

## **Morphological features and differential counts of *Plasmodium knowlesi* parasites in naturally acquired human infections**

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### **Abstract**

**Background:** Human infections with *Plasmodium knowlesi*, a simian malaria parasite, are more common than previously thought. They have been detected by molecular detection methods in various countries in Southeast Asia, where they were initially diagnosed by microscopy mainly as *Plasmodium malariae* and at times, as *Plasmodium falciparum*. There is a paucity of information on the morphology of *P. knowlesi* parasites and proportion of each erythrocytic stage in naturally acquired human infections. Therefore, detailed descriptions of the morphological characteristics and differential counts of the erythrocytic stages of *P. knowlesi* parasites in human infections were made, photographs were taken, and morphological features were compared with those of *P. malariae* and *P. falciparum*.

**Methods:** Thick and thin blood films were made prior to administration of anti-malarial treatment in patients who were subsequently confirmed as having single species *knowlesi* infections by PCR assays. Giemsa-stained blood films, prepared from 10 randomly selected patients with a parasitaemia ranging from 610 to 236,000 parasites per  $\mu$ l blood, were examined.

**Results:** The *P. knowlesi* infection was highly synchronous in only one patient, where 97% of the parasites were at the late trophozoite stage. Early, late and mature trophozoites and schizonts were observed in films from all patients except three; where schizonts and early trophozoites were absent in two and one patient, respectively. Gametocytes were observed in four patients, comprising only between 1.2 to 2.8% of infected erythrocytes. The early trophozoites of *P. knowlesi* morphologically resemble those of *P. falciparum*. The late and mature trophozoites, schizonts and gametocytes appear very similar to those of *P. malariae*. Careful examinations revealed that some minor morphological differences existed between *P. knowlesi* and *P. malariae*. These include trophozoites of *knowlesi* with double chromatin dots and at times with two or three parasites per erythrocyte and mature schizonts of *P. knowlesi* having 16 merozoites, compared with 12 for *P. malariae*.

**Conclusion:** *Plasmodium knowlesi* infections in humans are not highly synchronous. The morphological resemblance of early trophozoites of *P. knowlesi* to *P. falciparum* and later erythrocytic stages to *P. malariae* makes it extremely difficult to identify *P. knowlesi* infections by microscopy alone.