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Human Behavioral and Ecological Risk Factors for Lyme Disease Infection on Block Island, Rhode Island

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Abstract

Peridomestic exposure to infected *Ixodes scapularis* nymphs is considered the dominant means of infection with tick-borne pathogens in the eastern United States. Previous studies of risk of developing tick-borne infection established a positive association between the density of infected nymphs and Lyme disease cases at the population level. Studies examining the effectiveness of personal protective behaviors have not included measures of tick exposure. This study simultaneously assesses the effect of tick exposure and human behavior in Lyme disease infection risk using a longitudinal serosurvey study on Block Island, RI. Tick exposure risk at all Island properties was estimated by identifying remotely-sensed landscape proxies that most strongly correlated with tick density at the individual property level. Landscape metrics associated with lawn and shrub edge, patch density, percent land, class area, and the number of patches were found to be most associated with positive serology. Human behavior related risk factors included the average number of hours spent daily outside in tick habitat, and owning a cat that spends time both indoors and outdoors. Age at the time of test was also found to increase risk. Wearing protective clothing during outdoor exposure was protective. A multivariate model including peridomestic shrub patch density (decreased risk), wearing protective clothing (decreased risk), and owning a cat (increased risk) was determined to be the best model based on the lowest Akaike Information Criterion. Our findings emphasize that both environmental risk and human behavior contribute significantly to risk of tick-borne infection. They highlight the importance of accounting for environmental exposure to accurately ascertain the effectiveness of personal protective behaviors. A better understanding of the relative roles of environmental and behavioral risk factors in driving infection with tick-borne pathogens should guide future intervention studies to reduce the risk of these infections.

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Introduction

Lyme disease, caused by the spirochete *Borrelia burgdorferi,* is the most commonly reported vector-borne disease in the US, with greater than 20,000 cases reported annually (Bacon et al. 2008). The black-legged tick, *Ixodes scapularis,* serves as the principal vector in transmission to humans and is responsible for maintenance of the spirochete in natural reservoirs. Human risk of infection with tick-borne pathogens is determined primarily by the interaction between environmental or 'acarological' risk (density of *I. scapularis* nymphs that are actively host-seeking) (Kitron and Kazmierczak 1997, Stafford et al. 1998, Falco et al. 1999, Diuk-Wasser et al. 2012), peridomestic landscape features associated with human exposure (Falco and Fish 1988, Klein et al. 1996, Dister et al. 1997), and use of personal protective measures (Vazquez et al. 2008a, Connally et al. 2009a).

Since the Lyme disease vaccine was removed from the market in 2002, (Malouin et al. 2003, Nigrovic and Thompson 2007), strategies to reduce the number of human cases of Lyme disease have focused on ways to minimize contact between humans and infected ticks, either by reducing acarological risk or by modifying human behaviors to reduce the number of tick bites (Hayes and Piesman 2003b, Daltroy et al. 2007, Gould et al. 2008, Vazquez et al. 2008a, Connally et al. 2009a). Area-wide acaricides can be highly effective in controlling tick populations (Schulze et al. 1991, Stafford 1991, Curran et al. 1993, Hayes and Piesman 2003a), but a majority of residents in hyperendemic areas are not willing to use them on their properties due to safety and environmental concerns (Piesman 2006). Methods to reduce the number of ticks on deer ('four-poster', Brei et al. 2009, Hoen et al. 2009, Pound et al. 2009a, Pound et al. 2009b) and small mammals ('bait boxes',Dolan et al. 2004) are not in widespread use. Reduction in white-tailed deer (*Odocoileus virginianus*) populations has been linked to reduced *I. scapularis* abundance in fencing studies (Daniels et al. 1993, Daniels and Fish 1995, Hayes and Piesman 2003a) and in an insular system (Rand et al. 2004) and to decreased incidence of Lyme disease in humans in one study (Wilson 1988) but not in others (Jordan et al. 2007). Given the limitations of these control measures in decreasing acarological risk, modifying human behavior has been proposed as an essential component of any effort to lower the incidence of tick-borne diseases. Personal protective measures against tick bites, such as tick checks following outdoor activity, wearing of protective clothing, application of insect repellents, and avoiding tick habitat have been widely discussed and there is some evidence supporting their effectiveness (Poland 2001, Hayes and Piesman 2003b), mostly in peridomestic settings (Vazquez et al. 2008b, Connally et al. 2009b).

Ascertaining the effectiveness of these methods has been limited, however, by the absence of peridomestic exposure estimates in studies of Lyme disease risk behaviors (Vazquez et al. 2008b, Connally et al. 2009b). Protective behaviors may appear ineffective if residents of high risk properties are more likely to perform them. Interactions between acarological risk and human protective behaviors may also partially explain differences in the magnitude and direction of association between acarological risk and Lyme disease incidence in aggregate population studies (Brownstein et al. 2005, Pepin et al. 2012).

To ascertain the relative roles that acarological risk and individual behavior contribute to the risk of developing Lyme disease, we retrospectively assessed environmental exposure and individual behaviors in participants of a biannual survey conducted on Block Island between 2005 and 2011 (Krause 2003). Exposure risk at all Island properties was estimated through landscape proxies for tick density derived from very high resolution remotely sensed data (Figure 1). Additionally, tick populations were sampled in 2012 by determining the density of nymphs (DON) on the properties of study participants. The relative role of environmental and behavioral factors in driving infection with tick-borne pathogens can guide future intervention studies to reduce human infection with tick-borne diseases.

Methods

Study site

Block Island is a 25.2 km^2 landmass located in Washington County, Rhode Island, 23 km south of mainland RI (Rosenzweig et al. 2000). The year-round population is around 1000, which increases during the summer months to approximately 12,000 (BICC 2012). Because the dominant mainland habitat type for *Ixodes scapularis* - deciduous forest, (Enser 2002) is limited on the island to a 4 ha site (Enser 2000), one of the objectives of this study was to evaluate the suitability of brushland and other land cover types as non-traditional habitats for *I. scapularis* ticks.

Study Cohort

A study cohort was established in 1991 on Block Island, RI, by inviting all Island residents to take part in biannual serological surveys. The study cohort was restricted to residents who spent more than one month on the island during the

Lyme disease transmission period (May through October) and did not have a history of Lyme disease (Krause et al. 2003, Krause et al. 2006). The serosurvey was announced by placing notices in the local newspaper, over a cable television network, and at the Block Island Medical Center (Krause et al. 2003, Krause et al. 2006). All subjects were asked to provide blood samples for serological and PCR analysis and to complete a questionnaire (Supplementary Figure 1) which assessed the history of the residents' tick-borne illnesses, peridomestic factors potentially linked to tick exposure, and the participants' protective behaviors and outdoor activities. Written informed consent was obtained from all study participants in accordance with the human investigation committees at the University of Connecticut School of Medicine and the Yale School of Public Health.

For this study, we used a subset of the original cohort composed of all subjects who participated in serosurveys from 2005 to 2011. We restricted our analysis to this subset because our measure of exposure risk – backyard landscape proxies for acarological risk, was assessed retrospectively and assumed to be constant during that period. Once a participant had a laboratory-confirmed positive test for diagnosis, they were dropped from further analysis to control for potential confounding from behavioral changes arising from the diagnosis.

Serological Exposure Assessment

Serological evidence of exposure to *B. burgdorferi* was detected by ELISA and confirmed by Western blotting using standardized procedures (Krause et al. 2006). A positive ELISA result consisted of an IgM or IgG response at a dilution of 1:320 or more. Positive or equivocal ELISA results were confirmed by Western blotting. Specimens were considered positive if 5 or more bands of the ten most prevalent *B. burgdorferi*-specific bands were present in the immunoblot (CDC 1995, Krause et al. 2006, Skogman et al. 2010). All antibody assays prior to the fall of 2008 were carried out at the University of Connecticut Health Center. Assays from the spring survey of 2009 until the fall of 2012 were performed by commercial laboratories in New England using standard Lyme serodiagnostic assays. Positive serology in the fall was considered evidence of exposure during the summer, while positive spring serology was considered evidence of infection during the summer of the previous year.

Identification of Landscape Metrics to Use as Proxies for Peridomestic Exposure to I. scapularis Nymphs

Class and landscape metrics for island properties were previously calculated for all properties on Block Island (Salim et al. 2011). A land use classification was performed using a Worldview 2 satellite image. Landscape metrics were calculated using Fragstats software for all properties, including metrics such as lawn and shrub class area, patch density, total edge, and percent land. An example is provided in Figure 1.

During the 2012 season, 135 study participants granted permission to have their yards surveyed to determine DON. Of the 135 participants, 76 attended the serosurvey with another person who lived at the same residence. A total of 105 properties were visited for tick collection from May 15th to August 23rd. Eight of the properties surveyed were not listed as the primary residence of the participant, and were either vacant or rented out to tourists or visiting family members. In addition to the residential properties, three natural areas owned by The Nature Conservancy were surveyed. The property survey consisted of dragging $1m^2$ corduroy cloths along the edge of the property, typically at the edge of the lawn and shrubland vegetation as outlined in previous studies (Tellenklint-Eisen 2000,Schulze et al. 1997, Daniels et al. 2000) . Between 2 and 5 transects of approximately 100 meters in length were dragged, depending on the size of the property. *I. scapularis* nymphs were the focus of tick collection, as previous studies have implicated their role in disease transmission to humans (Steere et al. 1978). Attached ticks were counted and placed in 70% ethanol for species confirmation in the laboratory. Flagging was used to collect ticks inaccessible by the dragging method. Species were confirmed using taxonomy and identification keys (Durden et al. 1996).

Risk Factors for Human Lyme Disease Exposure:

STATA version 12.0 was used for statistical analyses. Negative binomial regression (nbreg procedure) was used to assess the association between landscape metrics and the peridomestic density of *I. scapularis* nymphs. Logistic regression was used to assess the effect of property landscape metrics and self-reported behavior from the serosurvey questionnaire on the individual's serology. Models were built from variables which were significant at either $p<.05$ or demonstrated a linear relationship with serology. 2- and 3-variable combinations of the findings were then run in multivariate regressions. Mixed models were also assessed but provided virtually identical models, so the simpler logistic regression models were reported. All models were assessed and compared by the Akaike Information Criterion corrected for small sample sizes (AICc) (Akaike 1974, Hurvich and Tsai 1989, Burnham and Anderson 2004). The level of significance was $p<0.05$.

Results:

Of 611 patients participating in at least one serosurvey between 2005 and 2011, both blood samples and completed questionnaires were available from 520. There were 1132 records available from the 520 participants; however 136 were dropped after positive diagnosis. The seropositivity rate from all blood samples was 10.7% (107/996). The use of any form of tick protection was reported by 72.6% of the participants filling out a questionnaire (724/996); routine tick checks were the most commonly used protective measure, while use of repellent was practiced the least (Table 1). The average age of the participants during testing was 61.5.

In univariate regression analyses, wearing protective clothing (OR 0.508, p=0.004), owning a cat (OR=1.623, p=0.033), and the average number of hours spent in tick habitat daily (OR=1.349, p=0.04) significantly modified exposure risk (Table 2). There was also an increase of risk seen with increasing participant age at the time of the test (OR=1.019, p=0.011), although the increased risk was relatively low. Using repellent, avoiding brush, performing tick checks, using at least one protective measure including the three previously mentioned protective measures, and occupational exposures were not found to be associated with disease risk.

Similarly, univariate regressions were performed on all of the previously derived landscape metrics (Table 3). Although none of the landscape metrics achieved statistical significance at the p<0.05 level, shrub class metrics appeared to be associated with an increase in risk. Examples of high and low-class and landscape metrics with the correlation to positive Lyme serology are included in Figure 2. The lawn landscape shape index (OR 1.197, p=0.0062) and shrub class area (OR 1.166, p=0.088) reached significance at the p<0.1 level, and were included in multivariate models. A description of all the landscape metrics is provided in Supplementary Table 1.

Next, 2 and 3-variable models containing landscape metrics demonstrated a linear relationship in univariate models and human behaviors which achieved a significance of p<0.05 (Table 4). A pairwise correlation test was run to determine which metrics were least correlated (<0.2), and therefore could be added to the models as covariates (Supplementary Table 2). None of the metrics with a pairwise correlation coefficient of <0.2 were significant in 2 landscape metric models. The AIC was calculated for each model to determine their relative goodness of fit.. Although a few of the models reached statistical significance for all variables, one 3-variable model including wearing protective clothing (OR 0.508, p=0.006), owning a cat spending time outdoors (OR 1.606 p=0.0049) and the binary shrub patch density (OR 0.607, p=0.0310) was found to have a significantly lower AIC (560.398) than the other models (Table 5). This model was more than 30 units lower than the next closest model. Descriptions of the variable codes are provided in Supplementary Table 3.

During the tick collection season of 2012, 105 participant properties and three natural areas were dragged and flagged for ticks. A total of 1595 nymphs were collected by both the dragging and flagging method, and 475 of those were found on resident properties. The average number of nymphs found per transect was 1.84 (range 0-22).

To determine whether landscape metrics could serve as proxy measures of density of nymphal ticks, negative binomial regressions were performed on the previously determined linear and 2-category landscape metrics and property tick counts from 2012 (Tables 6 and 7). Many of the lawn and shrub landscape metrics reached statistical significance, with shrub metrics being associated with an increase in nymphal density, and lawn metrics demonstrating a decrease in nymphal density.

Discussion

Of the Block Island residents, 73% reported using at least one protective measure against Lyme disease. Tick checking (51%) and protective clothing (42%) were the most commonly used protective behaviors among residents, while wearing repellent was practiced the least (16%). Of the human behaviors, wearing protective clothing demonstrated the strongest protective effect against Lyme disease both in univariate analysis and multivariate models. Using any protective measure, use of repellent, avoiding brush, and performing tick checks also were found to be protective, however they failed to reach statistical significance in univariate analysis. None of the landscape metrics met the $p<0.05$ statistical significance level in univariate analyses; however lawn landscape shape index (p=0.062) and the shrub class area (p=0.0088) were significant at $p<0.1$ and increased the risk of Lyme disease. Including shrub patch density in multivariate models improved the model fit, indicating an interactive role of landscape in influencing human exposure risk.

Owning a cat that spends time both indoors and outdoors and the average number of hours spent outdoors in tick habitat each day were found to be associated with an increased risk of Lyme disease, and were also statistically significant. It is possible that cats may be the main source of exposure in individuals who did not report spending a significant amount of time outdoors, as cats often bring nymphal ticks into the household. Because our study population was relatively old, it is possible that these individuals may not have the visual acuity to see the nymphal ticks, and therefore are at greater risk. Unfortunately, no information was available as to whether or not topical repellents were applied to cats, which may result in a decrease in risk.

Only 16% of the residents reported tick repellent application. The most effective topical repellents contain the ingredient DEET (Herrington 2004). Multiple studies have demonstrated the safety (Fradin 1998, Fradin and Day 2002) and effectiveness of DEET in protecting against Lyme disease (Herrington et al. 1997, Herrington 2004), although our study and a study by Connally et al. (2009b) did not find it protective. Up to one third of Americans do not believe it is effective and 40% believe that DEET itself could results in user's sickness (Herrington 2004), which may explain the low rate of repellent use.

A few of our multivariate models reached statistical significance, with the model including owning a cat, wearing protective clothing, and the binary shrub patch density having the lowest AIC value, and therefore demonstrating the best fit The finding that increasing shrub patch density leads to a decrease in positive Lyme serology was not expected. It would seem that an increase in the patch density, or the number of patches per unit area, would increase risk. As patch density increases, we see a decrease in the size of the corresponding patches. Decreased patch size results in less suitable habitat for deer, mice, and other animals which can carry ticks. This in turn would lead to a reduction in the number of ticks found in shrub patches, and a subsequent decrease in the risk of developing Lyme disease. This model demonstrates that models using both ecological and behavioral variables can be used to explain Lyme disease risk, and further shows that Lyme disease risk cannot be accounted for from strictly a behavioral or ecological perspective.

Our study has additionally shown that shrubs are associated with increased nymphal density on Block Island and that an increase in the density of edges between shrubs and other land cover types is associated with increased risk of developing

B. burgdorferi antibody. At larger spatial scales, forest fragmentation which increases the amount of forest edge has been linked to increased tick density and infection prevalence of ticks due to increased densities of mice, the most competent host for immature ticks and *B. burgdorferi* (Ostfeld and Keesing 2000, Allan et al. 2003). Increased forest edge has also been linked to increased (Jackson et al. 2006) and decreased (Brownstein et al. 2005) Lyme disease cases. Our study is the first to describe the association between tick habitat edge and human exposure in the peridomestic setting. At this spatial scale, edges likely represent increased human contact with infected ticks because the biological processes driving increased density of mice in smaller patches typically operate at a larger scale (Allan et al. 2003).

Our study suffers from several limitations. First, although serology is the most effective method to assess exposure, antibodies to *B. burgdorferi* are not detectable for 2-3 weeks following the onset of infection. We also were unable to determine the site where exposure and subsequent infection occurred. Even though the study was restricted to people who lived on Block Island more than 3 months during the peak transmission period, Island residents might have acquired the infection on the mainland or away from their residence, reducing the expected association between peridomestic risk and exposure. Recall bias may represent another possible study limitation. Participants who have had Lyme disease may have been more enthusiastic about enrolling in our study and more likely to remember events related to Lyme disease. Finally, we did not investigate in detail the possible variability in the way protective behaviors are used. For instance, we did not inquire about the frequency of protective measure use, so we were not able to assess the protective effect seen with increasing use.

In conclusion, our findings emphasize the association between environmental risk and human exposure and highlight the importance of accounting for environmental exposure to accurately ascertain the effectiveness of personal protective behaviors. Wearing protective clothing when exposed to tick habitat appears to be the most effective method to reduce exposure to Lyme disease. Additionally, limiting the exposure of cats to the outdoors or applying topical insecticides to pets may also reduce the risk of developing Lyme disease. Employing landscaping strategies which reduce the amount of peridomestic shrub edge could serve to reduce exposure and lessen disease risk as well. Prospective cohort studies are necessary to understand the relative importance and interactions between acarological risk, landscape design, and protective behaviors in reducing Lyme disease risk in a community.

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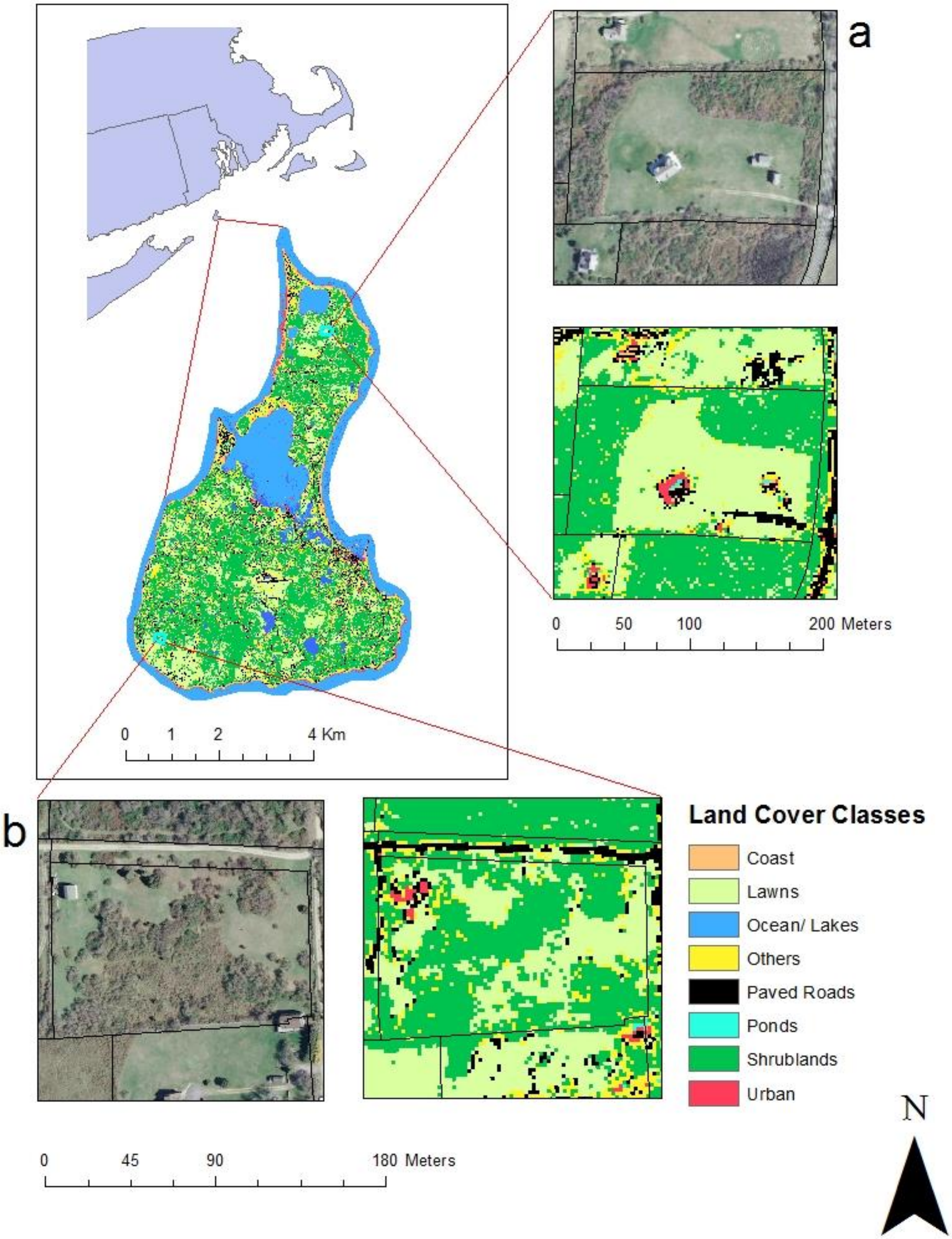
Appendix

Figure 1. Land cover classification of Block Island, Rhode Island. Examples of properties with a) low shrub edge density (454m/ha) and b) high edge density (959m/ha)



Supplementary Figure 2. Examples of properties with high (top row) and low (center row) landscape metrics used in three variable models, and their associated relationship with serology (bottom row)

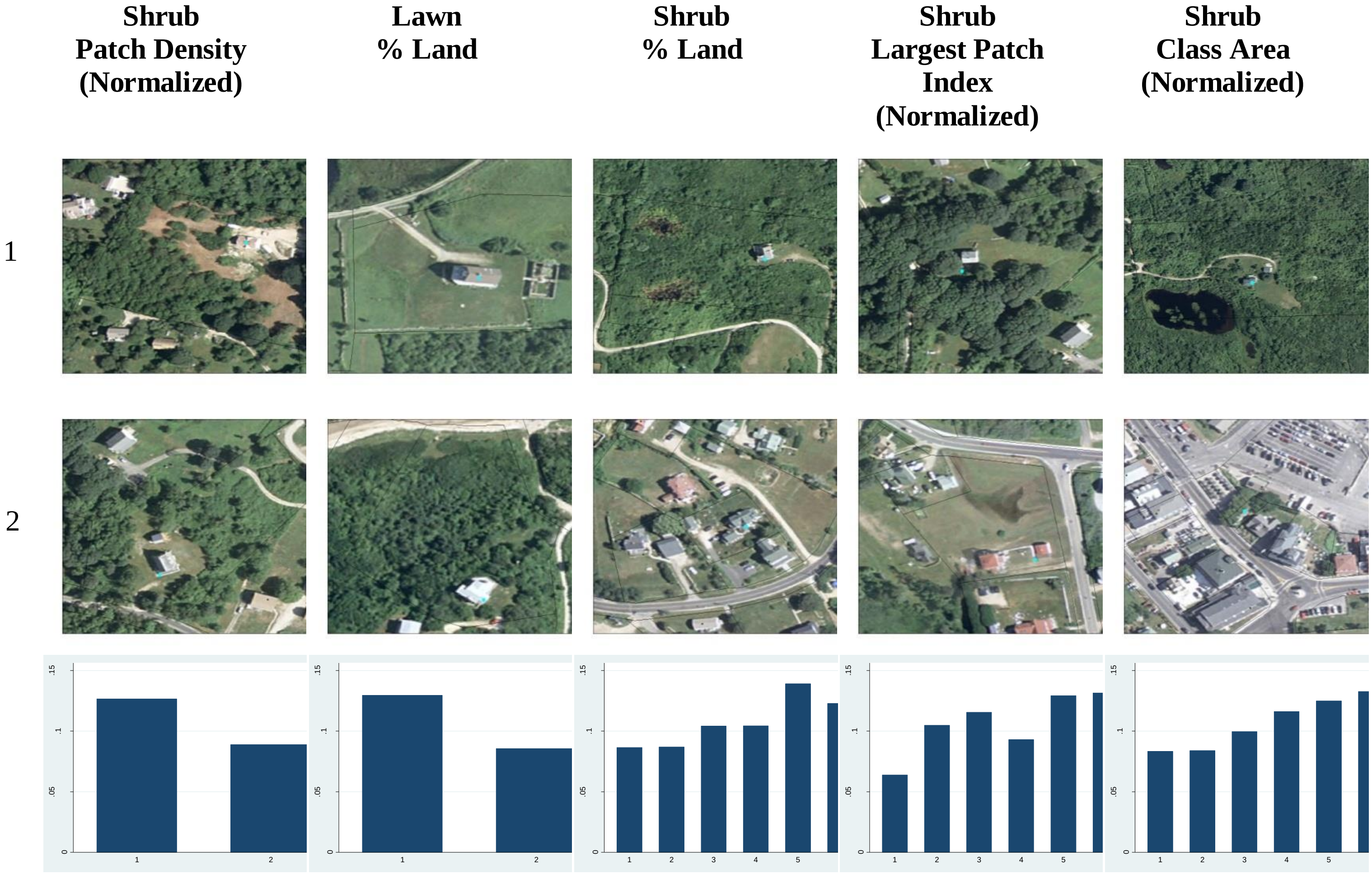


Table 1. Frequency (%) of behaviors reported by participants at serological surveys between 2005 and 2011. The total number of participants' use of each behavior is indicated.

Variable	**Yes**	**Total responses**
Any Tick Protection*	724 (72.6)	983
Protective Clothing	414 (41.6)	925
Repellent Use	159 (16.0)	925
Tick Checking	505 (50.7)	927
Avoiding Brush	351 (35.2)	923
Owning a Cat	246 (24.7)	893

*Any Tick Protection= use of either protective clothing, tick checking, repellent use or avoiding brush.

Table 2. Univariate logistic models of behaviors with Lyme serology

Variable	OR	P-value	LR chi2
Owning a Cat	1.62	0.033	4.37
Owning a Dog	1.09	0.699	0.15
Using Any Protective Measure	0.76	0.212	1.51
Use of Repellent	0.74	0.341	0.96
Use of Protective Clothing	0.51	0.004	8.95
Avoiding Brush	0.73	0.173	1.91
Routine Tick Checks	0.88	0.550	0.36
Occupational Exposure to Tick Habitat	0.60	0.216	1.73
Average Hours Spent in Vegetation Daily	1.35	0.040	4.27
Age at Test	1.02	0.011	7.19

Table 3. Univariate logistic models of landscape metrics with Lyme serology

Landscape Metric	**OR**	**P-value**	**LR chi2**
Lawn_CA	1.07	0.481	0.46
Lawn_PD	1.03	0.797	0.07
Lawn_NP	**1.10**	**0.321**	**0.93**
Lawn_LPI	0.91	0.388	0.77
Lawn_TE	1.12	0.201	1.52
Lawn_ED	0.94	0.530	0.39
Lawn_LSI	**1.20**	**0.062**	**3.36**
Shrub_LSI	1.10	0.325	0.93
Shrub_CA	**1.17**	**0.088**	**2.64**
Shrub_PLAND	**1.17**	**0.110**	**2.52**
Shrub_NP	1.01	0.951	0
Shrub_PD	**0.89**	**0.301**	**1.14**
Shrub_LPI	**1.16**	**0.137**	**2.16**
Shrub_TE	**1.11**	**0.234**	**1.27**
Shrub_ED	1.11	0.328	0.96
Lawn_PLAND	**0.99**	**0.215**	**1.58**

*Landscape metrics in bold demonstrated a linear relationship with serology and were subsequently used in multivariate models.

Table 4. Candidate models for landscape metrics and behaviors, ordered by Akaike information criterion

Variable	OR	P-Value	AIC	ΔAIC
Shrub_PD_2cat + Cat + Clothing	**0.61/1.61/0.51**	**0.031/0.049/0.006**	**560.3990**	**0**
Shrub_PLAND + Hrsveg + Cat	1.01/1.45/1.56	0.042/0.022/0.055	592.7549	32.3560
Shrub_LPI + Hrsveg + Cat	1.23/1.44/1.57	0.054/0.024/0.052	593.2725	32.8737
Shrub_CA + Clothing + Hrsveg	1.22/0.49/1.40	0.043/0.003/0.030	595.8768	35.4779
Shrub_PLAND + Cat	1.01/1.58	0.058/0.046	596.5307	36.1318
Shrub_PD_2cat + Hrsveg + Cat	0.57/1.45/1.69	0.011/0.020/0.022	597.2295	36.8305
Shrub_CA + Clothing	1.22/0.51	0.042/0.004	599.3159	38.9169
Lawn_PLAND_2cat + Hrsveg + Clothing	0.68/1.40/1.58	0.078/0.035/0.046	600.6702	40.2713
Shrub_PD_2cat + Cat	0.59/1.72	0.019/0.018	601.2322	40.8333
Lawn_PLAND_2cat + Hrsveg + Clothing	0.63/1.34/0.49	0.034/0.030/0.002	601.2397	40.8408
Lawn_NP_2cat + Cat	1.53/1.58	0.057/0.044	603.1832	42.7843
Lawn_PLAND_2cat + Cat	0.67/1.59	0.070/0.041	603.5440	43.1451
Shrub_TE_2cat + Cat	1.49/1.60	0.073/0.039	603.6155	43.2166
Lawn_PLAND_2cat + Clothing	0.62/0.51	0.030/0.004	604.5714	44.1724
Cat	1.62	0.033	604.8802	44.4813
Horse	1	1	605.6684	45.2695
Clothing	0.51	0.004	607.3657	46.9667
Opet	1.33	0.565	608.7142	48.3152
Dog	1.10	0.699	609.3261	48.9271
Abrush	0.73	0.173	613.9694	53.5704
Repellent	0.74	0.341	615.3524	54.9535
Tcheck	0.88	0.550	616.3906	55.9917
Occ_Exp	0.60	0.216	618.0345	57.6356
Lawn_LSI + Hrsveg	1.20/1.33	0.063/0.050	665.8926	105.4937
Lawn_PLAND_2cat + Hrsveg	0.65/1.35	0.04/0.042	670.4411	110.0421
Tprotect	0.76	0.212	670.5455	110.1466
Shrub_PD_2cat + Hrsveg	0.66/1.39	0.043/0.027	670.5809	110.1819
Lawn_NP_2cat + Hrsveg	1.47/1.32	0.067/0.058	671.2865	110.8876
Hrsveg	1.35	0.040	672.7122	112.3132
Shrub_CA	1.17	0.088	672.9716	112.5727
Shrub_PLAND	1.01	0.110	673.0849	112.6859
Shrub_PLAND	1.17	0.110	673.0849	112.6859
Deer	0.93	0.585	673.2375	112.8385
Shrub_LPI	1.16	0.137	673.4525	113.0535
Shrub_TE	1.11	0.234	674.3385	113.9396
Shrub_PD	0.89	0.301	674.4676	114.0686
Hrsvegbinary	1.43	0.112	674.5437	114.1447
Shrub_ED	1.11	0.328	674.6518	114.2528
Shrub_LSI	1.10	0.325	674.6794	114.2804
Lawn_LSI	1.20	0.062	674.7579	114.3589
Shrub_NP	1.01	0.951	675.6057	115.2067
Lawn_TE	1.12	0.201	676.5974	116.1984
Lawn_NP	1.10	0.321	677.1857	116.7867
Lawn_LPI	0.91	0.388	677.3450	116.9461
Lawn_CA	1.07	0.481	677.6494	117.2504
Lawn_ED	0.94	0.530	677.7182	117.3192
Lawn_PD	1.03	0.797	678.0474	117.6485
Lawn_PLAND_2cat	0.63	0.026	678.4560	118.0570
Shrub_PD_2cat	0.67	0.057	679.8057	119.4067
Lawn_NP_2cat	1.47	0.066	680.0337	119.6347
Shrub_PLAND_2cat	1.28	0.234	682.0645	121.6655
Shrub_TE_2cat	1.27	0.249	682.1499	121.7509

Table 5. Multivariate model containing cat, clothing, and 2 category shrub patch density covariates

	Odds Ratio	SE	Z-score	P-value	95% CI (Lower)	95% CI (Upper)
Cat	1.61	.387	1.97	0.049	1.002	2.575
Clothing	.51	.125	-2.76	0.006	0.314	0.822
Shrub_PD_2cat	.61	.140	-2.76	0.031	0.387	0.954

Table 6. Negative binomial models of lawn and shrub landscape metrics vs. density of nymphs

Landscape Metric	**Coefficient**	**P-value**	**AIC**
Lawn_CA	-0.383	0.572	359.6419
Lawn_PLAND	**-0.221**	**0.005**	**354.4727**
Lawn_PD	-0.247	0.160	358.2804
Lawn_NP	0.593	0.103	357.3679
Lawn_LPI	**-0.357**	**0.005**	**353.7905**
Lawn_TE	0.113	0.717	361.2919
Lawn_ED	**-0.347**	**0.022**	**350.6045**
Lawn_LSI	0.285	0.143	360.4392
Shrub_LSI	**0.344**	**0.038**	**361.3610**
Shrub_CA	**1.348**	**0.021**	**355.9406**
Shrub_PLAND	**0.674**	**0.001**	**343.5467**
Shrub_NP	**0.441**	**0.011**	**361.4086**
Shrub_PD	-0.354	0.100	350.9096
Shrub_LPI	**0.422**	**0.018**	**346.2717**
Shrub_TE	**0.857**	**0.016**	**359.4927**
Shrub_ED	**0.486**	**0.002**	**358.6309**

*Landscape metrics in bold were significant at $p<0.05$.

Table 7. Negative binomial modelss of 2 category lawn and shrub landscape metrics vs. density of nymphs

Landscape Metric	**Coefficient**	**P-value**	**AIC**
Lawn_CA_2cat	-0.083	0.746	1036.8289
Lawn_PLAND_2cat	**-0.809**	**0.001**	**1027.0910**
Lawn_PD_2cat	0.173	0.501	1036.4805
Lawn_NP_2cat	**0.548**	**0.033**	**1032.4047**
Lawn_LPI_2cat	**-0.893**	**0.001**	**1024.9379**
Lawn_TE_2cat	0.0634	0.804	1036.8725
Lawn_ED_2cat	-0.482	0.062	1033.4388
Lawn_LSI_2cat	**0.548**	**0.033**	**1032.4047**
Shrub_LSI_2cat	0.208	0.420	1036.2840
Shrub_CA_2cat	**0.538**	**0.037**	**1032.5681**
Shrub_PLAND_2cat	**1.251**	**0**	**1013.5554**
Shrub_NP_2cat	0.387	0.132	1034.6635
Shrub_PD_2cat	0.305	0.236	1035.5323
Shrub_LPI_2cat	0.223	0.389	1036.1883
Shrub_TE_2cat	**0.525**	**0.046**	**1032.9700**
Shrub_ED_2cat	**0.840**	**0.001**	**1026.3461**

*Landscape metrics in bold were significant at $p<0.05$.

Supplementary Figure 1. Questionnaire delivered during the biannual serosurveys

Study #_________

Name____ ___________Birth date ______________

Permanent address and telephone #_________________________Occupation______________

____

Vacation street address and telephone # ____

____Fire code # ____________

Have you received the Lyme Vaccine?Yes_______If yes, what year?_______ No_____

Which of these groups best describes your ethnic identification? Circle the number of your answer:

1) Asian, 2) Black, not of Hispanic origin, 3) Hispanic, 4) West Indian/Caribbean,

5) White, 6) American Indian, 7)Mixed, 8) Other, 9) Don't know

EXPOSURE HISTORY

1. How many years have you spent at your present address (permanent or vacation)? _________

During which months? All___If not all,check all that apply- Jan___Feb___Mar___Apr___ May ___ June ___

July ___ Aug ___Sept ___ Oct ___ Nov___Dec__

2. How many hours a day do you spend out of doors near vegetation?

Less than 1 ___ Several ___ 5 or more ___

3. Do you keep a pet? dog___ cat___ horse___ other___

4. How frequently do you see deer around your residence?

Daily ___ Weekly ___ Less frequently ___

5. Have you been bitten by a tick this year? Yes ___ No ___

If yes, was it a deer tick ___ wood/dog tick ___ tiny ___ large ___

If yes, was it in your town? ___ Elsewhere? ________________

6. Do your tick bites itch? Yes ___ No ___

7. When outdoors, what personal protection measures against ticks do you employ?

None ___ Repellent ___ Long pants/socks___ Avoid brush___Tick check ___

8. Do you try to control ticks around your residence? Yes ___ No ___

Chemical spray ___ Damminix ___ Brush control ___ Other ________

ILLNESS HISTORY

9. Have you ever been diagnosed with Lyme disease____ babesiosis ___anaplasmosis____?

If so, by symptoms ___ blood test ___ both ___

When? ________ What was your treatment? _____________

Name of physician _______________________ City ______________

How much did your illness cost you (time lost, physician and treatment costs, etc.)? ____

10. Have you had any of the following signs of illness this year?

A) rash B) chills C) fever D)headache E) muscle aches F) fatigue G) night sweats H) joint pains I) swelling J) nasal congestion K) cough L) sore throat

11. How many people do you know who have had Lyme disease?_______

12. Have you ever been diagnosed to have immunodeficiency?_______

13. Have you had problems with recurrent infections in the last 10 years?______________

14. Are you on long-term steroids or other immunosuppressive medication?__________

Supplementary Table 1. Class and landscape metric calculations in Fragstats.

Landscape Metric	Description
CA	CA equals the sum of the areas (m^2) of all patches of the corresponding patch type, divided by 10,000 (to convert to hectares); that is, total class area.
PLAND	PLAND equals the sum of the areas (m^2) of all patches of the corresponding patch type, divided by total landscape area (m^2), multiplied by 100 (to convert to a percentage); in other words, PLAND equals the percentage the landscape comprised of the corresponding patch type. Note, total landscape area (A) includes any internal background present.
NP	NP equals the number of patches of the corresponding patch type (class).
PD	PD equals the number of patches of the corresponding patch type divided by total landscape area (m^2), multiplied by 10,000 and 100 (to convert to 100 hectares). Note, total landscape area (A) includes any internal background present.
TE	TE equals the sum of the lengths (m) of all edge segments involving the corresponding patch type. If a landscape border is present, TE includes landscape boundary segments involving the corresponding patch type and representing 'true' edge only (i.e., abutting patches of different classes). If a landscape border is absent, TE includes a user-specified proportion of landscape boundary segments involving the corresponding patch type. Regardless of whether a landscape border is present or not, TE includes a user-specified proportion of internal background edge segments involving the corresponding patch type.
LSI	LSI equals the total length of edge (or perimeter) involving the corresponding class, given in number of cell surfaces, divided by the minimum length of class edge (or perimeter) possible for a maximally aggregated class, also given in number of cell surfaces, which is achieved when the class is maximally clumped into a single, compact patch.
LPI	LPI equals the area (m^2) of the largest patch in the landscape divided by total landscape area (m^2), multiplied by 100 (to convert to a percentage); in other words, LPI equals the percent of the landscape that the largest patch comprises. Note, total landscape area (A) includes any internal background present.
ED	ED equals the sum of the lengths (m) of all edge segments in the landscape, divided by the total landscape area (m^2), multiplied by 10,000 (to convert to hectares). If a landscape border is present, ED includes landscape boundary segments representing 'true' edge only (i.e., abutting patches of different classes). If a landscape border is absent, ED includes a user-specified proportion of the landscape boundary. Regardless of whether a landscape border is present or not, ED includes a user-specified proportion of internal background edge. Note, total landscape area (A) includes any internal background present.

Supplementary Table 2. Pairwise correlations of landscape metrics

	Shrub_CA	Shrub_PLAND	Shrub_PD_2cat	Shrub_PD_3cat
Shrub_CA	1			
Shrub_PLAND	0.530	1		
Shrub_PD_2cat	-0.421	-0.387	1	
Shrub_PD_3cat	-0.369	-0.380	0.821	1
Lawn_LSI	0.687	0.266	-0.1823	-0.116
Lawn_NP_2cat	0.409	0.316	-0.188	-0.101
PLAND_2cat	-0.361	-0.554	0.177	0.180
Shrub_TE_2cat	0.558	0.469	-0.314	-0.249
Shrub_LPI	0.480	0.953	-0.439	-0.451

	Lawn_LSI	Lawn_NP_2cat	Lawn_PLAND_2cat	Shrub_TE_2cat
Lawn_LSI	1			
Lawn_NP_2cat	0.681	1		
Lawn_PLAND_2cat	-0.136	-0.171	1	
Shrub_TE_2cat	0.635	0.645	-0.141	1
Shrub_LPI	0.156	0.228	-0.567	0.366

Supplementary Table 3. Descriptions of landscape metrics and variable codes

Variable	**Description**
Cat	Owning a cat that spends time indoors and outdoors
Horse	Owning a horse
Clothing	Wearing protective clothing during outdoor exposure
Opet	Owning a pet other than a cat, dog, or horse
Dog	Owning a dog that spends time indoors and outdoors
Abrush	Avoiding brush during outdoor exposure
Repellent	Wearing repellent during outdoor exposure
Tcheck	Performing tick checks after outdoor exposure
Occ_Exp	Employed in a profession that requires outdoor exposure
Tprotect	Using any protective measure against exposure
Hrsveg	Categorical variable of the number of hours spent outside daily (1,2,3,4,5 or more)
Shrub_CA	Shrub class area
Shrub_PLAND	Shrub percentage of land
Deer	If deer are frequently seen on the property
Shrub_LPI	Shrub largest patch index
Shrub_TE	Shrub total edge
Shrub_PD	Shrub patch density
Hrsvegbinary	Binary variable of the number of hours spent outside daily separated at the median
Shrub_ED	Shrub edge density
Shrub_LSI	Shrub landscape shape index
Lawn_LSI	Lawn landscape shape index
Shrub_NP	Shrub number of patches
Lawn_TE	Lawn total edge
Lawn_NP	Lawn number of patches
Lawn_LPI	Lawn largest patch index
Lawn_CA	Lawn class area
Lawn_ED	Lawn edge density
Lawn_PD	Lawn patch density
Lawn_PLAND_2cat	Binary variable of the lawn percentage of land separated at the median
Shrub_PD_2cat	Binary variable of the shrub patch density separated at the median
Lawn_NP_2cat	Binary variable of the lawn number of patches separated at the median
Shrub_PLAND_2cat	Binary variable of the shrub percentage of land separated at the median
Shrub_TE_2cat	Binary variable of the shrub total edge separated at the median