Performance Comparison of GeneXpert MTB/RIF and ProbeTec ET Tests for Rapid Molecular Diagnosis of Extrapulmonary Tuberculosis in a Low TB/MDR-TB Incidence Country

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Highlights of the Study

- GeneXpert MTB/RIF (Xpert) detected extrapulmonary tuberculosis (EPTB) in more specimens compared to ProbeTec ET (PTec-ET) in smear-positive and culture-positive samples.
- Xpert also detected EPTB in more specimens with histopathological evidence of tuberculosis (TB) and in cerebrospinal fluids for diagnosing TB meningitis.
- Xpert will help in better management of patients with EPTB, particularly in countries with low incidence of TB where EPTB is more common.

Keywords

Mycobacterium tuberculosis · Extrapulmonary tuberculosis · Molecular diagnosis · Xpert MTB/RIF assay · Probe Tec ET test · Kuwait

Abstract

Objective: This study evaluated the performance of GeneXpert MTB/RIF (Xpert) and ProbeTec ET (PTec-ET) assays in diagnosing extrapulmonary tuberculosis (EPTB) in Kuwait. **Materials and Methods:** We tested nonrespiratory clinical specimens ($n = 3,995$) collected from 3,995 patients suspected to have EPTB. These included cavitory fluids ($n = 2,054$), fine-needle aspirate (FNA)/pus/tissue biopsy ($n = 1,461$), urine ($n = 302$), cerebrospinal fluid (CSF, $n = 118$), and others ($n = 60$). All specimens were processed for acid-fast bacilli (AFB), culture in mycobacteria growth indicator tube 960 system,

and nucleic acid detection by Xpert and PTEC-ET according to manufacturer's instructions. **Results:** Of 3,995 specimens, 95 were AFB-positive, 403 were culture-positive, and an additional 86 samples had histopathology suggestive of TB. Using culture as reference, the sensitivity and specificity values were 88.33 and 97.3% for Xpert and 72.95 and 97.80% for PTEC-ET, respectively. Although performance of both tests was comparable in AFB-positive samples, Xpert detected significantly more cases in culture-positive samples. Among culture-negative samples, Xpert detected 18 more cases including 16 with histopathological evidence of TB. Lowest positivity was detected for both tests in cavitory fluids. Xpert performed better than PTEC-ET in culture-positive FNA/pus/tissue biopsy and CSF samples. **Conclusions:** Although performance of both tests was suboptimal for AFB-negative/culture-positive samples, Xpert performed better than PTEC-ET and also detected more cases of AFB-negative/culture-

negative/histopathology-positive samples. PTec-ET was positive in 3, while Xpert was positive in all 6 culture-positive CSF specimens for rapid diagnosis of TB meningitis. Xpert was thus superior to PTec-ET or smear microscopy in rapid diagnosis of EPTB.

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Introduction

Although the global incidence of tuberculosis (TB) and TB deaths is slowly declining, TB is still a formidable public health challenge. According to recent data released by the WHO, an estimated 10 million new active TB disease cases and ~1.41 million deaths occurred in 2019, making TB one of the top 10 causes of death worldwide and the leading cause of death from a single infectious agent [1]. Furthermore, an estimated 465,000 people developed TB that was resistant to rifampicin, and of these, nearly 363,000 (78%) were multidrug-resistant (MDR)-TB cases (defined as infection with a *Mycobacterium tuberculosis* strain resistant at least to rifampicin and isoniazid, the 2 most effective first-line drugs) which are much more difficult to treat than drug-susceptible TB [1–3]. Early detection of *M. tuberculosis* in clinical specimens, its susceptibility to anti-TB drugs, particularly rifampicin and effective treatment are essential for global TB control efforts [2–4].

Sputum smear-positive pulmonary TB is contagious as it spreads to close contacts, usually by coughing [5]. Extrapulmonary TB (EPTB) affects other parts of the body, mostly involving the pleura and lymph node, and is typically not infectious [6]. EPTB accounts for ~15% of all TB cases worldwide with the maximum prevalence of 24% reported from the WHO Eastern Mediterranean Region [1, 7]. The Eastern Mediterranean Region includes Kuwait, an Arabian Gulf country where ~95% of mycobacterial infections are caused by *M. tuberculosis* [8]. Kuwait has a low incidence of TB (~24 cases per 100,000 population), high (44%) prevalence of EPTB, and a low (~1.1%) incidence of MDR-TB [7, 9]. Other studies from nearby countries (Saudi Arabia and Turkey) have also reported high numbers of EPTB cases [10, 11].

The diagnosis of EPTB is problematic as it often involves invasive procedures that require special expertise and is limited in accuracy and time to diagnosis [6, 12]. Although culture is the gold standard for the diagnosis of active TB, it has low sensitivity (30–50%) for pleural fluid and even lower for cerebrospinal fluid (CSF) [4, 6, 12, 13]. Culture is also sometimes negative for biopsy material

even when there is histopathological evidence suggestive of active TB disease [6, 12, 14–16].

The GeneXpert MTB/RIF (Xpert) assay has been endorsed by the WHO as a rapid molecular test for the diagnosis of pulmonary TB in low- and middle-income countries and allows the simultaneous detection of *M. tuberculosis* complex and the most common mutations in the *rpoB* gene conferring resistance to rifampicin in <2 h [1, 17]. However, the performance of Xpert assay in non-respiratory samples for the diagnosis of EPTB is highly variable ranging from 83–96% in lymph node tissue and aspirates to 34–46% in pleural fluid with smear-positive samples yielding higher sensitivities [14–16]. Similarly, the diagnostic performance of ProbeTec ET (PTec-ET) assay with nonrespiratory samples is not well established [18]. This study was performed to evaluate the performance of Xpert and PTec-ET tests in diagnosing EPTB in Kuwait, an Arabian Gulf country with a low incidence of TB and MDR-TB [7].

Materials and Methods

Patients, Specimens, and M. tuberculosis Isolates

A total of 3,995 nonrespiratory specimens collected from 3,995 suspected TB patients during November 2011–November 2015 at Kuwait National TB Control Laboratory were used in this retrospective study. The clinical specimens were collected after obtaining verbal consent from suspected TB patients as part of routine diagnostic work-up and resistance surveillance, and the results are reported on de-identified samples. All samples were processed for smear microscopy for acid-fast bacilli (AFB), culture on liquid medium by mycobacteria growth indicator tube (MGIT) 960 system and detection of *M. tuberculosis* DNA with/without additional detection of resistance to rifampicin by Xpert and PTec-ET assays.

The smears for detection of AFB were prepared by Ziehl-Neelsen stain and the cultures from nonsterile samples were processed by using N-acetyl-L-cysteine and sodium hydroxide (NALC/NaOH), while sterile clinical specimens were directly processed according to the instructions supplied by the manufacturer of MGIT 960 system (Becton Dickinson, Sparks, MD, USA) and as described previously [9, 19]. The cultures flagged positive by MGIT 960 system were identified as *M. tuberculosis* by using the AccuProbe DNA probes, as described previously [9, 19]. All *M. tuberculosis* positive cultures were subjected to phenotypic drug susceptibility testing (DST) against 4 (streptomycin, isoniazid, rifampicin, and ethambutol) drugs (final drug concentrations: 1.0 mg/L for streptomycin, 0.1 mg/L for isoniazid, 1.0 mg/L for rifampicin, and 5.0 mg/L for ethambutol) by MGIT 960 system, as described previously [9, 19].

Cepheid GeneXpert MTB/RIF and ProbeTec ET Assays

The Xpert (Cepheid, Sunnyvale, CA, USA) assay was performed directly on clinical samples without prior extraction, and the results were interpreted by using the GeneXpert MTB/RIF cartridge and software according to the manufacturer's instructions and as de-

Table 1. Comparison of GeneXpert MTB/RIF (Xpert) and ProbeTec ET (PTec-ET) test results with smear microscopy for AFB and MGIT 960 system culture among 3,995 extrapulmonary specimens

Molecular test result	MGIT 960 system culture-positive		MGIT 960 system culture-negative	
	AFB-positive	AFB-negative	AFB-positive	AFB-negative
Xpert-positive/PTec-ET-positive	74	216	17 ^a	60 ^b
Xpert-positive/PTec-ET-negative	2	64	1	19 ^c
Xpert-negative/PTec-ET-positive	0	4	0	2
Xpert-negative/PTec-ET-negative	1 (NTM)	42	0	3,493
All Xpert-positive only	76	280	18	79
All PTEC-ET-positive only	74	220	17	62
κ	0.49	0.44	0	0.848
κ	0.46		0.87	
κ	0.875			

MGIT, mycobacteria indicator growth tube; AFB, acid-fast bacilli; NTM, nontuberculous mycobacteria; κ, kappa coefficient. ^a 16 samples showed histopathological evidence of TB. ^b 54 samples showed histopathological evidence of TB. ^c 16 samples showed histopathological evidence of TB.

scribed previously [20]. The PTEC-ET (Becton Dickinson, Sparks, MD, USA) assay was also performed by following the manufacturer's instructions and as described previously [21]. Briefly, the DNA released from decontaminated samples (150 µL) in duplicate was added to the priming wells of the microtiter plate containing primers and probes to rehydrate the reagents, and then 100 µL of the priming mix was added to the corresponding wells of another microtiter plate containing activated enzymes, buffer, and dNTPs to initiate amplification in the BD ProbeTec analyzer. Positive and negative controls were run with each assay including internal controls to avoid false-positive and false-negative results due to amplification inhibitors in clinical specimens and the results were interpreted according to the manufacturer's instructions.

Detection of Rifampicin Resistance by GenoType MTBDRplus Assay

Culture isolates of *M. tuberculosis* grown from clinical samples exhibiting rifampicin resistance by Xpert assay were also tested by reverse hybridization-based GenoType MTBDRplus assay (Hain Lifescience, Nehren, Germany) which detects resistance to rifampicin as well as to isoniazid [2]. Ten rifampicin-susceptible *M. tuberculosis* isolates were also tested for comparison. Genomic DNA from the MGIT 960 system cultures was prepared by using Chelex-100, as described previously [22]. The manual GenoType MTBDRplus assay was performed, and the results were interpreted according to the manufacturer's recommendations and as described in detail previously [23]. Negative controls (water instead of DNA) were included in each run. The GenoType MTBDRplus assay results were also confirmed by PCR amplification followed by DNA sequencing (PCR-sequencing) of hot-spot region of the *rpoB* and *katG* codon 315 regions, performed as described previously [19, 23].

Statistical Analyses

Categorical variables were expressed as absolute number. Statistical analysis was performed using Fisher's exact test and probability levels <0.05 by the 2-tailed test were considered as statistically

significant. Statistical analyses were performed using WinPepi software ver. 3.8 (PEPI for Windows, Microsoft Inc., Redmond, WA, USA) or GraphPad software (GraphPad, La Jolla, CA, USA). The strength of agreement between the different test results was assessed by using the robust kappa statistics. A kappa coefficient (κ) value of <0.2, 0.21–0.4, 0.41–0.6, 0.61–0.8, and 0.81–1.0 indicated slight agreement, fair agreement, moderate agreement, substantial agreement, and almost perfect agreement, respectively [24].

Results

Clinical Specimens

3,995 extrapulmonary specimens collected from 3,995 suspected TB patients were used. Clinical samples, obtained before initiation of anti-TB therapy, included cavity fluids (including pleural fluid, *n* = 1,340; ascitic fluid, *n* = 484; pericardial fluid, *n* = 45, and other body fluids, *n* = 185); fine-needle aspirate (FNA), *n* = 668; pus, *n* = 444; tissue biopsies, *n* = 349; urine, *n* = 302; CSF, *n* = 118; and other miscellaneous specimens including stools, swabs etc., *n* = 60. FNA samples were obtained from lymph nodes, but the source of tissue biopsies and pus samples were not available. Skin pus swabs were included among other miscellaneous specimens.

Smear Microscopy, MGIT 960 System Cultures, Xpert, and PTEC-ET Tests

The results for AFB and MGIT 960 system culture positivity and for molecular detection of *M. tuberculosis* by Xpert and PTEC-ET are presented in Table 1. 95 samples

Table 2. Positivity, sensitivity, specificity, PPV, and NPV of GeneXpert MTB/RIF (Xpert) and ProbeTec ET (PTec-ET) tests for the diagnosis of extrapulmonary TB with MGIT 960 system culture or CRS as a reference standard

Reference	Molecular test	Positives, <i>n</i>	Sensitivity, %	Specificity, %	PPV,* %	NPV,* %
Culture* (<i>n</i> = 403)	Xpert	356	88.33	97.30	65.56	98.67
	PTec-ET	294	72.95	97.80	78.82	96.99
CRS* (<i>n</i> = 489)	Xpert	442	90.39	99.69	95.57	98.67
	PTec-ET	364	74.44	99.74	97.59	96.55

PPV, positive predictive value; NPV, negative predictive value; MGIT, mycobacteria growth indicator tube.
* Culture, MGIT 960 culture; CRS which included MGIT 960 culture and/or clinical diagnosis of TB.

were positive for AFB by microscopy and 403 specimens yielded mycobacterial growth (including 1 nontuberculous mycobacteria, NTM culture) by MGIT 960 system. Among 3,592 culture-negative specimens, 86 samples exhibited evidence of histopathological TB including 18 AFB-positive samples.

Among 95 AFB-positive specimens, Xpert was positive in 94 (98.95%) samples, while PTEC-ET was positive in 90 (94.74) samples, but the difference was not significant ($p = 0.211$). However, among 403 culture-positive specimens, Xpert and PTEC-ET detected *M. tuberculosis* in 356 (88.34%) and 294 (72.95%) specimens, respectively, and the difference was statistically significant ($p = 0.001$). The performance of both Xpert and PTEC-ET was nearly same in AFB-positive, culture-positive and AFB-positive, culture-negative specimens (Table 1). However, in AFB-negative, culture-positive specimens ($n = 326$), Xpert and PTEC-ET detected *M. tuberculosis* in 280 (85.89%) and 220 (67.48%) samples, respectively, and the difference was statistically significant ($p = 0.001$). Although Xpert also detected *M. tuberculosis* in 79 (2.21%) samples while PTEC-ET detected it in 62 (1.73%) samples among 3,574 AFB-negative, culture-negative specimens (including 70 samples with histopathological evidence of TB), the difference did not reach statistical significance ($p = 0.148$) (Table 1).

Using culture as the gold standard, 356 (88.34%) samples were positive by Xpert among 403 culture-positive samples yielding a sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) of 88.34, 97.30, 65.56, and 98.67%, respectively, for the Xpert assay (Table 2). The sensitivity, specificity, PPV, and NPV for the PTEC-ET test were 72.95, 97.80, 78.82, and 96.99%, respectively (Table 2). The difference in the sensitivity values by the 2 tests was statistically significant

($p = 0.001$). Using a composite reference standard (CRS) that included laboratory (culture) and/or clinical (histopathological evidence of infection) diagnosis of TB ($n = 489$), the sensitivity of Xpert was also higher (90.39% positive) than PTEC-ET (364 positives among 489 samples, 74.44%) ($p = 0.001$). The specificity, PPV, and NPV values against CRS were 99.69, 95.57, and 98.67% for Xpert and 99.74, 97.59, and 96.55% for PTEC-ET, respectively (Table 2). The concordance between Xpert and PTEC-ET test results was examined by calculating κ values; the data showed almost perfect agreement (κ , 0.87) among all samples. However, only moderate agreement (κ , 0.46) was seen among MGIT culture-positive samples, while almost perfect agreement (κ , 0.87) was obtained for culture-negative samples.

Performance Comparison of Xpert and PTEC-ET Tests in Different Extrapulmonary Specimens

The results for the detection of *M. tuberculosis* by Xpert and PTEC-ET in various categories of clinical specimens are presented in Table 3. Among culture-positive samples, Xpert detected *M. tuberculosis* in more samples than PTEC-ET in cavitory fluids, urine, CSF, and other miscellaneous specimens, however, the differences were not statistically significant. On the contrary, in FNA/pus/tissue biopsy specimens, Xpert, and PTEC-ET detected *M. tuberculosis* in 283 (93.09%) and 233 (76.64%) samples and the difference was statistically significant ($p = 0.001$) (Table 3). Against culture, the sensitivity and specificity values for cavitory fluids, FNA/pus/tissue biopsy, urine, CSF and other (stool/swab, etc.) specimens were 69.7 and 99.6%, 93.1 and 92.3%, 88.9 and 100%, 100 and 100%, and 75 and 100%, respectively, for Xpert and 64.5 and 99.7%, 76.6 and 93.8%, 77.8 and 100%, 50 and 100%, and 25 and 100%,

Table 3. Comparison of GeneXpert MTB/RIF (Xpert) and ProbeTec ET (PTec-ET) test results with MGIT 960 system culture among various different extrapulmonary specimens

Molecular test result	Cavitary fluids, <i>n</i> = 2,054		FNA/pus/tissue biopsy, <i>n</i> = 1,461		Urine, <i>n</i> = 302		CSF, <i>n</i> = 118		Others (stool and swabs), <i>n</i> = 60	
	culture-positive	culture-negative	culture-positive	culture-negative	culture-positive	culture-negative	culture-positive	culture-negative	culture-positive	culture-negative
Xpert-positive/PTec-ET-positive	47	6	231	70 ^a	7	0	3	0	2	1
Xpert-positive/PTec-ET-negative	6	1	52	19 ^b	1	0	3	0	4	0
Xpert-negative/PTec-ET-positive	2	0	2	2	0	0	0	0	0	0
Xpert-negative/PTec-ET-negative	21	1,971	19 (1 NTM)	1,066	1	293	0	112	2	51
Xpert/culture positivity, %	53 (69.7)	na	283 (93.1)	na	8 (88.9)	na	6 (100)	na	6 (75)	na
PTec-ET/culture positivity, %	49 (64.4)	na	233 (76.6)	na	7 (77.7)	na	3 (50)	na	2 (25)	na
κ coefficient	0.76	0.92	0.34	0.87	0.61	No value	0	No value	0.2	1
κ coefficient	0.92		0.86		0.93		0.66		0.57	

FNA, fine-needle aspirate; NTM, nontuberculous mycobacteria; na, not applicable; CSF, cerebrospinal fluid; TB, tuberculosis; MGIT, mycobacteria growth indicator tube. ^a All 70 samples showed histopathological evidence of TB. ^b 16 samples showed histopathological evidence of TB.

respectively, for PTEC-ET. Against CRS, the sensitivity and specificity values for FNA/pus/tissue biopsy specimens were 94.6 and 99.7%, respectively, for Xpert and 77.7 and 99.8%, respectively, for PTEC-ET. Only fair agreement (κ , 0.34) was apparent between Xpert and PTEC-ET tests for culture-positive FNA/pus/tissue biopsy specimens, while substantial agreement was noted for culture-positive cavitary fluids (κ , 0.76) and urine specimens (κ , 0.61) (Table 3). Among all categories of culture-negative samples, Xpert detected *M. tuberculosis* in more samples than PTEC-ET; however, the differences were not statistically significant, and both tests exhibited almost perfect agreement for most sample types (κ range, 0.87–1.00). Overall, almost perfect agreement was seen between Xpert and PTEC-ET for cavitary fluids, FNA/pus/tissue biopsy and urine specimens (κ range, 0.86–0.93), while lower agreement was seen for CSF and other specimens (Table 3).

Phenotypic and Molecular Detection of Resistance to Rifampicin

Phenotypic DST data showed that only one *M. tuberculosis* isolate was resistant to rifampicin, while the remaining 401 (excluding nontuberculous mycobacteria) isolates were rifampicin-susceptible. The Xpert results were concordant with phenotypic DST data. The GenoType MTBDR_{plus} assay and PCR-sequencing studies performed on rifampicin-resistant and 10 selected rifampicin-susceptible isolates showed the presence of S456W mutation in the *rpoB* gene in rifampicin-resistant isolate (and S315T mutation in *katG* conferring resistance to iso-

niazid; MDR-TB) and wild-type *rpoB* sequence among all 10 rifampicin-susceptible isolates. Interestingly, the culture-positive FNA sample, testing positive for *M. tuberculosis* and its resistance to rifampicin by Xpert, tested negative by PTEC-ET assay.

Discussion

Rapid diagnosis of TB prevents the development of drug resistance and improves outcome, while delayed diagnosis increases illness and death, and incorrect diagnosis causes unnecessary treatment [1]. Routine tests for the diagnosis of EPTB are limited in sensitivity, accuracy, and time to diagnosis typically due to paucibacillary load [6, 12–16]. Although culture is often used as the gold standard for the definitive diagnosis of TB, it has low sensitivity for specimens from extrapulmonary sites, particularly in some types of specimens (e.g., cavitary fluids) [6, 12]. In some extrapulmonary samples with clear histopathological evidence of clinical TB disease, the culture remains negative [14, 16, 25]. Thus, some studies have used a combination of culture and clinical diagnosis of TB [25], or a CRS that includes a molecular diagnostic test, histology, smear, biochemical testing results, signs, and symptoms, or a response to treatment with anti-TB therapy in addition to culture [14]. Xpert has been approved for diagnosis of TB in respiratory specimens (particularly sputum samples); it has also been used with extrapulmonary specimens (off-label use) mainly due to limitations of other available tests for the diagnosis of

EPTB [14–16]. Similarly, PTec-ET is also approved only for respiratory specimens; however, some studies have also evaluated its performance with nonrespiratory specimens [14, 21, 26, 27].

We performed a head-to-head comparison of the performance of Xpert and PTec-ET tests for the diagnosis of EPTB using a large number ($n = 3,995$) of extrapulmonary samples and culture or a CRS comprising culture and/or histopathological evidence of TB as reference. The sensitivity values with respect to smear-positive samples for Xpert and PTec-ET were 98.9 and 94.7%, respectively. With culture as reference, the sensitivity, specificity, PPV, and NPV for the diagnosis of all forms of EPTB were 88.34, 97.30, 65.56, and 98.67%, respectively, for Xpert and 72.95, 97.80, 78.82, and 96.99%, respectively, for PTec-ET. Similarly, the sensitivity with respect to CRS was also higher for Xpert (90.39%) than PTec-ET (74.44%). Again, the specificity values for both the tests were nearly same (99.69% for Xpert vs. 99.74% for PTec-ET). Interestingly, using CRS as reference, the PPV for the diagnosis of EPTB for both Xpert (95.57%) and PTec-ET (97.59%) were nearly same. On the contrary, the PPV was relatively much lower for Xpert (65.56%) than for PTec-ET (78.82%) when culture was used as reference. This is largely due to the detection of several culture-negative specimens with histopathological evidence of active TB disease testing positive by Xpert assay. This phenomenon has also been reported previously with extrapulmonary samples in previous studies [14, 16, 25]. Only one previous study has performed head-to-head comparison of these 2 rapid molecular tests for the diagnosis of EPTB with a small number ($n = 23$) of nonrespiratory samples, and the sensitivity of Xpert observed to be higher (91%) than PTec-ET (65%) with culture as reference [18]. A few studies have also tested nonrespiratory specimens by PTec-ET and reported sensitivity values of 61.1% [21], 71.4% [26], or 100% [27] for the diagnosis of EPTB compared to culture. On the contrary, the Xpert has been extensively evaluated with nonrespiratory samples for the diagnosis of EPTB.

In an early large-scale study, the sensitivity and specificity of Xpert for the diagnosis of EPTB were reported as 81.3 and 99.8% against a reference standard that comprised culture and/or clinical diagnosis of TB [25]. Denking et al. [14] performed a meta-analysis of 18 studies based on 4,461 nonrespiratory samples and reported pooled Xpert sensitivity and specificity values against culture or a CRS comprising culture or a molecular diagnostic test (other than Xpert), histology, smear microscopy, biochemical testing results, clinical diagnosis,

or a response to treatment with anti-TB therapy. Their data showed that the sensitivity varied greatly among different types of specimens making overall estimates of accuracy difficult; however, the pooled sensitivity across all specimen types was 97.4% against smear-positive samples [14]. Our results (Xpert sensitivity of 98.9% against smear-positive specimens) are thus in close agreement with the metadata reported by Denking et al. [14]. Thus, Xpert can rapidly differentiate EPTB from other mycobacterial infections in smear-positive specimens.

In a more recent review based on 66 unique studies (50 studies carried out in low- /middle-income countries) and based on 16,213 specimens, Kohli et al. [16] have also concluded that pooled Xpert sensitivity varies greatly across different specimen types, varying from a low of 31% in pleural fluid to 97% in bone or joint fluid against culture. However, the specificity values, as in our study, were very high (range 79–99%) [16].

The data for various nonrespiratory specimens also showed that Xpert detected *M. tuberculosis* in more samples than PTec-ET including cavitory fluids, FNA/pus/tissue biopsy, urine, CSF, and other miscellaneous specimens, with reference to both, culture or CRS as reference. The Xpert test showed a sensitivity of only 69.7% for cavitory fluids against culture but a higher sensitivity of 93.1% for FNA/pus/tissue biopsy specimens against culture or 94.6% against CRS, while PTec-ET yielded a sensitivity of 64.5% for cavitory fluids and 76.6% for FNA/pus/tissue biopsy specimens against culture or 77.7% for FNA/pus/tissue biopsy specimens against CRS. A meta-analysis of Xpert data has also shown lower pooled sensitivity in cavitory fluids that varied from 50.9% in pleural fluid (27 studies, 4,006 specimens) to 59.2% in peritoneal fluid (16 studies and 712 specimens) to 65.7% in pericardial fluid (7 studies and 324 specimens) against culture [16]. Also, pooled Xpert sensitivity was higher among FNA/pus/tissue biopsy specimens which varied from 97.2% in bone or joint aspirates (5 studies, 385 specimens) to 91.8% in bone or joint tissue (7 studies and 618 specimens) to 87.6% in lymph node aspirate (17 studies and 1,710 specimens) to 78.9% in lymph node tissue (10 studies and 484 specimens) against culture [16]. Lower sensitivity (45%) was also obtained for cavitory fluid (pleural fluid, $n = 47$; ascitic fluid, $n = 18$; and other sterile fluids, $n = 17$) samples, while higher sensitivity of 72% was reported for FNA/pus specimens against culture in 1 PTec-ET study that tested a large number of nonrespiratory specimens [21]. Other studies using PTec-ET test have either used small number of extrapulmonary speci-

mens [18, 26] or the data were not tabulated for individual specimen types [27].

Although the number of positive samples was relatively low, the sensitivity of Xpert was very high for CSF (100%) and urine (89%) specimens with 100% specificity against culture in our study. Kohli et al. [16] have also reported a pooled sensitivity of 71.1% and specificity of 98% in CSF (29 studies and 3,774 specimens) and a pooled sensitivity of 82.7% and specificity of 98.7% in urine specimens (13 studies, 1,199 specimens). On the contrary, PTec-ET yielded a lower sensitivity of 50% for CSF and 77.8% for urine specimens with 100% specificity in our study. A previous study also showed a sensitivity of only 40% for the detection of *M. tuberculosis* by PTec-ET in CSF specimens using TB diagnosis as reference [21]. Other studies using PTec-ET test have either used very few CSF specimens ($n = 2$) [18] or the data were not tabulated for individual specimen types [27]. Taken together, our data showed higher sensitivity of Xpert than PTec-ET for the rapid diagnosis of EPTB, particularly in clinical specimens with histopathological evidence of TB and for the diagnosis of TB meningitis.

The Xpert also detected resistance to rifampicin in 1 specimen which is lower than the reported prevalence (<1%) of MDR-TB in extrapulmonary specimens in Kuwait [9]. However, it may be more useful for settings where MDR-TB is more prevalent, particularly in countries with a high burden of MDR-TB [1]. Both Xpert and PTec-ET tests have recently been updated as Xpert MTB/RIF Ultra and BD MaxTM MDR-TB, respectively [28, 29]. A preliminary evaluation of these 2 tests, mostly in respiratory specimens, found Xpert MTB/RIF Ultra to be more useful than BD MaxTM MDR-TB for the diagnosis of pulmonary TB in patients with paucibacillary load [30], it will be interesting to perform large scale evaluation of these 2 rapid tests for the diagnosis of EPTB and its resistance to rifampicin (and MDR-TB) in different settings.

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Conclusions

The data reported in this study have shown that Xpert detected EPTB in significantly more specimens compared to PTec-ET in both MGIT 960 system culture-positive samples as well as in specimens that yielded culture and/or presented with histopathological evidence of TB (CRS). These differences were particularly pronounced in AFB-negative/culture-positive and AFB-negative/culture-negative/histopathology-positive samples. The performance of Xpert was also uniformly superior compared to PTec-ET in nearly all categories of extrapulmonary specimens. More importantly, Xpert detected TB in all cases of culture-positive CSF specimens for rapid diagnosis of TB meningitis, while the performance of PTec-ET was markedly lower. Thus, Xpert is superior to PTec-ET for the rapid diagnosis of EPTB.

Statement of Ethics

This study was approved by the Health Sciences Center Ethics Committee, Faculty of Medicine, Kuwait University (Approval no. VDR/EC/2947 dated December 12, 2016).

Conflict of Interest Statement

The authors have no conflicts of interest to declare.

Funding Sources

This study was supported in part by Kuwait University Research Sector grant MI 03/16.

Author Contributions

E.M., S.A., and H.S.E. designed the study. H.S.E. performed the experiments and collected the data. E.M. and S.A. analyzed the data and wrote the first draft of the manuscript. All the authors edited and approved the final version of the manuscript.

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