Humasis

P.f P.f/P.v P.f/Pan

HUMASIS MALARIA ANTIGEN TEST

HIGH SENSITIVE DIFFERENTIAL DIAGNOSIS OF MALARIA INFECTION

References

1) World Malaria Report 2010, WHO
2) Rapid diagnostic tests for malaria parasites, Clin. Microbiol. Rev. 2002, 15(1):66-78
3) The role of rapid diagnostic tests in managing malaria, PLoS Medicine, 2009, 6(4):1-2
4) Malaria rapid diagnosis_making it work, WHO regional office for the western pacific, 2003

What is Malaria?

Malaria is a mosquito-borne infectious disease of humans caused by eukaryotic protists of the genus *Plasmodium*. It is widespread in tropical and subtropical regions, including much of Sub-Saharan Africa, Asia and the Americas. Malaria is prevalent in these regions because of the significant amounts of rainfall and consistent high temperatures; warm, consistent temperatures and high humidity, along with stagnant waters in which their larvae mature, provide mosquitoes with the environment needed for continuous breeding.

According to WHO in 2010, there were 247 million malaria cases among 3.3 billion people at risk in 2008 from 109 countries resulting in an estimated 881,000 deaths. These deaths were primarily in Africa (91%) and in children under 5 years of age (85%).

Malaria, countries or areas at risk of transmission, 2010

Figure 1. Geographical distribution of Malaria infection.

Four species of *Plasmodium* can infect and be transmitted by humans. Severe disease is largely caused by *Plasmodium falciparum*. Malaria caused by *Plasmodium vivax*, *Plasmodium ovale* and *Plasmodium malariae* is generally a milder disease that is rarely fatal.

Table 1. Differentiating features of *P.falciparum* and *P.vivax*

P.falciparum	*P.vivax*
Associated with serious complications e.g. cerebral malaria, jaundice, renal failure etc. including high mortality	Relatively benign and rarely produces serious complications or death
35% of RBC's involved	Less than 1% of RBC's are parasitized
Gametocytes persists in the blood for 30~60 days or more	Gametocytic stage persists in the peripheral blood for 2 days

Diagnosis of Malaria

Microscopy remains the gold standard for detection of malaria parasitemia as it can provide information on both the species of parasite and parasite density of infection. However, the procedure is labor-intensive and time-consuming, requiring substantial training and expertise due to fleeting skills. These problems are magnified in nonendemic regions where light microscopy to diagnose malaria is infrequently performed, resulting in missed diagnosis, misidentification of *Plasmodium* species, and therapeutic delays. Methods using advances in technology have been evaluated as alternatives to light microscopy. While these methods have varying strengths and weaknesses, they are limited by equipment, supplies, expertise, cost, time, applicability in acute infection, and/or availability. New immunochromatographic rapid diagnostic tests (RDTs) for malaria were introduced.

Table 2. Comparison of methods for diagnosing Malaria in blood

Parameter	Microscopy	PCR	Fluorescence	RDT HRP-II	RDT pLDH
Sensitivity (parasites/uL)	50	5	50	>100	>100
Specificity	All species	All species	*P.falciparum* good, others difficult	*P.falciparum* only	*P.falciparum*, *P.vivax*, *P.ovale* and *P.malariae* only with pLDH
Parasite density or parasitemuia	Yes	No	50	>100	>100
Time for result	30~60 min	24 h	30~60 min	15~30 min	15~30 min
Skill Level	High	High	Moderate	Low	Low
Equipment	Microscopy	PCR apparatus	Fluorescence microscopy	None	None
Cost/test	Low	High	Moderate	Moderate	Moderate

HUMASIS Malaria Antigen Test

Histidine Rich Protein-II (HRP-II)

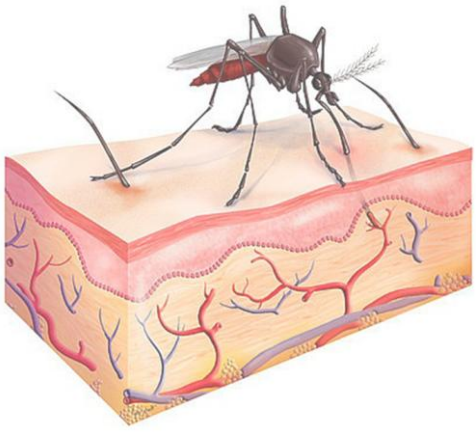
- The antigen is expressed only by *P.falciparum* trophozoites.
  The antigen can be detected in erythrocytes, serum, plasma, cerebrospinal fluid and even urine as a secreted water-soluble protein.
- It generally takes around two weeks after successful treatment for HRP-II-based tests to turn negative, but may take as long as one month, which compromises their value in the detection of active infection.
- Since HPR-II is expressed only by *P. falciparum*, these tests will give negative results with samples containing only *P. vivax, P. ovale*, or *P. malariae*; many cases of non-falciparum malaria may therefore be misdiagnosed as malaria negative (some *P.falciparum* strains also don't have HRP-II)

pLDH – panLDH

- *P. falciparum* lactate dehydrogenase (pLDH) is a 33 kDa oxidoreductase. It is the last enzyme of the glycolytic pathway, essential for ATP generation and one of the most abundant enzymes expressed by *P. falciparum*.
- pLDH is similar to pGluDH. LDH from *P. vivax, P. malariae*, and *P. ovale* exhibit 90~92% identity to pLDH from *P. falciparum*.

Table 3. Characteristics of HRP-II and pLDH antigen

Description	HRP-II	pLDH
Sensitivity	Approx. >99.9% (100 parasite/uL)	Approx. >95% (100 parasite/uL)
Specificity	Approx. >99.5%	Approx. >99.5%
False Result	False positive result for 7~14 days following chemotherapy	False negative result in low levels of parasites False positive result for gametocytes
Differential Detection	P.f only	P.f and other species (Pan)
Detection period	Human life cycle except matured gametocytes	All stage of human life cycle including gametocyte
Purpose	Suitable test for prevalence of P.f	For monitoring with therapy Suitable test for prevalence of *P.vivax*



HUMASIS Malaria Antigen Test

Humasis Malaria Antigen test can offer significant benefits in malaria management as followings:

- A clear benefit will occur in health outcomes
- Allow more rational use of anti-malarial drugs
- The accuracy of Humasis Malaria Antigen test is quality controlled
- Protected from high temperature
- Affordable Price

Potential uses for Humasis Malaria Antigen test

- Outbreak investigation and malaria prevalence surveys.
- Diagnosis by health workers distant from good microscopy services.
- 'After-hours' diagnosis in hospital or clinics.
- Remote diagnosis in organized workforces in endemic area.

Test Methods

Whole blood

1. FIRST, read carefully the instructions on how to use Humasis Malaria Antigen test kit.

2. Open the package and look for the following.

1) Test device individually packaged with a foil-pouch and with a silica gel in it.

2) Buffer

3) Disposable sample loop(5uL)

4) Lancet

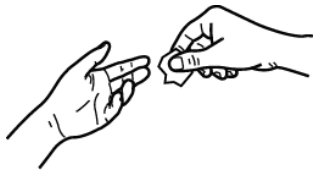
5) Alcohol prep pad

Open the foil pouch, and look for the following.

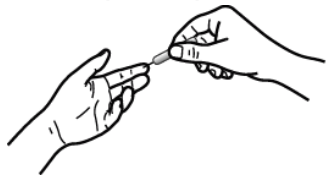
3. Next, look at the expiry date at the back of the foil pouch. Use anothe tet device if the expiry date has passed.

(For examlpe)

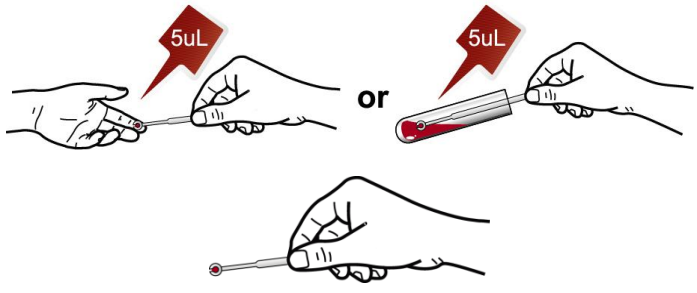
4. Clean the fingertip with an alcohol pad and let dry.



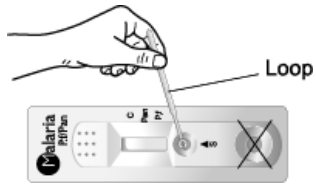
5. Take a lancet and make a quick deep stab on the side of the finger.



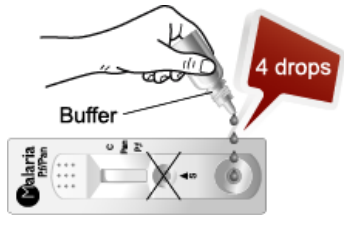
6. Take 5uL of whole blood by a loop.
In order to collect the correct amount of blood, lay the loop and fill a film of blood completely across the loop.



7. Drop the specimen in a specimen insertion-hole.



8. Add 4 drops of buffer(approximately 120uL).



9. Wait for 30 minutes and then read the result.
Do not interpret the test result after 30 minutes.

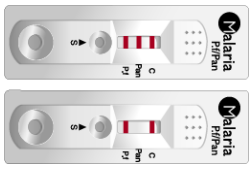
Interpretation of the results

Malaria P.f/Pan Antigen Test

Negative

Positive

1. Positive for P.f



The colored band in P.f region can be appeared after the medical treatment.

2. Positive for P.v or P.o or P.m

3. Positive for P.f and P.v or P.o or P.m

Invalid

Malaria P.f/P.v Antigen Test

Negative

Positive

1. Positive for P.f

2. Positive for P.v

Invalid

Specification

Specification	Humasis Malaria P.f/Pan Antigen Test	Humasis Malaria P.f/P.v Antigen Test	Humasis Malaria P.f Antigen Test
Test Method	Lateral flow test		
Result within	15-30minutes		
Specimen	Whole blood or finger punctured blood		
Semple volume	5uL		
Target antigen	HRP-Ⅱ for P.f and pLDH for P.f, P.v, P.o and P.m	HRP-Ⅱ for P.f and pLDH for P.v	HRP-Ⅱ for P.f
Sensitivity	P.f: 99.9% P.pan: 95.6%	P.f: 99.9% P.v: 95.6%	99.9%
Specificity	99.5%	99.5%	99.5%
Stability	24 months		
Storage	1~30'C		

Contents

- Humasis Malaria Antigen Test
  25 Devices
- Disposable sample dispensing loop (5uL)
  25 ea
- Dilution buffer
  1 bottle
- Alcohol prep pad
  25 ea
- Lancet
  25 ea
- Instruction Manual
  1 ea

Ordering Information

Test Device	Cat. No.
HUMASIS Malaria P.f/Pan Antigen Test	AMAL-7025
HUMASIS Malaria P.f/P.v Antigen Test	AMFV-7025
HUMASIS Malaria P.f Antigen Test	AMPF-7025

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