Title: Investigation of malaria diagnostic performance and antimalarial resistance markers in contrasting transmission settings in Cameroon

Background: Malaria remains a major public health challenge in Cameroon, where *Plasmodium falciparum* is the predominant species. The World Health Organization recommends universal parasitological confirmation prior to treatment, making the reliability of malaria rapid diagnostic tests (mRDTs) and the sustained efficacy of artemisinin-based combination therapies (ACTs) essential to malaria control. Emerging biological threats, including *pfhrp2/3* gene deletions associated with false-negative HRP2-based mRDT results and mutations in *pfk13* and other loci linked to antimalarial resistance—pose potential risks to both diagnosis and treatment outcomes. This study investigated post-market mRDT performance, the occurrence of *pfhrp2/3* deletions, and the distribution of key molecular markers of drug resistance in a low- and high-transmission setting in Cameroon.

Methods: A cross-sectional study was conducted in two health facilities representing contrasting transmission intensities in the Littoral region of Cameroon. Multiplex quantitative PCR was used to confirm *P. falciparum* infection, estimate parasite density, and assess the presence of *pfhrp2/3* deletions. Targeted amplicon sequencing using Oxford Nanopore Technology was performed to characterize genetic variation in established molecular markers associated with antimalarial drug resistance.

Findings: Genomic analyses confirmed the presence of parasites with *pfhrp2/3* deletions in the study area, including strains associated with discordant HRP2-based mRDT results. Drug resistance profiling identified circulating haplotypes linked to reduced susceptibility to commonly used partner drugs, alongside additional polymorphisms in *pfk13* and related loci. A detailed breakdown of mutation patterns, haplotype distribution, and transmission setting comparisons will be presented and discussed during the meeting.

Interpretation: The detection of *pfhrp2/3*-deleted parasites highlights the importance of continued diagnostic performance monitoring, even where HRP2-based mRDTs remain broadly effective. The presence of molecular markers associated with antimalarial resistance further underscores the need for integrated and routine genomic surveillance. Strengthening diagnostic and therapeutic monitoring systems will be essential to inform national malaria policy and sustain recent gains in malaria control in Cameroon.