Performance of a Rapid Immunochromatographic Screening Test for Detection of Antibodies to Human Immunodeficiency Virus Type 1 (HIV-1) and HIV-2: Experience at a Tertiary Care Hospital in South India

Theophilus-Sunder Vijayakumar, Shoba David, Kavitha Selvaraj, Thiruppavai Viswanathan, Rajesh Kannangai, and Gopalan Sridharan*

Department of Clinical Virology, Christian Medical College, Vellore, Tamil Nadu PIN 632004, India

Received 16 March 2005/Returned for modification 9 May 2005/Accepted 16 May 2005

The performance characteristics of a rapid immunochromatographic-screening test, SD Bioline HIV-1/2 3.0 (Standard Diagnostics Inc., Kyonggi-do, South Korea) on 23,754 sera and 30 plasma samples are reported. The sensitivity and specificity for the assay on serum samples are 100% and 99.4%, respectively. The assay detected antibodies in individuals infected with human immunodeficiency virus type 1 (HIV-1) genotypes A and C and HIV-2. This straightforward assay is a reliable diagnostic tool for screening HIV in resource-poor settings.

The human immunodeficiency virus (HIV) pandemic has come to stay. Attempts to curtail this infection have had some success, with diagnosis of infection and counseling playing a significant role in prevention and patient management. Detection of anti-HIV antibodies remains the mainstay for diagnosis of HIV infection, though molecular tests have their application under certain circumstances. Rapid screening assays are simple to perform, give easy visual readout of results, and do not require any equipment, as needed for enzyme-linked immunosorbent assay (ELISA), and batch testing is not required. Presumptive positive results known during a patient’s visit to a voluntary counseling and testing center (VCTC) can lead to early counseling to ensure risk-reducing behaviors, etc.

We had earlier evaluated SD Bioline HIV-1/2 3.0 (Standard Diagnostics Inc., Kyonggi-do, South Korea) using a panel of 100 sera (9) and have been using this assay since November 2002. We report here the real-time performance characteristics of this assay observed during a period of 23 months. One of the vital aspects of the performance characteristics of a kit, in addition to the accuracy indices, is its capability to detect locally prevalent types and subtypes (2, 8, 11, 14). In India the predominant subtypes are HIV type 1 (HIV-1) subtype C (13) and HIV-2 subtype A (10), and so testing this assay’s usefulness here was important. This assay needed to be evaluated against the ELISA, which is not available in small medical establishments and VCTCs in Asia and Africa.

HIV testing is done in our institution, Christian Medical College, Vellore, India, a tertiary care hospital in southern India, primarily to facilitate HIV infection management. General consent for all tests including blood tests is obtained when needed. No treatment/procedure is denied to anyone if found HIV infected, and patients are referred routinely to the infectious disease clinic in our hospital for counseling and follow-up.

A total of 23,754 serum samples from as many individuals received by our department for rapid screening for anti-HIV antibodies from November 2002 to September 2004 in situations like emergency invasive procedures/surgeries, labor room procedures for women whose HIV status is not known, dialysis, availability of a cadaver organ donor, needle stick injury, etc., were tested using SD BIOLINE HIV-1/2 3.0, followed by ELISA. All rapid-test-screened samples were further tested by ELISA, and the ELISA result is given as the final result for a given sample. All readings were taken by highly trained technicians without prior knowledge of the HIV status of the sample. A total of 90,000 HIV antibody ELISA tests were done, including rapid test follow-up in the 2-year study period reported here.

Fourteen plasma samples from HIV-2-infected individuals confirmed by an in-house HIV-2-specific ELISA (7) and by nested PCR (5) and 2 plasma samples with dual infection confirmed by immunoblot and nested PCR were tested by SD BIOLINE HIV-1/2 3.0 for the capability to detect HIV-2. Thirteen plasma samples from individuals positive for HIV-1 subtype C and one sample from an individual positive for HIV-1 subtype A were also tested to investigate the ability to detect locally prevalent subtypes.

SD Bioline HIV-1/2 3.0 (Standard Diagnostics, Kyonggi-do, South Korea) is an immunochromatographic test for the qualitative detection of antibodies of all isotypes (immunoglobulin G [IgG], IgM, and IgA) specific to HIV-1 and HIV-2 simultaneously, in human serum, plasma, or whole blood. Briefly the procedure of the assay consists of addition of 10 μl of the serum/plasma to the sample well of the membrane test assembled followed by 3 or 4 drops of assay diluent from the reagent dropper bottle. The test is read at an outer limit of 20 min. A colored band should appear in the region marked as “C” (control) in order for the test to be valid. In addition, for positive samples a band should be present in the result window marked “1” (for HIV-1) or “2” (for HIV-2) or both (dual infection).
TABLE 1. Performance characteristics of SD Bioline HIV-1/2 3.0 compared with ELISA results on 23,754 samples

<table>
<thead>
<tr>
<th>SD Bioline test result</th>
<th>No. of samples with ELISA result of:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Reactive</td>
</tr>
<tr>
<td>Rapid reactive</td>
<td>564</td>
</tr>
<tr>
<td>Rapid negative</td>
<td>23,190</td>
</tr>
</tbody>
</table>

* The results of 13 samples were inconclusive and hence were not included in the analysis.

The algorithm used for testing samples in this study has been described earlier (6). All samples tested by SD Bioline HIV-1/2 3.0 were also tested by ELISA screening. Samples found negative by rapid test and the ELISA were declared “negative.” Samples found to be reactive in the rapid test were tested in duplicate wells by two independent ELISAs, and the ELISA-concordant results were given as the final results (either “negative” or “ELISA reactive”). Any discordance in results between the two ELISAs was resolved by a confirmatory test (Western blot/immunoblot). All tests were performed by strictly adhering to the instructions of the manufacturers, and the kits were on the approved list of kits of the World Health Organization/UNAIDS program. The accuracy indices such as sensitivity, specificity, negative predictive value, and positive predictive value were calculated using Epi-info software, version 6.04d (January 2001).

Of the 23,754 samples, 23,190 were declared rapid negative and 564 samples were declared rapid reactive. On further ELISA testing, all of the 23,190 rapid-negative samples were found negative and declared so. The final results for 13 of the rapid-reactive samples were inconclusive and hence excluded from analysis. HIV RNA testing could not be done on these 13 samples, as only refrigerated serum samples were available. Of the remaining 551 rapid-reactive sera, 406 were declared ELISA reactive and 145 were declared negative. A summary of the findings and the accuracy indices are shown in Tables 1 and 2. The capability to detect locally prevalent types and subtypes was also evaluated. All the 30 plasma samples, i.e., 14 from individuals infected with HIV-1 only, 14 from individuals infected with HIV-2 only, and 2 from individuals infected with both HIV-1 and HIV-2, gave concordant results.

Our laboratory has had more than 16 years of experience in HIV care, ever since we diagnosed the first HIV-1-infected individuals in our state (3, 14) and neighboring states and HIV-2-infected individuals (1, 4). SD Bioline HIV-1/2 3.0 (Standard Diagnostics, Inc.) is a simple test, easy to perform and interpret, where the results are known within 15 to 20 min, making it a useful assay for on-site testing and rapid screening. It is a third-generation kit, which can detect both IgG and IgM, thereby considerably reducing the window period for serodiagnosis. The test can also discriminate between HIV-1 and HIV-2 infection. The temperature range for kit storage, 2 to 30°C, is an advantageous feature, facilitating easy transport to field sites. Cold storage has to be maintained during the warm and hot months in tropical countries like India for best results.

The data presented here clearly show the reliability of the kit as a screening test. The sensitivity and specificity of the kit are very high and satisfactory for screening purposes. The negative predictive value of 100%, with a sample size of 23,754, strongly favors the reliability of the assay in situations warranting quick clinical decision making, such as needle stick injury while testing index cases, HIV-infected women during delivery, and corneal and other transplants from cadaver to donor, etc. The positive predictive value is slightly low (73.7%) compared to other rapid tests like HIV Tridot and HIV spot (6) but is better than Capillus HIV [1/2], another third-generation rapid (particulate agglutination) test (12). Both these evaluations were carried out in a low-risk population, probably contributing to the low positive predictive value. Since SD Bioline is only a screening test, all positives have to be retested using another screening test and the concordant positivity in both screening tests has to be confirmed by a supplemental/confirmatory test before diagnosing a person as truly infected with HIV. Thus, the lower positive predictive value would not be a major disadvantage in clinical situations, as positive results will be confirmed as described above, before informing the individual, with appropriate counseling. Since the negative predictive value for this test is 100% as we saw in this extensive real-time evaluation study, in poor-resource settings the negative samples need not be repeat tested by an ELISA, and this will reduce the cost of testing to about U.S. $2.5.

Information on seroconversion dates is not available for our HIV-infected population. Such information is generally lacking in the country. The National AIDS Research Institute (Pune, India) now maintains a cohort of high-risk individuals for HIV vaccine evaluation. Seroconversion panels are very expensive to obtain, so it must be stated that the present evaluation has been done on samples obtained from individuals with established HIV infection. However, this does not detract the usefulness of the test in situations described earlier.

In conclusion, we find that SD Bioline HIV-1/2 3.0 is a rapid test, easy to perform and interpret, with very satisfactory accuracy indices. The demonstrated capability of detecting and distinguishing HIV-1 and HIV-2 infections and a 100% negative predictive value make this assay a valuable tool, especially in resource-poor settings and in VCTCs.

TABLE 2. Accuracy indices of SD Bioline HIV-1/2 3.0

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Indexa</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sensitivity</td>
<td>100 (97.4–100)</td>
</tr>
<tr>
<td>Specificity</td>
<td>99.4 (99.3–99.6)</td>
</tr>
<tr>
<td>Negative predictive value</td>
<td>100 (100–100)</td>
</tr>
<tr>
<td>Positive predictive value</td>
<td>73.7 (69.2–80.6)</td>
</tr>
</tbody>
</table>

a Indices are derived from data shown in Table 1. Values are percentages, calculated as follows: sensitivity = [TP/(TP + FN)] × 100, where TP is the number of true positives and FN is the number of false negatives; specificity = [TN/(TN + FP)] × 100, where TN is the number of true negatives and FP is the number of false positives; positive predictive value = [TP/(TP + FP)] × 100; negative predictive value = [TN/(TN + FN)] × 100. Values in parentheses are 95% confidence intervals.

REFERENCES