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See external label,



$\Sigma=25$ or 50 tests



Cat. #172103P-25



2°C-30°C for test card and 2°C-8°C for Sample tube

For the convenience of storage, test cards and sample tubes can be stored separately

Malaria P.f. **Malaria Plasmodium falciparum (P.f.) Rapid Test**

INTENDED USE:

This Malaria *Plasmodium falciparum* (P.f.) Rapid Test is a qualitative test for the detection of histidine-rich protein 2 antigen (HRP-2) of P.f. in human whole blood. This test is for In-Vitro Diagnostic use only.

INTRODUCTION:

Malaria is one of the world's most prevalent parasitic diseases and ranks third in the world among major infectious diseases in terms of mortality. The protozoal parasites that cause malaria are from the *Plasmodium* genus. Four species of *Plasmodium* protozoa cause malaria: *Plasmodium falciparum*, *Plasmodium vivax*, *Plasmodium malariae* and *Plasmodium ovale*. Transmitted principally by the Anopheles mosquito, malaria infections may also occur from contacting infected blood, such as from blood transfusions.

P. falciparum accounts for the majority of infections and is the most lethal. *P. vivax*, *P. malariae* and *P. ovale* cause a less severe form of malaria with intermittent fever which is usually neither debilitating nor fatal. Classic symptoms of malaria include fever, headaches, chills, vomiting, shivering and convulsions. In some rare forms of falciparum malaria, chills and fever may be absent and the patient may present with delirium or coma. Remission periods can last from a few weeks to several months. Severe anemia is often attributed to the cause of death from a malaria infection.

Malaria is a curable disease with a host of drugs that can be used in both its treatment and prevention. Two of the best known and most commonly used are chloroquine and quinine. The early detection of *P falciparum* malaria is of great importance due to rising levels of drug resistance now being associated with this disease.

TEST PRINCIPLE

This *Plasmodium falciparum* (P.f.) malaria test is a rapid, *in-vitro* immunodiagnostic test for the detection of circulating P.f. antigen in whole blood. The test uses antibodies that are specific for the histidine-rich protein 2 antigen (HRP-2) of P.f.

Whole blood (5 uL) is applied to the sample pad where the red blood cells are lysed with a specially formulated solution. The label pad that is next to the sample pad on the strip is impregnated with blue latex that has an anti- HRP-2 antibody coupled to it. The label pad is also impregnated with purple latex that is coupled to a control antibody. A second anti-HRP-2 antibody is immobilized on the test strip at the test line region. A control material is immobilized on the strip at the control line region. When a positive sample is applied to the sample pad, P.f. antigen in the sample contacts the latex-labeled antibody and binds to it. A washing reagent is then added to a test vial, and the strip is placed in the vial. As the liquid flows along the length of the strip, any antigen-latex complexes also migrate with the liquid. These complexes are captured by their respective antibodies at the test and control line regions. If a sample contains P.f. antigen, a blue line will form in the test region. If no P.f. antigen is present, a blue line will not form in the test region. A purple control line will always appear in the control region if the test has been properly performed.

KIT CONTENTS:

Each kit contains the following components in sufficient quantities to perform the number of tests indicated on the package label:

- 25 test devices packaged in individual foil pouches.
- 25 sample collection capillaries
- 1 bottle of Lysing/Wash reagent
- 1 product insert

MATERIALS REQUIRED BUT NOT SUPPLIED:

- Lancets
- Disinfecting, sterile wipe
- Timer capable of timing from 0 to 60 minutes

PRECAUTIONS:

1. Specimens should be handled as being potentially infectious. The Centers for Disease Control (CDC) and the National Institutes of Health (NIH) recommend that all potentially infectious agents be handled at a Biosafety Level 2.
2. Biological decontamination procedures should be followed for all equipment, containers, surfaces, etc. that come in contact with potentially infectious specimens. All disposables that come in contact with these samples should be disposed of as infectious waste.
3. For best results, strict adherence to these instructions is required. Be careful not to touch the tip of the wash bottle to the sample well when adding buffer to the device. This will greatly minimize the likelihood of contaminating the wash reagent.
4. The wash solution contains a low concentration of sodium azide as a preservative (less than 0.1 %). Sodium azide is toxic. Do not drink this buffer. Sodium azide may also react with lead and copper in plumbing to form explosive compounds. If you dispose of this buffer down a drain, flush the drain with excess amounts of water to minimize the accumulation of potentially explosive metal-azide compounds.
5. Do not use the test devices or Lysing/Wash reagent beyond the stated expiration date marked on the package label.
6. Store the test kits and buffer according to temperature and humidity conditions stated on the package label.
7. All test devices, buffers and specimens must be at room temperature (15-30°C) before running the assay.
8. Do not re-use the test devices.

STORAGE AND SHELF LIFE OF REAGENTS:

Store the kit between 2°C and 30°C. Do not store the kit in direct sunlight. Be sure to use the device immediately after removing it from its foil pouch. The test kit may be used until its expiration date, which can be found on the package label.

SPECIMEN COLLECTION:

1. Handle all specimens as capable of transmitting infectious diseases. Dispose of all materials that come in contact with the specimen as infectious waste.
2. Specimens should be collected aseptically by fingerstick or venipuncture according to standard methods such as those specified by the National Committee for Clinical Laboratory Standards (NCCLS). The use of grossly lipemic or turbid samples should be avoided.
3. Whole blood samples should be used immediately, if possible. NCCLS provides recommendations for storing blood specimens (Approved Standard - Procedures for the Handling and Processing of Blood Specimens, H1SA, 1990).
4. **Use the collection capillary provided to deliver a 5 µL sample or collect venous blood into EDTA tubes.** To obtain capillary blood, puncture a finger, heel or other appropriate site. First cleanse the area with a disinfecting sterile wipe. Use a lancet to puncture the skin. Allow a blood droplet to form. Touch the collection capillary to the blood droplet and transfer to the test strip immediately. To collect venous blood, use the standard venipuncture procedure and collect blood into an EDTA tube. If the test cannot be performed immediately, the blood may be stored for up to three days at 2°C to 8°C.

TEST PROCEDURE:

1. Just prior to use, remove a device from its foil pouch. Lay the test device flat on the work surface.
2. Using a sterile lancet and clean sample capillary, collect blood by puncturing an accessible site (e.g., finger or heel). Allow a blood droplet to form at the puncture site and touch the tip of the capillary to the blood droplet. Allow blood to fill about 3/4 of the capillary. Alternatively, 5 µL of EDTA venous blood may be used. Ensure that the blood sample warms to room temperature prior to use.
3. Transfer the blood sample from the capillary tube to the test device by holding the capillary vertically and gently touching the full end against the pad within the sample addition port until all of the blood has been transferred. Discard the capillary properly. If using a micro-pipetter, slowly apply 5 µL of blood to the sample pad.
4. Immediately add one drop of the Lysing/ Wash reagent to the sample port on top of the whole blood.
5. Add five drops of the Lysing/Wash reagent to the buffer well.
6. Using a timer, allow the reaction to proceed for 15 minutes. Do not pick up the device during this time.
7. When the 15-minute period is over, read the results. If there is still a reddish background, lay the device flat on the work surface and wait an additional 15 minutes. The results may be read from 15 to 30 minutes. Do not read results after 60 minutes.

Negative results must be confirmed at 30 minutes

IMPORTANT NOTICE:

This test only detects malaria infections caused by *Plasmodium falciparum*.

Occasionally, residual malaria antigen may be detected for several days following elimination of the parasite by anti-malarial treatment. The diagnosis of Malaria should be made using the results of this test together with the other clinical and laboratory

findings.

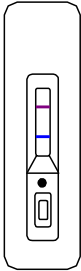
INTERPRETATION OF THE RESULTS:

A positive result is indicated when any visible line forms in the result window next to the test zone together with a line in the C zone. The test is positive even if the line in the test zone appears lighter or darker than the line in the C zone.

1. The test is not valid if the control line does not appear, regardless of the presence of line in the test line region. Repeat the test with a new device.
2. Positive results may appear as early as 5 minutes. Negative results must be confirmed after at 30 minutes.
3. The background of the strip should be pinkish-white, not red, prior to confirming a negative result.
4. Results should not be read after 60 minutes.

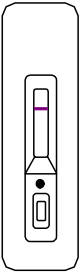
Positive Test Result

A visible blue test line on the strip located in the test zone indicates a positive test result for *Plasmodium falciparum*. The purple control line must also be present.



Negative Test Result

The test is negative if only the control line appears



Invalid Test Result

The test is invalid if a purple line does not appear in the control zone. If this occurs, the test should be repeated using a new test device.

Histidine Rich Protein 2 (HRP-2) is secreted by the *Plasmodium falciparum* species. Its presence usually indicates a malaria *P.f.* infection. Occasionally, residual HRP-2 may be detected for several days following elimination of the parasite by anti-malarial treatment. The diagnosis of *P.f.* Malaria should be made using the results of this test together with the other clinical and laboratory findings.

QUALITY CONTROL:

1. For the assay to be considered valid, the control line must appear. If it does not appear, the test results are not valid and the test must be repeated.
2. In addition to your laboratory's standard quality control procedures, the NCCLS recommends that a positive and negative external control be tested at least once within each 25- test kit and by each operator performing testing within a kit. This will verify that the reagents and test strips are working properly and the operator is able to correctly perform the test procedure. Please refer to this NCCLS publication C24- A for recommendations on appropriate Quality Control practices.

LIMITATIONS OF THE TEST:

1. This HRP-2 based Malaria *P.f.* tests may give positive malaria results for up to 2 weeks following chemotherapy and

parasite clearance as confirmed by microscopy.

2. As with all diagnostic tests, the result must be correlated with clinical findings. If the test result is negative and malaria infection suspicion still exists, additional follow-up testing using other clinical methods is recommended.
3. A negative result at any time does not preclude the possibility of an early malaria infection.
4. Strict adherence to the test procedure is required. Do not re-use negative devices. Do not adulterate the Lysing/Wash reagent.
5. This test cannot be used to monitor therapy or to estimate the titer of the infection.
6. A final diagnosis should be based on these test results in conjunction with other clinical and laboratory findings.

SENSITIVITY AND SPECIFICITY:

A clinical study using a total 370 whole blood samples was conducted at various sites in 3 countries. The results of the Merlin Labs malaria combo test were compared with the blood smear / microscopy method. The sensitivity and specificity of the *Pf* test results are given below:

<i>P.f.</i> Test Results	smear (+)	Blood smear (-)
Rapid Test (+)	44	8
Rapid Test (-)	0	318

Sensitivity = >99% Specificity = 98%

STABILITY:

This Malaria *P.f.* test has been found to be stable for up to 14 months from the date of manufacture when stored between 2 to 30°C. The expiration date of each test can be found on the Kit box label. No component or reagent of the test should be used beyond its printed expiration date.

REFERENCES:

1. Taylor, DW, Voller, A. The development and validation of a simple antigen detection ELISA for *Plasmodium falciparum* malaria. *Trans. R. Soc. Trop. Med* (1993), 87:29-31
2. Parra ME, et. al. Identification of *Plasmodium falciparum* histidine rich protein II in the plasma of humans with malaria. *J. Clin. Microbiol.* (1991), 29: 1629-1634
3. Beadle, C, et.al. Diagnosis of malaria detection of *Plasmodium falciparum* HRP-II antigen with rapid dipstick antigen capture assay. *Lancet* (1994), 343: 564-568.
4. World Health Organization Press Release (2001) May 23, "WHO and Novartis join forces to combat drug resistant malaria."
5. World Health Organization Fact Sheet (1998) , Malaria, No.94
6. CDC/NIH Guidelines. Biosafety in Microbiological and Bio-medical Laboratories. 2nd Edition, 1988
7. Soti-Strong, Diagnosis, prevention, and treatment of tropical disease, 7th ed., Philadelphia, The Ablakiston Company



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