UNIVERSITI TEKNOLOGI MARA

VARIANT ANALYSIS OF MULTI-DRUG-RESISTANT *Mycobacterium tuberculosis* CLINICAL ISOLATES

NORZULIANA
BINTI ZAINAL ABIDIN

MSc

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AUTHOR'S DECLARATION

I declare that the work in this thesis was carried out in accordance with the regulations of Universiti Teknologi MARA. It is original and is the results of my own work unless otherwise indicated or acknowledged as referenced work. This thesis has not been submitted to any other academic institution or non-academic institution for any degree or qualification.

I, hereby, acknowledge that I have been supplied with the Academic Rules and Regulations for Post Graduate, Universiti Teknologi MARA, regulating the conduct of my study and research.

Name of Student	:	Norzuliana Binti Zainal Abidin
Student I.D. No.	:	2017408544
Programme	:	Master of Science (Molecular Biology) – AS753
Faculty	:	Applied Sciences
Thesis Title	:	Variant Analysis of Multi-Drug-Resistant Mycobacterium tuberculosis Clinical Isolates.
Signature of Student	:	……..
Date	:	30 August 2021

ABSTRACT

Tuberculosis (TB) is a deadly infectious disease. One of the infection sources is the transmission of the bacilli of *Mycobacterium tuberculosis* (Mtb) between person to person through inhalation of contaminated air droplets aerosolized from a TB carrier. Unfortunately, the emergence of drug-resistant *Mycobacterium tuberculosis* (DR-Mtb) strain has posed concern in fighting the spread of TB. A total of twenty-four local clinical *Mycobacterium tuberculosis* complex (MTBC) isolated from patients diagnosed with TB in 2017 were sequenced using the next-generation sequencing. The whole-genome assembly and annotation were completed using an in-house developed bioinformatic pipeline. The mutation sites were identified and compared to the existing databases. Genomes with an average of 99.66% completion and 66× coverage were successfully assembled. Sequencing of the local clinical MTBC genomes revealed two species of *Mycobacterium*, *Mycobacterium tuberculosis* and *Mycobacterium bovis*. Besides that, from the *in silico* profiling of the MTBC lineage diversity, a total of five MTBC lineages were discovered, lineage 1 (n=11), lineage 2 (n=8), lineage 3 (n=3), lineage 4 (n=1) and *bovis* strain (n=1). The majority of the MTBC isolates were closely related to lineage 1 (Indo-Oceanic strain) and lineage 2 (Beijing strain), where the latter is highly associated with the DR-Mtb type. Twenty-six (26) single nucleotide variants (SNVs) that confer resistance to isoniazid (*fabG1, inhA, katG* and *kasA*), rifampicin (*rpoB*), ethambutol (*embB*), streptomycin (*rpsL* and *rrs*), pyrazinamide (*pncA*) and fluoroquinolones (*gyrA*) were profiled. Based from the crosscheck between the phenotypic DST (Phe-DST) and the next-generation sequencing-based drug susceptibility profile (NGS-based DSP), there are discrepant in drug susceptibility testing (DST) profiles which resulted in differences in characterizing the DR-Mtb types. While the microbial genome-wide association studies (mGWAS) of the local Mtb genomes, a total of twenty-four (24) novel variants with predicted deleterious effects and significantly associated with anti-TB drug resistance were determined and studied. The identified novel variants focused in this study were found on genes that functional for the growth, survival, and pathogenicity of *Mycobacterium tuberculosis* complex TB infection.

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