Review of: "The influence of biological, epidemiological, and treatment factors on the establishment and spread of drug-resistant Plasmodium falciparum"

Toshihiro Mita[1]

1 Juntendo University

Potential competing interests: The author(s) declared that no potential competing interests exist.

The authors investigated the usefulness of Targeted amplicon deep sequencing (TADS) as a molecular marker to distinguish recrudescences from reinfections. They constructed ama1 haplotypes and evaluated the presence/absence of haplotypes in the blood frequently obtained from treated patients using three different regimens. In 9 recurrent cases, haplotype composition was also compared before treatment and recurrence. Power to distinguish recrudescences from reinfections was compared between TADS and conventional molecular methods (msp1, msp2 and glurp). Despite that sample numbers were not so large, this study showed important findings as follows.

1. More haplotypes were detected in TADS with ama1 than those in other conventinal markers.
2. TADS showed similar level of COI as msp1 and msp2.
3. Stable number of TADS haplotypes were observed throughout the treatment.
4. They identified one haplotype showing parasite clearance after treatment.

Major

Since dead-parasites' DNA can persist in the human blood after treatment, the evaluate amount (also presence) of haplotype may not directly reflect the real situation. This would become an important confounding factor. Could you discuss this point?

Minor

1. Page4 “The DNA from 3D7 and Dd2, were mixed as follows to come up with sequencing controls: 100%:100%, 75%:25%, 85%:15%, 95%:5% and 100%:0% to determine the lowest limit of haplo-type detection.” “Does “100%:100%” mean “50%:50%”?
2. Page 5 “There was successful detection of the two expected ama1 haplotypes from the 3D7 and Dd2 laboratory isolates, as well as three mdr1 haplotypes, one from 3D7 and the 2 copies of Dd2”. I think some readers might feel curious about two haplotypes in the laboratory clone (Dd2). Could add some explanation?