Ticked off: An analysis of the inadequate diagnosis and treatment of Lyme disease

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Abstract

Lyme disease is the most common vector-borne disease in the United States and has a high prevalence among people in the northeast. Lyme disease can be a debilitating illness if not diagnosed early, and can lead to long-term health problems for many patients. This thesis serves as a review of scientific literature on Lyme disease, with the prevalence, symptomology, the bacterial mechanism of infection, the diagnostic process, transmission, and treatment therapies. Further research and development could lead to better primary care for those suffering with Lyme disease.

Ticked Off: An analysis of the inadequate diagnosis and treatment of Lyme disease

Lyme disease was first recognized in the 1970s as a debilitating disease, when several patients in Lyme, Connecticut suffered with swollen joints, paralysis, and severe chronic fatigue. The underlying cause of these symptoms was unknown and many patients were misdiagnosed as a result. Scientist Willy Burgdorfer began research on the infection and discovered the relationship between the common deer tick and the causative agent of Lyme disease. The bacterium causing the infection was given the name *Borrelia burgdorferi*, after the scientist who discovered it (Hyde, 2017; Tilly, Rosa, & Stewart, 2008).

Known as the Great Imitator, Lyme disease can mimic essentially every kind of disease; it can affect every tissue and organ system of the human body, making it incredibly difficult to recognize (Holtorf, n.d.; USA, 2011). The current diagnostics for Lyme disease, as recommended by the Centers for Disease Control and Prevention (CDC), have poor sensitivity and can show negative results more than 50% of the time in positively infected individuals ("Lyme Disease Diagnosis," n.d.). The poor diagnostic testing leads to a lack of antibiotic treatment and thus a progressive build-up of the parasitic infection and a greater manifestation of the disease as the organism multiplies. Once properly diagnosed, this prolonged infection leads to more extensive and possibly unsuccessful treatment of the disease. On average, patients will see five doctors over the course of two years before a diagnosis is made, making it more difficult for full recovery. Thus, 40% of Lyme patients suffer with long-term health problems (ILADS, n.d.). Because of possible late diagnoses, failed conventional treatment, and immune suppression, patients often seek alternative health care to find relief from their symptoms

(Gardner, 2000). This increase of Lyme patients seeking out alternative healthcare merits further research and study into the safety and efficacy of alternative therapies. Chief of epidemiology and surveillance for the CDC Lyme disease program, Paul Mead, referred to Lyme disease as a tremendous public health problem, which augments the need for further study and research of Lyme disease in effort to increase awareness and obtain early diagnosis (CDC, 2013).

Incidence

Lyme disease has been reported from forty-nine states, and has been found in more than fifty countries (Shapiro & Gerber, 2000). It is the most common vector-borne illness and is the fastest growing infectious disease in the United States. While it is mostly concentrated in the upper northeast region of the country, it has been reported in nearly every state and in several countries, and is extending into various regions (CDC, 2016a; Donohoe, Pennington-Gray, & Omodior, 2015). It is estimated there are 30,000 new cases of Lyme disease reported each year in the United States, with the true number of cases being three to twelve fold higher than the number of reported cases, making Lyme disease a larger epidemic than Avian Flu, West Nile Virus, and HIV combined ("CDC provides estimate," 2013; USA, 2011).

Lyme disease is transmitted to a person through the saliva of a deer tick, *Ixodes scapularis* or *Ixodes pacificus*, and transmission of the bacteria from the tick to the host requires a minimum of 72 hours. In rare cases a nymph-stage tick can transmit the bacteria in twenty-four hours (Barbour, n.d.; Fallon, Vaccaro, Romano, Clemente, 2006; Moore, Nelson, Molins, Mead & Schriefer, 2016). While all people are susceptible to Lyme disease, it is slightly more prevalent in males than in females, with the majority of

males infected between the ages five and nine. Cases of Lyme are seen mostly in young children and the middle-aged, who are between forty-five and fifty-five years of age (CDC, 2016b).

The risk factors for Lyme disease are mostly environmental. Spending time outdoors, especially in heavily wooded areas, increases one's risk for Lyme disease. While all persons are equally susceptible and there are no genetic predispositions to the contraction of Lyme disease, there are genetic abnormalities that could worsen the severity in which Lyme disease is manifested in a particular person. These genetic abnormalities can specifically enhance the chronic arthritic symptoms (Bramwell, Teuscher, & Weis, 2014). *B. burgdorferi* relies on cholesterol to maintain a firm cell membrane, and research shows that genetic predispositions to elevated cholesterol levels will cause an individual to have a higher susceptibility to a stronger manifestation and increased severity of Lyme disease, as represented by the higher numbers of bacteria found within the body (Toledo, Monzón, Coleman, Garcia-Monco & Benach, 2015).

Symptoms

Lyme disease symptoms continually vary as the disease progresses. Symptoms originate as local manifestations until it systemically spreads throughout the body. The first stage begins as the early localized stage, then progresses to the early disseminated, and evolves into late disseminated stage, which is commonly known as chronic Lyme disease (Goldstein, Harden, & Schachter, 2004). The early localized stage usually consists of flu-like symptoms, and most commonly, a red bull's-eye rash at the site of the bite, although, only approximately 19% of patients with Lyme disease actually develop a bulls-eye rash (Smith, Oertle, & Prato, 2014). Perhaps the beginning stage is most

difficult to diagnose because of the largely inconsistent symptoms, which is a hallmark of the disease, and the wide variety in which the symptoms can occur (Lyme Research Alliance, n.d.). As the disease progresses into the early disseminated stage, symptoms can develop into facial paralysis, tingling and numbness in extremities, severe fatigue, changes in vision, joint pain, and cardiac abnormalities. The late disseminated stage exhibits many of the same symptoms, but at a much higher severity. The late stage can progress into arthritis, neurological conditions such as disorientation, dizziness, short-term memory loss, inability to concentrate, or most commonly a mental "fog" (Barbour, n.d.). The late disseminated stage can occur months or years after the initial infection and usually causes the most severe symptoms. Additional symptoms of this late stage can include hallucinations, tremors, hypersensitivity to light and sound, seizures, sudden weakness and blackouts, nerve pain, and temporary amnesia (Goldstein et al., 2004; "Late stage," 2017). Lyme disease can mimic a wide variety of other diseases, and patients are commonly misdiagnosed with a variation of Alzheimer's disease, arthritis, systemic Lupus, Amyotrophic Lateral Sclerosis (more commonly known as ALS), Parkinson's disease, Multiple Sclerosis, Fibromyalgia, and many others, before an accurate diagnosis of Lyme disease is made (Lyme Research Alliance, n.d.). In 2001, patients with Lyme disease were surveyed to determine their overall quality of life and indicated that living with Lyme can be severely debilitating (Klempner et al., 2001). The results of the study showed that the average quality of life for a patient with chronic Lyme disease was worse than that of a patient with Type 2 diabetes, or recovery from a recent heart attack, and was equivalent to patients with congestive heart failure or osteoarthritis (ILADS working group, 2004; Johnson & Stricker, 2010). The CDC

conducted a study which showed that chronic Lyme patients suffer a worse quality of life when compared to patients with multiple sclerosis and other chronic diseases, combined with over 40% of patients being unemployed because of Lyme disease and 24% of patients identified having received some form of disability accommodations (Johnson, Wilcox, Mankoff, & Stricker, 2014). To further complicate the symptomatic ramifications of a *B. burgdorferi* infection is the Jarisch-Herxheimer reaction, in which a patient will have an intense worsening of symptoms following antibiotic treatment. The reaction is described as a shock-like state in response to destruction of the bacteria in the body as a result of antibiotic therapy. While the exact cause is unknown, it is suggested the worsened symptoms are due to the damaged spirochete releasing its endotoxins into the body (Maloy, Black, & Segurola, 1998). Overall, the symptoms of a Lyme patient can be highly variable and can range in intensity, especially if a Jarisch-Herxheimer reaction occurs and increases the severity of the symptoms for a patient.

Mechanism of infection

The obligate parasite, *B. burgdorferi*, is a member of Phylum *Spirochaetes.* Bacterial parasites of this phylum are characterized by their spiral shape, which allows for high motility throughout the body (Tilly et al., 2008). Spirochetes, because of their spiral shape, have the ability to burrow into soft tissue of the human host (Frazer, 2013). This bacterium chooses to burrow into privileged sites, such as the eye, synovial fluids, and the brain, which requires a more rigorous treatment approach to eradicate (Sticker & Middelveen, 2015). Additionally, *B. burgdorferi* has been identified in every organ of the body, which is not common amongst most bacterial pathogens. It also has a high affinity of attraction for the central nervous system (ILADS working group, 2004). The bacterium

is unique in that it has two flagella at both ends of the spirochete within their outer membrane, whereas most bacteria have flagella only at one end and extending out of the outer membrane cell wall (Smith et al., 2014). *B. burgdorferi* is one of the most aggressive pathogens to infect the human body and is unique from other spirochetes in that it has a flat-wave, sinusoidal, and planar appearance, in comparison to others which may have a helical, more cylindrical appearance. The undulating movement of the spirochetes as they travel throughout the body allow the bacterium to invade smaller spaces and denser environments, contributing largely to its pathogenic success (Harman, Vig, Radolf, & Wolgemuth, 2013).

The life cycle of *B. burgdorferi* is unique as it progresses through different hosts and maintains its infectivity within different reservoirs. Because of its survival in dual environments, the bacterium is incredibly adaptive to its environment. The ability of it to adapt so efficiently can be attributed to its progression through changes in its outer surface proteins (Osp), also known as antigenic variation. When the bacteria are active in a nymph tick, it resides in the midgut and expresses OspA to help maintain survival within the tick. When the nymph is feeding on a human, the proteins change and the bacteria down-regulates expression of OspA and upregulates expression of OspC. The expression of OspC on the bacteria is required for the infection of a healthy mammalian candidate. The OspC is recognized by the immune system and antibodies are formed as a result. In response, the bacteria then down-regulate OspC. The outer surface protein VIsE is then synthesized, which is consistent with the ability of the bacteria to maintain a long-term infection. VIsE remains on the outer surface, and while the exact function is still

unknown, the VIsE protein serves to continually undergo antigenic variation to evade immune system attempts of clearing the infection (Tilly, Bestor, & Rosa, 2013).

Perhaps a more concerning method of infection and evasion of the immune cells is the bacteria's ability to attack B lymphocytes. Research has shown that when the bacteria were surrounded by B-cells, the spirochete entered the B-cell and ruptured it. In addition to rupturing the lymphocytes, the spirochete then can surround itself with the outer layer of the B cell and conceal itself, thus preventing another immune attack (Grier, 2000).

The blood-brain barrier is a well-known protected environment, which has very limited permeability. The barrier serves as a safeguard for the brain and central nervous system from any drastic changes in internal conditions or from toxins that may be present in the body. Most bacteria, or large molecules are unable to pass through this barrier (R&D systems, n.d.). However, *B. burgdorferi* freely passes through it and then resides deep within the brain tissue. This progression of *B. burgdorferi* into the brain contributes mainly to the prominence of neurological symptoms associated with Lyme disease (Grab et al., 2005). Another study showed the specific ability of the spirochetes to hide in skin cells, the first organ it infects. Skin fibroblasts and keratinocytes were found to protect *B. burgdorferi* from the effects of antibiotics or immune system attempts. So the conclusion was made, in light of the evidence revealed, eukaryotic cell types serve as an ideal environment in which Lyme spirochetes can survive long-term (Georgilis, Peacocke, & Klempner, 1992).

Diagnosis

The diagnosis of Lyme disease is based on a two-tiered system in which an initial ELISA test is run to determine the presence of *B. burgdorferi* antibodies within the patient. If positive, a western blot is then conducted to confirm results by identifying the bacterial proteins recognized by those antibodies. However, this approach is somewhat unreliable, as the two-tiered system misses up to 90% of positive cases (ILADS working group, 2004). Because the testing for Lyme disease is so unreliable, most clinicians rely solely on the presentation of symptoms that coincide with the disease. However, this also presents a problem; while the telltale sign of Lyme disease is erythema migrans, more commonly called a Bulls Eye rash, only 80% of patients develop a general rash, and of that 80%, only 19% have the characteristic Bulls-eye rash that is consistent with a diagnosis. (Dayhoff-Brannigan, 2016; Smith et al., 2014). The ELISA test serves as a preliminary screening test in which the antibodies in the blood are measured. However, as discussed above, *B. burgdorferi* infects the body by specifically evading the immune system and preventing the body from forming antibodies, this can cause a false negative as detected by the ELISA test because the specific antibodies to *B. burgdorferi* will not be present in the blood. Because of the negative result, many patients do not receive further testing or treatment (Lyme Research Alliance, n.d.).

Additional diagnostic challenges present when a patient is bitten by *Ixodes scapularis*, particularly because this tick species transmits more than one type of infection. The acute diseases this tick species may contain is highly variable, which can contribute to a more difficult process of obtaining an accurate diagnosis. The contraction of a co-infection in addition to the manifestation of Lyme disease may contribute to

longer lasting and varied symptoms (Krause et al., 2002). The same *Ixodes* species that transmits Lyme can also be infected with the agent that causes human granulocytic anaplasmosis, babesiosis, or ehrlichiosis (Gary, 2006). In some regions, up to 40% of white-footed mice can be infected with 2 or more tick-borne pathogens. Similarly, in some regions, up to 66% of patients had antibodies against *B. burgdorferi* as well as to *Babesia microti* (Goldstein, Thompson, Spielman, Krause, 2001). A 2002 study concluded that concurrent infection by more than one tick-borne pathogen, is common (Krause et al., 2002).

Changes were recently made in 1995 to the interpretation of the Western blot test, and the antigen recognition requirements that were necessary for a positive diagnosis of Lyme disease. Under these changes, the CDC requires at least five out of the ten *B. burgdorferi* bands to be recognized by patient IgG antibody. These bands include 18 kDa, 21 kDa, 28 kDa, 30 kDa, 39 kDa, 41 kDa, 45 kDa, 58 kDa, 66 kDa, and 93 kDa. The changes also require evidence of patient IgM recognizing two out of three *B. burgdorferi* proteins to be present, 24 kDa, 39 kDa, and 41 kDa, for a positive diagnosis (CDC, 2001). While the western blot is generally deemed a more reliable test form than the ELISA test, because it indicates an immune response to a variety of *B. burgdorferi* antigens, it is still estimated to only be 80% sensitive because many patients do not produce antibody to *B. burgdorferi* antigens (Lyme Research Alliance, n.d.). Thomas Grier outlined an interesting case for the rejection of the modifications made to the interpretation of the Western blot test, in which the bands 22, 25, 31, and 34 are all missing from the list of positive indicators. These bands, however, are all clinically significant in the history of Lyme disease, as they correlate with the OSP antigens found

on *B. burgdorferi.* Additionally, these OSP antigens with the corresponding bands were targeted by the vaccine that was made and utilized in the 1990s (Angue, 2016; Grier, 2000). Interestingly enough, the bands which are clinically known to correlate with a positive indication of Lyme disease, are not listed on the bands necessary for a positive diagnosis. Grier also outlined a study that was conducted to test the changes made to the interpretation of the western blot test, and showed that under the previous criteria of tests, 66 patients with the history of a tick bite and symptoms consistent with Lyme disease, as well as a Bull's eye rash, were all considered to be positive. But with the new changes to the western blot bands required for a diagnosis, only 20 patients were now considered to be positive, leaving 46 patients that, according to the new criteria, were not considered to be positive for Lyme. Further, without a positive diagnosis for Lyme disease, despite the consistent symptoms, history of a tick bite, or the erythema migrans, the patients could no longer be treated, as insurance companies do not cover patients without a positive diagnosis (Grier, 2000).

The poor testing and false negatives consistent with Lyme disease can be attributed to the narrow range of antigens tested, despite the wide changing variety of antigens (Petrovic et al., 1998). An additional study was conducted to test the efficacy of the new updated criteria, and two performance panels were used, the criteria outlined by the CDC, and the criteria outlined by Boston Biomedica Inc. The sensitivity and specificity of the western blot were evaluated against each criteria guideline. The results showed the panel of western blot antigens outlined by Boston Biomedica Inc. were more sensitive in diagnosing Lyme disease than those used as outlined by the CDC; however, the specificity of recognizing those without the disease increased with the CDC criteria

(Tilton, Sand, & Manak, 1997). This research shows the low reliability of the current diagnostic practices and contributes to the high amount of underreported cases of Lyme disease.

B. burgdorferi, as outlined in the mechanism of infection section, can hide in the brain and the cerebrospinal fluid. It does so by altering its outer surface proteins and thus evading any immune system attempts to create antibodies against it. Because the body does not recognize the need to make antibodies, no antibodies are made and the patient will always respond with a seronegative result in any blood test. The lack of reliability of blood antibody tests is due to the nature of the bacteria itself and its impossibility to accurately detect. Thus, physicians that are educated about the complexity of Lyme disease rely largely on the clinical presentations and the wide range of symptoms that may present (Kaplan, 2004).

The incubation period may also contribute to the lower efficacy of testing. The amount of time required to show a set of symptoms after initial infection is usually 3-10 days ("Learn More," n.d.). Thus, patients may see a physician fairly quickly for their current symptoms. However, the production of IgM antibodies is not detectable until 1-2 weeks after the infection and IgG antibodies usually takes 4-6 weeks to form. Because of the need for both IgM and IgG antibodies to be present for a positive diagnosis, antibodies could take up to a month and a half to fully form before a positive diagnosis can be made (Borchers, Keen, Huntley, & Gershwin, 2015). The presence of antibodies, then, may not be fully matured at the time the patient is tested.

The discrepancy in accurate diagnoses of Lyme disease contributes to the gross underreporting of cases. It has been estimated the true number of cases is between 3-and

12-fold higher than what is reported, which would equate to 90,000- 360,000 actual true cases ("CDC provides estimate," 2013). Given the evidence, it can be concluded there is a very large number of patients that may be living with Lyme disease, but tested negative and thus could falsely be diagnosed with other diseases based on their symptoms.

Communicability

The CDC designates the transmission of Lyme disease to be transmissible mainly through the saliva of a tick bite, however, other possible forms of transmission can occur. Based on the knowledge that only about 2% of tick bites actually result in the formation of Lyme disease, as outlined by authors Sticker and Middelveen (2015), it would take 15 million tick bites per year to meet the current estimated rate of Lyme disease as expressed by the CDC (Hu, 2014). This implausible number of tick bites must suggest, then, there is another method of transmission (Stricker & Middelveen).

B. burgdorferi have been found in blood that is stored for donation, and thus, patients with Lyme disease are advised and instructed to not donate blood (CDC, 2015). Additionally, with the knowledge that other spirochetal diseases such as Syphilis and Relapsing fever cause maternal-fetal transmission, it raises concern for the possible maternal-fetal transmission of Lyme disease (Alexander & Cox, 1996). While it is infrequent, there have been several reports of Lyme disease being transmitted from mother to fetus during pregnancy (Lakos & Solymosi, 2010; Schlesinger, Duray, Burke, Steere, & Stillman, 1985; Weber, Bratzke, Neubert, Wilske, & Duray, 1988). Based on studies in animal models of Lyme disease, only acute infection at the beginning of pregnancy resulted in fetal loss, in contrast to chronic infections which did not increase abortion rate (Silver et al., 1994). The same study also concluded, though, that direct

transmission of *B. burgdorferi* to the fetus was not necessary for death, as the bacterium was not detected in the fetuses. Perhaps the bacterium targeted the fetus and caused death, then traveled back to the mother; complete transmission was not found to be necessary in cases of fetal death. While research on animal models gives much insight to the pathological effects of *B. burgdorferi*, results cannot be substituted for a human model.

In one reported case, a 28-year old female became pregnant, and early in her pregnancy she contracted Lyme disease. The baby was delivered at 35 weeks with respiratory difficulty, and the child died within 39 hours. Upon autopsy, the results showed a wide range of cardiovascular abnormalities. Additionally, spirochetes were found in the spleen, renal tubules, and bone marrow (Schlesinger et al., 1985). While complete transmission may not be required for fetal death, as concluded in the first study, complete transmission is certainly still a cause for fetal death as recorded in the second study. In another case, a pregnant woman in her first trimester received penicillin treatment for 7 days after showing the typical Bulls-eye rash. Less than 24 hours after birth of the child, the child died, and spirochetes were found in samples of the brain and the liver (Weber et al., 1988). While fetal death is not very common, it is perhaps more prevalent with infections during early pregnancy rather than third trimester pregnancies or cases with chronic Lyme.

In addition to maternal-fetal transmission of *B. burgdorferi*, little research has been conducted on the transmission of the bacteria through sexual transmission or the transmission from mother to baby through breast milk (Stricker & Middelveen, 2015). However, one study shows spirochetes were found in urine and in breast milk from two

lactating women. This suggests Lyme disease is present in bodily secretions and has the potential to be transmitted through breast milk, however, the viability of the bacteria was not recorded (Schmidt et al., 1995). Additionally, a study was conducted to test the hypothesis that *B. burgdorferi* could be transmitted through intimate contact. Vaginal and seminal secretions were obtained from all subjects and 11 out of the 13 secretions obtained from patients with a positive Lyme disease test had viable, motile spirochetes present. Further, the tests conducted from 3 couples participating in unprotected sex showed identical strains of bacteria in their genital secretions, suggesting the direct transmission through intimate contact (Middelveen et al., 2014). This study, however, received much criticism from its reviewers, based on the questioning reliability of the study. There were concerns from the reviewers about the sterility of the samples as well as the staining process and the need for cross-checking of blind samples to ensure the bacteria observed was *B. burgdorferi*. Nevertheless, the study should still be considered, and while not conclusive, it can strongly suggest the possibility of sexual transmission of Lyme disease.

One study was conducted to test the possible contact transmission of *B. burgdorferi* amongst the *Peromyscus leucopus* species, also known as the white-footed mouse, without the need of a tick vector. The study used a group of white-footed mice as a vector to inoculate with bacteria and house in an environment with uninfected mice. The results concluded contact transmission was possible, as antibodies were formed in the uninfected mice, and there were no indications it had been transferred through blood. This study could offer a probable explanation for the formation of Lyme disease among some patients with no tick exposure (Burgess, Amundson, Davis, Kaslow, & Edelman,

1986). In another study, however, with the same species of mice, spirochetes were found in the urine and feces of mice and the uninfected mice that came in contact with the infected mice contracted the spirochetes and became infected. The spirochetes were isolated mainly within the bladder of the mouse species. The study also concluded possible oral contact transmission through grooming, but also negated the possibility of maternal-fetal transmission (Wright & Nielsen, 1990). Overall, many of these studies show strong suggestions and implications, however it can be concluded that the results from small mammal models cannot definitively be applied to the human body.

Treatment

While the treatment of Lyme disease is highly variable, there are several treatment methodologies and scientific reasoning for their application. No attempt will be made to contribute to the ongoing Lyme Wars so prevalent in media today (Hoppel, 2008; Johnson, 2014; Stricker & Lautin, 2003). Lyme disease is said to present a public health threat of major proportions and the clinical complexity in patients with persistent symptoms has aroused more attention in the press than many other diseases (Kullberg, Berende, & van der Meer, 2011). The controversy lies between two organizations, Infectious Diseases Society of America (IDSA) and International Lyme and Associated Diseases Society (ILADS). These two organizations differ on the treatment approach, and the fundamental difference resides in whether or not chronic Lyme disease exists (Hoppel, 2008). The Institute of Medicine in Washington D.C. regards the Lyme disease guidelines controversy between IDSA and ILADS as perhaps the most contentious debate among medical conditions with differing guidelines. The IDSA concluded that the evidence for persistent, or chronic Lyme disease, is too weak, and thus treatment for it

cannot be justified. The ILADS, however, acknowledges the low quality of life of those with long-term symptoms, and deems the clinical presentation as enough evidence to support long-term treatment, with the goal of treating a persistent infection that may be present (Johnson, 2014; Johnson & Stricker, 2010). The treatment protocol as advised by ILADS, is to provide long-term antibiotic therapy for patients that still feel sick after the routine 30 days of treatment as outlined by the standards of IDSA. IDSA acknowledges their treatment approach is more generalized based on scientific evidence and it does not address an individualized approach (Hoppel, 2008). While the debate rages on, the patients are left with conflicting recommendations, no solid treatment protocol, and progressively sickened bodies. While the majority of patients with persistent symptoms seek medical aid, no consensus on adequate treatment, or approach to management of the disease can be made (Kullberg et al., 2011). This section is to serve as a review to provide a thought-provoking overview on the treatment efficacy and perhaps evaluate the gap in medical knowledge in treatment of this disease.

In order to discuss possible treatments, it is advantageous to first discuss the limitations in the treatment of Lyme disease. Perhaps the most challenging obstacle in treatment is approaching each patient differently, as not all strains of *Borrelia*, or presentations of Lyme disease, respond the same to therapy. This requires a very specialized and individualized approach for patients, and the most compelling difference in treatment resides in the duration of therapy. The standard treatment therapy for Lyme disease is a course of antibiotics, which prevents the manifestation of other diseases that may arise in a later stage (Borchers et al., 2015). The physiology of *B. burgdorferi*, however, allows it to change its outer surface proteins and cell wall shape. The bacterium

is able to reduce its cell wall and revert to a simpler, spherical form. In reducing its cell wall, antibiotics that target the cell wall are at a disadvantage when targeting the bacterium. Similarly, as discussed in the above section, *B. burgdorferi* tend to occupy privileged sites, where the immune system cannot access. Thus, if the bacteria remain in those spaces and lies dormant, antibiotics will be unable to target it, regardless of the length of treatment (Grier, 2000).

Therapy for early Lyme disease with no complications is fairly simple and has a high cure rate of 84-95% of cases. The difficulty, then, is presented in the patients with non-specific symptoms that cannot receive a reliable diagnosis early enough. Treatment success for patients with a delayed diagnosis is unknown and is underreported, and adequate treatment for patients with long-term symptoms needs to be further studied and researched (Kullberg et al., 2011).

Standard treatment guidelines for early Lyme disease in adults, as outlined by IDSA, consists of a 14-day regimen of doxycycline, amoxicillin, or cefuroxime axetil. In patients with neurological Lyme, presenting with symptoms consistent with an infection of the central nervous system, or patients with Lyme carditis that may have been hospitalized or have symptoms consistent with a heart condition, a 14-day regimen of intravenous ceftriaxone is recommended. Patients presenting with long-term symptoms, or an attributed chronic Lyme disease ($\geq$ 6 months) are recommended to not take antibiotics, yet no other guidelines are given for patients presenting with persistent symptoms (Wormser et al., 2006). The ILADS, however, do not have a standard treatment protocol, but rather take into account a variety of factors that would affect the method of treatment. ILADS evaluates each patient based on age, allergy, prior exposure,

clinical presentations, ability to tolerate side effects, and possible coinfections to determine the treatment protocol for that patient. Persistent Lyme is more likely to respond to intravenous therapy, but oral antibiotic therapy may be administered first to determine the patient's response. If antibiotics are the chosen course of treatment, many of the same antibiotics listed under IDSA's guidelines are also recommended by ILADS, which include amoxicillin, azithromycin, cefuroxime, clarithromycin, doxycycline and tetracycline (ILADS working group, 2004).

The controversy in treatment, then, resides with the duration of treatment. The duration of treatment is based solely on the persistent symptoms that may present in some patients. Persistent symptoms following a routine course of 2-4 weeks of antibiotics has been shown in 25-80% of patients, and a persistent infection that was diagnosed on the basis of culture or PCR was found in up to 40% of patients (Stricker, 2007). Symptoms may persist for several reasons, which include a low-grade persistent infection, sequestration of the bacterium in privileged areas where antibiotics cannot access, remaining damage from previous active infection, autoimmune response, or an underlying co-infection that presents with clinical manifestations similar to Lyme disease (Fallon et al., 2006).

Several studies have shown both the efficacy, and the failure of long-term antibiotic use in patients with persistent symptoms. In one study of 227 patients with chronic Lyme disease, all subjects were treated with tetracycline for eleven months and the outcomes were favorable. At the end of eleven months, 20% of the patients were cured, 70% of patients had improved conditions, and only 10% of patients had failed treatment. The symptoms also had improved from month 2 to month 3, which suggests

extended antibiotic therapy correlates with a better symptomatic outcome in patients (Donta, 1997). In another study conducted on patients with well-documented Lyme disease, those who had received previous treatment were administered IV ceftriaxone or IV placebo for 10 weeks. The result showed an improvement in cognitive function, with a relapse in cognition after discontinuation (Fallon et al., 2008). Another study conducted on 66 patients with long-term symptoms received 28 days of IV ceftriaxone, and despite the slight improvement of fatigue, there was no cognitive improvement among the patients, although four patients had adverse effects which resulted in hospitalization. This study concluded that additional antibiotic therapy with ceftriaxone, in treating those with long-term symptoms of Lyme disease, should not be advised or administered (Krupp et al., 2003). In another study, patients with a history of Lyme disease that tested negative for a persistent infection of *B. burgdorferi* were sampled, and no significant difference was concluded between the patients that received IV or oral antibiotics compared to those who received the placebo (Klempner et al., 2001). However, all the subjects used in the studies that had either no improvement or a negative outcome were seronegative for *Borrelia,* which may have contributed to the lack of effectiveness of antibiotics. On the contrary, perhaps the reason behind why some patients seem to improve on long-term antibiotics, aside from the placebo effect, is the anti-inflammatory effect of the drugs (Feder et al., 2007).

There is a discrepancy in results among all of these studies regarding the efficacy of long-term treatment. However, all patients respond differently, and the possible benefits must be assessed against the possible risks associated with taking long-term antibiotics (Dattwyler et al., 1997; Nadelman et al., 1992). Although, one research study

concluded that long-term antibiotics are worth the risk and are justified in the case of Lyme disease (Stricker, 2007). Additionally, it is also critical to understand the differences that occur in every patient with Lyme. The bacterium affects every human body differently, and thus every patient will respond to therapy differently (Borchers et al., 2015). Lastly, one study could not conclude any recommendations, and simply stated there was no answer for patients with long-term symptoms. But rather, the emphasis was placed on the need for different strategies and proven options to provide relief for Lyme patients (Melia & Auwaerter, 2016).

One study investigated the experiences of patients with long-term Lyme disease symptoms and reported the overwhelming majority of patients seek complementary and alternative medicine to assist in their healing and recovery process. The authors discuss that in light of Lyme disease being poorly understood and often resistant to conventional treatment, patients often seek alternative medicine to answer their medically unexplained condition of long-term symptoms. The complementary and alternative medicine often included over-the-counter products, mind-body practices, dietary modifications, acupuncture, and chiropractic care. None of the patients studied had experienced adverse reactions to the complementary and alternative medicine therapies and patients reported a greater satisfaction with the practitioners (Ali, Vitulano, Lee, Weiss, & Colson, 2014). These alternative treatment methods often provide patients more open-mindedness in treatment and cultivate an individualized approach, rather than the usual standard of care in a conventional treatment setting. Alternative medicine can be complementary to conventional medicine and more research should be conducted to further understand the efficacy and safety of these approaches.

The use of antibiotics for early treatment of Lyme disease is highly effective, but after long term symptoms evolve and patients have developed the late disseminated stage, antibiotic treatment has been shown to have some positive effects, but also possible negative effects and adverse reactions. The potential benefits and risks of antibiotics are different for every patient in the late disseminated stage, and there are alternative treatment methods that should be evaluated in the treatment of Lyme disease. Medical doctor, surgeon, and Chinese medicine practitioner, Shiroko Sokitch, discusses the importance of supporting the body throughout a treatment process rather than emphasize the use antibiotics to forcefully eradicate the infection. While eradicating the Lyme infection should be of optimal importance, the body and innate immune system should be well-supported using various methods such as, herbs, acupuncture, nutritional supplements, or diet, in order for the body to fully heal (personal communication, 2016). Alternative treatments that specifically target the eradication of disease, combined with supporting the body, can work together to facilitate the optimal immune function of the body.

In a mouse model study, Vitamin D was supplemented for mice that had developed chronic arthritis from a *B. burgdorferi* infection. The results showed vitamin D was successful in preventing arthritis from progressing into more severe forms, and also decreased the symptoms. The results also showed no increased risk of hypercalcemia from Vitamin D supplementation (Cantorna, Hayes, DeLuca, 1998). An *in vitro* study of *B. burgdorferi* showed grapefruit seed extract worked in inhibiting the bacterium's transitional forms, which is known for contributing largely to its pathogenic success. Grapefruit seed extract has been shown to be highly anti-microbial against bacteria and

fungi, and have higher therapeutic effects than other well-known anti-bacterial agents. The substance specifically prevented *B. burgdorferi* from transitioning into cystic forms which contribute to its ability to evade the immune system. Prevention of this transitional form of the bacteria can assist the body in fighting off the infection as it allows the bacteria to be more easily targeted. While this was an *in vitro* study, the study suggests the combination of grapefruit seed extract with antibiotics would be efficient in eradicating Lyme disease, with supporting evidence to show no severe side effects have been observed in the medicinal application of grapefruit seed extract (Brorson & Brorson, 2007). Another *in vitro* study tested the efficacy of whole leaf Stevia extract against *B. burgdorferi* spirochetes, biofilms, and persister cells. The effects of Stevia were compared to common antibiotics such as doxycycline, cefoperazone, and daptomycin, and their combinations. It was reported the antibiotics were not effective in eliminating the persister cells of *B. burgdorferi*, but Stevia was able to eliminate the spirochetes and the persister cells. The study reported that certain antibiotics promoted the biofilm formation of the bacterium, which is the cause of the antibiotic resistant nature of the bacterium. Stevia, however, worked to reduce the biofilm mass and attachment to the bacteria, thereby contributing to the opportunity to eradicate the infection. The study concluded the efficacy of Stevia against *B. burgdorferi* because of its strong antibacterial properties. The study merits further *in vitro* research in hopes of implementing this in clinical practice (Theophilus et al., 2015). Other studies report the safety of high dietary intake of Stevia, as well as a study on diabetic patients consuming Stevia with no reported adverse reactions (Hsieh et al., 2003; Thomas & Glade, 2010).

The maintenance of long-term Lyme disease, particularly those with inconsistent symptoms, largely has to do with nutritional and lifestyle factors. Many diet modifications of those with long-term Lyme disease can be made in effort to assist the healing process. Dr. Rath Research Institute in California tested the efficacy of naturopathic healthcare treatments, in response to the majority of Lyme patients seeking alternative options to conventional therapy (Dr. Rath Research Institute, 2015). Many practical applications were found as a result of the research conducted, with the general overview consisting of micronutrient supplementation, immune system support, decreasing inflammation, detoxification, and implementing a proper diet. Applying a proper diet can perhaps be the most immediate response to the maintenance of Lyme disease, and the recommendation as outlined by Dr. Rath Research Institute consist of decreasing inflammation by practicing a gluten and dairy free diet, which works to avoid further inflammation to a Lyme patient. Digestive functioning of a patient with Lyme disease is already inhibited, so a low allergen diet is important, especially for those patients that are on long-term antibiotics, which can cause yeast overgrowth. High volumes (1L for every 25kg) of clean, filtered water should be consumed to aid in the constant fluid flow of the body and the natural detoxification process. Lastly, since the pain experienced by Lyme patients often causes insomnia, adequate sleep is highly encouraged for healing, and melatonin or other herbs can be supplemented for proper rest. The overall goal of changing diet and lifestyle of patients with Lyme disease should be geared towards limiting artificial or processed foods, along with artificial ingredients that can be found in cleaning and skin-care products. Since Lyme disease downregulates

the body's immune system and causes such a wide range of debilitating symptoms, the importance of limiting any additional toxic load is highlighted by this research.

Treatment options for patients with Lyme disease are highly variable, and the research shows the need for an individualized approach to each patient. The efficacy of treatment in early stages are very positive, but if the diagnosis of the disease is delayed and Lyme disease sets into the late disseminated stage, treatment can require a much more aggressive approach and has a lower rate of efficacy. In the late disseminated stage, the focus should shift from treatment to the maintenance of its symptoms. Long-term Lyme disease should be managed until treatment options for these patients are fully researched and available.

Conclusion

Lyme disease presents several different challenges, for both the patient and the health care provider. Despite its growing incidence in the United States, Lyme disease is commonly misdiagnosed, which demands for increased awareness among the medical community, in effort to stress the importance of early diagnosis. The shrewd mechanism of infection used by *B. burgdorferi* classifies it as an excellent pathogen, but merits further study to increase the effectiveness of treatment methods. Additionally, more research into the transmission of Lyme disease is warranted. Perhaps the most important area of research should be on further treatment options for those experiencing long-term symptoms. Conventional and alternative healthcare practices can combine efforts to optimize the patient's well-being and facilitate healing. Overall, Lyme disease can be a debilitating disease with severe consequences if not treated early, and further research should be conducted in effort to provide early relief for those suffering.

References

Alexander, J. & Cox, S. (1996). Lyme disease and pregnancy. *Infectious Diseases in Obstetrics and Gynecology, 3,* 256-261. Retrieved from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2364450/pdf/IDOG-03-256.pdf

Ali, A., Vitulano, L., Lee, R., Weiss, T., & Colson, E. (2014). Experiences of patients identifying with chronic Lyme disease in the healthcare system: A qualitative study. *BMC Family Practice, 15,* 1-8. doi: 10.1186/1471-2296-15-79

Angue, L. (2016). Resurrecting a vaccine for Lyme disease. Retrieved from http://www.medicalnewstoday.com/articles/260471.php

Barbour, A. (n.d.) Lyme disease. Retrieved from http://www.aldf.com/lyme-disease/

Borchers, A., Keen, C., Huntley, A., & Gershwin, E. (2015). Lyme disease: A rigorous review of diagnostic criteria and treatment. *Journal of Autoimmunity, 57,* 82-115. doi: 10.1016/j.jaut.2014.09.004.

Bramwell, K., Teuscher, C., & Weis, J. (2014). Forward genetic approaches for elucidation of novel regulators of Lyme arthritis severity. *Cellular and Infection Microbiology, 4,* 45-52. doi: 10.3389/fcimb.2014.00076.

Brorson, Ø. & Brorson, S. (2007). Grapefruit seed extract is a powerful *in vitro* agent against motile and cystic forms of *Borrelia burgdorferi sensu lato*. *Infection, 35,* 206-208.

Burgess, E., Amundson, T., Davis, J., Kaslow, R., & Edelman, R. (1986). Experimental inoculation of *Peromyscus spp.* with *Borrelia burgdorferi*: Evidence of contact transmission. *American Journal of Tropical Medicine and Hygiene, 35,* 355-359.

Cantorna, M., Hayes, C., & DeLuca, H. (1998). 1, 25-Dihydroxycholecalciferol inhibits the progression of arthritis in murine models of human arthritis. *The Journal of Nutrition, 128,* 68-72.

CDC. (1995). Notice to readers: Recommendations for test performance and interpretation from the second national conference on serologic diagnosis of Lyme disease. Retrieved from https://www.cdc.gov/mmwr/preview/mmwrhtml/00038469.htm

CDC. (2013). CDC provides estimate of Americans diagnosed with Lyme each year. Retrieved from https://www.cdc.gov/media/releases/2013/p0819-lyme-disease.html

CDC. (2015). Transmission. Retrieved from https://www.cdc.gov/lyme/transmission/

CDC. (2016a). Data and statistics. Retrieved from https://www.cdc.gov/lyme/stats/index.html

CDC. (2016b). Lyme disease graphs. Retrieved from https://www.cdc.gov/lyme/stats/graphs.html

Dattwyler, R., Luft, B., Kunkel, M., Finkel, M., Wormser, G., Rush, T., Grunwaldt, E., Agger, W., Franklin, N., Oswald, D., Cockey, L., & Maladorno, D. (1997). Ceftriaxone compared with doxycycline for the treatment of acute disseminated Lyme disease. *The New England Journal of Medicine, 337,* 289-295. doi:10.1056/NEJM199707313370501

Dayhoff-Brannigan, M. (2016). Lyme disease: The first sign is not always a rash. Retrieved from http://center4research.org/uncategorized/diagnosing-and-treating-lyme-disease-the-first-sign-is-not-always-a-rash/

Donohoe, H. Pennington-Gray, L., & Omodior, O. (2015). Lyme Disease: Current issues, implications, and recommendations for tourism management. *Tourism management, 46,* 408-418.

Donta, S. (1997). Tetracycline therapy for chronic Lyme disease. *Clinical Infectious Diseases, 25,* S52-S56. doi: https://doi.org/10.1086/516171

Dr. Rath Research Institute. *Scientific guide in natural approach to Lyme disease information for health practitioners* [PDF document]. Retrieved from https://www.drrathresearch.org/images/attachments/Infectious%20Diseases/Lyme-guide_DRRI.pdf

Fallon, B., Keilp, J., Corbera, K., Petkova, E., Britton, C., Dwyer, E., Slavov, I., Cheng, J., Dobkin, J., Nelson, D., & Sackeim, A. (2008). A randomized, placebo-controlled trial of repeated IV antibiotic therapy for Lyme encephalopathy. *Neurology, 70,* 992-1003. doi: http://dx.doi.org/10.1212/01.WNL.0000284604.61160.2d

Fallon, B., Vaccaro, B., Romano, M., & Clemente, M. (2006). Lyme borreliosis: Neuropsychiatric aspects and neuropathology. *Psychiatric Annals, 36,* 120-122, 124-128.

Feder, H., Johnson, B., O'Connell, S., Shapiro, E., Steere, A., & Wormser, G. (2007). A critical appraisal of "chronic Lyme disease." *The New England Journal of Medicine, 357,* 1422-1430. doi: 10.1056/NEJMra072023

Frazer, J. (2013). On the curious motions of Syphilis and Lyme disease bacteria. Retrieved from https://blogs.scientificamerican.com/artful-amoeba/on-the-curious-motions-of-syphilis-and-lyme-disease-bacteria/

Gardner, C. (2000). Treating Lyme disease naturally & effectively. Retrieved from https://healthimpactnews.com/2013/success-treating-lyme-disease-naturally-without-antibiotic-drugs/

Gary, W., (2006). Early Lyme disease. *The New England Journal of Medicine, 354,* 2794-2801. Retrieved from http://search.proquest.com/docview/223929149?pq-origsite=summon&accountid=12085

Georgilis, K., Peacocke, M., & Klempner, M. (1992). Fibroblasts protect the Lyme disease spirochete, Borrelia burgdorferi, from ceftriaxone in vitro. *The Journal of Infectious Diseases, 166,* 440-444.

Goldstein, E., Thompson, C., Spielman, A., & Krause, P. (2001). Coinfecting deer-associated zoonoses: Lyme disease and ehrlichiosis. *Clinical Infectious Diseases, 33,* 676-685. doi: 10.1086/322681

Goldstein, M., Harden, C., & Schachter, S. (2004). Lyme disease. Retrieved from http://www.epilepsy.com/information/professionals/co-existing-disorders/infectious-states-seizures/lyme-disease

Grab, D., Perides, G., Dumler, S., Kim, K., Park, J., Kim, Y., Nikolskaia, O., Choi, K., Stins, M., & Kim, K. (2005). *Borrelia burgdorferi*, host-derived proteases, and the blood-brain barrier. *Infection and Immunity, 73,* 1014-1022. doi: 10.1128/IAI.73.2.1014-1022.2005

Grier, T. (2000). The complexities of Lyme disease (PDF document). Retrieved from http://www.borelioza.org/materialy_lyme/the_complexities_of_lyme_disease.pdf

Harman, M., Vig, D., Radolf, J., & Wolgemuth, C. (2013). Viscous dynamics of Lyme disease and syphilis spirochetes reveal flagellar torque and drag. *Biophysical Journal, 105,* 2273-2280. doi: 10.1016/j.bpj.2013.10.004

Holtorf, K. Lyme disease- the great imitator. Retrieved from https://www.holtorfmed.com/lyme-disease-the-great-imitator/

Hoppel, A. (2008). The Lyme wars: Debate rages about treatment. *Clinician reviews, 18,* 44-46.

Hsieh, M., Chan, P., Sue, Y., Liu, J., Liang, T., Huang, T., Tomlinson, B., Chow, M., Kao, P., & Chen, Y. (2003). Efficacy and tolerability of oral Stevioside in patients with mild essential hypertension: A two-year, randomized, placebo-controlled study. *Clinical Therapeutics, 25,* 2797-2808.

Hu, L. (2014). Patient education: What to do after a tick bite to prevent Lyme disease (beyond the basics). Retrieved from https://www.uptodate.com/contents/what-to-do-after-a-tick-bite-to-prevent-lyme-disease-beyond-the-basics

Hyde, J. (2017). *Borrelia burgdorferi* keeps moving and carries on: A review of Borrelial dissemination and invasion. *Frontiers in Immunology,* 8. doi: 10.3389/fimmu.2017.00114

ILADS. (n.d.). Retrieved from http://www.ilads.org/lyme/lyme-quickfacts.php

ILADS working group. (2004). The International Lyme and Associated Diseases Society: Evidence based guidelines for the management of Lyme disease. *Expert Review of Anti-infective Therapy, 2.*

Johnson, L. (2014). The Lyme wars: Guidelines, controversy, and the informed consent. Retrieved from https://www.lymedisease.org/lyme-basics/resources/lyme-wars-guidelines-controversy-informed-consent/

Johnson, L. & Stricker, R. (2010). The Infectious Diseases Society of America Lyme Guidelines: A cautionary tale about the development of clinical practice guidelines. *Philosophy, Ethics, and Humanities in Medicine, 5.* doi: 10.1186/1747-5341-5-9

Johnson, L., Wilcox, S., Mankoff, J., & Stricker, R. (2014). Severity of chronic Lyme disease compared to other chronic conditions: A quality of life survey. *PeerJ, 2.* doi: 10.7717/peerj.322

Kaplan, M. (2004). Interpreting the IgG & IgM western blot for Lyme disease. Retrieved from http://www.anapsid.org/lyme/wb.html

Klempner, M., Hu, L., Evans, J., Schmid, C., Johnson, G., Trevino, R., Norton, D., Levy, L., Wall, D., McCall, J., Kosinski, M., & Weinstein, A. (2001). Two controlled trials of antibiotic treatment in patients with persistent symptoms and a history of Lyme disease. *New England Journal of Medicine, 345,* 85-92. doi: 10.1056/NEJM200107123450202.

Krause, P., McKay, K., Thompson, C., Sikand, V., Lentz, R., Lepore, T., Closter, L., Christianson, D., Telford, S., Persing, D., Radolf, J., Spielman, A., & the Deer-Associated Infection Study Group. (2002). Disease-Specific diagnosis of coinfecting tickborne zoonoses: Babesiosis, human granulocytic ehrlichiosis, and Lyme Disease. *Clinical Infectious Diseases, 34,* 1184-1191. doi: 10.1086/339813

Krupp, L., Hyman, L., Grimson, R., Coyle, P., Melville, P., Ahnn, S., Dattwyler, R., & Chandler, B. (2003). Study and treatment of post Lyme disease (STOP-LD): A randomized double masked clinical trial. *Neurology, 60,* 1923-1930.

Kullberg, B., Bernede, A., & van der Meer, J. (2011). The challenge of Lyme disease: Tired of the Lyme qars. *The Journal of Medicine, 69,* 98-100.

Lakos, A. & Solymosi, N. (2010). Maternal Lyme borreliosis and pregnancy outcome. *International Journal of Infectious Diseases, 14,* 494-498. doi: 10.1016/j.ijid.2009.07.019

Late stage Lyme disease. (2017). Retrieved from http://www.lymedisease.org.au/about-lyme-disease/late-stage-lyme-disease/

Learn more about Lyme disease. (n.d.). Retrieved from http://www.lymemd.org/learn-about-lyme.html

Lyme disease diagnosis. (n.d.). Retrieved from https://www.lymedisease.org/lyme-basics/lyme-disease/diagnosis/

Lyme Research Alliance. (n.d.). Diagnostic dilemma. Retrieved from http://www.lymeresearchalliance.org/test-diagnostic.html

Maloy, A., Black, R., & Segurola, R. (1998). Lyme disease complicated by the Jarisch-Herxheimer reaction. *The Journal of Emergency Mediince, 3,* 437-438. doi: http://dx.doi.org/10.1016/S0736-4679(98)00011-0

Melia, M. & Auwaerter, P. (2016). Time for a different approach to Lyme disease and long-term symptoms. *New England Journal of Medicine, 374,* 1277-1278. doi:10.1056/NEJMe1502350

Middelveen, M., Burke, J., Sapi, E., Bandoski, C., Filush, K., Wang, Y., Franco, A., Timmaraju, A., Schlinger, H., Mayne, P., & Sticker, R. (2014). Culture and identification of *Borrelia* spirochetes in human vaginal and seminal secretions [version 1; referees: 1 not approved]. *F1000Research, 3*. doi: 10.12688/f1000research.5778.1

Moore, A., Nelson, C., Molins, C., Mead, P., & Schriefer, M. (2016). Current guidelines, common clinical pitfalls, and future directions for laboratory diagnosis of Lyme disease, United States. *Emerging Infectious Diseases*, 22, 1169-1177. doi: 10.3201/eid2207.151694

Nadelman, R., Luger, S., Frank, E., Wisniewski, N., Collins, J., & Wormser, G. (1992). Comparison of cefuroxime axetil and doxycycline in the treatment of early Lyme disease. *Annals of Internal Medicine, 114,* 273-280. doi: 10.7326/0003-4819-117-4-273

Petrovic, M., Vogelaers, D., Van Renterghem, L., Carton, D., De Reuck, J., & Afschrift, M. (1998). Lyme borreliosis-A review of the late stages and treatment of four cases. *Acta clinical belgica, 53,* 178-183. Retrieved from https://www.ncbi.nlm.nih.gov/pubmed/9701852

R&D systems. (n.d.). Blood-brain barrier permeability. Retrieved from https://www.rndsystems.com/research-area/blood--brain-barrier-permeability

Schlesinger, P., Duray, P., Burke, B., Steere, A., & Stillman, T. (1985). Maternal-Fetal transmission of the Lyme disease spirochete, *Borrelia burgdorferi*. *Annals of Internal Medicine, 103,* 67-68.

Schmidt, B., Aberer, E., Stockenhuber, C., Klade, H., Breier, & Luger, A. (1995). Detection of *Borrelia burgdorferi* DNA by polymerase chain reaction in the urine and breast milk of patients with Lyme borreliosis. *Bacteriology, 21,* 121-128. doi: 10.1016/0732-8893(95)000027-8

Shapiro, E., & Gerber, M. (2000). Lyme Disease. *Clinical Infectious Diseases, 31,* 533-542. doi: https://doi.org/10.1086/313982

Silver, R., Yang, L., Daynes, R., Branch, D., Salafia, C., & Weis, J. (1994). Fetal outcome in murine Lyme disease. *Infection and Immunity, 63,* 66-72.

Smith, A., Oertle, J., & Prato, D. (2014). Chronic Lyme disease: Persistent clinical symptoms related to immune evasion, antibiotic resistance and various defense mechanisms of Borrelia burgdorferi. *Open Journal of Medical Microbiology, 4,* 252-260. doi: 10.4236/ojmm.2014.44029.

Stricker, R. (2007). Counterpoint: Long-Term antibiotic therapy improves persistent symptoms associated with Lyme disease. *Clinical Infectious Diseases, 45,* 149-157. doi: https://doi.org/10.1086/518853

Stricker, R., & Lautin, A. (2003). The Lyme wars: time to listen. *Expert Opinion on Investigational Drugs, 12,* 1609-1614. doi: 10.1517/13543784.12.10.1609

Stricker, R., & Middelveen, M. (2015). Sexual transmission of Lyme disease: challenging the tickborne disease paradigm. *Expert review of Anti-infective Therapy, 13,* 1303-1306. doi: 10.1586/14787210.2015.1081056

Theophilus, P., Victoria, M., Socarras, K., Filush, K., Gupta, K., Luecke, D., & Sapi, E. (2015). Effectiveness of *Stevia rebaudiana* whole leaf extract against the various morphological forms of *Borrelia burgdorferi in vitro. European Journal of microbiology and Immunology, 5*, 268-280. doi: 10.1556/1886.2015.00031

Thomas, J., & Glade, M. (2010). Stevia: it's not just about calories. *The Open Obesity Journal, 2*, 101-109.

Tilly, K., Bestor, A., & Rosa, P. (2013). Lipoprotein success in *Borrelia burgdorferi*: Similar but distinct roles for OspC and VIsE at different stages of mammalian infection. *Molecular Microbiology, 89*, 216-227. doi: 10.1111/mmi.12271

Tilly, K., Rosa, P., & Stewart, P. (2008). Biology of infection with *Borrelia burgdorferi*. *Infectious disease clinics of North America, 22*, 217-234. doi: 10.1016/j.idc.2007.12.013.

Tilton, R., Sand, M., & Manak, M. (1997). The Western immunoblot for Lyme disease: Determination of sensitivity, specificity, and interpretive criteria with use of commercially available performance panels. *Clinical Infectious Diseases, 25,* S31-S34.

Toledo, A., Monzón, J., Coleman, J., Garcia-Monco, J., & Benach, J. (2015). Hypercholesterolemia and ApoE deficiency result in severe infection with Lyme disease and relapsing-fever *Borrelia*. *Microbiology, 112*, 5491-5496. doi: 10.1073/pnas.1502561112

USA, A. (2011). Lyme disease: Misdiagnosed, underreported-and epidemic. Retrieved from http://www.anh-usa.org/lymedisease/

Weber, K., Bratzke, H., Neubert, U., Wilske, B., & Duray, P. (1988). Borrelia burgdorferi in a newborn despite oral penicillin for Lyme borreliosis during pregnancy. *The Pediatric Infectious Disease Journal, 7,* 286-289. doi: 10.1097/00006454-198804000-00010

Wormser, G., Dattwyler, R., Shapiro, E., Halperin, J., Steere, A., Klempner, M., Krause, P., Bakken, J., Strle, F., Stanek, G., Bockenstedt, L., Fish, D., Dumler, S., & Nadelman, R. (2006). The clinical assessment, treatment, and prevention of Lyme disease, human granulocytic anaplasmosis, and babesiosis: clinical practice guidelines by the infectious diseases society of America. Retrieved from https://www.idsociety.org/uploadedfiles/idsa/guidelines-patient_care/pdf_library/lyme%20disease.pdf

Wright, S., & Nielsen, S. (1990). Experimental infection of the white-footed mouse with *Borrelia burgdorferi. American Journal of Veterinary Research, 12,* 1980-1987.