

## ***Anaplasma phagocytophilum***

### **Disease Agent:**

- *Anaplasma phagocytophilum* (formerly referred to as *A. phagocytophila*, *Ehrlichia phagocytophila* and *Ehrlichia* spp)

### **Disease Agent Characteristics:**

- Obligate, intracellular, Gram-negative bacterium with tropism for neutrophils
- Order: Rickettsiales; Family: Anaplasmataceae
- Size: 0.5-0.8 µm × 1.2-3 µm
- Nucleic acid: A circular DNA of about 1500 kb
- Physicochemical properties: The rickettsiae are susceptible to 1% sodium hypochlorite, 70% ethanol, glutaraldehyde, formaldehyde, and quaternary ammonium disinfectants and are sensitive to moist heat (121°C for at least 15 min) and dry heat (160-170°C) for at least 1 hour.

### **Disease Name:**

- Human granulocytic anaplasmosis (HGA); previously known as human granulocytic ehrlichiosis (HGE)

### **Priority Level:**

- Scientific/Epidemiologic evidence regarding blood safety: Moderate
- Public perception and/or regulatory concern regarding blood safety: Low
- Public concern regarding disease agent: Low/moderate in focal endemic areas

### **Background:**

- Emergent; first described in 1994 as the causative agent of HGE (now HGA). HGA became a reportable disease in 1999. The majority of cases reported to CDC (90%) come from New York, Connecticut, New Jersey, Rhode Island, Minnesota and Wisconsin. Incidence of reported cases has increased almost 5-fold from 2000 to 2010 probably due to increased awareness resulting in better diagnosis and surveillance, and/or spread of infected ticks.

### **Common Human Exposure Routes:**

- Tick-borne zoonosis; humans are accidental/incidental hosts.
- Transmitted by the same tick vectors as those that transmit Lyme disease and babesiosis: *Ixodes scapularis* ticks in the Northeast and upper Midwest (also known as the deer tick or the black-legged tick); incidence peaks synchronized to tick seasons in affected areas. The majority of cases occur during the summer months with a peak observed during June/July.
- *I. pacificus* (western black-legged tick) transmits the infection in the western US, while *I. ricinus* and *I. persulcatus* are the principal vectors in Europe and Asia, respectively.
- Reports of transfusion transmission increasing

### **Likelihood of Secondary Transmission:**

- None documented

### **At-Risk Populations:**

- Individuals at enhanced risk for exposure to infected ticks through outdoor activity, including those involved in hiking, gardening, clearing brush, etc.
- The frequency of reported cases is higher among males and anyone over 40 years of age.
- A compromised immune system due to cancer treatments, advanced HIV infection, prior organ transplants, or immune suppression may increase the risk of severe outcome.

### **Vector and Reservoir Involved:**

- Ticks of genus *Ixodes* (*I. scapularis*, *I. pacificus*, *I. ricinus*, *I. persulcatus*)
- The tick nymph is primarily responsible for transmission of Lyme disease, babesiosis, and HGA, although adult stages also can transmit infection. Because of its small size, the bite may not be noticed and consequently the tick may not be removed before disease transmission occurs.
- White-footed mice (*Peromyscus leucopus*), raccoons (*Procyon lotor*), gray squirrels (*Sciurus carolinensis*), Northern short-tailed shrews (*Blarina brevicauda*), Eastern chipmunks (*Tamias striatus*), and several other small mammals have demonstrated competency as reservoir hosts in the US.
- White-tailed deer (*Odocoileus virginianus*) harbor a similar strain of *A. phagocytophilum* not associated with human infections; therefore, they are not considered competent reservoirs for strains that cause HGA.

### **Blood Phase:**

- The bacteremia lasts for days to a few weeks after the occurrence of symptoms.
- The duration and frequency of asymptomatic bacteremia have not been documented.

### **Survival/Persistence in Blood Products:**

- Viable organisms have been recovered from anticoagulated, refrigerated whole blood from infected patients for 2 weeks and transmitted by transfusion in animal models (sheep) after 13 days. In a single case (abstract), RBCs stored for 30 days from a seropositive/PCR-negative donor transmitted the organism to a recipient who at diagnosis was seroconverting, PCR positive, and febrile. Among fully published cases, *A. phagocytophilum* survived for 8-15 days in packed RBCs.

### **Transmission by Blood Transfusion:**

- Eight cases have been reported; seven in the US and one in Europe. Specific details of these cases are presented in the attached Table. Key elements follow below.
  - Cases are being recognized more frequently: seven of eight cases reported since 2007.

- Patients ranged in age from 36 to 85 years of age, but majority (n = 6) were over 50 years of age. Gender distribution equally split; 4 female and 4 male.
- HGA generally suspected by identifying morulae (microcolonies of *Anaplasma*) on blood smear; confirmation by PCR and/or seroconversion
- Patients treated with doxycycline demonstrated rapid improvement and resolution of infection.
- Implicated donors ranged in age from 42 to 81 years of age and most reported risk factors for exposure to *A. phagocytophilum* including tick bites, time spent in an endemic area, or febrile illness prior to donation. For most cases, index donation was PCR positive.
- Leukoreduced, as well as non-leukoreduced products implicated in transmission. Products ranged in age from 5 to 30 days at the time of transfusion. Most recent case implicated a leukoreduced apheresis platelet product.
- A cluster of cases of apparent human-to-human transmission of *A. phagocytophilum* infection (and the first report of HGA in China) associated with blood contact was reported in 2008. The infection was not confirmed by blood smear or culture in the index patient, but *A. phagocytophilum* DNA was amplified from and sequenced from the patient who had been bitten by a tick, and nine family members or healthcare workers who had been in close contact. All nine reported contact with the patient's blood; seven had contact with respiratory secretions. The index patient died before seroconversion, but all nine contacts seroconverted.

#### Cases/Frequency in Population:

- Seroprevalence:
  - Blood donors: 11.3% of Westchester County, New York donors, 0.5% of Wisconsin donors, and 3.5% of Connecticut donors had antibodies to *A. phagocytophilum*.
  - Residents: 0.4% in northern California, 3.4% in New York, 14.9% in northwestern Wisconsin had antibodies to *A. phagocytophilum*.
- Incidence: During 2000-2010, the reported annual US incidence increased from 1.4 to 6.1 cases per million population. In 2010, 1,716 cases were reported to the CDC. The number of infected individuals and clinical cases are likely much higher due to underreporting and lack of recognition of this often nonspecific infection.

#### Incubation Period:

- 7-10 days from tick bite to bacteremia and acute symptoms

#### Likelihood of Clinical Disease:

- Low to moderate
- Males outnumber females by a 2:1 ratio
- Immunocompromised and elderly patients are at greater risk to develop more severe manifestations of disease.

#### Primary Disease Symptoms:

- Nonspecific febrile illness characterized by high-grade fever (> 39°C), rigors, generalized myalgias, severe headache, and malaise often accompanied by thrombocytopenia, leukopenia, and elevated liver transaminases occurs approximately 5-21 days after a bite from an infected tick.
- Anorexia, arthralgias, nausea, nonproductive cough, and rash are sometimes present.
- Median duration of illness is 9 days (1-60 days).

#### Severity of Clinical Disease:

- Generally not severe
- Severe cases are characterized by prolonged fever, acute renal failure, gastrointestinal bleeding, septic shock-like illness, rhabdomyolysis, respiratory insufficiency, and secondary opportunistic infections.

#### Mortality:

- <1% mortality

#### Chronic Carriage:

- Not documented in humans. Of interest, a chronic, subclinical infection of almost one year was reported in dogs inoculated with a human isolate from New York.

#### Treatment Available/Efficacious:

- Tetracyclines (e.g., doxycycline) are therapeutically effective.
  - Rifampin may be effective based on limited data in those who cannot take tetracyclines (e.g. pregnant women or those with allergy).

#### Agent-Specific Screening Question(s):

- No specific question is in use.
- Not indicated at this time because relatively few cases of transfusion transmission have been reported.
- No sensitive or specific question is feasible. In endemic areas, a question on exposure to tick bites has been shown to be ineffective in distinguishing *Babesia*-infected from *Babesia*-uninfected donors. This question probably also lacks sensitivity and specificity for *A. phagocytophilum*.

#### Laboratory Test(s) Available:

- No FDA-licensed blood donor screening test exists.
- Options for laboratory testing include blood smear microscopy, cell culture, IFA, EIA, and NAT
  - During the first week of infection, examination of peripheral blood smears may reveal morulae in the cytoplasm of white blood cells in 25-75% of patients. Morulae are most frequently observed in granulocytes.
  - Specialized cell culture techniques can be used to amplify the infection and observe infected cells. Sensitivity is > 55% in the first week, falling to 33% during week two.

- ELISA is considered the gold standard serologic test. A four-fold rise in IgG antibody levels to *A. phagocytophilum* antigen is considered diagnostic for a recent infection. Sensitivity ranges from 22-44% in the first week after onset to > 95% at 3 weeks and beyond.
- PCR detection primarily during first week of infection (67-90%); thereafter sensitivity rapidly declines, particularly after administration of appropriate antibiotics.

#### Currently Recommended Donor Deferral Period:

- No FDA Guidance or AABB Standard exists.
- In an individual who receives treatment, prudent practice would be to defer donor until signs and symptoms are gone and treatment is complete.
- Based on the natural history of infection, it may be prudent to defer an untreated individual for a minimum of 90 days (30 days beyond the longest duration of illness).

#### Impact on Blood Availability:

- Agent-specific screening question(s): Not applicable
- Laboratory test(s) available: Not applicable; serologic testing, if implemented in the future, could result in a deferral rate of 3-5% in selected collection areas.

#### Impact on Blood Safety:

- Agent-specific screening question(s): Not applicable
- Laboratory test(s) available: Not applicable

#### Leukoreduction Efficacy:

- Preliminary studies indicate that leukoreduction reduces, but does not eliminate transmission risk associated with *A. phagocytophilum*. Recent transfusion cases implicating leukoreduced products support this observation.

#### Pathogen Reduction Efficacy for Plasma Derivatives:

- Not expected to be transmitted by plasma derivatives

#### Other Prevention Measures:

- Tick avoidance measures (e.g., long pants, long sleeves, insect repellent)
- Riboflavin/Light has been effective in inactivating *Orientia tsutsugamushi*, a similar obligate intracellular pathogen.

#### Suggested Reading:

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**TABLE. Summary of reported cases of transfusion-transmitted anaplasmosis**

Case Characteristics	Case #1 Eastlund et al., 1999	Case #2 Kemperman et al./CDC, 2008	Case #3 Bachowski et al., 2009	Case #4 Annen et al., 2012
<b>Year of case</b>	1998	2007	2008	2007
<b>Location</b>	Minnesota	Minnesota	Minnesota	Wisconsin
<b>Recipient</b>	75-year-old male	68-year-old male	85-year-old female	51-year-old female
<b>Underlying Conditions</b>	Rheumatoid arthritis with GI bleeding	Chronic renal insufficiency, psoriatic arthritis, ankylosing spondylitis, and corticosteroid therapy—underwent elective knee arthroplasty and synovectomy	Double heart valve replacement surgery	Hypertension, supraventricular tachycardia, renal insufficiency, degenerative joint disease, and multiple myeloma. Received two stem cell transplants from sibling donor.
<b>Products transfused</b>	2 U non-LR RBCs	34 U non-LR RBCs 4 U LR apheresis platelets 14 U FFP 7 U Cryo	6 U LR RBCs 2 U LR apheresis platelets 5 U FFP	5 U granulocytes 19 U LR apheresis platelets 6 U LR RBCs *all units irradiated
<b>HGA Diagnosis</b>	Morulae in neutrophils on blood smear; confirmed by PCR and serology (1:512)	Morulae in neutrophils on blood smear; confirmed by PCR, weakly positive (1:64) by IFA	Morulae on blood smear; confirmed by PCR	Morulae on blood smear; confirmed by PCR, but serology was negative
<b>Treatment &amp; Recovery</b>	Doxycycline; symptoms resolved within 24 hr post-treatment.	Doxycycline; rapid improvement within 1 wk	Not known	Doxycycline; recovered and discharged 13 days after admission
<b>Recipient Risk Factors</b>	No history of recent tick bites. Pre-transfusion sample negative for <i>A. p.</i> DNA and antibodies.	Three wks before hospitalization traveled to location where blacklegged ticks endemic; did not spend times outdoors and reported no tick bites	No prior tick exposure	None, largely housebound due to underlying illness
<b>Implicated Donor</b>	Female; positive by serology (1:2048); history of Lyme disease (1997) and extensive deer tick bites 2 mo before donation in 1998; recalled no associated illness	64-year-old female; index donation PCR positive, follow-up samples positive by serology at 1:512 and 1:256. Did not recall tick bite, but spent time in endemic areas (MN) within the month before donation; no history of fever or illness in month before donation	81-year-old male; lived in endemic areas of central MN; hospitalized for anaplasmosis day after implicated donation and was positive for anaplasmosis by smear and serology (IgG 1:512, IgM < 1:20); subsequent sample 1 month later positive by serology (IgG 1:128 in serum, 1:256 in plasma); also positive for Lyme borreliosis	Not identified. All donors except 2 associated with RBC and 1 associated with an apheresis platelet provided follow-up samples that tested negative (samples collected 3 to 10 mo after donation). Two RBC donors denied illness or tick bites. Stem cell donor not tested.
<b>Type and Age of Implicated Product</b>	Non-LR RBC 30 days	Non-LR RBC 15 days	LR RBC 29 days	Not identified
<b>Index product Tested?</b>	Residual blood from RBC bag negative by PCR	Post-donation samples collected at time of donation positive by PCR	No	N/A

<b>Case #5 Annen et al., 2012</b>	<b>Case #6 Jereb et al., 2012</b>	<b>Case #7 Alhumaidan et al., 2013</b>	<b>Case #8 Townsend et al., 2013</b>
2010	2010	2011	2012
Wisconsin	Ljubljana, Slovenia	Rhode Island	Connecticut
81-year-old female	36-year-old female	64-year-old male	41-year-old male
Rheumatoid arthritis; admitted for hip fracture	Pregnant, at 29 wks experienced preeclampsia and restriction of intrauterine growth; elective cesarean section followed by hemorrhagic shock	Chronic obstructive pulmonary disease, coronary artery disease, and recurrent gastritis with iron deficiency anemia.	Trauma, multiple gunshot wounds
2 U LR RBCs	6 U LR RBCs 2 U FFP	5 U LR RBCs	13 U LR RBCs 3 U LR apheresis platelets 9 U FFP
Morulae on blood smear; confirmed by PCR	Morulae on blood smear and a positive PCR; later seroconversion. Morulae and <i>A. p.</i> DNA also detected in bone marrow biopsy.	Morulae in PMNs on blood smear; confirmed by PCR, but negative by ELISA	Morulae on blood smear (23 days post transfusion); confirmed by PCR, but negative by IgM/IgG ELISA
Not stated; fully recovered	Doxycycline; dramatic improvement beginning day 4 of treatment	Doxycycline; infection cleared within 1 wk	Unknown; died due to underlying trauma
No known risk for tick bites	Denied history of tick bites; had not left house for several weeks before hospitalization. Pre-transfusion samples negative for <i>A.p.</i> DNA and antibodies	No recent history of tick exposure or illness. One week before hospitalization spent weekend on Cape Cod, but remained indoors. Owns indoor cat. No prior history of tick-borne disease.	History not available
53-year-old male; index donation positive by PCR, follow-up sample showed IgM titer of 1:40 and IgG titer of 1:1040. No recollection of tick bites, but hunted in wooded area 1 wk before donation. Owned a dog and field dressed animals. No signs or symptoms of illness immediately before donation	42-year-old male; index donation positive by PCR and serology (1:1024); sample from 118 d before index donation tested negative; lived in endemic area and reported several tick bites every year; reported having an acute self-limited febrile illness 2-3 weeks before implicated donation	81-year-old male; healthy with outdoor activities in endemic areas, but no reported tick bites; index donation positive by PCR and serology using ELISA with IgM > 17.4 (normal, < 1.0) and IgG > 12.1 (normal, 1.0)	52-year-old female; lived in an endemic area, healthy and asymptomatic at the time of donation, but reported tick bites 1-2 months prior to donation; positive by PCR 105 days post transfusion, but subsequently resolved; elevated IgG titers in the absence of IgM titers
LR RBC Not stated	LR RBC 8 days	LR RBC 12 days	LR apheresis platelet 5 days
Segment from implicated RBC unit positive by PCR	Stored, frozen plasma sample taken on day of donation positive by PCR and serology (1:1024)	Segment from implicated RBC unit positive by PCR and serology (ELISA)	No