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## PROMOTING EFFECT OF ETHANOLIC EXTRACT OF ROOT TUBER OF *Gloriosa superba* L. ON MELON GROWTH AND FRUIT QUALITY\*\*

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### ABSTRACT

An increase in the demand of colchicine extracted from *Colchicum autumnale* for plant mutagenic and medicinal activities makes the search of locally plant based product for its substitution critically important and *Gloriosa superba* is considered one of them since this plant contains colchicine. For this purpose, a field experiment was done to determine the effective concentration of root tuber extract of *G. superba* L. on promoting melon growth and fruit quality. Hybrid melon seeds of Action 434 were soaked in water as a control, in 0.04 gL<sup>-1</sup> colchicine, and in 10%, 20%, and 30% (wv<sup>-1</sup>) root tuber extract of *G. superba*. The soaked seeds were then planted in October 2012 at Agriculture Faculty Bengkulu University research plot in a CRD arrangement. Results showed that the 20% and 30% root tuber extract of *G. superba* were highly effective in promoting melon growth as indicated by larger leaf size, taller plant, and larger flower size and fruit quality as indicated by heigher fruit weight, larger fruit diameter, thicker edible fruit with and without rind, and higher dissolved sugar content. The promoting effect of these two concentrations on growth and fruit quality was as effective as 0.04 gL<sup>-1</sup> pure colchicine. Overall, from these three treatments 20% root tuber extract of *G. superba* was considered the most promising treatment because at this concentration fruit weight and fruit diameter was almost 1.5 times higher and fruit was approximately 2 Brix sweeter.

**Keywords:** *alkaloid, growth analysis, fruit quality, mutagenic substance, biological active substance*

### Introduction

In recent years there has been increased interest in screening higher plants for biologically active products [16] and *Gloriosa superba* L. (Family Liliaceae) is considered one of them. *G. superba* L. also called "kembang sungsang" (Java, Indonesia) or Glory Lily (English) is a native of tropical Africa and is found growing naturally in many countries of tropical Asia including Indonesia. The plant is found in the wild on natural fences a decade back but now it has been domesticated for an economic gain. All its part contains valuable alkaloids viz., colchicine and colchicoside as the major constituents [12], gloriosin and colchicocides, which are very costly, being highly demanded by pharma industries [10]. Due to excessive use of this plant for diverse medicinal purposes and poor seed germination the species has become endangered [3].

Medicinal importance of *G. superba* is due to the presence of colchicine. Colchicine is an alkaloid drug, chemically known as N-[(7S)-1, 2, 3,10-tetramethoxy-9-oxo-5,6,7,9 - tetrahydrobenzo[a]heptalen-7-yl] acetamide, originally extracted from *Colchicum autumnale* (autumn crocus, meadow saffron) medicinal plants and widely used for the treatment of gout disease [6]. Colchicine has the high market value and consistent demand in the field of medicine [5]. The alkaloid is the drug of choice to relieve acute attack of gout and familial Mediterranean fever [1]. At present there is renewed interest in the use of colchicine as a possible cure for cancer related diseases due to the action of colchicoside on spindle fibre

formation during cell division [9]. Since the detection of colchicine in *G.superba* [7], a number of researchers have suggested that this species could serve as a commercial source of colchicine and be recommended as substitute plants for *C. autumnale* for the alkaloid colchicine because the colchicine content in the genera *Colchicum* has been reported to be lower than in *Gloriosa* [4].

In addition for the treatment of various diseases, colchicine is also used for induction of plant polyploidy, multiplication of the chromosome in cell nucleus, due to its potent affinity for tubulin. The main action of the colchicine for the induction is to prevent the formation of a spindle so the anaphase movement of the chromosomes does not take place and the cell fails to divide. When the daughter chromosomes finally divide, they are all included in one cell and the chromosome number is doubled [9].

Polyploid plants can be of much economic importance by increasing plant vegetative growth and fruit yield. Even some flower qualities are improved by polyploidization. For example, polyploids induced by mitotic chromosome doubling showed larger and deeper-colored flowers than diploids in carnation and cyclamen [22, 20]. In lily, the large flower and sturdy stem in the tetraploids were useful as compared with those in the diploids [21]. In potato, colchicine treatment enhanced plant height, fresh weight, and number of leaves, which consequently increase the number of tuber and finally increase total fresh weight of tubers [11]. In melon, from the literature cited very few works on the effect of colchicine extracted from *G. superba* tuber on its growth and fruit quality has been reported. Thus, the present study was undertaken to determine the effective concentration of ethanolic extract of root tuber of *G.superba* L. on promoting melon growth and fruit quality.

## Materials and Methods

The research was conducted in October 2012 - December 2012 at Agriculture Faculty Research Plot Bengkulu University. The experimental design used was a completely randomized design (CRD) with the concentration of ethanolic extract of *G. superba* tuber as a treatment. The concentration of the treatments were control, 0.04% colchicine (0.04 g 100 ml of water<sup>-1</sup>), 10% root tuber extract of *G. superba* (10 g 100 ml of water<sup>-1</sup>), 20% root tuber extract of *G. superba* (20 g 100 ml of water<sup>-1</sup>), and 30% root tuber extract of *G. superba* (30 g 100 ml of water<sup>-1</sup>).

*G.superba* tubers collected from the Tanjung Jaya villagers Bengkulu were used as a plant material for colchicine extraction. The powdered tuber was subjected to extraction by maceration method [13]. The dry extract was then dissolved in distilled water to prepare 10%, 20%, and 30% (w v<sup>-1</sup>) concentrations. For 0.04% colchicine solution, 0.04 g colchicine (Sigma) was dissolved in 100 ml distilled water. These solutions were then tested for their agronomic effects by soaking F1 hybrid (Action 434) melon seed in each solution in a petridish for 24 hours. As a control, the seed was only soaked in distilled water. The soaked seeds were then planted in a polybag containing 10 kg media, one seed per polybag. The growing media was a mixture of soil, manure, and paddy bract. Plants were fertilized with 8.3 g urea, 7.1 g SP-36, 7.1 g KCl, and 2.4 g NPK (15-15-15) each polybag. The growing plants were kept upward by lining stem on wool string and tie it in upright position. Cutting apical meristems was undertaken on the 22<sup>nd</sup> node of stem, fruits were thinned and one selected fruit was left until harvest. Fruits were harvested at 65 – 70 days after planting when fruit maturity was attained.

Data from the observations were statistically analyzed by analysis of variance (F test level of 5%) [17] and significant differences among the treatments were then analyzed with Duncan's multiple range test (DMRT).

## Results and Discussion

Results of analysis of variance showed that a significant increase in leaf area, stem length, and flower diameter due to root tuber extract of *G. superba* was observed, but not in root fresh weight, stem fresh weight, leaf fresh weight, stem diameter, number of nodes, root length, and leaf greenness level (data not shown). Further statistical analysis with DMRT showed that 20% and 30% root tuber extract of *G. superba* caused leaf area almost three times larger than control (Table 1) and considered the most effective concentration for promoting leaf area. A similar result was also reported by Ernawati *et al.* [8] on chilli.

According to Suryo [19], an increase in leaf area occurred because colchicine transformed a diploid plant with a characteristic of larger size of polyploid cells. For plant stem length 30% root tuber extract of *G. superba* was considered the most effective treatment for promoting plant stem length, while 10% root tuber extract of *G. superba* was the worst. Plant stem at 30% root tuber extract of *G. superba* was about one third longer than that at 10% root tuber extract of *G. superba*. For flower diameter 0.04% pure colchicine was considered the most effective treatment but as effective as 20% root tuber extract of *G. superba*. At 0.04% colchicine flower diameter was approximately 1.50 times the width of flower diameter at control. An increase in flower diameter due to root tuber extract of *G. superba* was also reported by Sulistianingsih [18] on orchid.

Table 1. Mean of leaf area, stem length, and diameter of flower of melon treated with pure colchicine and root tuber extract of *G. superba*

Treatment	Leaf Area [cm <sup>2</sup> ]	Stem Length [cm]	Diameter of Flower [cm]
Control	1481.00 c	131.25 ab	2.73 c
0.04% pure colchicine	2650.75 b	146.75 a	4.25 a
10% root tuber extract	1882.00 bc	114.25 b	2.95 bc
20% root tuber extract	4205.75 a	139.00 a	3.90 ab
30% root tuber extract	5064.50 a	153.50 a	3.63 abc

Values within the same column that are not sharing the same letter differ significantly at  $P \leq 0.05$  (DMRT)

Table 2. Mean of fruit characteristics as affected by pure colchicine and root tuber extract of *G. superba*

Treatment	Fruit weight [g]	Fruit diameter [cm]	Fruit thickness [cm]	Thickness of edible part [cm]	Fruit sweetness [Brix]
Control	1187.50 bc	11.30 c	3.30 b	3.20 b	7.63 c
0.04% pure colchicine	1552.00 ab	14.50 b	4.65 a	4.55 a	9.13 ab
10% root tuber extract	1050.00 c	11.53 c	3.28 b	3.18 b	8.00 bc
20% root tuber extract	1925.00 a	16.20 a	4.50 a	4.40 a	9.38 a
30% root tuber extract	1587.50 a	13.78 b	4.50 a	4.40 a	9.65 a

Values within the same column that are not sharing the same letter differ significantly at  $P \leq 0.05$  (DMRT).

Analysis of variance showed that a significant increase in fruit weight, fruit diameter, fruit flesh thickness, and the sweetness of the fruit due to root tuber extract of *G. superba* was observed, but not in the number of seeds (data not shown). Further statistical analysis with DMRT showed that 20% root tuber extract of *G. superba* was the most effective treatment in promoting fruit weight followed by 30% root tuber extract of *G. superba* (Table 2). The fruit weight at these two concentrations was approximately 1.5 to 2 times the fruit weight at 10% concentration and control and was as heavy as the fruit weight at 0.04% pure colchicine. Therefore, colchicinoid substances from tuber extract of *G. superba* was as effective as pure colchicine in promoting fruit weight. This result was in agreement with the one reported by Anggraito [2]. He stated that colchicine increased the size of the fruit. For fruit diameter 20% root tuber extract of *G. superba* was considered the most effective treatment (Table 2). At this concentration its fruit diameter was about 1.5 times the fruit diameter at control, about 1.2 times the fruit diameter at 20% root tuber extract of *G. superba* and 0.04% pure colchicine. This result was in accordance with the one reported by Anggraito [2]. He stated that colchicine increased the thickness of fruit mesocarp. For thickness of edible part 20% and 30% root tuber extract of *G. superba* and 0.04% pure colchicine were considered the most effective treatment and 20% and 30% root tuber extract of *G. superba* were as effective as 0.04% pure colchicine (Table 2). At these three concentrations the thickness of fruit edible part was about 1.25 times the thickness of fruit edible part at control. According to Ramachandran [14], an increase in these fruit quality indicators were probably due to the fact that colchicine made the treated plant experienced an increasing cell size and the metabolic activity of this cell also increased. This was ultimately able to improve fruit quality. However, the same treatment was not able to increase the number of seeds. This occurred because colchicine only affected the size of the cell, but not to the acceleration of

cell division, including seed cells. Therefore, for the next study it was suggested that measuring the weight of 1000 seeds is more emphasized than measurement of the number of seeds.

In addition to fruit weight, fruit diameter, and thickness of fruit, an increase in fruit sweetness due to root tuber extract of *G. superba* was observed. Further statistical analysis with DMRT showed that 30% root tuber extract of *G. superba* was the most effective treatment for promoting fruit sweetness (Table 2). In addition, this treatment was as effective as 20 % root tuber extract of *G. superba* and 0.04% pure colchicine in promoting fruit sweetness. With these three treatment fruit with 9.65 Brix of dissolved sugar content was produced. Based on melon fruit quality developed by Rubatzky and Yamaguchi [15], melon fruit with 9.65 Brix of dissolved sugar content was categorized as high quality melon because sugar content was in the range of 9 – 11 Brix. According to Rubatzky and Yamaguchi [15], an increase in fruit sweetness occurred as the impact of enlargement of stomata due to colchicine application. The increase in the stomata size subsequently improved the process of photosynthesis due to increased CO<sub>2</sub> that entered through it.

### Conclusion

Application of 20% root tuber extract of *G. superba* effectively promoted melon growth as indicated by larger leaf size, taller plant, and larger flower size and melon fruit quality as indicated by higher fruit weight, larger fruit diameter, thicker edible fruit part with and without rind, and higher dissolved sugar content.

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