Chapter 1

Introduction

1.1 Background

The field of medicine is currently facing a major challenge of antibacterial resistance. It is giving the impact such as reducing the effectiveness of antibacterial in eliminating infectious bacteria. As a result, infections are becoming increasingly difficult to manage. Beside the clinical impact, antibacterial resistance poses a serious ecological threat and has been recognized as a global public health crisis. It is estimated around 700,000 people die each year due to infections caused by resistant bacteria (Khan et al., 2021). However, more recent global analysis reported that antibacterial resistance caused approximately 1.14 million deaths in 2021 (Naghavi et al., 2024). Therefore, developing effective strategies are important to enhance antibacterial penetration to eliminate bacterial infections.

One of the potential strategies involves exploring the endophytic bacteria which is the microorganism inhabiting plant without causing harm. These bacteria form beneficial impacts within the hosts, enhancing plant health by suppressing the pathogen, and adapting to competitive microbial environments. The ability to survive suggests the presence of bioactive metabolites, the compounds help to interact and outcompete with the other microorganisms, supporting the host's resilience (Afzal et al., 2019). In this study, *Piper crocatum*, known as a red betel leaf, is selected due to its traditional use for treating various diseases, including infections. *Piper* species are grown widely in the tropical and sub-tropical region, making Indonesia with its warm climate and high humidity become an ideal habitat (Siswina et al., 2019). Additionally, according to Dong et al. (2018), different parts of a plant host distinct endophytic bacterial communities because each plant organ represents a unique ecological niche for endophytes. Although *Piper crocatum* itself has strong medicinal potential, this study focuses on its bacterial endophytes. Because in the future application, working

directly with the plant would require a long cultivation period, while the endophytic bacteria can be isolated and cultured much more efficiently in the laboratory. Since *Piper crocatum* plants are already known for antibacterial activity, exploring their associated bacteria provides a practical and promising approach to discover potential bioactive compounds (Astuti et al., 2014).

However, most of the previous study of *Piper crocatum* mainly focused on exploring the fungal endophytes, particularly for the antifungal (Siswina et al., 2023). In contrast, the study on bacterial endophytes within *Piper crocatum* remains scarce and unexplored. This lack of information creates a knowledge gap, as bacterial endophytes may play an equally important function in the plant defense system, producing a diversity of bioactive chemicals with potential antibacterial properties. From the rising concern, investigating these bacterial endophytes of *Piper crocatum* will provide new opportunities for discovering novel bioactive compounds effective to inhibit pathogens.

1.2 Objective

Main objective:

Explore and characterize the antibacterial properties of endophytic bacteria isolated from *Piper crocatum*.

Specific objectives:

- Isolate and characterize the endophytic bacteria from *Piper crocatum*
- Assess the antibacterial effects of endophytic bacteria through cross-streaking, secondary metabolite extraction, and secretome against selected Gram-positive and Gram-negative bacteria.

1.3 Hypothesis

The hypothesis of this study is endophytic bacteria isolated from *Piper crocatum* are able to produce secondary metabolites that exhibit wide-spectrum antibacterial activity against selected Gram-positive and Gram-negative bacteria, through cross-streaking, secondary metabolite extraction, and secretome.