Ecological and Evolutionary Trends of Lyme disease in the Northeastern United States

Introduction

Since the first reported outbreak in 1975 in Old Lyme Connecticut, Lyme disease has gained increasing prevalence as a major emerging infectious disease comparable to HIV and malaria and is currently the most common vector-borne illness in the United States (Steere et al., 2004; Centers for Disease Control and Prevention, 2013). Lyme disease is a blood-borne pathogen caused by the bacterial spirochete *Borrelia burgdorferi* which, when transferred from the blacklegged tick (*Ixodes scapularis*) to humans can cause skin lesions, chronic oligoarthritis, cardiac abnormalities, and in some cases fatal neurological disorders such as meningitis (Steere et al., 1983). Different strains of Lyme disease have been found in Europe and Asia, making the activity of this pathogen a global concern (Schmid, 1985). With over 20,000 nationally reported cases of Lyme disease in the US in 2012 and the increasing spread of this disease, it is more important than ever to investigate the cause of Lyme disease to begin efforts in reducing its incidence locally and internationally (Centers for Disease Control and Prevention, 2013).

The cause of the high incidence in Lyme disease in the Northeastern US within the past thirty-seven years can be explored by analyzing both the pathogen's ecological and evolutionary trends within its environment. However, to provide the fullest understanding of the increasing prevalence in Lyme disease, it is important to recognize that the pathogenic bacteria is dependent on ecological as well as evolutionary strategies for survival and that there often exists an overlap between these two strategies to meet the common goal of survival. With this in mind, the content of this review will be presented with the goal of containing and reducing future outbreaks of Lyme disease by understanding *Borrelia burgdorferi's* ecological role with other organisms in its environment as well as the bacteria's ability to evolve and adapt to its surroundings.

Life Cycle

Before analyzing the ecological and evolutionary trends of the spirochete *Borrelia burgdorferi*, it is necessary to first describe its life cycle to gain a better understanding of how the basic needs of this organism have impacted its ecological and adaptive behavior in its environment. The life cycle of the spirochete *Borrelia burgdorferi* parallels the life cycle of the blacklegged tick (*Ixodes scapularis*), since it is this tick that displaces the spirochete from its preferred natural reservoir in the white-footed mouse (*Peromyscus leucopus*). Once inside the white-footed mouse, *Borrelia burgdorferi* evades its host's immune system while absorbing basic nutrients and simple carbon compounds (Singh and Girschik, 2004). According to Ostfeld's (1997) research in tick ecology and the transmittance of Lyme disease, the blacklegged tick takes three blood meals over the course of its lifetime, progressing through stages from larvae to nymph to adult. The first blood meal occurs at the larval stage shortly after hatching, when the tick is born free of bacterial infection. The larval tick acquires a blood meal from a small mammal and detaches from its host molting into its nymph phase. The following summer, the nymphal tick takes its second blood meal from another mammal. After molting into its adult phase, the blacklegged tick takes its third and final blood meal from the white-tailed deer (*Odocoileus virginianus*), where it also fertilizes female eggs (Steere et al., 2004).

During this life cycle, the spirochete agent for Lyme disease can be transmitted to the tick at any one of its life stages through feeding on an infected organism, which can then be transmitted to humans. However, given its life cycle and the generalist preference of the blacklegged tick for a blood meal among an array of species including the white-footed mouse, white-tailed deer, chipmunk (*Tamias striatus*), robin (*Turdus migratorius*), common shrew, (*Sorex araneus*) and short-tailed shrew (*Blarina brevicauda*), it can be concluded that human

interaction with ticks resulting in Lyme disease is unintentional as the blacklegged tick does not need a blood meal from a human host to survive (Steere et al., 2004; Hanincova et al., 2006). An increase in the prevalence of Lyme disease in humans may therefore indicate a close association with the blacklegged tick, specifically from human encroachment on tick habitat. Human and community interactions with the blacklegged tick and *Borrelia burgdorferi* will be discussed shortly, but it is first important to note how the spirochete has evolved through its unique survival strategies.

Molecular Strategies and Signs of Evolution

Competent reservoirs suitable for *Borrelia burgdorferi* to survive have been found in many of the same organisms that blacklegged ticks parasitize, including white-footed mice, white-tailed deer, chipmunks, and robins (Ostfeld and Keesing, 2000). This suggests the spirochete is also a host generalist like its vector the blacklegged tick and may have coevolved with its tick vector to increase survival (Hanincova et al., 2006). Comparison of the genomes of the Northeastern *Borrelia burgdorferi* strain and the Midwestern US strain by DNA sequencing revealed these two bacteria diverged from a common ancestor, possibly displaying the degree of human interaction and influence over pathogenic evolution (Brisson, et al., 2010). In other words, the difference in spirochete genomes in the Northeastern and Midwestern US may have something to do with human interaction with the blacklegged tick and the decline in this organism's forest habitat with the development of houses. Analysis of the spirochete genome and its molecular structure may further help explain spirochetal interactions with macroscopic organisms such as the blacklegged tick and the bacteria's competent reservoir organisms.

Borrelia burgdorferi contains a linear genome 950kb long with an astonishing 21 recombinant DNA plasmids. This is the largest number of plasmids found in any single bacterium (Fraser et al., 1997). Moreover, the genetic redundancy of its genome and plasmid DNA coding for similar proteins suggests genotypic plasticity in adapting to the internal conditions of multiple organisms (Fraser et al., 1997). Further research on the spirochete's genome and molecular structure has revealed multiple outer surface proteins that vary in their roles of bacterial transmittance (Liang et al., 2002). Outer Surface Protein A (OSP-A), for example is known to anchor the spirochete into the cell membrane of its tick vector, likely protecting the spirochete from moving down the tick's digestive tract and becoming consumed (Singh and Girschik, 2004). It has also been found that bacterial reservoirs such as white-footed mice exhibit an immune response to infections with *Borrelia burgdorferi*. (Schwan et al., 1989). However, despite this response, this pathogen still manages to evade detection by the host's immune system with exception to the Western Fence Lizard (*Sceloporus occidentalis*), which has evolved to secrete a toxin that kills the spirochete (Lane and Quistad, 1998). The subject of exactly how the spirochete evades the host's immune system remains controversial. Early researchers such as Posey and Gherardini (2000) believe the spirochete manages evasion of the immune system by circumventing the need for iron, an essential nutrient vital for most bacteria to survive inside a host, while others such as Liang et al. (2002) believe the variability of outer surface proteins is responsible for evasion of the host immune system. All of these findings related to the genomic and molecular structure of *Borrelia burgdorferi* help explain the dynamic association between this spirochete and its host species.

Community Interactions

Since the spirochete lives primarily in white-footed mice and blacklegged ticks, the incidence of Lyme disease depends heavily on the survival and the community interactions between these two organisms. Studies show that the relative increase in prevalence of Lyme disease within the past four decades can be greatly attributed to human interaction and disturbance of the woodland and forest ecosystems of the Northeastern US, specifically through significant deforestation and suburbanization in the late 1970s (Brownstein et al., 2005). These two simultaneous events led to a subtle change in forest landscape that resulted in a dramatic spike in the incidence of Lyme disease. The two main factors resulting from this change in forest habitat were an increase in the white-footed mouse population and an increase in tick exposure to humans.

The increase in population density of white-footed mice from the change in forest habitat has been investigated in a number of ways. LoGiudice et al. (2000) determined the effect of mammal biodiversity on spirochete population by trapping and counting the number of spirochetes present in engorged ticks and by then comparing field results to a mathematical model predicting theoretical population densities for white-footed mice. Results showed that an increase in mammal biodiversity (such as an increase in chipmunks, shrews, and squirrels) led to a decrease in the white-footed mouse population, likely through competition for limited resources; this resulted in a corresponding decrease in Lyme disease incidence, displaying the theory of the "dilution effect." This dilution effect states that increasing biodiversity helps to "dilute" pathogenic incidence by increasing the population of other non-competent reservoir organisms such as the gray and red squirrels or the chipmunk (LoGiudice et al., 2003). These non-competent organisms decrease the risk of Lyme disease by killing off the harmful pathogen that could have been stored in the competent white-footed mouse or white-tailed deer, later

transmitting the disease to humans by the blacklegged tick. In this same study, LoGiudice et al. (2003) also observed that in small populations of white-footed mice, incidence of the spirochete increased by 61% in the typically non-competent common and short-tailed shrews, indicating the presence of a "rescue species." Although the exact mechanism of this reversal of competency is unknown, this survival strategy can most logically be attributed to the spirochete's genetic diversity and its resultant environmental plasticity (Kurtenbach et al., 2006).

The increase in the white-footed mouse population and the incidence of Lyme disease as a result of deforestation has also been found to increase heterogeneity in the forest landscape. By monitoring the population density, sex ratio, age, adult masses, and reproductive activity of white-footed mice in fifteen separate woodlots ranging in various size and three areas of continuous forest, Nubb and Swihart (1998) determined that the white-footed mouse population increased with increasing patch-wood area, or area of discontinuous forest. Nubb and Swihart (1998) concluded that this increase in population density was most likely due to a lack of both competition and predation. In 2005, Brownstein et al. built off of Nubb and Swihart's research and concluded that a change in forest landscape increased the heterogeneity of the forest, allowing for greater diversity in landmark features such as fields, rocks, and with suburbanization, houses. Brownstein et al. (2005) also determined that landscape heterogeneity was inversely related to biodiversity, supporting the dilution effect. Since the white-footed mouse and white-tailed deer are both regarded as generalist species that can adapt to a changing environment by transitioning from a forest to a patch-wood and suburban environment, these two organisms proliferated and were unmet by both competition and predation just as Nubb and Swihart had predicted. Brownstein et al. (2005) further deduced that a suburban environment made for an ideal "edge habitat" for white-tailed deer, as the deer could graze on lawn and

ornamental vegetation and quickly escape from danger by bounding back into the edge of the forest. Given this change in landscape heterogeneity, the decline in predation, and the proliferation of the white-footed mouse and white-tailed deer populations, it is no wonder why a Lyme disease outbreak affected 20,000 people in upstate New York during the summer of 1998 (Orloski et al., 1998).

Abiotic Factors

Just as molecular strategies, evolution, and community interactions influence spirochete and Lyme disease prevalence, abiotic factors such as temperature, pH, and water also have an impact on pathogenic incidence. On the microscopic level of *Borrelia burgdorferi*, temperature and pH play an important role in bacterial regulation. As a frequently transmitted pathogen, *Borrelia burgdorferi* must constantly adapt to the internal conditions of its host. When an infected tick receives a blood meal its temperature rises and pH decreases, causing the spirochete pathogen to change its expression of its outer surface proteins, specifically by down-regulating OSP-A and up-regulating OSP-C (Singh and Girschik, 2004). Although the exact purpose of this adaptation has yet to be researched, it is known that the spirochete undergoes similar OSP changes upon transmission to different organisms, especially from arthropods to mammals (Singh and Girschik, 2004). As a parasite, *Borrelia burgdorferi* is dependent on the health of the white-footed mouse, blacklegged tick, and the white-tailed deer populations, indicating that temperature and pH conditions must be optimal for life of these three species in order to maintain life for the spirochete bacteria as well.

Although the relative amount of water as a nutrient source to sustain life for the spirochete bacteria is unknown, the role of water for the spirochete's macroscopic host species is

better understood. In 2001, Ostfeld et al. studied the effect of acorn abundance on blacklegged tick and white-footed mouse populations after a six year period of increased seasonal rainfall in Duchess County, New York from 1996-2000. Results showed that an increase in acorn abundance from increased seasonal precipitation attracted both the white-footed mouse and white-tailed deer, increasing the incidence of Lyme disease. Ostfeld et al. (2001) reported that an outbreak of Lyme disease could be expected in high population densities of the white-footed mouse and white-tailed deer two summers after rainfall given the tick life cycle. This study supports the development of a "Lyme disease forecast" that can be very valuable in notifying and preparing a local community of an outbreak to reduce the magnitude of human infection.

Climate Change

Assessing the role of future climate change on the spread of Lyme disease may lead to important insight on containment and prevention. According to a report published by the Centers for Disease Control and Prevention in 2012, 95 percent of Lyme disease was found in Connecticut, Delaware, Maryland, Massachusetts, Michigan, Minnesota, New Hampshire, New Jersey, New York, Pennsylvania, Vermont, Virginia, and Wisconsin. In 2005, Brownstein et al. developed a logistic model with 89% accuracy to make predictions about the population density of blacklegged ticks with an increase in future global temperature. Using this model, average global temperature is projected to increase between 3.85-4.91°C by the 2080s, leading to expansion of tick habitat, specifically through the southern and Midwestern states of North Carolina, Georgia, Minnesota, Iowa, and northern Michigan as well as expansion into southern Canada (Brownstein et al., 2005; Ogden et al., 2009). There is also an anticipated retraction of blacklegged ticks from Texas, Mississippi, and Florida as increased temperature becomes intolerable. Despite these projections, the study by Brownstein et al. (2005) does not take into

account the migrational activity of the white-footed mouse or the white-tailed deer let alone changes in forest landscape habitat in response to an increase in global temperature, limiting the scope of this study. Tick species, however, may be adapting to temperature changes by developing resistance to increased humidity and moisture with infection of *Borrelia burgdorferi*, allowing for increased survival and proliferation of tick vectors for Lyme disease (Herrmann and Gern 2010). Although it is not guaranteed that blacklegged tick populations will expand to the states described by Brownstein et al. (2005), precautionary measures can be taken to reduce or prevent the outbreak of Lyme disease in these regions by increasing public awareness, opening vaccination clinics, and by incorporating non-competent reservoir organisms such as the Western Fence Lizard to outcompete and dilute disease incidence.

Conclusion

Lyme disease is a recent and serious emerging infectious disease concentrated in the Northeastern US that can cause a range of mild to debilitating arthritis along with cardiac and neurological dysfunction. Given the recent peak of 20,000 reports of Lyme disease cases in 1998 and trends towards a warmer environment spurring tick habitat expansion, it is imperative that efforts are made for prevention, containment, and reduction of this number one vector-borne illness in the country. Both ecological and evolutionary research should continue to be conducted, while initiating carefully planned field experiments to identify strategies to help reduce disease incidence. Lastly, in the event of experiencing heavier rainfall as a result of stronger storms such as Superstorm Sandy brought on by global warming, a Lyme disease weather forecast system should be established to warn residents of the risk of a high infection year and adequate preparations should be made, including authorizing vaccination clinics in

regions of high disease risk and potentially incorporating non-competent host species to dilute disease incidence.

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