ABSTRAK

Latar Belakang Kenaikan kasus *Multi Drug Resistant/Rifampicin Resistant-Tuberculosis* (MDR/RR-TB) menyebabkan adanya kebutuhan obat antimikobakterium baru. Indonesia merupakan negara dengan biodiversitas yang besar, namun ekplorasi *Actinomycetes* Indonesia untuk isolasi senyawa antimikobakterium masih belum banyak dilakukan. **Metode** Penelitian ini melakukan eksplorasi sistematik terhadap *Actinomycetes* Indonesia untuk menghasilkan isolasi senyawa antimikobakterium baru. Eksplorasi tersebut meliputi penggunaan dereplikasi biologi, dereplikasi kimia, serta optimasi fermentasi untuk meniadakan re-isolasi senyawa metabolit sekunder. Penelitian juga dilanjutkan dengan melakukan proses *bioassay guided isolation* untuk mendapatkan senyawa yang mempunyai aktivitas antimikobakterium dari InaCC A758. **Hasil** Berdasarkan hasil skrining, didapatkan isolat *Actinomycetes* InaCC A758 yang mampu menghambat pertumbuhan *M. tuberculosis* strain H37Rv. Berdasarkan analisis taksonomi, InaCC A758 diketahui merupakan *Actinomycetes* yang pernah ditemukan dan dieksplorasi sebelumnya (mempunyai kedekatan homologi dengan *S. parvus* strain NBRC 14599 (99,64%)). Selain itu, gen NRPS dan PKS InaCC A758 diketahui mempunyai kedekatan dengan gen NRPS milik *S. parvulus* (93%), serta borrelidin type-PKS seperti halnya *S. rochei* dan *S. parvulus* (99%). Sementara itu, hasil dereplikasi kimia menggunakan HR-MS, diketahui bahwa InaCC A758 berpotensi untuk menghasilkan senyawa baru. Selanjutnya, berdasarkan hasil analisis multivariat, didapatkan hasil bahwa terdapat peningkatan potensi keragaman profil metabolit sekunder dari ekstrak InaCC A758 hasil optimasi fermentasi dan co-kultur. Namun, berdasarkan hasil *bioassay guided isolation*, didapatkan dua senyawa re-isolasi yang berpotensi sebagai antimikobakterium dari InaCC A758, yaitu 1) A758p1 dengan aktivitas hambatan pertumbuhan *M. tuberculosis* strain H37Rv sebesar 0,78 µg/mL. Senyawa tersebut berdasarkan hasil pemeriksaan LC-MS, HR-MS, UV-Vis, FTIR, dan NMR teridentifikasi sebagai actinomycin D ($C_{62}H_{86}N_{12}O_{16}$; m/z 1255,6426) dan Actinomycin V ($C_{62}H_{84}N_{12}O_{17}$; m/z 1269,6131), dan 2) A758p4 dengan aktivitas hambatan pertumbuhan *M. tuberculosis* strain H37Rv sebesar 100 µg/mL. Senyawa tersebut berdasarkan hasil pemeriksaan LC-MS dan HR-MS diduga adalah (a) (2-chlorophenyl){2-[(-methoxyphenyl) imino] -4-methylidene-3-thia-1-azaspiro[4,5]dec-1-1yl} methanone ($C_{12}H_9N_3OS$; m/z 243,04573 / $C_{23}H_{23}ClN_2O_2S$; m/z 426,959), (b) dimethenamid ($C_{12}H_{18}ClNO_2S$; m/z 276,07228), atau (c) timoprazole/orbencarb ($C_{13}H_{11}N_3OS$; m/z 257,06151). **Kesimpulan** Metode eksplorasi sistematik menggunakan dereplikasi biologi, dereplikasi kimia, dan optimasi fermentasi terhadap *Actinomycetes* Indonesia dapat berpotensi untuk meningkatkan keragaman metabolit sekunder dari InaCC A758. Namun metode sistematik tesebut masih belum dapat menghasilkan isolasi senyawa baru yang mempunyai potensi sebagai antimikobakterium.

Kata kunci : Antimikobakterium, *Actinomycetes* Indonesia, Actinomycin, Metabolit Sekunder *Streptomyces sp*.

ABSTRACT

Backgrounds The increase in Multidrug Resistant/Rifampicin Resistant-Tuberculosis (MDR/RR-TB) cases has led to the need for new antimycobacterial drugs. Indonesia is a country with a large biodiversity, but the exploration of Indonesian *Actinomycetes* as an antimycobacterial is still rarely done. **Methods** This study carried out a systematic exploration of Indonesian *Actinomycetes* to produce new antimycobacterial. Exploration carried out by the use of biological dereplication, chemical dereplication, and optimization of fermentation to eliminate re-isolation of secondary metabolites. The research also continued by conducting bioassay guided isolation to obtain compounds that have antimycobacterial activity from InaCC A758. **Results** Based on screening results, *Actinomycetes* InaCC A758 was able to inhibit the growth of *M. tuberculosis* strain H37Rv. InaCC A758 is known to be an Actinomycetes that has been found and explored before (homology closeness with *S. parvus* strain NBRC 14599 (99.64%)). In addition, InaCC A758 isolates are known to be close to the NRPS gene of *S. parvulus* (93%), as well as PKS-type borrelidin of *S. rochei* and *S. parvulus* (99%). The result of chemical dereplication using HR-MS states that InaCC A758 isolate has the potential to produce new compounds. Furthermore, based on the results of multivariate analysis, it was found that there was an increase in the chemical diversity of the secondary metabolite profile of the InaCC A758 extract resulting from optimization of fermentation and co-culture. However, based on the results of bioassay guided isolation, two re-isolation compounds that have potential as antimycobacterial obtained from InaCC A758, namely 1) A758p1 with activity to inhibit the growth of *M. tuberculosis* strain H37Rv, MIC value 0,78 µg/mL. The compound based on LC-MS, HR-MS, UV-Vis, FTIR, and NMR data is identified as actinomycin D ($C_{62}H_{86}N_{12}O_{16}$; m/z 1255,6426) and actinomycin V ($C_{62}H_{84}N_{12}O_{17}$; m/z 1269,6131), and 2) A758p4 with activity to inhibit the growth of *M. tuberculosis* strain H37Rv, MIC value 100 µg/mL. The compound based on LC-MS and HR-MS data is predicted as (a) (2-chlorophenyl){2-[(-methoxyphenyl) imino] -4-methylidene-3-thia-1-azaspiro[4,5]dec-1-1yl} methanone ($C_{12}H_9N_3OS$; m/z 243,04573 / $C_{23}H_{23}ClN_2O_2S$; m/z 426,959), (b) dimethenamid ($C_{12}H_{18}ClNO_2S$; m/z 276,07228), or (c) timoprazole/orbencarb ($C_{13}H_{11}N_3OS$; m/z 257,06151). **Conclusions** Systemic exploration methods using biological dereplication, chemical dereplication, and optimization of fermentation of Indonesian *Actinomycetes* can potentially increase the chemical diversity of secondary metabolites from InaCC A758. However, the systematic method still cannot produce isolation of new compounds that have potential as antimycobacterial.

Keywords : Antimycobacterial, Indonesian *Actinomycetes*, Actinomycin, Secondary Metabolite of *Streptomyces sp*.