Climate Change, Lyme Disease, and Other Tickborne Diseases in Vermont

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VERMONT
DEPARTMENT OF HEALTH

Executive Summary

A review of current scientific literature and Lyme disease data shows that **the** risk for Lyme disease and other diseases transmitted by blacklegged ticks is likely to increase in the future as temperatures in Vermont warm due to climate change. Currently, Lyme disease incidence is lower in areas in the cooler northeastern portion of the state, as well as areas with higher elevation. Warming temperatures due to climate change may make these areas more suitable for blacklegged ticks in the future, presenting new areas where Lyme disease and anaplasmosis may be transmitted.

Nearly all tickborne infections in Vermont (99%) are caused by bites from blacklegged ticks (*Ixodes scapularis,* also commonly known as deer ticks), which are also the most commonly encountered ticks in the state[1]. Currently, Lyme disease (*Borrelia burgdorferi*) and anaplasmosis (*Anaplasma phagocytophilum*) are the most common of these tickborne infections. The incidence of both diseases has been rising in recent years (Figures E.1 and E.2). Potential explanations for this rise include: better diagnosis and reporting of the disease by primary care providers, and true changes in disease incidence, spurred by long-term changes in forest cover (including reforestation throughout the 20th century and forest fragmentation), warming temperatures due to climate change that affect ticks and their animal hosts, as well as other factors that may affect populations of rodents and deer, which play integral roles in the transmission cycles of tickborne pathogens.

Key information:

In areas where tickborne diseases are present (like Vermont), the likelihood of a person getting a tickborne disease (like Lyme disease) depends on three factors: 1) how many ticks are in the area, 2) how many of those ticks are infected with the pathogen, 3) how often people come into contact with those ticks. To a varying degree, climate change can affect all three of these factors.

1) Ticks can only live in areas where the climate is suitable for them (ie. the right temperatures and the right amount of moisture). Warming temperatures due to

[1] There are four types of ticks that can transmit diseases that may be encountered Vermont: the blacklegged tick (Ixodes scapularis, also known as the deer tick), the American dog tick (Dermacentor variabilis), the woodchuck tick (Ixodes cookei), and the lone star tick (Amblyomma americanum).

climate change may make Vermont more hospitable to blacklegged ticks. This may mean that areas that were not previously infested with ticks (e.g. colder areas, and areas at higher elevations) can become infested, and that tick populations may increase in areas that already have them.

2) Work done by our partners at Lyndon State College suggests that areas more densely populated with blacklegged ticks in Vermont also tend to have higher rates of infection with *Borrelia burgdorferi* among those ticks. This suggests that in areas where blacklegged tick populations may grow due to climate change, a greater proportion of these ticks may be infected with the bacteria in the future as these populations grow.

3) Blacklegged ticks are typically not active at temperatures below freezing. Warming temperatures due to climate change mean more days when ticks are active and looking for blood meals, which means more opportunities for encounters between ticks and humans.

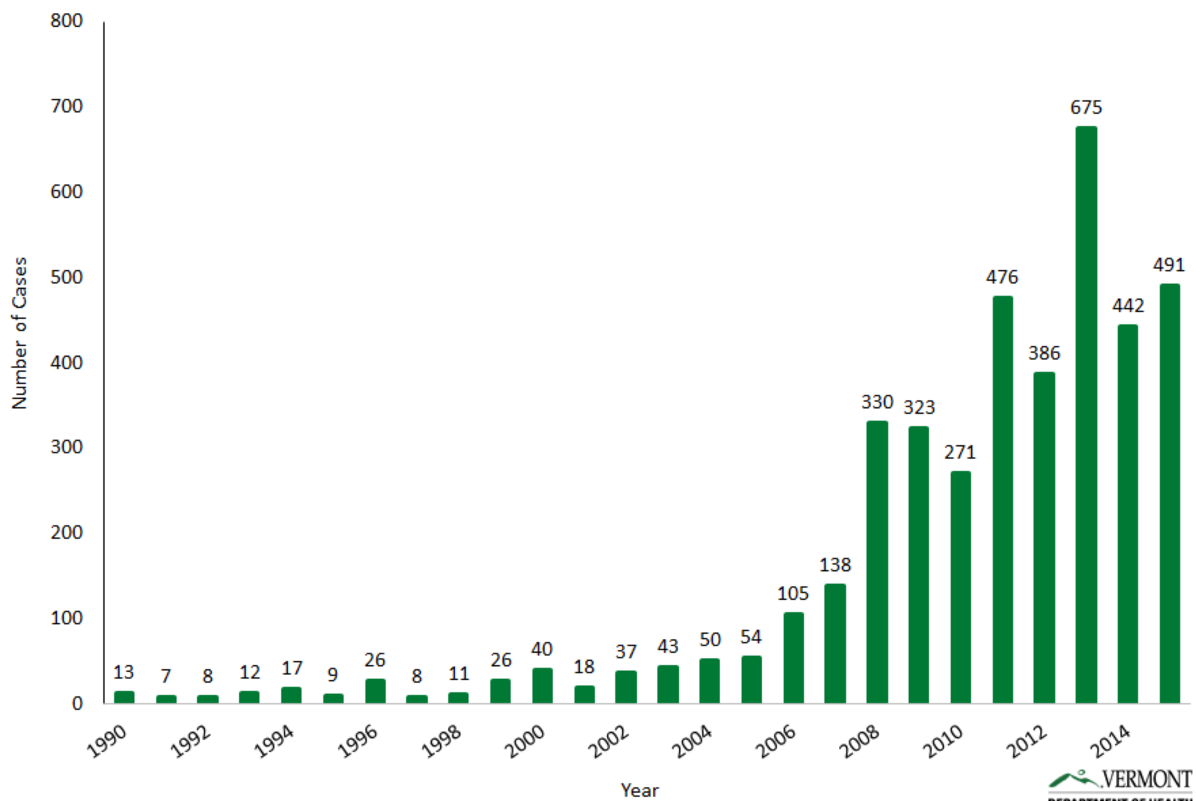


Figure E.1 – Number of confirmed Lyme disease cases reported to the Vermont Department of Health, 1990 to 2015.

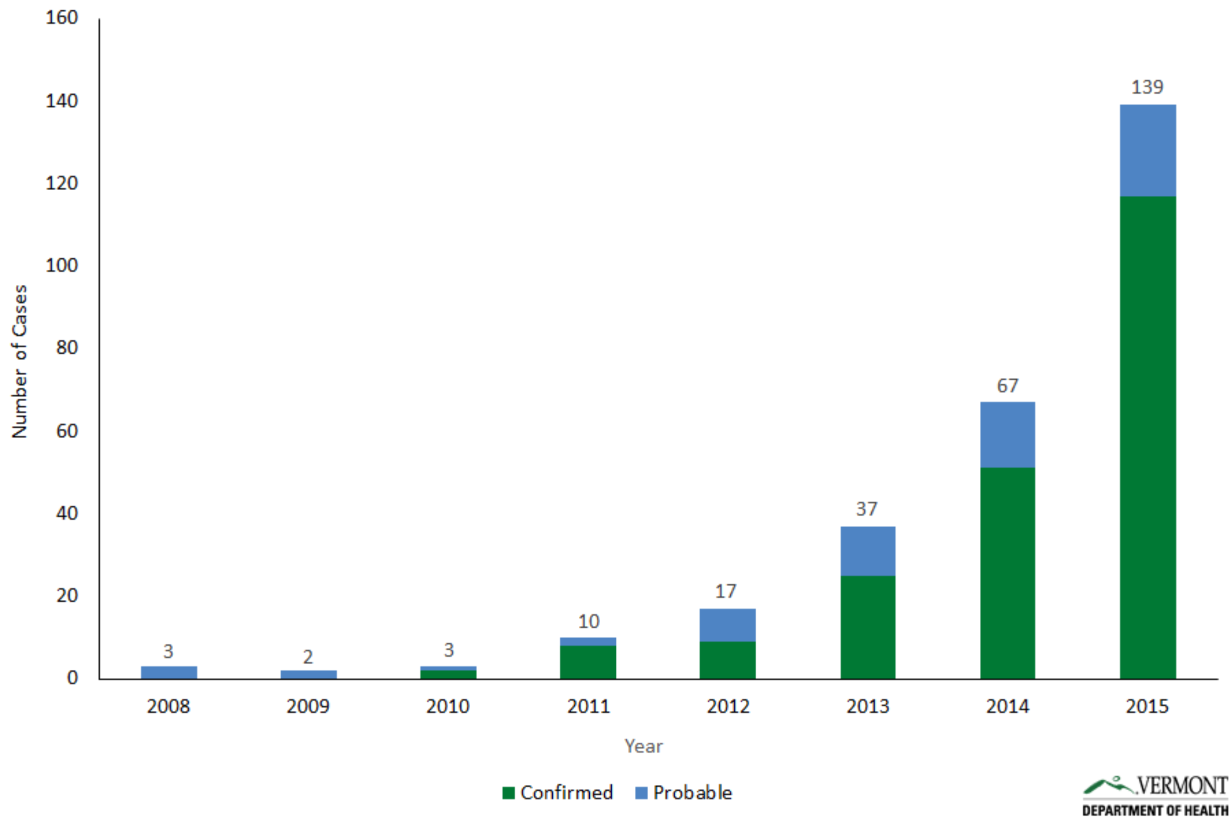


Figure E.2 – Number of confirmed & probable cases of anaplasmosis reported to the Vermont Department of Health, 2008 to 2015.

Climate and Lyme Disease Analysis Findings:

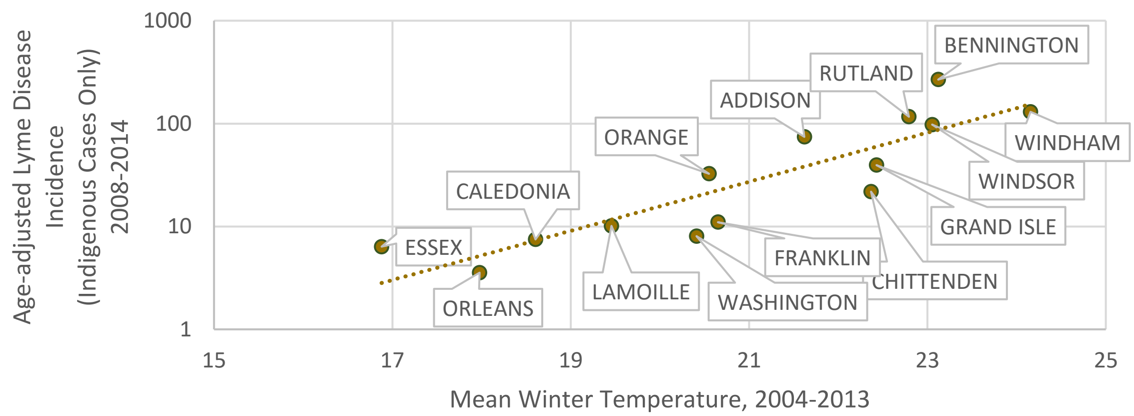
The original analysis presented in this report indicates that:

1) The incidence of Lyme disease in Vermont counties for the years 2008-2014 was closely correlated to ten-year average winter temperatures (2004-2013) of those counties ($p>0.001$, $R^2=0.77$).
   a. This seasonal effect was stronger than those of other seasonal and annual meteorological variables analyzed. Those included spring, fall, and summer mean temperature; winter, spring, summer, and fall precipitation; average annual growing degree days, heating degree days, and cooling degree days; total annual precipitation, and mean annual temperature.
   b. Winter temperatures may be linked to both the overwintering survival of ticks, as well as rodents such as white-footed mice that act as reservoirs for the *Borrelia burgdorferi* pathogen that causes Lyme disease. Over the long-term, this could

mean that winter temperatures act as an important limiting factor for the *B. burgdorferi* transmission cycle.

c. If winter temperatures are an important limiting factor for Lyme disease transmission, the risk for being bitten by infected ticks in the northeastern portions of the state, and in areas of higher elevation that are typically cooler, may increase as winter temperatures continue to rise with climate change.

Figure E.3 – Graph of age-adjusted Lyme disease incidence (2008-2014, indigenous, within-county cases only) plotted against mean winter temperature (2004-2013)



2) More growing degree days (i.e. warmer temperatures) in the first 80 days of the year are significantly associated with an earlier start to peak Lyme disease season in Vermont counties. Conversely, increased precipitation during the same period of the year was associated with a later start to the season. Given that both temperatures and overall precipitation are expected to rise during this portion of the year, and due to limitations in our climate projections, we are currently unable to determine precisely how future changes in temperature and precipitation will affect the timing of peak Lyme disease season in the future.

Contents

Introduction

This report documents the relationship between tickborne diseases and climate in Vermont. We reviewed both current scientific literature and Vermont data to assess the current state of tickborne diseases in Vermont and to project how global climate change may affect the incidence of these diseases in the future.

Background

Vector-borne diseases are one of the primary health concerns associated with a changing climate. Warming temperatures are expected to expand the suitable range for ticks and their hosts while also extending the activity season of ticks.

Nearly all tickborne infections in Vermont (99%) are caused by bites from blacklegged ticks (*Ixodes scapularis*). Currently, Lyme disease (*Borrelia burgdorferi*) and anaplasmosis (*Anaplasma phagocytophilum*) are the most common of these tickborne infections. Other pathogens transmitted by blacklegged ticks include *Babesia microti,* the parasite that causes babesiosis, a malaria-like illness, and deer tick virus (DTV), a strain of Powassan virus (POWV) that can cause fever and encephalitis. Blacklegged ticks are currently the most commonly encountered tick species in Vermont (VDH 2015a*)*.

The incidence of both Lyme disease and anaplasmosis has increased substantially in recent years in Vermont. Confirmed Lyme disease cases have risen from 50 or fewer per year before 2005 to more than 380 each year from 2011 onward (Figure 1). While some of this may be due to changes in disease reporting[2], it is likely that there has been a true increase in the incidence of this disease. Anaplasmosis case reports have also increased steadily in the past five years, from only 3 cases in 2010 to 67 cases in 2014 (Figure 2).

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[2] The Centers for Disease Control and prevention case definition for Lyme disease included only confirmed cases until 2007, and both confirmed and probable cases from 2008 to the present. Additionally, the CDC case definition criteria for laboratory evidence of infection for confirmed Lyme disease cases was changed in both 2008 and 2011. All of these factors, as well as awareness of the disease, may affect physicians' reporting of cases to the State of Vermont and thus reported case counts.

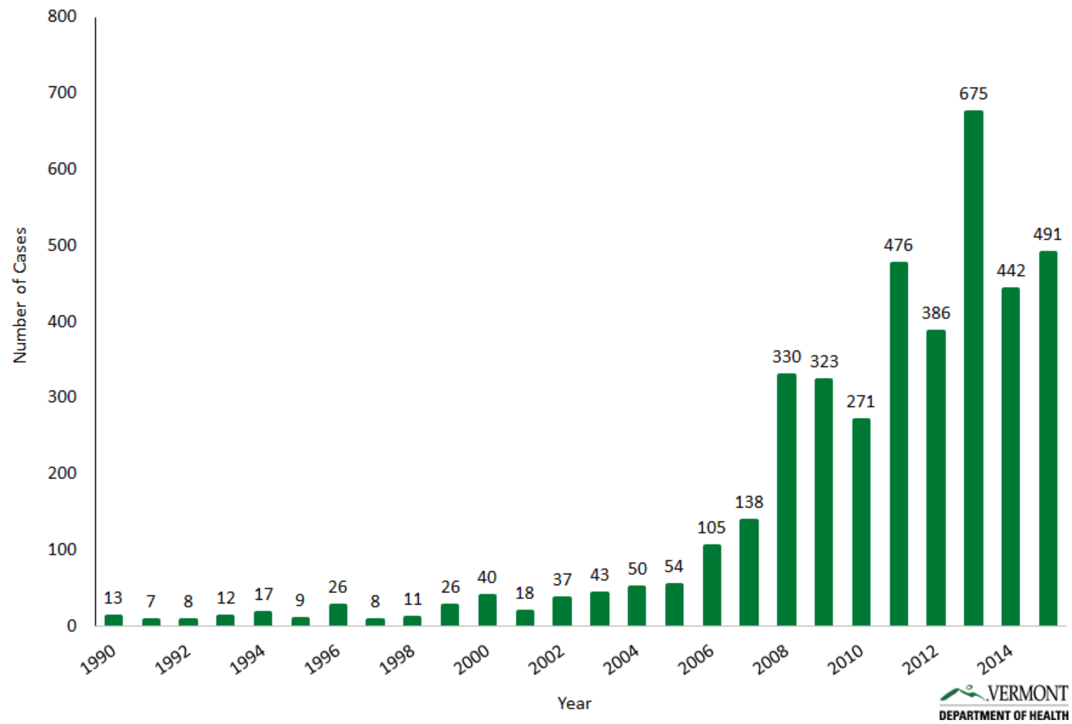


Figure 1 – Confirmed cases of Lyme disease in Vermont, 1990 to 2015.

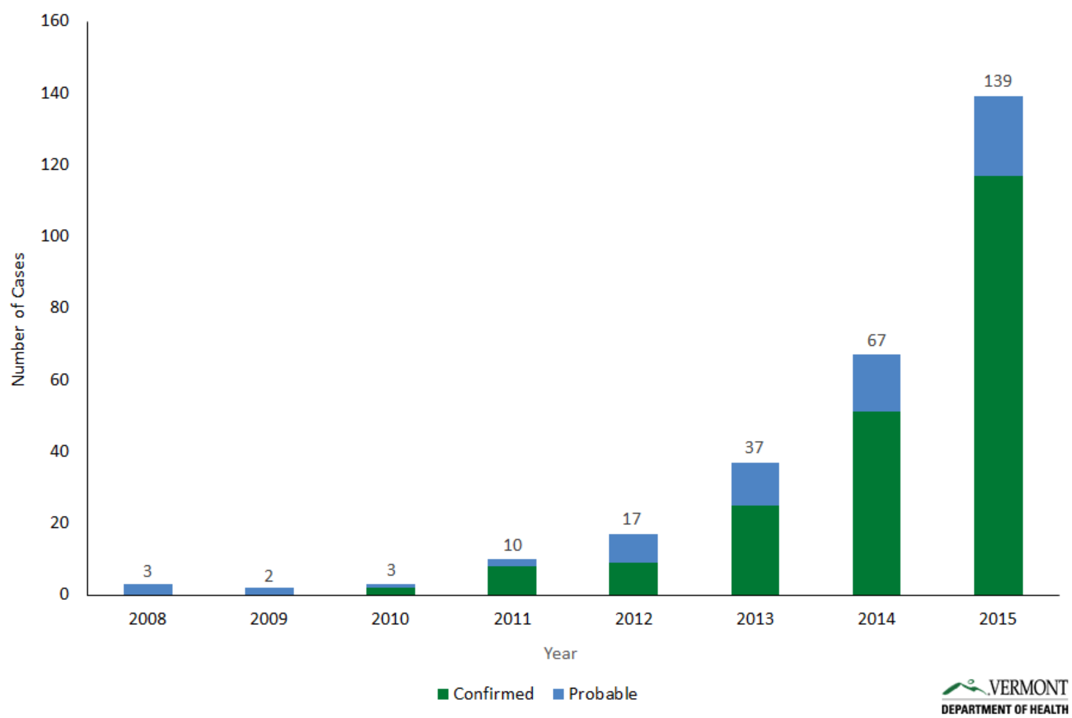


Figure 2 - Reported confirmed cases of anaplasmosis in Vermont, 2010 to 2014

Climate change is one of a number of hypothesized factors that may explain this increase in Lyme disease and anaplasmosis incidence (Brownstein et al. 2010). Others include reforestation over the 20th century (Barbour and Fish 1993), increasing forest fragmentation (which can affect the abundance of ticks and white-footed mice, as well as the amount of interaction people have with them), rises in deer populations which blacklegged ticks depend on as hosts for reproducing adults (Brownstein et al. 2010), and better detection, diagnosis, and reporting when cases develop. Regarding deer populations, in Vermont, the statewide pre-hunt population estimate of white-tailed deer has fluctuated between roughly 100,000 and 160,000 since the year 2000 (Figure 3).

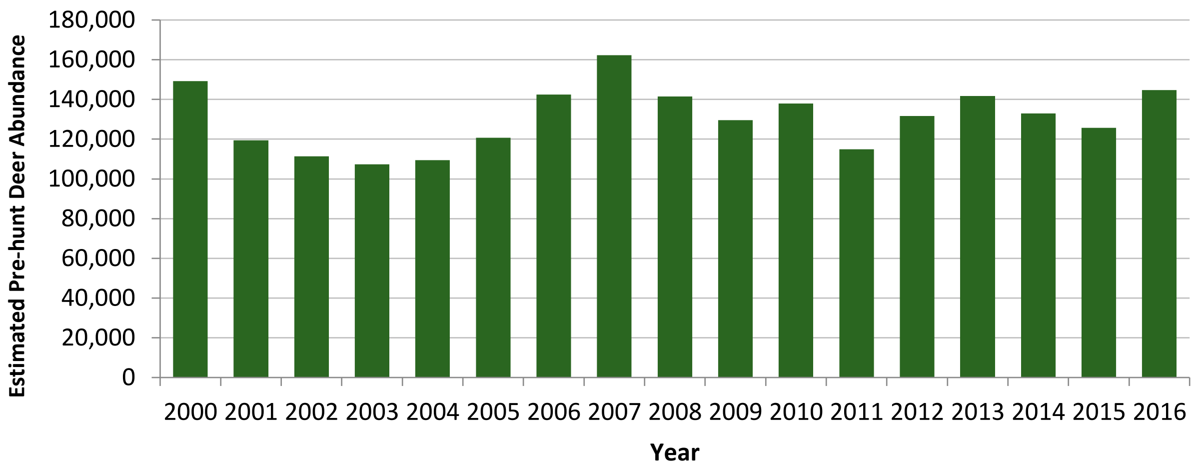


Figure 3 – Vermont statewide pre-hunt deer population estimates, 2000-2016. Population estimates are based on VT-DEOPOP, Mark Removal, and Sex-Age-Kill modeling.

The hypothesis that climate change may be responsible for some of the increase in Lyme disease incidence is supported by 1) the steady warming trend over the past several decades, which improves the likelihood of tick survival over winter, and 2) the fact that the increases in Lyme disease incidence have been observed first in the southern part of the state and later in the northern part of the state. It is unlikely that any given hypothesized explanation for the increase in tickborne disease incidence in Vermont is a sole contributor acting alone. Rather, it is likely that many, or all, of these hypothesized explanations have played a role.

Most importantly, climate change may increase the risk for Vermonters to contract tickborne diseases in the future. Warming temperatures, especially during the winter months, may make conditions more suitable for Lyme disease transmission in areas where the risk has been historically low, such as the northeastern portion of the state. The purpose of this report is to evaluate the current evidence on climate, ecology, seasonality, and tickborne diseases and to assess the potential impact of climate change on the risk tickborne diseases in Vermont.

Report Overview

This report reviews the major tickborne diseases of concern in the Northeastern United States, important tick species, and the connection between climate and tickborne diseases.

The report contains:

- a review of the current science of how tickborne diseases may be affected by climate change;
- a summary of tick surveillance activity that has occurred in Vermont to-date;
- descriptive statistics and analysis of the seasonality of Lyme disease in Vermont;
- an analysis of the geographic distribution of Lyme disease in Vermont as it relates to climate and ecological factors;

and an appendix containing:

- background on the ecology and epidemiology of tickborne diseases of importance in the Northeastern United States; and
- an overview of the tick species that are currently found in Vermont and those that may establish themselves in the state as temperatures warm.

While this report has a broad objective of characterizing the current science on tickborne disease and climate change in Vermont, the sections concerning Vermont-specific data and analysis focus only on Lyme disease, as the relatively large number of Lyme cases in Vermont allows sufficient statistical power for quantitative analysis.

Existing Literature on Climate Effects on Tickborne diseases

Climate change is expected to alter the transmission cycles of tickborne diseases, and there is evidence that these changes are already occurring. Generally, climate change may lead to an increase in tickborne diseases that already exist in an area, or it may facilitate the expansion of diseases into areas which they did not exist in recent history. Conversely, it may make areas that were previously suitable for the transmission of a tickborne disease less suitable for transmission of that disease (Brownstein et al. 2005). The U.S. Global Change Research Program stated, in 2016, "[b]ased on the evidence, there is high confidence that climate change, especially temperature change, is likely to cause shifts in the geographical distribution of ticks capable of carrying *B. burgdorferi* to more northern latitudes, the timing of host-seeking activity of ticks, and the timing of Lyme disease case occurrence", although the authors stress that uncertainty in the literature make it difficult to determine how this may affect human disease (USGCRP, 2016). In Vermont, there is some evidence to suggest that climate change may make Vermont more suitable for the transmission of Lyme disease and anaplasmosis, and potentially other infections transmitted by blacklegged ticks, although more study is needed to determine the extent to which this is the case.

Within locations where *B. burgdorferi* is transmitted, three key factors are important in determining human risk for contracting the disease: 1) the abundance of tick vectors (especially host-seeking nymphs) 2) prevalence of *B. burgdorferi* infection among those ticks 3) the frequency of contact between ticks and humans (USGRP, 2016). These factors are similarly important for other tickborne diseases. Climate change and its accompanying effects (e.g., warmer temperatures and greater precipitation) can affect the potential for tickborne disease transmission in a variety of ways described in more detail below.

Extreme Winter Temperature and White-footed Mice

White-footed mice (*Peromyscus leucopus*) are one of the most abundant mammals in North America, are a reservoir for both *Borrelia burgdorferi* and *Anaplasma phagocytophilum* in nature, and are also competent reservoir hosts for *Babesia microti*, *Borrelia miayamotoi*, and deer tick virus (DTV) (Cook and Barbour, 2015; Levin et al. 2002). White-footed mice are important hosts for blacklegged ticks, and a study in southern Quebec found that they were the most common hosts for blacklegged ticks among several different small mammal species that were trapped (Bouchard et al. 2011).

Vermont is near the northern habitat limit for white-footed mice in the Northeast, which currently extends into southern Québec (Roy-Dufrense et al. 2013). The northern range of this species has been expanding in recent years which has been documented in historical records from both the Upper Peninsula of Michigan and southern Quebec (Roy-Dufrense et al. 2013). Modeling efforts in Quebec predict that milder and shorter winters will continue to expand the northern distribution of white-footed mice. Average winter length and maximum temperature were the strongest predictors of white-footed mouse distribution, and the authors noted that where average maximum winter temperatures were above -5°C (23°F), the probability for white-footed mice to be in the area increased substantially (Roy-Dufrense et al. 2013). Because white-footed mice play such an important role in the transmission cycles of tickborne diseases in the northeast, their dependence on mild winter temperatures may play an important factor in determining location-specific risk for transmission of *B. burgdorferi*, *A. phagocytophilum*, and other pathogens.

Extreme Winter Temperature and Blacklegged Ticks

There is some evidence that winter temperatures may affect overwintering survival of newly molted nymphs and gravid adult female blacklegged ticks (Ostfeld and Brunner 2015). Laboratory evidence indicates that short term exposure to extreme cold (below -15°C) can be lethal to these ticks (Burks et al 1996, Vandyk et al. 1996, as summarized in Brunner et al 2012). In contrast, a limited experimental study of blacklegged tick nymphs in their natural environment found that about 80% of ticks survived regardless of winter conditions at the two sites tested (Brunner et al. 2012). One reason given for this survival rate is that overwintering ticks rarely experience the coldest temperatures of winter due to their ability to insulate themselves under leaf litter, soil, and snow (Brunner et al. 2012). However, that study was limited in scope, with only two sites tested, and minimum temperatures did not get lower than approximately -15°C (~5°F) at either site. In contrast, temperatures in Essex and Bennington Counties in Vermont reached as low as -23°C (~ -10°F) and -17°C (~1°F), respectively, in 2014. So, the results of the study by Brunner and others do not necessarily indicate that winter temperatures are a non-factor for ticks in Vermont, and more research is needed to understand the effects of extreme winter temperatures and insulating snowpack on blacklegged tick populations. Additionally, extreme cold temperatures in the winter are associated with fewer numbers of days overall above freezing, which may mean fewer days when ticks can be out and biting, which in turn may place constraints on tick populations.

Elevation and Blacklegged Ticks

In Vermont, a mountainous state, one very important factor to consider is the relationship between tick abundance and elevation. Generally speaking, areas of higher elevation in a given region are colder than those of lower elevation, and more closely resemble areas of higher latitude (i.e., in the northern hemisphere, the ecology of areas on the tops of high elevation hills and mountains more closely matches that of areas much farther north but at relatively lower elevations.) Increasing elevation is correlated with lower temperatures; this effect is roughly equivalent to a decrease in 5.7° C (10.3°F) per kilometer increase elevation (Linacre 1992, as summarized in Sutherst 2004). Mills and others (2010) note evidence that plant and animal species have had habitat range shifts toward higher elevations, in addition to higher latitudes, in response to warming temperatures. This has important implications for disease vectors like ticks.

Studies have demonstrated that both the density of ticks and the prevalence of *Borrelia burgdorferi* infection in those ticks are elevation dependent, although these have been conducted in areas outside of the United States. In a study of *Ixodes ricinus* ticks in Europe (the European tick vector of Lyme disease), Jouda and others (2004) found decreasing tick population density, decreasing *B. burgdorferi* infection prevalence, and delayed onset of tick season with increasing elevation. Additionally, Burri (2007) found that *B. burgdorferi* infection prevalence of *I. ricinus* nymphs and adults both decreased with increasing elevation. Studies in central Europe have found that *I. ricinus* ticks have been sampled in field studies in areas at considerably higher elevations in 2001 and 2002 than they had been at the same locations in field studies in 1977 and 1979-80 (Gray et al. 2009).

Based on these assertions, it is reasonable to expect that tick vectors in the United States will likely be able to survive at higher elevations as temperatures warm, increasing the risk for tick bites in higher elevation areas where previously ticks were not commonly encountered.

Northern Range Expansion of Blacklegged Ticks

Climate change has been broadly projected to expand the maximum latitude and elevation at which important disease vectors can thrive, due to warming temperatures (Sutherst 2004). This may facilitate the expansion of new vector species into areas that they did not previously exist, or only existed sparsely.

A recent modelling study of tick dynamics suggests that the number of offspring that each blacklegged tick generates may increase substantially in the Northeastern United States, the Midwest, Quebec, and Ontario. In the Northeast specifically, (quantified by the authors in Lyme, CT) this reproductive number is expected to increase by more than double: from 3.1 offspring per tick to 7.1 by 2051-2069, under a higher greenhouse gas emissions scenario (A2).[3]

[3] Emissions scenarios describe how variations in future releases of greenhouse gases into the atmosphere will affect climate patterns. These are used because climate change in the future is not only dependent on the net amount heat-trapping greenhouse gases that humans have already emitted into the atmosphere, but also on the amount of these gases that are emitted or sequestered in the future. The A2 greenhouse gas emissions scenario, described by Nakicenvoic et al. (2000) in the International Panel on Climate Change (IPCC) Special Report on Emissions Scenarios (SRES), is at the higher end of the SRES emissions scenarios (though not the highest). The Vermont Climate and Health Program utilizes the A2 and B1 (a lower emissions scenario) in assessing future effects of climate on health.

This type of increase was relatively consistent across the Northeast in the modeled maps provided in the study (Ogden et al. 2014). This modelling effort supports the hypothesis that warming temperatures helped drive the spread of Lyme disease, along with reforestation, rising deer populations and other factors (Ogden et al. 2014). The projected increase in the number of offspring that each tick generates implies that climate and ecosystem changes carry three important implications:

1) Areas that are, at present, climatically unsuitable as blacklegged tick habitat may become infested in the future (i.e. areas currently too cold or dry to have sustainable blacklegged tick habitat may become warmer and wetter in the future, improving their suitability to harbor sustained blacklegged tick populations);
2) In regions already suitable for blacklegged ticks, but not yet infested, the rate of invasion will accelerate; and
3) Tick abundance may increase in areas where the ticks are already present (Ogden et al. 2014).

Based on these findings, blacklegged ticks in Vermont may become more abundant where they are already present and may become more widely distributed as they move into higher latitudes. A greater number and distribution of ticks could lead to greater risk for disease transmission (Ogden et al. 2014).

Seasonal Activity of Blacklegged Ticks and Mammal Hosts

In addition to an expansion of tick habitat and an increase in tick abundance, climate change may affect the length and timing of seasonal activity for ticks, as well as the mammal hosts that act as reservoirs for pathogens. Blacklegged ticks are not active when temperatures are below freezing (Ogden et al. 2008). With warming average temperatures, potential days of activity and by extension potential days of possible human exposure are expected to increase. However, the dynamics of Lyme disease transmission are extremely complex and dependent on multiple factors, such as the population dynamics of rodents that act as reservoirs for pathogens like *Borrelia,* and the availability of food for these hosts (Ostfeld et al. 2006). These factors may also be affected by temperature changes (Ogden et al. 2008).

Seasonal temperature changes may affect the general seasonality of the tick life cycle. For example, tick nymphs may begin to be active and start biting earlier in the spring than they did previously. Higher temperatures have been associated with more rapid development rates for *Ixodes* ticks (Ostfeld and Brunner, 2015). Temperature also affects tick questing (host-seeking) activity. On the low end of temperatures, Duffy and Campbell (1994) found that adult blacklegged ticks on Long Island, NY, began questing when temperatures reached or exceeded 4^{o} C (~40^{o}F). However, very high temperatures in summer may reduce nymph tick questing activity and may affect the immune response in some ticks against pathogens, reducing their vector capacity (Ostfeld and Brunner, 2015). So, increases in temperature may increase the risk early in the season, but potentially lead to a decreased risk of tick bites during very hot days in the summer. In general, the U.S. Global Change Research Program concluded with high confidence that it was likely that warming temperatures associated with climate change in the winter and spring will to earlier blacklegged tick activity, which would correspond to an earlier annual onset of Lyme disease cases (USGRP, 2016).

Humidity

Changes in precipitation, humidity, and soil moisture may also have effects on tick survival, reproduction, and activity, although these mechanisms are less well understood than those related to temperature. For instance, Vail and Smith (1998) found that ticks become more active when relative humidity is higher. This finding was corroborated by Berger and others (2014), who found that the seasonal abundance of blacklegged ticks was associated with the relative humidity in the leaf litter where ticks rest. Specifically, the number of long dry events (more than 8 hours below 82% relative humidity in leaf litter) was associated with decreased tick abundance (Berger et al. 2014). The hypothesized reasoning for this was that blacklegged ticks are susceptible to desiccation and can die off if they dry out. We are unable to project how climate change will affect relative humidity at the leaf-litter level, due to the complex interactions of precipitation, vegetative cover characteristics, soil saturation, and temperature that go into such a projection. This is further complicated because while we expect a greater overall amount of precipitation in Vermont, we expect more precipitation to fall in heavy events and less in light events. During heavy rain events, a greater proportion of the water tends to become runoff compared to an equivalent amount of rain in light rain events, where more water can sit and soak into soil and leaf litter. This may have implications for soil and leaf litter humidity and, in turn, tick activity and survival.

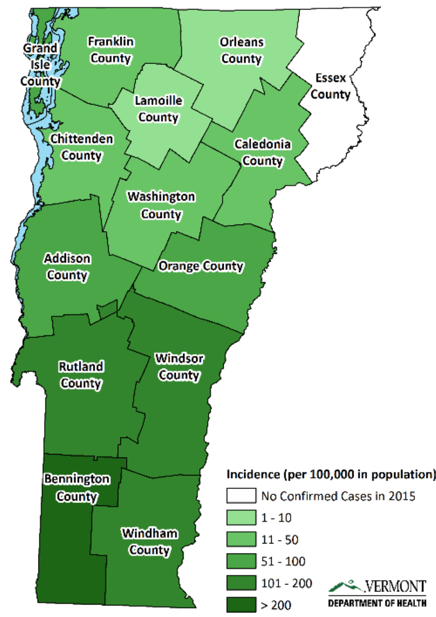
Analysis of climate associations with Lyme disease

Geographic Analysis

Background

Lyme disease incidence rates in Vermont are geographically heterogeneous, with county-level incidence being higher in more southern portions of the state historically (Figure 4). However, there have been no clear explanations for this heterogeneity. This analysis attempts to identify how different ecological factors may influence Lyme-disease rates in small geographic areas of the state (either individual towns or groups of towns, depending on population size). To do this, we analyzed associations between geographic Lyme disease case data for the years 2008 to 2014 and a variety of meteorological, environmental, and topographic indicator variables, at the county and sub-county level.

Figure 4 – County-Level Incidence of Confirmed Lyme Disease Cases Reported to the Vermont Department of Health, 2015



Methods

Data sources

Historic, daily temperature and precipitation data were provided by the PRISM Climate Group at Oregon State University to the Vermont State Climate Office at the University of Vermont. These data were averaged at the county level and used to derive meteorological variables representing average conditions over a 10-year period from 2004 (starting on 12/21/2003 to capture the start of the Winter season) to 2013 (ending on 12/20/2013). An average of ten years, starting earlier than the years where the Lyme disease incidence rates used as our outcome variable of interest, was used with the reasoning that some of the effects of average seasonal temperature and precipitation may be lagged for multiple years (*e.g.* warm conditions in a particular winter may lend themselves to more favorable conditions for white-footed mice that affect Lyme prevalence in ticks the following year, which may in turn cascade into effects in future years.) Ideally, this 10-year average captures possible cumulative effects of seasonal trends in temperature and precipitation. Data from 2014 was not included in our seasonal weather averages because full PRISM data for that year wasn't available at the time of analysis. However, because this analysis is concerned with long-term averages, we do not believe that this poses a major issue. Future updates to this analysis may reflect additional PRISM data availability. From these data, the following variables were derived:

- Mean seasonal temperatures: these variables represent the mean of the county-average daily temperature for all dates belonging to a particular season (e.g. winter, spring, summer, and fall) during the 10-year period.
- Mean annual temperature: this variable is the average of the four mean seasonal temperature variables for each county.
- Mean seasonal precipitation: these variables represent the average total precipitation during each season (e.g. winter, spring, summer, and fall) during the 10-year period for each county.
- Mean annual precipitation: this variable is the sum of the four seasonal precipitation variables for each county.
- Mean annual growing degree days (GDD), heating degree days (HDD), and cooling degree days (CDD): these variables were derived from county average temperatures, using the same formulas as are described in the Seasonality section below, but calculated as annual averages over the 10-year period.

Median latitude and elevation for each sub-county area (SCA, defined in the section below) were calculated using ArcGIS 10 (NAD 1983 State Plane Vermont FIPS 4400 Projection). Elevation data was adapted from a statewide digital elevation model (DEM) available from the Vermont GIS Clearinghouse. Forest cover data for each SCA was calculated using data from the National Land Cover Database, U.S. Geological Survey (2011 Edition, amended 2014).

Lyme disease cases for the years 2008-2014 were from the Vermont Department of Health reportable disease database. Seven-year annual incidence rates for indigenous cases only (*ie.* cases where the suspected infection occurred within the individual's county of residence) were calculated from these data. All cases reported between these years that had county of residence information and that were suspected to be indigenous to the county of residence (*n*=2,757, 72.6% of all reported cases during the time period) were included in the calculation of county-level indigenous incidence rates.

Sub-county Areas (SCAs)

In order to better understand how smaller scale conditions (e.g. forest cover and average elevation) affect Lyme disease rates, sub-county areas (SCAs) were also included in a secondary analysis. These areas were originally developed for use in reporting cancer incidence at a higher geographic resolution than the county level. The populations of these areas range from ~3,500 to ~42,000. These areas are designed to be as small as possible, but still have a large enough sample size to be able to calculate accurate rates. The areas used in this analysis include individual towns or groups of towns. For the purpose of this analysis, because the town of residence was reported as a free-text field, cities and towns where the same name might cause ambiguities in the precision of reporting (Barre City &Barre Town; Rutland City & Rutland Town; Newport City & Newport Town, and St. Albans City & St. Albans Town) were grouped together.

Incidence Rate Calculation

Crude and age-adjusted incidence rates were calculated for both counties and sub-county areas. Rates were calculated for the seven-year period of 2008 to 2014 and represent average annual incidence. This seven-year period was used to maximize statistical power, starting from the first year that both confirmed and probable cases were counted (2008), and extending to 2014, which was the most recent year for which summary data were available at the time of the analysis. Age-adjusted rates were adjusted to the 2000 U.S. Standard Population, consistent with the age-adjustment methods used in the Environmental Public Health Tracking (EPHT) and Healthy Vermonters 2020 online portals. Age-adjustment was based on 17 age categories, in five-year increments (0-4, 5-9, 10-14, … 80-85, 85+). County population estimates from the 2010 U.S. Census were used as denominators to calculate all rates.

Models

We used linear regression analysis to determine the association between Lyme disease incidence rates at both the county level (primary analysis) with meteorological indicator variables and the sub-county level (secondary analysis) with local environmental characteristics. Incidence rates were log-normalized so that they would be appropriate for use in linear regression.

Variables used in the analysis were:

- County-level variables: mean seasonal precipitation (winter, spring, summer, fall), mean seasonal temperature (winter, spring, summer, fall), mean annual growing degree days (GDD), mean annual heating degree days (HDD), mean annual cooling degree days (CDD)[4], mean annual precipitation, and mean annual temperature. All averages were for the ten-year range described in the "Meteorological Variables" section above.
- SCA-level variables: median latitude, median elevation, percent forest canopy coverage.

County-level models:

Simple univariate linear regression models were used to determine the association between log-normalized age-adjusted county Lyme disease incidence with each of our meteorological variables of interest.

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[4] Degree days are the sum of the difference, in degrees, of the average ambient air temperature on a given day beyond a particular threshold temperature. Traditionally, degree days use these threshold temperatures and caps on both the minimum and maximum temperatures, which are averaged to provide the growing degrees (or heating degrees, etc.) for a given day.

So, in our case the growing degrees on a given day can be defined as: (continued on next page)

$$Growing\ Degrees = \frac{T_{min}+T_{max}}{2} - Base\ Temperature$$

where T_{min} is the minimum temperature and T_{max} is the maximum temperature, which are both capped at a minimum of 50°F and a maximum of 86°F, respectively. Growing degree days are traditionally based on agricultural production of corn crops, which do not grow below 50°F (the base temperature). Additionally, temperatures above 86°F may be stressful to corn plants, and provide no additional benefit to growth.
CDD are calculated similarly to GDD. However, the CDD calculation uses a base temperature of 65˚F, with no upper threshold, whereas the GDD calculation uses an upper threshold of 80˚F.

A similar formula is applied to heating degree days, which acts as a metric for the overall severity of winter temperatures, representing how much indoor heating must be typically done to maintain comfortable temperatures:

$$Heating\ Degrees = Base\ Temperature - \frac{T_{min} + T_{max}}{2}$$

SCA-level models:

We performed the SCA-level model analysis in multiple steps. The first involved univariate linear regression analyses to determine the strength of association between our SCA-level topographic and environmental variables (median latitude, median elevation, and percent forest canopy coverage) with Lyme disease incidence. After this, we used a multivariate analysis including both latitude and elevation variables, because the association between elevation and Lyme disease incidence may be confounded by latitude, as the distribution of mountainous areas is not equal across the northern and southern portions of the state. This was followed by another multivariate analysis including all three of these variables.

Beta values, confidence intervals, p-values, and coefficients of determination (R^2) were used to assess the strength and statistical significance of the associations for all models.

Results

County-level Analysis

Figure 5: Age-adjusted annual Lyme disease incidence by county, 2008-2014. Includes only indigenous cases (cases suspected to have been acquired within the county of residence, n=2757).

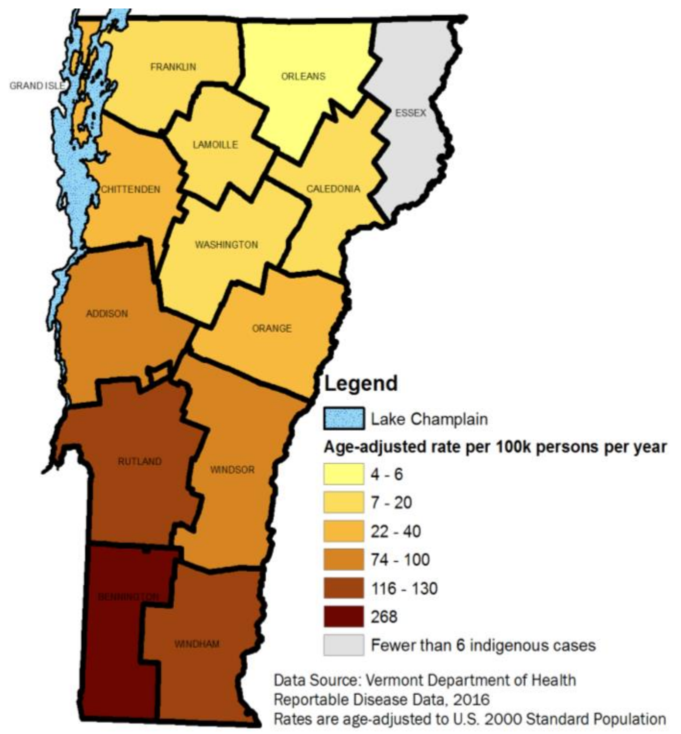


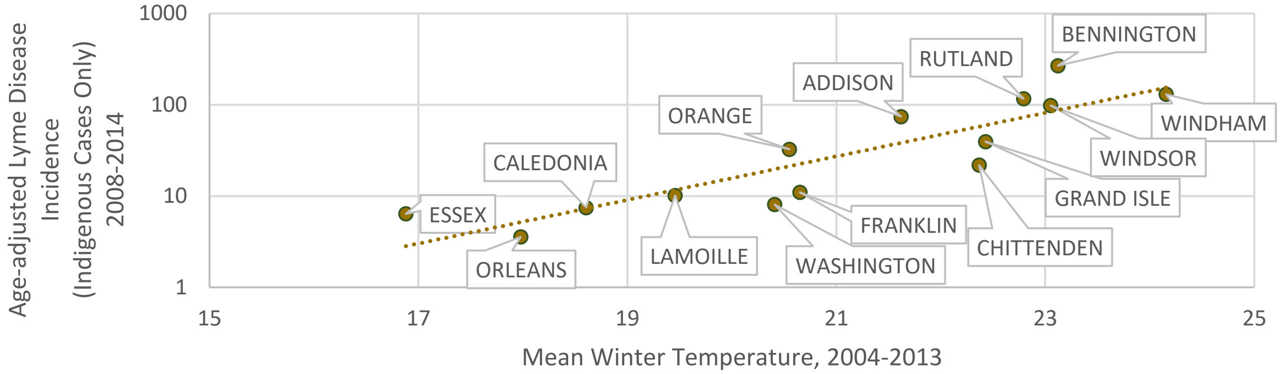
Figure 5 shows a map of county-level age-adjusted annual incidence of Lyme disease for the years 2008 to 2014. These are the values that were used in the dependent variables in the county-level models detailed below.

Table 1 shows the results of the univariate linear regression models using 10-year meteorological averages to predict log-normalized, age-adjusted Lyme disease incidence. Among temperature indicators, mean winter temperature ($p>0.0001$) had the strongest association with Lyme disease incidence, with warmer average temperatures being associated with higher incidence. Other temperature indicators that are associated with winter temperatures had significant, but weaker associations with Lyme incidence. These included: heating degree days ($p=0.0026$), mean fall temperature ($p=0.0068$), mean spring temperature ($p=0.0374$) and mean annual temperature ($p=0.0084$). In addition, mean winter precipitation had a weakly significant ($p=0.0452$) positive association with Lyme disease incidence.

Table 1 - Model Parameters for Weather Averages Predicting Log-normalized Age-Adjusted Lyme incidence, 2008-2014. Rates used in models only contain cases suspected to have been contracted within the county of residence. Variables with statistically significant associations ($p < 0.05$) are shown in bold.

Covariate	Parameter Estimate	95% Confidence Bounds of Parameter Estimate		P-value	Model R^2
		Lower	Upper		
Mean Seasonal Temperature					
Winter	**0.550**	**0.363**	**0.737**	**<.0001**	**0.77**
Spring	**0.473**	**0.033**	**0.914**	**0.0374**	**0.31**
Summer	0.297	-0.184	0.779	0.2031	0.13
Fall	**0.568**	**0.189**	**0.947**	**0.0068**	**0.47**
Mean Seasonal Precipitation					
Winter	**0.547**	**0.014**	**1.080**	**0.0452**	**0.29**
Spring	0.190	-0.832	1.210	0.6931	0.01
Summer	-0.293	-1.210	0.627	0.5012	0.04
Fall	0.523	-0.089	1.130	0.0872	0.22
Mean Annual Temperature and Precipitation					
Mean Growing Degree Days	0.003	0.000	0.007	0.0674	0.25
Mean Heating Degree Days	**-0.002**	**-0.003**	**-0.001**	**0.0026**	**0.54**
Mean Cooling Degree Days	0.007	-0.001	0.015	0.0914	0.22
Mean Annual Temperature	**0.542**	**0.167**	**0.917**	**0.0084**	**0.45**
Mean Annual Precipitation	0.111	-0.098	0.320	0.2697	0.10

Figure 6 – Graph of age-adjusted Lyme disease incidence (2008-2014, indigenous, within-county cases only) plotted against mean winter temperature (2004-2013)

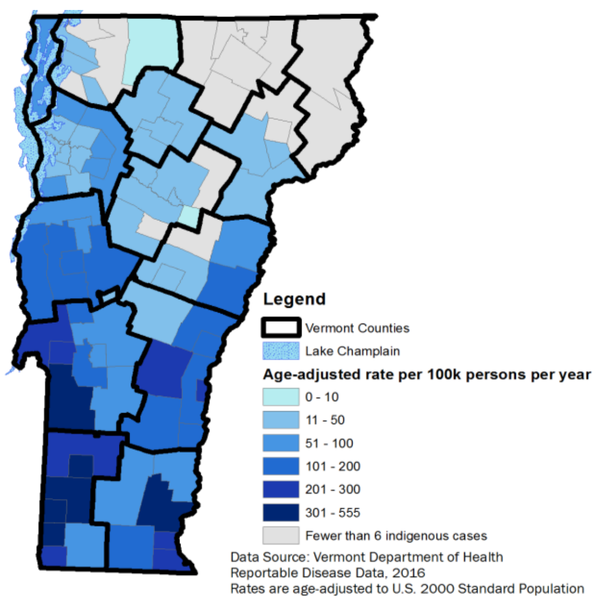


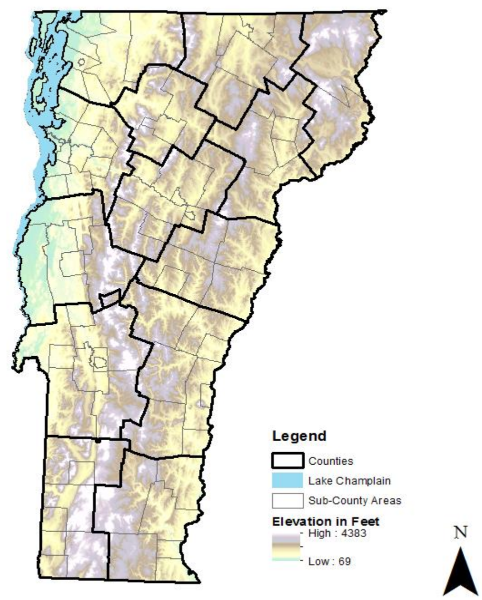
Sub-county-level Analysis

Figure 7 shows the age-adjusted annual Lyme disease incidence of Vermont SCAs for the years 2008 to 2014, the dependent variable in in the sub-county models analysis. For comparison, Figure 8 shows the sub-county areas superimposed over a Vermont elevation map.

Figure 7 - Vermont annual Lyme disease incidence for indigenous, within county cases, by sub-county area, 2008-2014, age-adjusted to the 2000 U.S. Standard Population. Darker blue represents areas with higher incidence. Because incidence rates in this map include only indigenous cases, they should not be compared to other data within or outside of the state.

Figure 8 – Map of Vermont elevations. White denotes areas of highest elevation, green denotes areas of lowest elevation. Heavy black outlines indicate counties, while grey outlines indicate sub-county area boundaries.





In univariate linear regression models, median latitude was strongly associated with Lyme disease incidence, but median elevation was not (Table 2). This was not surprising, given that latitude is such a strong predictor of Lyme disease incidence in Vermont, and there is a wide range of elevations in both northern and southern areas of the state (Figure 8). When including both elevation and latitude in the same model (i.e., the effect of elevation was tested while adjusting for latitude as a potential confounder), resulting in both variables being highly significant (Table 3), and the model fit (adjusted $R^2 = 0.665$) improving over that of the univariate model for elevation in Table 2. Both variables had inverse correlations with incidence, indicating that Lyme disease incidence is typically higher in southern areas of the state and in areas of lower elevation.

Percent forest coverage was not significantly associated with Lyme disease incidence in the univariate analysis (p=0.1345, Table 2). When included in a multivariate model with elevation and latitude, the association of forest coverage with Lyme disease was slightly stronger, but still not statistically significant (p=0.0747,Table 4). The inclusion of forest cover in the model with median elevation and latitude did not produce a much better fit (Adjusted Model $R^2 = 0.7298$, Table 4) than the model with median elevation and latitude alone (Adjusted Model $R^2 = 0.7224$, Table 3).

Table 2 - Univariate linear regression model parameters for variables predicting log-normalized, age-adjusted Lyme incidence, (indigenous cases only) 2008-2014.

Covariate	Parameter Estimate	95% Confidence Bounds of Parameter Estimate		P-value	Model R^2
		Lower	Upper		
Median Elevation (thousands of feet)	0.04964	-0.48173	0.58100	0.8531	0.0004
Median Latitude	-1.58120	-1.83278	-1.32961	<.0001	0.6531
Percent Forest Cover	0.00949	-0.00300	0.02198	0.1345	0.0268

Table 3 - Model parameters for multivariate linear regression analysis using elevation and latitude to predict log-normalized, age-adjusted Lyme disease incidence (indigenous cases only), 2008-2014

Covariate	Parameter Estimate	95% Confidence Bounds of Parameter Estimate		P-value
		Lower	Upper	
Median Elevation (thousands of feet)	-0.715	-1.01	-0.418	<.0001
Median Latitude	-1.77942	-2.01783	-1.54100	<.0001

Adjusted Model R^2 **= 0.7224**

Table 4 - Model parameters for multivariate linear regression analysis using forest cover, elevation, and latitude to predict log-normalized, age-adjusted Lyme disease incidence (indigenous cases only), 2008-2014

Covariate	Parameter Estimate	95% Confidence Bounds of Parameter Estimate		p-value
		Lower	Upper	
Median Elevation (thousands of feet)	-1.10	-1.61	-0.584	<.0001
Median Latitude	-1.735	-1.975	-1.495	<.0001
Percent Forest Cover	0.0114	-0.00116	0.0239	0.0747

***Adjusted Model R^2* = 0.7298**

Discussion

The county-level portion of this analysis indicates that the geographic pattern of Lyme disease incidence in Vermont may be partially explained by variations in meteorological conditions and topography. Specifically, counties with warmer winters tended to have higher Lyme disease incidence.

Among meteorological variables, average winter temperature, had the strongest association with Lyme disease incidence, while average fall temperature, heating degree days, and average annual temperature also had strong correlations. These variables are highly correlated with one another, as HDD is an indicator of average temperatures across the cold season (typically from late fall into early spring), and annual temperatures are related to all other temperature indicators.

These results are somewhat consistent with scientific literature, and there are two ways in which cold temperatures during the winter months may affect the *Borrelia* transmission cycle: 1) Reduced tick populations, 2) reduced white-footed mice populations, and 3) reduced tick activity.

> 1) Short-term exposure to extreme cold, below -15^{o} C, can be lethal to blacklegged ticks (Burks et al 1996, Vandyk et al. 1996, as summarized in Brunner et al. 2012). However, other factors such as the availability of insulation in the form of snow, leaf litter, and soil, may allow ticks to survive such cold temperatures (Brunner et al. 2012).
>
> 2) Extremely cold winters have been associated with decreased white-footed mouse survival (Subak 2003). Decreases in rodent population due to extreme cold may reduce the probability that tick larvae hatching in spring will find an infected rodent host to feed upon, leading to a reduction in the density of infected nymphs the following year (Subak 2003).
>
> 3) Ticks may reduce activity at lower temperatures: Duffy and Campbell (1994) found that adult blacklegged ticks on Long Island, NY, began questing for bites when temperatures reached or exceeded 4^{o} C (~40^{o}F), and did not quest at lower temperatures. Longer cold periods during winter may reduce the overall amount of time during the year that blacklegged ticks can quest for bloodmeals.

In the sub-county analysis, median latitude was a strong predictor of Lyme disease incidence, and median elevation was also a significant predictor, when included in a model with latitude. In this model, southern areas with lower median elevations were expected to have the highest incidence of Lyme disease. When included in models with elevation and latitude, forest cover was not a significant predictor of Lyme disease and only slightly increased the model fit. This suggests that the percent forest cover of a given SCA is not a good predictor of Lyme disease incidence within that area. This could be because forest cover in the area does not necessarily correlate to an individual's exposure to forest habitats in the county where blacklegged ticks are active. Additionally, areas with less total forest cover may merely have more fragmented forests, providing more forest edge habitat where people can get bitten by ticks.

The results of this analysis are limited by the fact that we did not have sufficient data to look at other potentially important factors, such as the annual crop of acorns produced by oak trees (which act as food sources for white-footed mice), and geographic trends in white-tailed deer and white-footed mouse populations may influence overall Lyme disease risk in a given area. Both acorn crop abundance (lagged two years) as well as mouse and chipmunk abundance (lagged one year) were significantly associated with the density of *B. burgdorferi*-infected blacklegged tick nymphs (an indicator of Lyme disease risk) in a 13-year study (1991-2004) of field sites in Duchess County, New York (Ostfeld et al. 2006).

There are two important implications of this analysis. The first is that, if winter temperatures are a limiting factor for Lyme disease transmission in Vermont, then warming winter temperatures due to climate change may increase the risk for Lyme disease transmission in areas that have been historically at low risk like Essex and Orleans counties. This may also be the case for other tickborne diseases that rely on similar disease transmission cycles, like anaplasmosis.

In a 1981-2010 baseline period, the western climate region of Vermont had average daily minimum temperatures during the winter months (December, January, and February) of 10.4˚F, and average daily maximum temperatures of 29.6 ˚F. In comparison, the cooler northeast climate region had average daily minimum temperatures of 6.9 ˚F and average daily maximum temperatures of 27.1 ˚F. However, by midcentury (2041-2070 projection period) we expect temperatures in the northeast region to rise to average daily minimums and maximums as high as 13.0 ˚F and 30.8˚F, respectively (Vermont Department of Health, 2016 - Appendix). This indicates that winter temperatures in Vermont's northeastern counties will be similar to those currently in western counties where the incidence of Lyme disease is currently higher, which may lead to a corresponding increase in tickborne disease risk.

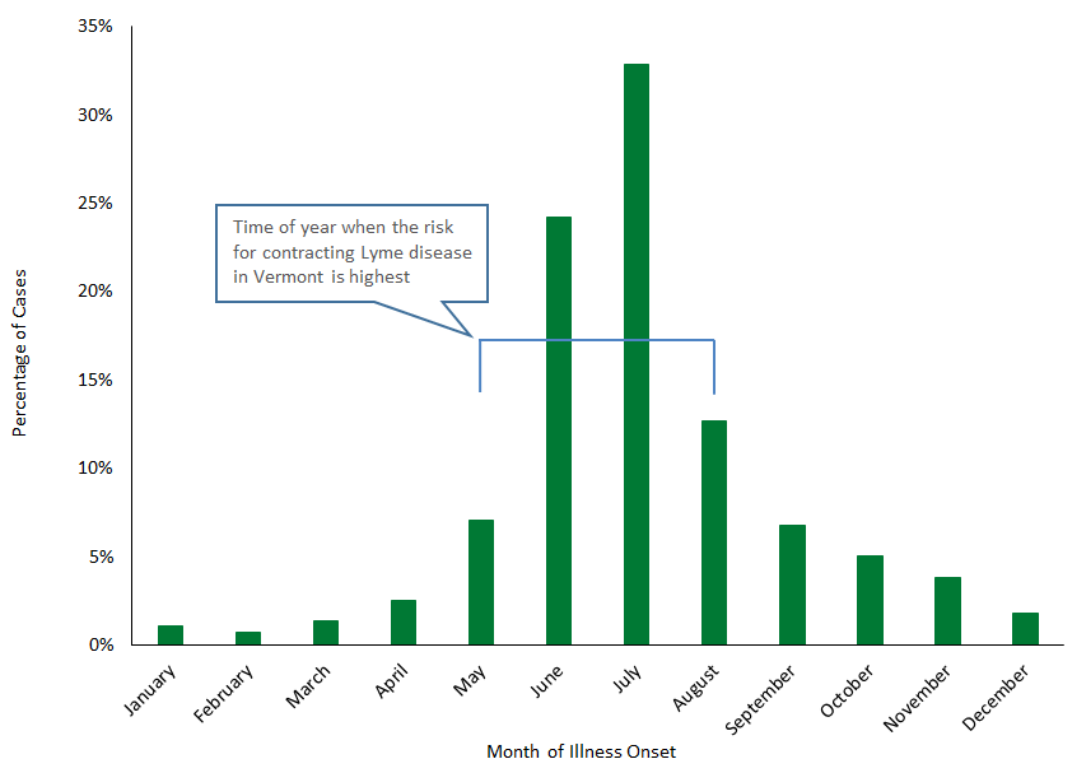
In areas that currently have the comparatively higher incidence of Lyme disease and other tickborne diseases in Vermont, like Bennington county, warmer temperatures may also have a similar effect of increasing this risk. However, if temperatures reach or exceed optimal levels for tick survival and reproduction in these areas, other environmental variables may become limiting for tickborne disease risk.

The second important result from this analysis is that higher elevation areas appear to have lower Lyme disease incidence. This result is limited by the fact that we do not know whether individuals living in areas with a higher median elevation were bitten and infected at those higher elevations. However, warmer temperatures may put higher elevation areas, which currently experience lower incidence rates of Lyme disease, at higher risk for tickborne diseases in the future. While it is generally expected that disease vectors may reach higher elevations than they previously did due to warmer temperatures from climate change (Mills et al. 2010), there have been no published scientific studies on this matter with regard to North American blacklegged ticks (*I. scapularis*). Because of this, understanding how climate change may affect risk for Lyme disease across elevation gradients may be an important realm for study in Vermont and the rest of the Northeast.

Seasonality

Lyme disease exhibits a distinct seasonal pattern in case reporting, as shown below for Vermont in Figure 9. From 2005-2015, onset dates of reported Lyme disease cases peaked in June and July. However, nymphs are active and host-seeking in Vermont as early as April or May, when temperatures start to become warm enough for consistent activity. This seasonal pattern of disease is related to the life cycle of blacklegged ticks. In the northern United States, it typically takes 2 years for these ticks to complete their life cycle. In this cycle, tick nymphs are most active and seeking blood meals from hosts in the spring and early summer. Nymphs are most important to the seasonal disease pattern because they are difficult to see on a person's body or clothes (more so than adults, which are far larger), and thus present the greatest risk for human infection.

Figure 9: Confirmed Cases of Lyme Disease Reported to the Vermont Department of Health by Month of Illness Onset, 2005-2015



However, reported symptom onset dates occur year-round, even in winter months. There are many possible explanations for cases with symptom onset in winter months: 1) infection may be occurring during particularly warm periods in these months, when temperatures reach or exceed thresholds for tick activity and ticks become active; 2) symptom onset may occasionally be delayed far greater than the typical 3-28 days; 3) individuals travel to southern states where warmer temperatures mean ticks are still active, and become infected in those states; 4) onset dates may occasionally be misreported to the state.

The precise timing of the start and peak of activity for tick nymphs depends on seasonal weather variables, including air and soil temperature, precipitation, and soil moisture. Blacklegged ticks may become active at relatively low temperatures: Duffy and Campbell (1994) found that adult blacklegged ticks on Long Island, NY, began questing when temperatures reached or exceeded 4˚ C (~40˚F). Ticks are also more active when humidity is higher, and the risk for desiccation is low (Vail and Smith 1998). Seasonal changes in meteorological conditions like temperature and precipitation are thus key determinants of the general activity of ticks and the timing of the emergence of nymphs, and therefore the risk of tickborne disease. On the human end of the tickborne disease cycle, seasonal weather factors can affect the amount of time a person spends outside in areas infested with ticks. The period for peak nymph activity corresponds to times when people spend the most time out of doors, adding to the potential for contact between humans and questing ticks.

While it is important to communicate the fact that the risk of tickborne diseases is present year-round so long as it is warm enough for ticks to be active, understanding the dynamics of the peak season when the majority of Lyme disease (and anaplasmosis) cases occur is critical. Meteorological factors that may be affected by climate change have been associated with the annual timing of peak nymph activity and Lyme disease case reports. Moore et al. (2014) found that the start of Lyme disease season (defined approximately as when the week-by-week increase in cases is highest) in 12 states[5] was significantly associated with a variety of temperature and precipitation indicators. Specifically, a higher number of growing degree-days

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[5] *North:* Maine, Massachusetts, New Hampshire; *East:* Connecticut, Rhode Island, New Jersey, New York, Pennsylvania; *South:* Maryland, Virginia; *Midwest*: Minnesota, Wisconsin

(GDD[6]) above 10˚C (~50˚F) in the first 20 weeks of the year was associated with an earlier start to the Lyme disease season. The authors hypothesized that the relationship they found between higher cumulative GDD and an earlier start to the season was due to increased development rates of larvae into nymphs, as well as increased ambient temperatures in early spring, which is associated with greater host-seeking activity (Moore et al. 2014). In contrast, higher cumulative precipitation after the eighth week of the year was associated with a later start to the Lyme disease season. The hypothesized reason for this was that heavy rainfall events may impede the ability for tick nymphs to quest for bloodmeals, which may delay the start of the Lyme disease season in humans (Moore et al. 2014). When this analysis was stratified into four geographic regions[5], the results varied by region. In particular, the association between cumulative GDD and the start of Lyme disease season was not present in the Northern states, though Vermont was not included in the analysis. The authors hypothesized that this may have been due to the lower inter-annual variability in GDD in the northern states, compared to other states. The authors did not find an association between meteorological variables and the end of the Lyme disease season (Moore et al. 2014).

The findings on GDD early in the season are consistent with observations by Diuk-Wasser and others, who found earlier timing of peak host-seeking activity in ticks in warmer southern states than in northern states (Diuk-Wasser et al. 2006, as referenced in Eisen et al. 2016). Other factors, such as the precipitation and saturation deficit of specific weeks early in the year were also associated with the timing of the start of Lyme disease season (Moore et al. 2014).

To test these associations in Vermont, we conducted an analysis of the association between the timing of Lyme disease and meteorological variables, using a methodology similar to that used by Moore and others (2014).

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[6] Growing degree days are typically calculated by summing the daily number of degrees above a certain threshold temperature, in this case 50°F, with a maximum capped at 86°F, during a particular period of time.

if $T_{mean} \geq 50^{o}\text{F}$, $GDD = T_{mean} - 50^{o}\text{F}$

if $T_{mean} < 50^{o}\text{F}$, $\text{GDD} = 0$, *Methods*

Weekly Lyme disease case counts were calculated for the years 2005 to 2014 for each county in Vermont. Data from 2005 to 2007 were included, in spite of the lack of probable case inclusion, for the purpose of maximizing statistical power. Because understanding the timing of Lyme disease cases was necessary for this analysis, only cases with a reported onset date of symptoms (n=3283, 80% of all reported cases) were included in the analysis. If there were fewer than 10 cases with onset dates in a given year for a county, data for that county and year were excluded from analysis, to ensure that each data point was able to be representative of a sufficient number of cases to be methodologically valid. In all, there were data from 50 county-years included in the analysis. For each county-year, the week where 20% of that year's cumulative number Lyme disease cases was reached was determined, as an approximation of the start of the peak Lyme disease season. For the sake of brevity, this point will hereafter be referred to as the start of peak Lyme disease season.

Meteorological variables included in the analysis were adapted from PRISM Climate Group data (Oregon State University; http://www.prism.oregonstate.edu/) provided to us by the Vermont State Climate Office at the University of Vermont. These data included daily, historic temperature and precipitation data that were averaged to the county level. These data were used to derive cumulative growing degree days (GDD) in the first 80, 100, and 120 days of the year, cumulative heating degree days (HDD) in the first 80, 100, and 120 days of the year, and cumulative precipitation in the first 80, 100, and 120 days of the year[7]. In addition, we used a

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[7] Degree days are the sum of the difference, in degrees, of the average ambient air temperature on a given day beyond a particular threshold temperature. Traditionally, degree days use these threshold temperatures and caps on both the minimum and maximum temperatures, which are averaged to provide the growing degrees (or heating degrees, etc.) for a given day.

So, in our case the growing degrees on a given day can be defined as

$$Growing\ Degrees = \frac{T_{min}+T_{max}}{2} - Base\ Temperature$$

where T_{min} is the minimum temperature and T_{max} is the maximum temperature, which are both capped at a minimum of 50°F and a maximum of 86°F, respectively. Growing degree days are traditionally based on corn plants, which do not grow below 50°F (the base temperature). Additionally, temperatures above 86°F may be stressful to corn plants, and provide no additional benefit to growth. These growing degrees are then summed for the first 80, 100, and 120 days of the year to obtain our GDD metric.

variation on GDD, which will be called "tick degree days (TDD)", as a measure designed to be appropriate to tick activity, rather than plant growth, as is the case for GDD[8]. We used three different lengths in the beginning of the year for the purposes of comparison and sensitivity testing.

Univariate multilevel linear regression analysis (using PROC MIXED in SAS 9.3[9]) was used to determine the association between meteorological variables with the week of the year when 20% of cases were reached. Because there was a change in Lyme disease reporting from 2008 onwards, we had to adjust our models for whether the county-year of interest belonged to the year 2005 to 2007. We did this using a dummy indicator variable. Only confirmed cases of Lyme disease were reported to the state prior to 2008, whereas since 2008, both probable and confirmed cases have been reported. Because our analysis included both probable and confirmed cases for the years 2008 to 2014, we had to adjust for this issue with prior years. Each variable was analyzed separately, in initial models, and county-years were nested within the counties which they belonged to. Model fit was determined using Aikake Information Criterion

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A similar formula is applied to heating degree days, which acts as a metric for the overall severity of winter temperatures, representing how much indoor heating must be typically done to maintain comfortable temperatures:

$$Heating\ Degrees = Base\ Temperature - \frac{T_{min} + T_{max}}{2}$$

Where T_{min} and T_{max} are both capped at 65°F, which is also the base temperature. These are then summed for the first 80, 100, and 120 days of the year to obtain our HDD metric.

[8] TDD are calculated identically to GDD, however the base temperature is 40°F, representing the lower limit for tick questing activity observed by Duffy and Campbell (1994), on which Where T_{min} and T_{max} are both capped. There is no maximum cap on T_{min} and T_{max} in the TDD metric.

[9] Multilevel mixed models were designed to account for the nesting of data, in this case, multiple years of data for each county. An example of SAS code used in our analysis is provided in the footnote on the following page:

```
SAS Code Example:
proc mixed data = stats  covtest ;
  class county;
  model start_week = heating_degree_days_80a Year2005_2007
                / solution ddfm = bw notest cl;
  random intercept / subject = county type = un;
run;
```

(AIC) values. Meteorological variables that were significantly associated with the start of the Lyme disease season in univariate models were grouped into a final multivariate model.

Results

Statewide, the start of peak Lyme disease season was reached, on average, 23 weeks into the year (median=23). Among the 50 county-years included in the analysis, this point was reached as early as 17 weeks into the year, and as late as 27 weeks.

Table 5 shows which Vermont counties had ten or more Lyme disease cases in a given county-year during our study period. Table 6 shows the descriptive statistics for the meteorological variables used in the regression models.

Table 5 - County-years with 10 or more Lyme disease cases that include symptom onset date information

County	Number of County-Years
Addison	7
Bennington	10
Chittenden	7
Franklin	1
Orange	3
Rutland	7
Windham	8
Windsor	7

Table 6: Descriptive Statistics for Meteorological Variables (n=50 county-years)

Variable	Time Frame	Mean	Median	Maximum	Minimum	Standard Deviation
Heating Degree Days	First 80 Days	3484.2	3494.0	4164.9	2718.5	324.6
	First 100 Days	4062.0	4098.0	4844.1	3186.4	381.9
	First 120 Days	4470.4	4506.8	5355.4	3527.1	402.2
Growing Degree Days	First 80 Days	12.4	6.6	61.5	0.0	15.1
	First 100 Days	36.6	21.4	126.1	1.0	35.7
	First 120 Days	121.7	113.6	241.8	57.7	44.0
Tick Degree Days	First 80 Days	59.2	52.3	149.5	9.6	30.5
	First 100 Days	629.7	611.9	789.2	538.1	62.3
	First 120 Days	307.7	298.6	523.9	172.6	77.6
Total Precipitation (inches)	First 80 Days	7.7	7.2	17.1	3.7	2.4
	First 100 Days	9.6	9.6	19.5	4.7	2.5
	First 120 Days	12.2	11.8	21.9	6.3	3.1

Table 7 shows the model results for each of the meteorological variables used to predict the timing of the start of peak Lyme disease season. Total precipitation in the first 80 days of the year had the strongest association with the timing of the start of peak Lyme disease season (p=0.0009, AIC=211.9), such that more precipitation was associated with a later start of the peak Lyme disease season. More precipitation in the first 100 and 120 days were similarly associated with a later start of the peak Lyme disease season, but had slightly weaker associations, based on their higher AICs and p-values (Table 7).

In contrast, cumulative growing degree days in the first 80 days had a significant negative association with this timing, (p=0.0021, AIC=220.8). So, more growing degree days were associated with an earlier start to the peak Lyme disease season. Similar but slightly weaker associations were found for cumulative growing degree days in the first 100 and 120 days. Cumulative heating degree days were not significantly associated with the timing of the start of peak Lyme season.

Table 7 – Model results for meteorological variables predicting the start of peak Lyme disease season. Statistically significant variables with the lowest AIC values for their categories are shown in bold

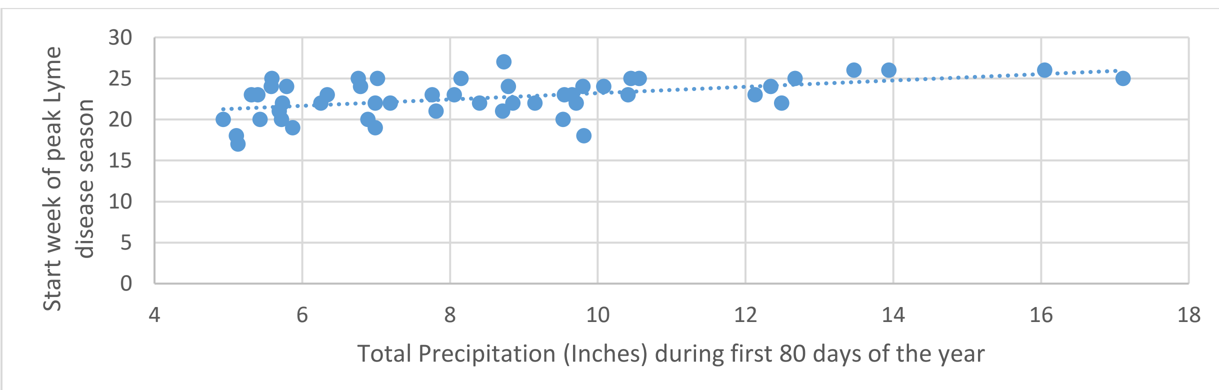
Variable	Time Frame	Parameter Estimate	95% Confidence Bounds of Parameter Estimate		p-value	AIC
			Lower	Upper		
Heating Degree Days	First 80 Days	0.00087	-0.00147	0.003212	0.4565	231.8
	First 100 Days	0.00114	-0.00081	0.0031	0.2438	231.4
	First 120 Days	0.00097	-0.00098	0.002908	0.3213	231.8
Growing Degree Days	**First 80 Days**	**-0.0552**	**-0.08602**	**-0.0243**	**0.0008**	**215.5**
	First 100 Days	-0.0211	-0.03491	-0.00724	0.0037	220
	First 120 Days	-0.0137	-0.02624	-0.00106	0.0343	224.4
Tick Degree Days	**First 80 Days**	**-0.0256**	**-0.04259**	**-0.00853**	**0.0042**	**219.9**
	First 100 Days	-0.0124	-0.02091	-0.00397	0.005	221.6
	First 120 Days	-0.0092	-0.01676	-0.00162	0.0185	224.3
Total Precipitation	**First 80 Days**	**0.3584**	**0.1567**	**0.5601**	**0.0009**	**211.9**
	First 100 Days	0.3104	0.1262	0.4947	0.0015	213.1
	First 120 Days	0.2714	0.1048	0.438	0.0021	213.9

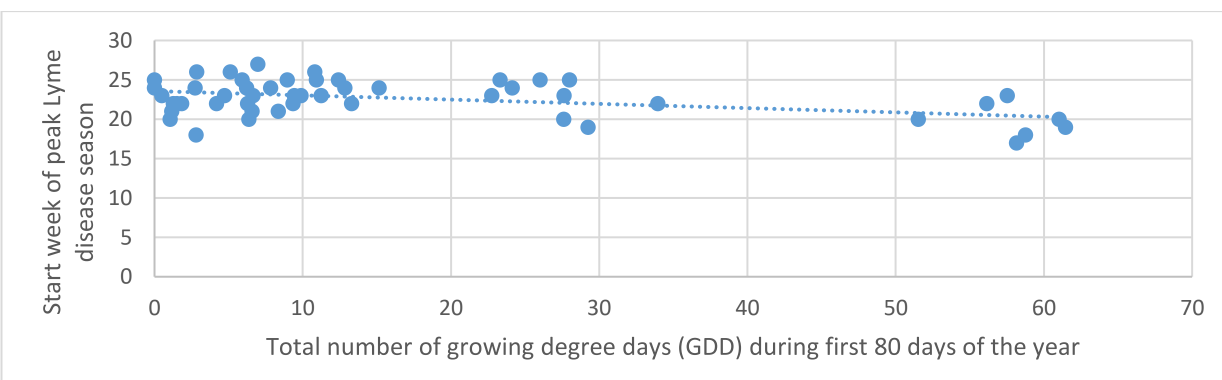
We used a multivariate linear regression model to assess the association both of total precipitation and cumulative GDD in the first 80 days of the year (Table 8). In this model, total precipitation still had a strong, statistically significant, positive association with the timing of tick season (p= 0.011). The parameter estimate in this model indicates that for roughly every four (3.76) inches of precipitation that occurred in the first 80 days of the year, the expected start of the peak tick season was delayed by a week. Cumulative GDD in the first 80 days of the year also still had a significant negative association with the timing of the tick season (p=0.011, Table 8). This indicates that for every ~24 (24.34) additional GDD that occurred in the first 80 days of the year, the peak Lyme disease season started one week earlier.

Table 8 – Combined linear regression model results: Total precipitation and cumulative growing degree days in the first 80 days of the year

Variable	Parameter Estimate	95% Confidence Bounds of Parameter Estimate: Lower	95% Confidence Bounds of Parameter Estimate: Upper	p-value	Model AIC
Intercept	20.9312	18.57	23.29	<.0001	211.5
Growing Degree Days (First 80 Days)	-0.041	-0.072	-0.010	0.011	
Total Precipitation (First 80 Days)	0.266	0.064	0.47	0.011	
Year 2005-2007	1.852	-0.18	3.88	0.073	

Figure 10 – Association between start of peak Lyme disease season with total precipitation and growing degree days (GDD) during the first 80 days of the year





Our results are consistent with other literature on the seasonality of Lyme disease. Moore and others (2014) also found similar associations between more growing degree days early in the year and an earlier start to the Lyme disease season. Similarly, they also found that increased total precipitation was associated with a later start to the tick season. Unlike that study, we were unable to assess saturation deficit and humidity in this analysis due to a lack of suitable data available to us at the time of analysis. These results are limited, in that we were not able to distinguish between snowfall and rainfall in our historic data, although most precipitation during the first 80 days of the year in Vermont during the years studied would occur predominantly as snow.

These results indicate that climate change may have conflicting effects on the start of the Lyme disease season in Vermont. In the future, we expect warmer winters (increasing the number of GDD), as well as greater total precipitation. With climate change, by the end of the century precipitation in December, January, and February (a close proxy for our first 80 days of the year indicator) is expected to increase by about 1 inch compared with the current period. We are currently not able to project the increase in GDD that will occur in the first 80 days of the year due to data limitations. The interactions between warmer temperatures and increased precipitation are also difficult to predict, because even if there is an increased amount of snow, it may tend to melt more rapidly due to a trend of warmer temperatures.

Conclusions

Currently, the scientific literature surrounding the effects of climate change on tickborne diseases (with a focus on Lyme disease) in the United States supports the hypothesis that temperature (especially winter minimum temperature) is a strong predictor of where blacklegged ticks can live, and the effect of warming temperatures on blacklegged tick populations has been a northward expansion of the ticks' range into Canada (USGCRP, 2016). Furthermore, winter temperatures may also be a limiting factor in Vermont and surrounding regions for population density of white-footed mice. With shorter winters also resulting in a longer tick activity season, these changes may contribute to an increased risk for diseases transmitted by blacklegged ticks in the future and may be a contributing factor to the observed increases in Lyme disease and anaplasmosis that have occurred in recent years. Other contributing factors may include forest fragmentation, other factors that may influence population of tick hosts like white-footed mice and white-tailed deer, as well as increased recognition of cases by physicians and reporting of cases to the Department of Health.

In our Vermont-specific analysis, we found a strong correlation between Vermont county Lyme disease incidence with warmer average winter temperatures. This indicates that if winter temperatures are a limiting factor for *Borrelia* transmission, there is an opportunity for tick populations to further increase in number and expand in range Vermont, especially in areas such as the Northeast Kingdom, where extreme winter temperatures may currently be a limiting factor for sustained Lyme disease transmission. Additionally, we found that higher elevation areas tend to have lower Lyme disease incidence. As temperatures warm, these areas may become suitable habitats for blacklegged tick populations, presenting more areas where people and their pets may encounter them.

The rapid increase in cases of anaplasmosis, which is occurring several years after the increase in cases of Lyme disease, also raises concerns about the introduction and expansion of other tickborne pathogens into the state. For example, ticks infected with *Babesia microti* are just starting to be detected in ticks collected and tested through Vermont's surveillance efforts (see Appendix). In addition, the spread of ticks, such as the lone star tick (*Ambloymma americanum*), which transmits ehrlichiosis (*Ehrlichia spp.*) may be of concern as temperatures continue to rise, increasing the likelihood that the species becomes established in Vermont. Developments in other New England states may provide additional insight into how climate change may affect the abundance of different species of tick in Vermont.

Given the complexity of this system, there is some uncertainty as to how much warming temperatures and changing precipitation patterns due to climate change will affect the actual incidence of tickborne diseases in the future (USGCRP, 2016). However, the literature review and the analyses highlighted in this document support the importance of actions currently being taken to help prevent Lyme disease, anaplasmosis, and other tickborne diseases in Vermont. Ongoing actions include raising awareness, providing education about tick bite prevention and what to do in the event of getting bitten by a tick, as well as surveillance of tick distributions and tickborne pathogens in partnership with the Vermont Department of Agriculture and Vermont academic institutions. In the future, these actions can be supplemented with additional analyses to improve our understanding of current and future risks, and to inform the design of effective interventions for reducing these risks.

Appendix I - Ticks and Tickborne Diseases of Concern in Vermont

Ticks that Can Carry Disease in Vermont

Currently, there are four species of tick in Vermont that can cause diseases in humans (Table 9). These are the blacklegged tick (*Ixodes scapularis*, also commonly called the "deer tick"), the American dog tick (*Dermacentor variablis*), the woodchuck tick (*Ixodes cookei*, also commonly called the "groundhog tick" or "Packard tick"), and the Lone star tick (*Ixodes scapularis*), which is not commonly found Vermont.

This section gives a brief overview of these four species of tick, and diseases of concern that are associated with them.

Table 9 – Tickborne diseases of concern in the Northeastern United States

			Transmitted by			
Disease	**Pathogen**	**Reported Cases from tick bites in Vermont in last 10 years (through 2016)?**	**Blacklegged tick**	**American dog tick**	**Woodchuck tick**	**Lone star tick***
Lyme Disease	*Borrelia burgdorferi*	Yes	X			
Anaplasmosis	*Anaplasma phagocytophilum*	Yes	X			
Babesiosis	*Babesia microti*	Yes	X			
Powassan virus disease	Powassan Virus (POWV) / Deer Tick Virus (DTV)	No	X		X	
Erlichiosis	*Erlichia* spp.	Yes				X

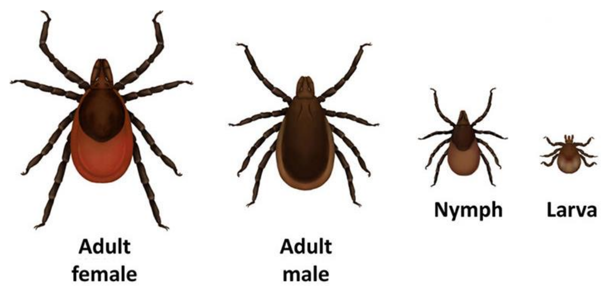
Rocky Mountain Spotted Fever	*Rickettsia rickettsia*	No		X		
Tularemia	*Franciscella tularensis*	No				X
Spotted Fever Group Rickettsiosis (SFGR)*	*Rickettsia spp.*	Yes		X		Possibly
Borrelia miyamotoi	*Borrelia miyamotoi*	Yes				

*No description included in this appendix. For more information on SFGR, please consult the Centers for Disease Control's website on SFGR diseases at: https://www.cdc.gov/otherspottedfever/ . For more information on *Borrelia miyamotoi* please consult the Health Department's website on *B. miyamotoi* at: http://www.healthvermont.gov/disease-control/mosquito-tick-zoonotic/borrelia-miyamotoi.

Blacklegged Ticks or "Deer Ticks" (Ixodes scapularis)

Blacklegged ticks are the primary vectors of Lyme disease, babesiosis, and anaplasmosis in the eastern United States. They also transmit the deer tick virus (DTV), a strain of the Powassan virus. Blacklegged ticks are widespread throughout the eastern United States and the Midwest (Figure 12). They are the most commonly encountered ticks in Vermont.

Figure 11 – Diagram of blacklegged tick life stages (CDC 2015)



Adult female black legged ticks have reddish-brown colored bodies with dark brown plates behind their heads and are about 3.5mm long. Adult males are slightly smaller (~2.5mm long) and are entirely dark brown. Larvae and nymphs are much smaller than adults (0.8mm and 1.5mm, respectively). Because of their small size, which makes them hard to see, and the fact that they can be infected with pathogens like *Borrelia burgdorferi* (Lyme disease) and *Anaplasma phagocytophilum* (anaplasmosis), the poppy seed-sized nymphs represent the greatest risk for human infection.

Blacklegged ticks feed once each life cycle stage and can feed on many different birds and mammals. Larvae and nymphs usually take blood meals from smaller animals, like birds and white-footed mice (*Peromyscus leucopus*). Adults prefer to feed on larger mammals such as white-tailed deer (*Odocoileus virginianus*). Given the opportunity, both nymphs and adults will also feed on other mammals, including dogs and people.

The lifecycle of the blacklegged tick comprises four successive stages, egg, larva, nymph, and adult. This lifecycle generally spans two years but is dependent on climatic factors (CDC 2011). Progression to each successive life stage after hatching requires one blood meal from a single host (CDC 2011). The lifecycle is summarized in Figure 13 and Figure 14, which show the life cycle and seasonal activity of blacklegged ticks. Eggs hatch into larvae, which are typically uninfected. Larvae feed on mice and other small animals. If one of their hosts is infected with *Borrelia burgdorferi,* the larvae may become infected and then stay infected throughout their lifecycle (CDC 2011). After a larva takes a blood meal, it falls off its host and over time molts into a nymph. A nymph also takes a single blood meal, often from bigger mammals such as deer or humans (CDC 2011). Nymphs are most common in the spring and summer. After taking their blood meal, nymphs drop from their hosts and develop into adults. Adult female ticks will take a final blood meal, typically during the fall, during which they can transmit *Borrelia burgdorferi* if they are infected. Adult male blacklegged ticks do not take blood meals but will hop onto to hosts to wait for females and find an opportunity to mate. Because of this, adult male ticks do not transmit pathogens. After mating and taking their final blood meal, adult females will lay eggs, restarting the cycle. So, only nymphs and adult females can transmit *B. burgdorferi*. Nymphs are the most likely to transmit disease to humans because they are small (about the size of a poppy seed) and can stay attached for longer periods without being noticed (CDC 2015).

Figure 12: Geographic range of black-legged ticks in the United States (CDC 2015b)

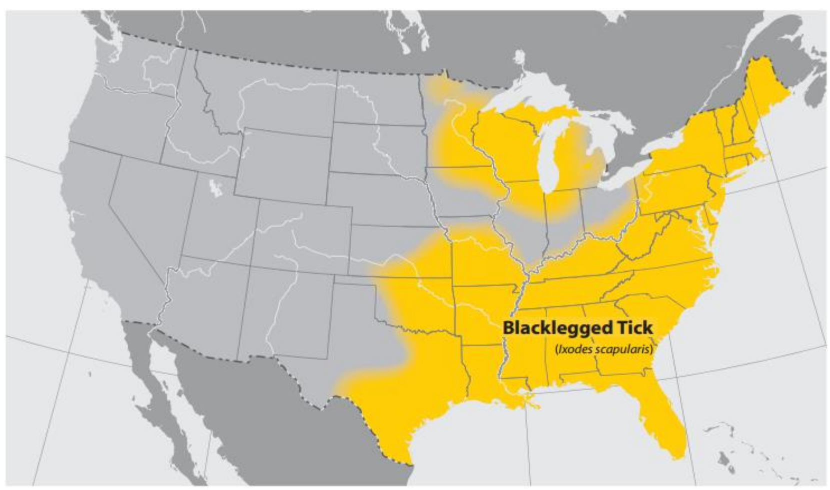


Figure 13: Lifecycle of the blacklegged tick, *Ixodes scapularis* (CDC 2011)

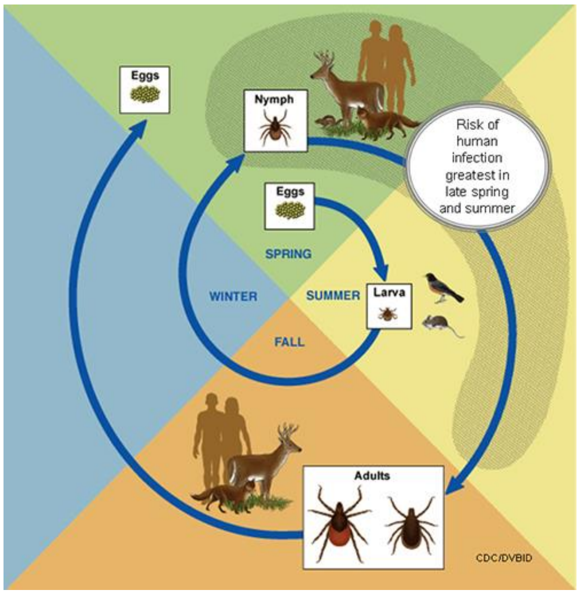
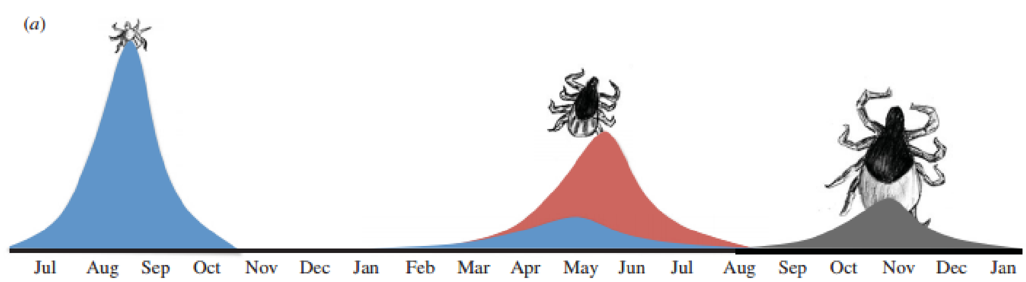


Figure 14 - Tick activity by month and life cycle stage (larva, nymph, adult) - from Levi 2015. Blue signifies larval activity, red signifies nymph activity, and dark grey signifies adult activity.

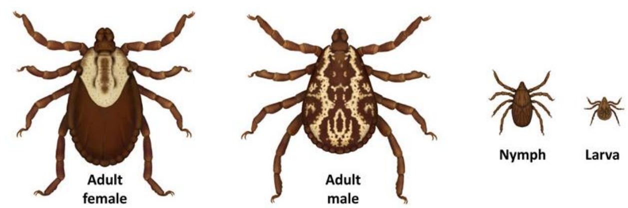


American Dog Ticks (Dermacentor variabilis)

Dermacentor variabilis, also known as the American dog tick or wood tick, is found predominantly in the United States, east of the Rocky Mountains. The tick also occurs in certain areas of Canada, Mexico, California, and the Pacific Northwest of the U.S (Figure 16, Mcnemee et al. 2003). This tick is a vector of the pathogens causing Rocky Mountain spotted fever (RMSF) and tularemia and can cause canine tick paralysis.

Figure 16: Geographic extent of American dog ticks in the United States (CDC 2015b)

Figure 15: Diagram of American dog tick life stages (CDC 2015)



Adult dog ticks have brownish-red bodies with white markings on their back. On female ticks, these are limited to a small area near the top of the back, while males have markings over their entire back (Figure 15). Male and female dog ticks are both roughly 6.5mm long, but females, when engorged after feeding, may reach as large as 13mm long by 10mm wide (VDH 2014a).

American dog ticks feed on three hosts throughout their life cycle, targeting smaller mammals as a larva and nymph and larger mammals as an adult. As its name suggests, it is most commonly found on dogs as an adult (but other ticks, including blacklegged ticks will also readily feed on dogs). However, this tick will also feed on other mammals, including larger animals, such as cattle, horses, and humans.

In the northeast, dog ticks become active in April and remain a nuisance through mid-August, with peak activity occurring in late June. These ticks are abundant in the tall grass and weeds that border roads and trails. Dog ticks are also present in other areas where their hosts live, which include pastures, meadows, marshes, and the edges of lakes and streams. Although ticks of this type do not cause household infestations, they can be brought into a home by a person or a pet and can survive inside for days without feeding (VDH 2014a).

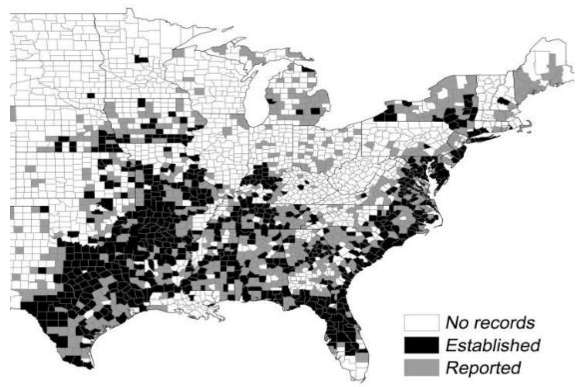
Lone Star Ticks (Amblyomma americanum)

Lone star ticks are the primary vectors of numerous infections in humans and domestic animals, including ehrlichiosis, tularemia, and STARI (Southern Tick Associated Rash Illness), among others. This species of tick is primarily distributed in the eastern and central United States and is more common in the south. While these ticks are established in other areas of the Northeast, this tick is not yet definitively established within Vermont. However, climate change may make conditions in Vermont more favorable for this species of tick in the future.

Figure 17 - Diagram of lone star tick life stages (CDC 2015)

Lone star ticks are slightly larger than deer ticks and woodchuck ticks and have wide, tannish-red bodies (3-4mm, unfed; Figure 17). Female ticks are easily identified, since they have a prominent light-colored spot in the center of their backs. Males have faint, lighter-colored markings on the edges of their backs. Nymphs are smaller and redder in color than adults.

Figure 18: Spatial distribution of counties in which Lone star ticks are known to be established or reported, cumulative from the 1890s to 2014, from Springer et al. 2014



Lone star ticks will feed on most mammals. Nymphs and adults are more likely to be found on deer and other large mammals, while larvae and nymphs usually feed on birds and medium-sized mammals such as skunks, opossums, raccoons, squirrels and foxes. Larvae, nymphs and adult females may bite people, but only nymphs and adults may be infected with pathogens.

Figure 19: Geographic extent of Lone star ticks in the in the United States (CDC 2015b)

Adults are most active in the springtime when they are looking for a blood meal and preparing to lay eggs. Larvae hatch from the eggs laid in spring and take their first blood meal during late summer and early fall. Nymphs are active between April and June since they must find a host and take a blood meal before molting into an adult. High temperatures and low humidity usually decrease this tick's activity.

Woodchuck Ticks (Ixodes cookei)

Woodchuck ticks (also known as “groundhog ticks” or “Packard ticks”) are found east of the Rocky Mountains. They are especially common in the New England, the Midwest, and certain southern portions of Canada. Woodchuck ticks are a vector of Powassan virus, which can cause serious disease in humans (deer tick virus, a strain of Powassan virus, is transmitted by the blacklegged tick). Human cases of Powassan virus disease are relatively rare, and only one illness due to Powassan virus has ever been reported in Vermont (VDH 2014b).

An adult woodchuck tick is about the size of a sesame seed and has a tan body with a reddish-tan plate on its back behind its head. Nymphs and larvae are a lighter tan color and are much smaller than adults (VDH 2014).

These ticks typically feed on small mammals, including woodchucks, raccoons, foxes, skunks, weasels, cats, and dogs. They have also been found to feed on birds and will occasionally feed on humans (VDH 2014).

Woodchuck ticks may be found in brushy areas and along trails bordered by tall grass or weeds. They are also common in unused human dwellings since these environments are nesting places for small mammals (VDH 2014).

Tickborne diseases in the Northeastern United States

Most tickborne diseases that affect humans have only been formally identified by the scientific community since the mid-20th century. Among these diseases in the northeast, Lyme disease and anaplasmosis have the highest incidence rates. However, babesiosis, erlichiosis, tularemia, Powassan/deer tick virus, and Rocky Mountain spotted fever are also of concern. These are diseases that currently have tick vectors that exist in Vermont or other areas of the northeastern United States and which may become established in Vermont given appropriate climatic and ecological conditions. This section outlines the basic ecology and epidemiology of these diseases.

Diseases that can be transmitted by blacklegged ticks (Ixodes scapularis)

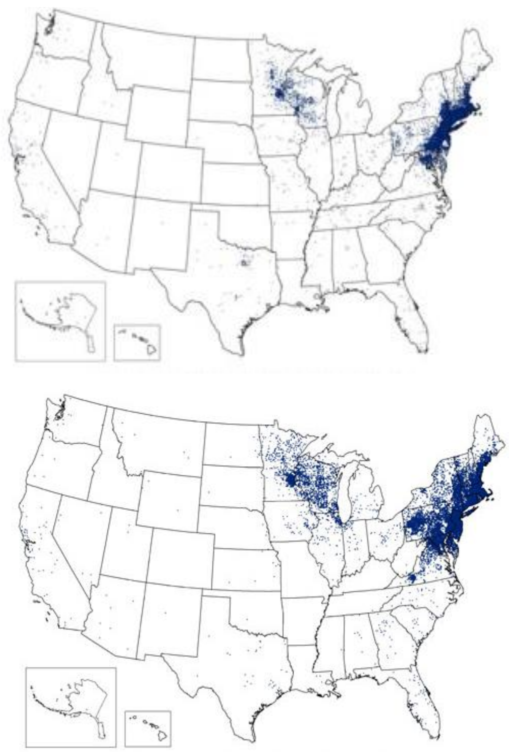
Lyme disease

Lyme disease is caused by infection with the bacteria *Borrelia burgdorferi* (VDH 2014c). In the eastern United States, the infection is transmitted by the bite of *Ixodes scapularis* ticks, commonly known as blacklegged ticks or deer ticks. The *B. burgdorferi* pathogen is primarily maintained in ecosystems by transmission between infected ticks and rodent hosts, which act as the reservoir for the pathogen. These primarily are white-footed mice (*Perocyscus leucopus*), but chipmunks (*Tamias striatus*), shrews (*Blarina brevicauda* and *Sorex cinereus*), and eastern gray squirrels (*Sciurus carolinensis*) (Salkeld et al. 2008) may also act as hosts. When tick larvae and nymphs bite these infected hosts, they may then become infected with the pathogen, and pass it on to future hosts in later life stages.

Early symptoms usually start within 3 to 32 days after infection (usually within ten days) from a tick bite. The symptoms of Lyme disease can vary because various body parts can be affected, including the skin, heart, nerves, or joints. Symptoms can include fatigue, chills, fever, muscle and joint pain, headache, swollen lymph nodes and a characteristic expanding rash, usually near the bite location, known as erythema migrans (Heyman 2010; VDH 2014c). Lyme disease is treatable with antibiotics. If the disease is not recognized and treated in the early stages, it can spread to other parts of the body, in what is known as disseminated Lyme disease. Symptoms of disseminated Lyme disease can occur days to months after initial infection and can include numbness and pain in the arms and legs and paralysis of facial muscles, usually on one side of the face. In some cases, meningitis may occur, resulting in fever, stiff neck and severe headache. In rare cases, disseminated Lyme results in an abnormal heartbeat. Up to 60% of people who do not receive treatment develop bouts of arthritis, characterized by severe pain and swelling at the joints. A small number of cases develop chronic nervous system problems months to years after infection. These chronic nervous problems include shooting pains, numbness or tingling in the hands and feet and problems with concentration and short-term memory.

Lyme disease was first identified in an outbreak in Lyme, Connecticut in the 1970s (NIAID 2012). However, the disease itself appears to have been described in Europe medical literature in the early 20th century (NIAID 2012). It is unclear when or how the pathogen made its way to North America: one genetic study of *B. burgdorfi* suggests that it may have already been present in pre-colonial times (Hoen et al. 2009).

Figure 20: Cases of Lyme disease by county, 2001 (top) and 2012. (bottom). One dot placed at random in county of incidence per case. Note the multi-directional spread of Lyme disease from one epicenter in the Northeast and one in the Midwest (adapted from CDC 2015g).



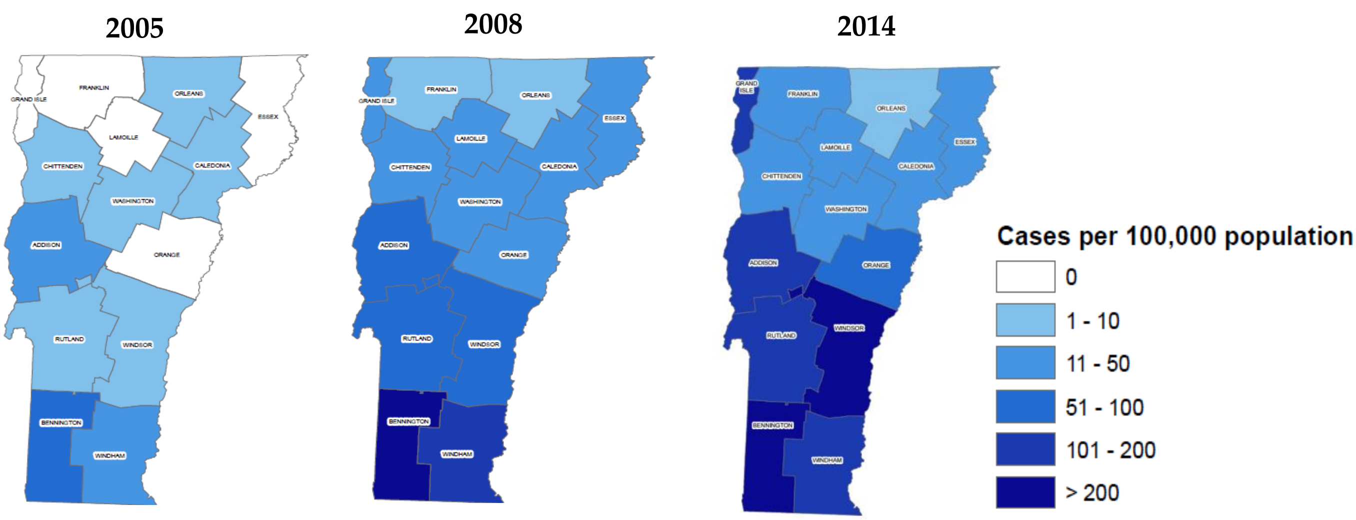
Lyme disease incidence in the United States is heavily concentrated in the Northeast and in the northern Midwest. Figure 20 shows this distribution, and illustrates the expansion of Lyme disease, in geographic scope and number of cases from 2001 to 2012.

The number of cases of Lyme disease reported in Vermont has increased dramatically since the turn of the century. The rise in incidence of the disease in Vermont has occurred largely from south to north (Figure 21).

It is not clear exactly which factors have contributed to the increase in incidence of Lyme disease in recent years. Some portion of the spread may be due to better detection and reporting of the disease by physicians. Warming temperatures due to climate change may also be a contributing factor. Milder winters may make conditions more favorable for blacklegged ticks that transmit the disease. While there have been anecdotal accounts of increases in blacklegged tick populations, no long-term tick counts are currently available in Vermont to verify such an increase. Another potential contributing factor in the surge of Lyme in the northeast is the region's reforestation and associated rise in white-tail deer population (Hoen et al. 2009). By the end of the 19th century, only about 30% of Vermont was forested (Albers 2000), while today, about 78% is forested (FPR 2016). Such an increase in forest cover, while beneficial in many capacities, could provide a more suitable habitat for ticks and tick hosts, particularly white-tailed deer and white-footed mice. Furthermore, there are indications that fragmented land uses, in which forested and residential areas mix, create higher risk for Lyme disease transmission. For instance, Brownstein et al. found an association with fragmented land use in Connecticut with higher tick densities and higher rates of tick infection, although these did not correspond to higher infection rates among humans (Brownstein et al. 2005). Tran and Waller, on the other hand, found a positive association between fragmented land use and county-level human infection rates in thirteen states in the Northeast (Tran and Waller 2014).

In the future, climate change may increase Lyme disease risk in Vermont by providing a better habitat for both the survival of blacklegged ticks as well as rodent hosts that act as reservoirs of the disease. Furthermore, warmer temperatures mean more days of the year during which ticks can be active and humans may be outside recreating, increasing the opportunity for tick bites.

Figure 21: Lyme disease cases 2005-2014. Only confirmed cases are included in the 2005 map. The 2008 and 2014 maps include both confirmed and probable cases.



Anaplasmosis

Human granulitic anaplasmosis (also referred to as HGA, formerly as human granulocytic erlichiosis (HGE) [prior to a taxonomic reorganization of the organism in 2001], and also simply 'anaplasmosis') is a disease caused by infection with the bacterium, *Anaplasma phagocytophilum*. *Anaplasma phagocytophilum* has a life cycle similar to that of *Borrelia burgdorferi*. It cycles naturally among small and large mammals via the bite of ticks in the *Ixodes* genus. In the northeast United States, the tick responsible for this transmission is the blacklegged tick (*Ixodes scapularis*). This is the same tick that can transmit Lyme disease (CDC 2016).

Symptoms of anaplasmosis can range in severity and can be similar to those of the flu. These may include headache, fever, chills, muscle aches and fatigue. Less commonly, people may experience abdominal pain, nausea, vomiting, diarrhea, joint aches, and a rash. Signs of illness typically appear 5 to 21 days following a tick bite and tend to last about 1 to 2 weeks. Many people experience mild illness and recover fully without persistent complications. More severe cases of the disease are associated with older age and compromised immune systems (CDC 2016). Nationwide, around five to seven percent of patients require intensive care due to severe complications, including septic or toxic shock-like syndrome, atypical pneumonitis/acute respiratory distress syndrome (ARDS) and other breading difficulties, hemorrhage, acute renal failure, and opportunistic infections from other pathogens, among others (CDC 2016a). About 1% of cases die as a result of these complications (CDC 2016a). Anaplasmosis is treatable with antibiotics. Not all individuals who are infected with *A. phagocytophilum* will develop noticeable symptoms, and not all symptomatic cases will be diagnosed and reported, so it is likely that the true rate of infection is higher than what ultimately ends up in reportable disease data.

Like Lyme disease, the incidence of anaplasmosis is highest in the Northeast and northern Midwest (CDC 2016b). The incidence of anaplasmosis in Vermont has risen markedly since 2010, when the first confirmed cases in the state were reported.
Among tickborne diseases in Vermont, anaplasmosis incidence is second only to that of Lyme disease. Like Lyme disease, the exact cause of the increase in anaplasmosis incidence in Vermont may be due to a number of factors, including tick and host population dynamics (which may be affected by climate change), and recognition, diagnosis, and reporting of the disease by physicians. Anaplasmosis has only been listed as CDC nationally notifiable condition since 2008 (Code 11090, confirmed and probable). In the future, warming conditions due to climate change, which are favorable for blacklegged tick survival and potentially rodent reservoir hosts, may increase the risk for anaplasmosis in Vermont.

Babesiosis

Babesiosis is caused by the parasite *Babesia microti* and is transmitted to humans by the blacklegged tick (*I. scapularis*). It may also be transmitted between rodents by mouse ticks (*Ixodes muris*), which do not bite humans. Because it is a parasite, it has a more complex life cycle than the bacterial pathogens discussed in this document, like *A. phagocytophilum* or *B. burgdorferi*. The parasite's life cycle begins when infected ticks introduce *Babesia* sporozoites into a rodent host. Sporozoites enter the red blood cells of the rodent and undergo an asexual reproduction phase. They will eventually break out of the cells and emerge, differentiating into male and female gametes. These gametes are then ingested by another tick consuming a blood meal from the same rodent. The gametes, once inside the midgut of the tick, will undergo a sexual reproduction phase (sporogony), and make a new set of sporozoites, which enter the tick's salivary gland, ready to infect another rodent upon the tick's next blood meal. This life cycle is summarized in Figure 22 (CDC 2015),

If humans are infected by a tick, the sporozoites will enter the human red blood cells and go through the same asexual reproduction phase. This kills red blood cells. The anemia associated with this, as well as subsequent releasing of toxins from the dying cells is responsible for the clinical manifestations of the disease. In a severe infection, as many as 6-8% of a person's red blood cells may become infected.

Many people who are infected with *B. microti* do not experience any symptoms at all, while others may develop nonspecific symptoms including headache, fever, chills, and fatigue. Babesiosis can have a much longer incubation period than many other tickborne diseases, and symptoms can appear from about one week to several months after infection. Because these parasites infect and destroy red blood cells, infection can lead to hemolytic anemia which can result in jaundice and dark urine. Babesiosis can be a potentially life-threatening illness for the elderly and people with weakened immune systems. People with health conditions, such as liver, spleen or kidney dysfunction, are also at higher risk for developing a serious illness (VDH 2013c). Babesiosis is treatable with certain antimicrobials.

Infection with babesiosis in Vermont has been uncommon. Only one case has been documented in a person that did not travel out of the state prior to developing symptoms, which occurred in 2012. However, in other areas of the Northeast, including areas in Connecticut, Massachusetts, Rhode Island, southern New York, and New Jersey, it is endemic (VDH 2014).

As can be seen in Figure 23, the geographic incidence of babesiosis has a similar pattern to that of Lyme disease (Figure 20), with most cases occurring in the Northeast and northern Midwest.

Figure 22 - *Babesia microti* life cycle (CDC 2014)

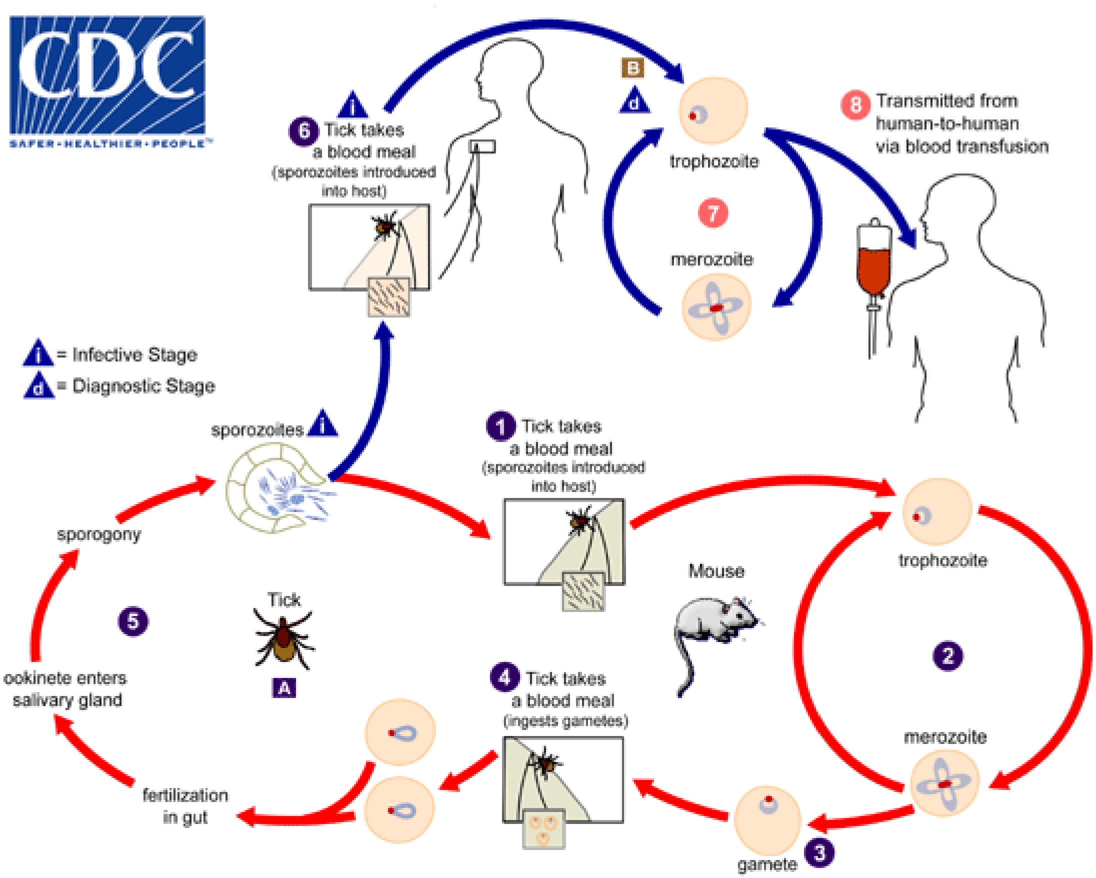
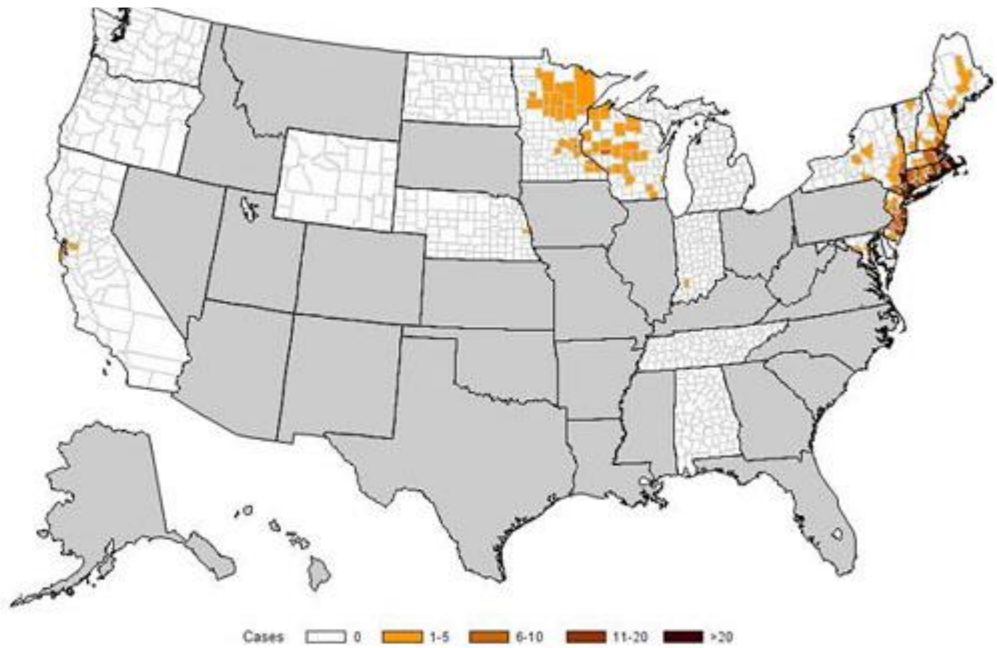


Figure 23: Babesiosis case counts, by county, in the United States, 2012 (CDC 2014)



Powassan / Deer tick Virus

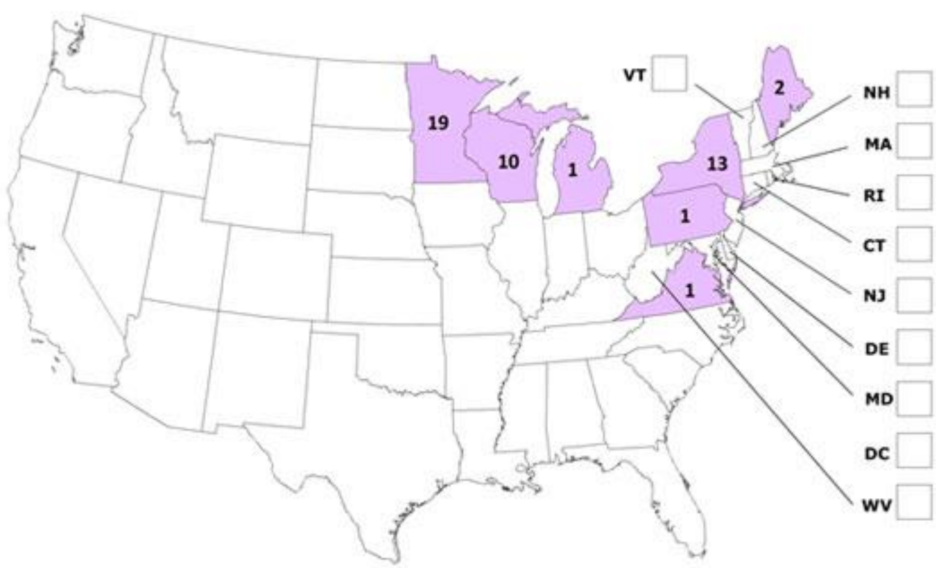
Powassan (POW) virus is a rare but serious disease, transmitted through the bite of an infected tick, most often *Ixodes cookei*, commonly known as the groundhog tick or woodchuck tick. A separate strain of the virus is transmitted by blacklegged ticks (*Ixodes scapularis*). For the sake of clarity and differentiation, this strain is known as *deer tick virus* (DTV) (Ebel 2010, CDC 2015). The two strains are nearly identical genetically, and both may infect humans (Ebel 2010, CDC 2015). Powassan is an RNA virus in the genus *Flavivurus*. Transmission of the virus occurs in three primary cycles in nature: 1) between woodchuck ticks (*Ixodes cookei*) and woodchucks, 2) between *Ixodes marxi* ticks and squirrels, 3) and between blacklegged ticks (*Ixodes scapularis*) and mice (CDC 2015).

The virus was first identified in 1958 and named after Powassan, Ontario, the town of its discovery. POW virus is related to some mosquito-borne viruses, including West Nile virus (VDH 2013f). Most people who become infected with POW virus do not feel ill, but symptoms of the disease can be severe. For those that do become ill, symptoms typically begin to occur between one week and a month after being bitten by an infected tick (CDC 2015). Symptoms include fever, headache, vomiting, muscle weakness, drowsiness, confusion, loss of coordination, speech difficulties and memory loss. The virus can also infect the central nervous system and cause encephalitis (inflammation of the brain) or meningitis (inflammation of the membranes surrounding the brain and spinal cord). About 10% of POW virus infections result in death. Some patients who recover from the initial illness have continuing neurological problems (VDH 2013f). There is no specific treatment for POW virus disease. Treatment consists of supportive care, rest, and increased fluid intake to prevent dehydration (VDH 2013f).

Cases of Powassan virus disease are rare in the United States. There have only been approximately 60 cases of the disease in the country over the past 10 years (CDC 2015). In 1999, one Vermont resident was diagnosed with Powassan virus disease, but it is uncertain as to whether the infection occurred in-state (CDC 2001). There have been no reports of POW virus disease in Vermont since then. Cases of POW virus disease have been reported from New York, Maine, and other northern states in the US, as well as the Canadian provinces of Ontario, Quebec and New Brunswick (VDH 2013f). Figure 24 shows the distribution of neuroinvasive Powassan virus disease cases in the United States from 2002 to 2012.

Woodchuck ticks and blacklegged ticks that transmit Powassan and DTV are both established in Vermont. Warming conditions due to climate change can affect tick activity, reproduction, survival, and tick season length, which may in turn affect tick populations and the infection rates of those ticks. However, it is unknown at this time how common the virus is among tick vectors. Because of this, we cannot make any assertion that climate change will have a notable effect on the risk of humans being infected with Powassan virus.

Figure 24: Distribution of cases of neuroinvasive Powassan virus, 2001 to 2012 (CDC 2013)



Diseases transmitted by other ticks

Ehrlichiosis

Erlichiosis (human monocytic erlichiosis, or HME) is a term that is applied to illnesses caused by from several different bacteria in the *Erlichia* genus; in the United States, these are *Ehrlichia chaffeensis*, *Ehrlichia ewingii*, and *Erlichia muris*. Of these, infection with *E. chaffeensis* is by far the most common in humans. In the United States, erlichiosis is spread by the bite of an infected lone star tick (*Amblyomma americanum*).

Symptoms of ehrlichiosis usually appear one to two weeks after a bite from an infected tick. The bacteria target white blood cells, and the most common symptoms are fever, headache, fatigue and muscle aches. A rash can occur in up to 25% of adults and 60% of children. Patients can also experience nausea, vomiting, diarrhea, joint pain and confusion. Infection usually produces a mild to moderately severe illness, and early treatment with antibiotics usually results in full recovery. Typically, symptoms of the disease last from one to two weeks. Occasionally complications occur, including respiratory problems, blood and kidney abnormalities, meningitis and other central nervous system problems. Hospitalization is sometimes necessary for people with more severe illnesses (VDH 2013d). Ehrlichiosis can occasionally be a life-threatening disease with an estimated 1.8% of illnesses resulting in death (VDH 2013d).

Figure 25 shows an incidence map for ehrlichiosis, which largely corresponds with the geographic distribution of the lone star tick (CDC 2010). Reports from the Vermont Tick Tracker and veterinarian tick submissions to the state indicate that lone star tick populations may currently exist in Vermont, albeit in small numbers, although they have not yet been found in active tick surveillance efforts (i.e. systematic trapping efforts). Warming temperatures will likely lend to more favorable environments for lone star ticks in Vermont and may facilitate the introduction of erlichiosis as an endemic disease in the state. Modeled simulations by Ludwig and others (2015) suggest that *A. americanum* tick populations could survive as far north as Montreal under current climatic conditions, indicating that areas of Vermont may also be suitable habitat for these ticks. Given that, it is unclear as to why lone star tick populations have not yet been firmly established in Vermont. Warming conditions due to climate change may facilitate the expansion of lone star ticks into Vermont and may increase the risk of Vermonters contracting ehrlichiosis within the state.

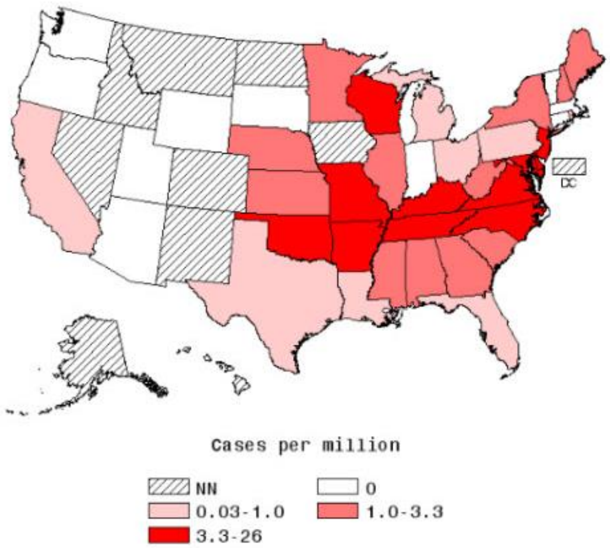


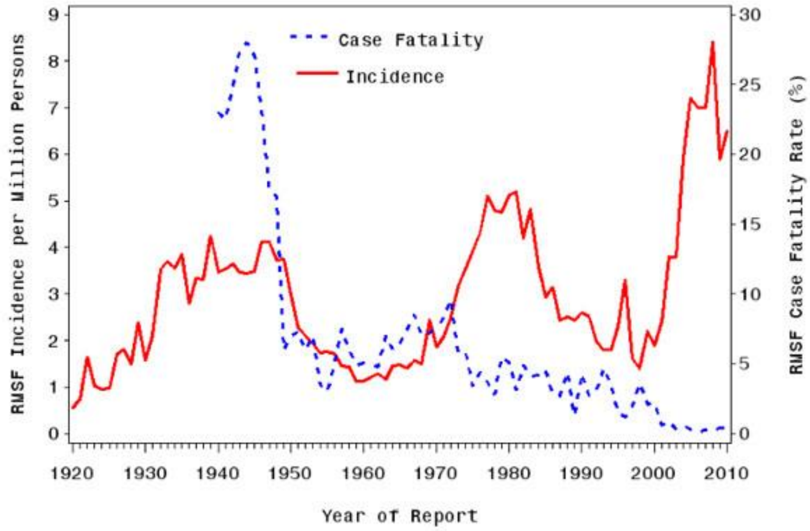
Figure 25: Incidence of Ehrlichiosis in 2010 (CDC 2013)

Rocky Mountain spotted fever

Rocky Mountain spotted fever (RMSF) is an illness caused by infection with the bacterium *Rickettsia rickettsii*. The pathogen cycles naturally between ticks and rodents. In the eastern United States, RMSF is spread primarily by the bite of infected American dog ticks (*Dermacentor variabilis*). Unlike *Borrelia burgdorferi,* it can be transmitted vertically between a mother tick and her eggs, so larvae can be infected with the bacterium. (Niebylski, et al. 1999). Transmission from a tick to a human can occur as soon as four to six hours after the start of a bloodmeal (Wyoming Department of Health, 2016). Symptoms of RMSF usually begin of 2-14 days after the bite of an infected tick. Early symptoms are similar to the flu although frequently more severe (VDH 2012). Most people (90%) with RMSF will develop a rash at some point during the course of the illness; this characteristic rash gives the illness its namesake. Other, flu-like, symptoms include fever, headache, body aches, nausea, acute abdominal pain, and vomiting (CDC 2013). More serious consequences such as damage to the lungs, heart and kidneys, may occur if the condition is left untreated (CDC 2013). Patients who are promptly treated may recover quickly through outpatient medication, while others may require hospitalization and intravenous antibiotics (CDC 2013). Patients with particularly severe infections that require long hospitalizations may have prolonged health problems caused by the disease, including damage to blood vessels, which results in loss of circulation to the extremities (CDC 2013). While the disease can be very severe, especially in those with compromised immune systems, the proportion of RMSF cases that result in death in the United States decreased to less than 0.5% in

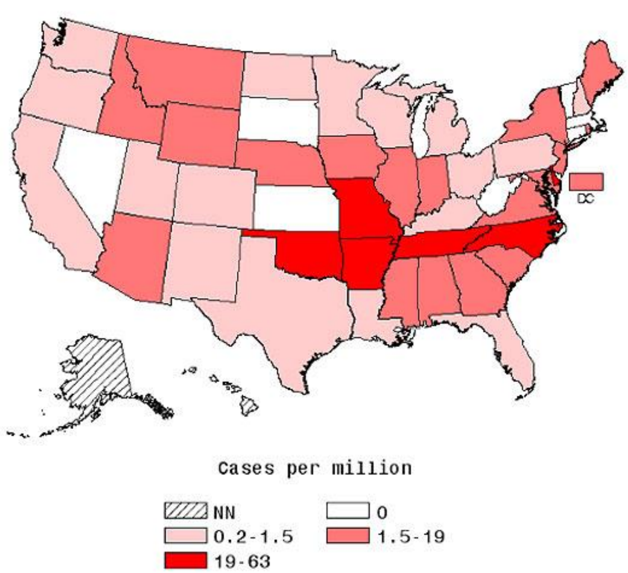
2010 (CDC 2013). RMSF is treatable with antibiotics. However, the incidence of the disease has increased in recent years. Figure 26 shows the increasing incidence and decrease in case fatality rate that has occurred in the United States during the past century.

Figure 26 - Reported incidence (red line) and case fatality (blue dashed line) of Rocky Mountain Spotted Fever in the United States, 1920–2010 (CDC 2013)



RMSF has been reported from all areas of the country, but five states (North Carolina, Oklahoma, Arkansas, Tennessee, and Missouri) account for over 60% of cases. Figure 27 shows

Figure 27 - Incidence of Rocky Mountain Spotted Fever in 2010. While Vermont does not have any recorded cases, New York, New Hampshire and Maine have had cases (CDC 2013c)



the case distribution of RMSF in the United States. There have been no illnesses reported in Vermont, although the American dog tick is established in the state.

Tularemia

Tularemia, also known as rabbit fever or deerfly fever, is caused by infection with the bacterium *Francisella tularensis*. Unlike the other diseases discussed in this document, it is not solely transmitted by ticks. There are several ways that people can become infected with tularemia, including tick and deerfly bites, contact with infected animals, drinking contaminated water or breathing in contaminated dusts or aerosols. Tularemia is included in this section because, while tick bites are not the only mode of transmission, they may be the most climate-sensitive one. Tularemia can be transmitted by both the American dog tick (*Dermacentor variabilis*) and the lone star tick (*Amblyomma americanum*). Tularemia cases in humans are relatively rare and annual case counts have diminished drastically since the 1950s (Figure 28). States in the central United States currently exhibit the majority of cases (Figure 29). The reasons for the decline in cases is not known, however, it has been hypothesized that it may be due to a declining frequency of human exposure to rodents, rabbits, and hares, which may be due to a decrease in the number of hunters and a decrease in the percentage of people living in rural areas (WHO 2007). The symptoms of tularemia vary depending upon the route of transmission though frequently they include sudden onset of fever, chills, headache, muscle aches, chest pain and coughing. Tularemia is treatable with antibiotics and most people recover completely (VDH 2013h).

Only one case of tularemia has been documented in Vermont since 2001, and the last major outbreak in Vermont occurred in 1968 and was related to the trapping of muskrat (VDH 2013h, Young et al. 1969). Figure 29 shows the distribution of Tularemia cases in the United States. Nakazawa and others (2007) evaluated spatial patterns of tularemia in the United States as they related to climate change and found that shifts in the distribution of cases from the 1960s to 1990s could be attributed to changes in climate, however northward shifts have been slight. There is no compelling evidence to date demonstrating that climate change will increase the risk of tularemia in Vermont in the coming century.

Figure 28: Yearly reported tularemia cases in the United States, 1950-2014 (CDC 2015)

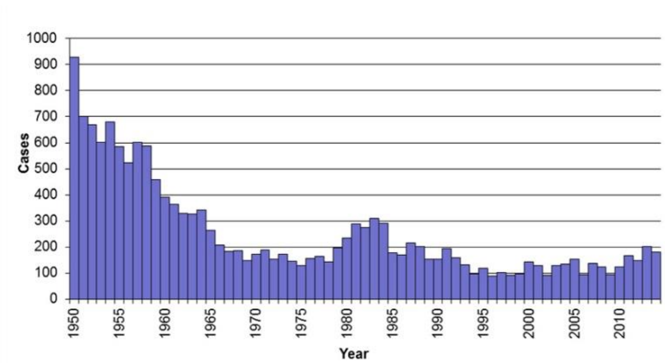
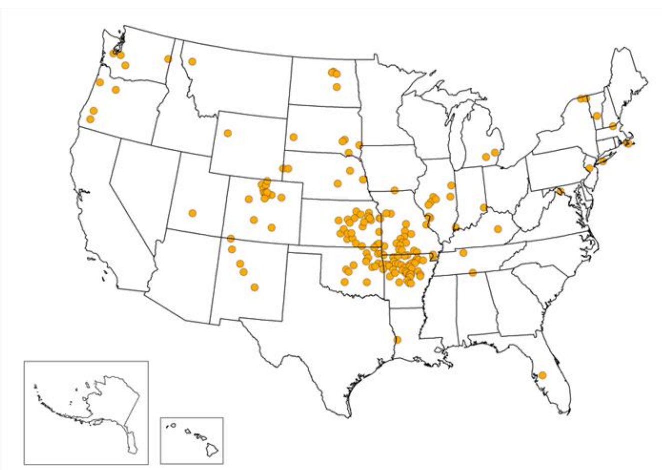


Figure 29: Distribution of Tularemia cases in the United States, 2003-2014 (CDC 2015). One dot is placed randomly within the county of residence for each reported case.



Appendix II - Tick Surveillance in Vermont

General Background

The State of Vermont has engaged in tick surveillance since the early 2000s, using several methods to answer a variety of questions. These include both passive surveillance methods where tick reports and specimens come from members of the public, as well as active surveillance methods, where researchers go to find and collect ticks, which are then identified and tested for pathogens. These surveillance methods have been used to determine that blacklegged ticks (*Ixodes scapularis*) and American dog ticks (*Dermacentor variabilis*) are the most commonly encountered ticks in Vermont. Other species, such as brown dog ticks (*Rhipicephalus sanguineus*), moose ticks (*Dermacentor albipictus*) and groundhog ticks (*Ixodes cookei*) exist in small populations. Lone star ticks (*Amblyomma americanum*) are also occasionally found within the state.

Active Surveillance in Conjunction with Lyndon State College

Background

Since 2013, the Vermont Department of Health has been involved in a tick surveillance effort with Dr. Alan Giese, a biologist at Lyndon State College. Sampling efforts detailed in this section occurred twice annually, in the spring and in the fall, from 2013 to 2015. In all, ticks were sampled from sites in a total of thirteen towns during the surveillance effort. This investigation helped to better understand:

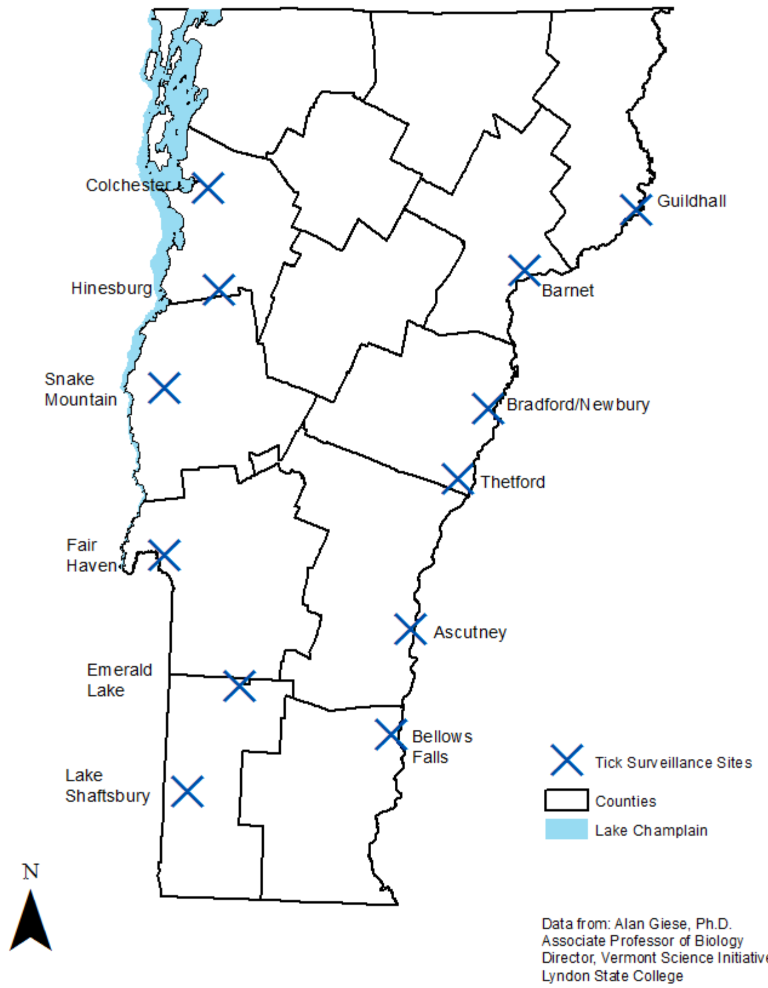
1) the overall prevalence of *Borrelia burgdorferi*, *Anaplasma phagocytophilum*, and *Babesia microti* in Vermont's blacklegged tick populations; and
2) the effect of latitude of the sampling sites on tick abundance and the prevalence of pathogens within local tick populations.

Additionally, this investigation served to establish baseline data of tick abundances at sites for comparison against future repeated samples at the same sites.

Methods

In 2013, twelve relatively low-elevation collection sites were identified, six along the eastern border of VT, and six along the western border (Figure 30). Some sites could not be sampled in all seasons, or had to be changed, depending on circumstances. Eleven of these sites were sampled in the spring of 2013. The Bellows Falls site (southern-most of the six Eastern sites) was identified at the same time but was not sampled until subsequent seasons. The Newbury site was not sampled after 2013 due to an issue that limited access to the site. In 2015, this site was replaced with a site in the neighboring town of Bradford.

Figure 30 – Location of sites sampled during Fall and Spring tick surveillance, 2013-2015



Ticks were collected using flagging (dragging an approximately 1-meter-wide piece of cloth, which host-seeking ticks attach to) in defined 200 meter transects. Both forest and meadow transects were identified at each site and sampled in 2013. Due to low tick yields from the meadow transects and issues in accommodating full 200 meter transects at some meadow sites, the decision was made to instead focus predominantly on sampling forest transects in 2014 and 2015. Most sites were sampled twice in a given season, although some were sampled once if circumstances did not allow for a second sample. Typically, sequential sampling efforts at individual sites were separated by a minimum of 72 hours, although for the first session in Spring 2013, a minimum of 24 hours was used. While attempts were made to keep sampling methods consistent across sites and over time, some modifications to sites or methods were required in response to a lack of ticks found at certain sites, some sites having forest or meadow areas too small to accommodate full 200-meter tick collection transects, and the need to collect blacklegged ticks for pathogen testing.

During the spring, transects were surveyed during the expected peak of blacklegged tick activity in April, May, and June. During the fall, transects were surveyed in October and November. At certain sites, the number of ticks collected on transects was relatively low. At these sites, off-transect flagging was also added to survey efforts, in order to increase the number of ticks available for pathogen testing. These off-transect surveys were conducted within 1.5 miles of the original transect.

Over three years of Spring and Fall sampling from 2013 to 2015, the Lyndon State College team collected a total of 1,611 *I. scapularis* ticks, which included 569 adult females, 591 adult males, 307 nymphs, and 144 larvae. A total of 1,234 of these ticks were tested for four pathogens: *Borrelia burgdorferi* (the bacteria that causes Lyme disease), *Borrelia miyamotoi* (a bacterium related to *Borrelia burgdorferi* that can cause fever, chills, headache, joint pain, and fatigue), *Anaplasma phagocytophilum* (the bacteria that causes anaplasmosis), and *Babesia microti* (the bacteria that causes babesiosis), by the Laboratory of Medical Zoology at the University of Massachusetts, Amherst.

Findings

There was a high level of variation in the numbers and densities of ticks collected per site. In general, the data from this surveillance effort do not support the idea that tick abundance is necessarily higher in southern parts of the state than it is in northern areas, in spite of the higher incidence of Lyme disease in southern areas. The sites with the highest numbers of ticks collected on surveillance transects were Thetford, in the middle of the state on the eastern side, followed closely by Lake Shaftsbury in the southwestern portion of the state, and Colchester, the northernmost site in the west (Table 10).

Table 10 – Total number of blacklegged ticks collected by site, Spring 2013 to Fall 2015. Values include both the # of ticks collected in study transects and off-transect flagging. Numbers in parentheses indicate the number of ticks collected in study transects only.

Western Sites	Total # of Ticks Collected (# collected in study transects)	Eastern Sites	Total # of Ticks Collected (# collected in study transects)
Colchester	269 (166)	**Guilford**	0 (0)
Hinesburg	124 (39)	**Barnet**	35 (5)
Snake Mountain	37 (17)	**Newbury**	7 (2)
Fair Haven	176 (94)	**Thetford**	306 (236)

Emerald Lake	176 (94)	**Ascutney**	129 (66)
Lake Shaftsbury	194(169)	**Bellows Falls**	102 (33)

There were also season-to-season variations in the collection rates of blacklegged ticks. Densities of adults were the highest in the Spring 2013 field season, after which densities of adults collected varied between roughly 2 and 6 per 1000 square meters (Figure 31). Similarly, density of nymphs was also highest in the Spring 2013 season (Figure 32).

Figure 31 - Collection rate for adult blacklegged ticks, by field season

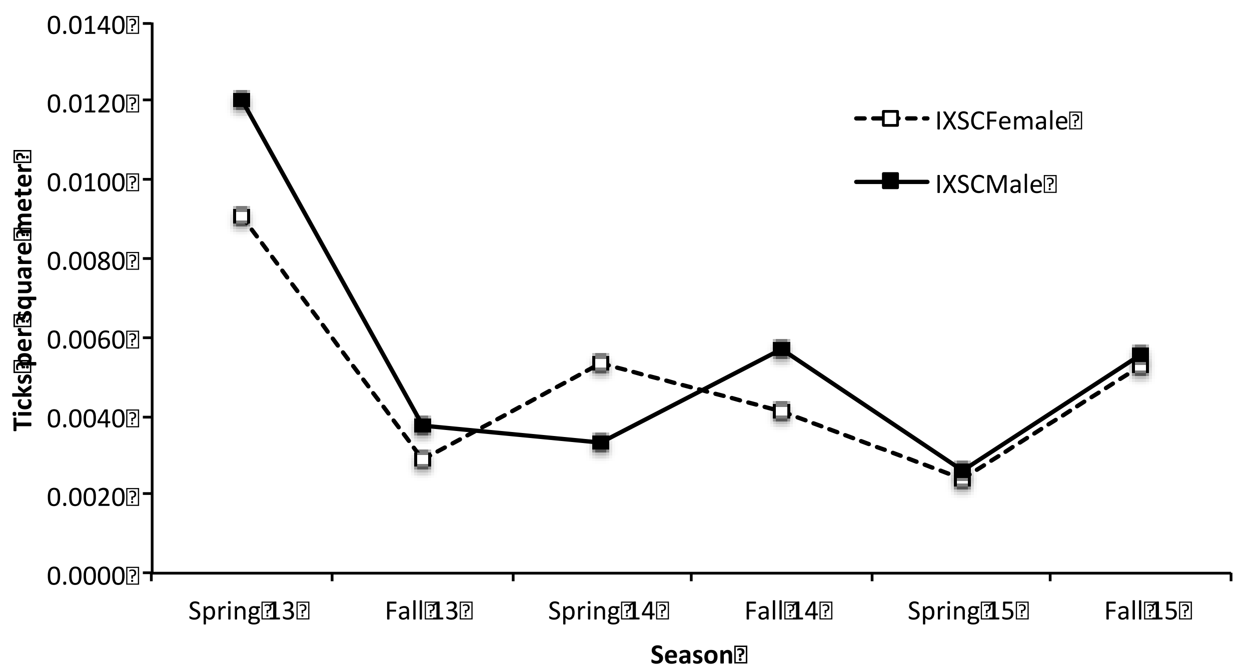
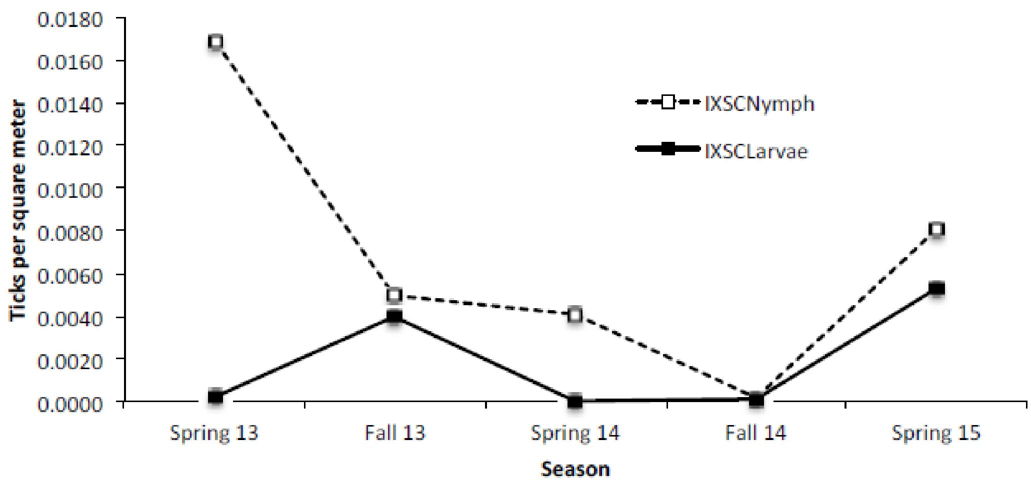
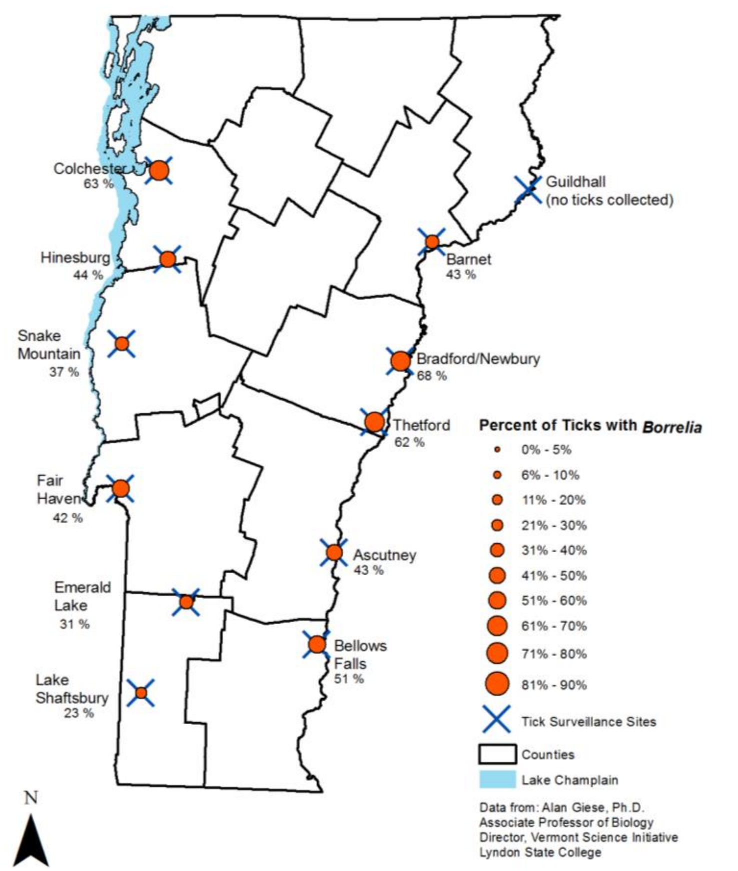


Figure 32 - Collection rate for nymph and larval blacklegged ticks, by field season (note no nymph or larval ticks were collected in Fall 2015 season)



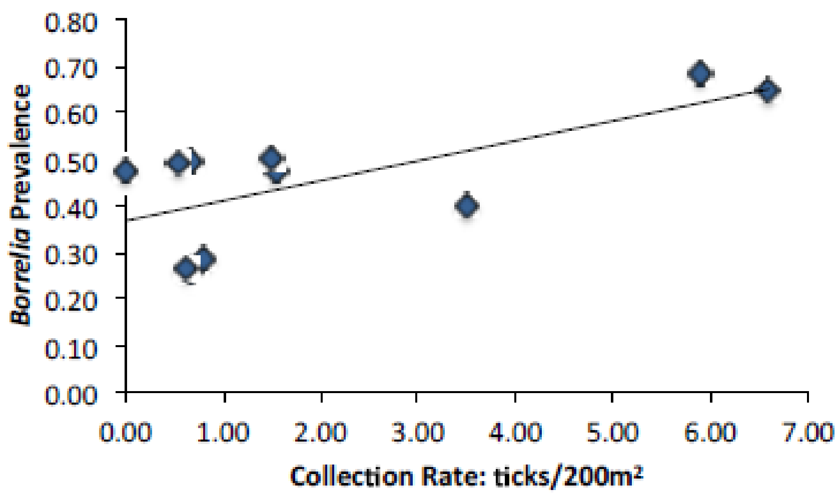
The overall *Borrelia burgdorferi* infection rate over three years of sampling was 50%. There was no strong north-south trend in *B. burgdorferi* infection prevalence at different sites, which varied overall between 23 and 63 percent (Figure 33). This result, in conjunction with the finding of no strong north-south trend in tick abundance, may be due to the limited number of sites sampled, especially in the northernmost portions of the state.

Figure 33 - Borrelia infection prevalence in adult and nymph blacklegged ticks (combined), Spring 2013 to Fall 2015



An analysis of the 2013-2014 data indicated a relationship between tick density and *B. burgdorferi* prevalence[10] (Figure 34), although this relationship is not particularly strong (R^2=0.41). Some areas with low collection rates had high prevalence (although some of this may be attributable to small sample size and natural statistical variation as opposed to a truly high prevalence). The prevalence of *B. burgdorferi* was not below 20% in any of the areas where ticks were collected.

Figure 34 – *B. burdgorferi* infection prevalence by Black-legged tick density, 2013-2014, Spring and Fall Seasons



In a 2014 study funded by the Vermont Agency of Agriculture, the sites in Barnet and Colchester were also sampled for mice (*Peromyscus spp.*), in order to understand the *B. burgdorferi* infection prevalence in mammal populations that tick larvae and nymphs feed on (Figure 35). At the Colchester site, an area with high tick density, the infection prevalence among mice was far higher than the infection prevalence among mice at the Barnet site, an area with low tick density. This is consistent with the finding in Figure 34, indicating that areas more densely populated with ticks will have a higher infection prevalence in competent hosts (in this case, white-footed mice), which will also support more ticks becoming infected.

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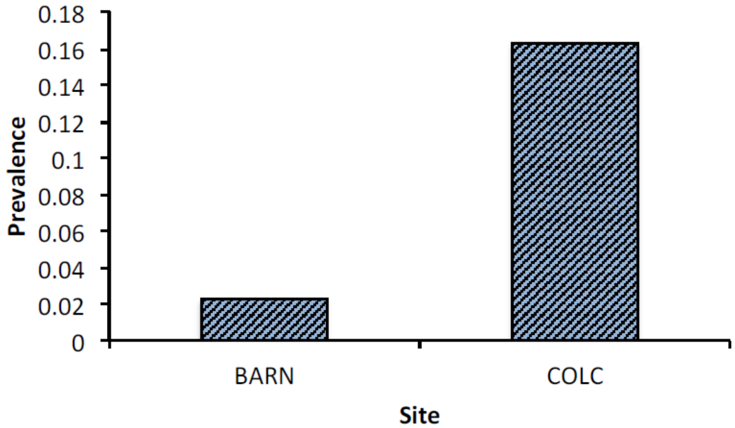
[10] (*Note: Borrelia* prevalence includes ticks collected off-transect. So, the tick collection rate at a given transect may be zero, but testing of off-transect ticks allows for the calculation of a *Borrelia* prevalence)

Figure 35 - Prevalence of *Borrelia burgdorferi* in white-footed mice collected from the Barnet (n=44) and Colchester (n=43) sites. Tick collection rates (indicative of tick density) at the sites were 0.00 (i.e. no ticks collected were on official flagging transects; all ticks were collected off-transect) and 5.89 ticks / 200m^2, respectively.



Overall, The *Anaplasma* infection prevalence among sites was far lower than that of *Borrelia burgdorferi*. Prior to the Spring of 2015, *Anaplasma* infection prevalence was about 1% among nymphs and adults. However, nymphs tested during the Spring 2015 season had an infection prevalence of 16%. Relatively higher prevalence of *Anaplasma* was found at the Ascutney, Colchester, and Bellows Falls sites, where infection prevalence in nymphs ranged from 50% to 61.5%. The relatively higher prevalence at these sites persisted into the Fall 2015 season. Table 11 shows a summary of all pathogen tests conducted to date, organized seasonally, for blacklegged ticks that were collected.

The dramatic increase in *Anaplasmosis* prevalence over a single year at these sites is difficult to explain. It may simply be an anomaly, however, if the results reflect a true and persistent rise in *Anaplasma* prevalence among blacklegged ticks at these sites, it may be evidence of the establishment of *Anaplasma* in an area where it had previously not been.

Additionally, *Babesia* was first detected in collected ticks in Spring 2015, and became more prevalent by Fall 2015, although it was still relatively rare, being detected in 6.6% of ticks that were tested. (Table 11). All *Babesia*-infected ticks (one in Spring 2015, eight in Fall 2015) were collected at the Fair Haven site, and eight of the nine ticks were also infected with *B. burgdorferi*.

Table 11 - Summary of all pathogen tests to date for the years 2013 to 2015, organized seasonally. Proportion of I. scapularis ticks (nymphs and adults) testing positive for three pathogens.

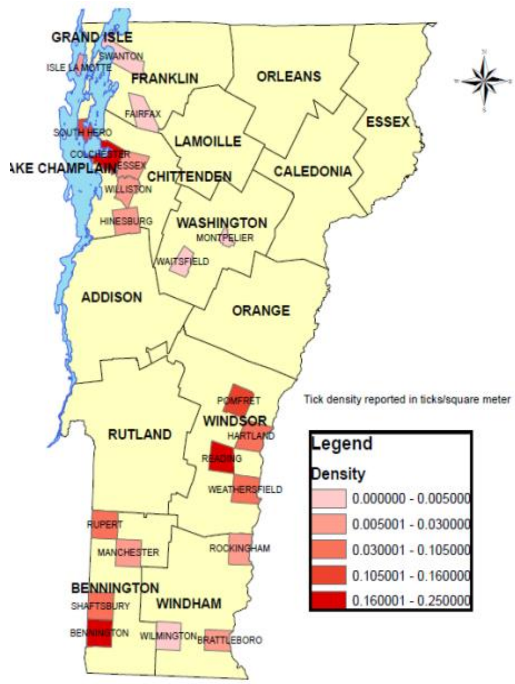
Season	N	*Borrelia burgdorferi*	*Borrelia miyamotoi*	*Anaplasma phagocytophilum*	*Babesia microti*
Spring 2013	395	53.9%	N/A*	0.5%	0.0%
Fall 2013	151	51.0%	N/A*	1.3%	0.0%
Spring 2014	190	43.7%	N/A*	0.0%	0.0%
Fall 2014	85	48.2%	N/A*	0.0%	0.0%
Spring 2015	291	42.6%	0.3%	16.8%	0.3%
Fall 2015	122	60.7%	4.1%	10.7%	6.6%
Total	1234	49.6%	0.5%	5.3%	0.7%

**Babesia miyamotoi* was only tested for in the 2015 samples.

Vermont Agency of Agriculture 2015 Surveillance

In the spring and fall of 2015, the Vermont Agency of Agriculture surveyed twenty towns in seven counties for blacklegged ticks. These ticks were tested for *Borrelia burgdorferi, Anaplasma phagocytophilum,*and *Babesia microti*. All sites, save for one (in Bennington), were selected randomly. Two or four 100m transects were flagged for ticks, which were subsequently tested. A total of 659 ticks were collected from transect flagging, (72 in spring, 589 in fall). Among these, 300 were adult females, 353 were adult males, and 5 were nymphs. Six larvae were also collected but were tested in a single pooled batch. All ticks from transects were tested for pathogens. (Graham 2016).

Figure 36 - Tick density at sites surveyed by the Vermont Agency of Agriculture in spring and fall, 2015 (Graham 2016)



Among adult females, 62% were infected with *B. burgdorferi*, 10% were infected with *Anaplasma*, and 1% were infected with *Babesia*. Among adult males, 56% were infected with *B. burgdorferi*, 11% were infected with *Anaplasma*, and only one (0.28%) was infected with *Babesia*.

Site-specific tick density and pathogen infection prevalence in ticks varied considerably. Tick density was highest at the sites in Reading, Colchester, and Bennington. Among sites with 10 or more ticks collected, *B. burgdorferi* infection prevalence ranged from 4.5% to 81%. In these sites, *Anaplasma phagocytophilum* infection prevalence ranged from 0% to 27%. *Babesia microti* was only detected in ticks from the Bennington site, with five of the 2016 ticks (2.3%) found to be infected. All five of these *Babesia*-infected ticks were also infected with other pathogens: 3 with *B. burgdorferi,* 1 with *A. phagocytophilum*, and 1 with both *B. burgdorferi* and *A. phagocytophilum* (Graham 2016).

For a full summary of this surveillance activity, please consult the Vermont Entomological Society's Spring 2016 newsletter (Available at: http://www.vermontinsects.org/vesnews/VES%20News%20-%20Spring%202016.pdf)

Vermont Tick Tracker

Since 2013, the Vermont Department of Health has operated an interactive "Tick Tracker" web-based tool as a means of raising awareness and collecting important data from the public. Anyone encountering a tick on their body or in the wild is encouraged to report the location and species of the tick on the Tick Tracker. The tool has allowed the state to estimate the relative abundances of different tick species that members of the public encounter day to day, and is also useful in reporting sightings of rarer tick species in Vermont that are medically important, such as the lone star tick and the woodchuck tick. Additionally, the tracker has utility in helping to identify the start of the "tick season" in Vermont, when encounters begin to occur, as well as within-season variation in tick reporting. These reports are not validated by the Health Department, and must be interpreted cautiously, as the majority of submissions likely represent adult ticks, which are easiest to find, and tick identifications by members of the lay public may not be accurate, and enthusiasm for such reporting may vary throughout the season. Among the reports, over two-thirds (69.5% in 2015) of the ticks encountered by Vermonters reporting to the tick tracker are blacklegged ticks, giving further indication that these may be among the most commonly-encountered ticks in the state. This is consistent with the findings of the other surveillance methods in the state.

Veterinarian Tick Submissions

From 2002 to 2004, the Vermont Department of Forests, Parks, and Recreation collected tick specimens that were removed from pets by veterinarians. These ticks were not tested for pathogens. In the course of this effort, 1,671 ticks were collected. The majority of these (~74%) were blacklegged ticks (*I. scapularis*); 17% were dog ticks (*Dermacentor variabilis*). Other species identified were woodchuck ticks (*I. cookei*), lone star ticks (*Amblyomma americanum*), *Ixodes angustus*, Moose ticks (*Dermacentor albipictus*), and *Ixodes muris*. Some of these ticks were likely to have been picked up by dogs outside of Vermont, although a few specimens of rarer species of ticks (ex. lone-star ticks) were reported to have been acquired in-state.

Deer Season Survey

From 2000 to 2004, the Vermont Department of Forests, Parks, and Recreation and the Department of Fish and Wildlife collaborated on a baseline study to try to determine the abundance of blacklegged ticks (*I. scapularis*) and moose ticks (*D. albipictus*) in the state. Over the five-year course of the study, 1,939 deer were examined at 18 deer check stations, and 639 ticks were collected from these deer. Roughly 9% (n=181) of deer had blacklegged ticks on them. In 2004, blacklegged ticks that were collected from deer were tested for the Lyme disease pathogen *Borrelia burgdorferi*. Among the 88 adult blacklegged ticks tested, eight (7%) tested positive for *B. burgdorferi*. If this is an accurate assessment of the statewide *Borrelia* prevalence at the time, it indicates that the prevalence of *Borrelia* has increased greatly in Vermont's tick populations over the past several years, as it is now closer to 50% (see Active Surveillance section). However, there are limitations in using this as an indicator of statewide *Borrelia* prevalence of ticks, given that this is a small sample of adult ticks collected in a short portion of the fall season, whereas estimates from active surveillance efforts include both nymphs and adults taken in wider time periods during the spring and fall. Despite this, given the increase in Lyme disease cases since the early 2000s, it very plausible that there has been a rise in *Borrelia* prevalence among blacklegged ticks in the state.

Grand Isle and Bennington County Active Surveillance, 2004

Staff from the Health Department and the Department of Forests, Parks, and Recreation collected ticks in Grand Isle County during the October of 2004, in response to reported Lyme disease cases in the area. A total of 69 blacklegged ticks were collected in Grand Isle county, and another 56 were collected at three sites in in Bennington county. In total, 34 of the 69 ticks (49%) in Grand Isle county and seven of the 56 ticks (12.5%) in Bennington county tested positive for *Borrelia burgdorferi* spirochetes.

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