Proposed guidelines for malaria antigen testing

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To the Editor: The use of rapid immunochromatographic antigen detection tests (RDTs) to diagnose malaria infection in South African pathology laboratories has been both a blessing and a confounder over the years. RDTs are sensitive and have probably saved many lives. On the other hand laboratory personnel tend to become overly reliant on antigen tests instead of using them in conjunction with microscopy, and pan-malarial antigen tests have sometimes led to confusion and misdiagnosis. We propose the following guidelines for malaria antigen testing in the hope of improving the use of malaria antigen tests in South Africa.

Many rapid malaria antigen tests use immunochromatographic technology. Rapid antigen testing provides a simple, quick, sensitive method for determining the presence of malaria parasites. Their sensitivity for detecting *Plasmodium falciparum* infections is usually high (> 90%) compared with light microscopy in a routine laboratory setting.[1] However, any malaria antigen test should first be validated for the setting in which it will be performed, then regularly quality-assured thereafter. In addition, these tests should not be considered a complete substitute for direct microscopic examination of thick and thin Giemsa-stained blood smears, which remains the international gold standard for the detection and identification of malaria parasites.

Circumstances under which malaria antigen testing should be performed

- Antigen testing should be performed only by suitably trained staff.
- Storage conditions, expiry dates and methods should be strictly adhered to.
- Antigen testing is useful to obtain a quick preliminary result, and the laboratory report should reflect that a limited test was performed.
- Antigen test results should be analysed in conjunction with thick and thin Giemsa-stained blood smears where possible.
- Antigen testing is useful to confirm the presence of *P. falciparum* in a mixed infection with another malaria species.
- Antigen testing is useful in situations where there is no experienced, competent microscopist or equipment available to perform blood smears.

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Limitations and misconceptions about malaria antigen tests

- Persistence of antigenaemia despite parasite clearance following treatment has been observed with the histidine-rich protein 2 (HRP II) antigen tests. This limits their use in terms of monitoring response to treatment and may cause confusion in the evaluation of treated patients.[1]
- RDTs that detect both *P. falciparum* and non-falciparum species cannot distinguish pure *P. falciparum* infections from co-infections with *P. falciparum* and the other malaria species, as the test line configurations are limited.[1]
- RDTs that detect both *P. falciparum* and non-falciparum species cannot differentiate between *P. vivax*, *P. ovale* and *P. malariae*.[1]
- False-negative results, even in the face of high parasitaemias, have been described.[2]
- False-positive results, especially in patients who are rheumatoid factor-positive, are possible.[3]
- Many antigen tests that claim to detect *P. falciparum* and *P. vivax* in fact do not specifically detect *P. vivax*, but rather a pan-malarial antigen common to all four human malaria species. This can lead to confusion in the laboratory. However, the sensitivity of pan-malarial antigens for *P. ovale* and *P. malariae* infections has been reported to be low.[4]
- *P. falciparum* accounts for more than 95% of the malaria cases in southern Africa, so an antigen test detecting only this species is the most cost-effective solution. *P. vivax* is the least common of the four human malaria species occurring in southern Africa, making antigen testing for this species scientifically questionable.
- The sensitivity of antigen tests decreases at low parasitaemias and may only be 50 - 70% compared with microscopy at parasite loads less than 100/µl.[5]
- A practical limitation is lack of parasite load quantitation, which is regarded as integral to laboratory diagnosis of malaria.
- Batch quality variability of RDTs has been reported.[6]

Malaria antigen tests are a valuable additional tool when used under the correct circumstances and in conjunction with smear microscopy. It is important that the abovementioned limitations of RDTs are well understood to allow the correct interpretation and use of these tests.[6] In laboratories with inexperienced malaria microscopists and where reliable microscopy may not be available, it would be in the patient's best interests to perform both malaria antigen testing and blood smear examination.

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