Human Ehrlichiosis: A Case Study

JACK G SCHNEIDER

Ehrlichiosis is an infection of white blood cells that affects various mammals, including mice, cattle, dogs, and humans.[1] It was first reported in dogs in 1935, and the first human case was documented in the United States in 1986. Ehrlichia are obligate, intracellular bacteria that are transmitted by ticks to humans. They grow as a cluster (morula) in neutrophils (*Anaplasma phagocytophilum* and *E. ewingii*) and in monocytes (*E. chaffeensis*).[2,3] The infection may cause prolonged fever and general aches, and is characterized by leukopenia, cytopenia, and elevated liver transaminases.[4] In the first week of infection, ehrlichiae can be detected by finding intracellular aggregates on the blood/body fluid smears and various other laboratory findings. Immunofluorescent antibodies (IFA) titers and PCR are generally needed for confirmation and a definitive diagnosis. Early diagnosis is necessary as antibiotic treatment with doxycycline is very effective.[3,5]

ABBREVIATIONS: ALP = alkaline phosphatase; ALT, SGPT = alanine aminotransferase; AST, SGOT = aspartate aminotransferase; BUN = blood urea nitrogen; PTT = partial thromboplastin time; WBC = white blood cell count.

INDEX TERMS: amblyomma americanum; anaplasmosis; ehrlichiosis; ixodes scapularis.

Clin Lab Sci 2009;22(1):3

Jack G Schneider BS MT (ASCP) *is of Clarian Health Partners/ Indiana University School of Medicine, Division of Clinical Microbiology, Department of Pathology & Laboratory Medicine.*

The peer-reviewed Clinical Practice Section seeks to publish case studies, reports, and articles that are immediately useful, are of a practical nature, or contain information that could lead to improvement in the quality of the clinical laboratory's contribution to patient care, including brief reviews of books, computer programs, audiovisual materials, or other materials of interest to readers. Direct all inquiries to Bernadette Rodak MS CLS(NCA), Clin Lab Sci Clinical Practice Editor, Clinical Laboratory Science Program, Indiana University, Clarian Pathology Laboratory, 350 West 11th Street, 6002F, Indianapolis IN 46202. brodak@iupui.edu

Address for correspondence: *Jack G Schneider BS MT (ASCP), Clarian Pathology Laboratory, Clinical Laboratory Sciences Program, 350 W 11th Street, Indianapolis IN 46202. (812) 707-1773, (317) 491-6163 (fax). jgschneid@gmail.com.*

ACKNOWLEDGEMENTS: *The author would like to acknowledge J Stephen Dumler MD, Division of Medical Microbiology, Department of Pathology, The Johns Hopkins Medical Institutions; Michael Sever MD, Indiana University School of Medicine, Department of Pathology and Laboratory Medicine; and the Indiana University Clinical Laboratory Sciences faculty at Indiana University School of Medicine.*

CASE PRESENTATION

A 52-year-old avid outdoorsman has had multiple tick bites throughout his adult life. He experienced nonspecific malaise and fatigue for about one month. There were no fevers, chills, sweats, or weight loss. He was witnessed to have a generalized seizure and a marked decrease in mental status and was admitted into the hospital for thorough evaluation.

Due to sepsis-like presentation and clinical picture, he was treated empirically with broad-spectrum antibiotics, including vancomycin and doxycycline. Serum creatinine, blood urea nitrogen (BUN) levels, and hepatic enzymes were extremely high as a result of the patient's acute renal and liver failure respectively. As his illness progressed, pulmonary function diminished and mechanical ventilation was required, as was hemodialysis. Multiple serology tests revealed that the patient was negative for *Rickettsia*, *Borrelia* (Lyme disease), histoplasmosis, West Nile virus, and viral hepatitis.

Because of his history of frequent tick bites, doxycycline was initiated upon admission and continued throughout his work-up. Further evaluation revealed positive serology for *Ehrlichia chaffeensis* with an initial titer of 64 during the first week's test, 128 for the second week, and a maximum of 1024 by the third week.

The patient was eventually extubated and made an outstanding recovery. By the time of discharge, he was fully cognizant with clear thinking and normal speech. Liver function tests

improved and hematologic parameters returned to normal levels. Creatinine and BUN levels dropped to 2.7 and 44 respectively after the first week of dialysis. Hemodialysis was again implemented following discharge due to elevated BUN (84) and creatinine (7.0) (Table 1).

History of the disease

Since the end of the 20th century, several tick-borne diseases have been identified throughout the United States, including babesiosis, Lyme disease, and ehrlichiosis.[1]

Initially recognized in 1935, ehrlichiosis is caused by bacterial species in the genus *Ehrlichia*. Over time, veterinary pathogens within this genus were found to cause disease in a variety of animals. Currently, one species in Japan and three in the United States are known to cause disease in humans.[1] With the advent of new diagnostic methods, such as PCR and gene sequencing, other disease-causing species of *Ehrlichia* may be identified in the near future.

The genus *Ehrlichia* was named after the German bacteriologist Paul Ehrlich, who surprisingly did not contribute to its classification and discovery. It is a more recently recognized disease, with the first diagnosed case in the United States occurring in 1986 in a 51-year-old man from Detroit who had been exposed to ticks in a rural area of Arkansas.[1,6,7] Prior to this finding, the first ehrlichial pathogen was identified in Japan as causing Sennetsu Fever. This disease, caused by

Table 1. Selected laboratory data from patient on admission and discharge

	Patient admission laboratory values	Patient discharge laboratory values	Reference ranges
WBC	3.2 x 10^9/L	6.5 x 10^9/L	4.5-11.5 x 10^9/L
Hemoglobin	10.0 g/dL	10.2 g/dL	14-18 g/dL
Platelets	60 x 10^9/L	230 x 10^9/L	150 450 x 10^9/L
Partial thromboplastin time (PTT)	60.5 seconds	31.3 seconds	20-35 seconds
BUN	122 mg/dL	84 mg/dL	6-20 mg/dL
Serum creatinine	9.6 mg/dL	7.0 mg/dL	0.7-1.3 mg/dL
Aspartate aminotransferase (AST, SGOT)	1334 U/L	40 U/L	8-20 U/L
Alanine aminotransferase (ALT, SGPT)	300 U/L	77 U/L	10-40 U/L
Alkaline phosphatase (ALP)	274 U/L	360 U/L	53-128 U/L
Albumin	1.8 g/dL	1.5 g/dL	3.5-5.2 g/dL

Neorickettsia (Ehrlichia) sennetsu, is characterized by swollen lymph nodes and fever. Most cases involving this species are very rare outside East and Southeast Asia and have only been reported in parts of western Japan and Malaysia.[1,2]

Ehrlichiosis species represent a group of clinically similar, yet etiologically and epidemiologically distinct, diseases caused by *A. phagocytophilum, E. ewingii,* and *E. chaffeensis.* The former *Ehrlichia phagocytophila* is an organism that is similar to *E. equi* and is now referred to as *Anaplasma phagocytophilum* (which will be used throughout the rest of the discussion*).*[1,2,3] The remainder of this paper will focus on the types of ehrlichiosis that occur in the United States, along with common treatments and preventative measures.

Bacteriology/taxonomy

The *Ehrlichia* and *Anaplasma* genera are classified as small, gram-negative, and obligate intracellular cocci that infect leukocytes in humans and various animal species. They range from 1 to 3 µm in diameter and generally divide within white blood cells to form morulae, characteristic of this bacterial pathogen.[1,4,8]

These genera are found in the Anaplasmataceae family and include the following recognized species relevant to human infection: *E. ewingii, A. phagocytophilum,* and *E. chaffeensis.* Initially these species were classified according to the blood cell most commonly infected (i.e., monocyte or granulocyte). However, it has been found that more than one species may be responsible for the generic clinical presentation called "ehrlichiosis". [1,4]

Epidemiology and ecology

Through 2003, the CDC recorded at least 1508 cases of Human Monocytic Ehrlichiosis (HME) in the United States, yet there may be underreporting since the disease is not reportable in all states. Endemic regions, such as southeastern Missouri, estimated the annual incidence rates of HME to be as high as 158 cases per 100,000 population.[9]

The occurrences of these diseases seem to reflect the geographic distributions and seasonal activities of the tick vectors. Areas of the southeast, south-central, and mid-Atlantic regions correspond to places where *Amblyomma americanum* (Lone Star) ticks, the vector of *E. chaffeensis*, are plentiful. Most infections occur between May and July, which correspond to the peak feeding times of Lone Star ticks. Transmission occurs most often after bites of adult ticks, which generally are painless and leave no inflammatory skin lesion. This finding is quite different than in patients with Lyme disease where tick bites (*Ixodes scapularis*) tend to cause inflammation, followed by radial extensions (erythema migrans). [1,9]

Erlichiosis infection is quite distinct from several other tick-transmitted diseases in that it generally infects adults greater than 40 years of age, unlike Lyme disease and Rocky Mountain spotted fever, which occur often in children. Men generally are diagnosed with HME more frequently than women by a near ratio of two to one, possibly due to occupational and recreational exposure. Even though the average age of infection is 48, fatalities have also been reported in children. [1,9]

PATHOPHYSIOLOGY

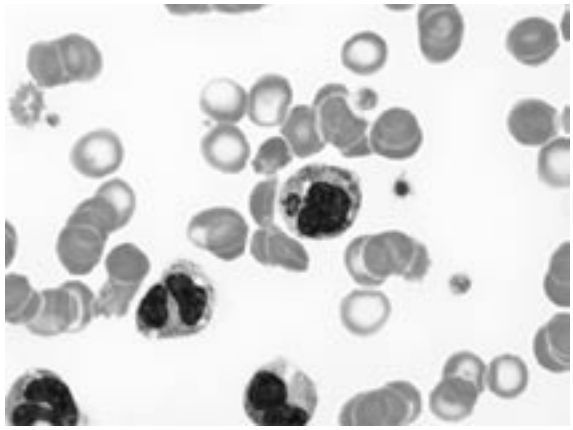
Means of replication

Generally, the *Ehrlichia* species tend to infect leukocytes, where they enter the cell by endocytosis and once inside the host cell, they inhibit the fusion of the phagosome and lysosome. They develop within the host cell vacuoles first as reticulate cells and then as dense-core cells, eventually being released by lysis of the cell. The inclusion body that contains the organism is called a morula, due to its "mulberry-shaped" appearance (Figure 1).[1,9-10]

Human Monocytic Ehrlichiosis

Caused by *E. chaffeensis*, animals in the southeastern and mid-Atlantic United States such as white-tailed deer, dogs,

Figure 1. *Anaplasma*



From Girgis G, Marler LM, Siders JA. Hematology Image Atlas CD-ROM, Indiana Pathology Images, 2008.

and foxes tend to serve as the animal hosts of this species. *E. chaffeensis* predominantly affects monocytes and macrophages, but it may also be seen rarely in the granulocytes of some patients with severe disease. The disease resembles Rocky Mountain spotted fever, except that a rash does not develop in over 67% of the cases.[6,10]

Human Granulocytic Anaplasmosis (HGA, formerly Granulocytic Ehrlichiosis-HGE)

Anaplasma phagocytophilum is the causative agent of this disease and is carried by the deer tick (*Ixodes scapularis*). Common animal hosts throughout the spring and summer include the white-footed mouse, among many other mammals. The pathogen that causes HGA primarily infects granulocytes (mainly neutrophils and rarely eosinophils) and is distributed in the upper Midwest and northeast United States.[9,11] This disease is similar to Human Monocytic Ehrlichiosis, except that mortality rates may be 10% higher.[10]

Ehrlichiosis Ewingii

This disease is named for its causative agent, *E. ewingii,* and is also carried by the Lone Star tick. Like HME, its animal hosts include white-tailed deer, dogs, and foxes throughout the spring and summer. It is commonly found in the southeastern, south-central, and mid-Atlantic United States, and primarily infects neutrophils and occasionally eosinophils.[9-10] The symptoms and signs are similar to HME and HGA, but infected patients usually have pre-existing medical conditions that cause immunosuppression such as splenectomies, HIV, and transplantations.[1]

CLINICAL FEATURES

Signs and symptoms

A person infected with ehrlichiosis generally presents with nonspecific signs and symptoms. In fact, it is possible that many infected people do not even seek treatment because the development of the illness may be mild and display no symptoms.[1]

Initial manifestation of infected patients commonly include fever, headaches, malaise, and muscle aches after an incubation period of approximately 5 to 10 days after the tick bite.[1] Other signs and symptoms include nausea, vomiting, cough, confusion, and occasionally a rash. Rashes are uncommon in adult patients with HME and HGA, but 60% or more of infected pediatric patients display this rash when infected with *E. chaffeensis*. Because most patients with ehrlichiosis present with fever and a severe headache, meningitis may be in the differential diagnosis.[1,9] The CSF profile in patients with ehrlichiosis is usually normal, but about 20% with signs of meningitis will have CSF pleocytosis similar to patients with viral or bacterial meningitis. Other diagnostic possibilities include malaria, babesiosis, Lyme disease, and Rocky Mountain spotted fever, suggested by a fitting epidemiological/travel history.[1,10]

If left untreated, ehrlichiosis can become very severe and possibly lead to renal failure, disseminated intravascular coagulation (DIC), respiratory distress, and prolonged fever. With *E. chaffeensis* infection being the most common and most severe, only two to three percent of patients actually die from this infection. [1,4]

Overall, the seriousness of HME is dependent on the immune status of the patient. Corticosteroid therapy, chemotherapy, and HIV infection are examples that may put the patient at a greater risk of fatal outcomes.[1,10,12]

Laboratory detection

Availability of confirmatory assays presents a huge challenge in diagnosing ehrlichial infections. Therefore, doctors tend to make their treatment decisions based on clinical and epidemiologic clues. In general, clinical laboratory tests supportive of ehrlichiosis include a low white blood cell count, low platelets, and elevated liver enzymes. The biggest clue is the peripheral blood smears containing morulae in the leukocytes on a Giemsa stain. [1,9-10]

Leukopenia, which involves decreased numbers in both neutrophils and lymphocytes, is usually resolved after the first few weeks of illness with proper treatment. The cytopenias cannot be explained by bacterial lysis of infected cells because they do not infect erythrocytes and platelets. However, the frequent detection of macrophage-rich inflammatory infiltrates within the liver, often along with hemophagocytic cells, suggests macrophage activation as the mechanism for both the cytopenias and the frequency of increased serum transaminases (ALT and AST). [8,13,14]

Standard media used in the microbiology laboratory is generally not successful in isolating Anaplasmataceae (*Anaplasma* and *Ehrlichia*). Therefore, laboratory confirmation requires serologic, molecular, or cell culture based methods. Serologic evaluations use indirect immunofluorescence assays (IFA), but these may not always detect the organism. Most patients demonstrate increased IgM or IgG titers by the second week of the illness, and IgG levels usually stay elevated up to 2.5 years after the acute illness. One disadvantage of this testing

is the likelihood of cross reactivity between the different species, hindering the epidemiological distinction between the ehrlichial infections.[1]

Polymerase chain reaction (PCR) may be used to detect the organism. The detection of *E. chaffeensis* DNA is based upon the amplification of specific genomic DNA sequences. The diagnosis of infection should not rely solely upon the PCR assay; however, a positive PCR result should be considered in conjunction with clinical presentation and additional diagnostic tests.[1,9,15] Above all, direct isolation is the most desired method but can be very time-consuming. *E. chaffeensis* and the *A. phagocytophilum* have been isolated from blood samples when inoculated to canine DH82 or human HL-60 cells. The organism usually grows within 7 to 36 days.[1]

Diagnosis

The diagnosis of ehrlichiosis requires not only laboratory evidence, but also a well-matched clinical history characteristic to the disease. Significant laboratory evidence includes a single IgG antibody titer of at least 256, a 4-fold rise in titer between the acute and convalescent stage, recovery of *E. chaffeensis* or *A. phagocytophilum* in culture, or detection of their nucleic acids in blood by PCR. These specific tests, along with common laboratory values and characteristics, help the physician to determine a definitive diagnosis. The patient case history previously described demonstrates why physicians should consider ehrlichiosis when patients present with fever, cytopenias, myalgia, leukopenia, thrombocytopenia, elevated liver enzymes, and above all, a history of tick bites or exposure.[1,4,6,9-10,15]

Treatment/prognosis

Most patients with ehrlichiosis respond favorably to treatment, particularly if the therapy is initiated early in the disease. Doxycycline is the antibiotic of choice and within 24 to 72 hours after treatment, fever generally goes down. After one week, white blood cell and platelet counts usually return to normal. Fatigue may persist for weeks to months, but depending on the health status of the individual, recovery time is generally short.[1,9-10]

Prevention and control

Ehrlichiae is spread by tick bites. Therefore, limiting one's exposure to ticks reduces the chances of infection. Common measures include using insect repellent, wearing clothing to

Table 2. Selected epidemiological and ecological features of ehrlichioses[9]

	Human Monocytic Ehrlichiosis (HME)	Human Anaplasmosis (Granulocytic Ehrlichiosis)	Ehrlichiosis "Ewingii"
Organism	*Ehrlichia chaffeensis*	*Anaplasma phagocytophilum*	*Ehrlichia ewingii*
Vector	Lone Star tick (*Amblyomma americanum*)	Deer tick/black-legged tick (*Ixodes scapularis*)	Lone Star tick (*Amblyomma americanum*)
Animal host	White-tailed deer, dogs, foxes, wolves	White-footed mouse, other mammals	White-tailed deer, dogs, foxes, wolves
Cells infected	Monocytes	Neutrophils	Neutrophils
Time of year	Spring/summer	Spring/summer	Spring/summer
Location	Southeastern, south-central, and mid-Atlantic US	Upper Midwest and northeast US, California, and Europe	Southeastern, south-central and mid-Atlantic US

cover skin, avoiding dense brush and long grasses, and constantly checking for ticks and removing any that be found on the body after being outside, especially in endemic areas. Studies have shown that a tick must be attached to the body for at least 24 hours in order to cause disease, so early removal may prevent the spread of infection (Table 2). [1,10,12,16]

CONCLUSION

Human ehrlichiosis is a rising disease that generally provides few clinical clues for diagnosis and may cause severe infections or even death in infected patients. It is imperative to be mindful of this disease and acknowledge common risk factors associated with it, such as traveling to endemic areas and tick bites. Fever, elevated liver enzymes, and cytopenias are all important laboratory findings when dealing with ehrlichiosis. Even if suspected patients are unaware of vector contact or merely traveling in endemic areas, administering doxycycline therapy is the best treatment when ehrlichiosis is suspected but not yet confirmed. [8,11,13]

Clin Lab Sci encourages readers to respond with thoughts, questions, or comments regarding this article. Email responses to brodak@iupui.edu. In *the subject line, please type "CLIN LAB SCI 22(1) CP SCHNEIDER". Selected responses will appear in the Dialogue and Discussion section in a future issue. Responses may be edited for length and clarity. We look forward to hearing from you.*

REFERENCES

1. CDC. Human Ehrlichiosis in the United States. Available from http://www.cdc.gov/ncidod/dvrd/ehrlichia/Q&A.htm. Accessed 2007 Mar 17.
2. Dumler JS. Reorganization of genera in the families Rickettsiaceae and Anaplasmataceae in the order Rickettsiales: unification of some species of Ehrlichia with anaplasma, Cowdria with Ehrlichia and Ehrlichia with Neorickettsia, descriptions of six new species combinations and designation of Ehrlichia equi and 'HE agent' as subjective synonyms of Ehrlichia phagocytophila. Int J Syst Evol Micr. 2001; 51:2145-65.
3. Reddy VB: Extracorpuscular defects leading to increased erythrocyte destruction-nonimmune causes. In: Rodak BF, Fritsma GA, Doig K, editors. Haematology: clinical principles and applications, 3rd edition. St. Louis, MO: Elsevier; 2007:319.
4. MedlinePlus Medical Encyclopedia. Ehrlichiosis. Available from http://www.nlm.nih.gov/medlineplus/ency/article/001381.htm. Accessed 2007 Mar 17.
5. Dumler JS, Walker DH: Tick-borne ehrlichioses: more of them higher incidences, and greater clinical diversity. Lancet Infect Dis 2001; 21-28 (preview edition).
6. Nauschuetz WF. Zoonotic and rickettsial infections. In: Mahon CR, Lehman DC, Manuselis G, editors. Textbook of diagnostic microbiology, 3rd edition. St. Louis, MO: Elsevier, 2007:1068-9.
7. Maeda K, Markowitz N, Hawley RC, and others. Human infection with Ehrlichia canis, a leukocytic rickettsia. N Engl J Med 1987;316(14):853-6.
8. Cunha BD. emedicine. Ehrlichiosis. Available from http:// www.emedicine.com/ med.htm. Accessed 2007 Mar 17.
9. Stone JH, Dierberg K, Aram G, Dumler JS. Human monocytic ehrlichiosis. JAMA 2004; 292:2263-70.
10. Microbiology and immunology online/University of South Carolina. Rickettsia, Ehrlichia, Coxiella and Bartonella. Available from http:// pathmicro.med.sc.edu. Accessed 2007 Mar March 17.
11. Dumler, JS. Anaplasma and Ehrlichia infection. NY Acad Sci 2005; 1063:361-73.
12. Mayo Clinic. Ehrlichiosis. Available from http://www.mayoclinic.com/health.ehrlichiosis. Accessed 2007 Mar 17.
13. Dumler JS, Dawson JE, Walker DH. Human ehrlichiosis: hematopathology and immunohistologic detection of Ehrlichia chaffeensis. Hum Pathol 1993;24:391-5.
14. Henderson AR, Moss DW: Enzymes. In: Burtis CA, Ashwood ER, editors. Tietz fundamentals of clinical chemistry, 5th edition. St. Louis, MO: Saunders;2001: 354-89.
15. Walker DH. Diagnosing human ehrlichiosis. ASM News. 2000;66:287-290.
16. Maender J, Tyring S. Treatment and prevention of rickettsial and ehrlichial infections. Dermatol Ther 2004; 17:499-504.