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Distribution of the deer tick (Ixodes scapularis) in a corridor between Marquette and Menominee in the Upper Peninsula of Michigan

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DISTRIBUTION OF THE DEER TICK (*IXODES SCAPULARIS)* IN A CORRIDOR BETWEEN MARQUETTE AND MENOMINEE IN THE UPPER PENINSULA OF MICHIGAN

By

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ABSTRACT

DISTRIBUTION OF THE DEER TICK (*IXODES SCAPULARIS)* IN A CORRIDOR BETWEEN MARQUETTE AND MENOMINEE IN THE UPPER PENINSULA OF MICHIGAN

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The distribution of *Ixodes scapularis* was studied in a corridor of Upper Michigan between Menominee and Marquette counties by flagging and trapping. Sixty-nine sites were chosen for macroenvironmental characteristics, and microenvironmental characteristics (soil type, moisture, and pH and leaf litter moisture) were investigated as indicators of the presence of deer ticks. Results indicate that deer ticks have not expanded their population within the corridor in 14 years. Soil type, pH and leaf litter moisture were not significant indicators of the presence of deer ticks. There was a significant interaction between soil moisture and tick presence, with a higher proportion of deer ticks in areas with low available water capacity. There was no significant difference in prevalence and intensity of deer ticks on *Peromyscus leucopus* (intensity = 1.6 ticks/infected mouse, prevalence = 100 % mice infested) and *P. maniculatus* (intensity = 4.0 ticks/ infected mouse, prevalence = 75 % mice infested).

DEDICATION

This thesis is dedicated to my family

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INTRODUCTION:

The deer tick (*Ixodes scapularis)* has been steadily increasing its range across the United States and has the potential to continue increasing its range (Brownstein, Holford, & Fish, 2003). These ticks can transmit human pathogens causing infectious diseases such as Lyme disease, babesiosis and granulocytic ehrlichiosis when humans act as hosts for the nymphs and adult ticks (Mitchell, Reed & Hofkes, 1996). The life cycle of *I. scapularis* is typically 2-4 years. Larvae hatch, feed once on a host and then drop off and overwinter in the leaf litter. In the spring, immature ticks molt into nymphs, find a new host, feed once, and drop off into the leaf litter to mature into adults the same year. The following spring they feed once on a host and mate. After feeding and mating, ticks drop off the host and females lay their eggs in leaf litter. The amount of time between stages varies from months to one year depending on diapause events and environmental conditions (Padgett & Lane, 2001).

Larvae and nymphs can feed on small mammals, birds and reptiles. Their typical hosts include chipmunks (*Tamias* spp.), squirrels (*Sciurus* spp.) and mice, with the most common being the white-footed mouse (*Peromyscus leucopus).* Adults feed on larger mammals, such as white-tailed deer (*Odocoileus virginianus*), raccoons (*Procyon lotor),* red foxes (*Vulpes vulpes),* and feral and domesticated dogs (*Canis lupus familiaris;* Duffy, Campbell, Clark, DiMotta & Gurney, 1994; James & Oliver, 1990; Van Buskirk & Ostfeld, 1998).

Deer ticks are established in counties in 43 states, with the most common and largest populations in Connecticut, Delaware, Maine, Maryland, Massachusetts, Minnesota, New Hampshire, New Jersey, New York, Pennsylvania, Rhode Island, and

Wisconsin (Nicholson & Mather, 1996). In the Upper Peninsula (U.P.) of Michigan, an established population was first reported in Menominee County by Strand, Walker and Merritt (1992). A tick population is considered established if 6 ticks, or 2 of the 3 stages are identified within an area (typically counties within the state). A reported population is when 1 to 5 ticks or just one life stage are identified within the area (Centers for Disease Control and Prevention, 2005).

Environmental characteristics associated with the distribution of the deer tick

Macroenvironmental factors

Habitat type, through its impact on the abundance of and utilization patterns by suitable hosts, may influence tick densities by directly and indirectly affecting tick survival (Ginsberg & Ewing, 1989; Lindsay, Mathison, Barker, McEwen & Surgeoner, 1999; Ostfeld, Cepeda, Hazler & Miller, 1995; Talleklint-Eisen & Lane, 2000; Van Buskirk & Ostfeld, 1998). The distribution of larval deer ticks was determined by the movement patterns of white-tailed deer the previous year, since adult females lay their eggs after they drop off. The distribution of nymphs and adults relies on the movement of small mammals (Ginsberg & Ewing, 1989).

In Maine, deer ticks were positively associated with deciduous to mixed forests that had ample leaf litter and a dense shrub layer (Lubelczyk, Elias, Rand, Holman, Lacombe & Smith, 2004) where questing ticks could be protected from desiccation due to direct sunlight and wind movements (Adler, Telford, Wilson & Pielman, 1992). In Illinois and Wisconsin, the presence of deer ticks was positively associated with dry to mesic deciduous forests (Guerra et al., 2002). These forests are commonly oak (*Quercus* spp.) dominated with a dense canopy layer and ample underlying vegetation providing suitable habitat for questing, molting, diapause and oviposition for ticks. However, abundance of *I. scapularis* is not solely influenced by forest type. In Ontario, Canada, maple (*Acer* spp.) forests had a higher infestation of ticks than oak forest in some years, but oak forests had a greater infestation than maple forests in other years, indicating that other factors also influenced the distribution and infestation levels of deer ticks (Lindsay et al., 1999).

Because ticks spend most of their time off a host they are strongly affected by the moisture that is held within leaf litter and soil, the depth of the leaf litter, and the ground level relative humidity. Ticks survive best at high humidity because they are sensitive to desiccation (Padgett & Lane, 2001). Larval ticks, due to their smaller size, are particularly sensitive to exposure and low relative humidity (Talleklint-Eisen & Lane, 2000). In drier areas questing ticks must find a host quickly or go back into the leaf litter before they die. The drier the area, the quicker this would happen.

Microenvironmental factors

Vegetation, soil type, soil moisture, topography, and climate are all interrelated (Curtis, 1959). In the Mid-Atlantic region of the United States of America (Delaware, Maryland, New Jersey, Pennsylvania, and Virginia), deer ticks were significantly associated with acidic, coarse-textured soil within a narrow range of low available water capacity (Bunnell, Price, Das, Shields & Glass, 2003). Available water capacity is a measure of soil moisture, which is determined by various soil properties. Increased rock fragment content decreases available water capacity, but increased organic matter content increases the available water capacity. Soil texture also affects available water capacity. Soils with a coarser texture (sandy or sandy-loam content) have a lower available water capacity compared to soil with finer texture (loam, silt or clay content; Berndt, 1989; Linsemier, 1989; Schwenner, 1989; United States Department of Agriculture Natural Resources Conservation Service (USDA NRCS), 1995, 1997).

When ticks are not on a host or questing they spend their time in the leaf litter at depths with adequate moisture. In Midwestern habitats, the presence of deer ticks was positively associated with soils of sandy or loam-sand texture overlying sedimentary rock, and with oak and maple forest (Guerra et al., 2002). Sites with sandy soils supported deer tick populations better than sites with loamy soils due to their lower available water capacity (Bunnell et al., 2003). Excessive moisture in the soil may harm ticks while they are hiding in it when not questing, and while they overwinter. Additionally, higher levels of moisture enhance the growth of fungi and are favorable for nematodes, which may have adverse effects on the tick population (Zhioua, Ginsberg, Humber & Lebrun, 1999).

Soil pH typically ranges from 3.5 to 9.0. Soil pH can fluctuate within the year since it is dependent on minerals present, climate and weathering in the area (USDA NRCS, 1998). For example, rainfall lowers the pH when positive ions (Ca^{2+}, Mg^{2+}, K^+ and Na^+) are leached out of the soil and replaced with H^+ ions (USDA NRCS, 1999). A pH of 6 to 7 is generally most favorable for plant growth because most plant nutrients are readily available, but some plants have lower or higher pH requirements. A higher pH of 6.6 to 7.3 is favorable for microbial activity (USDA NRCS, 1998), with various types of nematodes typically found in soil with a pH ≥ 4.5 (Gerrard, 2000). A lower pH may be favorable for tick development due to the plants that grow there (Bunnell et al., 2003) and there are fewer microbes that could be detrimental to tick development (Yoder et al., 2003; Zhioua et al., 1999).

Host

White-footed mice (*Peromyscus leucopus)* and deer mice (*P. maniculatus)*, which are common hosts for immature deer ticks, are found throughout the northeastern United States and portions of southeastern Canada. They are considered to be habitat generalists (Barry & Francq, 1980; Kilpatrick, Rich & Crowell, 1994; Witt & Huntly, 2001) and are frequently found in locations with habitats suitable for tick development (e.g., abundant trees, stumps, logs, rocks, and other debris; Barry & Francq, 1980). Both species of mice are active during the evening hours of the summer. *Peromyscus leucopus* is active earlier in the evening when it is still warm, and *P. maniculatus* is active during later evening hours when it becomes slightly cooler (Drickamer & Capone, 1977; Drickamer, 1987).

Immature *I. scapularis* also feed on reptiles and birds (Sonenshine, 1991). In a laboratory experiment, *I. scapularis* larvae had significantly higher feeding success rates on mice (68-86%) than on lizards (44-58%) or chickens (8-16%; James & Oliver, 1990). Nymphs showed no significant difference in feeding ability on mice (67-93%) and lizards (50-80%), but fed better on mice and lizards than they did on chickens (20-70%).

Adult deer ticks typically feed on white-tailed deer (Duffy et al.. 1994; James & Oliver, 1990; Van Buskirk & Ostfeld, 1998) and the geographic distribution of *I. scapularis* is closely related to deer presence (Stafford, Denicola & Kilpatrick, 2003; Wilson, Ducey, Litwin, Gavin & Spielman, 1990). White-tailed deer are commonly found in forests, forest meadows, and woodland clearings with ample cover (Kurta, 1995). They forage mostly during sunrise and sunset, but are active during much of the day, feeding as they walk (Kurta, 1995). Duffy et al. (1994) found that the density of *I. scapularis* increases as densities of white-tailed deer increase. Adult *I. scapularis* also parasitize many species of medium and large mammals (Duffy et al., 1994; James & Oliver, 1990; Van Buskirk & Ostfeld, 1998).

Tick increase in range

Because ticks do not travel themselves colonization of new areas depends on host's movements. Adult female ticks generally spend from 1 to 3 weeks on a white-tailed deer before they drop off and die after oviposition (Glavanakov et al., 2001). White-tailed deer home ranges are 0.5-5 km^2 during the tick questing season (Rand et al., 2003). Fawns disperse 5 km to 20 km from where they were born, with males dispersing farther than females (Nelson & Mech, 1984). White-footed mice and deer mice have home ranges of around 500 m^2. They disperse 250 m to 450 m from their original home range, but not over 1000 m (Bowman, Forbes & Dilworth, 2001; Witt & Huntly, 2001).

In an endemic are of Westchester County in New York deer the tick population expanded north and west following the Hudson River (Daniels et al., 1998; Glavanakov et al., 2001). Tick surveillance showed that the range of *I. scapularis* expanded up to 384 km from the original known endemic area of Long Island, NY in 7 years (White et al., 1991).

Madhav, Brownstein, Tsao and Fish (2004) modeled a hypothetical landscape loosely based on the northeastern United States. Their model included only three hosts: deer, mice and the American robin (*Turdus migratorius)*. In their model, the range of *I. scapularis* expanded non-linearly, increasing slowly at first then more quickly as time progressed. In this model, hosts with high tick burdens and large home ranges (e.g., deer) played an important role in deer tick expansion. Hosts with small home ranges (e.g., mice) limited range expansion if they diverted a sufficient number of ticks from feeding on more mobile hosts (e.g., robins) that migrate annually. The authors concluded that their model is a good, but simplistic example of tick expansion. Unfortunately, little is known about complex factors that affect ticks and their hosts. Thus, they lacked data for estimating (1) on-host tick mortality, yearly host tick burdens, and host behavior and home range dynamics where deer ticks are established; (2) all host seasonal movements (migration and juvenile dispersal) into unpopulated areas; (3) geographical barriers to *I. scapularis;* (4) length of the *I. scapularis* life cycle in different areas of the United States.

Upper Peninsula of Michigan tick population

During a study on the distribution of the deer tick in the southwestern U.P. of Michigan, Strand et al. (1992) found clusters of *I. scapularis* only in southern Menominee County. The clusters mostly occurred along the Menominee River bordering Wisconsin, Little Cedar River which runs southwesterly through south-central Menominee, and along the Walton River in southeastern Menominee County. Friedrich (2003) tested sites throughout Menominee County and into central Delta County; she also found deer ticks only in southern Menominee County.

Macroenvironmental factors in a corridor between Menominee and Marquette in the Upper Peninsula

Deer tick establishment and activity can be regulated by temperature, humidity, sunlight, endogenous rhythms and the presence of vertebrate hosts (Sonenshine, 1993). Since 98 % of the *I. scapularis* life cycle occurs off the host (Fish, 1993), various abiotic factors, including temperature and moisture, are likely to regulate off-host tick survival

(Bertrand & Wilson, 1996; Needham & Teel, 1991). The average temperature and rainfall of areas within the corridor between Marquette and Menominee fall within the same range as areas (such as Wisconsin and Menominee County) where deer ticks have established populations (Table 1; Brownstein et al., 2003; Kitron & Kazmierczak, 1997; National Climatic Data Center, 2002).

Brownstein et al. (2003) showed that average monthly low temperatures, monthly high temperatures and vapor pressure significantly contributed to the maintenance of deer ticks in the United States, and predicted that an average monthly minimum temperature below -7 °C in the winter would prevent an area from maintaining established populations. The average minimum temperatures present within Marquette and the corridor are below the -7 °C. However, other areas within Wisconsin that have established tick populations also are below the -7° C average minimum temperature during the winter months (Table 2; National Climatic Data Center, 2002). Ticks over-winter in the leaf litter where they are blanketed by snow and are not subjected to the air temperature at all times throughout the winter; therefore, they are protected from air temperatures throughout much of the winter (Ogden et al., 2004).

Land use distribution is similar in the corridor from Marquette County to Menominee County (Figure 1; Michigan Geographic Data Library Center for Geographic Information, 2001; Leatherberry & Spencer, 1993). Each county is heavily wooded (Menominee = 79 %, Delta= 80 %, Dickinson= 81 %, Marquette= 88 %), with the other land consisting of urban or crop usage. Menominee County, Marquette County and Delta County's forests are primarily fir or spruce species and maple species. Dickinson County forests are primarily fir or spruce species and aspen species (Table 3; Leatherberry & Spencer, 1993).

Host species in the western half of the Upper Peninsula

Deer populations are lower in size and density within the northern portion compared to the southern portion of the U.P. The southern portion (Menominee County, southern Dickinson County, and western Delta County) has an estimated 30.4 deer/ km^2 (202,319 deer). The middle portion (southern Marquette County and northern Dickinson County) has an estimated deer population of 18.1 deer/ km^2 (172,276 deer); and the northern portion (northern Marquette County) has an estimated 9.2 deer/ km^2 (100, 824 deer; Mayhew, 2003).

White-footed mice are widely distributed throughout central and eastern North America. The northern boundary of their distribution is the Great Lakes basin along the southern part of the U.P. (Kurta, 1995). Another host of immature ticks is the deer mouse (James & Oliver, 1990). This species of mouse has an extensive range that includes the entire U.P. (Kurta, 1995). The deer tick is able to feed on both species of mice in the laboratory (Hazler & Ostfeld, 1995) and in the field (Rand et al., 1993). Two studies have compared tick infestation on *P. leucopus* and *P. maniculatus* in the U.P. (Forst, 2003; Friedrich, 2003). In a study covering one year within a small area of Menominee County, Forst (2003) found equal numbers of both species of mice and no significant difference in number of ticks feeding on *P. leucopus* compared to *P. maniculatus.* In a two-year study that covered much of Menominee County, Friedrich (2003) found that significantly more larval deer ticks fed on *P. leucopus* ($n = 193$) than on *P. maniculatus* in one out of two years ($n = 222$; $p = 0.001$). However, in the first year, *P. leucopus* (n =

103) did not have significantly more deer tick larvae than *P. maniculatus* (n = 84). The number of nymphs per mouse did not differ between *P. leucopus* and *P. maniculatus* in either year. Friedrich (2003) hypothesized that the difference in levels of infestation between the two *Peromyscus* species may be due to variation in habitat preferences, mouse activity and host density.

White-tailed deer and deer tick movement in the Upper Peninsula

In the U.P., some white-tailed deer migrate biannually between summer and winter ranges that can be up to 50 km apart. Fawns learn these migration routes from their mothers, and tend to use the same ranges throughout their lives (Van Deelen, Campa, Hamady & Haufler, 1998). Northern portions of the U.P. receive more annual snowfall than southern portions throughout the winter. During a 29-year study (1971-2000) by the National Climatic and Data Center (2002), Marquette received on average 466 cm of snowfall annually, Iron Mountain (Dickinson County) received on average 164 cm of snowfall annually, Escanaba (Delta County) received an average of 129 cm of snowfall annually, and Menominee received an average of 168 cm of snowfall annually. The high snowfall and lack of food drive white-tailed deer south where less snow cover results in more available food. Traditionally, deer within Minnesota move south in December and move back north in April (Nelson, 1998).

The combination of mild winters, low snowfall, land cover distribution (deciduous stands mixed with lowland conifers and agriculture), and timber harvest patterns makes most of Menominee County a favorable area for deer (Michigan Department of Natural Resources ((MI DNR), 2005a). In spring, when the snow begins to melt, white-tailed deer move back north to their summer ranges. Usually snow depths of 10-30 cm and temperatures of 0.3-8.1° C are indicators for when deer depart their winter ranges (Nelson, 1995, 1998). Tagged deer from central Menominee County have moved to northern Menominee County and as far north as central Marquette County during the spring and into the summer (MI DNR, 2005a). This northward migration during tick season is a method that could move ticks out of Menominee County and into Dickinson, Delta and Marquette counties.

Adult ticks emerge in the spring when questing conditions are met. Host-seeking activities are influenced by a variety of factors including temperature, humidity, sunlight and presence of hosts (Vail & Smith, 2002). Although deer ticks have higher survival rates at higher temperatures (24-28 ° C), they are able to survive at lower temperatures (4-8 ° C) for short periods (Ogden et al., 2004). They can become active and start questing with air temperatures as low as -2 °C and 70 % snow cover (Carroll & Kramer, 2003). Ticks that emerge from the leaf litter at these lower temperatures quest briefly but then return to the leaf litter if they did not find a host (Ogden et al., 2004). In southern Menominee County, these threshold temperatures and snow cover levels are typically reached within the months of March and April (National Climatic and Data Center, 2002). Therefore, there is an overlap in adult tick questing and deer migration; and adult deer ticks may start questing at the same time as white-tailed deer start migrating north to their summer ranges.

Adult deer ticks feed for up to several weeks (Glavanakov et al., 2001) and white-tailed deer typically travel between their summer and winter ranges within a 14-day period (Nelson, 1995). Adult deer ticks also typically mate on the host. Thus, when an engorged female tick drops off, she would lay her eggs in that area. If it is an area where the tick's environmental needs (host presence, adequate soil and air moisture levels, and adequate soil and air temperature levels) were met, the eggs would hatch, the larvae may survive through adult stage, and then mate and produce more ticks within that area. In this manner, the deer tick could expand its range within the U.P.

OBJECTIVES:

First, because deer migrate from deer tick endemic regions north, I estimated the current range of *Ixodes scapularis* in the Upper Peninsula of Michigan in a corridor extending from the city of Menominee to the city of Marquette. Second, in order to determine if microenvironmental characteristics are potential limiting factors in the expansion of *I. scapularis* in the corridor of the U.P., I investigated soil characteristics (soil type, soil moisture, soil pH and leaf litter moisture) that the deer tick as been associated with on large, multi-state studies within chosen macroenvironmental characteristics (i.e., deciduous based forest and ample leaf litter). Third, I used published data on white-tailed deer movements in the U.P. to hypothesize what factors influence the distribution of deer tick population in the U.P.

MATERIALS AND METHODS:

The study was conducted May through September of 2004. A corridor (7410 km^2) between Menominee and Marquette was partitioned into 4 study areas: north, north central, central and south. Within each study area, flagging and trapping took place at multiple study sites on state forest land (MI DNR use permit 32-007-04), areas surrounding public campgrounds, and on private land. Sites as close as possible to those of Strand et al. (1992) were included (Figure 2). If an established deer tick population was found the first time it was flagged, that site was not sampled again. If only one deer tick, one life stage, or if no deer ticks were found, then the same site was sampled again or another site nearby (within 1 km) was sampled. This helped compensate for the patchy distribution of deer ticks.

Study site criteria

Study sites were chosen based on the following criteria: ≥ 60 % deciduous forest with ≥ 4 cm of leaf litter on the ground, ≤ 800 m of a body of water, and limited human disturbances (i.e., no campsites, houses, agricultural fields; French, 1995). Edges between field and forest were flagged as long as there was ≥ 4 cm leaf litter and low lying shrubby vegetation, because this is what adult and nymphal deer ticks climb up when questing for a host (Ginsberg & Ewing, 1989).

Flagging

Study areas were flagged for questing ticks once every four weeks from the end of May through the end of September 2004. Because deer tend to travel along trails (Kurta, 1995), flagging was limited to walking trails within the selected site. A white, flannel cloth (86 cm x 110 cm) was dragged across the ground and over low lying vegetation. Between 800 m and 1600 m of trail were flagged in each site, depending on the size of suitable area. The cloth was closely examined for ticks every 10 m (Schultz & Jordan, 2001). All ticks that had latched onto the cloth or me were removed and placed in labeled vials containing 70 % ethanol for later identification. Study sites were flagged for ticks in the morning (before 1200) or in the early evening (after 1700) when ticks were more likely to be questing (Kollars, Oliver, Kollars & Durden, 1999). Flagging was not done when it was raining because ticks do not quest during this time (Vail & Smith, 2002).

Trapping

Feeding ticks were sampled by live trapping small mammals from June through the end of September at various sites using techniques approved by Northern Michigan University Institute Animal Care and Use Committee #011 (Appendix C). All sites trapped also were flagged. Twenty-five to thirty-five Sherman live traps (25 cm x 7.5 cm x 7.5 cm) were set up in the late afternoon (after 1800) along an 800 m to 1600 m trail 5-20 m off of the trail, baited (oatmeal, peanut butter and bacon grease mixture) and checked the next morning. The number of traps depended on the number of available locations for traps (i.e., next to burrow openings, fallen tree stumps, areas with abundant brush, and other areas that provided ample food and hiding places for rodents) and security of each site for leaving traps overnight.

Trapped animals were anesthetized with halothane mixed with mineral oil (Forst, 2003). A few drops of the mixture was put on cotton balls and placed in the tip of a medium centrifuge tube, 25 mL or 50 mL depending on the size of the animal. The tube was then placed over the head of the animal and kept there until there was no response when its hind foot was pinched. The head and neck of trapped animals were carefully examined. All ticks were removed and placed into labeled vials containing 70 % ethanol. Saliva samples were collected from all mice using a cotton swab dipped in physiological saline and placed in labeled vials in order to identify species of mouse by salivary amylase testing. Samples were put on ice, transferred to the lab and stored at -20 °C. External characteristics (tail length, hind foot length, and ear length, color pattern on tail and body, and degree of pencillation on the tail) were noted in case salivary amylase identification was unsuccessful.

Sampling procedure

During the first week of the four-week period, four to six sites in the north area were sampled. In the morning of day one, the first site was flagged. In the afternoon, the second site was flagged and traps were set. On day two, traps were checked in the morning and site three was flagged and traps were set. This pattern continued throughout the week. Thus, not all sites were both trapped and flagged due to location of the site and time constraints. The following week, four to six sites were sampled in the north central area. This procedure continued until four to six sites in each area of the corridor had been sampled each month. Once an established deer tick population was found within a specific site it was not checked again. At the time of each sampling, air temperature and soil temperature were measured using field thermometers; soil pH, available water capacity and leaf litter moisture was measured with a Kelway Soil Moisture and pH Tester (Kel Instruments Co., Inc.). Forest tree composition (oak (*Quercus* spp)., maple (*Acer* spp.), aspen (*Populus* spp.), pine (*Pinus* spp.) or cedar (*Cedrus* spp.)) was determined at each site by walking the sampled trail and counting each tree that was ≥ 1.5 m tall, ≥ 5.0 cm dbh, and had limbs that stretched over the flagged trail. These were trees that likely contributed the most to the ground cover within the study area. Available water capacity (soil moisture) and pH measurements obtained at each site were compared to soil surveys conducted by the USDA NRCS to reduce uncertainty from a one time sample because pH and moisture levels can fluctuate within the year due to weather conditions (temperature and rainfall; USDA NRCS, 1999). In this way I could determine if the samples I obtained were typical for the area or were abnormal measurements.

Each site's soil was classified as sandy, loamy or other by exactly locating each site on USDA NRCS maps (Berndt, 1989; Linsemier, 1989; Schwenner, 1989; USDA NRCS, 1995, 1997). The soil was then classified according to its percentage of sand, silt and clay. "Sandy" soil was ≥ 90 % sand, "loamy" soil was 50-60 % sand, and around 25% silt and clay. "Other" soil consisted of soil that was 60-90 % sand, and 10-30% silt and/or clay. Sites were allocated into one of five moisture categories (very low to very high) based on soil available water capacity defined by the USDA NRCS (Berndt, 1989; Linsemier, 1989; Schwenner, 1989; USDA NRCS, 1995, 1997).

At the end of July, halfway through the study period, no deer ticks had been found in the north, north central or central area. During the last half of the study (August and September), more intense sampling took place within the southern area (Menominee County and southern Dickinson and Delta County) to help define more specifically the extent of deer tick's range in the corridor.

Laboratory work

Ticks were identified using various classification keys for adults, nymphs and larvae (Clifford, Anastros & Elbl, 1961; Keirans & Litwak, 1989; Keirans, 1990). Morphological identification of *Peromyscus leucopus* and *P. maniculatus* (e.g., tail length, ear length, hind foot length, and tail and body colorization) is difficult because these species overlap in these measurements. For example, tail lengths of adult white-footed mice range from 50 to 88 mm, whereas tail lengths of adult deer mice range from 70 to 105 mm (Kamler, Pennock, Welch & Pierotti, 1998; Lindquist, Aquadro, McClearn & McGowan, 2003). A more accurate way to identify *Peromyscus* is by electrophoresis of salivary amylase (Lindquist et al., 2003; Long & Long, 1993; Palas, Schwartz &

Vivas, 1992). Four electromorphs of this enzyme (*Amy*-1^{50}, *Amy*-1^{65}, *Amy*-1^{76}, and *Amy*-1^{85}) are unique to the deer mouse. Two electromorphs of this enzyme (*Amy*-1^{100} and *Amy*-1^{109}) are unique to the white-footed mouse (Aquadro & Patton, 1980; Lindquist et al., 2003).

Saliva samples were analyzed using modified procedures of Aquadro and Patton (1980). In the laboratory, saliva samples were thawed in 15 µL of physiological saline, and then centrifuged at 3000 x g for approximately 5 minutes. Ten µL of salivary sample were then added to 5 µL of Tris-glycine native sample buffer (2x; 4 M Tris HCl, 10 % glycerol, 0.1 % bromophenol blue, pH = 8.6). Nine µL of the dyed sample were then placed in a well of 8 % Tris-glycine gels (1.0mm x 10 well, Invitrogen Novex pre-cast gels) with 10x native running buffer (25 mM Tris base, 192 mM glycine, pH = 8.6) diluted to 1x in an upright gel apparatus and run at a constant 45 volts for 20 minutes. The gel was then run at a constant 25 mA until bromophenol blue tracking dye bands were ¾ of the way down the gel. The gel was then immersed in a starch solution (2.0 % Tris-HCl, 1.5 % calcium chloride, and 10 g/L soluble potato starch) and stored at 4 °C for approximately 12 hours. The gel was then rinsed three times with distilled water and stained with a tri-iodine solution (6.34 g iodine and 4.15 g potassium iodide per 500 ml) for approximately 3 minutes. After staining, gels were rinsed again with distilled water and placed in a clearing solution (5 parts methanol: 5 parts distilled water: 1 part glacial acetic acid) until desired clearing of the gel was obtained. The gel was then photographed and migration of each protein band was measured and compared to controls to determine species.

Statistical analysis

All statistical tests were run using SPSS version 13.0 (SPSS Inc., Chicago, IL), with a significance level determined at $p \leq 0.05$. Data sets for analyses included only sites within the southern area of the corridor, since this was where established deer tick populations were found. Forward stepwise (conditional) logistic regression was used to investigate the importance of soil type (sandy, loamy, or other), soil moisture class (available water capacity), soil pH and leaf litter moisture (%) in characterizing sites with an established deer tick population. Log-linear analysis was used to test the interaction between available water capacity class and deer tick establishment. The Mann-Whitney U-test was used to test whether soil pH differed between sites that had an established deer tick population and sites that did not. Contingency table analysis was used to test whether the proportion of established deer tick sites was independent of soil type. Chi-square analysis was used to compare prevalence and intensity of deer ticks on *P. leucopus* and on *P. maniculatus.*

RESULTS:

Study sites

Sixty-nine sites were flagged and/or trapped for deer ticks in the corridor between Menominee and Marquette cities (Figure 3). Within the north area, a total of 9 sites were each flagged twice, 4 were each trapped twice. Within the north-central area, 7 sites were each flagged twice; 3 were each trapped twice. Within the central area, 10 sites were sampled; 4 of the 10 sites were flagged twice and 3 of the 4 were trapped twice. Within the south area 43 sites were sampled. Six sites were flagged twice, 15 sites were trapped with 5 sites trapped twice.

Only 2 species of ticks were found, *I. scapularis* and *Dermacentor variabilis* (American dog tick). A total of 81 *I. scapularis* were collected (40 adults, 12 nymphs, 29 larvae) at 13 of 69 sites. Twelve of the 13 had established deer tick populations (at least 6 ticks collected, or 2 of the 3 life stages identified); one adult deer tick was located while flagging at 1 site and therefore the site was considered to have a reported tick population. All established and reported deer tick sites were in the south study area (Figure 4). A total of 541 *D. variabilis* (183 adults, 321 nymphs, and 37 larvae) were collected in 35 out of 69 sites. These ticks were collected in all 4 areas of the corridor (5 out of 9 in the north area, 7 out of 10 in north-central area, 5 out of 7 in central area, and 18 out of 43 in the south area. All sites with *D. variabilis* had established populations. All adult ticks were collected by flagging, nymphs and larvae were collected by both flagging and trapping. Twenty-nine deer ticks (7 nymphs and 22 larvae) were collected through trapping.

Environmental factors

Of the sites studied within the southern area, soil pH ranged from 3.5 to 7.0, with established sites ranging from 4.2 to 7.8. Of the 12 sites with established deer tick populations; 7 had low available water capacity class, 3 had moderate available water capacity class, and 2 sites had very high available water capacity class. There were no established sites within the high available water capacity class (Table 4). The site with reported deer tick populations had a very high available water capacity. Leaf litter moisture ranged from 0.1 % to 80 %, with established sites ranging from 30 % to 80 %, and the reported site at 25 %. Of the 12 established sites; 5 were classified as sandy soil, 6 were classified as loamy soil, and 1 was classified as other soil. The site with a reported deer tick population had loamy soil. All "other" soil was either sandy/loam or loamy/sand soil and had 60-90 % sand, and a mixture of silt and clay.

Within ranges exhibited by these 43 sites, soil type, leaf litter moisture, and soil pH were not important factors for the establishment of deer ticks in a site (Table 5). Only available water capacity class had a significant effect on whether deer tick populations were established at a site, and a model with available water capacity class present fit the data better than a model without available water capacity class (change in -2LL G = 10.335, df = 3, p = 0.016). In a reduced model (ln (odds of established deer tick population) = constant + available water capacity class), the effect of available water capacity on the odds of the event was significantly different from 0 (Wald = 8.203, df = 3, p = 0.042). Thirty percent of the variation of established sites could be explained by available water capacity class (Nagelkerk $R^2 = 0.296$).

In general, the proportion of established deer tick sites in each available water capacity class appears to decrease as moisture level increases, with most established deer tick sites found within areas with low available water capacity and the most un-established sites found within areas with high available water capacity (Z = 2.936, p = 0.003, odds ratio = 2.987; Figure 5). There was no association between tick establishment and pH (n = 43, U = 197.5, p = 0.918); or tick establishment and soil type ($X^2 = 3.638$, df = 2, p = 0.162).

Hosts

A total of 46 small mammals (43 mice, 1 chipmunk and 2 voles) were trapped during 580 trap nights (12.6 % success) at 26 sites. All mice were identified through salivary amylase electrophoresis. Amylase bands for *P. leucopus* were identified at an average distance of 2.8 cm, while amylase bands for *P. maniculatus* were identified at an average distance of 2.1 cm from the top of the gel, respectively. Only mice were infested with deer ticks. Trapping took place at 5 out of the 12 established deer tick sites, and a total of 17 mice were trapped within these established sites (8 *P. maniculatus* and 9 *P. leucopus*). Four out of 8 trapped *P. maniculatus* were infested with *I. scapularis;* all 9 trapped *P. leucopus* were infested with *I. scapularis.* Both species of mice were trapped at 3 of the 5 sites (4 *P. maniculatus,* 5 *P. leucopus*). These were the mice used to compare the prevalence and intensity of deer ticks on *Peromyscus* spp. Infestation intensity of *P. maniculatus* was 4.0 ticks/infested mouse whereas *P. leucopus* intensity was 1.6 ticks/ infested mouse. Tick prevalence for *P. maniculatus* was 3 infested mice/ 4 mice tested, and *P. leucopus* was 5 infested mice/ 5 mice tested. There was no significant difference in the intensity ($X^2 = 0.667$, df = 1, p = 0.414) and prevalence ($X^2 = 3.571$, df = 1, p = 0.059) of *I. scapularis* on either species of mice.

DISCUSSION:

According to my findings, the deer tick has not expanded its range in the U.P. within the corridor from Menominee to Marquette, nor was it found in any sites further north from its range in 1990. Though people have been diagnosed with Lyme disease within the corridor in Menominee, Marquette and Delta Counties (Centers for Disease Control and Prevention, 2004), no deer ticks were found outside of Menominee County throughout my study, nor in a 2 year study by Friedrich (2003). Lyme disease cases in a locale are not definitive evidence of deer tick populations in that locale because infection can occur elsewhere, but be diagnosed in a deer tick free area. However, the deer tick does have a patchy distribution. Therefore, deer ticks may have been present outside of

Menominee County within sites that were not sampled and therefore were not found throughout this study. Expansion of the deer tick range requires three conditions. (1) Free living stages must be able to survive within the habitat (i.e., suitable environmental conditions) and find hosts for all stages. (2) Infected hosts must move ticks to new areas. (3) Adult deer ticks must be able to find mates within the new area.

Environmental characteristics

My study indicated that deer ticks have suitable environmental factors and hosts within the corridor. Lack of association of 3 soil characteristics (soil type: sandy, loamy, or other; leaf litter moisture; and pH) with the presence of deer ticks among sampled sites suggests that deer ticks are able to survive within the ranges of these conditions present in the corridor, and therefore are not limited by environmental factors within the corridor in the U.P.

Deer tick sites could be negative due to any of 3 situations: (1) Sites could be negative because some unrecognized factor prevents the establishment of deer ticks. Many factors, such as habitat type (Adler et al., 1992; Ginsberg & Ewing, 1989; Guerra et al., 2002; Lindsay et al., 1999; Lubelczyk et al., 2004; Ostfeld et al., 1995; Padgett & Lane, 2001; Talleklint-Eisen & Lane, 2000; Van Buskirk & Ostfeld, 1998), soil characteristics (Bunnell et al., 2003; Guerra et al., 2003), host presence (Barry & Francq, 1980; Duffy et al., 1994; Glavanakov et al., 2001; James & Oliver, 1990; Kilpatrick et al., 1994; Witt & Huntly, 2001), elevation (Bunnell et al., 2003; Nicholson & Mather, 1996), glacial outwash (Jackson & DeFoliart, 1970) and climate (Brownstein et al., 2003) have been studied as possible factors in the establishment of deer ticks in an area. Various elements of habitat type, soil characteristics, and host presence were examined in this study. Elevation and glacial outwash were not examined, since the elevation (Bunnell et al., 2003; Nicholson & Mather, 1996) and glacial outwashes (Jackson & DeFoliart, 1970; Kitron & Kazmierczak, 1997) present within the corridor are the same as areas within Wisconsin that have established deer tick populations. The climate (average monthly minimum, average monthly maximum and average precipitation) within the corridor of the U.P. is similar to areas in Wisconsin where there are established deer ticks (National Climatic Data Center Records, 2002). (2) A negative site could be a false negative site where deer ticks were established but not detected. However, Friedrich (2003) also did not find deer ticks outside Menominee County within her Delta County sites. The sampling techniques used (flagging and trapping) appeared adequate for detecting ticks, since dog ticks were found in sites where deer ticks were not found, and in sites where deer ticks were established. Dog tick established sites were located throughout all 4 sampling areas. (3) A negative site could be a site that can sustain a deer tick population, but deer ticks had not yet been introduced. The expansion of the deer tick is strongly associated with the increase in white-tailed deer in the United States (Dennis, Nekomoto, Victor, Paul & Piesman, 1998; Rand et al., 2003). This expansion suggests that all suitable deer tick habitat is not yet occupied (Brownstein et al., 2003). The range of environmental factors present within the corridor in the U.P. appears sufficient to support deer ticks. Therefore, the un-established sites could be suitable, but deer ticks have not yet been introduced into the area.

Deer ticks were more common within dry to mesic forests with soil with a lower available water capacity (Bunnell et al., 2003; Guerra et al., 2002). There must be enough moisture within the soil and leaf litter for deer ticks to avoid desiccation, but too much can be detrimental. High water capacity soils can reduce tick populations by fostering fungal colonization in ticks (Yoder et al., 2003; Zhioua et al., 1999). Available water capacity class was a better indicator than leaf litter moisture for tick presence. Leaf litter moisture was a one time measurement that can fluctuate constantly throughout the tick season. Available water capacity class is a range, which encompasses the fluctuation of moisture levels that occurs throughout the season.

In contrast to my study, Guerra et al. (2002) and Bunnell et al. (2003) reported that soil texture and acidity of soils were important factors in the distribution of the deer tick. However, Guerra et al. (2002) and Bunnell et al. (2003) selected sites to represent various regions and habitats in the Midwest and Mid Atlantic (Midwest: 113 in Wisconsin, 3 in Michigan, and 22 in northern Illinois); (Mid Atlantic: 12 in southern New Jersey, 12 in southern Pennsylvania, 10 in Delaware, 38 in Maryland, 8 in Virginia). Because both studies chose sites at random within plots or transects, study sites within less suitable habitat (i.e., fields) for deer ticks were sampled. In contrast, my sampling strategies selected for suitable sites for deer ticks based on macroenvironmental characteristics. Thus, their study included a wider range of soil characteristics (soil type, soil moisture and soil pH) compared to mine.

There are deciduous and mixed forested areas within the corridor between Marquette and Menominee that can provide the required habitat for free-living deer ticks. A highly fragmented habitat with large plots of open land between the forested areas could be detrimental to the deer tick establishment, since if they drop off in the open area they have a lower chance of survival. The land distribution map (Figure 1) of the corridor shows that there are deciduous and mixed forested lands throughout the corridor in continuous or slightly fragmented distribution. Deer tend to travel through deciduous based forested areas (Nelson, Mech & Frame, 2004), which increases the probability that deer ticks will drop off in an area where they will have a higher survival rate.

Hosts

Deer ticks were found on both deer mice and white-footed mice throughout this study, which supports Freidrich (2003) and Forst (2003). Intensity and prevalence of deer ticks on *P. leucopus* (1.6 ticks/ infected mouse, 100 % infected) and *P. maniculatus* (4 ticks/infected mouse, 75 % infected) were similar. Forst (2003) and Friedrich (2003) also found that deer ticks parasitized both species of mice in the U.P. (Table 6). Deer ticks feed on both species of mice in the field (Rand et al., 1993) and in the laboratory (Hazler & Ostfeld, 1995). Therefore, there are suitable hosts to support immature ticks across the U.P.

Because deer ticks do not move more than a meter away from where they have fallen off of their host, far-ranging hosts such as migratory birds and white-tailed deer have a higher chance of expanding the deer tick's range than small ranged hosts (i.e., small rodents; Glavanakov et al., 2001; Madhav et al., 2004). Deer ticks typically quest during the morning hours after dew has evaporated and during late evening hours (Vail & Smith, 2002), which is the time of day that white-tailed deer are foraging for food and moving around (Nelson et al., 2004). The major overlaps in times and places for deer feeding and for deer tick questing increase the chances of deer ticks finding hosts. Migratory birds can transport immature ticks great distances, but the timing of bird migration (spring and summer) with immature tick questing would tend to disperse larvae in a southward direction and nymphs in a northward direction; adult deer ticks are seldom found on birds (Madhav et al., 2004). Transported larvae and nymphs would have a lower probability of populating a new area than adults since immature ticks have to molt 1 to 2 times before becoming an adult, finding a mate, and surviving until eggs are laid. This would take time, and the mortality rate of ticks is high. In contrast, a transported engorged adult female tick lays an average clutch size of 2400 eggs (Sonenshine, 1993), and therefore has the potential to begin a new generation within that area in a shorter amount of time.

White-tailed deer move out of the Menominee area north into Delta, Dickinson and Marquette counties during tick season (MI DNR, 2005a). Out of 39 deer that were tagged within Menominee County and then spotted between 1990 and 2001, at least 4 (~10%) traveled from deer tick established sites north to un-established sites within the corridor at times when they could be carrying deer ticks (Figure 6). Of the 4 tagged deer that moved out of the tick endemic area, 3 moved into northern Menominee County and 1 moved into southeastern Marquette County. There are an estimated 19.1 deer/km^2 (37,002 deer) in southern Menominee County within the deer tick endemic area (MI DNR, 2005b). If 10 % of deer move out of Menominee County in 11 years (1990-2001), then roughly 3700 deer (336 deer/year) moved out of the tick endemic area at a time when they might be carrying feeding deer ticks. Assuming that 25 % of these deer migrated out of Menominee County, 925 deer (84 deer/year) would have moved from the tick endemic area of Menominee County into other counties within the corridor. These are very rough estimates of the number of deer moving out the of the endemic area of Menominee County, since the white-tailed deer population within the U.P. fluctuated from 1990 to 2005, when the estimated deer count was taken (MI DNR, 2005b).

However, even though it is a rough estimate of deer movement, it shows that white-tailed deer may be moving out of the endemic area of Menominee County. Deer ticks have the chance of attaching to and feeding on deer that are moving out of the current endemic area of Menominee County and therefore being moved into a new area within the corridor.

The third condition needed for deer tick expansion is creating new generations within the new area; therefore, adult ticks must be able to find a mate. Birth and death rates determine the abundance of populations at any particular point in time. Tick fecundity is dependent on the female mating with a fertile male, engorging sufficiently, and surviving until eggs are laid (Fish, 1993). An overall mortality as high as ~ 99.9 % would allow each adult deer tick to be replaced by another adult in the next generation. Actual mortality of ticks in the field is high, but is probably lower than that (Fish, 1993). Therefore, even though deer ticks have a high mortality rate from one stage to the next, an adult female tick produces enough offspring to populate a suitable area.

In deer ticks' mating occurs more often while on the host than off the host (Fish, 1993). Mates find each other with sex attractant pheromones (Wilson & Deblinger, 1993). Mated females engorge rapidly, detach and drop off. They then lay their eggs in a sheltered area where they dropped off. Mated males usually remain, reattach, feed again, and mate with other females (Sonenshine, 1991). If the habitat the female drops into meets the requirements of the deer tick (e.g., appropriate moisture levels, forest, soil characteristics, host density), a new generation may hatch. These new larvae will have to survive until adulthood and find mates in order to produce offspring. Therefore, the number of deer moving out of Menominee County is not the only important factor.

Prevalence and intensity of infestation on deer is also important, since this would determine the density of deer ticks in the area and the probability of deer ticks finding a mate. The size of the corridor studied between Menominee and Marquette was 7410 km^2 (non-endemic portion of corridor = 5035 km^2). If roughly 84 deer move out of the endemic area of Menominee County per year into other counties of the corridor, the relative density of deer that could be carrying a deer tick into the corridor is 1.6 deer/ 100 km^2, assuming 100 % prevalence. A low density of deer infested with deer ticks moving into new areas contributes to a low probability of deer ticks becoming established there.

Densities of white-tailed deer in the western U.P. (9.2 to 30.4 deer/ km^2 within the corridor; Mayhew, 2003) appear high enough to sustain deer tick populations. Bluff Point Coastal Reserve in Groton, Connecticut sustained deer tick populations with a white-tailed deer density of 73.4 deer/ km^2 in 1993, decreased to 13.5 deer/ km^2 in 1997 and again decreased to 10.42 deer/ km^2 in 2001 (Stafford et al., 2003). Duffy et al. (1994) found that areas on Long Island, New York with no observed deer maintained established deer tick populations because medium sized animals (i.e., raccoons, foxes, opossums and feral and domestic dogs) acted as hosts. Therefore, white-tailed deer are not needed in order to sustain a deer tick population. The deer population in the U.P. is large enough to sustain a deer tick population, but may not be large enough to move enough deer ticks into new areas to create an established population.

Another component of tick expansion is tick abundance. The number of ticks that are introduced in an area depends on the number of deer ticks that attach onto hosts that move there. As deer ticks decrease in abundance the probability of a deer tick being on a deer that moves out of Menominee County decreases. The deer tick population within

Menominee County is considered a low density population (Guerra et al., 2002). Deer tick abundance is determined using an index of the number of ticks collected while flagging/ hour, ticks collected/certain distance, or the number of immature ticks found on small rodents. The abundance of deer ticks within Menominee County between 1990 and 2004 has stayed relatively stable, with only a slight increase in the density of deer ticks in the last 14 years. This can be seen using the index level of the number of immature ticks on rodents trapped. In 1990 the relative abundance of deer ticks was 0.9 larvae/ mouse trapped, 0.1 nymphs/ mouse trapped (237 *Peromyscus* spp. where deer ticks were found; Strand et al. 1992). In 2004 the relative abundance of deer ticks was 1.3 larvae/ mouse trapped and 0.4 nymphs/ mouse trapped (17 *Peromyscus* spp. within areas where deer ticks were found). The number of ticks collected/ drag hour (10.4/ hour; Strand et al. 1992) and ticks collected/distance flagged (1.2 ticks/ 500 m) could not be compared. Abundances may be limited in Menominee County by minimum monthly temperatures in the winter (Brownstein et al. 2003). The low deer tick density within Menominee County may result in few deer ticks feeding on relatively few deer moving out of the endemic area. Therefore, deer ticks may be able to survive within the site where they dropped off the deer, but are unable to find a mate once they become adults. The deer ticks are then unable to produce offspring, the local population of deer ticks goes extinct, and the deer ticks range has not expanded.

In summary, deer ticks were not found outside of Menominee County throughout my study and it seems they have not expanded their range northward from southern Menominee County over the past 14 years. The deer tick's range does not appear limited in the Upper Peninsula by environmental characteristics (forest type, distance to water, hosts, soil type, soil moisture and soil pH). Because deer ticks feed on both the deer mouse and the white-footed mice, the white-footed mouse is unlikely to be limiting the deer tick's range. However, low number of deer ticks attaching to a low number of deer migrating from the endemic sites to new areas could be major factors preventing the spread of the deer tick in the U.P. If the white-tailed deer population increases, then more deer may migrate in order to find food (Nelson, 1998; Van Deelen et al., 1998). The increase in white-tailed deer migration may increase the probability of deer ticks being moved to new areas.

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APPENDICES

APPENDIX A

TABLES

Table 1: Average temperature and rainfall for selected cities of the western Upper Peninsula and Wisconsin (National Climatic Data Center 2002)

Cities/towns	Avg. precipitation (cm)	Avg. temperature (low) °C	Avg. temperature (high) °C
Marquette, MI	92.3	-7.7	19.1
Sawyer, MI	92.2	-11.4	18.0
Rock, MI	84.4	-8.9	19.2
Felch, MI		-10.8	19.7
Escanaba, MI	72.5	-8.8	19.2
Spalding, MI	80.7	-10.7	19.4
*Menominee, MI	158.1	-10.7	19.6
*Hazelhurst, WI	77.9	-2.1	9.8
*Gordon, WI	79.6	-2.1	10.8
*Sparta, WI	83.8	0.9	13.1

* Areas that have an established deer tick population

Table 2: Average minimum temperatures for winter months in selected cities of the western Upper Peninsula and Wisconsin (National Climatic Data Center 2002)

	November	December	January	February	March	April
Marquette, MI	-2.0	-8.3	-11.7	-10.2	-5.4	0.4
Sawyer, MI	-5.3	-12.1	-15.9	-14.8	-9.8	-2.8
Escanaba, MI	-2.8	-9.5	-13.9	-13.4	-7.9	-1.0
*Stephenson, MI	-5.0	-12.5	-17.2	-15.1	-8.4	-1.4
*Hazelhurst, WI	-6.2	-14.7	-33.5	-16.9	-10.4	-2.6
*Gordon, WI	-6.6	-15.5	-34.7	-17.1	-9.4	-2.1
*Sparta, WI	-4.0	-12.1	-16.1	-12.6	-5.4	1.2

* Areas that have an established deer tick population

Table 3: Land usage in Marquette, Delta, Dickinson, and Menominee Counties in the Upper Peninsula of Michigan (Leatherberry and Spencer 1993).

Land usage (%)	Marquette County	Delta County	Dickinson County	Menominee County
Forested land	88	80	81	79
Maple	45	59		27
Fir and spruce	23	39	32	39
Aspen			30	
Other	12	20	19	21

Table 4: Proportion of established deer tick sites for each class of available water capacity defined by the United States Department of Agriculture Natural Resources Conservation Service (Berndt 1989, Linsemier 1989, Schwenner 1989, USDA NRCS 1995, 1997)

Available water capacity class	Available water capacity (cm/cm)	Proportion of positive sites
Very low	0.000 - 0.050	0/ 0
Low	0.051 - 0.100	7/12
Moderate	0.101 - 0.150	3/13
High	0.151 - 0.200	0/ 1
Very high	> 0.201	3/17

Table 5: Beginning logistic regression model variables and their scores of importance on the establishment of deer ticks in an area. Within soil type "other" soil acted as the referent, as did high available water capacity class. Only variables with a p value ≤ 0.05 was added into the final equation.

Variables	Score	d.f.	P-value.
Soil type	1.724	2	0.422
Sandy	1.724	1	0.189
Loamy	0.146	1	0.703
Moisture class	10.320	3	0.016
Low	9.778	1	0.002
Moderate	0.354	1	0.552
Very high	4.011	1	0.045
Leaf litter moisture	0.232	1	0.630
Soil pH	0.039	1	0.844

Table 6: Intensity and prevalence of *Ixodes scapularis* on *Peromyscus maniculatus* and *Peromyscus leucopus* within sites where both species of mice were trapped in 3 studies conducted in the Upper Peninsula of Michigan (Forst (2003), Friedrich (2003), and my study).

Study	Species	# mice infested	Prevalence** (%)	# ticks	Intensity*
My study	*P. leucopus*	5	1.00	8	1.6
	P. maniculatus	4	0.75	12	4.0
Friedrich(2003)	*P. leucopus*	101	0.52	659	6.5
	P. maniculatus	80	0.36	269	3.4
Forst (2003)	*P. leucopus*	24			8.2
	P. maniculatus	16			6.9

*Intensity = number of ticks per infested mouse
**Prevalence = number of infested mice/ number of mice examined

APPENDIX B

FIGURES

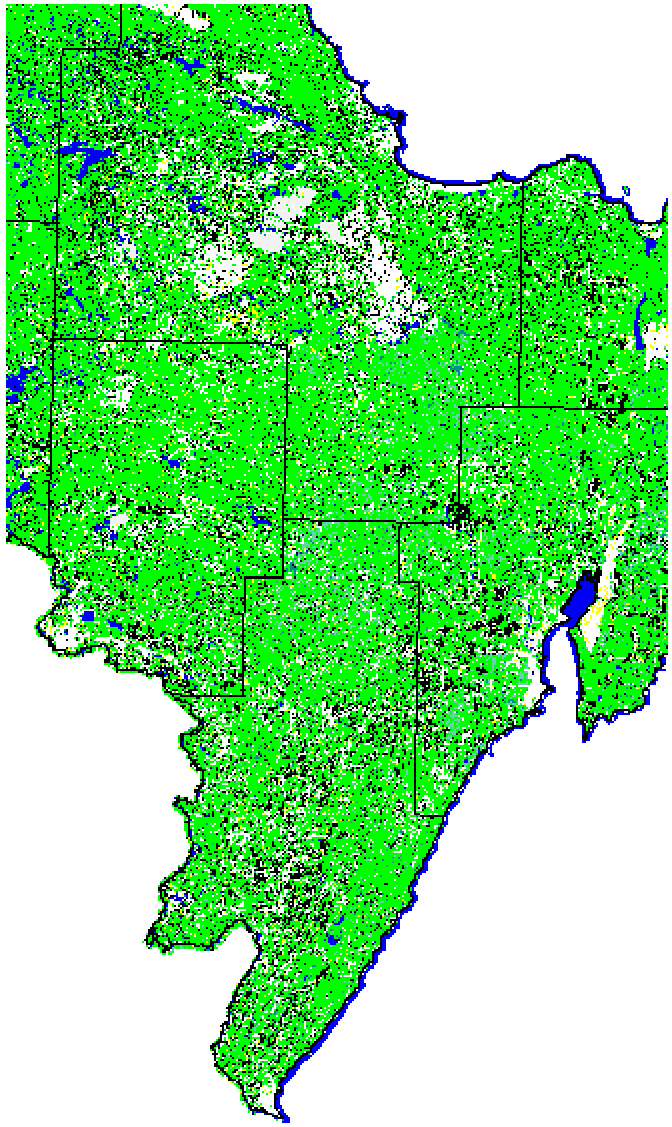

Figure 1: Land distribution map of the corridor between Marquette and Menominee Counties in the Upper Peninsula of Michigan.
Black: deciduous forests (upland and lowland), green: mixed forests (upland and lowland), white: coniferous forests, urban and open rangeland, blue: water. (Michigan Geographic Data Library, Center for Geographic Information 2001, http://www.mcgi.state.mi.us/mgdl).

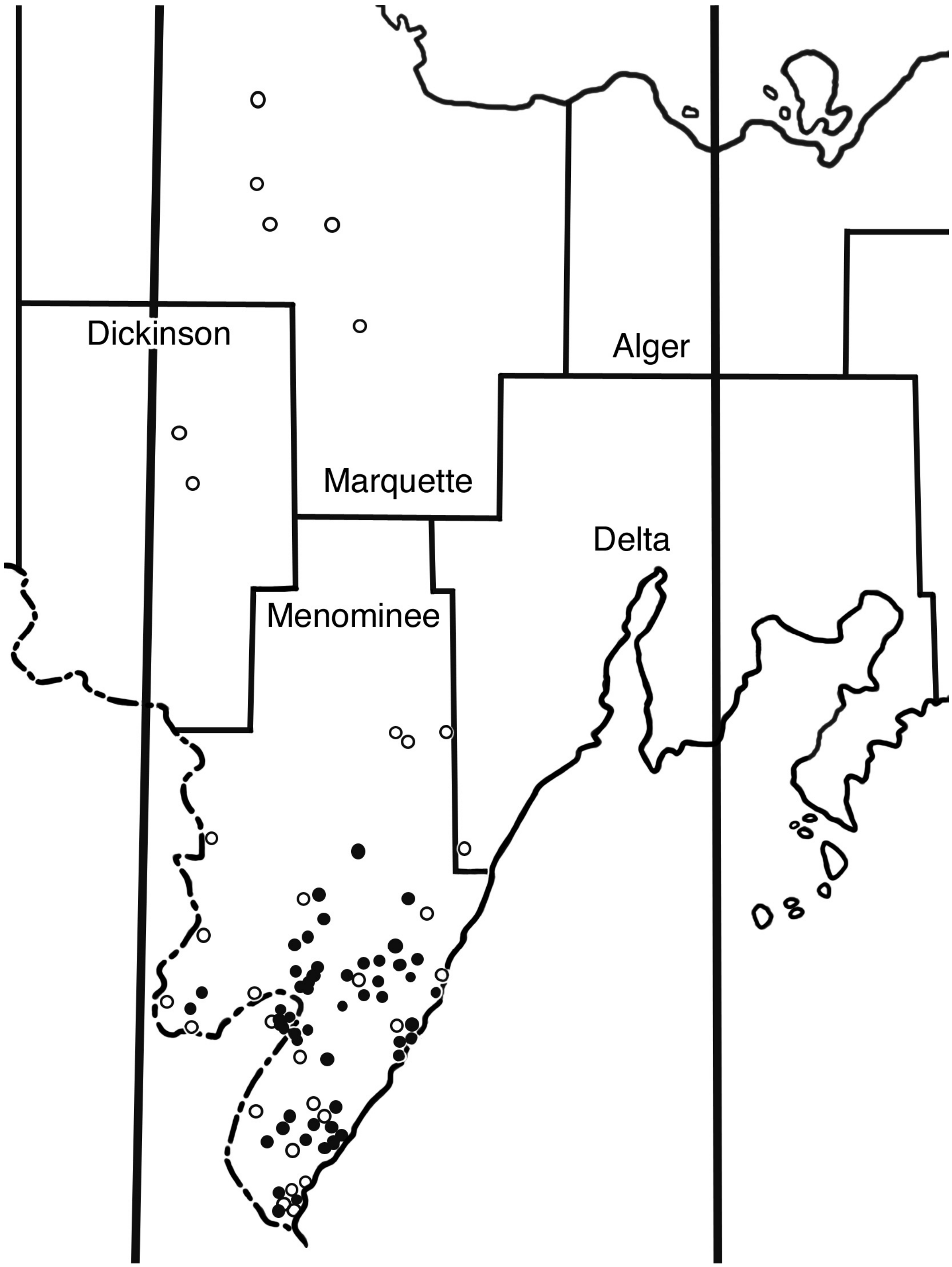

Figure 2: Sites sampled for *Ixodes scapularis* by Strand et al. (1992).
o represents negative sites for deer ticks (n = 36), and ● represent positive sites for deer ticks (n = 43).

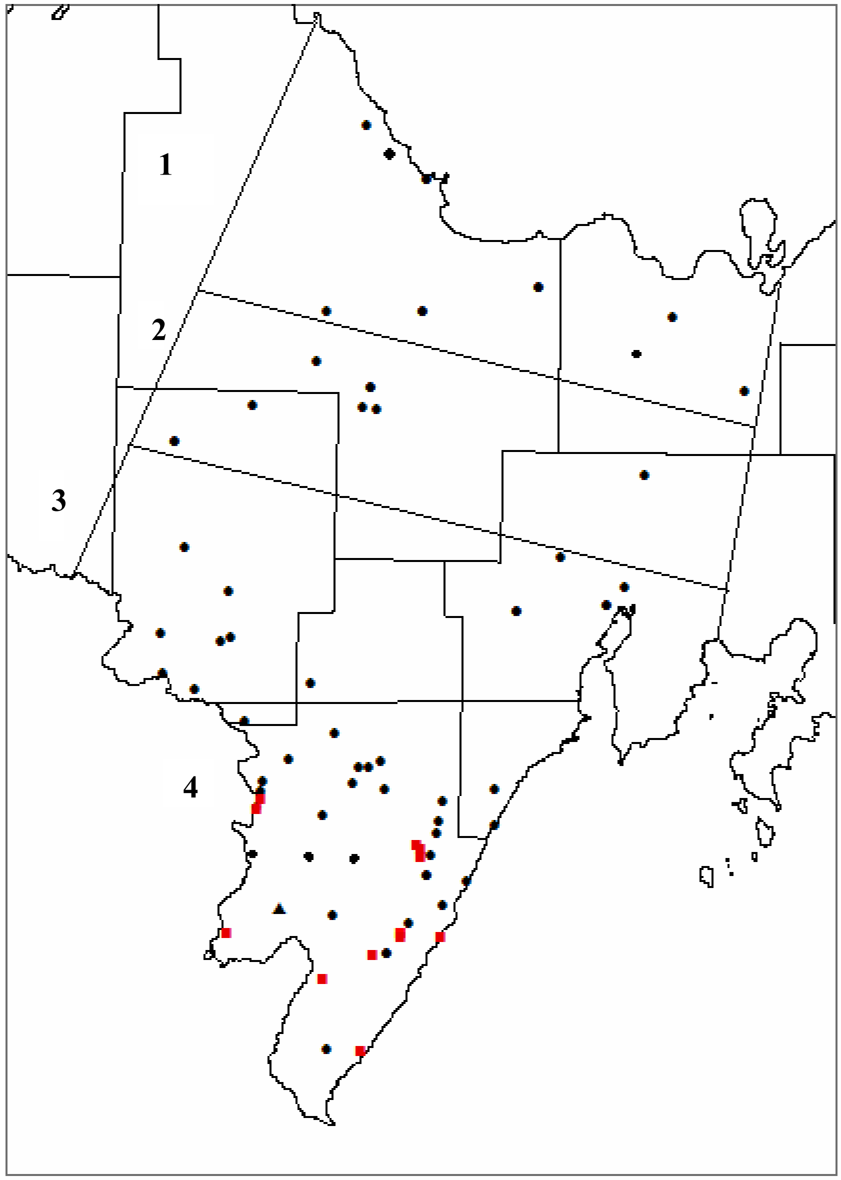

Figure 3: Sixty-nine study sites within the corridor from Menominee to Marquette, Michigan.
1= North area, 2= North-central area, 3= Central area, 4= South area.
● represents sites that were negative for deer tick populations, ■ represents sites that had an established deer tick populations, ▲ represents the site that had a reported tick population.

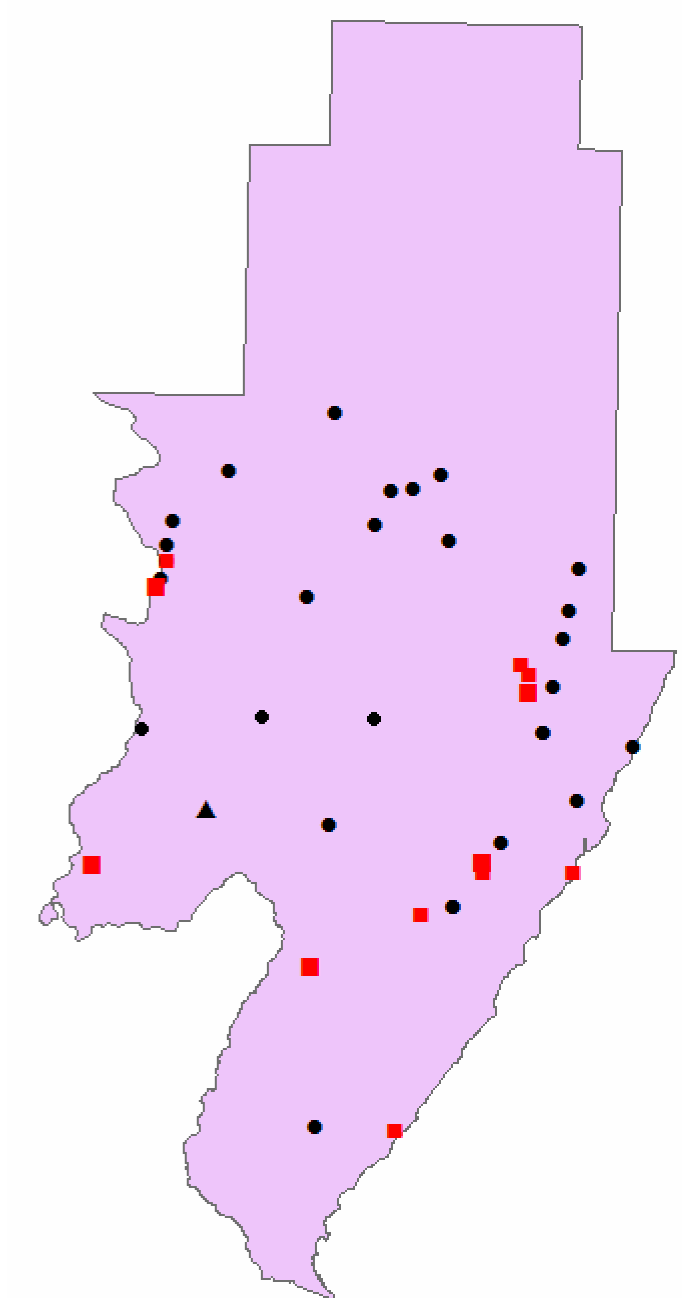

Figure 4: Thirty-six study sites within Menominee County, Michigan.
● represents sites un-established for deer ticks, ■ represents areas with an established deer ticks, and ▲ represents the site with the reported tick population.

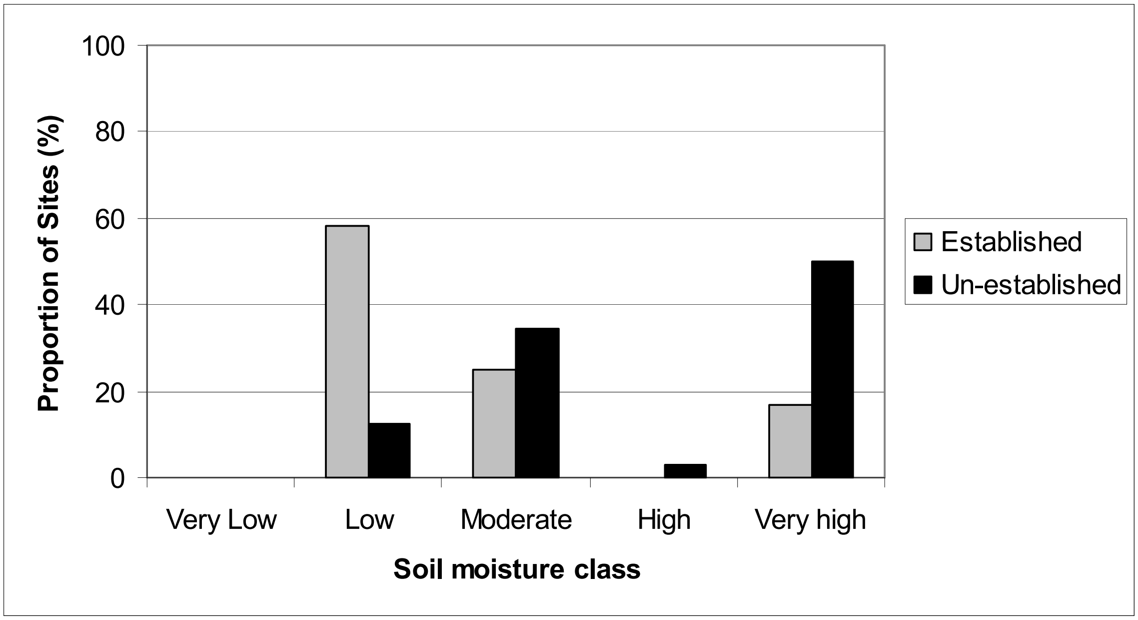

Figure 5: Soil moisture levels for 12 sites with established deer tick populations and for 32 sites with un-established deer tick populations within the south section of the corridor. Soil moisture levels are based on available water capacity levels indicated by the United States Department of Agriculture Natural Resources Conservation Service.
Refer to Table 4 for moisture level values.

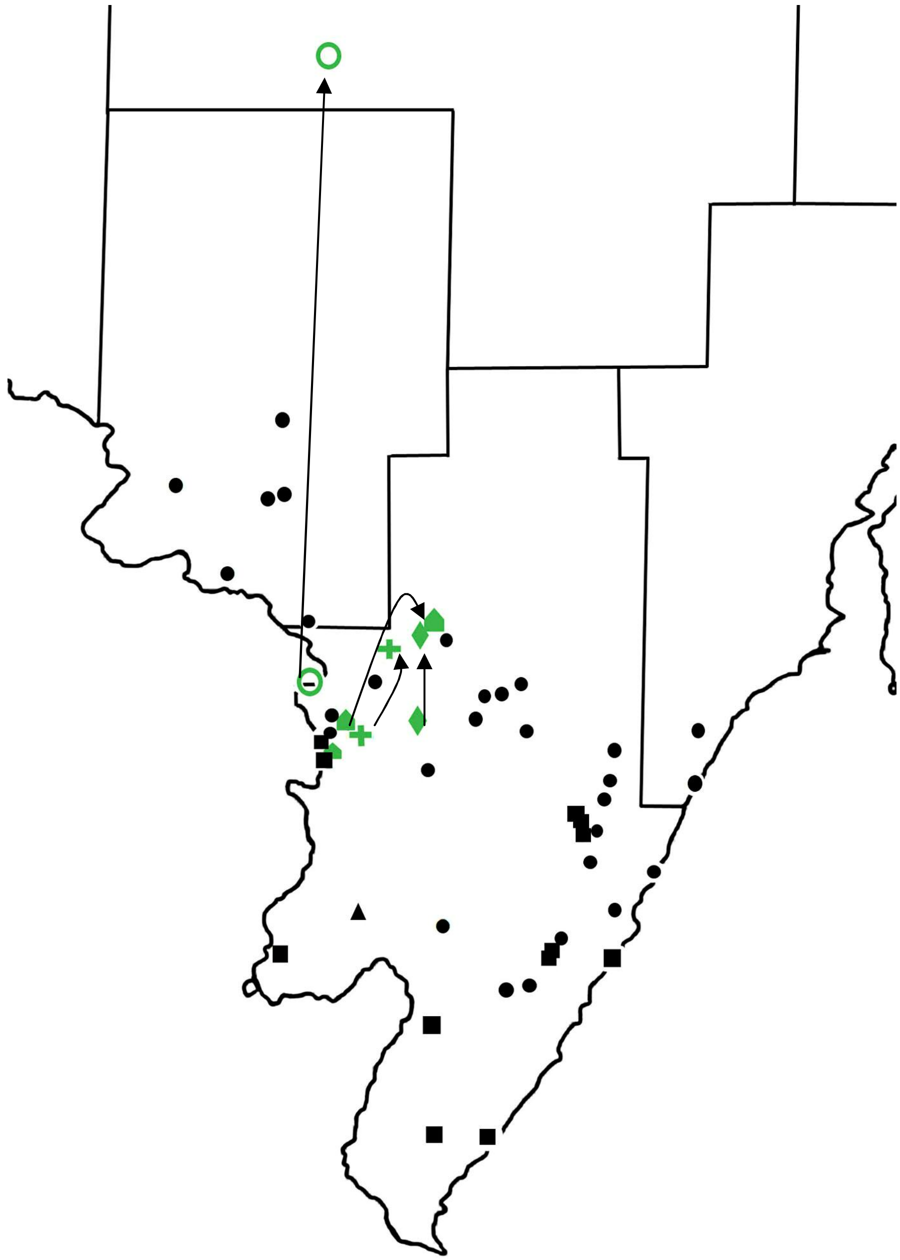

Figure 6: Movement of 4 deer from sites with established deer tick populations to negative sites within the Upper Peninsula of Michigan.
■ represents deer tick established sites, ● represents sites with un-established deer tick populations, ▲ represents the reported tick population. **o**: Movement north between May and November 1999, +: Movement between June and November 1999, ⬟: Movement between August and November 2000, ♦: Movement between November 1999 and November 2000. ↑ connect the two sites where the white-tailed deer was seen (Michigan Department of Natural Resources 2005a).

Appendix C

Institute of Animal Care and Use Committee approval form

Northern
Michigan
University

Continuing Education & Sponsored Progran
1401 Presque Isle Aven
Marquette, MI 49855-53

June 6, 2004

TO: Jackie Bird/Kimberly Miedema
Biology Department

FROM: Leonard Heldreth, Associate Provost
Academic Affairs

RE: **Application to use Vertebrate Animals**
Application # IACUC 011
Approval Period: May 20, 2004 – October 30, 2004

The Institutional Animal Care and Use Committee has approved your application to use vertebrate animals in research entitled "The Range Expansion of the Deer Tick North of Menominee toward Marquette."

If you have any questions, please contact me.

ljh

cc: Biology Department