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Comparison of the Performances of Five Primer Sets for the Detection and Quantification of *Plasmodium* in Anopheline Vectors by *Real-Time* PCR

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Abstract

Quantitative real-time polymerase chain reaction (qrtPCR) has made a significant improvement for the detection of *Plasmodium* in anopheline vectors. A wide variety of primers has been used in different assays, mostly adapted from molecular diagnosis of malaria in human. However, such an adaptation can impact the sensitivity of the PCR. Therefore we compared the sensitivity of five primer sets with different molecular targets on blood stages, sporozoites and oocysts standards of *Plasmodium falciparum* (Pf) and *P. vivax* (Pv). Dilution series of standard DNA were used to discriminate between methods at low concentrations of parasite and to generate standard curves suitable for the absolute quantification of *Plasmodium* sporozoites. Our results showed that the best primers to detect blood stages were not necessarily the best ones to detect sporozoites. Absolute detection threshold of our qrtPCR assay varied between 3.6 and 360 Pv sporozoites and between 6 and 600 Pf sporozoites per mosquito according to the primer set used in the reaction mix. In this paper, we discuss the general performance of each primer set and highlight the need to use efficient detection methods for transmission studies.