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SYSTEMATIC REVIEW: EVALUATION OF DIAGNOSTIC ACCURACY AND PRACTICAL USEFULNESS OF RAPID DIAGNOSTIC TESTS FOR MALARIA

Tinjauan Sistematis: Evaluasi Akurasi Diagnostik dan Kegunaan Praktis Tes Diagnostik Cepat untuk Malaria

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ABSTRAK

Malaria masih menjadi masalah kesehatan global yang signifikan, dengan 249 juta kasus dan 608.000 kematian dilaporkan pada tahun 2022. Deteksi dini, terutama di daerah endemis dan terbatas sumber daya, sangat penting untuk menghentikan penularan. Tes Diagnostik Cepat (RDT) banyak digunakan karena kecepatan dan kepraktisannya, tidak memerlukan infrastruktur laboratorium. Tinjauan ini bertujuan untuk mengevaluasi akurasi diagnostik (sensitivitas dan spesifisitas) RDT konvensional (HRP2, pLDH, kombinasi) dan RDT ultra-sensitif (uRDT), dan membandingkan efektivitasnya di berbagai populasi dan pengaturan klinis. Pencarian literatur dilakukan di PubMed, ScienceDirect, dan Cochrane Library (2020–2025), termasuk studi primer dalam bahasa Inggris atau Indonesia yang melibatkan kasus malaria yang dikonfirmasi. Sebanyak 30 studi disertakan berdasarkan pedoman PRISMA dengan populasi yaitu anak-anak, neonatus, dewasa, ibu hamil, pasien klinis, serta komunitas di daerah endemis maupun impor (total lebih dari 50.000 individu). Sebagian besar RDT menunjukkan spesifisitas tinggi (>90%), tetapi sensitivitas sangat bervariasi (0 hingga >95%), dipengaruhi oleh tingkat parasitemia dan karakteristik populasi. RDT berbasis HRP2 efektif untuk Plasmodium falciparum, tetapi delesi gen HRP2 menyebabkan hasil negatif palsu. RDT berbasis pLDH bekerja lebih baik pada infeksi multispesies. uRDT meningkatkan deteksi parasitemia berdensitas rendah, terutama pada wanita hamil dan asimtomatik. Namun, sensitivitas menurun pada neonatus dan orang dewasa dengan parasitemia rendah. Sementara beberapa RDT mengungguli mikroskopi dalam sensitivitas klinis, PCR menjadi standar emas, untuk mendeteksi infeksi subklinis. Sebagai kesimpulan, RDT memberikan deteksi cepat dan spesifik, terutama untuk infeksi P. falciparum berdensitas sedang hingga tinggi. Namun, pada kasus berdensitas rendah atau tanpa gejala, sensitivitasnya yang terbatas memerlukan pengujian konfirmasi menggunakan mikroskopi atau PCR.

Kata kunci: *akurasi diagnostik, malaria, rapid diagnostic test*

ABSTRACT

Malaria remains a significant global health concern, with 249 million cases and 608,000 deaths reported in 2022. Early detection, especially in endemic and resource-limited settings, is crucial to interrupt transmission. Rapid Diagnostic Tests (RDTs) are widely used due to their speed and practicality, requiring no laboratory infrastructure. This review aimed to evaluate the diagnostic accuracy (sensitivity and specificity) of conventional RDTs (HRP2, pLDH, combination) and ultra-sensitive RDTs (uRDTs), and compare their effectiveness across different populations and clinical settings. Literature searches were conducted in PubMed, ScienceDirect, and Cochrane (2020–2025), including studies in English or Indonesian. A total of 30 studies were included based on the PRISMA guidelines, with study populations encompassing children, neonates, adults, pregnant women, clinical patients, and community members in both endemic and imported settings (involving more than 50,000 individuals in total). Most RDTs demonstrated high specificity (>90%), but sensitivity varied widely (0 to >95%), influenced by parasitemia level and population characteristics. HRP2-based RDTs were effective for Plasmodium falciparum, but HRP2 gene deletions caused false negatives.

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pLDH-based RDTs performed better in multi-species infections. U-RDTs improved the detection of low-density parasitemia, especially in pregnant women and asymptomatic individuals. However, sensitivity declined in neonates and adults with low parasitemia. While some RDTs outperformed microscopy in clinical sensitivity, PCR remains the gold standard, especially for detecting subclinical infections. In conclusion, RDTs provide rapid and specific detection, particularly for moderate to high-density P. falciparum infections. However, in low-density or asymptomatic cases, their limited sensitivity necessitates confirmatory testing using microscopy or PCR.

***Keywords**: diagnostic accuracy, malaria, rapid diagnostic test*

INTRODUCTION

According to WHO data from 2022, malaria remains a highly impactful infectious disease globally, with 249 million cases and approximately 608,000 deaths. This disease poses a significant burden, particularly in sub-Saharan Africa and Southeast Asia, and carries a high risk of serious complications if not diagnosed and treated promptly and appropriately.[1] Rapid malaria diagnosis is crucial in preventing disease progression and breaking the chain of transmission, particularly in resource-limited endemic areas where laboratory facilities are not always available.[2]

The use of Rapid Diagnostic Tests (RDTs) has expanded globally due to their practical nature, requiring no complex laboratory infrastructure, and being able to provide results in less than 30 minutes. RDTs work by detecting specific antigens from Plasmodium in a patient's blood. Several types of RDTs are commonly used, namely Histidine-Rich Protein 2 (HRP2), Plasmodium Lactate Dehydrogenase (pLDH), combinations, and ultra-sensitive RDTs (uRDT). HRP2 specifically detects Plasmodium falciparum and is known to have high sensitivity, but is prone to producing false-positive results due to the presence of antigens that persist after the infection has resolved, as well as false-negative results due to genetic mutations such as the deletion of the hrp2 gene.[2,3] Meanwhile, pLDH can detect all malaria species and has the advantage that this antigen disappears quickly after therapy, but its sensitivity tends to be lower, especially in cases with low parasitemia.[4] Another type, namely Aldolase, is used as a pan–Plasmodium marker, but its sensitivity and specificity are generally lower than HRP2 and pLDH.[3] To improve accuracy, combination RDTs (HRP2+pLDH or HRP2+aldolase) have been developed that are capable of detecting more than one Plasmodium species, although their use requires higher costs and more complex result interpretation. The latest innovation, the ultra-sensitive RDT (uRDT), is even capable of detecting parasitemia down to very low levels (±10-40 parasites/µL), but its application is still constrained by cost and limited availability.[5]

The diagnostic accuracy of RDTs remains a significant challenge because their sensitivity and specificity vary significantly depending on the test type, Plasmodium species, patient clinical condition, and the comparator method used (microscopy or PCR). Therefore, a systematic review of the diagnostic accuracy of various malaria RDTs is crucial. This can serve as a basis for diagnostic policymaking at both the global and national levels. This study aimed to evaluate the diagnostic accuracy of malaria RDTs, compare the performance of conventional RDTs (HRP2, pLDH, and combinations) and ultra-sensitive RDTs (uRDTs) in various populations, and identify limitations to their use in cases with low or asymptomatic parasitemia.

METHODS

Data Search Strategy

This research is a systematic literature review, a research and development methodology used to evaluate research on a specific topic. The study was conducted from June 14 to July 14, 2025. The research process included determining a data collection strategy by selecting studies through quality assessments based on eligibility criteria and quality assessment instruments, data extraction, and data synthesis.

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The keywords used in this research search were “malaria” AND “Rapid Diagnostic Test” AND “Diagnostic Accuracy” OR “Sensitivity” OR “Specificity” OR “Positive Predictive Value” OR “Negative Predictive Value”.

Information Sources

The database sources used to search for literature in this study were PubMed, ScienceDirect, and the Cochrane Library.

Eligibility Criteria

The eligibility criteria in this study include inclusion and exclusion criteria. The inclusion criteria in this study are 1) The literature used is in the form of primary studies or scientific journals with a Randomized Controlled Trial design, 2) The literature sources come from Pubmed, ScienceDirect, and the Cochrane Library, 3) The literature has open access, 4) The literature can be accessed in full text, 5) The literature uses English or Indonesian, 6) The year of publication of scientific articles is in 2020-2025, 7) The discussion in the literature includes the use of Rapid Diagnostic Tests in individuals suspected or diagnosed with malaria as a diagnostic test tool. While the exclusion criteria in this study are Literature in the form of Reviews, Case Reports, and Studies without primary data. In addition, the researcher also limited the scope of the study. The researcher used the PICO method (Population, Intervention, Comparison, Outcomes), as in the table below:

Table 1. PICO Summary

Component	Information
Population	Individuals suspected or diagnosed with malaria, including individuals with symptoms (symptomatic) or individuals without symptoms (asymptomatic), in all age groups and all settings (hospitals, clinics, communities)
Intervention	Use of Rapid Diagnostic Tests, including conventional RDTs (HRP2-based, pLDH-based, or combo) and ultra-sensitive RDTs (uRDTs)
Comparison	Polymerase Chain Reaction/PCR, Microscopy, or other diagnostic modalities as reference standards.
Outcomes	Sensitivity, Specificity, positive predictive value (PPV), negative predictive value (NPV)

1. Quality Assessment

Literature was selected using the PRISMA (Preferred Reporting Items for Systematic Reviews) method. The PRISMA Flow Diagram in this study is attached in Figure 1. After using the PRISMA method, the results obtained were a total of 44 scientific journals, 8 of which were excluded due to data duplication, 4 did not meet the exclusion criteria, and 2 did not have full-text access. The final results of the study included 30 inclusion criteria, which will be used in the literature.

2. Synthesis Data

In this study, the data synthesis process was conducted by comparing scientific journals that met quality assessments and inclusion and exclusion criteria. This section refers to the research objective, which is to determine the accuracy of the Rapid Diagnostic Test as a diagnostic tool for detecting individuals suspected or diagnosed with malaria. High heterogeneity between studies due to differences in methods, populations, RDT types, and settings means that results between studies cannot be directly compared or combined quantitatively (meta-analysis), so the analysis is descriptive.

3. Data Extraction

*T*he data extraction results are attached in a table consisting of the author's name, year of publication, population, type of RDT used, and diagnostic accuracy (sensitivity value, specificity, and the author's objective assessment).[4]

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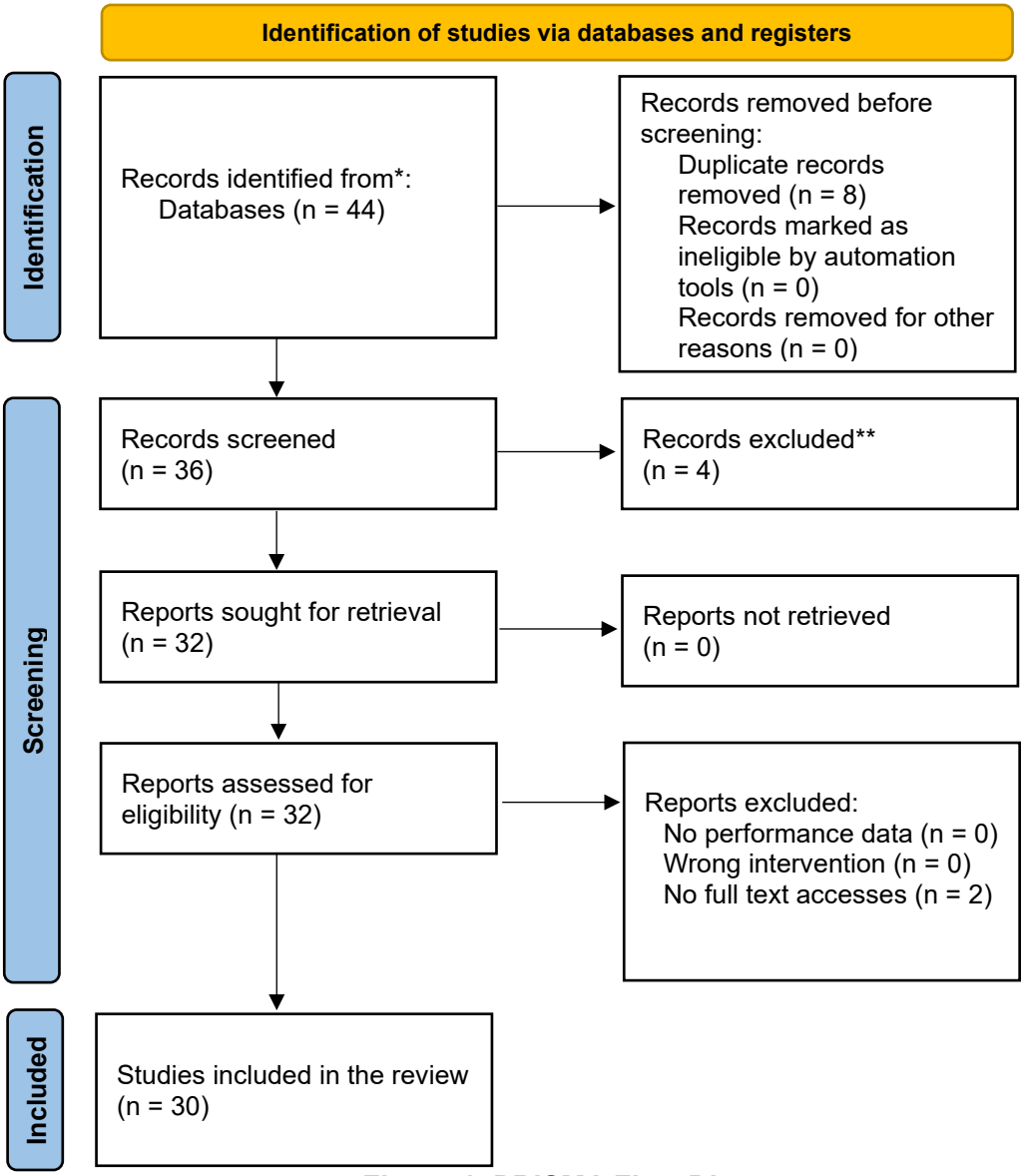

Figure 1. PRISMA Flow Diagram

RESULT

A total of 30 literature reviews were included in this study to evaluate the diagnostic accuracy and effectiveness of Rapid Diagnostic Tests (RDTs) in diagnosing malaria. The literature reviewed came from various countries, ranging from highly endemic areas such as Uganda, Ghana, Indonesia, and the Democratic Republic of the Congo, to non-endemic countries such as the United Kingdom and France. The study populations included neonates, children, pregnant women, and the general population with various clinical conditions, both symptomatic and asymptomatic.

The results of the literature reviewed in this study indicate that the specificity of RDTs is generally high, with most studies reporting values >90%, even reaching 100% in some populations. However, RDT sensitivity varies widely, depending on factors such as the type of RDT, the level of Plasmodium parasitemia, and the individual's clinical status.

In the pediatric population, as reported by several studies, the sensitivity of RDT reaches more than 93%.6, with a specificity above 90%7, making RDTs very useful for early diagnosis in primary health care facilities. In contrast, in neonates, a study by Adeniji showed RDT sensitivity of only 0%, indicating the ineffectiveness of RDTs

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for detecting malaria in this group, which is most likely due to low parasitemia, while the minimum limit of detection (LOD) of RDTs is generally in the range of 100–200 parasites/μL of blood.

Studies evaluating RDTs in the general population and asymptomatic adults, such as Benie et al showed very low sensitivity, only 17.8%, although specificity remained high.[3.8] This demonstrates the limitations of RDTs in detecting subclinical malaria infections or those with low parasitemia. In contrast, studies by Christian et al. and Yuniatun & Haryatmi showed sensitivity >90% and specificity of 100% in patients with moderate to high parasitemia.[3.8]

Several studies have also compared RDTs with other diagnostic methods. In Ghana, Afriyie et al.9found that RDTs were more sensitive than microscopy in detecting clinical malaria cases, but neither was capable of detecting submicroscopic infections that could only be identified by PCR. Parr found a similar finding in the DRC.10 Where RDT showed a sensitivity of 75% and a specificity of 92%, superior to microscopy.

Ultra-sensitive RDTs (uRDTs) have been reported to have higher sensitivity than conventional RDTs in populations such as pregnant women or in community screening, as shown in a study by Maketa and Kabalu.[11] However, other studies, such as Unwin et al.12showed that the sensitivity of uRDT remains low when used for the detection of low-density infections, and is not significantly different from conventional RDTs.

Table 2. Systematic Review of Sensitivity and Specificity of RDT Types Based on Populations in Various Countries

No	Writer	Country	Population	Types of RDT	Sensitivity	Specificity
1	Gorret et al., 2021.[13]	Uganda	250 Children with P. falciparum	HRP2/pLDH	92.7–95.2%	79.5–83.1%
2	Adeniji et al., 2024.[14]	Nigeria	131 Neonates	Combination	0%	100%
3	Opoku Afriyie et al., 2023.[15]	Ghana	1,040 Clinical Patients	HRP2	55.70%	98.20%
4	Aninagyei et al., 2024.[16]	Ghana	200 General samples	HRP2/pLDH	72.1–77.9%	98.3–100%
5	Benié et al., 2022.[17]	Ivory Coast	1,011 asymptomatic adults	HRP2	17.80%	100%
6	Bird et al., 2023.[6]	English	1,414 Children in the Emergency Room	HRP2/pLDH	93.6% (100% for Pf)	99.40%
7	Charpentier et al., 2020.[18]	French	331 imported malaria patients	HRP2/pLDH	86.30%	99.6–100%
8	Christian et al., 2025.[3]	Indonesia	317 General population	HRP2/pLDH	97.30%	87.30%
9	Yuniatun & Haryatmi, 2025.[8]	Indonesia	50 General population	HRP2/pLDH	93.50%	100%
10	Tshiongo et al., 2025.[19]	DRC	250 pregnant women	uRDT	88%	-
11	Maketa et al., 2022.[11]	DRC	242 pregnant women	uRDT	Higher than microscopy	-
12	Unwin et al., 2020.[12]	Indonesia	270 pregnant women	uRDT vs csRDT	Low	Tall

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No	Writer	Country	Population	Types of RDT	Sensitivity	Specificity
13	Irshad et al., 2024.[7]	Pakistan	140 Children	Combination	96.25%	90%
14	Ngalamie et al., 2025.[20]	Cameroon	260 General samples	HRP2/pLDH	45%	100%
15	Bosco et al., 2020.[21]	Uganda	359 Children (2–10 yrs)	HRP2	Low	Not mentioned
16	Feleke et al., 2021.[2]	Ethiopia	29,419 General population	Combination	High enough	Tall
17	Shankar et al., 2021.[22]	India	3,322 General population	Combination	49.90%	90.40%
18	Rogers et al., 2022.[23]	English	General population	HRP2/pLDH	High for Pf	Low for non-Pf
19	Kabbale et al., 2025.[4]	Uganda	6,354 Children and adolescents	HRP2/pLDH	91.7% (vs. microscopy), 91.6% (vs. qPCR)	56.7% (vs. microscopy), 64.0% (vs. qPCR)
20	Golden et al., 2024.[24]	Multinational (12 countries)	1,497 people Community population	HRP2/pLDH	94.4%	Very high
21	Kiemde et al., 2022.[25]	Burkina Faso	1,176 people General population	HRP2/pLDH	>95% (HRP2)	Height (pLDH)
22	Breton et al., 2025.[26]	Mali	1,164 people Community Population	HRP2	≥90% (literature)	≥95% (literature)
23	Parr et al., 2021.[10]	DRC	3,627 General population	HRP2	75%	92%
24	Shelus et al., 2022.[27]	Uganda	63 Rural communities	HRP2/pLDH	76.6%-86.9%	>90%
25	Slater et al., 2022.[28]	Multi-country	General population	HS-RDT vs conventional	56.10%	95%
26	Ali et al., 2021.[29]	Cameroon	Pregnant mother	HRP2/pLDH	91.67%	53.13%
27	Collins et al., 2024.[1]	Outpatients	1,126 General population	HRP2/pLDH	94%	91-94%
28	Mortazavi et al., 2025.[30]	Uganda	225 people aged 1-81 years	HRP2	66%	80%
29	Eyong et al., 2022.[31]	Cameroon	324 Population of Mount Cameroon	HRP2	99.7%	99.5%
30	Aidoo & Incardona, 2022.[32]	Sub-Saharan Africa	General population	Combination	HRP2>pLDH	tall

DISCUSSION

The results of this systematic review confirm that the Rapid Diagnostic Test (RDT) is a practical, rapid, and relatively accurate diagnostic tool, especially for detecting Plasmodium falciparum infections with moderate to high parasitemia. The high specificity in most studies indicates that the RDT is highly reliable for confirming positive malaria cases, thus supporting the "test and treat" policy at various levels of health care.

However, varying, and even very low, sensitivity in certain populations such as neonates, asymptomatic adults, or patients with low parasitemias suggests caution in interpreting negative RDT results. In a clinical context, a negative RDT result does

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not necessarily exclude the diagnosis of malaria, especially in patients with strong symptoms and epidemiological history.

It's important to note that there is significant heterogeneity in the results of various studies. This heterogeneity reflects differences in research settings, reference methods, populations studied, and cut-off values used to detect malaria. Heterogeneity can be biological (parasitemia level, immune status, gene mutations, species type) or technical (antigen type, kit quality, usage method, and storage conditions). These differences can influence the results and conclusions drawn from the studies.[14]

The setting or context in which research is conducted significantly influences RDT results. In endemic areas, RDTs are often used as the primary method due to their convenience and speed, while in non-endemic areas, RDTs are usually used as a complement to microscopy or PCR. In areas with limited access to diagnostic facilities, RDTs demonstrate superior sensitivity and specificity.

Furthermore, the reference method used in the research also plays a crucial role. Some studies use microscopy as a comparative method against RDTs, while others use PCR or a combination of both as a comparison.[2,9,15–17] These differences produce different results in terms of RDT sensitivity and specificity. Similarly, variations in the characteristics of the populations studied, such as children, asymptomatic adults, or neonates, have been reported. It has been suggested that children are more easily diagnosed with RDTs than neonates and asymptomatic adults, which can result in significant differences in RDT effectiveness.[7,17]

This difference occurs because children generally have higher parasitemia due to immature immune cells, making Plasmodium antigens more easily detected. Neonates tend to have very low parasitemia due to passive immunity from maternal antibodies, often below the detection limit of RDTs. In asymptomatic adults in endemic areas, partial immunity develops, suppressing parasitemia to submicroscopic levels, thus decreasing RDT sensitivity.[18]

One aspect of this study is the exploration of non-falciparum Plasmodium species. Although most research focuses on P. falciparum, as this species causes severe malaria, other species, such as P. malariae, P. ovale, and P. knowlesi, also have the potential to cause serious and sometimes recurrent infections.

*P. malariae and P. ovale*are often undetected by RDTs, primarily due to their low parasitemia and tendency to cause subclinical infections. The limitations of RDTs in detecting these species indicate the need to develop more sensitive RDTs for P. malariae and P. ovale, especially in asymptomatic patients with low parasitemia.

*Plasmodium vivax*tends to have a higher proportion of low-density infections than Plasmodium falciparum. In a study in a low-transmission area, 65.6% of microscopically detected P. vivax infections had densities <100/µL, while only 40% of P. falciparum infections were in this range.[35]

To improve malaria detection capabilities with RDTs, further research should develop RDTs that are more sensitive to non-falciparum Plasmodium, particularly P. malariae, P. ovale, and P. knowlesi. The addition of new detection targets in combination RDTs based on HRP2 + pLDH or HRP2 + aldolase could help improve sensitivity to non-falciparum Plasmodium species. Furthermore, the use of PCR or LAMP as complementary methods can also improve diagnostic accuracy, particularly for detecting infections with low parasitemia or asymptomatic infections.[36]

Comparisons between RDTs, microscopy, and PCR in several studies have shown that PCR remains the reference standard with the highest sensitivity, especially in detecting latent or submicroscopic infections. Microscopy still plays an important role, particularly for species identification and parasite density assessment.

This systematic review has the advantage of combining results from recent studies (2020–2025) covering diverse populations in both endemic and non-endemic areas, and comprehensively comparing the performance of

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conventional RDTs and ultra-sensitive RDTs (uRDTs). However, this study also has limitations, including high heterogeneity between studies, differences in reference methods used, inconsistent sample sizes, and limited data for non-falciparum species, which makes the conclusions descriptive in nature without quantitative meta-analysis. These findings imply that clinically, a negative RDT result cannot completely exclude the diagnosis of malaria in high-risk populations, requiring confirmation by microscopy or PCR. From a policy perspective, RDTs remain a valuable tool for a “test and treat” strategy in primary healthcare, but the development of RDTs with higher sensitivity for non-falciparum Plasmodium should be a priority in further research to close the existing diagnostic gap.

CONCLUSION

Rapid Diagnostic Test(RDT) is a rapid, practical, and specific diagnostic tool for detecting malaria, especially Plasmodium falciparum. HRP2-based RDTs have high sensitivity, but are susceptible to false-positive results after infection and false-negative results due to deletion of the pfhrp2 gene. pLDH-based RDTs are more accurate in detecting active and multiparasite infections, although their sensitivity decreases in cases with low parasitemia. Innovations in ultra-sensitive RDTs (uRDTs) improve the ability to detect low parasitemia, especially in pregnant women and asymptomatic cases, but their availability is still limited and less than optimal for neonates and non-falciparum species.

Compared with microscopy and PCR, RDTs excel in speed and ease of use, but their limited sensitivity in subclinical infections underscores the need for confirmation using reference methods. Therefore, RDTs remain an important tool in malaria test-and-treat strategies, but their use must be contextualized and supported by further research to improve the effectiveness of multi-species detection.

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