

Effects of Planting Depth and Sunshine Drying on Expression of Greenness at the Tip of Peeled ‘Hongsan’ Garlic Cloves

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Abstract: The expression of green color at the tip of peeled ‘Hongsan’ garlic cloves is one of the pertinent issues for growers to maximize cultivation area and support regional marketing of the product. The purpose of this study was to find the effects of planting depth and sunshine drying on the expression of greenness at the tip of peeled ‘Hongsan’ cloves. Garlics cloves were planted at three soil depths – 5 cm (control), 7.5 cm and 10 cm and the harvested garlics were assessed their greenness on the tip of peeled cloves at one, two, five and ten days after sunshine drying in the field. The results have revealed that planting depth at control, 7.5 cm and 10 cm following sunshine drying of one day after garlic harvest (AGH) showed the greenness (1–4 scale) values of 2.7, 1.9 and 1.7 in the tip of peeled cloves, respectively. The shallower the planting depth, greener the tip of the peeled cloves in ‘Hongsan’ garlic. As the number of days for sunshine drying prolonged AGH, the intensity of greenness increased at all planting depths. The intensity of greenness at the tip of peeled cloves increased from 2.7 to 3.1 as the number of sunshine drying days AGH increased from the first to the tenth at control. Meanwhile, garlic yield between the planting depths was non-significant ($p \leq 0.05$) though there are significant differences in emergence of the garlics. Thus, greenness at the tip of peeled ‘Hongsan’ garlics can be regulated by manipulating soil depth and sunshine drying days AGH in field conditions.

Keywords: Chlorophyll, Emergence, Fresh weight, Hardneck, Total phenols

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Introduction

Globally, garlic (*Allium sativum* L.) is the second most important *Allium* crop. It is well-known for medicinal and nutraceutical properties in addition to its common usage as a vegetable and condiment [1]. Garlic is rich in sugar, protein, fat, calcium, potassium, phosphorus, sulfur, iodine, fiber, silicon, and vitamins [2]. The demand of garlic in the world is increasing due to its medicinal benefits and economic significance. Korea is the fourth largest garlic producer after China, India

and Bangladesh. In Korea, garlic is cultivated in 24,700 ha with the total production of 3.18 million tons. There are many garlic cultivars cultivated across the Korea. Among them, 'Hongsan' is one of the popular hardneck cultivar, developed in 2016 by the National Institute of Horticulture and Herbal Science (NIHHS), Korea [3]. Garlic 'Hongsan' is cultivated both in cold and warm climates, and green color at the tip of peeled garlic cloves is a prominent feature for the identification. Higher contents of chlorophyll *a*, chlorophyll *b* and total chlorophyll have reported in 'Hongsan' cloves compared to 'Daeseo' and 'Namdo'. Chlorophyll is a bioactive molecule, exhibits wide range of health beneficial effects, such as antioxidant, antimutagenic, anti-genotoxic, anti-cancer, and anti-obesogenic activities [4]. 'Hongsan' is popularly grown in Hongseong region of Korea, covered 880 ha. Besides, it is cultivated in the central region, such as Seosan, Uiseong and Danyang, and known to be a long-dormant type with a strong spicy taste and good storage properties. Despite its popularity in Hongseong region, the persistence of greenness at tip of the peeled cloves is the prominent issues among garlic growers which is hindered its marketing and distribution.

Prior researches of the garlic 'Hongsan' have done in the planting depth, plant geometry, bulb sizes, and harvesting and sowing times [5,6]. The optimal harvesting time (June 20) has been identified to obtain the highest bulb weight in 'Hongsan' in the Midwest region of Korea. In the comparison of chemical properties of the 'Hongsan' and 'Hansan' cultivars, 'Hongsan' showed higher total phenol, flavonoid contents and antimicrobial activity than 'Hansan' [7]. Yoo et al. [8] also reported a higher content of total phenolic compounds in 'Hongsan' garlic than those of 'Namdo', 'Daeseo' and 'Uiseong'. Varieties, bulb size, soil moisture content, depth of ploughed layer and climatic conditions affect the planting depth [9,10]. The effect of planting depth on the morphology, physiology and flower qualities of lilies has been well-documented [11]. In addition, deep planting has been reported to improve the growth and productivity of several crop species including *Lilium*, *Crocus* and *Curcuma* [12].

Despite the many studies have been done on numerous aspects of the garlic 'Hongsan' in Korea, cultivation technology aimed at regulating green color of the peeled garlic tips has not yet been investigated. In the past, manipulation of the planting depth to reduce the amount of greenness in potato tubers were studied in different countries [13-15]. Mogren et al. [16] have studied field curing of the onions after lifting and leaving them for ten days, and they reported increased flavonoid content in bulbs. 'Hongsan' garlic has a dense peduncle structure and takes a long time to dry. As of yet, no research has been published to address the issue of greenness at the tip of peeled 'Hongsan' garlic after sunshine drying in the field. After harvesting, farmers often leave garlics in the field for sunshine drying in order to reduce rotting loss during the storage. The assessment for greenness appearance at the tip of peeled garlics cloves following sunshine drying over the time has poorly understood. Appearance of greenness at the tip of peeled 'Hongsan' garlics depends on the growing regions and growers' concerns are now increasing to retain greenness. Understanding the expression of greenness at the tip of peeled garlic cloves at appropriate soil depth, followed by appropriate sunshine drying time in the field would help growers for further expanding the territory of 'Hongsan' cultivation as well as also promote sales and distribution at the regional level. Therefore, the purpose of this study was to regulate the expression of greenness at the tip of peeled 'Hongsan' garlic cloves by adjusting planting soil depth and sunshine drying period AGH under field conditions.

Materials and Methods

Plant materials and field cultivation

The widely grown garlic 'Hongsan' was used in the study. After harvesting of preceding season garlic 'Hongsan', followed by proper drying, it was then stored at a well-ventilated room for two months. The day before planting, cloves were separated and treated them with Castelan (Sungbo Chemical Co. Ltd., Seoul, Korea), and Manroopo (Hankook Samgong Co. Ltd., Seoul, Korea) for an hour, followed by drying it under the shade. The experimental field was prepared at Muan-gun (latitude 34°55'55" N and longitude 126°25'39" E, altitude of 22 meter above sea level), Jeollanam-Do, Korea. The ridge bed was prepared and mulched by transparent plastic film with black-strip types after one week of fertilizer application. The pre-plant fertilization

(N-P₂O₅-K₂O) was done on September 23, 2022 at the rate of 90–77–45 kg/ha and it was top-dressed equally to the plots at the rate of 160–0–83 kg/ha [17]. The first top-dressing was done on February 23, 2023 and the second was applied one month after the first top-dressing. Prior to plant, the stick was marked at 5 cm (control), 7.5 cm and 10 cm with a meter scale and prepared the soil depth accordingly in the plots, and it was repeated four times in a randomized complete block design. At each treatment, a total of 400 uniform-sized cloves were planted manually on October 19, 2022 at the spacing of 12 cm × 16 cm. The recommended agronomic practices were followed for garlies outlined by the Rural Development Administration.

Observations and statistical analysis

At 40 days after planting (DAP), emergence of the plants was recorded and expressed in percentage as (total cloves planted/number of plants emerged) × 100. The garlic bulbs were harvested on June 9, 2023 when the plants turned dry and yellow. The harvested garlies were left in the field to dry under sunshine for one, two, five and ten days at each planting depth. Altogether, six persons were surveyed the garlies at each planting depth for each four times (one, two, five and ten days) of sunshine drying AGH. Right after harvesting, followed by each sunshine drying day, six individuals were assessed phenotypically on 15 garlic samples and evaluated the intensity of greenness into 1-4 scale (Fig. 1) at each replication, where 1, No appearance; 2, Slightly appearance; 3, Progressive appearance and 4, Distinct appearance. The individual rating scale for each sample and each replication was averaged. The total fresh weight produced as each plot was used to calculate the garlic yield.



Fig. 1. Intensity of greenness (1-4 scale) on the tip of peeled garlic cloves in 'Hongsan'.
1, No appearance; 2, Slight appearance; 3, Progressive appearance; 4, Distinct appearance.

Data were subjected to analysis of variance (ANOVA) using Minitab Statistical Software (version 20.0, Minitab Inc. State College, PA, USA). Tukey's HSD (Honest Significant Difference) test at $p \leq 0.05$ was employed to separate the means of traits and ordinal (rating) data. Correlation coefficients between the variables were estimated using Microsoft Excel (version 16.0, Microsoft, Redmond, WA, USA).

Results and Discussion

Effect of planting depth on emergence (%)

The planting depth significantly affected on the emergence ($p \leq 0.05$) of garlic. At 40 DAP, the garlic planted at control treatment exhibited higher emergence than those of 7.5 cm and 10 cm depths. The emergence between 7.5 cm and 10 cm planting depths showed non-significant (Fig. 2). Compared to control, plant emergence was decreased by 2.6% and 5.3% at 7.5 cm and 10 cm soil depths, respectively, whereas it was decreased by 2.7% at 10 cm soil depth as compared to 7.5 cm. A highly significant

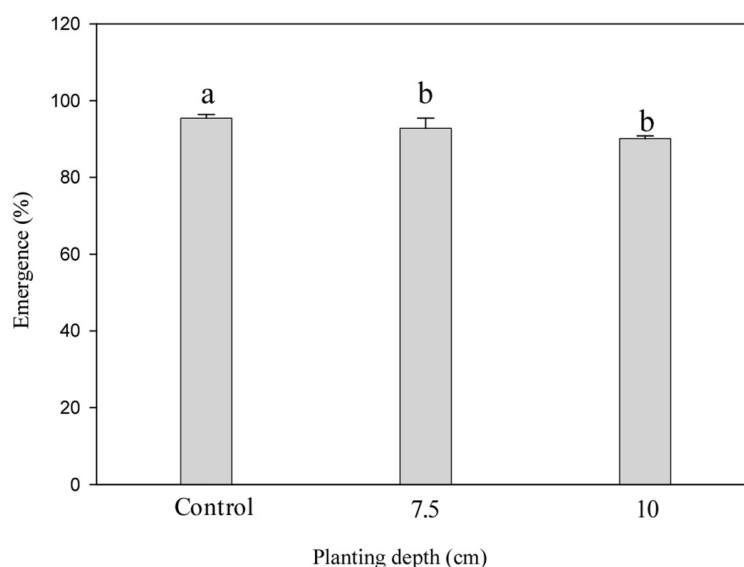


Fig. 2. Effect of planting depth on emergence (%) of garlic in the field at 40 DAP.

Data presented are the mean±SD of four biological replicates. Different letter in a bar signify significant differences between treatments, Tukey's HSD (Honest Significant Difference) test, $p \leq 0.05$.

($p \leq 0.01$) strong negative correlation ($r = -0.99^{**}$) was observed between planting depth and emergence of cloves (Table 1). This indicates that cloves planted at more soil depth tend to emerge less in the field and vice-versa. Light intensity, wavelength, photoperiod and temperature affect the seed germination [18]. Moreover, the depth of soil from at which a seed can germinate varies depending on plant species [19]. Planting depth has direct impact on seed-to-soil contact as well as seeds' access to adequate moisture and temperature. The emergence of garlic at increased planting depth was decreased that might due to lower soil temperatures and water content. The effect of planting depth on seed germination has also been studied in *Alternanthera tenella* previously [20]. The linear reduction of seedling emergence in deep seeding is reported in ginseng [21]. Our results are consistent with the findings of [9], who found a higher percentage of bulb emergence in shallow planting depth than deep planting. According to [22], planting onion bulbs deep in the soil profile has an impact on the metabolic process, which slows down the process of differentiation and microstalk formation. Furthermore, the adverse effect of deep planting has been reported on the bulb traits of *Ornithogalum saundersiae* [23].

Effect of planting depth and sunshine drying days on appearance of green color

Planting depth significantly affected on the appearance of green color (Fig. 3; $p \leq 0.001$). The garlic planted at control (5 cm) showed significantly higher green color at the tip of peeled cloves than those of others. The green color at control appeared more than 2.5 value (1-4 scale) indicated the progressive appearance. However, green color in the peeled garlic tips between 7.5 cm and 10 cm planting soil depth showed non-significant ($p \leq 0.05$). The differences in greenness in varied planting depths are shown in Fig. 4. Garlic harvested from control showed higher greenness (Fig. 4A) than those of 7.5 cm (Fig. 4B) and 10 cm (Fig. 4C). The garlic planted at shallow depth might be exposed to the above-ground temperature and light which affected

Table 1. Correlation coefficients between planting depth (cm) and emergence (%) of garlic 'Hongsan' cloves

Variable	Planting depth (cm)	Emergence (%)
Planting depth (cm)	1.0	-0.99**
Emergence (%)	-0.99**	1.0

**Significant at $p \leq 0.01$

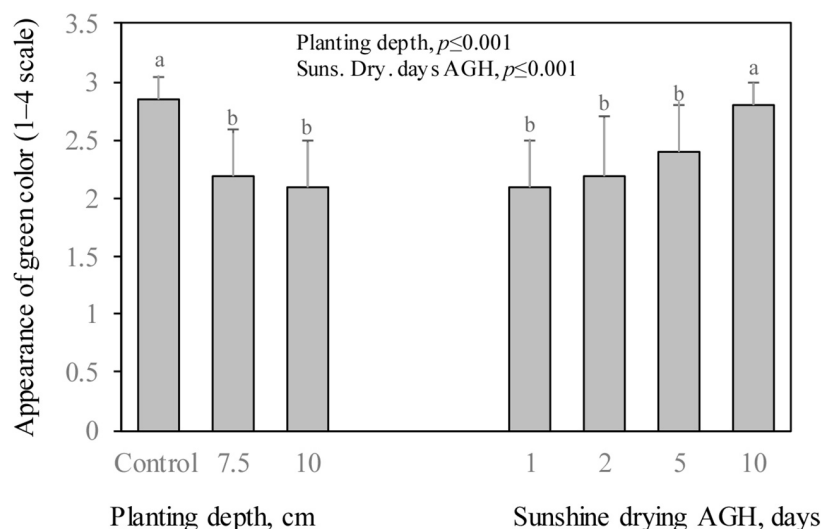


Fig. 3. Effect of planting depths and sunshine drying days AGH on appearance of green color (1-4) in tip of the peeled cloves.

Data presented are the mean±SD of four biological replicates. Different letters signify statistically significant differences between treatment Tukey's HSD Test, $p \leq 0.05$. Green color (1-4): 1, No appearance; 2, Slightly appearance; 3, Progressive appearance; 4, Distinct appearance.



Fig. 4. Appearance of green color (1-4 scale) in the tips of peeled garlics harvested at different planting depths in field.

Planting soil depths; Control (5 cm), 7.5 cm and 10 cm. 1, No appearance; 2, Slightly appearance; 3, Progressive appearance, 4, Distinct appearance; A, Distinct appearance; B, Slightly appearance; C, Progressive appearance.

the growth of the crop. The higher greenness in potato tubers has been reported at the lowest planting depth (10 cm) than that of planting at 25 cm depth [15]. Besides, a significant difference was observed in peeled color of the different garlic varieties [8]. It has been reported that total chlorophyll contents in the peeled 'Hongsan', 'Namdo' and 'Daeseo' and garlics were 6.29

µg/g, 3.71 µg/g and 1.76 µg/g, respectively. Similarly, variation in chlorophyll *a*, *b* and *ab* content has reported at cloves of red garlic 'Azarshahr' [24]. It is likely that shallow planting of cloves emerged first, exposed to light for prolong periods of time which expressed of genes involved in chlorophyll synthesis and maintained apparent greenness at the tip of peeled cloves. The light intensity affects gene expression, which is required for plants to synthesize a chlorophyll molecule. Light has a major impact on the properties of polysaccharides and the synthesis of phenolics, and flavonoids [25,26]. Furthermore, planting depth of 8 cm to produce high quality garlics has recommended in 'Namdo' and 'Danyang'. The number of sunshine drying days AGH showed the significant ($p \leq 0.001$) variation in greenness at tip of the cloves and it was observed the highest in ten days after sunshine drying. However, greenness of the cloves tip was insignificant between one, two and five days of sunshine drying AGH (Fig. 3). Amyloplasts are plastids, commonly found in vegetative plant tissues such as tubers (potatoes) and bulbs. Garlic bulbs contain a variety of pigments and secondary metabolites including lycopene, carotenoids, flavonoids and total phenols, which are influenced by temperature and light intensity [27]. The curing temperature affects the biochemical composition of the skin, particularly in flavonoids and anthocyanin, and their contribution to color change of the onions skins [28]. Prolong exposure of garlic bulbs to sunlight during field curing triggered enzymatic reaction such as maturation of plastids into chloroplasts that might have contributed to significant variation in green color after ten days of sunshine drying. During bulb formation, soluble sugars such as glucose, fructose and sucrose are stored which is synthesized in leaves by photosynthetic process, and are used as energy sources for the subsequent plant metabolic processes [29].

Soil depth significantly ($p \leq 0.05$) affected on the appearance of greenness at each one, two, five and ten days AGH (Table 2). The planting of garlic at control had the highest green color in one, two, five and ten days AGH. However, greenness of the peeled garlic tips was non-significant between 7.5 cm and 10 cm soil depths in all observations. Greenness of tuber reduces as planting depth increases [13] which also confirms to our results. The greenness in tip of the peeled garlic cloves increased progressively with increased sunshine drying days AGH in the field. It was recorded the highest (3.1 in 1-4 scale) in ten days AGH and the lowest (2.7 in 1-4 scale) in one day of sunshine drying AGH in the field. It is interesting to note that the longer the sunshine drying days AGH, more greenness appeared at tip of the cloves. The photoperiod and temperature in different durations in the field curing of garlics might influence on pigments contents, phytohormones and secondary metabolites including phenolic acids which could be reason of developing green color at tip of the peeled cloves. Light and temperature enhance phenolic acids, endogenous phytohormones, and allicin contents of the garlic cloves [30]. Field curing of onion bulbs could influence on their flavonols and anthocyanin levels [28]. The effect of temperature during field curing on the development of onion skin color has also been well-reported [28]. Mogren et al. [16] reported that field curing after lifting the onion, leaving for ten days significantly increases the flavonoid content of the bulbs.

Effect of soil depth on garlic yield

The effect of soil depth on garlic yield was non-significant ($p \leq 0.05$; Fig. 5). In general, morphological and biochemical proc-

Table 2. Effect of planting depth on appearance of green color (1-4 scale) at tip of peeled 'Hongsan' garlic cloves over the sunshine drying days AGH

Planting depth (cm)	Sunshine drying AGH, days			
	1	2	5	10
5 (Control)	2.7±0.4 a	2.8±0.3 a	2.9±0.3 a	3.1±0.2 a
7.5	1.9±0.4 b	2.0±0.3 b	2.2±0.3 b	2.7±0.2 b
10	1.7±0.1 b	1.8±0.1 b	2.1±0.4 b	2.6±0.2 b
F-Test	*	*	*	*

Means±Standard Deviation (n=4). Means at each column followed by same letter(s) are not significantly different between treatments, Tukey's HSD (Honest Significant Difference) Test, $p \leq 0.05$. Green color (1-4), 1: No appearance, 2: Slightly appearance, 3: Progressive appearance, and 4: Distinct appearance.

*Significant at $p \leq 0.05$.

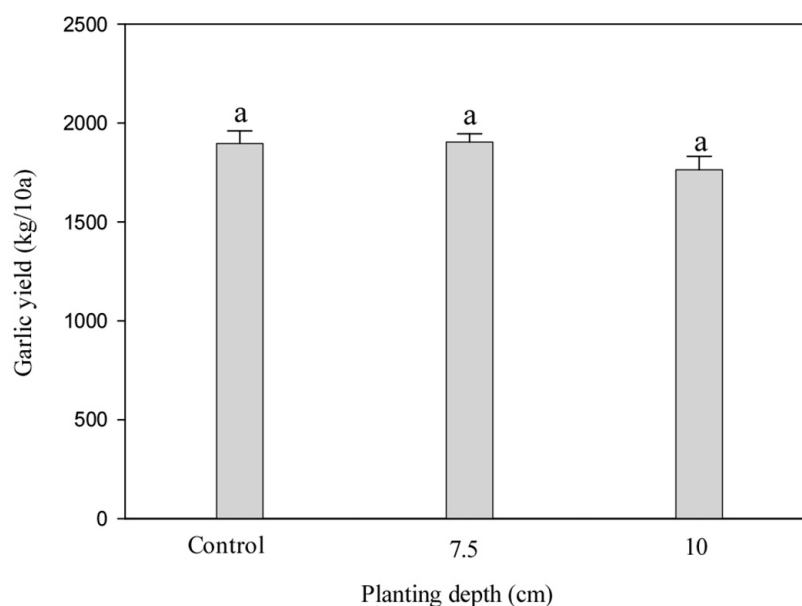


Fig. 5. Effect of planting depth in garlic yield (kg/10a).

Data presented are the mean $\pm$ SD of four biological replicates Same letters are not significantly different between treatments, Tukey's HSD Test, $p\leq 0.05$.

esses of the plants slow down at increasing planting depth [22]. Though the germination was higher at control, garlic yield at all planting depths found statistically non-significant. The initial phase of vegetative growth in garlics might be delayed due to increase depth of planting, but plants could recover their growth in later stages of development which might be contributed to obtain better garlic yield in all planting depths. Furthermore, lower germination at planting depth 7.5 cm and 10 cm may lessen plant competition for soil moisture and nutrition which in turn may increase bulb size and produced higher yield. Similar findings have been reported in garlic [31]. Besides, the unwavering formation of the assimilation by leaves of the garlic plants could have attributed to produce stable yields. In contrast, planting soil depth at 2 cm has been suggested to obtain higher bulb yield with better quality in onions [10].

Conclusion

Variations in planting depth showed the impact on emergence and appearance of the green tips on peeled garlic cloves. Green color was apparent when cloves were planted at a depth of 5 cm (control). After harvesting, tips of the garlic cloves becomes greener when it is exposed to sunlight for ten days AGH in the field. As the sunshine drying period progressed, the presence of green color intensified at all planting depths, but it was distinct at shallow planting depth (control). The effect of planting depth on garlic yield was non-significant. This study first described the result of greenness expression at tip of the peeled 'Hongsan' garlic based on phenotypic assessment that were grown under different soil depths, followed by sunshine drying AGF. Furthermore, the effects of planting depth and sun drying period after harvest on physico-chemical properties of the garlic should be investigated in detail in order to determine the degree of greenness at tip of the peeled 'Hongsan' variety.

Data Availability: All data are available in the main text or in the Supplementary Information.

Author Contributions: Luitel B.P. performed the analysis and wrote the manuscript. Lim, T.J. designed the experiment, collected

the data and wrote jointly the manuscript; Finally, Lim, T.J. Lee, H.J. and Choi. M.S. designed and edited the manuscript.

Notes: The authors declare no conflict of interest.

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Additional Information:

Supplementary information The online version contains supplementary material available at <https://doi.org/10.5338/KJEA.2024.43.13>

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