

Two cases of malaria due to *Plasmodium falciparum* resulting in death: postmortem identification

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Abstract: *Plasmodium falciparum*, a malaria agent, is an important cause of morbidity and mortality in people who travel to the regions where this parasite is endemic. A 53-year old Turkish man was sent for an autopsy after death due to malaria complications from Mozambique. A 65-year-old German man was intubated at the airport due to the impaired overall status and admitted to the intensive care unit. The patient could not be diagnosed and died. The next day, an autopsy was performed on the patient. Malaria pigment was seen within the blood cells at the Giemsa staining of blood in both cases. *Plasmodium falciparum* DNA was positive in the analysis performed with blood samples from the patients. Postmortem sampling and demonstration of plasmodium DNA is among the important methods for diagnosis in cases with sudden death and a history of travel to foreign countries, where the antemortem diagnosis cannot be made.

Key Words: malaria, *Plasmodium falciparum*, postmortem, polymerase chain reaction, travel, autopsy.

Malaria, which affects almost half of the world population, continues to be an important health issue [1]. Currently, approximately 300 million cases of malaria occur every year in the developing countries and almost 1 million of these cases die due to malaria [2]. Malaria is endemic in Southern and Southeastern Anatolia regions and sporadic in other regions in Turkey. However, it has been reported that there are no new local cases of malaria in the recent years and the new cases are returning from endemic regions abroad [3].

Plasmodium falciparum, one of the malaria agents, is an important cause of morbidity and mortality in people who travel to the regions where this parasite is endemic [4]. It causes a severe clinical picture upon

the development of acute respiratory distress and renal failure [1, 5]. The ineffectiveness of taking the malaria prophylaxis and visiting endemic regions without education result in the occurrence of foreign-based malaria cases [5]. Therefore, malaria must be considered in the cases with unexpected sudden deaths and a history of travel [6].

The current article presented the results of postmortem assessment of two foreign-based patients who died due to *Plasmodium falciparum* malaria.

CASE 1

A 53-year old Turkish man was sent to Istanbul Mortuary Specialization Office for an autopsy after

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death due to malaria complications in Mozambique. It was determined that the case was on postmortem day 8 and the patient had been working abroad four months ago, but did not receive prophylaxis for malaria. After a twenty-day stay at the hospital, the family of the patient was informed that the patient died, but the epicrisis report could not be obtained. However, quinine was found in the analysis of the blood taken from the patient, so it was assumed that the patient received antimalarial treatment. Malaria pigment was seen within the macrophage cells in the Giemsa staining of thin blood smear preparation (Fig. 1).

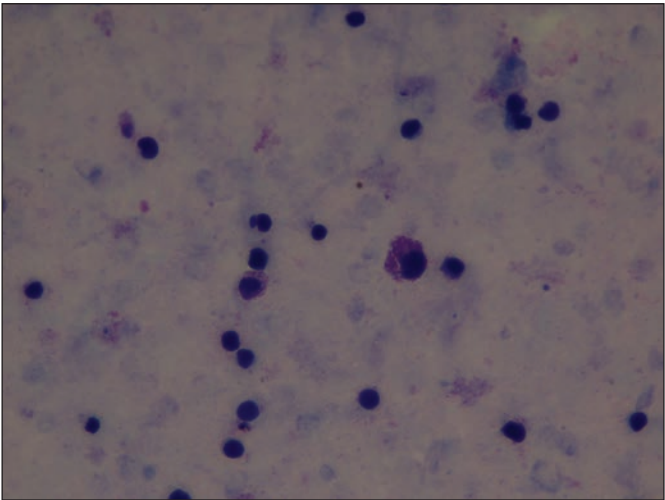


Figure 1. Malaria pigment within the macrophage cells, Case 1, (Giemsa boyama x 100).

CASE 2

A 65-year-old German man was intubated at the airport due to an overall impaired status and it was reported that the patient was unconscious and did not respond to painful stimulants. He was admitted to the intensive care unit of the hospital with the pre-diagnoses

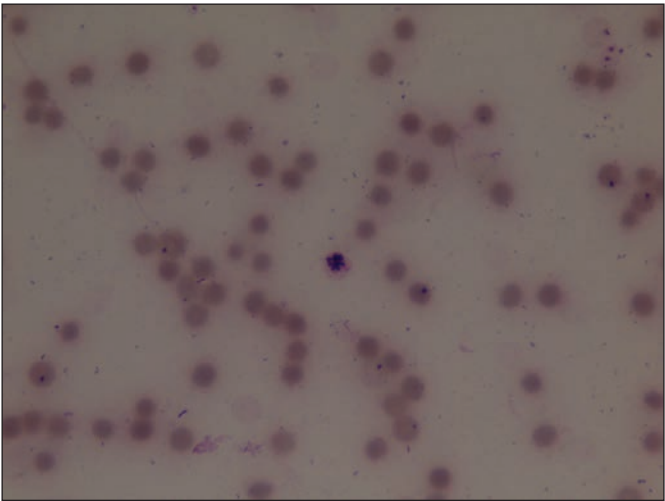


Figure 2. Malaria pigment within the erythrocyte cells, Case 2, (Giemsa boyama x 100).

of pneumonia and acute renal failure. Eight units of platelets and one unit of erythrocyte suspension were administered upon the findings of TA: 50/32 mm/Hg, hemoglobin: 7.2 gr/mm³, platelets: 14000/mm³. The chest radiography did not reveal any pneumonic infiltration. The patient, who could not be diagnosed and whose overall status did not improve, died in the morning. The patient was sent to the Istanbul Mortuary Specialization Office the next day with a record of suspicious death and an autopsy was performed on postmortem day 1. Malaria pigment was seen within the erythrocyte cells in the Giemsa staining of thin smear preparation of the blood from the patient (Fig. 2).

The presence of pigment was reported in the erythrocytes in the vessel lumens in myocardium, lungs, spleen, kidneys, brain, cerebellum and brain stem in the histopathological examination of postmortem tissues (Figs 3-5).

The polymerase chain reaction (PCR) method was used to analyze the plasmodium DNA in the blood and paraffin-embedded tissue samples (spleen and heart tissues) from the patients. Xylene and ethyl alcohol were used to deparaffinize the paraffin-embedded tissue samples. DNA isolation was performed on QIASymphony with DSP Virus/Pathogen Midi kit (Qiagen, Germany); and DNA amplification was performed on Rotor GeneQ (Qiagen, Germany) with the real-time PCR (RT-PCR) method using a Malaria RG PCR (Qiagen, Germany) kit. Upon the positive findings of Plasmodium spp. from RT-PCR, the blood samples were sent to the Referans Laboratory for species identification. At the laboratory, Plasmodium falciparum DNA was positive in both samples studied with Genesig P. falciparum RT-PCR (Primerdesign, UK), Genesig P. malaria RT-PCR (Primerdesign, UK), Genesig P. ovale RT-PCR (Primerdesign, UK), Genesig P. knowlesi RT-PCR (Primerdesign, UK), and Genesig P. vivax RT-PCR (Primerdesign, UK) kits.

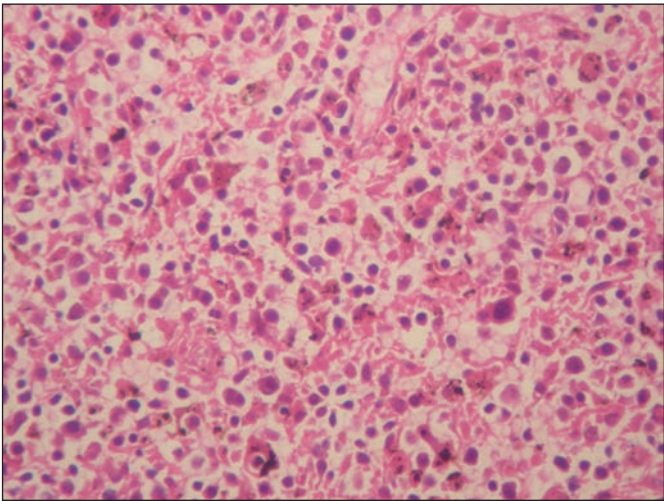


Figure 3. The presence of pigment in the erythrocytes in the vessel lumen of the spleen tissue, Case 1, (HE x 20).

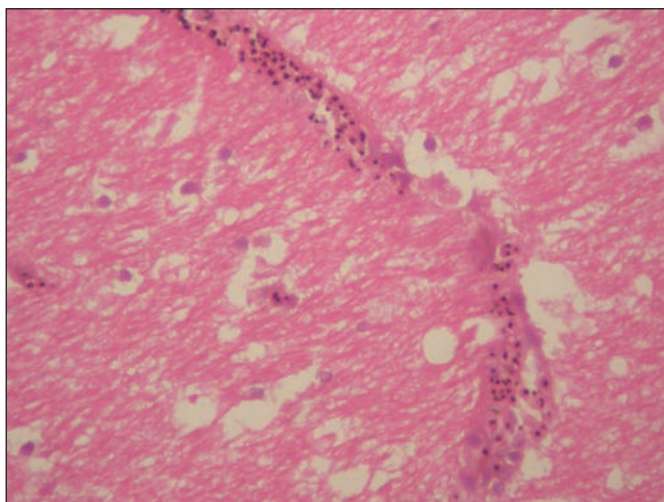


Figure 4. The presence of pigment in the erythrocytes in the vessel lumen of the brain tissue, Case 2, (HE x 20).

DISCUSSION

Malaria is a life-threatening and serious infection in people who travel to tropical and subtropical countries [7]. Although no new malaria cases have been seen in Turkey, *P. falciparum* malaria occurs more frequently due to the increased number of people visiting endemic regions [8]. The late diagnosis of the patients who have caught malaria after visiting these regions increases the mortality [9].

The other risk factors increasing mortality include the inappropriate use of chemoprophylaxis, age, gender, mistreatment, immunodeficiency, and *P. falciparum* infection [10]. Additionally, one of the most important causes increasing mortality is the increased number of malaria infections that developed resistance to the medication in the recent years [11]. The risk factors of the cases are not exactly known; however, the non-use of chemoprophylaxis in the first case presented and the *P. falciparum* finding in both cases are among the reported risk factors.

The standard diagnosis method for malaria is direct microscopic examination of thin smear or thick drop preparations of peripheral blood. The presence of parasite forms in the Giemsa staining of these preparations establishes the diagnosis [5]. In addition to the blood sampling in postmortem cases, the preparations made with hematoxylin eosin from the histopathological tissue sampling may allow visualization of trophozoite, schizont, or pigment structures of the malaria parasite [6, 12, 13]. However, the pigment structures were seen rather than the trophozoite forms of the parasite due to the extended postmortem interval or the antimalarial treatment [13]. The presence of infected cells with the parasite pigment in the preparations from the postmortem blood and tissue samples in the two presented cases increased the suspicion of malaria infection.

The PCR method, which has been increasingly used for the diagnosis of malaria in recent years, has been

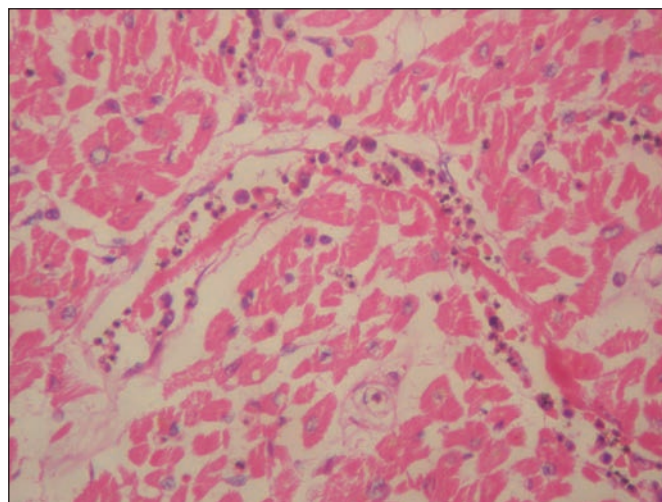


Figure 5. The presence of pigment in the erythrocytes in the vessel lumen of the heart tissue, Case 2, (HE x 20).

determined to be the best method because of detecting parasitemia < 5 parasites/ μ l, and a higher sensitivity and specificity [14, 15]. The antemortem and postmortem studies report that the PCR method has a high sensitivity [16-18]. Therefore, plasmodium-PCR is one of the important methods for use in postmortem cases where the microbiological and histopathological microscopic examinations are suspicious. Malaria cases, especially those due to *Plasmodium falciparum* that might progress to sudden death, have been reported in individuals visiting endemic regions of Africa [9, 19]. In both cases presented, *Plasmodium falciparum* DNA was found positive with the real-time PCR method.

Malaria is a fatal parasitic infection that can be generally diagnosed antemortem and less frequently postmortem [20]. As in the second case presented, the significance of postmortem identification emerges in cases with unexpected sudden death that could not be diagnosed antemortem. Therefore, the attentive performance of postmortem examinations (microbiological, histopathological) for diagnosing malaria in such cases is important for identifying this disease.

The increased volume of international travel and the increasingly improved medication resistance against malaria at present suggest that malaria will continue to be one of the important diseases in countries where malaria is not endemic. Therefore, malaria should be considered in the cases with unexpected sudden deaths and a history of travel to endemic regions [6].

In conclusion, appropriate malaria prophylaxis should be advised for protection from malaria to individuals who travel or go for work purposes to regions where malaria is endemic, such as Africa. In cases with sudden death and a history of travel to foreign countries, where antemortem diagnoses cannot be made, the analysis of plasmodium DNA is important to diagnose malaria if the postmortem microscopic examinations are suspicious.

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