BMC Infectious Diseases

RESEARCH Open Access

Asymptomatic malaria parasitaemia and virological non-suppression among children living with HIV in a low transmission area in Accra, Ghana: a cross-sectional study

Adwoa K. A. Afrane[1*], Yakubu Alhassan[2], Linda Eva Amoah[3], Mame Yaa Nyarko[4], Adolphina Addo-Lartey[5], Elijah Paintsil[6] and Kwasi Torpey[7*]

Abstract

Background Human Immunodeficiency Virus (HIV) and malaria are two major diseases in sub-Saharan Africa. Co-infection can significantly impact the clinical outcomes of both conditions. We assessed the proportion of HIV-infected children at Korle Bu Teaching Hospital (KBTH) and Princess Marie Louise Hospital (PML) with malaria parasites. The association between asymptomatic malaria parasitaemia and virological non-suppression was also determined in these children.

Methods This cross-sectional study of 277 asymptomatic malaria in children receiving care at paediatric HIV clinics at KBTH and PML was conducted from September to November 2022. Patients who had been on antiretroviral therapy (ART) for at least six months were eligible to participate. Structured questionnaires were used to collect socio-demographic information, malaria prevention behaviors, and ART-related data using in-person interviews. Microscopy and PCR were used to screen for malaria, and GeneXpert was used to determine viral load. To examine the determinants of malaria PCR positivity and virological non-suppression, chi-square tests and logistic regression were performed.

Results The median age of the participants was 9 years (range: 6–12 years). Males comprised 158 (57%) of the study population. We detected 10 (3.6%) and 21 (7.6%) malaria cases by microscopy and PCR, respectively. Virological non-suppression (VL > 1000 copies/ml) was observed in 82 (29.6%) of the 277 participants. Among the suppressed individuals, 62 (22.4%) exhibited low-level viraemia (VL level 40-1000 copies/ml) and 133 (48%) had undetectable viral load levels. No factors were associated with the presence of malaria PCR positivity carriage. Poor adherence to ART was associated with a five-fold increase in the risk of viral load non-suppression (AOR = 4.89 [CI = 2.00-11.98], $p = 0.001$).

Conclusion The proportion of children living with HIV with asymptomatic malaria parasitaemia was low. Approximately one-third of the study population had virological non-suppression. The interaction between malaria

*Correspondence:
Adwoa K. A. Afrane
akaafrane@ug.edu.gh
Kwasi Torpey
KTorpey@ug.edu.gh

Full list of author information is available at the end of the article

parasitemia and viral replication may not be the main cause for virological non-suppression in this low transmission area.

Keywords Antiretroviral therapy, Asymptomatic malaria, Parasitaemia, Paediatric HIV, Viral load, Virological non-suppression

Background

HIV and malaria have been significant global health challenges for decades [1]. HIV emerged in the mid-20th century, and malaria has plagued humanity for centuries. Despite advancements in prevention and treatment, HIV and malaria continue to be major public health issues, particularly in sub-Saharan Africa (SSA) [1]. This region accounts for over half of all HIV infections and AIDS-related deaths worldwide [2]. According to the World Health Organization (WHO), approximately 213 million malaria cases and 380,000 deaths are reported annually in Africa [3]. Simultaneous infection by HIV and malaria may be as high as 30% in children living with HIV (CLHIV) in SSA [4, 5].

The combined impact of HIV and malaria has severe health, economic, and social consequences, particularly for children [6] These diseases result in more severe illnesses, higher mortality rates, and significant burdens on the healthcare system [7]. The deaths of parents due to these diseases can lead to orphanhood, placing additional burden on families and communities. Furthermore, illnesses and caregiving responsibilities often lead to school absences and dropouts, limiting future opportunities for the affected children [7].

HIV and malaria have a synergistic relationship [8]. HIV impairs the immune system, making individuals, particularly children, more susceptible to infections like malaria in malaria-endemic areas [8, 9] In turn, malaria can exacerbate HIV symptoms and increase the risk of HIV transmission.

Some antiretroviral therapies (ARTs) have antimalarial properties and can work synergistically with antimalarials to combat Plasmodium falciparum [10] Additionally, daily treatment with cotrimoxazole and ARTs is associated with a reduced prevalence of clinical malaria [10].

The effect of malaria on HIV has been well-documented. HIV-infected patients with malaria show increased viral loads, decreased CD4 counts and worsening HIV outcomes [11]. Malaria also elevates viral load, while HIV weakens the body's immune response to malaria and reduces the effectiveness of antimalarial treatments. In CLHIV, clinical malaria episodes are more frequent and severe than in HIV-uninfected children, particularly in those with higher immunosuppression levels [1]. Malaria infection in CLHIV promotes rapid HIV-1 replication due to upregulated pro-inflammatory cytokines, leading to increased HIV-1 RNA levels, accelerated disease progression, and a higher transmission risk [4, 7].

The goal of combination antiretroviral therapy (cART) for CLHIV is long-term virological suppression, defined as a viral load (VL) of <1000 copies/ml in low- and middle-income countries, indicating treatment adherence and reduced HIV transmission risk [12]. Factors influencing virological suppression include socio-demographic factors, HIV comorbidities, HIV clinical severity, and ART regimen.

Asymptomatic malaria is defined as "the presence of parasites in the blood of an individual in the absence of malaria-related symptoms such as fever, which is a temperature greater than 37.5°C" [13]. Asymptomatic carriers, particularly children, serve as carriers of *Plasmodium falciparum* contributing to malaria transmission [14, 15]. The 2019 Ghana Malaria Indicator Survey showed a decline in symptomatic malaria among children under five, from 27% in 2014 to 14% in 2019, attributed to interventions such as long-lasting insecticide-treated nets (LLINs), Indoor Residual Spraying (IRS), and case management with Artemisinin Combination Therapy [16]. However, malaria remains a threat in high-transmission areas, especially in CLHIV [17, 18]. To reduce malaria transmission, epidemiology data on asymptomatic malaria is essential. This information helps control local transmission and its associated factors.

The World Health Organization recommends conducting surveys to characterize baseline malaria transmission, identify carriers, and target at-risk populations like CLHIV for control measures [19]. Despite the significance of this issue, few studies have been conducted on asymptomatic malaria in Ghana. By investigating the relationship between asymptomatic malaria and virological non-suppression in CLHIV, the study contributes to filling knowledge gaps and informing public health interventions.

This study therefore aimed to evaluate the prevalence of malaria parasites among HIV-infected children at Korle Bu Teaching Hospital and Princess Marie Louise Hospital. We also sought to investigate the relationship between asymptomatic malaria parasitemia and virological non-suppression in these children. By conducting this study within healthcare facilities, we assessed a well-defined population of children already receiving regular care, ensuring a representative sample and comprehensive data collection. As the participants were already engaged in the healthcare system, implementing

interventions based on the study findings can be done more efficiently, streamlining efforts to improve health outcomes.

Methods

Study design and setting

A cross-sectional study was conducted at the Korle Bu Teaching Hospital (KBTH) and Princess Marie Louise (PML) Hospital in the city of Accra, Greater Accra region of Southern Ghana. Recruitment was conducted between September and November 2022. The Paediatric HIV clinic at KBTH has been providing comprehensive HIV/AIDS care and management services since 2004. An average of 40 patients were examined per clinic day, which was run once a week. Patients are referred from primary and secondary health facilities as well as from other departments within the hospital. Children are seen from 6 weeks of age until approximately 15 years of age, when they would have had full disclosure and are ready to be transferred to the adolescent HIV clinic at KBTH. The HIV clinic at PML sees approximately 10–20 patients every clinic day, and the clinic runs twice a week. KBTH however has a clinic once a week and sees about 20–30 patients per clinic day. Referrals to the PML Paediatric HIV clinic are mainly from the southern part of Ghana. PML is a paediatric hospital that mainly sees paediatric patients up to 18 years of age.

According to the National AIDS Control Program (NACP) guidelines, VL is obtained at least once a year per patient, and the cost is borne by the NACP. National treatment guidelines updated with WHO recommendations are used by both HIV clinics. Antiretroviral drugs that are provided at the clinics are abacavir (ABC), nevirapine (NVP), lamivudine (3TC), tenofovir (TDF), zidovudine (AZT), and ritonavir-boosted lopinavir (LPV/r), efavirenz (EFV) and dolutegravir (DTG). The most popular ART regimens used in the clinic include: LPV/r based e.g. ABC+3TC+LPV/r and DTG based e.g. TDF+3TC+DTG.

In accordance with the clinical guidelines, all CLHIV, regardless of viral load level were administered cotrimoxazole. Cotrimoxazole is a combination of trimethoprim and sulfamethoxazole and is recommended for the prevention of Pneumocystis pneumonia, toxoplasmosis, bacterial infections, and malaria [20].

Study population

The study participants were CLHIV who had been on ART for at least 6 months at the Paediatric HIV clinic at KBTH and PML.

Inclusion criteria CLHIV under 15 years of age and who had been on ART for at least six months.

Exclusion criteria CLHIV under 15 years of age who had been on ART for at least six months and were symptomatic for malaria at the time of recruitment.

Sample size determination

Cochran's sample size formula for prevalence studies was used to calculate the sample size using a confidence level of 95%, an error margin of 5% and a design effect of 1.5 due to the multicenter design. The prevalence of HIV-malaria co-infection (12.3%) used in the sample size calculation was obtained from a study by Ikilezi et al. in Ugandan children in 2013 [21]. The sample size obtained was 277 participants. A total of 140 participants recruited from the PML and 137 from the KBTH.

Sampling method and data collection tools

The 277 study participants were sampled consecutively at each site until the total number of participants required was determined. Blood samples were collected from each participant to detect the viral load and carriage of malaria parasites. A questionnaire was designed to collect information from the study participants. The three components of the questionnaire were sociodemographic characteristics of the participants, malaria preventive practices, and ART-related practices. Sociodemographic data included age, gender, residence caregivers' occupation, and level of education. Data on the malaria preventive practices of the caregiver included the use of an insecticide-treated net, mosquito repellent coil mosquito repellent cream and mosquito repellent spray. Respondents were also asked whether their child was on cotrimoxazole.

ART-related factors, including the type of drug regimen and duration of ART, were derived from the participants' medical records. A section also documented the viral load and malaria parasite density on the day of the interview. A 14-day recall by the caregiver was used to gauge adherence. Participants were asked about their recent 14-day adherence by asking them to recollect their prescribed medications. Participants were questioned about their adherence during the day, the previous three days, the previous week, and the past 14 days prior to the interview. Using the following formula, the level of drug use compliance during the previous 14 days was calculated [22]:

$$\%\,14\,day\;Adherence = \frac{(\#\;doses\;should\;have\;taken - \#\;missed\;doses)}{\#\;doses\;should\;have\;taken} * 100$$

The interpretation of adherence was as follows: good adherence equivalent to ≥95%, fair adherence equivalent to 85–94%, and poor adherence equivalent to <85%.

Data collection procedure

Data was collected over eight weeks. The principal investigator and two research assistants conducted the interviews. Using the eligibility criteria, potential participants and their caregivers were identified, and received written and verbal information about the study, and those who were willing to participate endorsed a consent form either by signing or thumbprinting. Children above the age of nine years endorsed an assent form. All the children and their caregivers received a copy of the endorsed form. The questionnaire was administered through face-to-face interviews and additional information such as current drug regimen and ART duration, was extracted from the patients' medical records. Trained research assistants thoroughly reviewed all the questionnaires to validate and ensure that they had been fully completed.

Laboratory procedures

At the laboratory, 2–3 ml of blood was taken from each participant: 1–2 ml was used to measure viral load, and ~100 µl was used to make thick and thin films on a microscope slide to detect malaria parasites by microscopy. DNA was extracted from the remaining blood using PCR to detect *Plasmodium* parasites. Thin and thick blood smears were processed for Giemsa staining and evaluated using a WHO protocol; "Giemsa staining of malaria blood films," 2016 [23]. *Plasmodium* species were identified after evaluating the thin films, and parasite density was estimated using the thick films. To detect satisfactory fields, the whole smear was first screened at low magnification (10X x 40X objective lens) [23]. *Plasmodium* species identification and parasitaemia were determined using 100X oil immersion microscopy.

Estimation of parasite density

Parasite density was determined for each malaria positive slide. The total number of white blood cells (neutrophils, eosinophils, lymphocytes and basophils) and *Plasmodium* parasites was counted in each field on the slides using a tally counter until a total of 200 white blood cells were obtained, and the corresponding parasite count was recorded. *Plasmodium* parasites were counted per 200 leukocytes and used to estimate parasite density.

$$Parasite\ density = \frac{Total\ number\ of\ Plasmodium\ parasite}{Total\ number\ of\ white\ blood\ cells} x\ 8{,}000 \mu\, l$$

Two independent, WHO trained and certified microscopists, who were blinded to the patients' clinical status and the microscopy results, examined all coded smears for parasites.

PCR identification of Plasmodium falciparum isolates

The nested PCR method for Plasmodium species identification based on amplification of the small subunit ribosomal RNA (18SrRNA) gene was used [24]. The initial PCR involved the use of genus-specific rPLU5 and rPLU6 primers to amplify the gDNA fragment or sequence conserved in four human Plasmodium species. The primary amplified product was used as the template for secondary PCR using species-specific oligonucleotide primer pairs for the Plasmodium species. The primer pair used for *P. falciparum* was rFAL1 and rFAL2. Genomic DNA from *Plasmodium falciparum* 3D7 strain was used as a positive control and double-distilled water was used as a negative control in all amplification reactions [25, 26].

The gene amplifications were carried out in a 15 µL reaction volume composed of 200 nM dNTP, 2 mM $MgCl_2$, 200 nM of each primer, and 0.5 U of One Taq DNA polymerase (New England BioLAB, UK). Four microliters (>20 ng) of gDNA was used as a template for the primary reaction and 1 µL of the product was used as the template for the secondary reaction. The amplification cycling conditions were as follows: initial denaturation at 94 °C for 5 min, followed by 30 cycles at 94 °C for 30 s, 58 °C annealing temperature for 1 min, and 72 °C for 1 min, with a final extension at 72 °C for 5 min. All conditions remained the same for the primary and secondary amplifications, except for the secondary reaction annealing temperature, which was 55 °C [25, 26].

Malaria parasite detection was performed at the Immunology Department of the Noguchi Memorial Institute for Medical Research, University of Ghana.

Determination of viral load

The GeneXpert platform was used to estimate viral load. Plasma (1.2 ml) was transferred into an Xpert HIV-1 Viral Load cartridge using a calibrated pipette and loaded into the machine. Test results were observed and recorded after 90 min. The limit of detection was 40 cp/ml and the clinical cut-off was 1000 cp/ml. Viral load measurements were performed at the chest clinic laboratory at KBTH, which participates in an external quality assurance testing program at the South African Public Health Reference Laboratory.

Management of patients based on laboratory results

Asymptomatic participants who tested positive for malaria were referred to a clinician for management. Oral Artemether Lumefantrine medications were provided to the facility for dispensing to such patients. Clinicians were also aware of the participants' viral load levels. All patients were managed according to a standard protocol.

Statistical analysis

Standard descriptive analysis was performed using frequencies and percentages for categorical variables, means and standard deviations for normally distributed continuous variables, and medians and interquartile ranges for non-normally distributed continuous variables. Pearson's chi-squared test was used to determine the association between the carriage of malaria parasites (PCR positivity) and associated factors (socio-demographic characteristics, malaria preventive practices and ART). Pearson's chi-square test was also used to assess the association between carriage of malaria parasites and virological suppression (target not detected). A t-test was used to test the equality of means between children with malaria PCR positivity and those with malaria PCR negativity.

Finally, a binary logistic regression model was used to assess variables independently associated with the presence of Malaria PCR among CLHIV. The multivariate model of factors associated with viral load non-suppression included the study site, age of the child, adherence to medication, ART duration, and ART regimen. In the final logistic regression model, viral load suppression was included as a major exposure variable, while age and sex were included because of their relevance in the prevalence of health outcomes among children. Cotrimoxazole use and study site were included in the final adjusted model because their significance level was below 0.1000 in the unadjusted binary logistic regression model. Independent variables that showed a statistically significant association with the outcome variable in univariate analysis were subjected to multivariate logistic regression. The crude odds ratio (COR), adjusted odds ratio (AOR), the corresponding 95% confidence interval (CI) and p values are presented. In the final model, statistical significance was observed with a p-values below 0.05.

Results

Socio-demographic, malaria preventive practices and ART characteristics of participants

A total of 277 participants (140 from PML and 137 from KBTH), were enrolled. More than half of the participants were males 158 (57%). The age group 10–14 years had the highest number of participants 123 (41.5%). The mean age of the participants was 9.0±3.4 years. The primary caregivers of the participants were biological mothers, 150 (54.2%). Approximately 250 caregivers (90%) were employed (Table 1).

Overall, 27 (9.7%) participants did not practice any malaria-preventive method. Of the 277 participants, 217 (78.3%) were given cotrimoxazole. The majority of the participants 224 (80.9%) had good adherence to ART. About 250 (48%) participants had been on ART between 12 and 59 months. The ART regimens used by participants were EFV-based 104 (37.5%), followed by DTG-based 90 (32.5%), LPV/r-based 73 (26.4%) and NVP-based 10 (3.6%). Children sampled from the KBTH clinic significantly differed from children sampled from the PML clinic by demographics including age group ($p=0.005$), relationship of child to respondent ($p=0.036$), being given cotrimoxazole ($p=0.025$), duration on ART ($p=0.027$) and ART regimen ($p=0.003$).

Proportion of CLHIV with malaria parasites

Of 277 participants, microscopy detected 10 (3.6%, 95% CI: 1.9–6.6%), while PCR detected 21 cases of malaria (7.6%, 95% CI: 5.0-11.4%). The results are presented based on the PCR results. PCR positivity was higher among children from the PML facility 16/140 (11.4%) than among those from the KBTH facility 5/137 (3.6%).

Viral load levels among CLHIV

Of 277 participants, 82 (29.6%) had virological non-suppression (VL>1000 copies/ml). Of the participants who were virologically suppressed, 62 (22.4%) had low-level viraemia (VL level 40-1000 copies/ml), while 133 (48%) had undetectable viral load levels. (Fig. 1).

Malaria PCR positivity by ART regimen among participants

Of the 10 patients who were on a nevirapine-based regimen, none tested positive for malaria. The proportion of participants who had malaria parasites among participants on a DTG-based regimen was comparable to that among participants on LPV/r-based-regimen (8.9% vs. 9.6%) (Fig. 2).

There was no significant association between malaria PCR positivity and participant characteristics (Table 2).

Association between characteristics of participants and malaria PCR positivity

The mean age of the malaria PCR negative sample was 9.0 years (±3.3 years) which was not statistically different from mean age of 8.8 years (±3.9 years) among the PCR positive samples ($p=0.780$). There was no significant association between malaria PCR positivity and the characteristics of the participants (Table 2).

Association between characteristics of participants and virological non-suppression

The age of the child, adherence level and ART regimen were associated with virological non-suppression. The mean age of the children that were virologically suppressed was 9.3 years (±3.2 years) which was significantly higher than the mean age of 8.1 years (±3.7 years) among the non-virologically suppressed ($p=0.004$). More than half of the children younger than 5 years were virologically non-suppressed (56.4%), compared to 28.7% non-suppression among children aged 5–9 years and 22.0% non-suppression among children aged 10–14 years.

Table 1 Socio-demographic, malaria preventive practices, and ART characteristics of study participants

	Total	KBTH	PML	*P*-value
	N = 277	*N* = 137	*N* = 140	
Characteristics	*n* (%)	*n* (%)	*n* (%)	*n* (%)
Sex				0.840
Male	158 (57)	79 (57.7)	79 (56.4)	
Female	119 (43)	58 (42.3)	61 (43.6)	
Age of child				0.005
<5 years	39 (14.1)	11 (8.0)	28 (20.0)	
5–9 years	115 (41.5)	55 (40.1)	60 (42.9)	
10–14 years	123 (44.4)	71 (51.8)	52 (37.1)	
Relation of respondent to child				0.036
Mother	150 (54.2)	69 (50.4)	81 (57.9)	
Father	43 (15.5)	17 (12.4)	26 (18.6)	
Other relatives	84 (30.3)	51 (37.2)	33 (23.6)	
Employment status of caregiver				0.580
Unemployed	27 (9.7)	12 (8.8)	15 (10.7)	
Employed	250 (90.3)	125 (91.2)	125 (89.3)	
Using any malaria preventive method				0.890
Yes	250 (90.3)	124 (90.5)	126 (90.0)	
No	27 (9.7)	13 (9.5)	14 (10.0)	
Cotrimoxazole				0.025
Yes	217 (78.3)	115 (83.9)	102 (72.9)	
No	60 (21.7)	22 (16.1)	38 (27.1)	
Adherence				0.330
Good	224 (80.9)	115 (83.9)	109 (77.9)	
Fair	33 (11.9)	15 (10.9)	18 (12.9)	
Poor	20 (7.2)	7 (5.1)	13 (9.3)	
Duration on ART				0.027
<12 months	14 (5.1)	5 (3.6)	9 (6.4)	
12–59 months	134 (48.4)	57 (41.6)	77 (55.0)	
60–119 months	95 (34.3)	52 (38.0)	43 (30.7)	
120 + months	34 (12.3)	23 (16.8)	11 (7.9)	
ART regimen code				0.003
DTG based	90 (32.5)	47 (34.3)	43 (30.7)	
LPV/r based	73 (26.4)	24 (17.5)	49 (35.0)	
EFV based	104 (37.5)	58 (42.3)	46 (32.9)	
NVP based	10 (3.6)	8 (5.8)	2 (1.4)	

Virological non-suppression was highest among children with poor medication adherence (60.0%) compared to 25.9% non-suppression rate among children with good adherence and 36.4% non-suppression rate among children with a fair adherence level. Additionally, virological non-suppression rates were higher among children who had been on ART for less than 12 months (57.1%) compared to 35.1% among children aged 12–59 months on ART, 20.0% among children with 60–119 months on ART, and 23.5% among children with 120 or more months on ART. Virological non-suppression was highest among children on LPV/r -based regimen with 45.2% prevalence, followed by children on EFV based regimen (26.0%) then children on the DTG- based regimen (23.3%) and was lowest among children on the NVP-based regimen (10.0%) (Table 3).

Factors associated with malaria PCR positivity by logistic regression

None of the factors was significantly associated with *P. falciparum* PCR positivity (Table 4).

Factors associated with virological non-suppression by binary logistic regression

PCR positivity was not significantly associated with viral load non-suppression from the multivariable logistic regression model (AOR = 1.46 [CI = 0.49–4.33], $p = 0.495$). The odds of non-suppression were significantly lower among the older age groups relative to those aged < 5 years, with 57% lower odds among those aged 5–9 years (AOR = 0.43 [CI = 0.19–0.97], $p = 0.042$) and 63% lower among those aged 10–14 years (AOR = 0.37 [CI = 0.14–0.99], $p = 0.048$). Relative to those with good adherence

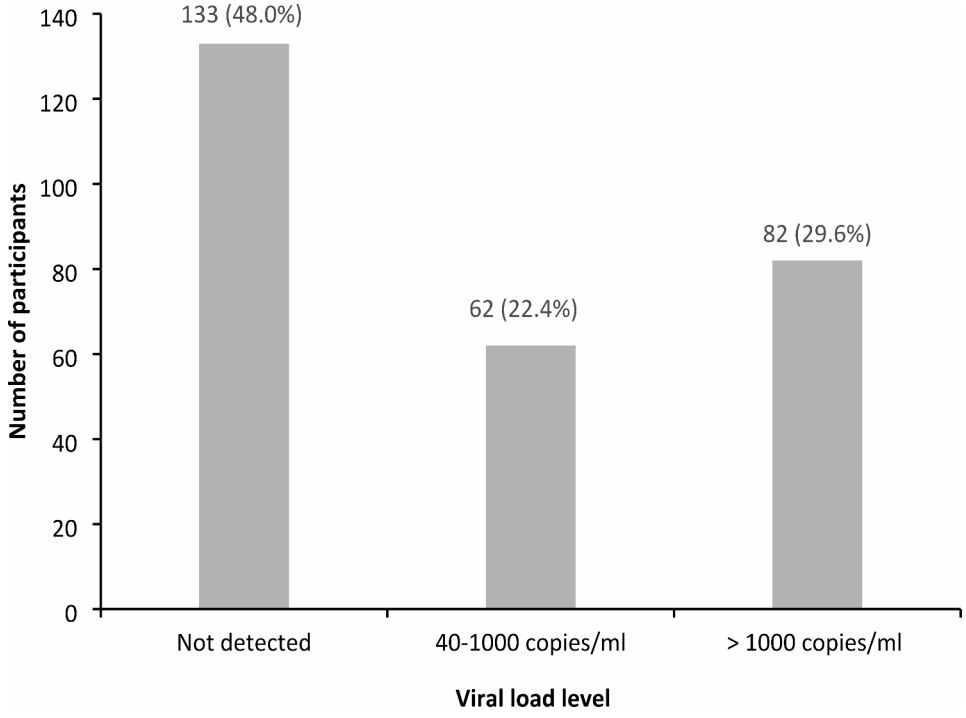

Fig. 1 The viral load levels of study participants at KBTH and PML

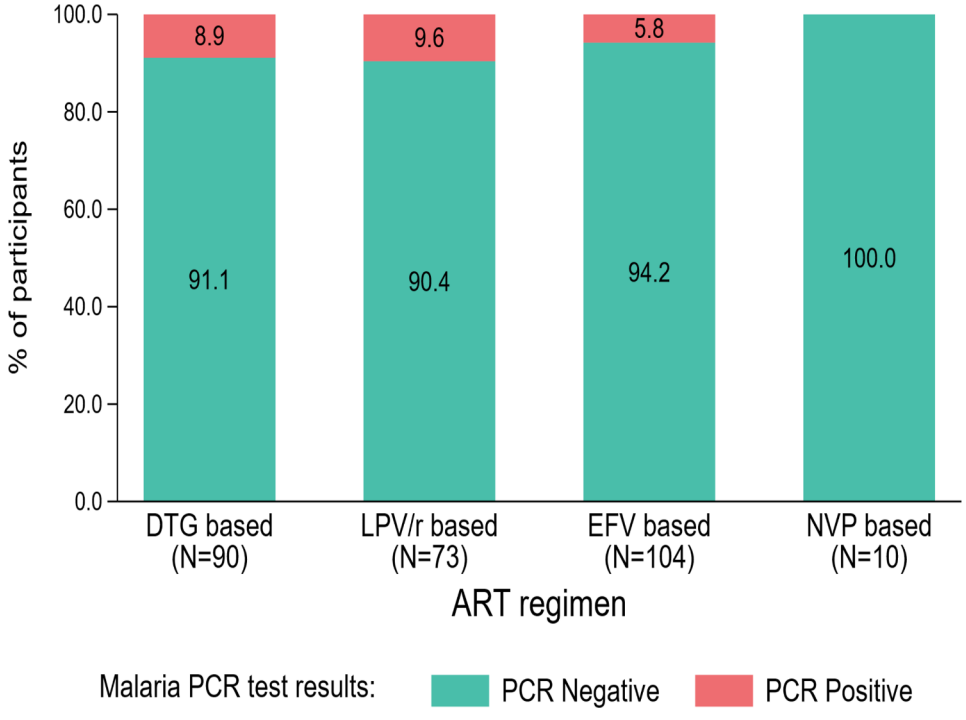

Fig. 2 Malaria PCR positivity by ART regimen among study participants at KBTH and PML

to medication, the odds of viral load non-suppression were approximately five times higher among those with poor adherence (AOR=4.89 [CI=2.00-11.98], p=0.001) (Table 5).

Discussion

This study determined the proportion of asymptomatic children living with HIV (CLHIV) with malaria parasites and identified the associated factors in paediatric HIV clinics at two hospitals in Accra, Ghana. The proportion

Table 2 Association between characteristics of participants and *P. Falciparum* PCR positivity

	***P.* falciparum PCR test**		
	PCR Negative	**PCR Positive**	***P* value**
Characteristics	***n* (%)**	***n* (%)**	
Overall	**256 (92.4)**	**21 (7.6)**	
Study site			**0.014**
KBTH	132 (96.4)	5 (3.6)	
PML	124 (88.6)	16 (11.4)	
Sex			0.350
Male	144 (91.1)	14 (8.9)	
Female	112 (94.1)	7(5.9)	
Age group of children			0.790
<5 years	35 (89.7)	4 (10.3)	
5–9 years	107 (93.0)	8 (7.0)	
10–14 years	114 (92.7)	9 (7.3)	
Relation of respondent to child			0.25
Mother	136 (90.7)	14 (9.3)	
Father	39 (90.7)	4 (9.3)	
Other relatives	81 (96.4)	3 (3.6)	
Caregiver's highest education			0.830
None	17 (89.5)	2(10.5)	
Primary	41 (93.2)	3 (6.8)	
JHS/JSS/Middle school	142 (93.4)	10(6.6)	
SHS or Higher	56 (90.3)	6 (9.7)	
Employment status			0.970
Unemployed	25 (92.6)	2 (7.4)	
Employed	231 (92.4)	19 (7.6)	
Cotrimoxazole			0.057
Yes	204 (94.0)	13 (6.0)	
No	52 (86.7)	8 (13.3)	
Using any malaria preventive method			0.120
No	27 (100.0)	0 (0.0)	
Yes	229 (91.6)	21 (8.4)	
Adherence			0.420
Good	208 (92.9)	16 (7.1)	
Fair	31 (93.9)	2 (6.1)	
Poor	17 (85.0)	3 (15.0)	
Duration on ART			0.970
<12 months	13 (92.9)	1 (7.1)	
12–59 months	124 (92.5)	10 (7.5)	
60–119 months	87 (91.6)	8 (8.4)	
120 + months	32 (94.1)	2 (5.9)	
ART regimen code			0.580
DTG based	82 (91.1)	8 (8.9)	
LPV/r based	66 (90.4)	7 (9.6)	
EFV based	98 (94.2)	6 (5.8)	
NVP based	10 (100.0)	0 (0.0)	

of CLHIV with malaria was low, as detected using both microscopy and PCR. No factors were significantly associated with the presence of malaria parasites. Over one-third of the participants were virologically non-suppressed. The odds of viral load non-suppression were approximately five times higher among those with poor adherence to antiretroviral therapy than among those with good adherence.

HIV increases the incidence of malaria in individuals living in malaria endemic areas, particularly in those with established immunosuppression [1]. According to data from the 2022 Ghana Demographic and Health Survey (GDHS), the prevalence of symptomatic malaria in urban

Table 3 Association between study participant characteristics and virological non-suppression

	Viral load level		
	Suppression	Non-suppression	*P* value
Characteristics	***n* (%)**	***n* (%)**	
Overall	195 (70.4)	82 (29.6)	
Study site			**0.005**
KBTH	107 (78.1)	30 (21.9)	
PML	88 (62.9)	52 (37.1)	
Sex			0.460
Male	114 (72.2)	44 (27.8)	
Female	81 (68.1)	38 (31.9)	
Age group of children			**< 0.001**
<5 years	17 (43.6)	22 (56.4)	
5–9 years	82 (71.3)	33 (28.7)	
10–14 years	96 (78.0)	27 (22.0)	
Relation of respondent to child			0.910
Mother	104 (69.3)	46 (30.7)	
Father	31 (72.1)	12 (27.9)	
Other relatives	60 (71.4)	24 (28.6)	
Caregiver's highest education			0.440
None	13 (68.4)	6 (31.6)	
Primary	28 (63.6)	16 (36.4)	
JHS/JSS/Middle school	113 (74.3)	39 (25.7)	
SHS or Higher	41 (66.1)	21 (33.9)	
Employment status			0.660
Unemployed	20 (74.1)	7 (25.9)	
Employed	175 (70.0)	75 (30.0)	
Cotrimoxazole			0.810
Yes	152 (70.0)	65 (30.0)	
No	43 (71.7)	17 (28.3)	
Using any malaria preventive method			0.380
No	21 (77.8)	6 (22.2)	
Yes	174 (69.6)	76 (30.4)	
Adherence			**0.004**
Good	166 (74.1)	58 (25.9)	
Fair	21 (63.6)	12 (36.4)	
Poor	8 (40.0)	12 (60.0)	
Duration on ART			**0.008**
<12 months	6 (42.9)	8 (57.1)	
12–59 months	87 (64.9)	47 (35.1)	
60–119 months	76 (80.0)	19 (20.0)	
120 + months	26 (76.5)	8 (23.5)	
ART regimen code			**0.005**
DTG based	69 (76.7)	21 (23.3)	
LPV/r based	40 (54.8)	33 (45.2)	
EFV based	77 (74.0)	27 (26.0)	
NVP based	9 (90.0)	1 (10.0)	

areas of Ghana is approximately 4.3% [27] .This rate is significantly lower compared to rural areas, highlighting a disparity in malaria prevalence between different geographic regions within the country. Despite this lower prevalence in urban areas, the presence of asymptomatic malaria cases remains considerable and poses a public health challenge by contributing to ongoing malaria transmission.

There are few and limited studies on asymptomatic malaria in HIV-infected people, especially in children in Ghana. CLHIV are at a higher risk of developing malaria parasitaemia than HIV-negative children. The higher risk of malaria parasitaemia among CLHIV could be due to

Table 4 Binary logistic regression model of factors associated with malaria PCR positivity among study participants

PCR positivity	Unadjusted model		Adjusted model	
Characteristics	**COR [95% CI]**	***P* value**	**AOR [95% CI]**	***P* value**
Sex				
Male	1.00 [reference]		1.00 [reference]	
Female	0.64 [0.25, 1.65]	0.358	0.56 [0.20, 1.55]	0.263
Age of child				
< 5 years	1.45 [0.42, 5.00]	0.559	0.88 [0.22, 3.59]	0.864
5–9 years	0.95 [0.35, 2.55]	0.914	0.97 [0.34, 2.75]	0.951
10–14 years	1.00 [reference]		1.00 [reference]	
Cotrimoxazole				
Yes	1.00 [reference]		1.00 [reference]	
No	2.41 [0.95, 6.14]	0.064	2.30 [0.88, 6.05]	0.091
Study site				
KBTH	1.00 [reference]		1.00 [reference]	
PML	3.41 [1.21, 9.59]	**0.020**	2.85 [0.92, 8.86]	0.070
Suppression (Copies/ml)				
Viral load suppression (≤ 1,000)	1.00 [reference]		1.00 [reference]	
Viral load non-suppression (> 1000)	1.88 [0.76, 4.66]	0.173	1.72 [0.63, 4.72]	0.289

Table 5 Binary logistic regression model of factors associated with viral load non-suppression among study participants

	Unadjusted model		**Adjusted model**	
Characteristics	**COR [95% CI]**	***P* value**	**AOR [95% CI]**	***P* value**
Malaria PCR				
Negative	**1.00 [reference]**		**1.00 [reference]**	
Positive	**1.88 [0.76, 4.66]**	**0.173**	**1.46 [0.49, 4.33]**	**0.495**
Study site				
KBTH	**1.00 [reference]**		**1.00 [reference]**	
PML	**2.11 [1.24, 3.59]**	**0.006**	**1.47 [0.80, 2.70]**	**0.212**
Age of child category				
<5 years	**1.00 [reference]**		**1.00 [reference]**	
5–9 years	**0.31 [0.15, 0.66]**	**0.002**	**0.43 [0.19, 0.97]**	**0.043**
10–14 years	**0.22 [0.10, 0.47]**	**< 0.001**	**0.37 [0.14, 0.99]**	**0.048**
Adherence				
Good	**1.00 [reference]**		**1.00 [reference]**	
Fair	**1.64 [0.76, 3.54]**	**0.211**	**1.53 [0.63, 3.72]**	**0.351**
Poor	**4.29 [1.67, 11.04]**	**0.003**	**4.89 [1.98, 12.08]**	**0.001**
ART duration				
<12 months	**1.00 [reference]**		**1.00 [reference]**	
12–59 months	**0.41 [0.13, 1.24]**	**0.113**	**0.55 [0.20, 1.49]**	**0.236**
60–119 months	**0.19 [0.06, 0.61]**	**0.005**	**0.36 [0.12, 1.07]**	**0.065**
120 + months	**0.23 [0.06, 0.87]**	**0.030**	**0.51 [0.13, 1.92]**	**0.317**
ART regimen				
DTG based	**2.74 [0.33, 22.98]**	**0.353**	**3.12 [0.28, 34.65]**	**0.355**
LPV/r based	**7.43 [0.89, 61.90]**	**0.064**	**5.10 [0.45, 57.99]**	**0.189**
EFV based	**3.16 [0.38, 26.18]**	**0.287**	**2.57 [0.24, 27.71]**	**0.436**
NVP based	**1.00 [reference]**		**1.00 [reference]**	
Clotrimazole				
No	**1.00 [reference]**		**1.00 [reference]**	
Yes	**0.92 [0.49, 1.74]**	**0.808**	**1.00 [0.49, 2.05]**	**0.993**

COR: crude odds ratio. AOR: adjusted odds ratio. CI: confidence interval

immune deficiencies and dysfunction attributable to HIV infection. A study by Okonkwo et al. (2018) looked at asymptomatic malaria parasite density in children under-five years in sub-Saharan Africa. Findings from their study showed that parasitaemia was higher in the HIV infected individuals as compared to the HIV uninfected individuals [28]. The observed prevalence of asymptomatic malaria in our study was 3.6% and may seem small, but it warrants attention given the severity of malaria in Ghana.

The low prevalence could be because the majority of the participants were using some form of malaria preventive method. The malaria preventive methods used included mosquito coils, mosquito repellent, and mosquito spray. The majority of the participants were also from urban Accra, where the transmission of malaria is relatively low. Other studies on asymptomatic CLHIV have reported higher rates of malaria parasitaemia: 8.8% in Douala City, Cameroon [29], and 34.1% in Benin City, Nigeria [7].

No factors were associated with the presence of malaria parasites in this study. Although not statistically significant, asymptomatic malaria parasitaemia was almost three times higher in participants who were not taking cotrimoxazole. This finding is consistent with previous studies that have established the dual benefits of cotrimoxazole in reducing morbidity and mortality arising from both HIV and malaria [30, 31].

In this study, older children (ages 10–14) had higher rates of asymptomatic parasitaemia than younger children (<5 years). While this difference was not statistically significant, it is known that younger children, who lack protective immunity, are at higher risk of getting sick from malaria and are more likely to be diagnosed and treated [32]. On the other hand, older children, who have developed more immunity, are more likely to have asymptomatic infections that often go untreated [33].

Some ART drugs have shown antimalarial activity, including protease inhibitors such as lopinavir and ritonavir, and nonnucleoside reverse transcriptase inhibitors (NNRTIs) such as efavirenz [34]. There was no association between ART regimen type and malaria parasitaemia in this study. Similarly, another multicenter study conducted in Kenya, Malawi, and Uganda found no difference between lopinavir/ritonavir and NNRTI based ART in the clearance of *Plasmodium falciparum* clinical parasitaemia [35].

The ultimate goal of combined ART for People Living with HIV (PLHIV)is to achieve long-term virological suppression. There was a high rate of virological non-suppression (29.6%). This translates to a suppression rate of 70.4%, which is far from the 95% target for virological suppression according to the UNAIDS framework for HIV program target milestones to be achieved by all countries by 2030. This highlights the need to closely follow and properly manage children on ART to suppress viral load in our setting. The rate of virological non-suppression obtained in this study is in agreement with other studies performed in Uganda (28%), Zimbabwe (30%) and Ghana (38.4%) [36–38]. However, other studies have reported lower rates in Ethiopia (12%) and South Africa (15%) [39, 40].

The odds of viral load non-suppression were approximately five times higher among those with poor adherence. This finding is similar to those of previous studies that have shown that virological non-suppression is significantly associated with poor adherence [36, 41, 42]. Poor adherence results in therapeutic plasma ART drug concentrations, leading to resistance to one or more drugs in a given regimen, and the possibility of cross-resistance to other drugs in the same class. Suboptimal adherence may include missed or late doses, treatment interruptions, and discontinuations, as well as therapeutic or partial dosing.

CLHIV require almost perfect levels of adherence to achieve long-lasting non-detectable viral load levels, with optimal adherence to ART being the most common cause of virologic failure. WHO's goal to stop the acquired immune deficiency syndrome (AIDS) epidemic as a public health threat by 2030 depends on multiple factors. These include universal access to testing and diagnosis, effective antiretroviral therapy (ART) coverage, prevention strategies, and addressing social determinants of health [43].

The interaction between HIV and malaria underscores the urgent need for comprehensive policy implementation to address both conditions effectively. Enhanced coordination between HIV and malaria control programs, along with targeted strategies for co-infected individuals, is essential to improve health outcomes and mitigate the impact of these diseases.

Limitations of the study

The study design was unable to resolve whether past clinical malaria left persistent parasites or whether these individuals were parasitaemic, because they were generally at a higher risk.

Overall, this study showed that the proportion of CLHIV with asymptomatic malaria parasitaemia was low. Approximately one-third of the participants had virological non-suppression. The odds of viral load non-suppression were approximately five times higher among those with poor adherence. The interaction between malaria parasitemia and viral replication may not be the main cause of virological non suppression in this low-transmission area.

Recommendations

Malaria preventive strategies should be emphasized during counselling at routine clinics to further decrease the prevalence of malaria. Adherence counselling should be strengthened for these children to ensure that their viral load levels remain suppressed. Longitudinal studies should be conducted to track the incidence and impact of asymptomatic malaria on CLHIV over time. This could help determine the true burden of asymptomatic malaria and its potential role in disease progression. There is also a need to utilize molecular techniques to characterize the genetic diversity of malaria parasites in asymptomatic CLHIV. This could provide insights into the transmission dynamics and potential drug resistance patterns. While the study focused on a specific population with a low prevalence of asymptomatic malaria, it highlights the importance of comprehensive surveillance. There is a need for tracking both symptomatic and asymptomatic cases to better understand the disease burden and identify emerging trends. Enhanced surveillance can inform targeted interventions and resource allocation for both the NACP and the Malaria Elimination Control Program.

Abbreviations

ABC	Abacavir
AIDS	Acquired Immunodeficiency Syndrome
AOR	Adjusted Odds Ratio
ART	Antiretroviral Therapy
CLHIV	Children Living with HIV
DTG	Dolutegravir
EFV	Efavirenz
HIV	Human Immunodeficiency Virus
KBTH	Korle Bu Teaching Hospital
LPV/r	ritonavir boosted lopinavir
NACP	National AIDS Control Programme
NVP	Nevirapine
PCR	Polymerase Chain Reaction
PI	Protease Inhibitor
SPSS	Statistical Package for Social Sciences
TDF	Tenofovir
COR	Crude Odds Ratio
WHO	World Health Organization
ZDV	Zidovudine
3TC	Lamivudine

Acknowledgements

Our gratitude goes to the participants from Korle Bu Teaching Hospital and Princess Marie Louis Hospital for participating in the study. We are also grateful to Seth Anim and Christodea Haizel who were involved in data collection. We thank Noguchi Memorial Institute for Medical Research and Mr. Godfrey Adams of the Chest Clinic, Korle Bu Teaching Hospital for laboratory support. We also acknowledge Fogarty International Center of National Institute of Alcohol Abuse and Alcoholism of the National Institutes of Health under Award Number D43 TW011526.

Author contributions

AKAA contributed to the study conception and design, acquisition of data, analysis and interpretation of data and drafting of the manuscript. KT contributed to the study design, interpretation of data and substantively revised it. EP contributed to the interpretation of the data and substantively revised it. YA contributed to the analysis of the data, interpretation of the data and substantively revised it. LEA substantively revised it. AAL substantively revised it. MN substantively revised it. All authors read and approved the final manuscript.

Funding

AKAA received funding from the Fogarty International Center of National Institute of Alcohol Abuse and Alcoholism of the National Institutes of Health under Award Number D43 TW011526. The content is the sole responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.

Data availability

The data sets used and analyzed during this study are available within the corresponding author on request.

Declarations

Ethics approval and consent to participate

Ethical approval to conduct research at the PML was granted by the Ghana Health Service Ethical Review Board, Accra (GHS-ERC: 047/07/22). Ethical approval to conduct research at KBTH was granted from the linstitutional Review Board of the Korle Bu Teaching Hospital Greater Accra region (KBTH-STC/IRB/000110/2022). Written informed consent/assent was obtained from study participants after the objectives of the study were explained. All procedures involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and the 1964 Helsinki Declaration and its later amendments or comparable ethical standards.

Consent for publication

Not applicable.

Competing interests

Findings of this study are not influenced by the finders or the position of the finders. The authors declare no competing interests.

Author details

[1]Department of Child Health, University of Ghana Medical School, Accra, Ghana
[2]Department of Biostatistics, School of Public Health, University of Ghana, Legon, Accra, Ghana
[3]Department of Immunology, Noguchi Memorial Institute for Medical Research, University of Ghana, Accra, Ghana
[4]Princess Marie Louise Children's Hospital, P.O. Box GP 122, Accra, Ghana
[5]Department of Epidemiology and Disease Control, School of Public Health, College of Health Sciences, University of Ghana, Accra, Ghana
[6]Boston University Chobanian and Avedisian School of Medicine, Boston, MA, USA
[7]Department of Population, Family and Reproductive Health, School of Public Health, College of Health Sciences, University of Ghana Legon, Accra, Ghana

Received: 30 December 2023 / Accepted: 22 September 2024
Published online: 11 October 2024

References

1. Kwenti TE. Malaria and HIV coinfection in sub-saharan Africa: prevalence, impact, and treatment strategies. Res Rep Trop Med. 2018;9:123. https://doi.org/10.2147/RRTM.S154501.
2. Moyo E, Moyo P, Murewanhema G, Mhango M, Chitungo I, Dzinamarira T. Key populations and Sub-saharan Africa's HIV response. Front Public Health. 2023;11. https://doi.org/10.3389/FPUBH.2023.1079990.
3. World malaria report. 2021 n.d. https://www.who.int/teams/global-malaria-programme/reports/world-malaria-report-2021 (accessed June 1, 2024).
4. Kwenti TE. Malaria and HIV coinfection in sub-saharan Africa: prevalence, impact, and treatment strategies. Res Rep Trop Med. 2018;9. https://doi.org/10.2147/rrtm.s154501.
5. Sandie SM, Sumbele IUN, Tasah MM, Kimbi HK. Malaria parasite prevalence and haematological parameters in HIV seropositive patients attending the regional hospital Limbe, Cameroon: a hospital-based cross-sectional study. BMC Infect Dis. 2019;19. https://doi.org/10.1186/S12879-019-4629-4.

6. Roberds A, Ferraro E, Luckhart S, Stewart VA. HIV-1 impact on Malaria Transmission: a complex and relevant Global Health concern. Front Cell Infect Microbiol 2021;11. https://doi.org/10.3389/FCIMB.2021.656938/FULL
7. Figueroa-Romero A, Saura-Lázaro A, Fernández-Luis S, González R. Uncovering HIV and malaria interactions: the latest evidence and knowledge gaps. Lancet HIV. 2024;11:e255–67. https://doi.org/10.1016/S2352-3018(24)00035-3.
8. Alemu A, Shiferaw Y, Addis Z, Mathewos B, Birhan W. Effect of malaria on HIV/AIDS transmission and progression. Parasit Vectors. 2013;6:18. https://doi.org/10.1186/1756-3305-6-18.
9. Flateau C, Le Loup G, Pialoux G. Consequences of HIV infection on malaria and therapeutic implications: a systematic review. Lancet Infect Dis. 2011;11:541–56. https://doi.org/10.1016/S1473-3099(11)70031-7.
10. Kasirye R, Baisley K, Munderi P, Grosskurth H. Effect of cotrimoxazole prophylaxis on malaria occurrence in HIV-infected patients on antiretroviral therapy in sub-saharan Africa. Trop Med Int Health. 2015;20:569–80. https://doi.org/10.1111/tmi.12463.
11. Chalwe V, Van Geertruyden JP, Mukwamataba D, Menten J, Kamalamba J, Mulenga M, et al. Increased risk for severe Malaria in HIV-1–infected adults, Zambia - 15, number 5—May 2009 - emerging infectious diseases journal - CDC. Emerg Infect Dis. 2009;15:749–55. https://doi.org/10.3201/EID1505.081009.
12. World Health Organization. Consolidated guidelines on HIV prevention, testing, treatment, service delivery and monitoring: recommendations for a public health approach. 2021.
13. Das NG, Dhiman S, Talukdar PK, Goswami D, Rabha B, Baruah I, et al. Role of asymptomatic carriers and weather variables in persistent transmission of malaria in an endemic district of Assam, India. Infect Ecol Epidemiol. 2015;5:25442. https://doi.org/10.3402/iee.v5.25442.
14. Danquah I, Ziniel P, Eggelte TA, Ehrhardt S, Mockenhaupt FP. Influence of haemoglobins S and C on predominantly asymptomatic Plasmodium infections in northern Ghana. Trans R Soc Trop Med Hyg. 2010;104:713–9. https://doi.org/10.1016/j.trstmh.2010.08.001.
15. Greenwood B, Marsh K, Snow R. Why do some African children develop severe malaria? Parasitol Today. 1991;7:277–81. https://doi.org/10.1016/0169-4758(91)90096-7.
16. Statistical Service Accra G. Ghana Malaria Indicator Survey 2019 Final Report 2020.
17. Alemu A, Shiferaw Y, Addis Z, Mathewos B, Birhan W. Effect of malaria on HIV/AIDS transmission and progression. Parasit Vectors. 2013;6:1–8. https://doi.org/10.1186/1756-3305-6-18/COMMENTS.
18. Mirzohreh ST, Safarpour H, Pagheh AS, Bangoura B, Barac A, Ahmadpour E. Malaria prevalence in HIV-positive children, pregnant women, and adults: a systematic review and meta-analysis. Parasit Vectors. 2022;15. https://doi.org/10.1186/s13071-022-05432-2.
19. WHO. From malaria control to malaria elimination: a manual for elimination scenario planning. Who. 2014;52:67.
20. WHO | 7. 3 Monitoring response to ART and the diagnosis of treatment failure n.d. http://www.who.int/hiv/pub/guidelines/arv2013/art/artmonitoring/en/index3.html (accessed January 11, 2017).
21. Ikilezi G, Achan J, Kakuru A, Ruel T, Charlebois E, Clark TD, et al. Short report: prevalence of asymptomatic parasitemia and gametocytemia among HIV-infected Ugandan children randomized to receive different antiretroviral therapies. Am J Trop Med Hyg. 2013;88:744–6. https://doi.org/10.4269/ajtmh.12-0658.
22. Suleiman IA, Momo A. Adherence to antiretroviral therapy and its determinants among persons living with HIV/AIDS in Bayelsa state. Nigeria Pharm Pract (Granada). 2016;14:0–0. https://doi.org/10.18549/PHARMPRACT.2016.01.631.
23. WHO. Giemsa staining of malaria blood films 2016. https://www.who.int/publications/i/item/HTM-GMP-MM-SOP-07a (accessed December 2, 2023).
24. Snounou G, Viriyakosol S, Xin Ping Zhu, Jarra W, Pinheiro L, do Rosario VE, et al. High sensitivity of detection of human malaria parasites by the use of nested polymerase chain reaction. Mol Biochem Parasitol. 1993;61:315–20. https://doi.org/10.1016/0166-6851(93)90077-B.
25. Ayanful-Torgby R, Quashie NB, Boampong JN, Williamson KC, Amoah LE. Seasonal variations in Plasmodium Falciparum parasite prevalence assessed by varying diagnostic tests in asymptomatic children in southern Ghana. PLoS ONE. 2018;13. https://doi.org/10.1371/JOURNAL.PONE.0199172.
26. Adjah J, Fiadzoe B, Ayanful-Torgby R, Amoah LE. Seasonal variations in Plasmodium Falciparum genetic diversity and multiplicity of infection in asymptomatic children living in southern Ghana. BMC Infect Dis. 2018;18:1–10. https://doi.org/10.1186/S12879-018-3350-Z/FIGURES/4.
27. Demographic G. and Health Survey 2022 Key Indicators Report n.d.
28. Okonkwo I, et al. A study of malaria parasite density in HIV-1 positive under-fives in Benin city. Nigeria J Trop Pediatr. 2018;64:289–96. https://doi.org/10.1093/tropej/fmx065.
29. Bayoï CN, Lehman LG, Tonga C, Kangam L, Wepnje GB, Tchanga FC, et al. Effects of Malaria and HIV infection on Anemia and T-cells levels in children in Douala City. Cameroon Cureus. 2022;14. https://doi.org/10.7759/CUREUS.32074.
30. Okonkwo IR, Ibadin MO, Omoigberale AI, Sadoh WE. Effect of HIV-1 Serostatus on the prevalence of asymptomatic Plasmodium Falciparum Parasitemia among Children Less Than 5 years of age in Benin City, Nigeria. J Pediatr Infect Dis Soc. 2015;5:21–8. https://doi.org/10.1093/jpids/piu093.
31. Mermin J, Ekwaru JP, Liechty CA, Were W, Downing R, Ransom R, et al. Effect of co-trimoxazole prophylaxis, antiretroviral therapy, and insecticide-treated bednets on the frequency of malaria in HIV-1-infected adults in Uganda: a prospective cohort study. Lancet. 2006;367:1256–61. https://doi.org/10.1016/S0140-6736(06)68541-3.
32. Carneiro I, Roca-Feltrer A, Griffin JT, Smith L, Tanner M, Schellenberg JA, et al. Age-patterns of malaria vary with severity, transmission intensity and seasonality in sub-saharan Africa: a systematic review and pooled analysis. PLoS ONE. 2010;5. https://doi.org/10.1371/JOURNAL.PONE.0008988.
33. Yeka A, Nankabirwa J, Mpimbaza A, Kigozi R, Arinaitwe E, Drakeley C, et al. Factors associated with malaria parasitemia, anemia and serological responses in a spectrum of epidemiological settings in Uganda. PLoS ONE. 2015;10. https://doi.org/10.1371/JOURNAL.PONE.0118901.
34. Kanters S, Vitoria M, Doherty M, Socias ME, Ford N, Forrest JI, et al. Comparative efficacy and safety of first-line antiretroviral therapy for the treatment of HIV infection: a systematic review and network meta-analysis. Lancet HIV. 2016;3:e510–20. https://doi.org/10.1016/S2352-3018(16)30091-1.
35. Shaffer D, Kumwenda J, Chen H, Akelo V, Angira F, Kosgei J et al. Brief Report: No Differences Between Lopinavir/Ritonavir and Nonnucleoside Reverse Transcriptase Inhibitor–Based Antiretroviral Therapy on Clearance of Plasmodium falciparum Subclinical Parasitemia in Adults Living With HIV Starting Treatment (A5297). J Acquir Immune Defic Syndr (1988) 2022;89:178–82. https://doi.org/10.1097/QAI.0000000000002839
36. Bulage L, Ssewanyana I, Nankabirwa V, Nsubuga F, Kihembo C, Pande G, et al. Factors Associated with Virological non-suppression among HIV-Positive patients on antiretroviral therapy in Uganda, August 2014-July 2015. BMC Infect Dis. 2017;17:326. https://doi.org/10.1186/s12879-017-2428-3.
37. Makadzange AT, Higgins-Biddle M, Chimukangara B, Birri R, Gordon M, Mahlanza T, et al. Clinical, Virologic, immunologic outcomes and emerging HIV Drug Resistance patterns in children and adolescents in Public ART Care in Zimbabwe. PLoS ONE. 2015;10. https://doi.org/10.1371/JOURNAL.PONE.0144057.
38. Afrane AKA, Goka BQ, Renner L, Yawson AE, Alhassan Y, Owiafe SN, et al. HIV virological non-suppression and its associated factors in children on antiretroviral therapy at a major treatment centre in Southern Ghana: a cross-sectional study. BMC Infect Dis. 2021;21. https://doi.org/10.1186/S12879-021-06459-Z.
39. Chandiwana N, Sawry S, Chersich M, Kachingwe E, Makhathini B, Fairlie L. High loss to follow-up of children on antiretroviral treatment in a primary care HIV clinic in Johannesburg, South Africa. Medicine. 2018;97. https://doi.org/10.1097/MD.0000000000010901.
40. Yihun BA, Kibret GD, Leshargie CT. Incidence and predictors of treatment failure among children on first-line antiretroviral therapy in Amhara Region Referral hospitals, northwest Ethiopia 2018: a retrospective study. PLoS ONE. 2019;14:e0215300. https://doi.org/10.1371/JOURNAL.PONE.0215300.
41. Moolasart V, Chottanapund S, Ausavapipit J, Likanonsakul S, Uttayamakul S, Changsom D, et al. The Effect of detectable HIV viral load among HIV-Infected children during antiretroviral treatment: a cross-sectional study. Children. 2018;5:6. https://doi.org/10.3390/children5010006.
42. Shumetie A, Moges NA, Teshome M, Gedif G. Determinants of virological failure among HIV-Infected Children on First-Line Antiretroviral Therapy in West Gojjam Zone, Amhara Region, Ethiopia. HIV AIDS (Auckl). 2021;13:1035. https://doi.org/10.2147/HIV.S334067.

43. Fonsah JY, Njamnshi AK, Kouanfack C, Qiu F, Njamnshi DM, Tagny CT, et al. Adherence to antiretroviral therapy (ART) in Yaoundé-Cameroon: Association with opportunistic infections, Depression, ART Regimen and Side effects. PLoS ONE. 2017;12. https://doi.org/10.1371/JOURNAL.PONE.0170893.

Publisher's note