Comparative study of Amplicor polymerase chain reaction and ligase chain reaction for direct detection of *M. tuberculosis* in clinical specimens

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**ABSTRACT**

**Objective:** To compare the sensitivity and specificity of two deoxyribonucleic acid amplification techniques, Amplicor polymerase chain reaction and ligase chain reaction in the detection of *M. tuberculosis* in clinical specimens.

**Methods:** At a 350-bed general hospital in Jeddah, Saudi Arabia, 326 patient samples were selected for this study on the basis of positive smear for acid fast bacilli, history of mycobacterial disease or culture positivity, or both. Each sample in addition to microscopy and culture, was tested by both Amplicor polymerase chain reaction and ligase chain reaction.

**Results:** Of the 326 patient samples, 74 specimens were culture positive for *M. tuberculosis* and 46/74 (62%) were acid fast bacilli smear positive. All the 46 smear positive- *M. tuberculosis* positive cultures were ligase chain reaction positive (100%) and 44 (95.6%) were positive by Amplicor polymerase chain reaction. In the 28 smear negative *M. tuberculosis* positive cultures, 25 (89.3%) were ligase chain reaction positive and 21 (75.0%) were Amplicor polymerase chain reaction positive. Of all specimens, 68/326 (20.9%) were culture positive for non-tuberculous mycobacteria and all were negative by Amplicor polymerase chain reaction and ligase chain reaction.

**Conclusion:** The high specificity of both methods allows for differentiation of *M. tuberculosis* from non-tuberculous mycobacteria. The use of deoxyribonucleic acid amplification techniques allows for rapid and specific diagnosis of *M. tuberculosis*, thus eliminating unnecessary administration of antituberculosis therapy and isolation measures for non-tuberculous mycobacteria patients.

**Keywords:** Amplicor PCR, LCR, Tuberculosis, DNA.


Conventional diagnosis of respiratory tract tuberculosis is made by first concentrating, digesting, and decontaminating respiratory tract specimens (expectorated or induced sputa, bronchoalveolar lavage or gastric lavage) and then performing both acid fast microscopy using fluorescein isothiocyanate, or Ziehl Neelsen (ZN) stain and culture of the processed specimens. Acid-fast microscopy results can be reported in 2 hours or less but the method lacks sensitivity and is unable to distinguish between different species of mycobacteria. It is estimated that 5,000 - 10,000 organisms per milliliter of sputum are required for smear positivity and that only 50% of clinically diagnosed cases are culture positive. Growth on selective agar is slow, with visible colony growth taking at least 3 weeks and usually 4-6 weeks. Due to the communicability of tuberculosis, the delay in

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confirmed diagnosis by culture contributes to the spread of this disease, hence as an alternative to slow growth of mycobacteria by culture, methods amplifying mycobacterial nucleic acid have been developed which can reduce the time for diagnosis to a single day.

Isothermal enzymatic amplification targeting repetitive sequences in ribosomal ribonucleic acid (rRNA), using the Gen-Probe Amplified Mycobacterium tuberculosis Direct Test (MTD) and Amplicor polymerase chain reaction (PCR), which uses PCR to amplify a deoxyribonucleic acid (DNA) target sequence in the 16S rRNA gene, have now been widely applied for the detection of Mycobacterium tuberculosis (MTB) nucleic acid in the processed sediments of respiratory specimens. Although these techniques are more sensitive than direct microscopy, discrepant results occur between these molecular methods and conventional culture. Ligase chain reaction (LCR) is a probe amplification technique first described in 1989 and modified in 1991 incorporating a DNA polymerase, is based on sequential rounds of template dependent ligation of two adjacent oligonucleotide probes. When a pair of probes has hybridized to the target sequence on a single strand of DNA, there is a gap of a few nucleotides between the probes. The addition of a DNA polymerase into the reaction acts to fill in this gap between the probes by incorporating nucleotides. Once the gap is filled ligase can covalently join the pair of probes to form an amplification product that is complementary to the original target sequence and can itself serve as a target in subsequent cycles of amplification. The target nucleic acid sequence for the LCR assay for MTB (Abbott LCX Probe system, Abbott Diagnostics, Chicago, Ill.) is found within the single copy chromosomal gene of MTB which encodes for protein antigen b. This gene sequence appears to be specific to the MTB complex and has been detected in all MTB complex strains examined to date. The aim of this study is to compare the sensitivity and specificity of the LCR and Amplicor PCR assays in comparison with culture, for the detection of MTB in clinical specimens.

Methods. This study was conducted in the Microbiology Division of the King Khalid National Guard Hospital (KKNGH), Jeddah, Saudi Arabia. KKNGH is a 350-bed general teaching hospital which provides primary and secondary health care to Saudi Arabian National Guardsmen and their dependants with access to all Saudis. The Microbiology Division is a referral center for specimens from 4 primary health care clinics both in Jeddah and Taif which investigates approximately 2000 respiratory specimens annually for the presence of mycobacteria. Specimens were collected between April and September 1996 from patients being investigated for tuberculosis or other pulmonary mycobacterial infections or from patients being followed during antituberculosis therapy. A total of 326 specimens, 320 respiratory specimens (sputum, bronchoalveolar lavage and gastric lavage) from 165 patients with up to a maximum of 3 specimens included per patient, and 6 specimens of pus from extrapulmonary abscesses were included in this study. Specimens were selected for this study on the basis of smear positivity, history of mycobacterial disease and culture positivity with molecular assays performed retrospectively. Four groups of specimens were investigated viz: 1. Seventy four specimens from 33 patients which were culture positive for MTB. 2. Sixty eight samples from 46 patients which were culture positive for non-tuberculous mycobacteria (NTM) on either solid or liquid culture media. 3. Sixty six respiratory samples from 30 patients with prior history of mycobacterial disease which were acid fast bacilli (AFB) smear positive but negative for culture on solid media alone, and 4. One hundred and eighteen specimens from 61 patients which were culture negative in both solid and liquid media and negative by acid fast stain.

Specimens were held at 4°C until processed by standard laboratory procedures. The majority of specimens were processed within 24 hours by a standard N-acetyl-L-cysteine sodium hydroxide method and were centrifuged at 3,000 x g for 20 minutes. The final pellet was resuspended to 1.5-2.0 ml in sterile saline. For each specimen, 2 Lowenstein-Jensen (LJ) slopes were inoculated with 0.2 ml of specimen and a smear was prepared for auramine-phenol staining. Fluorochrome staining was performed by standard procedures and all positive smears were counterstained by the ZN stain. Slope cultures were examined weekly for 8 weeks. Of the majority of specimens 241/326, 0.5 ml was inoculated into a MB/BacT (Organon Teknika, Durham, N.C.) process bottle containing Middlebrook 7H9 broth, Tween 80 (0.4% w/v), Glycerol (5% w/v), Amanth (0.002% w/v) and an antibiotic supplement containing Amphotericin B (0.018% w/v), Azlocillin (0.0034% w/v), Nalidixic acid (0.04% w/v), Polymixin B (10,000 units) and Trimethoprim (0.0105% w/v). Liquid cultures were monitored continuously in the MB/BacT 2400 cabinet (Organon Teknika) for up to 8 weeks. Eighty five/326 specimens with prior isolation of either MTB or NTM were inoculated onto LJ media but not into liquid media for cost effectiveness. All positive isolates were confirmed as Mycobacterium sp. isolates on the basis of ZN stain with characteristic morphological appearance. All isolates were sent to the Mycobacterium Reference Unit, Public Health Laboratory Service, University of Wales, Cardiff, UK, for full identification and appropriate sensitivity tests.

An aliquot from each sample (0.7-1.1 ml) was stored frozen at -20°C for up to 4 months until
molecular assays were performed. Of the processed sediment 0.1 ml was assayed by the Amplicor PCR Mycobacterium tuberculosis test (Roche Diagnostics Systems, Branchburg, N.J.) according to the manufacturers' instructions. Briefly, sample preparation required a single centrifugation step with a wash solution followed by incubation with a lysis reagent at 60°C. Positive and negative controls which were included in each run were prepared by adding 100 ul of TB positive and negative controls to 400 ul of sputum lysis reagent supplied by the manufacturers. Amplification of DNA was performed in a 37 cycle program in a Perkin-Elmer Thermocycler 2400 (Perkin-Elmer Cetus, Norwalk, Conn). The detection step required hybridization of amplified, denatured products to a MTB specific probe on a microwell plate and was detected by an enzyme immunoassay technique. The optical density was measured at a wavelength of 450 nm. Specimens with A450 ≥ 0.350 were considered positive. Selective amplification of target DNA from clinical specimens in the Amplicor MTB test is achieved by the use of amperase, which contains the enzyme Uracil N-Glycosylase (UNG), which recognizes and catalyses the destruction of deoxyuridine-containing DNA but not thymidine-containing DNA. The presence of deoxyuridine in amplicons renders contaminating amplicons susceptible to destruction by amperase prior to the amplification of the target DNA.

0.5ml of the sediments from processed clinical specimens were tested by the LCR Mycobacterium tuberculosis assay (Abbott LCX Probe system) according to the manufacturer's directions in the draft clinical brochure. The sample was added to a 1.5ml microcentrifuge tube containing glass beads and an acidic buffer solution. The MTB DNA preparation uses two centrifugation steps at 9000 x g for 10 minutes, heat treatment at 95°C for 20 minutes in a covered bath (Abbott LCX Probe system), and sonication in an LCX Lysor (Abbott LCX Probe system). Amplification was carried out using a 37 cycle program in a Perkin-Elmer thermal cycler (Abbott LCX Probe system). Detection was achieved by loading amplification vials directly into a carousel for automated detection in the LCX analyzer (Abbott LCX Probe system). Two negative and 2 positive reagent controls and one saline negative and positive (4000-6000 cfu/ml MTB) processing control were included in each batch of samples assayed. The cut-off value for the assay was determined at 0.3 times the mean positive reagent control rate. The sample results were calculated as the ratio of the sample rate (S) to the cut-off rate (CO) i.e. (S/CO). Samples in the ratio range S/CO 0.3-1.0 were considered equivocal, while ratios greater than 1.0 were considered positive for MTB. To eliminate the risk of amplification product contamination, at the end of LCx MTB assay, the amplification product is automatically inactivated using a two-reagent, chemical inactivation system. Both reagents are delivered into the LCx reaction cells by the LCx analyzer after the amplification product has been detected. The ensuing reaction results in the complete destruction of any nucleic acid present.

Results. Table 1 summarizes the results of the 326 patient samples investigated in this study. In Group 1, of 74 specimens culture positive for MTB, 71 specimens were LCR positive. The 3 samples MTB culture positive and LCR negative were AFB smear negative. Two of these samples gave equivocal S/CO ratios (0.89 and 0.87) and the 3rd sample (S/CO ratio 0.029) was culture positive in liquid media only after 30 days. 65/74 specimens culture positive for MTB were positive by the Amplicor PCR method. Nine specimens, 7 of which were AFB smear negative were Amplicor PCR negative from 5 patients. In one patient a previous specimen was Amplicor PCR positive. AFB smear

<table>
<thead>
<tr>
<th>Mycobacteria culture and acid fast bacilli (AFB) smear result</th>
<th>No. of samples</th>
<th>No. LCR positive</th>
<th>No. Amplicor PCR positive</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1: MTB (AFB smear positive).</td>
<td>46</td>
<td>46</td>
<td>44</td>
</tr>
<tr>
<td>MTB (AFB smear negative).</td>
<td>28</td>
<td>25</td>
<td>21</td>
</tr>
<tr>
<td>All MTB positive cultures.</td>
<td>74</td>
<td>71</td>
<td>65</td>
</tr>
<tr>
<td>Group 2: <em>M. avium intracellulare</em></td>
<td>40</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td><em>M. scotochromogen</em></td>
<td>14</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td><em>M. fortuitum</em></td>
<td>3</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td><em>M. chelonae</em></td>
<td>5</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td><em>M. terrae</em></td>
<td>5</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td><em>M. kansasi</em></td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>All MTM</td>
<td>68</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Group 3: No growth (on solid media alone) (AFB smear positive)</td>
<td>66</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Group 4: No growth (on solid media alone) (AFB smear negative)</td>
<td>118</td>
<td>2</td>
<td>1</td>
</tr>
</tbody>
</table>

TOTAL 326 75 67
was positive in 46 (62%) of the 74 samples culture positive for MTB. In Group 2, the 68 patient samples from 46 patients culture positive for NTM were all negative by both LCR and Amplicor PCR. In group 3, comprising 66 patient samples who had positive AFB smear and no growth on solid media alone, were 64 samples from 28 patients in which Mycobacterium avium complex (MAC) had been previously isolated and from 2 patients undergoing antituberculous therapy with previous positive cultures for MTB. LCR was positive only in the 2 samples with prior isolation of MTB. Amplicor PCR was positive in one of these samples. In group 4, of 118 patient samples AFB smear negative and culture negative, 2 samples were positive by LCR with one of these samples also Amplicor PCR positive. This sample was from known tuberculosis patients on antituberculosis therapy. One sample had a S/CO ratio 1.13 and was from a patient without clinical evidence of tuberculosis and was considered a false positive for LCR. In Table 2, the summaries of the sensitivity, specificity, accuracy and predictive values of the molecular methods and AFB smear result in relation to culture are presented. The NTM specimens (i.e Group 2 specimens) were excluded in the statistical analysis and comparisons were carried out on the remaining 258 specimens. In smear positive samples, LCR compared with culture showed a sensitivity of 100% and a specificity of 97.0%. Amplicor PCR versus culture had a sensitivity of 95.6% and specificity of 98.5% in smear positive specimens. In the 28 AFB smear negative, culture positive specimens, LCR had a sensitivity of 89.3% and a specificity of 98.3%, and Amplicor PCR had a sensitivity of 75% and a specificity of 99.2%. In all samples, LCR had the highest sensitivity of 95.9% while AFB smear had the lowest sensitivity of 62.2%. The sensitivity for PCR was 87.8%. The specificity of PCR was highest (98.9%), that of LCR 97.8%, and that of AFB smear 64.1%. LCR had the highest accuracy of 97.3% (i.e., the proportion of all specimens that were correctly identified). The accuracy of PCR was 95.7% while that of AFB smear was 63.6%. The proportion of test positives that were true positives (i.e., positive predictive value) was highest for PCR (97%), and was lowest for AFB smear (41.1%). That of LCR was 94.7%. Similarly, the proportion of test negatives that were true negatives (i.e., negative predictive value) was also highest for PCR (98.9%) and was lowest for AFB smear (80.8%). The negative predictive value for LCR was 98.4%. The results of the statistical signiﬁcant tests applying the McNemar test to compare each diagnostic test against the others, are shown in Table 3. The sensitivity of LCR was signiﬁcantly greater than that of PCR (0.025 < P < 0.05), but there was no signiﬁcant difference in their speciﬁcity (P > 0.10). Similarly, there was no difference in their accuracy (P > 0.05). However, both LCR and PCR are significantly superior to AFB smear in terms of sensitivity, speciﬁcity and accuracy (P < 0.001).

Table 2 - Sensitivity, speciﬁcity, accuracy and predictive values of LCR, Amplicor PCR and AFB smear in the diagnosis of M.tuberculosis.

<table>
<thead>
<tr>
<th></th>
<th>LCR (%)</th>
<th>PCR (%)</th>
<th>AFB smear (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Sensitivity</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AFB+</td>
<td>100.0</td>
<td>95.6</td>
<td></td>
</tr>
<tr>
<td>AFB-</td>
<td>89.3</td>
<td>75.0</td>
<td></td>
</tr>
<tr>
<td>All specimens</td>
<td>95.9</td>
<td>87.8</td>
<td>62.2</td>
</tr>
<tr>
<td><strong>Specificity</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AFB+</td>
<td>97.0</td>
<td>98.5</td>
<td></td>
</tr>
<tr>
<td>AFB-</td>
<td>98.3</td>
<td>99.2</td>
<td></td>
</tr>
<tr>
<td>All specimens</td>
<td>97.8</td>
<td>98.9</td>
<td>64.1</td>
</tr>
<tr>
<td><strong>Accuracy</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AFB+</td>
<td>98.2</td>
<td>97.0</td>
<td></td>
</tr>
<tr>
<td>AFB-</td>
<td>96.6</td>
<td>94.5</td>
<td></td>
</tr>
<tr>
<td>All specimens</td>
<td>97.3</td>
<td>95.7</td>
<td>63.6</td>
</tr>
<tr>
<td><strong>Positive predictive value</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AFB+</td>
<td>95.8</td>
<td>97.8</td>
<td></td>
</tr>
<tr>
<td>AFB-</td>
<td>92.6</td>
<td>95.5</td>
<td></td>
</tr>
<tr>
<td>All specimens</td>
<td>94.7</td>
<td>97.0</td>
<td>41.1</td>
</tr>
<tr>
<td><strong>Negative predictive value</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AFB+</td>
<td>100.0</td>
<td>97.0</td>
<td></td>
</tr>
<tr>
<td>AFB-</td>
<td>97.6</td>
<td>94.4</td>
<td></td>
</tr>
<tr>
<td>All specimens</td>
<td>98.4</td>
<td>98.9</td>
<td>80.8</td>
</tr>
</tbody>
</table>

Table 3 - Statistical test* of signiﬁcance of sensitivity and speciﬁcity of LCR, Amplicor PCR and AFB smear.

<table>
<thead>
<tr>
<th></th>
<th>LCR</th>
<th>PCR</th>
<th>AFB Smear</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>X²</td>
<td>P-Value</td>
<td>X²</td>
</tr>
<tr>
<td><strong>Sensitivity</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LCR</td>
<td>4.1</td>
<td>P &lt; 0.05</td>
<td>23.0</td>
</tr>
<tr>
<td>PCR</td>
<td></td>
<td></td>
<td>15.7</td>
</tr>
<tr>
<td><strong>Specificity</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LCR</td>
<td>0.5</td>
<td>P &gt; 0.10</td>
<td>56.4</td>
</tr>
<tr>
<td>PCR</td>
<td></td>
<td></td>
<td>60.1</td>
</tr>
<tr>
<td><strong>Accuracy</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LCR</td>
<td>2.25</td>
<td>P &lt; 0.05</td>
<td>85.0</td>
</tr>
<tr>
<td>PCR</td>
<td></td>
<td></td>
<td>60.1</td>
</tr>
</tbody>
</table>

* McNemar Test was used (n.s) not significant

Discussion. Saudi Arabia, still has a high incidence of tuberculosis despite the great decline from 135 per 100,000 in 1980 to 18.6 per 100,000 in 1990. Highest figures have been reported from the Jeddah health region with an incidence rate of 63.4 per 100,000. These figures represent an incidence 2-4 times greater than that of developed countries. In our patient population, patients with positive AFB smears and pulmonary symptoms with or without radiological findings are frequently started on antituberculous therapy pending the results of culture which may take up to 12 weeks. It has been observed in the Southern region of Saudi Arabia that up to 50% of healthy individuals harbor NTM in their
mounds and throats. The high incidence of NTM complicates the diagnosis of tuberculosis and frequently results in the unnecessary administration of antituberculosis drugs. Therefore tests which can provide a high specificity for MTB in smear positive specimens will prove cost effective in reducing use of antituberculosis drugs, contact investigations and isolation facilities. The sample groups 2 and 3, comprising predominantly of samples with either positive NTM culture or prior history of NTM isolation demonstrate the high specificity of both molecular methods as no false positive results were observed. These samples highlight our experience in NTM culture and the report of Hoffner, illustrating that liquid culture supports the growth of mycobacteria far more than Lowenstein Jensen alone, particularly in the isolation of Mycobacterium avium.

In this study AFB smear was positive in 62% (46/75) of culture positive specimens for MTB. The sensitivity of LCR was 100% and the specificity was 97.0% in the smear positive samples. In smear positive specimens Amplicor PCR showed a sensitivity of 95.6% and specificity of 98.5%. The high sensitivity and specificity of both the Ampligene PCR and LCR assays in smear positive specimens suggests that both can detect and differentiate MTB from NTM in a single respiratory specimen, hence greatly reducing the time of diagnosis of pulmonary tuberculosis. In contrast, the sensitivity of both assays was diminished in smear negative specimens. The sensitivity of the LCR assay was reduced to 89.3% and Amplicor PCR assay was reduced to 75.0% in smear negative specimens, however the specificity of both assays remained high at 98.3% and 99.2%. The LCR and Amplicor PCR positive but culture negative results observed in specimens from 2 patients, who had previously positive cultures and had antituberculosis therapy support the findings of other investigators that DNA amplification techniques can detect non-viable organisms. Both Amplicor PCR and LCR will differentiate equally and accurately between MTB and NTM which increases the reliability of these techniques.

Six extrapulmonary pus samples from patients with abscesses were investigated as part of this study. In 3 of these samples MTB was isolated and both PCR and LCR assays were positive. A sample of tissue from one of these patients (results not included) was culture positive and negative for both PCR and LCR. Previous studies on discrepant Amplicor PCR and culture results have attributed false negative results to the presence of inhibitors in both pulmonary and extrapulmonary specimens, sample variation, and reduced numbers of organisms. Moore and Curry in a retrospective and prospective study of 1009 specimens determined that only 7% of false negative PCR specimens were attributed to the presence of inhibitors. The 3 false negative LCR specimens were from AFB smear negative samples from 3 patients and were also negative by Amplicor PCR. Two of these samples which gave equivocal LCR results, were from patients that had prior positive LCR specimens. One culture positive sample, but LCR negative was only culture positive for MTB in liquid media after 30 days of incubation. This suggests that both false negative LCR and Amplicor PCR results may have been due to reduced numbers of organisms or sample variation. In patients with negative smear and radiological evidence of disease it would be advantageous to test multiple (at least 2 or 3) respiratory samples to reduce the risk of a false negative result due to the low numbers of organisms particularly in severely immunocompromized AIDS population where AFB smear has low sensitivity.

The ability of Amplicor PCR to detect MTB is reduced when the number of organisms in the sample is scanty, particularly in smear negative paucibacillary samples. In a study by Schirm et al. comparing in-house PCR and Amplicor PCR with culture, obtained a sensitivity of 90% with in-house PCR as opposed to 60% with Amplicor PCR in smear negative specimens. The authors correlated the higher sensitivity with the in-house PCR in smear negative specimens to the increased sample volume added to the amplification reaction in the in-house PCR (almost 5 times that of Amplicor PCR). The larger specimen volume (0.5ml) used in the LCR assay as recommended by the manufacturer probably contributes to the greater sensitivity observed. However, this may allow for higher concentrations of inhibitors in the reaction as shown by An et al. The use of 2 wash and centrifugation steps in the sample preparation for the LCR assay compared with the single sample wash in the Amplicor PCR assay is probably integral in the removal of inhibitors. We support the suggestion of Cartuyvels et al that the sensitivity of the Amplicor PCR assay may be enhanced by increasing the specimen volume. An increase in the sample volume e.g. from 0.1 ml to 0.5 ml would however necessitate a second wash step in the sample preparation of Amplicor PCR to aid inhibitor removal. In both methods, sample preparation is not technically demanding, although specimens must be initially processed in a Class 1 Biological Safety Cabinet with strict adherence to the prescribed methodology. In our study, and as previously reported, Amplicor PCR is easily performed and results can be reported within one 8 hour shift. Similarly, in the LCR assay a batch of 48 specimens including controls can be prepared within 2 1/2 hours. The amplification and detection steps are automated and require approximately one hour for each step. It is therefore possible to incorporate this assay into the normal laboratory work flow and report results within one 8 hour shift.

In this study, LCR assay for MTB shows greater
sensitivity than the Amplicor PCR assay (P < 0.05). The high specificity of both assays is of great benefit in differentiating MTB from NTM. In practical terms, the use of DNA amplification techniques allows for rapid and specific diagnosis of MTB, which in turn eliminates unnecessary administration of antituberculosis therapy and isolation measures for NTM patients.

Acknowledgment. We would like to thank Abbott Diagnostics for providing the LCR reagents and instrumentation for this study.

References