Serologic Survey for Antibodies to *Borrelia burgdorferi* in White-tailed Deer in Ontario

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ABSTRACT: Serum samples collected from 623 white-tailed deer (*Odocoileus virginianus*) in southern Ontario (Canada) from 1985 to 1989 were tested for antibodies to *Borrelia burgdorferi* using an indirect fluorescent antibody (IFA) staining method. Samples from 150 of the deer were also tested using an enzyme-linked immunosorbent assay (ELISA). At IFA titers of 1:64 and 1:128 deer with antibodies to *B. burgdorferi* appeared to be widespread throughout southern Ontario, with an apparent prevalence ranging from 3 to 47%. At IFA titres ≥1:256 and ELISA titres ≥1:160 deer with antibodies to *B. burgdorferi* were only present on Long Point which is the only known endemic focus of *Ixodes scapularis*, the primary vector for *B. burgdorferi*, in southern Ontario. At these titres the apparent prevalence of antibodies to *B. burgdorferi* on Long Point was only 5 to 7%, even though the mean intensity of infestation of adult *I. scapularis* on deer was >180, and 60% of the adult ticks are infected with *B. burgdorferi*. Based on these results, white-tailed deer do not appear to be a good sentinel species for the distribution of *B. burgdorferi*.

Key words: *Borrelia burgdorferi*, Lyme disease, *Odocoileus virginianus*, sentinel species, seroprevalence, survey, white-tailed deer.

White-tailed deer (*Odocoileus virginianus*) are the definitive host of *Ixodes scapularis* which is the primary vector for *Borrelia burgdorferi*, the etiologic agent of Lyme disease in eastern North America. Several studies (Magnarelli et al., 1984a, 1984b, 1986, 1991; Gill et al., 1993; Mahnke et al., 1993) have reported antibodies to *B. burgdorferi* in white-tailed deer from the eastern United States, and it has been suggested that serological surveys in white-tailed deer may be useful for determining the distribution of *B. burgdorferi* (Gill et al., 1993; Mahnke et al., 1993). In the present paper, we report the results of a serological survey for antibodies to *B. burgdorferi* in white-tailed deer from the southern part of the province of Ontario (Canada). The only known endemic focus for *I. scapularis* in southern Ontario is on Long Point (44°34′N; 80°10′W) on the north shore of Lake Erie (Barker et al., 1992). However, Lyme borreliosis has been reported in Ontario residents who have not travelled to areas where *I. scapularis* is endemic (C. LeBer, pers. comm.), and seropositive dogs which have not travelled to *I. scapularis* endemic areas have also been reported (Artsob et al., 1993). Our aim was to determine if antibodies to *B. burgdorferi* were widespread in white-tailed deer in southern Ontario.

Blood samples were collected from white-tailed deer at check stations during the fall hunting season, from animals immobilized during the winter months for studies on deer ecology, and from animals euthanized following motor vehicle collisions, in various locations throughout southern Ontario (Fig. 1) from 1985 to 1989. Serum was frozen at −70 °C and later analyzed. All of the samples were analyzed using an indirect fluorescent antibody assay (IFA) starting at a dilution of 1:32 as described by Artsob et al. (1993). One hundred and fifty samples were also analyzed using an enzyme-linked immunosorbent assay (ELISA) (Magnarelli et al., 1991). One hundred and twelve of these samples were from Long Point. The remaining 38 samples, which included all of the samples with IFA titres ≥1:64, were from other locations.
ELISA, with titres $\geq1.640$. Five of the samples which were seropositive using ELISA had IFA titres $\geq1.256$, as did three of the ELISA negative samples (titres $<1.160$). None of the 38 samples from other locations were seropositive using ELISA, although 20 had IFA titres $\geq1.64$.

At IFA titres $<1.256$, antibody to B. burgdorferi appears to be widespread throughout southern Ontario, but it is only common at Point Pelee and Long Point. The occurrence of deer with low antibody titres in areas outside of the known focus of I. scapularis may be caused by transmission from nymphs disseminated on migrating birds (Battalay et al., 1987). It may also be caused by cross-reaction with other agents, such as Leptospira interrogans. Antibodies to B. burgdorferi will cross-react with other spirochetes in both IFA and ELISA (Magnarelli et al., 1986, 1987), and may also cross-react with other bacterial proteins (Hansen et al., 1988). L. interrogans serovars pomona, grippotyphosa, and icterohemorrhagiae, which cross-react with B. burgdorferi at dilutions of 1:64 and 1:128 (Magnarelli et al., 1986), have been reported from white-tailed deer in southern Ontario (Abdulla et al., 1962).

At an IFA titre $\geq1.256$ and ELISA titre $\geq1.160$, the only seropositive deer were on Long Point. This correlates with the observation that Long Point is the only en-

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**TABLE 1.** Prevalence (%) of titres to the antibodies to *Borrelia burgdorferi* determined by indirect immunofluorescent assay from white-tailed deer at different locations in southern Ontario.

<table>
<thead>
<tr>
<th>Locations</th>
<th>Year</th>
<th>Months</th>
<th>$n^b$</th>
<th>1:64</th>
<th>1:128</th>
<th>1:256</th>
</tr>
</thead>
<tbody>
<tr>
<td>Long Point</td>
<td>1987–89</td>
<td>Oct/Nov</td>
<td>116</td>
<td>15</td>
<td>11</td>
<td>7</td>
</tr>
<tr>
<td>Rondeau</td>
<td>1986–87</td>
<td>Jan/Feb</td>
<td>37</td>
<td>14</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Point Pelee</td>
<td>1989</td>
<td>Jan–May</td>
<td>19</td>
<td>37</td>
<td>11</td>
<td>0</td>
</tr>
<tr>
<td>Wingham</td>
<td>1985–87</td>
<td>Nov–Feb</td>
<td>40</td>
<td>5</td>
<td>5</td>
<td>0</td>
</tr>
<tr>
<td>Maple</td>
<td>1985–86</td>
<td>Feb–July</td>
<td>9</td>
<td>11</td>
<td>11</td>
<td>0</td>
</tr>
<tr>
<td>Huronia</td>
<td>1985–87</td>
<td>Nov–Apr</td>
<td>70</td>
<td>7</td>
<td>4</td>
<td>0</td>
</tr>
<tr>
<td>South Canonto</td>
<td>1985–86</td>
<td>Nov</td>
<td>33</td>
<td>3</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>North Canonto</td>
<td>1985–86</td>
<td>Nov</td>
<td>84</td>
<td>5</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Loring</td>
<td>1986–87</td>
<td>Oct–Aug</td>
<td>215</td>
<td>4</td>
<td>1</td>
<td>0</td>
</tr>
</tbody>
</table>

*a* See Fig. 1 for locations.

*b* Number of deer.

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**GRAPH 1.** Map of southern Ontario showing the locations from which sera were collected from white-tailed deer. The locations are (1) Long Point National Wildlife Area (44°34'N; 80°10'W), (2) Rondeau Provincial Park (42°17'N; 81°51'W), (3) Point Pelee National Park (41°57'N; 82°31'W), (4) Wingham District (43°53'N; 81°10'W), (5) Maple District (43°51'N; 79°31'W), (6) Huronia District (44°27'N; 79°44'W), (7) South Canonto Township (45°05'N; 76°50'W), (8) North Canonto Township (45°10'N; 76°53'W), and (9) Loring District (45°56'N; 80°00'W).
The apparent prevalence of antibodies to *B. burgdorferi* in white-tailed deer on Long Point is somewhat surprising given that all of the deer are infested with adult *I. scapularis* from October to December with a mean intensity of infestation 180 ticks (Watson and Anderson, 1976; Lindsay, 1995), and that approximately 60% of the unfed adult ticks are infected with *B. burgdorferi* (Lindsay et al., 1991). Experimentally-infected deer develop an antibody response to *B. burgdorferi* (Gill et al., 1993; Mahnke et al., 1993; Lane et al., 1994; Luttrell et al., 1994), but the intensity of the antibody response appears to vary between the JD-1 and SH-2 strains (Luttrell et al., 1994), and the response to the same strain may vary widely between deer (Lane et al., 1994). Thus, the low apparent prevalence of antibodies in deer on Long Point may be caused in part by the strains of *B. burgdorferi* which are present.

Another factor causing the low apparent prevalence of antibodies to *B. burgdorferi* in white-tailed deer on Long Point may be the timing of the antibody response. Magnarelli et al. (1995) reported the highest prevalence of antibodies from February to April. The antibody response to *B. burgdorferi* in experimentally-infected deer takes 3 wk to develop and appears to be decreasing by 10 wk (Lane et al., 1994; Luttrell et al., 1994). White-tailed deer on Long Point support large numbers of larvae and adult *I. scapularis*, but few nymphs (Watson and Anderson, 1976; Lindsay, 1995). The prevalence of *B. burgdorferi* is <0.2% in larvae (Lindsay et al., 1991) which are found on deer from March to August (Watson and Anderson, 1976), and the prevalence in nymphs, which are found on deer from April to August, is 17%. Adult ticks are found on deer from October to April. Most of the deer from Long Point were collected in October (Table 1). Thus, deer which had been exposed to *B. burgdorferi* from the adult ticks may not have had time to develop a detectable antibody response. The prevalence of IFA titres did not differ between fawns, yearlings and adults (ch<sup>2</sup> = 4.34; P = 0.36; Zar, 1974), thus it is doubtful that the antibody response resulted from exposure the previous year.

Based on the results of this study, white-tailed deer do not appear to be a good sentinel species for serologic surveys for *B. burgdorferi*. The occurrence of seropositive deer with IFA titres of 1:64 and 1:128 in areas without endemic populations of *I. scapularis* suggests that the tests are not sufficiently specific when cross-reacting bacteria may be present in the deer herd. The low apparent prevalence at higher end-points on Long Point where deer were heavily infested with adult ticks, 60% of which are infected with *B. burgdorferi*, suggests that white-tailed deer do not develop a sufficiently strong antibody response to compensate for the loss of sensitivity associated with an increased specificity at higher cutoff values, even when they are heavily parasitized by ticks.

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**LITERATURE CITED**


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