



Morphological Characteristics of *Phalaenopsis* spp. by Colchicine Application Based on Qualitative

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Abstract. Indonesia has very diverse biodiversity, such as *Phalaenopsis*. *Phalaenopsis* have the potential to be cultivated in plant breeding as parents for new hybrids. One of the attempts to improve the potential of *Phalaenopsis* spp. is polyploidization using colchicine. Polyploidization using colchicine can change morphological characteristics of *Phalaenopsis* spp. Into the desired character. This study aimed to examine the morphological characteristics of *Phalaenopsis* spp. Resulted from polyploidy induction by using 1000 ppm colchicine. The research was conducted at greenhouse of the Faculty of Agriculture, Sebelas Maret University, Surakarta in December 2021-March 2022. Materials used in this research were *Doritaenopsis* Shu Long Purple Queen x Ox King ‘3534’, *Phalaenopsis* Chain Xen Moment, and *Phalaenopsis* Shu Long ‘TS2904’ which had been treated with 1000 ppm colchicine on the flower buds. Morphological observations based on the Orchid Ornamental Plant Characterization Guidebook. Observed variables consisted of 15 characters, including the characterization of leaves and flowers. Morphological diversities were analyzed descriptively based on direct observation and documentation of orchid flower. Results showed that the treatment of 1000 ppm colchicine on orchid flower buds could change the shape of dorsal sepal tip and lateral sepal tip.

Keywords: Flower morphology · Leaf morphology · Flower bud

1 Introduction

Orchids are ornamental plants that are in great demand for religious ceremonies or home decoration. Orchids are attractive ornamental plants because they have a variety

of shape, color, and size of flowers. Beside their beauty and attractiveness, orchids also have high economic value. Orchids can be marketed in the form of compote, juvenile plants, adults, and cut flowers. Some cultivated orchids are *Phalaenopsis*, *Dendrobium*, *Cattleya*, *Vanda*, and *Orcidium*.

One of the most popular orchid species in Indonesia is *Phalaenopsis*. Besides being found in Indonesia, *Phalaenopsis* can also be found in Malaysia, Philippines, and Thailand. *Phalaenopsis* have a large flower shape, varied flower color and a long blooming time.

Phalaenopsis are one of the local orchid species that are widely used as parent plants for new hybrids [1]. Therefore, *Phalaenopsis* have the potential to be developed in plant breeding. One of the attempts to improve the potential of *Phalaenopsis* is polyploidization using colchicine.

Colchicine is one of mutagens. Colchicine can produce polyploidized individuals by increasing the chromosome number in plants [2]. Increased chromosome number due to the use of colchicine can change the nature of the plant to the desired trait [3]. The treatment of colchicine in orchid flowers can give morphological characteristics of larger flowers than without colchicine treatment. The morphological characteristics of the plant become larger as a result of the increase in chromosomes due to colchicine treatment. Chromosomes in polyploidy plants due to colchicine treatment are more numerous, so the cell size and cell nucleus increase in size. The larger cell size will produce larger plant parts as well. Colchicine can cause significant changes in flower characteristics [4]. Colchicine-treated plants have an appearance superior to control plants such as the largest diameter of florets, the longest and thicker flower stalks, and an increase in the number of florets. Besides affecting quantitative characteristics, colchicine also affects the qualitative characteristics of the plants.

Morphological characterization is some of the approaches used to generate information about desired plant trait for farmers and breeders [5]. Morphological characterization of orchids is needed for selecting orchid germplasm varieties that have superior characters to be used as parents in crosses. This study aimed to examine the morphological characteristics of *Phalaenopsis* spp. Resulted from polyploidy induction by using 1000 ppm colchicine.

2 Materials and Methods

The experiment was conducted in December 2021 - March 2022 at greenhouse of the Faculty of Agriculture, Sebelas Maret University, Surakarta. Materials used in this research were stationary, camera, and Orchid Ornamental Plant Characterization Guidebook [6]. Plant materials in this research are *Doritaenopsis* Shu Long Purple Queen x Ox King '3534', *Phalaenopsis* Chain Xen Moment, and *Phalaenopsis* Shu Long 'TS2904'. Orchid flower buds were wrapped with cotton and treated with 1000 ppm colchicine using a pipette. The flower buds were then covered with carbon paper for 2 days to prevent evaporation. Furthermore, the flower buds were labeled with paper according to the treatment. This research was conducted by direct observation through recording primary data and documentation of orchid parts, especially on flowers.

Morphological observations were made based on the Orchid Ornamental Plant Characterization Guidebook [6]. Each accession consists of 2 samples to be observed. Data

collection techniques were based on the Orchid Ornamental Plant Characterization Guidebook [6] by observation and documentation. Observation of morphological characters amounts to 15 qualitative characters. Qualitative characters include flower shape, flower color, lateral sepal color pattern, dorsal sepal color pattern, petal color pattern, lateral sepal tip shape, dorsal sepal tip shape, petal tip shape, lateral dorsal sepal shape, petal shape, leaf shape, leaf color, leaf edge shape, leaf tip shape, and leaf surface texture. Observation data were analysed descriptively.

3 Results and Discussion

Individual plant traits can be identified in qualitative characters. Qualitative traits that can differentiate between individuals with contrast and clear boundaries controlled by major genes [6]. Changes in the results of colchicine treatment based on qualitative data are considered more accurate than quantitative data. Plant qualitative characters are not sensitive to environmental changes, so although planted in a different environment, these characters will always exist [7]. Qualitative character data of *Phalaenopsis* spp. Are presented in Table 1.

The observation shows that colchicine treatment on each orchid can cause different responses of plant qualitative characters. The result is that colchicine plays a role in doubling the number of chromosomes from diploid to polyploid which has a different effect on each plant. In accordance with another research which states that ploidy in plants will have a different effect according to the gene reaction and physiological reaction of the plant [8].

Polyploidy is a condition in an organism that has more than one set of chromosomes. Polyploidy induction can be obtained using chemicals such as colchicine. Colchicine is a mitotic inhibitor that can interrupt the work of microtubules thus preventing the formation of spindle thread. The failure to form spindle thread in the process of cell division leads to the formation of polyploidy individuals. One of the efforts in the development of *Phalaenopsis* orchid hybrids is polyploidy [9].

Colchicine ($C_{22}H_{25}O_6N$) is an alkaloid produced by the bulbs and seeds of the Autumn crocus plant (*Cholchicum autumnale* Linn.) of the Liliaceae family. Agriculture in the field of biotechnology has widely used colchicine to double the number of chromosomes. Diversity in plants can be created by doubling the chromosome number using colchicine.

Polyploidy of *Phalaenopsis* orchid varieties plays an important role in increasing large flower size and other characteristics [10]. However, colchicine applied to individual plants does not affect the mutation of all cells, but only the mutation of some cells. This is because colchicine can cause polyploidy in plants that have meristematic cells [11].

Based on the observations in Table 1, there are differences between the shape of the dorsal sepal tip and the shape of the lateral sepal tip between the results of colchicine treatment and without colchicine treatment on *Dtps.* Shu Long Purple Queen x OX King '3534' and *Phal.* Chain Xen Moment (Fig. 1 and Fig. 2). *Dtps.* Shu Long Purple Queen x OX King '3534' without colchicine treatment has a dorsal sepal tip shape is obtuse, while in colchicine treatment is retuse. The shape of the lateral sepal tip of *Dtps.* Shu

Long Purple Queen x OX King '3534' without colchicine treatment is acute, while in colchicine treatment is retuse. *D. nobile* tetraploid plants at first flowering showed

Table 1. Qualitative data of *Phalaenopsis* spp. Based on Orchid Ornamental Plant Characterization Guidebook

Qualitative Data	Varieties					
	<i>Doritaenopsis</i> Shu Long Purple Queen x Ox King '3534'		<i>Phalaenopsis</i> Chain Xen Moment		<i>Phalaenopsis</i> Shu Long 'TS2904'	
	C	WC	C	WC	C	WC
Leaf shape	Obovate	Obovate	Obovate	Obovate	Obovate	Obovate
Leaf Tip Shape	Emarginated	Emarginated	Emarginated	Emarginated	Emarginated	Emarginated
Leaf Edge Shape	Entire	Entire	Entire	Entire	Entire	Entire
Leaf Surface Texture	Glabrous	Glabrous	Glabrous	Glabrous	Glabrous	Glabrous
Leaf Color	Green	Green	Green	Green	Green	Green
Flower Shape	Round	Round	Round	Round	Round	Round
Lateral Dorsal Sepal Shape	Elliptic	Elliptic	Elliptic	Elliptic	Elliptic	Elliptic
Petal Shape	Semi-circular	Semi-circular	Semi-circular	Semi-circular	Semi-circular	Semi-circular
Dorsal Sepal Tip Shape	Retuse	Obtuse	Acute	Obtuse	Obtuse	Obtuse
Lateral Sepal Tip Shape	Retuse	Acute	Acute	Obtuse	Acute	Acute
Petal Tip Shape	Obtuse	Obtuse	Obtuse	Obtuse	Obtuse	Obtuse
Dorsal Sepal Color Pattern	Shaded	Shaded	Spotted	Spotted	Spotted	Spotted
Lateral Sepal Color Pattern	Shaded	Shaded	Spotted	Spotted	Shaded	Shaded
Petals Color Pattern	Shaded	Shaded	Spotted	Spotted	Spotted	Spotted
Flower Color	Purple	Purple	White	White	Pink	Pink

Note: C: Colchicine treatment. WC: Without colchicine treatment

imperfect petal, sepal, and lip shape in some of the flowers. However, tetraploid *D. nobile* growth visually flowered normally at seven years after the polyploidy induction experiment [12]. The observation results showed that *Dtps.* Shu Long Purple Queen x OX King '3534' treated with colchicine and without colchicine treatment had similar qualitative data parameters, such as leaf shape, leaf color, leaf tip shape, leaf surface texture, leaf edge shape, flower shape, flower color, petal shape, petal tip shape, petals color pattern, lateral dorsal sepal shape, lateral sepal color pattern, and dorsal sepal color pattern.

Phal. Chain Xen Moment without colchicine treatment has a dorsal sepal tip shape is obtuse, while in colchicine treatment is acute. The shape of the lateral sepal tip on *Phal.* Chain Xen Moment without colchicine treatment is obtuse, while the colchicine treatment is acute. *Phal.* Chain Xen Moment treated with colchicine and without colchicine treatment also have similar qualitative parameters on leaf shape, leaf tip shape, leaf edge shape, leaf surface texture, leaf color, flower shape, lateral dorsal sepal shape, petal shape, petal tip shape, dorsal sepal color pattern, lateral sepal color pattern, petals color pattern, and flower color. The difference in shape in orchid flowers is thought to be



Fig. 1. *Dtps.* Shu long purple queen x ox king '3534' with colchicine treatment (A) and without colchicine treatment (B).

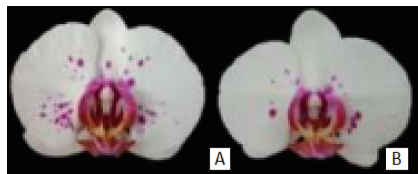


Fig. 2. *Phal.* Chain Xen Moment with colchicine treatment (A) and without colchicine treatment (B).



Fig. 3. *Phal.* Shu Long 'TS2904' with colchicine treatment (A) and without colchicine treatment (B).

caused by the effect of colchicine. According to another research that has been done, the effect of colchicine can show abnormalities in *Crocus sativus* L. plants, such as less asymmetrical tepals, irregular flower shapes, incomplete stigma, and angular tepals [10].

Table 1 showed that there was no difference between colchicine treatment and without colchicine treatment on *Phal.* Shu Long 'TS2904' based on qualitative data parameters. Polyploidy induction does not always reach the expected goal or does not always overcome the deficiencies of diploid plants. Tetraploids can also exhibit plants that are not twice as vigorous, resistant, and productive as diploid plants. This can be due to the occurrence of unfavorable chimera. Genetic instability and changes can be challenging for polyploidized plants. The disturbances produced by polyploidy induction still have the potential to produce new phenotypic and genotypic variations. The variation is useful for artificial selection in plant breeding programs [12]. However, although the basic color of the flowers that dominate in *Phal.* Shu Long 'TS2904' seems the same, the color of the flowers with colchicine treatment seems fainter than without colchicine treatment (Fig. 3). A color change is a response to colchicine treatment. Diploid plants tend to maintain the intensity of the original color, while polyploid plants can show the disappearance of the original color of the plant [11].

Table 1 showed the three accessions of orchids have similarities in flower shape, leaf shape, petal shape, lateral dorsal sepal shape, petal tip shape, leaf color, leaf edge shape, leaf tip shape, leaf surface texture. The similarity between orchid accessions is due to the same genus, *Phalaenopsis*, which has similar genetic traits. Orchid flower buds treated with colchicine are thought to produce tetraploid flowers. In general, the most common polyploidy is the doubling of the chromosome number from diploid (2x) to tetraploid (4x) [9]. Morphologically, diploid and tetraploid plants have different appearances. Tetraploid plants have taller and wider flowers that are significantly different from diploid plants [15]. Colchicine treatment on flower buds is thought to produce polyploid gametes. If pollination is performed, it will produce polyploidy orchids. Self-pollination of colchicine-treated orchids will produce tetraploid seeds, while colchicine-treated flowers crossed with flowers without colchicine treatment will produce triploid seeds [16]. Genetic diversity resulted from colchicine treatment can be used for plant breeding programs as a source of germplasm [17].

4 Conclusions

Based on the observation and analysis, it can be concluded that the application of 1000 ppm colchicine on flower buds can change the qualitative characteristics of *Phalaenopsis* spp. Based on qualitative characters, only *Phal.* Shu Long ‘TS2904’ has no difference between colchicine treatment and without colchicine treatment, while *Dtps.* Shu Long Purple Queen x Ox King ‘3534’ and *Phal.* Chain Xen Moment with colchicine treatment and without colchicine treatment have differences in the shape of the dorsal sepal tip and the shape of the lateral sepal tip.

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