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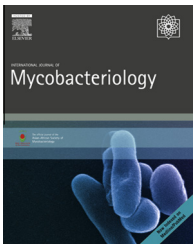

Short Communication

High prevalence of multidrug-resistant tuberculosis among patients with rifampicin resistance using GeneXpert *Mycobacterium tuberculosis*/rifampicin in Ghana

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ABSTRACT

Objective/Background: Drug-resistant strains of tuberculosis (TB) represent a major threat to global TB control. In low- and middle-income countries, resource constraints make it difficult to identify and monitor cases of resistance using drug susceptibility testing and culture. Molecular assays such as the GeneXpert *Mycobacterium tuberculosis*/rifampicin may prove to be a cost-effective solution to this problem in these settings. The objective of this study is to evaluate the use of GeneXpert in the diagnosis of pulmonary TB since it was introduced into two tertiary hospitals in Ghana in 2013.

Methods: A 2-year retrospective audit of clinical cases involving patients who presented with clinically suspected TB or documented TB not improving on standard therapy and had samples sent for GeneXpert testing.

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Results: GeneXpert identified 169 cases of TB, including 17 cases of rifampicin-resistant TB. Of the seven cases with final culture and drug susceptibility testing results, six demonstrated further drug resistance and five of these were multidrug-resistant TB.
Conclusion: These findings call for a scale-up of TB control in Ghana and provide evidence that the expansion of GeneXpert may be an optimal means to improve case finding and guide treatment of drug-resistant TB in this setting.

Introduction

The World Health Organization (WHO) recommends that drug susceptibility testing (DST) and culture be performed for all patients with suspected or known tuberculosis (TB) [1]. This encourages the identification, monitoring, and treatment of cases of drug resistance and has become standard practice in many high-income countries [1]. However, in low- and middle-income countries where TB burden is high and drug resistance constitutes a real threat to TB control, DST and culture are often unavailable or only performed on retreatment cases [1].

TB guidelines for Ghana currently target medium- and high-risk individuals for culture and DST. This includes cases of treatment failure, relapse, or default, contacts of multidrug-resistant TB (MDR-TB), healthcare workers, and patients with human immunodeficiency virus (HIV) [2]. However, resource constraints make it difficult to adhere to these guidelines. For example, in 2014, the incidence of TB infection in Ghana was estimated at 44,000, including 11,000 individuals with HIV coinfection; only 1799 cases were tested for further resistance. As a consequence of the limited availability and use of culture and DST, the true prevalence of MDR-TB in Ghana is not known. Moreover, second-line TB drugs are not always available to those who are identified as having drug resistance, including those with laboratory confirmed MDR-TB [3]. This allows for the ongoing spread of difficult-to-treat TB strains and results in avoidable morbidity and mortality.

Molecular assays such as GeneXpert are changing the landscape of the diagnosis and management of drug-resistant TB and may prove to be a cost-effective solution to this problem in a variety of settings [4–6]. GeneXpert uses real-time polymerase chain reaction to detect the specific sequence for *Mycobacterium tuberculosis* as well as that for rifampicin resistance and can supplement standard diagnostic tools (such as the acid-fast stain, which is the initial test routinely used in Ghana) [7,8]. Since the endorsement of GeneXpert by WHO in 2010, over 110 low- and middle-income countries have purchased GeneXpert, many of which have incorporated it into their diagnostic algorithms for TB [7]. Preliminary data have been promising, showing that more cases of TB are being identified (including MDR-TB), there is a shorter time-to-treatment initiation, and TB diagnosis is becoming decentralized [4,9,10].

This 2-year retrospective audit is the first to evaluate the use of GeneXpert since it was introduced as a diagnostic tool into two tertiary hospitals in Ghana in 2013. These findings call for a scale-up of TB control in Ghana and provide evidence that the expansion of GeneXpert may be an optimal means to improve case findings and guide management of drug-resistant strains in this setting.

Materials and methods

A retrospective audit of TB diagnosis by GeneXpert was conducted from February 2013 to January 2015 at two major hospitals in Ghana: Komfo Anokye Teaching Hospital (KATH) and Korle Bu Teaching Hospital (KBTH). During that time period, all patients with clinically suspected TB or documented TB not improving on standard therapy had samples sent for GeneXpert testing. Demographic data such as referral facility, age, and sex, as well as HIV status and the results of smear microscopy were obtained from patient notes or laboratory request forms. Contingency tables were prepared to determine the sensitivity of smear microscopy using GeneXpert as the reference standard.

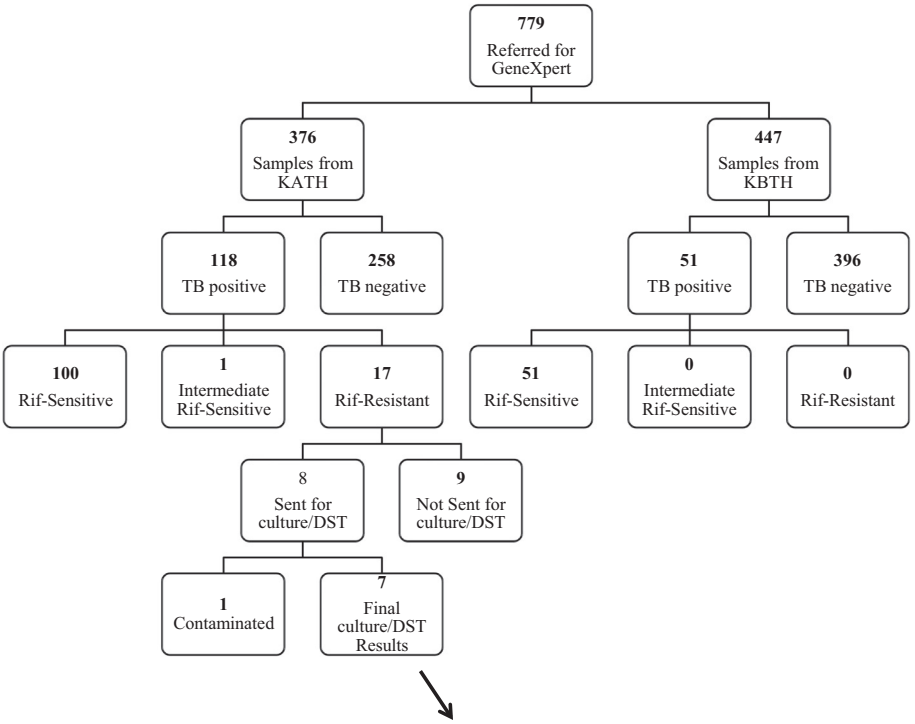
Samples from a subset of patients had been transported to Koforidua Regional Hospital for culture on Lowenstein–Jensen medium followed by DST to rifampicin, isoniazid, ethambutol, and streptomycin. Results were recorded from laboratory forms. These patients were also individually recalled to determine their treatment status and outcome.

This study was conducted in collaboration with the National Tuberculosis Control Program in Ghana. The protocol was approved by the Committee on Human Research, as well as Ethics of the Kwame Nkrumah University of Science and Technology, Kumasi, Ghana and KATH (CHRPE/AP/320/15). Written and verbal consent was obtained from patients who were recalled.

Results

Seven hundred and seventy-nine records were reviewed. Demographic data were available for all patients. Of the 376 cases of suspected TB who presented or were referred to KATH, 118 (31.4%) tested positive for TB by GeneXpert. Of those, 17 (14.4%) were categorized as rifampicin resistant, 100 (84.7%) as rifampicin sensitive, and one (0.8%) as intermediate rifampicin sensitive (Fig. 1).

In many cases, smear microscopy and HIV status were either not documented or the tests were never performed. Smear microscopy results were available for 190 of 376 patients from KATH. For these participants, the sensitivity of microscopy using GeneXpert as the standard was 45.3% (32.8–58.3%; 95% confidence interval). Of the 118 patients positive for TB by GeneXpert, HIV status was known for 22 of



No.	HIV status	DST results				Classification
		Rif	Inh	Emb	Str	
1	NK	R	R	R	R	MDR-TB
2	+	R	R	R	R	MDR-TB
3	NK	R	R	S	R	MDR-TB
4	-	R	R	S	R	MDR-TB
5	NK	R	R	S	S	MDR-TB
6	-	R	S	S	R	Poly-resistant
7	-	S	S	S	R	TB

Fig. 1 – Flow chart of samples tested for tuberculosis with GeneXpert, including antibiotic sensitivity of those determined to be rifampicin resistant. *Note:* DST = drug susceptibility testing; Emb = ethambutol; Inh = isoniazid; KATH = Komfo Anokye Teaching Hospital; KBTH = Korle Bu Teaching Hospital; NK = not known; Rif = rifampicin; Str = streptomycin; TB = tuberculosis; + = positive; − = negative.

them, and eight (36.4%) were HIV positive. Of the eight HIV coinfected patients, smear was available for six; three (50%) of those had negative smears.

Of the total 169 TB positive patients, only the 17 patients with rifampicin resistance had been considered for culture and DST, and eight of them were ultimately sent for testing. Culture and DST were not performed on the remaining patients due to difficulties in transporting the specimens, poor quality of specimens, or because a specimen was never collected. One of the eight specimens was contaminated. Fig. 1 shows the antibiotic sensitivity of the other seven samples. Six demonstrated further resistance: five (71.4%) new cases of MDR-TB and one case of polyresistant tuberculosis. The last sample was rifampicin resistant by GeneXpert but tested rifampicin sensitive by culture.

Three of the seven patients with culture-confirmed resistance were located during follow-up. One patient was cured after completion of second-line drugs, one was in the 10th month of treatment, and the third died while waiting to obtain treatment. The other four patients did not return to the clinic for follow-up and could not be located.

In KBTH, 51 (11.4%) of 447 had a positive GeneXpert result for TB, and all of these were rifampicin sensitive. Of 51 TB-positive patients, HIV status was available for 48 and all were negative. Smear microscopy was not available at KBTH during the time period audited.

Discussion

Currently, WHO estimates that 33% of cases of TB in Ghana are detected, and that 1.7% of new cases of TB and 17% of retreatment cases have MDR-TB [3]. This study provides new evidence that the expansion of GeneXpert would allow cases of drug-resistant TB to be identified, including MDR-TB. It also stresses the dire need for improved follow-up with TB patients and increased availability of second-line drugs to prevent the widespread transmission of drug-resistant TB throughout Ghana.

Sensitivity for smear microscopy in this population was low at 45.3%. A recent Cochrane review found that GeneXpert is more sensitive and specific than microscopy and increases

TB detection by 23% (95% confidence interval: 15–32%; 21 studies, 8880 participants) [11]. The accuracy of initial diagnostic testing is especially important in patients with HIV coinfection, as smears may be negative in up to 61% of these patients [12]. In this cohort, three (50%) patients with TB and HIV were smear negative. These findings reiterate the superiority of GeneXpert over smear microscopy as the initial diagnostic test for TB in this particular population.

GeneXpert identified 17 total cases of rifampicin resistance. Notably, the drug resistance was only found in patients presenting to KATH. This could be a result of several factors, such as higher prevalence of TB in that area or higher rates of HIV. As HIV testing had not been performed for the majority of TB positive patients at KATH, it was not possible to determine the significance of coinfection. In patients with culture results, a high concordance (86%) was observed between rifampicin resistance by GeneXpert and DST. This illustrates the ability of GeneXpert to accurately predict resistance and serve as a proxy for MDR-TB in this population.

A major advantage of GeneXpert is that it allows for the rapid initiation of second-line drugs while awaiting DST and culture [4]. This is the strategy advocated for by the WHO [1]; however, second-line drugs are not always readily available in Ghana. Ideally, all 17 patients with rifampicin resistance should have been started on second-line treatment while their samples were sent for DST and culture. In this cohort, only eight (47.1%) patients with rifampicin resistance had samples sent for DST and culture, and even patients with documented resistance to first-line medications were started or continued on standard TB therapy. Only two of the patients with resistance were able to access second-line drugs. This may reflect a national trend; in the past 2 years, laboratory-confirmed cases of resistance have increased in Ghana, while the number being started on second-line treatment has decreased [3,13]. The current practice of managing drug-resistant TB in Ghana allows for the dissemination of MDR-TB into the community and represents a major threat to TB control.

Of course, incorporating GeneXpert into the diagnostic algorithm for TB in Ghana will present some foreseeable challenges. Ghana would need to adjust its budget for TB to accommodate the installation and running costs of GeneXpert as well as the costs associated with additional culture, DST, and second-line drugs. Programs for MDR-TB treatment would need to be improved, as inadequate support structures result in poor adherence and treatment default [14]. Ultimately, a scale-up of Ghana's MDR-TB management will require a reallocation of TB funding as well as a strategic management plan. Despite these obstacles, this study's initial findings support the feasibility and critical importance of implementing GeneXpert in Ghana.

Additional research in Ghana is needed to better quantify the extent and regional distribution of MDR-TB, the relationship with HIV coinfection, and the prevalence of resistance among new cases compared to retreatment. One of the limitations of this study is that the proportion of resistance in new cases of TB versus treatment failure could not be calculated, as those data were not available for individual patients. In addition, because second-line drugs were not readily available and most patients were lost to follow-up, conclusions could not be drawn about the impact of GeneXpert on treatment outcomes or mortality. There was also a significant amount of incomplete documentation in patients' medical records, either due to human error or the unavailability of tests such as smear microscopy or culture and DST. In spite of these limitations, this study clearly demonstrates the need for continued operational research around TB in Ghana as well as the increased availability of GeneXpert, culture, and sensitivity testing, and second line anti-TB medications.

Conclusion

Ultimately, universal access to diagnosis and treatment of TB, including drug-resistant strains, is an achievable and well-warranted target for Ghana. This requires both the accurate detection of cases and the availability and induction of appropriate drug regimens. This is the first study to demonstrate the potential of GeneXpert *M. tuberculosis*/rifampicin to increase case findings, identify drug resistance, and guide treatment of TB in Ghana.

Conflicts of interest

All authors report no conflicts of interest.

Acknowledgments

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