Sp.spaniel	6 ára	Reykjavík	3	5,4	No	Neg
Fróði	Sp.spaniel	3 ára	Reykjavík	5,4	8,5	No	Neg
Tara	Irish setter	9 ára	Reykjavík	6,8	10,3	No	Neg
Þöll	Labrador	11 ára	Reykjavík	5,6	9	No	Neg
Bangsi	King pood	4 ára	Reykjavík	5,5	9	No	Neg
Kría	Border co	6 ára	Reykjavík	3,2	6,7	No	Neg
Kristófer	Golden ret	13 ára	Reykjavík	1,9	4,3	No	Neg
Patti	mixed br	10 ára	Reykjavík	3,4	5,5	No	Neg
Týr	mixed br	2 ára	Reykjavík	4,2	7,1	No	Neg
Lísa	mixed br	1 árs	Reykjavík	4,4	7	No	Neg
Hringur	I.sheepd	7 ára	Reykjavík	4,3	7,6	No	Neg
Kolur	I.sheepd	6 ára	Reykjavík	4,8	7,4	No	Neg
Hvatur	I.sheepd	5 ára	Reykjavík	5	8	No	Neg
Salvör	Rottweiler	6 mán	Reykjavík	2,7	5,9	No	Neg
Fúsi	I.sheepd	9 ára	Reykjavík	10,9	10,9	No	Borderline
Prins	Am.cocker	3 ára	Reykjavík	8,5	8,9	No	Borderline
Birta	mixed br	2 ára	Reykjavík	4,3	7,3	No	Neg
Daisy	Am.cocker	9 ára	Reykjavík	2,4	6,1	No	Neg
Gutti	mixed br	9 ára	Reykjavík	5,5	9,1	No	Neg
Nemó	Labrador	5 ára	Reykjavík	5,4	9,1	No	Neg
Nala	Rottweiler	3 ára	Selfoss	5,7	9,7	No	Neg
Strútur	Border co	4 ára	Selfoss	2,3	5,4	No	Neg
Sabina	Bulldog	2 ára	Selfoss	3,5	7	No	Neg
Sesar	Schaeffer	6 mán	Seltjarnarnes	4	7,2	No	Neg
Patti	mixed br	1 árs	Vestmannaeyjar	4,2	7,8	No	Neg
Steinunn	Boxer	2 ára	Vestmannaeyjar	7,9	9,7	No	Neg
Emil	Labrador	1 árs	Vestmannaeyjar	4,6	7	No	Neg
Hjördís	I.sheepd	5 ára	Vestmannaeyjar	8,8	8,6	No	Borderline
Lára	Poodle	7 ára	Vestmannaeyjar	4,1	5,6	No	Neg
Rósa	Labrador	3 ára	Vestmannaeyjar	7,7	8,7	No	Neg