

icterus developed, and the patient died of shock and multiorgan failure 9 hours after hospitalization, despite treatment with a mixture of antibiotics, which contained doxycycline.

Rickettsiae of the spotted fever group were isolated by the shell vial technique from the blood of the three patients. Sequences of polymerase chain reaction-amplified fragments of 16SrRNA (1440 bp), citrate synthase (382 bp), and *rompA* (590 bp) genes of the isolates show 100% similarity with the homologous sequence of Israeli spotted fever rickettsia (4,7,8).

All three patients lived in semirural areas, along the River Tejo (Setubal District). None had left Portugal during the previous year. Although none had a tache noire, contact with ticks cannot be excluded. The absence of tache noire is typical in Israeli spotted fever (6). These findings indicate that the geographic distribution of Israeli spotted fever is wider than had been thought and includes the Iberian Peninsula. Because initial signs and symptoms of the disease are particularly uncharacteristic and appropriate treatment may be delayed, this rickettsia can cause life-threatening disease.

**Fatima Bacellar,\* Lorenza Beati,† Ana França,‡  
José Poças,§ Russell Regnery,†  
and Armindo Filipe\***

\*Centro de Estudos de Vectores e Doenças Infecciosas, Instituto Nacional de Saúde Dr. Ricardo Jorge, Aguas de Moura, Portugal; †Centers for Disease Control and Prevention, Atlanta, Georgia, USA; ‡Hospital Garcia da Orta, Almada, Portugal; and §Hospital São Bernardo, Setubal, Portugal

### References

1. Tavares L, Botas J, Antunes F, Araújo FC. A febre escarotodular em Portugal. *O Médico* 1985;113:838-46.
2. Bacellar F, Regnery RL, Nuncio S, Filipe AR. Genotypic evaluation of Rickettsial isolates recovered from various species of ticks in Portugal. *Epidemiol Infect* 1995;114:169-78.
3. Regnery RL, Spruill CL, Plikaytis BD. Genotypic identification of *Rickettsiae* and estimation of interspecies sequence divergence for portions of two rickettsial genes. *J Bacteriol* 1991;173:1576-89.
4. Fournier PE, Roux V, Raoult D. Phylogenetic analysis of spotted fever group *Rickettsiae* by study of the outer surface protein *rOmpA*. *Int J Sys Bacteriol* 1998;48:839-49.
5. Roux V, Raoult D. Phylogenetic analysis and taxonomic relationships among the genus *Rickettsia*. In: *Rickettsiae and rickettsial diseases at the turn of the third millennium*. Raoult D, Brouqui P, editors. Paris: Elsevier; 1999. p. 52-66.
6. Goldwasser RA, Steiman Y, Klingberg W, Swartz TA. The isolation of strains of *Rickettsiae* of the spotted fever group in Israel and their differentiation from other members of the group by immunofluorescence methods. *Scand J Infect Dis* 1974;6:53-62.
7. Roux V, Raoult D. Phylogenetic analysis of the genus *Rickettsia* by 16S rDNA sequencing. *Res Microbiol* 1995;146:385-96.
8. Roux V, Rydkina E, Ereemeeva M, Raoult D. Citrate synthase gene comparison, a new tool for phylogenetic analysis and its application for *Rickettsiae*. *Int J Sys Bacteriol* 1997;47:252-61.

### **Avoiding Misdiagnosis of Malaria: A Novel Automated Method Allows Specific Diagnosis, even in the Absence of Clinical Suspicion**

**To the Editor:** We report three cases of malaria to illustrate a novel method that allows diagnosing the disease, even if clinicians do not suspect it or request malaria smears. Lack of clinical suspicion is a well-known factor for malaria misdiagnosis and may be responsible for almost 40% of deaths from *Plasmodium falciparum* infections in industrialized countries (1-3). A recent study from Canada showed that in 59% of cases malaria was initially misdiagnosed, and in 16% three or more physician contacts occurred before malaria smears were ordered (4).

Early diagnosis of malaria relies crucially on clinical suspicion. A clinician suspecting the disease has to explicitly request malaria smears. This problem has not been solved with the advent of several methods alternative to microscopy, including recently introduced rapid dipstick tests (5). Performing any of these tests blindly without a specific request is impractical. On the other hand, routinely performed laboratory tests in the work-up of febrile patients, e.g., automated full blood counts, have so far detected only nonspecific changes, such as anemia or thrombocytopenia, which are associated with many other conditions (6). These changes on their own are therefore not specific enough to trigger malaria smears without an explicit request.

New automated full blood counts-analyzers incorporate flow-cytometric principles. The Cell-Dyn 3500 (Abbott, Santa Clara, CA) uses scattered laser-light of leukocytes at four different angles to generate a white-blood-cell differential (7). Monocytes and neutrophils may

ingest birefringent depolarizing malaria pigment that can be detected by the instrument. The appearance of monocytes (purple dots) above the separation line, in the eosinophil area (green dots), is a highly specific sign of the presence of ingested malaria pigment and consequently malaria.

A study from South Africa investigating 224 directed samples for malaria diagnosis found a sensitivity of 72% and specificity of 96% (8). In Portugal, we observed 45 positives in 120 directed samples. So far, all cases identified by microscopy showed the typical changes in the full-blood-count plots, suggesting a near 100% sensitivity in imported malaria cases. Several thousand full-blood-count plots from patients with a wide range of underlying pathologic features did not show such changes, making them highly specific for malaria diagnosis. However, the changes may persist for some time despite clinical and parasitologic cure, as pigment-containing monocytes may remain in the circulation for 2 to 3 weeks (9). Consequently, the observed changes may not necessarily indicate acute disease but may persist during convalescence.

We report three cases in which clinical suspicion did not lead to the request of a malaria diagnostic test. The final diagnosis of malaria was made only because of the changes observed in the color monitor of the Cell-Dyn 3500. As part of a preliminary investigation of this new method, we reviewed all full-blood-count plots at 24-hour intervals. During a 2-week period, three full-blood-count granularity/lobularity plots compatible with malaria were identified and the full-blood-count results and clinical notes were reviewed. The principal symptoms in the three cases were fever and aches in bones and muscles, case 1; complications of assault, case 2; and feeling generally unwell (from drug abuse), case 3. In all cases, the full-blood-count results were within normal ranges, except for a thrombocyte count of 23,000 in case 2. In cases 1 and 3, the patients were discharged with a clinical diagnosis of flulike syndrome and drug abuse-related problems, respectively, while in case 2, the patient was to be admitted with a diagnosis of assault-related injuries. As attending clinicians had not requested malaria smears, we performed blood films on the recovered specimens that confirmed a diagnosis of malaria. (Case 1: *P. ovale*, 10,000  $\mu$ l; case 2: *P. falciparum*,

9,000  $\mu$ l; case 3: *P. falciparum*, 1,500  $\mu$ l). In case 2, our findings permitted appropriate treatment in the emergency room; in the other two cases, it allowed patients to be contacted at home. All three patients (two male, one female) were of Black African origin but lived in Portugal. They had returned to Portugal after visiting Africa (Angola and Guinea). None of them had taken malaria prophylaxis during their journey.

Anisotropic malaria pigment has been the basis for several microscopy methods for malaria diagnosis (10). However, sensitivities are similar to that of conventional microscopy, and these methods have to be ordered specifically. In contrast, automated full-blood-count is regarded as routine for febrile patients, and the new automated method has the potential to detect additional, unsuspected cases, in which clinical suspicion did not lead to requests for malaria testing. If further studies validate this technique, the instrument could be modified to specifically flag such results, which would alert laboratory staff to perform blood films on these samples, even in the absence of a clinician's request. Finally, if software algorithms are adjusted to enumerate pigment-containing leukocytes, the usefulness of this indicator as a prognostic marker (11) could be further evaluated. The instrument may greatly facilitate quantification of pigment-containing leukocytes, which have been determined by time-honored but cumbersome microscopy.

**Thomas Hänscheid,\* Bernadino G. Pinto,\*  
Isabel Pereira,\* José Melo Cristino,\* and  
Emilia Valadas†**

\*University Hospital Santa Maria, Lisbon, Portugal;  
and †London School of Hygiene and Tropical  
Medicine, London, United Kingdom

### References

1. World Health Organization. World malaria situation in 1994. *Wkly Epidemiol Rec* 1997;36:269-274.
2. Day KP. Malaria: a global threat. In: Krause RM, editor. *Emerging Infections*. New York: Academic Press; 1998. p. 463-97.
3. Greenberg AE, Lobel HO. Mortality from *Plasmodium falciparum* malaria in travelers from the United States (1959-1987). *Ann Intern Med* 1990;113:326-7.
4. Kain KC, Harrington MA, Tennyson S, Keystone JS. Imported malaria: prospective analysis of problems in diagnosis and management. *Clin Infect Dis* 1998;27:142-9.
5. Hänscheid T. Diagnosis of malaria: a review of alternatives to conventional microscopy. *Clin Lab Haematol* 1999;21:235-45.

6. Giacomini T, Lusina D, Foubard S, Baledent F, Guibert F, Le Penne MP. Dangers of hematological automated analysis for malaria diagnosis. *Bull Soc Pathol Exot* 1991;84:330-3.
7. de Grooth BG, Terstappen LW, Puppels GJ, Greve J. Light-scattering polarization measurements as a new parameter in flow cytometry. *Cytometry* 1987;8:539-44.
8. Mendelow BV, Lyons C, Nhlangothi P, Tana M, Munster M, Wypkema E, et al. Automated malaria detection by depolarization of laser light. *Br J Haematol* 1999;104:499-503.
9. Day NPJ, Thi Diep P, Thi Ly P, Xuan Singh D, Phu Loc P, Van Chuong L, et al. Clearance kinetics of parasite and pigment-containing leukocytes in severe malaria. *Blood* 1996;88:4694-700.
10. Lawrence C. Laveran remembered: malaria haemazoin in leucocytes. *Lancet* 1999;353:1852.
11. Phu NH, Day N, Thi Diep P, Ferguson DJP, White NJ. Intraleucocytic malaria pigment and prognosis in severe malaria. *Trans R Soc Trop Med Hyg* 1995;89:200-4.

### The First Reported Case of *Aerococcus* Bacteremia in a Patient with HIV Infection

**To the Editor:** We report the first case of *Aerococcus viridans* bacteremia in a patient with HIV infection. Two species in the genus *Aerococcus* have been implicated as rare pathogens in humans. *A. urinae* causes urinary tract infections; the other species, *A. viridans*, a gram-positive coccus considered a contaminant in cultures, has been associated with human infections that included bacteremia (1,2), septic arthritis (3), and infectious endocarditis (4,5). Widely distributed in the environment, the organism has been recovered from dust, vegetables, and crustaceans (6) and was isolated from different areas in a hospital (recovery room, intensive care unit, delivery room, treatment room, premature nursery) and from numerous objects (7).

We describe the first case of *A. viridans* bacteremia in a patient with HIV. A 34-year-old man without notable medical history sought medical attention after several weeks of epigastric midabdominal pain associated with a 15-lb weight loss; the pain did not respond to antacid medications. The patient said that he did not have fever, chills, night sweats, or history of transfusions and did not use alcohol, tobacco, or drugs. He had engaged in homosexual activity 2 to 3 years earlier.

Physical examination showed moderate cachexia and low-grade fever (38.8°C) associated with tachycardia, but the heart and lung

examination was otherwise normal. The abdomen was soft, flat, and tender to palpation in the midabdominal epigastric area, without hepatosplenomegaly, guarding, or rebound tenderness. No other abnormalities were identified. The patient was admitted to the hospital, and the initial set of routine blood cultures (Bactec 9240 instrument, Becton Dickinson, Sparks, MD) showed no growth. On hospital day 2, he began to have severe rigors, along with persistent fever. A second set of blood cultures drawn at that time grew paired gram-positive cocci in less than 24 hours. The patient was empirically started on penicillin G, and cefotaxime was added shortly thereafter because of the possibility of intermediately resistant pneumococcus. The rigors responded to antibiotic treatment, and a third set of blood cultures showed no growth. The negative blood cultures before and after appropriate antimicrobial therapy and the short time to detection (which suggests a large initial inoculum) led us to believe that the organism in this case was a true pathogen and not a contaminant.

The patient's work-up included a normal abdominal computer tomography; abdominal ultrasound showed nonobstructing cholelithiasis. Laboratory tests demonstrated anemia of chronic disease diagnosed by a hematocrit of 25% associated with a low reticulocyte production index, high serum ferritin, and an elevated erythrocyte sedimentation rate (91 mm/hr), with polyclonal hypergammaglobulinemia and hypoalbuminemia on serum protein electrophoresis. Stool samples were negative for occult blood, and serologic tests showed no *Helicobacter pylori* antibodies. The patient's total lymphocyte count was 300 cells/ $\mu$ l, HIV serologic testing by enzyme-linked immunosorbent assay and Western blot was positive, and flow cytometry revealed an absolute CD4+ T-lymphocyte count of 19 cells/ $\mu$ l, with an HIV-1 retroviral titer of 280,000 by polymerase chain reaction. Gallium scanning was negative for *Pneumocystis carinii* pneumonia and gastrointestinal lymphoma. A follow-up endoscopy showed esophageal ulcers, with disruption of the mucosal barrier. Blood cultures were negative for cytomegalovirus or mycobacteria, but the aerobic isolate initially reported as paired gram-positive cocci was later identified as *A. viridans*.

The identification of *A. viridans* was made on