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Predictors Of Babesia Microti Infection In Ixodes Scapularis Ticks In New England, Usa

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Introduction

Babesia microti is a tick-borne intraerythrocytic protozoan that is the primary etiological agent of human babesiosis [1]. The first well-defined human case of babesiosis in the United States was described in 1969 in a Massachusetts resident living on Nantucket Island [2]. Since the 1982 the number of human cases of babesiosis reported along with the geographic area in which cases were reported has expanded significantly to include mainland Massachusetts [3, 4], Connecticut [5], Maine [6], New Jersey [7], New York [8] and Rhode Island [9, 10]. The pathogen shares the same vector, *Ixodes scapularis* ticks, and the same primary reservoir, *Peromyscus leucopus* mice, as *Borrelia burgdorferi*, the etiological agent of Lyme disease. Both pathogens have followed a similar pattern of expansion, starting near coastal southeastern Connecticut and expanding northwards [5]. The geographic extent of *B. microti*, however, is limited to a subset of the geographic range of *B. burgdorferi* [11, 12] and the ratio of Lyme disease cases to babesiosis cases in the United States is around 25:1 [11]. Despite the slower spread of *B. microti* the pathogen is equally prevalent in ticks in certain areas where both *B. burgdorferi* and *B. microti* have been endemic for long periods, such as southeastern Connecticut [11]. The slower rate of babesiosis expansion as compared to Lyme disease has been attributed to a lower efficiency of transmission for *B. microti* [13]; however, this alone does not explain the similar prevalence of both microbes in areas long endemic to both pathogens.

It is possible that *B. microti* responds to similar ecological conditions as *B. burgdorferi*, such as the density of nymphal ticks (DON), host community composition, and climatic conditions, but with a temporal lag. Both babesiosis and Lyme cases are positively associated with increased forest fragmentation [14-17] and previous work has suggested that the DON is associated with the odds of zoonotic *B. microti* [9]. On the other hand, recent research suggests that co-infection of mice with *B. burgdorferi* improves the transmission efficiency *of B. microti* to ticks as compared to mice infected solely with *B. microti* [18]. Little research has been done to compare the risk factors for infection of *I. scapularis* nymphs by *B. burgdorferi* with the risk factors for infection by *B. microti*.

This study assesses the relative importance of ecological conditions and pathogen interactions in B. microti and B. burgdorferi prevalence in *I. scapularis* nymphs from a sample of 1514 nymph-stage ticks collected at 35 sites in eastern Connecticut, western Rhode Island, and southern Massachusetts. Variables considered included: infection status of the individual tick with the other pathogen, site level prevalence of *B. microti* and *B. burgdorferi*, the maximum density of *Ixodes scapularis* nymphs (DON) of the site during the sampling timeframe, and latitude and longitude. Our results show that the odds of a tick testing positive for *B. microti* is not associated to the density of nymphal ticks or the prevalence of *B. burgdorferi* at the site level, but it is associated with the presence of *B. burgdorferi* in the individual tick and the geographic location of the tick. None of the covariates tested showed a strong association with the odds of a tick testing positive for *B. burgdorferi*.

Materials and methods

Sample collection

A total of 64 sites were selected for sampling in eastern Connecticut, western Rhode Island, and southern Massachusetts (Figure 1). One state park was randomly selected among all parks in each of the towns. Babesiosis was first described in North Stonington, in the southeastern corner of Connecticut in 1989 and most southern towns in the study area became endemic for babesiosis in the early 1990s [5, 11]. Babesiosis progressively expanded northward and several towns remained free of babesiosis endemicity in 2010 [11]. All sites were sampled between one and five times from late May to September 2012, with at least one sample obtained during the nymphal season (late May/June).

Host-seeking *I. scapularis* nymphs at each site were collected via the dragging method [19]. Researchers dragged a $1m^2$ white corduroy cloth across the leaf litter of the forest floor in $100m^2$ transects. Every 20 meters any adults, nymphs, and larvae collected on the cloth were counted and placed in vials of 70% ethanol and latitude and longitude location information were recorded with reference to the WGS84 datum using handheld Garmin GPS devices. At each site at least 8 transects were completed on each date of sampling.

All ticks were identified by using a dissecting microscope and taxonomic keys [20]. DNA was extracted from all *I. scapularis* nymphs with QIAGEN DNeasy blood and tissue kit (QIAGEN Inc., Valencia, CA, USA) by using a modified protocol [21]. A real-time PCR was performed to amplify the 16S-23S rRNA intergenic spacer region of *B. burgdorferi* by using the primers and protocols developed by Liveris et al [22]. Amplicons were visualized on 1% agarose gel by using ethidium bromide. All positive samples were sequenced bi-directionally. *B. burgdorferi* amplicons were typed by comparing them with known genotypes by using BLAST [23]. Ticks were tested for *B. microti* infection by using a reverse transcription PCR that targets a sequence of the *B. microti* 18S rRNA gene (GenBank accession no. AY144696.1) [24].

For each site and visit, the density of nymphs (DON) was calculated by dividing the number of nymphs collected during each site visit by the total length (in meters) dragged during that visit. Information on the length dragged was calculated in ArcMap 10.1 (ESRI 2013. ArcGIS Desktop: Release 10.1. Redlands, CA: Environmental Systems Research Institute) using the location information collected by GPS receivers during each transect.

Data Analysis

Rather than selecting a single "best" fit model, multimodel inference (model averaging) was used to generate inferences about the relationship between the covariates and the dependent variable for each pathogen. Literature comparing the best-model selection strategy versus multimodel inference suggests that model averaging provides superior performance [25-28].

Using the R statistical language [29] with the lme4 package [30] and MuMIn package [31] two sets of logistic random effects models were constructed. One assessed the probability of a tick being infected with *B. microti*, and a second assessed the probability of a tick being infected with *B. burgdorferi*. The sample site was introduced as a random effect in order to account for autocorrelation of ticks collected within the same site. A series of nested models were constructed by adding fixed effects and comparing the AICc and BIC value of each model [27, 32].. An all-subsets approach to model selection was used where all possible combinations of covariates were evaluated. The AICc value for each parameter combination (model i) was defined as $AICc_i = -2\log L_i + 2V_i$, where $L_i$is the maximum likelihood for model i and V_i are the free parameters of model i [33]. The BIC value for each model i was defined as $BIC_i = -2logL_i + V_i\log(n)$ here n is the number of observations [34]. The suitability of using AICc for model selection with mixed-effects models has not been well tested [32] and AICc methods have been suggested to select more complex models than BIC [35]. We use both AICc and BIC methods for multi-model inference and report where the methods disagree

The AICc and BIC weights for model i (w_i) for each pathogen were calculated, where w_i can be interpreted as the probability that model i is the best model given the data and the set of model candidates [26, 35]. A 95% confidence set of models was created by for each pathogen by ordering the models by w_i and summing the weights until the cumulative weight exceeded 0.95. All models with a cumulative weight above 0.95 were rejected, with the remaining models considered to be the 95% confidence set of models [32]. Model averaging using the 'natural averaging' approach was employed to produce parameter and error estimates for each covariate [25, 27]. The model averaged parameter estimates ($\hat{\hat{\beta}}$) were calculated as follows:

$$\hat{\hat{\beta}} = \frac{\sum_{i=1}^{R} w_i \hat{\beta}_i}{\sum_{i=1}^{R} w_i}$$

where $\widehat{\beta_i}$ is the parameter estimate for model i, and w_i is the model weight [32]. The importance of each covariate t (I_t) was assessed by summing the weights over all the models in which each covariate was found [26]. The value of I_t can be interpreted as the probability that the covariate t is a component of the best model [32].

Sites were excluded from analysis if the total number of ticks collected at the site fell below a certain threshold. This threshold level was varied from 5, 10, 15, 20, and 30 and the model selection was carried out on each resulting dataset.

Moran's I was used to test for spatial autocorrelation between the mean deviance residuals of the best model for each pathogen at each site using R and the "ape" package [36].

Results

The prevalence of both pathogens across all sites ($n = 35$) at which at least 20 ticks were collected is shown in Figure 1. *B. burgdorferi* was present at all 35 sites, while *B. microti* was present at 28 sites. Site level prevalence of *B. burgdorferi* ranged from 0.06 to 0.43 ($median = 0.23,\ mean = 0.24 \pm 0.19$). Site level prevalence of *B. microti* ranged from 0.00 to 0.30 ($median = 0.09, mean = 0.09 \pm 0.14$). Site level coinfection prevalence ranged from 0.00 to 0.20 ($median = 0.04, mean = 0.05 \pm 0.10$). Among all ticks, more were coinfected than were infected with only *B.* microti (Table 1). The prevalence of *B. burgdorferi* was greater than that of *B. microti* in all sites except one, James Goodwin State Forest in northern CT, where the prevalence of *B. microti* was greater than that of *B. burgdorferi* (0.19 versus 0.15). For all 1514 ticks included in the analysis in the study region the ratio of *B. burgdorferi* infection to *B. microti* was 2.5:1.

The 95% confidence set of models to describe the presence or absence of *B. microti* at the tick level using either AICc (Table 2) or BIC (Table3) criteria do not present a clear "best" model. The model-averaged odds ratio showed that if a tick is infected with *B. burgdorferi* the tick is at 4.96 (95% CI: $3.34 - 7.36$) higher odds of being infected with *B. microti* as compared to an uninfected tick using AICc model averaging (Table 4). Model averaging using BIC produced similar results with an odds ratio of 5.03 (95% CI: $3.44 - 7.35$) (Table 5). Location was a significant predictor of tick infection status($OR = 0.89, 95\%\ CI: 0.82 - 0.96$) for AICc and ($OR = 0.89, 95\%\ CI: 0.82 - 0.98$)for BIC. Thus for every 10 km travel to the north, the odds of a tick testing positive for *B. microti* were 0.89 that of a tick 10 km further south. Site level prevalence of *B. burgdorferi* had a 0.57 and 0.06 probability of being in the best model according to AICc and BIC criteria respectively, while DON had a 0.35 and 0.00 probability.

The 95% confidence set of models to describe the presence or absence of *B. burgdorferi* indicate that the model with no covariates significantly outperforms any models with any of the potential covariates (Table 6, Table 7). The model-averaged odds ratios, 95% unconditional confidence intervals and the importance values (I_t) for the covariates confirm the low probability of any of the candidate covariates of appearing in the best model (Table 8, Table 9).

Using different thresholds for minimum number of total ticks collected did not significantly influenced the odds ratios for the covariates (Figure 2).Testing for spatial autocorrelation between the deviance residuals using Moran's I revealed no significant autocorrelation ($p = 0.89$).

Discussion

We investigated the extent to which the infection status of a tick with *B. burgdorferi*, DON, *B. burgdorferi* prevalence at the site and geographic location influence the odds of a tick testing positive for *B. microti*. We found that the *B. burgdorferi* infection status of the tick and its geographic location are the most important predictors of *B. microti* status in a tick, while nymphal infection prevalence and the DON at the site level had a reduced effect. In contrast and consistent with the endemic status of *B. burgdorferi*, none of the covariates we examined proved important in predicting the odds of a tick being infected with *B. burgdorferi*. Our results are consistent with previous laboratory experiments showing that host coinfection with *B. burgdorferi* and *B. microti* increases the odds of *B. microti* tick infection and may enhance the probability of *B. microti* invasion into *B. burgdorferi* endemic areas [18].

The odds of a tick testing positive for *B. microti* was most strongly influenced by the *B. burgdorferi* status of the individual tick and, to a much lesser extent, by the prevalence of *B. burgdorferi* in the tick population at the site. Nymphal infection prevalence of both pathogens is determined by dynamic interactions between the reservoir competence of the host community and other ecological determinants of transmission from the previous year, when these nymphs fed as larvae on all vertebrate tick hosts. While these ecological conditions may similarly influence the prevalence of both *B. burgdorferi* and *B. microti* at the site level, nymph coinfection can only result from larvae feeding on coinfected hosts. The increased odds of tick infection with *B. microti* when the tick is infected with *B. burgdorferi* may reflect one of two non-mutually exlusive effects: either a reduced host range of *B. microti*, such that only highly competent hosts for *B. burgdorferi* (such as *Peromyscus leucopus*) also transmit *B. microti*, or that there is enhanced transmission of *B. microti* transmission from coinfected hosts. Hersch et al. (2012) found a wide range of competent hosts for *B. microti*, similar to that of *B. burgdorferi*, rendering the first argument unlikely to explain the strong observed effect [37]. Observed enhancement of *B. microti* transmission from experimental coinfections [18] supports the premise that pathogen interactions may explain the observed increased odds. Conversely, the same experiment showed no reciprocal enhancement of *B. burgdorferi* transmission due to coinfection with *B. microti*. Consistent with the lack of enhancement due to *B. microti* infection we find that the *B. microti* status of a tick was not a strong predictor of the odds of a tick being infected with *B. burgdorferi*.

The site-level prevalence of *B. burgdorferi* is shown to have a marginal influence on the odds of a tick being infected with *B. microti*, with a 0.57 and 0.06 probability of being in the best model according to AICc and BIC criteria, respectively. An enhancement effect at the tick level should translate into higher odds of *B. microti* in areas with a greater prevalence of *B. burgdorferi*. The weak association we found was unexpected and warrants further investigation.

The spatial trend in *B. microti* infection is consistent with a pattern of ongoing emergence from southeastern portion of the state where the first case of babesiosis was reported in 1989 [5, 38]. The northerly and westerly expansion direction parallels the observed expansion in human cases [11]. The lack of spatial patterns in *B. burgdorferi* infection is consistent with its longer invasion history [11, 39].

Our findings that the DON does not influence the odds of tick infection with *B. microti* or *B. burgdorferi* in a tick is consistent with a sensitivity analysis for a global model of *I. scapularis* pathogen establishment that showed establishment was possible over a broad range of the number of ticks on hosts [40]. However, our results are counter to previous research indicating that the DON was associated with the odds of *B. microti* infection in *Peromyscus leucopus* hosts in Rhode Island in 1996 [9]. The lack of importance of DON may be due to the later stage of the invasion process of *B. microti* in this study compared to [9]. All but four sites of 32 we studied had *B. microti* infections, while Mather et al. (1996) found *B. microti* infected mice in only 8 out of 34 sites in 1996. Furthermore, Mather found that the positive sites were clustered in the coastal or near coastal regions of south-central Rhode Island [9]. This indicates that *B. microti* invasion may be more sensitive to tick density earlier in the emergence process. Since we excluded from the analysis sites with fewer than 20 ticks in order to improve the statistical power of our estimates of prevalence for each pathogens, it is possible that we biased our analysis by dropping sites with low DON. To test this, we performed the analysis while varying the minimum tick threshold between 5, 10, 15, 20, and 30 ticks. The lack of importance of DON remains for all thresholds tested (Figure 2).

The synergistic relationship that *B. microti* has with *B. burgdorferi* has been shown to vary by genotype of *B.* burgdorferi [13, 18]. Future work to genotype the *B. burgdorferi* samples derived from the infected ticks in this study would be productive in further investigating how the effect of coinfection varies across genotypes.

Although larval counts were collected during each dragging visit, the data was considered of too poor quality to include in the analysis. The overlap of host-seeking spring larvae and nymphs has been shown to be an important modifying factor in the effect that coinfection has on the transmission of *B. microti* [18]. Controlling for variations in the overlap of spring larvae is an important future direction for research.

Other ecological covariates, such as climate and land-use data were not included in the study since the focus was on the interaction between the two pathogens. Further, the study area was relatively small and has generally homogenous climatic conditions, with a possible exception of areas immediately proximate to the coastline.

Given that *B. microti* currently occupies only a subset of the geographic range of *B. burgdorferi* [11, 39], there is significant potential for *B. microti* expansion and more frequent interaction between the two pathogens. Our study area includes regions in an advanced state of *B. microti* emergence; it is expected that the relationship between *B. microti* and *B. burgdorferi* will be more pronounced in areas with newly emerging *B. microti,* where the zoonotic presence of *B. burgdorferi* could play a larger role in reducing the barriers for *B. microti* establishment [18].

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Figure Legends

FIGURE 1. Field data collection sites. Ticks were sampled from 64 sites from May to September 2012. Site at which at least 20 nymphal ticks were collected were included in the analysis and have pathogen prevalence information presented. All other sites are represented by a circle only.

TABLE 1. 2x2 contingency table showing the proportion of ticks infected with *B. microti*, *B. burgdorferi*, both pathogens, and neither pathogen.

TABLE 2. The 95% confidence set of models predicted *B. microti* infection in a tick using AICc selection criteria. Each row indicates the point estimates of every covariate included in that model. Black cells indicate covariates not included in that model.

TABLE 3. The 95% confidence set of models predicted *B. microti* infection in a tick using BIC selection criteria. Each row indicates the point estimates of every covariate included in that model. Black cells indicate covariates not included in that model.

TABLE 4. The model-averaged odds ratios, standard errors, 95% confidence intervals, and parameter weights for the 95% confidence set of *B. microti* models selected using AICc.

TABLE 5. The model-averaged odds ratios, standard errors, 95% confidence intervals, and parameter weights for the 95% confidence set of *B. microti* models selected using BIC.

TABLE 6. The 95% confidence set of models predicted *B. burgdorferi* infection in a tick using AICc selection criteria. Each row indicates the point estimates of every covariate included in that model. Black cells indicate covariates not included in that model.

TABLE 7. The 95% confidence set of models predicted *B. burgdorferi* infection in a tick using BIC selection criteria. Each row indicates the point estimates of every covariate included in that model. Black cells indicate covariates not included in that model.

TABLE 8. The model-averaged odds ratios, standard errors, 95% confidence intervals, and parameter weights for the 95% confidence set of *B. burgdorferi* models selected using AICc criteria.

TABLE 9. The model-averaged odds ratios, standard errors, 95% confidence intervals, and parameter weights for the 95% confidence set of *B. burgdorferi* models selected using BIC criteria.

FIGURE 2. Sensitivity to minimum tick threshold. Plots of the odds ratios and 95% confidence intervals of selected covariates when the analysis was performed using differing thresholds for the minimum number of nymphs needed at a site for inclusion in the analysis. Thresholds tested included 5, 10, 15, 20, and 30 nymphs.

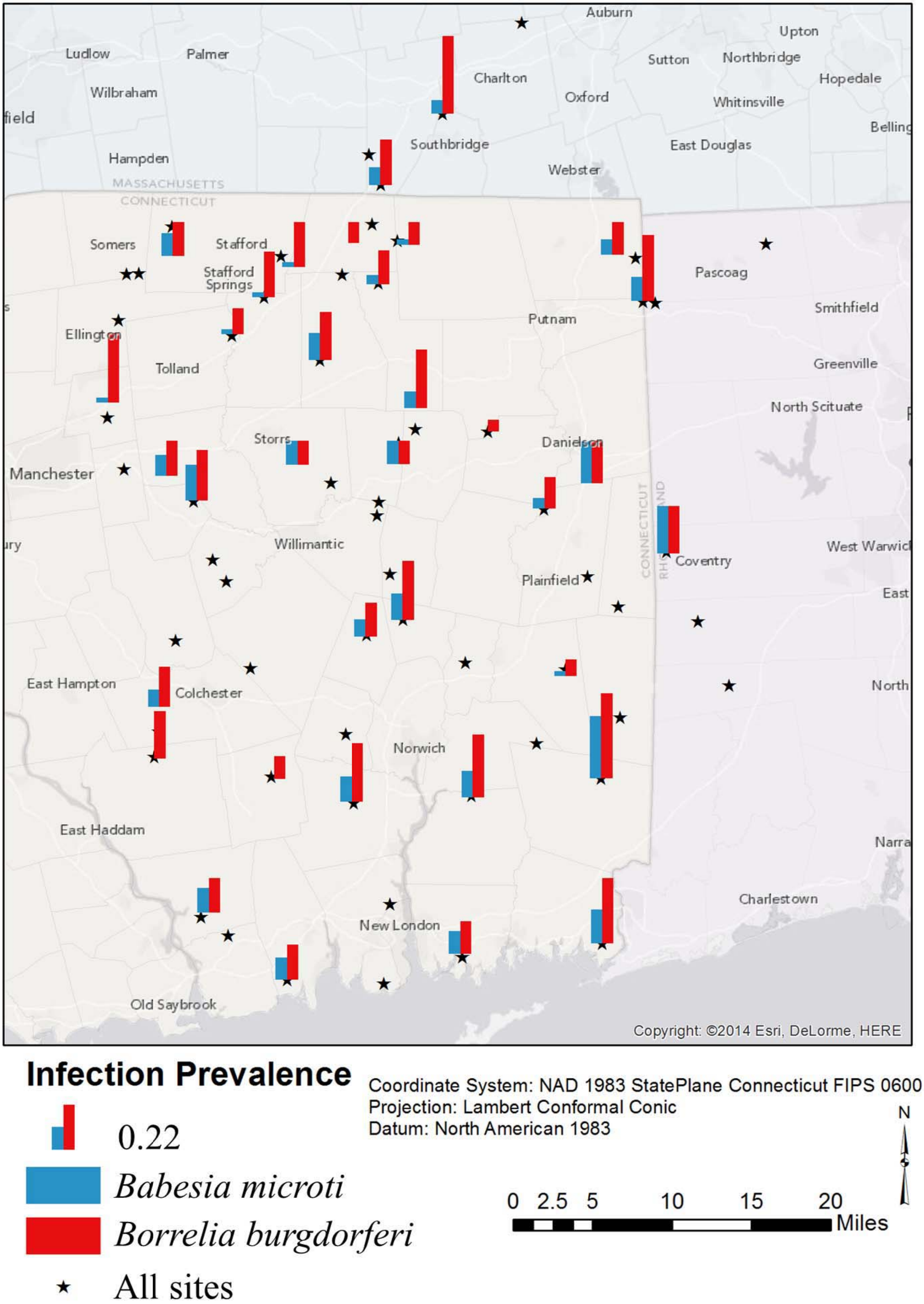

Auburn
Upton
Ludlow
Palmer
Sutton
Northbridge
Charlton
Hopedale
Wilbraham
Oxford
Whitinsville
field
Belling
Southbridge
East Douglas
Hampden
Webster
MASSACHUSETTS
CONNECTICUT
Somers
Stafford
Stafford Springs
Pascoag
Smithfield
Ellington
Putnam
Tolland
Greenville
North Scituate
Storrs
Danielson
Manchester
CONNECTICUT
RHODE ISLAND
Willimantic
Coventry
West Warwick
Plainfield
East
East Hampton
Colchester
North
Norwich
East Haddam
Narra
Charlestown
New London
Old Saybrook
Copyright: ©2014 Esri, DeLorme, HERE
Infection Prevalence
0.22
Babesia microti
Borrelia burgdorferi
All sites
Coordinate System: NAD 1983 StatePlane Connecticut FIPS 0600
Projection: Lambert Conformal Conic
Datum: North American 1983
N
0 2.5 5 10 15 20
Miles

Table 1

	B. microti -	*B. microti +*	Totals
B. burgdorferi -	1098	63	1161
B. burgdorferi +	275	78	353
Totals	1373	141	1514

Table 2

(Intercept)	Bb site prevalence	Bb tick status	X cordinate	Y cordinate	DON	AICc	ΔAICc	weight	Cu
-5.51	0.18	1.58	0.14	-0.12	[redacted]	855.40	0.00	0.24	
-6.47	[redacted]	1.65	0.18	-0.12	[redacted]	855.62	0.22	0.22	
-5.98	0.18	1.57	0.16	-0.12	-0.05	856.22	0.81	0.16	
-7.07	[redacted]	1.66	0.20	-0.12	-0.05	856.57	1.16	0.13	
-0.66	0.26	1.54	[redacted]	-0.12	[redacted]	857.47	2.07	0.09	
-0.55	0.27	1.54	[redacted]	-0.12	-0.02	859.30	3.90	0.03	
-0.16	[redacted]	1.63	[redacted]	-0.11	[redacted]	859.73	4.32	0.03	
-9.34	[redacted]	1.63	0.18	[redacted]	[redacted]	860.04	4.63	0.02	
-8.64	0.17	1.57	0.15	[redacted]	[redacted]	860.51	5.10	0.02	
-9.85	[redacted]	1.64	0.20	[redacted]	-0.05	861.44	6.03	0.01	

Table 3

(Intercept)	Bb site prevalence	Bb tick status	X coordinate	Y coordinate	DON	BIC	ΔBIC	weight	Cum. Weight
-2.99		1.61				889.30	0.00	0.49	0.4
-0.16		1.63		-0.11		891.60	2.30	0.16	0.6
-9.34		1.63	0.18			891.92	2.62	0.13	0.7
-6.47		1.65	0.18	-0.12		892.80	3.50	0.09	0.8
-3.58	0.25	1.55				893.52	4.22	0.06	0.9
-0.66	0.26	1.54		-0.12		894.65	5.35	0.03	0.9

Table 4

Covariate	OR Estimate	Lower CI	Upper CI	Importance
(Intercept)	0.00	0.00	2.73	NA
B. burgd. tick status	4.96	3.34	7.36	1.00
Latitude	0.89	0.82	0.96	0.95
Longitude	1.18	1.03	1.36	0.84
B. burgd. site prevalence	1.22	0.96	1.54	0.57
DON	0.95	0.86	1.05	0.35

Table 5

Covariate	Estimate	SE	Lower CI	Upper CI	Importance
(Intercept)	0.02	22.47	0.00	10.10	NA
Bb tick status	5.03	1.21	3.44	7.35	1.00
Latitude	0.89	1.05	0.82	0.98	0.26
Longitude	1.19	1.08	1.03	1.38	0.24
Bb site prevalence	1.29	1.15	0.98	1.69	0.06

Table 6

(Intercept)	Bm tick status	Bm site prevalence	X coordinate	Y coordinate	DON	AICc	ΔAICc	weight	Cu
-0.66						88.72	0.00	0.21	
-0.75	0.84					90.59	1.86	0.08	
-0.83		0.19				90.70	1.98	0.08	
-0.74					0.03	90.73	2.01	0.08	
-1.41			0.02			90.73	2.01	0.08	
-0.87				0.01		90.73	2.01	0.08	
-0.83	0.78	0.10				92.59	3.87	0.03	
-0.83	0.84				0.03	92.60	3.87	0.03	
-1.14	0.84			0.02		92.60	3.88	0.03	
-0.90	0.83		0.00			92.60	3.88	0.03	
-1.47		0.21		0.02		92.71	3.99	0.03	
-0.89		0.19			0.02	92.71	3.99	0.03	
-0.31		0.20	-0.02			92.71	3.99	0.03	
-0.97				0.01	0.03	92.74	4.02	0.03	
-1.22			0.01		0.02	92.74	4.02	0.03	
-1.61			0.02	0.01		92.75	4.03	0.03	
-1.46	0.78	0.12		0.02		94.60	5.88	0.01	
-0.91	0.78	0.09			0.03	94.61	5.88	0.01	
-0.34	0.78	0.11	-0.01			94.61	5.89	0.01	
-1.25	0.85			0.02	0.03	94.61	5.89	0.01	
-0.64	0.84		-0.01		0.03	94.61	5.89	0.01	
-1.27	0.84		0.00	0.02		94.62	5.90	0.01	
-1.56		0.21		0.03	0.02	94.72	6.00	0.01	
-0.79		0.24	-0.02	0.03		94.72	6.00	0.01	

Table 7

(Intercept)	Bm tick statu	Bm site prevalence	X coordinate	Y coordinate	DON	BIC	ΔBIC	Weight	Cum. Weight	c
-0.66						109.98	0.00	0.88	0.88	4.
-0.75	0.84					117.16	7.17	0.02	0.90	5.
-0.83		0.19				117.27	7.29	0.02	0.93	5.
-0.74					0.03	117.30	7.32	0.02	0.95	5.
-1.41			0.02			117.30	7.32	0.02	0.97	5.

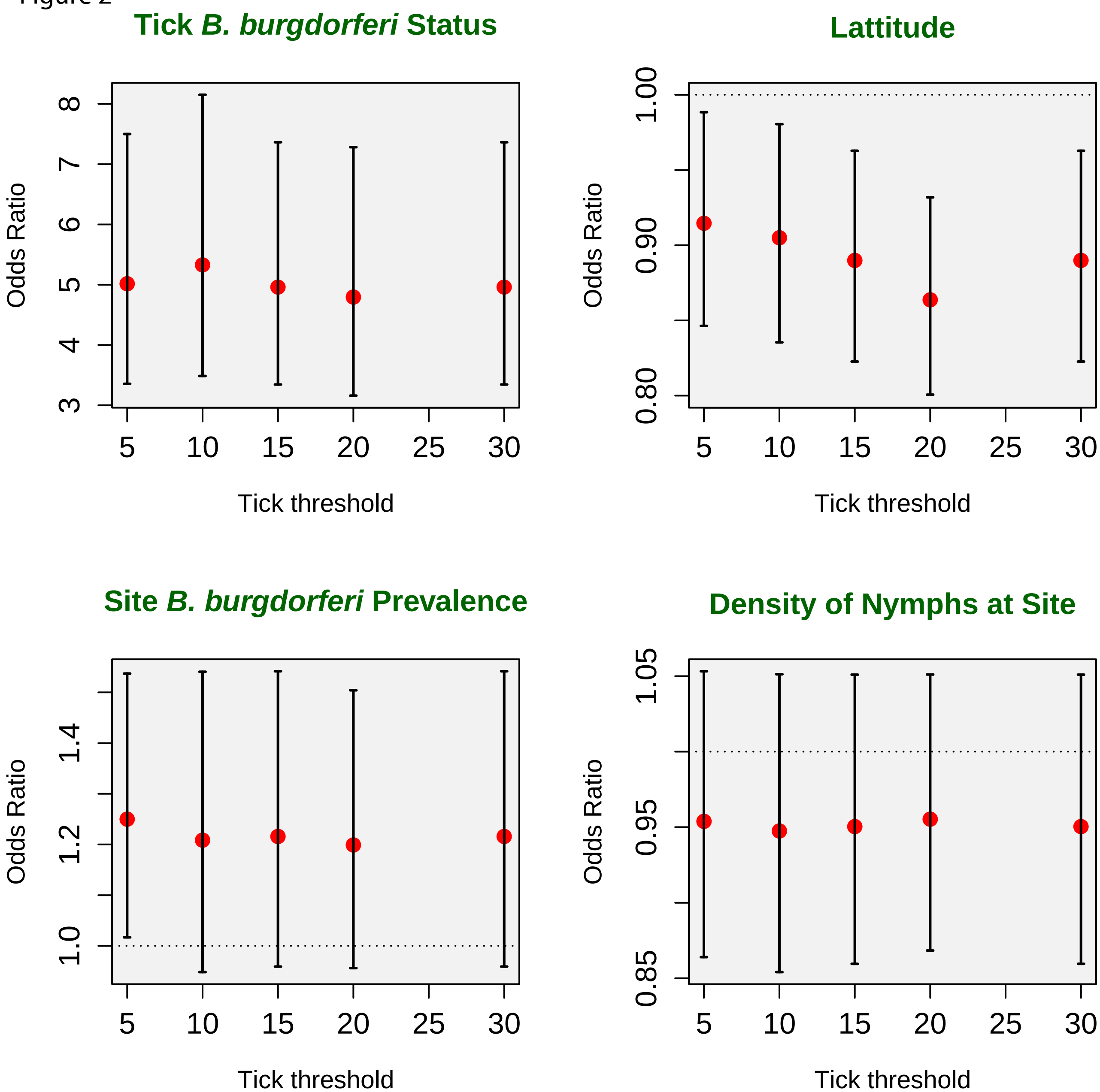
Table 8

Covariates	Estimate	SE	Lower CI	Upper CI	Importance
(Intercept)	0.38	1574965365.74	0.00	403132443973728000.00	NA
Bm tick status	2.28	132.07	0.00	32727.95	0.28
Bm site prevalence	1.19	9.85	0.01	105.15	0.25
DON	1.03	2.25	0.21	5.03	0.24
Y coordinate	1.01	1.85	0.30	3.40	0.24
X coordinate	1.01	2.70	0.14	7.07	0.23

Table 9

Covariates	Estimate	SE	Lower CI	Upper CI	Importance
(Intercept)	0.46	14.96	0.00	92.78	NA
Bm tick status	2.31	126.34	0.00	30328.51	0.03
Bm site prevale	1.21	9.14	0.02	92.53	0.02
DON	1.03	2.24	0.21	4.98	0.02

Figure 2


Tick B. burgdorferi Status
Odds Ratio
3
4
5
6
7
8
5
10
15
20
25
30
Tick threshold
Lattitude
Odds Ratio
0.80
0.90
1.00
5
10
15
20
25
30
Tick threshold
Site B. burgdorferi Prevalence
Odds Ratio
1.0
1.2
1.4
5
10
15
20
25
30
Tick threshold
Density of Nymphs at Site
Odds Ratio
0.85
0.95
1.05
5
10
15
20
25
30
Tick threshold