There is significant relationship between Beijing genotype family strains and resistance to the first-line anti-tuberculosis drugs in the Iranian population

Masoud Keikha\textsuperscript{a,b,c}

\textsuperscript{a} Antimicrobial Resistance Research Center, Mashhad University of Medical Sciences, Mashhad, Iran
\textsuperscript{b} Department of Microbiology and Virology, Faculty of Medicine, Mashhad University of Medical Sciences, Mashhad, Iran
\textsuperscript{c} Student Research Committee, Mashhad University of Medical Sciences, Mashhad, Iran

Dear Editor,

Tuberculosis is one of the most important global concerns, as it is considered as the second leading cause of death due to the infectious diseases after HIV. According to the WHO reports in 2020, it is estimated that around 10 million people have affected to TB and 1.5 million deaths occurred due to that in 2019 \cite{1,2}.

Iran is also one of the countries in the Middle-East region with a population of about 82 million; incidence of TB in Iran is estimated at about 11,000 people. Tuberculosis control and monitoring in Iran is very important due to its proximity to the countries such as Azerbaijan and Armenia (both high MDR-TB) and Afghanistan and Pakistan (with high-TB burden) \cite{1-3}. One of the most important strategies in designing the regional programs for the control and prevention of tuberculosis is the identification and tracking of community circulating Mycobacterium tuberculosis (Mt\textit{b}) strains by the molecular epidemiology techniques \cite{3,4}.

According to the literature review, the most common \textit{Mt\textit{b}} families in Iran are Beijing (Beijing / W), Haarlem (Haarlem I / HaarlemII and New-1), Delhi/Cas, East-Asian Indian, while TUR, Ural, LAM and bovis are less separable \cite{5,6}.

Evidence has been suggested in recent years about the relationship of drug resistance and some \textit{Mt\textit{b}} lineages \cite{5-7}. Thus, this study was performed aiming to determine the frequency of dominant lineages in Iran to evaluate the possible relationship of each of the Beijing, Delhi/ Cas, Haarlem and EAI lineages with the first-line drug resistance, especially isoniazid, rifampin and MDR-TB (Multi-drug resistant TB).

In order to fulfil the study, we attempted to investigate all the required studies of the Iranian population by searching PubMed, Scopus, Embase, Cochrane library, Google Scholar and Iranian databases Iranmedex, SID and ISC by March 2020. The search was done with the MeSH keywords including “Mycobacterium tuberculosis”, “TB”, “drug-resistance”, and “genotyping”. The inclusion criteria included: 1- Studies involving the prevalence of our genotypes in susceptible isolates and drug resistance, and 2- Studies in which genotyping was determined by the standard techniques of Spoligotyping, MIRU-VNTR, IS6110 RFLP, and Whole genome sequencing (WGS); and case reports, repetitive sample studies with, and nonclinical studies were included. Finally, the probable relation between the infection and each of the Beijing, Delhi/Cas, Haarlem and EAI genotypes with first-line drug resistance was measured by the Odds Ratio (ORs) with 95% confidence intervals and the Egger weighted regression method was also determined \cite{8}.

Based on our investigation, 28 original articles including Persian and English articles were included in the present quantitative analysis (Table 1). In the present study, 6671 \textit{Mt\textit{b}} strain data were evaluated. The studies were limited to 2006–2019 and were conducted in the areas including Tehran (National Research Institute of Tuberculosis and Lung Disease), Tabriz, Golestan, Kermanshah and Mashhad.

The studied \textit{Mt\textit{b}} strains included two groups of susceptible and drug-resistant TB, with drug-resistant TB strains being resistant to the antibiotics such as isoniazid, rifampin, streptomycin, ethambutol and pyrazinamide.

In general, the frequency of Beijing (22.5%), Delhi/Cas (66.7/66.7%), Haarlem (264/60.2) and EAI (223/514.5%) lineages were evaluated, while the distribution of lineages in first-line drug resistant strains were evaluated as Beijing (76.2%/398%), Delhi/Cas (19 / 129.5%), Haarlem (29/17.1%) and EAI (16/81%). The distribution of \textit{Mt\textit{b}} lineages between susceptible and drug-resistant TB is demonstrated in Fig. 1.

Based on statistical analysis by our random effects model, a significant relationship was observed between the infection with \textit{Mt\textit{b}} Beijing strains and drug resistance (ORs: 1.54 (with 95% CIs); 0.7–3.5; Q = 35.94; F = 61.04; P = 0.02). However, no significant relationship was observed between the infection with Delhi/Cas families (0.17; 0.11–0.22; Q = 131.4; F = 84.0; P = 0.00), Harrlem (0.18; 0.1–0.2; Q = 126.5; F = 84.9; EAI (0.15; 0.0–0.2; Q = 70.3; F = 81.5; P = 0.00) and drug resistance. Moreover, there was a significant relationship between first line drugs with \textit{Mt\textit{b}} Beijing strains and resistance to isoniazid and rifampin (2.19; 1.0–4.2; Q = 77.6; F = 88.4; P = 0.027 and 1.60; 0.75–3.41; Q = 59.1; F = 89.84; P = 0.2 respectively). Thus, according to the present meta-analysis results, the Delhi/Cas, Haarlem, and Beijing families were the three most common lineages in the tuberculosis patients in Iran, respectively, while Beijing

---

\textit{E-mail address: keikham971@mums.ac.ir.}

https://doi.org/10.1016/j.jctube.2020.100161

2405-5794/ © 2020 Published by Elsevier Ltd. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/BY-NC-ND/4.0/).
been several studies, so far, on the relationship of the infection with increasing MIC relative to the anti-tuberculosis drugs [42]. There have and presence of multiple SNPs in the resistance [39-41]. Based on WGS (whole genome sequencing) data, the relationship between the Beijing TB genotypes and drug resistance [37,38]. The incidence of Beijing genotypes in global distribution today [37,38]. The incidence of Beijing genotypes in East Asia but has worldwide distribution and was first introduced by Soolingen et al. (1995) as a predominant tuberculosis genotype of East Asia but has worldwide distribution and global distribution today [37,38]. The incidence of Beijing genotypes in recent years has been estimated at between 50% (East Asian countries) to 8.9% (Western countries) [6,38]. According to the review of the literature, the Beijing genotype is one of the predominant tuberculosis family in the Middle-East countries, especially in Iran [5,6,39]. Based on our analysis, the Beijing genotypes were the third most common genotype in Iran. Numerous studies have been published in recent years on the relationship between the Beijing TB genotypes and drug resistance [39-41]. Based on WGS (whole genome sequencing) data, the presence of multiple SNPs in the katG, rpoB, embB, rpsL, rrs, eis, gyrA and gyrB genes compared to other genotypes could be effective in increasing MIC relative to the anti-tuberculosis drugs [42]. There have been several studies, so far, on the relationship of the infection with Beijing genotypes and drug resistance. However, some other studies contradict this phenomenon [30-45]. According to Ramazanzadeh et al. (2014) about 80% of drug resistant Mtb isolates from the Iranian patients belonged to the Beijing genotype. Also, in the study by Tarashi et al. (2017), amongst the MDR-TB isolates of Iranian patients the frequency of Beijing and Haarlem genotypes were 19.3% and 18.7%, respectively [5,6]. We also showed in this study that the Beijing genotype was the third most common genotype in the Iranian tuberculosis (TB) patients, and the infection with the Beijing family had significant relationship with the resistance to isoniazid and rifampin. Farnia et al. (2006) showed that Haarlem I and Beijing genotypes are the most common TB lineages of the Afghan population residing in Iran, who are affected to MDR-TB [10].

Other similar studies have also found that the Beijing genotype has been isolated from a significant population of TB patients in Iran neighbouring countries, especially Afghanistan and Pakistan. Hence, in order to prevent the spread of Mtb Beijing family infection, it seems that designing the guidelines has a significant impact on reducing the DR-TB cases in the Iranian population [5,6,39].

**Ethical**

Ethical Statement is not applicable for this manuscript.

**References**

[1] Keikha M, Esfahani BN. The relationship between tuberculosis and lung cancer. Adv Biomed Res 2018;7:8–11.

[2] World Health Organization. Global tuberculosis report 2019. World Health Organization; 2020.

[3] Zaniani FR, Moghim S, Mirhendi H, Safaei HG, Fazeli H, Salehi M, et al. Genetic lineages of mycobacterium tuberculosis isolates in Isfahan, Iran. Curr Microbiol 2017;74(1):14–21.

[4] Supply P, Allix C, Lesjean S, Cardoso-Oelemann M, Rüsch-Gerdes S, Willery E, et al. Proposal for standardization of optimized mycobacterial interpersed repetitive unit-variable-number tandem repeat typing of Mycobacterium tuberculosis. J Clin Microbiol 2006;44(12):4498–510.
Shari Haeili M, Darban Jafarian M, Farnia P, Kargar M, Aghalimerza M, Ramazanzadeh R, Ahmadi M, et al. Merza MA, Farnia P, Salih AM, Masjedi MR, Velayati AA. The most predominant Mozafari M, Farnia P, Jafarian M, Razavi Deligani M, Kazempour M, Masjedi M, Keikha J Clin Tuberc Other Mycobact Dis 19 (2020) 100161 Amirmozafari N, Ramezanzadeh R, Farnia P, Ghazi F. The frequency of Beijing Farnia P, Masjedi MR, Nasiri B, Mirsaedi M, Sorooch S, Kazeampour M, Velayati AA. Diversity of Mycobacterium tuberculosis isolates from immigrant, Pakistani and Turkish tuberculosis patients: a comparative analysis. Indian J Med Microbiol 2010;58(2):123-32. Ahmad MO, Farnia P, Tajeddin E, Tabarsi P, Baghaei PA, Masjedi MR, Velayati A. Mycobacterium Tuberculosis Complex Strains Identification with Spoligotyping Method in Patients Attending to Masih Daneshvari Hospital. J Adv Med Biomed Res 2009 Aug 10;17(67):23-32. Mozafari M, Farnia P, Jafarian M, Razavi Deligani M, Kazempour M, Masjedi M, Velayati AA. Comparison of Mycobacterium tuberculosis Beijing genotype with other Mycobacterium tuberculosis strains Using MRU-VNTR method. ISMU 2012;15(1):1-2. Merza MA, Farnia P, Salih AM, Masjedi MR, Velayati AA. The most predominant spoligotypes of Mycobacterium tuberculosis isolates among Iranian, Afghan-immigrant, Pakistani and Turkish tuberculosis patients: a comparative analysis. Chemotherapy 2010;56(3):248-57. Jafarian M, Farnia P, Kargar M, Aghalimerza M, Ramazanzadeh R, Ahmad MO, et al. Study of genetic diversity of M.tuberculosis strains isolated by MIRU-VNTR technique in Isfahan province of Iran. Adv Biomed Res 2018;7:1. Afaghi GA, Moaddab SR, Darbouy M, Ansarini K, Banisaffar M, et al. Genotypic diversity of multi-and extensively drug-resistant Mycobacterium tuberculosis isolates in professor of medicine, Iran. Adv Biomed Res 2018;7. Vaziri F, Khol TA, Ghasvand H, Kamalaki M, Merker M, Hadifar S, et al. Genetic diversity of multi-and extensively drug-resistant Mycobacterium tuberculosis isolates from patients with tuberculosis in East Azerbaijan center by MIRU-VNTR. J Microbial World. 2009;11(4):320-31. Van Soolingen D, Qian L, De Haas PE, Douglas JT, Traore H, Portela F, et al. Predominance of a single genotype of Mycobacterium tuberculosis in countries of east Asia. J Clin Microbiol 1995;33(12):3234-8. Toumpoulou OK, Sotiriadis P, Markoupolou E, Zissimopoulos A, Voskoglou-Nomikos T. Polymorphisms of the rpoB gene in Mycobacterium tuberculosis strains from Greece and India. J Antimicrob Chemother 1993;32(2):217-21. Yuan L, Huang Y, Mi LG, Li YX, Liu PZ, Zhang J, Liang HY, Li F, Li H, Zhang SQ, Li AH, et al. Characterization of Mycobacterium tuberculosis Beijing genotype using Spoligotyping and SNPs. Microbiologyopen 2013;2(6):988-96. Tanveer M, Hasan Z, Siddiqui AR, Ali A, Kanji A, Ghebrimrechel et al. Genotyping of drug resistant or drug-susceptible strains of Mycobacterium tuberculosis in Pakistan. BMC Infect Dis 2008;8(1):171. Hoffner S, Sahebi L, Ansarini K, Sabour S, Mohajeri P. Mycobacterium tuberculosis of the Beijing genotype in Iran and the World Health Organization Eastern Mediterranean Region: a meta-analysis. Microbiol Drug Res 2018;24(6):693-8. Sun H, Zhang C, Xiang L, Pi R, Guo Z, Zheng C, Li S, Zhao Y, Tang K, Luo M, Rastogi N. Characterization of mutations in streptomycin-resistant Mycobacterium tuberculosis isolates in Sichuan, China and the association between Beijing-lineage and dual-mutation in gdh. Tuberculosis 2016;96:102-6. Nhu NT, Lan NT, Phuoc NT, van V, Chai N, Farrar J, Caws M. Association of streptomycin resistance mutations with level of drug resistance and Mycobacterium tuberculosis genotypes. Int. J. Tuberculosis Lung Dis 2015;19(1):31-9. Yuan L, Huang Y, Li LL, Li YX, Liu PZ, Zhang J, Liang HY, Li F, Li H, Zhang SQ, Li WJ. There is no correlation between sublineages and drug resistance of Mycobacterium tuberculosis Beijing-W lineage clinical isolates in Xinjiang, China. Epidemiol Infect 2015;143(1):141-9. Li Y, Cao X, Li S, Wang H, Wei J, Liu P, et al. Characterization of Mycobacterium tuberculosis isolates from Hebei, China: genotypes and drug susceptibility phenotypes. BMC Infect Dis 2016;16(1):107. Liu Y, Jiang X, Li W, Zhang X, Wang W, Li C. The study on the association between Beijing genotype family and drug susceptibility phenotypes of Mycobacterium tuberculosis in Beijing. Sci Rep 2017;7(1):1-7.