

I. INTRODUCTION

1.1 Background

Mycobacterium tuberculosis (MTb) is an obligate aerobic bacillus bacterium that causes a persistent disease known as tuberculosis (TB). Indonesia is second place for TB incident cases worldwide in 2021; additionally, there has been an increasing trend on TB prevalence with age in Indonesia in the population older than 15 years old (Noviyani et al., 2021; WHO, 2022). The MTb bacteria possesses a virulence factor on its atypical cell wall structure that forms strong impermeable barriers towards harmful drugs, causing intricate treatment options (Delogu et al., 2013). According to Getahun et al. (2015), MTb has the ability to switch between replicating and latent states, which increases the disease burden of drug-resistant tuberculosis (DR-TB) as the mycobacterium may be untreated by several antimycobacterial drugs.

The gold standard in detecting DR-TB still relies on culture-based drug susceptibility testing (DST) that needs a long period of time or through whole genome sequencing (WGS) that requires high investments (Zhao et al., 2022). Furthermore, there has been research that shows the resistance mechanisms of DR-TB are associated with several gene mutations; this leads to the resistance towards different antimycobacterial drugs. These two overlapping problems increase the hurdle in giving the most suitable treatment for each patient; hence, there is an urgent need to develop faster diagnostic methods in detecting gene mutations in DR-TB patients. With the advancements in sequencing technology, Oxford Nanopore Technology (ONT) offers real-time sequencing with rapid processing time that can be utilized in detecting DR-TB (Hall et al., 2023; Magdy et al., 2020). Aligned with that, there has not been any evaluation on the most prevalent DR-TB present in Jakarta; thus, by using ONT,

this research aims to genetically profile TB samples in detecting the most prevalent DR-TB type with its advancements in the sequencing protocol.

1.2 Research Aim

The aim of this research is to investigate the types and prevalence of drug-resistant tuberculosis from samples in Jakarta and West Java using Oxford Nanopore Technology sequencing.

1.3 Scope of Research

The scope of this research includes:

- 1) *Mycobacterium tuberculosis* DNA extraction using Phenol:Chloroform:Isoamyl alcohol method
- 2) Enrichment by PCR using provided primers from ONT
- 3) Library preparation using specialized Rapid Barcoding Kit 96 from ONT
- 4) Third Generation Sequencing using GridIon (Mk1) from Oxford Nanopore Technology

1.4 Hypothesis

The use of Oxford Nanopore Technology enables a more comprehensive and specific detection of drug-resistant tuberculosis strains in Jakarta and West Java compared to its gold-standard method, while allowing a better understanding on its distribution in the region.