Late relapse of imported Plasmodium ovale Malaria: A case report

Rechute tardive d'un paludisme à *Plasmodium ovale* : A propos d'un cas.

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RÉSUMÉ

Nous rapportons le premier cas tunisien d'une rechute d'un paludisme à *Plasmodium ovale* survenue trois ans après un séjour en Afrique subsaharienne.

Un tunisien âgé de 29 ans a consulté en Septembre 2011 pour une fièvre, des myalgies et des céphalées évoluant depuis huit jours et ce malgré un traitement par les antibiotiques. A l'interrogatoire, le patient a déclaré qu'il avait résidé trois ans auparavant en Côte-d'Ivoire pendant six mois, où il a eu un accès palustre de paludisme qui a été traité par l'artéméther-luméfantrine. Le patient a rapporté qu'il n'a pas eu d'autres séjours en pays endémiques et n'a pas eu de transfusion sanguine. Un autre examen microscopique du sang à la recherche du *Plasmodium* a été négatif. Le diagnostic a été établi 17 jours après l'apparition des symptômes. L'examen microscopique du frottis sanguin a confirmé la présence de *Plasmodium ovale* avec une parasitémie inférieure à 0,1%. Le patient a été traité par l'artéméther-luméfantrine, suivi de la primaquine. Cette observation met l'accent sur la possibilité d'une rechute de certaines espèces plasmodiales et souligne l'importance de répéter les examens microscopiques du sang en cas de suspicion de paludisme.

Mots-clés

Paludisme, *Plasmodium ovale,* rechute, primaquine.

SUMMARY

We report the first case of an imported *Plasmodium ovale* relapse in a Tunisian man who developed malaria three years after leaving sub-Saharan Africa.

A 29-year-old Tunisian man consulted in September 2011 because of a fever, myalgia, and headache that had begun eight days earlier and persisted despite treatment with oral antibiotics. On questioning, the patient stated that he had resided three years ago for six months in Ivory Coast, where he acquired malaria. He was treated with artemether-lumefantrine. The patient said he had no recent travel to any other malaria-endemic area and had not received a blood transfusion. A first microscopy of peripheral blood smears was negative for malaria parasites. The diagnosis was established 17 days after onset of symptoms. A repeat microscopic examination of blood smears confirmed the presence of *Plasmodium ovale* with a parasitemia lower than 0.1%. The patient was treated with artemether-lumefantrine, followed by primaquine.

This case emphasizes the possibility of relapse of some plasmodial species. It highlights the importance of repeating microscopic examination of blood when the diagnosis of malaria is suspected.

Key-words

Malaria, *Plasmodium ovale*, relapse, primaquine.

Malaria is an infectious disease caused by a protozoan parasite of the genus *Plasmodium* (*P.*) which includes five species (*P. falciparum*, *P. vivax*, *P. ovale*, *P. malariae* and *P. knowlesi*) infecting humans (1). *Plasmodium* is transmitted by the bite of an infective female *Anopheles* mosquito (2). Infections also can be acquired through exposure to infected blood products or by congenital transmission (2). Although malaria typically becomes clinically apparent within one month of infection, relapses can occur years after the last presumed exposure (3). We report the first case of an imported *P. ovale* relapse in a Tunisian man who developed malaria three years after the primary infection in sub-Saharan Africa.

OBSERVATION

A 29-year-old Tunisian man was admitted to the emergency department in September 2011 because of a high fever (up to 40°C), myalgia and headache that had begun eight days earlier and persisted despite treatment with several antibiotics and paracetamol. He reported shaking chills, nausea and dry cough. Fever episodes occurred every 48 hours, with profuse sweating followed by abrupt resolution. He had no other significant medical history. On presentation, he was pale. Physical examination was normal; the liver and spleen were not palpable. Blood pressure and other vital signs were normal. On admission, laboratory tests showed WBC at 5800/mm^3, with 71% neutrophils and 13% lymphocytes, a hemoglobin of 12 g/dL and hematocrit of 30.0%. Other laboratory abnormalities included a thrombocytopaenia of 54000/mm^3, an elevated C-reactive protein of 106 mg/L, and leukocyturia. Chest X-rays and abdominal ultrasound were normal. The Widal test was negative.

On questioning, the patient stated that he had resided 3 years ago for 6 months in Ivory Coast, where he acquired malaria that was confirmed with laboratory tests. The patient did not recall which specie was identified. He was treated with only artemether-lumefantrine. He said he had not recent travel to sub-Saharan Africa or any other areas endemic for malaria; he had no unexplained episodes of fever during the last 3 years since immigrating to Ivory Coast and had not received any blood transfusions. A first microscopy of peripheral blood smears was negative for malaria parasites. The diagnosis was established 17 days after onset of symptoms. A repeat microscopic examination of Giemsa-stained thick and thin blood smears in a specialized laboratory confirmed the presence of *P. ovale* with a parasitemia lower than 0.1%. Infected blood cells showed *P. ovale* characteristics, such as oval shape, slightly fimbriated aspects, and coarse Schuffner's stippling even in early trophozoite stage. The cytoplasm of the growing parasite was thick, compact and usually not ammoeboid (Figure 1).

The OptiMal-IT® test, a rapid diagnostic malaria test targeting the lactate dehydrogenase was negative. Diagnosis of *P. ovale* was confirmed by polymerase chain reaction (PCR). The DNA was extracted with QIAamp DNA Blood Kit (Qiagen,USA) according to the manufacturer's instructions. The small subunit ribosomal RNA gene 18S rRNA was amplified by a semi-nested PCR using primers described by Snounou (4). The PCR products were electrophoresed on 3% ethidium bromide stained agarose gel (Figure 2). The patient was treated with artemether lumefantrine (Coartem®), followed by primaquine. The screen for glucose-6-phosphate dehydrogenase (G6PD) deficiency was negative and he took 15 mg base primaquine daily for 14 days. Malaria parasites were not detected in the follow-up slide. The patient had a rapid and favorable response; symptoms resolved within 48 hours. He had no further febrile episodes after 24 months.

Figure 1 : Subject's malaria thin blood smear. Erythrocytes infected with *Plasmodium ovale*.

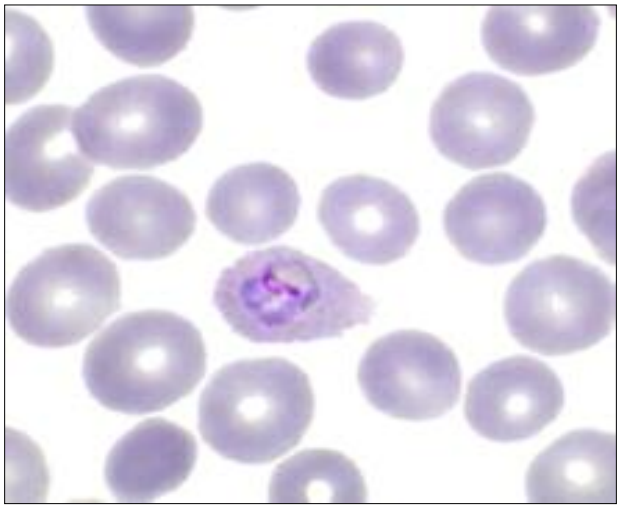
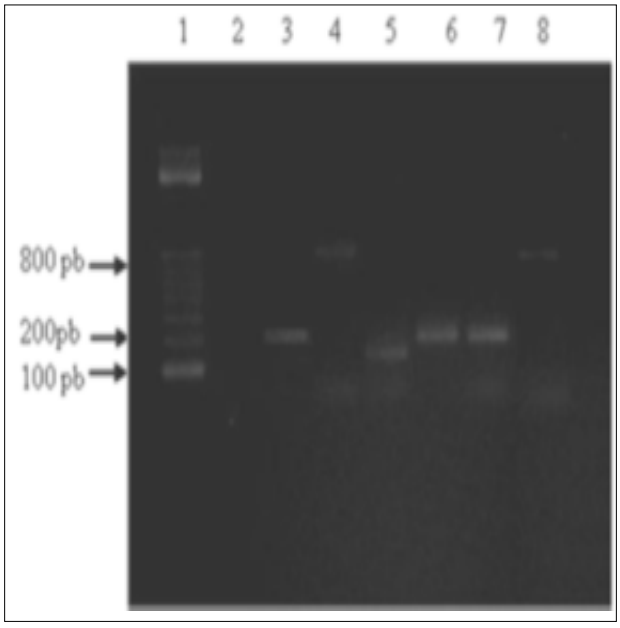


Figure 2 : Multiplex Polymerase chain reaction (PCR) results of analysis of the 18S ribosoma RNA genes of Plasmodium. Lane 1: 100-basepair DNA ladder, lane 2: PCR negative control, lane 3: control genomic DNA from P. falciparum (amplimer size = 205 base pairs), lane 4: control genomic DNA from P. ovale (amplimer size = 800 base pairs), lane 5: control genomic DNA from P. vivax (amplimer size = 120 base pairs), lane 6: DNA from a case amplified with Plasmodium falciparum-specific primers, lane 7: DNA from a case amplified with P. falciparum-specific primers, lane 8: DNA from the index case amplified with P. ovale-specific primers.



DISCUSSION

This is the first case report of a relapse of imported *P. ovale* malaria in Tunisia occuring three years after the primary infection. With *P. ovale* infections, after the sporozoites enter the hepatocytes not all parasites will develop into schizonts. Certain can remain dormant and susbsequently develop into mature schizonts and released invasive merozoites infecting erythrocytes and causing a relapse even many months after primary infection (5). Dormant stage occurs also in *P. vivax* infections (6). The mechanism behind the development of the sporozoites into either active schizonts or dormant hypnozoites is unknown (7). The authors suggested that the hypnozoites were programmed genetically to be activated at defined intervals or that they reacted on outside stimuli as cold or stress (7). *P. falciparum* and *P. malariae* do not have a dormant liver stage. Instead, *P. falciparum* can survive in the blood for months and *P. malariae* can cause long time chronic infections, which reoccur decades after the initial exposure (8,9).

P. ovale is responsible for rare travel-acquired infection (10). Globally, it causes 0.5-10.5% of all malaria cases (11). Few relapses have been published on *P. ovale* infections (12). The US CDC reported 27 cases of relapsing *P. ovale,* in the United States in 2004 which occurred 17 to 255 days after the primary attack (12). Other report described a relapse occurring 45 months after treatment of the primary attack of *P. ovale* (13). Relapses *with P. ovale* infections occur even after appropriate treatment of a blood-stage infection. The hypnozoites are insensitive to artemether lumefantrine, atovaquone-proguanil and to chloroquin, which are active against blood stage schizonts (14). Medicine to reduce the chance of such relapses is available and should follow treatment of the first attack. Primaquine is the most frequently used drug that kills hypnozoites (14). Because of the risk for hemolysis, patients must be screened for G6PD deficiency before starting treatment (15).

In our case no malarial parasites were found on initial blood smears. This examination is operator dependant and requiring considerable expertise. The accuracy of this test can be lower if microscopists are not well trained. It is also important to note that the negative results may be partly attributed to the low parasitemia (10). So when the diagnosis of malaria is suspected, microscopic blood smears should be repeated, up to three times and in an expert laboratory to detect malaria parasites (16). PCR marks real progress with a high sensitivity and specificity for molecular detection of *Plasmodium*. It should be utilized as a diagnosis tool especially to identify low parasitaemia and confirming the diagnosis in imported malaria cases (17). OptiMal-IT® test failed to detect *P. ovale* infection. All available rapid antigenic tests currently lack sensitivity to this specie (18). This could be due to the very low level of circulating antigen or to inadequate antigens used for these tests (19).

CONCLUSION

This report highlights the importance of considering malaria in differential diagnoses of febrile illnesses. It also underscores the importance of repeating microscopic examination of blood when the diagnosis of malaria is suspected.

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