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How far north are migrant birds transporting the tick *Ixodes scapularis* in Canada? Insights from stable hydrogen isotope analyses of feathers

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ABSTRACT

Lyme disease is emerging in Canada because of northward range expansion of the tick vector *Ixodes scapularis*. It is hypothesised that *I. scapularis* feeding on passerine birds migrating north in spring are important in founding new *I. scapularis* populations leading to northward range expansion. However, there are no studies on how far north *I. scapularis* may be carried, only inferences from passive tick surveillance. We used stable hydrogen isotope (δ^2H) analysis of rectrices collected from northward migrating, *I. scapularis*-carrying, passerine birds captured in Canada to estimate how far north *I. scapularis* may be carried. Rectrices are usually grown close to breeding sites and their δ^2H values reflect those in the environment, which vary strongly with latitude in North America. Passerines usually return to their breeding or natal sites so δ^2H values of rectrices of northward migrating birds can identify the likely latitudinal bands of their intended destinations. In 2006 we analysed δ^2H from rectrices of 73 *I. scapularis*-carrying birds captured at five migration monitoring stations, mainly from southern Ontario. Values of δ^2H ranged from −33 to −124‰, suggesting 19/71 (26.7%) birds were destined for latitude band B (the most southerly part of Ontario), 40/71 (56.3%) birds were destined for band C (which extends from southern Ontario, Quebec and the Maritimes to southern James Bay) and 12/71 (16.9%) birds were destined for bands D and E (which extend from northern Ontario and Quebec into the southern Canadian Arctic). This indicates that many *I. scapularis*-carrying migratory birds in spring have destinations far north in Canada, including some farther north than the current region of climatic suitability for *I. scapularis*. These findings support the hypothesis that *I. scapularis* may continue to be spread north by spring migrating passerines. Some thrush species may be particularly implicated in far northward dispersion of *I. scapularis*.

Introduction

Lyme disease is emerging in Canada due, in large part, to the northward spread of the tick *Ixodes scapularis*, which is the main vector in eastern and mid-western North America (Ogden et al., 2014). Ticks don't fly and can be dispersed only during the period they are attached to their animal hosts. *I. scapularis* ticks are host generalists, and feed on a wide range of vertebrates that can be involved in dispersion of the tick, as well as in maintenance of local tick populations. *I. scapularis* feed on birds, and numerous studies have identified ticks on birds both during the breeding season and on migration (e.g. Anderson et al., 1986; Weisbrod and Johnson, 1989; Battaly and Fish, 1993; Scott

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et al., 2012). In a previous study conducted in 2005 and 2006 (Ogden et al., 2008a), we identified that approximately 2% of northward migrating passerines at stopover locations in southeastern Canada carried immature *I. scapularis* ticks (mostly nymphs but also some larvae). Ticks were most common on ground-frequenting birds such as wrens, thrushes, some wood warblers and mimids. Although proportionally fewer ticks were present on species such as Red-winged Blackbirds (*Agelaius phoeniceus*) and sparrows, the large population sizes of many of these species mean they could still be important in dispersing *I. scapularis* during spring migration.

It has been hypothesised that migratory birds have been important in the northward range expansion of *I. scapularis* (Ogden et al., 2008b) and this was supported by analysis of surveillance data (Leighton et al., 2012). In passive tick surveillance involving voluntary submission of ticks collected from patients of participating medical and veterinary practitioners, ticks are not infrequently found long distances (>500 km) north of where *I. scapularis* populations are known to occur, further suggesting dispersion of ticks by migratory birds (Ogden et al., 2014). A factor favouring migratory birds in long-distance northward dispersion of *I. scapularis* into and across Canada is that during spring migration of many birds, nymphal *I. scapularis* are active in the two main regions of North America (the northeastern US and southeastern Canada, and the upper Midwestern US and south Central Canada) where this tick occurs (http://www.cdc.gov/lyme/stats/; Ogden et al., 2014). Nymphs may feed for four to five or more days on their hosts, so migrating passerines, which travel an average 85 km per day (Marra et al., 2005), could potentially disperse ticks 400 km or more northwards, assuming that they continue in that direction. We used this value to parameterise models of Lyme disease risk spread in the future (Ogden et al., 2008b).

A limitation of previous risk assessments (Ogden et al., 2008b), was the assumption that most birds keep migrating for several days after capture before reaching their breeding sites. However, if many of the tick-carrying birds were near their migration destination, the resultant risk maps may overestimate the rate of expansion of Lyme disease risk. Although some estimates are available of the densities of birds in the boreal forest (Cumming et al., 2010), the migration strategies and routes to different regions are not well known. As such, we cannot assume that the destinations of tick-carrying birds identified at Canadian banding stations close to the US border are distributed in a geographic pattern that is simply proportionate to the densities of breeding birds.

Stable isotope analysis (SIA) can help answer questions regarding migratory bird movements. The concentrations of some isotopes, particularly deuterium (δ^2H), in amount-weighted, growing-season average precipitation in North America (δ^2H_p), vary predictably geographically (Bowen et al., 2005). For most of North America east of the Rocky Mountains, δ^2H_p shows a strong latitudinal gradient with higher values in southern areas and progressively lower values toward the north. Birds acquire deuterium from the environment in the food and water they ingest, and this isotopic information is locked into metabolically inert keratin during feather formation (Wassenaar and Hobson, 2000; Hobson, 2005). The isotopic content (and in particular δ^2H content) of rectrices (tail feathers) can be used to define the breeding location of birds because, in most migratory bird species in North America, rectrices are moulted and regrow at the end of the breeding season, prior to, or shortly after, the start of southward migration (Pyle, 1997). On their return north during migration the following spring, the rectrices of these birds still carry the δ^2H content acquired from the location where they were grown. By comparing δ^2H values of rectrices (δ^2H_f) from birds identified as carrying ticks during spring migration in 2006 against predicted maps or "isoscapes" of δ^2H_f based on δ^2H_p isoscapes, we should be able to identify the broad latitude ranges within which the birds bred or were hatched in 2005 (Hobson, 2008; Hobson et al., 2014). It can be inferred that, since most adults of most species return to breed in the regions where they originated or bred the previous year (Greenwood and Harvey, 1982; Holmes and Sherry, 1992), δ^2H_f would reflect the approximate latitudinal destinations of birds carrying ticks captured in spring 2006.

Methods

Bird capture and examination

Eight eastern bird migration monitoring stations in the Canadian Migration Monitoring Network (CMMN; one in Nova Scotia, one in Quebec and six in Ontario: http://www.bsc-eoc.org/national/cmmn.html; Fig. 1) participated in a study of tick transport on birds (Ogden et al., 2008a). At each station, staff were asked to examine the head of as many captured birds as possible for the presence of ticks, using a binocular head loupe, during spring migration between March and June in 2006 (precise dates varied among stations). Staff removed all ticks observed using fine forceps, placed them in sterile tubes and despatched them to the National Microbiology Laboratory, Winnipeg, for identification using appropriate taxonomic keys as previously described (Ogden et al., 2008a). We also asked banding station staff to pluck one rectrix from each bird they identified as carrying a tick. In all cases, ID numbers were given to collected ticks and rectrices such that they could be cross-referenced with bird band numbers, ensuring that data on the birds from which they were obtained could be related to data on ticks and rectrices, which were sent to different laboratories. Birds were captured and rectrices removed under appropriate federal and provincial permits.

Stable isotope analysis

All feathers were cleaned of surface oils using a 2:1 chloroform: methanol solvent rinse and prepared for stable hydrogen isotope analysis at the Environment Canada stable isotope laboratory in Saskatoon. Stable-hydrogen isotope analyses of feathers were completed using the comparative equilibration method described in detail by Wassenaar and Hobson (2003) and through the use of calibrated keratin isotope reference materials. Stable hydrogen isotope measurements were performed on H_2 derived from high-temperature (1350 °C) flash pyrolysis of feathers using continuous-flow isotope-ratio mass spectrometry. All δ^2H results are expressed in the typical delta (δ) notation, in units of per-mil (‰), and normalized on the Vienna Standard Mean Ocean Water—Standard Light Antarctic Precipitation (VSMOW-SLAP) standard scale. Repeated analyses of hydrogen isotope inter-comparison material IAEA-CH-7 (−100‰) and keratin references yielded an external repeatability of ±2.0‰. Feather δ^2H values were compared to a δ^2H_f isoscape developed for migrant passerines by Hobson et al. (2014). The conversion of δ^2H_f to δ^2H_p for each species was based on the appropriate guild-specific algorithm as described in Hobson et al. (2014). In this process, the band of latitude corresponding to presumed destinations of birds carrying ticks was obtained by accounting for whether they birds migrate short-distances within North America, or are Neotropical migrants (those wintering at or south of 23°N), and for the latter, whether they are mostly ground or canopy feeding species (Hobson et al., 2014). This direct method of assignment to latitudinal bands was used, rather than more complex, spatially explicit Bayesian assignment depiction, because of the comparability of these methods and its adequacy for our study (Hobson et al., 2014).