

The Effects of Calcium and 6-Benzylaminopurine on the Growth of Pistachio Seedlings in Hydroponic Culture

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Information	Abstract
<p>Article Type: Original Article</p>	<p>Introduction: Pistachios are among the major horticultural products in Iran. The production of strong seedlings is required to give a high crop yield.</p> <p>Materials and Methods: A hydroponic culture medium was used in this study to control the environmental conditions of cultivation more effectively. For this purpose, pistachio seeds, from Badami Zarand cultivar, were planted in 18 pots, with each pot containing 3 seeds. Two quantities of 5 and 25 mM calcium acetate were added to a modified Hoagland solution in a hydroponic culture medium. Next, 60 ppm of 6-benzylaminopurine was sprayed 3 times per 2 weeks on 9 pots, including 3 replications of treatments. As a result, 6 treatments were produced, including a blank one, calcium 5, calcium 25, 6-benzylaminopurine, 6-benzylaminopurine and calcium 5, as well as 6-benzylaminopurine and calcium 25. After 4 months, the seedlings were harvested, with some growth factors, including stem height, stem diameter, as well as root and shoot dry and wet weight measured.</p> <p>Results: The effects of 6-benzylaminopurine were significant on most of the mentioned growth factors, except seedling height. There were different results concerning the treatments of the 2 amounts of calcium acetate, in which calcium 5 as well as 6-benzylaminopurine and calcium 5 yielded better results. In the end, the best treatment was determined to be 6-benzylaminopurine and calcium 5.</p> <p>Conclusion: According to the results of this study, using calcium at low concentrations is useful for the growth of pistachio seedlings. In addition, using growth regulators improves growth factors, such as root dry and wet weight as well as stem diameter. Applying the findings of this study could be useful in producing stronger plants under stressful conditions.</p>
<p>Article History: Received: 9 Dec. 2018 Accepted: 28 Feb. 2019 DOI: 10.22123/phj.2019.174473.1026</p>	
<p>Keywords: Calcium 6-Benzylaminopurine Pistachio Growth Factors</p>	
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► Please cite this article as follows:

Nadi M, Pakdaman N, Javanshah A. The effects of calcium and 6-benzylaminopurine on the growth of pistachio seedlings in hydroponic culture. *Pistachio and Health Journal*. 2019; 2 (1): 22-28

1. Introduction

Pistachios (*Pistacia vera* L.) are among the most important products planted widely in Iran. In recent years, there have been various environmental problems in the production of pistachios. Some of these problems have been the results of mismanagement, and some others have been due to climate changes. The production of strong seedlings could be a good solution under these conditions. Calcium is one of the prominent mineral elements useful in improving fruit quality and shelf life. Plants have evolved so as to rely on calcium properties for structural, enzymatic, and signaling functions [1]. It has been reported that calcium plays an essential role in strengthening cell membranes and preventing damage to plants, pollen germination, pollen tube growth, and fruit formation [2]. Calcium is a counter-cation that acts against organic and inorganic anions in the vacuole, and its cytosolic concentration is an obligate intracellular messenger, which coordinates responses to numerous developmental cues and environmental challenges [3]. Calcium pectate is a compound in plant cells' middle lamella, and when its amount is sufficient, it can prevent pectin wall demolition [4]. It has been reported that calcium protects cell membranes against damage inflicted by various stresses [5]. The problems of absorbing calcium from soil and calcium inactivity in the airstream have many physiological complications in fruit trees, which greatly reduce product quantity and quality [6]. Calcium inhibits sodium uptake, thereby reducing its harmful effects on seed

germination [7, 8]. It can also increase plant growth [9, 10]. In addition, calcium is effective in plant signaling, water relations, and cell wall interactions [11]. Ekinic [12] reported that calcium humate applied at different rates positively affected the total marketable yield, leaf dry matter, average fruit weight, fruit length, and fruit diameter in tomato and cucumber plants. It has been demonstrated that the spraying of some nutrients and calcium can improve the growth and increase the yield in pistachio trees in Fandoghi cultivar [13]. Another factor that affects the yield and quality of horticultural products is plant growth regulators [14]. Benzylaminopurine is a first-generation synthetic cytokinin that induces plant growth and development responses, sets blossoms, and stimulates fruit richness by promoting cell division. Cytokinins are crucial growth substances with various effects when applied to some plants. In addition, it is proven that they are the most important factor expediting shoot regeneration [15]. Benzylaminopurine promotes shoot and axillary bud proliferation [16]. Cytokinins play a significant role in plant adaptation to environmental stresses [17]. This study aims to investigate the effects of calcium and benzylaminopurine on some growth factors in pistachios.

2. Materials and Methods

The seeds of *P. vera* L. cv. at "Badami Zarand Pistachio Rootstock" were prepared from Pistachio Research Center (PRC) in Rafsanjan County, Kerman province, Iran. To grow the seedlings, firstly the dehisced seeds of pistachios were pre-chilled 10 days at 4°C

and then soaked 2 days in water. After covering the seeds in a wet cotton fabric [18], the germinated seeds were transferred into perlite hydroponic culture pots. Three germinated seeds were planted in each pot. Next, 2 g/L of modified Hoagland in the powder form, including 20.85% N, 2.62% P, 19.32% K, 19% Ca, 4.13% Mg, 0.045% B, 0.052% Mn, 0.004% Zn, 0.002% Cu, 0.001% Mo, 0.24% Fe, 5.54% S, and 0.4% fulvic acid was used as the nutrition solution, with calcium acetate added to it in 2 quantities. The nutrition solution was refreshed once in a month. At the next stage, 60 ppm of 6-benzylaminopurine (BA6) along with agricultural soap were sprayed 3 times in 2 weeks. Calcium acetate and 6-benzylaminopurine treatments were applied after two months from the growth of the seeds. In order to produce the BA6 solution, NaOH was used in a crystalline form to reach full solubility. The treatments included blank (B), two quantities (5 and 25 mM) of calcium acetate (Ca5 and Ca25), BA6 (H), calcium 5 and 6-benzylaminopurine (Ca5H), as well as calcium 25 and 6-benzylaminopurine (Ca25H) in three replications. After 4 months, the seedlings were harvested, and some growth factors, including stem height, wet and dry weight of root and shoot, as well as stem diameter were measured. Data were analyzed in Excel and SPSS. A *t*-test was used to evaluate the differences between the results.

3. Results

Fig. 1 shows the results for the stem diameter and height of seedlings. In fact, treatments Ca5 and Ca5H yielded better results (Fig. 1a). According to the *t*-test, there have been significant differences among treatments B, Ca5, Ca5H, and H. However, Ca5 and Ca5H showed no significant differences in the mentioned factors (Fig. 1a).

According to the results, there was not significant difference between stem height in treatments Ca5 and Ca5H (Fig. 1b). Treatment Ca5 showed significant differences in this factor with other treatments except Ca5H (Fig. 1b). In the case of root dry weight, treatments Ca5 and Ca5H resulted in higher weights and showed significant differences with all other treatments (Fig. 2a). However, there was no significant difference between treatments Ca5 and Ca5H in this factor (Fig. 2a). In addition, there were significant differences between treatments Ca5 and Ca5H and other treatments (Fig. 2b). Ca5H was the best treatment and showed significant differences with treatment Ca5 in this factor (Fig. 2b). Ca5H was the best treatment in root wet weight (Fig. 3a), and there were significant differences between Ca5H and all other treatments. In addition, Ca5 showed significant differences with other treatments except treatment B in this factor (Fig. 3a).

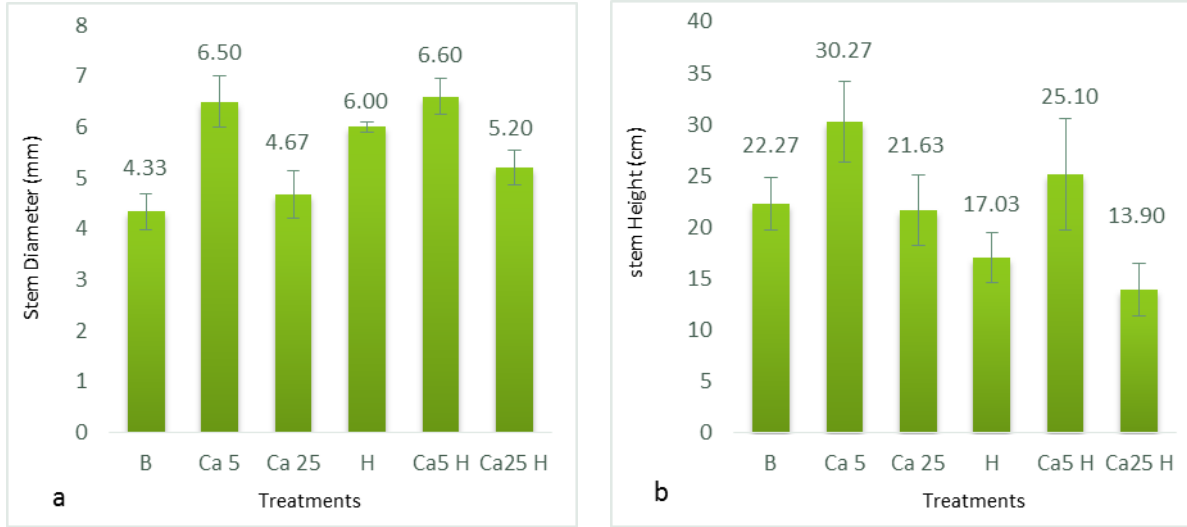


Fig. 1. The effects of the two quantities of calcium acetate and a hormone on the stem diameter (a) and stem height (b) of pistachio seedlings

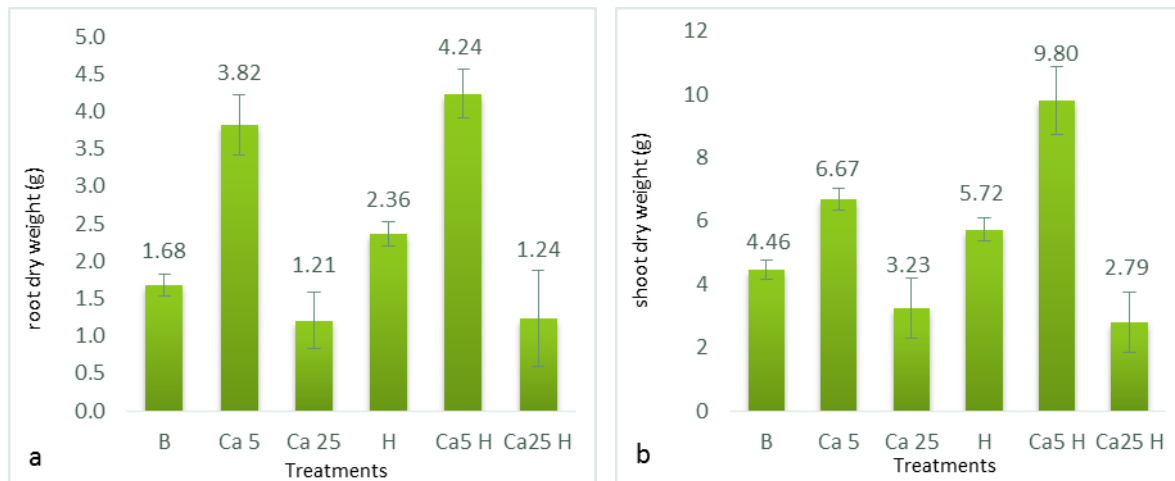


Fig. 2. The effects of the two quantities of calcium acetate and BA6 on root dry weight (a) and shoot dry weight (b) in pistachio seedlings

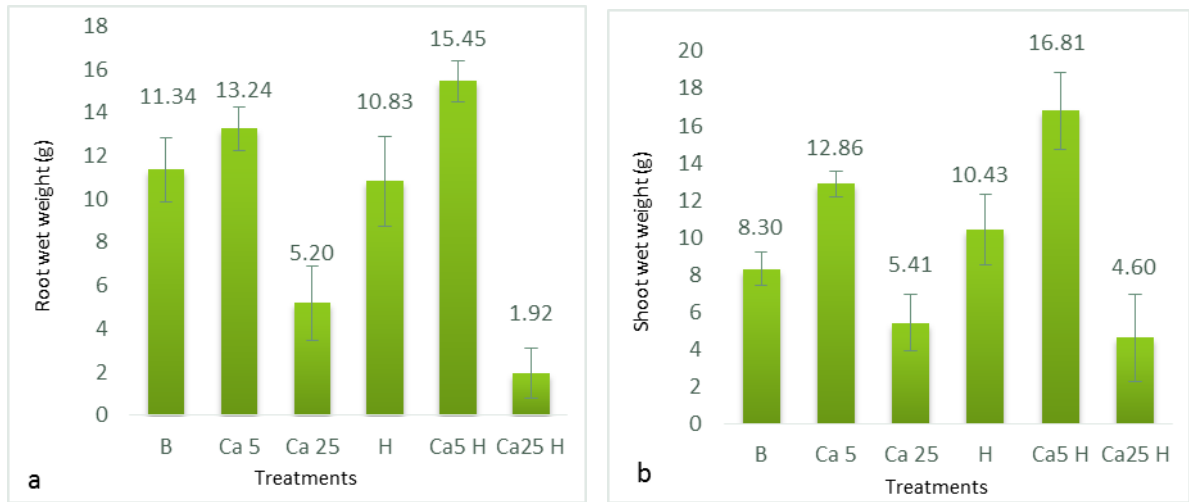


Fig. 3. The effects of the two quantities of calcium acetate and BA6 on root wet weight (a) and shoot wet weight (b) in pistachio seedlings

In the last factor studied, i.e. shoot wet weight, treatments Ca5 and Ca5H resulted in higher weights than others, and there were significant differences between these treatments with the others (Fig. 3b). Treatment Ca5H was the best treatment in this factor, and there was significant differences between it and treatment Ca5 (Fig. 3b). All and all, better results were achieved from treatments Ca5 and Ca5H than other treatments, with treatment Ca5H proved to be the best one.

4. Discussion

Calcium, as an important element, plays various roles in the growth of plants. In this study, the use of calcium in 2 quantities showed interesting results. The use of 5 mM calcium acetate led to significant results in most of growth factors. However, the use of 25 mM calcium acetate did not yield similar

results; this could be due to the effects of calcium on the increase in the concentration of the nutrient solution in the culture medium, which led to an increase in the osmotic pressure, a decrease in osmotic potentials, and some disturbances in the absorption of nutrients. According to Loneragan [19], an increase in the calcium concentration of the solution from 0.3 to 2.5 μM increased the crop yield to a large extent. In contrast, an increase in the calcium concentration from 10 to 1000 μM had small effects on the crop yield. Under excessive calcium conditions in the rhizosphere solution, plants could undergo calcium toxicity, thereby reducing plant growth [3]. Some factors, such as humidity, root pressure, and phytohormone activity could affect calcium distribution in plants [20]. In addition, the high concentration of calcium leads to the unbalanced absorption of some nutrients by plants, including phosphorus and

microelements [21]. The use of BA6 increased stem diameter as well as the dry weight of shoot and root but did not affect the height of seedlings. The increase in stem diameter as well as the dry weight of shoot and root could have happened because of BA6 effects on cell division. It has been proven that BA6, as a synthetic cytokinin, can induce plant growth and development by stimulating cell division [22]. It has been demonstrated that ascorbate and glutathione concentrations as well as their redox ratios are important factors in the survival of plants under stressful conditions [23]. According to some researchers, 6-benzylaminopurine can enhance the concentrations and redox states of ascorbate–glutathione as well as the related enzyme activities so as to improve plant tolerance under the conditions of oxidative damage [17, 24].

5. Conclusions

According to the results of the present study, the use of calcium at low concentrations is effective in the growth of pistachio seedlings. In addition, the use of growth regulators increases growth factors, such as root dry and wet weight as well as stem diameter. The results of the present study could be useful in cultivating stronger plants under stressful conditions.

Conflict of interest

The authors declare no conflicts of interest.

Acknowledgements

Here by appreciate the sincere cooperation of Pistachio Research Institute and Barafza Keshavarz Pars Company in carrying out this research.

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