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Correspondence

Sensitivity & specificity of combination testing algorithms for HIV in a tuberculosis clinic

Sir,

Expanding voluntary counselling and testing for HIV in a variety of health care settings calls for a simple and effective testing strategy. Several rapid and easy to perform non ELISA assays demonstrating a good level of sensitivity and specificity are available to circumvent the limitations of using conventional ELISA and Western blot assays[1,2]. A major advantage of rapid tests is the ability to use them in serial and parallel testing algorithms on a single specimen collected from the patient. However, parallel testing algorithms (where two tests are performed simultaneously) are about 2.5 times higher in terms of cost when compared to that of serial testing[3]. Hence, parallel testing is generally not recommended. We evaluated the sensitivity, specificity and predictive value of three indigenous rapid and three ELISA kits in a two step testing algorithm that uses a combination of two different tests in series. The predictive values at HIV prevalence rates of 1, 5 and 20 per cent in the population were calculated.

The study was performed between 2005 and 2006 with 150 known HIV-positive and 150 HIV-negative serum samples obtained from individuals attending the HIV/TB clinic at Tuberculosis Research Centre (TRC), Chennai, India. The study was approved by the Ethics Committee of the centre. Written informed consent was obtained from all study participants. HIV positivity was confirmed using the HIV Western Blot kit (J. Mitra & Co., New Delhi, India) having high performance characteristics (100% sensitivity & 100% specificity) similar to the FDA approved kit (HIV Blot 2.2, Gene labs, Singapore)[4]. Six aliquots of each serum sample were prepared, coded and randomized. The study was performed in a blinded fashion. The samples were tested with three indigenous HIV rapid test kits, namely, Combaids-RS HIV-1/HIV-2 (Span Diagnostics, Surat, India), HIV Tridot (J. Mitra & Co.) and Pareekshak HIV 1/2 Spot (Bhatt Biotech, Bangalore, India), and three indigenous ELISA kits, namely, Enzaids HIV 1+2 ELISA (Span Diagnostics), Microlisa HIV 1/2 (J. Mitra & Co.) and Pareekshak HIV 1/2 ELISA (Bhatt Biotech), frequently used in private and government laboratories in India, both singly and in series. A sample was considered positive if it was positive in both the tests. A negative sample by the first test was not retested.

The sensitivity and specificity of the rapid tests used ranged from 98-100 per cent and 96-100 per cent respectively, while the sensitivity and specificity of the ELISA kits employed ranged from 99-100 per cent and 93-100 per cent respectively. False positivity in ELISA may occur in a number of conditions including TB, rheumatoid arthritis and leprosy because of cross-reactive antibodies[5]. Though specificity of ELISA kits have improved over time, rapid tests used in this study performed better, as reported by others[6-8]. Although two of the kits used (HIV-1 Tridot and Microlisa HIV 1/2) demonstrated 100 per cent sensitivity and specificity, the performance in the field on large number of samples could be different, and therefore, it would be desirable to confirm a positive result with a second test before reporting HIV positivity. All the rapid-rapid combinations performed very well (100% specificity and 96-100% sensitivity) as did some of the ELISA-rapid combinations tested (Table).

We determined the impact of the different pre-test probabilities of HIV prevalence on the performance of these assays. The positive and the negative predictive values (PPV and NPV) of individual kits were determined for an HIV prevalence rate of 1 per cent (seen in many districts of India in the general population)[9], and 5-20 per cent (observed among tuberculosis patients)[10,11]. The clinical utility of PPV

Table. Sensitivity, specificity and predictive values of ELISA/rapid tests singly and in series

Assay		Sensitivity % (95% CI)	Specificity % (95% CI)	PPV for prevalence rate of 1%	PPV for prevalence rate of 5%	PPV for prevalence rate of 20%
Test 1	Test 2					
HIV Tridot		100	100	100	100	100
Combaids-RS HIV-1/HIV-2		98 (95.8 -100)	96 (92.8-99.1)	19.8	56.3	86
Pareekshak HIV-1/2 Spot		98.6 (96.7-100)	100	100	100	100
Microlisa HIV 1/2		100	100	100	100	100
Enzaids HIV 1+2 ELISA		100	93.3 (89.2-97.3)	13.1	44	78.9
Pareekshak HIV-1/2 ELISA		99.1 (97.5-100)	96.2 (93.1-99.3)	20.8	57.8	86.7
Microlisa HIV 1/2	HIV Tridot	100	100	100	100	100
Microlisa HIV 1/2	Combaids-RS HIV-1/HIV-2	98 (95.7-100)	100	100	100	100
Microlisa HIV 1/2	Pareekshak HIV-1/2 Spot	98.6 (96.7-100)	100	100	100	100
Enzaids HIV 1+2 ELISA	HIV Tridot	100	100	100	100	100
Enzaids HIV 1+2 ELISA	Combaids-RS HIV-1/HIV-2	98 (95.7-100)	99.8 (99.1-100)	83.2	96.3	99.2
Enzaids HIV 1+2 ELISA	Pareekshak HIV-1/2 Spot	98.6 (96.7-100)	100	100	100	100
Pareekshak HIV-1/2 ELISA	HIV Tridot	99.1 (97.5-100)	100	100	100	100
Pareekshak HIV-1/2 ELISA	Combaids-RS HIV-1/HIV-2	97 (94.2-99.7)	99.9 (99.3-100)	90.7	98.1	99.6
Pareekshak HIV-1/2 ELISA	Pareekshak HIV-1/2 Spot	97.6 (94.5-100)	100	100	100	100
HIV Tridot	Combaids-RS HIV-1/HIV-2	98 (95.8-100)	100	100	100	100
HIV Tridot	Pareekshak HIV-1/2 Spot	98.6 (96.7-100)	100	100	100	100
Combaids-RS HIV-1/HIV-2	Pareekshak HIV-1/2 Spot	96.6 (93.7-99.5)	100	100	100	100

PPV, Positive predictive value; C.I, Confidence Interval

is its ability to determine the likelihood that a positive rapid/ELISA test is a true positive. The negative predictive values were found to be close to 100 per cent for each of the kits but the positive predictive values were variable across different prevalence rates and were lower for ELISA compared to rapid tests. However, most of the serial combinations evaluated demonstrated high sensitivity, specificity and PPV.

To conclude, we identified several two step rapid HIV testing algorithms that provide good cumulative sensitivity (96.6-98.6%) and specificity (100%), and high positive and negative predictive values. These simple serial algorithms may be used to obtain HIV test results the same day and are therefore ideal for use in settings like TB clinics or Primary Health Centers in resource-poor countries. Besides providing quick and accurate diagnosis for HIV infection, the strategy offers cost savings over parallel testing algorithms as well as over algorithms that recommend three tests in series. The wider use of rapid tests could play a major role in reducing drop-outs from post-test counselling by providing results on the same day and thus improve the efficiency of HIV testing centres.

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Soumya Swaminathan[*‡], L. Antony[*] P. Venkatesan[**], Luke Elizabeth Hanna[*] B. Angayarkanni[*], C. Ponnuraja[**] Jim Robin[*], K. Lucia Precilla[*] & Ranjani Ramachandran[†]

[*]Division of HIV/AIDS

[**]Departments of Statistics &

[†]Bacteriology, Tuberculosis Research Centre

Mayor V.R. Ramanathan Road, Chetput

Chennai 600 031, India

[‡]*For correspondence*:

doctorsoumya@yahoo.com

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