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Review Article

Molecular Point-of-Care Diagnostics for Vector-Borne Pathogens: Biotechnological Innovations and Challenges in Field Implementation

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Abstract

Background: Centralized diagnostics can delay detection of vector-borne pathogens. Molecular point-of-care (mPOC) technologies may address this gap, but performance and field readiness remain uncertain.

Methods: This rapid systematic review followed Cochrane Diagnostic Test Accuracy guidance. PubMed, Scopus and Web of Science (Clarivate) were searched for 2020–2025 publications (13 September 2025). Two-by-two data were extracted or reconstructed, and sensitivity and specificity were recalculated with 95% Wilson confidence intervals. Studies were classified by setting, assessed using the real-time connectivity, ease of specimen collection, affordable, sensitive, specific, user-friendly, rapid and robust, equipment-free and deliverable to end-users (REASSURED) criteria, and appraised with Quality Assessment of Diagnostic Accuracy Studies–2. Heterogeneity led to narrative synthesis.

Results: 1,964 records yielded 22 studies: 11 malaria, 6 leishmaniasis and 5 arboviral infections. Twenty-one studies provided two-by-two data, yielding 26 non-overlapping analyses. One study reported sensitivity only (86.4%, 95% confidence interval 75.5–93.0%) and one analysis was not reconstructable. Median sample size was 100 (interquartile range 76.5–161.5; range 13–490). Thirteen analyses reported 100% sensitivity or specificity, but 9 had lower confidence limits below 90%. Four studies evaluated point-of-care tests. Based on lower confidence limits, 8 met sensitivity and 5 met specificity REASSURED thresholds; 6 met real-time connectivity and 7 met ease of specimen collection. Seventeen (77.3%) reported no cost data.

Conclusions: mPOC technologies are promising, but evidence is dominated by laboratory or near-laboratory studies with imprecise estimates. Multicenter field evaluations, standardized reporting including costs and complete two-by-two data are needed before integration.

Keywords: Rapid diagnostics; Diagnostic accuracy; Isothermal amplification; Vector-borne pathogens

Introduction

Vector-borne pathogens, a diverse range of bacterial, parasitic and viral agents transmitted by arthropod vectors, account for a substantial proportion of the global burden of infectious diseases and continue to pose a major threat to the sustainability of public health systems and health economies. The extensive geographical distribution of the infections they cause, together with their considerable human and economic burden, underscores that early diagnosis and rapid response are not merely desirable but funda mental prerequisites for their effective control (1–5).

Despite this critical need, conventional diagnostic methods, although still regarded as the gold standard, remain structurally distant from the point of care in many operational settings. Their dependence on centralized la-

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boratory infrastructure, skilled personnel, prolonged turnaround times and complex supply chains limits their accessibility and effectiveness. These constraints are particularly pronounced in resource-limited settings, which often bear the highest burden of vector-borne pathogens. Consequently, delays in life-saving treatment and reduced effectiveness of disease control programs may occur, further exacerbating existing health inequities (6–8).

In this context, molecular point-of-care (mPOC) diagnostics have emerged as a transformative biotechnological approach aimed at bringing diagnostic capabilities closer to patients and empowering frontline healthcare providers. By enabling rapid, simple and accurate testing, these technologies offer significant potential to bridge the gap between reference laboratory capacity and urgent field-based diagnostic needs. When aligned with essential performance criteria, including diagnostic accuracy, ease of use, rapid turnaround, environmental robustness and affordability, mPOC platforms may also satisfy the evolving requirements of real-time connectivity, ease of specimen collection, affordable, sensitive, specific, user-friendly, rapid and robust, equipment-free and deliverable to end-users framework (the REASSURED) (9–20).

Accordingly, the objective of this rapid review is to provide an up-to-date and critical synthesis of the available evidence regarding the diagnostic accuracy and operational performance of mPOC platforms for vector-borne pathogens, and to characterize the distance that remains between laboratory validation and genuine field deployment. Through a rapid systematic review approach, this study seeks to assess the current state of the field, identify critical evidence gaps and delineate the requirements necessary for the successful translation of these technologies from controlled laboratory environments to complex field settings (21).

Study objectives

The primary objective of this rapid review is to provide a structured synthesis, based on Cochrane rapid review guidance (53), of biotechnological innovations in mPOC diagnostics for vector-borne pathogens. With a particular emphasis on field applicability, implementation challenges and bench-to-field translation, this review aims to present a comprehensive overview of the current status of mPOC technologies, including isothermal nucleic acid amplification techniques (for example, loop-mediated isothermal amplification (LAMP), recombinase polymerase amplification (RPA)), clustered regularly interspaced short palindromic repeats (CRISPR)-based diagnostics and integrated microfluidic platforms, in the control of vector-borne pathogens.

The specific objectives are to assess the diagnostic accuracy of mPOC technologies for vector-borne pathogens, including malaria, leishmaniasis and arboviral infections against established reference standards, using extracted 2×2 contingency data and standardized confidence intervals to evaluate, at study level, the operational performance of these technologies against the nine criteria of the REASSURED framework (21) to characterize the evaluation settings in which the available evidence was generated, and thereby to distinguish laboratory validation from genuine field evaluation and to identify the methodological and reporting deficiencies of the current evidence base that constrain the translation of mPOC technologies from laboratory settings to field deployment.

Research questions

The research questions are: Based on evidence published between 2020 and 2025, what is the diagnostic accuracy of mPOC technologies for the detection of vector-borne pathogens, including malaria, leishmaniasis and arboviral infections, when compared with reference laboratory assays and how precise are the resulting estimates? To what extent do mPOC technologies for vector-borne pathogens satisfy the operational criteria of the REASSURED framework when assessed at study level and which domains demonstrate the greatest limitations?

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In what settings was the available evidence generated and to what extent does it support claims of readiness for deployment at the point of care? What methodological limitations, risks of bias and reporting deficiencies in the current evidence base hinder the bench-to-field translation of mPOC technologies?

Methodology

This study was conducted as a rapid systematic review to analyze technological innovations and evaluate the field applicability of molecular point-of-care (mPOC) diagnostics for vector-borne pathogens. The review followed current Cochrane guidance for the conduct of rapid reviews (53) and Cochrane guidance for diagnostic test accuracy (DTA) reviews, with a primary focus on diagnostic accuracy, operational performance and bench-to-field translation. Reporting follows the preferred reporting items for systematic reviews and meta-analyses of diagnostic test accuracy studies (PRISMA-DTA) (22).

A protocol specifying the research questions, eligibility criteria, search sources and data extraction framework was defined internally before the search was executed. The review was not prospectively registered in the international prospective register of systematic reviews (PROSPERO) or any other public registry. This is acknowledged as a limitation in the discussion. The internal protocol is available from the corresponding author on request.

Search strategy

A systematic literature search was performed on 13 September 2025 across three bibliographic databases: PubMed, Scopus and Web of Science (Clarivate). The complete search strategy for each database, including the full Boolean strings as executed, MeSH and field tags, the platform used, the applied publication-date limit (2020–2025), the date on which each search was run and the number of records retrieved from each source, is provided in Supplementary File S1. The reference lists of included studies and of relevant review articles were also screened. The search was not updated after 13 September 2025. Publications appearing after this date are cited in the discussion for contextual purposes only and did not contribute to study selection or to the evidence synthesis.

Eligibility criteria

Eligibility criteria were defined according to the population, intervention, comparison and outcome (PICO) framework. The population (P) comprised individuals with suspected vector-borne disease, including malaria, leishmaniasis, dengue fever and related infections, particularly in resource-limited settings. The index test (I) comprised mPOC technologies, including field-deployable PCR platforms, isothermal amplification assays such as LAMP, RPA and CRISPR-Cas-based assays. The comparator (C) was an established reference standard, including laboratory-based polymerase chain reaction (PCR), quantitative polymerase chain reaction (qPCR), nested PCR, microscopy or a composite reference standard. Outcomes (O) comprised diagnostic accuracy measures, including sensitivity, specificity and measures of agreement, together with any reported operational performance data relevant to the REASSURED criteria.

Studies were eligible if they evaluated a molecular point-of-care assay on human clinical specimens against a reference standard, irrespective of whether testing was performed in a field setting, in a laboratory using specimens collected in the field or in a laboratory using archived clinical specimens. Purely analytical studies were excluded, that is, studies reporting only analytical sensitivity or limit of detection using spiked, cultured or synthetic material without human clinical specimens, as were in silico and modelling studies, reviews, conference abstracts and non-peer-reviewed reports.

Because this review addresses bench-to-field translation, studies conducted under controlled laboratory conditions were deliberately retained rather than excluded, excluding them would remove one side of the comparison the

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review is designed to make. The evaluation setting of each study was instead recorded as a study-level variable (Table 1) and used to qualify the interpretation of the findings.

Evaluation setting

Each included study was classified into one of four categories according to where the index test was performed. Direct field or primary healthcare evaluation denotes testing at or near the point of care, in rural health facilities, primary care clinics, or antenatal clinics. Hybrid evaluation denotes parallel testing in a reference laboratory and a field facility or a study combining a field component with a laboratory component. Laboratory evaluation of prospectively collected clinical specimens denotes testing in a central laboratory using specimens collected prospectively from patients in endemic settings. Laboratory evaluation of archived material denotes testing under controlled laboratory conditions using biobanked specimens or previously extracted nucleic acid.

Study selection and data extraction

A total of 1,964 records were identified: 1,946 from the database searches (PubMed 596, Scopus 691, Web of Science 659) and a further 18 through screening the reference lists of included studies and relevant reviews. All records were imported into EndNote for reference management. After removal of 923 duplicate records, 1,041 records were screened by title and abstract against the eligibility criteria, and 1,018 records were excluded at this stage. The most frequent reason for exclusion was a focus on infections that are not vector-borne, with COVID-19 (87 records), SARS (29 records) and Ebola (14 records) accounting for 130 exclusions. Non-peer-reviewed publications and conference presentations were also excluded at this stage. Twenty-three reports were sought for full-text retrieval. One report could not be obtained and the full texts of the remaining 22 reports were independently assessed for eligibility by two reviewers. All 22 studies met the eligibility criteria and were included in the qualitative synthesis. Disagreements at any stage were resolved through discussion or by consultation with a third reviewer.

For each included study, the numbers of true positives, false positives, false negatives and true negatives were extracted from the published report, or reconstructed where the reported data determined them uniquely. Sensitivity and specificity were recalculated from the resulting 2×2 tables and are reported with 95% confidence intervals computed by the Wilson score method, irrespective of the interval method used in the original publication. Where a study reported more than one index test, specimen preparation, or evaluation setting, each was extracted as a separate analysis; subgroup analyses drawn from the same participants as a parent analysis are identified as such and were not counted as independent. Where a 2×2 table could not be recovered from the published report, the analysis was reported as not reconstructable and was excluded from the accuracy synthesis. Data on study characteristics, assay type, reference standard, evaluation setting and REASSURED indicators were extracted in parallel.

Real-time connectivity, ease of specimen collection, affordable, sensitive, specific, user-friendly, rapid and robust, equipment- free and deliverable to end-user assessment

Each included study was assessed at study level against the nine REASSURED criteria using a four-level scale (Met, Partially met, Not met, Not reported). Sensitivity and specificity were rated on the lower limit of the 95% Wilson confidence interval rather than the point estimate, so that estimates derived from small samples are not credited with precision they do not have: a criterion was rated Met where the lower limit was 90% or above, Partially met from 75% to below 90%, and Not met below 75%. These thresholds represent an operationalization adopted for this review and are not specified in the published framework. Ease of specimen collection was rated Met for non-invasive or minimally in-

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vasive specimens (urine, saliva, lesion swab or scraping, tape disc, dried blood spot, capillary blood), Partially met for venous whole blood, serum, plasma, or buffy coat and Not met where tissue biopsy was required. Real-time connectivity was rated Met only where a result could be transmitted digitally without manual transcription. Affordability was rated only where a cost figure was reported in the source publication; no cost was imputed. Where a study contributed more than one analysis, the criterion was rated on the weakest analysis relevant to intended field use.

Synthesis and quality assessment

No quantitative meta-analysis was undertaken. Pooling was precluded by heterogeneity along several distinct dimensions: clinical heterogeneity in target pathogen, parasite or viral load, patient population (symptomatic, asymptomatic, antenatal) and specimen matrix, and methodological heterogeneity in assay chemistry (isothermal amplification, clustered regularly interspaced short palindromic repeats (CRISPR)-based detection, cartridge PCR), nucleic acid extraction procedure, evaluation setting and above all reference standard, which ranged from Giemsa microscopy to qPCR, photo-induced electron transfer-polymerase chain reaction (PET-PCR) and composite standards. Subgroup analysis by pathogen group was considered but not performed, because the resulting strata would have contained too few studies with comparable reference standards to support stable pooled estimates. Findings were therefore synthesized narratively and are presented by pathogen group. Methodological quality and applicability were assessed using the quality assessment of diagnostic accuracy studies 2 (QUADAS-2) tool (54), across four risk-of-bias domains (patient selection, index test, reference standard and flow and timing) and three applicability domains, with each domain rated as low, high, or unclear risk based on the signalling questions. Judgments were made independently by two reviewers, and the rationale for each judgment is reported in Table 3.

Results

Study selection

The systematic search of PubMed, Scopus and Web of Science yielded 1,946 records, and a further 18 were identified through reference-list screening, giving 1,964 records in total. Following removal of 923 duplicates, 1,041 records were screened by title and abstract, of which 1,018 were excluded. Twenty-three reports were sought for full-text retrieval; one could not be obtained and the full texts of the remaining 22 were assessed for eligibility. All 22 studies were eligible and were included in the final qualitative synthesis (23–44). The selection process is summarized in the preferred reporting items for a systematic review and meta-analysis of diagnostic test accuracy studies (PRISMA-DTA) flow diagram (Fig. 1).

Study characteristics and evaluation settings

The included studies encompassed a broad spectrum of vector-borne pathogens. Eleven studies focused on malaria diagnostics, primarily targeting *Plasmodium falciparum* and *P. vivax*, with one study specifically addressing *P. malariae*. Six studies evaluated leishmaniasis diagnostics, including visceral, cutaneous and post-kala-azar forms, while five studies investigated arboviral infections, including dengue fever, chikungunya, Zika virus disease, West Nile virus infection and tick-borne encephalitis. From a technological perspective, isothermal amplification methods, predominantly LAMP and RPA, represented the most commonly investigated platforms, although CRISPR-based assays, portable PCR systems and automated cartridge-based platforms were also identified (23–44).

Considerable methodological heterogeneity was observed with respect to design, evaluation setting, specimen matrix and reference standard. Regarding evaluation settings, 4 studies were direct field evaluations (24, 30, 32, 42) and 3 were hybrid evaluations combining field and reference laboratory assess-

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ment (26, 29, 33), the remaining 15 studies were conducted in a laboratory setting, of which 10 tested clinical specimens collected prospectively from patients in endemic settings (23, 25, 27, 28, 31, 35, 37, 39, 41, 44) and 5 tested archived or biobanked material (34, 36, 38, 40, 43). Biological specimens included whole blood, dried blood spots, serum, urine, cutaneous lesion material and buffy coat. Reference standards comprised qPCR, nested PCR, PET-PCR, microscopy and composite diagnostic standards (Table 1).

Across the 22 studies, a complete 2×2 table was recovered for 21, yielding 26 non-overlapping analyses. One study reported sensitivity only, since no reference-standard-negative specimens were tested (34) and one analysis reported a sensitivity estimate for which no contingency table could be recovered from the published report (23). The median sample size was 100 (IQR 76.5–161.5, range 13–490), eight studies enrolled fewer than 100 participants and four fewer than 50. Extracted contingency data and recalculated accuracy estimates for all analyses are presented in Table 2.

Diagnostic accuracy of molecular point-of-care technologies

Diagnostic performance varied according to the target pathogen and is summarized below; all estimates are accompanied by 95 % Wilson confidence intervals in Tables 1 and 2.

Malaria

The clinical sensitivity of LAMP-based assays for malaria detection ranged from 68.6% to 100%, while specificity varied between 65.4% and 100%. The highest sensitivity values were reported in studies using archived specimens or conducted under near-laboratory conditions (40), in the largest such study, sensitivity was 98.9% (94.0–99.8%), although the corresponding specificity estimate rested on only ten reference-standard-negative samples and was correspondingly imprecise (100%, 72.2–100%). The lowest sensitivity was observed during field screening of afebrile pregnant women (30: 68.6%, 61.3–75.1%). Reduced specificity in some studies (30, 33) was largely attributable to the use of microscopy or lower-sensitivity PCR assays as reference standards, resulting in the identification of additional positive cases by LAMP, particularly submicroscopic infections, in one field trial, 46 of 133 microscopy-negative specimens were positive by cLAMP, yielding an apparent specificity of 65.4% (57.0–73.0%) (33). The analytical limit of detection varied substantially across studies (36, 44), and clinical performance declined markedly in infections with very low parasite densities (<100 parasites/μL) (24, 30). The CRISPR- and RPA-based platforms also demonstrated high sensitivity (24, 41). However, clinical evaluations were generally based on small sample sizes: in one field validation, a sensitivity of 95.2% rested on 21 reference-standard-positive specimens and a specificity of 87.5% on eight controls (24). A fully automated platform (34) and a confirmatory dbPCR-based algorithm (42) achieved sensitivities of 96% to 100% and substantially improved specificity by reducing false-positive results associated with persistent HRP2 antigenemia. Overall, parasite density and the nucleic acid extraction method strongly influenced the diagnostic sensitivity of molecular assays for malaria (36, 44) (Table 1).

Leishmaniasis

Diagnostic accuracy in leishmaniasis was highly dependent on parasite species and primer design. Studies employing optimized primer sets reported high diagnostic sensitivity (29, 39). However, the commercial Loopamp kit demonstrated poor performance for the detection of *L. aethiopica*, with a sensitivity of 48.4% (36.6–60.4%), primarily attributed to primer mismatch with the species-specific 18S rRNA sequence (32). More recently developed platforms, including smartphone-assisted RPA systems (38) and portable miniPCR devices (35), showed promising diagnostic performance, although in the latter both sensitivity and specificity of 100% were esti-

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mated from 24 positive and 24 negative specimens and carry lower confidence limits of 86.2%. Across all studies, nucleic acid extraction remained an essential prerequisite, and simplified extraction approaches, such as LAB buffer-based methods, were evaluated only to a limited extent (29, 35) (Table 1).

Arboviruses

Isothermal amplification technologies, particularly reverse transcription LAMP (RT-LAMP) and CRISPR-based platforms, demonstrated strong performance for arboviral diagnostics. For chikungunya virus, a lyophilized extraction-free RT-LAMP assay achieved 100% sensitivity (91.0–100%) and 96.7% specificity (88.8–99.1%) in a cohort of 100 clinical samples (26). A comparable approach yielded 100 % sensitivity (83.9–100%) and 93.8% specificity (86.2–97.3%) for Zika virus detection (27). In contrast, the performance of RT-LAMP for West Nile virus was highly dependent on the platform and sample preparation method: when applied directly to raw urine using a portable device, sensitivity declined to 65.0% (49.5–77.9%), whereas testing of extracted RNA on the same device achieved 100% sensitivity (91.2–100%) (25). A CRISPR/Cas13a-based platform for tick-borne encephalitis virus demonstrated sensitivity and specificity of 100% in a preliminary evaluation of 25 samples (37), the corresponding confidence limits, 79.6–100% and 72.2–100%, illustrate the limited precision of such small evaluations, and the reported limit of detection was higher than that of the reference RT-qPCR assay. The fully automated Standard M10 cartridge platform for dengue fever, evaluated in the largest study population included in this review, achieved an overall sensitivity of 95.0% (92.0–96.9%) and specificity of 100% (97.8–100%), reduced sensitivity was observed for the DENV-4 serotype (82.4%, 72.9–89.0%) (43) (Table 1).

Methodological quality

Risk of bias and applicability were assessed with QUADAS-2 (54). Domain-level judgments and the rationale for each are presented in Table 3, in which every study is identified by first author, year, and reference number so that each judgment is traceable to a named included study. Concerns were most frequent in the patient selection domain, reflecting the use of convenience and case-control sampling and in the flow and timing domain, where the interval between index test and reference standard was often not reported.

Compliance with real-time connectivity, ease of specimen collection, affordable, sensitive, specific, user-friendly, rapid and robust, equipment-free or simple and deliverable to end-users operational criteria

Results of the study-level REASSURED assessment are presented in Table 4. Applying the rating rules defined in the methods, sensitivity was rated Met in 8 of the 22 studies and specificity in 5, because rating was based on the lower confidence limit; several assays reporting point estimates of 100% were rated Partially met or Not met on account of imprecision. Nearly all platforms performed favourably with respect to turnaround time, typically under 60 minutes where separate nucleic acid extraction was not required. User-friendliness has improved in newer-generation systems through visual colorimetric readouts and smartphone-based applications (38, 40, 44). The Equipment-free criterion was rarely achieved, as all technologies required at least a basic heat source. Real-time connectivity was rated Met in only 6 studies, in which the result could be transmitted digitally without manual transcription. Ease of specimen collection was rated Met in 7 studies using non-invasive or minimally invasive specimens, 13 required venous blood, serum, or buffy coat and 2 required tissue biopsy. Affordability could be rated in only 5 studies: 17 of the 22 studies (77.3%) reported no cost or affordability data of any kind. Deliverability to end-users largely remained at the conceptual stage, with

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only a limited number of studies evaluating performance under real-world conditions involving community health workers (29, 33).

Barriers to bench-to-field translation

Multiple operational barriers were identified that hinder the successful deployment of mPOC technologies in resource-limited settings (44).

Nucleic acid extraction remains the most significant operational challenge. Conventional DNA and RNA extraction methods often require laboratory infrastructure, specialized equipment and cold-chain logistics, particularly when column-based protocols are used (29, 36). Although extraction-free approaches demonstrated promising results for certain viral infections (26, 27), substantial reductions in sensitivity were observed when applied to parasitic diseases and to unprocessed specimens (25, 36).

Sample and reagent stability represents another major challenge (40). DNA degradation during sample storage and transportation (33), together with cold-chain requirements for certain reagents (38), creates logistical constraints. Lyophilized and room-temperature-stable formulations (26) represent a promising advancement, although their implementation remains limited.

User training and inter-operator variability continue to affect performance (38). Even simplified platforms require operator training for accurate interpretation of faint test bands (24) and reliable execution of manual procedural steps (29).

Cost and access to diagnostic kits also remain important barriers (27). Shortages of RPA reagents following the COVID-19 pandemic (24) and the potentially high cost of cartridge-based systems (43) threaten long-term supply-chain sustainability; the near-absence of cost reporting across the evidence base compounds this uncertainty.

Pathogen-specific performance variability further limits widespread implementation. Failure to detect mixed infections (23), unequal sensitivity across viral serotypes (43) and species-specific performance deficiencies (32) indicate that broadly applicable primer designs and universally robust diagnostic performance have yet to be achieved.

Evidence gaps and emerging trends

The available evidence demonstrates a clear predominance of malaria-focused research within the mPOC diagnostic landscape, with 11 of the 22 included studies addressing malaria. In contrast, empirical evidence for viral hemorrhagic fevers, leishmaniasis and tick-borne diseases remains comparatively sparse and fragmented. Systematic economic evaluations were almost absent: 17 of 22 studies reported no cost data and none reported a formal cost-effectiveness analysis. Evidence on social acceptability, long-term usability and sustained adoption was likewise scarce.

Clinical sample sizes were small in many studies, particularly those evaluating CRISPR- and RPA-based technologies, and independent multicenter validation was uncommon. The consequence is visible in the accuracy estimates themselves: of the 13 analyses reporting a point estimate of 100% for sensitivity or specificity, 9 have a lower 95% confidence limit below 90%. Despite substantial technological advances, no platform simultaneously fulfilled all REASSURED criteria; equipment-free nucleic acid extraction and real-time connectivity remain largely unmet objectives.

Emerging trends include the progressive integration of artificial intelligence and mobile technologies for result interpretation, the development of fully integrated sample-to-answer cartridge systems, and the implementation of confirmatory diagnostic algorithms to enhance specificity. Nevertheless, the field readiness and large-scale operational maturity of these innovations remain at an early stage of development.

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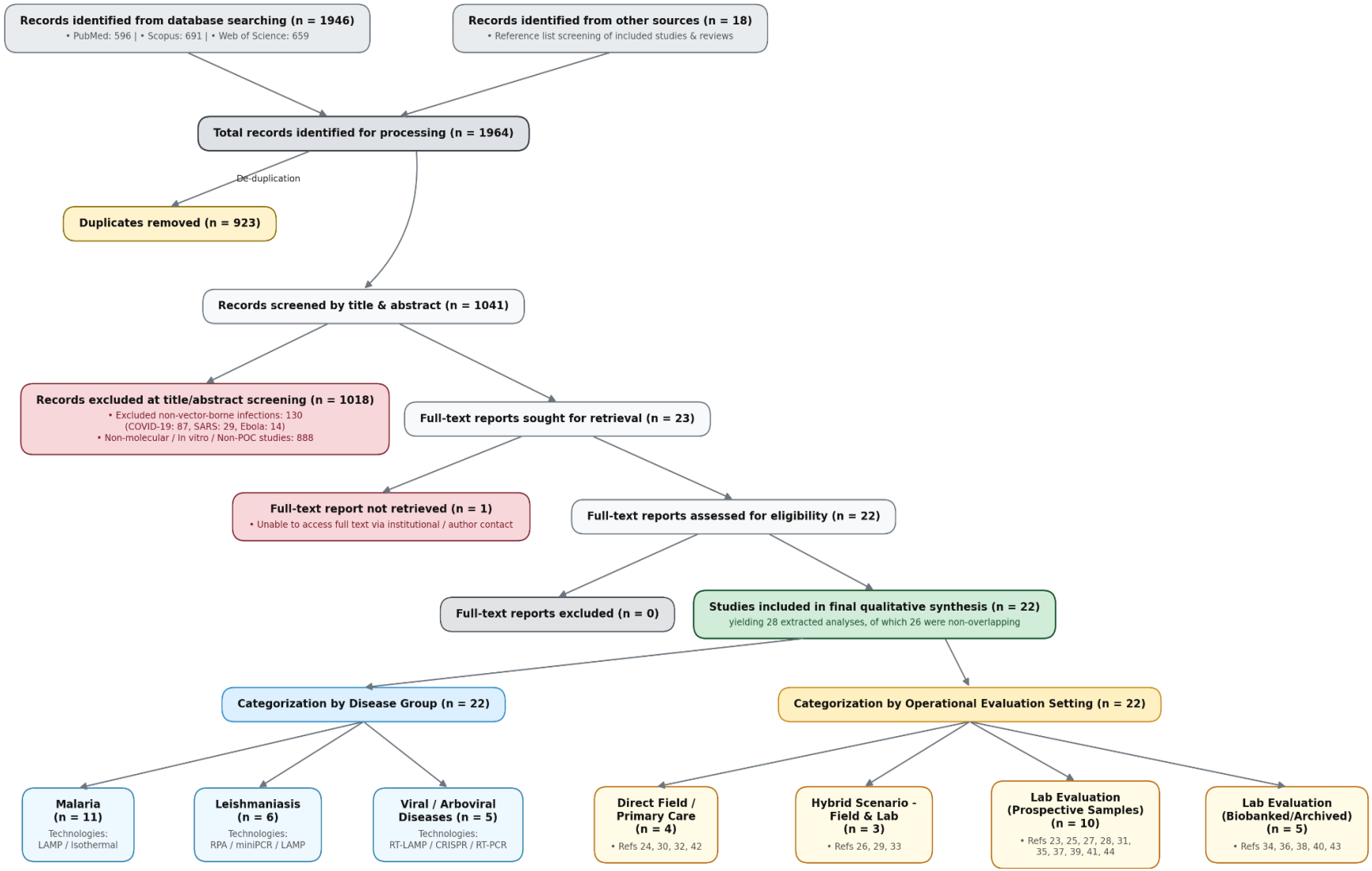


Fig. 1. Preferred reporting items for systematic reviews and meta-analyses of diagnostic test accuracy (PRISMA-DTA) flow diagram of study selection, showing identification from database searches and reference-list screening, de-duplication, screening with reasons for exclusion, full-text assessment, and stratification of the included studies by pathogen group and by operational evaluation setting

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Table 1. Characteristics and diagnostic performance of the 22 included studies of molecular point-of-care assays for vector-borne pathogens. Sensitivity and specificity are recalculated from the extracted 2×2 data and presented with 95% Wilson score confidence intervals. The evaluation setting classifies each study according to where the index test was performed

First author (Year) [Ref]	Disease or pathogen	Technology platform	Sn (%)	Sn 95% Wilson CI	Sp (%)	Sp 95% Wilson CI	Sample type	Reference standard	Evaluation setting	Notes
Zhang (2024) [37]	Tick-borne encephalitis virus (TBEV)	RT-RAA-CRISPR/Cas13a-LFD	100.0%	79.6%–100.0%	100.0%	72.2%–100.0%	Serum	RT-qPCR	Lab (prospective)	Fully reported clinical validation on 15 cases and 10 healthy controls.
Puri (2021) [39]	*L. donovani* (VL and PKDL)	Portable Fluorimetric LAMP	100.0%	92.9%–100.0%	98.0%	89.5%–99.6%	Blood and Dermal Biopsy	ITS-1 PCR	Lab (prospective)	Evaluated on 50 cases and 50 controls (1 FP in leprosy group).
da Silva (2024) [26]	Chikungunya virus (CHIKV)	One-step RT-LAMP	100.0%	91.0%–100.0%	96.7%	88.8%–99.1%	Serum	RT-qPCR	Hybrid	Validated on 100 clinical serum samples (39 positive, 61 negative).
Khedhiri (2024) [25]	West Nile Virus (WNV)	RT-LAMP / qcLAMP	65.0%–100.0%	49.5%–100.0%	94.1%–100.0%	80.9%–100.0%	Urine (Extracted RNA and Crude)	qRT-PCR	Lab (prospective)	Thermal cycler: Sens 90% (extracted) / 86.7% (crude); Spec 100% (extracted) / 96.9% (crude). Pebble-R device: Sens 100% (extracted) / 65% (crude); Spec 100% (extracted) / 94.1% (crude).
Assefa (2024) [24]	*Plasmodium malariae*	RPA-LF	95.2%	77.3%–99.2%	87.5%	52.9%–97.8%	Dried blood spots (DBS)	18S rRNA qPCR	Field	Field validation on 21 Pm+ field samples and 8 non-Pm controls.
Barazorda (2020) [23]	Malaria (*P. vivax* / *P. falciparum*)	LAMP (MG-LAMP)	100.0%	96.4%–100.0%	100.0%	89.3%–100.0%	Whole blood	18S rRNA nested-PCR	Lab (prospective)	MG-LAMP colorimetric assay (104/104 positive). RealAmp real-time fluorescence assay reported 92% sensitivity (unrecovered 2x2 table).
Colbert (2021) [44]	*Plasmodium falciparum*	PD-LAMP (Smartphone)	88.9%	56.5%–98.0%	100.0%	51.0%–100.0%	Whole blood	qPCR	Lab (prospective)	Asymptomatic community cohort (N=13; 8 TP, 1 FN, 4 TN).
Cossio (2021) [29]	Cutaneous Leishmaniasis (CL)	RPA-LF	74.7%–86.7%	64.4%–92.4%	85.7%–88.6%	70.6%–95.5%	Lesion swab / Whatman FTA	Composite Gold Standard	Hybrid	Ref Lab scenario: Sens 86.7% (77.8-92.4%), Spec 85.7% (70.6-93.7%). Field scenario: Sens 74.7% (64.4-82.8%), Spec 88.6%

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Table 1. Continued ...

										(74.0-95.5%).
Febrer-Sendra (2023) [33]	Malaria (*Plasmodium* spp.)	field-cLAMP (Colorimetric)	98.5%	92.0%–99.7%	65.4%	57.0%–73.0%	Venous whole blood	Giemsa Microscopy	Hybrid	Field trial in Cubal, Angola (N=200; 66 TP, 46 FP, 1 FN, 87 TN).
Hin (2021) [34]	Acute Febrile Illness Panel	FeverDisk (LAMP)	86.4%	75.5%–93.0%	NR	NR	Serum and Whole blood	Real-Time PCR	Lab (archived)	Proof-of-concept feasibility study across targets (51 TP, 8 FN, N=59). Specificity not estimable.
Kobialka (2024) [38]	*L. donovani* (VL and PKDL)	minoo RPA (Smartphone device)	88.1%	80.4%–93.1%	91.3%	82.3%–96.0%	Whole blood and Skin biopsy	Real-Time qPCR	Lab (archived)	Tested in India and Bangladesh (N=170; 89 TP, 6 FP, 12 FN, 63 TN).
Nguyen (2024) [31]	*Plasmodium falciparum*	cLAMP (A-type 18S rRNA)	100.0%	80.6%–100.0%	100.0%	94.7%–100.0%	Dried blood spots (DBS)	Microscopy / Nested PCR	Lab (prospective)	Clinical isolates from Malawi (N=84; 16 TP, 0 FP, 0 FN, 68 TN).
Nolasco (2021) [36]	*P. vivax* and *P. falciparum*	Colorimetric LAMP (PvCOX1 and Pfr364)	84.5%	77.3%–89.7%	97.3%	93.8%–98.8%	Whole blood	Nested PCR / qPCR	Lab (archived)	Dual-target screening cohort (N=312; 109 TP, 5 FP, 20 FN, 178 TN). Subgroup microscopic cases achieved 100% sensitivity.
Ruang-Areerate (2021) [28]	*Leishmania* spp.	Biotin-LAMP probe	94.1%	73.0%–99.0%	97.1%	89.9%–99.2%	Buffy coat	Nested PCR	Lab (prospective)	Evaluated on N=85 samples (16 TP, 2 FP, 1 FN, 66 TN). Note: Colorimetric closed-tube row in paper printed sum=84 due to minor typo.
Some (2025) [42]	*Plasmodium falciparum*	Sequential RDT + dbPCR-NALFIA	97.8%	95.2%–99.0%	94.6%	90.1%–97.2%	Whole blood	Expert Microscopy / qPCR	Field	Field trial in Burkina Faso (N=438; 264 TP, 9 FP, 6 FN, 159 TN).
Taye (2024) [32]	*Leishmania aethiopica* (CL)	Loopamp Leishmania Kit	48.4%	36.6%–60.4%	87.9%	77.1%–94.0%	Tape disc (Stratum corneum)	Skin-slit Microscopy	Field	Evaluated in Ethiopia (N=122; 31 TP, 7 FP, 33 FN, 51 TN). Reduced sensitivity due toprimer mismatch with *L. aethiopica*.
Santoso (2025) [43]	Dengue virus (DENV 1-4)	Standard M10 RT-PCR system	95.0%	92.0%–96.9%	100.0%	97.8%–100.0%	Serum	CDC DENV RT-PCR	Lab (archived)	Multi-center validation in Indonesia (N=490; 304 TP, 0 FP, 16 FN, 170 TN). Serotype DENV-4 subgroup sensitivity was

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Table 1. Continued ...

										82.4% (70/85; 72.9-89.0%).
da Silva (2021) [27]	Zika virus (ZIKV)	One-step RT-LAMP	100.0%	83.9%–100.0%	93.8%	86.2%–97.3%	Serum	RT-qPCR	Lab (prospective)	Clinical validation in Brazil (N=100; 20 TP, 5 FP, 0 FN, 75 TN).
Puri (2022) [40]	*Plasmodium falciparum*	Portable LED/photodiode LAMP	98.9%	94.0%–99.8%	100.0%	72.2%–100.0%	Whole blood	Microscopy / PCR	Lab (archived)	Evaluated in India (N=100; 89 TP, 0 FP, 1 FN, 10 TN).
Bel Hadj Ali (2024) [41]	*Leishmania major* (CL)	m22-RPA-LF	100.0%	90.8%–100.0%	100.0%	92.6%–100.0%	Lesion scraping / swab	Direct Exam + ITS1 PCR-RFLP	Lab (prospective)	Evaluated in Tunisia and Morocco (N=86; 38 TP, 0 FP, 0 FN, 48 TN); published in 2024 (Front Microbiol Vol 15).
Samuels (2022) [30]	*Plasmodium falciparum* (ANC)	Illumigene Malaria LAMP	68.6%	61.3%–75.1%	82.3%	77.6%–86.1%	Whole blood	PET-PCR	Field	Antenatal care screening in Kenya (N=482; 118 TP, 55 FP, 54 FN, 255 TN).
Castellanos-Gonzalez (2023) [35]	American Cutaneous Leishmaniasis	miniPCR-F (LAB buffer + P51 viewer)	100.0%	86.2%–100.0%	100.0%	86.2%–100.0%	Whatman FTA filter paper	Real-Time PCR	Lab (prospective)	Field samples from Peru (N=48; 24 TP, 0 FP, 0 FN, 24 TN).

Table 2. Extracted 2×2 contingency data and recalculated diagnostic accuracy for all 28 analyses across the 22 included studies. Sensitivity and specificity are presented with 95% Wilson score confidence intervals. Analysis status designates complete 2×2 reconstructions, subgroup analyses drawn from the same participants, sensitivity-only reporting, and unrecoverable tables

Ref	Study/analysis	Index test	Reference standard	Specimen	N	TP	FP	FN	TN	Sn (%)	Sn 95% CI	Sp (%)	Sp 95% CI	Analysis status
37	Zhang (2024)	RT-RAA-CRISPR/Cas 13a-LFD	RT-qPCR	Serum	25	15	0.0	0	10.0	100.0	79.6–100.0	100.0	72.2–100.0	Complete 2x2
39	Puri (2021)	Portable Fluorimetric LAMP	ITS-1 PCR	Blood and Dermal Biopsy	100	50	1.0	0	49.0	100.0	92.9–100.0	98.0	89.5–99.6	Complete 2x2
26	da Silva (2024)	One-step RT-LAMP	RT-qPCR	Serum	100	39	2.0	0	59.0	100.0	91.0–100.0	96.7	88.8–99.1	Complete 2x2
25	Khedhiri (Thermal Cycler, Extracted RNA)	RT-LAMP (Thermal Cycler)	qRT-PCR	Urine (RNA extract)	62	27	0.0	3	32.0	90.0	74.4–96.5	100.0	89.3–100.0	Complete 2x2
25	Khedhiri (Thermal Cycler, Crude Urine)	RT-LAMP (Thermal Cycler)	qRT-PCR	Urine (Crude 1:1 PBS)	62	26	1.0	4	31.0	86.7	70.3–94.7	96.9	84.3–99.4	Complete 2x2

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Table 2. Continued ...

25	Khedhiri (Pebble-R, Extracted RNA)	qcLAMP (Pebble-R portable)	qRT-PCR	Urine (RNA extract)	74	40	0.0	0	34.0	100.0	91.2–100.0	100.0	89.8–100.0	Complete 2x2
25	Khedhiri (Pebble-R, Crude Urine)	qcLAMP (Pebble-R portable)	qRT-PCR	Urine (Crude 1:1 PBS)	74	26	2.0	14	32.0	65.0	49.5–77.9	94.1	80.9–98.4	Complete 2x2
24	Assefa (2024)	RPA-LF	18S rRNA qPCR	Dried blood spots (DBS)	29	20	1.0	1	7.0	95.2	77.3–99.2	87.5	52.9–97.8	Complete 2x2
23	Barazorda (2020) - MG-LAMP Genus	MG-LAMP (Colorimetric)	18S rRNA nested-PCR	Whole blood	136	104	0.0	0	32.0	100.0	96.4–100.0	100.0	89.3–100.0	Complete 2x2
44	Colbert (2021)	PD-LAMP (Smartphone)	qPCR	Whole blood	13	8	0.0	1	4.0	88.9	56.5–98.0	100.0	51.0–100.0	Complete 2x2
29	Cossio (2021) - Ref Lab Scenario	RPA-LF (Ref Lab)	Composite (Smear+Culture+Histopathology+qPCR)	Lesion swab / Whatman FTA	118	72	5.0	11	30.0	86.7	77.8–92.4	85.7	70.6–93.7	Complete 2x2
29	Cossio (2021) - Field Scenario	RPA-LF (Field Health Facility)	Composite (Smear+Culture+Histopathology+qPCR)	Lesion swab / Whatman FTA	118	62	4.0	21	31.0	74.7	64.4–82.8	88.6	74.0–95.5	Complete 2x2
33	Febrer-Sendra (2023)	field-cLAMP (Colorimetric)	Giemsa Microscopy	Venous whole blood	200	66	46.0	1	87.0	98.5	92.0–99.7	65.4	57.0–73.0	Complete 2x2
34	Hin (2021)	FeverDisk (LAMP)	Real-Time PCR	Serum and Whole Blood	59	51	NR	8	NR	86.4	75.5–93.0	NR	NR	Sensitivity only
38	Kobialka (2024)	minoo RPA (Smartphone device)	Real-Time qPCR	Whole blood and Skin biopsy	170	89	6.0	12	63.0	88.1	80.4–93.1	91.3	82.3–96.0	Complete 2x2
31	Nguyen (2024)	cLAMP (A-type 18S rRNA)	Microscopy / Nested PCR	Dried blood spots (DBS)	84	16	0.0	0	68.0	100.0	80.6–100.0	100.0	94.7–100.0	Complete 2x2
36	Nolasco (2021) - Dual Target Cohort	PvCOX1 and Pfr364 LAMP	Nested PCR	Whole blood	312	109	5.0	20	178.0	84.5	77.3–89.7	97.3	93.8–98.8	Complete 2x2
28	Ruang-Areerate (2021) - AuNP Probe	AuNP-LAMP probe	Nested PCR	Buffy coat	85	16	2.0	1	66.0	94.1	73.0–99.0	97.1	89.9–99.2	Complete 2x2
42	Some (2025)	Sequential RDT + dbPCR-NALFIA	Expert Microscopy / qPCR	Whole blood	438	264	9.0	6	159.0	97.8	95.2–99.0	94.6	90.1–97.2	Complete 2x2
32	Taye (2024)	Loopamp	Skin-slit	Tape disc (Stratum	122	31	7.0	33	51.0	48.4	36.6–	87.9	77.1–94.0	Complet

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Table 2. Continued ...

		Leishmania Kit	Microscopy	corneum)							60.4			e 2x2
43	Santoso (2025) - Overall	Standard M10 RT-PCR system	CDC DENV RT-PCR	Serum	490	304	0.0	16	170.0	95.0	92.0–96.9	100.0	97.8–100.0	Complete 2x2
43	Santoso (2025) - DENV-4 Subgroup	Standard M10 RT-PCR system	CDC DENV RT-PCR	Serum	85	70	NR	15	NR	82.4	72.9–89.0	NR	NR	Subgroup (Sn only)
27	da Silva (2021)	One-step RT-LAMP	RT-qPCR	Serum	100	20	5.0	0	75.0	100.0	83.9–100.0	93.8	86.2–97.3	Complete 2x2
40	Puri (2022)	Portable LED/photodiode LAMP	Microscopy / PCR	Whole blood	100	89	0.0	1	10.0	98.9	94.0–99.8	100.0	72.2–100.0	Complete 2x2
41	Bel Hadj Ali (2024)	m22-RPA-LF	Direct Exam + ITS1 PCR-RFLP	Lesion scraping / swab	86	38	0.0	0	48.0	100.0	90.8–100.0	100.0	92.6–100.0	Complete 2x2
30	Samuels (2022) - Illumigene LAMP	Illumigene Malaria LAMP	PET-PCR	Whole blood	482	118	55.0	54	255.0	68.6	61.3–75.1	82.3	77.6–86.1	Complete 2x2
35	Castellanos-Gonzalez (2023)	miniPCR-F (LAB buffer + P51 viewer)	Real-Time PCR	Whatman FTA filter paper	48	24	0.0	0	24.0	100.0	86.2–100.0	100.0	86.2–100.0	Complete 2x2
23	Barazorda (2020) - RealAmp	RealAmp (Real-Time Fluorescence)	18S rRNA nested-PCR	Whole blood	NR	NR	NR	NR	NR	92.0	NR–NR	100.0	NR–NR	Not Reconstructable

Table 3. Methodological quality and risk of bias of the included studies assessed with quality and applicability were assessed using the quality assessment of diagnostic accuracy studies 2 (QUADAS-2), identified by first author, publication year and reference number, with domain-specific rationales

First author (Year) [Ref]	RoB: Patient selection	RoB: Index test	RoB: Reference standard	RoB: Flow and timing	Applicability: Patient selection	Applicability: Index test	Applicability: Reference standard	Summary of key QUADAS-2 concerns
Puri (2021) [39]	High	Low	Low	Low	High	Low	Low	Convenience sampling; possible selection bias in febrile patients
Zhang (2024) [37]	High	Low	Low	Low	High	Low	Low	Non-random patient enrollment; limited to symptomatic cases
da Silva (2024) [26]	Low	Low	Low	Low	Low	Low	Low	Well-defined prospective cohort; minimal concerns
Khedhiri (2024)	High	High	Low	Low	Low	Low	Low	Index test not pre-specified; threshold

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Table 3. Continued ...

[25]								issues
Assefa (2024) [24]	High	Unclear	Low	Low	High	Low	Low	Unclear index test execution; possible verification bias
Barazorda (2020) [23]	Low	Low	Unclear	Low	Low	Low	Low	Reference standard not fully described for all samples
Colbert (2021) [44]	High	Low	Unclear	Low	High	Low	Low	Inadequate reference standard details; high-risk population
Cossio (2021) [29]	Low	High	Unclear	High	Low	Low	Low	Index test interpretation not blinded; delays in testing
Febrer-Sendra (2023) [33]	Unclear	Unclear	Unclear	Low	Low	Low	Low	Insufficient reporting on recruitment and test procedures
Hin (2021) [34]	High	High	Low	High	High	Low	Low	Multiple protocol deviations; unclear test reproducibility
Kobialka (2024) [38]	Unclear	Unclear	Low	Low	Low	Low	Low	Incomplete demographic and test execution data
Nolasco (2021) [36]	Low	Low	Low	Low	Low	Low	Low	Robust design; no significant bias detected
Some (2025) [42]	High	Unclear	Low	Low	High	Low	Low	Selection based on prior RDT results; unclear index cut-off
Taye (2024) [32]	Low	Unclear	Unclear	Low	Low	Low	Low	Reference and index test details partially missing
Ruang-Areerate (2021) [28]	Low	Low	Low	High	Low	Low	Low	Delays between tests; potential verification bias
Santoso (2025) [43]	Unclear	Low	High	Low	Low	Low	Low	Reference standard had known limitations in field setting
da Silva (2021) [27]	High	Low	Low	Low	Low	Low	Low	Case-control design; possible spectrum bias
Puri (2022) [40]	High	Low	Low	Low	Low	Low	Low	Restricted to hospital-based patients; limited generalizability
Bel Hadj Ali (2024) [41]	High	High	Low	High	High	Low	Low	Index test not validated against gold standard uniformly
Samuels (2022) [30]	Low	Low	Low	Low	Low	Low	Low	Well-conducted; low risk across all domains
Castellanos-Gonzalez (2023) [35]	Low	Low	Low	Low	Low	Low	Low	Minimal methodological concerns; high applicability
Nguyen (2024) [31]	High	High	Low	Low	Low	Low	Low	Non-randomized selection; index test reproducibility not formally assessed; visual reading may introduce subjectivity

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Table 4. Study-level evaluation against the nine real-time connectivity, ease of specimen collection, affordable, sensitive, specific, user-friendly, rapid and robust, equipment-free and deliverable to end-users (REASSURED) criteria using a four-tier scale (Met, Partially met, Not met, Not reported). Sensitivity and specificity are rated on the lower limit of the 95% Wilson confidence interval (Met at 90% or above, Partially met from 75% to below 90%, Not met below 75%). NR indicates that the source publication reported no data for that criterion

Study	Specimen	Sn LCL	Sp LCL	1 Real-time connectivity	2 Ease of specimen collection	3 Affordable	4 Sensitive	5 Specific	6 User-friendly	7 Rapid and robust	8 Equipment-free or simple	9 Deliverable
Zhang (2024)	Serum	79.6	72.2	Not met	Partially met	NR	Partially met	Not met	Partially met	Met	Partially met	Partially met
Puri (2021)	Blood, dermal punch biopsy	92.9	89.5	Not met	Not met	NR	Met	Partially met	Partially met	Met	Met	Met
da Silva (2024)	Serum	91.0	88.8	Not met	Partially met	NR	Met	Partially met	Met	Met	Met	Met
Khedhiri (2024)	Urine (crude and extracted)	49.5	80.9	Met	Met	NR	Not met	Partially met	Met	Met	Met	Met
Assefa (2024)	Dried blood spots (DBS)	77.3	52.9	Not met	Met	$2.50-$3.50 per test	Partially met	Not met	Met	Met	Met	Met
Barazorda (2020)	Whole blood	96.4	89.3	Not met	Partially met	NR	Met	Partially met	Met	Met	Met	Met
Colbert (2021)	Whole blood	56.5	51	Met	Partially met	NR	Not met	Not met	Met	Met	Met	Met
Cossio (2021)	Lesion swab / Whatman FTA	64.4	74	Not met	Met	$0.03 (Boil and Spin) / $2.70 (column extraction)	Not met	Not met	Met	Met	Met	Met
Febrer-Sendra (2023)	Venous whole blood	92.0	57	Not met	Partially met	NR	Met	Not met	Met	Met	Met	Met
Hin (2021)	Serum and whole blood	75.5	NR	Met	Partially met	NR	Partially met	NR	Met	Met	Met	Met
Kobialka (2024)	Whole blood and skin biopsy	80.4	82.3	Met	Not met	NR	Partially met	Partially met	Met	Met	Met	Met
Nguyen (2024)	Dried blood spots (DBS)	80.6	94.7	Not met	Met	NR	Partially met	Met	Met	Met	Met	Met
Nolasco (2021)	Whole blood	77.3	93.8	Not met	Partially met	$0.03 (Boil and Spin)	Partially met	Met	Met	Met	Met	Met
Ruang-Areerate (2021)	Buffy coat	73.0	89.9	Not met	Partially met	NR	Not met	Partially met	Met	Partially met	Met	Partially met

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Table 4. Continued ...

Some (2025)	Whole blood	95.2	90.1	Not met	Partially met	NR	Met	Met	Partially met	Partially met	Partially met	Met
Taye (2024)	Tape disc (Stratum corneum)	36.6	77.1	Not met	Met	<$1.00-$5.30 per test	Not met	Partially met	Met	Met	Met	Partially met
Santoso (2025)	Serum	92.0	97.8	Met	Partially met	NR	Met	Met	Met	Met	Met	Met
da Silva (2021)	Serum	83.9	86.2	Not met	Partially met	NR	Partially met	Partially met	Met	Met	Met	Met
Puri (2022)	Whole blood	94.0	72.2	Not met	Partially met	NR	Met	Not met	Met	Met	Met	Met
Bel Hadj Ali (2024)	Lesion scraping / swab	90.8	92.6	Not met	Met	NR	Met	Met	Met	Met	Met	Met
Samuels (2022)	Whole blood	61.3	77.6	Not met	Partially met	NR	Not met	Partially met	Met	Met	Met	Met
Castellanos-Gonzalez (2023)	Whatman FTA filter paper	86.2	86.2	Met	Met	$0.01 per extraction (LAB buffer)	Partially met	Partially met	Met	Met	Met	Met

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Discussion

Our findings highlight the gap between laboratory innovations and the practical, field-based implementation of molecular point-of-care (mPOC) diagnostic technologies for vector-borne pathogens. A significant portion of the existing literature focused on malaria, aligning with prior evidence indicating that the development of point-of-care diagnostic tools has largely centered on this disease (30, 45). Although diagnostic accuracy in the reviewed studies was generally high, substantial heterogeneity was observed regarding assay platforms, nucleic acid extraction methods, detection techniques, sample types and evaluation conditions. This finding is consistent with the systematic review and meta-analysis by Feleke et al. (46), which demonstrated significant heterogeneity in the diagnostic performance of malaria detection methods, including LAMP and PCR. Notably, our review complements these observations by extending the scope of analysis beyond mere diagnostic accuracy to consider the operational characteristics and environmental conditions under which mPOC technologies were evaluated.

The high diagnostic accuracy reported in several studies must be interpreted with statistical precision in mind. Of the 26 analyses conducted, 13 reported point estimates of 100 % for sensitivity or specificity; however, in nine of these instances, the lower bound of the 95% confidence interval was below 90%, and in several cases, even below 75%. Consequently, these estimates should not be construed as evidence of flawless diagnostic performance. The median sample size was only 100 participants, and the small number of samples confirmed as positive or negative via the reference method (gold standard) resulted in wide confidence intervals across several studies. This distinction is particularly important when comparing our findings with previous reports that emphasize high point estimates but fail to adequately highlight the associated uncertainty (margin of error).

The reference standard was another significant source of variation in observed diagnostic performance. In studies using microscopy as the comparator, molecular assays could detect infections missed by microscopy. In a field evaluation, cLAMP detected 46 infections among 133 microscopy-negative samples, yielding a specificity of 65.4% (33). Rather than necessarily indicating poor specificity, this finding may reflect the higher analytical sensitivity of the molecular test for submicroscopic infections. In contrast, a study by Samuels et al. (30), which employed the more sensitive PET-PCR as the reference standard, reported significantly lower sensitivity for LAMP, particularly among pregnant women with predominantly low-density infections. These conflicting findings demonstrate that diagnostic accuracy estimates derived from different reference standards cannot be considered directly interchangeable. Similarly, composite reference standards used in several leishmaniasis and arbovirus studies (29, 41, 43) introduce additional interpretational complexity and potential incorporation bias.

Operational evidence further highlights the distinction between analytical performance and true point-of-care readiness. Numerous studies have reported on portable devices featuring rapid turnaround times, simple readouts or smartphone-assisted interpretation. However, significant challenges remain regarding sample preparation, nucleic acid extraction, equipment requirements, reagent stability, cost- effectiveness and user training. For instance, an evaluation of RT-LAMP for West Nile virus demonstrated that performance could decline when testing was conducted directly on raw, unextracted samples compared to extracted ones, underscoring the continued importance of sample preparation in field implementation (25). This observation aligns with broader literature emphasizing that point- of-care molecular diagnostics must account not only for an-

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alytical performance but also for technical complexity, infrastructure and workflow requirements (17, 55). Thus, our findings indicate that device portability alone should not be equated with operational readiness.

The REASSURED assessment reinforces this distinction. Only six studies met the predefined criterion for connectivity, and seven met the criterion for ease of sample collection, while 17 out of 22 studies (77.3%) reported no cost data. These shortcomings are significant because cost-effectiveness, connectivity, usability and suitability for local settings are integral components of point-of-care diagnostic implementation, rather than secondary considerations. The REASSURED framework specifically emphasizes these dimensions when evaluating the contribution of diagnostics to disease control and health system strengthening (47). More broadly, previous reviews have identified infrastructure, financing, training and health system integration as persistent barriers to the implementation of point-of-care diagnostics in resource-limited settings (20, 55, 56). Our review indicates that these implementation dimensions have been inadequately reported, even in recent evaluations of molecular technologies.

The underlying evidence also suffers from significant methodological limitations. Factors such as convenience or case-control sampling, incomplete reporting of blinding, uncertainty regarding the timing of index and reference tests, and variations in reference standards may have introduced bias, as reflected in the QUADAS-2 assessment. The review was not prospectively registered, and no formal framework for assessing the certainty of evidence, such as the grading of recommendations, assessment, development and evaluations (GRADE) approach, was applied. Furthermore, heterogeneity across pathogens, patient populations, samples, assay platforms and reference standards precluded quantitative pooling of data. Consequently, the present review offers a structured synthesis of existing evidence rather than a pooled estimate or a definitive comparison of technologies. Therefore, potential implications for disease control should be interpreted with caution. Portable molecular platforms could facilitate early diagnosis in settings with limited centralized laboratory infrastructure, and digital connectivity might support the integration of diagnostic data into surveillance systems (45, 47). However, these represent potential implementation benefits rather than outcomes demonstrated by the studies included in this review; none of the studies directly measured the impact on outbreak response, healthcare delivery, or population-level disease outcomes. Recent advancements, including portable PCR, LAMP-based assays and CRISPR-based molecular diagnostics, highlight the rapid technological evolution in this field (46, 48), yet their actual contribution to the control of vector-borne pathogens remains to be determined. Overall, evidence indicates that mPOC technologies can achieve high diagnostic performance in selected settings; however, the magnitude and reliability of that performance depend heavily on the reference standard, sample characteristics, sample processing requirements and evaluation settings. Therefore, future research should prioritize prospective, multicenter field evaluations involving representative populations, while reporting standardized 2×2 diagnostic data, confidence intervals, operational performance, usability and costs. Such evidence is essential to determine whether promising laboratory performance can translate into sustainable point-of-care diagnostic services.

Conclusion

Point-of-care molecular technologies hold promise for the diagnosis of vector-borne pathogens, but the available evidence remains limited due to small study volumes, inaccurate estimates of accuracy, and the predominance of laboratory assessments. Only four of the 22 studies reviewed evaluated testing directly at

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the point of care, while cost and other performance indicators were often not reported. Wider implementation requires prospective multicenter field validation, complete and standardized reporting of diagnostic performance, and evaluation of cost-effectiveness and operational feasibility. Until such evidence is available, the potential contribution of these technologies to routine surveillance and disease control should be interpreted with caution.

Policy and implementation recommendations

The gap between analytical performance and implementation readiness described above has direct implications for policy. Technological maturity cannot be judged on the basis of laboratory outcomes alone; it also depends on infrastructural preparedness, logistical capacity and the sustainable availability of essential consumables. Reliable electricity supply, consistent access to reagents and microfluidic chips, and resilient supply chains are therefore critical prerequisites for successful bench-to-field translation, and the recommendations set out below are organized around these requirements.

Support for research and development

Given the demonstrated potential of isothermal amplification technologies (particularly LAMP and RPA) and CRISPR-based diagnostic platforms, sustained investment in the development and optimization of locally adapted, field-resilient diagnostic systems is warranted. Support for research initiatives and technology development programmes should prioritize process simplification, particularly approaches that reduce dependence on complex nucleic acid extraction procedures.

Within this context, antimicrobial resistance (AMR) should be considered with appropriate conceptual caution. The studies included in this review did not evaluate AMR-related outcomes; AMR should therefore not be interpreted as an empirically demonstrated finding of the present review. Nevertheless, from a policy and implementation perspective, diagnostic technologies capable of shortening the time to diagnosis and facilitating treatment decisions at the point of care may contribute indirectly to more rational therapeutic use and the reduction of unnecessary empirical treatment. Antimicrobial resistance is accordingly presented as an implementation and research consideration rather than a direct outcome of this synthesis (52).

AI-assisted interpretation and smartphone-based decision support are promising developments that may make mPOC technologies more user-friendly, but firm evidence of their effectiveness in routine operational settings is still lacking.

Expansion of comprehensive field evaluations

Government agencies and relevant stakeholders should expand pilot programmes and comparative evaluation studies of mPOC technologies in high-risk settings, including malaria- and dengue-endemic regions. Such evaluations should be conducted through multicenter, longitudinal study designs to assess technology performance across diverse climatic, infrastructural and epidemiological conditions. Economic evaluations, including cost-effectiveness analyses, should be incorporated as an integral component of these field studies; the finding that 17 of 22 studies reported no cost data indicates how far current practice is from this standard.

Strengthening supporting infrastructure

Successful deployment of mPOC technologies requires the establishment of essential infrastructure prior to large-scale implementation. Key requirements include reliable electricity supplies or high-capacity battery sys-

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tems, secure access to consumables such as reagents and diagnostic cartridges through sustainable supply chains, and the development of user-friendly software solutions for on-site data analysis and interpretation. Training and capacity-building programmes for healthcare personnel and laboratory technicians should be recognized as critical components of implementation infrastructure.

Intersectoral collaboration and evidence-informed policymaking

Expert committees within ministries of health, regulatory agencies, and research organizations should use comprehensive evidence assessments, including systematic reviews and rapid reviews, to develop procurement and implementation frameworks for emerging diagnostic technologies. Incorporating the REASSURED framework into decision-making processes can assist policymakers in balancing analytical performance with operational feasibility.

Strengthening evidence dissemination and reporting standards

In alignment with the principles of evidence-informed policymaking, the findings of rapid reviews and related systematic reviews should be disseminated through formal reporting mechanisms and integrated into health system governance structures. Adherence to established reporting standards, including PRISMA-DTA flow diagrams and the complete reporting of 2×2 contingency data, would materially improve the usability of primary diagnostic accuracy studies: in the present review, one analysis had to be excluded from synthesis solely because its contingency table could not be recovered from the published report (50, 51).

Ethical considerations

Not applicable to this rapid review.

Conflict of interest statement

The authors declare that there is no conflict of interest.

Declaration of generative AI and AI-assisted technologies in the writing process

During the preparation of this rapid review, the authors used Grammarly and ChatGPT Plus to improve the clarity, readability, grammar and overall language of the manuscript. In addition, Elicit, NotebookLM and Semantic Scholar were used to assist with literature exploration, evidence organization, and data extraction from eligible studies. All outputs generated by these tools were carefully verified against the sources, critically reviewed by the authors, and revised where necessary. The authors retained full responsibility for the study design, literature selection, data interpretation, conclusions and the final content of the manuscript.

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