Repeat or persistent Lyme disease: persistence, recrudescence or reinfection with *Borrelia Burgdorferi*?

Eugene D. Shapiro

Address: Departments of Pediatrics, Epidemiology of Microbial Diseases and Investigative Medicine, Yale University Schools of Medicine and of Public Health and Graduate School of Arts and Sciences, New Haven, CT, USA

Email: Eugene.Shapiro@Yale.edu

F1000Prime Reports 2015, 7:11 (doi:10.12703/P7-11)

All F1000Prime Reports articles are distributed under the terms of the Creative Commons Attribution-Non Commercial License (http://creativecommons.org/licenses/by-nc/3.0/legalcode), which permits non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

The electronic version of this article is the complete one and can be found at: http://f1000.com/prime/reports/m/7/11

Abstract

Whether or not *Borrelia burgdorferi* can persist after conventional treatment with antimicrobials has been a very controversial issue. Two recent studies took different approaches to try to answer this question. In one, investigators showed that, in each of 22 instances in 17 patients with two consecutive episodes of culture-proved erythema migrans, the strains of *B. burgdorferi* were different based on their genotypes. This indicated that the repeat episodes were due to new infections rather than recrudescence of the original infection. In another study, in which persistence of *B. burgdorferi* was assessed by using xenodiagnosis, no viable *B. burgdorferi* were cultured from ticks fed on any of the patients. There continues to be no evidence that viable *B. burgdorferi* persist in humans after conventional treatment with antimicrobials.

Introduction

One of the most contentious issues related to Lyme disease has been whether infection with *B. burgdorferi* can be easily eradicated by conventional antimicrobial treatment or whether it is a persistent, recurrent and difficult to eradicate infection. Post-treatment Lyme disease syndrome refers to patients with persistent, non-specific symptoms, such as arthralgia, fatigue or perceived cognitive impairment ≥6 months after completion of treatment for Lyme disease. Many of those who believe in a condition termed “chronic Lyme disease” argue that documenting that the bacteria can survive a standard course of antimicrobial treatment will prove that chronic Lyme disease and post-treatment Lyme disease syndrome are indeed a consequence of persistent infection with *B. burgdorferi*. This belief persists, despite a mountain of scientific evidence that chronic Lyme disease, a label without a case definition that is applied to patients with non-specific symptoms more properly designated “medically unexplained symptoms”, does not exist [1–3].

Several studies in which investigators have claimed to have documented persistence of *B. burgdorferi* in humans, despite treatment with antimicrobials, have not been reproduced or have been shown to be due to laboratory contaminants [4–8]. Some studies in which models of the infection in animals (usually in either mice or non-human primates) have been performed have lent support to the argument that *B. burgdorferi* can persist in tissues despite antimicrobial treatment [9]. However, there are numerous problems with the animal models that have led many to question either the reliability of the results of these studies or the generalizability of the results to disease in humans [10,11]. Among the many problems with animal models is the difficulty of mimicking the route and infectious dose of *B. burgdorferi* in human infection, differences in the pharmacokinetic and pharmacodynamic parameters of antimicrobial treatment in the animals vs. humans, and differences in immune responses of different animal species (for example, mice are reservoirs for *B. burgdorferi*).
in nature). In addition, even in the rare instances in which
*B. burgdorferi* has been identified in treated animals, it has
not been shown to be viable (that is, able to replicate in
culture media). Moreover, whether it is producing symp-
tomatic disease in these animals is speculative. Several
recent studies have shed additional light on this topic
and have added to the evidence that viable *B. burgdorferi*
do not persist after antimicrobial treatment of humans.

**Recent studies**

**Repeat episodes of erythema migrans**

It is well recognized that subsequent episodes of
erythema migrans are not unusual in patients with an
initial episode who receive prompt antimicrobial treat-
ment. If *B. burgdorferi* persist despite antimicrobial
treatment, it would make sense that subsequent episodes
of erythema migrans might be due to the originally infect-
ing strain of the organism. In a study to assess whether
such episodes were new infections or recurrences, strains
of *B. burgdorferi* from 17 patients with two or more
episodes of culture-proved erythema migrans had 22
paired episodes with organisms available for analysis
(first and second episodes, and, for those with additional
episodes, third and fourth episodes) [12]. Strains were
compared by amplifying their DNA with polymerase
chain reaction (PCR) assay of each strain, followed by
genotyping of outer surface protein C of each strain. In
all instances, the strains in each of the paired episodes
were different, indicating that each of the subsequent
episodes of erythema migrans was due to a new infection
(presumably transmitted from a new tick bite) rather
than recrudescence of a persistent infection [12,13].
Moreover, investigators used statistical simulation based
on these data to conclude that patients treated for early
Lyme disease develop protective immunity that is strain
specific and that lasts for at least 6 years [14].

**Xenodiagnosis for B. Burgdorferi**

Proponents of the existence of “chronic Lyme disease”
postulate that the organism often persists despite conven-
tional treatment with an antimicrobial, and that very
prolonged treatment with antimicrobials is necessary to
eliminate the organism. However, other than a few
isolated claims (that could not be replicated) of recovery
of *B. burgdorferi* from patients with chronic symptoms,
there is no evidence that viable organisms persist as a cause
of chronic symptoms in humans, despite conventional
treatment with antimicrobials [4–8]. In infected patients,
*B. burgdorferi* is present only in low concentrations in
blood and spinal fluid and can be difficult to culture [15].

Xenodiagnosis is a method of documenting the presence
of a microorganism in tissue by allowing a vector to feed
on potentially infected tissue and then examining the
vector for the presence of the microorganism it may have
ingested. In the past, it was used extensively as a method
of diagnosing Chagas disease [16]. Some say it may be
the most sensitive test in mammals because of evolu-
tionary adaptations of the vector and the microorganism.
For example, there is evidence that a chemottractant is
secreted by *Ixodes scapularis* that can enhance migration
of *B. burgdorferi* to its mouthparts while feeding on a
host [17]. *I. scapularis* ticks have been used successfully
for xenodiagnosis in studies with animals [18]. The first
use of xenodiagnosis to try to detect *B. burgdorferi* in
humans was reported recently [16].

In this study [16], 36 human subjects (median age:
55 years) had 25–30 laboratory-raised, pathogen-free,
larval *I. scapularis* ticks placed on them under a retention
dressing and were then collected after feeding for 3 to
7 days. The 36 subjects included 10 patients who had
been treated for Lyme disease and still had high
concentrations of antibodies against the C6 peptide of
the membrane protein of *B. burgdorferi*, 10 patients
treated for Lyme disease who had post-treatment Lyme
disease syndrome with continuing symptoms severe
enough to impair their normal activities, 5 patients who
recently completed antibiotic therapy for erythema
migrans, 1 patient early in the course of antibiotic
therapy for erythema migrans, and 10 healthy controls.
The patients recently treated for erythema migrans were
meant potentially to be positive controls, since it was felt
to be unethical to delay treatment of infected patients,
but it was thought that these patients had the best chance
of testing positive by xenodiagnosis. Biopsies of skin at
the site where ticks fed and homogenates of the ticks
were cultured for *B. burgdorferi*. Ticks were also tested for
DNA of *B. burgdorferi* by PCR assay and/or by isothermal
amplification followed by PCR and electrospray ioniza-
tion mass spectroscopy. In addition, attempts were made
to infect immunodeficient mice by having the poten-
tially infected ticks feed on them or by inoculation of
homogenates of fed ticks into the mice.

No viable organisms were recovered from ticks or from
skin biopsies from any of the patients. DNA of *B. burgdorferi*
was identified from the ticks of 2 patients. One was the
patient who was early on in the course of antibiotic
treatment for erythema migrans. The other was one of
the patients with post-treatment Lyme disease syndrome.

What do these results mean? Most would say that only a
positive culture (recovery of viable organisms) would
constitute positive xenodiagnosis. It is well recognized that
fragments of DNA of *B. burgdorferi* can persist for a very
long period after successful antibiotic treatment and
killing of viable organisms [19]. A positive result of a
highly sensitive PCR assay of a tick from a patient with post-treatment Lyme disease syndrome certainly is provocative. Can persistence of antigens of organisms killed after antimicrobial treatment lead to prolonged non-specific symptoms? Perhaps persistent (non-viable) fragments of organisms can persist and provoke inflammation that leads to symptoms [20,21]. This is a single instance and needs to be replicated. Much more data are needed before any conclusions can be drawn. Certainly the results of this study do not provide evidence for the suppositions that viable B. burgdorferi persist after conventional antimicrobial treatment, or suggest that additional clinical trials of prolonged antimicrobial treatment of patients with only non-specific symptoms after Lyme disease are warranted [22].

Abbreviation
PCR, polymerase chain reaction.

Disclosures
The author receives royalties from UptoDate.

References
1. Feder HM, Johnson JB, O’Connell S, Shapiro ED, Steere AC, Wormser GP, Ad Hoc International Lyme Disease Group, Agger WA, Artssob H, Auwaerter P, Dumler JS, Bakken JS, Bockenstedt LK, Green J, Dattwyler RJ, Munoz J, Nadelman RB, Schwartz I, Draper T, McSweegan E, Halperin JJ, Klempner MS, Krause PJ, Mead P, Marshel M, Porwancher R, Radolf JD, Smith RP, Sood S, Weinstein A, Wong Sj et al.: A critical appraisal of “chronic Lyme disease”. N Engl J Med 2007, 357:1422-30.

2. Lantos PM: Chronic Lyme disease: the controversies and the science. Expert Rev Anti Infect Ther 2011, 9:787-97.

3. Hatcher S, Arroll B: Assessment and management of medically unexplained symptoms. BMJ 2008, 336:1124-8.

4. Phillips SE, Mattman LH, Hulinska D, Moayd H: A proposal for the reliable culture of Borrelia burgdorferi from patients with chronic Lyme disease, even from those previously aggressively treated. Infection 1998, 26:364-7.

5. Sapi E, Pabbati N, Datar A, Davies EM, Rattelle A, Kuo BA: Improved culture conditions for the growth and detection of Borrelia burgdorferi from human serum. Int J Med Sci 2013, 10:362-76.

6. Marques AR, Stock F, Gill V: Evaluation of a new culture medium for Borrelia burgdorferi. J Clin Microbiol 2000, 38:4239-41.

7. Johnson, Barbara J B, Pilgrim MA, Russell TM: Assessment of new culture method for detection of Borrelia species from serum of Lyme disease patients. J Clin Microbiol 2014, 52:721-4.

8. Klempner MS, Schmid CH, Hu L, Steere AC, Johnson G, McC cloud B, Noring R, Weinstein A: Intralaboratory reliability of serologic and urine testing for Lyme disease. Am J Med 2001, 110:217-9.

9. Embers ME, Barthold SW, Borda JT, Bowers L, Doyle L, Hodzic E, Jacobs MB, Hasenkampf NR, Martin DS, Narasimhan S, Phillippe-Falkenstein KM, Purcell JE, Raterrere MS, Philipp HT: Persistence of Borrelia burgdorferi in rhesus macaques following antibiotic treatment of disseminated infection. PLoS ONE 2012, 7:e29914.

10. Wormser GP, Schwartz I: Antibiotic treatment of animals infected with Borrelia burgdorferi. Clin Microbiol Rev 2009, 22:387-95.

11. Lantos PM, Auwaerter PG, Wormser GP: A systematic review of Borrelia burgdorferi morphologic variants does not support a role in chronic Lyme disease. Clin Infect Dis 2014, 58:663-71.

12. Nadelman RB, Hanincová K, Mulkerjee P, Liveris D, Nowakowski J, McKenna D, Brisson D, Cooper D, Bitzer S, Madison G, Holmgren D, Schwartz I, Wormser GP: Differentiation of reinfection from relapse in recurrent Lyme disease. N Engl J Med 2012, 367:1883-90.

13. Steere AC: Reinvestigation versus relapse in Lyme disease. N Engl J Med 2002, 346:1070-2.

14. Khatchikian CE, Nadelman RB, Nowakowski J, Schwartz I, Wormser GP, Brisson D: Evidence for strain-specific immunity in patients treated for early Lyme disease. Infect Immun 2014, 82:408-13.

15. Wormser GP, Bitzer S, Cooper D, Nowakowski J, Nadelman RB, Pavia C: Yield of large-volume blood cultures in patients with early Lyme disease. J Infect Dis 2001, 184:1070-2.

16. Marques A, Telford SR, Turk S, Chung E, Williams C, Dardick K, Krause P, Brandenburg C, Crowder CD, Carolan HE, Eshoo MW, Shaw PA, Hu LT: Xenodiagnosis to detect Borrelia burgdorferi infection: a first-in-human study. Clin Infect Dis 2014, 58:937-45.

17. Shih C, Cho L, Yu C: Chemotactic migration of the Lyme disease spirochete (Borrelia burgdorferi) to salivary gland extracts of vector ticks. Am J Trop Med Hyg 2002, 66:616-21.

18. Hua CM, Cheminade Y, Perret J, Weynants V, Lobet Y, Gern L: Early detection of Borrelia burgdorferi sensu lato infection in Balb/c mice by co-feeding Ixodes ricinus ticks. Int J Med Microbiol 2003, 293:421-6.

19. Straubinger RK: PCR-Based quantification of Borrelia burgdorferi organisms in canine tissues over a 500-Day postinfection period. J Clin Microbiol 2000, 38:2191-9.

20. Bockenstedt LK, Radolf JD: Xenodiagnosis for posttreatment Lyme disease syndrome: resolving the conundrum or adding to it? Clin Infect Dis 2014, 58:946-8.

21. Bockenstedt LK, Gonzalez DG, Haberman AM, Belperron AA: Spirochete antigens persist near cartilage after murine Lyme borreliosis therapy. J Clin Invest 2012, 122:2652-60.

22. Wormser GP, Nadelman RB, Schwartz I: The amber theory of Lyme arthritis: initial description and clinical implications. Clin Rheumatol 2012, 31:989-94.