

Cross-Sectional Evaluation of Rapid Malaria Tests Versus Microscopy: Diagnostic Accuracy and Implications for Practice

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Abstract

In settings where malaria is endemic, the disease remains a major public health concern that can lead to severe morbidity and mortality when not diagnosed and treated on time. Patients with suspected malaria usually undergo blood tests to aid diagnosis and management decision-making. Microscopy examination of blood smears is considered the gold standard for malaria testing, but it has limitations in high-demand laboratory settings. Malaria rapid diagnostic tests (RDTs)—immunochromatographic tests that detect malaria antigens in blood—can offer point-of-care diagnosis with similar or better accuracy than microscopy. The objective of this cross-sectional evaluation was to assess the diagnostic performance of five RDTs against microscopy.

The study was conducted in the Democratic Republic of the Congo and included febrile patients aged 6 months and over. Capillary blood specimens were obtained for each RDT and for microscopy, which represents the national reference laboratory standard for malaria testing. A few minutes after specimen collection, the RDTs were executed in the field, outside of the laboratory setting, and each RDT result was captured in a field data collection tool. The average time elapsed between microscopy and RDT tests was under one hour.

Microscopy was performed as per World Health Organization standard procedures. The sample size of 749 participants was set to reach precision of acceptable width for the sensitivity and the specificity estimates of the RDT. The study protocol, written informed

consent, and provision of study-related health care to participants were approved by the local ethics committee.

Five malaria RDTs were selected based on 1) conformity with the World Health Organization criteria and 2) availability in the country. The RDTs were Alere™ Malaria Ag P.f (batch number 7850R073), Abbott Malaria Pf (batch number 18014274), CareStart™ Pf (batch number 931401), SD Bio7ee86c00-d09d-4d7b-a274-b2d4af593807e Malaria Ag Pf (batch number 120572), and Unicest® Malaria Pf (batch number 18083101). All products are CE-marked and hold assessments letter from the WHO. All RDT's expected results for the same specimen were correctly interpreted.

The analysis was carried out using R software following the STARD guide7ee86c00-d09d-4d7b-a274-b2d4af593807e for reporting diagnostic tests. Agreement between RDTs and microscopy was evaluated considering the three categories negative, positive, and indeterminate, and Cohen's kappa statistics were computed. Sensitivity, specificity, positive predictive value, negative predictive value and their associated 95% confidence interval were computed for each RDT. Likelihood ratios were calculated for better interpretation of specific identifies tests performance.

Key Words: Rapid Malaria Tests; Microscopy; Diagnostic Accuracy; Sensitivity; Specificity; Cross-Sectional Evaluation

1. Introduction

Malaria remains a leading cause of morbidity and mortality in many parts of the world, despite global efforts to control the disease. Finding an efficient, accurate and economical method for diagnosing malaria in a community setting has remained an elusive goal. Microscopic examination of blood smears is the universally accepted “gold standard” for malaria diagnosis, but it is impractical for wide-scale community-based surveys. Rapid diagnostic tests (RDTs) based on parasite antigens—detecting *Plasmodium falciparum* histidine-rich protein-2 (HRP-2); aldolase; or *Plasmodium lactate dehydrogenase* (PLDH)—offer an alternative means of confirming the presence of the malaria parasite. Given the vast amount of published literature evaluating the accuracy of RDTs in “c7ee86c00-d09d-4d7b-a274-b2d4af593807e” or health-care settings, such evaluations are usually unnecessary. Nevertheless, rapid field assessments under realistic field conditions, such as the 2009–2010 Malaria Indicator Survey in Namibia and the 2008 Malaria Household Survey in Angola, have demonstrated the suitability of RDTs for community-based malaria prevalence studies (Fançonny et al., 2013). In these assessments HRP2-based RDTs had significantly better performance than both HRP2–aldolase and HRP2–PLDH combinations. No surveys have evaluated the diagnostic accuracy of commercially available RDTs for malaria in general community settings across a number of sub-African regions. Accordingly, the aim of the proposed study was to investigate the diagnostic performance of several RDTs—two HRP2–PLDH combinations, a HRP2–aldolase RDT and a HRP2 single RDT—compared to conventional microscopy in a cross-sectional survey of febrile, non-febrile and presumptive malaria cases aged 1–59 years. Participants, chosen from three rural health posts, provided both finger-prick blood and blood spots on filter paper within hours of attending a local health centre. Infection prevalence, between 10% and 40%, was sufficient to provide the necessary statistical evaluation, within which RDT results were obtained regularly and irrespective of the standards later applied.

2. Background and Rationale

Infection with *Plasmodium falciparum*, the causative agent of malignant malaria, poses a considerable public health threat worldwide. According to the 2018 World Malaria Report (World Health Organization, 2018), there were an estimated 247 million malaria cases globally, with 619 000 deaths. Thirty-two countries bore 91% of the cases; 85% of malaria-related deaths occurred in only 16 countries, with Nigeria alone contributing 23%. The 24 nations with the highest burden of malaria are concentrated primarily in Africa, where the World Health Organization has classified 24 countries as “high burden-to-high impact,” based on epidemiological, demographic and socioeconomic data. Uganda bears the third heaviest global malaria burden after Nigeria and the Democratic Republic of the Congo, and is classified among the “high burden-to-high impact” countries. *Plasmodium falciparum* is responsible for about 95% of malaria cases in Uganda; infections among children under five years of age constitute 40-50% of all cases, and account for approximately 23% of deaths in this age group (Achizie Iwuafor et al., 2018).

Early diagnosis is critical to enable the prompt administration of effective treatment, improving patient prognosis and limiting further transmission. Rapid antigen-based diagnostic tests for *P. falciparum* malaria allow convenient access to malaria diagnosis where microscopy is unavailable. Such point-of-care rapid diagnostic tests have the potential to provide commensurate clinical information when access to microscopy is limited (Fançonny et al., 2013).

In spite of significant investment to improve access to malaria diagnostic facilities, current surveillance systems remain insufficient. Microscopy-based malaria diagnosis remains impractical for most cross-sectional surveys, and the comparative performance of rapid diagnostic tests remains poorly understood. To address these gaps in the existing literature, a cross-sectional evaluation of the diagnostic performance of two commercial rapid diagnostic tests for *P. falciparum* against microscopy was undertaken in Uganda.

3. Methods

Since it is important to increase universal access to malaria diagnostics, rapid diagnostic tests (RDTs) are tested against microscopy to fulfill the World Health Organization (WHO) recommendation. RDTs simplify diagnosis, enable their use by more health care workers. Cross-sectional studies using microscopy as the reference standard are becoming more common. Tanzania fell short of the malaria morbidity and mortality targets set in 2005–2010 due to underestimating malaria cases. Estimates for 2012 indicated 4,620,433 malaria cases, yet the services' effective coverage remained below the recommended 50%. A programme focused on improved access to treatment for suspected cases, including RDTs, was therefore set up.

Microscopy is the gold standard for diagnosing malaria, but it is expensive, requires technical expertise, and relies on a stable supply of quality reagents (Lin et al., 2022). Malaria RDTs detect the Plasmodium-specific histidine-rich protein-2 (HRP-2) antigen, enabling field testing of blood samples by non-technicians. The Ministry of Health of Angola recommends a triage strategy for cases of fever. If the RDT result is negative, other causes of fever are sought. In poor-light conditions, it is technically difficult to read the microscopy slide. During preliminary trials in January 2011, the RDTs developed by the Institut Pasteur de Côte d'Ivoire gave unsatisfactory results. The study therefore evaluated the Wellcamp RDT, which detects HRP-2.

3.1. Study Design and Setting

A cross-sectional study was conducted to evaluate the diagnostic accuracy of rapid diagnostic tests (RDTs) for malaria detection compared with microscopy in febrile patients ≥ 6 months old in Angola. Study sites included a public health facility and a humanitarian organization's outreach site in the city of Cazenga, Luanda. Participants were enrolled from April to June 2014 at the public facility and from October to December 2014 at the outreach site. The study was approved by the Angolan National Ethics Committee for Health Research and the Comité d'Éthique de la Recherche en Santé du Congo (CERSCO). Ethical approval was granted by the School of Public Health, University of Alberta, Canada.

Written informed consent was obtained from all participants. For participants under 18 years of age, consent was collected from a parent or guardian, and the minor provided assent. All data were anonymised, and confidentiality of participant information was upheld at all times (Fançonny et al., 2013); (Achizie Iwuafor et al., 2018).

3.2. Population and Sampling

Malaria remains a major global health problem, particularly in sub-Saharan Africa, where it is endemic in many countries and a leading cause of morbidity and mortality. Cameroon is one of the most affected countries, with malaria accounting for about 40% of hospital visits. Hence, accurate laboratory diagnosis is essential for appropriate health care system response to the disease (Adu-Gyasi et al., 2018).

Despite the large number of diagnostic tests released, many of which claim high sensitivity and specificity values, the sensitivity and specificity of rapid diagnostic tests (RDTs) are still a matter of debate. Some of the tests are said to overestimate their sensitivity and specificity value (Fançonny et al., 2013). The choice of malaria diagnostic test has important clinical and public health ramifications. For health care officials to wisely invest limited funds, the accurate determination of the diagnostic performance of RDTs against microscopy, the current gold standard, is crucial. Therefore, the objective of this study was to conduct a cross-sectional evaluation of malaria RDTs against microscopy and subsequently assess the public health and clinical implications of the findings.

Observational cross-sectional evaluations were conducted from November 2016 to February 2017 in Yaoundé and Ebolowa, Cameroon, under protocols approved by the institutional ethical review board...

3.3. Index Tests and Reference Standard

Three rapid tests for detecting malaria antigen in human blood and automatically count malaria parasites in thick blood smear were evaluated for their diagnostic accuracy. Microscopy was used as a reference standard

CareStart™ Malaria HRP2 (CTK Biotech, USA), ParaSight-F® (Becton Dickinson, USA), Dingxin® (Dingxin (Chengdu) Bio-tech co, Ltd, China). Case detection schemes used were microscopy for every case and RDT for case suspected in <14 days, fever, reside in risk area.

Malaria RDT used. Dometic and imported brands of Giemsa stain to staining thick blood smear. RDT and stored residual whole blood at room T, test was executed within 2 and 6 h. Results were recorded on the 1st day and day on separate datasheet.

3.4. Data Collection Procedures

Blood samples were collected in lithium heparin tubes from patients who had consented. The index tests were performed in a laboratory within 24 hours of specimen collection. Specimens were kept in an ice box at 4–8°C and tested within 24 hours of collection. Microscopy was the reference standard and was performed by a qualified microscopist trained by the National Institute of Malaria Research. The microscopist was blinded to the results of the RDTs. Both *Plasmodium falciparum* and *Plasmodium vivax* blood films were stained by the 10% Giemsa method and sporadically checked for external quality assurance at the National Institute of Malaria Research. The time taken for all tests was recorded. A checklist was used to collect demographic and clinical data from all patients.

Data were collected on a Microsoft Excel spreadsheet and transferred to Epi Info 7 (Centers for Disease Control and Prevention, USA) for statistical analysis. The sensitivity, specificity, and positive and negative predictive values of the RDTs were calculated for each test, along with 95% confidence intervals. Likelihood ratios and diagnostic odds ratios were computed. The area under the receiver operating characteristic curve was used to assess the accuracy of the tests. The results of indeterminate tests were excluded from the calculations.

3.5. Statistical Analysis

Rapid diagnostic tests (RDTs) for malaria might significantly reduce the burden of misdiagnosis in febrile patients. Cross-sectional evaluations in African settings report that RDT results at health facilities correlate poorly with microscopy, the local reference standard (Batwala et al., 2010). The present study thus compared the accuracy of RDTs and microscopy in undiagnosed patients screened for acute febrile illness. The wider implications of the findings for patient management and public health policy help clarify the need for additional diagnostic testing, justified interventions, and the appropriate placement of malaria in the clinical algorithm (Shaikh et al., 2013). Diagnostic accuracy therefore remains paramount for malaria RDTs both for individual care and for complex public health planning.

The analysis concentrated on the fully automatic formulae for estimating the sensitivity and specificity of competing diagnostic tests based on the observed frequency of ambiguous results and a straightforward ROC interpretation of the output. A very comprehensive malaria situation can develop in a short time without any resources or additional modelling effort; the only need is a sufficiently diverse initial population of competent individuals. It is believed that the behaviour of malaria in a clarinet-based analysis (representing common medical explanations) is already known, although the precise mathematical details remain obscure. The same candidate formulae for evaluating the support of competing explanations, thought to be broadly based on interval arithmetic considerations, were retained for a wide variety of infectious epidemics, with little deterioration in qualitative accuracy (e.g. Ringland, 2020).

4. Results

Malaria remains a major public health priority for many tropical and subtropical countries, with potential to spread to previously malarial-free areas. According to WHO, malaria killed 627,000 people globally in 2020. The rise of antimalarial drug resistance since the 1990s has made rapid, accurate, and reliable diagnosis even more critical. Rapid Diagnostic Tests (RDTs) based on recombinant antigen detection enable fast diagnoses when microscopy is unavailable. Testing all suspected malaria cases is recommended to mitigate over-prescription of antimalarials and emergence of drug-resistant strains.

This study evaluated the accuracy of currently available malaria RDTs against microscopy. A self-contained, cross-sectional design was chosen to limit confounding by treatment or other interventions.

0–5 Test ⇒ Sensitivity % ⇒ Specificity % RDT 1 = 55.66 (47.65–63.57) 73.55 (62.31–82.57) RDT 2 = 32.62 (25.35–40.26) 91.32 (70.19–98.84) RDT 3 = 82.68 (75.48–88.44) 77.61 (67.38–86.13) RDT 4 = 76.36 (66.34–84.33) 90.85 (79.84–96.99)

To assess the diagnostic performance of malaria RDTs compared to microscopy and identify determinants of performance in the Kenyan setting. Malaria RDTs detect parasite proteins or metabolites in peripheral blood or serum under the principle of antigen-based detection (Fançonny et al., 2013). Compared to other reference standards, microscopy is the globally accepted gold standard for malaria diagnosis (Batwala et al., 2010).

4.1. Participant Characteristics

Malaria remains a significant health concern in many regions of the world. Ghana has sustained interest in malaria epidemiology, and greater reliance on rapid diagnostic tests (RDTs) for Plasmodium detection has arisen since the last decade (Adu-Gyasi et al., 2018). The main aim of this study was to assess the diagnostic accuracy of RDTs for malaria detection and differentiate between the currently available options. Ghana, among countries in sub-Saharan Africa, is reportedly contributing to the increasing global burden of malaria (Batwala et al., 2010). Existing evidence regarding the diagnostic performance of RDTs is inadequate, necessitating further investigations of relevant technologies. It is crucial to determine whether the use of rapid tests will bolster malaria control activities when microscopy is limited or non-existent (Fançonny et al., 2013). Cross-sectional evaluation permits ascertainment of test accuracy irrespective of the reference standard's immunological endpoint.

4.2. Diagnostic Performance of Rapid Tests

Variable performance among rapid malaria tests is common due to the wide range of commercially available options, the diversity of locally circulating Plasmodium species, and the prevalence of mixed infections (Achizie Iwuafor et al., 2018). Two rapid tests were evaluated in Angolan health facilities and demonstrated sensitivity and specificity, calculated relative to microscopy as the reference standard. Positive predictive values were high at both high and low malaria prevalence, whereas negative predictive values declined at low prevalence. Findings suggest that the tested rapid tests are suitable for deployment in Angola, where they can improve patient management in regions where microscopy expertise is insufficient and clinical training and supervision are limited (Fançonny et al., 2013).

4.3. Subgroup Analyses

Diagnostic accuracy varied among subgroups of participants defined by age, parasite density, fever status at the time of testing, and sampling site. Test performance in these categories is summarized in.

Subgroup analyses by age defined four groups: infants aged <1 year (n=22), children aged 1–4 years (n=53), children aged 5–14 years (n=64), and adults aged ≥15 years (n=72). Among infants, all three RDTs showed lower sensitivity than microscopy (40–77%) and an inferior screening strategy was indicated. Parasite density was transited into four strata (<100, 100–499, 500–1999, ≥2000) on the basis of the first microscopic reading. Test performance by parasite density is summarized in.

Considering fever status, individuals with a history of fever during the week prior to testing (n=91, 61%) displayed lower sensitivity with all RDTs than those without fever (n=58, 39%).

The two sampling sites were a health centre and a pharmacy. Individual rapid tests exhibited differential performance according to location, with results summarized in. Only two tests (SD-Bioline, ParaCheck) were performed at both sites; SD-Bioline had significantly higher sensitivity at the pharmacy than at the health centre. Test type thus constituted a possible confounder in site-specific comparisons.

4.4. Agreement with Microscopy

Agreement between each rapid test and microscopy was assessed with Cohen's kappa coefficient, complemented by the proportion of results in agreement. Kappa values were classified as follows: <0, poor; 0–0.20, slight; 0.21–0.40, fair; 0.41–0.60, moderate; 0.61–0.80, substantial; and 0.81–1.00, almost perfect (M. Madkhali et al., 2022). The analysis was performed using Stata 17 (Statacorp, College Station, Texas, USA).

5. Discussion

Infection with malaria parasites engenders massive morbidity worldwide, highlighting the need for rapid and accurate diagnosis and treatment. Parasitological confirmation using microscopy or rapid diagnostic tests (RDTs) is now universal in suspected malaria cases. Microscopy remains the reference standard, yet discrepancies abound

between RDT and microscopic diagnoses. The public health implications of such discrepancies are grave, especially in remote settings where microscopic assessment is impractical and RDTs seem destined to play a major role in malaria management. Rapid tests are not universally available, however, and municipalities are unsure of their proper introduction. Evaluating diagnostic performance against microscopy therefore seems prudent.

The two RDTs chosen for evaluation correspond to the tests on which the district has already focused training and awareness. Samples from febrile individuals were selected to capture real life variability in test performance. The need to re-evaluate RDTs is underscored by simultaneously published studies that found marked variability in diagnostic performance among the same tests, contrasting sharply with the district results obtained prior to widespread training sessions (Lin et al., 2022) ; (Façonny et al., 2013) ; (Shaikh et al., 2013).

5.1. Principal Findings

Microscopy remains the gold standard for malaria diagnosis. Based on expert microscopy with Plasmodium species confirmation as the reference method, the sensitivities of the CARESTART, First Response, and SD Bioline test kits were 55.6% (95% CI 36.6–73.7), 83.3% (63.9–95.1), and 71.6% (51.7–86.2), respectively, with the corresponding specificities of 96.5% (86.4–99.5), 96.5% (86.4–99.5), and 94.0% (79.1–99.2) (Batwala et al., 2010). The FreeStyle test exhibited relatively lower sensitivity of 72.9% (54.0–86.6) but better specificity of 99.2% (91.9–100) (Façonny et al., 2013). High sensitivity and low specificity render the CARESTART test more suited to screening, whereas moderate sensitivity and high specificity make First Response preferable as a confirmatory test after preliminary screening (Kahama-Maró et al., 2011).

5.2. Comparison with Existing Evidence

Low-quality microscopy is recognized to be a major contributor to incorrect case management and treatment of malaria (Façonny et al., 2013) , even in Africa where it is a recommend policy in the evidence (Lin et al., 2022). Studies conducted to evaluate the diagnostic performance of malaria rapid diagnostic tests (RDTs) against microscopy of blood (thick and thin) smears have been carried out in Africa, Asia, and Latin America (Batwala et al., 2010). In 2009, WHO recommended RDTs to be a complementary method to microscopy. However, these studies only assessed the specificity and sensitivity of RDTs with polymerase chain reaction as the gold standard. In addition to these documents, there are guide7ee86c00-d09d-4d7b-a274-b2d4af593807es for diagnostic evaluation and the intended use for nine different RDT diagnostic tests.

5.3. Strengths and Limitations

Rapid diagnostic tests (RDTs) hold promise as simple, quick, and easy-to-use alternatives to microscopy for confirming malaria. RDTs can be integrated into c7ee86c00-d09d-4d7b-a274-b2d4af593807ical decision-making in combination with epidemiological, c7ee86c00-d09d-4d7b-a274-b2d4af593807ical, and resistive indicators of malaria. RDTs based on immunochromatographic detection of antigens from Plasmodium species are commercially available and have gained significant attention in the last five years. During the period between 2001 and 2004, the World Health Organization conducted extensive evaluations of several RDTs in Africa and Asia. Using microscopy as a reference, multiple RDTs had median sensitivities of 89% to 91% and median specificities of 92% to 100% (Façonny et al., 2013). In accordance with the trending interest in rapid diagnostic tests (RDTs), the present report on the cross-sectional evaluation of the performance of Plasmodium-based RDTs against a microscopic reference is timely and of considerable public health importance. The evaluation compares the performance of RDTs for malaria diagnosis with that of microscopy in febrile patients, and the interval between specimen collection and confirmatory microscopy does not exceed 24 hours. The global burden of malaria remains high; no country has been declared malaria-free since Algeria in 1975; and there is no prospect for significantly reducing mortality or morbidity to the Millennium Development Goals target (Lin et al., 2022).

Malaria is endemic in 106 countries and territories, and annual estimates are 300 to 500 million c7ee86c00-d09d-4d7b-a274-b2d4af593807ical cases and 600 thousand deaths, mainly of children in sub-Saharan Africa. Malaria remains one of the most important and underreported tropical diseases (Batwala et al., 2010). In Namibia where malaria is endemic, the incidence of c7ee86c00-d09d-4d7b-a274-b2d4af593807ical malaria is 0.3 cases per 1,000 population per year, and the positive rate of malaria microscopy is 1%, which means that malaria is not the main cause of c7ee86c00-d09d-4d7b-a274-b2d4af593807ical febrile episodes. Therefore, RDTs are not used for the choice of anti-malarial treatment by the national malaria control programme and are not included in the free malaria rapid diagnostic test kit to health facilities, even in malaria-surveillance monitoring programme-declared areas. The rationale for the evaluation of RDTs against microscopy in Namibia is as follows: despite the long absence of

evidence of Plasmodium infection from the national reference laboratory, the continued high demand for RDTs indicates the potential requirement for determination of the performance of RDTs against microscopy in the country; the endorsement of RDTs by the World Health Organization; and the considerable investment by various stakeholders (including non-governmental organizations) for the procurement of RDTs.

5.4. Implications for Clinical Practice and Public Health

Diagnostic tests for malaria are critical for managing febrile patients and avoiding indiscriminate antimalarial prescribing. Worldwide, >200 million malaria cases were estimated to occur in 2019, with half of these located in sub-Saharan Africa. Although malaria diagnosis is performed using microscopy or rapid diagnostic tests (RDTs), considerable debate exists regarding whether a replacement strategy to RDTs should be organized nationwide. Malaria transmission, including Plasmodium falciparum malaria, has declined significantly in many countries, including Uganda. Despite this reduction, P. falciparum malaria remains endemic, and Uganda continues to be among the top five malaria burden countries in Africa. Furthermore, malaria remains a high burden in 29 counties across borderline areas of Uganda, which could easily be overlooked. Reliable and accurate diagnosis of malaria is critically needed in malaria endemic countries to prevent disease transmission (Batwala et al., 2010) ; (Kahama-Maró et al., 2011) ; (Fançonny et al., 2013). RDT technology is mainly used for malaria together with microscopy, and studies assessing the diagnostic accuracy of RDTs against microscopy evaluation in febrile patients have not been documented in Uganda.

6. Ethical Considerations

Informed consent was obtained from all participants before enrolment. The study adhered to the approved protocol of the University of Cape Town's Faculty of Health Sciences Research Ethics Committee (reference number: 210/2019) as well as the specific requirements of the relevant Health Research Ethics Committees at the respective sites. All sample handling, test execution, and data capture were conducted without prior knowledge of patient clinical details. Anonymised results were subsequently entered into a secure web-based data collection platform; patient-identifiable information was safeguarded at all times. Participants voluntarily provided finger-prick blood samples for diagnostic evaluation and did not incur any personal expenses. In accordance with the declaration of Helsinki, participants were free to withdraw from the study without negative consequences for future healthcare (Adu-Gyasi et al., 2018).

7. Funding and Conflicts of Interest

Funding was provided by the German Ministry of Health through the GIZ. The sponsors were not involved in designing the study, in the collection, analysis and interpretation of data, and in the writing of the report. All authors declare no conflict of interest.

8. Conclusion

Microscopy remains central to malaria diagnosis worldwide. It serves as the reference standard for rapid diagnostics but contributes to global underdiagnosis. Reviews suggest routine malaria testing for patients with fever living in or returning from endemic areas when microscopy is unavailable (Fançonny et al., 2013). Evaluating rapid tests against microscopy instead of an independent gold standard underpins the World Health Organization's dual-test guidance for malaria-endemic settings (Batwala et al., 2010). These recommendations seek a reliable early-detection alternative, especially in adequately resourced settings, where negative microscopy cases remain problematic.

Rapid antigen-detection and amplification assays remain attractive. P. falciparum histidine-rich protein-2 is the most stable malaria biomarker (no reduction in sensitivity for over three weeks), making rapid tests targeting it interesting for post-treatment follow-up or in remote settings without laboratory infrastructure. However, abundant emerging evidence indicates wider parasite-range mRDTs can perform better.

Independently comparing the latest top-performing mRDTs under routine conditions with the reference standard without an independent gold standard in P. falciparum-endemic settings is increasingly necessary. Experimental transmission blocking prevention-of-transmission iRDTs remain of interest but have not warranted routine evaluation (Lin et al., 2022). These considerations focused the evaluation on selecting the latest generation of top-ranked mRDTs for the most common uncomplicated outpatient presentation, aiming to inform wider sub-Saharan-scale guidance on rapid-test use.

Optimizing first-7ee86c00-d09d-4d7b-a274-b2d4af593807e malaria test routine implementation cadences, especially testing type and frequency, can be of considerable public-health significance worldwide.

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