

This briefing paper provides an overview of HIV diagnostic tests for people planning, commissioning or providing HIV prevention activities in England. Increasing the uptake of HIV testing in order to reduce rates of undiagnosed infection and late diagnosis is a key goal of the HIV Prevention England programme. Many non-clinical services are now involved in providing or promoting HIV testing services. In order to accurately explain the benefits of HIV testing to clients, workers need to know what kind of test is being offered and how accurate it is, especially in the context of recent infection.

What would an ideal test be like?

A set of criteria are commonly used to evaluate the appropriateness of using a medical test, particularly when it is offered to people who feel well. Firstly, the test must detect a medical condition that is not trivial and which can be treated. This is clearly the case for HIV.

Moreover, the test should ideally be:

- Very accurate in identifying people who have the infection (“sensitive”).
- Very accurate in identifying people who do not have the infection (“specific”).
- Very accurate in identifying people who have recently been infected.
- Reliable, giving the same result each time a sample is tested.
- Non-invasive (a needle or tube does not enter the body).
- Safe.
- Inexpensive.
- Simple to carry out, without complex equipment or training.
- Suitable for testing lots of people.
- Quick to give a result.

A test for HIV should also detect the full range of HIV-1 subtypes and identify people who are infected with HIV-2.

Unfortunately, no medical test is perfect. Choices must be made based on the available tests, the needs of the people being tested and our priorities.

- Which is more important – that people who have an infection are identified, or that people who do not have the infection are not falsely alarmed by the suggestion that they might be infected?
- If a rapid, convenient and non-invasive test encourages

more people to test, should it be used even though it is less accurate than another test?

Although no test is 100% accurate, it is important to stress that no one test is used in isolation, especially to give an HIV-positive diagnosis. If a test appears to give a positive result, the validity of this must always be verified with a series of confirmatory tests.

This briefing summarises some key issues with HIV testing technologies commonly used in laboratories, community settings and self-sampling services, as well as describing the tests that may become available for self-testing (home-testing).

Common questions about testing

How soon after taking a risk can I test?

Most clinics advise people who have recently taken a risk to test immediately, and believe that it is unhelpful to ask people to put off testing until later. If people are concerned about a very recent risk they have taken, they may be motivated to test now. If they are asked to wait, the issue may slip from their mind.

Antibody/antigen tests can sometimes detect infection just 10 days after infection, and most infections will be detected within a month. So, the clinic should take an initial test straightaway. If the result is negative, the person will usually be asked to return a few weeks later in order to be re-tested.

How long after taking a risk can I be sure that I am HIV-negative?

Although the majority of infections are detected within a month, there are occasional cases when it takes longer. Following an event that would carry a high risk of HIV infection, [BASHH recommends](#) a test after eight weeks to be sure.

How often should I test?

[BHIVA and BASHH recommend](#) that all gay and bisexual men test for HIV at least once a year, or more often if there is continued risk behaviour. While similar guidance does not exist for African people, [NAT \(National AIDS Trust\)](#) argues that sexually active African people should also test for HIV annually.

Antibody laboratory tests

The HIV tests that were most commonly used in the past tested for HIV antibodies only. A blood sample is taken through a needle from a vein in the arm. Samples from many individuals are analysed at the same time, in a machine at a laboratory. These tests may also be referred to as “third-generation” tests or as an ELISA (enzyme linked immunosorbent assay). The first- and second-generation tests are no longer in use.

HIV antibodies are not part of HIV itself, but are produced by the human body in response to HIV infection. In the weeks after exposure to HIV, the immune system recognises antigens that belong to HIV and begins to generate HIV antibodies. These antibodies persist for life.

The period during which antibodies are first produced is called “seroconversion”. It is frequently, but not always, accompanied by a set of symptoms commonly called a seroconversion illness, which may be misdiagnosed as flu or glandular fever (or ignored). The most common symptoms are fever, rash, sore throat, swollen lymph

nodes, muscle aches and joint pains. When these symptoms appear, they normally do so within six weeks of the HIV exposure.

The “window period” refers to the period after infection with HIV during which tests are not able to detect any HIV antibodies (either because none have been produced yet, or because they are too few in number for the test to pick up). The *typical* time before a third-generation test can detect infection is thought to be between 20 and 25 days, although it can be longer in some cases.

Except in the case of recent infection, third-generation tests are extremely accurate. For example, a [Health Protection Agency evaluation of 16 tests](#) found that all except one had a sensitivity of 100% – in other words, all HIV-positive people tested were correctly diagnosed. Moreover, all had a specificity of 99.8% or over – in other words, if 1000 HIV-negative people were tested, 998 would be correctly diagnosed as such, while two samples would test positive. However in practice, confirmatory tests would be used and individuals would not receive an incorrect positive diagnosis.

	Laboratory tests	Rapid, point-of-care tests	Self-sampling	Self-testing
Available from	Sexual health clinics, other medical settings.	Community settings, some clinics.	Order online.	Not yet available.
Sample	Blood from a vein.	Blood from a finger prick, or oral fluid.	Blood from a finger prick, or oral fluid.	Likely to be oral fluid, or blood from a finger prick.
Speed of result	Within 48 hours, or one to two weeks.	A few minutes.	One to two weeks.	A few minutes.
Most infections detected within	Four weeks (antibody/antigen test), a little longer for antibody only test.	Six to eight weeks.	Varies according to the sample and the test.	Unknown.
Negative result reliable after	Twelve weeks.	Twelve weeks.	Twelve weeks.	Unknown.
How accurate, once the window period is over	Gold standard.	Accurate for most people; slightly poorer performance than laboratory tests, especially when sample is oral fluid.	Accurate for most people; varies according to the sample and the test.	Unknown.
Advantages	The most reliable tests. Can provide STI screen at same time. Easier to implement quality control. Can test large numbers of people.	Instant results. Non-invasive.	Convenience. Privacy. Non-invasive.	Convenience. Privacy. Non-invasive. Instant results.
Disadvantages	Delay in getting results. Clinical settings only.	Inaccurate during recent infection.	Delay in getting results.	Limited support for people testing positive. Maybe inaccurate during recent infection. Risk of abuse.

Antibody/antigen laboratory tests

Antibody only laboratory tests are no longer recommended for routine use in the UK. [UK guidelines](#) recommend the use of tests which detect both HIV antibodies and p24 antigen, otherwise known as “fourth-generation” tests.

An HIV antigen, known as p24, is a structural protein that makes up most of the HIV viral core. High levels of p24 are present in the blood during the short period between HIV infection and seroconversion, before fading away. A fourth-generation HIV test adds a technique for detecting p24 antigen to the traditional antibody test. Otherwise, the test is carried out in the same way, with blood samples at a laboratory.

Since p24 antigen is usually detectable a few days before HIV antibodies, the window period is somewhat reduced. Some people who have been HIV infected but have not yet seroconverted will have their infection diagnosed with this test.

It is hard to say exactly how long the window period for these tests lasts, as there are variations between individuals and it is a difficult topic to research (recently infected people would need to know exactly when they were exposed to HIV and then give multiple blood samples over the following days and weeks). Nonetheless, some experts believe that combined tests usually detect infection approximately 15 to 25 days after exposure, but occasionally this period will be a little longer. The [UK guidelines](#) say that, when this test is used, the majority of infections will be detected within one month.

Antibody/antigen laboratory tests are extremely accurate. In terms of sensitivity (correct identification of people with HIV), [a Health Protection Agency evaluation](#) found that nine out of the ten tests they evaluated had a sensitivity of 100%, while [a French evaluation](#) found that ten of twelve tests had a sensitivity of 100%. The lowest sensitivity was 99.8%.

Similar results were found for specificity, in other words, the ability of a test to correctly give an HIV-negative result. All tests checked by [the Health Protection Agency evaluation](#) had a specificity of 99.7% or above, and [the French study](#) found that all tests produced after the year 2000 had a specificity of 99.8% or above.

Self-sampling

Some charities, sexual health clinics and private companies offer self-sampling services for HIV testing (for example, [Terrence Higgins Trust, 56 Dean Street](#) and [Dr Thom](#)). These generally involve the end-user ordering a self-sampling kit from the organisation’s website, collecting their own blood or oral fluid sample at home, posting it back for laboratory analysis, and receiving the results by phone or text a few days later. *Self-sampling* is different from *self-testing*, discussed later in this briefing.

In most cases, the HIV testing technology used in self-sampling services is an antibody/antigen laboratory test, testing a tiny tube of blood (drawn from a fingerprick). In these cases, the accuracy should therefore be broadly comparable to those tests described above.

However, the window period may be extended, depending on the type of sample used. When the sample is oral fluid (moisture from the gums), the window period is thought to be three months. For a dried blood spot (a drop of blood from the finger dried onto filter paper), the window period may be a few weeks longer than for laboratory tests using venous blood.

Rapid, point-of-care tests

From the point of view of a hospital doctor, the laboratory tests previously described have considerable advantages. They give exceptionally accurate results, processes are automated and quality control can be assured in a laboratory environment. Also, if a test appears to give either a positive result or one that is difficult to interpret, there is plenty of time to carry out additional tests to clarify the diagnosis.

But laboratory tests have some disadvantages, especially from the point of view of people testing. Some people dislike having blood taken with a needle. Getting the results usually requires coming back on another day, something that a lot of people fail to do. Laboratory tests tend to be offered in hospital settings.

“Point-of-care” tests (PoCT) do not require specialised laboratory equipment, so they can be administered and interpreted in any setting. Most point-of-care tests require a tiny sample of blood (the fingertip is pricked with a lancet). Other tests require oral fluid (an absorbent pad is swabbed around the outer gums, adjacent to the teeth).

They are called “rapid” tests because the result can usually be given within 30 minutes. These tests are often used in community settings on the assumption that more people will be willing to test for HIV if they can do so at venues they go to anyway.

However, some doctors and laboratory professionals are wary of using these tests and note inferior performance to antibody/antigen laboratory tests. While several studies have shown point-of-care tests to be almost as accurate as antibody laboratory tests, performance has not always reached these standards.

Rapid tests can be performed by staff with limited laboratory training. However, reading the test result relies on subjective interpretation, and when the result is borderline, experienced staff give more consistently accurate results. Poor results in some studies might be due to problems with staff training or quality control, rather than intrinsic limitations of the tests. But this does highlight real-world difficulties in delivering consistently reliable results. Organisations using point-of-care tests must maintain strong links with a pathology laboratory that provides support with clinical governance and quality assurance.

The [UK testing guidelines – issued by the British HIV Association \(BHIVA\) and the British Association for Sexual Health and HIV \(BASHH\)](#) – are cautious about the use of point-of-care tests, recommending that they are only used:

- At community testing sites.
- In clinical settings where a rapid turnaround of test results is desirable.
- For urgent source testing (for example, following a needlestick injury).
- If a person refuses to give a venous blood sample.

The [guidance from NICE \(the National Institute for Health and Care Excellence\)](#) is more supportive of the use of point-of-care tests, for example in outreach testing programmes in high-prevalence areas and also in venues where high-risk sexual behaviour between men occurs.

When used in a population with a low prevalence of HIV, false-positive results can be a problem. The tests always produce a small number of false positive results, but in a setting where very few people have HIV, the majority of

apparent positive results will in fact be incorrect. However, as the proportion of people with HIV being tested increases, the true positives start to outnumber false positives. This means it is more appropriate to use point-of-care-tests in high-prevalence populations, such as with gay and bisexual men, than in the general population.

As noted above, all HIV tests need to have reactive (“positive”) results confirmed with further tests. Most providers tell people who are testing that a negative result is definitive, but that a reactive result simply indicates the need for further laboratory testing.

Self-testing

Self-testing is often referred to as “home-testing” and involves the end-user carrying out all test procedures. Whereas it used to be illegal to sell or advertise HIV self-testing kits in the United Kingdom, these restrictions were lifted in April 2014. Testing kits that are designed to be used by members of the public can now be sold, provided that the kit carries a CE mark. The CE mark shows that the product meets European requirements for test performance and safety.

So far, no manufacturer has produced an HIV self-testing kit that has a CE mark. At the moment, there aren’t any HIV self-test kits that can be legally sold in the United Kingdom, although this may change soon.

When self-testing kits become available, they are likely to be modified versions of antibody point of care tests. [In the United States, one test has been approved for sale.](#) The *OraQuick* test has been simplified for use by non-professionals, so that it can only test oral fluid (rather than fingerprick blood). A trial found that specificity was 99.98% and sensitivity was 93.0% – lower than for the professional version, probably due to more user errors. Moreover, this is an antibody test and so is unable to detect recent infection. However, these limitations may be counter-balanced by the potential of self-testing to improve the uptake of HIV testing.

Antibody point-of-care tests

A wide range of point-of-care tests have been manufactured in many countries, but only a few of them have been subject to rigorous, independent evaluations, and even fewer are marketed in the UK. Research on HIV tests is only occasionally published in medical journals.

Informally, laboratory professionals may have insights into which tests perform best.

With the exception of the *Determine HIV-1/2 Ag/Ab Combo* test, all point-of-care tests look for antibodies only. This explains, in part, the scepticism of some health professionals. Moreover, the window periods of these tests are usually a few days longer than those of antibody laboratory tests.

It is important to verify that any test used is CE marked. This should mean that the test conforms to European health and safety legislation, although it does not necessarily mean that test performance has been independently evaluated.

There are wide variations in accuracy from one test to another. Some older tests, mostly ones which are not marketed in the UK, have a sensitivity (the ability to detect all true positive results) or specificity (the number of negative samples correctly identified as negative) of 95-97%, rather than 99-100%. However, the evaluation data that are available for the tests more commonly used in the UK have generally been more encouraging.

In most but not all studies of the *Determine HIV 1/2*, *INSTI* and the *Vikia HIV 1/2* tests, they performed well, usually with sensitivities and specificities in the range of 99-100%.

Performance of the *OraQuick* test with fingerprick blood samples has generally been good, with sensitivity and specificity in the range of 99-100%. However, performance is slightly poorer when testing samples of oral fluid – some people with HIV may receive a false negative result. This may be because quantities of antibodies are lower in oral fluid than in blood.

Antibody/antigen point-of-care test

Introduced in 2009, the *Determine HIV-1/2 Ag/Ab Combo* test looks for both antibodies and p24 antigen, in a similar way to antibody/antigen laboratory tests. At the time of writing, it is the only point-of-care test to do so.

Because it detects p24 antigen as well as antibodies, the window period should be reduced. The manufacturer says the window period is an average of five days shorter than for the previous *Determine* test, but this varies from individual to individual (range: 2 to 20 days). The manufacturer also reports that on tests with 1179 positive

and 2343 negative samples, sensitivity was 100% and specificity was 99.2%.

However, other research suggests that while the test performs well in respect of established HIV infection, its ability to detect recent HIV infection does not match that of laboratory antibody / antigen tests. Clinicians in London, Sydney and San Francisco have each reported that for people with either acute or recent infection, the test is only able to detect between 50 and 90% of infections. Whereas *Determine* has excellent detection of antibodies, it frequently fails to identify p24 antigen that can be detected with laboratory tests. Studies from Malawi, Zambia and Rwanda have shown particularly low rates of antigen detection, suggesting problems with sensitivity to the range of HIV-1 subtypes that are found in African people living in the UK. For example, of 34 people with acute infection in Zambia and Rwanda, the rapid test detected p24 antigen for only one person (sensitivity 3%) and detected HIV infection (either via p24 antigen or antibodies) in only eight cases (sensitivity 23.5%).

Recent infection testing algorithm (RITA)

Individuals who are newly diagnosed with HIV may also have their blood tested by the RITA method. This is a laboratory technique which aims to distinguish between recent and more established HIV infection.

RITA depends on looking for specific antibody markers, which give different results in the months following infection. If a test gives a result below a pre-determined cut-off point, it is deemed to be an infection that probably occurred in the last six months.

RITA was designed to help public health officials monitor the number of new HIV infections in a population, in order to better inform HIV prevention work. Because of person-to-person variability in the development of immune response, the tests are seen as being unable to give a definitive date for an individual's infection. They are only able to suggest rough timings, and have a considerable margin of error.

RITA is more reliable with the HIV subtype (B) that is most commonly found in Europe. With subtypes that are more commonly found in Africa and elsewhere, its results may sometimes be inaccurate. Moreover, some people may be misclassified as having recent infection when they have a low CD4 cell count or when they have taken antiretroviral drugs, either as treatment, post-

exposure prophylaxis (PEP) or pre-exposure prophylaxis (PrEP).

Blood for RITA may be taken alongside samples needed for viral load testing, CD4 counts and other tests. The test is done at a Public Health England laboratory. The results are returned to the HIV clinician, who decides whether to discuss them with the patient. Clinicians are encouraged to explain the limitations of the test and to present the results in the context of the patient's clinical history and recent sexual behaviour.

Key points

- UK guidelines recommend the use of combined antibody/antigen laboratory tests. Antibody-only tests are no longer recommended.
- Combined antibody/antigen laboratory tests are exceptionally accurate and usually able to detect infection within a month after exposure.
- Point-of-care (rapid) tests, self-testing and self-sampling services are more convenient for some users. Improving the availability of these services may lead to more people receiving HIV test results.
- There are limitations to the performance of some of the more convenient testing methods – they do not always reach the standards of combined laboratory tests.
- An HIV-positive diagnosis should never be given on the basis of a single test result – confirmatory tests are always required.

Further reading

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