Chapter 1

Introduction

1.1 Background

Abrus precatorius is an herbal species belonging to the Leguminosae family. Historically, *A. precatorius* has been extensively utilized for medicinal herbs in India, Africa, China, and several other tropical and subtropical countries (Das & Bhakta, 2020). Two unique triterpenoid saponins, glycyrrhizin and abrusosides, are present in *A. precatorius* (Garaniya & Bapodra, 2014). Glycyrrhizin and abrusosides were found to be extensively used in several dietary and pharmaceutical usage, such as sugar substitutes for diabetic individuals, herbal medicine, and cough syrup, and were recently reported as a potential COVID-19 immunotherapy (Bailly & Vergoten, 2020). The aforementioned benefits indicate that *A. precatorius*, together with the products derived from it, possess significant value for the pharmaceutical industry (Kaur et al., 2022; Das & Bhakta, 2020).

The inherent accumulation of triterpenoid saponins in *A. precatorius* is notably low. Choi et al. (1989) reported that extraction of triterpenoid saponins from *A. precatorius* yielded only about 0.39% (w/w). With this amount, to meet an industrial scale demand, relying on extraction from high biomass is almost guaranteed, resulting in an intensifying planting, which requires a large planting land.

Implementing a method to increase the production yield can effectively harness the restricted quantity of triterpenoid saponins from *A. precatorius*, therefore enabling their utilization in the fields of food and medicine. In correlation to that, treatments on plants can be done, one of them is using phytohormones. Phytohormone treatment is a method that has been employed to increase secondary metabolites in several plant species via induction of its defence mechanism. This is done by foliar treatment directly to the plants.

Plants face numerous challenges, including environmental stresses such as drought, flood, temperature fluctuations, and predation by insects and microorganisms. Empirical research has demonstrated that the oxidative condition of plants plays a crucial role in their capacity to endure different biotic and abiotic stress forces (Sachdev et al., 2021). Multiple signalling pathways enable plant defence against these stresses by activating the production of various non-protein compounds and defensive proteins (War et al., 2011). Plant phytohormones including salicylic acid (SA), ethylene, jasmonic acid, and abscisic acid are essential factors of several signalling pathways that is required in plant defence as well as immunity (War et al., 2011). Salicylic acid functions as an intermediary in the phenylpropanoid pathway and plays a crucial role in suppressing infections, some insect pests, and biological stresses. Furthermore, SA controls the elements of its own signalling pathway in addition to participating in bidirectional communication with other pathways that facilitate plant resistance.

One hypothesis is that SA influences plant growth during stressful conditions by influencing nutrient absorption, water interactions, stomatal control, photosynthesis, ion uptake, seed germination, growth rate, and leaf membrane response to electrolytes (Alam et al., 2022; Zahid et al., 2023). Application of salicylic acid treatments exogenously to plants usually comes to varying methods such as foliar sprays, soil drenches, or nutritional solutions. Foliar spray is one of the most effective methods as it avoids soil interactions which could reduce the efficacy. Instead, it allows direct absorption via the plant's epidermis and stomata. This method has been used widely to increase the production of the secondary metabolites, such as triterpenoid saponin, in varying plant species. It has been demonstrated in previous research by Lei et al. (2022), that salicylic acid may increase saponin accumulation, where foliar treatment of salicylic acid increased the biosynthesis of saponin by activating the defense related pathways. Although, on *Abrus precatorius*, the application of the treatment to the increase of saponin accumulation remains under-explored. Hence, why this study is crucial to expand the understanding of its mechanisms and efficacy.

Various analytical techniques can be employed to measure accumulation of triterpenoid saponin. Phytochemical techniques are often employed for qualitative examination. For instance, the Liebermann-Burchard test which involves the mixing of acetic anhydride with sulphuric acid can be employed to rapidly confirm the presence of saponins. Furthermore, semi-quantitative studies were employed to measure the approximate quantity of the terpenoid compound. The quantification of terpenes was performed by combining the Liebermann-Burchard test and UV-vis spectroscopy, with Quillaja serving as the standard (Sinurat et al., 2020). Assays using acetic anhydride with sulphuric acid and UV-vis spectroscopy with Quillaja saponin as the standard have been performed for semi-quantification of saponin (Wu et al., 2019; Le et al., 2018). Thus, this experiment will gather the measurement of the triterpenoid saponin accumulation in the leaves and roots of *A. precatorius* after treatment with different concentrations of salicylic acid using a semi-quantitative analysis.

1.2 Objective

To investigate the effects of foliar application of salicylic acid towards triterpenoid saponin content in saga leaves and root tissue of *Abrus precatorius*.

1.3 Hypothesis

Salicylic acid treatment affects the accumulation of saponin production in *Abrus precatorius* leaves and roots.