

Aktivitas antibakteri dan mekanisme kerja ekstrak daun *Lawsonia inermis* Linn. terhadap *Streptococcus pneumoniae* = Antibacterial activity and mechanism of action of *Lawsonia inermis* Linn. Leaves extract against *Streptococcus pneumoniae*

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Abstrak

Streptococcus pneumoniae merupakan bakteri patogen yang menyebabkan penyakit pneumonia, meningitis, bakterimia dan otitis media. Beberapa galur resisten antibiotik dilaporkan menyebabkan penyakit infeksi pneumokokus. Tanaman obat *Lawsonia inermis* dilaporkan memiliki aktivitas antibakteri terhadap beberapa bakteri patogen. Penelitian ini bertujuan untuk menjelaskan aktivitas antibakteri *L. inermis* terhadap *S. pneumoniae*. Penentuan aktivitas antibakteri dilakukan dengan difusi cakram, dan mikrodilusi. Penentuan lisis dan kebocoran sel dilakukan dengan perbandingan nilai absorbansi pada panjang gelombang 595 nm, 280 nm dan 260 nm. Penentuan aktivitas bakterisidal dilakukan dengan uji waktu bunuh. Kemudian perubahan ultrastruktur *S. pneumoniae* yang diakibatkan oleh ekstrak *L. inermis* dilakukan dengan pengamatan TEM. Pengukuran laju ekspresi relatif dilakukan dengan qRT-PCR. Berdasarkan nilai MIC ekstrak etil asetat *L. inermis* memiliki aktivitas antibakteri yang kuat terhadap ATCC 49619 dan galur resisten antibiotik, sedangkan ekstrak etanol menunjukkan aktivitas antibakteri dengan rentang lemah hingga moderat. Kedua ekstrak menunjukkan aktivitas bakterisidal 100% pada 1 jam pertama inkubasi. Ekstrak *L. inermis* dapat menyebabkan pelisisan dan kebocoran sel bakteri sejak 1 jam pertama inkubasi. Selain itu, ekstrak *L. inermis* juga memiliki aktivitas antibiofilm dan menyebabkan penurunan ekspresi protein toksin-antitoksin epsilon-zeta serta kenaikan ekspresi protein lisin A pada *S. pneumoniae*. Ekstrak *L. inermis* juga memberikan perubahan ultrastruktur dari sel *S. pneumoniae* meliputi kerusakan dinding sel, kebocoran sel, dan penurunan integritas membran sel yang menyebabkan pelipatan membran sel yang diamati dengan TEM. Hasil LC-MS menunjukkan beberapa senyawa fitokimia turunan fenol yang berperan sebagai antimikroba terdapat dalam ekstrak *L. inermis*. Hasil penelitian ini menunjukkan bahwa ekstrak *L. inermis* memiliki aktivitas antibakteri terhadap *S. pneumoniae*.

.....*Streptococcus pneumoniae* is a human pathogen causing diseases. There are several studies reporting that drug resistant strain caused diseases in patients. Meanwhile, *Lawsonia inermis*, as medicinal plant, is reported to have antimicrobial activity against some human pathogenic bacteria. However, reports on antimicrobial activity of *L. inermis* against *S. pneumoniae* are limited. Therefore, this study is aimed to examine antibacterial activity of *L. inermis* against *S. pneumoniae*. Antibacterial activity of *L. inermis* extract is determined by disc diffusion and microdilution. Moreover, cell lysis and leakage were observed by comparing the absorbance at 595, 280 and 260 nm, while antibiofilm was measured based on crystal violet staining. Ultrastructure changes were observed under TEM. Meanwhile, expression rate of epsilon-zeta toxin-antitoxin system and lysin protein were measured by qRT-PCR. This study found that ethyl acetate extract of *L. inermis* showed strong antibacterial activity against ATCC 49619 and drug resistant isolates while ethanol extract of *L. inermis* showed weak to intermediate antimicrobial activity according to MIC value. Both extracts showed 100% bactericidal activity at first hour of incubation. Moreover, *L. inermis* extract also caused bacterial lysis, leakage and impact to epsilon-zeta and autolysin protein expression on *S.*

pneumoniae. In addition, *Lawsonia inermis* extract also showed antibiofilm activity and caused ultrastructure changes including cell wall disorder, bacterial leakage, and cell membrane folding. LC-MS spectra analysis showed that *L. inermis* extract was consisted of phenolic compounds reported to have antimicrobial activity. These findings showed that *L. inermis* extract had antibacterial activity against *S. pneumoniae*