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# CHRONIC LYME DISEASE IN BRITISH COLUMBIA
## A Review of Strategic and Policy Issues

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## Abbreviations

| | |
|---|---|
| AD | - Alzheimer's Disease |
| B.b. | - Borrelia Burgdorferi, the bacteria that causes Lyme Disease |
| BCCDC | - BC Centre for Disease Control |
| CARAT | - Council for Appropriate and Rational Antibiotic Therapy |
| CDC | - US Centers for Disease Control and Prevention |
| CNS | - Central nervous system |
| ELISA | - Enzyme-Linked Immunosorbant Serum Assay – a type of test used in detecting antibodies to the Lyme Disease bacterium, B.b.. |
| EM rash | - Erythema migrans rash – a bulls eye rash on the body commonly found after a tick bite. |
| HGE | - Human Granulocytic Ehrlichiosis, a coinfective agent sometimes found with B.b. from a tick bite. Also called |
| I. | - Ixodes, a major classification of a tick species, including I. Pacificus, the tick most prevalent in BC responsible for spreading the B.b. bacteria to humans. |
| IgG | - Immunoglobulin G |
| ILADS | - International Lyme and Associated Diseases Society |
| ISDA | - Infectious Diseases Society of America |
| LD | - Lyme Disease, the infection caused by bacteria B.b. |
| PCR | - Polymerase chain reaction – a type of diagnostic test that could be used to detect Lyme Disease. |
| PHAC | - Public Health Agency of Canada |
| PLS | - Post Lyme-disease syndrome, a term used by the IDSA, but often called Chronic Lyme Disease by the members of ILADS. |
| TLR | - Toll like receptor |
| VDR | - Vitamin D Receptor |

## 1. Introduction to Lyme Disease (with a focus on Chronic Lyme Disease)

Lyme disease was recognized in 1981 as a condition associated with an infectious bacterium.
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*Borrelia burgdorferi* (*B.b.*) is transmitted primarily by ticks of the *Ixodes ricinus* complex. It is the agent that causes Lyme borreliosis, commonly known as Lyme disease. In North America, the ticks have larval, nymphal, and adult stages. The risk of infection in a given area depends largely on the density of these ticks as well as their feeding habits and animal hosts, which have evolved differently in different locations (Steere, 2001). Humans then acquire the bacteria when fed on by an infected tick.

In general, in situations where Lyme disease is diagnosed shortly after being contracted and treatment is commenced, most physicians believe traditional antibiotic therapy can be effective in eradicating the bacterial infection of Acute Lyme Disease. Late symptoms are also recognized as a category of Lyme disease, as well as post Lyme syndrome for patients who originally had a laboratory diagnosis. There are issues, however, relating to the prompt diagnosis and treatment of the disease. Where a *B.b.* infection is not diagnosed, clinically or with a laboratory test, the infection may be able to progress within the body, with or without symptoms, and in many cases will become much more difficult to treat. Due to the ongoing symptoms that patients sometimes experience, the condition is called "chronic Lyme disease".

Lyme disease is recognized as a significant medical problem that is difficult to identify, diagnose and treat. The political and social dimensions of the disease are as challenging as the medical and scientific.

The following is a categorization of the

| **Categories associated with Lyme disease** |
|---|
| |
| Early localized infection (1 to 4 weeks)<br>Minor symptoms occur, such as a rash. A critical period in the disease before dissemination of the bacteria begins. |
| |
| Early disseminated infection (1 to 4 months)<br>More significant symptoms. The bacteria disseminates into the body. Harm to the body if left untreated. |
| |
| Late Persistent Infection or Post-Lyme Disease Syndrome<br>On going symptoms after having received standard treatment. Also called Chronic Lyme Disease including patients that may not have had any initial treatment. |

Feder, et al. (2007) states that "although chronic Lyme disease clearly encompasses post-Lyme disease syndrome, it also includes a broad array of illnesses or

symptoms for which there is no reproducible or convincing scientific evidence of any relationship to *B.b.* infection. Chronic Lyme disease is used in North America and increasingly in Europe as a diagnosis for patients with persistent pain, neurological and cognitive symptoms, fatigue, or all of these symptoms; with or without clinical or serologic (diagnostic) evidence of previous early or late Lyme disease".

This is where the controversy begins.

While relatively few new cases of acute Lyme disease are diagnosed each year in BC, there are many people in BC who believe they have chronic Lyme disease, and also see very few avenues within the province to address their perplexing and serious condition.

This report will address the significant medical, scientific, and political issues that most would agree are in dispute or, at the very least, require resolution.

Physicians within the mainstream clinical practice medical community are very skeptical about chronic Lyme disease. This is understandable in that the condition has the symptomatic characteristics of a number of other common chronic diseases or ailments in Western society: arthritis, multiple sclerosis, chronic fatigue, to name a few. The symptoms are so common that it is possible patients may actually have other infectious or chronic diseases. In this group's opinion, a specific diagnosis is required by physicians to provide treatments which, due to the lack of a definitive diagnosis in a large proportion of the cases, is often difficult to obtain. Even psychosomatic illness is sometimes suggested when no definitive diagnosis can be made. When there is no clear medical diagnosis, the most conservative management guidelines - often suggesting no treatment- hold sway among the litigation averse. In North America and Europe the guidelines of the Infectious Diseases Society (ISDA) heavily influence the practices of most physicians.

Physicians who are more involved with and disposed to this patient group's symptomatology look at Lyme disease more organically and accept that chronic Lyme disease is a bona fide syndrome, based on the patient's symptoms and diagnostic tests (where possible) and evidence of the patient residing in, or having visited, an endemic area for *Ixodes* tick. The condition can be debilitating and is sometimes seen as a hopeless cause in the eyes of patients. This medical group also uses management guidelines, but ones that are developed by another medical society, the International Lyme and Associated Diseases Society (ILADS).

Both factions have their extremes, ranging from complete acceptance of sometimes insensitive laboratory results and denial of chronic Lyme disease on one side, to misuse of expensive diagnostics and extended IV antibiotic therapy for little gain on the other.

In the figurative sense, the battleground in the USA has been set by the intervention of a State Government of Connecticut, where scientific debate of standard medical practice guidelines for Lyme disease is currently taking place.

In a March 2009 Annals of Neurology Wordpress article, the authors point out that the controversy came to a head in 2006 with the publication of Treatment Guidelines for Lyme disease by the Infectious Diseases Society of America (IDSA). These guidelines promptly became the target of an antitrust investigation by the State of Connecticut. The IDSA guidelines stated that there was no convincing biologic evidence for Chronic Lyme Infection and recommended against antibiotic therapy beyond the short-term treatment of acute infections.

While there are many arguments in medicine between the medical mainstream and those with contrary opinions, this polarization concerns politically charged area of health care. Disagreement is strongest in the areas of diagnostic test accuracy and prolonged antibiotic therapy in symptomatic patients. The very existence of chronic Lyme disease has been questioned by IDSA affiliated doctors and scientists. Alternatively, the medical and scientific members of the International Lyme and Associated Diseases Society (ILADS) cite its guidelines as evidence that chronic Lyme disease does exist and prolonged antibiotic treatment for chronic symptoms is a reasonable approach under medical supervision. The ILADS guidelines reference a number of scientific articles demonstrating these assumptions. There is concern by IDSA affiliated physicians and scientists that the ILADS treatment guidelines are being used to justify antibiotic therapy for nebulous symptoms that are not linked to Lyme infection.

David Holtzman, at the State University of New York summarizes: On one side are the experts who dismiss the possibility of persistent infections and discount incontrovertible examples of Lyme Borrelia infection. On the other side are a large number of ailing people with a myriad of symptoms but no real proof of exposure to bacterial spirochete Borrelia Burgdorferi, who nonetheless desperately demand antibiotics and are supported by strong advocacy groups. What is needed is better diagnostic tools and objective science. (Rosa et al., 2005)

There is no doubt that large numbers of patients in North America and Europe are living with symptoms of a disease state whose cause cannot be easily identified. Legislators in regions of North America where *B.b.* is common are hearing from thousands of constituents and their families (as patients with a largely unidentified chronic disease) that it's time to look at real, practical solutions.

Approach to the Review

The literature on Lyme disease and associated infections is sizeable, as are the number of issues associated with the identification and treatment of Lyme disease. In this review, the focus will be on *chronic Lyme disease, with a focus on key references, review articles and recent literature, in the areas of:*

- Transmission of *B.b.* and related co-infections.
- Inflammation and the immune system in chronic Lyme disease.
- Bacterial persistence and Biofilm development in chronic Lyme disease
- Diagnosis of Lyme disease, and Treatment of chronic Lyme disease
- Relationship of chronic Lyme disease to other chronic diseases, and
- Political/Legislative/Guideline Issues in chronic Lyme disease

## 2. The Characteristics of Lyme Disease and Related Diseases

Transmissibility

Lyme disease is spread by ticks. Some believe it can also be transmitted by biting flies and mosquitoes but this has not been scientifically validated. Other sources of transmission are through blood transfusion and to a lesser degree, organ transplantation. The US Department of Health and Human Services, "Lyme disease - the facts and the challenges. July 2008", states that even pregnant women, if infected with Lyme disease, can pass on the infection to their unborn children.

West of the Rocky Mountains the *Ixodes pacificus* (*I. pacificus)* tick is the predominant mode of transmission. *Ixodes scapularis* (*I. scapularis*) is within the same tick family *Ixodes rincus* (*I. rincus*) and is the vector for Lyme infection in eastern North America. In its Lyme disease Bulletin (2008), the BC Centre for Disease Control (BCCDC) reports that less than 1% of ticks that are regularly tested have the *B.b.* bacterium. In Oregon State, its only study reported a 3.5% infection rate in the *pacificus* tick.

At National Lyme disease Meeting March 8-9, 2006, Update on the Ecology of Lyme disease and *Ixodes* species on the West Coast, Muhammad Morshed (Medical Microbiologist, BCCDC) gave an update on Lyme disease in British Columbia, where it was first documented in 1993. Over 21 tick species are documented in British Columbia, with *Ixodes pacificus* and D. andersonii as the two most common types. *I. pacificus* is mostly found in the Lower Mainland, Fraser Valley, and Vancouver Island, and from the *Ixodes* family, *D. andersonii* in the Interior. During the years 2000 to 2004, communities submitted ticks to the BCCDC. Annually, about 400–500 *I. pacificus*, about 50 *Ixodes angustus*, about 200–300 B. *andersonii* and about 20–30 other species were received. All *Ixodes* spp. ticks collected through field studies and received from veterinarians, physicians and the public were cultured for *B.b.*; of 3,148 tests, 7 (0.22%) were culture positive.

Morshed also reported at the 2006 Lyme disease meeting, that most of the positive ticks were found from the Lower Mainland and Vancouver Island. Ticks have been found as far north as Smithers and as far east as Cranbrook. In the northern extremes however there was no evidence that any tick species has carried the *B.b.* bacteria. Genetic analysis of 30 randomly chosen strains from over the years has identified both *B.b.. Sensu stricto* and *B. bissettii* present in the province. In addition, ticks have been collected from birds from across Canada to identify the distribution of avian-associated tick species. Among them *I. scapularis*, *I. pacificus*, and *I. auritulus* were found to carry *B.b.* spirochetes.

In extreme examples in North Eastern United States, recent epidemiologic data suggest that many infections go unrecognized. In endemic areas, such as Connecticut, incidence rates from 1997 to 1999 were 24 to 51 cases/100,000 population.
Symptomatic infection in Europe appears to be rare; 66 cases have been reported, despite a median rate of infectious ticks of 6.2% in *I. Rincus* ticks among 35

published reports. Similarly, another report cites a median prevalence of infectious ti[illegible] in European *I. ricinus* ticks is 3% (45 publications), a figure close to that observed among North American *I. scapularis* and *I. pacificus* ticks (median 4.7% among 42 publications). (Ballantyne, 2008)

The Canadian Public Health Agency states that "the black-legged tick, *I. scapularis*, has a wide geographical distribution in Ontario, Canada, with a detected range extending at least as far north as the 50th parallel, and four out of five regions of Ontario affected." Also according to Canadian authorities, there is a connecting link to common strains of Lyme disease found in the northeastern United States."

In 1994, British Columbia was declared an endemic region for Lyme borreolis (Lyme disease). The BCCDC (2008) reports that 5-7 cases of the Lyme disease is diagnosed annually, from the 400-600 reports of citizens being bitten (but generally not infected with *B.b.*) each year.

Morshed et al. (2005) found that ground dwelling birds are parasitized by certain ticks and play an important role across Canada in the wide dispersal of *B.b.* through infected ticks. Most commonly, ticks are infected through their feeding on deer mice.

At the National Lyme disease Meeting March 8-9, 2006, Update on the Ecology of Lyme disease, panel conversation suggested that the *I. pacificus* tick is a less competent vector, compared to *I. scapularis*, of *B.b.*. *I. pacificus* is a competent vector, but may be less efficient than *I. scapularis*. Also, laboratory tests for *I. pacificus* can be more difficult than for *I. scapularis*, as the nymphs tend not to feed on rodents in the laboratory. Furthermore, climate and feeding habits are at play, as the mammalian population differs in the west and as *I. pacificus* tends to feed on lizards, which are refractory for *B.b.*. While *I. pacificus* ticks in California are known for this, it was not clear from the discussion whether ticks in BC feed on lizards.

There have been concerns expressed in the past that Lyme disease is under reported in BC. This is because 90 million American citizens live in Border States with Canada where their population generally experience higher rates of the disease. In general, the awareness of tick borne infection is significantly higher in the eastern USA and eastern Canada because of the higher rates of infection. In a more complacent BC population where only 5-7 persons are diagnosed with Lyme disease annually, there is probably less current awareness of the bacterium, and signs and symptoms may go unnoticed. Even though increased awareness of Lyme disease is important, it appears from the table below that BC's rates, relatively speaking, are corroborated by BC's neighboring US States (see Table below). However, it does not appear from on-line information that the education programs in the States of Washington, Oregon, and Idaho, are particularly strong. Because of the significant variability in symptomatology of patients receiving tick bites, and general complacency, under reporting of cases could be occurring in all jurisdictions.

**Comparisons of Incidence of Lyme Disease in BC and the States of Washington, Oregon and Idaho, compared to Eastern US States.**

| **State/Province** | **Population** (millions) 2009 | **Reported Cases** 2009 (*see ALDF ref) ( "see State web sites for recent 4-5yr Avg.) | **Rate per 100,000** population |
|---|---|---|---|
| BC | 4.5 | *6 "7 | *0.13 "0.15 |
| Washington | 6.7 | *12 "13 avg | *0.17 "0.19 |
| Oregon | 3.8 | *6 "23avg | *0.15 "0.60 |
| Idaho | 1.5 | *9 "7 avg | *0.60 "0.46 |
| Eastern USA | | | *+ 68.0 |

Historically, Lyme disease incidence is on a general but shallow upward trend, but has a much higher level in north eastern regions of the US. In the Pacific north-western states, and BC, infection rates are well below 1 case per 100,000 people whereas rates in the eastern US States are much greater.

The three Pacific Northwest States reported that a good proportion of Lyme disease cases were reported to have been contracted out of their states. At the very least, more cases than are formally registered could be in British Columbia by virtue of contracting the disease elsewhere, or having manifested through:

1. asymptomatic presentations,
2. undiagnosed co-infections
3. persons migrating to the province to live
4. living with Lyme disease without treatment due to inconclusive presenting symptoms, such as EM (*erythema migrans* rash), etc.
5. false negative tests due to having the tests within a month of being bitten,
6. or false negative tests due to the problems of ELISA testing. eg. false negatives, etc.

There are three additional factors that need to be considered, related to the anticipated burden of Lyme and related diseases in BC.

Climate Change

In a Bulletin of the World Health Organization, Githeko, et al. (2000) project that climate variability will have a direct influence on the epidemiology of vector-borne (such as ticks) diseases. By 2100 it is estimated that average global temperatures will have risen by 1.0–3.5 °C, increasing the likelihood of many vector-borne diseases in new areas.

Ogden et al (2009) cites studies that indicate that ambient temperatures now constrain the establishment of *I. scapularis* in Canada to the warmer regions of southeastern Manitoba, southern Ontario and Quebec and some regions of the Maritimes. As such, projected increases in temperature with climate change are expected to permit and accelerate the expansion of *I. scapularis* into Canada. In BC the inference is that similar climate conditions will increase the range of the endemic areas further north, with anticipated increases in the incidence of the infectious diseases such as Lyme disease.

Brownstein, et al (2005) project that there will be a significant expansion of the *Ixodes* tick into Canada with an increase in suitable habitat of 213% by the 2080's. His model forecasts the emergence of higher levels of tick-borne infectious disease in Canada.

## Co Infections with Lyme disease

PCR analysis of *I. scapularis* ticks collected in New Jersey identified infections (in a proportion of ticks) with *B.b.* (33.6%), *Babesia microti* (8.4%), *Anaplasma phagocytophila (HGE)* (1.9%), and *Bartonella* spp. (34.5%). The coexistence of these bacterium has also been found in human sera (Magnarelli et al, 1995).

Tokarz et al. determined the prevalence of polymicrobial infection in *Ixodes* ticks with *B.b.*, *Anaplasma phagocytophilium*, *Babesia microti*, *Borrelia miyamotoi*, and Powassan virus in 286 adult ticks from the two counties in New York State 71% of the ticks harbored at least one organism; 30% had a polymicrobial infection. Infections with three microbes were detected in 5% of the ticks.

The *I. scapularis* tick is a potential pathogen vector that can cause co-infection and contribute to the variety of clinical responses noted in some tick-borne disease patients. *Bartonella* species (19.2%) were also detected in *I. pacificus* ticks collected in Santa Clara County, Calif. (Adelson et al, 2004).

In Connecticut, 11% of people with Lyme disease are also infected with *Babesia*, and in the northern Midwest, *Babesia* co-infection has been observed in 9% to 16% of patients with Lyme disease (Shoemaker et al., 2006). The Lyme bacterium is just one of many disease-causing bugs that ticks can carry. There are others, such as babesiosis, a malaria-like parasite that invades red blood cells and whose symptoms range from none at all to death (Vannier, 2008).

Bakken and Dumler (2002) report that six hundred cases have been diagnosed in North America and that several cases have been reported on the west coast. The disease has been found in association with *B.b.*. Teglas and Foley (2005) report on differences in the transmissibility of HGE (*anaplasma phagocytophilium*). The western *I. pacifica*, showed a significantly higher vector competence for the bacterium, compared to the eastern *I. scapularis*.

Although less frequent, Eskow and Mordechai (2001) conclude that their data implicates *Bartonella henselae* as a potential human tick-borne pathogen. Patients with a history of Neuroborreliosis who have incomplete resolution of symptoms should be evaluated for the infection.

Co-infection, which is generally manifested by more severe symptoms than occur with either disease by itself, represents a major therapeutic challenge (Shoemaker et al., 2006).

Humans co-infected with *B.b.*. and *babesiosis* appears to be more intense with prolonged symptoms than those with *B.b.* alone. Co-infected persons can also manifest diverse, influenza-like symptoms, and abnormal laboratory test results are frequently observed. Co-infecting pathogens might alter the efficiency of transmission, cause cooperative or competitive pathogen interactions, and alter disease severity among hosts. (Stephen et al., 2006)

The *Ixodes* families of ticks are vectors for many infectious agents. Javed et al. (2001), state that in Tick dominated areas, patients should always be tested for co-infection with *Ehrlichia*, *Babesia*, and *B.b.*.

Similarly, Krause et al. (2002) suggest physicians should consider use of tests designed to diagnose Neuroborreolis and HGE (*human granulocytic Ehrlichiosis*) in patients with Lyme disease who experience a prolonged flu like illness that fails to respond to appropriate Lyme disease therapy.

Swanson et al. (2006) conclude that the true prevalence of co-infecting pathogens among *Ixodes* ticks remains largely unknown for the majority of geographic locations. The prevalence of dually infected *Ixodes* ticks appears highest among ticks from regions of North America and Europe where Lyme disease is endemic, with reported prevalence of ≤28%. In North America and Europe, the majority of tick-borne co-infections occur among humans with diagnosed Lyme disease. Animal models demonstrate that certain co-infections can modulate the immune response. Clinicians should consider the likelihood of co-infection when pursuing laboratory testing or selecting therapy for patients with tick-borne illness.

Concerning the I. Pacificus tick that is endemic in some parts of western North America, including BC, Swanson summarizes studies that found approximately 1% of I. pacificus nymph ticks from deciduous woodlands and I. pacificus adult ticks from coastal regions were dually infected with B. burgdorferi and A.phagocytophilum. Simultaneous infection with three tick-borne pathogens is unlikely. Studies weakly suggest that molecular evidence from Ixodes ticks of dual infection with B.b. and A. phagocytophilum appears more common than B.b. and other co-infections. Regardless of the proportion of co-infectious agents involved, or the specific transmissibility characteristics of the *I. pacificus*, there is enough data, if not evidence, for A. phagocytophilum as a co-infection agent to suggest that clinicians should consider the likelihood of co-infection with this agent when pursuing laboratory testing or selecting therapy for patients (including the possibility of

patients who have moved from other endemic areas of North America where co-infection rates are higher) with tick-borne illness.

*Transmission of Bacteria through Blood Transfusion*

In addition to possible transmission of bacteria from ticks, flies and mosquito vectors, evidence has shown transmission through blood transfusion and organ transplantation. Three studies (Dobroszycki et al., 1999 and Gubernot et al., 2009, and Leiby, 2006), have demonstrated Babesia infection through blood transfusions.

Stramer et al. (2009), member of the American Association of Blood Banks, Transfusion Transmitted Diseases Committee, reviewed a large number of information sources in order to identify infectious agents (66) with actual or potential risk of transfusion transmission now or in the future in the US or Canada. Sixteen (16) high to medium risk infectious agents were named. Within the high risk category (6 agents), *Babesia* was identified as one of the three highest risk agents. *Anaplasma phagocytophilum* (HGE), a co-infection agent with *B.b.*, was identified as one of the medium risk group (10 agents) where two transmissions had been reported. The author's state that the extent of bacterial transmission through blood transfusion is a problem is of great concern in the United States and investigations are beginning to create plans to mitigate the problem.

SUMMARY

Lest we be complacent in BC, a report by the US Centers for Disease Control and Prevention (CDC) 2007, reported that its national Healthy People 2010 objective is to reduce the annual incidence of Lyme disease to 9.7 new cases per 100,000 population in 10 reference states where the disease is endemic (Connecticut, Delaware, Maryland, Massachusetts, Minnesota, New Jersey, New York, Pennsylvania, Rhode Island, and Wisconsin). Their report summarized surveillance data for 64,382 Lyme disease cases reported to CDC during 2003-2005, of which 59,770 cases (93%) were reported from the 10 reference states. The average annual rate in these 10 reference states for the 3-year period (29.2 cases per 100,000 population) was approximately three times the Healthy People 2010 target. The rate of infection in BC in currently 0.15 new cases per 100,000 population. While there is some explanation for these low rates, it is important to remind all British Columbians that we are nonetheless living in an endemic area, that is (globally) warming, to the benefit of *Ixodes* ticks, and the population must be continually reminded to take steps to reduce their risk for infection.

In the Executive Summary report of the PHAC 2006 Lyme disease meeting it was stated that the number of reported cases of Lyme disease in Canada is relatively low (20-60 new cases reported annually), with over half associated with travel outside of the country. The actual number of cases is undoubtedly higher given that not all clinical cases are captured by current surveillance approaches and there are indications that the incidence of the disease may be increasing. The recent inclusion of Lyme disease as a nationally notifiable disease may help address these issues

over time. The use of definitions specifically for surveillance may also help improve ascertainment of case numbers in Canada.

### 3. The Nature, Behavior and Environment of the Borrelia burgdorferi

Certain features of *B.b.* have impeded genetic investigations. The bacteria classification of *B.b.* as a Spirochete, represents a classification of bacteria that is distinct from many other bacterial groups. The infectious cycle and unusual genomic structure of *B.b.* presents several interesting basic biological and molecular questions. (Rosa et al., 2005).

In clinical presentation, in the acute phase, the bacterial infection from *B.b.* usually evolves in two stages: a primary red skin lesion called *erythema migrans* (EM); later on, invasive bacteria disseminate to distant sites inducing secondary manifestations (neuropathies, arthritis, carditis, late skin disorders). Legal et al. (2006) found a pattern of invasiveness, related to *B.b.*, using the mouse as an experimental host of *B.b.* The scientists confirmed a complex gene activated process that is used by *B.b.* to help it disseminate throughout the host. Similar mechanisms have also been implicated in the spread of cancer.

Johnson and Conly (2005) present a number of facts about the incidence of erythema migrans rash. In a clinical trial, 20 to 30% of study subjects did not have EM. In another group 17% did not have EM. In a final group 18% of patients had systemic symptoms without EM. Stricker and Phillips (2003) report that about 20 to 25% of Lyme disease patient's do not remember developing a characteristic *erythema migrans* rash. According to recent health department statistics in Texas, Connecticut and California, the same authors report the EM rash failed to appear in 41 to 65% of Lyme disease patient's. Recognition of the rash may even be lower depending on the location of the tick bite and the awareness of the person who was bitten.

Since the presence of an EM rash is the best evidence for Lyme disease it has become the most common criterion for admission into Lyme disease studies. Since most patients in the studies have an EM rash, the incidence of the rash becomes inflated in the medical literature. The literature then perpetuates the thinking that the vast majority of Lyme disease patients have an EM rash (Harvey and Solvato, 2003, and Lautin et al., 2002).

However, the current IDSA guidelines state that clinical findings are sufficient for the diagnosis of EM but clinical findings alone (without laboratory diagnosis) are not sufficient for diagnosis of Lyme disease, HGE or *babesiosis*. Therefore, for approximately 20% or perhaps greater proportion of patients who do not have EM they cannot be diagnosed with Lyme disease without a confirmatory test. Importantly, with the two-step testing regime recommended in the IDSA guidelines, the first stage of the test is not recommended until approximately four weeks after the tick bite. This is because the first step test for Lyme disease, requires the presence of an antibody response, which is not manifested for this period of time.

The ability of *B.b.* to move through highly viscous media is thought to be critical to the ability of this spirochete to penetrate tissues and disseminate within the mammal or tick. A significant proportion of the *B.b.* chromosome encodes proteins that are involved in motility and chemotaxis. (Rosa et al., 2005). As one example, Stanek et al. (1990) isolated *B.b.* from the myocardium of a patient with longstanding cardiomyopathy.

Early Lyme disease can readily be treated if it is detected early. However, early Lyme disease often goes undetected due to lack of awareness of the tick bite and an absence of the EM rash. Recent studies have shown that tick saliva carries immunosuppressive substances that allow tickborne agents to invade tissues while paralyzing the local immune response. The Lyme disease spirochete may rapidly disseminate and become entrenched and resistant early in the disease. Co-infections may alter the course of early Lyme disease and these co-infections may make the Lyme disease patient more difficult to treat. (Stricker and Phillips, 2003)

## Inflammation and the Immune System

While many tick bites, or other forms of bacteria transfer, may resolve on their own, *B.b.* can spread through the skin or other tissues. Steere (2001) notes that inflammatory innate immune responses are critical in the control of early, disseminated infection. Despite the body's immune responses *B.b.* may sometimes survive in certain sites.

Lyme disease is caused by infection with the spirochete *B.b.* and is characterized by bacterial persistence and inflammation of many host tissues. *B.b.* expresses proteins with inflammatory properties that can contribute to localized tissue inflammation. Neutrophils are the predominant first responder into the inflamed arthritic joints, and are crucial for controlling the spirochete infection. But they may also contribute to the joint pathology associated with Lyme arthritis. Thus, models of Lyme arthritis should include the possible contribution of direct activation of neutrophils (immune system cells) to both defense and disease. (Morrison et al., 1997)

Any explanation of how microbial infections might set off autoimmune diseases must take into account the observation that all individuals appear to harbor potentially autoreactive lymphocytes, but that these cells remain innocuous unless somehow activated. Epitope mimicry is when an antigen on one of the bacteria's proteins (for example) is structurally similar to a determinant on one of the proteins made by the host, although different enough to be recognized as foreign by the host's immune system. The immune response to the microbial (e.g. a bacteria) determinant would then cross-react with host tissue and eventually result in autoimmune destruction. A prominent late manifestation, particularly in North America, with *B.b.*is an inflammatory joint disorder that resembles rheumatoid arthritis. In about 10% of patients with Lyme arthritis, joint inflammation resists extended antibiotic therapy. This has prompted the hypothesis that antibiotic-resistant Lyme arthritis is an autoimmune disease (Benoist et al, 2001). Cascao (2010) refers to contribution of neutrophils in rheumatoid arthritis. Neutrophils are phagocytic (cell killing)

leukocytes that play crucial roles in the acute defense against pathogens while modulating the function of other immune cells and contributing to the perpetuation of an initial inflammatory response.

The agent of Lyme disease, *B.b.*, produces membrane lipoproteins possessing potent inflammatory properties linked to disease pathology. Data indicate that receptors associated with *B.b.*. facilitate the inflammatory events associated with Lyme arthritis. (Hirschfield et al., 1999).

In Fallon and Marconi (2009), Dr. Dantzer reports that Lyme disease patients often present with non-specific symptoms that include pain, fatigue, sleep disturbances, mood disorders and concentration problems. These symptoms are often viewed as the result of persistent psychological distress caused by the disease. However, there is now evidence that the organism itself or inflammation caused by tick bites and *B.b.* can either directly or indirectly induce the expression of inflammatory mediators in the brain. The initial inflammatory event from years earlier (due to infection or other biological trauma) may be reactivated at a later point and, because of that past event, fail to turn off after being reactivated – causing chronic sickness symptoms.

Mammalian enzymes in the blood play a critical role in mammalian infection by *B.b.*. The Lyme disease spirochete expresses several plasminogen-binding proteins (creates plasmin that degrades fibrin clots). In its conversion to plasmin it may facilitate the bacterium's dissemination throughout the host by degrading the extracellular matrix. Brissette et al. (2009) demonstrated plasminogen binding by three highly similar *B.b.* outer surface proteins, all of which are expressed during mammalian infection.

The Lyme disease spirochete, *B.b.*, is capable of infecting a wide variety of vertebrates, suggesting the ability to contravene their immune defenses. *B.b.*. produces multiple different proteins on its outer membrane during mammalian infection. The data suggest that the presence of these proteins on its surface can allow a single *B.b.* bacterium to resist being killed in any of the wide range of potential hosts that it might infect. Thus, these proteins likely contribute to the persistence of *B.b.* in nature and to the ability of this bacterium to cause Lyme disease in humans and other animals (Stevenson et al., 2002).

Bubeck-Martinez (2005) concludes that *B.b.* have adapted very well to both surviving and persisting in the mammalian host despite a strong host antibody response. It appears that outer surface proteins have contributed to this persistence. The spirochetes are able to bind proteins to their surface, resulting in decreased complement activation (a biochemical process to kill the cell). In addition, the organisms have taken advantage of components of tick saliva to aid in their initial immune evasion and dissemination.

Results of in vitro testing indicate that the toll like receptor (TLR) pathway mediates, at least in part, the release of inflammatory mediators in human white blood cells stimulated with live *B.b.* spirochetes and furthermore suggests that the

interaction between these cells and live spirochetes is mediated by spirochetal lipoproteins. (Vida et al., 2009)

Fallon et al. (2010) says Lyme disease, can cause multi-systemic signs and symptoms, including peripheral and central nervous system disease. This review examines the evidence for and mechanisms of inflammation in neurologic Lyme disease, drawing upon some human studies and controlled research with rhesus monkeys. He suggests that when Lyme disease affects the brain and spinal cord, it can look like multiple sclerosis. The brain MRI among patients with Lyme disease may at times be suggestive of a demyelinating disease. Hildenbrand et al. (2009) concludes that cytokine activation is created by proteins expressed by bacterial cells that interact with cells of the immune system in order to regulate the body's response to disease and infection. This function may represent one important contributor to the chronic persistent symptoms of fatigue, pain, and cognitive dysfunction that patients may continue to experience despite having been treated for Lyme disease. Directions for future human research are suggested that may help to clarify the role of inflammation as a mediator of the chronic persistent symptoms experienced by some patients despite antibiotic treatment for neurological Lyme disease.

## Bacterial Persistence

Phillips et al. (1998) note that the possibility of bacterial persistence has long been a contentious issue between IDSA and ILADS doctors. Although persistent infection has been, to date, strongly suggested in chronic Lyme disease by positive PCR and antigen capture, the authors suggest there were major problems with the tests.

Stricker (2008) provides a review of a relatively new book on the Molecular Biology of Spirochetes, edited by Cabello, Hulinska and Godfrey. In the book, information is provided on what is known about the factors that lead to persistence of this organism, and mechanisms of antibiotic resistance in *B.b.*. Spirochetal infection like *B.b.* can mimic many other diseases, and the 'great pretender' mantle handed down from syphilis to Lyme disease is well deserved.

In 2003, Keren et al. found that bacterial populations produce persister cells that neither grow nor die in the presence of antimicrobial antibiotics. Persisters are largely responsible for high levels of biofilm tolerance to antimicrobials. With antibiotic testing, they found that production of persister cells depends on growth stage. The elimination of persister cells could be related to growth stage of the bacterial population. Their data indicate that persisters are specialized survivor cells, and that biofilm survival is based on the presence of persisters, not on any biofilm-specific resistance mechanisms. Considering that biofilms are responsible for over 60% of all human infections, the study of persisters becomes highly significant.

Hodzic et al. (2008) state that Lyme borreliosis is a multisystem disorder that arises from tick-transmitted infection with *B.b.*. When infection is left untreated, *B.b.* can effectively evade host immune clearance, resulting in persistent infection that may

or may not be manifested as clinical disease. They summarize that studies of a wide variety of laboratory animals indicate that persistent infection is the norm in fully immunocompetent hosts. There is evidence for persistence in human cases as well (Stanek et al., 1990). In light of the mounting evidence, it is interesting that scientists such as Feder et al. (2007), in his Critical Appraisal of Lyme disease article, would continue the assertion that it is an unproven and very improbable assumption that chronic *B.b.*. infection can occur in the absence of antibodies against *B.b.*. Feder et al. (2007) also state that negative results of serologic tests are often attributed to previous antibiotic therapy or to the theory that chronic infection with *B.b.* suppresses humoral immune responses; neither theory is well supported by scientific data.

Moniuszko et al. (2009) states that recently more and more reports of so called "Post Lyme syndrome (PLS)" have appeared. PLS is connected with patients with a history of Lyme disease (who have received antibiotic treatment). It is suspected that main factors responsible for PLS are: slow regression of infection, its turning into chronic process and permanent destruction of tissues or induction of immunological response against *B.b.*. The author states that so far there is no treatment for PLS.

Regardless of the etiology, there are now treatment outcomes that demonstrate that a proportion of patients can develop persistent symptoms consistent with chronic Lyme disease (some physicians and scientists call it Post Lyme Syndrome (Category 4, page 3). Also see Section 5.

Cysts and Biofilms

Persistence is created in two forms. The bacteria can assume a cystic form that allows the bacteria to lie dormant in the human host, thus evading antibiotic therapy that targets replicating bacteria. The non-replicating cyst form is the key to persistence of infection, and any antibiotic approach to Lyme disease that fails to recognize this pathogenic entity is doomed to failure (Stricker and Lautin, 2003). The other form relates to the fact that bacteria in natural environments usually form communities of surface-adherent organisms embedded in an extracellular matrix. These are called biofilm.

In Fallon and Marconi, 2009, Dr. Marconi reports that spirochetal infections of humans can be chronic and in the absence of treatment can persist indefinitely. Molecular mechanisms are employed by Lyme disease to evade the innate immune system.

Biofilms represent potential unrecognized stages in the pathways from infectious agent exposure to chronic disease. In both situations cultures and even PCR results can be negative (O'Connor et al., 2006). Aparna and Yadav, (2008) confirm that transitioning from acute to chronic infection is frequently associated with biofilm formation. The current antibiotic therapies are of limited effectiveness in resolving biofilms infection.

Once the balance between colonization and infection has been tipped in favor of overt infection, biofilms constitute a peculiar problem that characterizes 65% of infections treated by physicians in the developed world (Costerton et al., 1999 and Marshall, 2007). Lewis (2007) suggests the proportion is closer to 80%. Marshall states that electron microscopy has shown that these bacterial communities can evade phagocytosis (cell death), and persist in the cytoplasm of monocytes, macrophages, lymphocytes and neutrophils. Three decades ago, Wirostko et al. (1989) found such cell communities in Crohn's disease, Juvenile Rheumatoid Arthritis and Sarcoidosis. However, the mechanism(s) by which such persistent bacteria could evade the immune system have remained elusive. Recently, species of gliding bacteria never thought to be able to survive in vivo, have been found in surgically removed biofilms. Studies need to be done to identify whether the genomes of these gliding bacteria might yield insight into mechanisms by which such persistent pathogens could evade phagocytosis (cell death).

Bacteria have traditionally been regarded as individual organisms growing in homogeneous planktonic (free floating) populations. However, bacteria in natural environments usually form communities of surface-adherent organisms embedded in an extracellular matrix, called biofilm. Current antimicrobial (antibiotic) strategies often fail to control bacteria in the biofilm mode of growth. Treatment failure is particularly frequent in association with inserted or implanted medical devices and compromised host immunity. (Fux et al., 2003).

Because bacteria apparently prefer a sedentary lifestyle to a nomadic existence free floating in the blood stream. Biofilms have been described as "a structured community of bacterial cells enclosed in a substance called a polymeric matrix and adheres to an inert or living surface. These compounds are created by microorganisms, such as *B.b.*, that may be residing in the body. These compounds are important in biofilm formation and the cells attachment within the body. Although biofilms have been well known for a long time, it has not been until recent years that their importance in human disease has been realized. The extracellular matrix (a medium in which the bacteria grow) creates a scavenging system for trapping and concentrating essential minerals and nutrients from the surrounding environment (del Pozo and Patel, 2007). It may provide a certain degree of protection against environmental threats. It is suggested that biofilms are present in a large proportion of all bacterial infections.

Biofilm formation is a crucial step in the creation of many sub acute and chronic bacterial infections. Biofilms are difficult to eradicate with conventional antimicrobial agents. Biofilms also have several potential antimicrobial resistance mechanisms. Bacteria that attach to a surface and grow as a biofilm are protected from killing by innate host defenses and antimicrobial agents. This is a type of resistance unique to biofilm-associated bacteria and very distinct from conventional antimicrobial resistance. Although some antibiotics can penetrate biofilms, they may not be able to kill the bacteria within them. Enzymes within the biofilm may destroy antibiotics as well. Bacterial cells that can survive antibiotic insult have been referred to as "persisters" and may be present in relatively high numbers in the deep biofilm.

Antimicrobial treatment of bacterial biofilms may lead to eradication of most of the susceptible population. But, a small fraction of persister cell variants could survive the assault and would be able to reconstitute the biofilm following discontinuation of antimicrobial therapy. This may depend on the specific antimicrobial agent being used. Thus, the presence of persister cells may explain the resistance of bacterial biofilms to certain types of antimicrobials. Persister cells play a major role in the tolerance of biofilm bacteria to antimicrobial agents. Understanding the mechanisms involved in biofilm-associated antimicrobial resistance is key to development of new therapeutic strategies (del Pozo and Patel, 2007, and Costerton JW et al., 1999).

In most natural environments and in chronic bacterial infections, the free floating bacterium generally exist only transiently, and usually as a minor population. Emerging evidence describes bacterial populations are embedded in a self-secreted matrix, that provides numerous advantages for persistence in the face of environmental and host challenges. Therefore, biofilms and the existence of a complex bacterial life cycle provide a new perspective through which to view infectious diseases. There is now evidence for the theory that chronic infections are fundamentally different than acute infections, and that different interventional approaches are necessary to treat these biofilm infections more efficiently. The biofilm provides a fortress for bacteria. A good example of a biofilm is dental plaque. Within the biofilm there are added defenses against antibiotics and the immune system. Biofilms have demonstrated the ability to persist in 100 to 1000 times the normal therapeutic concentrations of antibiotics. Changing the perspective about chronic infectious disease to include biofilm will likely be an important advancement in opening up new methods for detection and treatment. The authors suggest that with molecular tests now becoming a viable part of routine bacterial analysis, this should be considered (Wolcott and Ehrlich, 2008).

Based on direct observations, Costerton, et al. (2003), have established that the bacteria that cause *device-related* and *other chronic infections* grow in matrix-enclosed biofilms. They conclude that the diagnostic and therapeutic strategies of the past are not effective with biofilm diseases. Aparna and Yadav (2008), confirm this finding. If biofilms form on the surfaces of tissues or other natural surfaces (e.g., teeth) or artificial and extraneous inert materials in the body, they may grow slowly and be well controlled, in that the planktonic cells that are released may be killed by normal defense mechanisms. Although antibiotic therapy and activated host defenses can kill derived planktonic cells and often obviate symptoms, they cannot kill the biofilm cells that constitute the focal points of these chronic infections. Colonized medical devices must often be removed in order to effect a cure and patients with devices and tissues that cannot be removed must reconcile themselves to intermittent antibiotic therapy for the remainder of their lives. The most immediate hope in this dismal prognosis is the pace at which biofilm science discovers new agents that preclude biofilm formation and lock potential pathogens in the planktonic (free floating)mode of growth, in which they can be killed by antibiotics and host defense factors.

In a recent review article in the journal, Nature, (Lewis, 2007) concludes that there is increasing evidence to suggest that bacteria have the ability to enter into a

dormant (non-dividing) state. This enables a cell to survive being challenged by [illegible]tics without expressing or using resistance mechanisms (this is known as antibiotic tolerance). In experimentation, even though the cells within the biofilm are susceptible to antibiotics, a small subpopulation of cells nonetheless remains alive irrespective of the concentration of the antibiotics. Because of the biofilm matrix, persister cells that are contained within it can survive both the onslaught of antibiotic treatment and the immune system. When the concentration of an antibiotic declines, persister cells can repopulate the biofilm, which will shed off new free-floating cells, producing the relapsing biofilm infection (Lewis, 2001) that had been temporarily moderated by the antibiotic therapy.

Lewis (2007) considers that infections such as syphilis, Lyme disease, and tuberculosis can persist in the body for many years. In most chronic, persistent, infections he speculates that the pathogen is at least partially shielded from the immune system, where syphilis and Lyme disease migrate into the CNS. Although progress has been made towards understanding the metabolic functions that are required for persistence, no persister genes have been isolated for any other bacterial pathogen that causes latent (chronic) infection.

It is possible that a lengthy, lingering infection that is not eradicated owing to tolerance (the ability of cells to survive killing by antibiotics without expressing or using resistance mechanisms) is likely to provide a fertile ground for the emergence of resistant bacterial mutants, or for the acquisition of resistance (tolerance) through gene transfer from other species of bacteria (Levin and Rosen, 2006). They hypothesize that a new anti-persister drug based on traditional approaches could be produced by combining a conventional antibiotic, and an inhibitor of an essential persister protein. Lewis (2007) states that the entrance of cells into a dormant, persistent state is largely responsible for the multidrug tolerance of infections. Persisters are likely to be responsible for latent chronic diseases, which can be suppressed, but not eradicated, with existing antimicrobials. The need to develop novel therapeutics capable of killing persister cells and eradicating infections is acute in the opinion of Lewis. Finding genes responsible for persister formation and maintenance should lead to drugs that disable persister's hand might allow conventional antibiotics to eradicate an infection. Stewart (2002) suggests that each gene and gene product contributing to antibiotic resistance (tolerance) may be a target for the development of new chemotherapeutic agents. Disabling biofilm resistance may enhance the ability of existing antibiotics to clear infections involving biofilms.

## Summary of the literature on Biofilm and Lyme Disease Infection

The study of chronic Lyme disease, particularly which related to the existence of persistent infections of B.b. and other bacteria within the body, is a strongly expanding field. While there is strong evidence in animal models, most of the work has not been substantiated in humans. What is known is:

a. That biofilms containing bacterial infections exist within humans is found mainly in studies of other infections, confirm the presence of biofilm with bacterial infections in humans. Examples of such studies (Chole and Faadis,

2002 and Hall-Stoodley, 2006), identify common bacteria, but not *B.b.* at this time.

b. That biofilms containing bacterial infection with B.b. have been found in very limited and unrepeated circumstances in a few human cases in the myocardium and a joint, and there is no evidence that these infections were susceptible or resistant to antibiotic treatment at this time.

c. That *B.b.* commonly exists within biofilms within experimental and animal models, that have also been shown to be resistant to antibiotic treatment.

Genetic Variants of the Lyme disease Bacteria

Lyme disease is known to present with varied symptoms and illness characteristics. Qui et al. (2008) sequenced 68 isolates of the bacteria from Europe and North America. The researchers found that one outer surface protein on the *B.b.* clone (the ospC-A clone) appeared to be highly prevalent on both continents, and isolates of this clone were uniform in DNA sequences, which suggest a recent trans-oceanic migration. On the other hand, they found other isolates that were distinct to each continent. The researchers conclude that the *B.b.* bacteria (ospC-A clone) has dispersed rapidly and widely in the recent past. The spread of the *B.b.* variant with this particular outer surface protein may have contributed, and likely continues to contribute, to the rise of Lyme disease incidence.

With this new knowledge of a common strain of bacterium in North America, researchers can now look at whether different strains have different capacities to cause disease," and the diversity in strains could potentially explain why certain antibiotics don't work well for some people (Ballantyne, 2008).

A novel approach to the understanding of Persistent Infection

Autoimmunity occurs when the immune system recognizes and attacks host tissue. In addition to genetic factors, environmental triggers (in particular viruses, bacteria and other infectious pathogens) are thought to play a major role in the development of autoimmune diseases (Ercolini and Miller, 2009). Built on the mounting likelihood of the existence of biofilm environments in the body, a novel theory is based on the hypothesis that chronic disease with certain characteristics are the result of infections within cells that cannot be killed by the immune system. These infectious bacteria types have developed the ability to remain alive and proliferate undetected inside the cells they infect. The cells include the body's own immune cells, like macrophages, the cells of the immune system that the body uses to kill invading pathogens (like bacteria). Once inside these cells, they cause the body's own cells to release inflammatory products that can cause pain and fatigue, for example.

These bacteria are also hypothesized to sustain themselves by grouping into communities called biofilms. Inside a biofilm the bacteria produce a protective

matrix that allows them to more effectively evade the immune system and are able to resist the effect of antibiotics. Under certain conditions, they mutate from classical bacteria, losing their cell walls in the process. This thinking is based on research on the human microbiome (all the microorganisms in the body which are found in association with both healthy and diseased humans) which shows that bacteria are far more pervasive within the human body than previously thought, increasing the possibility that autoimmune disease is bacterial in origin (Albert et al., 2009). When the innate immune system cannot function, the body cannot keep the bacteria in their bodies under control. Mechanisms are needed to ensure that specific immune receptors are kept free of bacterial proteins so they can do their job. This mechanism is purported to improve the body's ability to turn on the innate immune system and produce cells that will kill the bacteria. The immune system is then able to kill the pathogens and manage infections.

SUMMARY
The scientific community needs to take a broad approach in thinking about the causes of infection in humans. The immune system may be very important in the development of treatment strategies for Lyme disease, in addition to understanding persistence and biofilm development.

With the growth in non-specific chronic symptomatology in the population, and the mounting evidence that persistence is now as plausible an argument as it is not, science in this area needs to be stimulated and encouraged.

## 4. Diagnosis of Chronic Lyme Disease

Perhaps, the most controversial area of the chronic Lyme disease debate surrounds the appropriate identification and diagnosis of Lyme disease. This discussion of Diagnosis in Lyme disease is intended to focus on chronic Lyme disease. However, many would argue that the reason for the existence of chronic (or late) Lyme disease is due in part to the inadequacies of laboratory testing, as well as the extremely variable presentation of symptoms to the doctor's office, with or without an EM rash.

Nonetheless, there is controversy about the frequency and even the existence of post Lyme disease syndrome or chronic Lyme disease. There is no standardized case definition for either of these terms. It is difficult to define syndromes based on such symptoms as fatigue and pain, given the high incidence of fatigue and vague pains without objective signs in the general population (Datwyller, PHAC Meeting, 2006).

According to the Control of Communicable Diseases Manual (CCDM) (17th Edition; James Chin, Ed. APHA 2000) Washington State Department of Health the late manifestations of Lyme disease are:
Musculoskeletal system - Joint swelling, followed by chronic arthritis, in one or a few joints.

Nervous system - lymphocytic meningitis; cranial neuritis, radiculoneuropathy; or rarely, encephalomyelitis, all or in combination.

Cardiovascular system - acute onset of high-grade atrioventricular conduction defects that resolve in days to weeks and are sometimes associated with myocarditis.

Regardless of the methods of diagnosis, controversy still exists around the appropriate methods for diagnosing chronic Lyme disease:

- Some physicians who see patients with suspected chronic (late) Lyme disease recommend that it can only be diagnosed on the basis of clinical symptoms, while the mainstream medical group believe that diagnosis can only be made with best practice diagnostic testing.

- The current best practice diagnosis uses serological testing that only detects the body's immune response to *B.b.*. There is however a distinct possibility that *B.b.* can become a persistent cystic form or contained within biofilm bacteria itself. It is possible that planktonic (free floating) bacteria are present in the blood stream to be tested.

- Best practice serological testing can only be used 4 to 5 weeks after the initial infection. With growing evidence that Lyme disease is a persistent infectious disease, immediate testing is required in the acute phase to prevent chronic disease. Serological testing detection levels of only 20-50% in the first stage of the disease is inadequate.

- Better diagnostic testing methods are needed for detection of *B.b.* infection.

- It is also apparent from the literature that the *Ixodes* tick is a vector for at least 3-4 other bacterial infections that also reside within the animal, bird or rodent host that harbors *B.b.*, and can manifest similar symptoms and debilitation. Testing for these organisms is only done on specific request. Based on epidemiologic findings, common co-infections with Lyme disease in British Columbia should become part of a testing panel when a tick bite is suspected.

The BCCDC (2008) advises that Lyme disease should be diagnosed through a clinical evaluation of the patient's symptoms and risk of exposure to infected ticks. The Center states that a laboratory blood test may be administered through the BCCDC, but this test should not be interpreted in the absence of a clinical diagnosis. In a June 2000 article in the Journal of the Canadian Medical Association, Odgen et al. (2009) state that because the specificity of serologic tests for Lyme disease may not be high, epidemiologic findings about the likelihood of exposure to ticks that transmit Lyme disease inform the serologic diagnosis, rather than the other way around.

Similarly, the Diagnosis and Laboratory Services, State of Washington (February 2010), state that the diagnosis of early Lyme disease is based primarily on clinical findings (e.g. presence of the *Erythema Migrans* rash) since serologic testing is insensitive during the first few weeks after onset. In the later stages of Lyme disease, the Department explains that the diagnosis of patients is commonly based on clinical findings with support from serologic tests. However, the State of Washington also states that about 20% of patients who contract Lyme disease may not have specific symptoms, including the characteristic bulls eye rash (*Erythema migrans* (EM)). Stricker and Lautin (2003) report that the EM rash could be absent in a much higher proportion of Lyme patients, although this is a finding which may be more relevant to other areas in North America.

Nowakowski et al. (2001), conclude that the clinical diagnosis of erythema migrans is highly accurate in an area where *B.b.* is endemic if it is made by experienced health care personnel, but some patients with this diagnosis may not have *B.b.* infection. No single diagnostic modality is suitable for detection of *B.b.* in every patient with erythema migrans rash.

Coulter (2005) states that Lyme disease is usually diagnosed and treated based on clinical manifestations. However, laboratory testing is useful for patients with confusing presentations and for validation of disease in clinical studies. Although cultivation of *B.b.* is definitive, prior investigations have shown that no single test is optimal for Lyme disease diagnosis. The highest sensitivity (100%) for diagnosis is obtained when culture, skin PCR, and serologic tests were used, although serologic testing with skin PCR was almost as sensitive (92%). Plasma PCR was infrequently positive and provided no additional diagnostic value. Although culture is definitive and has a relatively high sensitivity, the results required a mean of 3.5 weeks to recovery. Coulter suggests that the combination of acute-phase serology and skin PCR was 75% sensitive, offering a practical and relatively rapid alternative for confirming clinical impression. The full battery of tests could be useful for patients with confusing clinical signs or for providing strong laboratory support for clinical studies of Lyme disease.

At the Public Health Agency of Canada (PHAC) National Lyme disease Meeting 2006, Plenary on Diagnosis and Laboratory Testing, there were concerns expressed that current testing procedures could miss some patients with Lyme disease. It was acknowledged by all that issues such as the testing approach, stage of disease, the time between tick bite and test, persistence of antibody responses, interpretation of western blot tests, impact of early treatment, and the technical demands of the western blot test can affect test outcomes and that newer and developing test procedures should be evaluated. The meeting notes also recorded that the US CDC two stage approach to serologic testing is the most likely to give the best combination of sensitivity and specificity.

Serologic Testing

Bunikis and Barbour (2002) suggest that the detection of antibodies to *B.b.* is the most practical and common approach for laboratory work-up of a case of suspected

Lyme disease. Serologic assays fall short of 100% sensitivity and specificity, however, and examination of a single specimen in time does not discriminate between previous and ongoing infection. Due to a background false positivity even among healthy populations of non-endemic regions, *serologic testing is recommended only when there is at least a one in five chance, in the physician's estimation, that the patient has active Lyme disease*. The pretest likelihood of the disease is determined by the physician in the context of epidemiologic and clinical facts of the case. This estimate can serve to reassure patients who are at low risk of *B.b.* infection but are seeking a Lyme test for complaints of a more nonspecific nature. This is an unfortunate recommendation, given the variations in presentation of Lyme disease, and a physician's duty of care, especially if the patient resides in an endemic area.

Aguero-Rosenfeld et al. (2005) undertook large studies of patients where standard ELISA serological testing achieved a sensitivity of 38% during the acute phase, 67% during convalescence after antimicrobial treatment, to 87% in the serum of patients with early neuroborreliosis and to 97% in those with Lyme arthritis. Similar results have been found in other studies. Stricker and Phillips 2003 remark that the ELISA test is the preferred method to diagnose Lyme disease due to its reported sensitivity, adaptability to automation and ease of quantification. However, ELISA test misses at least 50% of Lyme disease cases due to the assay's insensitivity and variability with antibiotic treatment. Stricker and Phillips (2003) infer that the recommended two-tier testing system will mask 50% of (early) Lyme disease cases because a positive ELISA result is required to proceed to the confirmatory Western blot test. Aguero-Rosenfeld's et al. (2005) paper suggests that time-restricted use of ELISA test may reduce the sensitivity of two-tier testing for confirmation of the existence of antibodies to *B.b.* in the blood serum of treated patients evaluated slightly beyond the 4-week time point in early convalescence, before antibodies have fully developed. A concern is that many patients may move to a chronic persistent phase before they have had antibiotic therapy.

PCR Testing

Bunikis and Barbour (2002) state that cultivation of *B.b.* from a patient's skin or blood is the gold standard for demonstration of active infection, but it is expensive and lacks clinical sensitivity. Detection of spirochetal DNA in clinical samples by PCR has better sensitivity, but PCR for *B.b.* has not yet been standardized for more routine diagnostic testing.

To date, only in vitro cultivation of *B.b.* has been widely accepted to confirm clinical diagnosis of Lyme disease. Nevertheless, various PCR based molecular assays have shown increasing significance in the laboratory diagnosis of Lyme *B.b.* because of their high sensitivity, specificity and capability of quantification (Wang, 2002).

In a February 2009 study, it was reported that a real time quantitative PCR assay to identify and detect small numbers of *B.b.* in infected mouse tissues had been developed. They showed that the molecular beacons are effective, sensitive and

specific probes for detecting and estimating wide-ranging numbers of *B.b.* in the presence of mouse DNA. It was concluded that the high sensitivity and specificity of molecular beacons make them superior probes for the detection of small numbers of *B.b.*. Furthermore, the use of molecular beacons can be expanded for the simultaneous detection and quantification of multiple pathogens in the infected hosts, including humans, and in the arthropod vectors (Saidac, et al. 2009).

Although persistent infection has been, to date, strongly suggested in chronic Lyme disease by positive PCR and antigen capture, Phillips et al. (1998) state there are major problems with these tests (perhaps at that time).

Promising advances in diagnostics

A group of scientists in successive studies (Evans et al., 2005, and Mavin et al., 2007 & 2009) have studied the problem of significant numbers of patients with clinical symptoms of Lyme disease being reported in Scotland as seronegative by the ELISA test or equivocal by the confirmatory Western blot test. Through successive comparisons of US and European standards, improvements in the testing were made, reducing false negative rates. The 2007 work, went further by investigating Nine Scottish *B.b.* isolates in western blot tests. Compared to the reference isolate, two selected Scottish isolates were used in western blot testing of previously tested samples. In general the trial verified the positive or negative test results in initial tests, but converted 73% of the results that were equivocal to weak or strongly positive. In the 2009 study, scientists evaluated the use of two local *B.b.* strains in a single mixed antigen for in-house IgG western blots testing in the routine diagnostic setting by comparing it with the current in-house protocol. The results showed the positive predictive value of the two antigen preparations was the same at 96%. The negative predictive value of the mixed antigen with revised criteria was higher than the reference antigen (96% versus 88%), but the specificity was similar (97% versus 98%). As a result of this work, the mixed antigen and revised interpretation criteria have been successfully incorporated into the routine diagnostic testing service, increasing the sensitivity of the in-house IgG western blot test for Scottish patients.

Coleman and Pal (2009), report on a gene that is expressed at extremely low levels in vitro and in ticks but is dramatically induced by spirochetes once introduced into the host and is highly expressed throughout mammalian infection. Further work in this area could contribute to the development of novel serodiagnostic markers for detection of Lyme disease.

The search for improved serologic tests has stimulated the development of recombinant protein antigens and the synthesis of specific peptides for immunodominant antigens. The use of these materials alone or in combination as the source of antigen in a single tier immunoassay may someday replace the currently recommended two tier testing strategy (Aguero-Rosenfeld et al., 2005).

Weinstein (2008) summarizes that the 2-tier ELISA and Western immunoblot test is not always necessary, sufficient, properly used, or properly interpreted. An erythema migrans (EM) rash is recognized in (a minimum of) 80% of patients with early Lyme disease. The C6 peptide ELISA is a single, primary test for Lyme disease. Other studies (Cinco et al., 2006) support the overall equivalent performance of C6 peptide test, although Steere et al. 2008 expressed concern regarding the possible lower specificity of the C6 peptide ELISA. The results should stimulate research in Lyme disease testing toward eliminating one of the major causes of the misdiagnosis of Lyme disease. Tjernberg et al. (2008) conclude that the C6 serum test, together with clinical evaluation, is a powerful diagnostic tool in adult patients with suspected neuroborreliosis with a symptom duration of more than 30 days.

At the National Lyme disease Meeting March 8-9, 2006, Plenary on Diagnosis and Laboratory Testing, Nick Harris, President of IGeneX , reported on the difficulties with current diagnostic testing. Most laboratory patients do not remember an erythema migrans (EM); however many remember tick-bites throughout their life. The problem is that the ELISA has low sensitivity and specificity. An ideal test should have sensitivity and specificity of greater than or equal to 95%. At the same meeting in 2006, Harris also stated that about 20% of Lyme disease patients never make antibodies, and antibodies to *B.b.*, often are present at only low levels, or are even absent in culture or PCR-positive patients who have been suffering for years from symptoms compatible with Lyme Borreliosis. Therefore, in addition to serological testing, the use of PCR and Lyme antigen detection in diagnosis of Lyme disease is recommended. Harris stated that it was documented in the literature that the Lyme polymerase chain reaction (PCR) for whole blood and urine give false positive results. On close evaluation of these studies, it was clear that the problem was the specificity of the primers used for PCR. He stated that PCR tests have been developed that are highly specific (> 99%) for all clinical sample types, including urine. The assay sensitivity is 42% and specificity is 89%, and when confirmed by the reverse Western blot the specificity is greater than 96%.

At the same 2006 PHAC session, Barbara Johnson, Chief, Microbiology and Pathogenesis Laboratory, Centers for Disease Control (CDC), reported that ELISA is much improved but cannot be used for patients at all stages of their illness, and two-tier testing is still essential because the specificity of the ELISA alone is inadequate. CDC recommends retaining the ELISA as a first step, because Western blots are not quantitative, and because Western blots show false positives in samples from healthy donors/or patients with other illnesses. However, the US Department of Health and Human Services 2008 report indicates that tests for Lyme infection need to be improved. Scientists supported by the National Institutes of Health are re-evaluating current tests, as well as developing new tests that use the highly sensitive genetic engineering technique known as PCR (polymerase chain reaction) as well as a micro-array and high throughput to know what sequencing technology to detect extremely small quantities of the genetic material of the Lyme disease bacterium or its products and bought body tissues and fluids.

In the Executive Summary report of the PHAC 2006 Lyme disease meeting, it was noted that despite the problems with two tier testing, it is the test most likely to

give the best combination of sensitivity and specificity. However, it was clear in discussions that there were concerns that current testing procedures could miss some patients with Lyme disease. It was acknowledged by all that issues such as the testing approach, stage of disease, the time between tick bite and test, persistence of antibody responses, interpretation of western blot tests, impact of early treatment, and the technical demands of the western blot test can affect test outcomes and that newer and developing test procedures should be evaluated.

An opportunity for ideologically divided scientists to work together

In a recent article, Wormser and Schwartz (2009) conclude that there is no scientific evidence to support the hypothesis that Lyme spirochetes, should they exist in humans, are the cause of post-Lyme disease syndrome. Another respected scientist, Feder et al. (2009), states that it is an unproven and very improbable assumption that chronic *B.b.* infection can occur in the absence of antibodies against *B.b.*. However, in the March 2010 Journal of Brain, Behavior and Immunity, Chandra et al. (2010), including Dr. Wormser, very recently concluded that some Lyme disease patients report debilitating chronic symptoms of pain, fatigue, and cognitive deficits despite recommended courses of antibiotic treatment. The mechanisms responsible for these symptoms, collectively referred to as post-Lyme disease syndrome (or chronic Lyme disease by other scientists), remain unclear. Anti-neural antibody reactivity was found to be significantly higher in the post Lyme disease syndrome (chronic Lyme disease) group than in the normal healthy groups. The heightened antibody reactivity observed in PLS patients could not be attributed solely to the presence of cross-reactive anti-*Borrelia* antibodies. *They conclude by saying the results provide evidence for the existence of a differential immune system response in Post Lyme Syndrome, offering new clues about the origins and effects of the disease that may prove useful in devising more effective treatment strategies.*

SUMMARY
There is a common refrain from organized health care programs that have balanced costs, and quantitative result reporting, with significant issues in appropriateness and effectiveness of the tests currently being used. This is particularly the case with the diagnosis of chronic Lyme disease for which no effective test currently exists. With the rising growth in the incidence of Lyme disease in North America and Europe, better diagnostics must be found through targeted development in promising areas.

## 5. Treatment of Chronic Lyme Disease

As noted in the first sections of this paper, a significant proportion of individuals recognize they have been bitten by a tick. At lease 20% or more do not, because of the lack of an EM rash and non-specific symptoms that may be attributable to many

infectious or chronic diseases. Chronic Lyme disease patients primarily experience neurological and/or arthritic symptoms in later stages of the disease.

Halperin (2008) summarizes that there have been four randomized blinded trials of prolonged antimicrobial therapy in patients previously treated for Lyme disease. He states that all have demonstrated the absence of any lasting improvement in cognitive function. Given the considerable risk of serious adverse events from prolonged antibiotic treatment it is time to look elsewhere for an effective management strategy to help patients with persistent cognitive symptoms after treatment for Lyme disease.

It seems that colleagues of Halperin, (Wormser and Schwartz (2009)) may have found the basis for alternative strategies, reporting in their recent study that despite resolution of the symptoms of Lyme disease after early antibiotic treatment, a minority of patients have fatigue, musculoskeletal pain, and/or difficulties with concentration or short-term memory. They call this post-Lyme disease syndrome or "chronic Lyme disease." They undertook a review of several recent studies in which *B.b.*-infected animals were treated with antibiotic therapy for *B.b.*. Their findings confirmed the persistence of the bacteria after antibiotic therapy.

While persistent cells are not a sign of drug resistance, overall, the overuse and misuse of antibiotics continue to be a concern in general, which can lead to increasing bacterial resistance and decreasing development of new antibiotics. One study by Fallon et al. (2008) showed improvement in patient systems with long term administration of IV antibiotics. The researchers recommended that the therapy not be recommended due to the risks of infection for patients.

In 2005, Cairns and Godwin performed an extensive meta analysis of 5 studies with a combined total of over 1000 patients. The meta-analysis provided strong evidence that some patients with Lyme disease have fatigue, musculoskeletal pain, and neurocognitive difficulties that may last for years despite antibiotic treatment. Further, the research showed that the higher prevalence of certain neurocognitive symptoms but not others, in the same pattern as reported in the literature, is further confirmation of a Post-Lyme borreliosis syndrome (chronic Lyme disease). The pattern of symptoms appears to be different from that seen in fibromyalgia, depression, and chronic fatigue syndrome.

In response to the overuse and misuse of antibiotics, leading to increasing bacterial resistance and decreasing development of new antibiotics, the Council for Appropriate and Rational Antibiotic Therapy (CARAT) has developed criteria to guide appropriate and accurate antibiotic selection. The criteria, which are aimed at optimizing antibiotic therapy, include evidence-based results, therapeutic benefits, safety, optimal drug for the optimal duration, and cost-effectiveness. Slama et al. (2005) report on the guidelines and state that in choosing appropriate and accurate antibiotic therapy, the clinician should use the criterion of evidence-based results, therapeutic benefits, safety, optimal drug for the optimal duration, and cost-effectiveness. Appropriate aggressive short-course treatment is recommended for ensuring clinical and microbiologic cure, optimal patient adherence, and minimal

generation of antibiotic resistance. Ideally, institution of the 5 CARAT criteria will optimize safe and well-tolerated treatment regimens; curb unnecessary prescribing of antibiotics, decrease treatment costs, and increase adherence. This is important advice, yet it is equally important to note that the treatment of persistent bacterial infections are largely unexplored and involve bacterial persistence, not drug resistance issues.

Feder et al. (2007) criticize that once the diagnosis of chronic Lyme disease is made, patients are commonly treated for months to years with multiple antimicrobial agents, some of which are inactive in vitro against *B.b.*. Steere et al. (2004) also report on the medical implications of prolonged antibiotic therapy. In studies of patients with unsubstantiated Lyme disease, minor side effects were common. Prolonged ceftriaxone therapy sometimes resulted in biliary (gallbladder) complications. Kidney and liver complications have also been identified. Although rare, but extremely serious, a few cases of death have been reported in the literature from antibiotic use. Prolonged use of antibiotics was recently associated with an increased risk of breast cancer. The authors (Velicer et al., 2004) however state that it cannot be determined from this study whether antibiotic use is causally related to breast cancer or other factors. Although further studies are needed, these findings reinforce the need for prudent long-term use of antibiotics.

Stricker (2007) asserts that the complex persistent pathology of *B.b.* allows the spirochete to invade diverse tissues, elude the immune response, and establish long term infection. Commercial testing for Lyme disease is highly specific but relatively insensitive, especially during the later stages of disease. Numerous studies have documented the failure of standard antibiotic therapy in patients with Lyme disease. Previous uncontrolled trials and recent placebo controlled trials suggest that prolonged antibiotic therapy (duration, >4 weeks) may be beneficial for patients with persistent Lyme disease symptoms. Tickborne co-infections may increase the severity and duration of infection with *B.b.*. The author concludes that prolonged antibiotic therapy may be useful and justifiable in patients with persistent symptoms of Lyme disease and co-infection with tickborne agents

Nau et al. (2009), suggest that while different genospecies are found in Europe, the most frequent clinical manifestation of *Borrelia* infection is *erythema migrans* (EM), followed by neuroborreliosis, arthritis, acrodermatitis chronica atrophicans, and lymphocytosis benigna cutis. Diagnosis is made on the basis of the clinical symptoms, and in stages II and III by detection of *Borrelia*-specific antibodies. The cases are treated successfully, and in approximately 95% of neuroborreliosis cases are cured without long-term sequelae. When chronic borreliosis is suspected, other potential causes of the clinical syndrome must be painstakingly excluded. Many believe a variety of nonspecific symptoms and disorders result in overdiagnosis and overtreatment of suspected Lyme disease. Conversely, however, given that many infections do not present with classical symptoms, such as EM, it is possible to assume that the proportion of chronic cases would be as few as 5%?

Optimal treatment remains uncertain for patients with cognitive impairment that persists or returns after standard IV antibiotic therapy for Lyme disease. A major

study was undertaken by Fallon et al. (2008). The use of IV ceftriaxone therapy resulted in short-term cognitive improvement for patients with post treatment Lyme encephalopathy, but relapse in the condition occurred after the antibiotic was discontinued. Treatment strategies that result in sustained cognitive improvement are needed (Fallon et al., 2008). A number of research studies have reported initial success with antibiotic treatment for a short period of time, but then relapsing to the previous state of persistent Lyme disease symptoms.

Barthold el al. (2010) report on a clinical trial of patients with chronic Lyme disease, using a newer tetracycline drug, Tigecycline. The drug was ineffective against persistent *B.b.*, extending previous studies with ceftriaxone. The trial found that antibiotic treatment is unable to clear persisting spirochetes, which remain viable and infectious, but are non-dividing or slowly dividing. Novel strategies are required to target persistence mechanisms as well as the bacterium itself.

## Treatment of Lyme Disease within Biofilm

del Pozo (2007), assert that it is essential to extend our knowledge about the mechanisms involved in biofilm resistance to antimicrobial agents to develop new and effective treatment strategies for biofilm associated infections. New treatment strategies are being directed toward designing substances able to compromise biofilm formation, destabilize established biofilms, and/or target persister cells. del Pozo also states that genes responsible for persistence could also be identified to serve as targets for new drugs. It is suggested that an inhibitor of persistence development could be combined with a conventional antimicrobial to try to eradicate a biofilm. Development of drugs which disable the persister phenotype is likely to provide an effective therapy for biofilm-associated infections.

Bacterial biofilms are highly recalcitrant to antibiotic treatment, which hold serious consequences for therapy of infections that involve biofilms. Many researchers are currently trying to overcome this extreme biofilm antibiotic resistance by developing novel therapies aimed at disrupting biofilms and killing the constituent bacteria. These studies have led to the identification of several molecules that effectively disturb biofilm physiology, often by interrupting bacterial quorum sensing. In this manner, manipulation of innate and induced resistance pathways holds much promise for treatment of biofilm infections (Anderson and O'Toole, 2008).

Bacterial populations produce a small number of dormant persister cells that exhibit multidrug tolerance. The ability of a biofilm to limit the access of the immune system components, and the ability of persisters to sustain an antibiotic attack could then account for the recalcitrance of such infections in vivo and for their relapsing nature. Identification of persister genes opens the way to a rational design of anti-biofilm therapy. Combination of a conventional antibiotic with a compound inhibiting persister formation or maintenance may produce an effective therapeutic. (Lewis, 2008)

Amongst the generally disappointing results from antibiotic therapy for patients with persistent Lyme disease, there are some promising signs. Bernardino et al. (2009) suggest that tetracyclines can moderate inflammatory responses of various etiologies. They hypothesized that tetracyclines, in addition to their antimicrobial function, could exert control over the inflammation elicited by *B.b.*. In in vivo testing of mammalian tissue, both antibiotics significantly reduced the production of tumor necrosis factor-alpha, interleukin (IL)-6, and IL-8 in a dose-dependent manner in all cell types. They state their results suggest that tetracyclines may have a dual therapeutic effect in Lyme disease.

Brorson et al. (2009) also report on in vitro studies in late 2009 demonstrating the efficacy of Tigecycline for inhibition and destruction of the different forms of *B.b.*, especially the round body type. They comment that many unfavorable conditions, including penicillin treatment, do not injure *B.b.* round bodies or syphlis. Tigecycline is more effective than doxycycline in destruction of the *Borrelia* RB forms, and they advocate clinical studies that compare these drugs.

At the October 2009 Lyme disease Conference of ILADS, Dr. Eva Sapi, Associate Professor, Research Scholar, and Coordinator at the Cellular and Molecular Biology Program at the University of New Haven, reported on yet to be published work of her team on "An in vitro evaluation of antibiotic susceptibility of different morphological forms of *B.b.*." The study suggests that exposure of *B.b.* cultures to doxycycline and Plaquenil significantly reduced the spirochete population but increased the number of cystic (persistent) forms of the bacteria. However, similar treatment of and tinidazole led to reduction of cystic forms in the culture. Furthermore, when combinations of most effective concentrations of 5-nitroimidazoles and tetracyclines were tested in vitro, both cystic and spirochete forms of *B.b.*. were significantly eliminated. The study suggests that *B.b.* specific combination therapy for Lyme disease patients might provide treatment options with a better outcome.

Coates and Hu (2006) discuss the targeting of non-multiplying bacteria (such as those that exist in biofilm) and the potential to yield new antibacterials that would shorten the duration of therapy. This could reduce the incidence of adverse effects of treatment, and might reduce the emergence of antibacterial resistance. More information is needed, particularly the mechanisms that lie behind their profound antibacterial tolerance. In the future, it is likely that most antibacterial drug design will be based on existing antibacterial structures, but an increasing number of new molecular antibacterial structures may emerge from screening against multiplying and perhaps non-multiplying bacteria.

SUMMARY

Studies on longer term antibiotic therapy supported by scientists who endorse IDSA guidelines approach, did not show any demonstrable improvement in Lyme patients. Duration of treatment was only three months maximum, and patient selection was problematic. On the ILADS side, scientists undertook a longer term study that went up to six months. This 2008 study did not result in a statistically significant

improvement, although positive outcomes were found. Importantly, symptoms returned in the trial patients after treatment stopped.

The unpublished work of Dr. Eva Sapi at the University of New Haven may offer interesting opportunities for future combination drug therapies that reduce both the spirochete population overall and the number of cystic (persistent) forms of the bacteria.

The result of at least a decade of continued clinical trials between research groups supporting the IDSA philosophy or the ILADS philosophy is constant bickering and criticism of each other's positions. The gulf in ideology, noted earlier in this paper, is enormous and counter productive. Positively, some gains in knowledge for better treatments have occurred over the last five years in particular, including evidence that oral antibiotics can be as effective as IV antibiotics. There is also growing evidence, but no where near conclusive evidence, that combined therapies hold promise in treating persistent infections.

## A novel approach to the understanding of Persistent Infection

Autoimmunity occurs when the immune system recognizes and attacks host tissue. In addition to genetic factors, environmental triggers (in particular viruses, bacteria and other infectious pathogens) are thought to play a major role in the development of autoimmune diseases (Ercolini and Miller, 2009). Built on the evidence of the existence of Biofilm environments in the body, a novel theory is based on the hypothesis that chronic disease with certain characteristics are the result of infections within cells that cannot be killed by the immune system. These infectious bacteria types have developed the ability to remain alive and proliferate undetected inside the cells they infect. These cells include the body's own immune cells, like macrophages, the very cells of the immune system that the body uses to kill invading pathogens (like bacteria). Once inside these cells, they cause the body's own cells to release inflammatory products that can cause pain and fatigue, for example.

Inside a biofilm the bacteria are able to resist the effect of antibiotics. Under certain conditions, they mutate from classical bacteria, losing their cell walls in the process. This thinking is based on research on the human microbiome (all the microorganisms which are found in association with both healthy and diseased humans) which shows that bacteria are far more pervasive within the human body than previously thought, increasing the possibility that autoimmune disease is bacterial in origin (Albert et al., 2009).

When the innate immune system cannot function, the body can no longer keep the bacteria they harbor under control. Mechanisms are needed to ensure that specific immune receptors are kept free of bacterial proteins. This mechanism is purported to improve the body's ability to turn on the innate immune system and produce cells that will kill the bacteria.

## 6. The Possibility of other Chronic Diseases being associated with Chronic Lyme Disease

The literature is plentiful with supposition concerning whether or not Lyme borreolis is associated with other chronic diseases. This is because many of the symptoms of Lyme disease resemble many of the symptoms of other chronic diseases.

With varying levels of evidence, many scientists have weighed in on this question. Miklossy et al. (2009), report on an in vitro study where researchers tried to determine whether an analogous host reaction to that occurring in Alzheimer's disease could be induced by infectious agents. They exposed mammalian glial and neuronal cells in vitro to *B.b.* spirochetes and to an inflammatory bacterial lipopolysaccharide that is found on the membrane of bacteria like *Borrelia*. Their observations indicate that, by exposure to bacteria or to their toxic products, host responses similar in nature to those observed in Alzheimer's disease patients may be induced. This is not proof of a theory, but it does respond to the observations.

Fallon et al. (2010) states Lyme disease, can cause multi-systemic signs and symptoms, including peripheral and central nervous system disease. The brain MRI among patients with Lyme disease may at times be suggestive of a demyelinating disease. As well, in a European study of cardiomyopathy, *B.b.* was detected in 21% of the patients through PCR or electron microscopy. Serology only identified antibodies in 2 cases (vs. 8). Antibiotic therapy was effective. Because serological examination does not provide a tool for diagnosis, biopsies are recommended. (O'Connor et al., 2006)

The pathological hallmarks of Alzheimer's disease (AD) consist of β-amyloid plaques and neurofibrillary tangles in affected brain areas. The processes, which drive this host reaction, are unknown. Resulting from a laboratory experiment, there is evidence that non-communicable chronic diseases can stem from infectious agents. However, existing diagnostic tools may not be sensitive enough to link infectious agents with a chronic disease. Assays may not be able to identify bacteria that can hide within human cells. Testing may occur too long after the exposure, particularly when years of pathology precede diagnosis of the chronic condition, or persistent immune response to an already cleared infectious agent accounts for chronic disease. They suggest that inflammation is a clear potential link between infectious agents and chronic diseases. In concluding that chronic diseases do often stem from infections, people living with chronic disease might benefit from strategies designed to prevent or appropriately treat selected infections (O'Connor et al., 2006). However, the scientific association between common chronic diseases and infectious diseases specifically, is not strong.

## 7. Medico-Legal and Political Issues in Chronic Lyme Disease

With an abundance of caution driven through poor evidence, the medical community has been reticent to prescribe appropriate treatments for patients that may have been suffering from Lyme disease for many years. This matter has come to the attention of governments in the eastern United States where chronic Lyme disease has now become a political and legal issue. The controversy around chronic Lyme disease is highlighted in a 2009 review article by Susan Ronn (2009). Ronn's article, summarizes the conflict that was ultimately created by two sets of clinical practice guidelines that were created by the Infectious Diseases Society of America (IDSA), a mainstream body representing infectious disease specialists and related scientists in the USA, and the International Lyme and Associated Diseases Society (ILADS) representing scientists, researchers, patient's, M.D.'s and the public at large.

The key points in the article (Ronn, 2009) are:

- The Infectious Diseases Society of America (ISDA) has published the mainstream clinical practice guidelines for Lyme disease.
- There are other, conflicting, published, peer reviewed guidelines (by ILADS), and in practice, physicians vehemently disagree as to nearly every aspect of the disease.
- The Connecticut Attorney General initiated an investigation of IDSA, and found conflict of interest among the members of the panel that authored the guidelines, and that not all available scientific knowledge regarding Lyme disease had been considered (including a lack of consideration of information regarding the very existence of Lyme disease).
- A legal agreement between the Attorney General and IDSA requires a new panel to decide whether the guidelines need to be redrafted. Their results were published in April 2010.
- Disagreement surrounds the testing, diagnosis, treatment, and in particular, the existence of chronic Lyme disease.
- The IDSA clinical practice guidelines quickly became the standard of care and influence insurance company reimbursements.
- The IDSA (guidelines panel) did not consider information regarding the existence of chronic Lyme disease and blocked inclusion of scientists and physicians with divergent views.

## 8. The Two Opposing Views in Medicine concerning Chronic Lyme Disease

Despite extensive research into the complex nature of *B.b.*, controversy continues over the diagnosis and treatment of Lyme disease. A report (Johnson and Stricker, 2004) focused on two aspects of the treatment of Lyme disease. First, the medical basis for diagnostic and therapeutic uncertainty in Lyme disease, including variability in clinical presentation, shortcomings in laboratory testing procedures, and design defects in therapeutic trials. Second, the standard of care and legal issues resulting from the clinical uncertainty of Lyme disease diagnosis and

treatment. Medico-legal issues are outlined to support a more rational approach to the diagnosis and treatment of Lyme disease and related tick-borne illnesses.

The IDSA Lyme disease hearing of July 30, 2009 included patient groups, and two professional groups with diametrically opposed views on chronic Lyme disease. It is clear from reports on the hearings that little about chronic Lyme disease is conclusive or noncontroversial. These two positions are summarized with the following examples, formatted below as the views of the IDSA and the ILADS.

**Comparison of Issues and Philosophy of Care of Chronic Lyme Patients**

| | **Conservative Management with only the highest level of evidence for Dx and Treatment**<br><br>**IDSA Associated** | **New knowledge for more appropriate Diagnosis and Treatment, and more compassionate care.**<br>**ILADS Associated** |
|---|---|---|
| What is it? | A rare disorder. | A common disease cluster in a complex clinical and biological presentation. |
| Seeking the Truth | A conservative medical group focused on high levels of evidence based care. | A physician/scientist group searching for new knowledge in the area of persistent infection. |
| Differing Evidence | Using their own levels of evidence and biases to either completely refute or support the existence of chronic Lyme disease. | Using their own levels of evidence and biases to either completely refute or support the existence of chronic Lyme disease. |
| Is chronic Lyme disease Real, or Not | Perception by some that chronic Lyme disease is a psychological disorder (this may be changing) | Chronic Lyme disease as a recognized medical disorder that requires active management. |
| Use of guidelines | Staying the course with existing 2006 guidelines. | Need for a general reassessment of what is known about Dx and Txt of Lyme disease. |
| Perception of Guidelines | Poor levels of evidence used in the development of the IDSA guidelines. | Lack of evidence of the existence of persistence of *B.b.* in the human body. |
| Diagnosis | Support for the adequacy of the current best practice diagnostic process using two-tiered serological testing. | Under diagnosis of Lyme disease due conservative testing criteria and evidence that *B.b.* can be hidden from the immune system |
| Evidentiary basis of treatment decisions | A perceived over emphasis on the determination of objective findings through laboratory testing and treatment evidence. | A perceived over emphasis on the use of subjective symptoms such as pain, fatigue and cognition. |
| Co-Infections | Limited support and recognition for the existence and role of additional bacterial co-infections. | Strong views on the existence and higher morbidity of tick bites containing multiple bacterium. |
| Clinical Trials | Small sample sizes of studies | Case studies of shorter term |

| | | |
|---|---|---|
| | showing no benefit from long-term antibiotic therapy. Methodological difficulties. | success in treatment of chronic bacterial infections. Methodological difficulties. |
| Treatment | No support for long-term antibiotic therapy. | Patient choice relative to benefits and risks. Some evidence. |
| Voluntary use of Guidelines | State that guidelines are voluntary and not intended to supplant individual medical judgment. | Assertions of IDSA members conflict of interest in supporting private interests. Inadequacy of IDSA guidelines for chronic care. |

An International Perspective (Germany)

The controversy around the diagnosis and treatment of Lyme disease is not a North American phenomenon.

The Deutsche Borreliose-Gesellschaft (German Society of Lyme-Borreliosis) (German Society, 2009) made a submission to the State of Connecticut Hearings in July 2009. They stated that the fundamental basis for their objections is that *the implementation of the IDSA guidelines extends beyond the United States and into Europe. Accordingly, their ability to diagnose and treat patients with Lyme disease is being severely restricted by these guidelines, and they believe that the guidelines must be revised* to provide greater flexibility in the diagnosis and treatment of Lyme disease given the poor laboratory test sensitivity, the persistence of the organism despite adherence to IDSA protocols, and the seriousness of this illness.

> *Challenge to Lab Diagnostic Test Requirement*--Page 1090 :"Diagnostic testing performed in laboratories with excellent quality-control procedures is required for confirmation of extracutaneous Lyme disease...."
> *Challenge to Restrictions on the Use of Clinical Judgment*—Pages 1089-90 : "Clinical findings are sufficient for the diagnosis of erythema migrants, but clinical findings alone are not sufficient for diagnosis of extra-cutaneous manifestations of Lyme disease or for diagnosis of HGA or babesiosis."
> *Challenge to Persistence*--Page 1118 The Basis for Treatment Limitations for Early and Late Lyme disease and Post-Lyme Syndrome: "The notion that symptomatic, chronic *B.b.*. infection can exist despite recommended treatment courses of antibiotics (tables 2 and 3) in the absence of objective clinical signs of Disease, is highly implausible

With nearly 100 references in their brief, the Society provided evidence on the insensitivity of diagnostic tests, outdated antibiotic recommendations, such as lack of reference to third generation drugs and appropriate antibiotics that can penetrate the CNS, and the lack of recognition of the existence of chronic Lyme disease.

## 9. Actions taken by the State of Connecticut as a result of its Investigation of the IDSA

The State of Connecticut came to agreement with the IDSA that an independent panel overseen by an appointed ombudsman would undertake a complete review of its guidelines, and that hearings would be held from interested parties to inform the Review Panel. The state also ordered a two step voting procedure, with each step requiring a supermajority vote (6 of 8 panelists). The first vote asks the question whether each of the contested guideline recommendations is "medically/ scientifically justified in light of all of the evidence and information provided." This vote requires a supermajority of the panel (6 of 8) in order for a guideline recommendation to stand. The second vote, also by supermajority, determines whether the guidelines require no changes, partial revision or complete revision. Otherwise, all guidelines with less support will require review and rewriting. Hearings occurred on July 30, 2009, and the panel then began its deliberations on each of the IDSA Guidelines. (Connecticut Attorney General's Office, May 1, 2008 and February 1, 2009)

The results of the Panel review were anticipated before the end of 2009. However, on Monday, February 1, 2010, the Connecticut Attorney General sent a letter to the IDSA expressing concern over improper voting procedures used by the IDSA in the Lyme guidelines review voting process. The Attorney General requested that the IDSA redo the votes on each guideline element to comply with the initial out of court agreement. It appears the IDSA used an improper voting procedure, which contravened their agreement with the State of Connecticut. The IDSA had consented to the voting procedure in the Settlement Agreement.

On April 22, 2010 the Review Panel published its final report. The task of the panel was to determine whether or not the 2006 Lyme Guidelines were based on sound medical/scientific evidence and whether or not these guidelines required change or revision. Any recommendation for update or revision to the 2006 Lyme Guidelines will be conducted by a separate IDSA group.

The Review Panel concluded that all the recommendations were medically/scientifically justified in light of all of the evidence and information provided. This included the validation of the lack of proven effectiveness of prolonged antibiotic treatment in clinical trials. While there is still controversy around the design of these studies, and the nature of treatment benefits other than cure, application of strict criteria leads to the conclusion that treatment for chonic Lyme disease is not scientifically justified.

The Attorney General is currently reviewing the IDSA report to determine whether the IDSA has complied with the Settlement Agreement. The panel had been ordered to redo voting on all the IDSA guidelines, and did with the exception of q contentious guideline relating to the requirement that diagnosis of Lyme disease be confirmed by a positive laboratory blood test. In the initial round of voting, the vote was split meaning under their criteria that there was no consensus.

## 10. US State Hearings and Legislative in the United States

Most states in the Northeastern United States have passed Bills supporting Federal research in Lyme disease and Public Education in their States.

Connecticut
Connecticut Insurance Bill
In 1999 a limited Lyme disease Insurance Bill was passed that insurers must pay for 30 days of IV antibiotic treatment, or 60 days of oral antibiotics. Anyone requiring longer therapy had to obtain a second opinion approval from an infectious disease doctor or other appropriate specialist.

The General Assembly of the State of Connecticut (2009) passed legislation permitting licensed physicians, having determined the presence in a patient of signs or symptoms compatible with acute infection with *B.b.*, or with late stage or persistent or chronic infection with *B.b.* to prescribe long-term antibiotic therapy for the treatment of Lyme disease. In total, five states have passed legislation of this nature.

In a Viewpoints article published by the Journal of the Infectious Diseases Society of America, Jerome Klein criticizes the intrusion of an officer of the state into the business of a professional society. He notes that the action of the Connecticut government should raise concerns for all medical and scientific groups, as it related to the issue of practice recommendations and guidelines. However, the conflicts of interest of the society members were the issue of primary concern.

Pennsylvania
On December 2, 2009 the Pennsylvania House of Representatives, Democratic Policy Committee, conducted a public hearing on the subject of Lyme disease. Actions resulting from this Hearing are currently unknown.

Rhode Island
In 2003 passed the Lyme Insurance Law, which ensures that there is mandatory coverage for diagnostic testing and long-term antibiotics for treatment of Lyme disease when ordered by a treating physician who determines it is medically necessary after a thorough review of the patient's medical history and condition.

California
In 2005 the state of California added to an existing law preventing those doctors treating Lyme disease patients from disciplinary action solely on the basis that the treatment or advice rendered to a patient is alternative or complementary medicine, including the treatment of persistent Lyme disease, if that treatment or a device meets certain criteria such as including prior examination of the patient, informed consent, there is a medical indication for treatment, or it is provided for health or well-being. Also required is physician communication to the patient concerning conventional treatment. The level of education, experience and credentials of the physician related to the alternative or complementary medicine should be provided. In addition, the alternative or complementary medicine should not cause a delay in,

or discourage traditional diagnosis of, a condition of the patient. Finally, there is no risk that the treatment could cause death or serious bodily injury to the patient. For the purposes of this provision the definition of alternative or complementary medicine means "those healthcare methods of diagnosis, treatment, or healing that are not generally used but that provide a reasonable potential for therapeutic gain in a patient's medical condition that is not outweighed by the risk of the healthcare method."

U.S. Congress pending legislation

A Lyme and Tick Borne Disease Prevention, Education, and Research Act of 2007 was tabled at the 110th session of the U.S. Congress. The act was intended to provide for the coordination of all federal programs and activities related to Lyme and other tick borne diseases, including the development of more sensitive definitive diagnostic tests and tools, accurate determinations of the prevalence of Lyme and other tickborne diseases in the United States, access to peer-reviewed information on the subject, public education through the expansion of CDC community-based education programs, creation of physician education programs that include the full spectrum of scientific research, scientific conferences that report on and consider the full spectrum of clinically based knowledge, and establishment of epidemiological research objectives to determine the long-term course of illness for Lyme disease and determination of the effectiveness of different treatment modalities. This bill was strongly endorsed by the high Lyme disease incidence US states. However, while States continue to refer to the need for such legislation, there has been no further action on the proposed Act.

## 11. Provincial Legislation and Regulation in British Columbia

In BC, provincial legislation sets out the framework for the self-regulation of the medical profession. The College of Physicians and Surgeons is the Health Profession's College established under the Health Professions Act. The duty of the College is to serve and protect the public and to exercise its powers and discharge its responsibilities under all enactments in the public interest. The college also has a role outlined in section 16 (2)(d) "to establish, monitor and enforce standards of practice to enhance the quality of practice ... , and in 16(2)(e) to establish and maintain a continuing competency program to promote high practice standards among registrants. In Section 16(2)(k) there is language to "promote and enhance (k,iii) the ability of its registrants to respond and adapt to changes in practice environments, advances in technology and other emerging issues'.

With respect to the controversy surrounding the diagnosis and treatment of Lyme disease, in section 25.2 the board may appoint an investigating committee ... for the purpose of investigating whether a registrant has and applies adequate skill and knowledge to practice medicine,... in section (2) the act provides that an investigating committee may not be appointed ... "solely on the grounds that a registrant practices complementary medicine or uses non-traditional therapies".

Under section 25.4, titled Alternative Medicine, the act states that "the College must not act against a registrant or an applicant for registration solely on the basis that the person practices a therapy that departs from prevailing medical practice unless it can be demonstrated that the therapy poses a greater risk to patient care or safety than does prevailing medical practice.

Under section 18.1 of the Act, titled Inquiry, if the Minister considers it necessary in the public interest, the Minister may appoint a person to inquire into any aspect of the administration or operation of a college, or the state of practice of a profession in British Columbia, a locality, or a facility.

## 12. Policy Considerations and Recommendations

At the PHAC 2006 Lyme disease meeting, recommendations were made for greater knowledge covering three key areas:

i. the organism, *B.b.*, which causes Lyme disease; the disease itself; and the epidemiology of vector ticks.
ii. the genetic diversity and pathogenesis of *B.b.* were identified as current priorities for research.
iii. focus on the disease itself should include improvement in diagnostic technologies and approaches, improvement in understanding the clinical presentations and the burden of disease; and improvement in physician awareness of, and knowledge about, Lyme disease.

The internet and other public accounts are replete with testimonials of those living with chronic Lyme disease and/or related infectious diseases. At the same time, there are physicians who are willing to support the treatment of these patients to give them a chance of a more normal life. There are other physicians, who are also in the medical profession to improve health, but for a number of professional, medico legal, and ethical reasons would nonetheless prefer to prescribe no treatment to these patients. Currently a good deal of the literature on Lyme disease is based on *in vitro* experiments with other small and larger mammals. However, despite the constant rebuttals of the two medical groups, there has been progress in blinded clinical trials that do show some improvement after therapy in patients with established chronic Lyme disease (albeit not within the most strict scientific criteria). It should also be noted that with current diagnostic technologies it is difficult to determine if a patient has an infectious disease or some other kind of chronic ailment, highlighting the importance and relevance of a careful case history and geographic location of the patient.

Despite extensive research into the complex nature of *B.b.*, controversy continues over the diagnosis and treatment of Lyme disease.

- The medical basis for diagnostic and therapeutic decisions is uncertain in Lyme disease, due to the variability in clinical presentations, shortcomings in laboratory testing procedures, and unclear science around the best options for treatment.

- The standard of care and medico-legal issues that have resulted from the clinical uncertainty of Lyme disease diagnosis and treatment has polarized physicians, decision makers and patients.
- The existing diagnostic standards that are based more so on factors such as standardization and ease of quantification, together with lack of advancements in diagnostics, particularly for Chronic Lyme Disease, has created skepticism and concern.
- The divergent treatment standards for Lyme disease diverge between two medical factions, and the process of creating common treatment guidelines for the complex infection is fleeting. The work of del Pozo, 2007 suggests that in the future the genes responsible for chronic infection could be identified to serve as targets for new drugs. An inhibitor of the persistent bacterial infection mechanism could be developed, and could be combined with a conventional antimicrobial to kill the bacteria itself.
- Concerns continue about the rights of physicians to provide treatment advice to their patients, and the patients' rights to receive care. Despite the advice of regulating bodies to the contrary, many physicians are fearful of retribution.
- The literature still speaks to ongoing consideration of the rights and medico legal implications of physicians refusing to provide any treatment when some evidence of benefit exists, or conversely, those who prescribe care to ensure a level of evidence of their treatment decisions and informed consent relative to the possible effects of treatment. There needs to be a more rational approach to the diagnosis and treatment of Lyme disease and related tick-borne illnesses (Johnson and Stricker, 2004)

## No Treatment for Chronic Lyme Disease (Post Lyme Disease Syndrome)

A dominant issue in the chronic Lyme disease debate relates to the lack of a definitive diagnosis of Lyme disease and the prescribing of longer term antibiotics. In such cases the prevailing medical practice could be to provide no treatment at all (sometimes related to a lack of a definitive diagnosis, and related indications for antibiotic therapy). In the face of existing diagnostic knowledge, which is ambiguous at best, it is understandable that a significant proportion of the medical profession is hesitant to move away from the strong and appropriate tradition of only practicing within established standards of care. Hopefully, standards of care and better diagnostics will change with more evidence, but in the face of no treatment at all patients are in the middle of the issue.

Without comparing the burden of any disease relative to another, we see that increasing resources are being consumed on medications for many types of cancers for which long term survival is desirable, but not achievable for all patients. Relapse can occur after cancer treatment as well, just as relapse that has been shown to occur after antibiotic treatment. Significant funds are also spent for cancer drugs that prolong life for short periods of time, often with side effects. On the other hand, antibiotics are inexpensive and generally considered safe (Slama et al., 2005) if carefully chosen, delivered, and medically overseen. There is a limit however to

this statement, especially in the cases of specific long term antibiotic regimes and prolonged intravenous therapy, as well as issues around antibiotic resistance.

With mounting clinical evidence over the last 5 years, more is known about safe and supervised treatments for Lyme disease. These treatments may not at all times meet the highest tests for permanent cure, but in cases where there are no other options, or hope, can provide a temporary level of symptom relief. These treatments do not meet the very highest standards that allow entry into the IDSA guidelines. In the context of patient safety and the potential benefits, therapeutic choices should be identified and treatment decisions can be made between the doctor and patient. Doctors who may wish to be involved in Lyme Disease care could also consider the opportunities to contribute to knowledge in this area through involvement in formal or informal clinical information sharing networks. Such an approach could contribute to the development of better medications in the future, through advancing knowledge within organized community networks (Macaulay, 2006).

BC may find some benefit in learning from the experience of some of the American States (see Connecticut, Rhode Island and California descriptions in Section 10) who enable doctors and their patients to undertake a decision making process to balance the risks and benefits of providing therapy under a formal decision process. In the face of a life of misery and hopelessness (as many patient internet blogs describe) doing nothing should not be the only option.

### Patients who do not respond to antibiotics

It is known that many chronic Lyme patients who have been given extended courses of antibiotics are not cured of Lyme disease. In such cases, patients with Lyme and Lyme like symptoms should be made aware of non-antibiotic therapeutic approaches, such as strengthening the immune system, better sleep, vitamin supplementation, and reduced stress.

### Mechanisms for an organized approach to complex needs

Dr. Jack Chritchley, Chair of the Provincial Health Services Patient Care Quality Review Board, observes in a recent Board report that there does not appear to be currently in place in BC a mechanism readily available for diagnosing and treating patients with the type of complex signs and symptoms associated with Chronic Lyme Disease. The Board suggested that instead of dealing with this matter on the footing of whether chronic Lyme disease ought to be recognized, or not, addressing the symptoms of the complainant and other similarly situated persons who present with these complex symptoms and whose diagnosis and treatment appears to require a multidisciplinary approach, and suggests that guidelines and protocols should be established.

The Review Board comments likely arise from the knowledge that a number of other chronic conditions have symptoms similar to those of Chronic Lyme Disease. This group of symptoms is sometimes referred to as Chronic Lyme and Lyme-like symptoms. This is an important observation that highlights the need for a careful clinical history of the patient, including whether the patient resided or visited in a Lyme endemic area. A complex care clinic may have merit in the future, but as a first step, attention must be given to immediate action on the development of better diagnostic technologies to detect bacterial and other organisms that may be responsible for chronic Lyme and Lyme-like symptoms. Without better diagnostics there will be no certainty, and little basis for rational treatment decisions.

## Recommendations

a. The current state of diagnostic methods for Chronic Lyme and other related infections is inadequate. Standard laboratory diagnostic processes have not changed significantly in years. Urgent attention should be given to ensuring BC patients receive the best possible diagnosis through adoption of more advanced technologies that BC could lead the nation in developing. Work should begin on genetic based and other novel and effective non-serological diagnostic panels. Diagnostic tests for any co-infections found in BC should be a component of the standard diagnostic panel. This is the first and highest priority for BC.

b. The Province of BC should continue national and local efforts to improve the diagnosis and treatment of Acute and Chronic Lyme Disease. Diagnostic tests need to be sufficiently sensitive to recognize the difference between Chronic Lyme Disease and other diseases with Lyme-like symptoms. Any related work should contribute to sharing and advancing knowledge.

c. Chronic Lyme and related diseases present a significant burden on patients who are sometimes given few options or hope. A realistic but flexible approach is needed for each patient with their physician in determining the patient's management of Lyme disease or other related infectious diseases. This should include a range of treatment options that have the potential for symptom relief and quality of life improvement.

d. The Province of BC should satisfy itself that that a doctor may prescribe therapy to a patient that departs from prevailing medical practice, unless it can be demonstrated that the therapy poses a greater risk to patient care or safety than does prevailing medical practice.

e. The Province of BC should satisfy itself that that a doctor will not be investigated solely on the grounds that the registrant practices complementary medicine or uses non-traditional therapies.

f. In the face of global warming and other factors affecting the prevalence of tick borne diseases in BC, improvements are needed in public education and support for active surveillance and precautions among the population in BC. These mechanisms should include more professional education on atypical symptoms and presentations related to B.b. and related co-infections. Continuing medical education and updates for physicians and recreational officials and workers should occur as well.

g. The incidence of Lyme disease is rising in the Northern Hemisphere and, while at a lesser rate, is likely to increase in BC. Studies should be completed to more accurately estimate the burden of Lyme disease in the population, given all the factors that mitigate against comprehensive case finding. This could include diagnostic deficiencies, co-infections, migration of persons with Chronic Lyme disease and related co-infections from other provinces and countries, and proactive case finding of the prevalence and virulence of tick borne and other vectors of infection through proactive sampling.

h. Support for national mandatory reporting of Lyme disease should be provided.

## Appendix I - The Value of Animal Models in Lyme Disease Research

Many studies presented in sections 3, 4 and 5 involve significant basic research activity to understand the mechanisms of Lyme disease and the best ways that the disorder can be diagnosed and treated. A significant amount of this research work involves animals, mainly mice. The findings from four research teams described below support the importance of undertaking animal research, and its relevance to human disease. Animal studies are often an important first step in proving out experimental procedures and treatments before using them in human clinical trials.

Animal research using the mouse model is an ideal model organism for human disease. Not only are mice physiologically similar to humans, but a large genetic reservoir of potential models of human disease has been generated. In addition, a number of recent technological advances have dramatically increased our ability to create mouse models of human disease. In the last decade there has been a dramatic explosion in genome research. In the mouse this has meant the development of detailed linkage maps, the development of markers that can be typed by PCR and that are amenable to automation (Bedell et al., 1997). Bockamp et al. (2002) state that the ability to engineer the mouse genome has profoundly transformed biomedical research. During the last decade, various technologies have become invaluable experimental tools for modeling genetic disorders, assigning functions to genes, evaluating drugs and toxins, and by and large, helping to answer fundamental questions in basic and applied research. In addition, the growing demand for more sophisticated murine (mouse) models has also become increasingly evident. The practice, principles, and progress in the rapidly expanding area of conditional mouse technology is vital knowledge for researchers interested in this area.

Barthold et al. (1993) state that past studies have validated the laboratory mouse as a model of the common form of Lyme borreliosis in humans. The close parallels in clinical course and expanding immune response between mouse and human Lyme borreliosis strongly suggest the possibility that a high percentage of humans, like mice, may be persistently infected with *B.b.* despite apparent disease resolution. This has been demonstrated in a limited number of human patients, and should be further explored. As a specific example, Schaller, et al. (2001) have been able to show that the immunological events that lead to the development of autoimmune disease (in arthritis), in a certain mouse model, may also occur in human disease.

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