

The Effect of Vinca Alkaloids (Vincristine and Vinblastine) on the Morphological Characters of Jack Beans or Koro Pedang (*Canavalia ensiformis*)

Diah Asta Putri^{a,1,*}, Mita Rahyuni^{b,2}, Mega Sarwati^{b,3}

^aLaboratorium Botani, Program Studi Biologi, Fakultas Sains dan Teknologi Terapan, Universitas Ahmad Dahlan, Yogyakarta. Jl. Ringroad Selatan, Tamanan; Yogyakarta, 55191; Indonesia.

^bProgram Studi Biologi, Fakultas Sains dan Teknologi Terapan, Universitas Ahmad Dahlan, Yogyakarta. Jl. Ringroad Selatan, Tamanan; Yogyakarta, 55191; Indonesia.

^cProgram Studi Biologi, Fakultas Sains dan Teknologi Terapan, Universitas Ahmad Dahlan, Yogyakarta. Jl. Ringroad Selatan, Tamanan; Yogyakarta, 55191; Indonesia.

diah.putri@bio.uad.ac.id*

* corresponding author

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ABSTRACT (10PT)

Crop productivity and quality can be increased through polyploidy induced by antimitotic compounds such as vinca alkaloids. The use of vinca alkaloids has never been applied to jack beans (*Canavalia ensiformis*), a tropical legume belonging to the Fabaceae family. This study aims to find out the effects of vinca alkaloids treatment on the morphological characteristics of jack beans. The treatment consisted of four different vinca alkaloids concentrations (0.5%, 1.00%, 1.50%, 2.0%, and untreated (0.0%) as the control. The plants were arranged in a completely randomized design. Treated plants were characterized based on morphological traits such as plant height, leaf area and dry seed weight. The results obtained revealed significant differences ($p \leq 0.05$) in the morphological traits of the treated plants when compared to the control. Overall, a 2.0% vinca alkaloids concentration resulted in higher average plant height by 193.8 cm, leaf area increments by 115.1 cm² and the dry seed weight by 1.7 g. The improvement of the morphological traits depended on the concentration and increased along with increment in vinca alkaloids concentration. Thus, it is concluded that a 2.0% concentration should be employed in improving the growth and seed weight of jack beans.

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1. Introduction

Jack bean (*Canavalia ensiformis*) is a tropical legume with a protein content of more than 26% belonging to the Fabaceae family (Solomon *et al.*, 2017). Sword bean is a potential horticultural commodity to be developed in Indonesia and could serve as a potential substitute in providing sufficient protein. Increased protein consumption in Indonesia, with the relatively high cost of

protein imports especially from the United States (BPS, 2021), has led to the search for alternative sources of locally available and richer proteins. Advancement in biological and agricultural sciences has promoted crop productivity and quality through plant breeding. Plant breeding methods vary, but the essence is to select the best type among variants in terms of genotype and phenotype characters related to agronomic and economic value (Breseghello and Coelho, 2013).

Genetic improvement can be done by inducing polyploids. The morphological characteristic of polyploid plants is bigger plants including bigger plant parts such as roots, stems, leaves, flowers and fruit. Polyploid plants can be induced by various chemical compounds, one of which is vinca alkaloids. Vinca alkaloid is a toxic alkaloid extracted from the periwinkle plant (*Chataranthus roseus*) (Iskandar and Iriawati, 2015). Vinca alkaloids such as vincristine and vinblastine act as a mitotic inhibitor (Chung *et al.*, 2011). These alkaloids have a colchicine-like effect that can duplicate chromosomes. They act by preventing tubulin polymerization, so that the separation of chromosome sets is disrupted when mitosis and cells have multiplied the chromosomes (Purbosari and Etika, 2018). Currently, the antimitotic compounds vincristine and vinblastine are available in the form of pure compounds and are produced commercially (Daryono, 2015). Previous study of Faba bean, where the seed treatment was done by soaking, indicated that they were successful in giving a significant effect on some traits such as diameter of stem, number of pods/plant, plant length at first harvesting and in total seeds/plant (Esho, 2019).

There have been numerous studies conducted on vinca alkaloids-induced polyploidy in order to improve the characters and yields of plants. Soybean (Kusnuriyanti *et al.*, 2017), melon (Daryono *et al.*, 2018), cayenne pepper (Purbosari and Etika, 2018), lily (Wardana *et al.*, 2019), and spearmint (Viza, 2019) showed significant increase in several characteristics such as plant height, leaf area, fruit size, and seed weight. Therefore, the present study was carried out to investigate the mutagenic effect of vinca alkaloids on morphological traits in jack beans.

2. Findings and Discussion

In this newly created file, highlight all of the contents and import your prepared text file. You are now ready to style your paper; use the scroll down window on the left of the MS Word Formatting toolbar. The morphological characteristics of control and treated *C. ensiformis* plants were analyzed based on the plant habitus and seed. The effect of colchicine in the plant characters was significantly different for the plant height, leaf area and dry seed weight (Table 1).

Table 1. The effects of vinca alkaloids on plant height, leaf area, and seed dry weight of *C. ensiformis*

Table Head	Mean ± SD			Plant Height (cm)	Dry Seed Weight (g)
	Leaf Area (cm ²)				
	Left Leaf	Middle Leaf	Right Leaf		
0%	21.8 ± 3.7 ^a	40.5 ± 3.6 ^a	23.7 ± 3.5 ^a	96.5 ± 1.03 ^a	1.0 ± 3.0 ^a
0.5%	27.7 ± 6.1 ^a	42.5 ± 2.1 ^a	25.3 ± 2.2 ^a	142.3 ± 1.09 ^b	1.1 ± 3.3 ^a
1%	37.7 ± 4.3 ^a	47.3 ± 4.7 ^a	34.8 ± 4.2 ^a	156.5 ± 1.05 ^b	1.3 ± 2.1 ^a
1.5%	78.6 ± 6.8 ^b	106.2 ± 6.7 ^b	80.9 ± 6.4 ^b	164.1 ± 1.67 ^b	1.4 ± 4.5 ^a
2%	88.6 ± 5.1 ^b	115.1 ± 4.7 ^b	86.2 ± 3.8 ^b	193.8 ± 1.86 ^c	1.7 ± 3.6 ^b

Plants treated with 2% of vinca alkaloids showed the best height, with an average of 193.8 cm. According to Suciati (2012), the height of a *C. ensiformis* plant can reach 100 cm. Therefore, the plant height achieved in this study can be said to exceed the height of *C. ensiformis* plant in general. The antimitotic-compound treatment was also reported to show an increase in the plant height of *Pennisetum purpureum* (Kamwean *et al.*, 2017), *Vicia cracca* (Munzbergova, 2017) and *Capsicum annuum* (Tammu *et al.*, 2021).

Vinca alkaloids treated plants demonstrated a bigger size of leaves compared to the control plant (Tabel 1). In plants, vinca alkaloids act as an antimitotic that often induces the sizes of

various plant organs (Hadfield, 2014). Morphological changes are regularly reported in response to chromosome doubling. Traits such as leaf size, internode length and plant height have all been reported to be influenced by chromosome doubling (Touchell *et al.*, 2020). These confirmed the results shown in this study. The results of this study were in accordance with other research such as at *Polianthes tuberosa* (Rahayu *et al.*, 2014) and *Cucumis melo* (Daryono *et al.*, 2018). A study on spearmint (*Mentha spicata*) also found that treated plants had broader leaves compared to control plants (Viza, 2019).

Observation of the seeds showed that the dry weight of the vinca alkaloids treatments was higher than that of the control (Tabel 1). The treatment with a concentration of 2.0% resulted in the heaviest seeds. An increase in leaf size can indirectly lead to an increase in the rate of photosynthesis. The increased rate of photosynthesis can produce photosynthate in greater quantities, so that larger seeds can be produced as a storage place for photosynthate. Similar studies reported that antimitotic substances from periwinkle plants increase soybean (*Glycine max*) yield, the higher concentration increased the number of flowers, number of pods, and number of seeds by 16, 11 and 30 per plant, respectively (Kusnuriyanti *et al.*, 2017).

3. Method

2.1. Tools and Materials

The tools used in this study were a measuring flask, measuring glass, beakers, stirring rods, analytical balances, hygrometer, soil tester, shovel, sprayer, ruler, stationery, and camera. The materials used in this study were the seeds of *C. ensiformis*, Bio-Chatarantine powder containing the active substances of vincristine and vinblastine, goat manures, soil, 35 x 35 cm polybags, pot trays or seedling trays, label papers, 96% alcohol, and aquadest.

2.2. Procedures

- Seeds were harvested from a jack bean plantation in Sleman Regency, Yogyakarta, Indonesia. The viability of the seeds was tested by soaking seeds in water and floating seeds were discarded while sinking seeds were treated. Treatment solutions of vinca alkaloids were prepared by dissolving vinca alkaloids powder (Bio-Chatarantin, UGM, Indonesia) in distilled water to reach treatment concentration of 0.5%, 1%, 1.5% and 2% according to the research design (Daryono, 2015).
- Subsequently, a total of 15 *C. ensiformis* seeds were soaked in each of 10 ml vials filled with corresponding vinca alkaloid solutions for the treatments for 18 h. After the treatment, the seeds were washed with distilled water and planted in the seedling trays containing soil medium. After 10 days, the seedlings were transferred into 35 x 35 cm polybags containing a mixture of soil and manure with a 1:1 ratio. The seedlings were watered every day. Plant maintenance was done by adding fertilization. In this study, three replicates of plants from each treatment sample were used for further data analysis (Manzoor *et al.*, 2019)
- The plant morphological characteristic measurement was carried out after the first harvest of *C. ensiformis*, which was about 2-month-old. In the observation, the morphological characteristics were divided into two categories. The first was the plant habitus, including plant height and leaf area. The second was the fruit characteristic, namely the weight of dry seeds. All data obtained were analyzed using one-way ANOVA ($\alpha = 5\%$) and followed by the Duncan Multiple Range Test (DMRT).

4. Conclusion

The results of this study showed that 2.0% vinca alkaloids treatment had a significant effect on several morphological characteristics of *C. ensiformis*. The treatment increased plant height, leaf area and dry seed weight. Our findings provide essential information to obtain better performances of *C. ensiformis* plants through vinca alkaloids treatment. This study serves as a preliminary step in increasing the productivity and quality of the local *C. ensiformis* in Indonesia.

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