



Malaria Rapid Diagnostic Test Performance

Results of WHO product testing of
malaria RDTs: Round 1 (2008)

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Dr. John Barnwell	US Centers for Disease Control and Prevention / National Center for Infectious Diseases
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Dr. Qin Cheng	Army Malaria Institute, Australia
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Dr. Jane Cunningham	TDR, Special Programme for Research and Training in Tropical Diseases
Linda Dantes	WHO – Regional Office of the Western Pacific
Djibrine Djalle	Institut Pasteur Bangui, Central African Republic
Dionicia Gamboa	Universidad Peruana Cayetano Heredia Instituto de Medicina Tropical, Peru
Dr. Michelle Gatton	Queensland Institute of Medical Research Malaria Drug Resistance and Chemotherapy
Dr. Iveth Gonzalez	FIND, Switzerland
Beatrice Gordis	FIND, Switzerland
Sandra Incardona	Consultant, FIND
Jean Joly	TDR, Special Programme for Research and Training in Tropical Diseases
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Myat Phone Kyaw	Department of Medical Research, Myanmar
Jenny Luchavez	Research Institute of Tropical Medicine, Philippines
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Dr. James McCarthy	Queensland Institute of Medical Research University of Queensland
Didier Menard	Institut Pasteur de Madagascar, Madagascar
Claribel Murillo	Centro Internacional de Entrenamiento e Investigaciones Médicas (CIDEIM), Colombia
Bernhards Ogutu	Kenya Medical Research Institute (KEMRI), Kenya
Wellington Oyibo	University of Lagos
Dr. Rosanna Peeling	TDR, Special Programme for Research and Training in Tropical Diseases
Anita Pelecanos	Queensland Institute of Medical Research
Dr. Mark Perkins	FIND, Switzerland
Dr. Charuni Senanayake	WHO – Regional Office for the Western Pacific
Michael Valentine	US Centers for Disease Control and Prevention / National Center for Infectious Diseases
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ABBREVIATIONS

ACT	Artemisinin-based combination therapy
AMI	Army Malaria Institute
AusAID	Australian Agency for International Development
CDC	US Centers for Disease Control and Prevention
CLIA	Clinical Laboratory Improvement Amendments
FIND	Foundation for Innovative New Diagnostics
HRP2	Histidine-rich protein 2
HTD	Hospital for Tropical Diseases
pLDH	Plasmodium lactate dehydrogenase
p/μL	Parasites per microlitre
QA	Quality Assurance
QC	Quality Control
QMS	Quality Management Systems
RDT	Rapid Diagnostic Test (for the purposes of this report, this refers to immunochromatographic lateral flow devices for the detection of malaria parasite antigens)
SOP	Standard Operating Procedure
TDR	Special Programme for Research and Training in Tropical Diseases sponsored by UNICEF, UNDP, World Bank and WHO
USAID	United States Agency for International Development
WPRO	Western Pacific Regional Office
WHO	World Health Organization

1. EXECUTIVE SUMMARY

1.1. INTRODUCTION

The World Health Organization estimates that 3.3 billion persons were at risk of acquiring malaria in 2006, with 247 million of these developing clinical malaria (86% in Africa), and nearly 1 million (mostly African children) dying from the disease. Malaria remains endemic in 109 countries, and while parasite-based diagnosis is increasing, most suspected cases of malaria are still not properly identified, with accurate diagnosis and disease monitoring consequently remaining elusive (1).

WHO recommends that malaria case management be based on parasite-based diagnosis in all cases, with the exception of young children in areas of high transmission and where lack of resources or need for urgent response temporarily limits its application. The use of antigen-detecting rapid diagnostic tests (RDTs) forms a vital part of this strategy, providing the possibility of parasite-based diagnosis in areas where good quality microscopy can not be maintained. The number of RDTs available, and the scale of their use, has rapidly increased over the past few years. However, limitations of comparative field trials and the heterogeneous nature of malaria transmission has limited the availability of good quality performance data that national malaria programmes require to make informed decisions on procurement and implementation. To this end in 2006, WHO and FIND (Foundation for Innovative New Diagnostics) launched an evaluation program to assess the performance of commercially available malaria RDTs and allow direct product comparisons that would assist WHO, other UN agencies and national governments in making procurement decisions and would ultimately encourage improvement in the quality of manufacturing.

1.2. THE WHO PRODUCT TESTING PROGRAMME

This report, which presents the results of the first round of WHO product testing of malaria antigen-detecting RDTs, was completed in November 2008 in collaboration with FIND, the US Centers for Disease Control and Prevention (CDC) and other partners (Fig.2). All companies manufacturing under ISO-13485 Quality System Standard were invited to submit up to 3 tests for evaluation under the programme. In this first round of testing, 41 products from 21 manufacturers were evaluated against prepared blood panels of cultured *Plasmodium falciparum* parasites and patient-derived *P. falciparum* and *P. vivax* parasites, and a parasite-negative panel. Thermal stability was assessed after 2 months of storage at elevated temperature and humidity, and a descriptive ease of use assessment was recorded. Of the 41 products, 16 detect *P. falciparum* alone, 22 detect and

differentiate *P. falciparum* from non-*P. falciparum* malaria¹, and 3 detect *P. falciparum* and non-*P. falciparum* malaria without distinguishing between them. Manufacturers submitted 2 lots of each product for evaluation.

The evaluation is designed to provide comparative data on the performance of the submitted production lots of each product. Such data will be used to guide procurement decisions of WHO and other UN agencies and national governments. Product testing is part of a continuing programme of work to improve the quality of RDTs that are used, and to support broad implementation of reliable malaria diagnosis in areas where malaria is prevalent. A second round of product testing began in April 2009. It is anticipated that a further round will be undertaken in 2010, with a call for expressions of interest later in 2009.

1.3. RESULTS OF THE EVALUATION

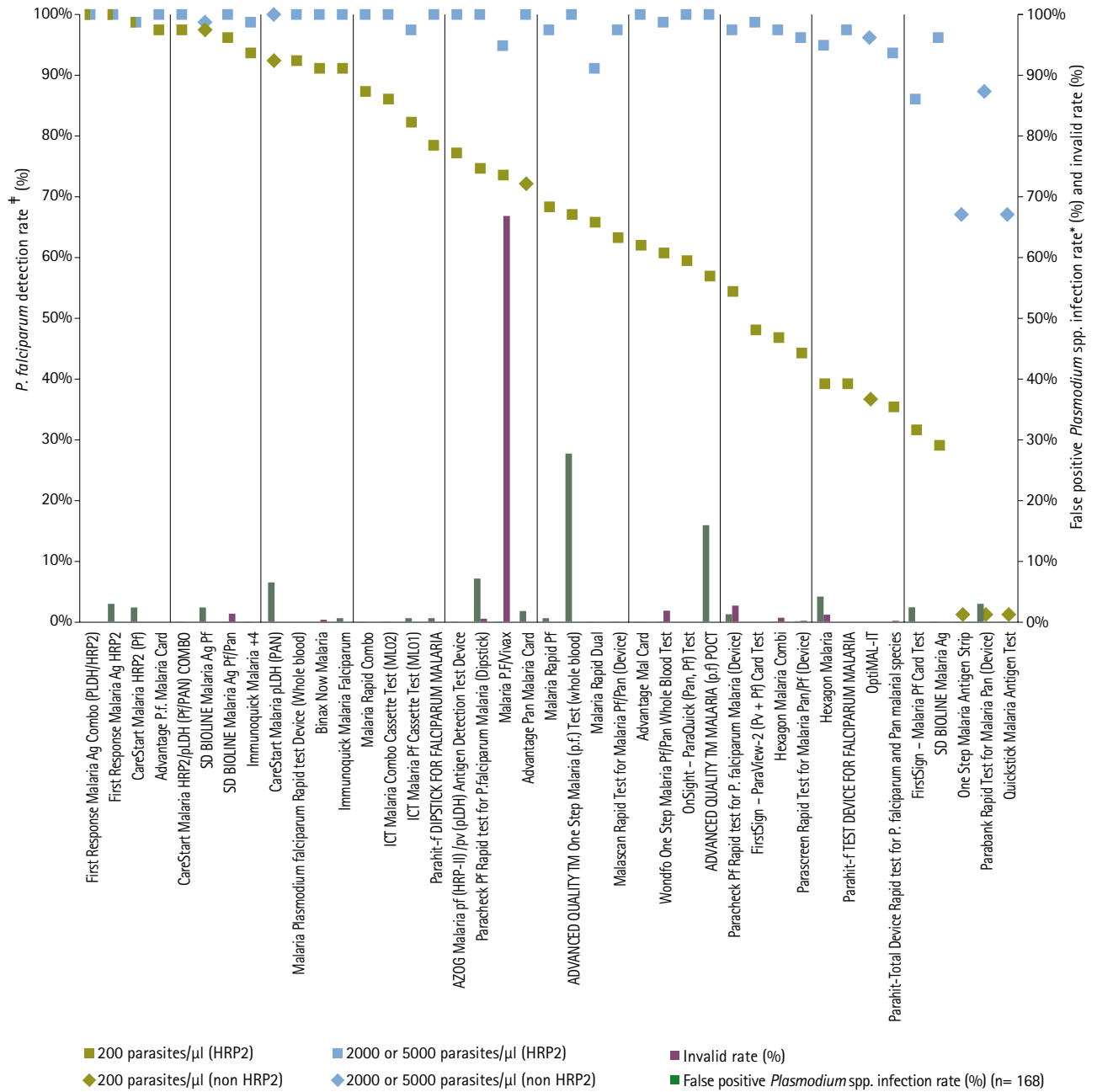
The results (summarized in Tables 3, 3a, 4, 4a and Figure E1) provide comparative data on 2 lots of products against a panel of parasite samples diluted to a low parasite density (200 parasites/ μ L) and a higher parasite density (2000 or 5000 parasites/ μ L). The former is below the mean parasite density found in many populations with endemic malaria. For the purposes of this report, 'detection rate' of a product is the percentage of malaria samples in the panel giving a positive result by two RDTs per lot at the lower parasite density, and a single RDT per lot at the higher parasite density. Thus, it is not a measure of RDT clinical sensitivity, or positivity rate against the panel but rather a combined measure of positivity rate, along with inter-test and inter-lot consistency.

The clinical sensitivity of an RDT to detect malaria is highly dependent on the local conditions, including parasite density, in the target population, and so will vary highly between malaria-endemic populations. The results in this report show comparative performance between RDTs, and give an idea of which products are likely to provide higher sensitivity in the field, particularly in populations with low-density infections. As the detection rate at 2000 parasites/ μ L indicates, the sensitivity of many of these products will be similar in populations with higher parasite densities, although a subset of any population will include vulnerable individuals who may develop illness at low parasite densities (e.g. young children, pregnant women, those well protected by bed nets) and must always be taken into account when interpreting RDT results.

Heat stability (summarized in Table 5) is vital to maintaining sensitivity of the test in the field. As a result, for procurement, it is essential that careful consideration be given to stability results to ensure that products to be used in areas with high temperatures of transport and storage have demonstrated great stability in the product testing programme. Requirements will vary between countries: for example, if tests are to be deployed in areas where temperatures rarely rise above 30°C, less emphasis needs to be placed on stability at high temperatures.

1 One is *P. vivax* specific

Figure E1: Summary performance of malaria RDTs against blood samples containing wild type *P. falciparum* at low (200) and high (2000 or 5000) parasite densities (parasites/ μ l) and malaria-negative samples.



Pf - *Plasmodium falciparum*
 Pv - *Plasmodium vivax*
 pan - *Plasmodium* species

† - A sample is considered detected only if all RDTs from both lots read by the first technician, at minimum specified reading time, are positive. Phase 2 Evaluation Panel: 79 *P. falciparum* wild type samples; 20 *P. vivax* wild type samples and 90 *Plasmodium* spp. negative samples. Rapid diagnostic tests (RDTs) - 2 tests x 2 lots at 200 parasites/ μ l and 1 test x 2 lots at 2000 parasites/ μ l

* - The total number of times a positive result for malaria was generated when it should not have been (Pf+; Pf +/-Pan or Pv -; Pf+/Pan or Pv+; Pf-/Pan or Pv+)

Ease of use requirements will also vary, depending on the extent of training and the work environment of the end-users. Particularly in primary health care settings, the simpler the tests, the easier it will be to avoid errors in preparation and interpretation.

1.4. SUMMARY OF OUTCOMES

- There is now a mechanism in place that allows laboratory-based evaluation of RDT performance in a standardized way to distinguish between well and poorly performing tests to inform procurement and prioritization for entry into WHO prequalification and procurement schemes.
- Several RDTs are available that demonstrated consistent detection of malaria at low parasite densities (200 parasites/ μ l), have low false positive rates, are stable at tropical temperatures, are relatively easy to use, and can detect *P. falciparum*, *P. vivax* infections, or both.
- Performance between products varied widely at low parasite density (200 parasites/ μ l); however, most products showed a high level of detection at 2000 or 5000 parasites/ μ l.
- *P. falciparum* tests targeting HRP2 antigen demonstrated the highest detection rates, but some tests targeting pLDH also exhibited high detection rates.

- Test performance varied between lots, and widely between similar products, confirming the advisability of lot-testing post purchase and prior to use in the field.
- The results underscore the need for manufacturers to have adequate reference materials for product development and lot-release. The WHO-FIND malaria RDT evaluation programme, in collaboration with the CDC, will soon offer quality standard panels to manufacturers to assist in this process.

1.5. USE OF THESE RESULTS

Ultimately, it is imperative that procurement decisions based on these results take into consideration local conditions of malaria transmission and illness where the tests will be used (e.g. *Plasmodium* species, target antigen variation, parasite densities, climate). Procurement of RDTs must not occur without programmatic and infrastructure preparation for proper use, including supply chain management, training on test usage and disposal, and training on patient management in response to results. This report provides an algorithm to assist in this decision-making process (Annex 5).

2. BACKGROUND

In 2006, WHO estimated that 3.3 billion persons were at risk of acquiring malaria. Of these, 247 million were infected (86% in Africa) and nearly 1 million (mostly African children) died of the infection. In 2008, malaria was still endemic in 109 countries worldwide, 45 of them in Africa. WHO estimates that approximately 1.1 million persons were still dying of malaria that year (1).

In the past decade, major new opportunities for the control of malaria have emerged, including implementation of long-lasting insecticidal nets, indoor residual spraying of insecticides and artemisinin-based combination therapy (ACT). These tools, in combination with increased coverage of malaria control programs, are likely to reduce the burden of malaria infection in countries where they are adequately implemented. In turn, the proportion of febrile episodes attributable to malaria is likely to decrease substantially.

Despite WHO recommendations for laboratory-confirmed diagnosis of malaria infections², diagnosis is often made

on clinical grounds (2). However, in most endemic areas malaria makes up a minority of 'malaria-like' febrile illness. Microscopy has been the cornerstone of diagnosis and remains the recommended method of malaria diagnosis at a central level, but the need for trained personnel, adequate reagents and equipment limit its availability and accessibility. Rapid, accurate and accessible diagnostic tools are becoming increasingly important, as programmes expand parasite-based diagnosis and the prevalence of malaria decreases. In recent years, rapid diagnostic tests (RDTs), which detect *Plasmodium*-specific antigens (proteins) in whole blood of infected people, have emerged as an attractive alternative to microscopy. Currently available RDTs come in various formats (dipstick, cassette or card) and contain bound antibodies to specific antigens such as histidine-rich protein-2 (HRP2) (specific to *P. falciparum*), pan-specific or species-specific *plasmodium* lactate dehydrogenase (pLDH) or aldolase (specific to all the major *Plasmodium* species: *P. falciparum*, *P. vivax*, *P. malariae*, *P. ovale* (Figure 1).

To be widely useful, a RDT must have high sensitivity to ensure all clinically-significant malaria infections are detected; high specificity to enable monitoring of low malaria prevalence and appropriate management of non-malarial fever; and high stability to allow transport and storage in ambient conditions in malaria-endemic areas. Published field trials of RDTs show high variability in performance, likely due to inadequate quality of manufacture, incorrect storage and handling, poor preparation and interpretation,

2 With the exception of children under 5 years of age in areas of high transmission in whom treatment may be provided on the basis of a clinical diagnosis

and sometimes poor study methods, analysis and reporting (3-11).

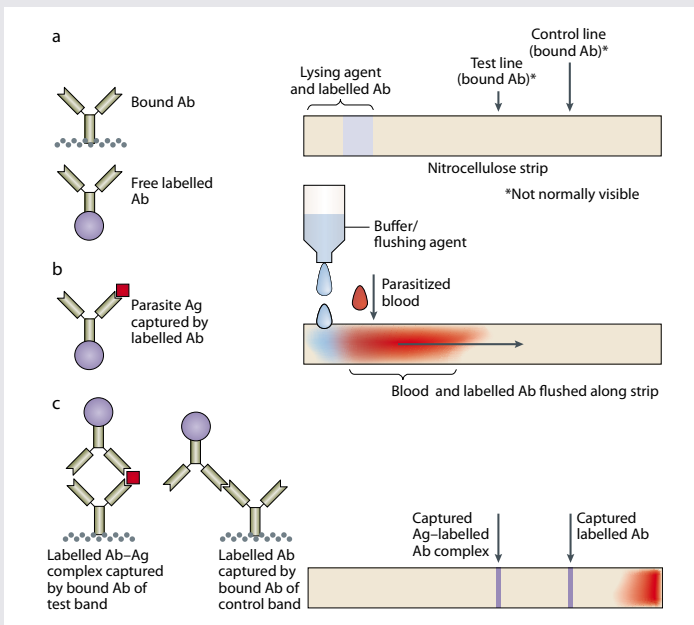
The number of RDTs available on the market has grown rapidly since their introduction in the late 1990s. It is estimated that there are 60 brands and over 200 tests commercially available today, with an estimated 50-70 million tests used in 2008³. However, regulatory oversight of diagnostics is often weak, and procurement agencies have faced considerable problems in selecting appropriate RDTs and ensuring quality. In view of the inconsistency in field study results and the inherent difficulties in

assessing large numbers of products in a standardized way through field trials, WHO and various partners embarked on a Malaria Rapid Diagnostic Test Product Evaluation Programme in 2002 to develop and employ standardized assessment of malaria RDT performance, and to guide procurement decisions and regulatory mechanisms. The Programme has been overseen by WPRO and TDR in partnership with FIND, and has been guided by a Steering Committee⁴ and several consultations from 2003 to 2008. A network of specimen collection sites was established to contribute specimens to a global bank at the US CDC and to facilitate local quality control activities (Figure 2).

3 WHO. Unpublished data

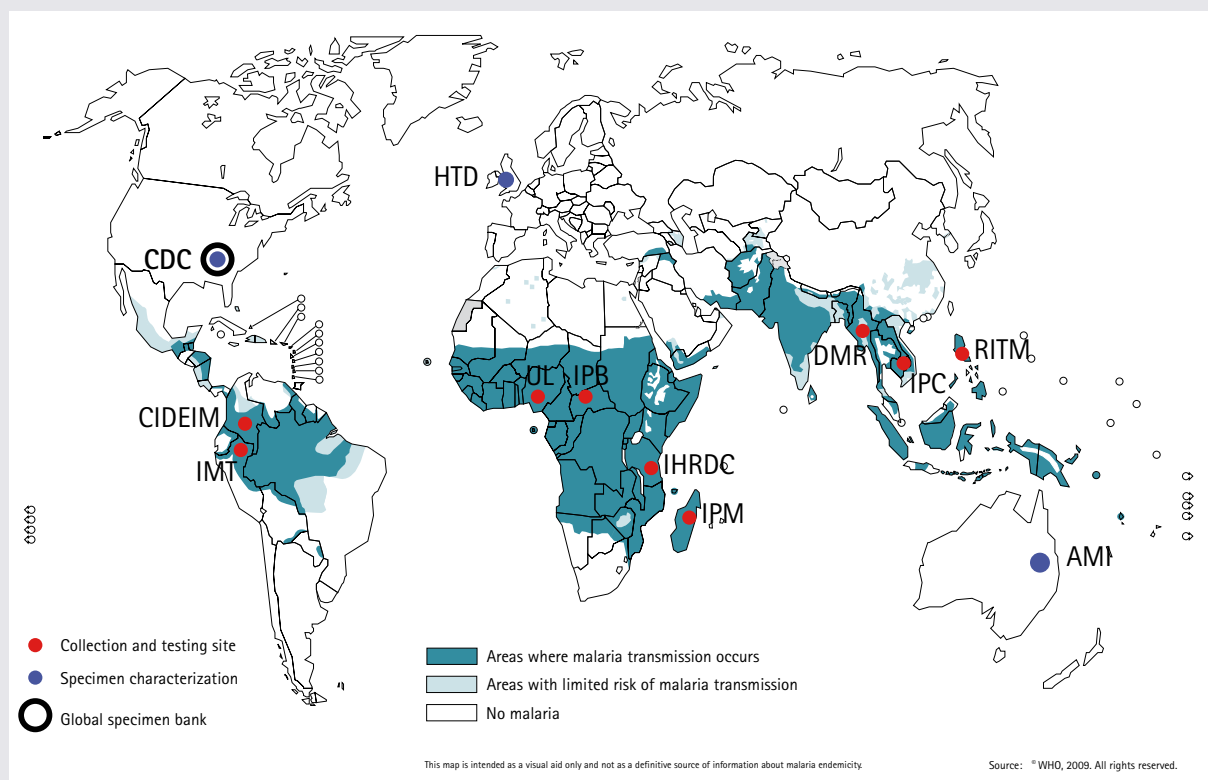
4 WHO/FIND/CDC, Methods manual for product testing of malaria rapid diagnostic tests (version 1). 2008

Figure 1: Mode of action of antigen-detecting malaria RDTs



Mode of action of common malaria RDT format: (a) Dye-labeled antibody (Ab), specific for target antigen, is present on the lower end of the nitrocellulose strip or in a well provided with the strip. Antibody, also specific for the target antigen, is bound to the strip in a thin (test) line, and either antibody specific for the labeled antibody, or antigen, is bound at the control line. (b) Blood and buffer, which have been placed on the strip or in the well, are mixed with the labeled antibody and are drawn up the strip across the lines of bound antibody. (c) If antigen is present, some labeled antibody will be trapped on the test line. Other labeled antibody is trapped on the control line.

Figure 2: Network of specimen collection, characterization and testing sites



Abbreviations: AMI Army Malaria Institute (Queensland, Australia); CDC Centers for Disease Control and Prevention (Atlanta, United States of America); CIDEIM Centro Internacional de Entrenamiento y Investigaciones Médicas (Cali, Colombia); DMR Experimental Medicine Research Division (Department of Medical Research, Yangon, Myanmar); HTD Hospital for Tropical Diseases (London, United Kingdom of Great Britain and Ireland); IHRDC Ifakara Health Research and Development Center (Bagamoyo, The United Republic of Tanzania); IMT Instituto de Medicina Tropical (Universidad Peruana Cayetano Heredia, Lima, Peru); IPB Institut Pasteur de Bangui (Bangui, Central African Republic); IPC Institut Pasteur du Cambodge (Phnom Penh, Cambodia); IPM Institut Pasteur de Madagascar (Antananarivo, Madagascar); RITM Research Institute of Tropical Medicine (Manila, Philippines); UL University of Lagos (Lagos, Nigeria).

3. OBJECTIVES

- Evaluate malaria RDTs to produce performance data that can guide procurement of RDTs for use in the field.

4. MATERIALS AND METHODS

4.1. TEST SELECTION

In August 2007, the WHO-FIND Malaria RDT Evaluation Programme issued a call for expression of interest to manufacturers of malaria RDTs along with information regarding the requirements for submission of a product and the conditions for participation in the Evaluation Programme⁵. Requirements included: ISO 13485:2003

5 http://www.wpro.who.int/sites/rdt/who_rdt_evaluation/ (accessed 08 April 2009)

certification, supply of sufficient quantities of products (1100 tests from each of 2 lots), and compliance with in-house real time stability testing protocol⁶.

After an initial call for expressions of interest, 21 manufacturers submitted a total of 41 products to be included in Round 1 (including 2 co-listed (identical) products).

In summary, of the 41 products: 16 detect *P. falciparum* alone, 22 detect and differentiate *P. falciparum* from non-*P. falciparum* malaria⁷, and 3 detect *P. falciparum* and non-*P. falciparum* malaria without distinguishing between them. Annexes 1 and 2 provide a comprehensive overview of product characteristics.

6 Methods Manual for Product Testing of Malaria RDT Version One 2008. SOP 2.2d; available at www.wpro.who.int/sites/rdt/documents.htm (accessed 08 April 2009)

7 One is *P. vivax*-specific

Table 1: Manufacturers and products accepted into Round 1 of WHO-FIND Malaria RDT Evaluation Programme

Manufacturer	Product name	Catalogue No.*	Target Antigen
Access Bio, Inc.	CareStart Malaria HRP2 (Pf)	G0141	HRP2
	CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	G0131	HRP2/pLDH
	CareStart Malaria pLDH (PAN)	G0111	pLDH
ACON Laboratories, Inc.	Malaria <i>Plasmodium falciparum</i> Rapid test Device (Whole blood)	IMA-402	HRP2
Amgenix International, Inc.	OnSight – ParaQuick (Pan, Pf) Test	536-25DB	HRP2/pLDH
AZOG, Inc.	AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	MFV-124R	HRP2/pLDH
Biosynex	Immunoquick Malaria Falciparum	0502_K25	HRP2
	Immunoquick Malaria +4	0506_K25	HRP2/pLDH
Diagnostics Automation/ Cortez Diagnostics, Inc.	Malaria P.F/Vivax	172110P-25	HRP2/aldolase
DiaMed AG	OptiMAL-IT	710024	pLDH/pLDH
Human GmbH	Hexagon Malaria	58051	HRP2
	Hexagon Malaria Combi	58024	HRP2/aldolase
IND Diagnostic Inc.	One Step Malaria Antigen Strip	820-1	pLDH
Innovatek Medical Inc.	Quickstick Malaria Antigen Test	--	pLDH
	(Co- listing with IND Diagnostics Inc. One Step Malaria Antigen Strip)**		
InTec Products, Inc.	ADVANCED QUALITY™ One Step Malaria (p.f.) Test (whole blood)	ITP11002TC40	HRP2
	ADVANCED QUALITY™ MALARIA (p.f.) POCT	ITP11002TC1	HRP2
Inverness Medical Innovations, Inc.	Binax Now Malaria	IN660050	HRP2/aldolase
J. Mitra & Co. Pvt. Ltd.	Advantage Pan Malaria Card	IR013025	pLDH
	Advantage Pf. Malaria Card	IR016025	HRP2
	Advantage Mal Card	IR221025	pLDH/pLDH
Orchid Biomedical Systems	Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	30301025	HRP2
	Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	30302025	HRP2
Premier Medical Corporation Ltd.	First Response Malaria Ag HRP2	I13FRC30	HRP2
	First Response Malaria Ag Combo (pLDH/HRP2)	I16FRC30	pLDH/HRP2
ICT Diagnostics	ICT Malaria Pf Cassette Test (ML01)	ML01	HRP2
	ICT Malaria Combo Cassette Test (ML02)	ML02	HRP2/aldolase
Span Diagnostics Ltd.	Parahit-f DIPSTICK FOR FALCIPARUM MALARIA	25977	HRP2
	Parahit-f TEST DEVICE FOR FALCIPARUM MALARIA	25975	HRP2
	Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species	25989	HRP2/pLDH /aldolase
Standard Diagnostics, Inc.	SD BIOLINE Malaria Ag	05FK40-02-5	pLDH/pLDH
	SD BIOLINE Malaria Ag Pf	05FK50-02-4	HRP2
	SD BIOLINE Malaria Ag Pf/Pan	05FK60-02-3	HRP2/pLDH
Unimed International, Inc.	FirstSign – Malaria Pf Card Test	--	HRP2
	FirstSign – ParaView-2 (Pv + Pf) Card Test	2102CB-25	HRP2/pLDH
Vision Biotech (Pty) Ltd.	Malaria Rapid Pf	VB01	HRP2
	Malaria Rapid Combo	VB011	HRP2/aldolase
	Malaria Rapid Dual	VB020	HRP2/pLDH
Guangzhou Wondfo Biotech Co., Ltd	Wondfo One Step Malaria Pf/Pan Whole Blood Test	W56-C (4.0 mm)	HRP2/pLDH
Zephyr Biomedicals	Parascreen Rapid Test for Malaria Pan/Pf (Device)	50310025	HRP2/pLDH
	Malascan Rapid Test for Malaria Pf/Pan (Device)	50402025	HRP2/aldolase
	Parabank Rapid Test for Malaria Pan (Device)	50301025	pLDH

* Some products may include different catalogue numbers for different box sizes, contact manufacturers for details.

** Co-listing: One product is submitted for assessment. Manufacturers confirm that second product is identical with different label.

Participating manufacturers received results of test performance and reproducibility prior to publication.

4.2. OUTLINE OF THE PRODUCT TESTING PROTOCOL

The testing process is outlined in Figure 3 and in the Methods Manual for Product Testing of Malaria Rapid Diagnostic Tests - Version One⁸. In brief, RDTs from each of two lots of each product were evaluated against a panel of parasite-positive and parasite-negative cryo-preserved blood samples, and a panel of parasite-negative samples. Both lots were also tested for heat (thermal) stability, evaluated before and after 2 months' storage at 4°C, 35°C and 45°C. Finally, an ease-of-use description was developed using a standard assessment format (Figure 3). The testing process and all results were overseen by the specimen bank steering committee, and manufacturers were given an opportunity to comment on individual product results prior to publication.

4.3. EVALUATION PANELS

RDTs were evaluated against three panels, specifically:

- i) *P. falciparum* culture lines (includes a subset, 'manufacturer's panel') at low (200 parasites/μl) and high parasite densities (2000 or 5000 parasites/μl).
- ii) Wild-type *Plasmodium* species (*P. falciparum*, *P. vivax*) from naturally infected humans and parasite-negative samples at low (200 parasites/μl) and high parasite densities (2000 or 5000 parasites/μl).
- iii) Parasite-negative panel ('clean' samples and disease-specific or blood factor-specific samples).

An overview of the sample collection and characterization process can be found in the "Outline of Product Testing and Associated Protocols"⁹ and detailed methods manuals prepared for laboratory quality control¹⁰ and product testing of malaria RDTs⁹⁻¹⁵. Characterization results can be found at www.wpro.who.int/sites/rdt.

9 Initiative for Quality Assurance of Malaria Rapid Diagnostic Tests: Outline of product testing and associated protocols; available at www.wpro.who.int/sites/rdt/documents.htm (accessed 08 April 2009)

10 Methods Manual for Laboratory Quality Control Testing of Malaria Rapid Diagnostic Test Version 5a available at www.wpro.who.int/sites/rdt/documents.htm (accessed 08 April 2009)

8 Available at www.wpro.who.int/sites/rdt/documents.htm

Figure 3: Malaria RDT Product Testing Overview

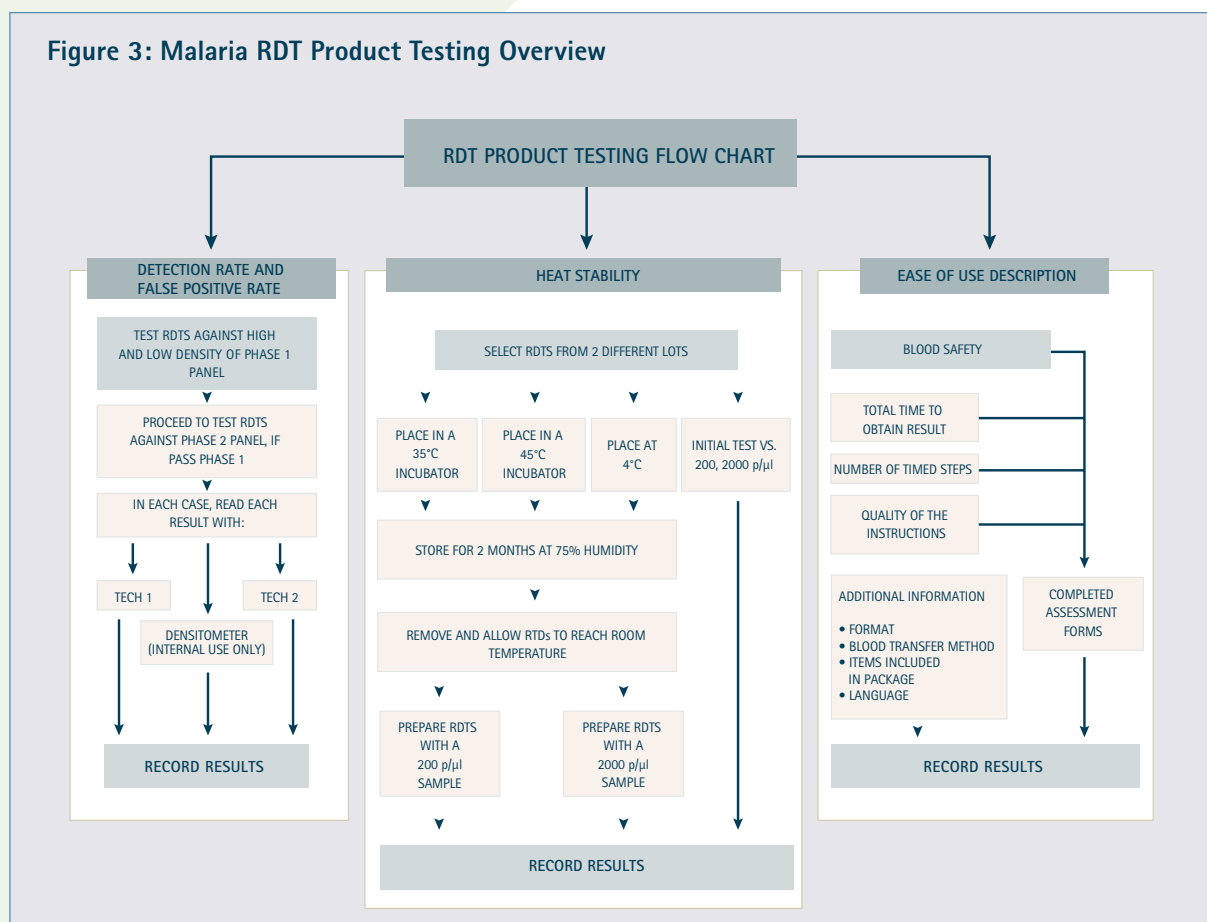


Figure 4a: Origin of *P. falciparum* wild type samples

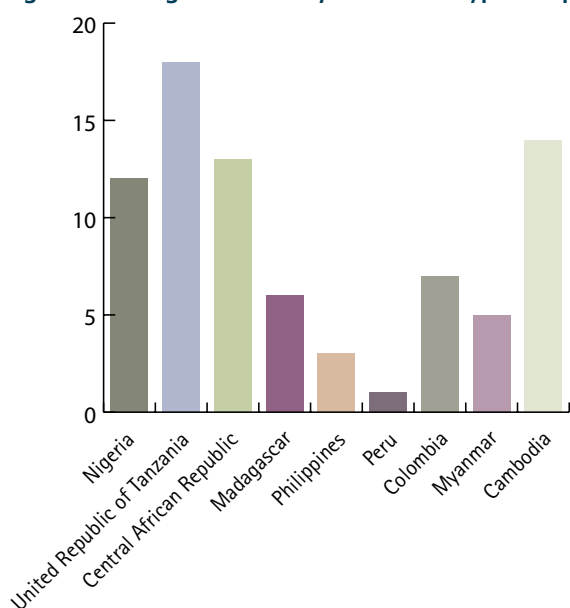
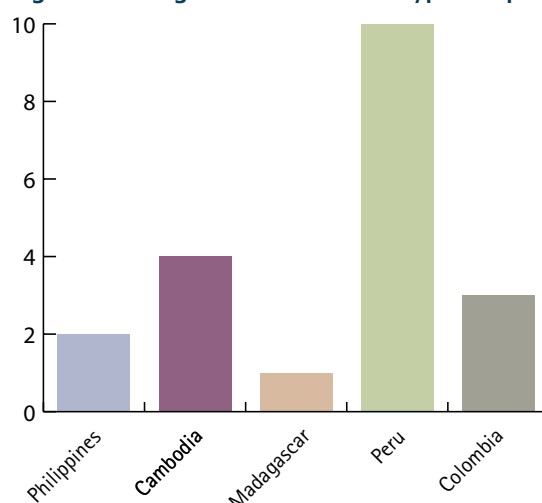


Figure 4b: Origin of *P. vivax* wild type samples



In summary, each panel specimen was characterized for:

- i) Geographical origin
- ii) Species by duplicate microscopy¹¹ (two microscopists) and confirmation by nested PCR of mono-species infection¹²
- iii) HRP2 sequence by PCR amplification¹³
- iv) Antigen concentration, determined by quantitative ELISA for HRP2, pLDH, aldolase¹⁴
- v) PCR for malaria and confirmatory testing for other pathology in the case of parasite-negative samples

Samples were collected from febrile patients and processed according to standardized methods designed to preserve target antigen concentration¹⁶. After dilutions and cryopreservation, samples were transferred to the global bank at US CDC for further characterization. The distribution of concentration of HRP2, aldolase and pLDH were determined on a larger sample, and a test panel developed that excluded samples with extremes of high or low antigen concentration.

Panel composition

P. falciparum-cultured parasites panel

Twenty culture-adapted strains of *P. falciparum* of varied geographical origin were selected, including 13 strains with type B HRP2 sequence, 5 with Type A, and 3 with Type C HRP2 sequence. All specimens were derived from the culture bank of US CDC, and diluted in O+ US donor blood¹⁵.

Wild type parasite panel

The parasite-positive wild type panel consisted of samples from 79 cases of *P. falciparum* and 20 cases of *P. vivax*, derived from 9 collection sites in Asia, Africa and South America (Figs 2, 4a and 4b).

Negative blood samples

The negative panel consisted of 'clean' parasite-negative samples from donor-derived blood banks in non-endemic areas of the Philippines, Madagascar, USA, and Nigeria, and parasite-negative samples from donors with diseases that may potentially be differential diagnoses of malaria, or with specific blood factors known to be common in the community or known to have the potential to cause false positive reactions on immunochromatographic tests (Table 2). Further details of the parasite-negative panel are found at <http://www.wpro.who.int/sites/rdt>.

11 Methods Manual for Product Testing of Malaria RDTs Version One 2008. Chapter 4 SOP 4.5, 4.6

12 Methods Manual for Product Testing of Malaria RDTs Version One 2008 Chapter 5 SOP 5.7, 5.8

13 Methods Manual for Product Testing of Malaria RDTs Version One 2008 Chapter 5 SOP 5.9

14 Methods Manual for Product Testing of Malaria RDTs Version One 2008 Chapter 5 SOP 5.1-5.6

15 Methods Manual for Product Testing of Malaria RDTs Version One 2008. Chapter 3 SOP 3.9

16 Methods Manual for Laboratory Quality Control Testing of Malaria Rapid Diagnostic Tests Version 5a Chapter 3; available at www.wpro.who.int/sites/rdt/documents.htm

Table 2: Characteristics of *Plasmodium* spp. negative specimens

Nature of negative sample ¹	No
'Clean' negative ²	42
Anti-nuclear antibody positive (sera)	13
Anti-mouse antibody positive (plasma)	1
Rheumatoid factor positive (whole blood and sera)	4
Rapid plasma reagin positive (sera)	9
Chagas' disease antibody positive (plasma)	2
Dengue antibody positive (whole blood and sera)	4
Leishmaniasis antibody positive (sera)	5
Schistosomiasis antibody positive (whole blood and sera)	10

1 Whole blood unless indicated. Sera and plasma samples were reconstituted packed cells

2 Healthy volunteers with no known current illness or blood abnormality

4.4. RDT REGISTRATION

The receipt of each shipment of RDTs at the evaluation centre was recorded in a dedicated RDT register. Temperature monitoring devices were offered to manufacturers free of charge, to accompany RDTs shipments to CDC. All RDTs were stored at $\leq 25^{\circ}\text{C}$ immediately and temperature monitors were labeled with receipt date and forwarded for downloading, when applicable.

4.5. SPECIMEN PANEL REGISTRATION

All panel specimens were assigned unique identification numbers at the collection sites and stored in aliquots of $50\mu\text{l}$ at -70°C until the time of testing. All data pertaining to specimen ID, storage location and characteristics was stored in a dedicated database.

4.6. TEST PHASES

The evaluation was divided into two testing phases:

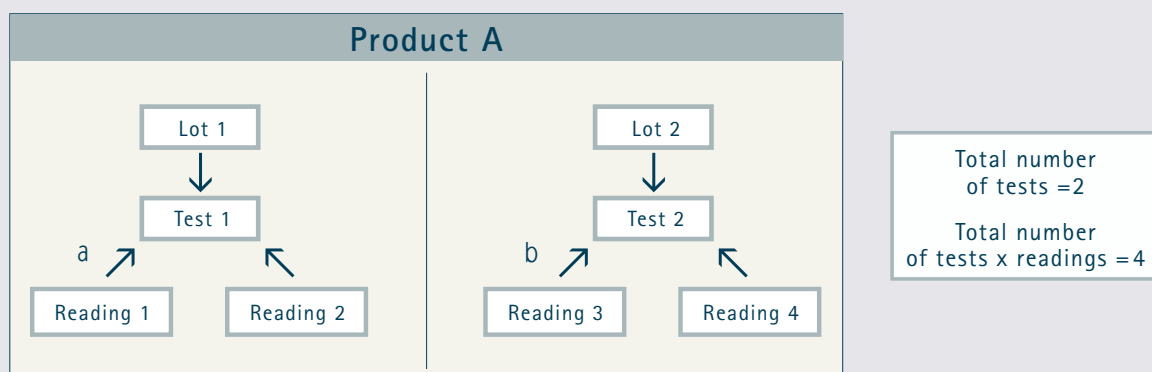
Phase 1 – A first screening step to allow the selection of RDTs meeting minimal quality requirements. Products from two lots were evaluated against a panel of 20 culture-derived *P. falciparum* samples at high (2000 or 5000 parasites/ μl) and low (200 parasites/ μl) parasite densities.

Phase 2 – Products from two lots were evaluated against a panel of blood samples containing wild type parasites and a parasite-negative panel, evaluated for heat (thermal) stability, and assessed for ease of use:

- The parasite-positive and parasite-negative panel was comprised of 79 *P. falciparum*, 20 *P. vivax* at two parasite densities (200 parasites/ μl and 2000 or 5000 parasites/ μl), and 90 parasite-negative controls.
- Heat stability evaluation: Baseline testing of 10 RDTs from each of two lots against a single culture-derived isolate (Nigeria XII strain, Pf HRP2 sequence type B with a typical antigen concentration) at 200 parasites/ μl and 2000 parasites/ μl and 4 RDTs from each lot against a negative sample. This procedure was repeated after RDTs were maintained for 60 days at 4°C , 35°C and 45°C at 75% humidity.

Figure 5: Testing procedure for Product A against a sample density of 2000 or 5000 parasites/ μl

The first reading was at the minimum time specified by the manufacturer; the second reading was up to one hour later and contributed to the determination of the final result only if the manufacturer specified that reading should be repeated if the initial reading was negative for malaria.



Based on positive results of the first test reading in each lot, the mean band intensity score = $a+b/2$

- c. Ease of use assessment: After becoming familiar with the test device, technicians jointly described the test for blood safety characteristics, quality of instructions, number of timed steps and total time to result, using a standard reference guide¹⁷.
- d. A stability assessment was also required to be conducted by manufacturers at the manufacturing site. Manufacturers were requested to assess real-time heat stability at three month intervals against high and low parasite densities supplied by WHO at the upper limit of their recommended storage temperature throughout shelf-life and at the end of shelf-life. Results are submitted to WHO at regular intervals, for internal use, only.

minimum specified reading time and by a second technician within 1 hour. Technicians were rotated, and blinded to sample type and to each other's results during Phase II. Annexes 1 and 2 contain a descriptive and illustrated summary of the test characteristics, steps and guide to interpretation of results.

4.7. PERFORMING RAPID TESTS

All RDTs were brought to room temperature prior to first use. Desiccant was inspected for colour changes and products were discarded if present. RDTs were labeled with patient ID, dilution, and the date when test was performed. Performance of rapid tests was in accordance with manufacturer's instructions, with the exception that blood transfer was carried out by micro-pipette from the sample tube. The result was recorded by a technician at the

4.8. INTERPRETATION OF RESULTS

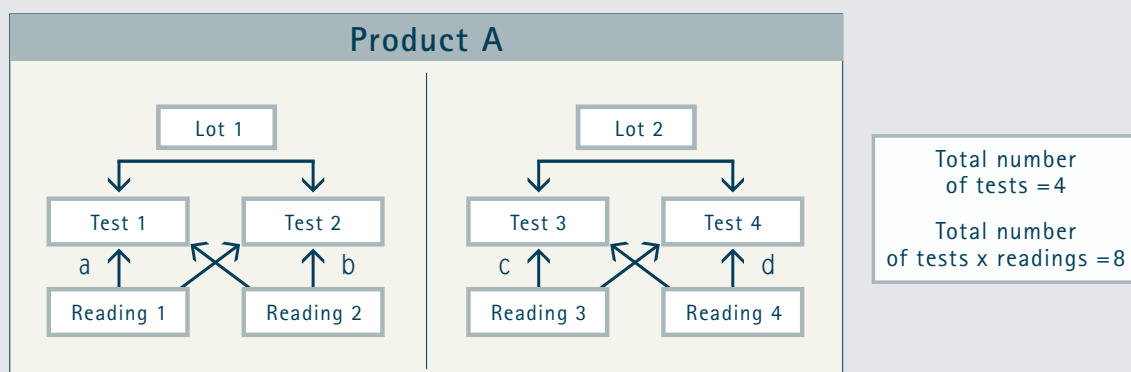
Results of control and test lines were recorded as negative or positive by each technician. Each test was read against a standard colour chart and the band intensity graded as 0 (no band), 1, 2, 3 or 4. If the control line is recorded as absent by either technician, the test is recorded as invalid.

Figures 5 and 6 illustrate the testing sequence at low and high parasite densities.

¹⁷ Methods Manual for Product Testing of Malaria RDTs Version One 2008. Chapter 7, available at www.wpro.who.int/sites/rdt/documents.htm

Figure 6: Testing procedure for Product A against a sample density of 200 parasites/ μ l

The first reading was at the minimum time specified by the manufacturer; the second reading was up to one hour later and contributed to the determination of the final result only if the manufacturer specified that reading should be repeated if the initial reading was negative for malaria.



Based on the positive results of first test reading (2 tests per lot), the mean band intensity score = $(a+b+c+d)/4$ (excluding negative results).

5. DATA MANAGEMENT

The receipt of products was hand recorded in an RDT register at the CDC as per Standard Operating Procedures (SOPs). Data associated with specimen collection and characterization was recorded first on hard copy report forms as per the SOPs at the collection sites (Figure 2), HTD (ELISA reporting) and CDC (PCR) and then entered directly into formatted excel spreadsheets that were subsequently imported into a specially developed database.

The results of the product panel testing and heat stability testing conducted at the CDC were recorded on report forms by each technician individually, as per the SOP.

These results were transferred to a Master Results Sheet and subsequently registered in the database via one of two mechanisms: i) direct database entry or ii) into an excel spreadsheet formatted for subsequent importation into the database.

In order to control for data entry errors, all data discrepancies between Reader 1 and Reader 2 were resolved through inspection of the original hard copy report forms.

All source documents and electronic records of study data have been kept in secure areas until the conclusion of the evaluation, data analysis and report publication.

Individual product testing reports and accompanying raw data were assembled for manufacturers' review 60 days prior to release of the published report.

6. QUALITY ASSURANCE

Product testing followed SOPs developed through prior testing experience and based on recommendations of expert consultations^{6,8}. A pilot phase using a limited number of tests and an abbreviated panel was conducted to refine these procedures. The quality of critical steps was controlled, as follows:

- Quality of the malaria RDTs and their use:

All RDTs were stored in a controlled environment at $\leq 25^{\circ}\text{C}$; the pouch was opened and desiccant checked immediately before use; manufacturer instructions were followed with the exception of use of the blood transfer device provided by the manufacturer (a micropipette was used to ensure correct blood volume).

A temperature-monitoring device was offered to be included with the RDTs for shipment to the testing site.

- Quality and objectivity of the RDT reading results:

Results were read in good lighting by trained technicians tested for visual acuity. Technicians were rotated. Readings of a second technician were reported for discrepant rates; it should be noted that these may have been affected by changes in presence or intensity of test lines with time.

All wild parasite samples at low parasite density were first randomized with a similar number of negative control samples and re-labeled for blinded reading of the RDT results.

- Quality of the specimen bank samples:

SOPs were established for the preparation of all specimen bank samples⁸. Culture lines of parasites and wild-type samples were selected taking into account previous evidence and data from specifically conducted studies. All diluted parasite samples were stored and transported at -70°C , and were used only once within 8 hours of thawing.

- Quality of the product testing site:

The Malaria Branch, Division of Parasitic Diseases at the Centers for Disease Control and Prevention (CDC, Atlanta, USA) is one of the major operating components of the Department of Health and Human Services (HHS) of the USA. The laboratory holds Clinical Laboratory Improvement Amendments (CLIA) accreditation and is monitored by internal Quality Management Systems (QMS) programmes.

7. ETHICAL CONSIDERATIONS

Each specimen collection site obtained approval from a WHO Research Ethics Review Committee and local institutional review board for specimen collection, transport and archiving of blood samples for the purpose of product testing, lot testing and quality assurance procedures.

8. DATA ANALYSIS

8.1. DEFINING SENSITIVITY AND SPECIFICITY/ DETECTION AND FALSE POSITIVE RATES

Malaria RDTs detect parasite-derived antigen. The relationship of the concentration of antigen available from the blood sample (after lysis of red cells and parasites) to the peripheral parasite density varies highly due to a series of host and parasite factors. The population frequency of specific factors that may result in false positive results may also vary. Therefore, field sensitivity and specificity of an RDT may change in different epidemiological situations. The evaluation reported here does not predict sensitivity or specificity in a given field situation. It reports comparative detection of target antigens and false positive rates of RDTs against a standardized panel, in a controlled, repeatable manner. As the panel is developed to be a close approximation of field samples, the comparative detection rates between products are expected to be reflected by similar comparative detection rates in the field. As the panel is designed to include a large number of samples close to the limits of detection of RDTs, the panel is likely to discriminate more clearly than a field trial. It follows that in some settings, such as where parasite density is very high, differences in detection rates between tests observed against the WHO evaluation panel may not be observed in patient populations. Furthermore, where parasite densities are very low, detection rates may be lower than those reported here.

Referring to Figure 6, to be classified as detecting a sample, a product must return 4 positive tests at the manufacturers' recommended minimum reading time (2 from Lot 1, 2 from Lot 2 at initial reading time) when tested using a parasite density of 200 parasites/μl. When tested against 2000 or 5000 parasites/μl (Figure 5) the product must return 2 positive tests at the manufacturers' recommended minimum reading time (1 from each lot). Thus, a "positive" result is a

measure of consistency of result, as well as ability to detect antigen.

The detection rate was defined as the percentage of *P. falciparum* wild type samples detected by the product. The non-*P. falciparum* detection rate is the percentage of *Plasmodium* positive/*P. falciparum* negative samples identified by the product.

8.2. FALSE POSITIVES

False positive results are analysed and reported as two separate groups; those that had incorrect species identification, and those that returned a positive result for samples containing no *Plasmodium* spp. parasites. Specifically, the false positive rate is the percentage of all tests that returned a positive test when it shouldn't have, based on reader 1 results.

8.2.1. Incorrect species identification

A test is considered as returning an incorrect species result if a positive *P. falciparum* test line appears with a sample containing non-*P. falciparum* parasites. *P. vivax* samples resulting in a visible line on a *P. falciparum*-specific test line are also considered to be false positives.

8.2.2. False positives from *Plasmodium*-negative samples

Any test that produces a positive reading to samples with no *Plasmodium* parasites is considered a false-positive. In Phase 2, parasite-negative samples consist of clean negative samples and also samples containing other infectious agents (e.g. Dengue, Leishmania, Chagas) and immunological factors (rheumatoid factor, anti-nuclear antibodies, heterophile antibodies).

8.3. BAND INTENSITY

All positive tests results were recorded according to the band intensity against a standard reference chart, matched closely to line colour. Based on Reader 1 results, the distribution of band intensity results is presented as the mean band intensity of positive results. In addition, the intensity was expressed for each possible result (0, 1, 2, 3 or 4)¹⁸ as the percentage recorded at that level.

8.4. LOT AGREEMENT

Disagreement between test lots is calculated from the number of samples that returned a positive result on both RDTs tested in that lot against parasite-positive samples at 200 parasites/ μ l, and on the single RDT from each lot tested against samples at 2000 or 5000 parasites/ μ l. Thus, high inter-lot agreement indicates consistency in detecting malaria parasites.

¹⁸ A standard intensity comparison chart is used, allowing matching to the closest of 4 common colour variants of labelled antibodies used on RDTs, each at 4 levels of intensity.

8.5. READER VARIABILITY

Two readings of each test are conducted, although only the first is certain to comply with the manufacturer's guidelines for reading test results. A second reading is performed within one hour. Detection rates for both readers are calculated to assess how sensitive the results are to the time of reading.

8.6. INVALID TESTS

The total number of tests that were deemed invalid during testing of both lots, using samples at 200 parasites/ μ l and 2000 or 5000 parasites/ μ l.

8.7. HEAT (THERMAL) STABILITY

The results of heat stability testing are reported as the number of positive tests (maximum 10) and mean band intensity (for positive tests only) per lot at baseline and after lots were stored at 4°C, 35°C and 45°C for 2 months against one *P. falciparum* parasite sample at 200 and 2000 parasites/ μ l.

9. LABORATORY VERSUS FIELD-BASED MALARIA RDT EVALUATIONS

Despite the strengths of the product testing programme, the evaluation is not completely analogous to field testing of malaria RDTs. In order to compose a panel that could be reproducibly used to evaluate RDTs, blood samples were diluted, frozen and stored below -70°C . Blood that has undergone a freeze thaw process may not have exactly the same characteristics as fresh blood, but as red cell lysis occurs as a first step on RDTs, the effect of this is limited. A further variation from field equivalence is the use of a micro-pipette to supply blood to the RDT device rather than the blood transfer device provided by the manufacturer. This was necessary because blood is collected from a cryo-tube rather than a finger-prick. This technique also ensured consistency of testing by reducing the likelihood of operator error.

10. RESULTS

10.1. SUMMARY

In Round 1 of the WHO-FIND Malaria RDT Evaluation Programme, 41 products were evaluated against *P. falciparum* culture samples, blood samples collected from parasitaemic patients from three continents and a large panel of parasite negative samples. Heat stability was assessed at temperatures commonly encountered in malaria endemic countries. Twelve research institutes were engaged in either sample collection or sample characterization to establish the evaluation panels. Between May and November 2008, in total, over 41 000 tests were performed at the US Centers for Disease Control and Prevention.

The results of the evaluation reveal the following key outcomes:

1. Several RDTs are available that demonstrated consistent detection of malaria at low parasite densities (200 parasites/ μ l), have low false-positive rates, are stable at tropical temperatures, are relatively easy to use, and can detect *P. falciparum*, *P. vivax* infections, or both.

2. Performance between products varied widely at low parasite density (200 parasites/ μ l); however, most products showed a high level of detection at 2000 to 5000 parasites/ μ l.

3. *P. falciparum* tests targeting HRP2 antigen demonstrated the highest detection rates, but some tests targeting pLDH also exhibited high detection rates.

4. Test performance varied between lots, and widely between similar products.

Tables 3 and 4 summarize the performance of 41 malaria RDTs against *P. falciparum* cultured parasites (Table 3) and blood containing wild type *P. falciparum* and *P. vivax* parasites and *Plasmodium* spp. negative samples (Table 4). Tables 3a and 4a present summary results for 7 products based on a delayed reading when specified in the manufacturers' instructions. Detailed information pertaining to all product testing results is included in Annex 3 (Phase 1) and Annex 4 (Phase 2). A graphical representation of this data follows below.

In summary tables, data is colour coded according to arbitrary categories, to ease the interpretation of results, and these do not imply limits of acceptable or unacceptable performance.

Table 3: Summary performance of malaria RDTs against 20 cultured *P. falciparum* lines at low (200) and high (2000 or 5000) parasite densities (parasites/µl)

Product	Manufacturer	Detection rate* (n=20) 200 parasites/µl	Detection rate* (n=20) 2,000 or 5,000 parasites/µl	False positive non-Pf infection † (%) 200 parasites/µl	False positive non-Pf infection † (%) 2,000 or 5,000 parasites/µl	Invalid rate (%)
Pf only						
ADVANCED QUALITY TM MALARIA (p.f) POCT	InTec Products, Inc.	15.0	90.0	N/A	N/A	0.0
ADVANCED QUALITY TM One Step Malaria (p.f.) Test (whole blood)	InTec Products, Inc.	10.0	90.0	N/A	N/A	0.0
Advantage Pf. Malaria Card	J. Mitra & Co. Pvt. Ltd.	85.0	100.0	N/A	N/A	0.0
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	100.0	100.0	N/A	N/A	0.0
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	100.0	100.0	N/A	N/A	0.0
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	0.0	80.0	N/A	N/A	0.0
Hexagon Malaria	Human GmbH	5.0	95.0	N/A	N/A	0.0
ICT Malaria Pf Cassette Test (MI01)	ICT Diagnostics	55.0	100.0	N/A	N/A	0.0
Immunoquick Malaria falciparum	Biosynex	55.0	100.0	N/A	N/A	0.0
Malaria Plasmodium falciparum Rapid test Device (Whole blood)	ACON Laboratories, Inc.	85.0	95.0	N/A	N/A	0.0
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	70.0	100.0	N/A	N/A	0.0
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	15.0	95.0	N/A	N/A	2.5
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	55.0	100.0	N/A	N/A	0.0
Parahit-F DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	45.0	100.0	N/A	N/A	0.0
Parahit-F TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	10.0	90.0	N/A	N/A	0.0
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	90.0	100.0	N/A	N/A	0.0
Pf & Pan/Pf & Pv						
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	0.0	100.0	1.25	0.00	0.0
AZOG Malaria pf (HRP-II) / pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	85.0	100.0	0.00	0.00	0.0
Binax Now Malaria	Inverness Medical Innovations, Inc.	80.0	100.0	0.00	0.00	0.0
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	95.0	100.0	1.25	0.00	0.0
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	95.0	100.0	0.00	0.00	0.0
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	15.0	95.0	2.47	0.00	0.0
Hexagon Malaria Combi	Human GmbH	10.0	95.0	0.00	0.00	0.0
ICT Malaria Combo Cassette Test (MI02)	ICT Diagnostics	75.0	100.0	1.25	0.00	0.0
Immunoquick Malaria + 4	Biosynex	85.0	100.0	0.00	0.00	0.0
Malaria Pf/Pv/vvax	Diagnostic Automation / Cortez Diagnostics, Inc.	0 (19)	6.67 (15)	82.69	91.67	36.7
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	50.0	95.0	0.00	0.00	0.0
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	75.0	100.0	0.00	0.00	0.0
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	15.0	100.0	0.00	0.00	0.0
One Step Malaria Antigen Strip	IND Diagnostic Inc.	0.0	70.0	0.00	0.00	0.0
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	65.0	100.0	0.00	0.00	0.0
OptiMAL-IT	DiaMed AG	0.0	100.0	1.25	0.00	0.0
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	5.0	90.0	0.00	0.00	0.0
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	30.0	100.0	0.00	0.00	0.0
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	0.0	70.0	0.00	0.00	0.0
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	0.0	85.0	0.00	0.00	0.0
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	85.0	100.0	0.00	0.00	0.0
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	30.0	90.0	0.00	0.00	0.0
Pan only						
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	0.0	95.0	N/A	N/A	0.0
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	60.0	100.0	N/A	N/A	0.0
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	0.0	65.0	N/A	N/A	0.0

Pf - <i>Plasmodium falciparum</i> Pv - <i>Plasmodium vivax</i>						
pan - Plasmodium species						
* - Pan or Pv line only positive indicates a false positive non <i>P. falciparum</i> infection						
† - a sample is considered detected only if all RDTs from both lots read by the first technician, at minimum specified reading time, are positive						
Detection rate (%)	≥95	85-94	50-84	< 50		
False positive rate (%)	<2	2-5	6-0	>10		
Invalid rate (%)	<1% of tests conducted	1-2% of tests conducted	2-5% of tests conducted	>5% of tests conducted		

Table 3a: Summary performance of malaria RDTs against 20 cultured *P.falciparum* lines at low (200) and high (2000 or 5000) parasite densities (parasites/µl) based on a delayed reading when specified by the manufacturer

Product	Manufacturer	Detection rate* (n=20)		False positive non-PF infection [†] (2,000 or 5,000 parasites/µl)	Invalid rate (%)
		200 parasites/µl	2,000 or 5,000 parasites/µl		
Pf & Pan/Pf & Pv					
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	50	95	0	0
Immunoquick Malaria +4	Biosynex	85	100	0	0
Malaria PF/Vivax	Diagnosics Automation/Cortez Diagnostics, Inc.	0 (19)	0 (17)	75.47	34.17
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	25	100	0	0
Parascreeen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	70	100	0	0
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	40	90	0	0
Pf only					
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	0	95	N/A	0

Pf - *Plasmodium falciparum*

Pv - *Plasmodium vivax*

pan - *Plasmodium* species

[†] - Pan or Pv line only positive indicates a false positive non *P. falciparum* infection

* - a sample is considered detected only if all RDTs from both lots read by the first technician, at minimum specified reading time, are positive

Table 4: Summary performance of malaria RDTs against wild type *P. falciparum* and *P. vivax* samples at low (200) and high (2000 or 5000) parasite densities (parasites/µl) and *Plasmodium* spp. negative samples.

Product	Manufacturer	Detection rate* (%)				False positive rates (%)		Total False positive rates* (%)		Invalid rate (%)	
		200 parasites/µl		2000 or 5000 parasites/µl		200 parasites/µl		2000 or 5000 parasites/µl			
		Pf samples (n=79)	Pv samples (n=20)	Pf samples (n=79)	Pv samples (n=20)	Pf samples False positive non Pf infection [†] (n=316)	Pv samples False positive non Pf infection [†] (n=80)	Pf samples False positive non Pf infection [†] (n=158)	Pv samples False positive PF infection [‡] (n=40)		
Pf only											
ADVANCED QUALITY™ MALARIA (p.f.) POCT	InTec Products, Inc.	5696	N/A	100	N/A	N/A	12.5	N/A	17.5	16.07	0
ADVANCED QUALITY™ One Step Malaria (p.f.) Test (whole blood)	InTec Products, Inc.	6709	N/A	100	N/A	N/A	48.75	N/A	45	27.98	0
Advantage P.f. Malaria Card	J. Mitra & Co. Pvt. Ltd.	97.47	N/A	100	N/A	N/A	1.25	N/A	2.5	0	0
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	98.73	N/A	98.73	N/A	N/A	5	N/A	7.5	2.38	0
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	100	N/A	100	N/A	N/A	0	N/A	0	2.98	0
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	31.65	N/A	86.08	N/A	N/A	12.5	N/A	15	2.41 (166)	0
Hexagon Malaria	Human GmbH	39.24	N/A	94.94	N/A	N/A	7.89 (76)	N/A	2.5	4.19 (167)	1.18
ICT Malaria Pf Cassette Test (ML01)	ICT Diagnostics	82.28	N/A	97.47	N/A	N/A	1.27	N/A	2.5	0.6	0
Immunoquick Malaria falciparum	Biosynex	91.14	N/A	100	N/A	N/A	0	N/A	0	0.6	0
Malaria <i>Plasmodium falciparum</i> Rapid test Device (Whole blood)	ACON Laboratories, Inc.	92.41	N/A	100	N/A	N/A	0	N/A	0	0	0
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	68.35	N/A	97.47	N/A	N/A	0	N/A	0	0.6	0
Paracheck PF Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	54.43	N/A	97.47	N/A	N/A	3.95 (76)	N/A	5	1.25 (160)	2.69
Paracheck PF Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	74.68	N/A	100	N/A	N/A	16.46	N/A	10	7.19 (167)	0.51
Parahit-f DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	78.48	N/A	100	N/A	N/A	0	N/A	0	0.6	0
Parahit-f TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	39.24	N/A	97.47	N/A	N/A	0	N/A	0	0	0
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	97.47	N/A	98.73	N/A	N/A	0	N/A	0	2.38	0

Product	Manufacturer	Detection rate* (%)				False positive rates (%)		False positive rates (%)		Total False positive rates* (%)		Invalid rate (%)
		200 parasites/µl (n=79)	Pf samples (n=20)	Pf samples (n=79)	Pv samples (n=20)	2000 or 5000 parasites/µl (n=80)	Pf samples (n=316)	200 parasites/µl (n=80)	Pv samples (n=40)	2000 or 5000 parasites/µl (n=158)	Pf samples (n=40)	
Pf & Pan/Pf & Pv												
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	62.03	100	100	100	2.53	0	0	0	0	4.17	0
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	77.22	30	100	95	1.9	0	0	2.5	0	9.52	0
Binax Now Malaria	Inverness Medical Innovations, Inc.	91.14	10	100	85	0.32	3.8	5	0	0	0.00	0.34
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	97.47	90	100	95	0.32	1.25	0	2.5	0	2.98	0
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	100	75	100	65	0	3.75	0	20	0	3.57	0
FirstSign - ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	48.1	0	98.73	85	0.95	3.75	5	0	0	0.00	0
Hexagon Malaria Combi	Human GmbH	46.84	0	97.47	50	0	3.75	2.63 (38)	0	0	2.99	0.67
ICT Malaria Combo Cassette Test (MLO2)	ICT Diagnostics	86.08	0	100	95	0.32	3.75	0	5	0	0.6	0
Immunoquick Malaria +4	Biosynex	93.67	30	98.73	100	0 (314)	0	0 (157)	0	0	0.60	0
Malaria Pf/Pvmax	Diagnostic Automation / Cortez Diagnostics, Inc.	73.58 (53)	0 (15)	94.87 (39)	30.77 (13)	1.03 (97)	0 (30)	2.08 (48)	0 (18)	0	1.56	67.51
Malaria Rapid Combo	Span Diagnostics Ltd.	87.34	10	100	90	0.32	7.5	0	7.5	0	7.74	0
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	75.95	5	98.73	90	1.27	0	0	0	0	10.12	0
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	63.29	0	97.47	75	0.63	6.25	0	17.5	0	5.36	0
One Step Malaria Antigen Strip	IND Diagnostic Inc.	1.27	0	67.09	60	2.22	3.75	1.9	0	0	1.80	0
OnSight - ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	59.49	50	100	100	0	1.25	0.63	0	0	0.00	0
OptiMAL-IT	DiaMed AG	36.71	95	96.2	100	3.8	0	0	0	0	0.00	0
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	35.44	0	93.67	50	0 (315)	0	0	2.5	0	0.00	0.17
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	50.63	25	100	95	0.63	3.8	0	0	0	1.19	0.17
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	1.27	0	67.09	60	2.22	3.75	1.9	0	0	1.80	0
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	29.11	50	96.2	100	0	1.25	0	0	0	1.79	0
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	96.2	35	100	95	0 (310)	0	0	2.56 (39)	0	1.19	1.35
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	60.76	30	98.73	95	3.52 (312)	1.3 (77)	0 (155)	2.56 (39)	0	6.59	1.85
Pan only												
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	72.15	100	100	100	N/A	N/A	N/A	N/A	N/A	1.79	0
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	92.41	100	100	100	N/A	N/A	N/A	N/A	N/A	6.55	0
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	1.27	30	87.34	100	N/A	N/A	N/A	N/A	N/A	2.99 (167)	0
Pf - <i>Plasmodium falciparum</i>												
Pv - <i>Plasmodium vivax</i>												
pan - <i>Plasmodium</i> species												
+ - For combination tests, Pan or Pv line, only, positive indicates a false positive non <i>P. falciparum</i> infection												
* - A sample is considered detected only if all RDIs from both lots read by the first technician, at minimum specified reading time, are positive												
o - Pf line positive indicates a false positive <i>P. falciparum</i> infection												
* - The total number of times a positive result for malaria was generated when it should not have been (Pf+, Pf -/Pan or Pv -, Pf+/Pan or Pv+, Pf-/Pan or Pv+)												

Detection rate (%)	≥95	85-94	50-84	< 50
False positive rate (%)	<2	2-5	6-10	> 10
Invalid rate (%)	<1% of tests conducted	1-2% of tests conducted	2-5% of tests conducted	>5% of tests conducted

Table 4a: Summary performance of malaria RDTs against *P. falciparum* lines at low (200) and high (2000 or 5000) parasite densities (parasites/μl) based on a delayed reading when specified by the manufacturer

Product	Manufacturer	Detection rate* (%)				False positive rates (%)		False positive rates (%)		Total false positive rates* (%)	
		200 parasites/μl		2000 or 5000 parasites/μl		200 parasites/μl		2000 or 5000 parasites/μl		Clean Negative samples	False positive <i>Plasmodium</i> sp. infection (n=168)
		Pf samples (n=79)	Pv samples (n=20)	Pf samples (n=79)	Pv samples (n=20)	Pf samples	Pv samples	Pf samples	Pv samples		
Pf & Pan/Pf & Pv											
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	64,56	0	100	100	0	2,5 (80)	0	2,5	0	0
Immunoquick Malaria +4	Biosynex	93,67	45	100	95	0	1,25 (80)	0	2,5	7,14	0,17
Malaria Pf/Vivax	Diagnostic Automation / Cortez Diagnostics, Inc.	71,43 (56)	0 (15)	92,5 (40)	45,45 (11)	0	0 (30)	4,17	0 (15)	3,23	67,17
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	68,35	0	98,73	85	1,58	7,5 (80)	0	12,5	6,55	0
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	65,82	55	98,73	100	0,63	0	0,63	0	2,38	0,09
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	68,35	35	98,73	90	1,92	3,9 (77)	0	5,13 (39)	7,38	1,85
Pan only											
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	13,92	75	97,47	100	N/A	N/A	N/A	N/A	N/A	0

* - For combination tests, Pan or Pv line, only, positive indicates a false positive non *P. falciparum* infection
 † - A sample is considered detected only if all RDTs from both lots read by the first technician, at minimum specified reading time, are positive
 ‡ - Pf line positive indicates a false positive *P. falciparum* infection

Detection rate (%)	≥95	85-94	50-84	< 50
False positive rate (%)	<2	2-5	6-10	>10
Invalid rate (%)	<1% of tests conducted	1-2% of tests conducted	2-5% of tests conducted	>5% of tests conducted

Table 5: Heat stability testing results for malaria RDTs on a cultured *P. falciparum* sample at low (200) and high (2000 or 5000) parasite density (parasites/μl). Positivity rate at baseline, and after 60 days incubation at 35°C and 45°C

Product	Manufacturer	Positive tests results for <i>P. falciparum</i> (Pf line) 200 parasites/μl		Positive tests results for <i>P. falciparum</i> (Pf line) 2000 parasites/μl		Positive tests results for <i>P. falciparum</i> (Pan line) 2000 parasites/μl	
		Baseline	45°C	Baseline	45°C	Baseline	45°C
		Number of tests positive (max. 20)		Number of tests positive (max. 20)		Number of tests positive (max. 20)	
Pf only							
ADVANCED QUALITY™ MALARIA (p.f.) POCT	InTec Products, Inc.	16	19	18	20	N/A	N/A
ADVANCED QUALITY™ One Step Malaria (p.f.) Test (whole blood)	InTec Products, Inc.	16	17	9	20	N/A	N/A
Advantage P.f. Malaria Card	J. Mitra & Co. Pvt. Ltd.	19	20	20	20	N/A	N/A
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	20	20	20	20	N/A	N/A
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	20	20	20	20	N/A	N/A
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	4	3	0	20	19	N/A
Hexagon Malaria	Human GmbH	10	7	12	20	N/A	N/A

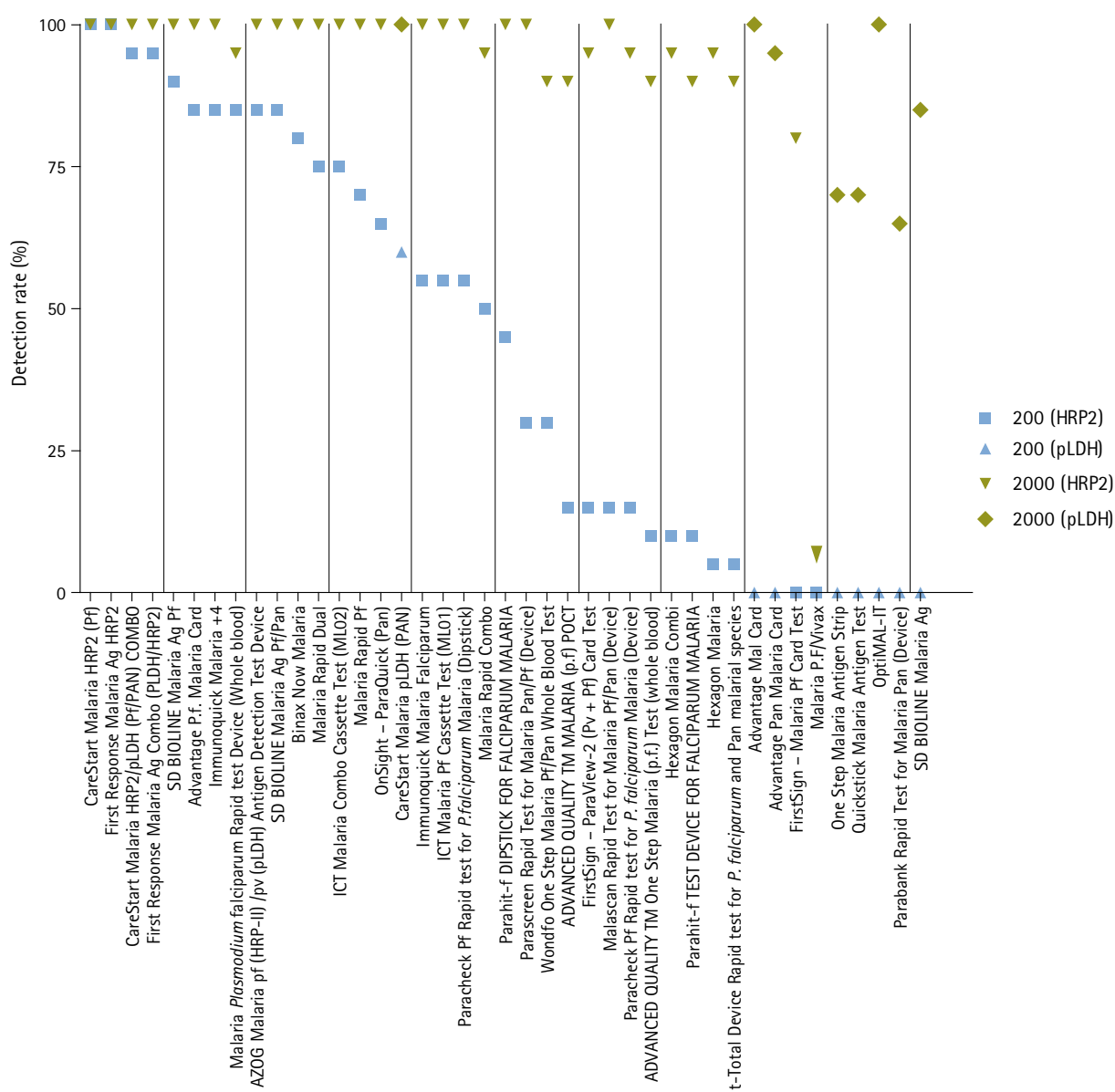
Product	Manufacturer	Positive tests results for <i>P. falciparum</i> (Pf line) 2000 parasites/µl			Positive tests results for <i>P. falciparum</i> (Pf line) 2000 parasites/µl			Positive tests results for <i>P. falciparum</i> (Pan line) 2000 parasites/µl							
		Baseline	35°C	45°C	Baseline	35°C	45°C	Baseline	35°C	45°C					
		Number of tests positive (max. 20)			Number of tests positive (max. 20)			Number of tests positive (max. 20)							
		Lots 1 and 2 combined			Lots 1 and 2 combined			Lots 1 and 2 combined							
ICT Malaria Pf Cassette Test (ML01)	ICT Diagnostics	20	20	19	20	20	20	20	20	20	N/A	N/A	N/A	N/A	
Immunquick Malaria Falciparum	Biosynex	20	20	20	20	20	20	20	20	20	N/A	N/A	N/A	N/A	
Malaria Plasmodium falciparum Rapid test Device (Whole blood)	ACON Laboratories, Inc.	20	20	20	20	20	20	20	20	20	N/A	N/A	N/A	N/A	
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	20	20	17	20	20	20	20	20	20	N/A	N/A	N/A	N/A	
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	18	14	10	20	17	20	20	20	20	N/A	N/A	N/A	N/A	
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	19	20	17	20	20	20	20	20	20	N/A	N/A	N/A	N/A	
Parahit-F DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	20	20	20	20	20	20	20	20	20	N/A	N/A	N/A	N/A	
Parahit-F TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	14	10	8	20	19	20	19	20	20	N/A	N/A	N/A	N/A	
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	20	20	20	20	20	20	20	20	20	N/A	N/A	N/A	N/A	
Pf & Pan/Pf & Pv															
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	20	20	11	19	20	20	19	20	20	11	9	8	20	20
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	12	13	7	20	20	20	20	20	20	8	7	5	18	4
Binax Now Malaria	Inverness Medical Innovations, Inc.	20	20	20	20	20	20	19	20	20	1	0	0	19	15
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	20	19	20	20	20	20	20	20	20	19	20	20	20	20
First-Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	20	20	20	20	20	20	20	20	20	19	14	20	20	20
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	19	14	0	20	19	15	0	0	0	0	0	0	0	0
Hexagon Malaria Combi	Human GmbH	13	11	10	20	17	19	0	0	0	0	0	0	0	0
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	20	20	20	20	20	20	20	20	20	1	1	0	18	15
Immunquick Malaria + 4	Biosynex	20	20	20	20	20	20	20	20	20	0	0	0	20	16
Malaria Pf/Vvax	Diagnostic Automation / Cortez Diagnostics, Inc.	13	3	4	13	9	1	0	0	0	0	0	0	0	0
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	20	20	20	20	20	20	20	20	20	3	6	0	19	15
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	20	20	20	20	20	20	20	20	20	0	3	0	12	5
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	19	18	17	20	20	20	20	20	20	0	1	0	13	11
One Step Malaria Antigen Strip	IND Diagnostic Inc.	3	0	0	13	10	0	0	0	0	3	0	0	13	10
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	20	18	12	20	20	20	20	20	20	0	0	0	20	19
OptiMAL-IT	DiaMed AG	6	2	0	20	19	0	0	0	0	6	3	0	20	2
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	13	15	5	19	20	20	19	20	20	1	0	0	0	0
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	19	16	9	20	20	19	19	20	20	1	0	0	17	14
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	3	0	0	13	10	0	0	0	0	3	0	0	13	10
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	7	12	15	20	20	20	20	20	20	0	9	15	11	20
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	20	20	20	20	20	19	0	0	0	1	16	18	9	18
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	20	19	20	19	20	20	20	20	20	14	18	14	19	20
Pan only															
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	10	13	14	20	20
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	20	20	18	20	20
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	1	0	0	17	14
Pf – Plasmodium falciparum pan – Plasmodium vivax * – pan line for pan-only tests															

10.2. PHASE 1 - *P. FALCIPARUM* CULTURE PANEL

The majority (73%) of tests consistently detected $\geq 95\%$ of *P. falciparum* cultured parasites at high parasite densities; however, detection rate was highly variable at low parasite densities (0-100%). At low parasite densities, the top ten products all targeted HRP2 (Figure 7).

Table 3a illustrates detection rates at low and high parasite densities based on a delayed second reading if it was specifically indicated in the package insert following initial negative results at the specified reading time. For 3 products, results were unchanged and for 4 products detection rates increased.

Figure 7: Phase 1 *P. falciparum* detection rate of malaria RDTs at low (200) and high (2000 or 5000) parasite densities (parasites/ μ l) according to target antigen type (HRP2 or pLDH)[†]



[†] - Phase 1 Evaluation Panel : 20 *P. falciparum* culture samples. RDTs - 2 tests x 2 lots at 200 parasites/ μ l ; 1 test x 2 lots at 2000 parasites/ μ l.

10.3. PHASE 2 - WILD TYPE *P. FALCIPARUM* AND *P. VIVAX* AND *PLASMODIUM* SPP. NEGATIVE SAMPLES

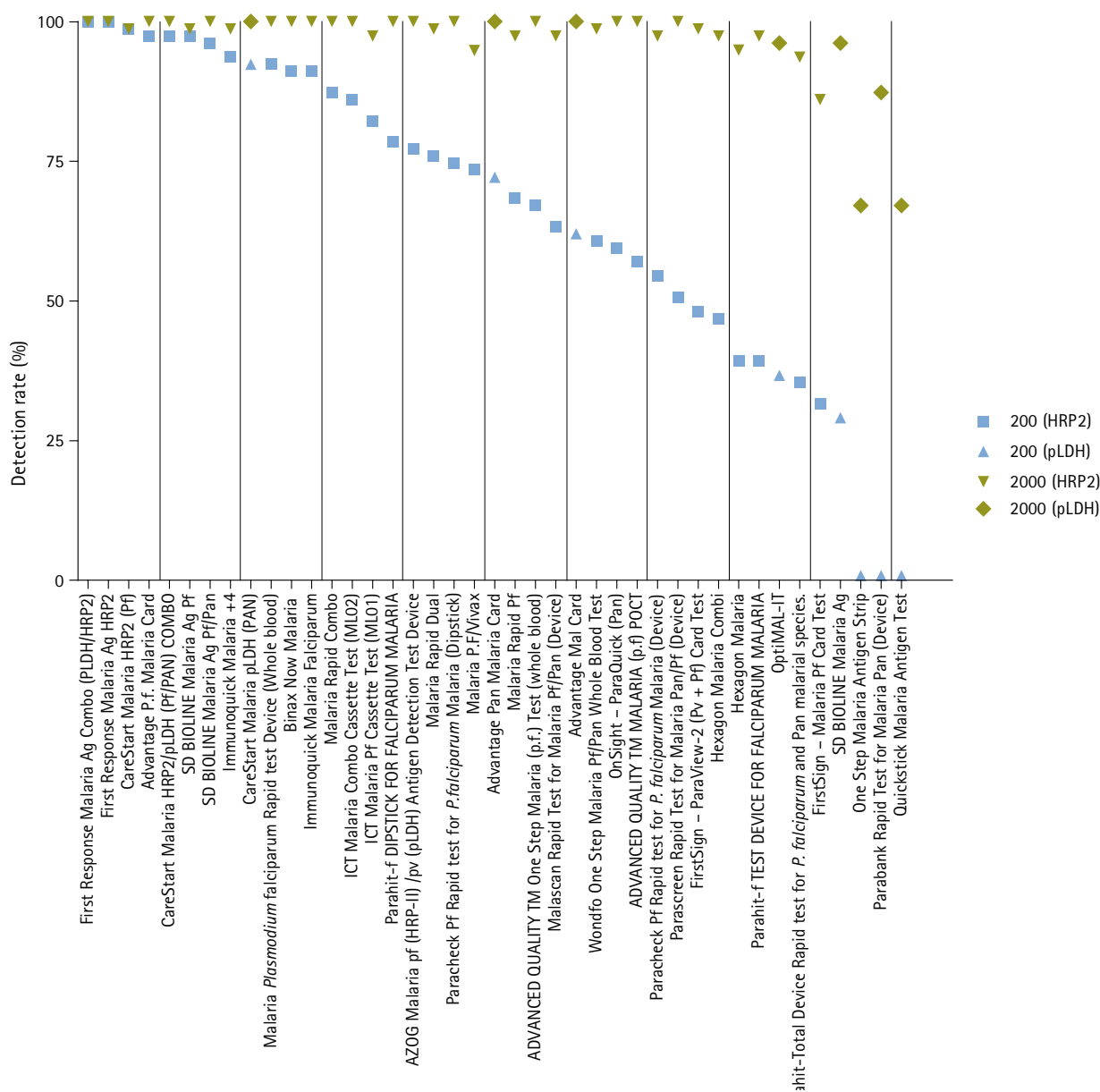
10.3.1. *P. falciparum* detection rate

Compared to the *P. falciparum* cultured parasite panel, *P. falciparum* detection rates of wild type samples were generally higher, reflecting the increased antigen content of wild type samples. As in Phase 1, the majority of tests (35; 85%) detected $\geq 95\%$ of *P. falciparum* samples at high

parasite densities but only 6 tests (15%) had this high level of detection at low parasite density (200 parasites/ μ l). All of these 6 products targeted HRP2; however 3 products targeting pLDH detected $> 60\%$ of *P. falciparum* samples. (Figure 8).

Table 4a illustrates detection rates at low and high parasite densities based on a delayed second reading if it was specifically indicated in the package insert following initial negative results at the specified reading time. At the low parasite density (200 parasites/ μ l), the *P. falciparum* detection rate increased for 5 of the 7 products, based on the delayed reading.

Figure 8: Phase 2 *P. falciparum* detection rate of malaria RDTs at low (200) and high (2000 or 5000) parasite density (parasites/ μ l) according to target antigen type (HRP2 or pLDH)[†]



[†] - Phase 2 Evaluation Panel included 79 blood samples containing wild type *P. falciparum*. RDTs - 2 tests x 2 lots at 200 parasites/ μ l and 1 test x 2 lots at 2000 parasites/ μ l

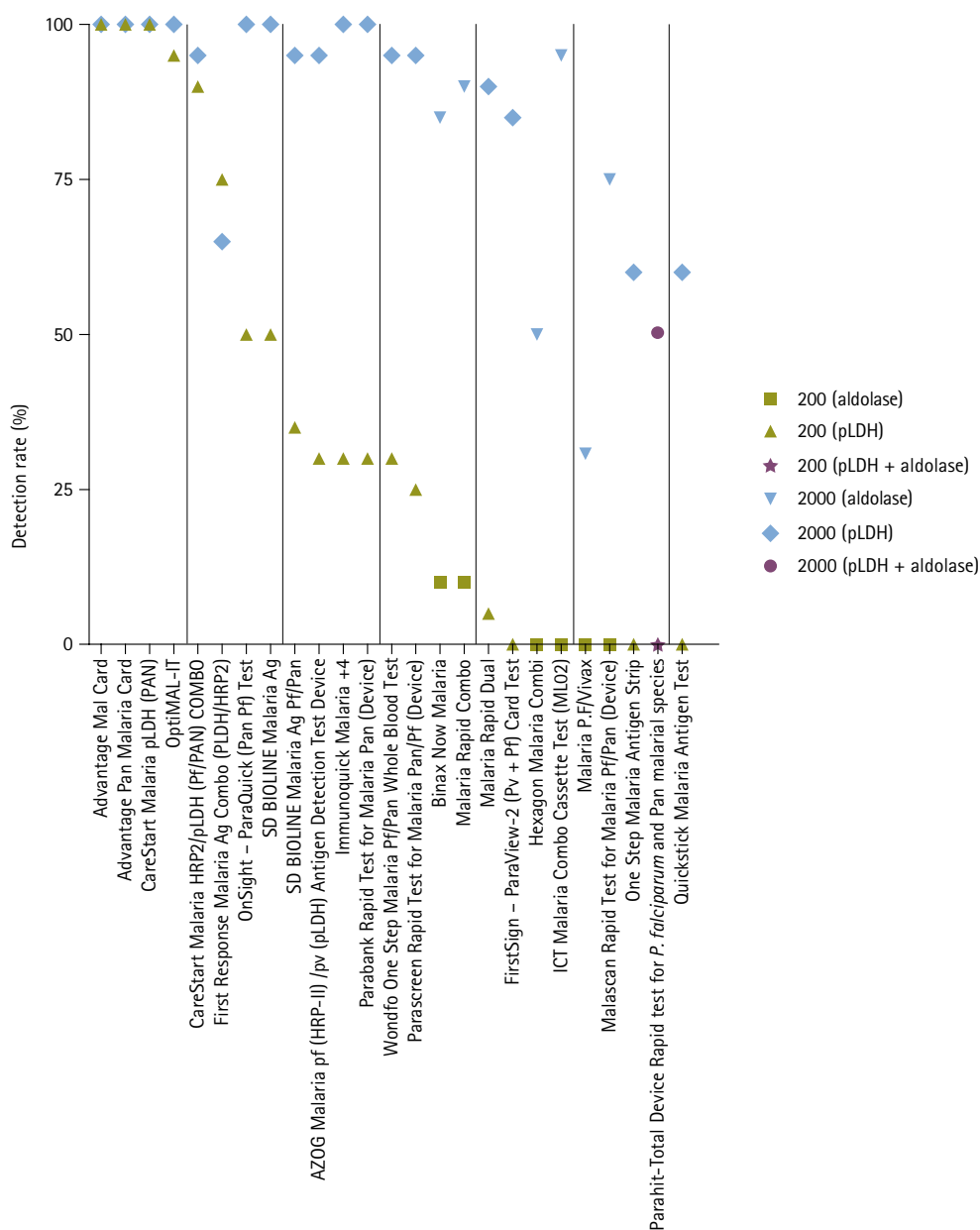
10.3.2. *P. vivax* detection rate

P. vivax was generally less well detected than *P. falciparum* in clinical samples. As Figure 9 illustrates, even at high parasite densities (2000 or 5000 parasites/μl) several products failed to consistently detect clinical samples infected with *P. vivax* samples and detection rates dropped at low parasite density, with only 5 products (25%) detecting > 90% of samples. Products targeting pLDH performed better than those targeting aldolase. One product had a higher detection rate at 200 parasites/μl compared to 2000 parasites/μl because false positive *P. falciparum* specific test lines at 2000 parasites/μl were counted as incorrect or negative results.

10.3.3. Combined detection of *P. falciparum* and *P. vivax*

Considering combination tests, 3 products had detection rates > 60% for both *P. falciparum* and *P. vivax* at the low parasite density (200 parasites/μl) (Tables 4. 4a). However, several performed well at high parasite densities. Two pan-specific tests showed high detection rates of both *P. falciparum* and *P. vivax*.

Figure 9: Phase 2 *P. vivax* detection rate of malaria RDTs at low (200) and high (2000 or 5000) parasite densities (parasites/μl) according to target antigen type (aldolase, pLDH, aldolase + pLDH)[†]



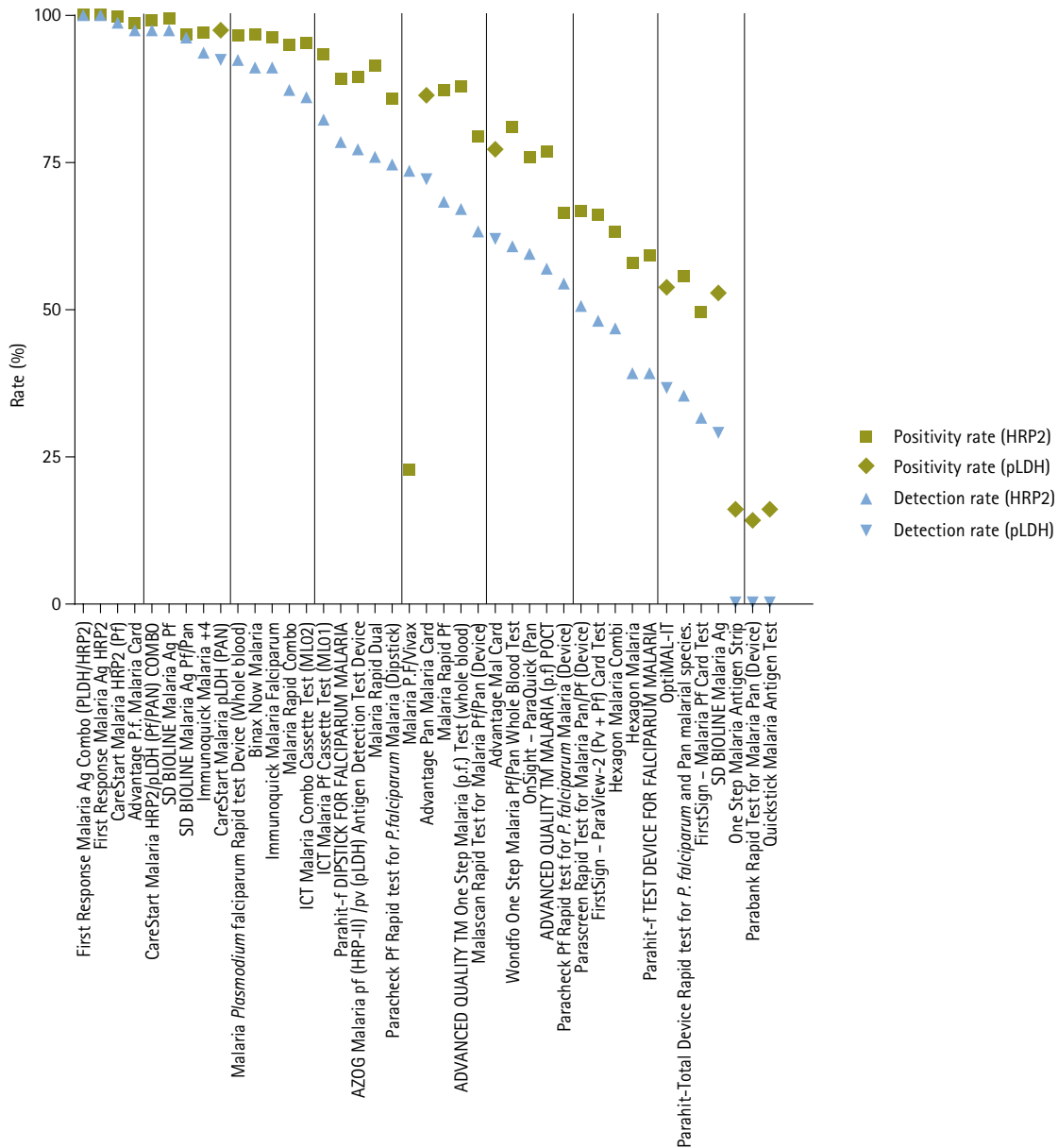
[†] - Phase 2 Evaluation Panel included 20 blood samples containing wild type *P. vivax*. RDTs - 2 tests x 2 lots at 200 parasites/μl and 1 test x 2 lots at 2000 parasites/μl

10.3.4. *P. falciparum* and *P. vivax* positivity rate

In addition to detection rate, positivity rate was also measured. This puts aside test and lot differences captured in detection rate and simply measures the total number of times a test returned a positive result. As expected,

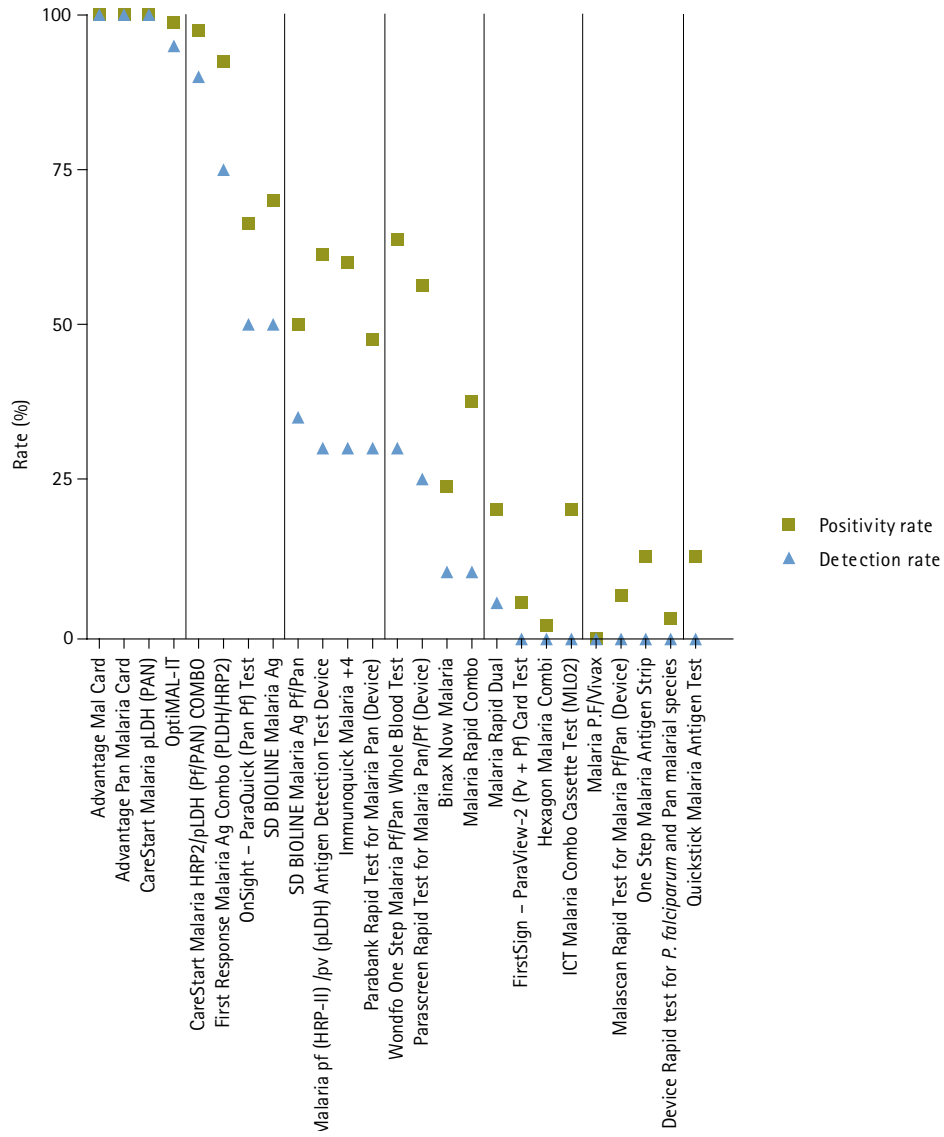
positivity rate was higher but mirrored detection rate of wild type *P. falciparum* and *P. vivax* samples (Figs 10, 11) with one exception. In this exceptional case, the product had a very high invalid rate against *P. falciparum* samples (Table 4) and invalid test results were included in the calculation of positivity rate but excluded from detection rate.

Figure 10: Phase 2⁺ wild type *P. falciparum* detection rate and positivity rate at 200 parasites/ μ l



*- Phase 2 Evaluation Panel included 79 blood samples containing wild type *P. falciparum*. RDTs - 2 tests x 2 lots at 200 parasites/ μ l and 1 test x 2 lots at 2000 parasites/ μ l

Figure 11: Phase 2⁺ wild type *P. vivax* detection rate and positivity rate at 200 parasites/ μ l



⁺ Phase 2 Evaluation Panel included 20 blood samples containing wild type *P. vivax*. RDTs - 2 tests x 2 lots at 200 parasites/ μ l and 1 test x 2 lots at 2000 parasites/ μ l

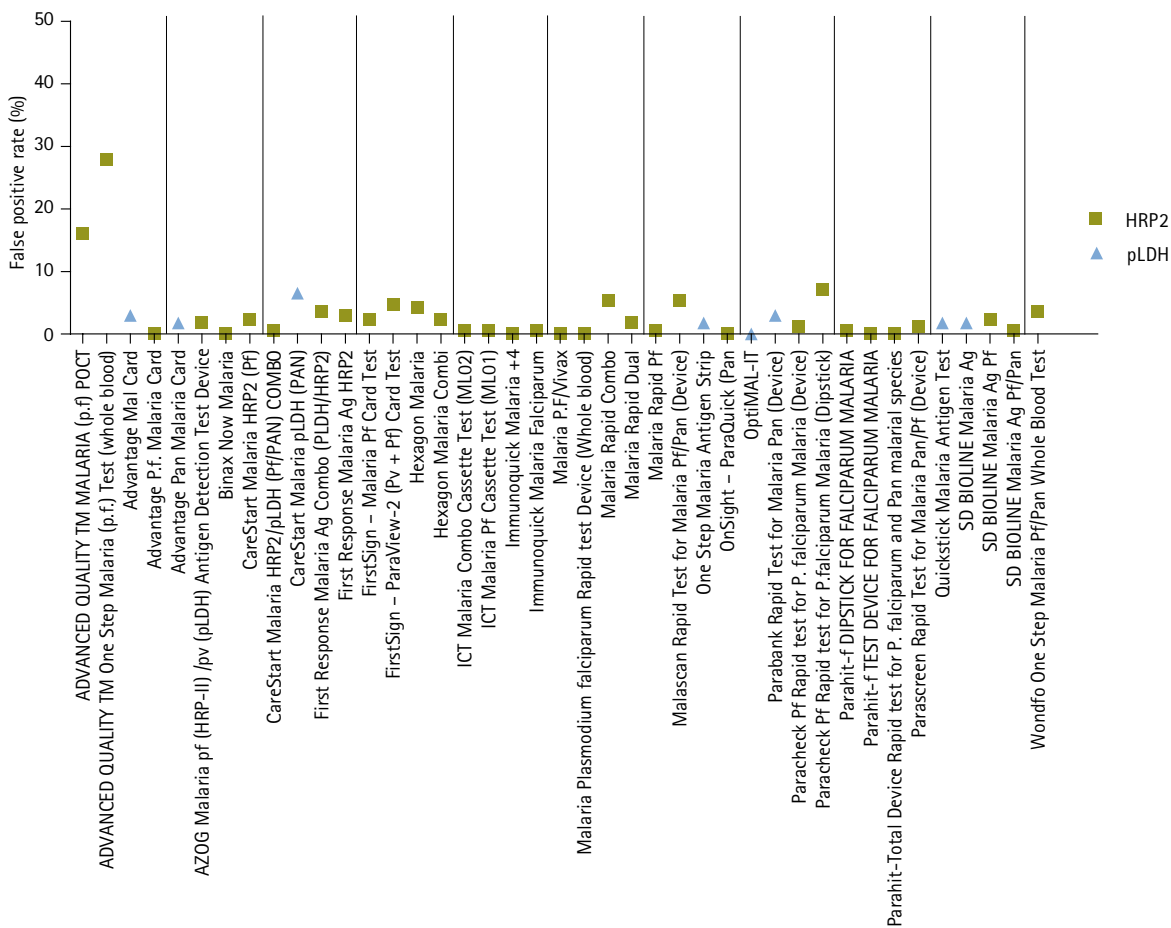
10.3.5. Band intensity

Although RDTs are not quantitative, technicians did grade positive results according to a standard chart and mean band intensity (for positive results) was calculated (Annex 4 - Tables A4.4, A4.5). There was a positive correlation between detection rate and band intensity and reader agreement; suggesting that, as expected, strong test bands are interpreted more reliably.

10.3.6. False positive rates

Overall false positive rates were modest, rarely > 10% on parasite-free blood bank specimens or *Plasmodium* spp. negative samples containing potentially cross-reacting factors (Figs 12-15). False positive results were seen on both *P. falciparum* and pan/*P. vivax* test lines. No consistently higher rate of false positive was seen with blood abnormalities compared to infectious pathogens; however, sample sizes were small. For detailed information regarding the blood abnormality or pathogen that generated false positive results for a specific product refer to Annex 4 (Tables A4.10-A4.12).

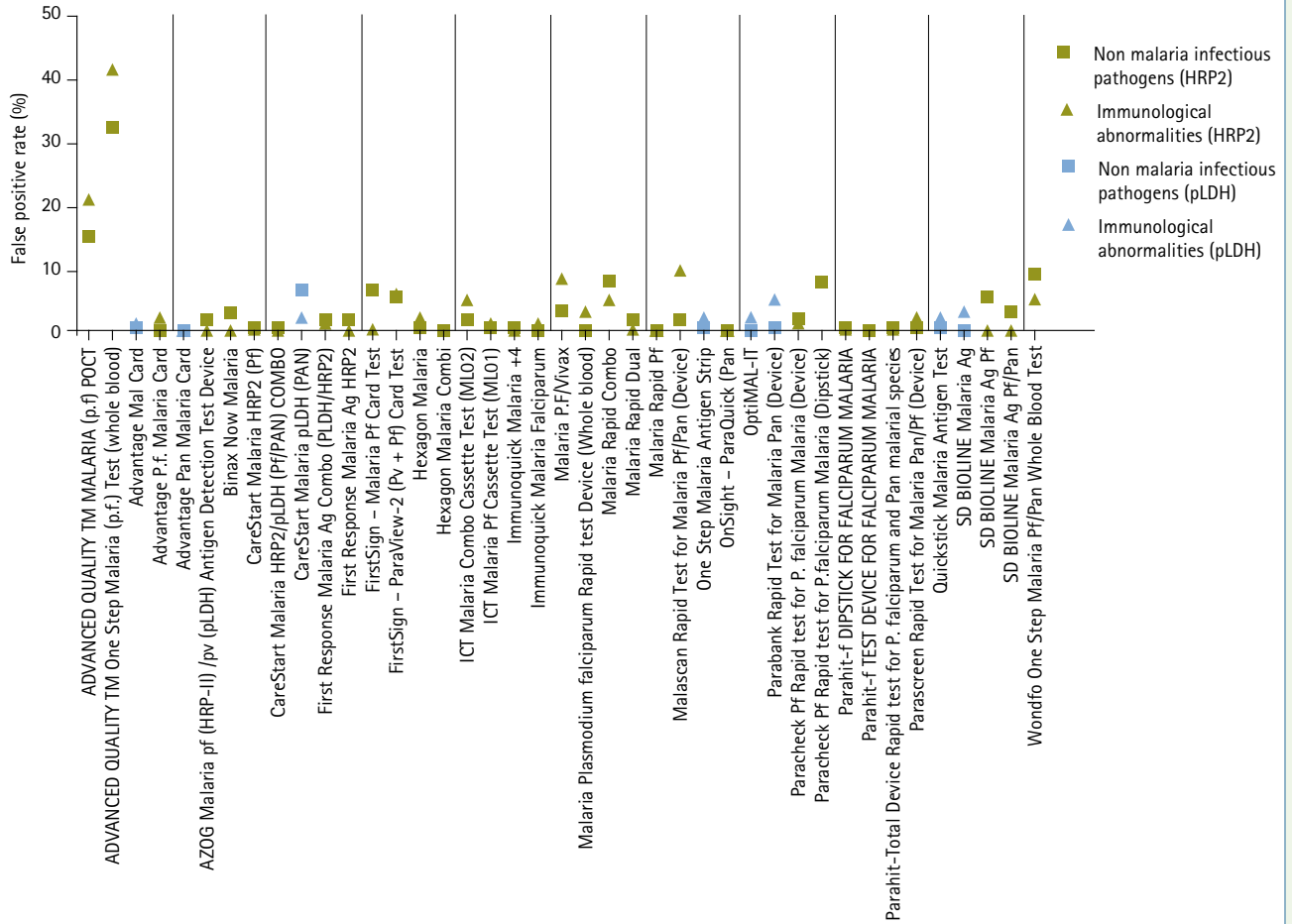
Figure 12: *P. falciparum* (*P. falciparum* test line*) false positive rate against clean negative samples[†]



[†] - Phase 2 Evaluation Panel included 90 *Plasmodium* spp. negative samples of which 42 were "clean" negatives. RDTs - 2 tests x 2 lots at 200 parasites/μl and 1 test x 2 lots at 2000 parasites/μl

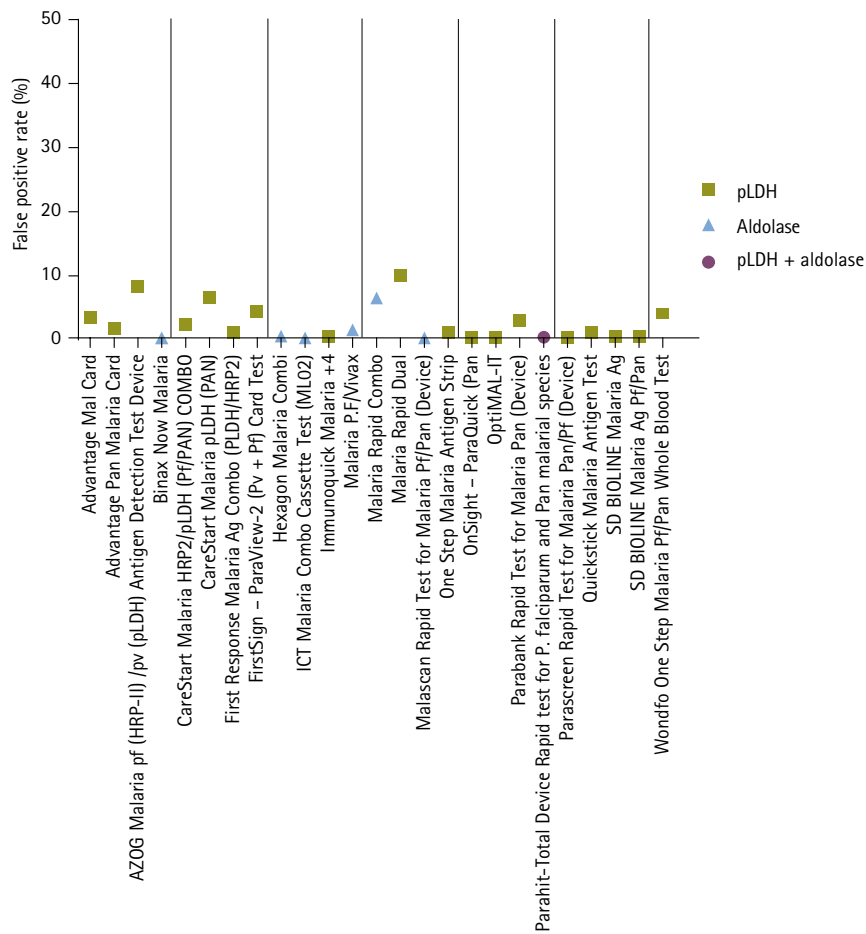
* - includes Pf specific lines on combination tests and pan-specific lines on pan-specific only products.

Figure 13: *P. falciparum* (*P. falciparum* test line*) false positive rate against *Plasmodium* spp. negative samples containing potentially cross-reacting blood immunological abnormalities or infectious pathogens[†]



[†] - Phase 2 Evaluation Panel included 90 *Plasmodium* spp. negative samples - 42 clean negatives; 27 samples with blood immunological abnormalities and 21 with non *Plasmodium* spp. infectious pathogens. RDTs - 2 tests x 2 lots at 200 parasites/ μ l and 1 test x 2 lots at 2000 parasites/ μ l
^{*} - includes Pf specific lines on combination tests and pan-specific lines on pan-specific only products.

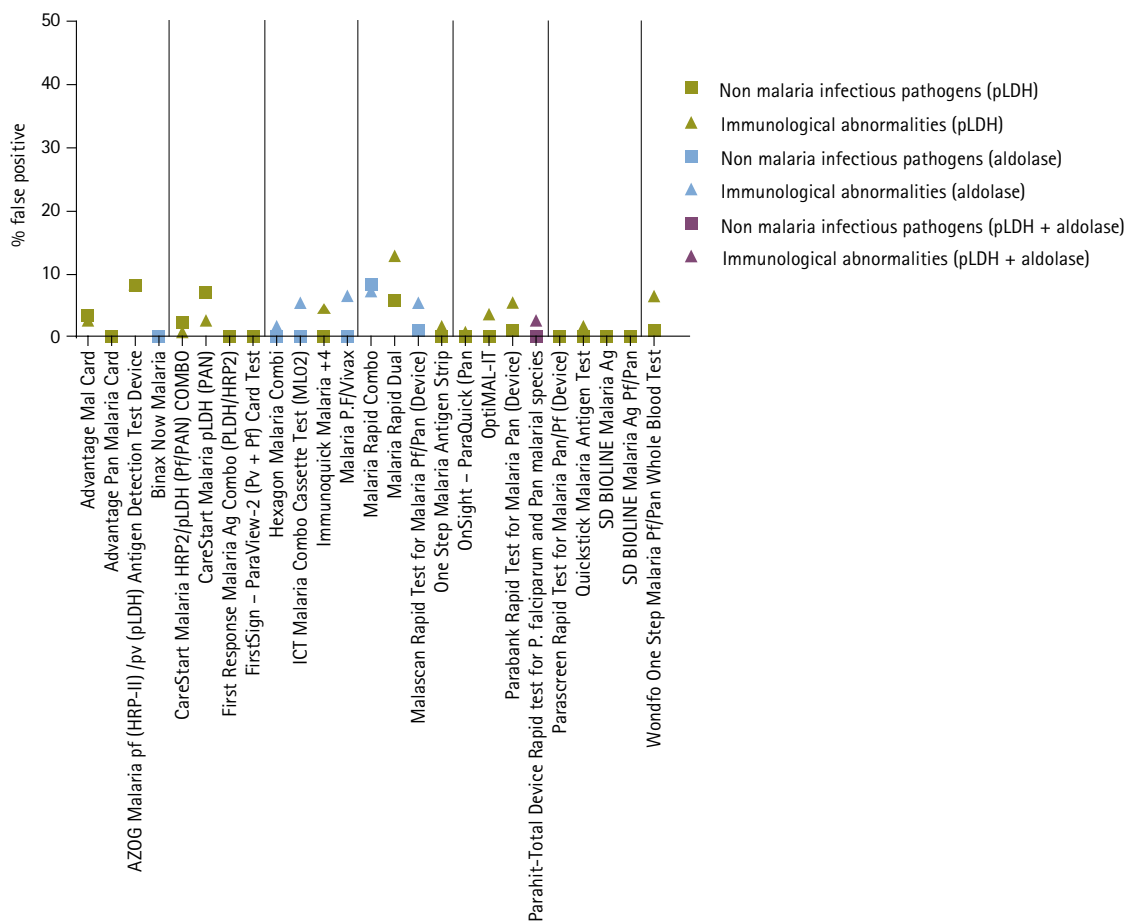
Figure 14: *Plasmodium* spp. (pan or *P. vivax* test line*) false positive rate against clean negatives[†]



[†]- Phase 2 Evaluation Panel included 90 *Plasmodium* spp. negative samples - 42 clean negatives; 27 samples with blood immunological abnormalities and 21 with non *Plasmodium* spp. infectious pathogens. RDTs - 2 tests x 2 lots at 200 parasites/ μ l and 1 test x 2 lots at 2000 parasites/ μ l

*- includes pan of pv line on combination tests and pan-specific line on pan-specific only products.

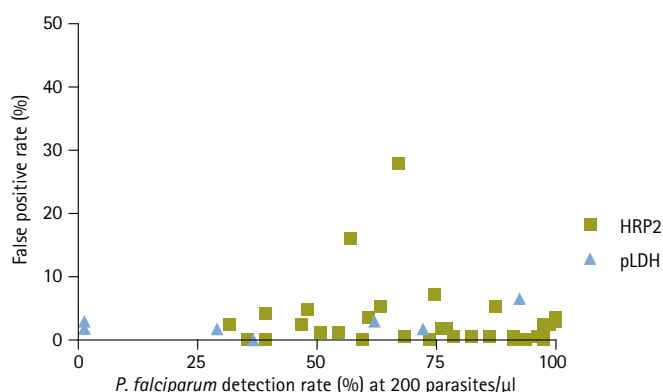
Figure 15: *Plasmodium* spp. (pan or *P. vivax* test line*) false positive rate against *Plasmodium* spp. negative samples containing potentially cross-reacting blood immunological abnormalities or infectious pathogens[†]



[†]- Phase 2 Evaluation Panel included 90 *Plasmodium* spp. negative samples - 42 clean negatives; 27 samples with blood immunological abnormalities and 21 with non *Plasmodium* spp. infectious pathogens. RDTs - 2 tests x 2 lots at 200 parasites/μl and 1 test x 2 lots at 2000 parasites/μl
^{*}- includes pan of pv line on combination tests and pan-specific line on pan-specific only products.

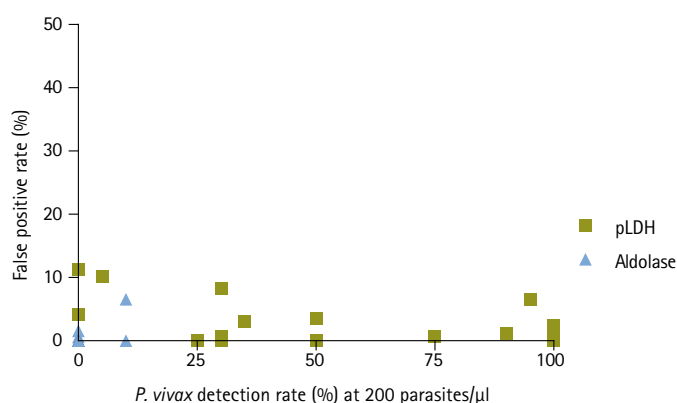
Figure 16 and 17 illustrates that high rates of detection of *P. falciparum* or of *P. vivax* were not associated with a significant overall increase in false positive rates.

Figure 16: *P. falciparum* false positive rate¹⁹ versus *P. falciparum* detection rate at low (200) parasite density (parasites/ μ l)



19 On the clean negative panel, only.

Figure 17: *P. vivax* false positive rate versus *P. vivax* detection rate at low (200) parasite density (parasites/ μ l)



10.3.7. Inter-reader variability

Each RDT result was read twice, the first, according to the manufacturers minimum reading time and a second reading was performed by a different technician within one hour. The mean time between readings for each product can be found in Tables A4.2, A4.3 (Annex 4). Figures 18 and 19 illustrate that for many products, the second reading yielded more positive results. However, for two products Reader 2 results returned fewer positive results secondary

to the fading of weak test band signals. In the case of 7 products²⁰ manufacturers specifically indicated that a second reading should be performed if the initial results were negative. Results are therefore reported on the basis of the delayed reading in Tables 3a, 4a and in Annex 4 (Tables A4.1a, A4.4a-A4.12a)

20 Parascreen Rapid Test for Malaria (Pan/Pf) Device, Parabank Rapid Test for Malaria Pan (Device), Malascan Rapid Test for Malaria Pf/Pan (Device), ImmunoQuick Malaria + 4, Malaria Pf/vivax, Wondfo One Step Malaria Pf/Pan Whole Blood Test, FirstSign – ParaView-2 (Pv + Pf) Card Test.

Figure 18: Wild type *P. falciparum* detection rate according to first and second readings at 200 parasites/ μ l

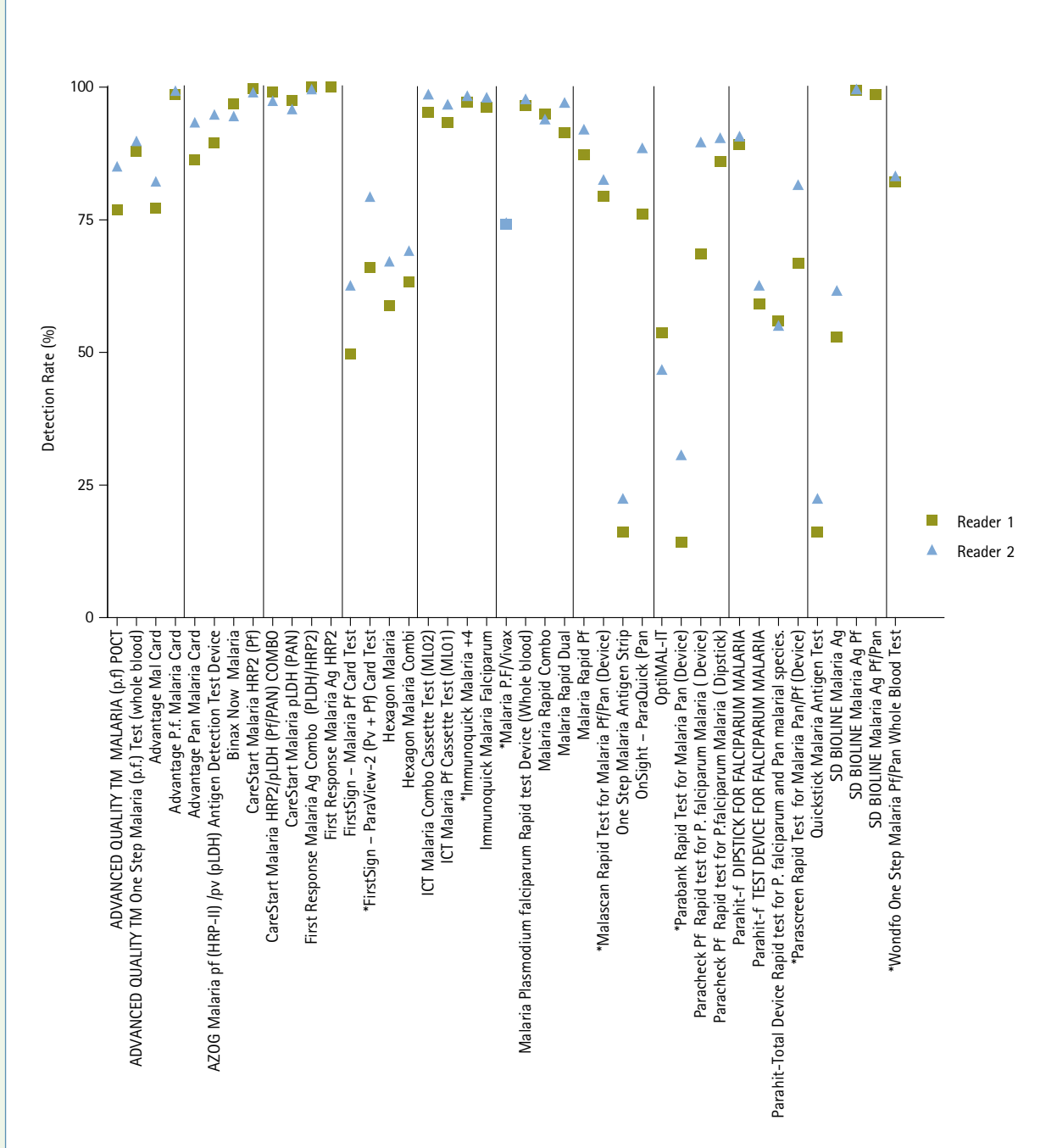
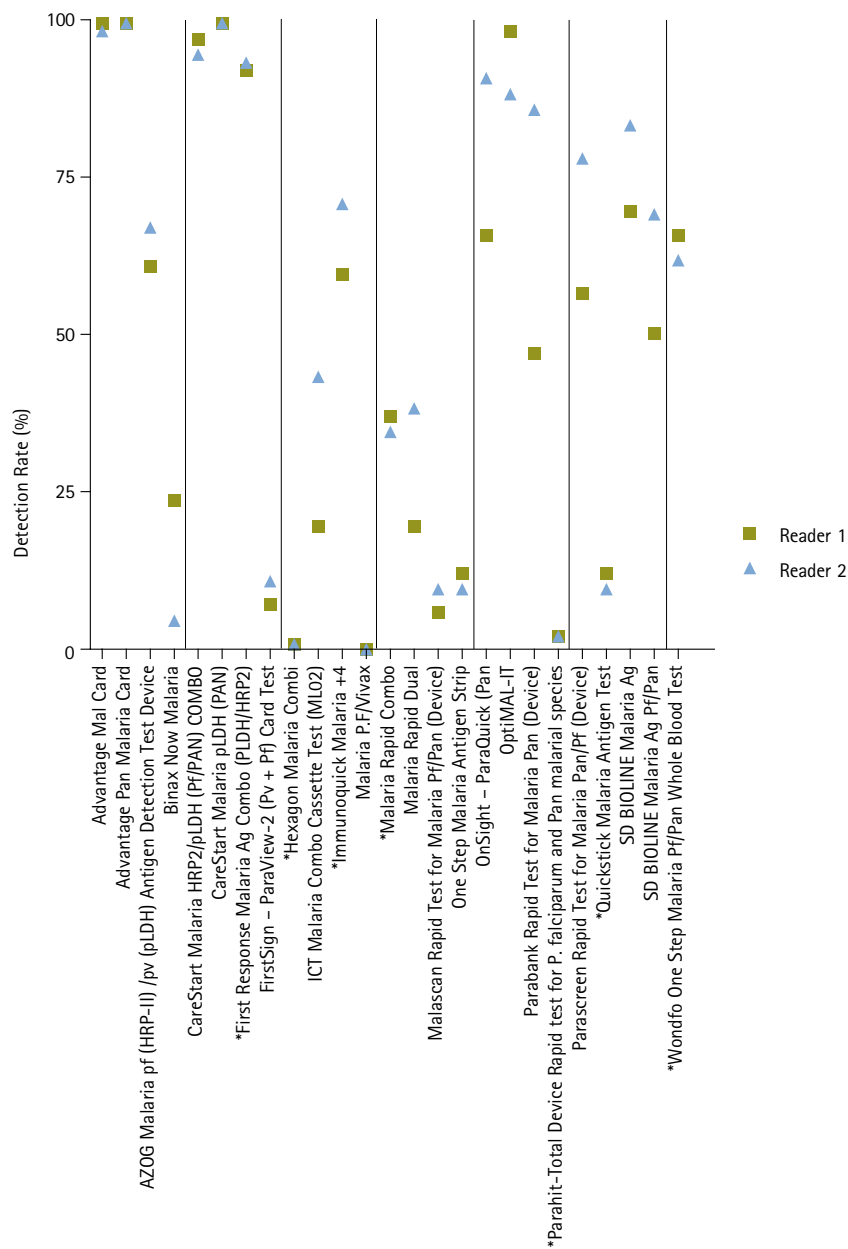


Figure 19: Wild type *P. vivax* detection rate according to first and second readings at 200 parasites/ μ l



11. HEAT STABILITY

A single *P. falciparum* culture sample was used as the reference sample for heat stability testing. Variations in baseline performance reflect inter-test variation as the sample at 200 parasites/ μ l was at the limit of detection of some products.

Several products were stable, meaning that they detected a *P. falciparum* cultured sample the same number of times at baseline and following incubation for 2 months (75% humidity) at 4°C, 35°C and 45°C. Detailed results are presented in Tables A4.13-A4.15 (Annex 4); in Table 5 and Figs 20-29, the results of both lots are combined (maximum score 20; 10 tests per lot).

Overall products (including HRP2 and pan test lines) were significantly more stable against samples with high (2000 or 5000) compared to low parasite densities, Figures 21, 23, 25 and Figures 20, 22, 24, respectively. For several, *P. falciparum* and combination tests, HRP2 test lines showed a high degree of stability at 35°C but several dropped off at 45°C. Some products showed an improved performance with incubation (Figs 22, 26). Baseline detection based on pan test lines was lower than baseline HRP2 test line results and the pan test lines of many products had poorer stability than HRP2 test lines.

Ultimately, there are products in each test category (Pf only, combination, pan only) that show good consistent baseline detection and heat stability up to 45°C for 2 months.

The summary results of heat/thermal stability testing are presented in Table 5.

11.1. *P. FALCIPARUM* TEST LINES

Figure 20: Heat stability of *P. falciparum* specific (HRP2) test line of *P. falciparum* only tests against a low density *P. falciparum* sample (200 parasites/ μ l). Positivity rate at baseline, and after 60 days incubation.

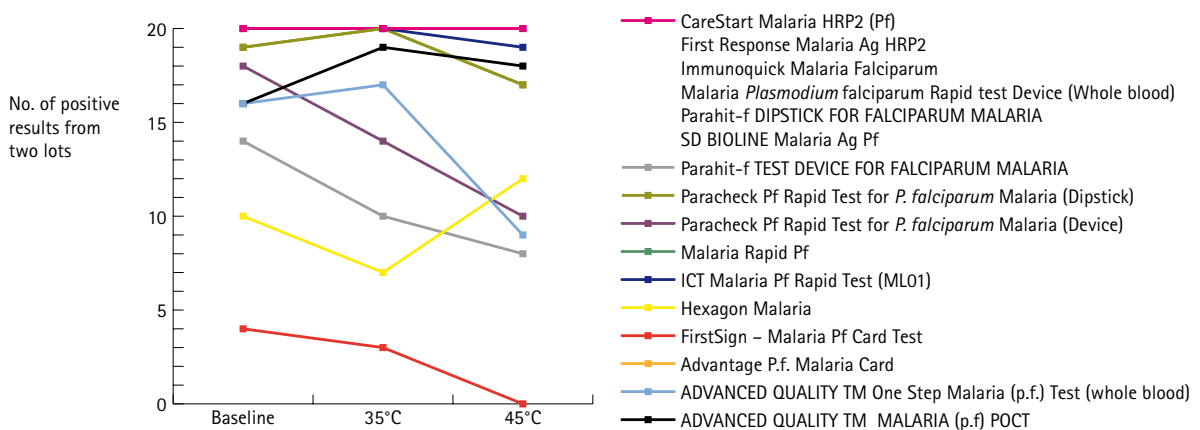


Figure 21: Heat stability of *P. falciparum* (HRP2) test specific line of *P. falciparum* tests against a high density *P. falciparum* sample (2000 parasites/ μ l). Positivity rate at baseline, and after 60 days incubation.

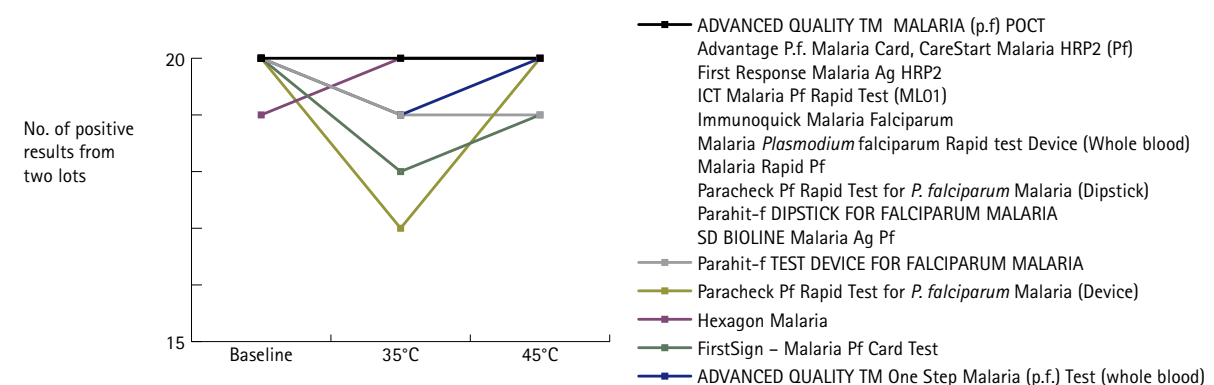


Figure 22: Heat stability of *P. falciparum* specific (HRP2) test line in combination tests against a low density *P. falciparum* sample (200 parasites/ μ l). Positivity rate at baseline, and after 60 days incubation.

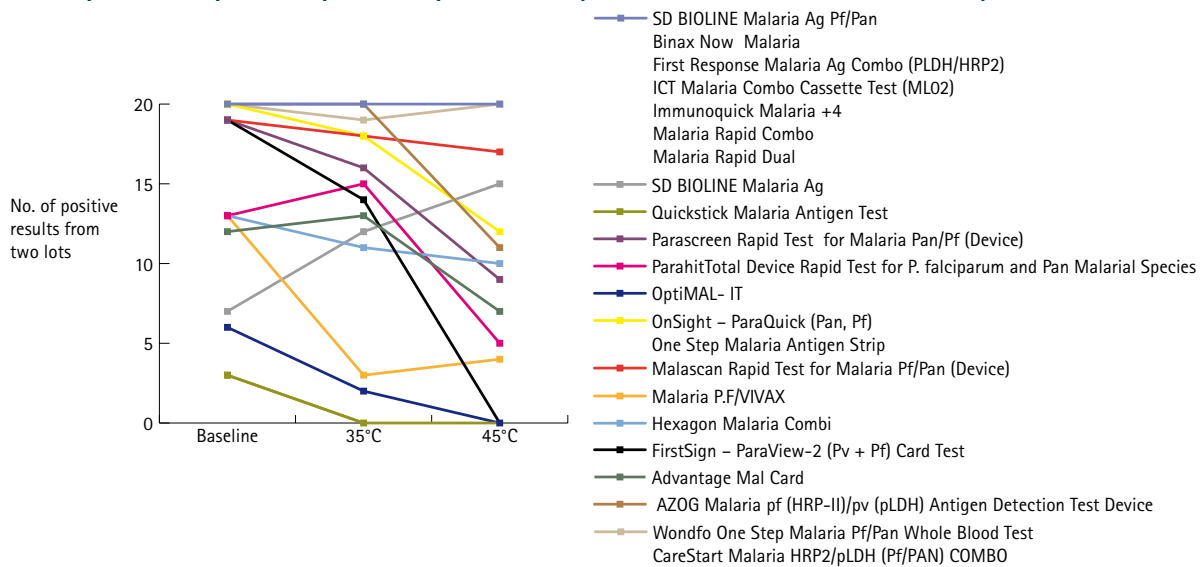


Figure 23: Heat stability of *P. falciparum* (HRP2) test specific line in combination tests against a high density *P. falciparum* sample (2000 parasites/ μ l). Positivity rate at baseline, and after 60 days incubation.

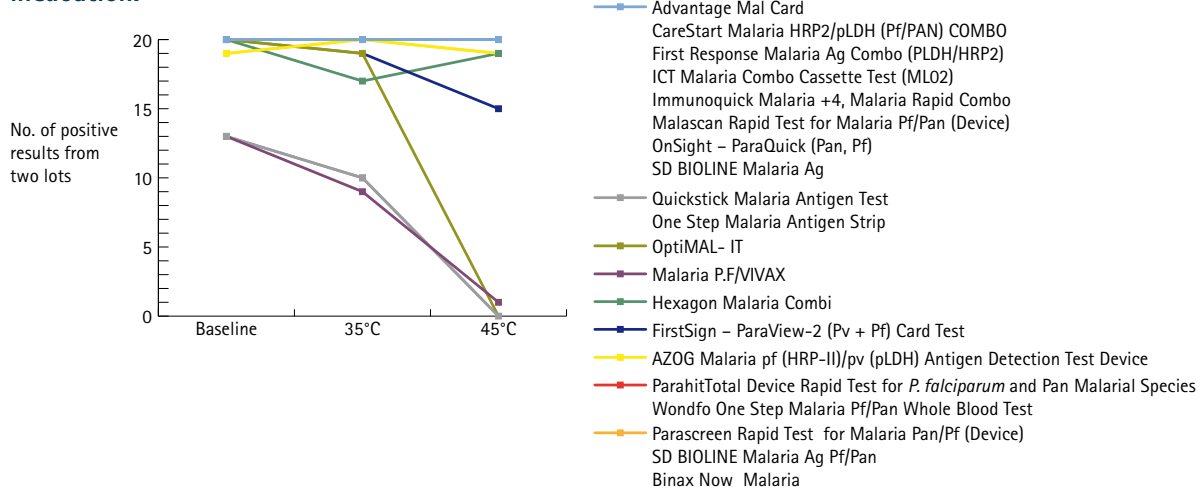


Figure 24: Heat stability of pan-line of pan-specific tests against a low density *P. falciparum* sample (200 parasites/ μ l). Positivity rate at baseline, and after 60 days incubation.

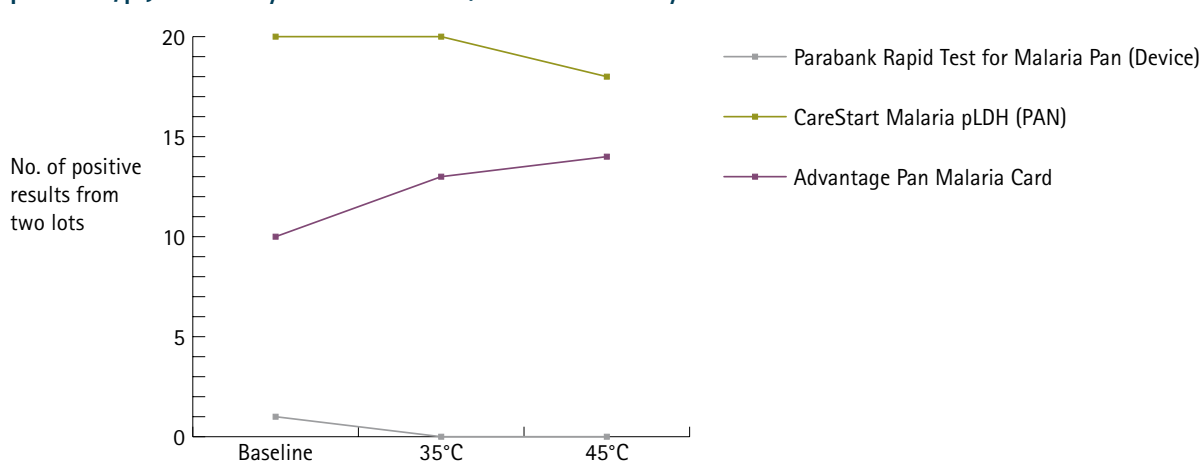
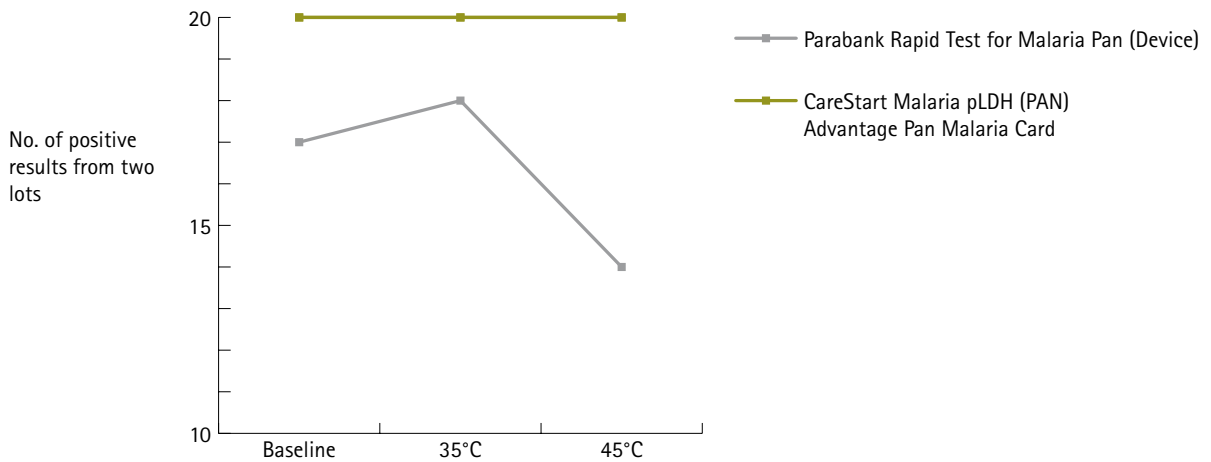


Figure 25: Heat stability of pan-line of pan-specific tests against a high density *P. falciparum* sample (2000 parasites/ μ l). Positivity rate at baseline, and after 60 days incubation.



11.2. PAN TEST LINES

Figure 26: Heat stability of pan-line of combination tests against a low density *P. falciparum* sample (200 parasites/ μ l). Positivity rate at baseline, and after 60 days incubation.

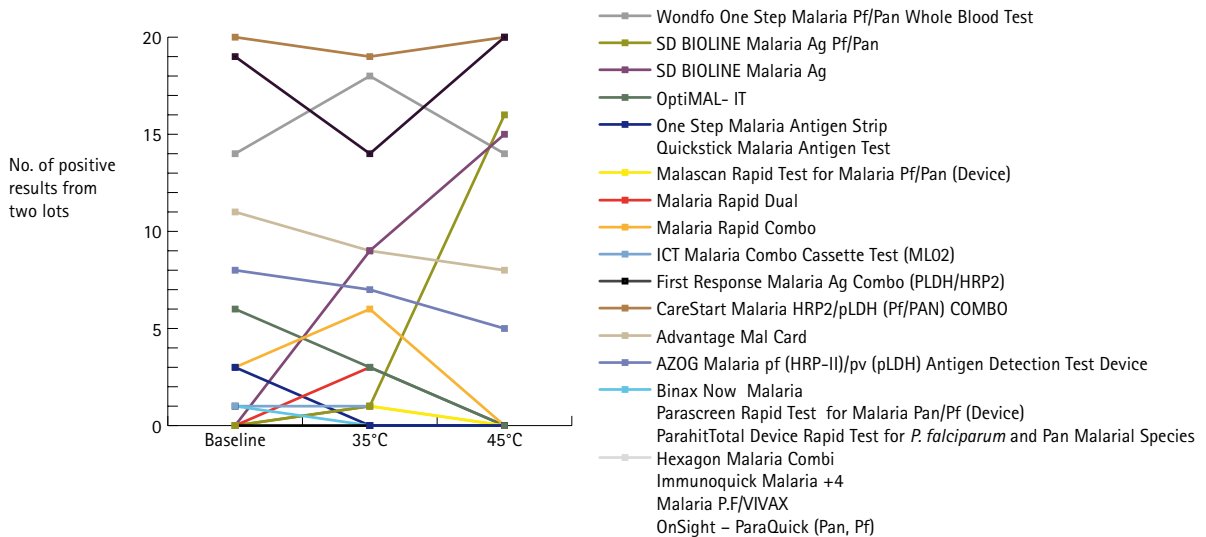


Figure 27: Heat stability of pan-line of combination tests against a high density *P. falciparum* sample (2000 parasites/ μ l). Positivity rate at baseline, and after 60 days incubation.

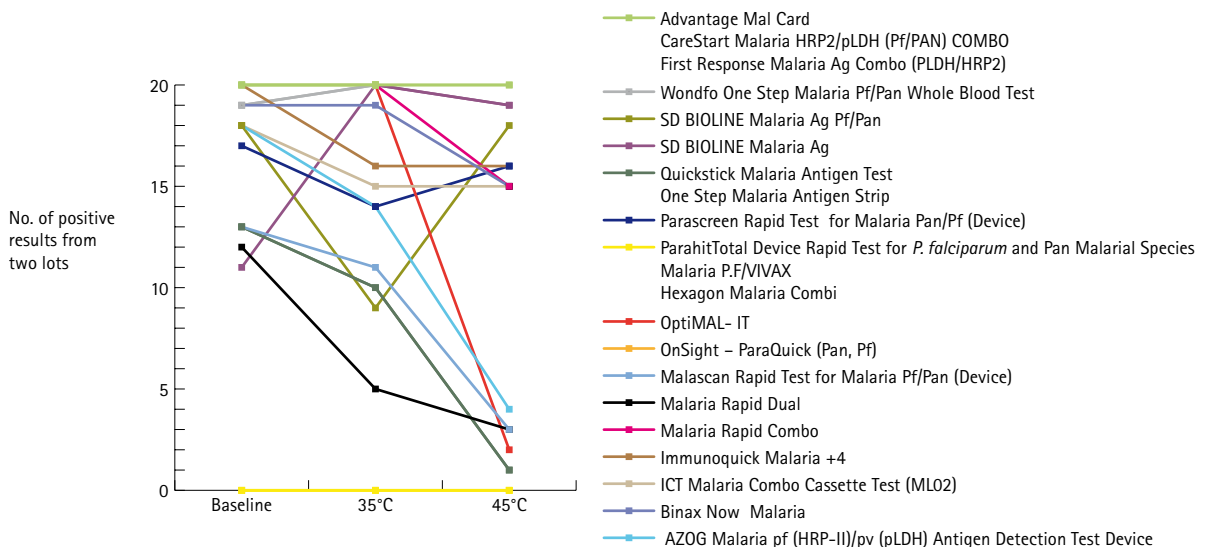


Figure 28: Heat stability of pan-line of pan-specific tests only against a low density *P. falciparum* sample (200 parasites/ μ l). Positivity rate at baseline, and after 60 days incubation.

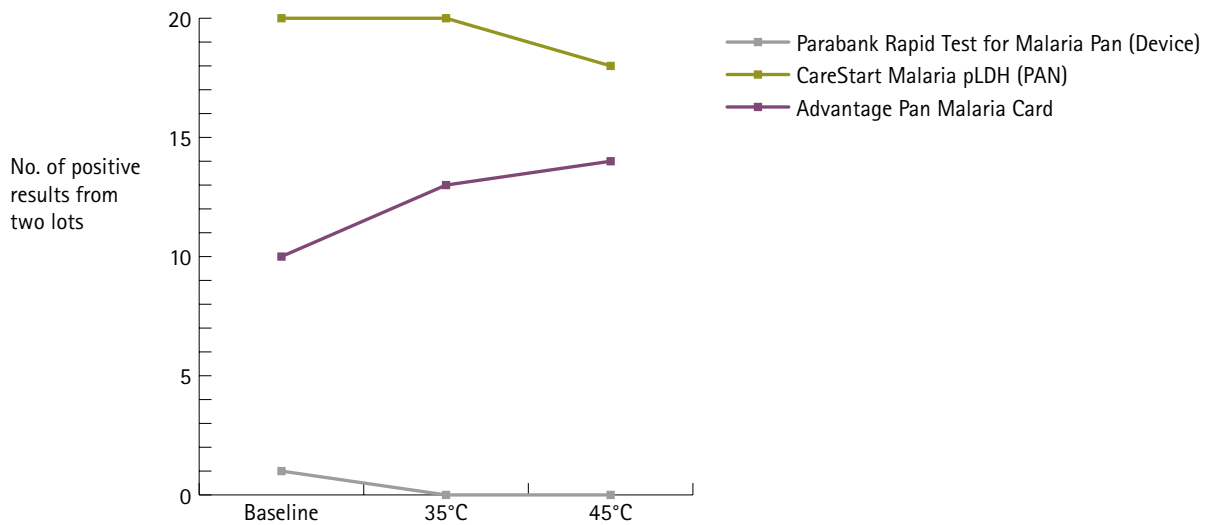
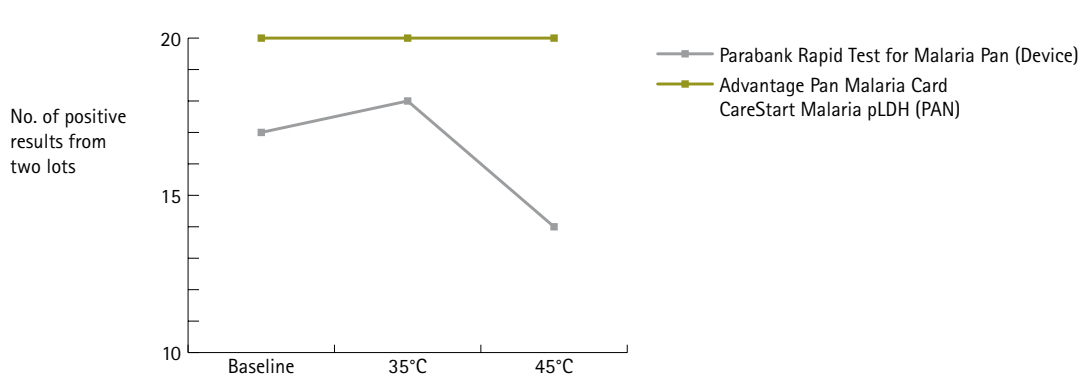


Figure 29: Heat stability of pan-line of pan-specific tests only against a high density *P. falciparum* sample (2000 parasites/ μ l). Positivity rate at baseline, and after 60 days incubation.



12. EASE OF USE DESCRIPTION

After becoming proficient at using a product, two technicians jointly produced an agreed assessment of product usability. The results are presented in Table 6.

Table 6: Ease of use description of 41 malaria rapid diagnostic tests

Product	Manufacturer	Blood safety*			Score (max. 3)	Diagram of result & method	Score (max.2)	Combined score (max. 5)	Number of timed steps	Total time to result	Blood transfer device	Format	Language of instruction	Items included in package*
		Mixing wells involved	Retractable needle	Strip exposed										
PF only														
ADVANCED QUALITY™ MALARIA (p.f.) POC	InTec Products, Inc.	1	1	1	3		4	1	15:00	pipette	cassette	English	test, buffer, lancet, pipette, alcohol swab	
ADVANCED QUALITY™ One Step Malaria (p.f.) Test (whole blood)	InTec Products, Inc.	1	1	1	3		4	1	15:00	pipette	cassette	English	test, buffer, lancet, pipette, alcohol swab	
Advantage P.f. Malaria Card	J. Mitra & Co. Pvt. Ltd.	1	0	1	2		4	1	20:00	loop	cassette	English	test, buffer, loop, lancet, alcohol swab	
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	1	0	1	2		4	1	20:00	capillary	cassette	English	test, buffer, swab, lancet, capillary	
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	1	0	1	2		4	1	20:00	capillary	cassette	English	test, buffer, lancet, pipette, alcohol swab	
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	1	0	1	2		3	1	15:00	loop	cassette	English	test, buffer, loop	
Hexagon Malaria	Human GmbH	1	0	1	2		3	1	15:00	capillary	cassette	English, German	test, buffer, capillary	
ICT Malaria Pf Cassette Test (ML01)	ICT Diagnostics	1	0	1	2		4	1	15:00	pipette	cassette	English	test, buffer, lancet, pipette, alcohol swabs	
Immunorquick Malaria Falciparum	Biosynex	1	0	0	1		3	1	10:00	none	dipstick	English, French	test, buffer, test tubes, rack	
Malaria Plasmodium falciparum Rapid test Device (Whole blood)	ACON Laboratories, Inc.	0	0	1	1		3	2	16:00	pipette	cassette	English	test, buffer, well, pipette	
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	1	0	1	2		4	1	15:00	pipette	cassette	English	test, buffer, lancet, alcohol swabs, pipette	
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	1	0	1	2		3	1	15:00	loop	cassette	English	test, buffer, loop	
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	1	0	0	1		2	1	15:00	loop	dipstick	English	test, buffer, loop	
Parahi-t-f DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	1	0	0	1		3	1	15:00	capillary	dipstick	English	test, buffer, capillary, lancet, alcohol swabs	
Parahi-t-f TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	1	0	1	2		4	1	15:00	capillary	cassette	English	test, buffer, capillary, alcohol swab, lancet	
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	1	0	1	2		4	1	15:00	loop	cassette	English	test, buffer, alcohol swab, lancet, loop	
Pf & Pan/Pf & Pv														
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	1	0	1	2		4	1	20:00	loop	cassette	English	test, buffer, loop, lancet, alcohol swabs	
AZOG Malaria pf (HRP-II) / pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	1	0	1	2		4	1	20:00	none	cassette	English	test, buffer	
Binx Now Malaria	Inverness Medical Innovations, Inc.	1	0	1	2		4	2	~16:00	capillary	card	English, Italian, Portuguese, French, German, Swedish, Dutch, Greek, Norwegian, Danish	test, buffer, capillary	
CareStart Malaria HRP2/ pLDH (Pf/PAN) COMBO	Access Bio, Inc.	1	0	1	2		4	1	20:00	capillary	cassette	English	test, buffer, swab, lancet, capillary	
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	1	0	1	2		4	1	20:00	capillary	cassette	English	test, buffer, lancet, pipette, alcohol swab	
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	1	0	1	2		3	1	15:00	loop	cassette	English	test, buffer, loop	
Hexagon Malaria Comb	Human GmbH	1	0	1	2		3	1	15:00	capillary	cassette	English, German	test, buffer, capillary	

Product	Manufacturer	Mixing wells involved	Blood safety [†]			Instruction quality [*]			Number of timed steps	Total time to result	Blood transfer device	Format	Language of instruction	Items included in package [*]
			Retractable needle	Strip Exposed	Score (max. 3)	No diagram	Diagram of result	Diagram of result & method						
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	1	0	1	2			4	15:00	pipette	cassette	English	test, buffer, lancet, pipette, alcohol swabs	
Immunoquick Malaria +4	Biosynex	1	0	0	1			3	10:00	none	dipstick	English, French	test, buffer, test tubes, test rack	
Malaria PFiVivax	Diagnostic Automation / Cortez Diagnostics, Inc.	1	0	1	2			4	15:00	none	cassette	English	test, buffer	
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	1	0	1	2			4	15:00	pipette	cassette	English	test, buffer, lancet, alcohol swabs, pipette	
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	1	0	1	2			4	15:00	pipette	cassette	English	test, buffer, lancet, alcohol swabs, pipette	
Malascan Rapid Test for Malaria PfiPan (Device)	Zephyr Biomedicals	1	0	1	2			3	15:00	loop	cassette	English	test, buffer, loop, alcohol swab, lancet	
One Step Malaria Antigen Strip	IND Diagnostic Inc.	0	0	0	0			2	19:00	pipette	dipstick	English	test, buffer, wells, pipette	
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	1	0	1	2			3	15:00	loop	cassette	English	test, buffer, loop	
OptiMAL-IT	DiaMed AG	0	0	0	0			2	22	capillary	hybrid	English, Spanish, French, Portuguese, German, Italian	test, buffer, lancet, capillary	
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species	Span Diagnostics Ltd.	1	0	1	2			4	15:00	capillary	cassette	English	test, buffer, capillary, alcohol swab, lancet	
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	1	0	1	2			3	15:00	loop	cassette	English	test, buffer, loop, alcohol swab, lancet	
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	0	0	0	0			2	19:00	pipette	dipstick	English	test, buffer, wells, pipette	
SD BIOLINE Malaria Ag PfiPan	Standard Diagnostics, Inc.	1	0	1	2			4	15:00	loop	cassette	English	test, buffer, alcohol swab, lancet, loop	
SD BIOLINE Malaria Ag PfiPan	Standard Diagnostics, Inc.	1	0	1	2			4	15:00	loop	cassette	English	test, buffer, alcohol swab, lancet, loop	
Wondfo One Step Malaria PfiPan, Whole-Blood Test	Guangzhou Wondfo Biotech Co., Ltd	1	1	1	3			5	15:00	none	cassette	English	test, buffer, alcohol swab, lancet	
Pan only														
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	1	0	1	2			4	20:00	loop	cassette	English	test, buffer, loop, lancet, alcohol swab	
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	1	0	1	2			4	20:00	capillary	cassette	English	test, buffer, swab, lancet, capillary	
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	1	0	1	2			3	15:00	loop	cassette	English	test, buffer, loop, alcohol swab, lancet	

* - These are not necessarily standard contents. Procurers should verify what materials accompany test kits with the manufacturer and ensure they procure all the required accessories at the same time.

+ - Retractable needle Yes=1; No=0
+ - Mixing wells involved Yes=0; No=1
+ - Strip Exposed, not within card or cassette Exposed=0; Covered=1
+ - No diagrams =0
Diagrams of result =1
Diagrams of result and method=2

13. DISCUSSION OF KEY FINDINGS

This report describes the performance of many of the available malaria antigen-detecting RDTs manufactured under the ISO-13485 quality standard. Malaria RDTs have the potential to provide a huge step forward in the management of febrile illness in malaria-endemic areas. To be useful in this context, malaria RDTs must have adequate:

1. **sensitivity**, to detect nearly all clinically-significant cases of malaria;
2. **specificity**, to accurately discriminate non-malarial febrile illness from malaria, to ensure appropriate management and accurate disease monitoring;
3. **stability**, for accuracy to be maintained after transport and storage in ambient conditions;
4. **ease of use and safety**, to allow safe and correct preparation, and correct interpretation of results.

In order to assist national malaria control programmes, and other procurement agencies to select products appropriate to their needs, malaria RDTs were evaluated in terms of these four major requirements. The blood panel used successfully discriminated between the RDTs evaluated, showing a considerable range of performance. Importantly, a number of products demonstrated a high rate of antigen detection combined with a low false-positive rate and good heat (thermal) stability, attributes essential if they are to be relied on as a basis for malaria treatment decisions in most endemic populations.

The principal results in this report are presented in Tables 3, 3a, 4, 4a. The table groups the RDTs by type, depending on what they aim to detect, e.g. *P. falciparum* only, *P. falciparum* and non-falciparum species, or all malaria species without discrimination. Detection rates at both high and low concentrations are presented, as are false-positive rates, and the percentage of invalid test results. Tests in each category are listed alphabetically, but the results are colour-coded to assist the reader in quick interpretation of the data. These colour codes are intended to be used to quickly compare performance in the different categories and not as performance cut-offs to guide test selection or procurement. When choosing an appropriate product, it is important to also review the stability results (Table 5) in the context of the expected conditions of transport and storage of the RDTs in the field.

This evaluation is performed against a standardized panel of frozen blood samples by experienced technicians in a research laboratory and is not a field evaluation of RDT accuracy in a specific epidemiological context in the hands of intended users. The panel is designed to mimic fresh blood samples from real cases as closely as possible, while allowing direct comparison of a large number of products

simultaneously in a manner that controls for confounding factors and is calibrated to a level likely to discriminate performance differences of various products. In interpreting the results, it is therefore important that the following notes are taken into account.

13.1. PARASITE DETECTION AND ITS RELATIONSHIP TO SENSITIVITY

Evaluation of the RDTs against the parasite-positive panel with parasite densities of 200 parasites/ μ L (Figs 8, 9) revealed a wide range of detection rates between products. Testing at higher parasite densities (2000 or 5000 parasites/ μ L) was less able to demonstrate small differences in performance. As two tests each from two different lots were tested, and as all four results had to be positive for a sample to be considered *detected* by an RDT, a positive result indicated both the ability of a product to detect the target antigen in the sample, and to do this consistently (both tests from both lots).

The rate of detection against the panel used in this evaluation is expected to differ from the sensitivity for detection of malaria in a specific clinical setting. There are five main reasons for this:

- i. Performance may vary between lots or batches of the same product. Variability in lot performance is an issue with all diagnostics, and it can not be guaranteed that the results found here will predict results from different RDT lots that are purchased. It is important to test lots prior to distribution to the field, to ensure that expected performance is maintained (Section 14.2).
- ii. In clinical settings, patients show a wide variety of parasite densities, the range of which will depend on the local epidemiology of the disease. The magnitude of the parasite density in the population tested affects the clinical sensitivity of the test. Detection rates in the test panel of blood samples diluted to 200 parasites/ μ L are likely to underestimate the clinical sensitivity of an RDT in many situations, especially in high-transmission areas where symptomatic patients often have much higher parasite densities in their blood. Many tests that showed only moderate sensitivity against the 200 parasite/ μ L panel may perform very well in such settings, as shown by the high performance of most products against the panel set at 2000 parasites/ μ L.

Importantly, when interpreting Figs E1, 7, 8 and 9 and the colour coding in Tables 3, 3a, 4, 4a the small differences in detection rates found among the better-performing RDTs in this evaluation are unlikely to result in noticeable differences in clinical sensitivity, and other issues such as stability, cost, or ease of use and manufacturing capacity may be more important factors in test selection.

When considering the parasite density of the target populations and the likely field sensitivity of RDTs, it is important to note that, even in areas with high transmission and strong malaria immunity, populations may include individuals with low parasite densities but

clinically significant infections (e.g. young children, pregnant women, others with low immunity). The ability to detect low parasite density infections reliably therefore remains important in these cases.

- iii. Performance of tests against the challenge panel may not directly relate to sensitivity in clinical testing as there is variability in the amino acid sequence of the HRP2 antigen of *P. falciparum* that may affect the ability of RDTs to detect it. In certain *P. falciparum* parasites the HRP2 antigen may not be detectable at all. Specifically, there is evidence that *P. falciparum* strains in some areas of South America may express HRP2 antigens very poorly or not at all.²¹ If a significant proportion of parasites in a given area do not express HRP2, it is necessary to use tests detecting other target antigens (pLDH or aldolase). The distribution of such strains is currently being mapped.
- iv. The methods used to transport and store tests can affect their field sensitivity. All the tests used in this evaluation were shipped and stored under conditions intended to safeguard against degradation caused by high temperature or other extreme conditions. If similar precautions are not taken with purchased RDTs, loss of performance could result. Ambient temperatures of storage conditions vary widely in settings where these tests are commonly used, as do temperatures during transport, and requirements for heat stability of a product will therefore differ.
- v. Diagnostic sensitivity and specificity are dependent on the quality of preparation and interpretation of the tests. Highly trained individuals performed all the testing in this product evaluation. In clinical settings, malaria RDTs will often be used by health workers with limited training and supervision. Simplicity of design and clearly-interpretable results will have an influence on ensuring that the technical proficiency of a product translates into accurate diagnosis in the field.

13.2. FALSE POSITIVE RATE AND SPECIFICITY

False positive rates are reported here against a panel of 'clean' negative samples taken from blood donated in low-transmission settings by people without malaria symptoms. Also calculated were false positive rates against a smaller number of samples with specific characteristics that affect the likelihood of a false-positive result from an immuno-diagnostic test (e.g. rheumatoid factor, anti-nuclear antibody), or that may be of significance in a specific population (e.g. leishmaniasis, dengue). The importance of these results will vary with the intended area of use. High false positive rates against samples of blood from dengue patients, for example, may not be a significant factor to consider in regions where dengue does not occur. In view of the small number of samples in each category in this evaluation, the results should be considered primarily as a guide, to highlight potential cross-reactions that will require

close monitoring if relevant to the target population.

In general, it is preferable to procure a product with a low rate of false-positive reactions. In the case of many diagnostic tests, a trade-off must be made between a preference for a high rate of antigen detection (sensitivity) and a low false positive rate (specificity). The context in which the test will be used will guide the relative importance of these two factors in choosing one product over another. In this evaluation there was no correlation of loss of sensitivity associated with high specificity, with a number of products attaining both a high detection rate (predicting a high sensitivity) and a low false-positive rate (predicting a high specificity).

13.3. HEAT (THERMAL) STABILITY

RDTs in this evaluation were held for two months at 35°C and 45°C and then retested to evaluate stability at these temperatures. The importance of thermal stability will vary according to the ambient conditions under which a product is expected to be transported and stored. Thus, stability at high temperatures will be vital if an RDT is to be stored at clinic level in a country where ambient temperatures can reach 45°C in the hot season, but less critical in a high-altitude or cooler tropical environment where temperatures rarely rise above 35°C. Most of the commercial RDTs list 30°C as the maximal storage temperature. Higher temperatures were used for this evaluation because it is unusual for malaria-endemic countries to have maximum ambient temperatures less than 35°C, though the use of cool storage methods will also allow the use of less heat-stable products. Where transport and storage of RDTs is likely to occur at high ambient temperatures, heat (thermal) stability should be seen as a significant factor in ensuring maintenance of sensitivity.

High humidity will accelerate the degradation of malaria RDTs and other lateral flow tests. All the products in this evaluation were packaged in individual envelopes that contain a desiccant and are designed to be moisture-proof. This allows the user to open the envelope of a specific test at the time of use, limiting exposure to high humidity. During the stability testing phase of this evaluation, RDTs were stored at 75% humidity. The packaging should, if in good condition, protect the contents from exposure to high humidity during storage. As such, the stability testing results presented here provide an assessment of both the stability of the RDT and the quality of its packaging.

Several products showed high stability at the temperatures and time periods used in this evaluation. In general, pan-specific lines (pLDH and aldolase) were less stable than HRP2 test lines, but there was overlap between the antibodies against these targets with some pLDH tests maintaining high positivity rates after 2 months at 45°C. A small number of products showed a consistent improvement in positivity rate after incubation, for reasons which are not clear.

Though temperature and humidity were held constant in this evaluation, in the field temperatures fluctuate with time of day and season. While two months' storage at a set temperature can not accurately predict long-term stability

²¹ WHO 2009, unpublished data. Clarification of these findings will be made in following publications of the WHO-FIND malaria RDT evaluation programme.

under field conditions, loss of parasite detection over this period indicates a likelihood that significant sensitivity will be lost when similar or higher storage temperatures comprise a significant amount of the storage time, and indicates likelihood of a higher susceptibility to degradation during short periods of exposure to much higher temperatures, such as during transport (12, 13).

13.4. EASE OF USE DESCRIPTION

The sensitivity and specificity of RDT results will depend to some extent on the quality of preparation and interpretation of the test. In general, a simpler format with fewer steps or fewer extraneous materials is likely to be prepared and interpreted more reliably. Thus, cassette-format RDTs are generally more reliably prepared and interpreted than products in dip-stick format (14). The extra cost involved in such a format may be offset by the advantages of increased accuracy and, in some cases, less additional equipment required to perform them.

The method of blood transfer from the patient to the test is important for the safety of the user, and for the accuracy of volume of blood transferred. Devices for blood transfer are supplied with RDTs, and vary widely in design. The performance of blood transfer devices was not formally assessed in this evaluation, as blood was transferred from a tube by a micro-pipette to ensure the manufacturer-specified volume was used. Programmes procuring RDTs should consider the adequacy of the blood transfer device supplied, including previous experience of health workers and the costs and time required for re-training. It may often be appropriate to discuss with manufacturers changing the blood transfer device from that normally supplied.

Clarity of results is important to test interpretation. A clearly visible (intense) test line is less likely to be overlooked than a line that is barely visible. While reading proficiency and adequate work places should always be ensured, health workers may sometimes have sub-optimal vision or work in conditions of inadequate lighting. The intensity of the line of the test band is closely associated with error rate and with the detection rate found in this report (Tables A4.4, A4.4a, A4.5, A4.5a).

The importance of format and simplicity of test design will depend on the intended end-users. Trained laboratory technicians may handle a complicated procedure more reliably than village-level volunteers with limited supervision. In all cases, specific proficiency-based training and adequate supervision should be included in any RDT-based diagnostic programme, and clear instructions should be provided in a language and format appropriate for the end-user²²(14–16).

13.5. INTER-LOT VARIABILITY

This testing programme evaluated only two production lots of each product. Malaria RDTs are complex biological

products made of components commonly supplied from multiple sources, and subject to various conditions during manufacture that may affect the quality of the final product. All manufacturers entered in this evaluation have current ISO 13485:2003 certification, a standard designed to give assurance of consistency of quality of final product if correctly implemented. The results presented here indicate that inter-lot variability does occur, and WHO strongly recommends that a sample of RDTs from each production lot be tested prior to dissemination to the field to ensure it meets an appropriate standard. This can be facilitated by WHO (see section 14.2).

Since inter-test variability also occurs, this will be detected to some extent by routine lot testing. Ensuring manufacturers have good manufacturing standards should minimize the likelihood of inconsistencies due to poor practice in the manufacturing process.

13.6. INTER-READER VARIABILITY

All RDTs in this evaluation were interpreted by two technicians, blinded to the other technicians' results. The first reading was performed at the minimum time specified by the manufacturer to obtain a result, and this reading is used as the primary determinant of RDT performance. The second reading was obtained later, usually within 45 minutes, always within 90 minutes. Where a manufacturer specifically instructed in the product insert that interpretation be deferred if a result was not visible at the time of the initial reading, the result of the second reader was also reported.

The difference between the two readings reported here was therefore subject to the stability of the result: if a test became positive after the minimum time specified by a manufacturer to obtain a result, this appeared as a discrepancy between the two readers. The intensity of the test lines of antigen-detecting RDTs was also dependent on the amount of available antigen. Therefore, at the low parasite-densities used in this evaluation where a test line intensity was close to the limit of human visual perception (i.e. close to the operational limits of the product), inter-reader discrepancies were more likely to occur.

Early, accurate and stable RDT results are important in the context of busy clinical work. Specifying a single reading time at which the result can be determined to be negative or positive is also an advantage in terms of training and consistency of use. It is recommended that low inter-reader variability and early availability of result be considered in making decisions on procurement, but that the stability of the result (an adequate time period during which the result may be read) is also ensured.

13.7. TARGET ANTIGENS AND SPECIES

Malaria RDTs included in this evaluation detect one or more of three parasite antigens (HRP2, pLDH, and aldolase) in various combinations. HRP2 is present only in *P. falciparum*, whereas aldolase and pLDH are present in all four species

22 See www.wpro.who.int/sites/rdt for generic examples

and may be used as pan or all-species targets. Some tests use differences in pLDH sequences between species as a means to differentiate *P. falciparum* from *P. vivax* and other species. There is considerable overlap in the detection rates of products targeting the different antigens in this evaluation. While the products with the highest detection rates for *P. falciparum* targeted HRP2, a number of pLDH-detecting products demonstrated high detection rates against *P. falciparum* and *P. vivax*. The stability of tests targeting these different antigens also overlapped.

The choice of RDT should take target antigen into account: HRP2-detecting RDTs should not be used in areas where high rates of HRP2 non-expression occur. Tests detecting only HRP2 (without pLDH or aldolase lines) will have limited utility where non-falciparum malaria is common. pLDH (and possibly aldolase) RDTs may have further advantages where antigen persistence (common with HRP2) may result in a high false-positive rate in areas where early re-testing in the weeks soon after treatment is common.

The required sensitivity of a test may also vary with species; a less sensitive test may be acceptable for detection of *P. vivax* compared to detection of *P. falciparum*, as severe outcomes due to missed diagnoses are less likely. Use of a sufficiently sensitive pan-specific test may be appropriate in areas where both *P. falciparum* and *P. vivax* occur, if all infections were to be managed initially as a *P. falciparum* infection with artemisinin-based combination therapy (ACT).

It should be noted that pan-species tests were not evaluated for detection of *P. ovale* or *P. malariae* due to lack of sources of suitable mono-species infections of these parasites.

14. ADDITIONAL MEASURES TO ENSURE QUALITY AND UTILITY OF RDT TESTING

This report provides data to guide programmes in selecting products likely to perform to a high standard in the particular contexts in which the programme operates. The final decision on product selection requires that this data be considered in a systematic way, often with other data obtained from the manufacturer and other sources. An algorithm to guide this process, adapted from the quality assurance guide of the WHO Global Malaria Programme (found at www.wpro.who.int/sites/rdt) is given in Annex 5.

While malaria RDTs have application in a number of settings, the area in which they have the greatest impact on public health is in extension of access to accurate, parasite-based diagnosis of malaria to regions and populations where good quality microscopy-based analysis is impractical to maintain. This currently applies to most people at risk of malaria in endemic countries (1). In many settings where RDTs have been introduced, the true rate of parasitaemia has been found to be dramatically lower than expected, allowing health systems to save on wasted RDTs and to focus

appropriately on the management of non-malarial causes of fever, including pneumonia and sepsis. A successful RDT programme must therefore address not just malaria but also the management of other common and severe febrile illness that occur locally as differential diagnoses of malaria, if the potential full public health impact of an RDT programme is to be achieved.

14.1. BEYOND PROCUREMENT

RDTs are often added to a procurement list that includes mainly chemical agents such as pharmaceuticals, bednets, and insecticides. Unlike these products, however, diagnostic tests are relatively more complex to implement, not least because they represent the starting point, not the end point, in a health system intervention, and their use presumes that appropriate patient management, based on testing, will follow. Thus, successful introduction of RDTs requires careful planning going beyond rational procurement to ensure consistent supplies of all necessary materials (including gloves, sharps disposal containers, and supplies required for further case management), training of users, and community sensitization. This extends beyond malaria management to management of other febrile diseases and health service delivery systems.

This report provides information to guide procurement of RDTs within this framework. A number of factors beyond performance characteristics reported here must influence procurement decisions. An example algorithm to guide these decisions is given in Annex 5. Details of implementation will vary widely between programmes according to local capacity and needs. Further recommendations on budgeting, planning and implementation can be found in Annex 6.

14.2. LOT TESTING

Complementary to the product testing programme, WHO and FIND currently support three laboratories that perform continual quality assurance of RDTs in the form of lot testing. These lot testing facilities respond to formal requests from national malaria programmes, manufacturers, and procurement bodies and assess the quality of RDT lots when they arrive in country, before distribution. Testing is performed with carefully prepared blood panels such as those used for this evaluation. A number of other national institutions have also developed this capacity. Lot-testing reassures countries that the product they have purchased is performing to a high standard and helps to ensure that manufacturers produce consistently good lots and improve their products.

Countries and/or manufacturers ship between 125–175 RDTs to the regional lot testing centre where they are evaluated against a small panel of parasites at high and low parasite

densities and negative samples. They are subsequently incubated at a temperature close to the manufacturer's specified storage temperature and retested every three months until their expiry date. Initial results are available after five days and then sent at regular intervals. Details of the protocol can be found in a published methods manual for lot testing.²³

All countries and procuring agencies are encouraged to participate in the lot testing programme and in the near future it is intended to publish overall performance of products submitted for lot testing.

23 WHO, Rapid Diagnostic Tests for Malaria: Methods Manual for Laboratory Quality Control Testing. Version 5a. 2008, WHO - Regional Office for the Western Pacific. Available at: www.wpro.who.int/sites/rdt/documents/list.htm

15. CONCLUSIONS

This study is a landmark in the field of malaria RDT evaluations because of the number of products evaluated and its comprehensiveness including, samples with low and high parasite densities, detailed parasite characterization (antigen quantification, HRP 2,3 characterization) multiple test lots and heat stability assessment. New laboratory methods were developed and validated to support parasite characterization and this work generated new findings regarding the variation in antigen content at similar parasite densities and the variation in the structure and expression of HRP proteins. The latter will critically inform RDT selection in several endemic countries.

16. REFERENCES

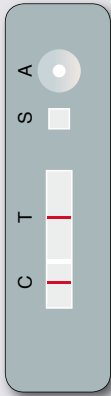
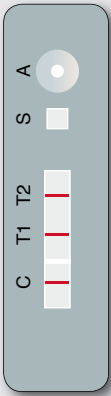
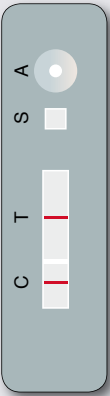
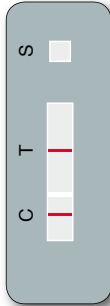
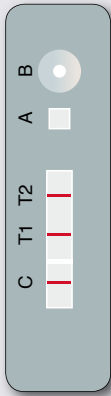
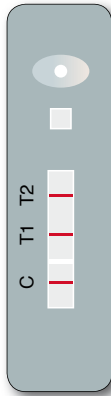


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ANNEXES

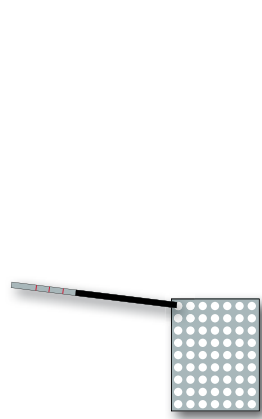


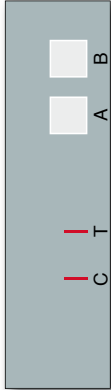
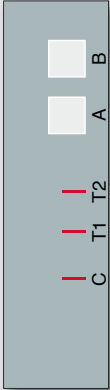
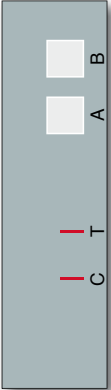
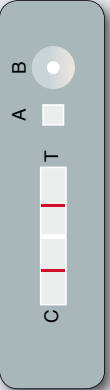

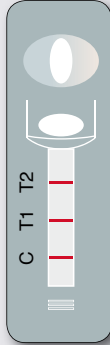
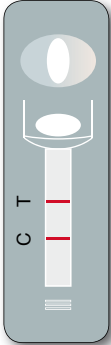
ANNEX 1: CHARACTERISTICS OF RAPID MALARIA TESTS IN THE EVALUATION

Manufacturer	Product name	Plasmodium species targeted (F = P. falciparum, V = P. vivax, O = P. ovale, M = P. malariae, P = PAN; major Plasmodium species)	Target Antigen ¹	Format	Sequence and type of bound antibody ²		Required volume (µl) of whole blood	Buffer volume (drops)	Intermediate Step	Time to results ³ (mins)	Maximum reading time ³	Results Interpretation (Type A-E) ⁴
					C	T1 T2						
Access Bio, Inc.	CareStart Malaria pLDH (PAN)	P	pLDH	Cassette	✓	pLDH	5	2 drops	-	20	-	
	CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	F, P	pLDH/HRP2	Cassette	✓	pLDH HRP2	5	2 drops	-	20	-	
	CareStart Malaria HRP2 (Pf)	F	HRP2	Cassette	✓	HRP2	5	2 drops	-	20	-	
ACON Laboratories, Inc.	Malaria Plasmodium falciparum Rapid Test Device (Whole Blood)	F	HRP2	Cassette	✓	HRP2	20	3 drops	Mixing time 1 min, squeeze tube 5 times	10	20	
Amgenix International, Inc.	OnSight - ParaQuick (Pan, Pf) Test	P, F	pLDH, HRP2	Cassette	✓	pLDH HRP2	5	6 drops	-	15	15	
AZOG, Inc.	AZOG Malaria pf (HRP-II) / pv (pLDH) Antigen Detection Test Device	F, P	"HRP2, pLDH	Cassette	✓	pLDH HRP2	5	2 drops	-	20	-	
Biosynex	Immunoquick Malaria Falciparum	F	HRP2	Dipstick	✓	HRP2	20 adults/5 children	6 drops	-	10	15	
	Immunoquick Malaria +4	F, P	HRP2/pLDH	Dipstick	✓	pLDH HRP2	20 adults/5 children	6 drops	Buffer in tube, Blood on stick, stick in tube	15	30	

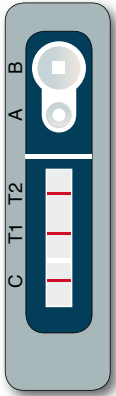
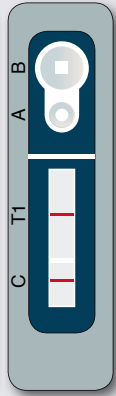
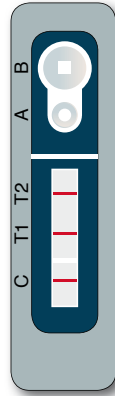
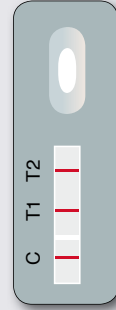
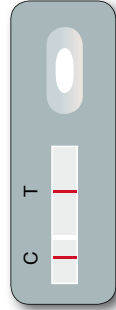
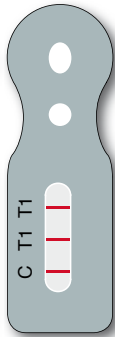
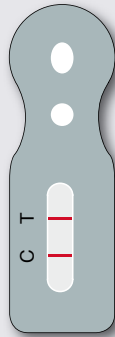
Manufacturer	Product name	Plasmodium species targeted (F = P. falciparum, V = P. vivax, O = P. ovale, M = P. malariae, P = PAN; major Plasmodium species)	Target Antigen ¹	Format	Sequence and type of bound antibody ²		Required volume (µl) of whole blood	Buffer volume (drops)	Intermediate Step	Time to results ³ (mins)	Maximum reading time ³ (min)	Results Interpretation (Type A–E) ⁴	
					C	T1 T2							
Diagnostics Automation/ Cortez Diagnostics, Inc.	Malaria Pf / Vivax (Malaria Combo Test)	F, P	HRP2/ Aldolase	Cassette	✓	HRP2	Aldolase	1 drops	1 drop in blood well, 5 drops in buffer well	15	60 min (30 min)	D	
DiaMed AG	OptiMal IT	F, P	pLDH	Dipstick-Cassette	✓	pLDH	10	Wait 1 min after buffer	Blood in well 1 stir, Wait 1 min, Stick in well 1 wait 10 min, Transfer stick to well 2, after 10 minutes remove from well 2	10	-	C	
Human GmBH	Hexagon Malaria	F	HRP2	Cassette	✓	HRP2	5	2 drops (3 drops)	-	15	-	A	
IND Diagnostic Inc.	One Step Malaria Antigen Strip	P,F	pLDH (par, pf)	Dipstick	✓	LDH (pf specific)	20	1 drop conjugate well, 3 drops washing well	Blood in conjugate well, mix and wait 1 min, stick in well, wait 5 min, Stick in washing well, wait 10 min"	15	-	C	

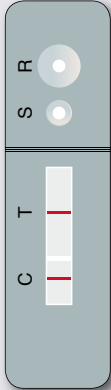
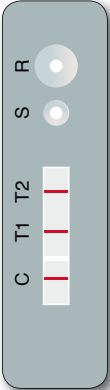
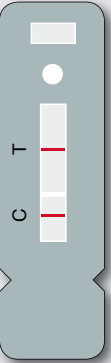


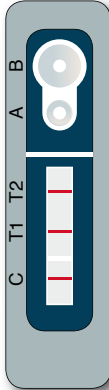
Manufacturer	Product name	Plasmodium species targeted (F = <i>P. falciparum</i> V = <i>P. vivax</i> O = <i>P. ovale</i> M = <i>P. malariae</i> P = PAN; major <i>Plasmodium</i> species)	Target Antigen ¹	Format	Sequence and type of bound antibody ²		Required volume (µl) of whole blood	Buffer volume (drops)	Intermediate Step	Time to results ³ (mins)	Maximum reading time ³	Results Interpretation (Type A-E) ⁴
					C	T1 T2						
Innovatek Medical Inc.	Quickstick Malaria Antigen Test	P, F	pLDH (pan, pf)	Dipstick	✓	pLDH (pf specific) pLDH	20	1 drop conjugate well, wait 1 min, stick in well, wash 3 drops	Blood in conjugate well, mix and wait 1 min, stick in well, wash 5 min, stick in washing well, wait 10 min	15	-	C
InTec Products, Inc.	ADVANCED QUALITY™ One Step Malaria (p.f.) Test (whole blood)	F	HRP2	Card/strip looks like a cassette!	✓	HRP2	10	3 drops (4 drops)	-	-	30	A
Inverness Medical Innovations, Inc.	Binax Now Malaria	F, P	HRP2/pan malaria antigen	Cassette	✓	HRP2 pan malaria antigen	15	2 drops (3 drops) Allow blood to run to almost upper pad, 4 drops upper pad, close the card	-	15	-	D
	ADVANCED QUALITY™ MALARIA (p.f.) POCT	F	HRP2	Card no separate product insert??	✓	HRP2	10	3 drops	-	-	30	A

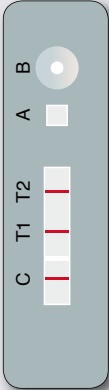
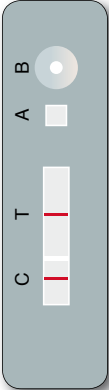
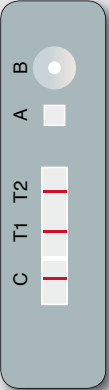


Manufacturer	Product name	Plasmodium species targeted (F = <i>P. falciparum</i> V = <i>P. vivax</i> O = <i>P. ovale</i> M = <i>P. malariae</i> P = PAN; major <i>Plasmodium</i> species)	Target Antigen ¹	Format	Sequence and type of bound antibody ²		Required volume (µl) of whole blood	Buffer volume (drops)	Intermediate Step	Time to results ³ (mins)	Maximum reading time ³	Results Interpretation (Type A-E) ⁴	Results Interpretation (Type A-E) ⁴
					C	T1 T2							
J. Mitra & Co. Pvt. Ltd.	Advantage Pf. Malaria Card	F	HRP2	Card	✓	HRP2	5	5 drops	-	20	+	A	
	Advantage Pan Malaria Card	P	pLDH	Card	✓	pLDH	5	5 drops	-	20	-	B	
	Advantage Mal Card	P,F	pLDH	Card	✓	pLDH HRP2	5	5 drops	-	20	-	C	
Orchid Biomedical Systems	Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	F	HRP2	Cassette	✓	HRP2	5	6 drops	-	15	15	A	
	Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	F	HRP2	Dipstick	✓	HRP2	5	4 drops in tube, Blood on stick, Stick in tube	-	15	15	A	
Premier Medical Corporation Ltd.	First Response Malaria Ag Combo (pLDH/HRP2)	F, P	pLDH/HRP2	Cassette	✓	pLDH HRP2	5	2 drops	-	20	-	C	
	First Response Malaria Ag HRP2	F	HRP2	Cassette	✓	HRP2	5	2 drops	-	20	-	A	

Manufacturer	Product name	Plasmodium species targeted (F = P. falciparum, V = P. vivax, O = P. ovale, M = P. malariae, P = PAN; major Plasmodium species)	Target Antigen ¹	Format	Sequence and type of bound antibody ²			Required volume (µl) of whole blood	Buffer volume (drops)	Intermediate Step	Time to results ³ (mins)	Maximum reading time ³	Results Interpretation (Type A-E) ⁴
					C	T1	T2						
ICT Diagnostics	ICT Malaria Pf Cassette Test (MLO1)	F	HRP2	Cassette	✓	HRP2		5	5 drops	-	15	15	A
	ICT Malaria Combo Cassette Test (MLO2)	F, P	HRP2/pan malaria Antigen	Cassette	✓	HRP2	pan malaria antigen	5	5 drops	-	15	15	D
	Parahit-f DIPSTICK FOR FALCIPARUM MALARIA	F	HRP2	Dipstick	✓	HRP2		8	4 drops	Buffer in tube, blood on stick, Stick in tube		15	30
Span Diagnostics Ltd.	Parahit-f TEST DEVICE FOR FALCIPARUM MALARIA	F	HRP2	Cassette	✓	HRP2		8	4 drops		15	30	A
	Parahit-Total Device Rapid test for P. falciparum and Pan malarial species	F, P	HRP2/ pLDH/ aldolase	Cassette	✓	HRP2	pLDH/ aldolase	8	4 drops		15	30	D
Standard Diagnostics Inc.	SD BIOLINE Malaria Ag	F, P	pLDH	"Cassette"	✓	pLDH (pan)	pLDH (Pf)	5	4 drops	-	15	30	C
	Dipstick"	✓	pLDH (pan)	pLDH (PF)	5	HRP2		5	4 drops	-	15	30	A
	SD BIOLINE Malaria Ag Pf/ Pan	F	HRP2	Cassette	✓	HRP2		5	4 drops	-	15	30	C



Manufacturer	Product name	Plasmodium species targeted (F = <i>P. falciparum</i> V = <i>P. vivax</i> O = <i>P. ovale</i> M = <i>P. malariae</i> P = PAN; major <i>Plasmodium</i> species)	Target Antigen ¹	Format	Sequence and type of bound antibody ²		Required volume (µl) of whole blood	Buffer volume (drops)	Intermediate Step	Time to results ³ (mins)	Maximum reading time ³	Results Interpretation (Type A-E) ⁴	Results Interpretation (Type A-E) ⁴
					C	T1 T2							
Unimed International, Inc.	FirstSign – Malaria Pf Card Test	F	HRP2	Cassette	✓	HRP2	5	6 drops	-	15	15	A	
	FirstSign – ParaView-2 (Pv + Pf) Card Test	F, V	pLDH/HRP2	Cassette	✓	pLDH (specific Pv) HRP2	5	4 drops	-	15	15	E	
Vision Biotech (Pty) Ltd.	Malaria Rapid Pf	F	HRP2	Cassette	✓	HRP2	5	5 drops	-	15	-	A	
	Malaria Rapid Combo	P, F	HRP2/ Aldolase	Cassette	✓	HRP2 Aldolase	5	5 drops	-	15	-	D	
	Malaria Rapid Dual	P, F	HRP2/pLDH	Cassette	✓	HRP2 pLDH	5	5 drops	-	15	-	D	
Guangzhou Wondfo Biotech Co., Ltd.	Wondfo One Step Malaria Pf/Pan Whole Blood Test	F, P	pLDH/ HRP2	Cassette	✓	pLDH HRP2	5	3 drops	-	15	30	C	

Manufacturer	Product name	Plasmodium species targeted (F = <i>P. falciparum</i> V = <i>P. vivax</i> O = <i>P. ovale</i> M = <i>P. malariae</i> P = PAN; major <i>Plasmodium</i> species)	Target Antigen ¹	Format	Sequence and type of bound antibody ²			Required volume (µl) of whole blood	Buffer volume (drops)	Intermediate Step	Time to results ³ (mins)	Maximum reading time ³	Results Interpretation (Type A-E) ⁴
					C	T1	T2						
Zephyr Biomedicals	Parascreen Rapid Test for Malaria Pan/Pf (Device)	P, F	pLDH, HRP2	Cassette	✓	pLDH	HRP2	5	4 drops	-	15-30	-	
	Parabank Rapid Test for Malaria Pan (Device)	P	pLDH	Cassette	✓	pLDH		5	4 drops	*	15-30	-	
	Malascan Rapid Test for Malaria Pf Pan (Device)	F, P	HRP2, Pan specific aldolase	Cassette	✓	Aldolase	HRP2	5	4 drops	-	15-30	-	

1 – pLDH = *plasmodium* lactate dehydrogenase ; HRP = histidine rich protein

2 – Sequence when test held in a horizontal position and the sample well at far right and control line far left

3 – From placement of buffer, or from 'intermediate step' if this is present

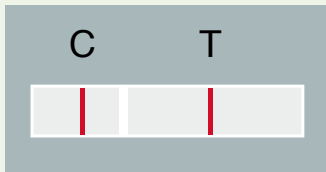
4 – See Annex 2

Each product should ideally be accompanied by all required materials (lancet, pipette etc.) Particularly when used at the village health worker level; however, this is often not the case and the contents depend on the request of procuring agent.

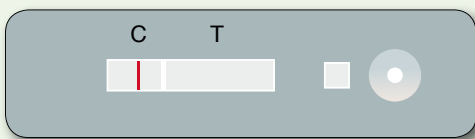
ANNEX 2: MALARIA RDT GUIDE TO RESULTS INTERPRETATION

Type A: Malaria Generic Pf RDT Results Guide

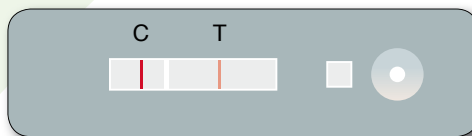
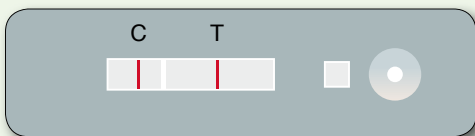
Results Window: C=control line; T=test line with bound HRP-2 antibody.



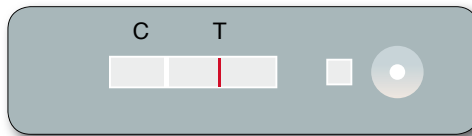
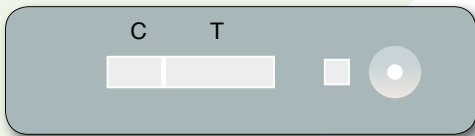
Negative Results: One line 'C' appears in the results window



Positive Results: *P. falciparum* infection. Two lines 'C' and 'T' appear in the results window. Test is positive even if the test line is faint

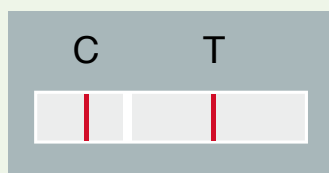


Invalid Results: No 'C' line appears in the results window. Repeat the test using a new RDT if no control line appears.

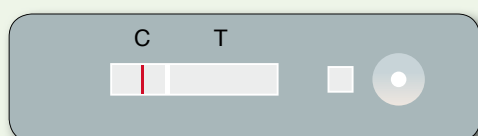


Type B: Malaria Generic Major *Plasmodium* species (PAN) RDT Results Guide

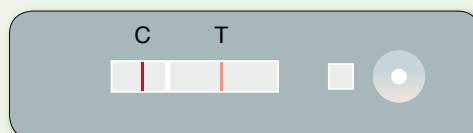
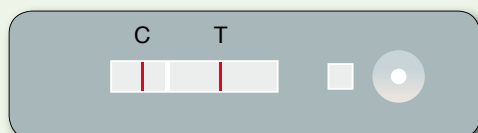
Results Window: C=control line; T=test line with bound HRP2 antibody.



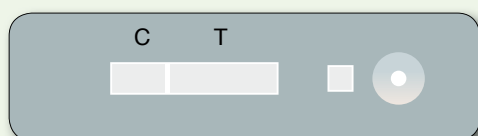
Negative Results: One line 'C' appears in the results window



Positive Results: *Plasmodium* species (*P. falciparum*, *P. vivax*, *P. malariae*, *P. ovale*) infection. Two lines 'C' and 'T' appear in the results window. Test is positive even if the test line is faint

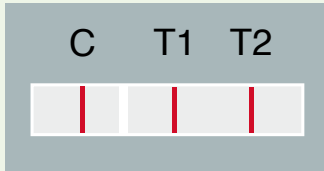


Invalid Results: No 'C' line appears in the results window. Repeat the test using a new RDT if no control line appears.

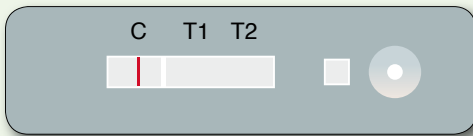


Type C: Malaria Generic Pan-Pf RDT Results Guide

Results Window: C=control line; T1=test line with bound pLDH or aldolase antibody; T2=test line with bound HRP2 or Pf specific pLDH antibody.

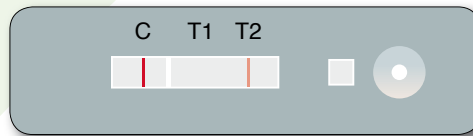
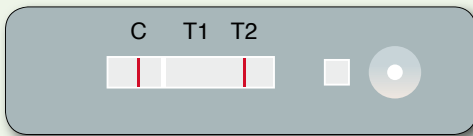


Negative Results: Only one line 'C' appears in the results window.

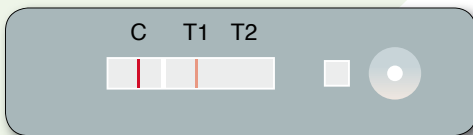


Positive Results:

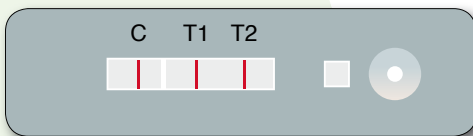
P. falciparum: Two lines 'C' and 'T2' appear in the results window.



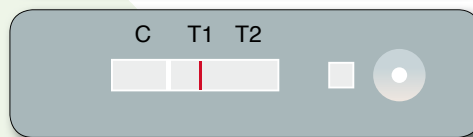
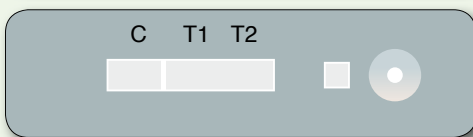
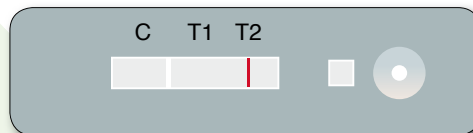
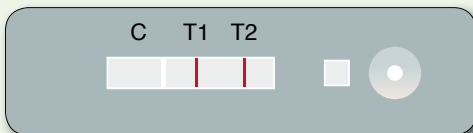
Non-falciparum infection (*P. vivax*, *P. ovale*, *P. malariae*) or mixed infection of these: Two lines 'C' and 'T1' appear in the results window.



P. falciparum or mixed infection. Three lines 'C', 'T1' and 'T2' appear in the results window.

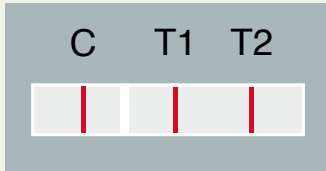


Invalid Results: No 'C' line appears in the results window. Repeat the test using a new RDT if no control line appears.

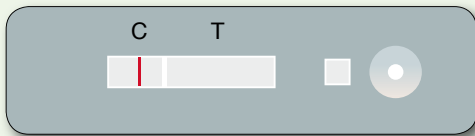


Type D: Malaria Generic Pf-Pan RDT Results Guide

Results Window: C=control line; T1=test line with bound HRP2 or Pf specific LDH antibody; T2=test line with bound pLDH or aldolase antibody

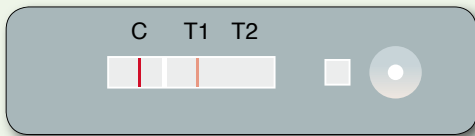


Negative Results: Only one line 'C' appears in the results window.

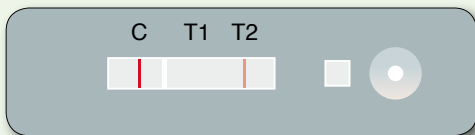


Positive Results:

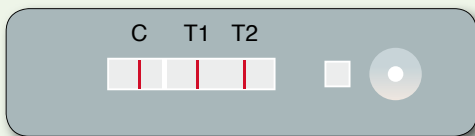
P. falciparum infection. Two lines 'C' and 'T1' appear in the results window.



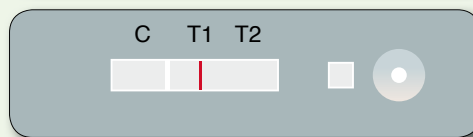
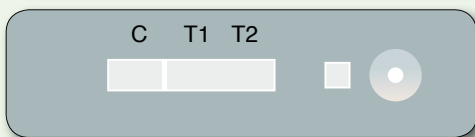
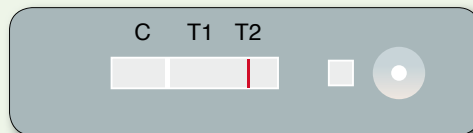
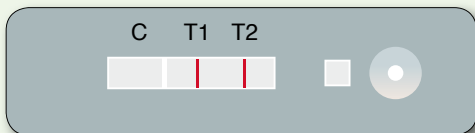
Non-falciparum infection (*P. vivax*, *P. ovale*, *P. malariae*) or mixed infection of these. Two lines 'C' and 'T2' appear in the results window



P. falciparum or mixed infection. Three lines 'C', 'T1' and 'T2' appear in the results window.

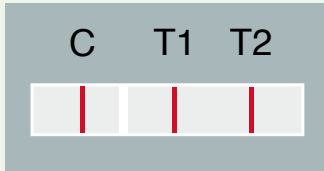


Invalid Results: No 'C' line appears in the results window. Repeat the test using a new RDT if no control line appears.

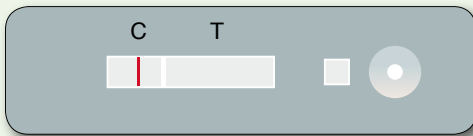


Type E: Malaria Generic Pv-Pf RDT Results Guide

Results Window: C=control line; T1=test line with bound *P. vivax* specific pLDH; T2=test line with bound HRP-2 antibody.

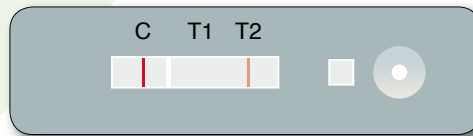
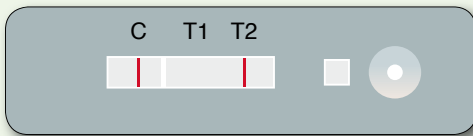


Negative Results: Only one line 'C' appears in the results window.

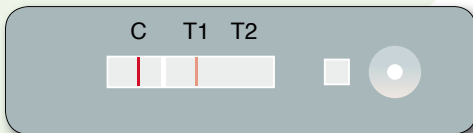


Positive Results:

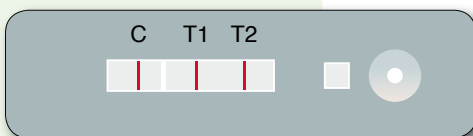
P. falciparum infection. Two lines 'C' and 'T2' appear in the results window



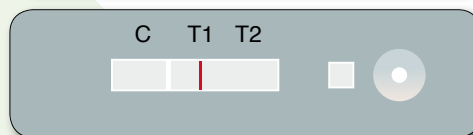
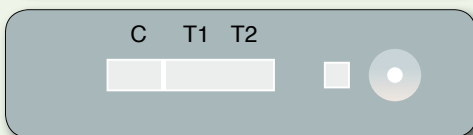
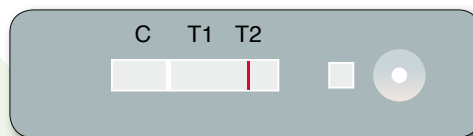
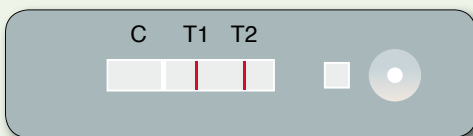
P. vivax infection. Two lines 'C' and 'T1' appear in the results window



P. falciparum and *P. vivax* mixed infection. Three lines 'C', 'T1' and 'T2' appear in the results window.



Invalid Results: No 'C' line appears in the results window. Repeat the test using a new RDT if no control line appears.



Product	Manufacturer	<i>P. falciparum</i> samples (n=20)												
		200 parasites/µl					2,000 or 5,000 parasites/µl							
		Total positive results returned		Lot 1		Lot 2		Total positive results returned		Lot 1		Lot 2		
		Test 1	Test 2	No. positive agreements (max=20)*	Test 1	Test 2	Test 1	Test 2	No. positive agreements (max=20)*	Test 1	Test 2	Test 1	Test 2	No. positive agreements (max=20)*
Pan only														
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	13	8	6	2	2	2	2	1	20	19	20	19	1
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	18	18	17	17	15	13	13	13	20	20	20	20	13
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	0	0	0	0	0	0	0	0	18	14	18	14	0
PF - <i>Plasmodium falciparum</i>														
Pv - <i>Plasmodium vivax</i>														
pan - <i>Plasmodium</i> species														
† results are based on the first readers interpretation according to manufacturers instructions.														
* Number of samples that returned a positive result for both tests. Where one test was invalid and the other positive, positive agreement was recorded.														

Table A3.1a: Lot variability in positive results against *P. falciparum* culture samples at low (200) and high (2000 or 5000) parasite densities (parasites/µl) based on a delayed reading when specified by the manufacturer

Product	Manufacturer	<i>P. falciparum</i> samples (n=20)												
		200 parasites/µl					2,000 or 5,000 parasites/µl							
		Total positive results returned		Lot 1		Lot 2		Total positive results returned		Lot 1		Lot 2		
		Test 1	Test 2	No. positive agreements (max=20)*	Test 1	Test 2	Test 1	Test 2	No. positive agreements (max=20)*	Test 1	Test 2	Test 1	Test 2	No. positive agreements (max=20)*
Pf & Pan/Pf & Pv														
FirstSign - ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	13	15	11	15	13	13	13	13	20	19	20	19	13
Immunoquick Malaria +4	Biosynex	19	19	18	19	19	18	18	18	20	20	20	20	18
Malaria Pf/Pv/vivax	Diagnostics Automation/Cortez Diagnostics, Inc.	1 (16)	0 (11)	1 (18)	1 (15)	2 (11)	1 (16)	1 (16)	1 (16)	1 (12)	0 (14)	1 (12)	0 (14)	1 (16)
Malaccan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	10	10	7	9	15	8	8	8	20	20	20	20	8
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	18	14	14	17	17	17	17	17	20	20	20	20	17
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	11	12	8	16	16	15	15	15	18	19	18	19	15
Pan only														
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	4	1	0	1	3	1	1	1	20	19	20	19	1
PF - <i>Plasmodium falciparum</i>														
Pv - <i>Plasmodium vivax</i>														
pan - <i>Plasmodium</i> species														
* Number of samples that returned a positive result for both tests. Where one test was invalid and the other positive, positive agreement was recorded.														

Table A3.2: Detection rate of 20 *P. falciparum* culture lines at low and high parasite densities based on initial reading according to manufacturers instructions and a delayed second reading performed within 1 hour.

Product	Manufacturer	200 parasites/ μ l				Detection rate*			
		Reader 1 [†] (n=80)	Reader 2 (n=80)	Percent agreement between readers (n=80)	Mean time between readings	Reader 1 [†] (n=40)	Reader 2 (n=40)	Mean time between readings	2,000 – 5,000 parasites/ μ l
Pf only									
Advanced Quality One Step Malaria (pf) Test (Whole Blood) ITP11002TC1	Intec Products Inc.	43.75	62.5	68.75	0.19:52	95	97.5	0.19:33	
Advanced Quality One Step Malaria (Pf) Test (Whole Blood) ITP11002TC40	Intec Products Inc.	36.25	57.5	68.75	0.22:18	95	97.5	0.21:39	
Advantage P.F. Malaria Card	J. Mitra Company Pvt Ltd	92.5	95	95	0.16:28	100	100	0.19:00	
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	100	100	100	0.13:44	100	100	0.13:13	
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	100	100	100	0.20:16	100	100	0.22:34	
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	15	8.75	83.75	0.20:10	85	82.5	0.19:03	
Hexagon Malaria	Human GmbH	22.5	38.75	78.75	0.20:58	97.5	95	0.20:13	
ICT Malaria Pf Cassette Test (ML01)	ICT Diagnostics	82.5	86.25	86.25	0.17:32	100	100	0.17:12	
Immunoquick Malaria Falciparum	Biosynex	70	90	80	0.13:06	100	100	0.15:36	
Malaria Plasmodium falciparum Rapid test Device (Whole blood)	ACON Laboratories, Inc.	92.5	87.5	92.5	0.20:16	97.5	100	0.24:18	
Malaria Rapid Pf	Vision Biotech	86.25	98.75	87.5	0.11:18	100	100	0.10:37	
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	50.63 (79)	72.15 (79)	65.38 (78)	0.15:04	97.37 (88)	100	0.13:26	
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	71.25	51.25	80	0.15:47	100	100	0.17:32	
Parahit-F DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics	67.5	62.5	82.5	0.20:13	100	100	0.21:57	
Parahit-F TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics	22.5	38.75	81.25	0.22:47	95	100	0.20:36	
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	93.75	90	96.25	0.28:49	100	100	0.32:18	
Pf & Pan/Pf & Pv									
Advantage Mal Card	J. Mitra Company Pvt Ltd	12.5	36.25	68.75	0.18:13	100	100	0.20:12	
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	91.25	97.5	93.75	0.12:26	100	100	0.13:00	
Binx Now Malaria	Inverness Medical Innovations	83.75	77.5	86.25	0.20:58	100	100	0.25:07	
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	98.75	98.75	97.5	0.09:50	100	100	0.09:20	
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	98.75	98.75	100	0.29:34	100	100	0.29:54	
FirstSign – Paraview-2 (Pv + Pf) Card Test	Unimed International, Inc.	41.25	70	71.25	0.16:33	97.5	97.5	0.15:28	
Hexagon Malaria Combi	Human GmbH	35	33.75	66.25	0.23:23	97.5	97.5	0.22:40	
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	90	91.25	96.25	0.19:00	100	100	0.19:17	
Immunoquick Malaria ++	Biosynex	91.25	95	91.25	0.23:25	100	100	0.22:36	
Malaria PFMvax	Diagnostic Automation/Cortez Diagnostics, Inc.	0 (52)	7.55 (63)	92.31 (52)	0.24:29	8.33 (24)	3.85 (26)	0.23:51	
Malaria Rapid Combo	Vision Biotech	72.5	87.5	85	0.16:28	97.5	100	0.15:45	
Malaria Rapid Dual	Vision Biotech	87.5	96.25	91.25	0.19:24	100	100	0.18:40	
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	35	55	75	0.33:02	100	100	0.33:00	
One Step Malaria Antigen Strip	IND Diagnostic Inc.	0	0	100	0.06:21	80	67.5	0.06:19	
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	78.75	85	93.75	0.19:04	100	100	0.18:51	
OptiMAL-IT	DiaMed	0	21.25	78.75	0.23:04	100	100	0.24:13	
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Parahit Diagnostics	23.75	32.5	86.25	0.23:48	92.5	95	0.22:22	
Parascan Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	55	82.5	72.5	N/A	100	100	N/A	
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	0	0	100	0.06:21	80	67.5	0.06:19	
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	2.5	3.75	96.25	0.23:37	92.5	97.5	0.25:51	
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	87.5	87.5	100	N/A	100	100	N/A	
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co, Ltd	56.25	68.75	77.5	0.13:31	92.5	92.5	0.10:14	
Pan only									
Advantage Pan Malaria Card	J. Mitra Company Pvt Ltd	31.25	61.25	47.5	0.18:25	97.5	100	0.19:52	
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	85	86.25	76.25	0.13:40	100	97.5	0.12:36	
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	0	11.25	88.75	0.32:52	80	97.5	0.32:29	

Pf – *Plasmodium falciparum*
Pv – *Plasmodium vivax*
† - Read according to manufacturers instructions
* - A sample is considered detected only if all RDIs from both lots read by the first technician, at minimum specified reading time, are positive
Those tests in which a manufacturer recommended a delayed reading if initial results were negative.

Table A3.3: Distribution of test band intensity scores (0-4) against 20 *P. falciparum* cultured parasites at low (200) and high (2000 or 5000) parasite densities (parasites/ μ)

Product	Manufacturer	200 parasites/ μ samples (n=20)				2,000 or 5,000 parasites/ μ samples (n=20)				200 parasites/ μ samples (n=20)				2,000 or 5,000 parasites/ μ samples (n=20)							
		0*	1	2	3	4	0	1	2	3	4	0	1	2	3	4	0	1	2	3	4
PF only																					
ADVANCED QUALITY TM MALARIA (p.f) POCt Test (whole blood)	InTec Products, Inc.	63.75	32.5	2.5	1.25	0	5	17.5	20	15	42.5	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
ADVANCED QUALITY TM One Step Malaria (p.f.) Test (whole blood)	InTec Products, Inc.	56.25	37.5	6.25	0	0	5	15	22.5	22.5	35	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Advantage Pf. Malaria Card	J. Mitra & Co. Pvt. Ltd.	7.5	46.25	35	8.75	2.5	0	2.5	5	15	77.5	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	0	25	32.5	36.25	6.25	0	0	2.5	12.5	85	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	0	36.25	35	20	8.75	0	2.5	10	87.5	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
FirstSign - Malaria Pf Card Test	Unimed International, Inc.	85	12.5	1.25	1.25	0	15	35	15	25	10	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Hexagon Malaria	Human GmbH	77.5	17.5	3.75	1.25	0	2.5	37.5	25	15	20	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
ICT Malaria Pf Cassette Test (ML01)	ICT Diagnostics	17.5	50	28.75	3.75	0	0	5	12.5	27.5	55	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Immunorquick Malaria falciparum	Biosynex	30	63.75	6.25	0	0	0	15	27.5	25	32.5	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Malaria Plasmodium falciparum Rapid test Device (Whole blood)	ACON Laboratories, Inc.	7.5	55	30	6.25	1.25	2.5	0	10	40	47.5	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	13.75	76.25	10	0	0	0	12.5	10	37.5	40	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	50	37.5	11.25	1.25	0	7.5	10	20	45	17.5	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	28.75	55	13.75	2.5	0	0	10	30	42.5	17.5	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Parahit-f DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	32.5	50	12.5	3.75	1.25	0	2.5	27.5	25	45	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Parahit-f TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	77.5	20	1.25	0	1.25	5	27.5	20	27.5	20	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	6.25	23.75	42.5	25	2.5	0	0	7.5	7.5	85	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
PF & Pan/PF & Pv																					
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	87.5	12.5	0	0	0	0	27.5	47.5	25	0	91.25	8.75	0	0	0	42.5	50	7.5	0	0
AZOG Malaria pf (HRP-II) jpv (pLDH) Antigen Detection Test Device	AZOG, Inc.	8.75	66.25	17.5	7.5	0	0	5	10	32.5	52.5	61.25	38.75	0	0	0	2.5	82.5	15	0	0
Binox Now Malaria	Inverness Medical Innovations, Inc.	16.25	67.5	13.75	2.5	0	0	10	15	45	30	100	0	0	0	0	12.5	87.5	0	0	0
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	1.25	21.25	47.5	25	5	0	0	5	10	85	2.5	96.25	1.25	0	0	0	0	35	45	20
First Response Malaria Ag Combo (pLDH/HRP2)	Premier Medical Corporation Ltd.	1.25	16.25	40	32.5	10	0	2.5	17.5	80	37.5	62.5	0	0	0	2.5	0	50	40	7.5	0
FirstSign - Paraview-2 (Pv + Pf) Card Test	Unimed International, Inc.	58.75	22.5	12.5	6.25	0	2.5	7.5	5	30	55	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Hexagon Malaria Combi	Human GmbH	65	33.75	1.25	0	2.5	27.5	22.5	25	22.5	25	100	0	0	0	100	0	0	0	0	0
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	10	35	40	13.75	1.25	0	7.5	15	77.5	97.5	2.5	0	0	0	22.5	67.5	10	0	0	0
Immunorquick Malaria +4	Biosynex	8.75	36.25	43.75	7.5	3.75	0	0	7.5	20	72.5	100	0	0	0	20	70	7.5	2.5	0	0
Malaria PF/vvax	Diagnostic Automation / Cortez Diagnostics, Inc.	100	0	0	0	0	95	5	0	0	0	45	11.25	25	15	3.75	32.5	5	12.5	7.5	42.5
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	27.5	61.25	8.75	2.5	0	2.5	7.5	10	40	40	98.75	1.25	0	0	0	20	80	0	0	0
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	12.5	52.5	26.25	7.5	1.25	0	5	12.5	10	72.5	88.75	11.25	0	0	0	27.5	72.5	0	0	0
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	65	31.25	3.75	0	0	0	20	35	27.5	17.5	100	0	0	0	0	52.5	35	12.5	0	0
One Step Malaria Antigen Strip	IND Diagnostic Inc.	100	0	0	0	0	20	72.5	7.5	0	0	100	0	0	0	0	22.5	70	7.5	0	0
OnSight - ParaQuick (Pan, Pf) Test	Amgen International, Inc.	21.25	47.5	22.5	5	3.75	0	2.5	10	20	67.5	98.75	1.25	0	0	0	2.5	47.5	35	12.5	2.5
OptiMAL-IT	DiaMed AG	100	0	0	0	0	0	67.5	25	7.5	0	98.75	1.25	0	0	0	65	27.5	7.5	0	0
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	76.25	21.25	1.25	0	1.25	7.5	20	27.5	22.5	22.5	100	0	0	0	0	100	0	0	0	0
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	45	28.75	22.5	1.25	2.5	0	10	25	30	35	100	0	0	0	0	35	45	15	5	0
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	100	0	0	0	0	20	72.5	7.5	0	0	100	0	0	0	0	22.5	70	7.5	0	0
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	97.5	2.5	0	0	0	7.5	55	32.5	5	0	100	0	0	0	0	75	25	0	0	0
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	12.5	57.5	25	2.5	2.5	0	5	5	10	80	100	0	0	0	0	55	45	0	0	0
Wondfo One-Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	43.75	51.25	5	0	0	7.5	12.5	27.5	32.5	20	90	10	0	0	0	10	45	30	12.5	2.5
Pan only																					
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	68.75	30	1.25	0	0	2.5	20	55	20	2.5	68.75	30	1.25	0	0	2.5	20	55	20	2.5
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	15	83.75	1.25	0	0	0	2.5	45	37.5	15	15	83.75	1.25	0	0	2.5	45	37.5	15	15
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	100	0	0	0	0	20	47.5	27.5	5	0	100	0	0	0	0	20	47.5	27.5	5	0

* denotes no band

Pf - *Plasmodium falciparum*

Pv - *Plasmodium vivax*

pan - *Plasmodium* species

† this includes Pan line intensity only for Pan-only tests

Table A3.3a: Distribution of test band intensity (0–4) scores against 20 *P. falciparum* cultured parasites at low (200) and high (2000 or 5000) parasite densities (parasites/ μ l) based on a delayed reading when specified by the manufacturer

Product	Manufacturer	200 parasites/ μ l samples (n=20)					2,000 or 5,000 parasites/ μ l samples (n=20)					200 parasites/ μ l samples (n=20)					2,000 or 5,000 parasites/ μ l samples (n=20)										
		Percentage distribution of test Pf band intensity* (n=80)					Percentage distribution of test Pf band intensity* (n=40)					Percentage distribution of Pan test band intensity (n=80)					Percentage distribution of Pan test band intensity (n=40)										
		0*	1	2	3	4	0	1	2	3	4	No band	1	2	3	4	No band	1	2	3	4	No band	1	2	3	4	
Pf & Pan/Pf & Pv																											
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	30	56.25	10	2.5	1.25	2.5	7.5	25	57.5	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Immunoquick Malaria +4	Biosynex	5	36.25	35	20	3.75	0	2.5	5	12.5	80	98.75	1.25	0	0	0	2.5	70	25	2.5	0	0	0	0	0	0	
Malaria P.F/Vivax	Diagnostics Automation/Cortez Diagnostics, Inc.	95	5	0	0	0	97.5	0	2.5	0	0	43.75	13.75	17.5	17.5	7.5	32.5	5	7.5	12.5	42.5	0	0	0	0	0	
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	45	46.25	7.5	0	1.25	0	20	30	35	15	96.25	3.75	0	0	0	25	62.5	12.5	0	0	0	0	0	0	0	
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	17.5	41.25	33.75	6.25	1.25	0	2.5	15	25	57.5	91.25	8.75	0	0	0	15	40	32.5	12.5	0	0	0	0	0	0	
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	31.25	60	6.25	2.5	0	7.5	7.5	30	20	35	53.75	43.75	2.5	0	0	7.5	30	32.5	12.5	17.5	0	0	0	0	0	
Pan only																											
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	88.75	11.25	0	0	0	2.5	42.5	45	10	0	88.75	11.25	0	0	0	2.5	42.5	45	10	0	0	0	0	0	0	

*=this includes Pan line intensity only for Pan-only tests

* denotes no band

Pf – *Plasmodium falciparum*

Pv – *Plasmodium vivax*

pan – *Plasmodium* species

ANNEX 4: PHASE 2 RESULTS

Table A4.1: Lot variability in positive results† against *P. falciparum* and *P. vivax* samples at low (200) and high (2000 or 5000) parasite densities (parasites/μl)

Product	Manufacturer	<i>P. falciparum</i> samples (n=79)										<i>P. vivax</i> samples (n=20)									
		Total positive results returned										Total positive results returned									
		200 parasites/μl					2,000 or 5,000 parasites/μl					200 parasites/μl					2,000 or 5,000 parasites/μl				
		Lot 1		Lot 2		No. positive agreements (max=79)*	Test 1	Test 2	Test 1	Test 2	No. positive agreements (max=79)*	Test 1	Test 2	Test 1	Test 2	No. positive agreements (max=20)*	Test 1	Test 2	Test 1	Test 2	No. positive agreements (max=20)*
PF only																					
ADVANCED QUALITY™ MALARIA (p.f) POCT	InTec Products, Inc.	59	61	51	63	60	54	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
ADVANCED QUALITY™ One Step Malaria (p.f.) Test (whole blood)	InTec Products, Inc.	67	70	62	72	69	62	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Advantage Pf. Malaria Card	J. Mitra & Co. Pvt. Ltd.	78	77	77	78	79	78	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	79	79	79	78	79	78	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	79	79	79	79	79	79	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	38	44	31	39	36	31	75	71	71	71	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Hexagon Malaria	Human GmbH	46	44	34	47	46	40	76	76	76	76	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
ICT Malaria Pf Cassette Test (MI01)	ICT Diagnostics	73	73	69	73	76	72	79	77	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Immunoquick Malaria Falciparum	Biosynex	75	76	73	77	76	75	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Malaria <i>Plasmodium falciparum</i> Rapid test Device (Whole blood)	ACON Laboratories, Inc.	74	77	73	76	78	76	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	61	62	55	76	77	74	77	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	53	48	45	55	54	51	72	78	78	78	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	68	66	62	70	67	65	78	78	78	78	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Parahit-F DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	69	72	67	72	69	67	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Parahit-F TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	54	45	43	45	43	38	79	77	77	77	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	79	79	79	78	78	77	79	78	78	78	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
PF & Pan/Pf & Pv																					
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	60	63	58	59	62	55	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	70	70	64	70	73	69	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Binox Now Malaria	Inverness Medical Innovations, Inc.	78	77	76	76	75	72	78	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	79	78	78	77	79	77	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	79	79	79	79	79	79	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
FirstSign – Paraview-2 (Pv + Pf) Card Test	Unimed International, Inc.	54	51	47	56	48	43	78	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Hexagon Malaria Combi	Human GmbH	50	50	43	51	49	45	78	77	77	77	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
ICT Malaria Combo Cassette Test (MI02)	ICT Diagnostics	74	75	71	75	77	74	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Immunoquick Malaria +4	Biosynex	77	77	77	76	77	76	78	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Malaria Pf/Pv/vivax	Diagnostic Automation / Cortez Diagnostics, Inc.	22	19	28 (39)	14	17	26 (36)	30	16	16	16	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	72	73	70	78	77	76	79	79	79	79	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	

Product	Manufacturer	<i>P. falciparum</i> samples (n=79)										<i>P. vivax</i> samples (n=20)										
		Total positive results returned										Total positive results returned										
		200 parasites/µl					2,000 or 5,000 parasites/µl					200 parasites/µl					2,000 or 5,000 parasites/µl					
		Lot 1		Lot 2		No. positive agreements (max=79)*	Test 1	Test 2	Test 1	Test 2	No. positive agreements (max=79)*	Test 1	Test 2	Test 1	Test 2	No. positive agreements (max=20)*	Test 1	Test 2	Test 1	Test 2	No. positive agreements (max=20)*	Test 1
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	71	69	65	75	74	71	78	79	71	78	79	1	2	1	10	3	18	20	3	18	20
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	64	61	56	63	63	58	78	78	58	78	78	1	2	0	1	1	17	16	0	17	16
One Step Malaria Antigen Strip	IND Diagnostic Inc.	10	9	4	13	19	9	57	66	9	57	66	2	3	1	4	1	13	16	0	13	16
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	60	66	57	55	59	51	79	79	51	79	79	14	15	13	12	12	20	20	10	20	20
OptiMAL-IT	DiaMed AG	47	47	36	39	37	33	77	77	33	77	77	19	20	19	20	20	20	20	20	20	20
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	42	47	37	48	39	37	75	78	37	75	78	0	1	0	0	1	13	14	0	13	14
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	54	53	46	50	54	48	79	79	48	79	79	13	13	10	9	10	20	19	6	20	19
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	10	9	4	13	19	9	57	66	9	57	66	2	3	1	4	1	13	16	0	13	16
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	46	44	32	38	39	30	77	77	30	77	77	16	14	12	12	14	20	20	11	20	20
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	74	78	77	76	78	78	79	79	78	79	79	11	10	9	10	9	18	20	9	18	20
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	62	59	54	68	67	64	78	76	64	78	76	13	12	10	14	13/12	18	18	10	20	19
Pan only		(77)	(77)	(77)	(78)	(78)	(78)	(78)	(77)	(78)	(78)	(77)	(19)	(19)	(19)	(18)	(18)	(19)	(19)	(10)	(19)	(19)
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	65	68	62	72	68	66	79	79	66	79	79	20	20	20	20	20	20	20	20	20	20
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	77	77	75	77	77	76	79	79	76	79	79	20	20	20	20	20	20	20	20	20	20
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	8	16	5	10	11	4	73	72	4	73	72	11	10	8	8	9	20	20	7	20	20

Pf - *Plasmodium falciparum*

Pv - *Plasmodium vivax*

* Results are based on the first readers interpretation according to manufacturers instructions.

* Number of samples that returned a positive result for both tests. Where one test was invalid and the other positive, positive agreement was recorded.

Table A4.1a: Lot variability in positive results against *P. falciparum* and *P. vivax* samples at low (200) and high (2000 or 5000) parasite densities (parasites/μl) based on delayed readings when specified by the manufacturer

Product	Manufacturer	<i>P. falciparum</i> samples (n=79)										<i>P. vivax</i> samples (n=20)									
		Total positive results returned										Total positive results returned									
		200 parasites/μl					2,000 or 5,000 parasites/μl					200 parasites/μl					2,000 or 5,000 parasites/μl				
Lot 1		Lot 2		No. positive agreements (max=79)*	Test 1	Test 2	Test 1	Test 2	Test 1	Test 2	Test 1	Test 2	Test 1	Test 2	Test 1	Test 2	Test 1	Test 2	No. positive agreements (max=20)*		
Test 1	Test 2	Test 1	Test 2																	Test 1	Test 2
Pf & Pan/Pf & Pv FirstSign – ParaView-2 (Pv + Pf) Card Test Immunoquick Malaria +4	Unimed International, Inc.	66	63	60	64	58	55	79	79	2	3	1	0	4	0	4	0	20	20	0	
	Biosynex	76	77	74	79	79	79	78 (78)	79	13	15	13	14	15	12	14	15	20	19	12	
Malaria Pf/Vivax	Diagnostic Automation / Cortez Diagnostics, Inc.	24	20	30 (40)	15	17	27 (39)	96.77 (31)	88.24 (17)	0 (8)	0 (8)	0 (10)	0 (9)	0 (5)	0 (12)	0 (9)	0 (5)	6 (10)	2 (5)	0 (12)	
	Zephyr Biomedicals	64	64	58	65	68	61	78	79	1	1	0	4	2	1	4	2	17	18	1	
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	64	64	59	65	65	59	78	79	16	15	13	17	14 (19)	12	17	14 (19)	20	20	12	
	Guangzhou Wondfo Biotech Co., Ltd	64	63	59	68	65	61	78 (78)	76 (77)	12	13 (19)	11	12	12	9	12	12	18	19	9	
Pan only Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	24	19	15	27	27	20	77	77	17	16	16	18	18	18	18	18	20	20	18	

Pf - *Plasmodium falciparum*

Pv - *Plasmodium vivax*

* Number of samples that returned a positive result for both tests. Where one test was invalid and the other positive, positive agreement was recorded.

Table A4.2: Reader variability in detection rate* of *P. falciparum* samples (n=79) based on initial reading according to manufacturers instructions and second reading within 1 hour

Product	Manufacturer	Detection rate of <i>P. falciparum</i> samples									
		200 parasites/ μ l					2,000 or 5,000 parasites/ μ l				
		Reader 1 (n=316)	Reader 2 (n=316)	No. invalid tests Reader 1	No. invalid tests Reader 2	Mean time between readings (h:m:s)	Reader 1 (n=158)	Reader 2 (n=158)	No. invalid tests Reader 1	No. invalid tests Reader 2	Mean time between readings (h:m:s)
Pf only											
ADVANCED QUALITY TM MALARIA (p.f) POCT	InTec Products, Inc.	76.9	85.13	0	0	0:24:40	100	100	0	0	0:22:10
ADVANCED QUALITY TM One Step Malaria (p.f) Test (whole blood)	InTec Products, Inc.	87.97	89.87	0	0	0:16:23	100	99.37	0	0	0:16:24
Advantage P.f. Malaria Card	J. Mitra & Co. Pvt. Ltd.	98.73	99.37	0	0	0:13:47	100	100	0	0	0:14:58
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	99.68	99.05	0	0	0:18:14	99.37	100	0	0	0:18:55
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	100	100	0	0	0:13:58	100	100	0	0	0:13:08
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	49.68	62.66	0	0	0:19:56	92.41	99.37	0	0	0:19:46
Hexagon Malaria	Human GmbH	58.84 (311)	67.2 (311)	5	5	0:27:08	97.44 (156)	97.44 (156)	2	2	0:28:22
ICT Malaria Pf Cassette Test (ML01)	ICT Diagnostics	93.35	96.84	0	0	0:17:42	98.73	100	0	0	0:17:21
Immunoquick Malaria Falciparum	Biosynex	96.2	98.1	0	0	0:17:36	100	100	0	0	0:17:38
Malaria <i>Plasmodium falciparum</i> Rapid test Device (Whole blood)	ACON Laboratories, Inc.	96.52	97.78	0	0	0:21:40	100	100	0	0	0:21:36
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	87.34	92.09	0	0	0:19:04	98.73	99.37	0	0	0:18:12
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	68.63 (306)	89.74 (312)	10	4	0:18:05	98.68 (152)	99.36 (156)	6	2	0:17:33
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	86.03 (315)	90.48 (315)	1	1	0:15:43	100 (156)	100 (157)	2	1	0:16:02
Parahit-F DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	89.24	90.82	0	0	0:17:46	100	100	0	0	0:16:15
Parahit-F TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	59.18	62.66	0	0	0:17:16	98.73	100	0	0	0:25:44
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	99.37	99.68	0	0	0:15:13	99.37	100	0	0	0:14:46
Pf & Pan/Pf & Pv											
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	77.22	82.28	0	0	0:13:22	100	99.37	0	0	0:13:36
AZOG Malaria pf (HRP-II) / pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	89.56	94.94	0	0	0:15:29	100	100	0	0	0:15:32
Binax Now Malaria	Inverness Medical Innovations, Inc.	96.84	94.62	0	0	0:17:32	100 (157)	100 (157)	1	1	0:17:19
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	99.05	97.47	0	0	0:16:01	100	98.73	0	0	0:15:16
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	100	99.68	0	0	0:17:45	100	99.37	0	0	0:17:49
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	66.14	79.43	0	0	0:24:36	99.37	100	0	0	0:21:49
Hexagon Malaria Combi	Human GmbH	63.29	67.72	0	0	0:20:39	98.73 (157)	98.73	1	0	0:19:36
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	95.25	98.73	0	0	0:16:31	100	100	0	0	0:17:32
Immunoquick Malaria +4	Biosynex	97.15	98.42	0	0	0:20:39	99.37	100 (157)	0	1	0:21:23
Malaria Rapid Combo	Diagnostic Automation / Cortez Diagnostics, Inc.	74.23 (97)	74.51 (102)	219	214	0:16:43	95.83 (48)	93.75 (48)	110	110	0:17:14
Malaria P-FMvax	Vision Biotech (Pty) Ltd.	94.94	93.99	0	0	0:15:49	100	99.37	0	0	0:15:38
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	91.46	97.15	0	0	0:13:08	99.37	98.73	0	0	0:13:23
Malasscan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	79.43	82.59	0	0	0:18:13	98.73	99.37	0	0	0:17:50
One Step Malaria Antigen Strip	IND Diagnostic Inc.	16.14	22.47	0	0	0:17:32	77.85	79.75	0	0	0:17:19
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	75.95	88.61	0	0	0:19:59	100	100	0	0	0:19:17
OptiMAL-IT	DiaMed AG	53.8	46.84	0	0	0:18:52	97.47	97.47	0	0	0:19:40
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	55.87 (315)	55.06	1	0	0:16:44	96.84	97.47	0	0	0:29:58
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	66.77	81.65	0	0	0:16:05	100	99.37	0	0	0:16:14
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	16.14	22.47	0	0	0:17:32	77.85	79.75	0	0	0:17:19
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	52.85	61.71	0	0	0:18:14	97.47	98.1	0	0	0:17:36
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	98.71 (310)	98.71 (311)	6	5	0:16:42	100	100	0	0	0:16:16
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	82.05 (312)	83.33 (312)	4	4	0:15:02	99.35 (155)	99.35 (155)	3	3	0:19:51
Pan only											
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	86.39	93.35	0	0	0:13:25	100	100	0	0	0:14:01
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	97.47	95.89	0	0	0:15:24	100	100	0	0	0:15:35
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	14.24	30.7	0	0	0:15:52	91.77	97.47	0	0	0:15:29
PF - <i>Plasmodium falciparum</i> Pv - <i>Plasmodium vivax</i> pan - <i>Plasmodium</i> species * - A sample is considered detected only if all RDTs from both lots read by the first technician, at minimum specified reading time, are positive											

Table A4.3: Reader variability in detection rate* for *P. vivax* samples (n=20) based on initial reading according to manufacturers instructions and second reading within 1 hour

Product	Manufacturer	Detection rate for <i>P. vivax</i> samples						Mean time between readings (h:m:s)		
		200 parasites/µl			2,000 or 5,000 parasites/µl					
		Reader 1 (n=80)	Reader 2 (n=80)	No. invalid tests Reader 1	No. invalid tests Reader 2	Reader 1 (n=40)	Reader 2 (n=40)	No. invalid tests Reader 1	No. invalid tests Reader 2	Mean time between readings (h:m:s)
PF & Pan/PF & Pv										
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	100	98.75	0	0	100	95	0	0	0:11:06
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	61.25	67.5	0	0	97.5	92.5	0	0	0:12:09
Binax Now Malaria	Inverness Medical Innovations, Inc.	24.05 (79)	5	1	0	92.5	95	0	0	0:12:37
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	97.5	95	0	0	97.5	97.5	0	0	0:11:35
First Response Malaria Ag Combo (pLDH/HRP2)	Premier Medical Corporation Ltd.	92.5	93.75	0	0	80	82.5	0	0	0:13:48
FirstSign - ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	7.5	11.25	0	0	90	100	0	0	0:15:27
Hexagon Malaria Combi	Human GmbH	1.27 (79)	1.27 (79)	1	1	60.53 (38)	57.89 (38)	2	2	0:22:15
ICT Malaria Combo Cassette Test (MI02)	ICT Diagnostics	20	43.75	0	0	95	92.5	0	0	0:16:56
Immunoquick Malaria +4	Biosynex	60	71.25	0	0	100	97.5	0	0	0:15:54
Malaria PF/Vvax	Diagnostic Automation / Cortez Diagnostics, Inc.	0 (30)	0 (30)	50	50	33.33 (18)	53.33 (15)	22	25	0:18:14
Malaria Rapid Combo	Malaria Biotech (Pty) Ltd.	37.5	35	0	0	92.5	95	0	0	0:15:16
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	20	38.75	0	0	95	97.5	0	0	0:13:23
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	6.25	10	0	0	82.5	87.5	0	0	0:16:02
One Step Malaria Antigen Strip	IND Diagnostic Inc.	12.5	10	0	0	72.5	65	0	0	0:10:18
OnSight - ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	66.25	91.25	0	0	100	100	0	0	0:16:51
OptiMAL-IT	DiaMed AG	98.75	88.75	0	0	100	95	0	0	0:17:36
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	2.5	2.5	0	0	67.5	55	0	0	0:17:09
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	56.96 (79)	78.48 (79)	1	1	97.5	100	0	0	0:17:00
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	12.5	10	0	0	72.5	65	0	0	0:10:18
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	70	83.75	0	0	100	97.5	0	0	0:17:03
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	50.63 (79)	69.62 (79)	1	1	97.44 (39)	97.44 (39)	1	1	0:16:33
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	66.23 (77)	62.34 (77)	3	3	97.44 (39)	94.87 (39)	1	1	0:17:57
Pan only										
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	100	100	0	0	100	100	0	0	0:11:18
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	100	100	0	0	100	97.5	0	0	0:11:42
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	47.5	86.25	0	0	100	100	0	0	0:16:59

PF - *Plasmodium falciparum*
 Pv - *Plasmodium vivax*
 pan - *Plasmodium* species
 * - A sample is considered detected only if all RDITs from both lots read by the first technician, at minimum specified reading time, are positive

Table A4.4: Distribution of test band intensity (0-4) scores against wild type *P. falciparum* samples (n=79) at low (200) and high (2000 or 5000) parasite densities (parasites/µl)

Product	Manufacturer	200 parasites/µl Percentage distribution of Pf test band intensity* (n=316)					2,000 or 5,000 parasites/µl Percentage distribution of Pf test band intensity* (n=158)					200 parasites/µl Percentage distribution of Pan test band intensity (n=316)					2,000 or 5,000 parasites/µl Percentage distribution of Pan test band intensity (n=158)				
		0*	1	2	3	4	0*	1	2	3	4	No band	1	2	3	4	No band	1	2	3	4
Pf only																					
ADVANCED QUALITY TM MALARIA (p.f) POCT	InTec Products, Inc.	23.1	32.91	34.49	7.59	1.9	0	1.27	29.11	29.11	40.51	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
ADVANCED QUALITY TM One Step Malaria (p.f) Test (whole blood)	InTec Products, Inc.	12.03	36.08	34.18	14.56	3.16	0	2.53	22.78	24.05	50.63	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Advantage Pf. Malaria Card	J. Mitra & Co. Pvt. Ltd.	1.27	12.03	42.09	30.38	14.24	0	0	1.9	22.15	75.95	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	0.32	6.01	30.7	29.11	33.86	0.63	0	3.16	12.66	83.54	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	0	6.33	32.91	27.53	33.23	0	0.63	1.27	10.13	87.97	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	50.32	25.95	16.14	6.01	1.58	7.59	10.13	27.22	31.65	23.42	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Hexagon Malaria	Human GmbH	42.09	29.11	23.1	4.11	1.58	3.8	6.96	32.28	37.34	19.62	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
ICT Malaria Pf Cassette Test (ML01)	ICT Diagnostics	6.65	27.53	34.81	24.37	6.65	1.27	0	13.29	29.11	56.33	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Immunoquick Malaria Falciparum	Biosynex	3.8	18.04	33.86	34.49	9.81	0	0	11.39	25.32	63.29	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Malaria <i>Plasmodium falciparum</i> Rapid test Device (Whole blood)	ACON Laboratories, Inc.	3.48	19.3	37.66	32.91	6.65	0	0.63	6.96	32.91	59.49	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	12.86	22.15	39.56	20.89	4.75	1.27	1.9	17.09	29.11	50.63	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	33.23	15.51	22.47	20.89	7.91	3.16	4.43	10.13	25.95	56.33	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	14.24	24.05	43.67	12.97	5.06	1.27	0.63	15.82	53.8	28.48	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Parahit-F DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	10.76	22.47	37.97	20.57	8.23	0	0	6.33	26.58	67.09	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Parahit-F TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	40.82	21.84	31.01	5.38	0.95	1.27	4.43	34.81	29.11	30.38	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	0.63	2.53	36.71	37.34	22.78	0.63	0	0	12.66	86.71	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Pf & Pan/Pf & Pv																					
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	22.78	51.58	22.47	2.85	0.32	0	1.9	24.05	56.96	17.09	31.01	52.85	14.56	1.58	0	4.43	53.16	31.65	10.76	
AZOG Malaria of (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	10.44	20.57	36.39	22.78	9.81	0	0.63	4.43	31.65	63.29	57.91	37.03	5.06	0	3.8	32.91	46.84	12.03	4.43	
Binax Now Malaria	Inverness Medical Innovations, Inc.	3.16	17.09	45.89	25.63	8.23	0	0	8.23	27.22	64.56	73.42	18.04	8.23	0.32	0	6.33	15.82	58.86	17.72	1.27
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	0.95	7.59	30.7	37.97	22.78	0	0	2.53	15.19	82.28	0.95	30.06	57.28	10.13	1.58	0	0.63	7.59	41.14	50.63
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	0	4.43	20.25	29.11	46.2	0	0	0.63	8.23	91.14	5.7	36.08	44.62	12.03	1.58	0	1.27	15.82	32.28	50.63
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	33.86	17.41	25.63	14.24	8.86	0.63	2.53	12.66	24.68	59.49	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Hexagon Malaria Combi	Human GmbH	36.71	28.16	24.05	9.81	1.27	1.9	4.43	27.85	36.71	29.11	99.68	0.32	0	0	80.38	17.72	1.9	0	0	
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	4.75	14.56	35.76	25.63	19.3	0	0	2.53	16.46	81.01	74.68	21.52	3.48	0.32	0	8.23	30.38	39.87	18.99	2.53
Immunoquick Malaria +4	Biosynex	2.85	7.28	28.8	35.44	25.63	0.63	0	0	13.29	86.08	74.52	21.34	3.5	0.64	0	4.46	14.65	42.04	37.58	1.27
Malaria Pf/Vvax	Diagnostic Automation / Cortez Diagnostics, Inc.	56.96	10.76	14.56	12.97	4.75	31.65	7.59	14.56	18.35	27.85	98.1	1.58	0.32	0	0	88.61	4.43	5.7	1.27	0
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	5.06	22.78	41.46	20.25	10.44	0	0.63	8.23	22.15	68.99	61.71	29.11	8.86	0.32	0	3.8	27.85	44.3	20.25	3.8
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	8.54	23.42	44.94	15.51	7.59	0.63	2.53	11.39	23.42	62.03	71.84	26.58	1.58	0	19.62	27.85	43.67	7.59	1.27	
Malerascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	20.57	28.16	39.56	10.76	0.95	1.27	1.9	19.62	43.04	34.18	91.77	6.96	1.27	0	18.99	32.28	31.01	16.46	1.27	
One Step Malaria Antigen Strip	IND Diagnostic Inc.	83.86	15.19	0.63	0.32	0	22.15	27.22	34.81	15.82	0	83.23	15.82	0.95	0	23.42	30.38	34.18	12.03	0	
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	24.05	13.61	25.32	21.84	15.19	0	0.63	3.8	22.78	72.78	77.53	14.24	6.96	1.27	0	8.86	6.33	32.28	26.58	25.95

Product	Manufacturer	200 parasites/µl					2,000 or 5,000 parasites/µl					200 parasites/µl					2,000 or 5,000 parasites/µl				
		Percentage distribution of Pf test band intensity* (n=316)					Percentage distribution of Pf test band intensity* (n=158)					Percentage distribution of Pan test band intensity (n=316)					Percentage distribution of Pan test band intensity (n=158)				
		0*	1	2	3	4	0*	1	2	3	4	No band	1	2	3	4	No band	1	2	3	4
OptiMAL-IT	DiaMed AG	46.2	41.77	11.08	0.95	0	2.53	10.13	39.24	32.91	15.19	49.05	40.19	9.81	0.95	0	2.53	13.29	40.51	31.65	12.03
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	44.3	20.57	25.63	7.59	1.9	3.16	6.33	28.48	27.22	34.81	99.68	0.32	0	0	0	72.78	17.72	9.49	0	0
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	33.23	14.56	26.9	16.14	9.18	0	0.63	15.82	20.89	62.66	86.08	7.59	5.7	0.63	0	10.13	13.29	42.41	21.52	12.66
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	83.86	15.19	0.63	0.32	0	22.15	27.22	34.81	15.82	0	83.23	15.82	0.95	0	0	23.42	30.38	34.18	12.03	0
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	47.15	40.82	11.71	0.32	0	2.53	3.8	57.59	31.65	4.43	96.2	3.8	0	0	0	26.58	44.3	29.11	0	0
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	3.16	14.24	38.92	26.27	17.41	0	0	3.8	14.56	81.65	91.14	7.91	0.95	0	0	12.03	37.34	46.84	3.8	0
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	18.99	37.34	29.11	10.13	4.43	2.53	3.8	18.99	35.44	39.24	34.81	45.57	14.56	4.75	0.32	3.16	6.33	36.08	34.18	20.25
Pan only																					
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	13.61	47.15	34.49	4.75	0	0	0	17.72	56.33	25.95	13.61	47.15	34.49	4.75	0	0	0	17.72	56.33	25.95
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	2.53	34.81	55.7	6.01	0.95	0	0.63	12.03	41.77	45.57	2.53	34.81	55.7	6.01	0.95	0	0.63	12.03	41.77	45.57
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	85.76	11.39	1.58	0.95	0.32	8.23	15.19	28.48	32.28	15.82	85.76	11.39	1.58	0.95	0.32	8.23	15.19	28.48	32.28	15.82

Pf - *Plasmodium falciparum*
Pv - *Plasmodium vivax*
pan - *Plasmodium* species

Table A4.4a: Distribution of test band intensity (0-4) scores against wild type *P. falciparum* samples (n=79) at low (200) and high (2000 or 5000) parasite densities (parasites/µl) based on a delayed reading when specified by the manufacturer

Product	Manufacturer	200 parasites/µl					2,000 or 5,000 parasites/µl					200 parasites/µl					2,000 or 5,000 parasites/µl				
		Percentage distribution of Pf test band intensity* (n=316)					Percentage distribution of Pf test band intensity* (n=158)					Percentage distribution of Pan test band intensity (n=316)					Percentage distribution of Pan test band intensity (n=158)				
		0*	1	2	3	4	0*	1	2	3	4	No band	1	2	3	4	No band	1	2	3	4
Pf & Pan/Pf & Pv																					
FirstSign - ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	20.57	14.24	25	25.63	14.56	0	1.27	3.16	22.78	72.78	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Immunoquick Malaria +4	Biosynex	1.58	4.75	27.22	39.87	26.58	0.63	0	0.63	15.19	83.54	55.73	35.03	7.96	0.96	0.32	1.27	8.92	33.12	50.96	5.73
Malaria r:PF/vivax	Diagnostic Automation / Cortez Diagnostics, Inc.	59.81	6.01	13.61	13.92	6.65	35.44	2.53	10.76	24.05	27.22	99.68	0.32	0	0	0	84.81	5.06	8.23	1.9	0
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	17.41	21.2	46.2	13.29	1.9	0.63	0.63	19.62	30.38	48.73	78.8	17.41	3.8	0	0	10.13	17.72	44.94	24.05	3.16
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	18.35	11.71	34.81	20.89	14.24	0.63	0	7.59	16.46	75.32	69.3	20.89	8.23	1.58	0	4.43	12.03	31.65	31.01	20.89
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	17.72	33.54	30.7	11.08	6.96	2.53	2.53	18.35	27.22	49.37	28.48	41.46	25	3.16	1.9	2.53	3.16	25.95	36.71	31.65
Pan only																					
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	69.3	20.89	8.54	0	1.27	2.53	6.96	29.11	32.91	28.48	69.3	20.89	8.54	0	1.27	2.53	6.96	29.11	32.91	28.48

Pf - *Plasmodium falciparum*
Pv - *Plasmodium vivax*
pan - *Plasmodium* species

Table A4.5: Distribution of pan/Pv test band intensity (0–4) scores for *P. vivax* (n=20) samples at low (200) and high (2000 or 5000) parasite densities (parasites/µl)

Product	Manufacturer	200 parasites/µl				2,000 or 5,000 parasites/µl						
		0	1	2	3	4	0	1	2	3	4	
Pf & Pan/Pf & Pv												
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	0	1.25	46.25	51.25	1.25	0	0	0	0	5	95
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	38.75	48.75	11.25	1.25	0	0	2.5	12.5	60	25	25
BinaX Now Malaria	Inverness Medical Innovations, Inc.	75	23.75	1.25	0	0	5	42.5	45	2.5	2.5	2.5
CareStart Malaria HRP2(pLDH) (Pf/PAN) COMBO	Access Bio, Inc.	3.75	13.75	71.25	13.75	0	0	0	5	22.5	72.5	72.5
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	3.75	18.75	66.25	10	1.25	0	0	0	22.5	77.5	77.5
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	92.5	6.25	1.25	0	0	10	5	52.5	32.5	0	0
Hexagon Malaria Combi	Human GmbH	98.75	1.25	0	0	0	40	37.5	22.5	0	0	0
ICT Malaria Combo Cassette Test (MI02)	ICT Diagnostics	77.5	22.5	0	0	0	0	2.5	52.5	37.5	7.5	7.5
Immunoquick Malaria +4	Biosynex	40	43.75	16.25	0	0	0	0	10	75	15	15
Malaria Pf/Vivax	Diagnostic Automation / Cortez Diagnostics, Inc.	100	0	0	0	0	85	0	15	0	0	0
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	55	37.5	7.5	0	0	0	2.5	55	40	2.5	2.5
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	80	20	0	0	0	5	42.5	45	7.5	0	0
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	93.75	5	1.25	0	0	2.5	5	50	40	2.5	2.5
One Step Malaria Antigen Strip	IND Diagnostic Inc.	86.25	13.75	0	0	0	27.5	15	32.5	20	5	5
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	32.5	20	38.75	8.75	0	0	0	5	20	75	75
OptiMAL-IT	DiaMed AG	1.25	31.25	53.75	13.75	0	0	0	0	22.5	77.5	77.5
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	97.5	2.5	0	0	0	32.5	37.5	27.5	2.5	0	0
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	40	17.5	38.75	2.5	1.25	2.5	2.5	7.5	25	62.5	62.5
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	86.25	13.75	0	0	0	27.5	15	32.5	20	5	5
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	28.75	36.25	35	0	0	0	0	22.5	57.5	20	20
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	50	35	15	0	0	2.5	0	35	50	12.5	12.5
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	35	45	20	0	0	2.5	2.5	15	45	35	35
Pan only												
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	0	10	73.75	16.25	0	0	0	0	10	90	90
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	0	5	61.25	33.75	0	0	0	0	5	95	95
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	52.5	16.25	26.25	5	0	0	0	2.5	12.5	85	85
Pf - Plasmodium falciparum												
Pv - Plasmodium vivax												
pan - Plasmodium species												

Table A4.5a: Distribution of pan/Pv test band intensity (0–4) scores for *P. vivax* (n=20) samples at low (200) and high (2000 or 5000) parasite densities (parasites/µl) samples based on a delayed reading when specified by the manufacturer

Product	Manufacturer	200 parasites/µl				2,000 or 5,000 parasites/µl						
		0	1	2	3	4	0	1	2	3	4	
Pf & Pan/Pf & Pv												
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.											
Immunoquick Malaria +4	Biosynex	27.5	37.5	35	0	0	0	0	7.5	52.5	40	40
Malaria Pf/Vivax	Diagnostic Automation / Cortez Diagnostics, Inc.	100	0	0	0	0	80	10	10	0	0	0
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	86.25	11.25	2.5	0	0	0	2.5	52.5	35	10	10
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	22.5	22.5	35	20	0	0	2.5	2.5	22.5	72.5	72.5
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	36.25	40	22.5	1.25	0	2.5	5	7.5	37.5	47.5	47.5
Pan only												
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	13.75	28.75	43.75	13.75	0	0	0	2.5	12.5	85	85
Pf - Plasmodium falciparum												
Pv - Plasmodium vivax												
pan - Plasmodium species												

Table A4.6 Detection rate* of *P. falciparum* in low (200) and high (2000 or 5000) parasite densities (parasites/µl) by continent

Product	Manufacturer	200 parasites/µl			2000 or 5000 parasites/µl		
		Africa (n=49)	Asia (n=22)	South America (n=8)	Africa (n=49)	Asia (n=22)	South America (n=8)
Pf only							
ADVANCED QUALITY™ MALARIA (p.f.) POCT	InTec Products, Inc.	57.14	63.64	37.5	100	100	100
ADVANCED QUALITY™ One Step Malaria (p.f.) Test (whole blood)	InTec Products, Inc.	71.43	59.09	62.5	100	100	100
Advantage P.f. Malaria Card	J. Mitra & Co. Pvt. Ltd.	97.96	100	87.5	100	100	100
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	97.96	100	100	97.96	100	100
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	100	100	100	100	100	100
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	34.69	31.82	12.5	85.71	90.91	75
Hexagon Malaria	Human GmbH	36.73	45.45	37.5	93.88	95.45	100
ICT Malaria Pf Cassette Test (ML01)	ICT Diagnostics	79.59	86.36	87.5	100	90.91	100
Immunoquick Malaria Falciparum	Biosynex	85.71	100	100	100	100	100
Malaria Plasmodium falciparum Rapid test Device (Whole blood)	ACON Laboratories, Inc.	89.8	95.45	100	100	100	100
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	65.31	77.27	62.5	97.96	100	87.5
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	55.1	59.09	37.5	95.92	100	100
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	73.47	77.27	75	100	100	100
Parahit-f DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	79.59	81.82	62.5	100	100	100
Parahit-f TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	42.86	36.36	25	100	95.45	87.5
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	100	90.91	100	97.96	100	100
Pf & Pan/Pf & Pv							
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	69.39	59.09	25	100	100	100
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	71.43	86.36	87.5	100	100	100
BinaX Now Malaria	Inverness Medical Innovations, Inc.	89.8	95.45	87.5	100	100	100
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	95.92	100	100	100	100	100
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	100	100	100	100	100	100
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	46.94	63.64	12.5	97.96	100	100
Hexagon Malaria Combi	Human GmbH	46.94	54.55	25	97.96	100	87.5
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	85.71	86.36	87.5	100	100	100
Immunoquick Malaria +4	Biosynex	91.84	95.45	100	97.96	100	100
Malaria Pf/Mvax	Diagnostic Automation / Cortez Diagnostics, Inc.	68.75 (32)	71.43 (14)	100 (7)	92.59 (27)	100 (8)	100 (4)
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	87.76	90.91	75	100	100	100
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	77.55	72.73	75	97.96	100	100
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	59.18	72.73	62.5	95.92	100	100
One Step Malaria Antigen Strip	IND Diagnostic Inc.	2.04	0	0	79.59	50	37.5
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	61.22	59.09	50	100	100	100
OptiMAL-IT	DiaMed AG	48.98	22.73	0	95.92	100	87.5
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	34.69	40.91	25	95.92	95.45	75
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	51.02	50	50	100	100	100

Product	Manufacturer	200 parasites/ μ l			2000 or 5000 parasites/ μ l		
		Africa (n=49)	Asia (n=22)	South America (n=8)	Africa (n=49)	Asia (n=22)	South America (n=8)
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	2.04	0	0	79.59	50	37.5
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	40.82	13.64	0	97.96	95.45	87.5
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	97.96	95.45	87.5	100	100	100
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	63.27	63.64	37.5	97.96	100	100
Pan only							
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	75.51	68.18	62.5	100	100	100
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	91.84	95.45	87.5	100	100	100
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	2.04	0	0	89.8	95.45	50

Africa - Tanzania, Central African Republic, Madagascar, Nigeria
 Asia - Myanmar, Philippines, Cambodia
 South America - Peru, Colombia

Pf - *Plasmodium falciparum*
 Pv - *Plasmodium vivax*

pan - *Plasmodium* species

* - A sample is considered detected only if all RDTs from both lots read by the first technician, at minimum specified reading time, are positive

Table A4-6a: Detection rate of *P. falciparum* in low (200) and high (2000 or 5000) parasite densities (parasites/ μ l) by continent based on a delayed reading as specified by the manufacturer

Product	Manufacturer	200 parasites/ μ l			2000 or 5000 parasites/ μ l		
		Africa (n=49)	Asia (n=22)	South America (n=8)	Africa (n=49)	Asia (n=22)	South America (n=8)
Pf & Pan/Pf & Pv							
FirstSign - ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	63.27	72.73	50	100	100	100
Immunoquick Malaria +4	Biosynex	95.92	86.36	100	100	100	100
Malaria Pf/Mvax	Diagnostic Automation / Cortez Diagnostics, Inc.	64.71 (34)	73.33 (15)	100 (7)	89.29 (28)	100 (8)	100 (4)
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	61.22	81.82	75	97.96	100	100
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	69.39	59.09	62.5	97.96	100	100
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	69.39	68.18	62.5	97.96	100	100
Pan only							
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	22.45	0	0	97.96	100	87.5

Africa - Tanzania, Central African Republic, Madagascar, Nigeria
 Asia - Myanmar, Philippines, Cambodia
 South America - Peru, Colombia

Pf - *Plasmodium falciparum*
 Pv - *Plasmodium vivax*

pan - *Plasmodium* species

* - a sample is considered detected only if all RDTs from both lots read by the first technician, at minimum specified reading time, are positive

Table A4.7 *P. falciparum* test line false positive results for *P. vivax* samples (n=20) at low (200) and high (2000 or 5000) parasite densities (parasites/μl)

Product	Manufacturer	<i>P. vivax</i> samples (n=20)					
		200 parasites/μl			2000 or 5000 parasites/μl		
		False positive Pf infection ^φ			False positive Pf infection ^φ		
	Lot 1 (n=40)	Lot 2 (n=40)	Overall (n=80)	Lot 1 (n=20)	Lot 2 (n=20)	Overall (n=40)	
Pf only							
ADVANCED QUALITY TM MALARIA (p.f) POCT	InTec Products, Inc.	7.5	17.5	12.5	20	15	17.5
ADVANCED QUALITY TM One Step Malaria (p.f.) Test (whole blood)	InTec Products, Inc.	52.5	45	48.75	50	40	45
Advantage Pf: Malaria Card	J. Mitra & Co. Pvt. Ltd.	2.5	0	1.25	5	0	2.5
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	10	0	5	10	5	7.5
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	0	0	0	0	0	0
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	5	20	12.5	15	15	15
Hexagon Malaria	Human GmbH	5.41 (37)	10.26 (39)	7.89 (76)	0	5	2.5
ICT Malaria Pf Cassette Test (MI01)	ICT Diagnostics	0 (39)	2.5	1.27 (79)	5	0	2.5
Immunoquick Malaria Falciparum	Biosynex	0	0	0	0	0	0
Malaria <i>Plasmodium falciparum</i> Rapid test Device (Whole blood)	ACON Laboratories, Inc.	0	0	0	0	0	0
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	0	0	0	0	0	0
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	2.63 (38)	5.26 (38)	3.95 (76)	5	5	5
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	7.5	25.64 (39)	16.46 (79)	10	10	10
Parahit-F DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	0	0	0	0	0	0
Parahit-F TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	0	0	0	0	0	0
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	0	0	0	0	0	0
Pf & Pan/Pf & Pv							
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	0	0	0	0	0	0
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	0	0	0	0	5	2.5
Binax Now Malaria	Inverness Medical Innovations, Inc.	2.56 (39)	5	3.8 (79)	0	10	5
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	2.5	0	1.25	5	5	2.5
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	2.5	5	3.75	25	15	20
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	0	7.5	3.75	0	10	5
Hexagon Malaria Combi	Human GmbH	0	0 (39)	0 (79)	5.26 (19)	0 (19)	2.63 (38)
ICT Malaria Combo Cassette Test (MI02)	ICT Diagnostics	5	2.5	3.75	5	5	5
Immunoquick Malaria +4	Biosynex	0	0	0	0	0	0
Malaria PfMvax	Diagnostic Automation / Cortez Diagnostics, Inc.	0 (14)	0 (16)	0 (30)	0 (10)	0 (8)	0 (18)
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	7.5	7.5	7.5	5	10	7.5
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	0	0	0	0	0	0
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	5	7.5	6.25	15	20	17.5
One Step Malaria Antigen Strip	IND Diagnostic Inc.	0	7.5	3.75	0	0	0
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	0	2.5	1.25	0	0	0
OptiMAL-IT	DiaMed AG	0	0	0	0	0	0
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	0	0	0	5	0	2.5
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	5	2.56 (39)	3.8 (79)	0	0	0
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	0	7.5	3.75	0	0	0
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	2.5	0	1.25	0	0	0
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	0 (39)	0	0 (79)	5.26 (19)	0	2.56 (39)
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	0 (39)	2.63 (38)	1.3 (77)	0	5.26 (19)	2.56 (39)
Pan only							
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	N/A	N/A	N/A	N/A	N/A	N/A
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	N/A	N/A	N/A	N/A	N/A	N/A
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	N/A	N/A	N/A	N/A	N/A	N/A
Pf - <i>Plasmodium falciparum</i>							
Pv - <i>Plasmodium vivax</i>							
pan - <i>Plasmodium</i> species							

^φ - Pf line positive indicates a false positive *P. falciparum* infection

Table A4.7a: *P. falciparum* test line false positive results for *P. vivax* samples (n=20) at low (200) and high (2000 or 5000) parasite densities (parasites/µl) based on a delayed reading when specified by the manufacturer

Product	Manufacturer	<i>P. vivax</i> samples (n=20)					
		200 parasites/µl		2000 or 5000 parasites/µl		2000 or 5000 parasites/µl	
		False positive PF infection [∅]		False positive PF infection [∅]		False positive PF infection [∅]	
	Lot 1 (n=40)	Lot 2 (n=40)	Overall (n=80)	Lot 1 (n=20)	Lot 2 (n=20)	Overall (n=40)	
PF & Pan/Pf & Pv							
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	2.5	2.5	2.5	0	5	2.5
Immunoquick Malaria +4	Biosynex	0	2.5	1.25	0	5	2.5
Malaria P.F/Vivax	Diagnostic Automation / Cortez Diagnostics, Inc.	0 (16)	0 (14)	0 (30)	0 (10)	0 (5)	0 (15)
Malacsen Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	5	10	7.5	15	10	12.5
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	0	0 (39)	0 (79)	0	0	0
Wondfo One-Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	2.56 (39)	5.26 (38)	3.9 (77)	10	0 (19)	5.13 (39)
Pan only							
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	N/A	N/A	N/A	N/A	N/A	N/A

Pf - *Plasmodium falciparum*

Pv - *Plasmodium vivax*

pan - *Plasmodium* species

∅ - PF line positive indicates a false positive *P. falciparum* infection

Table A4.8: Combination test pan line false positive non Pf infection on Pf samples (n=79) at low and high parasite densities (parasites/μl)

Product	Manufacturer	<i>P. falciparum</i> samples (n=79)				
		200 parasites/μl		2000 or 5000 parasites/μl		Overall (n=158)
		Lot 1 (n=158)	Lot 2 (n=158)	Lot 1 (n=79)	Lot 2 (n=79)	
Pf & Pan/Pf & Pv						
Advantage Mal Card	J. Mitra & Co. Pvt Ltd.	2.53	2.53	0	0	0
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	1.9	1.9	0	0	0
Binx Now Malaria	Inverness Medical Innovations, Inc.	0.63	0	0 (78)	0	0 (157)
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	0	0.63	0	0	0
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	0	0	0	0	0
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	0	1.9	0.95	0	0
Hexagon Malaria Combi	Human GmbH	0	0	0 (78)	0	0 (157)
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	0	0.63	0	0	0
Immunoquick Malaria +4	Biosynex	0	0 (156)	0 (314)	0	0 (157)
Malaria Pf/Vvax	Diagnostic Automation / Cortez Diagnostics, Inc.	1.79 (56)	0 (41)	1.03 (97)	0 (31)	5.88 (17)
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	0.63	0	0.32	0	0
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	0.63	1.9	1.27	0	0
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	0.63	0.63	0.63	0	0
One Step Malaria Antigen Strip	IND Diagnostic Inc.	1.9	2.53	2.22	1.27	2.53
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	0	0	0	0	0
OptiMAL-IT	DiaMed AG	4.43	3.16	3.8	0	1.27
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	0	0 (315)	0 (315)	0	0
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	0.63	0.63	0.63	0	0
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	1.9	2.53	2.22	1.27	2.53
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	0	0	0	0	0
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	0	0 (155)	0 (310)	0	0
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	3.21 (156)	3.85 (156)	3.52 (312)	0 (78)	0 (77)
Pf - Plasmodium falciparum						
Pf & Pan/Pf & Pv						
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	0	0	0	0	0
Immunoquick Malaria +4	Biosynex	0	0 (156)	0 (314)	0 (78)	0 (156)
Malaria Pf/Vvax	Diagnostic Automation / Cortez Diagnostics, Inc.	0 (58)	0 (44)	0 (102)	3.23 (31)	5.88 (17)
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	1.9	1.27	1.58	0	0
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	0.63	0.63	0.63	1.27	0
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	2.56 (156)	1.28 (156)	1.92 (312)	0 (78)	0 (77)
Pf - Plasmodium falciparum						
Pv - Plasmodium vivax						
pan - <i>Plasmodium</i> species						
1 - For combination tests, Pan or Pv line, only, positive indicates a false positive non <i>P. falciparum</i> infection						

Table A4.8a: Combination test pan line false positive non Pf infection on Pf samples (n=79) at low and high parasite densities (parasites/μl) based on a delayed reading when specified by the manufacturer

Product	Manufacturer	<i>P. falciparum</i> samples (n=79)				
		200 parasites/μl		2000 or 5000 parasites/μl		Overall (n=158)
		Lot 1 (n=158)	Lot 2 (n=158)	Lot 1 (n=79)	Lot 2 (n=79)	
Pf & Pan/Pf & Pv						
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	0	0	0	0	0
Immunoquick Malaria +4	Biosynex	0	0 (156)	0 (314)	0 (78)	0 (156)
Malaria Pf/Vvax	Diagnostic Automation / Cortez Diagnostics, Inc.	0 (58)	0 (44)	0 (102)	3.23 (31)	5.88 (17)
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	1.9	1.27	1.58	0	0
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	0.63	0.63	0.63	1.27	0
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	2.56 (156)	1.28 (156)	1.92 (312)	0 (78)	0 (77)
Pf - Plasmodium falciparum						
Pv - Plasmodium vivax						
pan - <i>Plasmodium</i> species						
1 - For combination tests, Pan or Pv line, only, positive indicates a false positive non <i>P. falciparum</i> infection						

Table A4.9: False positive rate of *P. falciparum* (or pan*) test line results on all malaria-negative samples

Product	Manufacturer	Percentage of false positive Pf (or Pan*) test lines on "clean" negative samples			Percentage of false positive Pf (or Pan*) test lines on samples containing non- <i>Plasmodium</i> spp. infectious agents			Percentage of false positive Pf (or Pan*) test lines on samples containing immunological factors		
		Lot 1 (n=84)	Lot 2 (n=84)	Overall (n=168)	Lot 1 (n=42)	Lot 2 (n=42)	Overall (n=84)	Lot 1 (n=54)	Lot 2 (n=54)	Overall (n=108)
Pf only										
ADVANCED QUALITY™ MALARIA (p.f.) POCT	InTec Products, Inc.	20.24	11.9	16.07	11.9	19.05	15.48	35.19	7.41	21.3
ADVANCED QUALITY™ One Step Malaria (p.f.) Test (whole blood)	InTec Products, Inc.	33.33	22.62	27.98	42.86	21.95 (41)	32.53 (83)	53.7	29.63	41.67
Advantage P.f. Malaria Card	J. Mitra & Co. Pvt. Ltd.	0	0	0	0	0	0	3.7	1.85	2.78
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	2.38	2.38	2.38	2.38	0	1.19	1.85	0	0.93
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	1.19	4.76	2.98	0	4.76	2.38	0	0	0
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	1.2 (83)	3.61 (83)	2.41 (166)	0	14.29	7.14	1.85	0 (53)	0.93 (107)
Hexagon Malaria	Human GmbH	3.57	4.82 (83)	4.19 (167)	2.44 (41)	0 (39)	1.25 (80)	3.7	1.85	2.78
ICT Malaria Pf Cassette Test (ML01)	ICT Diagnostics	0	1.19	0.6	0	2.38	1.19	0	3.7	1.85
Immunoquick Malaria Falciparum	Biosynex	1.19	0	0.6	0	0	0	1.85	1.85	1.85
Malaria <i>Plasmodium</i> falciparum Rapid test Device (Whole blood)	ACON Laboratories, Inc.	0	0	0	0	0	0	5.56	1.85	3.7
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	1.19	0	0.6	0	0	0	0	0	0
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	0 (81)	2.53 (79)	1.25 (160)	0 (39)	5 (40)	2.53 (79)	1.89 (53)	1.89 (53)	1.89 (106)
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	7.14	7.23 (83)	7.19 (167)	7.14	9.52	8.33	1.85	15.09 (53)	8.41 (107)
Parahit-F DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	0	1.19	0.6	2.38	0	1.19	0	1.85	0.93
Parahit-F TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	0	0	0	0	0	0	0	0	0
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	2.38	2.38	2.38	4.76	7.14	5.95	0	0	0
Pf & Pan/Pf & Pv										
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	2.38	3.57	2.98	2.38	0	1.19	1.85	1.85	1.85
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	2.38	1.19	1.79	2.38	2.38	2.38	0	0	0
Binx Now Malaria	Inverness Medical Innovations, Inc.	0	0	0	0	7.14	3.57	0	0	0
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	1.19	0	0.6	2.38	0	1.19	0	0	0
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	3.57	3.57	3.57	4.76	0	2.38	0	3.7	1.85
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	7.14	2.41 (83)	4.79 (167)	7.14	4.76	5.95	5.56	7.41	6.48
Hexagon Malaria Combi	Human GmbH	2.38	2.41 (83)	2.4 (167)	0	0	0	0	0	0
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	1.19	0	0.6	2.38	2.38	2.38	7.41	3.7	5.56
Immunoquick Malaria +4	Biosynex	0	0	0	0	2.38	1.19	0	0	0
Malaria Pf/Pv/vax	Diagnostic Automation / Cortez Diagnostics, Inc.	0 (35)	0 (29)	0 (64)	9.09 (11)	0 (15)	3.85 (26)	11.54 (26)	5.26 (19)	8.89 (45)
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	4.76	5.95	5.36	9.76 (41)	7.14	8.43 (83)	5.56	5.56	5.56
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	0	3.57	1.79	0	4.76	2.38	0	1.85	0.93
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	5.95	4.76	5.36	4.76	0	2.38	11.11	9.26	10.19
One Step Malaria Antigen Strip	IND Diagnostics Inc.	1.19	2.41 (83)	1.8 (167)	0 (41)	2.38	1.2 (83)	3.7	1.85	2.78
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	0	0	0	0 (41)	0	0 (83)	0 (53)	0 (53)	0 (106)
OptiMAL-IT	DiaMed AG	0 (83)	0	0 (167)	0 (41)	0	0 (83)	3.85 (52)	1.85	2.83 (106)
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	0	0	0	2.38	0	1.19	0	1.85	0.93
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	0	2.38	1.19	0	2.38	1.19	1.85	3.77 (53)	2.8 (107)
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	1.19	2.41 (83)	1.8 (167)	0 (41)	2.38	1.2 (83)	3.7	1.85	2.78
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	2.38	1.19	1.79	0	0	0	3.7	3.7	3.7
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	0	1.19	0.6	2.56 (39)	4.76	3.7 (81)	0 (53)	0	0 (107)
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	2.41 (83)	4.76	3.59 (167)	7.32 (41)	11.9	9.64 (83)	3.85 (52)	7.41	5.66 (106)
Pan only										
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	1.19	2.38	1.79	0	0	0	0	0	0
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	4.76	8.33	6.55	2.38	11.9	7.14	3.7	1.85	2.78
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	3.61 (83)	2.38	2.99 (167)	2.38	0	1.19	7.55 (53)	3.7	5.61 (107)

Pf - *Plasmodium falciparum*
 pan - *Plasmodium* species
 1 - For Pan-only tests

Table A4.9a: False positive rate of *P. falciparum* (or pan[†]) test line results on all malaria-negative samples based on a delayed reading where specified by the manufacturer

Product	Manufacturer	Percentage of false positive PF or Pan test lines on "clean" negative samples			Percentage of false positive PF or Pan test lines on samples containing non- <i>Plasmodium</i> infectious agents			Percentage of false positive PF or Pan test lines on samples containing immunological factors		
		Lot 1 (n=84)	Lot 2 (n=84)	Overall (n=168)	Lot 1 (n=42)	Lot 2 (n=42)	Overall (n=84)	Lot 1 (n=54)	Lot 2 (n=54)	Overall (n=108)
PF & Pan/PF & Pv										
FirstSign – ParaView-2 (Pv + PF) Card Test	Unimed International, Inc.	5.95	0 (83)	2.99 (167)	0	0	0	1.85	0	0.93
Immunoquick Malaria +4	Biosynex	1.19	13.1	7.14	0	11.9	5.95	1.85	24.07	12.96
Malaria P.F/Vivax	Diagnostic Automation / Cortez Diagnostics, Inc.	6.45 (31)	0 (31)	3.23 (62)	0 (9)	0 (14)	0 (23)	10.71 (28)	5.26 (19)	8.51 (47)
Malacian Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	8.33	4.76	6.55	7.14	0	3.57	14.81	3.7	9.26
Parascreen Rapid Test for Malaria Pan/PF (Device)	Zephyr Biomedicals	0	2.38	1.19	0	0	0	0	1.85	0.93
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	3.66 (82)	5.95	4.82 (166)	9.76 (41)	4.76	7.23 (83)	4 (50)	9.26	6.73 (104)
Pan only										
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	1.22 (82)	1.19	1.2 (166)	2.38	0	1.19	7.41	0	3.7

PF - *Plasmodium falciparum*

Pv - *Plasmodium vivax*

pan - *Plasmodium* species

† - for Pan-only tests

Table A4.10 False positive rate of *P. falciparum* (or pan[†]) test line results for samples containing specific non-malarial infectious pathogens

Product	Manufacturer	Percentage of false positive Pf (or Pan) test lines by infectious pathogen									
		Dengue		Schistosomiasis		Leishmania		Chagas			
		Lot 1 (n=8)	Lot 2 (n=8)	Lot 1 (n=20)	Lot 2 (n=20)	Lot 1 (n=10)	Lot 2 (n=10)	Lot 1 (n=4)	Lot 2 (n=4)		
Pf only											
ADVANCED QUALITY TM MALARIA (p.f) POCT	InTec Products, Inc.	0	0	10	10	30	30	0	75		
ADVANCED QUALITY TM One Step Malaria (p.f) Test (whole blood)	InTec Products, Inc.	50	0	40	5.26 (19)	30	50	75	75		
Advantage P.f. Malaria Card	J. Mitra & Co. Pvt. Ltd.	0	0	0	0	0	0	0	0		
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	0	0	0	0	10	0	0	0		
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	0	0	0	0	0	10	0	25		
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	0	12.5	0	5	0	10	0	75		
Hexagon Malaria	Human GmbH	12.5	0	0 (19)	0 (19)	0	0 (9)	0	0		
ICT Malaria Pf Cassette Test (ML01)	ICT Diagnostics	0	0	0	0	0	10	0	0		
Immunoquick Malaria falciparum	Biosynex	0	0	0	0	0	0	0	0		
Malaria <i>Plasmodium falciparum</i> Rapid test Device (Whole blood)	ACON Laboratories, Inc.	0	0	0	0	0	0	0	0		
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	0	0	0	0	0	0	0	0		
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	0	0	0 (18)	0 (18)	0 (9)	10	0	25		
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	0	12.5	5	5	20	20	0	0		
Parahit-F DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	0	0	0	0	10	0	0	0		
Parahit-F TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	0	0	0	0	0	0	0	0		
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	0	0	0	0	20	30	0	0		
Pf & Pan/Pf & Pv											
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	0	0	0	0	10	0	0	0		
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	0	0	5	0	0	10	0	0		
Bimax Now Malaria	Inverness Medical Innovations, Inc.	0	0	0	0	0	30	0	0		
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	0	0	5	0	0	0	0	0		
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	12.5	0	5	0	0	0	0	0		
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	12.5	0	10	0	0	10	0	25		
Hexagon Malaria Kombi	Human GmbH	0	0	0	0	0	0	0	0		
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	12.5	12.5	0	0	0	0	0	0		
Immunoquick Malaria +4	Biosynex	0	0	0	5	0	0	0	0		
Malaria P.f/Vivax	Diagnostic Automation / Cortez Diagnostics, Inc.	0 (2)	0 (3)	25 (4)	0 (9)	0 (4)	0 (3)	0 (1)	N/A		
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	0	12.5	5.26 (19)	0	20	20	25	0		
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	0	0	0	0	0	10	0	25		
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	25	0	0	0	0	0	0	0		
One Step Malaria Antigen Strip	IND Diagnostic Inc.	0 (7)	0	0	0	0	10	0	0		
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	0	0	0 (19)	0	0	0	0	0		
OptiMAL-IT	DiaMed AG	0	0	0 (19)	0	0	0	0	0		
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	0	0	5	0	0	0	0	0		
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	0	0	0	0	0	10	0	0		
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	0 (7)	0	0	0	0	10	0	0		
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	0	0	0	0	0	0	0	0		
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	0	0	0 (19)	0	12.5	20	0	0		
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	0	0	5.26 (19)	10	20	20	0	25		
Pan only											
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	0	0	0	0	0	0	0	0		
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	0	0	0	5	10	30	0	25		
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	0	0	0	0	10	0	0	0		

Pf - *Plasmodium falciparum*
Pv - *Plasmodium vivax*
pan - *Plasmodium* species
† - for Pan-only tests

Table A4.10a: False positive rate of *P. falciparum* (or pan[†]) test line results for samples containing specific non-malarial infectious pathogens based on a delayed second reading when specified by the manufacturer

Product	Manufacturer	Percentage of false positive Pf (or Pan) test lines by infectious pathogen							
		Dengue		Schistosomiasis		Leishmania		Chagas	
		Lot 1 (n=8)	Lot 2 (n=8)	Lot 1 (n=20)	Lot 2 (n=20)	Lot 1 (n=10)	Lot 2 (n=10)	Lot 1 (n=4)	Lot 2 (n=4)
Pf & Pan/Pf & Pv									
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	0	0	0	0	0	0	0	0
Immunoquick Malaria +4	Biosynex	0	12.5	0	15	0	10	0	0
Malaria Pf/Wvax	Diagnostic Automation / Cortez Diagnostics, Inc.	0 (2)	0 (3)	0 (2)	0	0 (4)	0 (3)	0 (1)	N/A
Malacian Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	37.5	0	0	0	0	0	0	0
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	0	0	0	0	0	0	0	0
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	0	0	15.79 (19)	0	10	10	0	25
Pan only									
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	0	0	0	0	10	0	0	0

Pf - *Plasmodium falciparum*

Pv - *Plasmodium vivax*

pan - *Plasmodium* species

[†] - for Pan-only tests

Table A4.11: False positive rate of *P. falciparum* (or pan[†]) test line results for samples containing potentially cross-reacting blood immunological factors

Product	Manufacturer	Percentage of false positive PF (or Pan) test lines by blood immunological factor																	
		Rheumatoid factor		Anti-nuclear antibodies		Anti-mouse antibodies		Rapid plasma reagin (RPR) positive											
		Lot 1 (n=8)	Lot 2 (n=8)	Lot 1 (n=26)	Lot 2 (n=26)	Lot 1 (n=2)	Lot 2 (n=2)	Lot 1 (n=18)	Lot 2 (n=18)										
PF only																			
ADVANCED QUALITY TM MALARIA (p.f) POCT	InTec Products, Inc.	25	25	30.77	0	0	0	0	0	0	0	50	11.11						
ADVANCED QUALITY TM One Step Malaria (p.f.) Test (whole blood)	InTec Products, Inc.	62.5	62.5	46.15	19.23	50	50	50	50	61.11	27.78								
Advantage Pf. Malaria Card	J. Mitra & Co. Pvt. Ltd.	0	0	0	0	100	0	0	0	0	0	0							
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	12.5	0	0	0	0	0	0	0	0	0	0							
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	0	0	0	0	0	0	0	0	0	0	0							
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	0	0	3.85	0 (25)	0	0	0	0	0	0	0							
Hexagon Malaria	Human GmbH	25	12.5	0	0	0	0	0	0	0	0	0							
ICT Malaria Pf Cassette Test (ML01)	ICT Diagnostics	0	0	0	0	0	0	50	0	0	0	5.56							
Immunoquick Malaria Falciparum	Biosynex	12.5	0	0	3.85	0	0	0	0	0	0	0							
Malaria <i>Plasmodium falciparum</i> Rapid test Device (Whole blood)	ACON Laboratories, Inc.	12.5	0	0	0	100	50	0	0	0	0	0							
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	0	0	0	0	0	0	0	0	0	0	0							
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	12.5	12.5	0	0 (25)	0	0	0	0	0 (17)	0	0							
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	0	12.5	3.85	8 (25)	0	50	0	50	0	22.22	0							
ParahiT-DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	0	0	0	0	0	0	0	50	0	0	0							
ParahiT-TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	0	0	0	0	0	0	0	0	0	0	0							
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	0	0	0	0	0	0	0	0	0	0	0							
Pf & Pan/Pf & Pv																			
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	12.5	0	0	3.85	0	0	0	0	0	0	0	0						
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	0	0	0	0	0	0	0	0	0	0	0	0						
Binax Now Malaria	Inverness Medical Innovations, Inc.	0	0	0	0	0	0	0	0	0	0	0	0						
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	0	0	0	0	0	0	0	0	0	0	0	0						
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	0	0	0	3.85	0	0	0	0	0	0	5.56							
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	0	0	7.69	15.38	0	0	0	0	5.56	0	0							
Hexagon Malaria Combi	Human GmbH	0	0	0	0	0	0	0	0	0	0	0							
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	12.5	0	7.69	3.85	0	50	5.56	0	0	0	0							
Immunoquick Malaria +4	Biosynex	0	0	0	0	0	0	0	0	0	0	0							
Malaria Pf/Mvax	Diagnostic Automation / Cortez Diagnostics, Inc.	0 (7)	0 (4)	25 (12)	0 (9)	0	N/A	0 (5)	16.67 (6)										
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	0	0	3.85	11.54	100	0	0	0	0	0	0							
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	0	0	0	3.85	0	0	0	0	0	0	0							
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	0	0	23.08	15.38	0	0	0	0	0	5.56	0							
One Step Malaria Antigen Strip	IND Diagnostic Inc.	25	0	0	0	0	0	50	0	0	0	0							
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	0	0	0 (25)	0 (25)	0	0	0	0	0	0	0							
OptiMAL-IT	DiaMed AG	0	12.5	0 (24)	0 (24)	100	0	0	0	0	0	0							
ParahiT-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	0	0	0	0	0	0	0	0	0	0	5.56							
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	0	12.5	0	3.85	0	0	5.56	0 (17)	0	0	0							
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	25	0	0	0	0	0	50	0	0	0	0							
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	25	25	0	0	0	0	0	0	0	0	0							
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	0	0	0 (25)	0	0	0	0	0	0	0	0							
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	0	0	0 (25)	11.54	50	0	5.88 (17)	5.56										
Pan only																			
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	0	0	0	0	0	0	0	0	0	0	0	0						
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	12.5	0	0	0	0	0	5.56	0	5.56	0	0							
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	12.5	12.5	7.69	0	0 (1)	0	5.56	5.56										
Pf - <i>Plasmodium falciparum</i>																			
Pv - <i>Plasmodium vivax</i>																			
pan - <i>Plasmodium</i> species																			
t - For Pan-only tests																			

Table A4.11a: False positive rate of *P. falciparum* (or pan^t) test line results for samples containing potentially cross-reacting blood immunological factors based on a delayed reading when specified by the manufacturer

Product	Manufacturer	Percentage of false positive PF (or Pan) test lines by blood immunological factor							
		Rheumatoid factor		Anti-nuclear antibodies		Anti-mouse antibodies		Rapid plasma reagin (RPR) positive	
		Lot 1 (n=8)	Lot 2 (n=8)	Lot 1 (n=26)	Lot 2 (n=26)	Lot 1 (n=2)	Lot 2 (n=2)	Lot 1 (n=18)	Lot 2 (n=18)
PF & Pan/PF & Pv									
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	0	0	3.85	0	0	0	0	0
Immunoquick Malaria +4	Biosynex	0	0	0	19.23	50	50	0	38.89
Malaria P-F/Vivax	Diagnostic Automation / Cortez Diagnostics, Inc.	0 (7)	0 (4)	23.08 (13)	0 (9)	0	N/A	0 (6)	16.67 (6)
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	0	0	30.77	7.69	0	0	0	0
Parascreen Rapid Test for Malaria Pan/PF (Device)	Zephyr Biomedicals	0	0	0	3.85	0	0	0	0
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	0	0	4.35 (23)	11.54	0	0	5.88 (17)	11.11
Pan only									
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	0	0	11.54	0	50	0	0	0

PF - *Plasmodium falciparum*

pv - *Plasmodium vivax*

pan - *Plasmodium* species

t - For Pan-only tests

Table A4.12: False positive rate of pan/*P. vivax* test line results on all malaria-negative samples

Product	Manufacturer	Percentage of false positive pan test lines on "clean" negative samples			Percentage of false positive pan test lines on samples containing non- <i>Plasmodium</i> infectious agents			Percentage of false positive pan test lines on samples containing immunological factors		
		Lot 1 (n=84)	Lot 2 (n=84)	Overall (n=168)	Lot 1 (n=42)	Lot 2 (n=42)	Overall (n=84)	Lot 1 (n=54)	Lot 2 (n=54)	Overall (n=108)
Pf & Pan/Pf & Pv										
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	3.57	3.57	3.57	2.38 (42)	4.76 (42)	3.57	1.85	3.7	2.78
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	2.38	14.29	8.33	4.76 (42)	11.9 (42)	8.33	3.7	12.96	8.33
Binax Now Malaria	Inverness Medical Innovations, Inc.	0	0	0	0 (42)	0 (42)	0	0	0	0
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	2.38	2.38	2.38	2.38 (42)	2.38 (42)	2.38	1.85	0	0.93
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	0	2.38	1.19	0 (42)	0 (42)	0	0	0	0
FirstSign - ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	5.95 (84)	3.61 (83)	4.79 (167)	7.14 (42)	0 (42)	3.57 (84)	3.7 (54)	7.41 (54)	5.56 (108)
Hexagon Malaria Combi	Human GmbH	0	1.2 (83)	0.6 (167)	0 (42)	0 (42)	0	0	3.7	1.85
ICT Malaria Combo Cassette Test (MI02)	ICT Diagnostics	0	0	0	0 (42)	0 (42)	0	5.56	5.56	5.56
Immunoquick Malaria +4	Biosynex	0	1.19	0.6	0 (42)	0 (42)	0	5.56	3.7	4.63
Malaria P/F/vivax	Diagnostic Automation / Cortez Diagnostics, Inc.	0 (35)	3.45 (29)	1.56 (64)	0 (11)	0 (15)	0 (26)	7.69 (26)	5.26 (19)	6.67 (45)
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	5.95	7.14	6.55	9.76 (41)	7.14 (42)	8.43 (83)	7.41	7.41	7.41
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	0	20.24	10.12	0 (42)	11.9 (42)	5.95	5.56	20.37	12.96
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	0	0	0	2.38 (42)	0 (42)	1.19	3.7	7.41	5.56
One Step Malaria Antigen Strip	IND Diagnostic Inc.	0	2.41 (83)	1.2 (167)	0 (41)	0 (42)	0 (83)	1.85	1.85	1.85
OnSight - ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	0	0	0	0 (41)	0 (42)	0 (83)	0 (53)	1.89 (53)	0.94 (106)
OptIMAL-IT	DiaMed AG	0 (83)	0	0 (167)	0 (41)	0 (42)	0 (83)	3.85 (52)	3.7	3.77 (106)
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	0	0	0	0 (42)	0 (42)	0	3.7	1.85	2.78
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	0	0	0	0 (42)	0 (42)	0	0	0 (53)	0 (107)
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	0	2.41 (83)	1.2 (167)	0 (41)	0 (42)	0 (83)	1.85	1.85	1.85
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	1.19	0	0.6	0 (42)	0 (42)	0	0	0	0
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	0	1.19	0.6	0 (39)	0 (42)	0 (81)	0 (53)	0	0 (107)
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co, Ltd	2.41 (83)	5.95	4.19 (167)	0 (41)	2.38 (42)	1.2 (83)	5.77 (52)	7.41	6.6 (106)
Pan only										
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	1.19	2.38	1.79	0 (42)	0 (42)	0	0	0	0
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	4.76	8.33	6.55	2.38 (42)	11.9 (42)	7.14	3.7	1.85	2.78
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	3.61 (83)	2.38	2.99 (167)	2.38 (42)	0 (42)	1.19	7.55 (53)	3.7	5.61 (107)
Pf - <i>Plasmodium falciparum</i> Pv - <i>Plasmodium vivax</i> pan - <i>Plasmodium</i> species										

Table A4.12a: False positive rate of pan test line results on all malaria-negative samples based on a delayed second reading when specified by the manufacturer

Product	Manufacturer	Percentage of false positive pan test lines on "clean" negative samples			Percentage of false positive pan test lines on samples containing non- <i>Plasmodium</i> infectious agents			Percentage of false positive pan test lines on samples containing immunological factors		
		Lot 1 (n=84)	Lot 2 (n=84)	Overall (n=168)	Lot 1 (n=42)	Lot 2 (n=42)	Overall (n=84)	Lot 1 (n=54)	Lot 2 (n=54)	Overall (n=108)
Pf & Pan/Pf & Pv										
FirstSign - ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.									
Immunoquick Malaria +4	Biosynex	1.19	11.9	6.55	0	16.67	8.33	9.26	25.93	17.59
Malaria P-F/Vivax	Diagnostic Automation / Cortez Diagnostics, Inc.	0 (31)	0 (31)	0 (62)	0 (9)	0 (14)	0 (23)	7.14 (28)	10.53 (19)	8.51 (47)
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	1.19	0	0.6	0	0	0	3.7	7.41	5.56
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	1.19	1.19	1.19	0	0	0	0	0	0
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	3.66 (82)	5.95	4.82 (166)	0 (41)	9.52	4.82 (83)	10 (50)	9.26	9.62 (104)
Pan only										
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	1.22 (82)	1.19	1.2 (166)	2.38	0	1.19	7.41	0	3.7

Pf - *Plasmodium falciparum*
Pv - *Plasmodium vivax*
pan - *Plasmodium* species

Table A4.13: Heat stability testing results for Pf test line on *P. falciparum* samples at low parasite density (200 parasites/ μ l). Positivity rate at baseline, and after 60 days incubation at 35°C and 45°C.

Product	Manufacturer	Baseline testing						35°C			45°C								
		Lot 1 (n=10)			Lot 2 (n=10)			Lot 1 (n=10)			Lot 2 (n=10)								
		No. positive	Mean band intensity	No. invalid	No. positive	Mean band intensity	No. invalid	No. positive	Mean band intensity	No. invalid	No. positive	Mean band intensity	No. invalid						
Pf only tests																			
ADVANCED QUALITY TM MALARIA (p.f.) POCT (whole blood)	InTec Products, Inc.	10	1.1	6	1	9	0	1.22	10	0	2	8	0	1.13	10	0	1.4		
ADVANCED QUALITY TM One Step Malaria (p.f.) Test (whole blood)	InTec Products, Inc.	10	0	6	0	1	7	0	1	10	0	7	0	1.14	2	0	1		
Advantage P.f. Malaria Card	J. Mitra & Co. Pvt. Ltd.	10	0	9	0	2.78	10	0	2.1	10	0	2.8	10	0	2.8	10	0	2.4	
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	10	0	3.7	10	0	2	10	0	3.6	10	0	1.7	10	0	3	10	0	2.6
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	10	0	3	10	0	3	10	0	2	10	0	3.1	10	0	2.5	10	0	2.8
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	4	0	1.25	0	N/A	1	0	1	2	0	0	1	0	N/A	0	2	N/A	
Hexagon Malaria	Human GmbH	6	0	1.17	4	0	1	0	N/A	7	0	1	7	0	1	5	0	1.2	
ICT Malaria Pf Rapid Test (ML01)	ICT Diagnostics	10	0	1.8	10	0	1.8	10	0	2.3	10	0	1.8	10	0	2	9	0	1.78
Immunquick Malaria Falciparum	Biosynex	10	0	2.1	10	0	2	10	0	2	10	0	2.3	10	0	2	10	0	2.6
Malaria Plusmodium falciparum Rapid test Device (Whole blood)	ACON Laboratories, Inc.	10	0	2	10	0	2.3	10	0	2.1	10	0	2.4	10	0	2	10	0	2.3
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	10	0	1.6	10	0	1.9	10	0	1.2	10	0	1.8	7	0	1	10	0	1.4
Paracheck Pf Rapid Test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	10	0	2	8	0	1.63	9	0	1.78	5	2	1.4	4	0	1	6	0	1.33
Paracheck Pf Rapid Test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	10	0	1.4	9	0	1.11	10	0	1.8	10	0	1.4	8	0	1.5	9	1	1.33
Parahi-F DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	10	0	1.2	10	0	2	10	0	1	10	0	2.1	10	0	1.8	10	0	2
Parahi-F TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	9	0	1	5	0	1	6	0	1	4	0	1	6	0	1	2	0	1
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	10	0	3	10	0	2.6	10	0	3	10	0	3	10	0	2.2	10	0	2.9
Pf line of combination tests																			
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	10	0	1.5	10	0	1.8	10	0	1.7	10	0	2	1	0	1	10	0	1.4
AZOG Malaria pf (HRP-II)/pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	8	0	1	4	0	1	7	0	1	6	0	1	6	0	1	1	0	1
Binx Now Malaria	Inverness Medical Innovations, Inc.	10	0	1.8	10	0	2	10	0	2.3	10	0	2	10	0	2.1	10	0	2.5
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	10	0	2.8	10	0	3	9	1	3	10	0	1	10	0	2.5	10	0	2.2
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	10	0	3	10	0	3	10	0	3	10	0	3	10	0	2.5	10	0	3
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	10	0	1.5	9	0	1	5	0	1	9	0	1.33	0	1	N/A	0	2	N/A
Hexagon Malaria Combi	Human GmbH	8	0	1	5	0	1	4	1	1	7	0	1	5	0	1	5	0	1
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	10	0	2.2	10	0	2.5	10	0	2.4	10	0	2.1	10	0	2	10	0	2.6
Immunquick Malaria +4	Biosynex	10	0	2.7	10	0	2.9	10	0	3	10	0	2	10	0	2.9	10	0	3.1
Malaria P/F/V/VAX	Diagnostic Automation / Cortez Diagnostics, Inc.	8	2	2.5	5	5	2.4	1	9	4	2	8	3.5	3	7	3	1	9	3
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	10	0	2.4	10	0	2	10	0	2.2	10	0	2.1	10	0	2.2	10	0	2.2
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	10	0	1.9	10	0	1.7	10	0	2.1	10	0	2.3	10	0	1.8	10	0	1.9
Malasscan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	10	0	1	9	0	1.44	9	0	1.67	9	0	1.33	10	0	1	7	0	1.71
One Step Malaria Antigen Strip	IND Diagnostic Inc.	3	0	1	0	0	N/A	0	0	N/A	0	4	N/A	0	0	N/A	0	2	N/A
OnSight – ParaQuick (Pan, Pf)	Amgenix International, Inc.	10	0	1.3	10	0	2.3	10	0	2	8	0	1.75	8	0	1.75	4	0	1
OptiMAL- IT	DiaMed AG	6	0	1	0	0	N/A	1	0	1	1	0	1	0	0	N/A	0	9	N/A
ParahiTotal Device Rapid Test for <i>P. falciparum</i> and Pan Malarial Species	Span Diagnostics Ltd.	8	0	1.13	5	0	1	8	0	1	7	0	1	3	0	1	2	0	2
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	10	0	1.6	9	0	2.22	8	0	1.75	8	0	1.88	7	0	1	2	0	1
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	3	0	1	0	0	N/A	0	0	N/A	0	4	N/A	0	0	N/A	0	2	N/A
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	2	0	1	5	0	1	8	0	1	4	0	1	9	0	1	6	0	1
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	10	0	2.8	10	0	2.3	10	0	1.7	10	0	2	10	0	2	10	0	2.9
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	10	0	1	10	0	1.1	9	1	1.89	10	0	2.1	10	0	2	10	0	1.8

Table A4.13a: Heat stability testing results for pan test line of combination RDTs on *P. falciparum* samples at low parasite density (200 parasites/ μ l). Positivity rate at baseline, and after 60 days incubation at 35°C and 45°C.

Product	Manufacturer	Baseline testing						35°C						45°C					
		Lot 1 (n=10)			Lot 2 (n=10)			Lot 1 (n=10)			Lot 2 (n=10)			Lot 1 (n=10)			Lot 2 (n=10)		
		No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity
PF & Pan/Pv & Pan																			
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	8	0	1	3	0	1	5	0	1	4	0	1	8	0	1	0	0	N/A
AZOG Malaria pf (HRP-III)/pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	0	0	N/A	8	0	1	0	0	N/A	7	0	1	0	0	N/A	5	0	2
Binox Now Malaria	Inverness Medical Innovations, Inc.	0	0	N/A	1	0	1	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	10	0	1	10	0	1	9	1	1	10	0	1.8	10	0	1.4	10	0	1
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	9	0	1	10	0	1	6	0	1	8	0	1	10	0	1	10	0	1
FirstSign - ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A	0	1	N/A	0	2	N/A
Hexagon Malaria Combi	Human GmbH	0	0	N/A	0	0	N/A	0	1	N/A	0	0	N/A	0	0	N/A	0	0	N/A
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	0	0	N/A	1	0	1	1	0	2	0	0	N/A	0	0	N/A	0	0	N/A
Immunoquick Malaria +4	Biosynex	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A
Malaria P.FM/VAX	Diagnostic Automation / Cortez Diagnostics, Inc.	0	2	N/A	0	5	N/A	0	9	N/A	0	8	N/A	0	7	N/A	0	9	N/A
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	3	0	1	0	0	N/A	2	0	1	4	0	1	0	0	N/A	0	0	N/A
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	0	0	N/A	0	0	N/A	0	0	N/A	3	0	1.333	0	0	N/A	0	0	N/A
Malascan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	0	0	N/A	0	0	N/A	0	0	N/A	1	0	1	0	0	N/A	0	0	N/A
One Step Malaria Antigen Strip	IND Diagnostic Inc.	3	0	1	0	0	N/A	0	0	N/A	0	4	N/A	0	0	N/A	0	2	N/A
OnSight - ParaQuick (Pan, Pf)	Amgenix International, Inc.	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A
OptiMAL - IT	DiaMed AG	6	0	1	0	0	N/A	1	0	1	2	0	1	0	0	N/A	0	9	N/A
ParahiTotal Device Rapid Test for <i>P. falciparum</i> and Pan Malarial Species	Span Diagnostics Ltd.	0	0	N/A	1	0	1	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	1	0	1	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	3	0	1	0	0	N/A	0	0	N/A	0	4	N/A	0	0	N/A	0	2	N/A
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	0	0	N/A	0	0	N/A	4	0	1	5	0	1	9	0	1	6	0	1
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	0	0	N/A	0	0	N/A	0	0	N/A	1	0	1	9	0	1	7	0	1.43
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	9	0	1	5	0	1	8	1	1	10	0	1	10	0	1	4	0	1.25
Pan only																			
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	8	0	1	2	0	1	6	0	1	7	0	1	9	0	1	5	0	1
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	10	0	1.1	10	0	1.1	10	0	1	10	0	1	8	0	1	10	0	1
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	0	0	N/A	1	0	1	0	1	N/A	0	0	N/A	0	1	N/A	0	0	N/A
Pf - <i>Plasmodium falciparum</i>																			
Pv - <i>Plasmodium vivax</i>																			
pan - <i>Plasmodium</i> species																			

Table A4.14: Heat stability testing results for Pf test line on *P. falciparum* samples at high parasite density (2000 parasites/ μ l) Positivity rate at baseline, and after 60 days incubation at 35°C and 45°C.

Product	Manufacturer	Baseline testing						35°C						45°C					
		Lot 1 (n=10)			Lot 2 (n=10)			Lot 1 (n=10)			Lot 2 (n=10)			Lot 1 (n=10)			Lot 2 (n=10)		
		No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity
Pf only tests																			
ADVANCED QUALITY TM MALARIA (p.f.) POCT	InTec Products, Inc.	10	0	3.6	10	0	2.9	10	0	3.1	10	0	3.7	10	0	2.9	10	0	2.8
ADVANCED QUALITY TM One Step Malaria (p.f.) Test (whole blood)	InTec Products, Inc.	10	0	2.5	10	0	3.3	9	0	2.67	10	0	2.6	10	0	3.1	10	0	2.4
Advantage P.f. Malaria Card	J. Mitra & Co. Pvt. Ltd.	10	0	4	10	0	3.9	10	0	4	10	0	4	10	0	4	10	0	3.9
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	10	0	4	10	0	4	10	0	4	10	0	4	10	0	4	10	0	3.8
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	10	0	4	10	0	4	10	0	3.9	10	0	4	10	0	3.9	10	0	4
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	10	0	2.8	10	0	2.8	9	0	1.11	9	0	2.67	10	0	1.4	9	0	2.67
Hexagon Malaria	Human GmbH	9	0	2.11	10	0	2.6	10	0	2.3	10	0	2.5	10	0	2.3	10	0	2.9
ICT Malaria Pf Rapid Test (MLO1)	ICT Diagnostics	10	0	3.8	10	0	4	10	0	3.7	10	0	4	10	0	3.2	10	0	3.5
Immunoquick Malaria Falciparum	Biosynex	10	0	4	10	0	4	10	0	3.9	10	0	3.9	10	0	4	10	0	4
Malaria Plasmodium falciparum Rapid test Device (Whole blood)	ACON Laboratories, Inc.	10	0	3.2	10	0	4	10	0	4	10	0	3.9	10	0	3	10	0	3.9
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	10	0	3.1	10	0	3.8	10	0	2.9	10	0	3.2	10	0	2.4	10	0	3.1
Paracheck Pf Rapid Test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	10	0	1.9	10	0	3.7	7	3	3.71	10	0	3.5	10	0	3.1	10	0	3.8
Paracheck Pf Rapid Test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	10	0	3	10	0	3	10	0	3	10	0	3.3	10	0	2.4	10	0	3.4
Parahi-f-DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	10	0	3.9	10	0	3.9	10	0	3	10	0	4	10	0	3.3	10	0	4
Parahi-F-TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	10	0	2.7	10	0	2.7	9	1	2.44	10	0	2.4	9	0	2.67	10	0	2.9
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	10	0	4	10	0	4	10	0	4	10	0	4	10	0	4	10	0	4
Pf line of combination tests																			
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	9	1	3.33	10	0	3.8	10	0	3.9	10	0	3.8	9	0	2.44	10	0	3.2
AZOG Malaria pf (HRP-III)/pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	10	0	2	10	0	2.7	10	0	2.2	10	0	2.7	10	0	2.9	10	0	2.4
Binx Now Malaria	Inverness Medical Innovations, Inc.	10	0	3.6	10	0	3.8	10	0	4	10	0	3.8	10	0	3.9	9	1	4
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	10	0	4	10	0	4	10	0	4	10	0	4	10	0	3.9	10	0	3.9
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	10	0	4	10	0	4	10	0	4	10	0	4	10	0	4	10	0	4
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	10	0	4	10	0	3.4	9	1	2.44	10	0	3.9	10	0	2.3	5	1	2.6
Hexagon Malaria Combi	Human GmbH	10	0	2.7	10	0	2.8	10	0	3.1	7	2	2.57	10	0	2.1	9	0	3
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	10	0	4	10	0	4	10	0	4	10	0	4	10	0	3.8	10	0	4
Immunoquick Malaria +4	Biosynex	10	0	4	10	0	4	10	0	4	10	0	4	10	0	3.9	10	0	3.7
Malaria P-FIVVAX	Diagnostic Automation / Cortez Diagnostics, Inc.	6	4	4	7	3	4	3	6	3.67	6	4	3.67	1	9	1	0	9	N/A
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	10	0	4	10	0	4	10	0	4	10	0	3.9	10	0	3.9	10	0	4
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	10	0	4	10	0	3.8	10	0	3.3	10	0	3.2	10	0	3.7	10	0	3.4
Malasscan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	10	0	2.8	10	0	3	10	0	3.3	10	0	3.3	10	0	3	10	0	3
One Step Malaria Antigen Strip	IND Diagnostics Inc.	10	0	1	3	0	1	10	0	1	0	0	N/A	0	0	N/A	0	5	N/A
OnSight – ParaQuick (Pan, Pf)	Amgenix International, Inc.	10	0	3.8	10	0	3.9	10	0	3.8	10	0	4	10	0	3.8	10	0	4
OptiMAL- IT	DiaMed AG	10	0	2	10	0	1.6	10	0	1.9	9	0	1.33	0	0	N/A	0	8	N/A
Parahitotal Device Rapid Test for <i>P. falciparum</i> and Pan Malaria Species	Span Diagnostics Ltd.	10	0	3.4	9	0	2.56	10	0	3.3	10	0	2.5	10	0	1.8	10	0	2.7
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	10	0	3.9	10	0	4	10	0	3.6	10	0	3.7	10	0	3.4	9	1	3.33
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	10	0	1	3	0	1	10	0	1	0	0	N/A	0	0	N/A	0	5	N/A
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	10	0	2	10	0	1.9	10	0	2	10	0	1.9	10	0	1.9	10	0	2
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	10	0	4	10	0	4	10	0	3.9	10	0	4	9	1	4	10	0	4
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	10	0	4	9	1	2.89	10	0	3.8	10	0	3.9	10	0	2.7	10	0	4
Pf - Plasmodium falciparum																			
Pv - Plasmodium vivax																			
pan - Plasmodium species																			

Table A4.14a: Heat stability testing results for pan test line of combination RDTs on *P. falciparum* samples at high parasite density (2000 parasites/ μ l). Positivity rate at baseline, and after 60 days incubation at 35°C and 45°C.

Product	Manufacturer	Baseline testing						35°C						45°C					
		Lot 1 (n=10)			Lot 2 (n=10)			Lot 1 (n=10)			Lot 2 (n=10)			Lot 1 (n=10)			Lot 2 (n=10)		
		No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity	No. positive	No. invalid	Mean band intensity
Pan line of combination tests																			
Advantage Mai Card	J. Mitra & Co. Pvt. Ltd.	10	0	1.6	10	0	1.8	10	0	2	10	0	2	10	0	1.9	10	0	2
AZOG Malaria pf (HRP-II)/pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	8	1	1	10	0	1	4	0	1	10	0	1	0	0	N/A	4	0	2.5
Binx Now Malaria	Inverness Medical Innovations, Inc.	10	0	1	9	0	1.56	9	0	1.44	10	0	1.8	6	0	1	9	1	1.56
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	10	0	2.9	10	0	2.4	10	0	2.6	10	0	2	10	0	2.3	10	0	2.5
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	10	0	2.9	10	0	2.8	10	0	1.8	10	0	2.4	10	0	2.2	10	0	2.4
FirstSign - ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	0	0	N/A	0	0	N/A	0	1	N/A	0	0	N/A	0	0	N/A	0	1	N/A
Hexagon Malaria Combi	Human GmbH	0	0	N/A	0	0	N/A	0	0	N/A	0	2	N/A	0	0	N/A	0	0	N/A
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	9	0	1	9	0	1.22	10	0	1.9	5	0	1	10	0	1.1	5	0	2
Immunoquick Malaria +4	Biosynex	10	0	1	10	0	1	9	0	1.11	7	0	1	6	0	1	10	0	1.7
Malaria P:PF/IVAX	Diagnostic Automation / Cortez Diagnostics, Inc.	0	4	N/A	0	3	N/A	0	6	N/A	0	4	N/A	0	9	N/A	0	9	N/A
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	10	0	1.4	9	0	1	10	0	1.4	10	0	1.6	6	0	1.33	9	0	1.33
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	9	0	1	3	0	1	2	0	1	3	0	1.33	0	0	N/A	3	0	3.33
Malaccan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	5	0	1	8	0	1	6	0	1	5	0	1	1	0	1	2	0	5
One Step Malaria Antigen Strip	IND Diagnostic Inc.	10	0	1	3	0	1	10	0	1	0	0	N/A	0	0	N/A	1	5	5
OnSight - ParaQuick (Pan, Pf)	Amgenix International, Inc.	10	0	1.5	10	0	1.6	10	0	1.8	10	0	1.6	9	0	1.11	10	0	1.2
OptiMAL- IT	DiaMed AG	10	0	2	10	0	1.6	10	0	1.6	10	0	1.2	1	0	1	1	8	2
ParahitTotal Device Rapid Test for <i>P. falciparum</i> and Pan Malarial Species	Span Diagnostics Ltd.	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A	0	0	N/A
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	8	0	1	9	0	1.67	6	0	1.33	8	0	1.5	8	0	1.25	8	1	1.5
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	10	0	1	3	0	1	10	0	1	0	0	N/A	0	0	N/A	1	5	5
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	7	0	1	4	0	1	10	0	1	10	0	1	10	0	1	9	0	1.2
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	10	0	1	8	0	1	2	0	1	7	0	1	9	1	1	9	0	1.33
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	10	0	3	9	1	2	10	0	3.6	10	0	3.7	10	0	1.8	10	0	3
Pan only																			
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	10	0	2.4	10	0	2.6	10	0	2.1	10	0	2.7	10	0	2.8	10	0	2.8
CareStart Malaria pLDH (PAN)	Access Bio, Inc.	10	0	2.8	10	0	3.4	10	0	2.4	10	0	2.5	10	0	2	10	0	2.9
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	7	0	1	10	0	1.8	10	0	1.4	8	0	1.25	8	1	1.62	6	0	1.5
Pf - <i>Plasmodium falciparum</i> Pv - <i>Plasmodium vivax</i> pan - <i>Plasmodium</i> species																			

Table A4.15: Heat stability testing results for Pf (or pan*) test line on parasite negative samples. Positivity rate at baseline, and after 60 days incubation at 35°C and 45°C.

Product	Manufacturer	Baseline testing						35°C				45°C					
		Lot 1 (n=4)		Lot 2 (n=4)		Lot 1 (n=4)		Lot 2 (n=4)		Lot 1 (n=4)		Lot 2 (n=4)		Lot 1 (n=4)		Lot 2 (n=4)	
		No. positive	No. invalid	No. positive	No. invalid	No. positive	No. invalid	No. positive	No. invalid	No. positive	No. invalid	No. positive	No. invalid	No. positive	No. invalid	No. positive	No. invalid
Pf only																	
ADVANCED QUALITY™ MALARIA (p.f.) POCT	InTec Products, Inc.	2	0	0	0	2	0	4	0	2	0	0	0	0	0	3	0
ADVANCED QUALITY™ One Step Malaria (p.f.) Test (whole blood)	InTec Products, Inc.	2	0	0	0	1	0	1	0	0	0	0	0	0	0	0	0
Advantage P.f. Malaria Card	J. Mitra & Co. Pvt. Ltd.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
CareStart Malaria HRP2 (Pf)	Access Bio, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
First Response Malaria Ag HRP2	Premier Medical Corporation Ltd.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
FirstSign – Malaria Pf Card Test	Unimed International, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Hexagon Malaria	Human GmbH	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
ICT Malaria Pf Cassette Test (ML01)	ICT Diagnostics	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Immunoquick Malaria Falciparum	Biosynex	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Malaria Plasmodium falciparum Rapid test Device (Whole blood)	ACON Laboratories, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Malaria Rapid Pf	Vision Biotech (Pty) Ltd.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Device)	Orchid Biomedical Systems	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Paracheck Pf Rapid test for <i>P. falciparum</i> Malaria (Dipstick)	Orchid Biomedical Systems	2	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
Parahit-F DIPSTICK FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Parahit-F TEST DEVICE FOR FALCIPARUM MALARIA	Span Diagnostics Ltd.	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
SD BIOLINE Malaria Ag Pf	Standard Diagnostics, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Pf & Pan/Pf & Pv																	
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
AZOG Malaria pf (HRP-II) /pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Binox Now Malaria	Inverness Medical Innovations, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
FirstSign – ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2
Hexagon Malaria Combi	Human GmbH	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
ICT Malaria Combo Cassette Test (ML02)	ICT Diagnostics	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Immunoquick Malaria +4	Biosynex	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Malaria Pf/Vivax	Diagnostic Automation / Cortez Diagnostics, Inc.	0	0	0	0	0	0	4	0	0	0	3	0	0	0	0	2
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Malasscan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
One Step Malaria Antigen Strip	IND Diagnostic Inc.	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	1
OnSight – ParaQuick (Pan, Pf) Test	Amgenix International, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
OptiMAL-IT	DiaMed AG	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4
Parahit-Total Device Rapid test for <i>P. falciparum</i> and Pan malarial species.	Span Diagnostics Ltd.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	1
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	0	0	0	0	0	0	0	0	2	0	1	0	0	0	1	0
Pan only																	
Advantage Pan Malaria Card	J. Mitra & Co. Pvt. Ltd.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Parabank Rapid Test for Malaria pLDH (PAN)	Access Bio, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0
Parabank Rapid Test for Malaria Pan (Device)	Zephyr Biomedicals	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Pf - Plasmodium falciparum																	
Pv - Plasmodium vivax																	
pan - Plasmodium species																	
* - For pan-only tests																	

Table A4.15a: Heat stability testing results for pan test line of combination RDTs on negative samples. Positivity rate at baseline, and after 60 days incubation at 35°C and 45°C.

Product	Manufacturer	Baseline testing						35°C				45°C					
		Lot 1 (n=4) No. positive	No. invalid	Lot 2 (n=4) No. positive	No. invalid	Lot 1 (n=4) No. positive	No. invalid	Lot 2 (n=4) No. positive	No. invalid	Lot 1 (n=4) No. positive	No. invalid	Lot 2 (n=4) No. positive	No. invalid				
Pan line of combination tests																	
Advantage Mal Card	J. Mitra & Co. Pvt. Ltd.	1	0	0	0	0	0	0	0	0	0	0	0	0			
AZOG Malaria pf (HRP-II)/pv (pLDH) Antigen Detection Test Device	AZOG, Inc.	0	0	4	0	0	0	0	0	0	0	0	0	2			
Binax Now Malaria	Inverness Medical Innovations, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0			
CareStart Malaria HRP2/pLDH (Pf/PAN) COMBO	Access Bio, Inc.	0	0	0	0	0	0	0	0	4	0	0	0	0			
First Response Malaria Ag Combo (PLDH/HRP2)	Premier Medical Corporation Ltd.	0	0	0	0	0	0	0	0	0	0	0	0	0			
FirstSign - ParaView-2 (Pv + Pf) Card Test	Unimed International, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	2			
Hexagon Malaria Combi	Human GmbH	0	0	0	0	0	0	0	0	0	0	0	0	0			
ICT Malaria Combo Cassette Test (MI02)	ICT Diagnostics	0	0	0	0	0	0	0	0	0	0	0	0	0			
Immunoquick Malaria +4	Biosynex	0	0	0	0	0	0	0	0	0	0	0	0	0			
Malaria P.F/WVAX	Diagnostic Automation / Cortez Diagnostics, Inc.	0	0	0	0	0	4	0	3	0	0	0	0	2			
Malaria Rapid Combo	Vision Biotech (Pty) Ltd.	0	0	0	0	0	0	1	0	0	0	0	0	0			
Malaria Rapid Dual	Vision Biotech (Pty) Ltd.	0	0	0	0	0	0	0	0	0	0	0	0	1			
Malasscan Rapid Test for Malaria Pf/Pan (Device)	Zephyr Biomedicals	0	0	0	0	0	0	0	0	0	0	0	0	0			
One Step Malaria Antigen Strip	IND Diagnostic Inc.	0	0	0	0	0	0	0	4	0	0	0	0	1			
OnSight - ParaQuick (Pan, Pf)	Amgenix International, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0			
OptiMAL- IT	DiaMed AG	0	0	0	0	0	0	0	0	0	0	0	0	4			
ParahitTotal Device Rapid Test for <i>P. falciparum</i> and Pan Malarial Species	Span Diagnostics Ltd.	0	0	0	0	0	0	0	0	0	0	0	0	0			
Parascreen Rapid Test for Malaria Pan/Pf (Device)	Zephyr Biomedicals	0	0	0	0	0	0	0	0	1	0	0	0	0			
Quickstick Malaria Antigen Test	Innovatek Medical Inc.	0	0	0	0	0	0	0	4	0	0	0	0	1			
SD BIOLINE Malaria Ag	Standard Diagnostics, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0			
SD BIOLINE Malaria Ag Pf/Pan	Standard Diagnostics, Inc.	0	0	0	0	0	0	0	0	0	0	0	0	0			
Wondfo One Step Malaria Pf/Pan Whole Blood Test	Guangzhou Wondfo Biotech Co., Ltd	0	0	0	0	0	0	0	0	0	0	0	1	0			
Pf - <i>Plasmodium falciparum</i>																	
Pv - <i>Plasmodium vivax</i>																	
pan - <i>Plasmodium</i> species																	

ANNEX 5: EXAMPLE ALGORITHM FOR SELECTING A MALARIA RDT

Select (from Table 4, 4a) an RDT with sufficiently high detection rate, low false positive rate and invalid rate, then...

Parameter	Decision process	Decision
1. Plasmodium species to be targeted	<p>Distinguish non-Pf from Pf or mixed?</p> <p>Yes</p> <p>No</p> <p>Combination test HRP2 - pLDH HRP2 - aldolase pLDH - pLDH</p> <p>Pf-only test HRP2 pLDH (may be combined with pan-specific pLDH)</p>
2. Expected areas of use	<p>Store and use in tropical environment without temperature control</p> <p>Yes</p> <p>No</p> <p>High-stability High stability demonstrated, and specified temperature e.g. $\geq 35^{\circ}\text{C}$</p> <p>Storage temperature less critical Accept lower stability and $< 35^{\circ}\text{C}$ specified storage</p>
3. Likely clinical scenarios	<p>Likely to use for re-testing soon after treatment / treatment-monitoring</p> <p>Yes</p> <p>No</p> <p>Target non-persistent antigen pLDH-detecting for <i>P. falciparum</i></p> <p>Antigen persistence not critical HRP2 or pLDH for <i>P. falciparum</i></p>
4. End-user	<p>For use by health workers outside medical laboratories</p> <p>Yes</p> <p>No</p> <p>Simple format, all-inclusive Cassette design, few steps, lancet, swabs etc. included in package</p> <p>Design less critical Include dipsticks Test-only packaging</p>
5. Other considerations:	<p>Box size: How many fever cases expected in 3 months (if less than one box, request reduced box size)</p> <p>Next Step: Finalize short-list of suitable RDTs</p> <p>Sometimes preferences will be incompatible, in which case, prioritize the importance of the categories above for your programme.</p>

CHOOSING SPECIFIC PRODUCT FROM RDT SHORT-LIST

1. Contact manufacturers and request:
 - a. Quoted price (include necessary accessories, delivery to country, and staggered delivery in 2-3 batches over 12 months)
 - b. Request heat stability data as evidence of manufacturer's stated storage temperature and shelf-life
 - c. Request sample of product to assess format, ease of use, compatibility of other materials with health system requirements



2. Assess experience of others:
If possible, obtain written assessments of field experience from other countries / programmes that have experience in using the product



Make preliminary procurement decision, then consider...



3. Options to improve implementation:
 - a. Negotiate replacement of product if supplied product fails lot-testing after delivery by a method approved by manufacturer / WHO-coordinated laboratory
 - b. Develop appropriately-formatted instructions in appropriate language and consider their inclusion of these in kits at the manufacturing site
 - c. Negotiate delivery dates for staggered delivery to reduce in-country storage times, ensure long (e.g. 18 months) shelf-life after delivery



4. Organize lot-testing prior to dispersal to the field

ANNEX 6: INTRODUCING RDT-BASED MALARIA DIAGNOSIS INTO NATIONAL PROGRAMMES

As parasite-based diagnosis is introduced at smaller clinics and village level for case management, a large number of challenges arise not only in logistical administration but also in managing the health-seeking and health-providing behaviour of patients and health workers. These can be addressed by a systematic approach to planning, implementation, monitoring and evaluation of the diagnostic programme; a process that must commence well before RDTs are procured. The following information is derived from existing WHO documents addressing this area.²² More information may be obtained at www.wpro.who.int/sites/rdt.

Many health workers and communities will have been taught that "fever equals malaria unless proven otherwise". Introducing RDTs will demonstrate that this is not the case. To have an impact on anti-malarial diagnosis and treatment, RDTs must be seen to provide an accurate diagnosis by both health workers and patients alike, that is, they must be as good or better than those relied on previously. A health worker will also need a good alternative to anti-malarial medicines for the management of parasite-negative febrile patients. To achieve and maintain confidence in RDT-based diagnosis, a good quality assurance system must be in place (detailed elsewhere on this website). There must be satisfactory education of health workers, and widespread community sensitization. Knowledge of other causes of fever will be necessary to develop appropriate management algorithms for parasite-negative cases.

At the national level, regulatory requirements may need to be developed to control the importation and

use of malaria RDTs, and new procedures for storage, distribution and inventory management, such as those used for medicines, may need to be developed. If changing from a different product or mode of diagnosis, an adequate phase-out plan for this must also be developed.

This requires a clear strategic plan to be developed well in advance of RDT introduction, with a clear timeline to ensure that the various components of the RDT programme are in place at the right time. A focal person, or persons, will be needed to coordinate the overall implementation plan and ensure that the various agencies that may be involved understand the process and their particular roles. To achieve this, funding for the programme must include a significant component for planning and coordination, sensitization/IEC, training, quality assurance, monitoring and supervision, and logistics, in addition to procurement. Without this, much of the funds expended on RDTs may be wasted, and a loss of confidence in RDT-based diagnosis may hinder the process of strengthening appropriate malaria case management.

An example of a national implementation plan is shown below. This will need to be modified considerably for each programme, preferably through a collaborative process involving all the major agencies concerned in its implementation. Budgeting for all the components of the programme at the outset is vital. An example of components to be considered in an overall budget is shown in Figure A6.1.

22 Developed by WHO Regional Office for the Western Pacific and the WHO Global Malaria Programme, with support from the Uganda Ministry of Health (National Malaria Control Programme), Management Sciences for Health (MSH), and other partners.

Summary of introduction plan (see following page)

Program planning and management

- Identify key stakeholders, and secure commitment for introduction of RDTs
- Establish working group and develop terms of reference
- Identify specific focal person(s) responsible for day to day oversight of the implementation plan
- Develop a timeline, scope, and budget for implementation
- Identify human and other resource needs, and a strategy for accessing them
- Review and update, if needed, case-management algorithms for malaria and other causes of febrile illness

Policy and regulatory issues

- Develop appropriate regulatory documents if required
- Register RDT products

Procurement of RDTs

- Develop product specifications and packaging requirements
- Develop product short-list
- Conduct quantification (estimation of needs)
- Procure RDTs
- Procure sharps boxes, gloves etc.

Logistics

- Develop distribution plan
- Train logistics and storage personnel in handling and distribution of RDTs
- Implement a system for data collection and information flows
- Arrange for appropriate transport and storage
- Review and strengthen inventory management, as needed
- Develop a plan for discontinuation and disposal of other diagnostic supplies, if appropriate

Quality Assurance

- Develop mechanisms for assessing samples at a national level (lot-testing), and regular (and random) testing at the level of use (e.g. microscopy-sentinel sites)
- Implement post-marketing surveillance

Training and communication

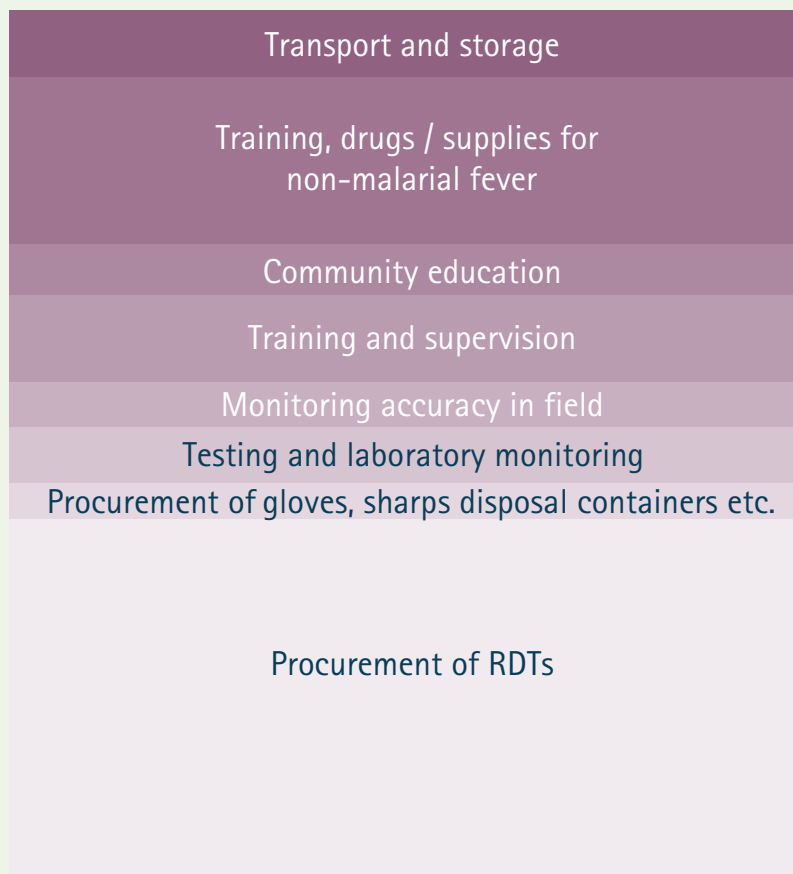
- Develop appropriate training and supervision materials
- Train health workers in case management and managing commodities
- Train in RDT use
- Develop and implement a program for community education/ sensitization

Monitoring and Evaluation

- Implement effective supervision and monitoring
- Strengthen recording and reporting procedures

Figure A6.1 Example malaria RDT implementation budget

Below is an example of major components of a programme budget to be considered when introducing RDTs into a malaria programme. Without adequate provision for each of these factors, it is likely that an RDT-based diagnostics programme will fail to achieve its goals. These components should therefore be addressed in proposals for programme funding, or provisions should be made for them in collaborating programmes.





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