Estimating Annual Fluctuations in Malaria Transmission Intensity and in the Use of Malaria Control Interventions in Five Sub-Saharan African Countries

RTS,S Epidemiology EPI-MAL-005 Study Group

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Abstract. RTS,S/AS01E malaria vaccine safety, effectiveness, and impact will be assessed in pre- and post-vaccine introduction studies, comparing the occurrence of malaria cases and adverse events in vaccinated versus unvaccinated children. Because those comparisons may be confounded by potential year-to-year fluctuations in malaria transmission intensity and malaria control intervention usage, the latter should be carefully monitored to adequately adjust the analyses. This observational cross-sectional study is assessing Plasmodium falciparum parasite prevalence (PPR) and malaria control intervention usage over nine annual surveys performed at peak parasite transmission. Plasmodium falciparum parasite prevalence was measured by microscopy and nucleic acid amplification test (quantitative PCR) in parallel in all participants, and defined as the proportion of infected participants among participants tested. Results of surveys 1 (S1) and 2 (S2), conducted in five sub-Saharan African countries, including some participating in the Malaria Vaccine Implementation Programme (MVIP), are reported herein; 4,208 and 4,199 children were, respectively, included in the analyses. Plasmodium falciparum parasite prevalence estimated using microscopy varied between study sites in both surveys, with the lowest prevalence in Senegalese sites and the highest in Burkina Faso. In sites located in the MVIP areas (Kintampo and Kombewa), PPR in children aged 6 months to 4 years ranged from 24.8% to 27.3%, depending on the study site and the survey. Overall, 89.5% and 86.4% of children used a bednet in S1 and S2, of whom 68.7% and 77.9% used impregnated bednets. No major difference was observed between the two surveys in terms of PPR or use of malaria control interventions.

INTRODUCTION

Substantial investment to expand existing malaria interventions has resulted in a reduction in the global incidence rate of malaria between 2010 and 2017.1 However, between 2015 and 2017, after stagnation, a slight upward trend in malaria incidence was observed. Malaria remains a major cause of death worldwide, with approximately 93% of all malaria deaths in 2017 occurring in Africa.1 To reach the Global Technical Strategy for Malaria 2016–2030 target of reducing global malaria incidence and mortality rates by at least 90% by 2030,2 the need for safe and effective malaria vaccines that prevent disease and death and decrease transmission to enable eradication was endorsed and documented in the WHO Malaria Vaccine Technology Roadmap.3

Gliaxosmithkline (GSK) has developed, in partnership with the PATH Malaria Vaccine Initiative, a pre-erythrocytic Plasmodium falciparum malaria vaccine, RTS,S/AS01E (GSK™, Wavre, Belgium), for routine immunization of infants and children living in sub-Saharan African (SSA) malaria-endemic countries. In 2015, the European Medicines Agency adopted a positive opinion for the use of the vaccine in children aged 6 weeks to 17 months at the first dose.4 In January 2016, the WHO recommended a pilot implementation of RTS,S/AS01E in children as of 5 months of age in three to five epidemiological SSA settings with moderate to high malaria transmission.5 In April 2017, the WHO announced the vaccine introduction based on a cluster-randomized design in pilot areas of Ghana (GH), Kenya (KE), and Malawi through the national expanded programs on immunization, in the framework of the Malaria Vaccine Implementation Programme (MVIP).6 Today, RTS,S/AS01E is the first vaccine implemented for the prevention of malaria.

To assess vaccine safety, effectiveness, and impact, GSK designed a pre- and a post-vaccine introduction observational study (ClinicalTrials.gov identifiers: NCT02374450 and NCT03855995, respectively), allowing comparison of the occurrence of malaria cases and adverse events in vaccinated versus unvaccinated children. In parallel with those studies, the present observational cross-sectional annual study (NCT02251704) is estimating P. falciparum parasite prevalence (PPR) and the use of malaria control interventions during up to nine consecutive years, applying standardized methodologies and multiple diagnostic testing. More specifically, considering the WHO recommendation to operate the MVIP in moderate to high transmission areas of SSA, this study will allow 1) characterizing the malaria transmission intensity (MTI) before RTS,S/AS01E vaccine introduction in different countries/areas, including the ones participating in the MVIP; 2) monitoring overtime fluctuations of MTI and of the use of malaria control interventions before and after vaccine
introduction in those areas to adjust the pre- and post-vaccine introduction comparison analyses for potential year-to-year and/or geographical variations.

We present here the results of the first two annual surveys that were conducted before vaccine introduction. On completion of all surveys, the data collected in this study involving approximately 50,000 participants representing multiple sites in various SSA countries will provide a unique perspective on malaria prevalence variations across Africa.

MATERIALS AND METHODS

Ethics. The study was approved by national independent Ethics Committees and local institutional review boards in Burkina Faso (BF), GH, KE, Senegal (SN), and Tanzania (TZ), and conducted in accordance with the provisions of the International Conference on Harmonisation and Good Clinical Practice guidelines.

Study population. Individuals aged 6 months to < 10 years, whose parents or legally acceptable representative had provided informed consent for study participation, were randomly selected each year in each of the study sites (see Study design section) using population listings generated from local Health and Demographic Surveillance Systems (HDSS) and following a stratification by age-group (see the Supplemental Appendix Section 1). Children in care, or actively participating in any trial involving the administration of an investigational malaria vaccine and/or drug, were excluded.

Study design. Malaria transmission intensity levels are consensus indicators developed by the Global Malaria Eradication Programme to measure malaria endemicity7 using a standardized methodology. There are several methods for estimating MTI, including entomological inoculation rates (EIRs), serological conversion rates (SCRs), and blood parasite prevalence. Although EIR is a standard method, the measure is challenging, and the interpretation and comparability of the setting may be difficult because of vector heterogeneity.8 The methodology and interpretation of SCRs to classify the intensity of malaria is still not commonly used. Plasmodium falciparum prevalence, despite requiring trained staff for slide reading, provides a standardized and relatively easy to implement method to assess MTI in study sites of varied transmission intensity, and has often been used in previous epidemiological studies.7,9,10 Therefore, PPR was the selected method to assess potential variations in MTI in the present study.

The study is multicentric with study sites corresponding to geographically limited catchment areas located in low, moderate, or high MTI regions of SSA, and having an HDSS in place.

Up to nine annual cross-sectional surveys will be conducted during the malaria peak transmission period in each study site (from mid-September to mid-December in Western African sites and from mid-April to mid-August in Eastern African sites). To optimize the detection of the peak transmission, each study site has been equipped with a weather station to record meteorological data such as rainfall, temperature, and humidity. Surveys were conducted during the course of the rainy season preferably when rains decrease, which should correspond to the period of highest malaria transmission. In this article, results of the first two annual surveys are presented. Survey 1 (S1) and survey 2 (S2) were conducted in seven study sites in five SSA countries: BF (Nouna, Saponé), GH (Kintampo), KE (Kombewa), SN (Keur Socé, Niakhari), and TZ (Korogwe). Sites in GH (Kintampo) and KE (Kombewa) are part of the areas where the RTS,S/AS01 vaccine will be implemented through the national Expanded Programs on Immunization in the framework of the MVIP. It is important to note that this study is also conducted in SSA areas where the RTS,S/AS01 vaccine will not be introduced in the framework of the MVIP because GSK initiated the study before the WHO recommendation.5,11

Data collection. Demographic details (age and gender), medical history, and information on care-seeking behaviors (hospitalization for malaria within the last 3 months, visits to healthcare provider for fever or malaria treatment in the previous 14 days, anti-malaria therapy received within the last 14 days); malaria control intervention usage (bednets [new (not older than a year), pierced/torn, impregnated], indoor residual spraying [IRS]); the usage of coils, repellents, and local herbs; and malaria potential risk factors (rural/urban area, house construction materials, use of electricity, and open/closed water source) were recorded for all participants at the time of the survey. Axillary body temperature was measured and recorded during the survey visit.

To assess within-site heterogeneity of PPPR, study areas were mapped by villages using grid referencing and divided into 3–14 segments with a minimum of 10 enrolled individuals per segment. Segments will remain unchanged for the duration of the study.

Assessment of parasitemia. Both microscopy and Nucleic Acid Amplification Tests (NAATs) were used in parallel on all participants to assess parasitemia. The latter are expected to be more sensitive and specific, particularly in cases of low parasite density.12,13 In brief, a blood sample was collected by finger prick for thin and thick blood films for the microscopy assessment and filter paper blood spots for NAATs. Blood smears were examined by two local independent microscopists, and any discrepancies were settled by a third reader. Parasitemia was measured by the examination of 100 high-powered fields on thick smear to determine the presence of parasites; in the case of a positive result, additional 100 fields were examined to assess the presence of multiple species. Plasmodium species and sexual forms were identified on thin blood film. Parasite density was computed as the geometric mean of two readings, counting parasites against 200 white blood cells on thin blood film, assuming 8,000 white blood cells/μL. Parasite density was categorized as low (< 2,500 parasites/μL), medium (2,500–9,999), high (10,000–19,999), or very high (> 20,000). In the case of low density (< 10 parasites against 200 leukocytes), parasite count was conducted against 500 white blood cells. The parasite count technique was replicated to count gametocytes. In parallel with microscopy, asexual and sexual parasitemia was assessed by NAATs using both real-time quantitative PCR (QT-PCR) assay and real-time nucleic acid sequence-based amplification (QT-NASBA) assay. Quantitative PCR allowed detection of asexual and sexual parasites combined, the final output being qualitative (positive or negative) and semi-quantitative (high, medium, low, and negative). Real-time nucleic acid sequence-based amplification allowed detection of gametocytes, the final output being qualitative (positive or negative). Details for blood slide and NAAT assessment of parasitemia are available in the Supplemental Appendix (Sections 2 and 3, respectively).
If fever (i.e., axillary temperature ≥ 37.5°C) was recorded at the time of the visit or reported to have occurred within 24 hours before the visit, a malaria rapid diagnostic test (RDT) was conducted using blood from the finger prick sampling. If the RDT was positive, then treatment was given according to national guidelines. Moreover, any participant identified as being parasite positive following microscopy was traced to receive treatment according to national guidelines.

**Statistical methods.** The planned sample size was 600 participants per study site and per survey distributed as 400 participants between the ages of 6 months to 4 years and 200 participants between the ages of 5 and 9 years. The sample size was calculated to ensure sufficiently narrow CIs around study site–specific *PfPR* estimates (with a maximum residual standard error of 0.25).

*Plasmodium falciparum* parasite prevalence and prevalence of gametocytes were estimated as the proportion of participants infected, or carrying gametocytes, respectively, among participants tested. Prevalences were estimated by age and by site. The agreement between the two diagnostic methods used in the framework of this study (parasitemia measured by microscopy versus NAATs) was described using the Cohen’s kappa coefficient and assessed using the Landis and Koch scale.\(^{14}\) The prevalence of *Plasmodium* species other than *P. falciparum* was estimated as the proportion of infected participants among participants tested. The within-site heterogeneity between segments was tested using Cochran’s Q-test based on inverse variance weights.

Malaria control intervention coverage was estimated as the proportion of users among participants for which this information was available. The care-seeking behaviors (treatment sought for malaria or fever in the 14 days before the visit and hospitalization for malaria in the last 3 months before the visit) were described as the proportion of participants having sought for health care among all participants. In addition, a risk factor analysis for malaria infection (dependent variable: *P. falciparum* parasitemia as measured by microscopy) was conducted using a multivariable logistic regression with study site as cluster and using a backward strategy for the selection of significant explanatory variables, that is, predefined potential risk factors and/or the use of malaria control interventions (Supplemental Appendix Section 4). Age was computed as a continuous variable.

**RESULTS**

Survey 1 and S2 data were collected between October 2014 and August 2015, and between September 2015 and July 2016, respectively. During S1 and S2, 4,215 and 4,204 children were enrolled and 4,208 and 4,199 were included in the analyses (Figure 1), with a balanced distribution across the seven study sites (Supplemental Table 1). Across all sites, 51.3% of participants in S1 and 50.3% in S2 were males (Supplemental Table 2).

**Year-to-year variation in *P. falciparum* prevalence.** *Plasmodium falciparum* parasite prevalence estimated using microscopy varied between study sites in both surveys, with the lowest prevalence figures in Senegalese sites and the highest in BF (Table 1, Figure 2). In Kintampo and Kombewa sites that are located in the MVIP areas, *PfPR* in children aged 6 months to 4 years ranged from 24.8% to 27.3% depending on the study site and the survey. In both surveys and across all sites except in SN, *PfPR* was lower in the 6-month to < 5-year than in the 5-year to < 10-year age-group. *Plasmodium falciparum* parasite prevalence was similar in S1 and S2, except for a higher prevalence in S2 in participants from the 6-month
to < 5-year age-group in BF Nouna and lower prevalence in S2 in both the 6-month to < 5-year and 5-year to < 10-year age-groups in participants from TZ Korogwe. Significant within-site heterogeneity was detected in all sites, except in SN: BF Nouna (S1 \( P < 0.0001 \), S2 \( P = 0.0184 \)), BF Saponé (S1 \( P = 0.0321 \), S2 \( P < 0.0001 \)), TZ Korogwe (S1 \( P = 0.0037 \), S2 \( P = 0.0275 \)), KE Kombewa (S1 \( P = 0.0220 \), S2 \( P < 0.0001 \)), and GH Kintampo (S2 only \( P = 0.0065 \)).

Similar trends were observed when measured by QT-PCR, with \( PfPR \) varying from 2.7% in SN Niakhar to 69.9% in BF Nouna (not measured in SN Keur Socé) in S1 and from 1.3% in SN Keur Socé to 76.3% in BF Nouna in S2 (Supplemental Table 3).

The prevalence of gametocytes measured by microscopy ranged from 0.5% to 4.0% in S1 and from 0.0% to 5.3% in S2 in all sites, except in BF (15.8% and 22.7% in S1 and 12.8% and 18.2% in S2 in Nouna and Saponé, respectively). Among participants tested positive for asexual parasites, 7.5–66.7% carried gametocytes in S1 (Kintampo and Keur Socé, respectively) and 0.0–83.3% in S2 (Niakhar and Keur Socé, respectively) (Table 2). The proportion of infected participants carrying gametocytes as estimated by QT-NASBA ranged from 13.3% in SN Niakhar to 59.2% in BF Saponé in S1 (not measured in Keur Socé) and from 7.1% in SN Niakhar to 66.7% in SN Keur Socé in S2 (Supplemental Table 3).

### Agreement between diagnostic tests

Across surveys, approximately 21.6% of participants with a positive QT-PCR result had a negative microscopy reading (285 of 1,268 positive participants per QT-PCR in S1 and 296 of 1,417 in S2) and around 8.8% of participants positive for microscopy had a negative result with QT-PCR (92 of 1,075 positive participants per microscopy in S1 and 110 of 1,231 in S2) (Table 3). The proportion of participants with a negative result by microscopy among participants with a positive QT-PCR ranged

![Figure 2](image-url)
between 42.9% and 78.6% in sites with low MTI (Niakhar, Keur Socé, and Korogwe).

Across all sites, Cohen’s kappa coefficient between qualitative results of the two methods of measurement of parasitemia (microscopy versus QT-PCR) using the Landis and Koch scale showed a substantial agreement in both surveys (S1: kappa = 0.75 [95% CI: 0.73; 0.78]; S2: kappa = 0.78 [95% CI: 0.75; 0.80]). Cohen’s kappa coefficient between semi-quantitative results for parasitemia measured by microscopy versus QT-PCR also showed a substantial agreement in both S1 (kappa = 0.65 [95% CI: 0.63; 0.68]) and S2 (kappa = 0.66 [95% CI: 0.64; 0.68]).

Between 25.0% (192 of 767 in S2) and 33.8% (185 of 548 in S1) of participants tested positive for gametocytes by QT-NASBA were also detected positive by microscopy (Supplemental Table 4).

Prevalence of Plasmodium species other than P. falciparum. Infection with Plasmodium malariae was observed in 1.5% of participants in S1 and 3.0% of participants in S2 (Supplemental Table 5). Coinfection with both P. malariae and P. falciparum was more frequent than single infection with P. malariae alone in both surveys (S1: 4.6% versus 0.2%; S2: 7.4% versus 1.2%). Of the 62 participants in S1 and 128 participants in S2 infected with P. malariae, 55 (88.7%) and 91 (71.1%) were also infected with P. falciparum, respectively. Infection with Plasmodium ovale was low in both S1 and S2 (0.5% and 0.2% of participants). Plasmodium vivax was not observed in S1 and in only one participant in S2. Infections with species other than P. falciparum were mostly observed in sites of medium-to-high P. falciparum prevalence.

Year-to-year variation in the use of malaria control interventions. Overall, 89.5% and 86.4% of children used a bednet the night before the survey in S1 and S2, respectively (Table 4, Supplemental Table 6). The highest use of bednets was in KE Kombewa (97.5%) in S1 and TZ Korogwe (99.2%) in S2, and the lowest in GH Kintampo (70.2%) in S1 and SN Niakhar (69.6%) in S2 (Table 4). A decrease in usage between the two surveys was observed in Kombewa, Keur Socé, and Niakhar, whereas an increase was observed in Kintampo and Korogwe.

Table 2. Gametocyte results measured by microscopy according to P. falciparum infection status per study site and survey

| Study site, n (%) | Participants positive for P. falciparum by microscopy | Participants tested for P. falciparum by microscopy |
|------------------|------------------------------------------------------|--------------------------------------------------|
|                  | Survey 1 | Survey 2 | Survey 1 | Survey 2 |
| Nouna, BF        | N = 403  | N = 488  | N = 606  | N = 600  |
| Positive         | 89 (22.1)| 76 (15.6)| 96 (15.8)| 77 (12.8)|
| Saponé, BF       | N = 299  | N = 316  | N = 604  | N = 599  |
| Positive         | 111 (37.1)| 84 (26.6)| 137 (22.7)| 109 (18.2)|
| Kintampo, GH     | N = 214  | N = 212  | N = 600  | N = 600  |
| Positive         | 16 (7.5 )| 23 (10.9)| 18 (3.0 )| 32 (5.3 )|
| Kombewa, KE      | N = 196  | N = 189  | N = 599  | N = 599  |
| Positive         | 19 (9.7 )| 8 (4.2 ) | 24 (4.0 )| 9 (1.5 ) |
| Keur Socé, SN    | N = 3    | N = 6    | N = 600  | N = 600  |
| Positive         | 2 (66.7 )| 5 (83.3 )| 5 (8.8 ) | 5 (8.8 ) |
| Niakhar, SN      | N = 9    | N = 3    | N = 598  | N = 601  |
| Positive         | 1 (11.1 )| 0 (0.0 ) | 3 (0.5 ) | 0 (0.0 ) |
| Korogwe, TZ      | N = 63   | N = 18   | N = 601  | N = 600  |
| Positive         | 7 (11.1 )| 1 (5.6 ) | 8 (1.3 ) | 1 (0.2 ) |

Overall, 70–80% of the bednets were impregnated, 60–70% were new, and approximately 25% were torn (Supplemental Table 6). Details for the characterization of bednet usage (new, impregnated, and pierced/torn) by study site are shown in Figure 3.

Participant’s recall of IRS in the past 12 months was recorded for a very low number of participants (across surveys, 4.1% overall), mainly in SN (Supplemental Table 7).

Overall, usage of coils and repellents was limited, around 10% of the population in both surveys with variations per site between 1% and 40% according to the survey (Supplemental Table 7).

Year-to-year variation in reported fever and care-seeking behaviors. Fever in the last 24 hours was reported for approximately a quarter of the participants in both surveys, with differences across study sites ranging from 3.6% in BF Saponé to 63.8% in KE Kombewa in S1 and from 3.0% in SN Keur Socé to 71.6% in KE Kombewa in S2 (Supplemental Table 8). Across all sites, occurrence of fever was higher in P. falciparum–infected versus non-infected participants (35.6% versus 21.1% in S1 and 33.5% versus 22.5% in S2, respectively). In both surveys, fever was more frequently reported by participants with higher parasite densities (Supplemental Table 9).

In S1, 15.7% of participants had sought treatment against malaria or fever in the 14 days before the survey compared with 12.8% in S2, ranging from 0.0% to up to 33.3% depending on the study site and on the survey (Table 5, Supplemental Table 10). Plasmodium falciparum–infected children sought treatment against malaria or fever more often than non-infected children (20.1% versus 14.0% in S1 and 20.0% versus 9.8% in S2, respectively).

The proportion of participants hospitalized for malaria was 2.6% in S1 and 2.8% in S2, with no marked difference between P. falciparum–infected and non-infected participants 2.8% versus 2.5% in S1 and 3.4% versus 2.6% in S2, respectively; (Table 5, Supplemental Table 10).

Association between potential risk factors and P. falciparum infection. An exploratory multivariable model was used to assess the association between potential risk factors or malaria control interventions and P. falciparum infection. Across both surveys, houses equipped with electricity (odds ratio [OR]: S1 0.75 [95% CI: 0.61; 0.93]; S2 0.89 [95% CI: 0.80; 0.98]), cement/plaster walls versus mud (OR: S1 0.81 [95% CI: 0.69; 0.96]; S2 0.87 [95% CI: 0.76; 0.99]), and nets on all windows (OR: S1 0.73 [95% CI: 0.63; 0.84]; S2 0.79 [95% CI: 0.66; 0.94]) were associated with a lower risk of infection with P. falciparum. In addition, older age was associated with a higher risk of infection (OR: 1.14 [95% CI: 1.05; 1.24] in S1 and 1.09 [95% CI: 1.02; 1.16] in S2; Supplemental Tables 11 and 12).

Figure 4 represents a plain language summary, which elaborates on the epidemiologic study relevance that could be shared with patients by healthcare professionals.

**DISCUSSION**

Characterizing MTI in different SSA settings. Using PPR as a proxy, this study aims at characterizing MTI in different SSA settings, including areas in GH and KE where the RTS,S/AS01e malaria vaccine is currently introduced in the framework of the MVIP. More specifically, considering the WHO
recommendation to operate the MVIP in moderate to high transmission areas of SSA, PPiR in children aged 6 months to 4 years in Kintampo and Kombewa sites was high, ranging from 24.8% to 27.3% depending on the study site and the survey. Similar PPiR was estimated by Drakeley et al., indicating a stable mesoendemic MVI level (PPiR = 10–50%) in those areas.\textsuperscript{9,16} Plasmodium falciparum parasite prevalence varied largely between sites as it was expected from various preselected transmission intensity areas. In both surveys, the two sites in BF had the highest PPiR, and the two sites in SN, the lowest. Plasmodium falciparum parasite prevalence rates recorded in this study are in line with previous findings.\textsuperscript{9,16–18} Across most sites, the PPiR was lower in the 6-month to <5-year than in the 5-year to <10-year age-group. In addition, a risk factor analysis highlighted an association between older age and higher risk of infection. Those results corroborate the findings of other studies, identifying increasing age as a risk factor for carrying malaria blood stage parasites.\textsuperscript{9,16,19,20} This may potentially be explained by the fact that younger children benefit from a more focused usage of control interventions (bednets).\textsuperscript{20} Another explanation would be increased immunity in older children due to repeated exposure to the parasite.

### Table 3: Agreement between microscopy and QT-PCR test results by survey

| Study site | P. falciparum measured by microscopy | QT-PCR | Microscopy | Microscopy | Total N
|------------|-------------------------------------|--------|------------|------------|---------|
|            | Positive (N = 1,268) | Negative (N = 2,069) | Positive (N = 1,417) | Negative (N = 2,737) | Total N |
|            | n | % | n | % | N | n | % | n | % | N |
| Nouna, BF  | 347 | 90.1 | 38 | 9.9 | 385 | 400 | 82.1 | 87 | 17.9 | 487 |
| Spaïné, BF | 59 | 30.1 | 137 | 69.9 | 196 | 57 | 50.9 | 55 | 49.1 | 112 |
| Kintampo, GH | 34 | 16.0 | 178 | 84.0 | 212 | 78 | 27.6 | 205 | 72.4 | 283 |
| Kombewa, KE | 34 | 16.0 | 178 | 84.0 | 212 | 78 | 27.6 | 205 | 72.4 | 283 |
| Keur Socé, SN | 188 | 90.0 | 21 | 10.0 | 209 | 201 | 94.8 | 11 | 5.2 | 212 |
| Niakhar, SN | 43 | 12.1 | 313 | 87.9 | 356 | 80 | 20.6 | 308 | 79.4 | 388 |
| Korogwe, TZ | 8 | 1.4 | 549 | 98.6 | 557 | 28.5 | 96.6 | 2 | 3.4 | 3 |
| Overall total | 938 | 81.4 | 231 | 18.6 | 1,119 | 956 | 81.4 | 231 | 18.6 | 1,119 |

**BF** = Burkina Faso; **GH** = Ghana; **KE** = Kenya; **SN** = Senegal; **TZ** = Tanzania; n = number of participants in a given category; P. falciparum = Plasmodium falciparum; QT-PCR = quantitative PCR; % = percentage of participants with available results.

### Table 4: Bednet usage the night before the survey by study site and survey

| Site, country | Survey 1 (N = 4,208) | Survey 2 (N = 4,199) |
|--------------|----------------------|----------------------|
| Nouna, BF    | N | % (95% CI) | N | % (95% CI) |
| Spaïné, BF   | 547 | 90.3 (87.6; 92.5) | 530 | 88.3 (85.5; 90.8) |
| Kintampo, GH | 557 | 92.2 (89.8; 94.2) | 534 | 89.1 (86.4; 91.5) |
| Kombewa, KE  | 421 | 70.2 (66.3; 73.8) | 497 | 82.8 (79.6; 85.8) |
| Keur Socé, SN | 584 | 97.5 (95.9; 98.6) | 534 | 89.1 (86.4; 91.5) |
| Niakhar, SN  | 568 | 94.7 (92.6; 96.3) | 520 | 86.7 (83.4; 89.3) |
| Korogwe, TZ  | 544 | 90.3 (87.3; 92.7) | 595 | 99.2 (98.1; 99.7) |
| Overall      | 3,788 | 89.5 (88.6; 90.5) | 3,629 | 86.4 (85.3; 87.4) |

BF = Burkina Faso; GH = Ghana; KE = Kenya; SN = Senegal; TZ = Tanzania; n = number (% of children using a bednet the night before the visit in each site; N = total number of participants; 95% CI = exact 95% CI.)
leading to asymptomatic carriage and a lower probability to be treated than in symptomatic children.\textsuperscript{21,22}

Infection with \textit{P. malariae} was observed in a few participants, and infection with \textit{P. ovale} was rare, which supports observations from other studies conducted across SSA.\textsuperscript{23–26} Across all sites and surveys, only one \textit{P. vivax} infection was identified by microscopy in TZ, which differs from Twohig et al.’s\textsuperscript{27} recent findings of growing evidence of this species in SSA. A high proportion of co-infections with \textit{P. falciparum} was observed in \textit{P. malariae}–infected participants (between 70\% and 90\%) and \textit{P. ovale}–infected participants (around 60\%). Similarly, high percentages of \textit{P. falciparum} co-infection in \textit{P. malariae}–infected participants was previously reported in Guinea (97\%),\textsuperscript{23} Uganda (91\%),\textsuperscript{24} and in the Democratic Republic of the Congo (90\%),\textsuperscript{25} with lower percentages reported in rural BF (67\%)\textsuperscript{26} and Benin (34\%).\textsuperscript{23}

Approximately 20\% of participants positive for \textit{P. falciparum} by QT-PCR were undetected by microscopy. Classifying the study sites by MTI, this proportion was higher in low MTI sites. This might be explained by a relatively lower sensitivity of microscopy readings than nucleic acid related techniques, particularly in low parasite density infections, which are more frequently observed in low MTI areas.\textsuperscript{12,13} Nevertheless, the kappa statistic estimated a substantial agreement between microscopy and NAAT techniques using either qualitative or semi-quantitative real-time PCR.

Monitoring year-to-year variations in MTI and in the use of malaria control interventions. In addition, the present results are establishing a standardized baseline for further estimation of the year-to-year variations in \textit{PfPR} and in the use of malaria control interventions, which are key variables to be monitored before and after vaccine introduction in the MVIP areas. Little variation in \textit{P. falciparum} prevalence between the first two annual surveys was observed among most sites. Usage of bednets as a malaria control intervention was high in all sites and in both surveys (ranging 70–99\%). Some variation in the usage of bednets was observed in all sites, except in BF.

In S2, participants not using mosquito coils, insecticide sprays, or repellents against malaria vectors were significantly less likely to be infected with \textit{P. falciparum} (OR: S2 0.88 [95\% CI: 0.782; 0.988]); however, no such association was observed in S1. Various entomological studies have questioned the efficacy of repellents and coils as effective malaria prevention measures and highlighted the false sense of protection perceived by the user.\textsuperscript{28–30} Our data do not allow to draw robust conclusions at this stage, and the trend observed in S2 should be closely monitored in the subsequent cross-sectional surveys.

Study limitations. As with all interview–questionnaire-based studies, this study could have been subject to information or recall bias as data related to bednet usage, control interventions usage, and care-seeking behaviors were collected from parents’ recollection as opposed to objective observation. The impact of these potential information biases on the study results is estimated to be limited because of the proximity in time between the occurrence of the event for

\begin{table}[h]
\centering
\caption{Care-seeking behaviors (treatment sought for fever and hospitalization for malaria) according to \textit{P. falciparum} infection status by microscopy and survey.}
\begin{tabular}{lcccccc}
\hline
Care-seeking behavior & Survey & \multicolumn{2}{c}{\textit{P. falciparum} infected} & \multicolumn{2}{c}{\textit{P. falciparum} not infected} & \multicolumn{2}{c}{Total} \\
& & \textit{N} & \textit{N} & \% (95\% CI) & \textit{n} & \textit{N} & \% (95\% CI) & \textit{n} & \textit{N} & \% (95\% CI) \\
\hline
Treatment sought for malaria or fever in the previous 14 days & S1 & 238 & 1,187 & 20.1 (17.8; 22.4) & 424 & 3,021 & 14.0 (12.8; 15.3) & 662 & 4,208 & 15.7 (14.6; 16.9) \\
& S2 & 247 & 1,232 & 20.0 (17.8; 22.4) & 290 & 2,967 & 9.8 (8.7; 10.9) & 537 & 4,199 & 12.8 (11.8; 13.8) \\
Hospitalization for malaria in the past 3 months & S1 & 33 & 1,187 & 2.8 (1.9; 3.9) & 76 & 3,021 & 2.5 (2.0; 3.1) & 109 & 4,208 & 2.6 (2.1; 3.1) \\
& S2 & 42 & 1,232 & 3.4 (2.5; 4.6) & 77 & 2,967 & 2.6 (2.1; 3.2) & 119 & 4,199 & 2.8 (2.4; 3.4) \\
\hline
\end{tabular}
\label{tab:5}
\end{table}

\textit{n} (\%) = number (percentage) of children in each group; \textit{N} = total number of participants; \textit{P. falciparum} = \textit{Plasmodium falciparum}; S1 = survey 1; S2 = survey 2; 95\% CI = exact 95\% CI.
which information is collected and the interview itself. Another study limitation may be related to both sensitivity and specificity of microscopy slide readings. Slide reading performances may indeed vary depending on the parasite density and species identification. However, the kappa statistic estimated a substantial agreement between microscopy and qPCR, the latter being able to detect a higher number of low-density infections than microscopy and RDT. Moreover, this should mainly impact low transmission settings due to the high proportion of low-density infections.

**CONCLUSION**

The present article summarizes the results of the first two annual surveys of a larger malaria prevalence study aiming at characterizing MTI in light of the use of malaria control interventions and other environmental factors. Our results confirm that the high PPR observed in study sites that are part of the MVIP is in line with the WHO recommendation to operate the program in moderate to high transmission areas. In addition, our results are key to inform on the potential occurrence of annual fluctuations in MTI that may influence the assessment of the RTS,S/AS01 vaccine safety, effectiveness, and impact. The observations based on the first two surveys of this study do not indicate major temporal changes in terms of Plasmodium prevalence or use of malaria control interventions, but more surveys are needed to confirm this trend. The data generated in this study will be used to create variables to adjust the temporal and concurrent comparison analyses of the RTS,S/AS01 vaccine safety, impact, and effectiveness study. More specifically, the year- and site-specific PPR computed on unvaccinated study participants will be included as covariates in the regression models of the safety, impact, and effectiveness study to assess annual fluctuations and/or changes due to other malaria control interventions.

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GSK makes available anonymized individual participant data and associated documents from interventional clinical studies which evaluate medicines, upon approval of proposals, submitted to www.clinicalstudydatarequest.com. To access data for other types of GSK sponsored research, for study documents without patient level data, and for clinical studies not listed, please submit an enquiry via the website. All authors reviewed and commented on a draft version of the manuscript and gave their final approval for it to be submitted for publication.

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The following are supplemental files and will be published online only

**Supplemental Appendix**

This appendix has been provided by the authors to give readers additional information about their work.

Appendix “Estimating annual fluctuations in malaria transmission intensity and in the use of malaria control interventions in 5 sub-Saharan African countries”
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1. **SELECTION OF CHILDREN PARTICIPATING IN THE SURVEY**

The participant selection process was repeated each year independently meaning that the individuals were different in each cross-sectional survey except if they were re-selected in a subsequent survey by chance. The population listings generated from the demographic surveillance allowed for sampling of the required individuals according to stratification by age group as follows (the number of individuals was approximately plus or minus 5 children):

- 60 children aged 6 months to <1 year
- 120 children aged 1 year
- 120 children aged 2 years
- 50 children aged 3 years
- 50 children aged 4 years
- 40 children aged 5 years
- 40 children aged 6 years
- 40 children aged 7 years
- 40 children aged 8 years
- 40 children aged 9 years.

2. **DETERMINATION OF PARASITEMIA BY MICROSCOPY**

**Assessing parasite presence**

A 100-field examination of the thick film was conducted to assess presence of parasites and species.¹

*Negative result:* 100 fields free of parasites were to be read before a slide was declared negative.

*Positive result:* If parasites were present within reading of 100 fields, the slide was positive. Positive slides were examined for a further 100 fields to ensure all species present were detected.

**Identification of *Plasmodium* species**

Positive parasitemia identified on any thick blood film was always identified to species. This was done on thin blood film except in cases of low parasitemia.

**Parasite density counting against assumed 8000 leukocytes per microliter**

In this method of estimating parasite density, it was assumed that there were 8000 leukocytes per microliter of blood.
• If upon counting 200 leukocytes, 10 or more parasites had been counted, the results were to be recorded as the parasites per 200 leukocytes.

• If upon counting 200 leukocytes, 9 or fewer parasites had been recorded, the reader was to continue counting until 500 leukocytes had been counted and the number of parasites per 500 leukocytes were to be recorded.

• It should be noted that the count was to be by species, and counts for *P. falciparum* were to be made for both gametocytes and asexual parasites.

**Criteria for concordance for double reading of slides**

All slides were read twice, by two independent readers to quantify the *P. falciparum* parasite presence and density. If slides were judged to be discordant, a third independent read was to be organized in the following cases:

A. The result from one reader was negative and the one of the other was positive.

B. For high and medium positive parasitemia results (blood parasitemia $>400/\mu L$), the higher count divided by the lower count was $>2$.

C. For low parasitemia (blood parasitemia $\leq 400/\mu L$), the highest reading density was more than one log$_{10}$ higher than the lowest reading.

If the parasitemia result was high or medium in one slide and the result from the other slide reading was low, i.e. one was $>400/\mu L$ and the other was $\leq 400/\mu L$, criterion (C) was to be applied.

**Determination of final result**

If there were two concordant results, the final result was the geometric mean of the two readings.

If the first two readings were discordant then the final result was to follow the following principles:

• For cases of positive/negative discrepancy (A), the majority decision was to be adopted. If the decision was positive, the final result was the geometrical mean of the two positives.

• For cases of three positive reads (B and C), the final result was to be the geometric mean of the two geometrically closest readings.
3. DETERMINATION OF PARASITEMIA BY NUCLEIC ACID AMPLIFICATION TEST (NAAT)

Determination of parasitemia by nucleic acid amplification test (NAAT) in the study used the following tests:

- quantitative polymerase chain reaction (QT-PCR) for the detection of both DNA (asexual parasites) and RNA (gametocytes);
- quantitative nucleic acid sequence-based amplification (QT-NASBA) for the specific detection of RNA, identifying sexual stages parasites (gametocytes).

Both were performed at AMC (Academic Medical Center, Amsterdam, The Netherlands). Details for QT-PCR are presented in Error: Reference source not found et al, 2001 and for QT-NASBA in Error: Reference source not found et al, 2004.
4. **LIST OF PRE-DEFINED MALARIA CONTROL INTERVENTIONS AND POTENTIAL RISK FACTORS**

List of malaria control intervention explanatory variables tested in the multivariable logistical regression model:

| Variable                                                   | Comparison                        |
|------------------------------------------------------------|-----------------------------------|
| Malaria or fever treatment sought for in the past 14 days  | Yes vs. No                        |
| Malaria hospitalization in the last 3 months               | Yes vs. No                        |
| Antimalarial or any other medication consumed within 14 days prior to study visit | Yes vs. No                        |
| Antimalarial drug consumed in the past 14 days             | Yes vs. No                        |
| Other medication consumed over 14 days prior to study visit| Yes vs. No                        |
| Subject sleep under a bednet last night                    | Yes vs. No                        |
| New net (less than 1 year)                                 | No Bednet vs. No                  |
|                                                           | Yes vs. No                        |
| Impregnated bednet                                         | Yes vs. No                        |
|                                                           | No Bednet vs. No                  |
| Pierced/torn bednet                                        | Yes vs. No                        |
|                                                           | No Bednet vs. No                  |
| Number of holes                                            | Less than 5 vs. More or equal to 5|
|                                                           | No Pierced Bednet vs. More or equal to 5|
|                                                           | No Bednet vs. More or equal to 5  |
| Use of mosquito coils over 7 days                         | Yes vs. Missing/No                |
| Use of insecticide sprays over 7 days                     | Yes vs. Missing/No                |
| Use of commercial repellents over 7 days                  | Yes vs. Missing/No                |
| Use of traditional repellents over 7 days                 | Yes vs. Missing/No                |
| Use of none of above over 7 days                          | Yes vs. Missing/No                |
| Use of indoor residual spraying (IRS) in the past 12 months to spray interior | Yes vs. No                        |
| Use of indoor residual spraying - number of months ago     | >4 vs. 1-2                        |
|                                                           | No Residual Spray vs. 1-2         |
|                                                           | 3-4 vs. 1-2                       |

IRS = Indoor Residual Spraying, application of a residual insecticide to internal walls and ceilings of housing structures.²
List of potential risk factor explanatory variables tested in the multivariable logistical regression model:

| Variable                                                                 | Options                                                                                                                                 |
|-------------------------------------------------------------------------|----------------------------------------------------------------------------------------------------------------------------------------|
| Age (in years)                                                          | Continuous                                                                                                                             |
| Gender                                                                  | Male vs. Female                                                                                                                        |
| Number of persons living in the same part of the house                  | 4-5 vs. ≤3, >5 vs. ≤3                                                                                                                  |
| Number of persons enrolled into the study                                | 1 vs. 2, 3 vs. 2, >3 vs. 2                                                                                                              |
| Localization                                                            | Urban area vs. Rural area, Semi-Rural Area vs. Rural area                                                                                |
| Type of location                                                         | Town (>10,000 and < 50,000 habitants) vs. Countryside, Small city (>50,000 and < 1mil. habitants) vs. Countryside, Large city (>1 mil. habitants) vs. Countryside (<10,000) |
| Main house construction material: Walls                                  | Clay vs. Mud, Cement/Plaster vs. Mud, Brick vs. Mud, Other vs. Mud, Cement /Paint vs. Mud                                               |
| Main house construction material: Floor                                  | Carpet vs. Natural floor*, Ceramic tiles vs. Natural floor*, Parquet/ polished wood vs. Natural floor*, Clay vs. Natural floor*, Cement vs. Natural floor* |
| Main house construction material: Roof                                   | Tiles vs. Grass/Palm, Other vs. Grass/Palm, Iron sheet vs. Grass/Palm, Clay vs. Grass/Palm                                           |
| Main house construction material: Windows/eaves                          | Other vs. Open, No Windows vs. Open, Closed vs. Open, Partially open vs. Open                                                          |
| Main house construction material: Nets                                   | Nets present on some windows vs. Nets not present, Nets present on all windows vs. Nets not present, Other vs. Nets not present        |
| Main source of drinking water                                           | Closed water source† vs. Open water source††                                                                                         |
| Is the open source in the compound                                       | No open water vs. No, Yes vs. No                                                                                                      |
| Presence of electricity                                                  | Yes vs. No                                                                                                                             |

* Natural floor = earth, sand, dung.
** Rudimentary floor = wood, palm, bamboo.
† Closed water source (piped water, tube well, dug well, protected well).
†† Open water source (unprotected well, spring water, rainwater, tanker truck, surface water).
## 5. SUPPLEMENTAL TABLES

### 5.1. Study Population and Demographic Characteristics

Supplemental Table 1  Number of individuals included in the analysis by study site, age group and survey

| Study site       | 6M–<5Y |          | 5–<10Y |          | All ages |          |
|------------------|--------|----------|--------|----------|----------|----------|
|                  | Survey 1 | Survey 2 | Survey 1 | Survey 2 | Survey 1 | Survey 2 |
|                  | n       | n        | n       | n        | n        | n        |
| Nouna, BF        | 404     | 400      | 202     | 200      | 606      | 600      |
| Saponé, BF       | 403     | 399      | 201     | 200      | 604      | 599      |
| Kintampo, GH     | 400     | 400      | 200     | 200      | 600      | 600      |
| Kombewa, KE      | 402     | 401      | 197     | 198      | 599      | 599      |
| KeurSocé, SN     | 400     | 397      | 200     | 203      | 600      | 600      |
| NiakharSN        | 398     | 397      | 200     | 204      | 598      | 601      |
| Korogwe, TZ      | 401     | 400      | 200     | 200      | 601      | 600      |
| **All**          | 2,808   | 2,794    | 1,400   | 1,405    | 4,208    | 4,199    |

6M–<5Y = Individuals aged 6 months to less than 5 years at informed consent.
5–<10Y = Individuals aged 5 years to less than 10 years at informed consent.
n = number of individuals in a given category.
BF = Burkina Faso; GH = Ghana; KE = Kenya; SN = Senegal; TZ = Tanzania.
### Supplemental Table 2  Summary of demographic characteristics of individuals by study site and survey

| Study site | Characteristics | Survey 1 | Survey 2 |
|------------|----------------|----------|----------|
| Nouna, BF  | N=606          | N=600    |          |
|            | Age at informed consent (years) | Mean±SD: 3.99±2.69 | 4.00±2.74 |
|            |                  | Range: 0.53–9.90 | 0.56–9.97 |
|            | Age group (years) | 6M–<5Y; n (%): 404 (66.7) | 400 (66.7) |
|            |                  | 5–<10Y; n (%): 202 (33.3) | 200 (33.3) |
|            | Gender           | Female; n (%): 304 (50.2) | 300 (50.0) |
|            |                  | Male; n (%): 302 (49.8) | 300 (50.0) |
| Saponé, BF | N=604           | N=599    |          |
|            | Age at informed consent (years) | Mean±SD: 4.05±2.77 | 4.06±2.75 |
|            |                  | Range: 0.51–9.93 | 0.50–9.89 |
|            | Age group (years) | 6M–<5Y; n (%): 403 (66.7) | 399 (66.6) |
|            |                  | 5–<10Y; n (%): 201 (33.3) | 200 (33.4) |
|            | Gender           | Female; n (%): 295 (48.8) | 283 (47.3) |
|            |                  | Male; n (%): 309 (51.2) | 316 (52.8) |
| Kintampo, GH| N=600           |          | N=600    |
|            | Age at informed consent (years) | Mean±SD: 3.99±2.74 | 4.02±2.74 |
|            |                  | Range: 0.55–9.81 | 0.58–9.94 |
|            | Age group (years) | 6M–<5Y; n (%): 400 (66.7) | 400 (66.7) |
|            |                  | 5–<10Y; n (%): 200 (33.3) | 200 (33.3) |
|            | Gender           | Female; n (%): 278 (46.3) | 297 (49.5) |
|            |                  | Male; n (%): 322 (53.7) | 303 (50.5) |
| Kombewa, KE| N=599           |          | N=599    |
|            | Age at informed consent (years) | Mean±SD: 4.05±2.78 | 4.08±2.75 |
|            |                  | Range: 0.57–10.00 | 0.58–9.97 |
|            | Age group (years) | 6M–<5Y; n (%): 402 (67.1) | 401 (66.9) |
|            |                  | 5–<10Y; n (%): 197 (32.9) | 198 (33.1) |
|            | Gender           | Female; n (%): 279 (46.6) | 315 (52.6) |
|            |                  | Male; n (%): 320 (53.4) | 284 (47.4) |
| KeurSocé, SN| N=600           |          | N=600    |
|            | Age at informed consent (years) | Mean±SD: 4.04±2.74 | 4.07±2.76 |
|            |                  | Range: 0.55–9.96 | 0.60–9.92 |
|            | Age group (years) | 6M–<5Y; n (%): 400 (66.7) | 397 (66.2) |
|            |                  | 5–<10Y; n (%): 200 (33.3) | 203 (33.8) |
|            | Gender           | Female; n (%): 303 (50.5) | 307 (51.2) |
|            |                  | Male; n (%): 297 (49.5) | 293 (48.8) |
| Niakhar, SN| N=598           |          | N=601    |
|            | Age at informed consent (years) | Mean±SD: 4.02±2.77 | 4.08±2.77 |
|            |                  | Range: 0.54–9.99 | 0.70–9.83 |
|            | Age group (years) | 6M–<5Y; n (%): 398 (66.6) | 397 (66.1) |
|            |                  | 5–<10Y; n (%): 200 (33.4) | 204 (33.9) |
|            | Gender           | Female; n (%): 309 (51.7) | 280 (46.6) |
|            |                  | Male; n (%): 289 (48.3) | 321 (53.4) |
| Korogwe, TZ| N=601           |          | N=600    |
|            | Age at informed consent (years) | Mean±SD: 4.01±2.75 | 4.03±2.73 |
|            |                  | Range: 0.50–9.95 | 0.54–9.97 |
|            | Age group (years) | 6M–<5Y; n (%): 401 (66.7) | 400 (66.7) |
|            |                  | 5–<10Y; n (%): 200 (33.3) | 200 (33.3) |
| Study site | Characteristics | Survey 1 | Survey 2 |
|------------|-----------------|----------|----------|
|            | Gender          |          |          |
|            | Female; n (%)   | 282 (46.9) | 306 (51.0) |
|            | Male; n (%)     | 319 (53.1) | 294 (49.0) |
| Overall    | N=4,208         |          |          |
|            | Age at informed consent (years) | Mean±SD |          |          |
|            | Range           | 0.50–10.00 | 0.50–9.97 |
|            | Age group (years) |          |          |
|            | 6M–<5Y; n (%)   | 2,808 (66.7) | 2,794 (66.5) |
|            | 5–<10Y; n (%)   | 1,400 (33.3) | 1,405 (33.5) |
|            | Gender          |          |          |
|            | Female; n (%)   | 2,050 (48.7) | 2,088 (49.7) |
|            | Male; n (%)     | 2,158 (51.3) | 2,111 (50.3) |

N = total number of individuals overall or per site.
n = number of individuals in a given category.
SD = standard deviation.
Range = minimum and maximum values.
6M–<5Y = Individuals aged 6 months to less than 5 years at informed consent.
5–<10Y = Individuals aged 5 years to less than 10 years at informed consent.
BF = Burkina Faso; GH = Ghana; KE = Kenya; SN = Senegal; TZ = Tanzania.
## 5.2. *Plasmodium* parasite prevalence

**Supplemental Table 3**  
*P. falciparum* results (asexual parasites and gametocytes) by study site and survey measured by NAAT (QT-PCR and QT-NASBA)

| Study site       | Survey 1 | Presence of gametocytes measured by QT-NASBA* | Survey 2 | Presence of gametocytes measured by QT-NASBA* |
|------------------|----------|-----------------------------------------------|----------|-----------------------------------------------|
|                  | *P. falciparum measured by QT-PCR* |                             | *P. falciparum measured by QT-PCR* |                             |
| Nouna, BF; n (%) | N=581    | N'=406 (69.9) | N=599    | N'=456 (76.3) |
| Positive         | 406 (69.9) | 205 (50.5)    | 457 (76.3) | 268 (58.8)    |
| Negative         | 175 (30.1) | 201 (49.5)    | 142 (23.7) | 188 (41.2)    |
| Saponé, BF; n (%) | N=426    | N'=245 (57.7) | N=599    | N'=390 (65.1) |
| Positive         | 246 (57.7) | 145 (59.2)    | 390 (65.1) | 229 (58.7)    |
| Negative         | 180 (42.3) | 100 (40.8)    | 209 (34.9) | 161 (41.3)    |
| Kintampo, GH; n  | N=565    | N'=229 (40.9) | N=600    | N'=281 (46.8) |
| Positive         | 231 (40.9) | 94 (41.0)     | 281 (46.8) | 134 (47.7)    |
| Negative         | 334 (59.1) | 135 (59.0)    | 319 (53.2) | 147 (52.3)    |
| Kombewa, KE, n   | N=599    | N'=262 (45.7) | N=599    | N'=236 (41.0) |
| Positive         | 262 (45.7) | 81 (30.9)     | 236 (39.4) | 120 (50.8)    |
| Negative         | 337 (54.3) | 181 (69.1)    | 363 (60.6) | 116 (49.2)    |
| KeurSocé, SN**; n| N=565    | N'=15 (2.7)   | N=601    | N'=14 (2.3)   |
| Positive         | -        | -              | -        | -              |
| Negative         | -        | -              | 7 (1.3)  | 4 (66.7)      |
| Niakhar, SN; n   | N=565    | N'=10 (2.7)   | N=601    | N'=32 (5.3)   |
| Positive         | 108 (18.0) | 21 (19.4)     | 32 (5.3) | 11 (34.4)     |
| Negative         | 493 (82.0) | 87 (80.6)     | 568 (94.7) | 21 (65.6)     |

NAAT = Nucleic Acid Amplification Test.  
QT-PCR = Quantitative Polymerase Chain Reaction.  
QT-NASBA = Quantitative Nucleic Acid Sequence-Based Amplification.  
BF = Burkina Faso; GH = Ghana; KE = Kenya; SN = Senegal; TZ = Tanzania.  
N = number of individuals tested by QT-PCR for detection of *P. falciparum DNA and RNA* per site.  
N' = number of individuals tested positive by QT-PCR being tested by QT-NASBA for detection of *P. falciparum gametocytes RNA* per site.  
n = number of individuals in a given category.  
*Test for detection of gametocytes, QT-NASBA (RNA specific), was only performed on samples positive by QT-PCR (detecting both DNA and RNA).  
**KeurSocé, SN had no valid NAAT results in Survey 1.  
† Only individuals with available results are included in N or N'.
Supplemental Table 4 Number of individuals carrying gametocytes measured by microscopy compared to gametocytes results measured by QT-NASBA by study site and by survey

| Presence of gametocytes measured by microscopy, n (%) | Presence of Gametocytes (measured by QT-NASBA) | Total |
|------------------------------------------------------|-------------------------------------------------|-------|
|                                                      | Positive | Negative |       |
| **Study site**                                       |          |          |       |
| Nouna, BF                                            |          |          |       |
| Survey 1                                             | N=205    | N=268    | N=406 |
| Survey 2                                             | N=201    | N=188    | N=456 |
| Presence of gametocytes                              | 66 (32.2)| 69 (25.7)| 86 (21.2)| 76 (16.7)|
|                                                      | 20 (10.0)| 7 (3.7)  | 96 (39.2)| 105 (26.9)|
| Saponé, BF                                           |          |          |       |
| Survey 1                                             | N=145    | N=229    | N=245 |
| Survey 2                                             | N=100    | N=161    | N=390 |
| Presence of gametocytes                              | 81 (55.9)| 89 (38.9)| 96 (39.2)| 105 (26.9)|
|                                                      | 15 (15.0)| 16 (9.9) | 13 (5.7) | 29 (10.3) |
| Kintampo, GH                                         |          |          |       |
| Survey 1                                             | N=94     | N=134    | N=229 |
| Survey 2                                             | N=135    | N=147    | N=281 |
| Presence of gametocytes                              | 12 (12.8)| 24 (17.9)| 13 (5.7) | 29 (10.3) |
|                                                      | 1 (0.7)  | 5 (3.4)  | 22 (8.4) | 9 (3.8)    |
| Kombewa, KE                                          |          |          |       |
| Survey 1                                             | N=81     | N=120    | N=262 |
| Survey 2                                             | N=181    | N=116    | N=236 |
| Presence of gametocytes                              | 18 (22.2)| 7 (5.8)  | 22 (8.4) | 9 (3.8)    |
|                                                      | 4 (2.2)  | 2 (1.7)  | 0       |
| KeurSocé, SN*                                        |          |          |       |
|                                                      | -        | N=4      | -      |
|                                                      | 3 (75.0) | -        | 3 (50.0)|
| Niakhar, SN                                          |          |          |       |
| Survey 1                                             | N=2      | N=1      | N=15   |
| Survey 2                                             | N=13     | N=14     | N=14   |
| Presence of gametocytes                              | 1 (50.0)| 0        | 2 (13.3)| 0        |
|                                                      | 1 (7.7)  | 0        |
| Korogwe, TZ                                          |          |          |       |
| Survey 1                                             | N=21     | N=87     | N=108  |
| Survey 2                                             | N=21     | N=32     | N=108  |
| Presence of gametocytes                              | 7 (33.3)| 0        | 8 (7.4)| 1 (3.1)  |
|                                                      | 1 (1.1)  | 1 (4.8)  | 0      |
| Overall                                              | N=548    | N=767    | N=1,265| N=1,415  |
|                                                      | N=717    | N=648    | N=1,265| N=1,415  |
| Presence of gametocytes                              | 185 (33.8)| 192 (25.0)| 227 (17.9)| 223 (15.8)|
|                                                      | 42 (5.9) | 31 (4.8) | 0   |

QT-PCR = Quantitative Polymerase Chain Reaction.
QT-NASBA = Quantitative Nucleic Acid Sequence-Based Amplification.
N = number of individuals with positive parasitemia as measured by QT-PCR in the given category.
n = number of gametocyte positive individuals measured by microscopy in the given category.
*KeurSocé, SN had no valid QT-PCR results in Survey 1.
BF = Burkina Faso; GH = Ghana; KE = Kenya; SN = Senegal; TZ = Tanzania.
Supplemental Table 5  Prevalence of *Plasmodium* species other than *P. falciparum* measured by microscopy by study site, *P. falciparum* infection status and survey

| Study site   | *Plasmodium* parasitemia; n (%) | *Pf* infected | *Pf* not infected | Total |
|-------------|----------------------------------|---------------|-------------------|-------|
|             | Survey 1 | Survey 2 | Survey 1 | Survey 2 | Survey 1 | Survey 2 | Survey 1 | Survey 2 |
| Nouna, BF   | N=403     | N=488     | N=203     | N=112     | N=606     | N=600     |
|             | *P. malariae* | 2 (0.5) | 30 (6.1) | 2 (1.0) | 27 (24.1) | 4 (0.7) | 57 (9.5) |
|             | *P. vivax* | 0 | 0 | 0 | 0 | 0 | 0 |
|             | *P. ovale* | 1 (0.2) | 0 | 2 (1.0) | 0 | 3 (0.5) | 0 |
| Saponé, BF  | N=299     | N=316     | N=305     | N=283     | N=604     | N=599     |
|             | *P. malariae* | 28 (9.4) | 29 (9.2) | 3 (1.0) | 27 (24.1) | 31 (5.1) | 35 (5.8) |
|             | *P. vivax* | 0 | 0 | 0 | 0 | 0 | 0 |
|             | *P. ovale* | 1 (0.3) | 0 | 0 | 0 | 1 (0.2) | 0 |
| Kintampo, GH| N=214     | N=212     | N=386     | N=388     | N=600     | N=600     |
|             | *P. malariae* | 9 (4.2) | 15 (7.1) | 2 (0.5) | 3 (0.8) | 11 (1.8) | 18 (3.0) |
|             | *P. vivax* | 0 | 0 | 0 | 0 | 0 | 0 |
|             | *P. ovale* | 3 (1.4) | 1 (0.5) | 5 (1.3) | 1 (0.3) | 8 (1.3) | 2 (0.3) |
| Kombewa, KE | N=196     | N=189     | N=403     | N=410     | N=599     | N=599     |
|             | *P. malariae* | 16 (8.2) | 16 (8.5) | 0 | 1 (0.2) | 16 (2.7) | 17 (2.8) |
|             | *P. vivax* | 0 | 0 | 0 | 0 | 0 | 0 |
|             | *P. ovale* | 8 (4.1) | 4 (2.1) | 0 | 3 (0.7) | 8 (1.3) | 7 (1.2) |
| KeurSocé, SN| N=3       | N=6       | N=597     | N=594     | N=600     | N=600     |
|             | *P. malariae* | 0 | 0 | 0 | 0 | 0 | 0 |
|             | *P. vivax* | 0 | 0 | 0 | 0 | 0 | 0 |
|             | *P. ovale* | 0 | 0 | 0 | 0 | 0 | 0 |
| Niakhar, SN | N=9       | N=3       | N=589     | N=598     | N=598     | N=601     |
|             | *P. malariae* | 0 | 0 | 0 | 0 | 0 | 0 |
|             | *P. vivax* | 0 | 0 | 0 | 0 | 0 | 0 |
|             | *P. ovale* | 0 | 0 | 0 | 0 | 0 | 0 |
| Korogwe, TZ | N=63      | N=18      | N=538     | N=582     | N=601     | N=600     |
|             | *P. malariae* | 0 | 1 (5.6) | 0 | 0 | 0 | 1 (0.2) |
|             | *P. vivax* | 0 | 1 (5.6) | 0 | 0 | 0 | 1 (0.2) |
|             | *P. ovale* | 0 | 1 (5.6) | 3 (0.6) | 0 | 3 (0.5) | 1 (0.2) |
| Overall     | N=1,187   | N=1,232   | N=3,021   | N=2,967   | N=4,208   | N=4,199   |
|             | *P. malariae* | 55 (4.6) | 91 (7.4) | 7 (0.2) | 37 (1.2) | 62 (1.5) | 128 (3.0) |
|             | *P. vivax* | 0 | 1 (0.1) | 0 | 0 | 0 | 1 (0.02) |
|             | *P. ovale* | 13 (1.1) | 6 (0.5) | 10 (0.3) | 4 (0.1) | 23 (0.5) | 10 (0.2) |

*Pf* infected = Individuals infected with *P. falciparum* parasitemia measured by microscopy.
*Pf* not infected = Individuals not infected with *P. falciparum* parasitemia measured by microscopy.
N = total number of individuals overall or per site.
n = number of individuals in a given category.
BF = Burkina Faso; GH = Ghana; KE = Kenya; SN = Senegal; TZ = Tanzania.
### 5.3. Malaria control interventions

Supplemental Table 6 Number of individuals having slept under a bednet the night before the visit and characterization of bednets by study site and survey

| Study site | Survey 1 | Survey 2 |
|------------|----------|----------|
| Nouna, BF  | N=606    | N=600    |
| Participant slept under a bednet the night before the visit | n=547 | 530 |
| % (95% CI) | 90.3 (87.6;92.5) | 88.3 (85.5;90.8) |
| New bednet (less than 1 year)* | n=428 | 218 |
| % (95% CI) | 78.2 (74.5;81.6) | 41.1 (36.9;45.5) |
| Impregnated*bednet* | n=525 | 501 |
| % (95% CI) | 96.0 (94.0;97.5) | 94.5 (92.2;96.3) |
| Pierced/torn bednet* | n=59 | 158 |
| % (95% CI) | 10.8 (8.3;13.7) | 29.8 (25.9;33.9) |
| Saponé, BF  | N=604    | N=599    |
| Participant slept under a bednet the night before the visit | n=557 | 534 |
| % (95% CI) | 92.2 (89.8;94.2) | 89.1 (86.4;91.5) |
| New bednet (less than 1 year)* | n=496 | 303 |
| % (95% CI) | 89.0 (86.2;91.5) | 56.7 (52.4;61.0) |
| Impregnated*bednet* | n=548 | 521 |
| % (95% CI) | 98.4 (97.0;99.3) | 97.6 (95.9;98.7) |
| Pierced/torn bednet* | n=40 | 61 |
| % (95% CI) | 7.2 (5.2;9.7) | 11.4 (8.9;14.4) |
| Kintampo, GH | N=600    | N=600    |
| Participant slept under a bednet the night before the visit | n=421 | 497 |
| % (95% CI) | 70.2 (66.3;73.8) | 82.8 (79.6;85.8) |
| New bednet (less than 1 year)* | n=217 | 492 |
| % (95% CI) | 51.5 (46.7;56.4) | 99.0 (97.7;99.7) |
| Impregnated*bednet* | n=167 | 495 |
| % (95% CI) | 39.7 (35.0;44.5) | 99.6 (96.6;100) |
| Pierced/torn bednet* | n=83 | 15 |
| % (95% CI) | 19.7 (16.0;23.8) | 3.0 (1.7;4.9) |
| Kombewa, KE | N=599    | N=599    |
| Participant slept under a bednet the night before the visit | n=584 | 534 |
| % (95% CI) | 97.5 (95.9;98.6) | 89.1 (86.4;91.5) |
| New bednet (less than 1 year)* | n=410 | 239 |
| % (95% CI) | 70.2 (66.3;73.9) | 44.8 (40.5;49.1) |
| Impregnated*bednet* | n=273 | 52 |
| % (95% CI) | 46.7 (42.6;50.9) | 9.7 (7.4;12.6) |
| Pierced/torn bednet* | n=153 | 188 |
| % (95% CI) | 26.2 (22.7;30.0) | 35.2 (31.2;39.4) |
| KeurSocé, SN | N=600    | N=600    |
| Participant slept under a bednet the night before the visit | n=568 | 520 |
| % (95% CI) | 94.7 (92.6;96.3) | 86.7 (83.7;89.3) |
| New bednet (less than 1 year)* | n=423 | 212 |
| % (95% CI) | 74.5 (70.7;78.0) | 40.8 (36.5;45.1) |
| Impregnated*bednet* | n=314 | 271 |
| % (95% CI) | 55.3 (51.1;59.4) | 52.1 (47.7;56.5) |
| Pierced/torn bednet* | n=50 | 85 |
| % (95% CI) | 8.8 (6.6;11.4) | 16.3 (13.3;19.8) |
| Study site | Survey 1 | Survey 2 |
|------------|----------|----------|
| Niaakar, SN |          |          |
| N=598      | N=601    |          |
| Participant slept under a bednet the night before the visit | n=547 | 418 | 91.5 (88.9;93.6) | 69.6 (65.7;73.2) |
| New bednet (less than 1 year)* | n=482 | 60 | 88.1 (85.1;90.7) | 14.4 (11.1;18.1) |
| Impregnated*bednet* | n=481 | 418 | 87.9 (84.9;90.5) | 100 (99.1;100) |
| Pierced/torn bednet* | n=25 | 296 | 4.6 (3.0;6.7) | 70.8 (66.2;75.1) |
| Korogwe, TZ |          |          |
| N=601 | N=600 |
| Participant slept under a bednet the night before the visit | n=544 | 595 | 90.5 (87.9;92.7) | 99.2 (98.1;99.7) |
| New bednet (less than 1 year)* | n=68 | 557 | 12.5 (9.8;15.6) | 93.6 (91.3;95.4) |
| Impregnated*bednet* | n=282 | 567 | 51.8 (47.5;56.1) | 95.3 (93.3;96.9) |
| Pierced/torn bednet* | n=496 | 69 | 91.2 (88.5;93.4) | 11.6 (9.1;14.4) |
| Overall | N=4,208 | N=4,199 |
| Participant slept under a bednet the night before the visit | n=3,768 | 3,628 | 89.5 (88.6;90.5) | 86.4 (85.3;87.4) |
| New bednet (less than 1 year)* | n=2,524 | 2,081 | 67.0 (65.5;68.5) | 57.4 (55.7;59.0) |
| Impregnated*bednet* | n=2,590 | 2,825 | 68.7 (67.2;70.2) | 77.9 (76.5;79.2) |
| Pierced/torn bednet* | n=906 | 872 | 24.0 (22.7;25.4) | 24.0 (22.7;25.5) |

95% CI = Exact 95% confidence interval.
N = total number of individuals overall or per site.
n = number of individuals in a given category.
*Denominator = number of individuals who slept under a bednet the night before the visit.
†Dipped in liquid insecticide before or after purchase.
BF = Burkina Faso; GH = Ghana; KE = Kenya; SN = Senegal; TZ = Tanzania.
| Study site      | Total | Survey 1 | Survey 2 |
|----------------|-------|----------|----------|
|                |       | N=606    | N=600    |
| **Nouna, BF**  |       |          |          |
| Use of mosquito coils over 7 days | n | 109 | 211 |
| % (95% CI)     |      | 18.0 (15.0;21.3) | 35.2 (31.3;39.1) |
| Use of insecticide sprays over 7 days | n | 14 | 14 |
| % (95% CI)     |      | 2.3 (1.3;3.8) | 2.3 (1.3;3.9) |
| Use of commercial repellents over 7 days | n | 9 | 14 |
| % (95% CI)     |      | 1.5 (0.7;2.8) | 2.3 (1.3;3.9) |
| Use of traditional repellents over 7 days | n | 1 | 2 |
| % (95% CI)     |      | 0.2 (0.0;0.9) | 0.3 (0.0;1.2) |
| Use of none of the above over 7 days | n | 473 | 364 |
| % (95% CI)     |      | 78.1 (74.5;81.3) | 60.7 (56.6;64.6) |
| Use of IRS in past 12 months | n | 0 | 14 |
| % (95% CI)     |      | 0 (0.0;0.6) | 2.3 (1.3;3.9) |
| **Saponé, BF** |       | N=604    | N=599    |
| Use of mosquito coils over 7 days | n | 1 | 16 |
| % (95% CI)     |      | 0.2 (0.0;0.9) | 2.7 (1.5;4.3) |
| Use of insecticide sprays over 7 days | n | 3 | 0 |
| % (95% CI)     |      | 0 (0.0;0.6) | 0.5 (0.1;1.5) |
| Use of commercial repellents over 7 days | n | 1 | 3 |
| % (95% CI)     |      | 0.2 (0.0;0.9) | 0.5 (0.1;1.5) |
| Use of traditional repellents over 7 days | n | 4 | 0 |
| % (95% CI)     |      | 0.7 (0.2;1.7) | 0 (0.0;0.6) |
| Use of none of the above over 7 days | n | 598 | 577 |
| % (95% CI)     |      | 99.0 (97.9;99.6) | 96.3 (94.5;97.7) |
| Use of IRS in past 12 months | n | 0 | 0 |
| % (95% CI)     |      | 0 (0.0;0.6) | 0 (0.0;0.6) |
| **Kintampo, GH** |     | N=600    | N=600    |
| Use of mosquito coils over 7 days | n | 84 | 30 |
| % (95% CI)     |      | 14.0 (11.3;17.0) | 5.0 (3.4;7.1) |
| Use of insecticide sprays over 7 days | n | 25 | 24 |
| % (95% CI)     |      | 4.2 (2.7;6.1) | 4.0 (2.6;5.9) |
| Use of commercial repellents over 7 days | n | 0 | 0 |
| % (95% CI)     |      | 0 (0.0;0.6) | 0 (0.0;0.6) |
| Use of traditional repellents over 7 days | n | 1 | 0 |
| % (95% CI)     |      | 0.2 (0.0;0.9) | 0 (0.0;0.6) |
| Use of none of the above over 7 days | n | 490 | 560 |
| % (95% CI)     |      | 81.7 (78.3;84.7) | 93.3 (91.0;95.2) |
| Use of IRS in past 12 months | n | 2 | 0 |
| % (95% CI)     |      | 0.3 (0.0;1.2) | 0 (0.0;0.6) |
| **Kombewa, KE** |     | N=599    | N=599    |
| Use of mosquito coils over 7 days | n | 23 | 7 |
| % (95% CI)     |      | 3.8 (2.4;5.7) | 1.2 (0.5;2.4) |
| Use of insecticide sprays over 7 days | n | 5 | 0 |
| % (95% CI)     |      | 0.8 (0.3;1.9) | 0 (0.0;0.6) |
| Use of commercial repellents over 7 days | n | 1 | 2 |
| % (95% CI)     |      | 0.2 (0.0;0.9) | 0.3 (0.0;1.2) |
| Use of traditional repellents over 7 days | n | 2 | 0 |
| % (95% CI)     |      | 0.3 (0.0;1.2) | 0 (0.0;0.6) |
| Use of none of the above over 7 days | n | 568 | 590 |
| % (95% CI)     |      | 94.8 (92.7;96.5) | 98.5 (97.2;99.3) |
| Use of IRS in past 12 months | n | 0 | 0 |
| % (95% CI)     |      | 0 (0.0;0.6) | 0 (0.0;0.6) |
| **KeurSocé, SN** |     | N=600    | N=600    |
| n | 8 | 4 |
| Study site       | Total                          |
|------------------|--------------------------------|
|                  | Use of mosquito coils over 7 days | % (95% CI) | 1.3 (0.6;2.6) | 0.7 (0.2;1.7) |
|                  | Use of insecticide sprays over 7 days | % (95% CI) | 0.8 (0.3;1.9) | 0 (0.0;0.6)  |
|                  | Use of commercial repellents over 7 days | % (95% CI) | 0.2 (0.0;0.9) | 0.2 (0.0;0.9) |
|                  | Use of traditional repellents over 7 days | % (95% CI) | 1.8 (0.9;3.3) | 13.5 (10.9;16.5) |
|                  | Use of none of the above over 7 days | % (95% CI) | 96.8 (95.1;98.1) | 85.7 (82.6;88.4) |
|                  | Use of IRS in past 12 months | % (95% CI) | 47.7 (43.6;51.7) | 0 (0.0;0.6)  |

**Niakhar, SN**

|                  | N=598                          | N=601                          |
|------------------|--------------------------------|--------------------------------|
|                  | Use of mosquito coils over 7 days | % (95% CI) | 2.2 (1.2;3.7) | 1.8 (0.9;3.3) |
|                  | Use of insecticide sprays over 7 days | % (95% CI) | 0.5 (0.1;1.5) | 0.7 (0.2;1.7) |
|                  | Use of commercial repellents over 7 days | % (95% CI) | 7.4 (5.4;9.8) | 0 (0.0;0.6)  |
|                  | Use of traditional repellents over 7 days | % (95% CI) | 17.1 (14.1;20.3) | 3.7 (2.3;5.5) |
|                  | Use of none of the above over 7 days | % (95% CI) | 79.8 (76.3;82.9) | 94.0 (91.8;95.8) |
|                  | Use of IRS in past 12 months | % (95% CI) | 2.0 (1.0;3.5) | 4.7 (3.1;6.7) |

**Korogwe, TZ**

|                  | N=601                          | N=600                          |
|------------------|--------------------------------|--------------------------------|
|                  | Use of mosquito coils over 7 days | % (95% CI) | 1.3 (0.6;2.6) | 4.3 (2.8;6.3)  |
|                  | Use of insecticide sprays over 7 days | % (95% CI) | 0.0 (0.0;0.6) | 3.3 (2.0;5.1)  |
|                  | Use of commercial repellents over 7 days | % (95% CI) | 0.0 (0.0;0.6) | 0.3 (0.0;1.2)  |
|                  | Use of traditional repellents over 7 days | % (95% CI) | 0.2 (0.0;0.9) | 0.2 (0.0;0.9)  |
|                  | Use of none of the above over 7 days | % (95% CI) | 98.5 (97.2;99.3) | 92.0 (89.5;94.0) |
|                  | Use of IRS in past 12 months | % (95% CI) | 0 (0.0;0.6) | 0 (0.0;0.6)  |

**Overall**

|                  | N=4,208                         | N=4,199                         |
|------------------|---------------------------------|---------------------------------|
|                  | Use of mosquito coils over 7 days | % (95% CI) | 5.8 (5.2;6.6) | 7.3 (6.5;8.1)  |
|                  | Use of insecticide sprays over 7 days | % (95% CI) | 1.2 (0.9;1.6) | 1.5 (1.2;2.0)  |
|                  | Use of commercial repellents over 7 days | % (95% CI) | 1.3 (1.0;1.7) | 0.5 (0.3;0.8)  |
|                  | Use of traditional repellents over 7 days | % (95% CI) | 2.9 (2.4;3.5) | 2.5 (2.1;3.0)  |
|                  | Use of none of the above over 7 days | % (95% CI) | 89.8 (88.9;90.7) | 88.6 (87.6;89.6) |
|                  | Use of IRS in past 12 months | % (95% CI) | 7.1 (6.4;7.9) | 1.0 (0.7;1.3)  |

*N = total number of individuals overall or per site.*

*n = number of individuals in a given category.*

*95% CI = Exact 95% confidence limits.*

*IRS = Indoor Residual Spraying, application of a residual insecticide to internal walls and ceilings of housing structures.*

*BF = Burkina Faso; GH = Ghana; KE = Kenya; SN = Senegal; TZ = Tanzania.*
### Care-seeking behaviours

**Supplemental Table 8**  Number of individuals presenting with fever reported in the last 24 hours and measured at visit by study site. *P. falciparum* infection status and survey

| Study site | Survey 1 | Survey 2 | Survey 1 | Survey 2 | Total |
|------------|----------|----------|----------|----------|-------|
|            | *Pf* infected | *Pf* not infected | *Pf* infected | *Pf* not infected |       |
| Nouna, BF  | N=403     | N=488    | N=203    | N=112    | N=606 |
| Fever in the last 24 hours* | n=138 | 120 | 38 | 29 | 176 |
| % (95% CI) | 34.2 (29.6;39.1) | 24.6 (20.8;28.7) | 18.7 (13.6;24.8) | 25.9 (18.1;35.0) | 29.0 (25.5;32.8) |
| Fever at visit** | n=29 | 32 | 3 | 5 | 32 |
| % (95% CI) | 7.2 (4.9;10.2) | 6.6 (4.5;9.1) | 1.5 (0.3;4.3) | 4.5 (1.5;10.1) | 5.3 (3.6;7.4) |
| Saponé, BF | N=299 | N=316 | N=305 | N=283 | N=604 |
| Fever in the last 24 hours* | n=15 | 32 | 7 | 8 | 22 |
| % (95% CI) | 5.0 (2.8;8.1) | 10.1 (7.0;14.0) | 2.3 (0.9;4.7) | 5.3 (3.0;8.6) | 3.6 (2.3;5.5) |
| Fever at visit** | n=13 | 25 | 8 | 8 | 21 |
| % (95% CI) | 4.4 (2.3;7.3) | 7.9 (5.2;11.5) | 2.6 (1.1;5.1) | 2.8 (1.2;5.5) | 3.5 (2.2;5.3) |
| Kintampo, GH | N=214 | N=212 | N=386 | N=388 | N=600 |
| Fever in the last 24 hours* | n=102 | 105 | 83 | 142 | 185 |
| % (95% CI) | 47.7 (40.8;54.6) | 49.5 (42.6;56.5) | 21.5 (17.5;25.9) | 36.6 (31.8;41.6) | 30.8 (27.2;34.7) |
| Fever at visit** | n=22 | 22 | 11 | 8 | 33 |
| % (95% CI) | 10.3 (6.6;15.2) | 10.4 (6.6;15.3) | 2.8 (1.4;5.0) | 2.1 (0.9;4.0) | 5.5 (3.8;7.6) |
| Kombewa, KE | N=196 | N=189 | N=403 | N=410 | N=599 |
| Fever in the last 24 hours* | n=127 | 146 | 255 | 283 | 382 |
| % (95% CI) | 64.8 (57.7;71.5) | 77.2 (70.6;83.0) | 63.3 (58.4;68.0) | 69.0 (64.3;73.5) | 63.8 (59.8;67.6) |
| Fever at visit** | n=18 | 15 | 14 | 4 | 32 |
| % (95% CI) | 9.2 (5.5;14.1) | 7.9 (4.5;12.8) | 3.5 (1.9;5.8) | 1.0 (0.3;2.5) | 5.3 (3.7;7.5) |
| KeurSocé, SN | N=3 | N=6 | N=597 | N=594 | N=600 |
| Fever in the last 24 hours* | n=0 | 3 | 37 | 15 | 37 |
| % (95% CI) | 0 (0.0;70.8) | 50.0 (11.8;88.2) | 6.2 (4.4;8.4) | 2.5 (1.4;4.1) | 6.2 (4.4;8.4) |
| Fever at visit** | n=1 | 2 | 42 | 62 | 43 |
| % (95% CI) | 0 (0.0;70.8) | 50.0 (11.8;88.2) | 6.2 (4.4;8.4) | 2.5 (1.4;4.1) | 6.2 (4.4;8.4) |
| Study site | PF infected | PF not infected | Total |
|------------|-------------|----------------|-------|
|            | Survey 1    | Survey 2       | Survey 1 | Survey 2 | Survey 1 | Survey 2 |
| Niakhar, SN| % (95% CI)  |                |         |         |         |         |
|            | N=9         | N=3            | N=589   | N=598   | N=598   | N=601   |
| Fever in the last 24 hours* | 33.3 (0.8;90.6) | 7.0 (5.1;9.4) | 10.4 (8.1;13.2) | 7.2 (5.2;9.5) | 10.7 (8.3;13.4) |
| Fever at visit** | 44.4 (13.7;78.8) | 22.2 (18.9;25.8) | 23.6 (20.2;27.2) | 22.6 (19.3;26.1) | 23.6 (20.3;27.2) |
| Korogwe, TZ | % (95% CI)  |                |         |         |         |         |
|            | N=63        | N=18           | N=538   | N=582   | N=601   | N=600   |
| Fever in the last 24 hours* | 57.1 (44.0;69.5) | 16.2 (13.2;19.6) | 7.6 (5.5;10.0) | 20.5 (17.3;23.9) | 8.3 (6.2;10.8) |
| Fever at visit** | 38.1 (26.1;51.2) | 2.8 (1.6;4.6) | 0.9 (0.3;2.0) | 6.5 (4.7;8.8) | 1.0 (0.4;2.2) |
| Overall    | % (95% CI)  |                |         |         |         |         |
|            | N=1,187     | N=1,232        | N=3,021 | N=2,967 | N=4,208 | N=4,199 |
| Fever in the last 24 hours* | 35.6 (32.8;38.4) | 21.1 (19.7;22.6) | 22.5 (21.1;24.1) | 25.2 (23.9;26.5) | 25.8 (24.5;27.1) |
| Fever at visit** | 9.1 (7.5;10.9) | 3.5 (2.9;4.3) | 3.4 (2.8;4.2) | 5.1 (4.5;5.8) | 4.8 (4.1;5.5) |

*Fever in the 24h prior to the visit reported during the visit.
**Temperature recorded at visit after axillary conversion ≥ 37.5°C.

BF = Burkina Faso; GH = Ghana; KE = Kenya; SN= Senegal; TZ = Tanzania.
### Supplemental Table 9  
Number of individuals presenting with fever reported in the last 24 hours and measured at visit by study site, parasite density measured by microscopy and survey

| Study site | Parasitemia | Number of individuals presenting with fever reported in the last 24 hours* | Fever at visit** |
|------------|-------------|---------------------------------------------------------------------------|------------------|
|            |             | Survey 1  | Survey 2 | Survey 1  | Survey 2 | Survey 1  | Survey 2 | Survey 1  | Survey 2 | Survey 1  | Survey 2 | Survey 1  | Survey 2 |
| Nouna, BF  |             |           |          |           |          |           |          |           |          |           |          |           |          |
| Fever in the last 24 hours* | n | 38 | 29 | 44 | 67 | 39 | 18 | 24 | 13 | 31 | 22 |
| Fever at visit** | % | (95% CI) | 18.7 | (13.6; 24.8) | 25.9 | (18.1; 35.0) | 22.2 | (16.6; 28.7) | 21.8 | (17.3; 26.9) | 37.1 | (27.9; 47.1) | 18.0 | (11.0; 26.9) | 50.0 | (35.2; 64.8) | 33.3 | (19.1; 50.2) | 59.6 | (45.1; 73.0) | 52.4 | (36.4; 68.0) |
| Saponé, BF |             | N=305 | N=283 | N=167 | N=151 | N=81 | N=93 | N=32 | N=31 | N=19 | N=41 |
| Fever in the last 24 hours* | n | 7 | 15 | 6 | 6 | 5 | 11 | 0 | 4 | 4 | 11 |
| Fever at visit** | % | (95% CI) | 2.3 | (0.9; 4.7) | 5.3 | (3.0; 8.6) | 3.6 | (1.3; 7.7) | 4.0 | (1.5; 8.4) | 6.2 | (2.0; 13.8) | 11.8 | (6.1; 20.2) | 0 | (0.10; 9.9) | 12.9 | (3.6; 29.8) | 21.1 | (14.2; 42.9) |
| Kintampo, GH |             | N=386 | N=388 | N=123 | N=118 | N=39 | N=38 | N=16 | N=23 | N=36 | N=33 |
| Fever in the last 24 hours* | n | 83 | 142 | 49 | 43 | 19 | 23 | 8 | 14 | 26 | 25 |
| Fever at visit** | % | (95% CI) | 21.5 | (17.5; 25.9) | 36.6 | (31.8; 41.6) | 39.8 | (31.4; 49.1) | 36.4 | (27.8; 45.8) | 48.7 | (32.4; 65.2) | 60.5 | (43.4; 76.0) | 50.0 | (24.7; 75.3) | 60.9 | (38.5; 80.3) | 72.2 | (54.8; 85.6) | 75.8 | (57.7; 88.9) |
| Kombowa, KE |             | N=403 | N=410 | N=100 | N=84 | N=42 | N=44 | N=25 | N=15 | N=29 | N=46 |
| Fever in the last 24 hours* | n | 255 | 283 | 64 | 61 | 21 | 33 | 16 | 13 | 26 | 39 |
| Fever at visit** | % | (95% CI) | 63.3 | (58.4; 68.0) | 69.0 | (64.3; 73.5) | 64.0 | (53.8; 73.4) | 72.6 | (61.8; 81.8) | 50.0 | (32.4; 66.8) | 75.0 | (59.7; 86.8) | 64.0 | (42.5; 82.0) | 86.7 | (59.5; 98.3) | 89.7 | (72.6; 97.8) | 84.8 | (71.1; 93.7) |
| Kombowa, KE |             | N=403 | N=410 | N=100 | N=84 | N=42 | N=44 | N=25 | N=15 | N=29 | N=46 |
| Fever in the last 24 hours* | n | 14 | 4 | 2 | 1 | 4 | 3 | 2 | 2 | 10 | 9 |
| Fever at visit** | % | (95% CI) | 3.5 | (1.9; 5.8) | 1.0 | (0.3; 2.5) | 2.0 | (0.2; 7.0) | 1.2 | (0.0; 6.5) | 9.5 | (2.7; 22.6) | 6.8 | (1.4; 18.7) | 8.0 | (1.0; 26.0) | 13.3 | (1.7; 40.5) | 34.5 | (17.9; 54.3) | 19.6 | (9.4; 33.9) |
| Study site         | Parasitemia | Negative | Low     | Medium | High   | Very high |
|-------------------|-------------|----------|---------|--------|--------|-----------|
|                   |             | Survey 1 | Survey 2 | Survey 1 | Survey 2 | Survey 1 | Survey 2 | Survey 1 | Survey 2 | Survey 1 | Survey 2 |
| KeurSocé, SN      | N=597       | N=594    | N=2     | N=2    | -      | N=3      | -        | -        | N=1      | -        | N=1      |
| Fever in the last 24 hours* | n=37       | 15       | 0       | 0      | -      | 2        | 0        | -        | 1        | -        | 1        |
|                   | % (95% CI)  | 6.2 (4.4;8.4) | 2.5 (1.4;4.1) | 0 (0.0;8.42) | 0 (0.0;8.42) | - (9.9;99.2) | 0 (0.0;97.5) | - (2.5;100) | 100 (2.5;100) | - (2.5;100) | 100 (2.5;100) |
| Fever at visit**  | n=42       | 62       | 0       | 1      | -      | 1        | 1        | -        | 1        | -        | 1        |
|                   | % (95% CI)  | 7.0 (5.1;9.4) | 10.4 (8.1;13.2) | 50.0 (0.0;84.2) | 0 (0.0;70.8) | - (0.0;70.8) | 100 (2.5;100) | - (2.5;100) | 100 (2.5;100) | - (2.5;100) | 100 (2.5;100) |
| Niakhar, SN       | N=589       | N=598    | N=6     | N=1    | N=1   | -        | N=1      | N=1      | N=1      | N=1      |
| Fever in the last 24 hours* | n=131      | 141      | 2       | 0      | 0      | -        | 1        | 0        | 1        | 1        |
|                   | % (95% CI)  | 22.2 (18.9;25.8) | 23.6 (20.2;27.2) | 33.3 (4.7;77.1) | 0 (0.0;97.5) | 0 (0.0;97.5) | - (2.5;100) | 0 (0.0;97.5) | 100 (2.5;100) | 100 (2.5;100) | 100 (2.5;100) |
| Fever at visit**  | n=14       | 10       | 0       | 0      | -      | 0        | 0        | -        | 0        | 0        | 1        |
|                   | % (95% CI)  | 2.4 (1.3;4.0) | 1.7 (0.3;3.1) | 0 (0.0;45.9) | 0 (0.0;97.5) | - (0.0;97.5) | 100 (2.5;100) | 100 (2.5;100) | 100 (2.5;100) | 100 (2.5;100) | 100 (2.5;100) |
| Korogwe, TZ       | N=538       | N=582    | N=24    | N=10   | N=18  | N=2      | N=3      | N=2      | N=18     | N=4      |
| Fever in the last 24 hours* | n=87       | 44       | 9       | 3      | 10    | 1        | 2        | 1        | 1        | 15       |
|                   | % (95% CI)  | 16.2 (13.2;19.6) | 7.8 (5.5;10.0) | 37.5 (18.8;59.4) | 30.0 (6.7;65.2) | 56.6 (30.8;78.5) | 50.0 (1.3;98.7) | 66.7 (9.4;99.2) | 50.0 (1.3;98.7) | 83.3 (58.6;96.4) | 25.0 (6.6;80.6) |
| Fever at visit**  | n=15       | 5        | 3       | 1      | 6     | 0        | 1        | 0        | 14       |
|                   | % (95% CI)  | 2.8 (1.6;4.6) | 0.9 (0.3;2.0) | 12.5 (2.7;32.4) | 10.0 (0.3;44.5) | 33.3 (13.3;59.0) | 0 (0.0;84.2) | 0 (0.0;84.2) | 0 (52.4;93.6) | 0 (0.0;60.2) |
| Overall           | N=3,021     | N=2,967  | N=620   | N=673  | N=286 | N=290    | N=126    | N=111    | N=155    | N=168    |
| Fever in the last 24 hours* | n=638      | 669      | 174     | 180    | 94    | 88       | 51       | 45       | 103      | 100      |
|                   | % (95% CI)  | 21.1 (19.7;22.6) | 22.5 (21.1;24.1) | 28.6 (24.3;31.8) | 26.7 (23.4;30.3) | 32.9 (27.5;38.6) | 31.4 (26.0;37.2) | 40.5 (31.8;49.6) | 40.5 (31.3;50.3) | 66.5 (58.4;73.8) | 59.5 (51.7;67.0) |
| Fever at visit**  | n=107      | 102      | 23      | 22     | 17    | 16       | 14       | 13       | 13       | 54       |
|                   | % (95% CI)  | 3.5 (2.9;4.3) | 3.4 (2.8;4.2) | 3.7 (2.4;5.5) | 3.3 (2.1;4.9) | 5.9 (3.5;9.3) | 5.7 (3.3;9.1) | 11.1 (6.2;17.9) | 11.7 (6.4;19.2) | 34.8 (27.4;42.9) | 28.0 (21.3;35.4) |

Low = <2,500 parasites/μL; Medium = 2,500 – 9,999 parasites/μL; High 10,000 – 19,999 parasites/μL; Very high ≥ 20,000 parasites/μL.
N= total number of individuals overall or per site.
n= number of individuals in a given category.
95%CI = Exact 95% confidence limits.
*Fever in the 24h prior to the visit reported during the visit.
**Temperature recorded at visit after axillary conversion ≥ 37.5°C.
BF = Burkina Faso; GH = Ghana; KE = Kenya; SN = Senegal; TZ = Tanzania.
Supplemental Table 10  Number of individuals having sought treatment for malaria or fever in the past 14 days and individuals hospitalized for malaria in the last 3 months by study site, *P. falciparum* infection status and survey

| Study site       | *Pf* infected | *Pf not infected | Total |
|------------------|---------------|------------------|-------|
|                  | Survey 1      | Survey 2         | Survey 1 | Survey 2 | Survey 1 | Survey 2 |
| Nouna, BF        |               |                  |        |        |        |        |
| Seeking malaria or fever treatment in past 14 days | n=403         | n=488            | n=203  | n=112  | n=606  | n=600  |
|                   | % (95% CI)    |                  |        |        |        |        |
| Malaria hospitalization in the last 3 months | n=7           | n=12            | n=5    | n=3    | n=12   | n=15   |
|                   | % (95% CI)    |                  |        |        |        |        |
| Saponé, BF       | n=299         | n=316            | n=305  | n=283  | n=604  | n=599  |
|                   | % (95% CI)    |                  |        |        |        |        |
| Kintampo, GH     | n=214         | n=212            | n=386  | n=388  | n=600  | n=600  |
|                   | % (95% CI)    |                  |        |        |        |        |
| Kombewa, KE      | n=196         | n=189            | n=403  | n=410  | n=599  | n=599  |
|                   | % (95% CI)    |                  |        |        |        |        |
| KeurSocé, SN     | n=3           | n=6              | n=597  | n=594  | n=600  | n=600  |
|                   | % (95% CI)    |                  |        |        |        |        |
|                  |               |                  |        |        |        |        |
| Study site | Survey 1 | Survey 2 | Survey 1 | Survey 2 | Survey 1 | Survey 2 |
|------------|----------|----------|----------|----------|----------|----------|
| Niakhar, SN |          |          |          |          |          |          |
| Seeking malaria or fever treatment in past 14 days | n | 0 | 0 | 6 | 0 | 6 | 0 |
| % (95% CI) | 0 (0.0;33.6) | 0 (0.0;70.8) | 1.0 (0.4;2.2) | 0 (0.0;0.6) | 1.0 (0.4;2.2) | 0 (0.0;0.6) |
| Malaria hospitalization in the last 3 months | n | 0 | 0 | 0 | 0 | 0 | 0 |
| % (95% CI) | 0 (0.0;33.6) | 0 (0.0;70.8) | 0 (0.0;0.6) | 0 (0.0;0.6) | 0 (0.0;0.6) | 0 (0.0;0.6) |
| Korogwe, TZ | N=63 | N=18 | N=538 | N=582 | N=601 | N=600 |
| Seeking malaria or fever treatment in past 14 days | n | 20 | 4 | 79 | 25 | 99 | 29 |
| % (95% CI) | 31.7 (20.6;44.7) | 22.2 (6.4;47.6) | 14.7 (11.8;18.0) | 4.3 (2.8;6.3) | 16.5 (13.6;19.7) | 4.8 (3.3;6.9) |
| Malaria hospitalization in the last 3 months | n | 4 | 0 | 12 | 6 | 16 | 6 |
| % (95% CI) | 6.3 (1.8;15.5) | 0 (0.0;18.5) | 2.2 (1.2;3.9) | 1.0 (0.4;2.2) | 2.7 (1.5;4.3) | 1.0 (0.4;2.2) |
| Overall | N=1,187 | N=1,232 | N=3,021 | N=2,967 | N=4,208 | N=4,199 |
| Seeking malaria or fever treatment in past 14 days | n | 238 | 247 | 424 | 290 | 662 | 537 |
| % (95% CI) | 20.1 (17.8;22.4) | 20.0 (17.8;22.4) | 14.0 (12.8;15.3) | 9.8 (8.7;10.9) | 15.7 (14.6;16.9) | 12.8 (11.8;13.8) |
| Malaria hospitalization in the last 3 months | n | 33 | 42 | 76 | 77 | 109 | 119 |
| % (95% CI) | 2.8 (1.9;3.9) | 3.4 (2.5;4.6) | 2.5 (2.0;3.1) | 2.6 (2.1;3.2) | 2.6 (2.1;3.1) | 2.8 (2.4;3.4) |

*Pf* infected = Individuals infected with *P. falciparum* parasitemia measured by microscopy.

*Pf* not infected = Individuals not infected with *P. falciparum* parasitemia measured by microscopy.

N = total number of individuals overall or per site.

n = number of individuals in a given category.

95% CI = Exact 95% confidence limits.

BF = Burkina Faso; GH = Ghana; KE = Kenya; SN = Senegal; TZ = Tanzania.
### Supplemental Table 11  
**Risk factors of being infected with *P. falciparum* (as assessed by microscopy) derived from the fitted logistic regression model with study site as cluster (Survey 1)**

| Characteristics                                      | Category                                    | Reference category | Odds ratio (OR) | 95% CI of OR |
|------------------------------------------------------|---------------------------------------------|--------------------|-----------------|--------------|
| Age (in years)*                                      | Continuous                                 |                    | 1.141           | [1.048;1.242]|
| Antimalarial drugs consumed in the last 14 days       | Yes                                         | No                 | 0.614           | [0.376;1.002]|
| Antimalarial or any other medication within 14 days   | Yes                                         | No                 | 1.033           | [0.772;1.383]|
| Main house construction material: Nets                | Nets present on all windows                 | Nets not present   | 0.729           | [0.631;0.842]|
| Main house construction material: Nets                | Nets present on some windows                | Nets not present   | 1.003           | [0.688;1.463]|
| Main house construction material: Nets                | Other                                       | Nets not present   | 1.130           | [1.019;1.252]|
| Main house construction material: Roof                | Iron sheet                                  | Grass/Palm         | 0.888           | [0.753;1.046]|
| Main house construction material: Roof                | Tiles                                       | Grass/Palm         | 1.090           | [0.786;1.513]|
| Main house construction material: Walls               | Clay                                        | Grass/Palm         | 1.824           | [1.593;2.088]|
| Main house construction material: Walls               | Other                                       | Grass/Palm         | 1.175           | [1.063;1.300]|
| Main house construction material: Windows/eaves       | Closed                                      | Open               | 0.984           | [0.845;1.144]|
| Main house construction material: Windows/eaves       | No Windows                                  | Open               | 0.959           | [0.810;1.137]|
| Main house construction material: Windows/eaves       | Partially open                              | Open               | 0.987           | [0.794;1.228]|
| Main house construction material: Windows/eaves       | Other                                       | Open               | 1.374           | [1.234;1.530]|
| Number of holes                                       | < 5                                         | ≥5                 | 0.921           | [0.712;1.191]|
| Number of holes                                       | No bednet                                   | ≥5                 | 1.044           | [0.740;1.473]|
| Number of persons living in the same part of the house| 4-5                                         | ≤3                 | 1.168           | [1.000;1.364]|
| Number of persons living in the same part of the house| >5                                          | ≤3                 | 1.317           | [1.165;1.488]|
| Pierced/torn bednet                                   | No bednet                                   | No                 | 1.000           | [1.000;1.000]|
| Presence of electricity                               | Yes                                         | No                 | 1.000           | [1.000;1.000]|
| Use of traditional repellents over 7 days            | Yes                                         | No                 | 0.752           | [0.611;0.926]|
| Use of traditional repellents over 7 days            | Missing/No                                  |                    | 0.994           | [0.907;1.088]|

*Note: Odds ratios were adjusted by a cluster variable.  
95% CI = Exact 95% confidence limits.  
The fitted logistic regression model was only applied on significant variables produced by the backward selection.  
The variables ‘Floor’, ‘number of persons enrolled into the study’ and ‘localisation’ were not included in the model due to convergence issue.  
*Age was introduced in the model as a continuous variable. The corresponding OR is given for a 1 year increase of age.*
Supplemental Table 12  Risk factors of being infected with *P. falciparum* (as assessed by microscopy) derived from the fitted logistic regression model with study site as cluster (Survey 2)

| Characteristics                                      | Category                        | Reference category | Odds ratio (OR) | 95% CI of OR |
|------------------------------------------------------|---------------------------------|--------------------|-----------------|--------------|
| Age (in years)*                                      | Continuous                      |                    | 1.087           | [1.018;1.161]|
| Antimalarial drug consumed in the past 14 days       | Yes                             | No                 | 0.457           | [0.324;0.645]|
| Impregnated Bednet                                   | No Bednet                       | No                 | 1.000           | [1.000;1.000]|
|                                                      | Yes                             | No                 | 1.093           | [0.887;1.345]|
| Localization                                         | Semi- Rural Area                | Rural area         | 0.741           | [0.419;1.308]|
|                                                      | Urban area                      | Rural area         | 0.185           | [0.109;0.314]|
| Main house construction material: Nets                | Nets present on all windows     | Nets not present   | 0.787           | [0.660;0.939]|
|                                                      | Nets present on some windows    | Nets not present   | 0.947           | [0.901;0.995]|
|                                                      | Other                           | Nets not present   | 1.007           | [0.886;1.143]|
| Main house construction material: Roof               | Iron sheet                      | Grass/Palm         | 0.953           | [0.818;1.111]|
|                                                      | Tiles                           | Grass/Palm         | 0.995           | [0.703;1.407]|
|                                                      | Clay                            | Grass/Palm         | 1.011           | [0.913;1.120]|
|                                                      | Other                           | Grass/Palm         | 1.236           | [0.786;1.943]|
| Main house construction material: Walls              | Brick                           | Mud                | 0.922           | [0.783;1.085]|
|                                                      | Cement/Plaster                  | Mud                | 0.866           | [0.757;0.992]|
|                                                      | Cement /Paint                   | Mud                | 0.990           | [0.853;1.148]|
|                                                      | Clay                            | Mud                | 1.197           | [1.120;1.278]|
|                                                      | Other                           | Mud                | 0.658           | [0.415;1.044]|
| Main source of drinking water                        | Closed water source †           | Open water source † | 0.921           | [0.835;1.015]|
| Malaria or fever treatment sought for in the past 14 days | Yes                             | No                 | 1.227           | [1.047;1.439]|
| New net (less than 1 year)                           | No Bednet                       | No                 | 1.095           | [0.901;1.330]|
|                                                      | Yes                             | No                 | 0.929           | [0.839;1.029]|
| Pierced/torn bednet                                  | No Bednet                       | No                 | 1.000           | [1.000;1.000]|
|                                                      | Yes                             | No                 | 0.912           | [0.798;1.042]|
| Presence of electricity                              | Yes                             | No                 | 0.885           | [0.803;0.976]|
| Use of traditional repellents over 7 days           | Yes                             | Missing/No         | 0.910           | [0.747;1.109]|
| No use of mosquito coils–insecticide sprays–commercial or traditional repellents over 7 days | Yes                             | Missing/No         | 0.879           | [0.782;0.988]|

Note: Odds ratios (ORs) were adjusted by a cluster variable. 95% CI = Exact 95% confidence limits.

The fitted logistic regression model was only applied on significant variables produced by the backward selection. The variables ‘Number of persons living in the same part of the house’, ‘Type of location’, ‘Floor’, ‘Windows/eaves’ and ‘Use of Insecticide sprays over 7 days’ were not included in the model due to convergence issue.

*Age was introduced in the model as a continuous variable. The corresponding OR is given for a 1 year increase of age.

†Closed water source (piped water, tube well, dug well, protected well).

††Open water source (unprotected well, spring water, rainwater, tanker truck, surface water).
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