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Mycobacterium tuberculosis genotype Beijing: About 15 strains and their part in MDR-TB outbreaks in Algeria

Malika Ifticene *, *Fatma Zohra Gacem, Djamel Yala, Fadela Boulahbal*

Laboratory of Tuberculosis and Mycobacteria, Pasteur Institute of Algeria, Algeria

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ABSTRACT

Within the framework of epidemiological surveillance by molecular typing tools conducted in the NRL on multi-resistant strains, the application of spoligotyping on a group of 390 strains consisting of 389 DR-MTB strains and 1 susceptible strain isolated from patients made it possible to detect the presence of 15 strains belonging to the Beijing genotype. All 15 strains were genotyped by MTBDR*plus*. Among the 15 strains, 11 were typed by RFLP and 9 by MIRU-VNTR.

After analysis of the profiles obtained by the software MIRU-VNTRplus, two spoligotypes (st No. 1 and st No. 265) and four RFLP profiles and a complete identity profile by MIRU-VNTR, information collected on patients allowed the research team to detect a family tie among patients of three different families, as well as one nosocomial TB transmission.

The percentage of Beijing strains found among the patients in this study was 3.8%; this figure does not reflect the reality because it was calculated from an essay on MDR-TB.

To get an idea of the prevalence of Beijing TB strains in this country, a more extensive study is currently being considered.

Introduction

The distribution of multidrug-resistant tuberculosis (TB) in general is unknown, because the majority of countries are not equipped with diagnostic laboratories and only a minority of laboratories are controlled by the supranational laboratories. The dissemination of strains, especially multidrug-resistant strains, is a real obstacle that compromises both the effectiveness of treatment and the efforts of the programs against TB.

Several epidemics associated with multidrug-resistant strains have been described worldwide, but the most important are those related to the Beijing strains. The Beijing genotype is widespread, especially in Asian countries, where it represents more than 80% of isolated strains and more than 40% in other countries [1–6].

The power of mutation in the genomic regions involved in the resistance, the rapid evolution of resistance, the low immune response related to their adaptation to the host, compared with other genotypes, gives these strains a particularly virulent character [7]. Therefore, many studies have been conducted towards the identification of factors that promote their dissemination and mechanisms that allow them to adapt to the host and to its defense mechanisms [8].

Since the introduction of a molecular typing tool in the laboratory used by the researchers, the existence of 15 Beijing genotype strains have been identified in a batch of 390 strains of MTB isolated between 1997 and 2011.

* Corresponding author. Address: National Tuberculosis Reference Laboratory, Pasteur Institute of Algeria, Route du Petit Staoueli, Dely Brahim 16000, Algeria. Tel./fax: +213 21 67 35 22; mobile: +213 773 971 281.

E-mail addresses: ifticene.ma@yahoo.fr (M. Ifticene), fatmazohragacem@yahoo.fr (F.Z. Gacem), djamyala@yahoo.fr (D. Yala), fadiboulboul@yahoo.fr (F. Boulahbal).

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Materials and methods

Materials

Within the framework of an epidemiological survey on multi-drug-resistant tuberculosis (MDR-TB), 389 MDR-TB strains of *tuberculosis* were studied by spoligotyping. This technique allowed the researchers to identify the existence of 14 Beijing genotype strains. During the course of the research, a strain of MTB genotype Beijing was isolated from a Chinese expatriate in Algeria in a professional framework. This latter strain was susceptible to TB drugs unlike 14 strains isolated in Algerian patients.

Geo-demographic information of patients with Beijing genotype strains such as age at diagnosis, sex, geographical origin or the setting of diagnosis, date of diagnosis, treatment history and outcome of treatment were obtained from the patients' files and the laboratory register, as well as some of the information provided by the treating physician. These data are shown in Table 1.

Methods

Determination of the drug susceptibility

A microscopic examination well as a culture on solid medium was performed on the samples. The study of strain susceptibility to anti-TB drugs was performed by two methods: the proportion method on solid media (Isoniazid, Rifampicin, Ethambutol, Streptomycin, Kanamycin and Ofloxacin) and the molecular biology method as MTBDR*plus* of HAIN [9,10].

Genotyping of strains

The 15 strains were typed by spoligotyping [11,12]. The RFLP technique [13,14] was performed on 11 strains and the MIRU-VNTR technique [15] has been applied on 9 strains owing to lack of culture material.

Results

Geo-demographic characteristics of patients

Among the 390 strains which were studied by spoligotyping, 15 Beijing genotype strains have been identified. All patients presented with pulmonary TB, including 4 females and 11 males. The age range was between 19 and 58 years.

Of the 15 patients, 2 were cured under treatment (P1, P7), 1 of the 2 in combination with surgical treatment (P1); 4 patients (P6, P10, P13, P14) lost their sight; and the follow-up of the disease was fatal after an average of 2 years for 8 patients (P2, P3, P4, P5, P8, P11, P12, P15) – a 53% death rate.

Regarding the origin of the patients, all patients harboring the Beijing strain were diagnosed in the center region of Algeria covering an area of approximately 200 km; 4 (P10, P12, P13 and P15) are from Dellys, a region located in the province of Tizi Ouzou about 90 km east of Algiers. One patient (P11) is from Miliana, located 60 km south of Algiers. Six patients (P1, P2, P3, P4, P5 and P6) are from Blida, 40 km west of Algiers. Three patients are from Algiers (P7, P8 and P14). The 15th patient (P9) is from China, arrived in Algeria in a professional setting and whose disease was diagnosed in a TB center of Algiers.

Links between the 15 patients

A family tie was found between 3 patients from Blida: P1, P2 and P3. Four other cases were identified among members of two families; one family in Algiers (P7, P8), and the second one in Dellys (P12, P13). A case of nosocomial TB infection (P4) has been identified. It concerns the nurse of the care unit attended by P2.

Drug susceptibility testing

The drug susceptibility testing performed on the 15 strains showed that those isolated in the Algerian patient are resistant to at least RH (study material of investigation MDR-TB).

Table 1 – Geo-demographic data of patients.

Patient No.	Age	Sex	Origin and date of diagnostic	Treatment history	Patient outcome and relationship with other patients of the group
P1	21	F	Blida, 2000	UT	Cured after lobectomy (niece of P2 and P3)
P2	39	M	Blida, 1997	UT	Deceased (uncle of P1)
P3	24	F	Blida, 1998	UT	Deceased
P4	28	F	Blida, 1999	UT	Deceased (nurse in the healthcare center of P2)
P5	42	M	Blida, 2000	UT	Deceased
P6	29	M	Blida, 2002	T	Loss of sight since 2003
P7	22	F	Algiers, 2002	UT	Cured (sister of P8)
P8	24	M	Algiers, 2004	UT	Deceased on post surgical in 2012 (operation on the affected lung)
P9	19	M	China, 2002	UT	Expatriate, returned to China immediately after diagnosis
P10	24	M	Dellys, 2004	UT	Loss of sight since 2005
P11	58	M	Miliana, 2002	T	Deceased
P12	45	M	Dellys, 2004	T	Deceased (brother of P13)
P13	27	M	Dellys, 2007	UT	Loss of sight since 2008
P14	54	M	Algiers, 2003	UT	Loss of sight since 2004
P15	35	M	Dellys, 2001	T	Deceased

H: Isoniazid, S: Streptomycin, R: Rifampicin, E: Ethambutol, M: male, F: female, T: treated, UT: untreated.