INT J TUBERC LUNG DIS 6(7):635–640
© 2002 IUATLD

Evaluation of the *FASTPlaque*TB assay for direct detection of *Mycobacterium tuberculosis* in sputum specimens

R. Muzaffar,* S. Batool,* F. Aziz,† A. Naqvi,‡ A. Rizvi‡

* Laboratory of Molecular Biology & Immunology, Sindh Institute of Urology and Transplantation (SIUT), Dow Medical College and Civil Hospital, Karachi, Pakistan; † VML Laboratories, Toronto, Ontario, Canada; ‡ Department of Urology, Sindh Institute of Urology and Transplantation (SIUT), Dow Medical College and Civil Hospital, Karachi, Pakistan

SUMMARY

SETTING: Sputum samples were collected from suspected tuberculosis patients attending out-patient clinics at the Ojha Institute of Chest Diseases, Karachi, Pakistan.

OBJECTIVE: To evaluate the performance of the *FASTPlaque*TB assay for rapid diagnosis of pulmonary tuberculosis.

DESIGN: A comparative study of 584 sputum samples using acid-fast smear microscopy, Löwenstein-Jensen culture and *FASTPlaque*TB.

RESULTS: A total of 514 samples yielded complete results. Seventy specimens were lost to analysis due to the overgrowth of contaminants. The addition of antimicrobials inhibited growth of gram-positive contaminants, and reduced the overall contamination rate from 18.2% to 7.2%. *Mycobacterium tuberculosis* was isolated from 175 smear-positive and 70 smear-negative specimens. *FASTPlaque*TB detected *M. tuberculosis* in 81.6% of specimens, with a specificity of 97.7%. The sensitivity and specificity of the assay for smear-positive specimens were respectively 87.4% and 88.2%. For smear-negative specimens, the sensitivity of the assay was 67.1% and the specificity was 98.4%. The combined sensitivity of smear and *FASTPlaque*TB for *M. tuberculosis* was 90%. Test results were available in 48 hours.

CONCLUSION: *FASTPlaque*TB is a sensitive and specific test for rapid diagnosis of pulmonary tuberculosis in high prevalence areas. The test is sensitive enough to confirm a large number of clinically suspected smear-negative cases and improve case finding.

KEY WORDS: smear-negative; tuberculosis; *FASTPlaque*; Pakistan

PAKISTAN is a low-income area with a high prevalence of tuberculosis (TB).[1] Microscopic examination of direct smears for the presence of acid-fast bacilli (AFB) in the sputum is the most commonly used method for diagnosis. Infection due to non-tuberculous mycobacteria (NTM) is not common and the specificity of smear remains high. But the method lacks sensitivity, requiring $>10^3$–10^4 organisms per ml of specimen to be present for the smear to be positive.[2]

It is believed that only one half to three quarters of active TB cases investigated are positive on smear microscopy.[3] According to World Health Organization (WHO) estimates, currently less than 20% of the roughly 8 million predicted annual cases of TB are identified as smear-positive.[4] Thus, when smear microscopy is the only diagnostic test used, a significant number of infected cases are likely to be missed. In Pakistan, which has an overall tuberculosis incidence of 181 per 100 000 population, 55% of expected TB cases are smear-negative.[1] Many of these patients are not likely to be investigated further if clinical and radiological findings are not clearly suggestive of TB. Although these patients are considered to be less infectious than smear-positive cases, they have nevertheless been shown to transmit disease.[5]

There is clearly a great need for rapid tests that can accurately detect smear-negative TB cases. In recent years, rapid nucleic acid amplification-based assays have been successfully used for diagnosing TB.[6,7] Many of these tests are too technically demanding and expensive to be used in routine diagnostic laboratories in developing countries.

The aim of this study was to evaluate the performance and utility of the *FASTPlaque*TB assay (Biotec Laboratories Ltd, Ipswich, UK) for rapid and specific detection of *Mycobacterium tuberculosis* in sputum samples in a laboratory in Pakistan. The *FASTPlaque*TB assay is based on phage amplification technology, in which bacteriophage D29 is used as an indicator to detect the presence of viable tubercle bacilli in sputum samples.[8–11] *M. tuberculosis* present in decontaminated sputum is infected with the phage (Actiphage).

Correspondence to: Dr Rana Muzaffar, PhD, Laboratory of Molecular Biology & Immunology, Sindh Institute of Urology and Transplantation (SIUT), Dow Medical College and Civil Hospital, Baba-e-Urdu Road, Karachi-74200, Pakistan. Tel: (+9221) 921 5752, 921 5718. Fax: (+9221) 921 5469. e-mails: rmuzaffar@global.net.pk, mblsiut@yahoo.com

Article submitted 17 November 2001. Final version accepted 5 April 2002.

After infection, addition of a virucide (Virusol) results in destruction of residual extra-cellular phage. The phage replication in infected cells results in cell lysis and release of progeny. Addition of a rapid growing *Mycobacterium* strain, *M. smegmatis* (Sensor cells), which is also sensitive to the phage, results in amplification of the progeny which can be visualised as zones of clearing (plaques) in a lawn of Sensor cells. Results are available in 48 hours. The results of the assay were compared with findings on conventional culture, which was used as the 'gold standard'.

METHODS AND MATERIALS

Single sputum samples were collected from 584 patients attending the out-patient clinics at the Ojha Institute of Chest Diseases with previously undiagnosed tuberculosis. The Ojha Institute is a public sector hospital serving a very large and diverse TB patient population of Karachi and the adjoining areas of Sindh Province. Specimens were collected between June 2000 and May 2001. Samples were processed by the standard N-acetyl-L-cysteine-NaOH method and concentrated by centrifugation at 3000 × g for 15 min. Sediment was resuspended in 15 ml of *FASTPlaque*TB (FPTB) medium and centrifuged again at 3000 × g for 15 min. The supernatant was discarded, and the pellet (usually 0.5 to 1 ml) was resuspended in 1 ml of FPTB medium. One ml of the suspension was transferred to a FPTB reaction tube and the remainder used to prepare smears (Ziehl-Neelsen acid-fast staining) and for culture isolation. Duplicate Löwenstein-Jensen (LJ) slopes were inoculated with 0.2 ml of suspension and incubated for 10 weeks at 37°C. Cultures were confirmed by p-nitro benzoic acid (p-NBA) testing[12] and a polymerase chain reaction (PCR) assay specific for *M. tuberculosis* complex.[13]

The *FASTPlaque*TB assay was performed according to the manufacturer's instructions. Resuspended sediment was incubated overnight at 37°C. Next morning 100 μl of phage reagent was added followed by 1 h incubation at 37°C. One hundred μl of the virucide was added to inactivate exogenous phage, followed by the addition of 5 ml of FPTB medium to neutralise the virucide. Next, 1 ml of Sensor cells was added. Cell suspension was mixed with 5 ml of molten FPTB agar and plated in Petri dishes. Plates were incubated at 37°C for 18–24 h, and the number of plaques (zones of clearing in the lawn of Sensor cell growth) was recorded by eye.

Quality control of the assay was assured by including positive and negative controls with every batch of specimens tested, as described in the assay protocol. Positive control (PC) consisted of Sensor cells infected with phage and treated with the virucide, resulting in 20–300 plaques. Negative control (NC) consisted of FPTB medium plus Actiphage only, treated with the virucide. To monitor for cross-contamination during specimen processing, sterile water samples (batch controls) were processed along with test specimens as negative controls. Results were considered valid only if the PC values were within the specified range, the number of plaques on NC plate was less than 10, and batch controls were negative.

The first 219 samples were tested without the addition of any antimicrobial agent to either the FPTB or LJ media. For the next 169 samples, the test was performed with the addition of penicillin (final concentration 50 μg/ml) in both the FPTB medium used for overnight resuscitation and the LJ medium to control the growth of contaminants. Forty-eight samples were split and tested in parallel, with both penicillin and Microclens, an anti-microbial agent provided by the manufacturer. The next 148 samples were tested with only Microclens, and cultures were isolated on LJ slopes containing penicillin.

RESULTS

We evaluated the performance of the *FASTPlaque*TB assay by comparing the assay results with smear and culture findings. A total of 584 sputum samples were tested. Results for 70 specimens were lost to analysis due to the overgrowth of contaminants either on assay plates or on LJ slopes. Table 1 shows the overall *FASTPlaque*TB and culture results for the remaining 514 samples. Of these, 245 (47.6%) were positive by culture. Analysis by p-NBA testing and PCR confirmed these as *M. tuberculosis*. For all specimens tested, *FASTPlaque*TB had a sensitivity of 81.6% and a specificity of 97.7% (Table 1).

We then looked at the correlation of *FASTPlaque*TB results with AFB smear and culture findings. As shown in Table 2, 192 (37.3%) of the specimens tested were smear-positive. Of these, 175 (91%) were positive by culture. The sensitivity of the *FASTPlaque*TB assay for these specimens was 87.4%, and the specificity was 88.2%. Of the 322 specimens that were smear-negative, 70 (21.7%) were culture-positive. The sensitivity and specificity of the assay for smear-negative specimens were respectively 67.1% and 98.4%.

Forty-five specimens were *FASTPlaque*TB-negative and culture-positive. Of these, 22 (11.44%) were smear-positive. Eighteen of these specimens had suffi-

Table 1 Performance of *FASTPlaque*TB for all specimens compared to LJ culture (n = 514)

FPTB assay	Culture +ve	Culture −ve	Sensitivity %	Specificity %	PPV	NPV
FPTB +ve	200	6	81.6	97.7	0.97	0.85
FPTB −ve	45	263				
Total	245	269				

FPTB = *FASTPlaque*TB; LJ = Löwenstein Jensen; PPV = positive predictive value; NPV = negative predictive value.

Table 2 Comparison of *FASTPlaque*TB results with AFB smear and LJ culture findings (n = 514)

	Culture +ve	Culture −ve	Sensitivity %	Specificity %	PPV	NPV
Smear positive (n = 192)						
FPTB +ve	153	2	87.4	88.2	0.99	0.40
FPTB −ve	22	15				
Smear negative (n = 322)						
FPTB +ve	47	4	67.1	98.4	0.92	0.91
FPTB −ve	23	248				

AFB = acid-fast bacilli; LJ = Löwenstein Jensen; PPV = positive predictive value; NPV = negative predictive value FPTB = *FASTPlaque*TB.

cient AFB in the smears (4+ to 2+), while the remaining four were of ± category (low AFB numbers). Cultures from 17 specimens were pure *M. tuberculosis* isolates, five cultures were *M. tuberculosis* mixed with NTM (four were identified as *M. gordonae* and one remained unidentified). *FASTPlaque*TB assay was then repeated on pure *M. tuberculosis* cultures obtained from these 22 specimens and also for the five NTM isolates. Assay was positive for all *M. tuberculosis* isolates, while no plaques were seen on the NTM assay plates. No other *Mycobacterium* species were isolated from the 23 smear-negative specimens, and all cultures were identified as *M. tuberculosis*. These 45 results were considered *FASTPlaque*TB false-negatives.

Fifteen (7.8%) of the 192 smear-positive specimens tested *FASTPlaque*TB-negative and culture-negative. Of these, 11 specimens were ± to 1+, and four were 2+ to 3+ on microscopy. No other mycobacteria grew from these specimens, and these results were considered as true negatives.

There were six (2.9%) specimens that were *FASTPlaque*TB positive and culture-negative. Of these, two were smear-positive and four were smear-negative. The smear-positive specimens had less than 10 AFB per 300 fields by ZN staining. For one smear-negative specimen, two colonies of *M. gordonae* were recorded only on a single LJ slope. The *FASTPlaque*TB assay performed on this isolate was negative. These results were considered as *FASTPlaque*TB false-positives.

Results were analysed to determine the sensitivity of smear and/or *FASTPlaque*TB for culture findings. As shown in Table 3, the sensitivity of smear with respect to a positive culture was 71.4%, and specificity was 93.6%. When positive results for smear and *FASTPlaque*TB were combined to predict a positive culture, the sensitivity was 90.6% (Table 4).

Table 3 Comparison of AFB smear performance with LJ culture (n = 514)

Smear	Culture +ve	Culture −ve	Sensitivity %	Specificity %	PPV	NPV
Positive	175	17	71.4	93.6	0.91	0.78
Negative	70	252				
Total	245	269				

AFB = acid-fast bacilli; LJ = Löwenstein Jensen; PPV = positive predictive value; NPV = negative predictive value.

Table 4 Analysis of combined *FASTPlaque*TB and AFB smear results with LJ culture (n = 514)

FPTB and/or smear	Culture +ve	Culture −ve	Sensitivity %	Specificity %	PPV	NPV
FPTB and/or smear +ve	222	21	90.6	92.1	0.91	0.91
FPTB and/or smear −ve	23	248				

AFB = acid-fast bacilli; LJ = Löwenstein Jensen; FPTB = *FASTPlaque*TB; PPV = positive predictive value; NPV = negative predictive value.

There were 70 specimen results lost to analysis due to the overgrowth of contaminants on assay plates and/or on LJ slopes. In the early part of the study, the assay was performed without the addition of any antimicrobial agent to the FPTB or LJ media. Of the 219 samples tested in this manner, 40 (18.26%) were lost due to contamination. We analysed the contaminants on the assay plates by gram-staining, and found the majority to be gram-positive rods and cocci. It appeared that the contaminating bacteria were multiplying during the overnight resuscitation step of the assay and were therefore growing on the assay plates also. To control this problem, modification was made in the assay protocol; penicillin was added to both the FPTB and LJ media, and 169 samples were tested accordingly. Addition of penicillin inhibited the growth of gram-positive contaminants, and decreased the contamination rate from 18.26% to 5.3%. The remaining contaminants were identified as gram-negative rods, mainly *Pseudomonas* species. Forty-eight samples were split and tested in parallel with penicillin and Microclens. Microclens was equally effective in inhibiting the growth of gram-positive contaminants, but also failed to inhibit growth of gram-negatives. The final 148 specimens were then tested with the addition of Microclens only. Of these, 14 (9.4%) specimens were lost to analysis due to the overgrowth of gram-negative bacilli. These were identified as *Pseudomonas* and *Serratia* species resistant to N-acetyl-L-cysteine-NaOH decontamination. As shown in Table 5, the addition of antimicrobials did not have an adverse effect on assay performance, and the sensitivity and specificity values were comparable when performed with or without the addition of antimicrobials.

DISCUSSION

The ability to rapidly detect *M. tuberculosis* in sputum specimens has important implications in the treatment of pulmonary TB. In Pakistan, laboratory diagnosis of TB is mostly based on direct smear-microscopy results. Treatment decisions are generally based on smear, clinical and radiological findings.

Table 5 Assay performance with and without antimicrobials

Specimens	Culture +ve	Culture −ve	Sensitivity %	Specificity %	PPV	NPV
Without antimicrobial (n = 179)*						
FPTB +ve	91	0	79.1	100	1.0	0.72
FPTB −ve	24	64	.			
With penicillin (n = 160)†						
FPTB +ve	47	3				
FPTB −ve	13	97	78.3	97.0	0.94	0.88
With microclens (n = 134)‡						
FPTB +ve	45	2	84.9	97.5	0.95	0.90
FPTB −ve	8	79				

* 219 samples tested, 40 lost.
† 169 samples tested, 9 lost.
‡ 148 samples tested, 14 lost.
PPV = positive predictive value; NPV = negative predictive value; FPTB = *FASTPlaque*TB.

These indicators are well characterised for patients with smear-positive pulmonary TB, the most infectious form of the disease.[14] These predicting characteristics are not as well defined in smear-negative patients, as they may have clinical and radiological findings different from those with AFB-positive smears.[15] Thus, a significant number of active smear-negative TB cases are likely to be missed even when clinical suspicion is high. For these reasons, there is an increasing interest in using new techniques that can rapidly diagnose and confirm TB.

*FASTPlaque*TB is the first commercialised test based on the phage amplification technology. This kit was introduced in Pakistan in 2000 for clinical evaluation. In the present study, 584 sputum samples were tested, and complete results were available for 514. The assay was easy to perform, results were available in 48 hours, and were easy to interpret. When compared to culture, *FASTPlaque*TB had a sensitivity of 81.6% and a specificity of 98%. The low number of false-positive results seen in this study strongly suggests that a positive *FASTPlaque*TB result corresponds to active TB.

Sixty-two per cent of the samples tested in this study were negative on smear. The high sensitivity (67%) and specificity (98%) of the assay, coupled with the rapid availability of the results (48 hours versus 6–8 weeks for culture), suggests a very effective role for *FASTPlaque*TB in early diagnosis of smear-negative cases. Our results are comparable to those obtained from similar studies carried out in other high TB prevalence areas. In a study conducted in Cape Town, South Africa, *FASTPlaque*TB had a sensitivity and specificity of respectively 48% and 98% for smear-negative TB.[16]

For 11.4% of the smear-positive specimens tested, *FASTPlaque*TB gave false-negative results when compared to culture findings. The cultures that grew from these specimens were confirmed as *M. tuberculosis*, and the assay when performed on the culture was positive. The *FASTPlaque*TB-negative results obtained for the four specimens with low AFB numbers (±) can perhaps be explained by the non-uniform distribution of the bacterial load in the test suspension. But for specimens that had sufficient numbers of AFB (18/22), factors other than AFB burden must be contributing to the failure of the assay. Similar findings were reported by Eltringham et al., who reported a primary failure rate of 8% when performing indirect drug susceptibility testing of *M. tuberculosis* isolates using D29 bacteriophage. Although they were also able to obtain valid results on retesting, the primary failure in their study also remained unexplained.[17]

The low number of false-positive *FASTPlaque*TB results (2.9%) observed in this study shows that the assay has a very high specificity for the *M. tuberculosis* strains present in our population. The smear-positive, *FASTPlaque*TB-positive specimens that were negative on culture had ± AFB scores. The much larger volume (1 ml) of specimen used in the *FASTPlaque*TB assay as compared to the culture inocula (0.2 ml) is likely to improve the sensitivity of the assay when testing low AFB burden specimens, and may explain the discrepant results.

The results of this study show a very high specificity of the AFB smear (94%) for *M. tuberculosis*. The majority of false-positive smear results (*FASTPlaque*TB-negative, culture-negative) were for specimens that contained small numbers of AFB (±). These results could be described as microscopy artefacts due to non-specific staining of other microorganisms. The unexplained false-positive rate for smear microscopy varies between 10% and 20%.[18] The low incidence of NTM seen in this study also strongly supports the specificity of a positive smear. Similar results have been reported from other high prevalence TB areas, where serious infections with NTM are not common. Conde et al., in a study from Brazil, showed AFB smear to have a specificity of 98% for *M. tuberculosis*.[19] Our data show that when smear and *FASTPlaque*TB results are combined to make a TB diagnosis, 90% of the culture-confirmed

cases can be rapidly identified with a high degree of specificity (92%).

In the initial part of the study, 18.6% of the samples were lost to analysis due to overgrowth of contaminating bacteria on assay plates during the overnight incubation of treated samples. These contaminants may be resistant to the sputum decontamination procedure used, or may be introduced accidentally during specimen collection and handling. As a large number of contaminants seen were gram-positive bacteria, specially Bacillus and Staphylococcus species, penicillin was added to the assay medium to inhibit their growth. This resulted in the inhibition of gram-positive contaminants, and brought down the contamination rate from 18.6% to 5.3%. Comparable results were obtained with Microclens, which also effectively inhibited gram-positive contaminants. The higher rate of contamination seen with Microclens (9% versus 5.3% with penicillin) was due to the presence of NaOH-resistant gram-negative bacteria and did not reflect differences in the antibacterial effectiveness of the two agents used. As Microclens is relatively more stable and easier to use than penicillin, it was preferred for the latter part of the study.

An important point of our study is that growth of contaminating bacteria in test samples must be controlled for effective application of this methodology in routine diagnosis. This is perhaps more relevant for developing countries like Pakistan, where sputum collection may not always be performed under optimal conditions, and laboratory facilities are not ideal. Isolation of Bacillus and Staphylococcus species from a large number of samples in this study is strongly suggestive of contamination being introduced during sample collection and handling.

The data presented in this study demonstrate that *FASTPlaque*TB is a sensitive and specific assay that allows rapid detection of *M. tuberculosis* in sputum samples. The assay is not technically demanding, and routine microbiology laboratory staff can be easily trained to perform the test. As there is no special equipment requirement, the test can be easily incorporated into the routine of a laboratory involved in AFB smear microscopy.

The test is relatively safe to perform, as the phage replication results in lysis of the viable TB bacilli present in the specimen. This makes the phage amplification methodology safer than the rapid culture techniques which directly or indirectly detect increase in the number of viable cells. As positive cultures must be further identified for a confirmed diagnosis, these procedures still require 2 to 6 weeks. The ability of the *FASTPlaque*TB assay to detect and identify *M. tuberculosis* directly in the clinical specimen makes it a very useful tool in TB diagnosis.

Rapid diagnostic methods based on PCR technology are sensitive and specific, but require special skills and are expensive when compared to routine microscopy and culture. The requirement for dedicated equipment and special technical skills has restricted diagnostic PCR to a few commercial laboratories in Pakistan. The *FASTPlaque*TB assay is less expensive than PCR ($8 versus $24 per test for *M. tuberculosis* PCR). A major advantage of the new technology is that it detects only live TB bacilli and the problems associated with PCR detection of non-viable or clinically irrelevant pathogens are avoided.

In conclusion, the present study shows that *FASTPlaque*TB offers a potential for an accurate diagnosis within 48 hours after samples are received in the laboratory. The high specificity and sensitivity of the assay would allow efficient TB diagnosis in the majority of suspected cases. Its low-tech nature is likely to facilitate its routine application in the routine diagnostic laboratory.

Acknowledgements

We are very grateful to Drs Ashraf Sadique and Sabira Tehseen and staff of the Ojha Institute of Chest Diseases for providing the sputum samples for this study.

These data were presented as a poster at the 32nd IUATLD World Conference on Lung Health, 1–4 November 2001, Paris, France.

References

1 World Health Organization. Global Tuberculosis Program. Global Tuberculosis Control. WHO Report 1999. 181. Geneva: WHO, 1999.
2 Hall G S, Howard B J. Mycobacteria. In: Howard B J, Keiser J F, Smith T F, Weissfeld A S, Tilton R C, eds. Clinical and pathogenic microbiology. St. Louis, MO: Mosby-Year Book Inc., 1994: pp 503–528.
3 Dutt A K, Stead W W. Smear-negative pulmonary tuberculosis. (published erratum appears in Semin Respir Infect 1994; 9:261). Semin Respir Infect 1994; 9: 113–119.
4 World Health Organization. Global Tuberculosis Control. WHO Report 2000. WHO/CDS/TB/2000, 275. Geneva: WHO, 2000.
5 Behr M A, Warren S A, Salamon H, et al. Transmission of *Mycobacterium tuberculosis* from patients smear negative for acid fast bacilli. Lancet 1999; 353: 444–449.
6 Roth A, Schaberg T, Mauch H. Molecular diagnosis of tuberculosis: current clinical validity and future prespectives. Eur Respir J 1997; 10: 1877–1891.
7 Jonas V, Longiaru M. Detection of *Mycobacterium tuberculosis* by molecular methods. Clin Lab Med 1997; 15: 119–128.
8 Riska P F, Jacobs W R Jr, Bloom B R, McKitrick J, Chan J. Specific identification of *Mycobacterium tuberculosis* with the luciferase reporter mycobacteriophage: use of p-nitroacetyl-amino-beta-hydroxy propiophenone. J Clin Microbiol 1997; 35: 3225–3231.
9 McNerney R, Kiepiela P, Bishop K S, Nye P M, Stoker N G. Rapid screening of *Mycobacterium tuberculosis* for susceptibility to rifampicin and streptomycin. Int J Tuberc Lung Dis 2000; 4: 1–7.
10 Wilson S M, al-Suwaidi Z, McNerney R, Porter J, Drobniewski F. Evaluation of a new rapid bacteriophage based method for the drug susceptibility testing of *Mycobacterium tuberculosis*. Nat Med 1997; 3: 465–468.
11 McNerney R. TB: the return of the phage. A review of fifty years of mycobacteriophage research. Int J Tuberc Lung Dis 1999; 3: 179–184.

12 Collins C H, Grange J M, Yates, M D. Identification of species. In: Collins C H, Grange J M, Yates, M D, eds. Tuberculosis bacteriology: organization and practice. 2nd ed. Oxford: Butterworth-Heinemann, 1997: pp 69–97.
13 Eisenach K D, Cave M D, Bates J H, Crawford J T. Polymerase chain reaction amplification of a repetitive DNA sequence specific for *Mycobacterium tuberculosis*. J Infect Dis 1990; 61: 977–981.
14 Tattevin P, Casalino E, Fleury L, et al. The validity of medical history, classic symptoms, and chest radiographs in predicting pulmonary tuberculosis: derivation of a pulmonary tuberculosis prediction model. Chest 1999; 115: 1248–1253.
15 Kanaya A M, Glidden D V, Chambers H F. Identifying pulmonary tuberculosis in patients with negative sputum smear results. Chest 2001; 120: 349–355.
16 Albert H, Heydenrych A, Brookes R, et al. Performance of a rapid phage-based test, *FASTPlaque*TB™, to diagnose tuberculosis from sputum specimens in South Africa. Int J Tuberc Lung Dis 2002; 6: 529–537.
17 Eltringham I J, Drobniewski F A, Mangan J A, Butcher P D, Wilson S M. Evaluation of reverse transcription-PCR and a bacteriophage-based assay for rapid phenotypic detection of rifampin resistance in clinical isolates of *Mycobacterium tuberculosis*. J Clin Microbiol 1999; 37: 3524–3527.
18 Roberts G D, Koneman E W, Kim Y K. *Mycobacterium*. In: Balows A, Hausler W J, Hermann K L, Isenberg H D, Shadomy H J, eds. Manual of clinical microbiology. 5th ed. Washington, DC: American Society for Microbiology, 1991: pp 304–340.
19 Conde M B, Figueira C M, Moraes R, Fonseca L S, Deriemer K, Kritski A L. Predictive value of the acid fast smear for detection of *Mycobacterium tuberculosis* in respiratory specimens in a reference center of HIV/AIDS in Rio de Janeiro, Brazil. Mem-Inst-Oswald-Cruz 1999; 946: 787–790.

RÉSUMÉ

CONTEXTE : Des échantillons d'expectoration ont été recueillis chez des patients suspects de tuberculose (TB) fréquentant les dispensaires externes à l'Institut des Maladies Thoraciques Ojha à Karachi, Pakistan.

OBJECTIF : Evaluer les performances du test *FASTPlaque*TB pour le diagnostic rapide de la TB pulmonaire.

SCHÉMA : Etude comparative de 584 échantillons d'expectoration explorés par l'examen microscopique des bacilles acido-résistants, la culture sur Löwenstein-Jensen et le test *FASTPlaque*TB.

RÉSULTATS : Au total, on a obtenu des résultats complets pour 514 échantillons. Soixante-dix échantillons n'ont pu être analysés par suite de contaminations. L'addition d'antibiotiques a inhibé le développement de contaminants à Gram-positif et a pu réduire le taux global de contamination de 18,2% à 7,2%. On a isolé *Mycobacterium tuberculosis* dans 175 échantillons à bacilloscopie positive et 70 à bacilloscopie négative. Le test *FASTPlaque*TB a détecté *M. tuberculosis* dans 81.6% des échantillons, avec une spécificité de 97,7%. La sensibilité et la spécificité du test ont été respectivement de 87,4% et de 88,2% pour les échantillons à bacilloscopie positive. Pour les échantillons à bacilloscopie négative, la sensibilité du test a été de 67,1% et sa spécificité de 98,4%. La sensibilité combinée du frottis et du test *FASTPlaque*TB pour *M. tuberculosis* a été de 90%. Les résultats des tests étaient disponibles après 48 heures.

CONCLUSION : *FASTPlaque*TB est un test sensible et spécifique pour le diagnostic rapide de la tuberculose pulmonaire dans les zones à haute prévalence. Le test est suffisamment sensible pour confirmer un grand nombre de cas à bacilloscopie négative et cliniquement suspects et améliorer ainsi le dépistage des cas.

RESUMEN

MARCO DE REFERENCIA : Se recolectaron muestras de esputo de pacientes sospechosos de tuberculosis (TB) que se presentan a consultorios externos del Instituto de Enfermedades Respiratorias Ojha, Karachi, Paquistán.

OBJETIVO : Evaluar el rendimiento del test *FASTPlaque*TB para el diagnóstico rápido de la tuberculosis pulmonar.

MÉTODO : Estudio comparativo de 584 muestras de esputo utilizando el examen microscópico para bacilos ácido-resistentes, el cultivo en medio Lowenstein-Jensen y el test *FASTPlaque*TB.

RESULTADOS : Se obtuviron resultados completos para un total de 514 muestras de esputo. Se perdieron 70 muestras debido a la contaminación. La adición de antibióticos inhibió el crecimiento de contaminantes Gram positivos y redujo la tasa global de contaminación de 18,2% a 7,2%. Se aisló *Mycobacterium tuberculosis* en 175 muestras con baciloscopia positiva y en 70 con baciloscopia negativa. El test *FASTPlaque*TB detectó *M. tuberculosis* en el 81,6% de las muestras, con una especificidad de 97,7%. La sensibilidad y especificidad del test para las muestras con baciloscopia positiva fueron de 87,4% y de 88,2%, respectivamente. Para las muestras con baciloscopia negativa la sensibilidad del test fue de 67,1% y la especificidad de 98,4%. La sensibilidad combinada de la baciloscopia y de *FASTPlaque*TB para *M. tuberculosis* fue de 90%. Los resultados del test se encontraban disponibles a las 48 horas.

CONCLUSIÓN : *FASTPlaque*TB es un test sensible y específico para el diagnóstico rápido de la TB pulmonar en áreas de alta prevalencia. La sensibilidad del test es suficiente como para confirmar un gran número de casos con baciloscopia negativa y mejorar así la detección de casos.