Answer to Photo Quiz: Tertian Malaria

(See page 1509 in this issue [[doi:10.1128/JCM.00093-12](https://doi.org/10.1128/JCM.00093-12)] for photo quiz case presentation)

The patient suffered from tertian malaria. The thin Giemsa-stained blood smear revealed several stages of *Plasmodium vivax* infection (see Fig. 1A, B, and C in the Photo Quiz): young trophozoites (delicate ring form) in normal-sized erythrocytes without Schüffner's dots (labeled "a" in Fig. 1 in the Photo Quiz), adolescent trophozoites with a vacuole (labeled "b"), male microgametocytes with a dispersed nucleus (labeled "c"), and more-developed trophozoites prior to the initiation of schizogony (nuclear division) (labeled "d"). Stages b to d were seen in enlarged erythrocytes with Schüffner's dots. At first sight, the filiform structures (labeled "e") looked like spirochetes; a concurrent bacterial infection with *Borrelia recurrentis* was suspected. Therefore, treatment with doxycycline was added to the treatment with chloroquine, which was used against intraerythrocytic stages of *P. vivax*. In some areas of North Africa, including Ethiopia and Sudan, *B. recurrentis* is endemic and the symptoms of relapsing fever are similar to those observed in malaria (4). However, the slender structures were later identified as microgametes. The presence of a nucleus (dark area) is the main characteristic, which helps in the differentiation since in borreliae no nucleus is visible (6). In plasmodiae, eight microgametes develop from one microgametocyte by exflagellation, and this usually takes place in the gut of an *Anopheles* mosquito. But a significant time lag between blood collection and blood smear preparation may favor the induction of the gametogenesis (exflagellation) outside the mosquito (2). *In vitro*, exflagellation is activated by a drop in temperature and a concomitant pH increase (1). These changes occur when blood is collected and left exposed to air either in an unstopped tube or on a microscopic slide (2, 6). As the CO₂ level in the blood rapidly falls, to equilibrate with the surrounding air, the pH of the blood rises and exflagellation may begin (6). *In vivo*, the drop in temperature has been identified as an obligatory inducer, but there is at least one further gametocyte-activating factor, namely, xanthurenic acid, which may contribute to the exflagellation in the mosquito (1). Microgametes are not observed in freshly prepared blood smears, and their presence may suggest a poor handling of the blood specimen in terms of timing (2, 6). Therefore, it is crucial that specimens are processed within less than 1 h after blood collection in order to avoid artifacts (3). In addition to spirochetes, microgametes may also be confused with trypanosomes. Laboratory staff should know their appearance in order to avoid errors in diagnosis and the unnecessary usage of antibiotics.

Following treatment with chloroquine for 2 days, the patient rapidly improved, fever disappeared, and C-reactive protein decreased within 3 days to 59 mg/liter. After exclusion of glucose-6-phosphat-dehydrogenase deficiency, primaquine was given for 2 weeks to eradicate the parasite at dormant stages in the liver (5). The identification of the slender structures as microgametes and a

negative broad-range PCR from blood ruled out the suspected relapsing fever, and doxycycline was discontinued.

REFERENCES

1. Billiker O, Miller AJ, Sinden RE. 2000. Determination of mosquito blood-meal pH in situ by ion-selective microelectrode measurement: implications for the regulation of malarial gametogenesis. *Parasitology* 120:547–551.
2. Bullock-Iacullo S. 2010. Detection of blood parasites: parasitology, p 9.8.1.4. In Garcia LS (ed), *Clinical microbiology procedures handbook*, 3rd ed. ASM Press, Washington, DC.
3. Bullock-Iacullo S. 2010. Preparation of thin blood films: parasitology, p 9.8.2.1. In Garcia LS (ed), *Clinical microbiology procedures handbook*, 3rd ed. ASM Press, Washington, DC.
4. Cadavid D, Barbour AG. 1998. Neuroborreliosis during relapsing fever: review of the clinical manifestations, pathology, and treatment of infections in humans and experimental animals. *Clin. Infect. Dis.* 26:151–164.
5. Karplus R, Johnston SP. 2006. Images in clinical medicine. *Plasmodium vivax* microgametes. *N. Engl. J. Med.* 354:1064.
6. Tembhare P, Shirke S, Subramanian PG, Sehgal K, Gujral S. 2009. Exflagellated microgametes of *Plasmodium vivax* in human peripheral blood: a case report and review of the literature. *Indian J. Pathol. Microbiol.* 52:252–254.

Annette Blaich

Division of Clinical Microbiology
University Hospital
Basel, Switzerland

Stefan Erb

Division for Infectiology and Hospital Epidemiology
University Hospital
Basel, Switzerland

Paul Bourbeau

Division of Laboratory Medicine
Geisinger Medical Center
Danville, Pennsylvania, USA

Reno Frei

Division of Clinical Microbiology
University Hospital
Basel, Switzerland

Alicia Rovó

Division of Diagnostic Hematology
University Hospital
Basel, Switzerland

Address correspondence to Annette Blaich, blaicha@uhbs.ch.

Copyright © 2012, American Society for Microbiology. All Rights Reserved.
[doi:10.1128/JCM.00100-12](https://doi.org/10.1128/JCM.00100-12)