Ultra-sensitive detection of Asymptomatic sub-microscopic Plasmodium falciparum Malaria infection amongst Children in Ngali, Center Region of Cameroon
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INTRODUCTION

Malaria remains the world's most important life-threatening parasitic infectious disease and, Plasmodium falciparum the leading cause of malaria-related mortality in sub-Saharan Africa. In spite of the great decline in the burden of malaria, residual transmission is ongoing at levels undetectable by microscopy and rapid diagnostic test (RDT). Highly sensitive novel primers have been designed for the detection of malaria by PCR. However, their diagnostic performances have not been evaluated on DNA isolated using the hot-chelex method, a highly preferred and economic DNA extraction method used in LMICs. This study aimed to evaluate the diagnostic performance of traditional 18SrRNA and, novel Mitochondrial (Cox3) and varATS primers in the detection of Plasmodium falciparum malaria infection.

METHODS

Archival samples collected from 314 asymptomatic children dwelling in the high malaria transmission locality of Ngali and aged 1-15 was used. RBC pellet was blotted on filter papers and allowed to air dry for 24 hours. DNA was extracted using the hot-chelex method and samples were examined for the presence of Plasmodium falciparum DNA using 18SrRNA, Cox3 in a Nested PCR and varATS primers in a single step PCR. Diagnostic performance was calculated using MedCalc statistical package with our composite test as the gold standard.

RESULTS

Overall, the general prevalence of P.falciparum malaria detected by thick film microscopy, Rapid Diagnostic Test, 18SrRNA, Mito(cox3) and varATS was 76.8%, 76.8%, 75.8%, 76.8% and 86.0% respectively. However, several bands resulting from the use of varATS were non-conclusive. Out of the 314 samples, 70 samples were negative for P.falciparum by TFM with temperatures ≤37.5oC. The prevalence of submicroscopic infection detected by RDT, 18SrRNA, Mito (cox3) and varATS was 0.0%, 35.7%, 45.7% and 74.3% respectively. The Sensitivity and specificity of 18SrRNA primer was 65.7% and 94.3%, Cox3 primer was 80% and 88%, and varATS was 100% and 51.4% using the composite test as a gold standard. With respect to the percentage of discordance, 24.2% of the samples were detected by all three primers.

CONCLUSION

These findings recommend Cox primers for hot-chelex applications. Despite being sensitive, varATS produced indeterminate results. Discrepancies with slide positives demand additional research.