The potential of several compounds in *Peperomia pellucida* as a solution to rifampicin resistance in tuberculosis disease

Iwang Davi Setiawan¹, Muhammad Reza Putra²-³, Aditya Wardhana⁴, and Sista Werdyani⁵,⁶*

¹Magister Programme of Pharmacy, Department of Pharmacy, Universitas Islam Indonesia
²The Institute of Pharmacology, College of Medicine, National Yang Ming Chiao Tung University, Taiwan
³Department of Medical Research, Taipei Veterans General Hospital, Taiwan
⁴Pharmacist Professional Study Program, Department of Pharmacy, Universitas Islam Indonesia
⁵Pharmaceutical Science Laboratory, Department of Pharmacy, Universitas Islam Indonesia
⁶Doctorate Programme of Biotechnology, Universitas Gadjah Mada

*Corresponding author: sista.werdyani@uii.ac.id*

**Abstract**

**Background:** Tuberculosis is a lung disease caused by *Mycobacterium tuberculosis*. In 2018, the incidence of tuberculosis in Indonesia was 1,017,290. The number of cases was accompanied by an increase in the number of resistances to first-line antibiotics (rifampicin) used due to patient non-compliance as well as the long duration of treatment. The increase in resistance around the world is always increasing every year by about 2%. Therefore, we need another alternative to existing plants in Indonesia that has the potential to be a solution to the problem and overcome resistance to these treatments.

**Objective:** This review was conducted to find out the potential of *Peperomia pellucida* as an antituberculosis drug that can be a solution to rifampicin resistance in tuberculosis disease.

**Results:** The results of the literature review showed that the *Peperomia pellucida* or suruhan plants have antibacterial potential against *Mycobacterium tuberculosis* because it contains a class of alkaloid compounds that function to inhibit deprenyl-phosphoryl-β-D-ribose-2 oxidase (an enzyme responsible for the biosynthesis of arabinogalactan in the cell walls of *Mycobacterium tuberculosis*), a flavonoid that has activity inhibition of fatty acid synthase II (FAS-II) enzymes, tannins that can precipitate proteins in *Mycobacterium tuberculosis*, and triterpenoids that have an activation mechanism of intracellular killing cascades in host cells during tuberculosis bacterial infection.

**Conclusion:** *Peperomia pellucida* or suruhan plants have the potential to be further investigated as a solution to overcome rifampicin resistance in tuberculosis disease.

**Keywords:** *Mycobacterium tuberculosis*, rifampicin resistance, *Peperomia pellucida*

1. Introduction

   Tuberculosis, which is a lung disease caused by *Mycobacterium tuberculosis*, was reported to have a high incidence rate by RISKESDAS in 2018 (Dheda *et al.*, 2016). The incidence rate reached 0.42%, or a total of 1,017,290 (Tim Riskesdas, 2019). The management of tuberculosis necessitates a relatively extended duration, which is typically accompanied by diverse treatment-related adverse effects, leading to frequent discontinuation of treatment by patients during the recovery phase. This fact is exacerbated by the high incidence of Multi Drug-Resistant Tuberculosis (MDR-TB). MDR-TB, or *Mycobacterium tuberculosis* bacteria has resistance to the first-line anti-tuberculosis drugs such as rifampicin (Falzon *et al.*, 2015). The occurrence of MDR-TB is a result of non-compliance with drug use in patients and causes
patients to have to take second-line drugs, which of course have a longer duration of treatment. Therefore, it is necessary to find alternative compounds or new drugs as a solution to the occurrence of resistance by utilizing the potential of plants in Indonesia.

*Peperomia pellucida* Kunth. is a shrub-type plant with succulent leaves that have a distinctive aroma and grow in moist soil and wet rock crevices (De Oliveira *et al.*, 2017). *Peperomia pellucida* Kunth., also called suruhan, is a member of the *Piperaceae* family. This plant is an herbaceous plant that is widely found in South America and Asia. This plant species has a history of ethnomedicinal use, which includes the treatment of abdominal pain, abscesses, acne, ulcers, colic, fatigue, gout, headaches, kidney dysfunction, rheumatic pain, and to treat breast cancer, impotence, measles, anxiety disorders, and smallpox (Oloyede & Onocha, 2011). In addition, *Peperomia pellucida* Kunth. also has potential as an antibacterial for several types of pathogenic bacteria, including *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumonia*, *Salmonella typhi*, and *Pseudomonas aeruginosa* (Idris *et al.*, 2016).

Previous research stated that the essential oil of *Peperomia pellucida* Kunth. has an antimicrobial effect. *In vitro* testing using the bacterium *Bacillus subtilis* showed inhibition zones of 11.6 mm and 17.3 mm. This bactericidal ability may be influenced by the presence of various secondary metabolites contained in the *Piperaceae* family, including flavonoids and phytosterols such as acacetin, apigenin, isovitexin, pellucidatin, campesterol, and stigmasterol (Oloyede & Onocha, 2011). Besides that, *Peperomia pellucida* Kunth. also contains alkaloids, flavonoids, saponins, tannins, and triterpenoids, which are reported to have anti-tuberculosis activity (Truong *et al.*, 2011; Radji *et al.*, 2015; Mishra *et al.*, 2017; Okoh *et al.*, 2017; Kaczmarek, 2020; Rabaan *et al.*, 2022). This indicates *Peperomia pellucida* Kunth. has the potential to be utilized in the treatment of MDR-TB.

Several types of secondary metabolites can be used in the treatment of MDR-TB. The first is the piperine alkaloids found in plant extracts, which are known to induce the proliferation of T and B cells, increase Th1 cytokines (IFN-γ and IL-2), and increase macrophage activation (Okoh *et al.*, 2017). Second, flavonoid compounds are known to help fight tuberculosis by removing hydroxyl radicals and superoxide anion radicals. This is because the increasing free radicals is related to the severity of the disease. Flavonoids also produce inhibitors of the fatty acid synthase II (FAS) enzyme (Idris *et al.*, 2016). Third, tannin compounds as anti-tuberculosis agent work by damaging bacterial cell membranes and can
induce the formation of tannin complexes with metal ions that can increase the toxicity of tannins. Moreover, tannins can create crenation of cell walls and membranes, which can interfere with cell permeability so that their growth can be inhibited and induce death (Chung et al., 1998). The fourth component is triterpenoid, which also has anti-tuberculosis activity by activating intracellular killing cascades in host cells during infection with tuberculosis bacteria or by increasing the main pre-inflammatory cytokines, namely INF-γ and TNF-α (Wei et al., 2011). The four secondary metabolites above indicated Peperomia pellucida Kunth. can be used as a potential anti-tuberculosis agent for the treatment of MDR-TB in the future.

2. TB drug resistance

*Mycobacterium tuberculosis* is a bacterium that causes the infectious disease tuberculosis. Most of these bacteria attack the lungs, but they can also attack other organs of the body. This disease can be transmitted through the air, but the bacteria that cause it can die in the presence of direct sunlight and can survive for several hours in a dark and humid place (Kemenkes RI, 2014). The government’s efforts to overcome this disease include providing free treatment. In order to help people affected by the disease, the drugs are rifampin, ethambutol, pyrazinamide, streptomycin injection, and INH (Kemenkes RI, 2014). The structure of the anti-tuberculosis drug can be seen in Figure 1.

WHO updated the therapy standard for adult patients and stated that those who have drug-susceptibility pulmonary TB may have a 4-month regimen of isoniazid, rifapentine, moxifloxacin, and pyrazinamide (2HPMZ/2HPM) (WHO, 2022). Resistance is a condition when bacteria change while they resist antibiotics. Resistance has been reported to antibiotics against tuberculosis. This is a global crisis that poses the greatest threat to the health of the population. This problem occurs due to several factors, namely the use of antibiotics that are not appropriate, patients are not compliant, drugs are not available at health care facilities, patients cannot tolerate side effects, the treatment period is so long, and the patient feels he is already healthy. This can lead to antibiotic resistance, leading to increased morbidity, mortality, and treatment costs (Harris et al., 2014; Mason et al., 2018).

Updated information from WHO (2022) stated that there were cases named XDR-TB, which means patients resistant to any fluoroquinolone and at least one additional group A drug (such as levofloxacin or moxifloxacin, bedaquiline, and linezolid). Meanwhile, antituberculosis
drug resistance is one of the antibiotic resistances that is always increasing, with an average increase of 2% per year worldwide. Indonesia is included in the 27 countries with the highest levels of antituberculosis drug resistance in the world (World Health Organization, 2013). There were 442,000 TB cases in 2017, and an estimated 8,600-15,000 of them suffered from antituberculosis drug resistance, but the coverage that was treated only reached 27.36% (Kemenkes RI, 2021). Anti-TB drugs with poor quality DOTS (Directly Observed Treatment Short-course), patient non-adherence to treatment, patient age 40-60 years, poverty, disease duration of more than a year, previous treatment, and smoking have a close relationship with the incidence of tuberculosis resistance (Zhao et al., 2012).

![Figure 1](image.png)

**Figure 1.** The structure of the antituberculosis drug
A. Isoniazid, B. Rifampicin, C. Pyrazinamide, D. Ethambutol, E. Streptomycin

According to the research to date, it shows resistance to antituberculosis drugs such as rifampicin (RIF), isoniazid (INH), ethambutol (ETB), pyrazinamide, and streptomycin (STR), as shown in Figure 2. These data show that rifampicin resistance is the highest compared to other anti-TB drugs, with a resistance percentage of 80%, while other antituberculosis drugs were 66.7% resistant to isoniazid, 78.7% resistant to pyrazinamide, 65.9% resistant to streptomycin, and 65.9% resistant to ethambutol (Asif et al., 2014; Hu et al., 2017; Tam et al., 2017; Shrestha et al., 2020). Rifampicin, the drug with the greatest resistance, is a serious problem at the global
level because there are about 558,000 new cases of rifampin-resistant TB worldwide (WHO, 2013).

Figure 2. Percentage of resistance to anti-tuberculosis drugs

Resistance is also reported when antituberculosis drugs are given in combination, also known as MDR. The combination of antituberculosis drugs that are most potentially resistant when taken together are rifampicin and isoniazid, but resistance has also been reported with other combinations of first-line antituberculosis drugs (Figure 3) (Kemenkes RI, 2021). Among cases of RR TB (rifampicin resistance) worldwide, as many as 82% are cases of MDR TB. WHO estimates that there are 23,000 MDR/RR cases in Indonesia (WHO, 2013; Kemenkes RI, 2021).

Figure 3. Percentage of MDR-TB against Mycobacterium tuberculosis

MDR-TB resistance data using 70 samples was reported by Nikmawati et al. (2018). The data shows that the percentage of INH and rifampicin is higher than that of other MDR-TB. This shows that treatment of MDR-TB cases needs to be considered, especially with rifampicin and isoniazid, so that there is no failure in therapy that can cause death (Nikmawati et al., 2018). So far, no effective procedure for dealing with this resistance has been found. Clinicians still have difficulty determining therapeutic regimens for patients in the absence of antibiotics that can
overcome the problem of resistance (Bhatia & Narain, 2010). To be able to overcome antibiotic resistance, other solutions are needed that can be effective as alternatives to using plants that have the potential to treat tuberculosis.

3. Compounds that have potential as anti-tuberculosis

There are some mechanisms that are exposed as newly developed anti-tuberculosis drugs, including targeting DNA replication and protein synthesis, cell wall biosynthesis, energy metabolism, and proteolysis (Mittal et al., 2023). Sutezolid, as one example of a drug that acts as a DNA replication and protein synthesis inhibitor, is now in phase II drug development (Tenero et al., 2019). Meanwhile, some drugs are still in preclinical development, such as Capuramycin (SQ641), which acts as a peptidoglycan biosynthesis inhibitor, and TBAJ-587 (2nd generation diaryquinoline), which can inhibit ATP synthase (Tran et al., 2017; TB Alliance, 2023). Some of the newly developed drugs are isolated from natural products; one example is cyclomarin A, which acts as a proteolysis inhibitor (Li et al., 2010). Besides that, several compounds have also been reported to have anti-tuberculosis activity and can be seen in Table 1.

Alkaloids are natural chemical compounds that contain nitrogen atoms. Alkaloids are produced by a wide variety of organisms, including fungi, bacteria, plants, and animals, and are part of a group of natural products also called secondary metabolites (Khakia, 2012). Alkaloids stop deprenyl phosphoryl-β-D-ribose-2 oxidase from doing its job. This enzyme makes the cell wall of arabinogalactan Mycobacterium tuberculosis (Tiwari et al., 2013). Alkaloids also have a role in inhibiting the mycolic acid cyclopropane synthase enzyme and the ketoacyl-ACP KasA synthase enzyme, which can cause death in Mycobacterium tuberculosis (Rollando, 2017). In addition, alkaloids can inhibit the cleavage of 32-single-stranded DNA in Mycobacterium tuberculosis (García et al., 2018). Plants that contain alkaloids include suruhan (Peperomia pellucida), Javanese chili (Piper chaba), and Ziziphus mauritiana (Panseeta et al., 2011; Mishra et al., 2017).

Flavonoids consist of a large group of polyphenolic compounds that have a benzo-γ-pyron structure and are present in plants (Kumar & Pandey, 2007). The enzyme fatty acid synthase II (FAS II) is released by flavonoids, which stops the growth of Mycobacterium tuberculosis (Idris et al., 2016). There are several plants and also microbiologists that contain
flavonoid compounds, including Suruhan plants (*Peperomia pellucida*), *Erythrina schliebenii*, and *Cnidoscolus chayamansa* (Angelina *et al*., 2015; Nyandoro *et al*., 2017; Pérez-González *et al*., 2017).

**Table 1.** The mechanism for several compounds

<table>
<thead>
<tr>
<th>No</th>
<th>Compound group</th>
<th>Mechanism</th>
<th>Basic structure</th>
<th>Reference</th>
</tr>
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<tbody>
<tr>
<td>1</td>
<td>Alkaloid</td>
<td>Inhibits decaprenylphosphoryl-β-D-ribose 2' oxidase</td>
<td></td>
<td>(Tiwari <em>et al</em>., 2013; Rollando, 2017; García <em>et al</em>., 2018)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Inhibits mycolic acid cyclopropane synthase and -ketoacyl-ACP KasA synthase enzymes</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Inhibits cleavage of 32-single-stranded DNA</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Flavonoid</td>
<td>Inhibits mycobacterial fatty acid synthase II (FAS-II)</td>
<td></td>
<td>(Idris <em>et al</em>., 2016)</td>
</tr>
<tr>
<td>3</td>
<td>Triterpenoid</td>
<td>Penetrates the cell membrane and changes its composition, thereby affecting the fluidity or plasticity of the membrane and affecting the signaling by many ligands. Activation of intracellular killing cascades in host cells during tuberculosis bacterial infection. In addition, the anti-tuberculosis effect also appears with an increase in the main pre-inflammatory cytokines, namely INF-γ and TNFα.</td>
<td></td>
<td>(López-García <em>et al</em>., 2015; Netala <em>et al</em>., 2015)</td>
</tr>
<tr>
<td>4</td>
<td>Tannin</td>
<td>Inhibits the DNA-topoisomerase enzyme, resulting in the inhibition of the replication process of <em>Mycobacterium tuberculosis</em> bacteria. Makes imperfect formation of cell walls by targeting cell wall polypeptides so that this causes bacterial cells to become lysed due to osmotic or physical pressure so that bacterial cells will die</td>
<td></td>
<td>(Chung <em>et al</em>., 1998)</td>
</tr>
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Tannins are phenolic compounds that can react, and agglomerate proteins or other compounds containing amino acids and alkaloids and give a bitter and astringent taste. Tannins, which are divided into two groups, namely hydrolyzed and condensed tannins, can act as an antibacterial by precipitating proteins in *Mycobacterium tuberculosis* bacteria. Tannins are found in many types of plants. This compound has the function of protecting plants from predation by pests and herbivores (Julianto, 2019). Plants that contain tannins include starfruit leaves (*Averrhoa bilimbi* L.), which are known to have a Flavan-3,6,7,4',5'-pentaol structure, and
suruhan plants (*Peperomia pellucida* L.) with the ellagitannin group of tannins, which are known to have potential as anti-tuberculosis (Fri & Rantelino, 2018; Hidjrawan, 2018).

4. Conclusion

According to the previous description, it is proven that the suruhan plants contain four compounds that play a role in the inhibition of *Mycobacterium tuberculosis*, which include alkaloids, flavonoids, tannins, and triterpenoids. Therefore, suruhan plants have enormous potential as an alternative to MDR TB, although not much research has been done in this field. It is very promising for further research as an alternative to MDR-TB.

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