The effect of light and gibberellic acid concentrations on breaking dormancy of potato micro tuber

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Abstract. Potato (Solanum tuberosum L) is one of Indonesia’s priority crops considering its nutritive benefit as a carbohydrates source in food diversification. Potato tubers are classified as stem tubers, so the tubers such as micro tuber can appear in the stem of in vitro cuttings. The research aimed to determine the effect of GA3 concentrations (0, 5, 10, 15, 20 ppm) and storage conditions, e.g., Dark (T1) and light (T2), on breaking dormancy of potato micro tubers. This experiment was conducted in the Tissue culture Laboratory of IVEGRI from April until September 2018. The experiment used a randomized block design with 3 replications; each treatment consisted of 40 micro tubers of Granola (var.) with size ranged >3 - <4 g per each. The result showed that storage conditions gave significant differences to the percentage of breaking dormancy for 4-6 weeks, and GA3 concentration did not affect the average number of shoot, average shoot length in concentration 5 until 20 ppm. The average shoot number, shoot lengths were 0.63 – 2.20 per tubers, 7.50 – 20.13 mm, respectively. There was no interaction between treatment. Visual observation concluded that storage in light conditions produced shoots with better quality than ones in dark conditions.

1. Introduction

Potato (Solanum tuberosum L) is an important crop globally, and it is the fourth largest source after rice, wheat, and maize. In Indonesia, potatoes are vegetables in tubers rich in vitamin C and potassium [1].

Tubers of potato are the stem tuber so that it can be produced in the stem of in vitro cutting, i.e., micro tubers [2]. Since the micro tuber is pathogen-free, it can minimize the high failure rate in plantlet acclimatization. Moreover, it has several advantages, such as easier handling of storage materials and planting [3] [4].

According to [5], [6], [7], a successful in vitro micro tuber production depends on the ability of a variety to produce tubers conventionally. Potato micro tubers develop from the swelling part at the end of the stolons to the node or axillary leaves.

When the potato tubers are harvested in a dormant state, the length of dormancy depends on the variety, the plants' age, and the tubers' quality when harvested. The dormancy period of potato tubers is the beginning of tuber initiation, e.g., slower growth. This kind of ‘inactive’ period is rather difficult to determine because it is very dependent on the variety, plant growth, and tuber maintenance after harvest [8]. Potato tubers will not sprout even when placed in a supportive environment. The potato dormancy period occurs for 4 - 15 weeks after harvest. The signs of dormancy breaking in potato is when 80% of the tubers have sprouted and produced ≥ 2 mm shoots. The length of the shoots is influenced by the variety, tuber size, and the tubers' growing environment [9]. Meanwhile, according to [10], [11] the
dormancy period depends on the plant growth's environmental conditions, the age of the plant, and the quality of the tubers at harvest.

Gibberellic acid (GA3) is a growth hormone that can accelerate the dormancy breaking of potato tubers [12]. It is a growth regulator that can affect plant growth and morphogenesis [13]. To break the dormancy of the micro-tubers, a precise concentration of GA3 is required. GA3 treatment can stimulate shoot growth of potato micro tubers [14]. It is also able to stimulate dormant shoots to grow [15].

This research aimed to determine the effect of GA3 concentration and storage for breaking dormancy of micro tuber cv. Granola. The hypothesis was proposed that the concentration of GA3 and storage would help to break micro-tuber dormancy.

2. Methods
The research was carried out at the Tissue Culture Laboratory of the Indonesia Vegetable Research Institute / IVEGRI from April to September 2018. Micro tubers cv. Granola was initiated by inducing plantlets in MS Media (1962) plus 80 g / l of sucrose. Plant cultures were maintained in an incubation room with a temperature of 21 - 24 °C in a photoperiod of 16 hours light 8 hours dark. The micro tubers were harvested three months after tuber induction.

The treatments included concentrations of GA3 (0, 5 ppm, 10 ppm, 15 ppm, 20 ppm), and storage treatments, e.g., dark (T1) condition and light (T2) condition, with three replications in a Randomized block design. Each treatment consisted of 40 micro tubers with size > 3 - <4 g tuber. The micro tubers were soaked in GA3 solution for 15 minutes and dried at room temperature, then placed in storage treatment. The results were recorded on 4,6,8,10 WAT (Weeks After Treatment) e.g., percentage (%) of shoot growth, an average of shoot numbers, and an average of shoot length.

3. Results and discussion
The statistical analysis on the observation of breaking dormancy micro tuber dormancy of cv. Granola, there was no interaction between treatments.

| Treatment | Percentage of growing shoots (%) |
|-----------|----------------------------------|
|           | 4 WAT   | 6 WAT   | 8 WAT   | 10 WAT  |
| T1(dark)  | 4.75 A  | 26.50 A | 40.74 A | 47.00 A |
| T2 (light)| 2.00 B  | 16.00 B | 40.00 A | 48.00 A |
| K1        | 0.63 c  | 10.63 b | 23.13 b | 31.25 b |
| K2        | 5.63 a  | 25.63 a | 48.88 a | 53.13 a |
| K3        | 5.00 a  | 23.13 a | 41.25 a | 48.13 a |
| K4        | 3.13 b  | 23.13 a | 40.63 a | 46.88 a |
| K5        | 2.20 b  | 23.75 a | 50.00 a | 58.13 a |

Note: 1) WAT: Weeks After Treatment. 2) The average value followed by the same letter are no significantly different according to Duncan's multiple test of 5%. K1-K5: GA3 (0,5,10,15,20 ppm)

In Table 1, the GA3 treatment was only significantly different at the observation on 4.6 WAT, dark treatment (T1) the growth percentage was higher than the light treatment (T2). For treatment, GA3 concentration was statistically different at the age of 4.6 WAT, then not really different. However, visually, it can be seen that the highest GA3 concentration (20 ppm) resulted in the highest growth percentage at 6.8 10 WAT.

The percentage of sprouting tubers with a length of ≥2mm, starting at 4 WAT and 10 WAT, reached 31.25 - 58.13%. The variety and size of tubers strongly influenced the growth percentage of micro tubers. The low percentage of sprouting was probably due to the sub-optimal concentrations of GA3 or the quality of the cv. Granola micro-tubers. Where the small size of the micro tubers, will lead to loss
of weight. This situation is possible because the tubers were harvested late or earlier so it affected dry matter content, especially low content of carbohydrates [16], [14].

The micro tuber can germinate due to a chemical compound in the tuber, especially starch, protein, and shrinkage of micro tuber due to water losses. [17], [18], [19], [20].

The percentage of sprouts from micro tubers is influenced by genotype, and several factors include growing conditions and tuber storage after harvest [21], [22]. At 4-12th weeks after harvest, tubers continue another physiological process called external dormancy. Potato tubers in this condition will sprout if they are placed in a favorable environment for sprouting.

### Table 2. Average number of shoots at 4,6,8,10 WAT

| Treatment  | Average number of shoots |
|------------|--------------------------|
|            | 4 WAT | 6 WAT | 8 WAT | 10 WAT |
| T1 (dark)  | 0.70 A | 1.65 A | 1.85 A | 1.95 A |
| T2 (light) | 0.45 B | 1.60 A | 1.90 A | 1.95 A |
| K1         | 0.00 b | 1.25 b | 1.50 b | 1.75 b |
| K2         | 1.00 a | 1.38 a | 2.00 a | 2.10 a |
| K3         | 1.50 a | 1.63 a | 1.88 a | 2.00 a |
| K4         | 0.75 a | 1.88 a | 2.00 a | 2.20 a |
| K5         | 0.63 a | 2.00 a | 2.00 a | 2.20 a |

Note: 1) WAT: Weeks After Treatment. 2) The average value followed by the same letter are not significantly different according to Duncan’s multiple test of 5%. K1-K5: GA3 (0,5,10,15,20 ppm)

The statistical analysis showed no interaction between storage place and GA3 concentration treatment and no significant difference between treatments for GA3. Visually, dark (T1) or light (T2) storage did not affect the number of shoots per tuber. From several studies, it was found that micro tubers, the tubers’ size, affected the number of shoots, which depend on the number of eyes on the tubers. The number of eyes is one of the characteristics of the variety. Generally, micro tubers only have one eye [23].

The average number of shoots up to 10 WAT was 1.75 - 2.20 per tuber. There was no effect on GA3 treatment. According to [24], [9], [14]. The number of shoots per tuber was strongly influenced by the genetics of the potato plant and the micro tubers’ size. Several factors, e.g., temperature, photoperiod, the concentration of carbohydrate sources, growth regulators used, and nitrogen content in the growth media, gave the effect of micro tuber quality [26].

### Table 3. Average shoot length (mm) at age 4,6,8, 10 WAT

| Treatment  | Average shoot length (mm) |
|------------|----------------------------|
|            | 4 WAT | 6 WAT | 8 WAT | 10 WAT |
| T1 (dark)  | 8.50 A | 16.00 A | 16.25 A | 19.15 A |
| T2 (light) | 5.50B | 15.50 A | 15.75 A | 18.25 A |
| K1         | 0.00 b | 10.0 b | 13.13 b | 14.88 b |
| K2         | 10.63 a | 18.25 a | 18.75 a | 19.00 a |
| K3         | 8.75 a | 15.63 a | 17.50 a | 20.13 a |
| K4         | 8.13 a | 18.13 a | 18.88 a | 19.00 a |
| K5         | 7.50 a | 16.25 a | 17.00 a | 18.50 a |

Note: 1) WAT: Weeks After Treatment. 2) The average value followed by the same letter are not significantly different according to Duncan’s multiple test of 5%. K1-K5: GA3 (0,5,10,15,20 ppm)

In the observation of 4,6,8,10 WAT, the average shoot length ≥ 2 mm, GA3 treatment (5, 10, 15,20 ppm) statistical analysis results were not significantly different. Although visually there was a difference
where at 10 WAT, the average shoot length was 18.50 mm - 20.13 mm, the higher GA3 concentration did not produce the highest average shoot length. The treatment that produced the highest average shoot length was GA3 with a concentration of 10 ppm. According to [25], [23], [27], the average shoot length of potato tubers is influenced by tuber size and storage after treatment.

Treatment of micro tuber storage after GA3 treatment, dark (T1) and light (T2) treatment results of the statistical analysis no different in observations 6 to 10 WAT. Visually, it can be seen that the average length of shoots in a dark place (T1) looks longer, with the quality of shoots not vigorous [12], [10]. It was shown that in the dark places, the micro tubers' shoots were etiolated [20].

Micro tubers are tubers that are produced or obtained from plantlets in vitro. These tubers are easy to handle during the distribution process due to their relatively small size. However, the utilization is not optimal because there are still many constraints on the information of potential yields, solving dormancy. However, there have been many publications that report the use of micro tubers in exchange for germplasm as basic seeds in seed production [28], [29], [30], [31], [32], [33].

4. Conclusions
The results obtained that micro tuber storage treatment after GA3 treatment in breaking dormancy did not interact between treatments. The average length of shoots in the dark (T1) was higher, but the quality was not good; the percentage of shoot growth was treated with GA3 46.88 -58.13 %. The storage place and GA3 concentration were not optimal for breaking the dormancy of cv. Granola micro tubers.

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