BABESIOSIS,
HUMAN EHRLICHIOSIS AND
HUMAN ANAPLASMOSIS:
POTENTIAL TRANSFUSION COMPLICATIONS

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BABESIOSIS, HUMAN EHRLICHIOSIS AND HUMAN ANAPLASMOSIS: POTENTIAL TRANSFUSION COMPLICATIONS

BABESIOSIS

Community Acquisition

Babesiosis is a zoonotic infection in which ticks transmit *Babesia* organisms from a vertebrate reservoir to an incidental human host, where the protozoan parasitizes erythrocytes, causing fever and hemolytic anemia. In New York State, babesiosis is typically caused by *Babesia microti*, a small (1.0 – 2.5µm) protozoan that is transmitted to humans, as incidental hosts, primarily by nymphal deer ticks (*Ixodes scapularis*), whose bites may not be noticed. It appears that attachment for many hours is necessary for transmission to humans. The white-footed mouse is the primary enzootic reservoir; tick larvae acquire organisms through feeding on infected mice, and continue to carry parasites as they develop into nymphs. Although mice are responsible for propagation of the parasites, deer nourish and transport adult ticks, spreading them from place to place. Many patients who require hospitalization are aged 50 to 89. Risk factors for community acquisition include outdoor activities, such as gardening, hunting, fishing and playing golf; exposure to wildlife; and lack of insect repellent.

The first case in New York was identified in 1980 and the disease was designated a reportable communicable disease by the New York State Department of Health in 1986. The incidence of babesiosis in New York State has increased significantly in recent years, more than doubling from 95 cases in 2004 to 269 in 2010. Shelter Island and the South Fork of Long Island, east of the Shinnecock Canal, have been and continue to be hyperendemic foci. More than half of the community-acquired cases have been concentrated in Suffolk, Dutchess and, to a lesser extent, Westchester Counties. However, cases have been reported in ever expanding areas. Outside New York, other high-incidence areas include coastal regions of southern New England, especially Martha’s Vineyard and Nantucket Island in Massachusetts and Block Island in Rhode Island. Cases have also been reported in the Midwest, especially in Minnesota and Wisconsin, and Northwest. Some cases from the Northwest appear to be due to *B. duncani*, which can be distinguished from *B. microti* by polymerase chain reaction (PCR) and indirect fluorescent antibody (IFA) assay, but not by morphologic criteria. Seven states are considered to have endemic babesiosis (Connecticut, Rhode Island, New York, Massachusetts, New Jersey, Minnesota, and Wisconsin). Sporadic cases of babesiosis have also been reported in Europe, Africa, Asia and South America. Seroprevalence in blood donors (based on IFA) in hyperendemic foci has been reported to be about 1% (endemic areas in Connecticut) to 4% (Shelter Island). Increasing incidence may reflect the human habitat
encroaching on the tick habitat, but increasing awareness and reporting may contribute to tabulated incidence figures.

Transfusion Transmission

Transmission of *Babesia* by transfusion of blood or blood components obtained from apparently healthy donors occurs rarely, but with increasing frequency. Many infected donors are asymptomatic, febrile, and have a normal hemoglobin/hematocrit on the day of donation. Donors are questioned about a possible history of babesiosis and a positive response or linkage to a transfusion-transmitted case results in indefinite deferral, but most affected persons are never aware of their infection. No *B. microti* assay has been approved by the FDA for blood donor screening. Transfusion-associated cases have been linked to (liquid stored) red blood cells, frozen deglycerolized red blood cells, and platelet concentrates (presumably from residual red cells). More than 160 transfusion-associated cases have been reported in the U.S. While the incidence per population is higher in Connecticut and Rhode Island, the plurality of cases has been in New York - 45 - more than twice as any other state. Nationwide, the vast majority of transfusion-associated cases have been linked to a donor who resides in, or has traveled to, an endemic area and is infected, but asymptomatic. Parasitemia in asymptomatic persons is usually seasonal, but may be protracted (durations exceeding a year), and transfusion transmission may occur year round. New York State saw 22 cases of transfusion-associated babesiosis from 2004 to 2009. Although most of these have occurred in the downstate area, patients throughout the state should be considered to be at risk.

Risk Factors for Symptomatic Disease

*Babesiosis* should be considered in transfusion recipients who present with a posttransfusion febrile illness in areas, such as New York State, where blood donors may be at risk for babesiosis. All recipients are at risk, but physicians should be aware of the increased risk for the disease to be symptomatic in elderly, very young (<1 year), asplenic, and immunocompromised patients. Because *Ixodes* ticks can carry the agents of Lyme disease and human anaplasmosis, these diagnoses should also be considered in patients in whom babesiosis is suspected. Transfusion transmission of Lyme disease has not been reported. The blood of one donor subsequently found to be co-infected with babesiosis and Lyme disease transmitted babesiosis, but not Lyme disease. A few cases of human anaplasmosis have been linked to transfusion, and the incidence of reported cases of the disease, caused by the bacterium *Anaplasma phagocytophilum*, is nearly that of babesiosis in New York.

Symptoms

Reported incubation periods generally range from 2.5 to 9 weeks among transfusion recipients, and 1 to 6 weeks following a tick bite in community-acquired cases. Many immunocompetent infected adults and children experience asymptomatic infection or a viral-like illness so mild that infection is only diagnosed incidentally by laboratory testing for other reasons or a generic screen. The initial clinical features of babesiosis, if they do present, are similar to those of malaria and some viral infections. Symptoms of babesiosis are nonspecific and include fever, chills, pallor, diaphoresis, fatigue, myalgias, anorexia, back and abdominal pain, nausea, vomiting, and diarrhea. Splenomegaly, hepatomegaly and/or jaundice may be observed upon physical examination. Possible complications in severe babesiosis (occurring almost exclusively in immunocompromised patients, often in association with high-density parasitemia) can include acute respiratory failure, disseminated intravascular coagulation, congestive heart failure, renal failure, and coma.
Laboratory Findings

Parasitemia on peripheral smear is generally 1 to 2% in symptomatic patients, but may be as high as 10% in severe cases, or even higher in patients who have significant immunosuppression due to a comorbidity. Laboratory findings include hemolytic anemia with reticulocytosis; thrombocytopenia; elevated liver enzymes and bilirubin; and decreased or absent haptoglobin. Serum protein electrophoresis usually demonstrates a polyclonal gammopathy, consistent with B-cell hyperactivity in response to T-cell suppression by Babesia. Renal dysfunction, demonstrated by proteinuria and elevated BUN and creatinine, can occur in cases of severe hemolysis.

Diagnosis

Babesiosis should be considered in the differential diagnosis of unexplained posttransfusion hemolytic anemia or thrombocytopenia. Diagnosis is based on clinical and laboratory findings. A specific diagnosis of acute babesiosis can generally be made by microscopic identification of the organism on Wright- or Giemsa-stained peripheral blood smears; multiple blood smears should be examined. Pathognomic tetrad forms of merozoites are diagnostic, but may not always be seen (see Figure 1). The ring-like forms of Babesia (see Figure 2) can be confused with those of Plasmodium falciparum malaria if the microscopy is not performed by trained personnel. Some patients may be initially treated for the wrong parasite.

Detection of B. microti DNA in blood by PCR or real-time PCR is more sensitive than peripheral blood smear. Molecular testing is useful when parasitemia is below the limit for visual detection or when trained personnel are not available to perform microscopic identification. A negative PCR result does not necessarily rule out infection because PCR may not be sufficiently sensitive to detect organisms at very low levels. Detection of antibodies to Babesia by IFA indicates infection at some time during the past year, but may not detect a recent infection.

Figure 1. Babesia microti (tetrad form) in human peripheral blood (courtesy of the Wadsworth Center Parasitology Laboratory)
Physicians are required to report confirmed cases of babesiosis to the local health department of the patient’s county of residence. Laboratories must report cases to the State Department of Health using the electronic reporting system and submit all positive specimens to the Wadsworth Center for confirmation. In January 2011, babesiosis became nationally notifiable, which means that state health departments, in states where babesiosis is a reportable communicable disease, should report confirmed cases to CDC in a non-identifying fashion, for surveillance purposes, using a standardized case definition.

Treatment

The preferred treatment for babesiosis is a combination of atovaquone and azithromycin given orally for seven to ten days; this approach is better tolerated than previous regimens. For immunocompromised patients with babesiosis, successful outcomes have been reported with administration of atovaquone combined with higher than usual doses of azithromycin. Clindamycin and quinine are the treatment of choice for patients with severe Babesia infections, but often have side effects.

Supplementation with partial or complete red blood cell exchange transfusion is indicated in patients with high-density parasitemia (>10 percent), significant hemolysis, or renal, hepatic or pulmonary compromise; such treatment may be lifesaving in these situations. Mortality rates of 3 to 5% have been reported, but may be as high as 20% in severely affected immunocompromised patients. None of the transfusion-associated cases in New York since 2004 were fatal. Outcomes in New York patients may be better than elsewhere because our clinicians are more likely to recognize infection because the incidence is so high. Ample professional educational information has been widely distributed or made available electronically, and testing is readily available free of charge at the Wadsworth Center.

Prevention

In the absence of donor screening, implicated donors are identified only after transmission has occurred in at least one recipient. Some assays are in development with an eye toward use of an automated platform suitable for mass testing. Serologic assays under development include IFA and ELISA; NAT by PCR may also be used. The first IND for donor screening is underway at Rhode Island Blood Center, which is employing a CMV-like approach performing IFA on selected units for use in at-risk recipients, including infants and pediatric sickle cell disease patients. Other centers are pursuing other possible approaches, in collaboration with industry.
HUMAN EHRLICHIOSIS

Community Acquisition

Human ehrlichiosis formerly known as human monocytic ehrlichiosis, is caused by rickettsial bacteria, including *Ehrlichia chaffeensis* and *Ehrlichia ewingii*. Like Lyme disease, ehrlichiosis is believed to be spread by ticks, such as the Lone Star tick (*Amblyomma americanum*). Organisms enter the skin by tick bite inoculation and spread through lymphatic and hematogenous routes. The bacteria then invade their target cells – macrophages and monocytes (*E. chaffeensis*) or granulocytes (*E. ewingii*).

People who spend time outdoors in tick-infested areas from April through November are at greatest risk for exposure; peak incidence is from May to July. Three-quarters of ehrlichiosis patients are males whose exposure risk stems from recreational and/or occupational activities. Cases of ehrlichiosis have been reported in 47 states; most have occurred in the southeastern or south central United States. In New York State, most diagnosed cases of ehrlichiosis have occurred on Long Island and in the Hudson Valley. In the Northeast, *E. chaffeensis* is the predominant causative organism.

Transfusion Transmission

Transmission via transfusion of infected human blood and blood components is theoretically possible, but has not been reported.

Symptoms

The incubation period for ehrlichiosis is generally four to ten days after a tick bite, with a mean of seven days. The most common symptoms are fever, myalgias, chills, malaise, anorexia and headache. A rash is present in about half of adult patients with ehrlichiosis. In later stages of the illness, other symptoms, such as nausea, vomiting, arthralgia, and confusion may arise. Severe complications include acute renal insufficiency, central nervous system abnormalities (20 percent) and gastrointestinal hemorrhage. The median duration of the illness is 23 days. Ehrlichiosis may occasionally be life-threatening in immunocompromised patients. The fatality rate for diagnosed ehrlichiosis cases is three percent, even despite antibiotic treatment.

Laboratory Findings

Thrombocytopenia (50,000 – 140,000/µL) and leukopenia (1,300 – 4,000/µL) are common, often striking, laboratory findings. Patients may also exhibit elevated liver enzymes and anemia.
Diagnosis

PCR performed on EDTA-anticoagulated blood is rapidly becoming the test of choice because of the timeliness of its results. Its sensitivity is 60 to 80 percent. However, it should be noted that most labs do not test for *E. ewingii*. The most sensitive method to confirm a diagnosis of ehrlichiosis is detection of seroconversion or a fourfold increase in antibody titer during the convalescent phase. Serologic testing for antibodies to *E. chaffeensis* and PCR testing are available free of charge at the New York State Department of Health’s Wadsworth Center. (See page 10 for information on specimen submission.) Because *E. ewingii* has not been cultured, antigens are not available for use in serologic testing. Therefore, *E. ewingii* infection is diagnosed by molecular methods (PCR).

In Wright- or Giemsa-stained peripheral blood smears, clusters of *E. chaffeensis* may occasionally (< 7 percent of patients) be observed in cytoplasmic vacuoles of monocytes called morulae because the microcolonies resemble mulberries (see Figure 3). Blood smears oruffy coat smears should be prepared within four hours of collection, preferably at the facility where the patient is receiving care.


Physicians are required to report confirmed cases of ehrlichiosis to the local health department of the patient’s county of residence. Laboratories must report cases to the State Department of Health using the electronic reporting system.

Treatment

Doxycycline is the treatment of choice and is generally well tolerated. Patients usually recover rapidly. Because of the risk of adverse effects, children under eight years of age and pregnant women should be treated in consultation with an infectious disease expert. Serologic tests can confirm the diagnosis, but patients with symptoms and/or laboratory findings consistent with ehrlichiosis should be treated empirically. However, prophylactic post-exposure treatment following a tick bite is not recommended in the absence of symptoms.
HUMAN ANAPLASMOSIS

Community Acquisition

Human anaplasmosis, formerly known as human granulocytic ehrlichiosis and human granulocytic anaplasmosis, is caused by the bacterium *Anaplasma phagocytophilum*, formerly known as *Ehrlichia phagocytophila*. Humans usually acquire anaplasmosis through the bite of a tick that has fed on an infected rodent. Major proven reservoir hosts are the white-footed mouse and wood rats. After a tick bite, the bacterium migrates to the bone marrow and spleen, and then invades its target cells – neutrophils.

Anaplasmosis was first recognized as a disease in the United States in the mid-1990s, but did not become a reportable disease until 1999. In New York, the incidence of reported cases of the disease is nearly that of babesiosis. In 2010, there were 231 cases of anaplasmosis reported in New York State. According to the Centers for Disease Control and Prevention, the number of anaplasmosis cases has increased steadily. The number of reported anaplasmosis cases in 2010 was more than double the number of cases in 1999. There have been more than 1,700 cases in New York reported from 2001 to date.

People who spend time outdoors in tick-infested areas between April and November are at risk for exposure. Anaplasmosis peaks in July and again in November, coinciding with the nymphal and adult stages of the *Ixodes scapularis* tick in the eastern U.S. Endemic areas in the U.S. (northeastern, mid-Atlantic, upper midwest and Pacific northwest states), Europe (Slovenia, Czech Republic, Sweden, Norway, Switzerland) and Asia all correspond to the territory of *Ixodes scapularis* ticks. Male anaplasmosis patients outnumber female patients by two to one. Anaplasmosis is infrequently diagnosed in children. In New York State, most diagnosed cases of anaplasmosis have occurred on Long Island and in the Hudson Valley. Because *Ixodes* ticks can carry the agents of Lyme disease and babesiosis, these diagnoses should also be considered in patients in whom anaplasmosis is suspected.

Transfusion Transmission

Although none occurred in New York, at least four cases of human anaplasmosis have been linked to transfusion. In December 1998, a case of probable transfusion-transmitted anaplasmosis was reported in Minnesota, linked to an asymptomatic donor who had a history of Lyme disease in 1997 and extensive deer tick bites in the fall of 1998. In November 2007, a case of probable transfusion-transmitted anaplasmosis in Minnesota was reported, linked to an asymptomatic donor. The donor, who resided in an endemic area, had no known tick bites, but recalled various outdoor activities in wooded areas before the donation. The implicated red blood cell (RBC) unit was donated 15 days prior to transfusion. Twenty days after the RBC transfusion, the patient began experiencing symptoms, and a peripheral blood smear demonstrated cytoplasmic inclusions of *A. phagocytophilum* morulae in the patient’s neutrophils. Whole blood specimens collected from the patient at this time were positive for *A. phagocytophilum* infection by
PCR and IFA. Retained segments from the implicated RBC unit were positive for evidence of *A. phagocytophilum* infection by both PCR and IFA. A confirmed case of anaplasmosis linked to transfusion was reported in 2010 in Wisconsin. Ten days after transfusion of two units of leukocyte-reduced RBCs, the patient began experiencing symptoms, and a peripheral blood smear demonstrated intracytoplasmic morulae. PCR testing performed on the patient’s blood confirmed *A. phagocytophilum* DNA. Segments were available for both of the units received. One was positive for *A. phagocytophilum* DNA by PCR; the other was negative. Serologic testing of the implicated donor for anti-*A. phagocytophilum* was positive (IgM titer 40 and IgG titer 1,040), consistent with a recent infection. In a presumptive case in Wisconsin, a patient was transfused with 19 units of leukocyte-reduced apheresis platelets from 18 donors, six units of leukocyte-reduced RBCs and five units of granulocytes. Seven days after discharge, the patient developed symptoms and a peripheral blood smear demonstrated intracytoplasmic morulae. PCR testing confirmed *A. phagocytophilum* DNA. The patient had no risk factors for community acquisition. No implicated donor was identified, but only 26 of the 29 donors could be tested. The implicated units were all leukoreduced, indicating that leukoreduction cannot be relied upon to prevent transmission.

**Symptoms**

The incubation period for anaplasmosis is generally four to ten days after a tick bite, with a mean of seven days. Common symptoms include fever, myalgias, severe headache, malaise, and arthralgias. More than one-third of patients complain of nausea and some have a nonproductive cough or stiff neck. A rash is rare – present in <10 percent of patients with anaplasmosis. Central nervous system infections are also rare in anaplasmosis, but severe complications can arise, such as respiratory insufficiency, septic shock-like illness, hemorrhage, and multiorgan failure. In most patients, clinical symptoms resolve within 30 days. However, nearly half of recognized cases require hospitalization, and up to 17 percent require intensive care. Anaplasmosis is rarely fatal (< 1 percent of cases), but it is associated with opportunistic pathogens, such as fungi and viruses.

**Laboratory Findings**

Laboratory features include thrombocytopenia, leukopenia, mild anemia and elevated liver enzymes in the first seven days of illness. Serum creatinine may also be elevated.

**Diagnosis**

Diagnosis should be attempted during the active phase of disease to facilitate early treatment that can prevent complications. Observation of characteristic cytoplasmic inclusions (morulae) in neutrophils may assist in diagnosis (see Figure 4). However, they may be few in number or may be absent, especially after the first week. PCR performed on EDTA-anticoagulated blood is rapidly becoming the test of choice because of the timeliness of its results and its sensitivity. Serologic testing for antibodies to *A. phagocytophilum* and PCR testing are available free of charge at the New York State Department of Health’s Wadsworth Center. (See page 10 for information on specimen submission.)
Physicians are required to report confirmed cases of anaplasmosis to the local health department of the patient’s county of residence. Laboratories must report cases to the State Department of Health using the electronic reporting system.

**Treatment**

Doxycycline is the treatment of choice and is generally well tolerated. Patients usually recover rapidly. Because of the risk of adverse effects, children under eight years of age and pregnant women should be treated in consultation with an infectious disease expert. Serologic tests can confirm the diagnosis, but patients with symptoms and/or laboratory findings consistent with anaplasmosis should be treated empirically. However, prophylactic post-exposure treatment following tick bites is not recommended in the absence of symptoms.
SEROLOGIC AND MOLECULAR TESTING AT THE NEW YORK STATE DEPARTMENT OF HEALTH’S WADSWORTH CENTER

Serologic testing for antibodies to *B. microti*, *E. chaffeensis* and *A. phagocytophilum*, as well as PCR testing, are available free of charge at the New York State Department of Health’s Wadsworth Center.

- Each specimen should be submitted in an individual New York State laboratory mailer available through local county health departments. The Wadsworth Center also can supply lists of acceptable specimens and mailing containers with all required documentation forms.

- Specimens submitted to the Parasitology Laboratory for morphological and real-time PCR testing for *B. microti* or to the Tick-borne Bacteria Laboratory for PCR testing for *E. chaffeensis* and *A. phagocytophilum*, should be EDTA-preserved whole blood (purple-top tube).

- For all serology, acute serum specimens drawn prior to treatment should be submitted to the Diagnostic Immunology Laboratory. Convalescent specimens, drawn at least three weeks after an acute (or other previous specimen) should be collected, for serology only. Specimens for serology should be collected in a red-top tube, spun to remove cells and serum poured off into a separate tube for submission. Acute specimens should be sent to the Wadsworth Center immediately.

- For shipping address or for questions about specimen submission, contact the Wadsworth Center Parasitology Laboratory, Tick-borne Bacteria Laboratory, or Diagnostic Immunology Laboratory at (518) 474-4177. You may also visit [http://www.wadsworth.org/docs/infectious_phs.shtml](http://www.wadsworth.org/docs/infectious_phs.shtml) for more detailed instructions and an Infectious Disease Requisition (IDR) form.
PERTINENT LITERATURE


Linden JV, Olkowska D, Grima KM, Manning ML. A series of 23 transfusion-associated babesiosis cases (abstract). Transfusion 2010;50(Suppl 2S):40A.


