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Perspective

What Could Be a Primary Cause of Multiple Sclerosis: Is It an Autoimmunity Triggered by Chronic Protozoan Infection?

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Received 4 January 2013; Revised 22 March 2013; Accepted 13 April 2013

Abstract The generally accepted paradigm of multiple sclerosis is the autoimmune one; still, a body of evidence suggests that this disease may actually be triggered by an infectious factor. In this paper, it is hypothesized that multiple sclerosis may actually be a rare complication of a protozoan infection, which is usually asymptomatic but in some susceptible individuals is accompanied by autoimmune attack against the nervous tissue. If multiple sclerosis were actually caused by such an infection, then a microorganism responsible should exhibit several properties: it (i) is transmitted by an arthropod vector; (ii) is characterized by specific metabolism of the lipids; (iii) should be dependent on iron; (iv) should be associated with an autoimmune response of the host; and (v) should be susceptible to pharmaceutical agents used for the treatment of multiple sclerosis but not to the degree that would allow its eradication. A combination of these properties suggests a role of a blood-residing protozoan.

Keywords epidemiology; multiple sclerosis; protozoan infections

1. Introduction

For the time being, multiple sclerosis (MS)—a chronic disease of the central nervous system—is a pathology of unknown background. The generally accepted paradigm of MS is the autoimmune one, which means that the disease is caused by autoimmune attack against nervous tissue primarily due to myelin-reactive T cells. It is suspected that a combination of genetic and environmental factors may be responsible. However, it remains unclear how such an autoimmune reaction could be initiated. Also, a number of uncertainties cast doubt on traditional autoimmune models of MS. Although the relative efficacy of immunomodulatory drugs used for the treatment of MS supports the idea that the immune system plays a significant role in the pathophysiology of the disease, several findings from human studies do not fit into the autoimmune paradigm. These experiments suggest that inflammatory processes responsible for formation of MS plaques are not necessarily primarily autoimmune [12,15,17,18,28,33,39,42,59,87,99,100,104,121,122].

2. Is MS actually caused by an infectious factor?

A number of papers suggest that MS may actually be triggered by an infectious factor [53,81,109]. Yet, in spite of several decades of intensive research, no definitive evidence has emerged to identify a single microorganism as a cause of the disease. In this review, I will examine which microorganism might theoretically be responsible. I will demonstrate that it is likely that MS is a rare complication of a protozoan infection, which is usually asymptomatic but in some susceptible individuals is accompanied by an autoimmune reaction against nervous tissue antigens. I will examine such a hypothetical scenario, first summarizing the results of epidemiological studies that suggest a role of infection in the pathogenesis of MS. Then, the findings of histological and immunological studies on MS will be discussed, followed by a review of pharmacological trials that support a potential role for a protozoan. Finally, I will focus on diagnostic strategies that may be successful in identifying such a microbe.

3. Evidence coming from epidemiological studies: an environmental and not genetic background of MS

The relatively low concordance rate of identical twins indicates a contribution of nongenetic factors to MS etiology [72,81]. Currently, it is also well known that MS is characterized by uneven geographic distribution. It is widely accepted that people living at higher geographic latitudes, with reduced solar exposure, and consequently manifesting vitamin D deficiency are at higher risk of developing MS. However, even if it is taught that the risk of MS is related to the geographic latitude, with a higher risk in countries located far away from the equator and a lower risk in the tropics, the actual geographic distribution of the disease is a bit different [50]. Although the distribution of MS is following the latitude paradigm, the high-risk
areas comprise northern Europe, the northern United States, Canada, eastern Siberia, southeastern Australia, and New Zealand; while countries located close to the equator are very rarely affected by MS [81, 94]. Similarly located at high latitudes, are China, Japan, northern Scandinavia, central Siberia, and Alaska, which belong to low- or moderate-risk areas [94]. The Mediterranean region and Middle East have long been considered low-/moderate-risk areas, but recent studies indicate that these regions are at moderate/high risk [52, 94]. A similar situation, with increasing risk for MS, is seen in the southern states of the United States, South Africa, and eastern Australia [4]. In the beginning of the 20th century, MS was primarily affecting Caucasians. The majority of MS patients was living in rural areas, and the female: male ratio was close to 1 [3, 81]. Nowadays, the disease is found mostly in women (female: male ratio ranging from 2 to 5), while racial and urban/rural differences are less pronounced [4, 116]. Since these changes occurred within a few (sometimes a single) generations, it is rather unlikely that genetically driven factors can primarily be responsible. Still, if MS were caused by a microorganism, a possibility that such a microbe has changed genetically within a century cannot be excluded.

Other observations also cast doubt on the genetic source of MS. These studies examined the fate of people migrating from high- to low- and from low- to high-risk areas. It has been revealed that adult people migrating from Europe (a high-risk area) to South Africa, Australia, and Israel (low-risk areas) retained the high risk for MS of their motherlands. However, those migrating as children (a borderline age was 15 years) presented with a risk similar to the native population of the new country [24, 27, 52, 53, 73]. Also, people migrating from continental Turkey (a low-risk area) to Cyprus (a high-risk area) were still at low risk of developing MS [25]. A similar situation was seen in the case of adults migrating from the Indian subcontinent, sub-Saharan Africa, and the Caribbean region (low-risk areas) to the United Kingdom (a high-risk area). The immigrants did not exhibit a high prevalence of MS. Still, while the adult immigrants to the United Kingdom infrequently developed MS, their UK-born children (obviously of the same genetic profile as their parents) presented with the same high risk of MS as native people of Britain [32]. Similarly, although MS is nearly unknown in black Africans [26], it is quite common in black Americans [50, 52]. In recent study on US soldiers, the risk of MS was even higher in blacks than in whites, contrary to the widely accepted genetic paradigm [116]. Of note, there are several studies that have demonstrated that the susceptibility to MS was associated with genes of the major histocompatibility complex (MHC) [41, 62]. Still, this effect was not very strong. On the contrary, the aforementioned epidemiological observations indicate that genetic susceptibility plays a minor role in MS pathogenesis and point toward an environmental factor that is responsible.

A number of epidemiological studies have shown that in several areas, the probable epidemics of MS took place. The best known and best documented example of such an epidemic comes from the Faroe Islands [52, 53, 117]. Importantly, medical records in the Faroe Islands—a north Atlantic territory of Denmark—were scrupulously carried out throughout the 20th century. Then the medical documentation of all Faroese was meticulously reviewed by MS researchers in a search of any anomalous epidemiological trend, thus minimizing a potential bias resulting from inadequate methodology or incomplete data. It appears that the Faroe Islands were free of MS until the islands were occupied by British troops during World War II. A few years after the arrival of British soldiers, the first cases of MS ever found among native Faroese were described. This first outbreak of MS was followed by two possible secondary epidemics [52, 53]. Similar epidemics of MS—yet, not so well documented—occurred in other relatively isolated areas: Iceland [53], the Orkneys [53, 70], and Sardinia [95]. Well-documented clusters of MS in some areas were also reported [31]. Besides, it seems that there is a spread of MS that affects new areas and new groups of humans (women, nonwhites, older people, urban populations) [4]. Although the prevalence of MS is growing in almost all countries studied, there are some areas (e.g., the Orkneys) [53] where the prevalence is falling, maybe due to an increased resistance of the population to an infectious agent.

### 4. MS as an epidemic

The epidemic of the disease does not necessarily mean that the pathology is caused by an infectious factor. Quite the contrary, a lot of epidemics are caused by nonbiological factors (tobacco, toxins, air pollutants, radiation, etc). Some researchers claim a role of environmental factors associated with the so-called western lifestyle (western diet, physical inactivity, inadequate sleep, chronic psychological stress, smoking, pollution, etc.). But if it were the case, then a high prevalence of MS should be expected not only in Europe and North America but also in Japan. Besides, MS should not have been known before the 20th century, which is not the case. (Actually, the first description of probable MS comes from the 15th century.) Considering the fact that MS is affecting very different environments (urban and rural areas, highly polluted and pristine territories, countries with high and low levels of background radiation, etc), it is very unlikely that MS is caused by nonbiological factors. Some epidemiological studies point toward a microorganism as the cause of MS epidemics [31, 51, 52, 53, 70, 95, 117]. Still, it is also known that despite many efforts, not a single microorganism has been definitely identified. If this were a simple issue, however, someone would have already
solved the problem. Perhaps the biology of MS may give us more information about this elusive microbe. Although MS is the most common neurodegenerative disease in young adults in western countries, it is nonetheless a rare pathology, with the prevalence in high-risk areas, ranging from 100 to 200 per 100,000 and being much less common in the rest of the world. Importantly, epidemiological models tell us that if an infection is symptomatic in the majority of infected individuals (like influenza virus) and the prevalence is low, it is impossible that such an epidemic is active for a longer time with such a low prevalence. It is due to the fact that the number of infected individuals in a population is too small to enable effective transmission of the pathogen. Still, some epidemics with such a low prevalence of symptomatic patients may persist, but an alternative reservoir of microorganisms is required, either in humans (in this scenario, the disease is clinically overt in a minority of infected people) or in animals. Sometimes a reservoir of infectious agent may exist both in nonsymptomatic humans and animals. Probably, the hypothetical infectious agent of MS had to be residing initially in animals. All, except for only a few, human infectious diseases are caused by microorganisms that originally were residing in animals, primarily domesticated ones, such as pigs, cattle, and poultry. Since the geographic distribution of MS is uneven, with the lowest prevalence in Africa, it points against microorganisms that have been infecting humans for millennia. Otherwise, MS would be distributed differently, either with the highest prevalence in Africa (the motherland of the mankind) or with the same prevalence worldwide. MS is also uncommon in the main centers of animal domestication: the Middle East (until the recent increase in prevalence), India (except for some non-Indian ethnic groups), and China [94]. It should also be mentioned that in the past, a canine source of MS was considered (canine distemper virus); but this possibility has been ruled out by epidemiological and virological research [6]. In conclusion, this elusive microbe is unlikely to come from typical domestic animals.

5. MS and historical data
Historical records of possible MS cases suggest that the disease is quite new. Contrary to other diseases, not a single description of possible MS can be found in ancient and early medieval texts (respecting the fact that the first medical description of MS as an entity actually comes from the second part of the 19th century) [16]. The first known person with possible MS was St. Lidwina, a Dutch woman who got sick in 1385 [74]. Some researchers claim that a primary focus of an MS epidemic might be situated in the Scandinavian peninsula [51,52,56] (far away from the main centers of animal domestication). If MS outbreaks actually began in Scandinavia, it is tempting to suggest that such an event took place around the 13th to 14th century. It is rather unlikely that it occurred much earlier, considering the geographic distribution of MS and a lack of ancient and early medieval descriptions of possible cases of the disease (even respecting the scarcity of old medical records). Moreover, a suggested chronology is in line with the nonexistence of MS in the Faroe Islands before the 1940s. These islands were settled by the Norsemen (the Vikings) in the early Middle Ages; and from that time (until World War II), the Faroese population, except for sporadic contact, was largely isolated. In general, the Faroese are probably the descendants of those Norsemen who had no contact with a hypothetical infectious agent of MS (interestingly, the only Faroese who developed MS before 1940 were those living for several years in mainland Denmark) [8]. There is also another fact pointing toward the 14th century. One may ask, what has happened in 14th-century Scandinavia that made the jump of a hypothetical infectious agent from an animal reservoir to humans possible? Indeed, the 14th century was somewhat unique. First, at that time, a dramatic climate cooling, the so-called Little Ice Age, took place. It resulted in the migration of nomadic tribes of northern Scandinavia, the Sami people (the Lapps), southward and their intensified contact with Norsemen. Second, at the same time, the Danish, Swedish, and Russian states became stronger and more centralized. Consequently, taxing the people, including the nomads, intensified, again resulting in more contact between the Lapps and Norsemen. Beginning from the 14th century, the Sami people, due to climate change and the necessity of paying taxes, intensified reindeer herding. Reindeer husbandry began in central Norway in the 11th century, but until the 14th century, reindeer hunting prevailed. The question of whether an infectious agent of MS has jumped from domesticated reindeer or whether another arctic animal has played a role will remain, of course, unanswered. Still, it is tempting to speculate that MS originally developed in the Sami people, perhaps long before the 14th century. It is known that the prevalence of MS among the Lapps is much lower than that of their Swedish and Norwegian neighbors [94] (despite the fact that they live in the northernmost part of Scandinavia, thus contrary to the latitude model of MS). But if an infectious agent of MS has been circulating in this population for a longer time, one may expect a decreased prevalence of the disease due to increased resistance and/or less intensive immune responses associated with the infection.

6. Other epidemiological data
In addition to the already-presented epidemiological data, there are also some other data that can help us to identify a hypothetical factor of MS. Positive correlations have been found between MS prevalence and the bovine population, as well as with milk consumption. Although
these phenomena could be interpreted differently, perhaps a link between MS and cows exists [67,68,81]. In a recent epidemiological study on US soldiers, the authors revealed an unexpectedly higher prevalence of MS in the Army and Air Force and significantly lower prevalence in the Navy and Marine Corps [116]. The researchers were not able to find a reasonable explanation for this fact. Was service on the sea somewhat protective? It is tempting to speculate that if a hypothetical microorganism responsible for MS were transmissible by an arthropod vector (for example, mosquito), one should expect to find such a phenomenon, since those militaries serving on the sea were less likely to meet blood-sucking arthropods.

There are also studies revealing obesity in adolescents as a risk factor of developing MS [80]. Although this association of increased body mass index with a higher susceptibility to MS is unclear, and although the possibility that obesity promotes MS through increased inflammation associated with a higher production of adipokines and proinflammatory cytokines cannot be ruled out, perhaps metabolism of lipids plays a role. Such a link is not very likely in the case of bacterial or viral infections. Still, it may be considered if protozoa were responsible. It is well known that many parasitic protozoa (including Plasmodium, Trypanosoma, and Trichomonas) cannot synthesize lipids and have to use these fatty compounds produced by the host [21,35,36,71,86]. Besides, some parasites (e.g., Trypanosoma cruzi) are using adipocytes as a safe reservoir, where these microbes can survive and evade the immune responses of humans [34,82]. Interestingly, a low-fat diet exhibits protective effects against MS progression, as has been demonstrated in some longitudinal clinical studies [106,107,118].

7. Histology of MS plaques and immune responses in the setting of the disease

MS plaques are characterized by profound heterogeneity of the lesions. Several types of these foci of demyelination can be distinguished. Still, all these types primarily present with CD8+ lymphocyte and macrophage infiltration [10,58,102], which is not typical in classic autoimmune disorders, characterized predominantly by CD4+ lymphocytic infiltrates. Deposition of immunoglobulins and complement antigens, suggesting an important role for autoimmunity, can only be seen in class II lesions; while in class I and III lesions, a destructive process is probably induced by another undetermined mechanism [64,69]. Immunological studies have also revealed that MS-associated autoantibodies are produced by nervous tissue- and cerebrospinal fluid-residing B cells. Interestingly, these cells comprise a limited number of clonotypes and are not found in the peripheral blood. This antibody response is stable over a long time [42]. Other studies have demonstrated clonally expanded CD8+ lymphocytes and persistence of these cells in the cerebrospinal fluid over a period of months [102]. Such an immune response, in addition to MS, is typical for a variety of acute and chronic infections of the central nervous system. CD8+ lymphocytic infiltration may suggest a protozoan infection (such a scenario has already been discussed by Murrell et al., who called attention to similarities between histological characteristics of MS and African trypanosomiasis; still, the authors found such a scenario impossible, considering the geographic distribution of African trypanosomes) [81]. Yet trypanosomes, as well as other parasitic kinetoplastids and apicomplexans, are actually also common in temperate and subarctic climate zones. Of note, parasitic trypanosomes induce the production of autoantibodies against myelin basic protein and galactocerebrosides [2,9,43,88]—a known hallmark of MS [75,84,89]. In general, the histological and immunological picture of MS is neither stereotypic of an autoimmune disease nor representative of a typical bacterial or viral infection. It rather suggests an atypical viral infection or a disease triggered by parasitic protozoa or spirochetes [81].

The histology of early MS plaques may also be helpful. Newly forming plaques are not infiltrated by lymphocytes. Such an infiltrate is typical for viral encephalitides and experimental autoimmune encephalomyelitis (the animal model disease of MS). On the contrary, early plaques primarily present with microglial activation [13,65,91,112]. Besides, early cortical lesions are associated with inflammation of the meninges [65]. Interestingly, such microglial activation, accompanied by infiltration by macrophages, can also be seen in the settings of several protozoan infections of the central nervous system [11,19,60]. Besides, meningoencephalitis, although histologically different from that seen in MS, can be found in humans infected with trypanosomes [20].

It is also known that MS plaques are preferentially found in the parts of the brain containing high amounts of iron [1,22,40]. Such a link may be indicative of viral infections, since some viruses selectively infect iron-acquiring cells. Still, the excess of iron within MS plaques seems to be located extracellularly, thus pointing against a role for viruses. Interestingly, T. cruzi requires an iron source for an optimal growth rate, and in hypoferremic hosts, the parasite is much less pathogenic [7,55,63,76]. Perhaps, other parasitic protozoa are also iron-dependent.

8. Which suspected microorganisms cannot be causing MS?

Epidemiological studies suggest that MS may be caused by an infectious factor. Yet, in spite of intensive research, no definitive evidence has emerged to identify a single microorganism that is unequivocally responsible. Since such a microorganism needs to meet several criteria...
from histological and biochemical studies (lymphocytic composition of perivascular and parenchymal infiltrate responses, as well as cerebrospinal fluid characteristics), this excludes most bacteria and viruses, with their typical pyrogenic activity. However, it is possible that MS emerges as a result of so-called molecular mimicry \[115\]. Epstein-Barr virus (EBV) has long been suspected to be such a causal factor. It is known that MS is found more often in EBV-seropositive individuals compared with those who are seronegative. MS patients have also been found to reveal increased immune responses against EBV \[8,45\]. Besides, circulating EBV-specific CD8\(^+\) cells can be found in these patients. However, EBV infection is very common in China and Japan (infection rates in these countries are even higher than in the West) \[108\]; still, MS prevalence in these countries is low, pointing against a role for EBV in MS pathogenesis. Moreover, recent studies that have used RNA in situ hybridization and real-time polymerase chain reaction (PCR) assays have revealed that EBV is very unlikely to be a direct trigger of MS \[85\]. A similar role in MS pathogenesis, with the same unconvincing results, has been considered for other microorganisms: Chlamydia pneumoniae, measles virus, adenoviruses, respiratory syncytial virus, human herpes virus-6, parainfluenza viruses, and others. Yet extensive microbiological studies have failed to demonstrate any consistent elevated antibody titers to all those microorganisms studied \[42,81\]. Similarly, epidemiological studies, especially migrant data or other aspects of MS epidemiology, are not supporting the idea that these microorganisms could be responsible for triggering MS \[8\].

9. What do pharmacological trials tell us?

The currently ruling paradigm is that MS is an autoimmune disease. Consequently, therapeutic strategies predominantly target the inflammatory cascade or modify the immune response. Interestingly, only a fraction of chemical compounds that exhibit such immunosuppressive or immunomodulatory activity and have been proven to be effective in experimental autoimmune encephalomyelitis have also revealed beneficial effects in humans \[18\]. These drugs, recommended for the treatment of MS, comprise interferon-beta, glatiramer acetate, and mitoxantrone. It is thought that all these drugs decrease or modify autoimmune reactions in the central nervous system. Then, if MS were actually driven by an infectious agent and not by autoimmunity, one should expect a deleterious clinical effect of such immunomodulating drugs, which is not the case. Maybe the aforementioned pharmaceutical agents are not active through immunomodulation but rather through antimicrobial activity. Indeed, all of them—interferon-beta \[48,49\], glatiramer acetate \[54\] and mitoxantrone \[29\]—in addition to their immunomodulating properties, exhibit antiprotozoan activity. Besides, it should be mentioned that some other drugs that are used for symptomatic treatment of MS—tetracyclines \[77,78,119\] and naltrexon \[23\]—also exhibit antiprotozoan activity \[5,46\]. For the time being, the antiprotozoan efficacy of a new anti-MS drug, natalizumab, has not been studied. Perhaps such research should be performed. The other new drug, fingolimod, increased the susceptibility to T. cruzi infection in an animal experiment, elevated parasitemia, and accelerated the mortality of infected mice \[30\]. However, another study has shown that fingolimod modified lipid metabolism and protected mice from high-fat-diet obesity \[47\]. Thus, maybe this pharmaceutical agent, in addition to its known immunomodulating effect, is effective in MS through the modification of lipid metabolism.

A well-known latitude paradigm of MS may also point toward microorganisms. Vitamin D, which is suspected to be responsible for the unique geographic distribution of MS, may also be involved. Still, its role is not necessarily related to its immunomodulatory property, as usually thought. It has been found that high levels of vitamin D exhibit a protective effect against T. cruzi and Plasmodium falciparum \[37,98,114\]. If MS were caused by a similar protozoan, a lot of sun, resulting in an adequate vitamin D level, might be protective against MS through its antimicrobial effects. Still, it should be remembered that vitamin D appears to contribute in defense against a number of viruses as well \[120\]. Besides, other light-dependent mechanisms may play a role (interestingly, apicomplexan and kinetoplastid protozoa are equipped with an organelle that probably developed from the chloroplast yet is no longer capable of photosynthesis). Some of these parasitic protozoa seem to be light-susceptible through melatonin-driven mechanisms, and light suppresses their replication \[66\]. Perhaps a hypothetical agent of MS exhibits similar properties.

10. Biological properties of the hypothetical microorganism

In summary, MS may be a rare complication of a highly prevalent (at least in high-risk areas) infection that is low or asymptomatic in the majority of infected individuals \[53,117\]. Most likely, the microorganism responsible is also infecting other mammals, and probably such an infection in animals is also asymptomatic. Epidemiological, histological, and clinical studies on MS suggest that if the disease were caused be an infectious agent, such a microorganism should exhibit several unique properties: it (i) is transmitted by an arthropod vector; (ii) should be characterized by specific metabolism of lipids; (iii) should be dependent on iron; (iv) should be associated with autoimmune responses of the host; and (v) should be susceptible to interferon-beta, glatiramer acetate, mitoxantrone, tetracyclines, and naltrexone but not to the degree that would allow its eradication.
In addition, this hypothetical microorganism, at least in the chronic stage of the infection, should exhibit tropism toward the brain. Although some of these properties may suggest very different microorganisms, a combination of them is unique for protozoa, especially trypanosomes and other blood-residing unicellular parasites. Of course, such an infectious agent of MS has not yet been identified. But it is also known that standard histological examinations cannot reveal many protozoa [14,20,60,93,111], especially in the chronic phase of the disease. Tissue samples of MS patients have never been studied using immunohistochemical staining against protozoan antigens, and neither has a search for protozoan antigens been performed using PCR assays.

As has already been suggested in this paper, Scandinavian reindeer may be the primary animal reservoir of this elusive agent of MS. Indeed, reindeer are infected by a legion of different unicellular parasites: trypanosomes (Trypanosoma cervi, Trypanosoma theileri) and apicomplexans (Babesia divergens, Babesia capreoli, Besnoitia sp., Eimeria sp., and Sarcosporidia sp.) [92]. Probably, other species of parasitic protozoa infecting these animals are still undiscovered. Faroe Islands studies suggest that the first contact with this hypothetical infectious agent of MS may manifest with acute gastroenteritis [117]. This may mean that the microorganism is attacking the intestines. Perhaps, if it is actually fat-dependent, as suggested by some epidemiological studies, it is initially residing in the intestinal lymphatics, with easy access to the lipid-rich chyle. Some parasitic trypanosomes preferentially invade and replicate in the lymphatic vessels of the abdominal cavity [90,103]. If such a scenario is correct, then the microorganism causing MS, after replication in the intestinal lymphatics, should spread into the bloodstream through the thoracic duct. This may account for the high prevalence of malformed valves of the left internal jugular vein seen in MS patients [101], since the thoracic duct is joining the venous system in proximity to the left jugular valve.

There are, however, some data pointing against a potential role for trypanosome-like protozoans in the pathogenesis of MS. Epidemiological studies on MS suggest that children under the age of 15 are resistant to this disease [24,52,73]. Still, trypanosomes are usually more virulent in children [79,96]. This means that trypanosomes (at least those exhibiting a similar biology to African and American species) are unlikely to be responsible for MS. On the contrary, another genus of parasitic protozoa, Babesia, is characterized by selective infection of adult hosts. Young animals, as well as young humans, are protected against these parasitic apicomplexans through innate immunity mechanisms [38,57,61,110,113]. Interestingly, proper function of the spleen is needed [38,113], and splenectomy of immature animals abolishes their resistance against these parasites [57]. It is well known that MS is rarely seen in children. Then, if such a low prevalence of MS in children were due to a similar mechanism as in the case of babesiosis, one should expect a higher prevalence of MS in those children who underwent a splenectomy. Yet such a surgical procedure is rarely performed in children, and no published evidence supports this idea. There is, however, an intriguing report on comorbidities in children suffering from immune thrombocytopenia. This pathology is associated with splenic dysfunction. Of note, an unexpectedly high prevalence (25 times more than anticipated) of pediatric MS has been found in this particular group of children [97].

11. How to test this hypothesis?
In order to test this protozoan hypothesis of MS, extensive research should be performed. It should be expected that simple blood smears will be negative. The microbe, if it really exists and inhabits the vascular system, is probably not available for standard screening, since it is residing in atypical locations (like the intestinal lymphatics and cerebral venules). It is also possible that after the first episode of infection, it is no longer present in the bloodstream but escapes into immunoprivileged organs (a specific intrathecal immune reaction in MS patients suggests a response against such brain-residing microorganisms). Moreover, it should be remembered that standard tissue staining may be inadequate to reveal many protozoa [60,93,111]. Rather, such techniques as immunohistochemical staining and PCR assays should be used. Yet with the microbe not yet known, this will not be an easy task. Modern diagnostic techniques, like multiplex molecular technology, should probably be used. Such molecular probe technology does not require growth of a microorganism in culture. The test requires only a sequence of sequential bases unique to the genome of a pathogen of interest. Perhaps, a screen should be performed using the antigens of known parasitic protozoa that infect domestic and wild animals in high-risk areas for MS. There are also some common protozoan epitopes that could be used in such research [44,83,105].

References
[1] C. W. Adams, Perivascular iron deposition and other vascular damage in multiple sclerosis, J Neurol Neurosurg Psychiatry, 51 (1988), 260–265.
[2] A. Al-Sabbagh, C. A. Garcia, B. M. Diaz-Bardales, C. Zacarias, J. K. Sakurada, and L. M. Santos, Evidence for cross-reactivity between antigen derived from Trypanosoma cruzi and myelin basic protein in experimental Chagas disease, Exp Parasitol, 89 (1998), 304–311.
[3] R. S. Allison and J. H. Millar, Prevalence of disseminated sclerosis in Northern Ireland, Ulster Med J, 23 (1954), 1–27.
[4] A. Alonso and M. A. Hernán, Temporal trends in the incidence of multiple sclerosis: a systematic review, Neurology, 71 (2008), 129–135.
[5] D. N. Amin, W. Masocha, K. Ng’andwe, M. Rottenberg, and K. Kristensson, Suramin and minocycline treatment of
experimental African trypanosomiasis at an early stage of parasite brain invasion, Acta Trop, 106 (2008), 72–74.

[6] A. M. Parce, A. F. Alifri, and A. A. Alifri, Canine distemper virus and multiple sclerosis: A real or an anecdotal association?, in Current Research, Technology and Education Topics in Applied Microbiology and Microbial Biotechnology, A. Méndez-Vilas, ed., Formatex Research Center, Badajoz, Spain, 2010, 737–745.

[7] J. M. Arantes, M. L. Pedrosa, H. R. Martins, V. M. Veloso, M. de Lana, M. T. Bahia, et al., Trypanosoma cruzi: treatment with the iron chelator desferrioxamine reduces parasitemia and mortality in experimentally infected mice, Exp Parasitol, 117 (2007), 43–50.

[8] A. Ascherio and K. L. Munger, Environmental risk factors for multiple sclerosis. Part I: the role of infection, Ann Neurol, 61 (2007), 288–299.

[9] T. Asonganyi, G. Lando, and J. L. Ngu, Serum antibodies against human brain myelin proteins in Gambian trypanosomiasis, Ann Soc Belg Med Trop, 69 (1989), 213–221.

[10] H. Babbe, A. Roers, A. Waisman, H. Lassmann, N. Goebels, R. Hohlfeld, et al., Clonal expansions of CD8+ T cells dominate the T cell infiltrate in active multiple sclerosis lesions as shown by micromanipulation and single cell polymerase chain reaction, J Exp Med, 192 (2000), 393–404.

[11] W. Baetas-da Cruz, R. M. Macedo-Silva, A. Santos-Silva, A. Henriques-Pons, M. F. Madeira, S. Corte-Real, et al., Destiny and intracellular survival of leishmania amazonensis in control and dexamethasone-treated glial cultures: protozoa-specific glycoconjugate tagging and TUNEL staining, J Histochem Cytochem, 52 (2004), 1047–1055.

[12] M. H. Barnett, A. P. Henderson, and J. W. Prineas, Relapsing and remitting multiple sclerosis: Pathology of the newly forming lesion, Ann Neurol, 55 (2004), 458–468.

[13] L. A. Benvenuti, A. Roggerio, H. F. Freitas, A. J. Mansur, A. Fiorelli, and M. L. Higuchi, Chronic American trypanosomiasis: parasite persistence in endomyocardial biopsies is associated with high-grade myocarditis, Ann Trop Med Parasitol, 102 (2008), 481–487.

[14] W. Brück, Inflammatory demyelination is not central to the pathogenesis of multiple sclerosis, J Neurol, 252 (2005), v10–v15.

[15] J. Charcot, Histologie de la sclérose en plaques, Gaz Hop, 41 (1868), 554–566.

[16] A. Chaudhuri and P. O. Behan, Multiple sclerosis is not an autoimmune disease, Arch Neurol, 61 (2004), 1610–1612.

[17] A. Chaudhuri and P. O. Behan, Multiple sclerosis: looking beyond autoimmunity, J R Soc Med, 98 (2005), 303–306.

[18] S. Chianella, M. Semprevivo, Z. C. Peng, D. Zaccheo, M. Bentivoglio, and G. Grassi-Zucconi, Microglia activation in a model of sleep disorder: an immunohistochemical study in the rat brain during Trypanosoma brucei infection, Brain Res, 832 (1999), 54–62.

[19] L. Chimelli, A morphological approach to the diagnosis of protozoal infections of the central nervous system, Patholog Res Int, 2011 (2011), 290853.

[20] L. Coppen, T. Levade, and P. J. Courtoy, Host plasma low density lipoprotein particles as an essential source of lipids for the bloodstream forms of Trypanosoma brucei, J Biol Chem, 270 (1995), 5736–5741.

[21] W. Cruelaus, M. W. Migdal, C. P. Luessenhop, A. Sugar, and I. Mihalakis, Iron deposits surrounding multiple sclerosis plaques, Arch Pathol Lab Med, 106 (1982), 397–399.

[22] B. A. Cree, E. Kornyeeye, and D. S. Goodin, Pilot trial of low-dose naltrexone and quality of life in multiple sclerosis, Ann Neurol, 50 (2001), 145–150.

[23] G. Dean, Annual incidence, prevalence, and mortality of multiple sclerosis in white South-African-born and in white immigrants to South Africa, Br Med J, 2 (1967), 724–730.

[24] G. Dean, H. Akson, T. Akalin, L. Middleton, and K. Kyriallis, Multiple sclerosis in the Turkish- and Greek-speaking communities of Cyprus. A United Nations (UNHCR) Bicomunal Project, J Neurol Sci, 145 (1997), 163–168.

[25] G. Dean, A. I. Bhigjee, P. L. Bill, V. Fritz, I. C. Chikanza, J. E. Thomas, et al., Multiple sclerosis in black South Africans and Zimbabweans, J Neurol Neurosurg Psychiatry, 57 (1994), 1064–1069.

[26] G. Dean and J. F. Kurtzke, On the risk of multiple sclerosis according to age at immigration to South Africa, Br Med J, 3 (1971), 725–729.

[27] O. C. DeLuca, G. C. Ebers, and M. M. Esiri, Axonal loss in multiple sclerosis: a pathological survey of the corticospinal and sensory tracts, Brain, 127 (2004), 1009–1018.

[28] A. Deterding, F. A. Dungey, K. A. Thompson, and D. Steverding, Anti-trypanosomal activities of DNA topoisomerase inhibitors, Acta Trop, 93 (2005), 311–316.

[29] M. R. Domínguez, J. Ersching, R. Lemos, A. V. Machado, O. Bruna-Romero, M. M. Rodrigues, et al., Re-circulation of lymphocytes mediated by sphingosine-1-phosphate receptor-1 contributes to resistance against experimental infection with the protozoan parasite Trypanosoma cruzi, Vaccine, 30 (2012), 2882–2891.

[30] P. T. Donnan, J. D. Parratt, S. V. Wilson, R. B. Forbes, J. I. O’Riordan, and R. J. Swingler, Multiple sclerosis in Tasmania, Scotland: detection of clusters using a spatial scan statistic, Mult Scler, 11 (2005), 403–408.

[31] M. Elian, S. Nightingale, and G. Dean, Multiple sclerosis among United Kingdom-born children of immigrants from the Indian subcontinent, Africa and the West Indies, J Neurol Neurosurg Psychiatry, 53 (1990), 906–911.

[32] N. Evangelou, D. Konz, M. M. Esiri, S. Smith, J. Palace, and P. M. Matthews, Size-selective neuronal changes in the anterior optic pathways suggest a differential susceptibility to injury in multiple sclerosis, Brain, 124 (2001), 1813–1820.

[33] A. V. Ferreira, M. Segatto, Z. Menezes, A. M. Macedo, C. Gelape, L. de Oliveira Andrade, et al., Evidence for Trypanosoma cruzi in adipose tissue in human chronic Chagas disease, Microbes Infect, 13 (2011), 1002–1005.

[34] E. Folly, N. L. Cunha e Silva, A. H. Lopes, M. A. Silva-Neto, and G. C. Atella, Trypanosoma rangeli uptakes the main lipoprotein from the hemolymph of its invertebrate host, Biochim Biophys Res Commun, 310 (2003), 555–561.

[35] M. P. Gillett and J. S. Owen, Characteristics of the binding of human and bovine high-density lipoproteins by bloodstream forms of the African trypanosom, Trypanosoma brucei brucei, Biochim Biophys Acta, 1123 (1992), 239–248.

[36] D. G. Godfrey, Antiparasitic action of dietary cod liver oil upon Plasmodium berghei and its reversal by vitamin E, Exp Parasitol, 6 (1957), 555–565.

[37] W. L. Goff, W. C. Johnson, S. M. Parish, G. M. Barrington, W. Troo, and R. A. Valdez, The age-related immunity in cattle to Babesia bovis infection involves the rapid induction of interleukin-12, interferon-gamma and inducible nitric oxide synthase mRNA expression in the spleen, Parasite Immunol, 23 (2001), 463–471.

[38] E. M. Hacking, Chronic cerebral spinal venous insufficiency in multiple sclerosis, Expert Rev Neurother, 11 (2011), 5–9.

[39] E. M. Hacking, M. Makki, Y. Ge, M. Maheshwari, V. Sehgal, J. Hu, et al., Characterizing iron deposition in multiple sclerosis
lesions using susceptibility weighted imaging, J Magn Reson Imaging, 29 (2009), 537–544.

[41] J. L. Haines, H. A. Terwedow, K. Burgess, M. A. Pericak-Vance, J. B. Rimmler, E. R. Martin, et al., Linkage of the MHC to familial multiple sclerosis suggests genetic heterogeneity. The Multiple Sclerosis Genetics Group, Hum Mol Genet, 7 (1998), 1229–1234.

[42] B. Hemmer, J. J. Archelos, and H. P. Hartung, New concepts in the immunopathogenesis of multiple sclerosis, Nat Rev Neurosci, 3 (2002), 291–301.

[43] C. A. Hunter, F. W. Jennings, J. F. Tierney, M. Murray, and P. G. Kennedy, Correlation of autoantibody titres with central nervous system pathology in experimental African trypanosomiasis, J Neuroimmunol, 41 (1992), 134–148.

[44] R. W. Hyman, R. P. St Onge, H. Kim, J. S. Tamaresis, B. Hemmer, J. J. Archelos, and H. P. Hartung, Beta-interferon inhibits epidemiology of multiple sclerosis. Does this J. F. Kurtzke, Further features of the Fennoscandian focus of multiple sclerosis, Acta Neurol Scand, 111 (2005), 238–246.

[45] J. F. Kurtzke, Epidemiology of multiple sclerosis. Does this really point toward an etiology? Lectio Doctoralis Health Nations Health, 54 (1964), 588–597.

[46] J. F. Kurtzke, Further considerations on the distribution of multiple sclerosis in Sweden, Acta Neurol Scand, 111 (2005), 238–246.

[47] R. J. Langley and J. S. Gray, Age-related susceptibility of the gerbil, Meriones unguiculatus, to the bovine parasite, Babesia divergens, Exp Parasitol, 64 (1987), 466–473.

[48] H. Lassmann, The pathology of multiple sclerosis and its evolution, Philos Trans R Soc Lond B Biol Sci, 354 (1999), 1635–1640.

[49] H. Lassmann, W. Brück, and C. F. Lucchinetti, The immunopathology of multiple sclerosis: an overview, Brain Pathol, 17 (2007), 210–218.

[50] K. R. Lemos, L. C. Marques, L. P. Aquino, A. C. Alessi, and R. Z. Zacarias, Astrocitic and microglial response and histopathological changes in the brain of horses with experimental chronic Trypanosoma evansi infection, Rev Inst Med Trop Sao Paulo, 50 (2008), 243–249.

[51] M. G. Levy, G. Clabough, and M. Ristic, Age resistance in bovine babesiosis: role of blood factors in resistance to Babesia bovis, Infect Immun, 37 (1982), 1127–1131.

[52] M. R. Lincoln, A. Montpeit, M. Z. Cader, J. Saarela, D. A. Dyment, M. Tislar, et al., A predominant role for the HLA class II region in the association of the MHC region with multiple sclerosis, Nat Genet, 37 (2005), 1108–1112.

[53] V. Loo and R. Lalonde, Role of iron in intracellular growth of Trypanosoma cruzi, Infect Immun, 45 (1984), 726–730.

[54] C. Lucchinetti, W. Brück, J. Parisi, B. Scheithauer, M. Rodriguez, and H. Lassmann, Heterogeneity of multiple sclerosis lesions: implications for the pathogenesis of demyelination, Ann Neurol, 47 (2000), 707–717.

[55] C. F. Lucchinetti, B. F. Popescu, R. F. Bunyan, N. M. Moll, S. F. Roemer, H. Lassmann, et al., Inflammatory cortical demyelination in early multiple sclerosis, N Engl J Med, 365 (2011), 2188–2197.

[56] M. Macías, M. N. Rodríguez-Cabezas, R. J. Reiter, A. Osuna, and D. Acuña Castroviejo, Presence and effects of melatonin in Trypanosoma cruzi, J Pineal Res, 27 (1999), 86–94.

[57] D. Malosse and H. Perron, Correlation analysis between bovine populations, other farm animals, house pets, and multiple sclerosis prevalence, Neuroepidemiology, 12 (1993), 15–27.

[58] D. Malosse, H. Perron, A. Sasco, and J. M. Seigneurin, Correlation between milk and dairy product consumption and multiple sclerosis prevalence: a worldwide study, Neuroepidemiology, 11 (1992), 304–312.

[59] C. Marik, P. A. Felts, J. Bauer, H. Lassmann, and K. J. Smith, Lesion genesis in a subset of patients with multiple sclerosis: a role for innate immunity?, Brain, 130 (2007), 2800–2815.

[60] J. R. Martin, Troop-related multiple sclerosis outbreak in the Okrshes?, J Epidemiol Community Health, 41 (1987), 183–184.

[61] J. Mazumdar and B. Striepen, Make it or take it: fatty acid metabolism of apicomplexan parasites, Eurkaryot Cell, 6 (2007), 1727–1735.

[62] H. F. McFarland, Twin studies and multiple sclerosis, Ann Neurol, 32 (1992), 722–723.

[63] J. G. McLeod, S. R. Hammond, and J. F. Kurtzke, Migration and multiple sclerosis in immigrants to Australia from United Kingdom and Ireland: a reassessment. I. Risk of MS by age at immigration, J Neurol, 258 (2011), 1140–1149.

[64] R. Medaer, Does the history of multiple sclerosis go back as far as the 14th century?, Acta Neurol Scand, 60 (1979), 189–192.

[65] T. Menge, P. H. Lalive, H. C. von Büdingen, B. Cree, S. L. Hauser, and R. Z. Zacarias, Trends after the interruption of vectorial and transfusional bovine babesiosis: role of blood factors in resistance to Babesia microglial response and histopathological changes in the brain of horses with experimental chronic Trypanosoma evansi infection, Rev Inst Med Trop Sao Paulo, 50 (2008), 243–249.

[66] A. Moncayo, Chagas disease: current epidemiological trends after the interruption of vectorial and transfusional
transmission in the Southern Cone countries, Mem Inst Oswaldo Cruz, 98 (2003), 577–591.

[80] L. Mungar, T. Chitnis, and A. Ascherio, Body size and risk of MS in two cohorts of US women, Neurology, 73 (2009), 1543–1550.

[81] T. G. Murrell, L. S. Harbig, and I. C. Robinson, A review of the aetiology of multiple sclerosis: an ecological approach, Ann Hum Biol, 18 (1991), 95–112.

[82] F. Nagayoshi, M. S. Desruisseaux, L. A. Jelicks, F. S. Machado, S. Chua, P. E. Scherer, et al., Perspectives on adipose tissue, chagas disease and implications for the metabolic syndrome, Interdiscip Perspect Infect Dis, 2009 (2009), 824324.

[83] J. T. Nazeer, K. El Sayed Khalifa, H. von Thien, M. M. El-Sibaaei, M. Y. Abdel-Hamid, R. A. Tawfik, et al., Use of multiplex real-time PCR for detection of common diarrhea causing protozoan parasites in Egypt, Parasitol Res, 112 (2013), 595–601.

[84] K. Ota, M. Matsui, E. L. Milford, G. A. Mackin, H. L. Weiner, and D. A. Haffer, T- cell recognition of an immune-dominant myelin basic protein epitope in multiple sclerosis, Nature, 346 (1990), 183–187.

[85] G. P. Owens and J. L. Bennett, Trigger, pathogen, or bystander: the complex nexus linking Epstein-Barr virus and multiple sclerosis, Mult Scler, 18 (2012), 1204–1208.

[86] K. M. Peterson and J. F. Alderete, Trichomonas vaginalis is dependent on uptake and degradation of human low density lipoproteins, J Exp Med, 160 (1984), 1261–1272.

[87] L. K. Peterson and R. S. Fujinami, Inflammation, demyelination, neurodegeneration and neuroprotection in the pathogenesis of multiple sclerosis, J Neuroimmunol, 184 (2007), 37–44.

[88] K. Petry, P. Voisin, T. Baltz, and J. Labouesse, Epitopes common to trypanosomes (T. cruzi, T. dionisii and T. vespertilionis (Schizotrypanum)): astrocytes and neurons, J Neuroimmunol, 16 (1987), 237–252.

[89] M. Pette, K. Fujita, D. Wilkinson, D. M. Alltmann, J. Trowsdale, G. Giegerich, et al., Multiple sclerosis: why does the number and morphology of T cells persist as clonal expansions in the cerebrospinal fluid and blood, Proc Natl Acad Sci U S A, 104 (2004), 2428–2433.

[90] C. Schmid, M. Richer, C. M. Bilenge, T. Josenando, F. Chapuis, C. R. Manthelot, et al., Effectiveness of a 10-day melarsoprol schedule for the treatment of late-stage human African trypanosomiasis: confirmation from a multinational study (IMPEML II), J Infect Dis, 191 (2005), 1922–1931.

[91] J. B. Segal and N. R. Powe, Prevalence of immune thrombocytopenia: analyses of administrative data, J Thromb Haemost, 4 (2006), 2377–2383.

[92] G. Rosati, The prevalence of multiple sclerosis in the world: an update, Neurol Sci, 22 (2001), 117–139.

[93] G. Rosati, I. Aiello, E. Granieri, M. I. Pirastru, S. Becciu, G. Demontis, et al., Incidence of multiple sclerosis in Macomer, Sardinia, 1912–1981: onset of the disease after 1950, Neurology, 36 (1986), 14–19.

[94] C. Schmid, M. Richer, C. M. Bilenge, T. Josenando, F. Chapuis, C. R. Manthelot, et al., Effectiveness of a 10-day melarsoprol schedule for the treatment of late-stage human African trypanosomiasis: confirmation from a multinational study (IMPEML II), J Infect Dis, 191 (2005), 1922–1931.

[95] J. T. Nazeer, K. El Sayed Khalifa, H. von Thien, M. M. El-Sibaaei, M. Y. Abdel-Hamid, R. A. Tawfik, et al., Use of multiplex real-time PCR for detection of common diarrhea causing protozoan parasites in Egypt, Parasitol Res, 112 (2013), 595–601.

[96] K. Ota, M. Matsui, E. L. Milford, G. A. Mackin, H. L. Weiner, and D. A. Haffer, T-cell recognition of an immune-dominant myelin basic protein epitope in multiple sclerosis, Nature, 346 (1990), 183–187.

[97] G. P. Owens and J. L. Bennett, Trigger, pathogen, or bystander: the complex nexus linking Epstein-Barr virus and multiple sclerosis, Mult Scler, 18 (2012), 1204–1208.

[98] K. M. Peterson and J. F. Alderete, Trichomonas vaginalis is dependent on uptake and degradation of human low density lipoproteins, J Exp Med, 160 (1984), 1261–1272.

[99] L. K. Peterson and R. S. Fujinami, Inflammation, demyelination, neurodegeneration and neuroprotection in the pathogenesis of multiple sclerosis, J Neuroimmunol, 184 (2007), 37–44.

[100] K. Petry, P. Voisin, T. Baltz, and J. Labouesse, Epitopes common to trypanosomes (T. cruzi, T. dionisii and T. vespertilionis (Schizotrypanum)): astrocytes and neurons, J Neuroimmunol, 16 (1987), 237–252.

[101] M. Pette, K. Fujita, D. Wilkinson, D. M. Alltmann, J. Trowsdale, G. Giegerich, et al., Multiple sclerosis: why does the number and morphology of T cells persist as clonal expansions in the cerebrospinal fluid and blood, Proc Natl Acad Sci U S A, 104 (2004), 2428–2433.

[102] C. Schmid, M. Richer, C. M. Bilenge, T. Josenando, F. Chapuis, C. R. Manthelot, et al., Effectiveness of a 10-day melarsoprol schedule for the treatment of late-stage human African trypanosomiasis: confirmation from a multinational study (IMPEML II), J Infect Dis, 191 (2005), 1922–1931.

[103] J. B. Segal and N. R. Powe, Prevalence of immune thrombocytopenia: analyses of administrative data, J Thromb Haemost, 4 (2006), 2377–2383.
[117] M. T. Wallin, A. Heltberg, and J. F. Kurtzke, Multiple sclerosis in the Faroe Islands. 8. Notifiable diseases, Acta Neurol Scand, 122 (2010), 102–109.

[118] B. Weinstock-Guttman, M. Baier, Y. Park, J. Feichter, P. Lee-Kwen, E. Gallagher, et al., Low fat dietary intervention with omega-3 fatty acid supplementation in multiple sclerosis patients, Prostaglandins Leukot Essent Fatty Acids, 73 (2005), 397–404.

[119] M. A. Yenari, L. Xu, X. N. Tang, Y. Qiao, and R. G. Giffard, Microglia potentiate damage to blood-brain barrier constituents: improvement by minocycline in vivo and in vitro, Stroke, 37 (2006), 1087–1093.

[120] D. A. Youssef, C. W. Miller, A. M. El-Abbassi, D. C. Cutchins, C. Cutchins, W. B. Grant, et al., Antimicrobial implications of vitamin D, Dermatoendocrinol, 3 (2011), 220–229.

[121] P. Zamboni, R. Galeotti, E. Menegatti, A. M. Malagoni, G. Tacconi, S. Dall’Ara, et al., Chronic cerebrospinal venous insufficiency in patients with multiple sclerosis, J Neurol Neurosurg Psychiatry, 80 (2009), 392–399.

[122] P. Zamboni, E. Menegatti, B. Weinstock-Guttman, M. G. Dwyer, C. V. Schirda, A. M. Malagoni, et al., Hypoperfusion of brain parenchyma is associated with the severity of chronic cerebrospinal venous insufficiency in patients with multiple sclerosis: a cross-sectional preliminary report, BMC Med, 9 (2011), 22.