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Diagnostic parasitology

Serological responses against species-specific recombinant MSP-10 antigens as markers of malaria exposure in the Peruvian Amazon

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Background: Accurate estimates of malaria exposure are essential to understand the spatial and temporal variations in disease transmission and to target and evaluate malaria control efforts. Most estimates have been based on responses to a small number of antigens that are primarily vaccine candidates. Recent reports using protein microarrays in Peru and Uganda have identified candidate antigens as potential biomarkers of recent vs. remote exposure. This study aimed to assess serological responses against the highly immunogenic merozoite surface protein-10 for *P. vivax* (PvMSP10) and for *P. falciparum* (PfMSP10) in terms of recent species-specific malaria exposure of individuals in the low transmission setting of the Peruvian Amazon.

Material/methods: From a 5-month (August-September 2012-January 2013) cohort of 960 Amazonian individuals, we selected those with monthly follow-up by light microscopy (LM). Individuals with negative LM and positive PCR (processed later) at the enrollment, and those who had no data of self-reported clinical malaria episodes in the previous year (SRCM) were excluded. Individuals with positive LM were treated according to national guidelines. Specific IgG antibody responses to HEK-293 mammal cells-produced recombinant PfMSP10 and PvMSP10 proteins were analyzed by ELISA. Chi-squared tests were used to compare seropositivity among participants, stratified by age (<=15,>15 years), SRCM (SRCM+,SRCM-), and infection status (no infection, infection within<30 days, 30-60 days or >60 days before sample collection). Multivariate logistic regression assessed association of seropositivity status and infection, adjusting for age and SRCM.

Results: Of 437 subjects, 278 remained uninfected, 128 acquired *P. vivax*, and 31 acquired *P. falciparum* infections. Median times between infection and sample collection were 45 d (range: 0-163) and 101 (range: 0-175) for *P. vivax* and *P. falciparum*, respectively. Age>15 years (p<0.001) and SRCM+ (p=0.028) were associated with *P. vivax* antibody responses. *P. vivax* seropositivity was highest in individuals with *P. vivax* infections of <30 days (74.4%), and decreased significantly with the time since last infection (p<0.001): 30-60 days (60.0%), >60 days (47.5%), no infection (18.7%). Individuals with *P. vivax* infections of <30 days were 8-times (AOR=8.2, 95%CI [3.7,17.9]) more likely to be seropositive than those without infections, after adjustment for age and SRCM. The low number of *P. falciparum* infections limited further analysis, but age>15 years (p<0.001) and *P. falciparum* infections (p=0.026) were associated with *P. falciparum* seropositivity.

Conclusions: In the low transmission setting of the Peruvian Amazon, serological responses against PvMSP10 are good markers of recent *P. vivax* malaria exposure. Comparison of anti-MSP10 responses with responses to other antigens and studying additional well-characterized subjects with recent new vs. old infections would further confirm PfMSP10 as a marker of *P. falciparum* exposure in

this region. Accurate determination of malaria exposure is essential for evaluating control and elimination measures, which is of global importance.