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Emergence of Garlic (*Allium sativum* L.) as Influenced by Low Storage Temperature and Gibberellic Acid Treatments

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Authors' contributions

This work was carried out in collaboration between all authors. Author FW designed, analyzed, interpreted and prepared the manuscript. Authors KW and GT corrected the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

The cost of garlic cloves together with inconsistent emergence due to dormancy issues which needs rest or dormant period of more than three months, has contributed to the reduction of garlic production cycle in a year. Thus, pre-planting garlic clove treatment with Gibberellic acid (GA_3) plant hormone and cold temperature storage duration are an important agronomic concern for dormancy breaking of local garlic (*Allium sativum L.*) variety. Greenhouse experiment was conducted in the spring cropping season of 2013 to investigate the effect of GA_3 concentration (0, 125, 250 and 375 ppm) and cold storage ($7^{\circ}C$) durations (10, 20, and 30 days) on emergence of local garlic cultivar. The treatments were laid out in factorial arrangement with three replications using completely randomized design. The interaction effect of GA_3 and cold storage duration

significantly increased on emergence percentage (96.66%), emergence rate (1.09) and number of sprouted cloves 0.447(100%) at GA_3 125 ppm for 20 days of cold storage duration. In conclusion the result of the experiment revealed that treatment GA_3 125 ppm with 20 days of cold storage duration is required for attaining optimum emergence performance of the local garlic variety in Haramaya Eastern Ethiopia.

Keywords: Cold temperature; gibberellic acid and emergence of garlic.

1. INTRODUCTION

Garlic (Allium sativum L.) is a member of Alliaceae family. It is an erect biennial herb normally grown as annual crop. It originated in the northwestern side of the Tien-Shan Mountains of Kirgizia in the arid and semi-arid areas of central Asia [1]. It is used for seasoning in many foods worldwide. It also has higher nutritive value compared to other bulb crops in addition to antibiotics like garlicin and allistatin it contains [2].

In tropical Africa, garlic is grown during the cold season in the Sahel region and high elevations in east Africa and in South Africa [3]. In Ethiopia, the total area under garlic production in 2012 was 21,558 ha and production was about 222,548 metric ton with average yield of 10.5t/ha. Garlic is produced for home consumption and as a source of income to many peasant farmers in many parts of the country [4]. The per capita consumption of garlic crops was estimated at about 1.74 kg and 5.9 kg in the rural and urban areas, respectively [5].

Even though garlic production throughout the year under rain- fed and irrigation conditions is possible, its potential has not been fully exploited due to scarcity of planting material, in addition to genetic, environmental and management factors. Garlic production is limited by planting material scarcity, due to the seed clove requiring resting period of more than three months after harvest as well as relatively short shelf life that do not sustain cloves to next cropping season. Garlic cloves require more than three months of ambient temperature and relative humidity storage for more uniform sprouting [6]. Despite beneficial effects on storing and availability of bulbs for human consumption, garlic dormancy creates problems for immediate use as planting material [7]. Use of cloves as planting material before completion of their innate dormancy period leads to garlic uneven sprouting.

Clove sprouting and emergence are controlled mainly by temperature and its storage period

[8,9]. During storage, sucrose and starch are consumed at high content and a part of non-consumed carbohydrates causes glucose accumulation of garlic tissue, enhancing sprouting [10]. Cloves treated at 10°C for 30 days storage temperature increased sprouting and percentage of emergence [11]. The inductive effect of cold storage depends not just on temperature regime but also on its duration depending on cold requirements for breaking bulb dormancy [12].

Plant growth regulators have been also known to play a vital role in sprouting of garlic [13]. Gibberellic acid is today known as a very efficient stimulating factor for breaking dormancy and increasing sprouting in many agronomic and horticultural crops [14].

The promoting effect of GA_3 treatment is often attributed to mobilization of stored reserves [15]. It was observed that the percentages of sprouting were higher at 250 ppm than at 125 or 500 ppm [13].

Farmers normally use garlic cloves as planting material after long storage period at room temperature that extend its dormancy to the next cropping year. Thus, it restricts the production cycle twice a year under rain fed and irrigation condition. Future prospects of Ethiopian small scale and large scale irrigation scheme is to increase vegetable production, including garlic, through increasing the frequency of production cycle per year. Thus, an increase in demand for garlic planting material is expected. To supplement this using freshly harvested garlic clove seed as a planting material after treating with growth promoter hormones and low storage temperature would be imperative.

Many studies have indicated that the application of growth regulators and low temperature storage period can shorten dormancy period of bulb crops, including garlic. However, there is scarce information regarding the effects of Gibberellic acid and low storage temperature treatments on sprouting of local garlic cultivars. Thus, the

objective of this study was to evaluate the effectiveness of application of different concentrations of GA_3 and low storage temperature on emergence of garlic.

2. MATERIALS AND METHODS

2.1 Experimental Site Description

The research was conducted at Haramaya University glasshouse research site during 2013 spring cropping season. The study area is located in the Eastern Hararghe zone of Oromia Regional State, 14 km west of Harar town and 508 km east of Addis Ababa, Ethiopia. Geographically, it is located between latitudes 9° 24' 53.13" N and 9° 24' 51.34" N, and longitudes 42° 01'55.69" E and 42° 01' 56.62" E at an altitude of 1980 meter above sea level.

2.2 Treatments and Experimental Design

The experiment consisted of 4 x 3 factorial combinations of Gibberellic acid concentrations (0, 125, 250 and 375 ppm) and storage durations (10, 20, and 30 days) stored at low temperature (7°C) (Table 1). The experiment was carried out in a glasshouse to evaluate sprouting and emergence parameters of garlic seed cloves. The treatments were laid out as a completely randomized design in a factorial arrangement where each treatment was replicated three times.

2.3 Experimental Procedures

Uniform and fresh bulbs of a local garlic variety, harvested at physiological maturity, were obtained from a farm in Haramaya University area. The bulbs were cured for one week and separated into cloves. The cloves were stored at low temperature storage (7 $^{\circ}$ C) for 10, 20, and 30 days. Then those stored cloves were soaked in Gibberellic acid solutions at concentrations of 0,125, 250, and 375 ppm for 24 hours. Gibberellic acid solution was made by dissolving Gibberellic acid powder in a drop of 70% ethanol enough to wet the powder for each concentration. Then it was mixed with distill required water to get the treatment concentrations.

A controlled experiment was undertaken in a glasshouse. A total of 20 cloves were planted on a seed tray per experimental unit. Planting was made on bench filled with seedbed media (3:2:1 soil, manure and sand mix ratio, respectively). Then proper watering of the seedbed was done until the trial was terminated. Sprouted and emerged cloves were recorded daily starting from one week after planting until one and half month, the period when cloves were expected to completely emerged.

2.4 Data Collection

Data on sprouted and unsprouted cloves and days to emergence were recorded from all planted cloves (20 cloves) in each experimental unit. The cloves establishment count was determined as percentages and rate of emerged cloves. As suggested by [16], emergence percentage (%) and emergence rate was recorded for each treatment as follows:

2.4.1 Emergence (%)

Determined as (Number of normal seedlings per total number of cloves planted) *100.

Table 1. Treatments used in the study

No	Treatments								
	Gibberellic acid (ppm)	Cold temperature storage (Days)	Combinations						
1	0	10	GA ₃ 0 x Day 10						
2	0	20	GA ₃ 0 x Day 20						
3	0	30	GA ₃ 0 x Day 30						
4	125	10	GA ₃ 125 x Day 10						
5	125	20	GA ₃ 125 x Day 20						
6	125	30	GA ₃ 125 x Day 30						
7	250	10	GA ₃ 250 x Day 10						
8	250	20	GA₃ 250 x Day 20						
9	250	30	GA ₃ 250 x Day 30						
10	375	10	GA ₃ 375 x Day 10						
11	375	20	GA ₃ 375 x Day 20						
12	375	30	GA ₃ 375 x Day 30						

2.4.2 Emergence rate

Determined as Number of normal seedlings/ (days of first count) +...+ Number of normal seedlings / (days of final count).

2.4.3 Sprouted cloves

It was determined as the sum total of all sprouted cloves.

2.4.4 Unsprouted cloves

It was determined as the sum total of all non-sprouted cloves (Total number planted – total number sprouted).

2.5 Statistical Analysis

Data collected for all parameters were subjected to analysis of variance (ANOVA) using statistical analysis system institute package (SAS, 2010 version 9.1). Comparisons of treatment mean values were made by least significant difference (Fisher's LSD) and Duncan's Multiple Range Test (DMRT) at 5% probability.

3. RESULTS AND DISCUSSION

3.1 Sprouted Cloves

Data presented in Table 2 indicated that the interaction effects of GA3 and cold storage duration was highly significant (P< 0.01) for the number of sprouted cloves of garlic. The interaction effect data showed that combinations of GA₃ at 125 ppm and 20 days of cold storage. GA₃ at 250 ppm with 20 and 30 days of cold storage were equally effective in enhancing sprouting of all cloves of garlic planted, 0.447 (100%). Cloves that received 250 GA₃ for 10 days cold storage treatments gave the lowest percentage, 0.420 (88.33%) of sprouted cloves. In general, cloves treated with GA₃ at 250 ppm, except those treated with cold for 10 days. appeared to be better in improving percent sprouted cloves. This result is in agreement with those of [13] who observed that the percentages of sprouting in garlic were higher at 250 ppm than at 125 ppm or 500 ppm. Similarly, author [14] found that Gibberellic acid is a very efficient stimulating factor for breaking dormancy and increasing sprouting in many horticultural crops.

Cold storage of cloves for 20 or 30 days also significantly increased sprouting, especially with supplement of GA₃ at 250 ppm. Temperature below 10 was effective in breaking dormancy of

garlic [17]. Similarly, [11] reported that at storage of 10°C for 30 days increased sprouting of cloves. Garlic sprouting at low temperature is attributed to a complex of biochemical reactions which is carried out by interposition of different enzymes [18,19] that increase glucose content associated with sprouting, as glucose increment means sprouting initiation of garlic [10,18].

3.2 Unsprouted Cloves

percentage of unsprouted cloves The 0.141(10.00%) was found to be higher in the treatment 10 days of cold treatment alone, 125 ppm GA₃ concentrations with 30 days prior cold storage treatments and 375 ppm GA₃ concentrations with 10 days of cold storage treatment. This might be due to dormancy in freshly harvested garlic bulbs whose sprouting depends on air temperature and storage period [18,19] as well ineffectiveness of low rates of GA₃ to replace cold treatment or probably toxicity at increased dose of GA₃ [20,21].

3.3 Emergence Percentage (%)

The results obtained from the greenhouse experiment exhibited that the effect of GA_3 concentration and its interaction with cold storage duration were highly significant (P<0.01) and the effect of cold storage duration was also significant (P<0.05) on seedling emergence percentage of garlic (Table 2).

Cloves stored under cold for 20 days and treated with GA_3 125 ppm gave the highest percentage (96.66%) of seedling emergence, which was statistically similar to values recorded in all cold stored cloves for 20 or 30 days regardless of GA_3 treatment, except those stored for 30 days and treated with either 125 ppm or 375 ppm GA_3 . Moreover, combination GA_3 125 and 250 ppm with 10 days of cold storage had significantly similar percentage of germination. On the other hand, cloves treated with GA_3 125 ppm with 30 days of cold storage had the lowest (66.66%) seedling emergence percentage.

In general, cloves treated with GA_3 250 ppm at all level of cold storage duration showed better seedling emergence percentage. This result agreed with observation of [13] who reported that concentrations of GA_3 in the same magnitude significantly improved percentage emergence in garlic. From the effect of cold storage duration extending from 10 to 30 days improved emergence percentage of garlic seedlings that agree with results of [11] who reported that the

Table 2. Interaction effects of Gibberellic acid and cold storage temperature duration on seedling emergence percentage (%) and rate, sprouted and unspotted cloves of garlic

Treatment	Sprouted cloves (%)		Un sprouted cloves (%)		Emergence	
	Transformed Mean	Original Mean	Transformed Mean	Original Mean	Percentage	Rate
GA ₃ 0 x Day 10	0.435c	95.00c	0.100e	5.00e	71.66cd	0.53def
GA ₃ 0 x Day 20	0.424de	90.00de	0.141cd	10.00a	83.33a-d	0.61de
GA ₃ 0 x Day 30	0.443ab	98.33ab	0.033f	1.66f	91.66ab	0.86bc
GA ₃ 125 x Day 10	0.439bc	96.66bc	0.100e	5.00e	85.00abc	0.56def
GA ₃ 125 x Day 20	0.447a	100.00a	0.000g	0.00f	96.66a	1.09a
GA ₃ 125 x Day 30	0.424de	90.00de	0.141cd	10.00a	66.66de	0.42efg
GA ₃ 250 x Day 10	0.420ef	88.33ef	0.152cd	11.66ab	85.00abc	0.69cd
GA ₃ 250 x Day 20	0.447a	100.00a	0.000g	0.00f	88.33abc	0.56def
GA ₃ 250 x Day 30	0.447a	100.00a	0.000g	0.00f	80.00a-d	0.84bc
GA ₃ 375 x Day 10	0.424de	90.00de	0.141cd	10.00a	78.33bcd	0.61de
GA ₃ 375 x Day 20	0.435c	95.00c	0.100e	5.00e	80.00a-d	0.51d-g
GA ₃ 375 x Day 30	0.428d	91.66d	0.127de	8.33cd	76.66bcd	0.45efg
LSD(0.05)	0.0067**	3.17**	0.0276**	2.94**	17.14 **	0.20 **
CV (%)	0.9	2.04	18.0	25.71	13.03	19.31

^{**} significantly different at 1% probability level; Means followed by the same letter within a column are not significantly different, at 5 % or 1% levels of significance (LSD); CV=coefficient of variation. Data transformed by square root

highest percentage of emergence was found in bulbs stored at 10℃ for 30 days. Similar with this study, [22] reported that pre-chilled cloves at 4℃ for three weeks gave increased emergence percentage (95.6%) than non-chilled (74.6%) cloves of garlic. Cold storage for 30 and 40 days reduced the dormant period and increased field emergence [23]. The proportion of seed clove germination was raised by the use of gibberellins and/or cold treatment could probably be due to rise of amino acid content in embryo, which is the source hydrolytic enzyme needed for digestion of endospermic starch when seeds restart growth at germination [24,15,25]. Such greater seedling emergence by GA₃ and/or cold storage would enable faster treatments crop development, early maturity and higher yields.

3.4 Emergence Rate

Significant difference on emergence rate of garlic seed clove were observed due to GA_3 concentration (P<0.01), cold storage duration (P<0.05) and their interaction (p<0.01) (Table 2). Cold storage of cloves for 20 days and treatment with GA_3 at 125 ppm resulted on the greatest seedling emergence rate (1.09). The lowest seedling emergence rate (0.42) was recorded from cloves that treated for 30 days cold storage with GA_3 at 125 ppm treatment. Increase in cold storage durations also increased at the rate of seedling emergence when no GA_3 was applied. The higher value indicated that 30 days cold storage duration had good ability to break garlic

dormancy earlier which could be important for successful field establishment as well as good yield than late emerged cloves. In agreement with the present observation [9,18] reported that seedling emergence in garlic has been enhanced by low temperature treatments. Storage of garlic at temperatures of 5 to 10°C than 25°C had promoted respiration rate during storage and the emergence of seedlings [10].

This exhibited that GA₃ could replace the cold storage requirement of dormant garlic for early sprouting and other early growth of the crop.

The result corroborates earlier reports by [24,15,25] that indicated presence of adequate levels of this acid, which accumulates in dormant seeds during cold storage and/or when exogenously applied, stimulates the synthesis, activation and secretion of hydrolytic enzymes, releasing reducing sugars and amino acids which are essential for embryo growth.

4. CONCLUSIONS

Greenhouse experiment was conducted to determine the effect of cold storage duration and GA₃ concentration on emergence performance of garlic. The results showed that the combination of 125 ppm GA₃ with cold storage for 20 days resulted in a highly significant difference in all of those emergence parameters. GA₃ at 250 ppm combined with 20 and 30 days of cold storage produced highly significant improved number of

sprouted cloves while extending cold storage duration alone highly significantly improved emergence percentage and rate.

Generally, the results of the present study showed highly significant effects of GA₃ concentration and cold storage duration on emergence of garlic. It could, thus, be concluded that using treatments of GA₃ at 125 ppm with cold storage duration at 20 days could be optimum for breaking dormancy of the local garlic variety and production of two cropping a year under rain-fed and irrigation conditions around Haremaya for the aim of supplementing planting material. However, it is difficult to reach a conclusive recommendation since the experiment was conducted at one location and one season. Therefore, this research should be repeated at different area using improved and high-yielding local varieties of garlic with clove cutting and other dormancy breaking methods is suggested for possibility of improving emergence of fresh seed cloves.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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