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GeneXpert contribution in the diagnosis of multi-resistant tuberculosis in new cases at the National University Hospital Centre of Pneumo-Phtisiology of Cotonou (Benin)

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Abstract

The diagnosis of bacteriologically confirmed pulmonary tuberculosis in Benin and in other developing countries relies on the search for acid-fast bacilli through microscopy despite its limited sensitivity. The objective of this study is to evaluate the contribution of the Xpert test in the diagnosis of multidrug-resistant tuberculosis in all new suspects received at the Centre National Hospitalier Universitaire de Pneumo-Phtisiologie de Cotonou (CNHU-PPC) of Benin. The biological material consisted of sputum from new suspected TB patients received at CNHU-PPC during the third quarter of 2018. A total of two thousand three hundred and seventy-five (2375) suspected patients were included in the study, of which 52% were men and 48% were women. The most representative age range was 15-24 years, 28%. GeneXpert was able to diagnose pulmonary tuberculosis in 10.02% of patients, including 9.7% rifampicin-susceptible and 0.3% rifampicin-resistant patients who could not be detected by microscopy. This molecular diagnostic tool is of great value for the diagnosis of multidrug-resistant tuberculosis in new suspects.

Keywords: Tuberculosis; Multidrug-resistant; GeneXpert; Benin

1. Introduction

Tuberculosis prevails in classes of unfavorable socio-economic conditions. A cosmopolitan disease known since the dawn of time (time immemorial), tuberculosis remains a social scourge and a problem of great concern in developing countries. It is the fifth leading cause of death worldwide due to communicable diseases and the second leading cause of death from a single infectious agent [6]

Tuberculosis incidence has been increasing in recent years and particularly in developing countries. The pandemic of HIV/AIDS is one of the main causes of increasing TB cases. Poverty, population growth, migration, economic and social conditions, diabetes, and malnutrition are other related causes to lack of accepted indicators in the disease detection and successful treatment of TB. Furthermore, rates of multi-drug-resistant TB and extensively-drug resistant TB are increasing [11].

Multidrug-resistant tuberculosis remains a public health crisis and a threat to health security. In 2016, the World Health Organization estimates that there will be 600,000 new cases with resistance to rifampicin, the most effective first-line anti-tuberculosis drug. Given the infectious nature of the disease, treatment and control rely heavily on rapid and accurate diagnosis [3]. However, in (developing) resource-limited countries, confirmation of tuberculosis and detection of resistance to anti-tuberculosis drugs is often problematic due to the lack of appropriate equipment and inconsistency

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in carrying out the various confirmatory tests. Nevertheless, the introduction of new tools, mainly molecular biology tools such as the Xpert MTB/RIF or GeneXpert test, approved for use since December 2010 by the WHO, have increased sensitivity and, above all, to shorten the time required to confirm tuberculosis quantitatively, as well as rifampicin resistance [3].

In Benin, the diagnosis of tuberculosis was based on microscopy and culture. In new cases, such tests are slow to diagnose MDR-TB. However, with the recent support from the Global Fund and the Ministry of Health, some TB diagnosis and treatment centres in the 12 departments have been equipped with GeneXpert, which has come to strengthen the technical platform of these Diagnosis and Treatment Centres. The general objective of our study is to evaluate the contribution of this tool in the diagnosis of multidrug-resistant tuberculosis in new cases at the CNHU-PP of Cotonou.

2. Material and methods

2.1. Study location

This was a prospective and descriptive study conducted at the Mycobacteria Reference Laboratory (LRM) of the Centre National Hospitalier Universitaire de Pneumo-Phtisiologie de Cotonou. It is a national laboratory that ensures the diagnosis and follow-up of tuberculosis patients (throughout the national territory) by carrying out several examinations. As a multi-purpose clinical biology laboratory, it supervises the activities of the laboratories of the Tuberculosis Diagnostic and Treatment Centres (CDT) throughout the country, trains laboratory technicians from the microscopy network in both Benin and other sub-regional countries, and conducts research activities focusing on mycobacteria.

Finally, MRI has been involved for several years in monitoring the resistance of bacteria of the "*Mycobacterium tuberculosis* complex" throughout the country.

2.2. Materials

The biological material consisted mainly of sputum from new suspected TB patients received at CNHU-PPC during the third quarter of 2018. Indeed, an exhaustive sampling of sputum from new cases received from July to October 2018 was carried out. A total of 2375 sputum samples were included in this study.

2.3. Methods

2.3.1 Collection and transport of samples

Upon receipt of the patient's request for examination form, a sterile spittoon was given to the patient, followed by strict instructions for sputum collection. Once the sputum was collected, the sample was identified by labelling and then the patient's request for examination form was registered. At this stage, macroscopic examination of the sample was carried out. The samples were then immediately transported to the handling room using a tray with a tightly closed lid. However, in case of delayed handling, the samples were stored at a maximum of 35°C within 3 days and at 4°C between 4 and 10 days.

2.3.2 Samples processing

After following the safeguards for handling a clinical sample under a fume hood, the necessary materials were placed in the fume hood. 2 mL of sputum was transferred into a conical tube with a cap on it using a transfer pipette for the Xpert MTB/RIF test. Alternatively, the entire sample can be processed in the original tube. A quantity of 4 ml of sample reagent was then added to the contents of the conical tube. The mixture was vortexed for at least 10 seconds and incubated for 10 minutes at room temperature. This suspension was again well homogenised for 10 seconds by vortexing and incubated at room temperature for 5 minutes. This preparation is then ready for the Xpert test. For this purpose, the sample identification number was written on the sides of the Xpert MTB/RIF cartridge. After opening the cartridge lid, the tube containing the preparation was opened and the contents were aspirated with the transfer pipette provided to the mark indicated on the pipette. The assembly was then transferred to the sample chamber of the Xpert MTB/RIF cartridge and the cartridge lid was closed tightly. The cartridge was finally loaded into the GeneXpert Dx instrument. The test started within 5 hours following the preparation of the cartridge.

2.3.3 Data collection

The data sources used were bacilloscopy registries for the third quarter of 2018 and the MRI database for suspected new cases. The technique consisted of using a card for each patient on which the sputum identification number, the results of theXpert gene test, and the patients' socio-demographic and clinical data were reported.

2.3.4 Statistical analysis

The data were entered using Microsoft Excel 2010 calculation software and entered into the software's database. A value of $p \leq 0.05$ was considered significant.

3. Results and discussion

Tuberculosis is a chronic infectious disease that remains a major public health problem in Benin, affecting mainly the most disadvantaged sections of the population [4]. New technologies like GeneXpert provide an opportunity for early identification of TB [5]. The overall objective of this study was to evaluate the contribution of GeneXpert in the diagnosis of multidrug-resistant tuberculosis in new cases at CNHU-PPC. From July to September 2018, the Mycobacteria Reference Laboratory of the CNHU-PPC registered 2375 new suspected tuberculosis cases. The male sex was the most represented 52% (Fig.1) with a sex ratio of 1.09 in favour of males and P <1%. This male predominance was found in several other studies in Africa such as: in Senegal 75%, in Marrakech in Morocco 60.3%, in Conakry in Guinea 63.7% etc. [1]. It could be explained by the lifestyle (consumption of alcohol, cigarettes, etc.) observed among men.

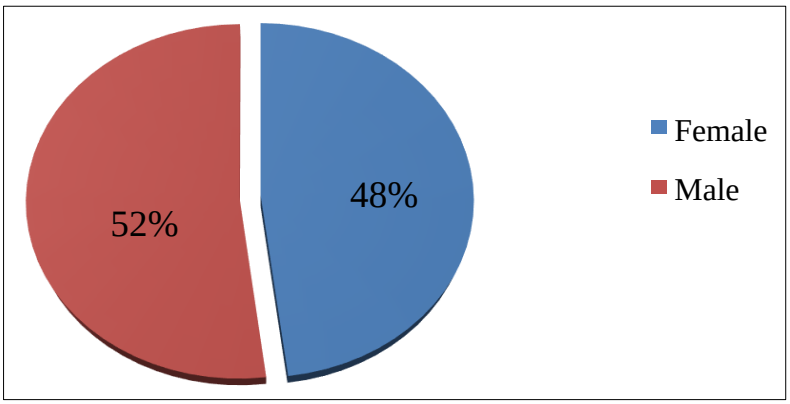

Figure 1 Distribution of suspected cases by gender

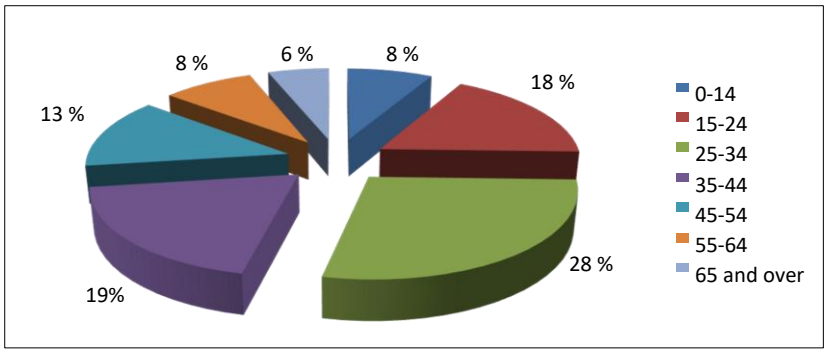

Figure 2 Distribution of suspected cases by age group

A high rate of suspicious patients was observed in the 25-34 age group with 665 cases 28%, followed by far respectively by age groups 35-44 years 19% and 15-24 years 18%. Subjects aged 65and over were less represented (Fig 2.). These two age groups in society are represented by young people who are considered active and highly sociable, which would easily expose them to risk factors [4].

In previous years, MRI encountered difficulties in diagnosing MDR-TB in new suspects because microscopy only confirmed the presence of *Mycobacterium tuberculosis*. However, according to the WHO report in 2016, MDR-TB remains a public health crisis and a threat to health security.

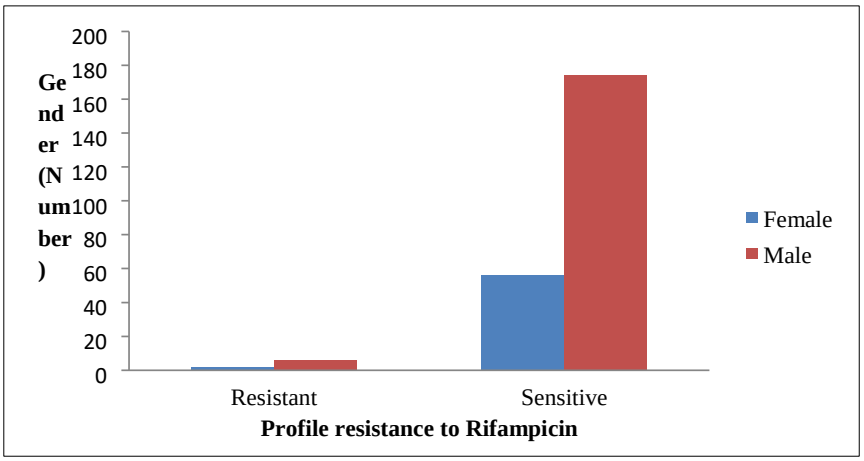

Figure 3 Gender distribution of GeneXpert results

In addition, the identification of *Mycobacterium tuberculosis* species by culture took 2 to 6 weeks followed by tuberculosis drug susceptibility testing for 3 weeks. However, with the Xpert test, tuberculosis is quantitatively detected in addition the genetic mutations associated with resistance to the drug rifampicin in less than 2 hours. Thus, in the present study, 10.0% new cases were identified, of which 9.7% were rifampicin-sensitive and 0.3% were rifampicin-resistant (Fig 3.). A study conducted in Conakry on the introduction of the Xpert system in the diagnosis of multidrug-resistant tuberculosis over a 2-year period found 258 cases of resistance to RIF [1]. This molecular test thus allows rapid and appropriate therapeutic management of MDR-TB cases without waiting for the results of culture and sensitivity tests. The WHO estimates that 600,000 new cases are resistant to rifampicin, the most effective first-line antituberculosis drug [3]. Most studies on GeneXpert in Africa confirm its performance compared to bacilloscopy. In October 2013, a multi centre trial comparing the use of the Genexpert to microscopy concluded that using the Genexpert meant that more patients had a same day diagnosis and same day traitement initiation, but the benefits did not translate into lower TB morbidity [9].

Tuberculosis affects both sexes. Nevertheless, in this study, male patients were the most affected by this disease with 7.3% of susceptible rifampicin and 0.3% of resistant rifampicin compared to 2.4% of susceptible rifampicin and 0.1% of resistant rifampicin for the female sex (Fig 3.). The high rate to this infection in males has also been recently confirmed by a Nepal study [8]. This predominance could be explained by socioeconomic status, demographic characteristics and organizational factors of care structures [4].

Table 1 Xpert test versus microscopy test

	Microscopy	**Xpert Test**
Period of use	T3 2017	T3 2018
MTB detected	232	238
RIF resistance detected 0		8

Moreover, the 25-34 age group was the most affected by tuberculosis with 70 susceptible cases versus 2 resistant cases. It is followed by the 35-44 age group with 52 susceptible cases for 4 resistant cases (Figure4.). These age groups, representing the young population of African nations, are much more affected by this disease. Therefore, tuberculosis is considered a young adults disease [2].

GeneXpert is of great value in the diagnosis of tuberculosis, specifically multidrug-resistant tuberculosis in new cases. It is a secure, automated, easy-to-use test that is fast and reliable. Although a comparative study has not been done with microscopy in this study, the performance and the sensitivity of the xpert test has been emphasized by several autheurs. The great contribution in our context is the ability of the genexpert to detect cases of resistance to rifampicin in record time.The introduction of GeneXpert in the diagnosis of tuberculosis in Benin has led to the detection of cases of rifampicin resistance that would not have been detected by microscopy (Table 1).

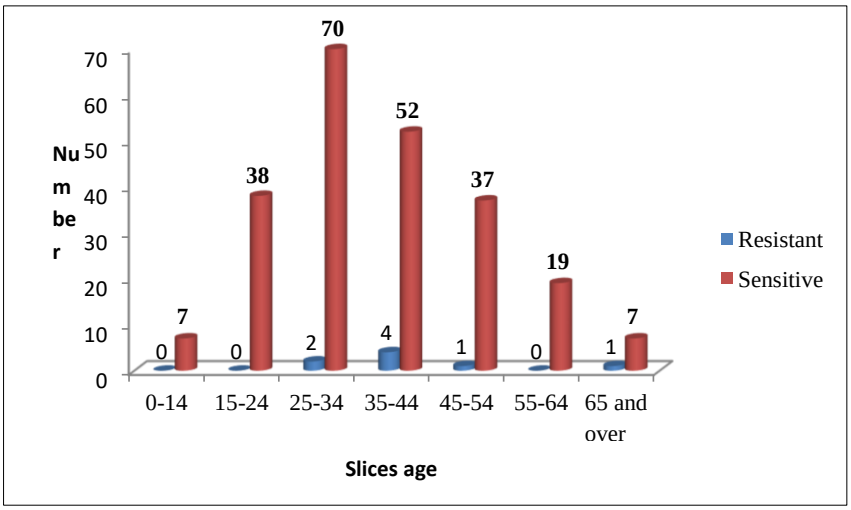

Figure 4 Age Distribution of GeneXpert Results

4. Conclusion

This study allowed us to diagnose cases of multidrug-resistant tuberculosis in a few suspected patients. It will contribute to early detection of tuberculosis in all its forms through the deployment of the geneXpert tool in all CDTs.

Compliance with ethical standards

Disclosure of conflict of interest

There is no conflict of interest declared.

Statement of informed consent

Informed consent was obtained from all individual members of this study

References

[1] Anagonou S, Ade G, Affolabi D, et al. Guide du Programme National contre la Tuberculose:4th edition Benin. 2017; 14-17.

[2] Canu A. et al. Chapter 4: Microbiology in Le préparateur en pharmacie 2nd edition. Éditions Lavoisiertech et doc. 2013; 273-360.

[3] Compendium of WHO guidelines and associated standards: Ensuring optimal delivery in the chain of care for tuberculosis patients. Second edition - June 2018.

[4] Francis V, et al. Tuberculosis: a practical guide for doctors, nurses, laboratory technicians and health care workers. Edition Partners in Health. 2014; 307.

[5] Lawn DD, Nicol MP. Xpert® MTB/RIF assay: development, evaluation and implementation of a new rapid molecular diagnostic for tuberculosis and rifampicin resistance. Future Microbiol. Sep 2011; 6(9): 1067-1082.

[6] Molimard M, et al. Tuberculosis: a new priority. Presse Med. 1994; 23: 1166-1171.

[7] Ninnet B, et al. New tests for the diagnosis of tuberculosis. Rev Mal Resp. 2011; 28(6): 823-833.

[8] Priyatam Khadka, Januka Thapallya, Basista Parsad Rijal. Diagnosis of tuberculosis rom smear-negative presumption TB cases using Xpert MTB//Rif assay: a cross-sectional study from Nepal. BMC Infectious Diseases. 2019; 19.

[9] Theron G. Feasibility accuracy, and clinical effect of point-of-care Xpert MTB /RIF testing for tuberculosis in primary-care settings in Africa: a multicentre, randomised, controlled trial, The Lancet. 2013.

[10] WHO: Global Tuberculosis Report. 2016. Geneva: World Health Organization; 2016