Concurrent Lyme Disease and Babesiosis

Evidence for Increased Severity and Duration of Illness

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Objective.—To determine whether patients coinfected with Lyme disease and babesiosis in sites where both diseases are zoonotic experience a greater number of symptoms for a longer period of time than those with either infection alone.

Design.—Community-based, yearly serosurvey and clinic-based cohort study.

Setting.—Island community in Rhode Island and 2 Connecticut medical clinics from 1990 to 1994.

Study Participants.—Long-term residents of the island community and patients seeking treatment at the clinics.

Main Outcome Measures.—Seroreactivity to the agents of Lyme disease and babesiosis and number and duration of symptoms.

Results.—Of 1156 serosurvey subjects, 97 (8.4%) were seroreactive against Lyme disease spirochete antigen, of whom 14 (14%) also were seroreactive against babesial antigen. Of 240 patients diagnosed with Lyme disease, 26 (11%) were coinfected with babesiosis. Coinfected patients experienced fatigue ($P=.002$), headache ($P<.001$), sweats ($P<.001$), chills ($P=.03$), anorexia ($P=.04$), emotional lability ($P=.02$), nausea ($P=.004$), conjunctivitis ($P=.04$), and splenomegaly ($P=.01$) more frequently than those with Lyme disease alone. Thirteen (50%) of 26 coinfected patients were symptomatic for 3 months or longer compared with 7 (4%) of the 184 patients with Lyme disease alone from whom follow-up data were available ($P<.001$). Patients coinfected with Lyme disease experienced more symptoms and a more persistent episode of illness than did those ($n=10$) experiencing babesial infection alone. Circulating spirochetal DNA was detected more than 3 times as often in coinfected patients as in those with Lyme disease alone ($P=.06$).

Conclusions.—Approximately 10% of patients with Lyme disease in southern New England are coinfected with babesiosis in sites where both diseases are zoonotic. The number of symptoms and duration of illness in patients with concurrent Lyme disease and babesiosis are greater than in patients with either infection alone. In areas where both Lyme disease and babesiosis have been reported, the possibility of concomitant babesial infection should be considered when moderate to severe Lyme disease has been diagnosed.

(JAMA. 1996;275:1657-1660)

HUMAN BABESIOSIS (caused by *Babesia microti*) and Lyme disease (caused by *Borrelia burgdorferi*) are emergent in many of the same sites in the northeastern and Great Lakes regions of the United States.[1,2] The risk of human infection by these pathogens, which originally was most intense on particular New England islands and in certain nearby mainland sites, now exists in more densely populated regions.[3-6] Babesial parasites appear to reside solely in red blood cells, and the resulting illness is characterized mainly by fever, chills, sweats, arthralgias, headache, and lassitude.[7] The Lyme disease spirochete, in contrast, resides mainly in fixed tissues and causes a flulike illness, rash, arthritis, and, less frequently, carditis or neuropathy.[2,8] Other infections, including a recently discovered rickettsiosis due to a granulocytic *Ehrlichia* species, also are zoonotic in the same sites.[9,10]

Persons residing or vacationing in these sites are now increasingly affected by these tick-transmitted zoonoses. Because the piroplasm that causes human babesiosis perpetuates in the same vector ticks (*Ixodes dammini*) and same reservoir mice (*Peromyscus leucopus*) as the Lyme disease spirochete, human coinfection may be prevalent.[1] Indeed, antibabesial antibody is present in about a tenth of Connecticut and Rhode Island residents who have experienced Lyme disease and in as many as two thirds of Long Island Lyme disease patients.[6,11]

Despite the naturally close associations between these pathogens, only 3 episodes of coinfection have been described in detail.[12-14] Each of the coinfected patients experienced a particularly severe illness, and 1 died. Disease in experimentally coinfected animals appears to be more severe than in those infected by the agent of Lyme disease alone, perhaps because babesial infection in animals is associated with immune suppression.[15,16] However, to our knowledge, no systematic analysis of the clinical consequences of coinfection has yet been described.

Human disease resulting from coinfection by the agents of human babesiosis and Lyme disease may be more severe than the sum of symptoms accompanying either infection alone. To explore this suggestion, we compared the clinical manifestations of patients infected by 1 of these agents with those of patients infected by both agents in a 5-year prospective, longitudinal study.

METHODS

Epidemiologic Analyses

We obtained epidemiologic information from a community-based serosurvey of 1156 of the 1200 residents of Block Island, Rhode Island, who resided there for at least 1 to 2 months during the Lyme disease transmission season. Residents were invited to participate in a biannual Lyme disease and babesiosis serosurvey through announcements in the local newspaper, over a cable television network,

and through notices posted at the medical center. A sample of blood obtained from residents during the autumn or spring of each year was used for serologic testing. For subjects participating more than once, only the first serologic test result was used to determine seroprevalence. The number of subjects participating each year was as follows: 1990, 550; 1991, 559; 1992, 389; 1993, 391; and 1994, 358. We also obtained epidemiologic information about Lyme disease and babesiosis from a clinic-based cohort study on Block Island.

Case Findings and Clinical Evaluation

We sought all cases of Lyme disease and babesiosis during the months of May through September from 1990 to 1994 in a clinic-based cohort study on Block Island and from 1992 to 1994 in a region of southeastern Connecticut, using the facilities of the Pequot Clinic in Groton, Conn, and the Seaport Clinic in Mystic, Conn. The staff of the sole medical center on Block Island, supplemented by a research nurse, sought to identify all episodes of Lyme disease or babesiosis occurring there throughout the study. At each study site, a study physician obtained a history and performed a physical examination on each patient when an illness resembling Lyme disease or babesiosis was first noted. Information on disease characteristics was standardized by asking all subjects whether they had experienced a particular array of symptoms. A medical history was obtained again within a month of the initial illness and was repeated at least monthly until the patient had become asymptomatic, and the findings from a physical examination were recorded for each subject.

We described the severity of disease by recording the frequency of each of the symptoms and signs that were evident at presentation or were reported by the patient. A sample of blood was obtained during both the acute and the convalescent stage of illness for specific serologic and polymerase chain reaction (PCR) tests for both the spirochetal and the babesial infections. Serologic testing of serial specimens was done in parallel. The PCR testing for B burgdorferi and B microti began in 1992 and 1993, respectively. The procedures followed were approved by the Harvard School of Public Health Human Subjects Committee. Written informed consent was obtained from all subjects.

Diagnosis of acute Lyme disease required either a physician diagnosis of erythema migrans (expanding, ringlike erythematous rash at least 5 cm in diameter) or the presence of symptoms consistent with Lyme disease accompanied by either a 4-fold elevation in anti–B burgdorferi antibody or a positive B burgdorferi-

Table 1.—Characteristics of Subjects Infected by the Agents of Lyme Disease (*Borrelia* Organisms) Alone, Human Babesiosis (*Babesia* Organisms) Alone, or Both Pathogens Simultaneously

Characteristic	Borrelia	Babesia	Both
No. of subjects*	214 (183)	10 (10)	26 (23)
Age, median (range), y	43 (2-86)	42 (6-73)	42 (6-86)
Male, No. (%)	109 (51)	6 (60)	16 (62)
Days from onset until antibiotic treatment, median (range)	5 (1-49)	NA	5 (1-60)

*Number refers to the total patient group from Block Island, Rhode Island, and Connecticut, with number of subjects in parentheses indicating patients from Block Island alone. Age, sex, and antibiotic data refer to the total patient group. NA indicates antibiotic not administered.

specific PCR test from blood. Diagnosis of acute babesiosis required presence of symptoms consistent with babesiosis and laboratory evidence of B microti infection, including microscopic identification of the pathogen in a Giemsa-stained thin blood smear, a positive B microti–specific PCR test from blood, or a 4-fold elevation in anti–B microti antibody. Evidence of acute babesiosis in 2 coinfected patients consisted of a convalescent serum sample that was positive for B microti by indirect immunofluorescence assay for IgG and IgM antibody.

Laboratory Assays

Preparation of Blood Smears.—Piroplasms were diagnosed microscopically in Giemsa-stained films of blood anticoagulated with ethylenediaminetetraacetic acid.[17] At least 100 fields (400×) were examined before declaring the sample free of piroplasms.

Indirect Immunofluorescence Assay for Antibabesial Antibody.—Babesial infection was diagnosed serologically by an indirect immunofluorescence assay as previously described.[18] Slides were examined at 630× magnification under epifluorescence. A positive specimen was defined as reacting at a dilution of 1:32 or greater.

Enzyme-Linked Immunosorbent Assay for Antispirochetal Antibody.—Serologic evidence of exposure to the Lyme disease spirochete was detected by enzyme-linked immunosorbent assay.[4] A reactive serum was defined as one reacting at a dilution equal to or greater than 1:320.

Western Blot Assay for Antispirochetal Antibody.—All borderline or reactive serum samples were further characterized by immunoblot.[19] Specimens were considered positive if the IgG immunoblot contained 5 or more of the 10 most common B burgdorferi-specific bands.[19]

PCR Assay for Spirochete and Piroplasm DNA.—Whole blood samples were analyzed and processed by personnel blinded to the clinical status of the donor.[20] With each DNA extraction, blood from coinfected subjects was processed simultaneously with blood from subjects with Lyme disease or babesiosis. A 294–base pair portion of the B burgdorferi OspA gene was targeted for amplification

using a recently described PCR protocol.[20] A 238–base pair portion of the B microti nuclear small subunit ribosomal gene was targeted for amplification using a PCR protocol described previously, except that the volume of blood analyzed was 0.5 mL rather than 0.2 mL.[21]

Data Analysis

A χ^2 test (or the Fisher exact test when expected cell frequencies were <5) was used to examine differences in the occurrence of symptoms between groups of subjects. Two-way comparisons were tested between the coinfection group and each single disease group independently. Group mean differences in the total number of symptoms per patient were evaluated with the Student t test (unpaired) following tests for homogeneity of group variances. Confidence intervals for proportions were calculated using standard formulas. All P values reported are 2-tailed.

RESULTS

Epidemiologic Features of Spirochetal and Babesial Infections

The frequency of exposure of residents to the agents of Lyme disease or human babesiosis at the Block Island study site was estimated in a community-based serosurvey. Of 1156 subjects surveyed (mean [SD] age, 43 [20] years; 605 women and 551 men), 83 (7%) had antibody to the spirochete, 56 (5%) to the piroplasm, and 14 (1%) to both pathogens. Of the 97 participants with antibody to the spirochete, 14 (14%) (95% confidence interval [CI], 12%-16%) also had antibody to the piroplasm.

Subjects in each diagnostic category in our clinic-based cohort were similar with respect to age and sex (Table 1), and virtually all became ill between May and September. Therapy was similar in the coinfected and Lyme disease groups: amoxicillin or doxycycline was administered for a minimum of 10 days to 94% of subjects in the Lyme disease group and 92% of subjects in the coinfected group. The duration of illness before treatment began did not differ between categories of diagnosis. No therapy for babesiosis was administered to any of the subjects; diagnosis generally was delayed. Al-

Table 2.—Clinical Manifestations and Duration of Illness in Symptomatic Patients From Block Island, Rhode Island, and Connecticut Infected by the Agents of Lyme Disease (*Borrelia* Organisms) Alone, Human Babesiosis (*Babesia* Organisms) Alone, or Both Pathogens*

Manifestation of Disease	Individual Diagnosis, No. (%)			P	
	Borrelia (n=214)	*Babesia* (n=10)	Both (n=26)	Both vs *Borrelia*	Both vs *Babesia*
Individual					
Fatigue	102 (48)	6 (60)	21 (81)	.002	.23
Headache	85 (40)	7 (70)	20 (77)	<.001	.69
Erythema migrans*	186 (88)	0 (0)	16 (62)	.002	<.001
Fever	89 (42)	8 (80)	15 (58)	.14	.27
Sweats	22 (10)	2 (20)	12 (46)	<.001	.26
Chills	46 (22)	4 (40)	11 (42)	.03	>.99
Myalgia	63 (29)	2 (20)	10 (38)	.37	.44
Anorexia	28 (13)	1 (10)	8 (31)	.04	.39
Arthralgia	78 (36)	4 (40)	7 (27)	.39	.45
Emotional lability	15 (7)	0 (0)	6 (23)	.02	.16
Nausea	10 (5)	1 (10)	6 (23)	.004	.65
Neck stiffness	45 (21)	3 (30)	6 (23)	.80	.69
Multiple erythema migrans	31 (14)	0 (0)	5 (19)	.56	>.99
Cough	20 (9)	2 (20)	4 (15)	.31	>.99
Sore throat	19 (9)	2 (20)	4 (15)	.29	>.99
Conjunctivitis	5 (2)	0 (0)	3 (12)	.04	.55
Splenomegaly	0 (0)	1 (10)	2 (8)	.01	>.99
Vomiting	9 (4)	0 (0)	2 (8)	.34	>.99
Joint swelling	6 (3)	0 (0)	1 (4)	.56	>.99
Overall					
Symptoms and signs per patient, mean (SD)	3.9 (3)	4.4 (2)	5.9 (3)	<.001	.16
Duration >1 mo	42/184 (23)	4/10 (40)	20/26 (77)	<.001	.05
Duration >3 mo	7/184 (4)	2/10 (20)	13/26 (50)	<.001	.14

*Erythema migrans rash was more frequent in patients with Lyme disease than in coinfected patients (P=.002) or in those with babesiosis alone (P<.001).

though 3 of the *Babesia*-infected subjects had a history of preexisting malignancy, no other evidence of immunosuppression was noted in any of the other subjects in the study.

The frequency of clinical Lyme disease and human babesiosis on Block Island was estimated from the clinic-based cohort at the medical center. Lyme disease was diagnosed in 206 residents of the island during the 5-year study period, of whom 23 (11%) (95% CI, 7%-15%) had been infected simultaneously with the piroplasm. Based on the coinfection rates derived from the serosurvey and clinic-based cohort study, we conclude that Block Island residents are frequently exposed to *B burgdorferi* and that about 1 in 10 patients with Lyme disease also become infected by the agent of human babesiosis.

Number of Symptoms and Signs of Disease

We compared the number of symptoms and signs of disease in subjects from the clinic-based cohort studies on Block Island and in Connecticut who sought medical care after experiencing Lyme disease, babesiosis, or both infections simultaneously. Subjects with evidence of both infections reported a greater array of symptoms than those infected by the spi-

rochete or piroplasm alone (Table 2). Fatigue (P=.002), headache (P<.001), nausea (P=.004), sweats (P<.001), anorexia (P=.04), chills (P=.03), emotional lability (P=.02), conjunctivitis (P=.04), and splenomegaly (P=.01) were significantly more frequent in the coinfected subjects than in those with Lyme disease alone. Coinfected subjects experienced more than 1.5 times as many different symptoms and signs of disease as subjects with Lyme disease alone (P<.001) (Table 2). Coinfected subjects have a more diverse episode of illness than subjects infected by only 1 of these pathogens.

Duration of Disease

Thirteen (50%) of the 26 coinfected subjects had at least 1 symptom that lasted for 3 months or more compared with only 7 (4%) of the 184 patients experiencing Lyme disease alone from whom follow-up data were available (P<.001) (Table 2). Fatigue was the most common symptom that persisted for more than 1 month. None of the coinfected subjects reported persistent fatigue prior to the onset of their illness. All coinfected subjects experiencing fatigue reported that at some time during their illness at least 25% of their daily activities were limited. More coinfected subjects (19 [73%] of 26) expe-

rienced fatigue for more than 1 month than did Lyme disease subjects (42 [23%] of 184; P<.001) or babesiosis subjects (2 [20%] of 10; P=.005). Nine (35%) of the coinfected subjects experienced fatigue for more than 6 months in comparison to 3% of patients with Lyme disease (P<.001) and 20% of patients with babesiosis (P=.69). Neither age nor sex accounted for the differences in fatigue among patients with coinfection, Lyme disease, or babesiosis. In contrast, arthritic, cardiac, and neurologic symptoms of more than 2 weeks' duration were experienced by as many subjects diagnosed with Lyme disease alone as by those diagnosed with both infections. Persistent and debilitating fatigue characterized coinfection.

Duration of Spirochetemia or Parasitemia

We also compared the persistence of *B burgdorferi* DNA in coinfected subjects with that in subjects apparently infected by only 1 of the pathogens. Blood was obtained for PCR testing after administration of antibiotic but at least 1 month before the onset of the following tick transmission season. The number of days between onset of illness and PCR testing was similar among the 3 groups. Spirochete-specific DNA was detected by PCR in 3 (27%) of 11 of the coinfected subjects, compared with 5 (6%) of 81 of those with Lyme disease alone (P=.06) and none of 7 patients with babesiosis alone. Spirochete-specific DNA was detected later in the course of illness in coinfected patients (mean, 91 days; range, 1-265 days) than in patients with Lyme disease alone (mean, 12 days; range, 1-49 days). *Babesia*-specific DNA was detected by PCR testing in almost equal proportion in patients with coinfection (9 of 18) and babesiosis alone (3 of 7) and in none of 98 patients with Lyme disease alone. Spirochete DNA was evident more often and remained in the circulation longer in coinfected subjects than in those experiencing either infection alone.

Silent Infection

We compared the number of subjects from Block Island participating in the serosurvey sample each year who had a 4-fold rise in antibody titer and reported no relevant illness during the previous year (as evidence of silent infection) with the number of Block Island subjects identified by case finding with symptomatic illness. Of subjects infected by both pathogens from 1991 to 1994, 1 (5%) of 19 was asymptomatic compared with 28 (16%) of 175 of those infected by the spirochete alone (P=.23) and 30 (79%) of 38 of those infected by the piroplasm alone (P<.001). Coinfected subjects were more likely to manifest clinical symp-

toms than were those infected by either pathogen alone.

COMMENT

The severity of Lyme disease varies greatly, ranging from a mild flulike illness in some patients to a disseminated disease that may include fever, multiple erythema migrans lesions, atrophy of the skin, and arthritic, cardiac, or neurologic sequelae. Variations inherent to the spirochete population may explain part of this diversity.[22] Host properties, including variation in the D-locus alleles of the major histocompatibility complex (HLA-DR type), may contribute further to the unusually variable expression of disease.[2] Our observations indicate that coinfection by at least 1 other tick-borne pathogen also may contribute to the biologic variation of Lyme disease.

The symptoms that tend to be exacerbated in coinfected patients generally are those that characterize both infections. In addition, coinfected patients experience particularly severe episodes of intense fatigue that significantly interfere with their daily activities and are more prolonged than in patients with either infection alone, generally persisting for more than 1 month. The presence of both agents tends to synergize a variety of other symptoms as well, especially headache, chills, and sweats. Although not all patients experience severe illness, coinfection generally results in more intense acute illness and a more prolonged convalescence than accompany either infection alone.

The mechanism by which human babesiosis and Lyme disease may each potentiate the severity of the other has not been defined. The inflammatory response to each organism may be additive or synergistic. Babesial infection may impair human host defense mechanisms, as it does in cattle and mice.[16,23] Such an immunosuppressive effect may explain our finding that spirochetal DNA is present more frequently and remains in circulation longer in coinfected subjects than in subjects infected by the spirochete alone. This finding implies the presence of living spirochetes, because spirochete DNA in blood is amplifiable only when these pathogens remain viable.[24] It also suggests a synergistic inflammatory response to both a parasitemia and an increased spirochetemia. In addition, babesial infection enhances Lyme disease myocarditis in mice,[15] which suggests that coinfection might also synergize spirochete-induced lesions in human joints, heart, and nerves. No such enhanced pathogenesis was observed in our patient population, perhaps because of the promptness of antibiotic therapy for Lyme disease in most cases.

Physicians caring for patients with moderate to severe Lyme disease should consider obtaining diagnostic tests for babesiosis and possibly other tick-borne pathogens in regions where these diseases are zoonotic, especially in patients experiencing episodes of "atypical Lyme disease" or patients in whom the response to antibiotic treatment is delayed or absent. Specialized diagnostic methods are required to detect babesial and borrelial infection and to distinguish infections that are transmitted simultaneously or sequentially. We found no evidence of spirochetal and babesial serologic cross-reactivity. As is common in the case of babesial infection, parasites frequently cannot be seen in blood films. Improved rapid diagnostic techniques for babesiosis are needed so that patients with Lyme disease who are also infected with B microti may be given appropriate treatment, including antibabesial therapy consisting of clindamycin and quinine.

Because the agents of Lyme disease and human babesiosis often are concurrently enzootic, human coinfection is relatively common. Serologic evidence of exposure to B microti is present in approximately 10% of people with Lyme disease in southern New England.[4,6] Coinfection is even more frequent in an enzootic site in New York.[11] The geographic range of babesiosis and of Lyme disease, both emerging infections, has yet to be defined, but enzootic sites have been identified in the northern Midwest, the western United States, and in Europe and Asia.[1,17] Recognition of the apparent synergistic effects of babesiosis and Lyme disease should help intensify efforts to identify regions in which babesiosis and other potentially cotransmissible diseases, such as human granulocytic ehrlichiosis, are enzootic.

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