The Effect of GA₃ Concentration, Micro Tubers Size, and Dark or Light Storage on Breaking Potato Dormancy

A K Karjadi* and N Waluyo
Indonesia Vegetable Research Institute /IVEGRI
Jl Tangkuban Perahu No. 517 Lembang – Bandung

*Email: asihkk@yahoo.com.

Abstract. Potatoes (Solanum tuberosum L) are included in priority crops considering their function as a source of carbohydrates in food diversification. One of the techniques in producing quality seed is through tissue culture continued within in vitro tuber production that is micro tubers. The aim of this research was to observe the effect of GA₃ concentration, storage, and size of micro tubers cv. Granola, i.e. large (>4 g); small (<1 g); medium (>1 g - <4 g), and storage system dark (G), light (T). The experimental design was a complete randomized block design with 3 replications, each treatment consists of 20 micro tubers. Results of experiment micro tubers size and storage were significantly different, GA₃ concentration does not affect the percentage shoot growth which initializes the breaking dormancy of tubers at 8 to 10 WAS (Weeks After Storage). Average shoot number 0.3 – 1.25 per tuber and shoot length 0.30 – 0.80 cm. In general, each micro tuber only grows one shoot. On visual observation, the large size of micro tubers will have quality and length of shoot better than small size micro tubers.

1. Introduction
Potatoes (Solanum tuberosum L) are vegetables in the form of tubers that were rich in vitamin C and potassium. Potato is one of Indonesia’s top commodities of priority because of its alternative function as carbohydrate sources in food diversification.

Potato tubers are stem tubers; thus, the tubers can appear in the stem of in vitro cuttings. Manipulated media composition, incubation, temperature, duration of light could be used to induce the micro tubers.

Producing micro tubers can be solved the high failure rate of plantlets acclimatization, besides that micro tubers have several advantages such as easier handling for storage materials and planting. Micro tubers production represents an efficient method for obtaining healthy materials [1, 2].

The success of micro tuber formation depends on the ability of variety in producing tubers with conventional methods [3-5]. The formation of micro tubers starts from swelling stolon growing end of node/axillary. Several factors influence the formation of micro tubers in the incubation room e.g., temperature, photoperiod duration, the concentration of carbohydrates, nitrogen content, and growth regulators used in media composition. Micro tubers can be the best...
means to propagated potatoes. It had many advantages, while it saves time and space, greater output, and diseases free [6, 7].

The potato tubers that will be used for seeds are dormant when they are harvested. The length of dormancy depends on the variety, age of tuber, and quality when harvested. The dormancy period is a period that starts at the tuber initiation, and rather difficult to determine because it depends on the variety, plant growth, and maintenance of the tuber after harvest [8]. Micro tubers dormancy is various and it depends on genotype, media composition, size of micro and may be optimized by producing larger tuber on induction [9]. In other word, producing larger micro tubers by modifying the producing procedures [10, 11].

They were two kinds of potato dormancy namely internal dormancy, that is potato tuber will not germinate even if it is placed in a supportive environment. Period of dormancy called periods of rest for 4 – 15 weeks after harvest. Moreover, this condition is influenced by several factors such as the condition of growth and storage of tuber after harvest [12, 13]. After 4 – 12 weeks the potato tubers still in the period of dormancy that is in the period of external dormancy. Potato tubers that are in period external dormancy will germinate if put in an environmental condition that will support sprouting.

According to [14], potatoes' broken dormancy is the condition where 80% of the tubers have sprouted with 2 mm length-shoots. And the length of the sprout is strongly influenced by the variety, tuber size, and environmental growth of tubers. Meanwhile, according to [15], the period of tubers to planting time depends on the environmental condition during growth, age of the plant when harvest, as well as the quality of tubers at harvesting.

Gibberellic acid (GA3) is a growth regulator that can assist in the solved period of potato dormancy [16]. Breaking dormancy of micro tubers required an appropriate concentration of growth regulator, where growth regulator can be affected to plant growth and morphogenesis [17]. The treatment GA3 can stimulate the growth of shoot, also able to induce shoot growth by changing the nature shoot growth in tuber that is still dormant [18].

The aim of this research was to observe the effect of GA3 concentration, storage, and size of micro tubers on breaking dormancy. The hypothesis proposed was GA3 treatment and storage could induce the breaking dormancy of micro tubers.

2. Material and Methods
The activities were carried out in the tissue culture laboratory of Indonesia Vegetable research Institute/IVEGRI from April to July 2018. The treatment was the concentration of GA3 (10, 15, 20 ppm) and the size of microtubes cv Granola Large (>4 g), small (<1 g), and medium (1 to <4 g), storage was dark (G) and light (T). Design of experiment CRBD (Completely Random Block Design) with 5 replication each treatment consisted of 20 tubers.

The experiment was carried out throughout the following steps, micro tubers have been soaked in GA3 solution for 15 minutes and dried at room temperature. Then placed in storage dark (G) or light (T). The observation was made on 4,6,8,10 weeks after storage (WAS) e.g. (a) percentage (%) of tubers growth shoot initialize breaking dormancy (b), an average of shoot numbers and (c) an average of shoot length (cm).

3. Results and Discussion
The statistical analysis showed shoot growth which initialize the breaking dormancy micro tubers cv. Granola at 4 until 10 weeks after storage (WAS). In Table 1, the observation in 10 WAS of the percentage of sprouting tubers, was the increase in the treatment high concentration of GA3 were 50.33 - 78.33%. The statistical analysis of storage results was not different, but visually the percentage in dark conditions was higher than light there was 65.56 - 77.04%. For the micro tuber size, the large size of the MT had a lower percentage of spouted tuber higher than the small tuber.
Statistical analysis showed that there was no interaction between treatment concentration GA$_3$, micro tuber size, and storage in the percentage of breaking dormancy, storage treatment there was no effect in the percentage of breaking dormancy.

The percentage of sprouting with the length of shoots ≥ 0.2 cm start at 4 WAS continued until 10 WAS were 79.45.% to 90.80 %, where micro tuber size was no effect of the percentage shoot growth. In general, the small size of micro tubers will lead to weight loss, and that micro tubers are not yet sufficiently grown, with less dry matter content, especially low content carbohydrates [19, 20]. The micro tuber can germinate due to remobilization of chemical compounds in tuber especially starch protein and shrinkage of micro tubers weight due to water losses [21, 22, 23, 24].

Table 1. Percentage of tuber number shoot growth which initialize breaking dormancy at 4 to 10 WAS.

| Treatment | Percentage of breaking dormancy |
|-----------|---------------------------------|
|           | 4 WAS  | 6 WAS  | 8 WAS  | 10 WAS |
| GA$_3$ concentration |         |        |        |        |
| 10 ppm    | 3.78 b  | 23.25 b | 37.78 b | 50.33 b |
| 15 ppm    | 6.11 a  | 33.33 a | 50.10 a | 77.22 a |
| 20 ppm    | 6.67 a  | 37.33 a | 62.11 a | 78.33 a |
| Storage   |         |        |        |        |
| Dark (G)  | 2.85 A  | 32.33 A | 59.63 A | 77.04 A |
| Light (T) | 2.52 A  | 23.33 A | 52.22 A | 65.56 A |
| Micro tuber size |         |        |        |        |
| Large     | 9.44 C  | 58.89 B | 79.56 B | 90.80 A |
| Medium    | 5.50 D  | 53.89 B | 79.45 B | 80.56 B |
| Small     | 3.89 E  | 51.11 B | 77.78 B | 79.45 B |

Note: 1. WAS = Weeks after Storage. 2. The average value followed by the same letter are not significantly different according to Duncan's multiple range test of 5 %.

Table 2. The average numbers of the shoot at 4 to 10 Weeks after Storage (WAS).

| Treatment       | Number of shoots |
|-----------------|------------------|
|                 | 4 WAS  | 6 WAS  | 8 WAS  | 10 WAS |
| GA$_3$ concentration |         |        |        |        |
| 10 ppm          | 0      | 0.35 a | 0.50 c | 0.75 b |
| 15 ppm          | 0      | 0.40 a | 0.60 b | 0.80 b |
| 20 ppm          | 0      | 0.50 a | 0.75 a | 1.20 a |
| Storage         |         |        |        |        |
| Dark (G)        | 0      | 0.55 A | 0.70 A | 0.80 A |
| Light (T)       | 0      | 0.30 B | 0.50 B | 0.70 B |
| Micro tuber size |         |        |        |        |
| Large           | 0      | 0.75 C | 1.00 C | 1.25 C |
| Medium          | 0      | 0.50 D | 0.75 C | 0.85 D |
| Small           | 0      | 0.40 E | 0.50 D | 0.60 E |

Note: 1. WAS = Weeks after Storage. 2. The average value followed by the same letter are not significantly different according to Duncan's multiple range test of 5 %.

On the statistical analysis of storage in dark (G) and light (T), there were significant differences in GA$_3$ concentration and micro tuber size. The high concentration and large size will affect the higher numbers of shoots.

In table 2, shoots growth started at age 6 WAS, the concentration of GA$_3$ visually affected the average number of shoots per tuber. In general, each micro tuber grows only have one shoot.
The treatment of dark and light storage and micro tuber size statistically affected the average number of shoots per tuber. In the observation of 10 WAS the number of shoots, the higher GA$_3$ concentration in the average number of shoots (0.75 - 1.20), micro tuber with the bigger size, average number of shoots (0.60 - 1.25). High GA$_3$ concentration and large tuber size will increase the average number of shoots per tuber.

The average number of eyes in the micro tuber was generally one and not all will be sprouting. According to [25] and [26], the number of the eye on potato tubers was the character of the variety. Besides the number of shoots also depends on the size of tubers. With the large size of micro tubers, generally there will be more than one number of eyes. Observation of the numbers of the number of shoots increases depend on micro tubers size, in the general number of eyes will more than one for micro tubers size $\geq$ 5 g [27, 28].

### Table 3. The average shoot length on 4 – 10 Weeks after storage (WAS).

| Treatment          | The shoot length (cm) | 4 WAS | 6 WAS | 8 WAS | 10 WAS |
|--------------------|-----------------------|-------|-------|-------|--------|
| **GA$_3$ concentration** |                       |       |       |       |        |
| 10 ppm             |                       | 0     | 0.30  | 0.42  | 0.56   |
| 15 ppm             |                       | 0     | 0.45  | 0.51  | 0.63   |
| 20 ppm             |                       | 0     | 0.47  | 0.53  | 0.70   |
| **Storage**        |                       |       |       |       |        |
| Dark (G)           |                       | 0     | 0.55  | 0.65  | 0.75   |
| Light (T)          |                       | 0     | 0.40  | 0.56  | 0.60   |
| **Micro tuber size** |                      |       |       |       |        |
| Large              |                       | 0     | 0.50  | 0.60  | 0.80   |
| Medium             |                       | 0     | 0.41  | 0.48  | 0.61   |
| Small              |                       | 0     | 0.40  | 0.40  | 0.52   |

Note: 1. WAS = Weeks After Storage. 2. The average value followed by the same letter are not significantly different according to Duncan’s multiple range test of 5%.

For the average shoot length (cm), it was seen that the higher GA$_3$ concentration and large tuber size will have a high average shoot length. However, dark storage had shoot longer than light storage. The results of statistical analysis at 10 WAS, GA$_3$ treatment of 10 ppm and 15 ppm were not significantly different for the average shoot length.

Generally, the size of micro tubers will produce a length difference of shoot, and the large micro tubers have longer shoots. According to Khadige [29] and [30], the length of shoots in micro tubers was influenced by micro tuber size and storage treatment [31].

On observation of shoot length on 6,8,10 WAS statistical analysis on GA$_3$ treatment with concentration 15,20 ppm. There were no significant differences although in visual observation the high concentration has results in an average shoot length longer than low concentration but the quality of shoot not good (worse).

The treatment of micro tuber storage between dark (G) and light (T) statistical analysis was significantly different on 8, 10 weeks after storage. According to the references of [29] and [24] stated that in the dark condition the shoot condition the etiolated and bigger size of micro tubers will have an average shoot length higher than other treatment, there was no interaction between treatment.

### 4. Conclusion

Treatment of GA$_3$ concentration, micro tuber size, and light or dark storage. The high concentration of GA$_3$, large size micro tuber gave a positive effect on the percentage of shoot growth which initializes the breaking dormancy of micro tubers cv. Granola. Visual observation
large size micro tubers were had the quality and average shoot length better than small size.

References
[1] Westermann D T 2005 *Journal of Potato Research* **82** 301-307.
[2] Mani F, Mhamdi M, Bettaib T, Klannachi C 2014 *Journal of New Science* **7**(2).
[3] Wattimena G A 1983 *Micropropagation as an alternative technology for potato production in Indonesia* PhD dissertation: University of Wisconsin, Madison.
[4] Husey G, Stancy N J 1981 *Annals of Botany* **48** 787-796.
[5] Otrosky M, Struik P C 2006 *Utilization of tissue culture technology in a seed potato tuber production scheme* PhD thesis: Wageningen University, The Netherlands.
[6] Saha S, Ahmed M, Islam M M, Remme R N, Ali M R 2013 *Journal of Agriculture and Veterinary Sciences* **4**(6) 58-62.
[7] Islam M S, Roni M Z K, Jamaluddin A F M, Shimasaki K 2017 *Plant Omics* **10** 15-19.
[8] Aksenova N B, Sergaev I L, Konstantinova I M, Golya Nouskaya O O, Romannova G A 2013 *Journal Physiology* **60** 301-312.
[9] Ranalli P 2007 *Journal of Potato Research* **50** 301-304.
[10] Seabrook J E A, Douglass L K, Arnold D A 2004 *Journal of Potato Research* **81** 1-5.
[11] Zakaria M, Hossain M M, Khalequemam M A, Sossain T, Uddin M Z 2008 *Journal of Agricultural Research* **33**(3) 419-425.
[12] Wiltshire J J, Cobbs A H 1996 *Annals of Applied Biology* **129** 553-59.
[13] Turnbull N G N, Hankel D E 1985 *Planta* **165** 359-365.
[14] Suttle J C 2004 *Journal of Potato Research* **81** 253-262.
[15] Ezekiel R, Singh B 2003 *Journal of Plant Physiology* **8** 141-144.
[16] Farnshin H, Abbas Z, Enayat R 2014 *Journal of Experimental Biology* **4** 90-102.
[17] Mohammadi M S, Kashani A, Vazan S, Hasan F 2014 *Journal of Biological Science* **4** 100-108.
[18] Ashayi M M, Kurrazi A, A Sharifi, Mehrvar M 2012 *Journal of Biology and Environmental Sciences* **6** 175-180.
[19] Galun E 2010 *Phytohormones and patterning: the role of hormones in plant architecture* World Scientific Publishing Company.
[20] Khazace S R, Nezami A 2009 *Journal of Horticultural Sciences* **23**(2) 61-67.
[21] Coleman W K 1987 *Journal of Potato Research* **64** 57-68.
[22] Sonnewald V 2001 *Plant Science* **6** 333-335.
[23] Bronhe F, Sonnewald U, Biemelt S 2007 *Heidelberg Springer* 297-315.
[24] Hassan P D, Yarni M, Khorsidi B 2007 *Journal of Agricultural Sciences* **4** 81-84.
[25] Struik P C 2007 *Potato Research* **50** 375-377.
[26] Haque M 2010 *Plant Journal Documents* **3**(1) 7-11 ISSN 1836 -3644.
[27] Sharad S C 2008 *Potato production, Processing and Marketing* Biotech Book: England.
[28] El S A, Matter M A, Girgis N D 2015 *Journal of Agriculture and Environmental Science* **15**(10) 134-139.
[29] Khadige T, Alireza R, Alireza S 2014 *Journal of Agricultural Sciences* **59**(3) 255-264.
[30] Classens M M, Vreugdenhil D 2000 *Potato Research* **43** 347-369.
[31] Hemberg T 1985 *Potato Rest In: Li PH* (ed) Potato Physiology Academic Press: NY 353-388.