ImmunoScience INDIA

ImmunoQuick

Malaria Pf

Rapid Malaria Pf (HRP 2) Antigen Test - Device

For *In-Vitro* Diagnostic Use Only **Store at 4°C to 40°C**

OVERVIEW

Malaria is a serious parasitic disease characterized by fever, chills, and anemia and is caused by a parasite that is transmitted by the bite of infected Anopheles mosquitoes. There are four kinds of malaria parasites that can infect humans: Plasmodium falciparum, P. vivax, P. ovale, and P. malariae. In humans, the parasites (called sporozoites) migrate to the liver where they mature and release another form, the merozoites. At present Malaria is diagnosed microscopically using thick and thin blood films. These require expert knowledge to correctly identify the species, not always available 24 hrs a day. This is where a reliable support test becomes invaluable.

INTENDED USE

This is a rapid, in vitro, qualitative lateral flow immunoassay for the detection of P. falciparum specific histidine rich protein-2 (Pf. HRP-2) from human whole blood samples. The test may also be used for the differentiation of P. falciparum and P. vivax infection.

PRINCIPLE

The Rapid Malaria Pf HRP-2 Antigen test-device contains a membrane strip, which is pre-coated with P. falciparum specific monoclonal antibody to HRP-2 antigen as test line (Pf) and Goat anti-rabbit IgG as control line (C).

After addition of the blood sample and the assay buffer to the respective wells on the device containing a test strip, the whole blood gets lysed and moves on to the conjugate pad containing HAMA blocking reagent and colloidal gold particles conjugated with malaria Pf specific HRP-2 antibodies, rabbit IgG If the sample contains detectable levels of the Pf HRP-2 antigen it reacts with the gold conjugated malaria Pf specific HRP-2 antibodies to form a complex. This complex moves further and reacts with the malaria Pf specific HRP-2 antibodies coated on the nitrocellulose membrane in test area to form a coloured band (Test line). The unbound complex and the rabbit IgG conjugated colloidal gold particles move further to the goat-anti rabbit IgG coated control area to form a coloured band (Control line). The appearance of test line and control line in respective area indicates the positive result. Appearance of only control line indicates a negative result. The control line acts as a procedural control. Control line should always appear if the test is performed as per the procedure and reagents are working properly.

MATERIAL PROVIDED

1. Test: Nitrocellulose Membrane assembly pre-dispensed with monoclonal anti-Pf (HRP-2) antibody, Goat anti-rabbit IgG, Conjugate strip containing HAMA blocking reagent and colloidal gold conjugated monoclonal anti-Pf (HRP-2) antibody and rabbit IgG at the respective regions.
2. Desiccant pouch
3. Disposable 5µl sample applicator
4. Package Insert
5. Assay Buffer

OPTIONAL MATERIAL REQUIRED

1. Calibrated micropipette capable of delivering 5 µl sample Accurately.
2. Stop watch.
3. Disposable gloves

PRECAUTIONS/KIT STORAGE AND STABILITY

1. Please read all the information in this package insert before performing the test. Pay particular attention to the position of the Control and Test lines.
2. Do not use after the expiration date printed on the foil pouch.
3. Store in the sealed pouch in a dry place in between temperature 4°C to 40°C. Do not freeze.
4. Do not use if pouch is torn or damaged.
5. Do not open the foil pouch until you are ready to start the test.
6. Keep out of the reach of children.

WARNINGS

1. Do not reuse the test.
2. Follow the instruction to get accurate results.
3. Use appropriate personal protective equipment.
4. Dispose of hygienically as per local regulatory requirements.
5. Do not touch the membrane.
6. Treat blood samples and used tests as potentially infectious. Avoid contact with skin.
7. For in vitro diagnostic use. Not to be taken internally.
8. Do not eat the desiccant in the package.
9. Do not mix the specimen sample or interchange the different specimen.

SPECIMEN COLLECTION

Fresh anti-coagulated whole blood should be used as a test sample. EDTA or Heparin or Oxalate or Tri-sodium Citrate can be used as suitable anticoagulants. The specimen should be collected in a clean glass or plastic container. If immediate testing is not possible then store the specimen at 2°C to 8°C for up to three days before testing. Clotted or contaminated blood samples should not be used for performing the test. Fresh blood from finger prick/ puncture may also be used as a test specimen.

TEST PROCEDURE

1. Bring the kit components to room temperature before testing.
2. Open the pouch and retrieve the test and desiccant pouch. Check the color of the desiccant. It should be blue, if it has turned colorless or pink, discard the test and use another test. Once opened, the test must be used immediately.
3. Label the test with patient's identity.
4. Tighten the vial cap of the assay buffer provided with the kit in the clockwise direction to pierce the dropper bottle nozzle.
5. Evenly mix the anti-coagulated blood sample by gentle swirling. Dip the sample loop into the sample. Ensuring that a loop full of blood is retrieved, blot the blood so collected in the sample port 'S'. (This delivers approximately 5µl of the whole blood specimen).

   OR

   In case finger prick blood is being used, touch the sample loop to the blood on the finger prick. Ensuring that a loop full of blood is retrieved, immediately blot the specimen in the sample port 'S'. (Care should be taken that the blood sample has not clotted and the transfer to the sample port is immediate).

   OR

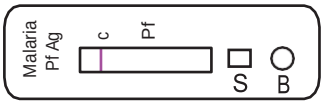
   Alternatively, 5µl of the anti-coagulated or finger prick specimen may be delivered in the sample port 'S' using a micro pipette.

Note: Ensure that the blood from the sample loop has been completely taken up at the sample port 'S'.

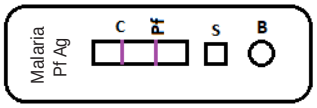
6. Immediately dispense three drops of assay buffer (Approx. 90µl) into buffer port 'B', by holding the plastic dropper bottle vertically.
7. Read the results at the end of 20 minutes. Do not read the result after 30 minutes.

INTERPRETATION OF RESULTS

NEGATIVE for Malaria: If colored band appears at the control region 'C' only,



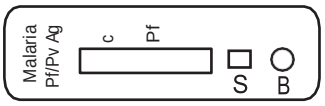
POSITIVE for P. falciparum infection: In addition to the control band, Colored band appear at regions 'Pf' in the test window.



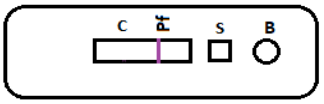
INVALID: The test should be considered invalid if

A) No line appears at 'C' and 'Pf' regions.

B)



C) No line appears at 'C' region and line appear at Pf region.



PERFORMANCE CHARACTERISTICS

Internal Evaluation:

In an in-house study, total 200 samples were evaluated for sensitivity and specificity. We found the relative sensitivity was 100 % (i. e. 52/52) and the relative specificity was 100 % (i. e. 148/148).

The results are summarized in the following table:

following table:

Sample	Total Number of Samples Tested	Rapid Malaria Pf (HRP 2) Antigen Test		Sensitivity (%)	Specificity (%)
		Positive	Negative		
Malaria Pf Positive Whole Blood Samples	52	52	0	100	-
alaria Negative Whole Blood Samples	148	0	148	-	100

Cross reactivity was studied using RF positive samples and no cross reactivity was observed.

External Evaluation:

In an external study, total 200 samples were evaluated for sensitivity and specificity. Relative sensitivity was 100 % (i. e. 19/19) and the relative specificity was 100 % (i. e. 181/181). Positive Predictive Value (PPV) and Negative Predictive Value (NPV) for the test was 100%.

The results are summarized in the following table:

Sample	Total Number of Samples Tested	Rapid Malaria Pf (HRP 2) Antigen Test		Sens-itivity %	Speci-ficity %	PPV (%)	NPV (%)
		Positive	Negative				
Malaria Pf Positive Whole Blood Samples	19	19	0	100	-	100	-
Malaria Negative Whole Blood Samples	181	0	181	-	100	-	100

LIMITATIONS

1. As with all diagnostic tests, the test result must always be correlated with clinical findings.
2. The results of test are to be interpreted within the epidemiological, clinical and therapeutic context. When it seems indicated, the parasitological techniques of reference should be considered (microscopic examination of the thick smear and thin blood films).
3. Any modification to the above procedure and/or uses of other reagents will invalidate the test procedure.
4. The test is limited to the detection of antigen to Malaria Plasmodium sp. Although the test is very accurate in detecting pLDH and HRP-2, a low incidence of false results can occur. Other clinically available tests are required if questionable results are obtained. As with all diagnostic tests, a definitive clinical diagnosis should not be based on the results of a single test, but should only be made by the physician after all clinical and laboratory findings have been evaluated.

REFERENCES

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2. Quintana M., et. al.,(1998) Malaria diagnosis by dipstick assay in a Honduran Population with coendemic Plasmodium falciparum and Plasmodium vivax. Am. J. Trop. Med. Hyg. 59(6) 868-871.
3. Hunte-Cooke A., et. al., (1999) Comparison of a Parasite Lactate Dehydrogenase-based Immunochromatographic Antigen Detection assay (OptiMAL®) with Microscopy for the Detection of Malaria Parasites in Human Blood Samples. Am J.Trop Med 60(2). 173-176.
4. John, S. M., et. al.,(1998) Evaluation of OptiMAL , a dipstick test for the diagnosis of malaria. Ann. Trop. Med. Parasitol., 92, 621-622.

	Manufacturer
	Manufacturing Date
	Expiry Date
LOT	Lot Number
	Number of tests in the pack
	Do not use if pouch or kit damaged
	This side Up

MANUFACTURED BY

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