

Neurobrucellosis Presenting as Leukoencephalopathy

The Role of Cytotoxic T Lymphocytes

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● A 65-year-old man developed a leukoencephalopathy associated with neurobrucellosis. The disease followed a 15-month progressive course with neurologic symptoms, and magnetic resonance imaging revealed bilateral symmetrical T2 signal hyperintensities in the white matter. Biopsy of the cerebral cortex and white matter was significant for nongranulomatous meningoencephalitis with reactive microgliosis and astrogliosis. The inflammatory infiltrate was predominantly composed of T lymphocytes, including numerous cytotoxic T cells. There was no evidence of significant myelin destruction. No organisms were detected microscopically, but elevated immunoglobulin G titers to *Brucella* were found in the cerebrospinal fluid. An abscess formed at the biopsy site, and *Brucella melitensis* was cultured from abscess contents. Neurobrucellosis is difficult to diagnose outside endemic regions and is associated with leukoencephalopathy-like pathology. Cytotoxic T lymphocytes and microglia activation play an immunopathogenic role in this rare disease.

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Human brucellosis is an infection of the reticuloendothelial system produced by *Brucella melitensis* and *Brucella abortus*.¹ Brucellosis is an endemic acute and chronic infection in much of the world but is quite rare in the United States. The nervous system is involved in up to 13% of brucellosis cases.² Some patients present with a neurologic disorder but no systemic signs or symptoms.³ The brain is rarely biopsied in brucellosis cases, and relatively few microscopic descriptions of central nervous system pathology have been published. In the present case, pathologic examination revealed white matter diffusely infiltrated by cytotoxic T lymphocytes and relative preservation of the myelin. This pattern of clinical and radiologic presentation appears unique and exposes the role of immunopathogenic mechanisms such as T-cell-mediated cytotoxicity in neurobrucellosis.

REPORT OF A CASE

A 65-year-old Iranian immigrant man living in the United States was in his usual state of good health until a return visit

to Iran 15 months prior to his diagnosis. In Iran, he developed right upper quadrant pain accompanied by a fever. An abdominal ultrasound examination revealed gallstones, and a cholecystectomy was performed. His postoperative course was uncomplicated, but 2 months later he developed right flank pain and bilateral knee pain. He then developed bitemporal headaches without constitutional symptoms, followed by otalgia and subtotal hearing loss on the right. Within weeks, there was progression and extension of hearing loss to the left. His symptoms persisted despite antibiotic treatment for presumed ear infection. He developed horizontal diplopia along with anorexia, nausea, and vomiting in association with his headaches. Evaluation of a lumbar puncture cerebrospinal fluid sample was "normal," although no results were available for review. The patient experienced a gradual, slowly progressive neurocognitive decline characterized by mild confusion, dysarthria, low mood, anhedonia, insomnia, and sexual dysfunction.

By the time he returned to the United States 9 months after his initial presentation, the patient had suffered a 13.6-kg weight loss and had persistent gastrointestinal and neurologic signs. An audiogram confirmed bilateral sensorineural hearing loss. There were mild cognitive impairments in verbal fluency, sequencing, and copying of written figures. Other neurologic functions were normal. A magnetic resonance image of the brain revealed diffuse, bilateral white matter T2 hyperintensities without parenchymal enhancement (Figure 1, A and B). Cerebrospinal fluid studies revealed a mild pleocytosis (white blood cells, 13/mm³, predominantly mononuclear cells) with normal protein (35 mg/dL) and glucose (54 mg/dL) concentrations. There were 2 oligoclonal bands, and the immunoglobulin (Ig) G index was 0.7 (normal range, up to 0.9). Serologic studies for human immunodeficiency virus, syphilis, neuroborreliosis, Ehrlichiosis, and Whipple disease were negative, as were routine blood and cerebrospinal fluid cultures. Serologic studies for known autoimmune and rheumatologic disorders were negative. Serologic studies for hepatitis C, A, and B surface antigens were negative, but there was evidence of anti-hepatitis B surface antibodies. Because of the magnetic resonance image evidence of a diffuse and symmetric white matter involvement, including prominent involvement of the subcortical u-fibers, and lack of enhancement, a panel of studies directed at late-onset leukodystrophies was obtained, again without diagnostic benefit.

Despite clinical improvement in cognitive deficits with antidepressant treatment, the magnitude of white matter lesions led to a decision to obtain a brain biopsy. Perioperative magnetic resonance images revealed an evolving, leptomeningeal enhancing lesion in the left Sylvian fissure. Following pathologic interpretation of the biopsy, neurobrucellosis entered the differential diagnosis, and the patient was found to have mildly elevated (1:64) cerebrospinal fluid titers of IgG antibodies directed against *B abortus*, with negative serum titers. The patient's postoperative course was uneventful until 4 weeks after the biopsy, when he presented to the emergency room with headaches, confusion, and urinary incontinence. A head computed tomography scan re-

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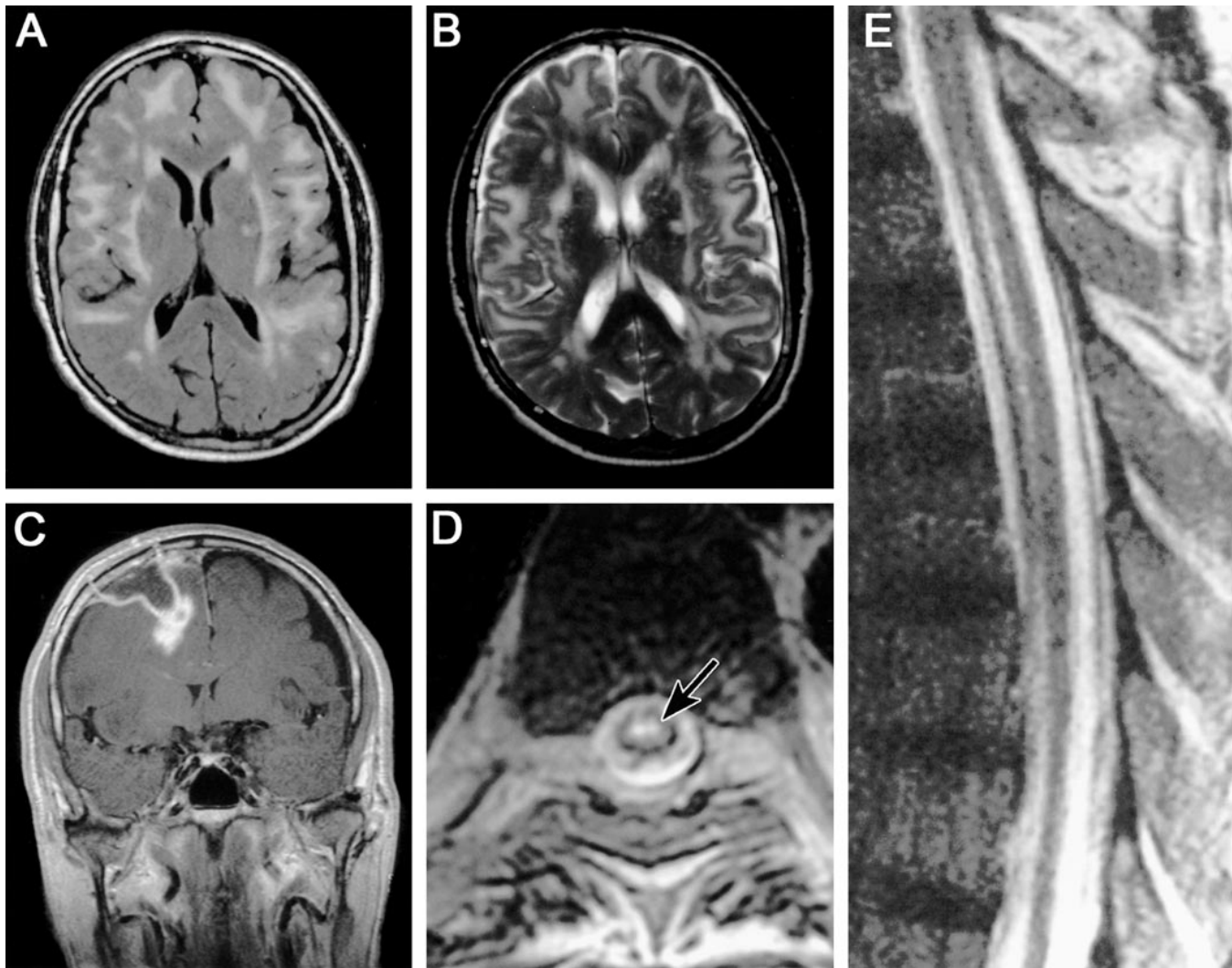


Figure 1. A and B, Diffuse, bilateral T2 signal hyperintensity is present throughout the white matter. C, An abscess is visible at the biopsy site after surgery. D and E, Myelitis is present in the thoracic spinal cord.

vealed a 2.0-cm abscess in the region of the previous biopsy (Figure 1, C). A right frontal craniotomy and abscess drainage was performed, and *B. melitensis* was cultured from the abscess contents. The patient significantly improved after 2 months of intravenous antibiotics (gentamicin and ceftriaxone) and 4 months of oral antibiotics (rifampin and doxycycline). However, 6 months after the brain biopsy, the patient presented to the hospital with fever, back pain, and rapidly progressive paraplegia over 1–2 days. He had myelitis with a central cord distribution that extended through the thoracic cord (Figure 1, D and E). A diagnosis of *Brucella* myelitis was made, and the patient received treatment with antibiotics and a 5-day course of intravenous methylprednisolone, followed by a tapering dose of prednisone. The patient has partially recovered from the weakness and is in the process of neurologic rehabilitation.

PATHOLOGIC FINDINGS

Examination of the biopsy specimen of the right frontal cortex and white matter revealed meningoencephalitis. A lymphocytic infiltrate was present in the leptomeninges and surrounding cortical vessels (Figure 2, A). Inflammatory cells were especially abundant in the white matter around vessels and infiltrating through the parenchyma (Figure 2, B). No evidence of vasculitis or granulomatous inflammation was detected. Methenamine silver, periodic

acid-Schiff, Brown and Hopps, and tissue Gram stains failed to reveal any microorganisms. No toxoplasma, papovavirus, adenovirus, or herpes virus antigens were detected immunohistochemically. Marked reactive astrogliosis was seen throughout the cortex and white matter (Figure 2, C). Macrophage markers such as Ham 56 highlighted numerous microglia, but macrophages were rare and predominantly located in perivascular spaces. Marked activation of microglia was demonstrated by immunostaining of major histocompatibility complex class II antigens (human leukocyte antigen DR marker) (Figure 2, D). B- and T-cell immunostains (CD20 and CD3) revealed the inflammatory infiltrate to be predominantly T cells, with only scattered B cells in the leptomeninges and perivascular spaces. CD4 and CD8 immunophenotyping of the T-cell infiltrates in the white matter and cortex revealed that the majority of the T cells were CD8⁺ lymphocytes (Figure 2, E). Luxol fast blue stains showed no significant myelin destruction (Figure 2, F). Microscopic examination of material resected from the abscess revealed necrotic tissue with acute and chronic inflammation and numerous macrophages. Although special stains were used, no microorganisms were identified in the abscess material.

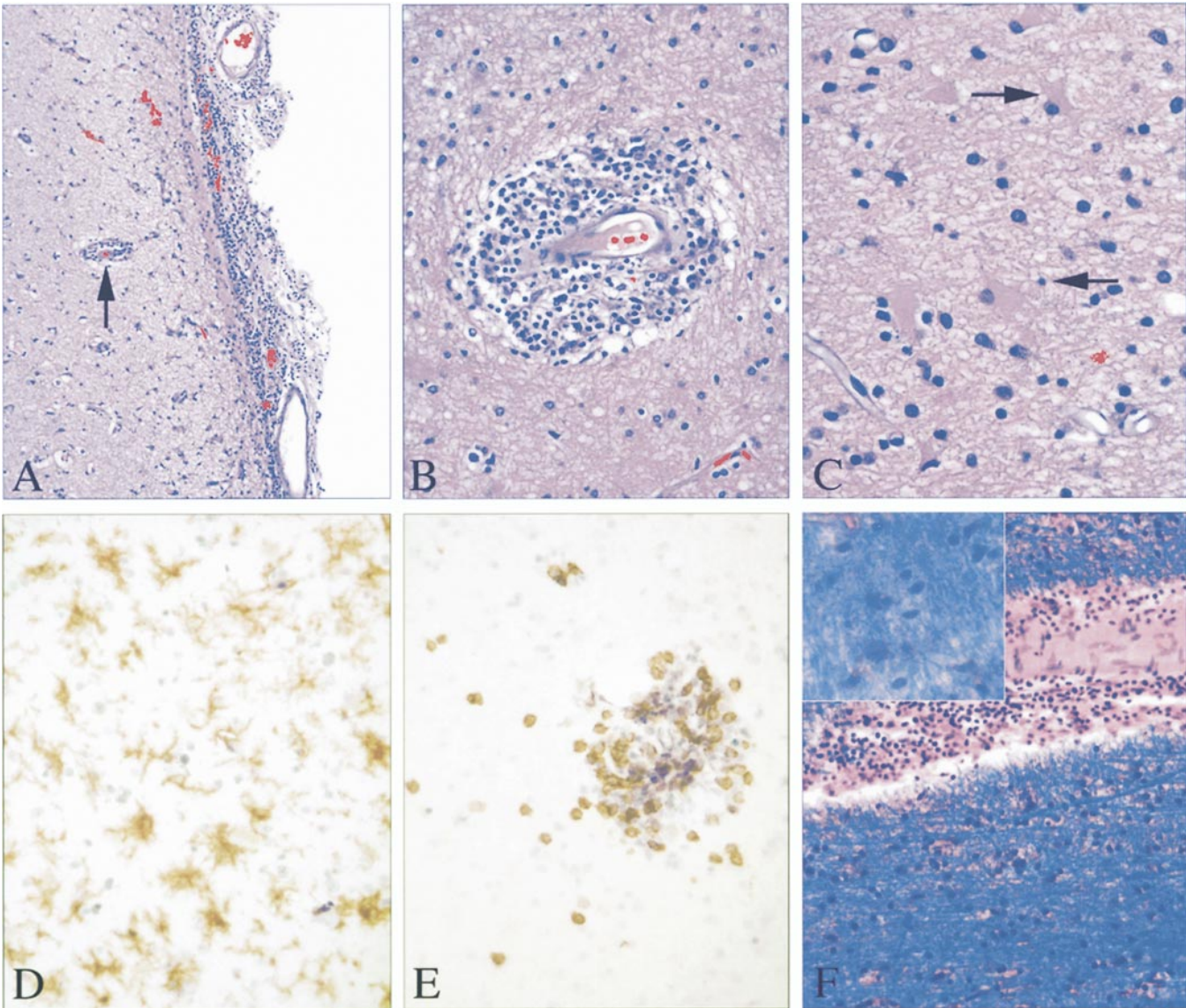


Figure 2. A, A lymphoplasmacytic infiltrate is present in the meninges and surrounding cortical vessels (arrow). The cortical surface is to the right (hematoxylin-eosin, original magnification $\times 20$). B, Perivascular and parenchymal inflammatory cells are especially prominent in the white matter (hematoxylin-eosin, original magnification $\times 200$). C, Reactive gliosis is prominent in the white matter (hematoxylin-eosin, original magnification $\times 400$). D, Human leukocyte antigen-DR-positive activated microglia are present throughout the white matter (original magnification $\times 200$). E, Cytotoxic CD8⁺ T lymphocytes are prominent around vessels and infiltrating through brain parenchyma (original magnification $\times 200$). F, Luxol fast blue stains highlight the preservation of myelin in the white matter, even in the presence of perivascular and parenchymal (inset) inflammation and gliosis (original magnification $\times 200$).

COMMENT

Brucellosis is a major zoonosis infamous for its protean clinical manifestations.^{1,4} *Brucella melitensis* remains an important human pathogen in endemic regions, most notably the Mediterranean basin, Arabian Peninsula, and Indian subcontinent. Brucellae are facultative intracellular, nonmotile, gram-negative coccobacilli that are capable of replication within mononuclear phagocytes. Most commonly, ingestion of infectious products is followed by hematogenous dissemination, residence in the reticuloendothelial system, and subsequent involvement of any organ system. Accordingly, the most common symptoms are nonspecific (fever, malaise, anorexia, and headaches), gastrointestinal (abdominal pain, nausea, vomiting, and diarrhea), and hematologic (anemia, leukopenia, thrombo-

cytopenia, and clotting disorders). Pathologic changes are most often identified in the reticuloendothelial system.

Involvement of the central nervous system is seen in 4% to 13% of patients with brucellosis.^{2,3,5} Meningitis, meningoencephalitis, polyradiculoneuritis, and cranial nerve palsies are the most frequent features of neurobrucellosis. To our knowledge, diffuse involvement of the white matter with a pattern resembling a leukoencephalopathy has not been described. Involvement of the white matter is rare in neurobrucellosis. In 2 earlier reports, discrete white matter lesions were initially confused with multiple sclerosis.^{6,7} One research group hypothesized that the infection resulted in a demyelinating process.⁷ However, the white matter involvement in our case was more diffuse and symmetric than that in these earlier cases, and de-

myelination was not an important histopathologic feature. Instead, we found a florid reactive astrogliosis and marked activation of microglia in association with an extensive infiltration by cytotoxic T lymphocytes. Cytotoxic T lymphocytes play a key role in the immune response to *Brucella* in murine models and in humans.^{8,9} Although generally beneficial, excessive or autoimmune cytotoxic T-cell responses have also been documented in a wide range of central nervous system diseases.¹⁰ The abundance of immune cells and the paucity of organisms in this case suggest that the clinical deficits and magnetic resonance image abnormalities associated with neurobrucellosis may be mediated at least in part by immunopathogenic mechanisms associated with direct T-cell-mediated cytotoxic injury of the white matter and cerebral cortex with concomitant microglial activation.

Our inability to detect organisms directly in the infected tissue is not unusual. Gram stains of tissues are generally negative in cases of brucellosis, and cultures are positive in less than 25% of cases. Currently, identification of antibodies against *Brucella* in the serum or cerebrospinal fluid is the most sensitive and specific way to detect the presence of the organism. Enzyme-linked immunosorbent assays are considered superior to agglutination assays, particularly for the detection of chronic disease or neurobrucellosis.⁵ In the present case, cerebrospinal fluid titers of antibodies directed against *B abortus* (the organism most commonly screened for in a search for antibodies directed against *Brucella*) were positive despite the absence of such antibodies in serum.

This case of meningoencephalitis in a 65-year-old Iranian man was caused by *B melitensis* infection and was complicated by the formation of a brain abscess and the

development of acute myelitis. Although neurobrucellosis is extremely rare in the United States, it should be considered in the differential diagnosis for patients suffering from chronic neurologic diseases, particularly patients with an appropriate travel history. The present case highlights the chronic, slowly progressive nature of the disease in some patients, the possibility of predominantly white matter involvement in the central nervous system, and the absence of specific systemic symptoms, signs, or laboratory findings in some cases. In addition, the prominence of CD8⁺ T cells suggests that direct cytotoxic damage could mediate some of the observed white matter changes in cases of chronic disease.

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