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Author(s): Muhammad G. Morshed, John D. Scott, Keerthi Fernando, Lorenza Beati, Daniel F. Mazerolle, Glenna Geddes, and Lance A. Durden
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# MIGRATORY SONGBIRDS DISPERSE TICKS ACROSS CANADA, AND FIRST ISOLATION OF THE LYME DISEASE SPIROCHETE, *BORRELIA BURGDORFERI*, FROM THE AVIAN TICK, *IXODES AURITULUS*

**Muhammad G. Morshed, John D. Scott*, Keerthi Fernando†, Lorenza Beati‡, Daniel F. Mazerolle§, Glenna Geddes†, and Lance A. Durden‖**

Laboratory Services, British Columbia Centre for Disease Control, Vancouver, British Columbia, Canada V5Z 4R4 and Department of Pathology and Laboratory Medicine, 655 West 12th Avenue, University of British Columbia, Vancouver, British Columbia, Canada V5Z 4R4. *e-mail: mmorshed@interchange.ubc.ca*

ABSTRACT: During a 3-yr comprehensive study, 196 ixodid ticks (9 species) were collected from 89 passerine birds (32 species) from 25 localities across Canada to determine the distribution of avian-associated tick species and endogenous Lyme disease spirochetes, *Borrelia burgdorferi* Johnson, Schmid, Hyde, Steigerwalt, and Brenner. We report the following first records of tick parasitism on avian hosts: the rabbit-associated tick, *Ixodes dentatus* Marx, from Manitoba and Ontario; the mouse tick, *Ixodes muris* Bishopp and Smith, from British Columbia; and the blacklegged tick, *Ixodes scapularis* Say, from New Brunswick. Moreover, we provide the first record of the Neotropical tick, *Amblyomma humerale* Koch (1 nymph), in Canada and its parasitism of any bird. This tick was compared morphologically with nymphs of other Neotropical *Amblyomma* spp., and genetically, using a 344-bp fragment of the 12S rDNA sequence of 41 New World *Amblyomma* species. The first collections of the western blacklegged tick, *Ixodes pacificus* Cooley and Kohls, from passerine species in Alberta and British Columbia, are also reported. Notably, we further report the first isolation of *B. burgdorferi* from the bird tick, *Ixodes auritulus* Neumann, collected from an American robin, *Turdus migratorius* L., on Vancouver Island. Furthermore, *B. burgdorferi*-positive *I. auritulus* larvae were collected from a reservoir-competent fox sparrow, *Passerella iliaca* (Merrem). Our findings indicate that ground-dwelling passerines, in particular, are parasitized by certain ixodid ticks and play an important role across Canada in the wide dispersal of *B. burgdorferi*-infected ticks and increased risk of Lyme disease exposure.

Ground-dwelling birds that forage among leaf litter and low-lying vegetation are especially susceptible to parasitism by certain ixodid ticks (Wright et al., 2000). Avian ticks normally attach to the skin of the head, neck, and ventral feather track of passerine birds as they take a blood meal (Nicholls and Callister, 1996; Durden et al., 2001; Gregoire et al., 2002). Some tick species harbor tick-borne pathogens that can be transmitted to humans and domestic animals resulting in debilitating or fatal outcomes. In eastern and central Canada, the blacklegged tick, *Ixodes scapularis* Say (northern populations previously considered *I. dammini* Spielman, Clifford, Piesman and Corwin) (Oliver et al., 1993; Keirans et al., 1996), is a vector of several tick-borne pathogens. The Lyme disease spirochete, *Borrelia burgdorferi* Johnson, Schmid, Hyde, Steigerwalt, and Brenner (Burgdorfer et al., 1982) is harbored by *I. scapularis*, which acts as a competent vector (Burgdorfer and Gage, 1986; Piesman and Sinsky, 1988; Sanders and Oliver, 1995). The *Borrelia* genospecies that cause Lyme disease are collectively referred to as *B. burgdorferi* sensu lato, and this multisystem illness was first described clinically in North America in a Wisconsin physician who was bitten locally by a tick in October 1969 (Scimenti, 1970). Immature stages (larvae, nymphs) of *I. scapularis* are known to parasitize several species of birds, some of which serve as competent reservoirs of *B. burgdorferi* (Anderson et al., 1984, 1986, 1990; Weisbrod and Johnson, 1989; Stafford et al., 1995; Rand et al., 1998; Richter et al., 2000; Durden et al., 2001). The first isolation of *B. burgdorferi* from the liver of a veery, *Catharus fuscescens* (Stephens), and from attached larval *I. scapularis* (reported as *Ixodes dammini*) documented infectivity in passerines and transmission of spirochetes (Anderson et al., 1984).

Certain *Ixodes* spp. ticks that parasitize birds may carry and transmit multiple tick-borne pathogens that can result in coinfections of hosts. *Ixodes scapularis* acts as a vector of other zoonotic pathogens that cause human granulocytic anaplasmosis (formerly ehrlichiosis) (Pancholi et al., 1995; des Vignes and Fish, 1997), human babesiosis (Piesman et al., 1986; Mather et al., 1990) and deer tick virus (Telford et al., 1997; Ebel et al., 1999), which is a variant of Powassan virus (Kuno et al., 2001). The causal organism of cat scratch disease has also been detected in *I. scapularis* (Eskow et al., 2001; Adelson et al., 2004). In the Baltic Region of Russia, Alekseev et al. (2001) reported the microorganisms that cause Lyme disease, human granulocytic anaplasmosis, and human monocytic ehrlichiosis in immature *Ixodes ricinus* (L.) removed from songbirds, and some of these ticks had dual or triple infections. Similarly, multiple tick-borne pathogens are common in focal areas in the United States, which serve as origins of vector ticks that can be transported to Canada. Despite the potential for dispersal of pathogen-carrying ticks by migratory birds, health professionals often disregard tick-borne diseases in their differential diagnosis when patients live in nonendemic areas.

In far-western Canada, the western blacklegged tick, *Ixodes pacificus* Cooley and Kohls, acts as a competent vector of *Borrelia* spp. that cause Lyme disease (Banerjee et al., 1994). Gregson (1956) reported *I. pacificus* on a grouse, a gallinaceous landbird, in British Columbia, but none was noted on any passerine birds studied. *Ixodes angustus* Neumann also plays a role

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* Lyme Disease Association of Ontario, 365 St. David Street S., Fergus, Ontario, Canada N1M 2L7.
† Laboratory Services, British Columbia Centre for Disease Control, Vancouver, British Columbia, Canada V5Z 4R4.
‡ Yale University School of Medicine, Department of Epidemiology and Public Health, 60 College Street, P.O. Box 208034, New Haven, Connecticut 06520. Current Address: Institute of Arthropodology and Parasitology, Georgia Southern University, Statesboro, Georgia 30460-8056.
§ Department of Biology, University of Saskatchewan, 112 Science Place, Saskatoon, Saskatchewan, Canada S7N 5E2.
‖ Department of Biology and Institute of Arthropodology and Parasitology, Georgia Southern University, Statesboro, Georgia 30460-8042.

in the enzootic transmission of *B. burgdorferi* between small mammals in Pacific northwest coastal areas, but it is not known to parasitize birds.

In northeastern California, Wright et al. (2000) found that passerine birds play an important role in the pathogen–vector–host cycle of *B. burgdorferi* between *I. pacificus* and rodents at a tick-infested site located in the Sierra Nevada foothills. At this site, the highest prevalence of *B. burgdorferi* in *I. pacificus* nymphs occurred during April, which coincides with the spring migration of many Neotropical bird species, i.e., birds that breed in temperate areas and overwinter in tropical areas, such as Central and South America.

Worldwide, birds are involved in the long-range distribution of ticks, especially during peak migration periods. In North America, spring migrants move northward from the Neotropics and the southern United States via flight paths to areas extending as far as boreal forests in the Canadian North, transporting ticks thousands of kilometers (Scott et al., 2001). In Europe, *I. ricinus* ticks are commonly found on migratory songbirds, and these avian hosts are partly responsible for the heterogenous distribution of *Borrelia* spp. spirochetes (Humair et al., 1993; Olsén, Jaenson, and Bergström, 1995). Based on global studies, Olsén, Duffey et al. (1995) detected *B. burgdorferi* sensu lato in the seabird tick, *Ixodes uriae* White, collected from islands in the northern and southern hemispheres. These authors proposed that this tick and its seabird hosts are involved in transhemispheric exchange of Lyme disease spirochetes.

In Canada, songbirds play a role in the dispersal of ixodid ticks, especially along major migratory flyways. On the Atlantic flyway, *I. scapularis* was initially reported on a common yellowthroat, *Geothlypis trichas* (L.) in Nova Scotia (Bell et al., 1992). Later, *I. scapularis* was found on migratory passerines from northern Alberta to Nova Scotia, some of which were infected with *B. burgdorferi* (Morshed et al., 1999; Scott et al., 2001). On the Mississippi flyway, *I. scapularis* was documented at Thunder Cape, near Thunder Bay, Ontario (Klich et al., 1996,; Scott et al., 2001). Previously, Scott et al. (2001) reported migratory songbirds in Canada with attached *Amblyomma* spp. ticks, which are indigenous to the Neotropics. Although Lyme disease has been reported in far-western Canada (Banerjee et al., 1994), minimal information has been available for ticks on songbirds in the Pacific flyway area.

The aim of this broad-based study was to determine the presence of ixodid ticks on passerine birds in far-western Canada, in particular, British Columbia; expand our findings of tick–spirochete associations in central and eastern Canada; and identify potential tick vectors of *B. burgdorferi* that parasitize migratory passerines.

## MATERIALS AND METHODS

### Tick collections

Ticks were collected by bird banders and wildlife rehabilitators from songbirds in 25 localities across Canada spanning the period 2001–2003, with sampling from April to October. Emphasis was placed on spring migration; however, other submissions were included to provide expanded representation. The selection of participants was based on the willingness of bird banders and wildlife rehabilitators to collect ticks from birds. Bird captures were made by bird banders using Japanese mist nets. Injured or sick birds submitted by the public were examined by wildlife rehabilitators. Due to the elusive nature of tiny, immature ticks, e.g., unengorged *I. scapularis* larvae (0.8 mm), birds were scanned by moving or blowing the feathers to observe the skin. Ticks were removed using fine-pointed tweezers, retained in polyethylene vials capped with tulle netting, and placed in a ziplock bag with moist paper towel. These ticks were sent directly by courier for identification using a binocular dissecting microscope (×8 to ×40) to determine the species, developmental stage, and status of engorgement. Some of the ticks were sent to Georgia Southern University for identification, and in 1 case, an *Amblyomma* sp. tick was forwarded to Yale University School of Medicine for DNA gene sequencing for attempted species identification. For this study, a tick occurrence consists of 1 tick species on an individual bird. Because several ticks represented first-time occurrences, they were kept intact as museum specimens. We did not assess tick occurrence by season, as some banding stations do not operate during spring migration. Bird nomenclature follows the checklist of the American Ornithologists' Union (1998).

### Spirochete detection

At the British Columbia Centre for Disease Control, live ticks were surface sterilized using 10% hydrogen peroxide, followed by 70% isopropyl alcohol, and rinsed with sterile water. Part of the idiosoma contents were excised aseptically on a glass slide, placed into a drop of sterile phosphate buffered saline (PBS) and examined by phase-contrast microscopy at a magnification of ×400. Midgut contents were cultured for spirochetes in Barbour–Stoenner–Kelly (BSK) II medium, as described previously (Barbour, 1984), incubated at 34 C, and checked weekly by dark-field microscopy for 30 days. Culture tubes were checked for live spirochetes weekly. Thirty-one ticks were not tested and were kept as voucher specimens at the Lyme Disease Association of Ontario. One *Amblyomma* sp. nymph (03-5A18, RML123602), putatively identified as *Amblyomma humerale* Koch, has been deposited in the U.S. National Tick Collection (USNTC) at Georgia Southern University, Statesboro, Georgia.

### DNA extraction and PCR amplification

DNA was extracted from pure or contaminated cultures using Qiagen tissue kits (QIAGEN, Mississauga, Ontario). PCR was performed to amplify a portion of the variable spacer region between 2 conserved structures, the 3′ end of the 5S rRNA (*rrf*) and the 5′ end of the 23S rRNA (*rrl*), as described previously (Postic et al., 1994), and, similarly, a portion of the outer surface protein A (*Osp*A) gene (Persing et al., 1990).

The PCR mixture for the variable spacer region consisted of 1 commercial bead containing 1.5 units of *Taq* polymerase (Roche Diagnostics, Quebec, Canada), 10 mM Tris-HCl (pH 9.0 at room temperature), 50 mM KCl, 1.5 mM $MgCl_2$, 200 μM of each deoxynucleoside triphosphate (dNTP) (Roche Diagnostics), and stabilizers, including bovine serum albumin (Amersham Pharmacia Biotech, Quebec, Canada), 1 μl (20 pmol) primer 1 (CTGCGAGTTCGCGGGAGA) and 1 μl (20 pmol) primer 2 (TCCTAGGCATTCACCATA), both from the same supplier (Sigma, Oakville, Ontario), and 10 μl extracted DNA in a total volume of 30 μl. Thermal cycling consisted of 5 min at 94 C, 50 cycles for 1 min at 94 C, 1 min at 52 C, and 2 min at 72 C, and a final 7-min extension at 72 C.

The PCR mixture for the *Osp*A gene consisted of 1 commercial bead containing 1.5 U of *Taq* polymerase, 10 mM Tris-HCl (pH 9.0 at room temperature), 50 mM KCl, 1.5 mM $MgCl_2$, 200 μM of each dNTP, and stabilizers, including bovine serum albumin, 1 μl (20 pmol) primer 3 (TTCTGACGATCTAGGTCAAA), and 1 μl (20 pmol) primer 4 (GCAGTTAAAGTTCCTTCAAG), and 10 μl extracted DNA in a total volume of 30 μl. Thermal cycling consisted of 5 min at 95 C, 50 cycles for 1.5 min at 95 C, 1 min at 55 C, and 1.83 min (110 sec) at 72 C, and a final 7-min extension at 72 C.

Negative and positive controls were used in all PCR reactions. The negative control was sterile water, and the positive control used purified *B. burgdorferi* strain B31. Amplification products were analyzed by electrophoresis in 2.0% agarose gels followed by staining with ethidium bromide and ultraviolet light illumination.

### Gene sequencing of *B. burgdorferi*

The PCR-amplified fragments of the *rrf* (5S)-*rrl* (23S) intergenic spacer region were selected using the *Mse*I endonuclease as previously

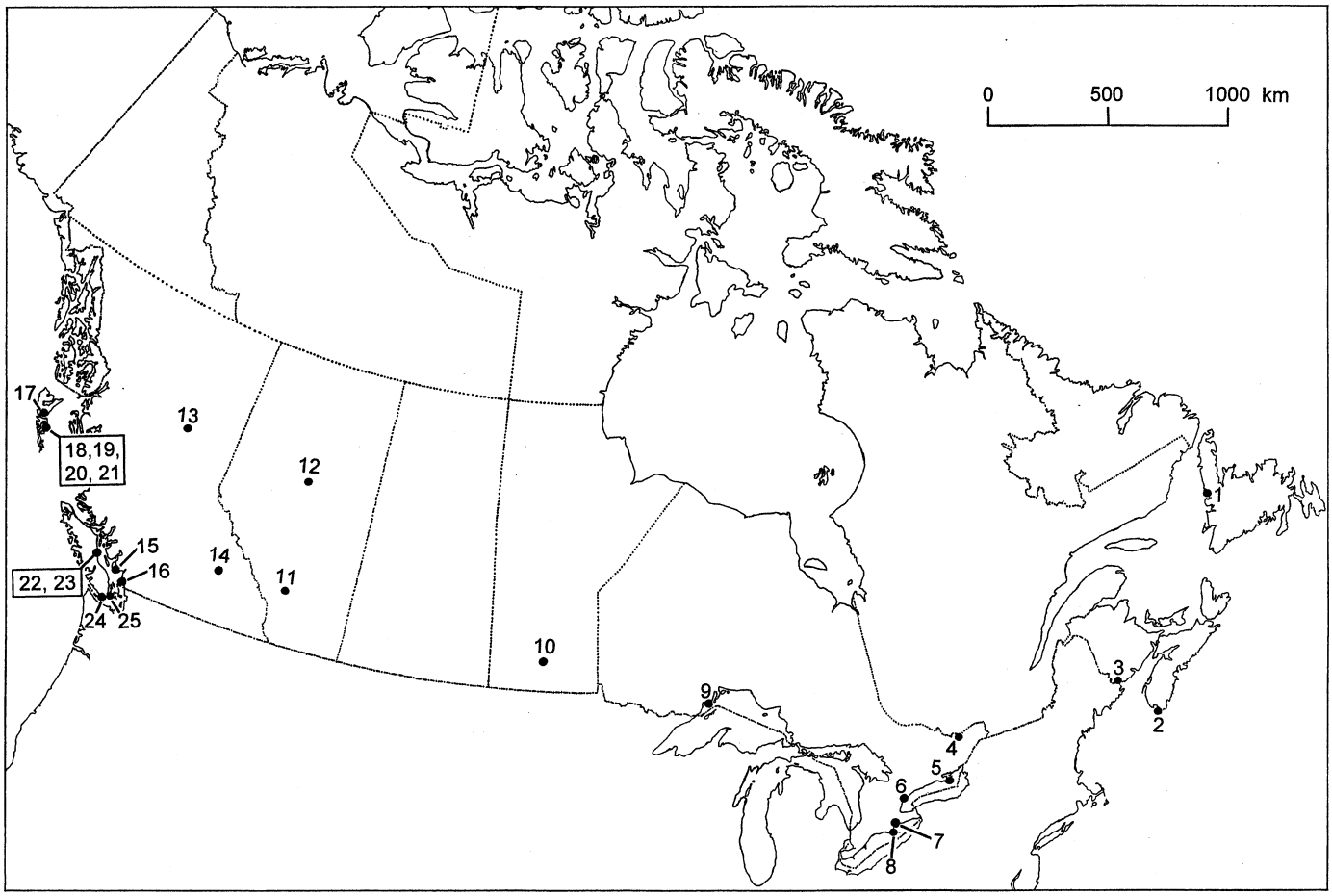


FIGURE 1. Sites in Canada where ticks were collected from songbirds. 1. Gros Morne National Park Migration Monitoring Station, Lobster Cove Head, Newfoundland and Labrador. 2. Atlantic Bird Observatory (Wolfville), Bon Portage Island, Nova Scotia. 3. Huntsman Marine Science Centre, St. Andrews, New Brunswick. 4. Innis Point Bird Observatory, Ottawa, Ontario. 5. Prince Edward Point Bird Observatory, Prince Edward Point (Picton), Ontario. 6. Fatal Light Awareness Program, Toronto, Ontario. 7. Haldimand Bird Observatory, Selkirk Provincial Park, Selkirk, Ontario. 8. Long Point Bird Observatory, Long Point (Port Rowan), Ontario. 9. Thunder Cape Bird Observatory, Sibley Peninsula (Pass Lake), Ontario. 10. Delta Marsh Bird Observatory, Delta (Portage la Prairie), Manitoba. 11. Inglewood Bird Sanctuary, Calgary, Alberta. 12. Lesser Slave Lake Bird Observatory, Slave Lake, Alberta. 13. Mackenzie Nature Observatory, Mackenzie, British Columbia. 14. Mount Revelstoke and Glacier National Parks of Canada, Revelstoke, British Columbia. 15. Wilson Creek, British Columbia. 16. British Columbia Alaksen Wildlife Refuge, Westham Island, British Columbia. 17. Queen Charlotte City, Queen Charlotte Islands (Q.C.I.), British Columbia. 18. Low Island, Q.C.I., British Columbia. 19. Reef Island, Q.C.I., British Columbia. 20. West Skedans, Q.C.I., British Columbia. 21. East Limestone, Q.C.I., British Columbia. 22. Mountainaire Avian Rescue Society (Courtenay), Black Creek, Vancouver Island, British Columbia. 23. Mountainaire Avian Rescue Society, Merville, Vancouver Island, British Columbia. 24. Rocky Point Bird Observatory, Rocky Point, Vancouver Island, British Columbia. 25. Victoria, British Columbia. Mailing addresses are listed in parentheses.

described (Postic et al., 1994). PCR products were purified using Microcon PCR centrifugal filter columns (Millipore Corp., Billerica, Massachusetts). Dye-terminated fragments were produced using ABI BigDye Terminator Sequencing kits (Applied Biosystems Co.). Fragments were purified by ammonium acetate-ethanol precipitation before running on an ABI Prism 310 DNA sequencer (Applied Biosystems Co., Foster City, California). DNA sequence data were analyzed using SeqMan and Megalign modules within the Lasergene Sequence Analysis software (DNASTAR Inc., Madison, Wisconsin).

### Sequencing of putative *A. humerale* specimen

DNA was extracted from the tick specimen as previously described (Beati and Keirans, 2001), and a 344-bp portion of its mitochondrial 12S rDNA was amplified and sequenced with primers T1B and T2A (Beati and Keirans, 2001). The cuticle of the tick was preserved and mounted on a slide with Euparal (BioQuip, Rancho Dominguez, California) as a voucher specimen (RML123602) in the United States National Tick Collection (USNTC). The sequence was manually aligned to an unpublished 12S rDNA data matrix, which contains 64 homologous sequences from 41 identified adult Neotropical and Nearctic *Amblyomma* spp. Pairwise distances were calculated by Megalign v.5.51 (DNASTAR Inc.).

## RESULTS

### Ticks on birds

A total of 196 ticks (9 species) were collected from 89 passerine birds (32 species) during a 3-yr period (1 May 2001 to 31 October 2003) at 25 locations across Canada (Fig. 1). From coast to coast, 6 different *Ixodes* spp. ticks (*I. auritulus* Neumann [n = 81], *Ixodes brunneus* Koch [n = 7], *Ixodes dentatus* Marx [n = 3], *Ixodes muris* Bishopp & Smith [n = 5], *Ixodes pacificus* [n = 3], *I. scapularis* [n = 32]) were retrieved from these infested passerine hosts, and Table I provides the number

of ticks recovered from each bird. Adult ticks for 3 tick species were recorded for *I. auritulus* (n = 13), *I. brunneus* (n = 5), and *I. muris* (n = 4). Table II lists the rabbit tick, *Haemaphysalis leporispalustris* Packard (n = 63).

The mean intensity of ticks per infested bird was 2.1 (range, 1–15). The heaviest infestation occurred on a fox sparrow, which had 15 subadult *I. auritulus* (12 larvae, 3 nymphs) collected on 8 October 2003 at Rocky Point, British Columbia. For *I. scapularis*, the mean intensity was 1.5 (range, 1–5). Ticks on birds, which were collected at Delta Marsh, Manitoba, were found primarily on the anterior part of the body: 60% around the ears, 15% around the eyes, 15% on the neck, 8% around the mouth, and 2% just above the furcular cavity. The majority of ticks reported in this study were found on birds that forage primarily on the ground and in the shrub layer, such as common yellowthroat (n = 12), Swainson's thrush, *Catharus ustulatus* (Nuttall) (n = 11), hermit thrush, *Catharus guttatus* Pallas (n = 7) (Tables I, II). The earliest detection of a tick on migratory birds during the study was an *I. brunneus* female that was collected from a hermit thrush on 16 April 2003 at Long Point, Ontario. Most *I. scapularis* ticks occurred on birds during May and early June; however, 1 nymph was collected in August. The presence of *I. scapularis* on orange-crowned warblers, *Vermivora celata* (Say), collected in Manitoba, represents a new host record. Our data provide the first report of *I. scapularis* on a bird in New Brunswick; it was collected from a mourning warbler, *Oporornis philadelphia* (Wilson), on 2 June 2001 at St. Andrews, which is situated within the Atlantic migratory flyway. Of note, we received 2 *I. scapularis* nymphs from a hermit thrush, which hit an illuminated, mirrored, multistory building at night in Toronto, Ontario, on 5 May 2002 during peak migration.

For the 92 tick occurrences, simultaneous infestations by 2 different tick species were observed on 3 individual birds: *I. auritulus* (4 nymphs, 1 female) and 1 *I. pacificus* nymph on a Swainson's thrush at Rocky Point, British Columbia; a *H. leporispalustris* nymph and an *I. dentatus* larva, both rabbit-associated ticks, occurred on a Swainson's thrush at Delta Marsh, Manitoba; and 1 *I. brunneus* female and 1 *I. muris* nymph on a veery at Delta Marsh, Manitoba. Nine occurrences of coinfestations of 2 or more active developmental stages were recorded for *I. auritulus*. All 3 motile stages of *I. aurtitulus* were found concurrently on an American robin, *Turdus migratorius* L., at Merville, British Columbia, on 11 June 2002, and *B. burgdorferi* was cultured from 1 of the fully engorged females. Because many of the ticks are documented for the first time on passerine birds at several new locations, they are listed individually in chronological order (Tables I, II).

### Spirochete detection

Three isolates were cultured from live *B. burgdorferi*-positive ticks that were attached to a total of 9 different songbirds parasitized by infected ticks. In Atlantic Canada, *B. burgdorferi* was isolated from engorged *I. scapularis* nymphs removed from spring migrants at Bon Portage Island, Nova Scotia; namely, white-throated sparrow, *Zonotrichia albicollis* (Gmelin), on 20 May 2002 (GenBank AY594657), and common yellowthroat on 24 May 2002 (GenBank AY594656). In central Canada, a combined pool of larval and nymphal *I. scapularis*, which was removed from a Swainson's thrush at Delta Marsh, Manitoba, on 27 May 2002, was positive for *B. burgdorferi*. Similarly, in Ontario, we report a *B. burgdorferi*-positive *I. scapularis*, which was collected from a common yellowthroat at Long Point on the north shore of Lake Erie on 19 May 2003. Five occurrences of *B. burgdorferi*-infected *I. auritulus* are reported for 11 coastal British Columbia sites. We report the first isolation of *B. burgdorferi* from an *I. auritulus* (female), which was removed from a fledgling American robin at Merville, British Columbia, on 11 June 2002, and this bird died 5 days later after 20 ticks were removed (8 presented for identification) (GenBank AY363396). Subsequently, we recorded a *B. burgdorferi*-positive *I. auritulus* (female) collected from an American robin at Westham Island, British Columbia, on 28 July 2002. At Rocky Point, British Columbia, *B. burgdorferi*-infected *I. auritulus* subadults were collected from 3 individual birds: song sparrow, *Melospiza melodia* (Wilson), Swainson's thrush, and fox sparrow on 26 July 2003, 26 July 2003, and 8 October 2003, respectively. In the latter occurrence, visible spirochetes from a pool of *I. auritulus* larvae attached to a fox sparrow were observed by dark-field microscopy and were subsequently PCR positive for *B. burgdorferi*. In addition to our study of passerine birds, we observed and removed 10 *I. auritulus* (3 nymphs, 7 females) from a juvenile blue grouse, *Dendragapus obscurus* (Say), a nonpasserine landbird, which died on 11 Aug 2002, a day after tick removal. All of the *H. leporispalustris* that were tested were negative for *B. burgdorferi*.

Based on *rrf* (5S)-*rrl* (23S) intergenic spacer region DNA sequencing, 2 isolates from Nova Scotia and 1 isolate from British Columbia cultured from ticks attached to 3 individual songbirds were found to be *B. burgdorferi* sensu stricto.

### Gene sequencing of putative *A. humerale* specimen

Two *Amblyomma* spp. ticks were recovered from Neotropical migrants at Delta Marsh, Manitoba. An *Amblyomma longirostre* nymph was collected on 24 May 2001 from a yellow-bellied flycatcher, *Empidonax flaviventris* (Baird and Baird), and identified morphologically. Subsequently, a nymphal *Amblyomma humerale* Koch was removed on 15 May 2003 from a gray-cheeked thrush, *Catharus minimus* (Lafresnaye), and because it was submitted dead, it could not therefore be reared through the nymph–adult molt. This nymph was a morphological match with an *A. humerale* nymph collected in Trinidad in 1962 that is deposited in the USNTC (accession no. RML39442). The nymphal stage of *A. humerale* has never been formally described, but the RML39442 specimen was identified by either C. M. Clifford or G. M. Kohls, both highly renowned tick taxonomists. Because the nymphal stages of several other Neotropical *Amblyomma* spp. are also undescribed (Guglielmone et al., 2003), this nymph was analyzed genetically by DNA sequencing. The nymphal 12S rDNA sequence was most closely related to that of an adult *A. humerale* collected in Venezuela in 1985 (USNTC accession no. RML118042). The pairwise distance between the 2 sequences was 3.8%. The sequence of our *Amblyomma* sp. nymph also differed by 12.2–19.2% from all other available *Amblyomma* spp. sequences. Therefore, we feel justified in identifying this tick as *A. humerale*. The 12S rDNA gene sequence of our *Amblyomma* sp. nymph has been placed in GenBank (AY766150).

TABLE I. Occurrence of *Ixodes* spp. ticks on passerine birds in Canada, by province, and presence of *Borrelia burgdorferi* in ticks, 2001–2003.*

| Bird species | Site† id. no. | Date tick removed | *I. auritulus* | | | *I. brunneus* | | *I. dentatus* | | *I. muris* | | *I. pacificus* | | *I. scapularis* | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | L | N | F | L | F | L | N | N | F | L | N | L | N |
| **Alberta** | | | | | | | | | | | | | | | |
| Gray-cheeked thrush, *Catharus minimus* | 12 | 16 May 2001 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 2 | 0 |
| Swainson's thrush, *Catharus ustulatus* | 12 | 31 May 2001 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 |
| Wilson's warbler, *Wilsonia pusilla* | 11 | 31 May 2002 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 2 | 0 | 0 |
| **New Brunswick** | | | | | | | | | | | | | | | |
| Mourning warbler, *Oporornis philadelphia* | 3 | 02 Jun 2001 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 |
| **Nova Scotia** | | | | | | | | | | | | | | | |
| Hermit thrush, *Catharus guttatus* | 2 | 02 May 2001 | 0 | 0 | 0 | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| White-throated sparrow, *Zonotrichia albicollis* | 2 | 20 May 2002 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 2‡ |
| Common yellowthroat, *Geothlypis trichas* | 2 | 24 May 2002 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1§ |
| | 2 | 21 May 2003 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 |
| | 2 | 22 May 2003 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 |
| **Manitoba** | | | | | | | | | | | | | | | |
| Veery, *Catharus fuscescens* | 10 | 20 May 2001 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 |
| | 10 | 28 May 2001 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 |
| Common yellowthroat | 10 | 28 May 2001 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 4 | 1 |
| Orange-crowned warbler, *Vermivora celata* | 10 | 17 May 2002 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 |
| | 10 | 17 May 2002 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 2 |
| Swainson's thrush | 10 | 27 May 2002 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1‡ | 1‡ |
| Swainson's thrush‖ | 10 | 05 May 2003 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Gray-cheeked thrush | 10 | 23 May 2003 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 |
| Gray catbird, *Dumetella carolinensis* | 10 | 23 May 2003 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 |
| Northern waterthrush, *Seiurus noveboracensis* | 10 | 25 May 2003 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 |
| **British Columbia** | | | | | | | | | | | | | | | |
| Song sparrow, *Melospiza melodia* | 24 | 17 Aug 2001 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Bewick's wren, *Thryomanes bewickii* | 24 | 18 Aug 2001 | 0 | 3 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Lincoln's sparrow, *Melospiza lincolnii* | 13 | 21 Aug 2001 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| Common yellowthroat | 24 | 29 Aug 2001 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| | 14 | 30 Aug 2001 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| American robin, *Turdus migratorius* | 23 | 11 Jun 2002 | 3 | 2 | 3§ | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Hermit thrush | 19 | 01 Jul 2002 | 6 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Varied thrush, *Ixoreus naevius* | 20 | 09 Jul 2002 | 2 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Hermit thrush | 19 | 09 Jul 2002 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Winter wren, *Troglodytes troglodytes* | 18 | 13 Jul 2002 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Hermit thrush | 17 | 15 Jul 2002 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| American robin | 16 | 28 Jul 2002 | 0 | 0 | 1§ | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Common yellowthroat | 13 | 05 Sep 2002 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| American robin | 22 | 03 Jun 2003 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Hermit thrush | 21 | 29 Jun 2003 | 1 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Swainson's thrush | 24 | 24 Jul 2003 | 0 | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Song sparrow | 24 | 26 Jul 2003 | 0 | 1§ | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Swainson's thrush | 24 | 26 Jul 2003 | 0 | 4‡ | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 0 |

TABLE I. Continued

| Bird species | Site† id. no. | Date tick removed | *I. auritulus* | | | *I. brunneus* | | *I. dentatus* | | *I. muris* | | *I. pacificus* | | *I. scapularis* | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | L | N | F | L | F | L | N | N | F | L | N | L | N |
| Golden-crowned sparrow, *Zonotrichia atricapilla* | 24 | 17 Sep 2003 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Song sparrow | 15 | 23 Sep 2003 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| White-crowned sparrow, *Zonotrichia leucophrys* | 15 | 27 Sep 2003 | 0 | 2 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| American robin | 15 | 28 Sep 2003 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| | 15 | 01 Oct 2003 | 0 | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Song sparrow | 15 | 01 Oct 2003 | 1 | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Fox sparrow, *Passerella iliaca* | 15 | 02 Oct 2003 | 1 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Oregon junco, *Junco oreganus* | 24 | 03 Oct 2003 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| | 15 | 05 Oct 2003 | 0 | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Fox sparrow | 24 | 08 Oct 2003 | 8‡ | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| | 24 | 08 Oct 2003 | 12 | 3 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Winter wren | 24 | 10 Oct 2003 | 0 | 2 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Varied thrush | 25 | 27 Oct 2003 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| | | | Ontario | | | | | | | | | | | | |
| Song sparrow | 7 | 04 May 2002 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| Hermit thrush | 6 | 05 May 2002 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 2 |
| | 8 | 16 Apr 2003 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| White-throated sparrow | 5 | 02 May 2003 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| | 7 | 03 May 2003 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Gray catbird | 7 | 09 May 2003 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Common yellowthroat | 5 | 18 May 2003 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 |
| | 8 | 19 May 2003 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1§ |
| Blue jay, *Cyanocitta cristata* | 8 | 22 May 2003 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 |
| Common yellowthroat | 9 | 05 Jun 2003 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 2 | 1 |
| Swainson's thrush | 9 | 06 Jun 2003 | 0 | 0 | 0 | 0 | 0 | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Common yellowthroat | 8 | 02 Aug 2003 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 |
| Total birds 62 (22 species) | | | 34 | 34 | 13 | 2 | 5 | 3 | 0 | 1 | 4 | 0 | 3 | 9 | 23 |

* L, larva(e); N, nymph(s); F, female(s). Bird nomenclature follows check-list of the American Ornithologists' Union (1998).
† See Figure 1 for sites.
‡ Positive for *Borrelia burgdorferi*, ticks pooled.
§ Single tick positive.
‖ This bird was also parasitized by a *H. leporispalustris* nymph.

TABLE II. Occurrence of rabbit ticks, *Haemaphysalis leporispalustris*, on passerine birds in Canada, by province, 2001–2003.*

| Bird species | Site† id. no. | Date ticks removed | Larva(e) | Nymph(s) |
|---|---|---|---|---|
| | Nova Scotia | | | |
| Northern parula, *Parula americana* | 2 | 12 May 2001 | 4 | 0 |
| Common yellowthroat, *Geothlypis trichas* | 2 | 22 May 2001 | 5 | 0 |
| White-throated sparrow, *Zonotrichia albicollis* | 12 | 15 Jul 2001 | 3 | 1 |
| Magnolia warbler, *Dendroica magnolia* | 12 | 16 Jul 2001 | 1 | 0 |
| Swainson's thrush, *Catharus ustulatus* | 12 | 03 Jun 2002 | 0 | 1 |
| Ovenbird, *Seiurus aurocapillus* | 12 | 14 Jul 2002 | 2 | 0 |
| | Manitoba | | | |
| White-throated sparrow | 10 | 04 May 2003 | 1 | 0 |
| Swainson's thrush‡ | 10 | 05 May 2003 | 0 | 1 |
| White-throated sparrow | 10 | 07 May 2003 | 0 | 1 |
| Swainson's thrush | 10 | 21 May 2003 | 3 | 0 |
| Swainson's thrush | 10 | 25 May 2003 | 1 | 0 |
| | Ontario | | | |
| Nashville warbler, *Vermivora ruficapilla* | 8 | 02 Aug 2002 | 1 | 0 |
| Veery, *Catharus fuscescens* | 9 | 03 Jun 2003 | 0 | 1 |
| Song sparrow, *Melospiza melodia* | 8 | 02 Aug 9003 | 0 | 1 |
| Carolina wren, *Thryothorus ludovicianus* | 8 | 04 Aug 2003 | 0 | 2 |
| Brown thrasher, *Toxostoma rufum* | 4 | 09 Aug 2003 | 1 | 6 |
| House wren, *Troglodytes aedon* | 8 | 15 Aug 2003 | 0 | 1 |
| Carolina wren | 8 | 17 Aug 2003 | 0 | 1 |
| House wren | 8 | 18 Aug 2003 | 0 | 1 |
| | Newfoundland and Labrador | | | |
| Yellow warbler, *Dendroica petechia* | 1 | 15 Jul 2003 | 1 | 0 |
| Northern waterthrush, *Seirus noveboracensis* | 1 | 30 Jul 2003 | 1 | 0 |
| Lincoln's sparrow, *Melospiza lincolnii* | 1 | 31 Jul 2003 | 0 | 1 |
| Northern waterthrush | 1 | 31 Jul 2003 | 1 | 3 |
| Lincoln's sparrow | 1 | 04 Aug 2003 | 4 | 1 |
| Lincoln's sparrow | 1 | 04 Aug 2003 | 3 | 7 |
| Black-and-white warbler, *Mniotilta varia* | 1 | 06 Aug 2003 | 2 | 0 |
| Total birds 26 (16 species) | | | 34 | 29 |

* Bird nomenclature follows check-list of the American Ornithologists' Union (1998).
† See Figure 1 for location of each site.
‡ This bird was also parasitized by an *I. dentatus* larva.

## DISCUSSION

The billions of birds that migrate into and out of Canada each year (Rich et al., 2004) play an important role in the dispersal of ixodid ticks, including ticks infected with Lyme disease spirochetes. Based on a comprehensive study of ticks on migratory passerines, we have uncovered novel bird–tick relationships and potential vectors of *B. burgdorferi*. Notably, we report the first occurrence of *A. humerale*, a species native to South America, on a bird in Canada, and the first *I. scapularis* infected with *B. burgdorferi* collected from avian hosts in Manitoba and Ontario. Consistent with previous studies, tick infestation was common among birds that forage primarily on the ground and in the shrub layer, and occurred in both short- and long-distance migrants. It is clear that North American migratory birds are important dispersers of ticks that are vectors of *B. burgdorferi* and that migratory birds can and do disseminate *B. burgdorferi*-positive ticks to nonendemic areas. The dissemination of such ticks has important medical implications that should be considered.

As songbirds move northward during spring migration to breeding and nesting areas, they disperse ticks with endogenous pathogens. These ticks may be acquired before departure from the wintering grounds or at stopover sites, as songbirds primarily follow 3 main migration routes. Along the Atlantic flyway, songbirds acquire larval and nymphal *I. scapularis* and can distribute these ticks a few days later in eastern Canada. Likewise, songbirds traveling the Mississippi flyway acquire immature *I. scapularis* mainly in the upper Midwest, and these ticks are disseminated in central Canada. Similarly, subadult *I. pacificus* are picked up along the West Coast and transported to far-western Canada. If a migrating bird is parasitized by several immature *I. scapularis*, they can drop in 1 location to form an established population, such as Point Pelee (Banerjee et al., 2000), Rondeau Provincial Park (Morshed et al., 2003), Long Point (Barker et al., 1988; Lindsay et al., 1991; Banerjee et al., 2000), and Turkey Point (Scott et al., 2004). The latter 3 areas are now endemic for Lyme disease. En route, songbirds make stopovers at wooded and grassy areas to replenish food reserves, where ticks can detach and drop to a new microhabitat. Because humidity is vital to survival, many ixodid ticks seek a

cool, moist microclimate often in the vegetative mat of the forest floor or meadow (Sonenshine, 1993).

Interestingly, the peak spring migration of many songbirds coincides with the main questing period of *I. pacificus* and *I. scapularis* nymphs. During May and early June, *I. scapularis* nymphs have a precipitous rise in host-seeking activity in most regions and attach to spring migrants (Battaly et al., 1987). At the beginning of May, early migrants, such as hermit thrush, white-throated sparrow, song sparrow, and gray catbird, *Dumetella carolinensis* (L.), which are reported in our study, are moving through Lyme disease-endemic areas in the northern United States. Similarly, from mid-May to early June, relatively late migrants, such as Swainson's thrush, common yellowthroat, Wilson's warbler, *Wilsonia pusilla* (Wilson), and veery are in high numbers until mid-June. Consistent with our findings, Hyland et al. (2000) also reported a high prevalence of tick parasitism on common yellowthroats, especially by *I. scapularis*.

In Newfoundland and Labrador (site 1), the bird-banding program commenced after 5 June 2003 and, consequently, the main influx of spring migrants had occurred and we did not receive any incoming, attached *Ixodes* spp. ticks. Only *H. leporispalustris* was collected during the breeding and nesting season.

We report *I. pacificus* for the first time on passerine birds at both coastal and inland sites, namely, Rocky Point, British Columbia, and Calgary, Alberta; these discoveries constitute new host records in Canada. To the best of our knowledge, the collection of 2 *I. pacificus* nymphs on a Wilson's warbler collected on 31 May 2002 at Calgary, Alberta, is the first report of this tick species on a bird east of the Rocky Mountains. With earlier discoveries of *I. scapularis* in Alberta (Scott et al., 2001), both *I. scapularis* and *I. pacificus* have now been reported in this province. Typically, activity of *I. pacificus* subadults along the West Coast within the Pacific flyway is prominent from April to June (Manweiler et al., 1990).

*Ixodes auritulus*, which was initially reported on passerines and galliforms (Cooley and Kohls, 1945; Gregson, 1956), is widely distributed along the western coast of the Western Hemisphere (Durden and Keirans, 1996). As a unique event, we cultured the first *B. burgdorferi* isolate from a fully engorged *I. auritulus* female collected from an American robin. Based on reservoir competency studies of *B. burgdorferi* in American robins, Richter et al. (2000) found that this avian species could remain infected for up to 6 mo. For our isolation of *B. burgdorferi*, only 1 of 8 *I. auritulus* female cohorts was infected, which indicates that the spirochete was acquired during larval and/or nymphal blood meals. Importantly, a Swainson's thrush captured at Rocky Point, British Columbia, on 26 July 2003 was simultaneously coinfested by an *I. pacificus* nymph; however, it was only slightly engorged and apparently had not been attached long enough to acquire infection. Notably, this coinfestation shows a potential bridging link for *B. burgdorferi* between *I. auritulus* and *I. pacificus*.

*Borrelia burgdorferi* has been found in 3 species of ticks on Vancouver Island. Previously, Banerjee et al. (1994) isolated *B. burgdorferi* from *I. angustus* and *I. pacificus*. Even though *I. angustus* does not parasitize birds, it is involved in the epizootiology of Lyme disease in British Columbia. Five *B. burgdorferi*-positive *I. auritulus* collected from songbirds in our study indicate this tick species is involved in the enzootic cycling of spirochetes at coastline habitats. Significantly, the recovery of visible *B. burgdorferi*-positive spirochetes removed from engorged *I. auritulus* larvae attached to a fox sparrow at Rocky Point, British Columbia, shows direct transfer of *B. burgdorferi* from the host bird to larval ticks, and this passerine species is likely a competent host because this would have been the first blood meal for these ticks. Using indirect immunofluorescence, Wright et al. (2000) reported *B. burgdorferi*-postive blood smears from fox sparrows employing the monoclonal antibody H5332 that is directed against OspA. However, no *I. pacificus* were removed from these birds captured in the California study. Earlier, Gregson (1956) listed the meadow mouse, *Microtus* sp. as a host for *I. auritulus* in coastal British Columbia habitats. In fact, some avian and murine fauna in lower coastal British Columbia act as competent reservoir hosts. Based on the presence of *B. burgdorferi* in 3 different tick species in focal areas, several vertebrate hosts are likely involved in the enzootic cycle. Pragmatically, reservoir-competent birds that circulate locally could act as a bridging link for *B. burgdorferi* within these coastal areas.

We report several tick species in new areas of Canada. We provide the first evidence of *I. muris* in far-western Canada; namely, on a Lincoln's sparrow, *Melospiza lincolnii* (Audubon), and a common yellowthroat at McKenzie, British Columbia, and also on a common yellowthroat at Revelstoke, British Columbia. With these *I. muris* discoveries, we have added another species to the diversity of ticks in British Columbia. An *I. dentatus* larva on a Swainson's thrush collected on 5 May 2003 at Delta Marsh, Manitoba is a new record for this tick species on any host in Canada. Similarly, 2 *I. dentatus* larvae are reported on Swainson's thrush at Thunder Cape, Ontario, collected on 6 June 2003; they constitute the first record on an avian host in this province. To the south, Kollars and Oliver (2003) reported larval and nymphal *I. dentatus* on a Carolina wren, *Thryothorus ludovicianus* (Latham), in Missouri, which is located within the Mississippi migratory flyway. In the Midwest, *I. dentatus* larvae have bimodal activity starting in May, which corresponds to spring migration of passerines moving northward when this tick species manifests heightened questing activity. We provide the first record of the larval stage of the bird tick, *I. brunneus* in Canada, which was collected from a hermit thrush on Bon Portage Island, Nova Scotia. Earlier, *I. brunneus* females were reported on songbirds in Canada (Durden and Keirans, 1996; Scott et al., 2001). Further south on the Atlantic flyway, Durden et al. (2001) reported larval *I. brunneus* on a Carolina wren during the summer on St. Catherine's Island, Georgia.

Our study provides new evidence of migrating songbirds dispersing *B. burgdorferi*-infected *I. scapularis* in eastern and central Canada and of these ticks subsequently parasitizing terrestrial animals. *Borrelia burgdorferi*-positive *I. scapularis* have been collected in eastern Canadian provinces from humans and companion animals with only local recent travel (Banerjee et al., 2000; Morshed et al., 2003; Scott et al., 2004). Additionally, we identified *I. scapularis* adults collected in Manitoba and Saskatchewan from canine and feline hosts, which had no out-of-province travel; the most likely source is birds. Concomitantly, Lyme disease cases have been reported across Canada in humans and domestic animals that had no history of travel to endemic areas (Banerjee et al., 1994, 2000). Our findings demonstrate the potential for migratory birds to disseminate *B. burgdorferi*-positive ticks to nonendemic areas across Canada

and increase the risk of Lyme disease transmission regardless of whether sufficient ticks are present to establish a population.

Some host birds had weakness and fatal outcomes after being parasitized by attached *I. auritulus* ticks. One fledgling American robin had difficulty grasping and walking and died within 5 days after 20 *I. auritulus* (8 identified) were collected; *B. burgdorferi* was isolated from 1 of the fully engorged females. Similarly, another American robin died after a fully engorged *I. auritulus* was removed; however, all of the attached ticks were negative for *B. burgdorferi*. For global comparison, the seabird tick, *I. uriae*, which is also found on the West Coast of North America, has also been implicated in death by exsanguination from large tick burdens on juvenile birds (Chastel, 1988). In the upper Midwest, a tick–bird study revealed that a few of the passerine birds were heavily infested with ticks and experienced weakness, and 2 of the birds eventually died (Nicholls and Callister, 1996). *Ixodes auritulus*, the most commonly recorded avian tick from the Pacific Coast in this study, does not parasitize humans. Further ecological investigation between Lyme disease spirochetes, *I. auritulus* ticks and their hosts is needed.

Based on morphological and sequencing analyses of a nymphal specimen, we report *A. humerale* from Canada for the first time. This tick species is indigenous in Trinidad and northern South America, extending as far south as Bolivia; adults are ectoparasites of land tortoises. Although *A. humerale* nymphs have been reported from terrestrial vertebrates, this stage has not been formally described (Labruna et al., 2002; Robbins et al., 2003). Ecologically, the distribution of *A. humerale* in Colombia, Ecuador, and Venezuela overlaps the migratory flight path of the host bird, a gray-cheeked thrush, which has a winter range as far south as Peru (Godfrey, 1986). Ultimately, the flight distance from northwestern South America to Delta Marsh, Manitoba, for the gray-cheeked thrush would be at least 5,000 km. For the nymphal *A. humerale* to sustain such a long-range flight, it must be a slow-feeding tick. Combining slow engorgement and long flight, Neotropical migrants show the potential to transport tick-borne pathogens between continents. Scott et al. (2001) documented immature Neotropical *Amblyomma* spp. on spring migrants in Canada: *A. longirostre*, which primarily parasitizes tree porcupines from Brazil to Panama (Jones et al., 1972) and *A. sabanerae*, which, as adults, normally parasitizes turtles in Central America (Barnard and Durden, 2000). Because molecular data sets provide objective identification tools for ticks with diverging lineage and speciation events, we sequenced a recognized 344-bp fragment of mitochondrial 12S rDNA. *Rhipicephalus turanicus* Pomerantsev ticks collected in Greece, Zimbabwe, Israel, and France showed levels of sequence variability for this fragment of 1.5–7.7% (Beati and Keirans, 2001). Thus, the 3.8% difference between our *A. humerale* nymph and that of a confirmed, conspecific adult specimen is within an acceptable range. Furthermore, our nymphal specimen morphologically matched an *A. humerale* nymph from Trinidad.

In conclusion, passerine birds are implicated in local- and long-distance distribution of several tick species across Canada. Based on our avian–tick studies, *I. auritulus* is a candidate vector for *B. burgdorferi* for coastal British Columbia. The premiere discoveries of *B. burgdorferi* in *I. scapularis* collected from tick-infested birds in Manitoba and Ontario during spring migration expands tick–spirochete relationships for songbirds in Canada. The movement of *Amblyomma* spp. ticks on Neotropical migrants from South America to Canada shows strong potential for intercontinental transmission of tick-borne pathogens. *Borrelia burgdorferi*-positive ticks on songbirds provide clear evidence as an introductory source of infection and public health risk. Not only do passerine birds play a role in the enzootic cycle within endemic areas, but they also disseminate infected ticks to nonendemic areas across Canada.

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