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Impact of treating bitter almond seeds with dormancy-breaking applications on germination percentage and growth of the produced seedlings

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This research was conducted in the National Research Centre (NRC), Dokki, Giza, Egypt during the period 2017/2018 and 2018/2019, in order to study the effects of cold stratification (CS) periods and gibberellic acid (GA_3) concentrations on seeds germination percentage, mean germination time, and growth of produced seedling of bitter almond which used as rootstock for almond cultivars. Seeds were stratified at 5 ± 1°C for 0 (control), 2, 4, 6 weeks. At the end of every cold stratification period, seeds were soaked in GA_3 solutions at 0, 500, 1000 and 1500 ppm concentrations. Treated seeds were sown in polyethylene bags containing sandy loam soil and placed in the greenhouse to determine the germination percentage and following up the vegetative growth of the produced seedlings. The highest germination percentage and lowest mean germination time were obtained from the combination of CS periods for 6 weeks with GA_3 at 1500 ppm. The best growth of seedlings was obtained from CS for 4 and 6 weeks at the same concentrate without significant differences between them. As a result, it can be concluded that CS for 4 weeks and applications GA_3 at 1500 ppm is effective in breaking dormancy of bitter almond seeds and increasing germination percentage and growth of the produced seedlings.

Keywords: Bitter almond, cold stratification, GA_3, seed dormancy, seed germination, seedling growth.

1. Introduction

Almond (*Prunus amygdalus* L.) is one of the most important stone fruit belongs to Rosaceae family and *Prunus* genus for its nutritious and highly valued seeds. *Prunus* seeds require special treatments to overcome the dormancy and promote their germination (Rajjou et al., 2012; Hassan and Fetouh, 2014), so before germination certain chemical and physiological changes must take place in seeds to contribute germination (Ghayyad et al, 2010; Tewari et al., 2011; Pipinis et al., 2012; Ikramullah et al., 2017).

Cold stratification which exposes seeds to treatment cold moist allows breakage of embryo dormancy and enhancing the germination of seeds (Hartman et al., 1997; Hartman et al., 2002). In order to accelerate these methods, it can be combined with some treatment such as chemical applications or mechanical seeds coat treatments (Martínez-Gómez and Dicenta, 2001).

Exogenous application of gibberellic acid (GA_3) has been reported to be effective in breaking dormancy and substituting of chilling requirement in seeds of many species (Pipinis et al., 2011; 2012; Dhupper, 2013). GA_3 is an exogenous growth regulator that promotes enzymes (Hartman and Kester, 1983; Hilborst and Karssen, 1992). Seed germination is a complex process that is stored with the absorption of water after a short pause the enzyme is activated (Matilla and Matilla-Vazquez, 2008). The use of GA_3 has been studied in fruit culture as a way to increase seed germination and therefore to obtain a uniform seedling size in the nurseries (Dhupper, 2013).

Many researches dealing with the effect of stratification and GA_3 application on seed germination of different almond species because the seeds of almond species are surrounded by a hard endocarp that makes it difficult to germinate, germination rate in these species is low (Isfendiyaroglu and Ozeker, 2001). The combination of cold stratification and GA_3 pretreatments affect physiological and metabolic activities of seeds resulting in early germination (Amooaghaie, 2010; Pipinis et al., 2012; Abou Rayya et al., 2021).

The objective of this study was to determine the effect of cold stratification periods and different gibberellic acid concentrations on the germination percentage of bitter almond and produced seedling growth.

2. Materials and Methods

This experiment was carried out during two growth seasons (2017/2018 & 2018/2019) and data was represented as average of two seasons. A factorial experiment was conducted to study two factors; the first was four periods of cold stratification (CS), the second factor was four concentrations of gibberellic acid (GA_3) solutions on seeds germination percentage and subsequent seedlings growth of bitter almond rootstock.

2.1. Seeds materials and treatments

Seeds were collected from mature bitter almond fruits (*Prunus amygdalus* L.) grown in Saint Catherine located at South Sinai of Egypt. Fruits were harvested in August during both seasons and packed in plastic bags and transported to the laboratory. The fleshy outer layer (exocarp and mesocarp) was removed by hand, then the clean seeds (with endocarp) stored in burlap bags in dry place and well ventilated at room temperature till used in the experiment. Seeds were surface sterilized in 10 % chlorox for 10 min and rinsed with distilled water three times, afterwards were mixed with sand moister and placed in plastic boxes (15 × 10 × 5 cm), then kept in a cold storage at 5 ± 1°C for 0 (control), 2, 4 and 6 weeks. During stratification periods, sand moisture was checked and added distilled water whenever necessary to keep it moist 60-70%.

At the end of every cold stratification (CS) periods seeds were soaked in GA_3 solutions at 0 (control), 500, 1000 and 1500 ppm concentrations with sixteen treatments and five replicates per each treatment (ten seed per replicate). All the treatments left for 24 hours in dark condition at room temperature.

On 15th December (Thanaa et al., 2020) from each seasons the seeds disinfected in a 0.01% Rizolix fungicide solution for 10 min and sown at depth equal to three times of the seed size in polyethylene bags (30 cm diameter, 50 cm height) containing sandy loam soil as showed in Table (1) Page et al., (1982), then placed in a private nursery at Kalubia governorate, Egypt and irrigated with water daily.

Table 1. Physical and chemical properties of the experiment soil

Soil pH	**EC $ds.m^{-1}$**	**OM (%)**	**Sand (g/kg)**	**Silt (g/kg)**	**Clay (g/kg)**
7.42	0.66	3.82	482.1	153.2	64.23
Soluble cation (meq/kg soil)			**Soluble anions (meq/kg soil)**		
Ca^{++}	**Mg^{++}**	**K^{+}**	**Na^{+}**	**HCO^{-3}**	**Cl^{-}**
3.5	1.05	0.25	1.1	1.58	0.3
Available macronutrients (mg/kg soil)			**Available micronutrients (mg/kg soil)**		
N	**P**	**K**	**Fe**	**Zn**	**Mn**
0.49	0.98	0.15	1.18	1.4	1.2

2.2. Measurements

2.2.1. Germination percentage (GP)

Germinated seeds were counted every day for 30 days, from the beginning of the emergence of the cotyledons above the soil (Ghayyad et al., 2010). The germination percentage per treatment was calculated by the equation:

$$Germination\ percentage\ (GP) = \frac{Number\ of\ germinated\ seeds}{total\ number\ of\ seeds} \times 100$$

2.2.2. Mean of germination time (MGT)

Mean of germination time was calculated by the equation described by (Isfendiyaroglu and Özeker, 2002):

Where t: time in days starting from day zero, n: number of seeds completing germination on day (t).

2.2.3. Seedling growth parameters

$$MGT = \frac{t \times n}{n}$$

At the end of September from both seasons the seedlings vegetative properties were determined in terms of stem and root length (cm), diameter (mm), seedlings fresh and dry weight (g), number of lateral shoots and roots/ seeding and number of leaves/seedling.

Seedling vigor index was measured according to the formula described by (Abou Rayya et al., 2018) as follows:

Leaf chlorophyll contents index were measured by using portable chlorophyll meter model (SPAD-502 Minolta Sensing Inc., Japan) (Acar et al., 2017).

$$Seedling\ vigor\ index = germination\ percentage \times seedling\ dry\ weight$$

Statistical analysis

The obtained data were subjected to analysis of variance (ANOVA) according to Gomez and Gomez (1984**),** using CoStat Software Program Version 6.303 (2004), and LSD at 0.05 level of significance was used for the comparison between means.

3. Results and Discussion

3.1. Germination percentage (GP)

Results in Fig. 1 show the interaction effect of cold stratification periods and GA_3 concentrations on germination percentage of almond seeds; generally, GP was increasing significantly with the increase of concentrations of GA_3 from 500 to 1500 ppm during the stratification different periods. The highest germination rate 95.6% were obtained from seed stratified for 4 weeks with GA_3 application at 1500 ppm followed by 6 weeks at the same concentrate without significant differences between them. The lowest seed germination was observed for the control treatment (zero cold stratification plus zero GA_3 application) with 25.13%.

From the above results, it can be notice that the present study confirmed that dormancy-breaking treatments had an important effect on seed germination. The high concentration of GA_3 was a significant effect than the low concentrations in increasing GP seeds. This result agrees with those obtained by Aygun et al. (2009); Mohammadi et al. (2014); Suja et al. (2016) who mentioned that the germination percentage increased with increasing GA_3 concentration up to a certain point. With regard to the CS period, treated seeds exhibited higher GP than the control (0 weeks) without stratification. This result is in line with Yamauchi et al. (2004); Abu-Qaoud (2007) who mentioned that stratification is commonly used to promote and synchronize seed germination. Therefore, it means the important role of cold stratification (Al-Imam and Qrunfleh, 2007) notice that the stratification of bitter almond at 5°C was very effective for cracking the endocarp and that due to the imbibition of water seeds which in turn caused an expansion of the embryo cells of the tissue.

Also, statistical analysis indicated that GP with the long period (4 weeks) was more effective than the short period of CS (2 weeks) or control (without stratification) in increasing GP and there were no significant results between (4 and 6 weeks) when the seeds were soaked by the same concentrations of GA_3, this may due to the effect of GA_3 in shorting the chilling periods. This result is in line with Ghayyad et al. (2010) who reported that GA_3 is effective in shortening the chilling requirement.

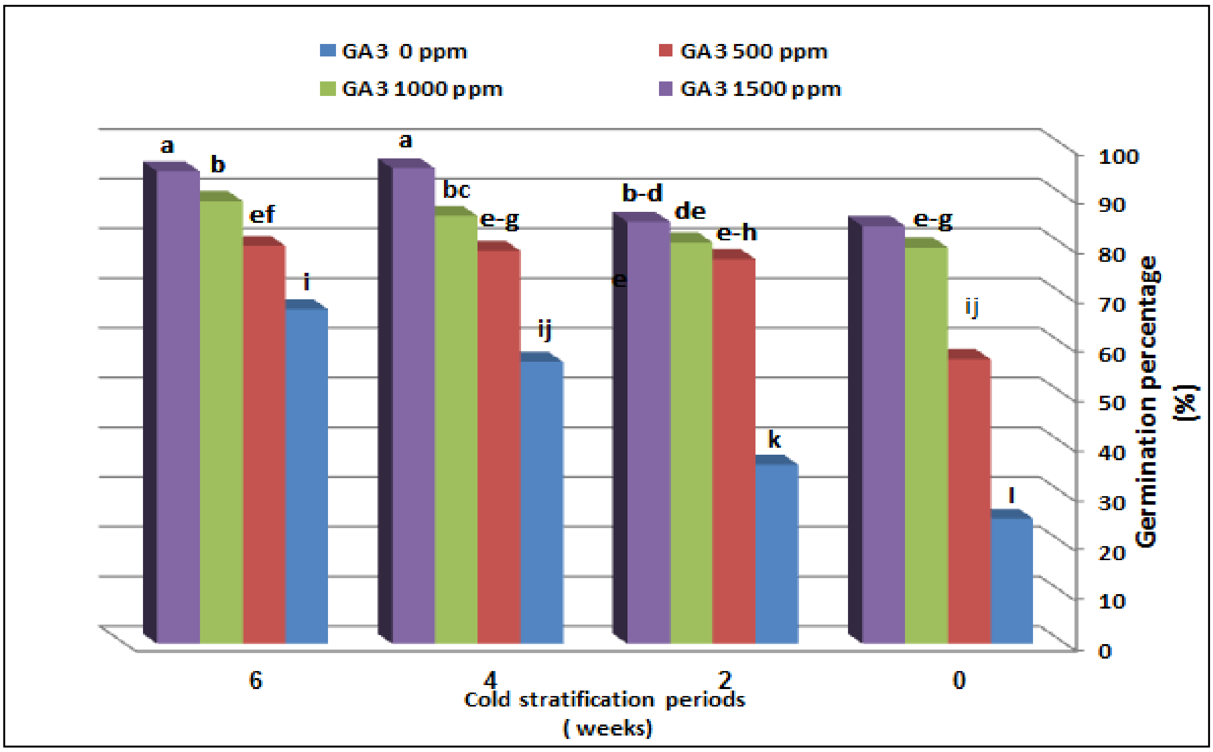

Figure 1. Interaction effects of cold stratification periods and gibberellic acid concentrations on germination percentage of bitter almond seeds.

3.2. Mean germination time (MGT)

Mean germination time decreased by combined treatments of stratification periods and GA_3 concentrations in almond seeds Fig. 2 compared to the control treatment (zero cold stratification plus zero GA_3 application). The longest MGT of complete germination was obtained from 0 CS plus GA_3 at 0, 500, 1000 ppm (28.67, 28.33 and 26.67 days), respectively as compared with other treatments. The shortest MGT of complete germination was obtained by CS for (4 weeks) combined with GA_3 at 1500 ppm, followed by CS for (6 weeks) at the same concentration (18 and 18.67 days), respectively without significant differences between them. It means that MGT significantly decreased by increasing the stratification periods until CS (4 weeks) and the high concentration of GA_3 is effective in shortening the germination time of almond seeds.

This result is near to those obtained by Sedaghat and Rahemi (2011); Esmailpour and Van Damme (2016); Thanaa et al. (2020) who mentioned that MGT significantly decreased by pre-treatments for breaking seed dormancy. Abu-Qaoud (2007) noticed that the longest mean germination time of three pistacia species was obtained from the control (0 stratifications).

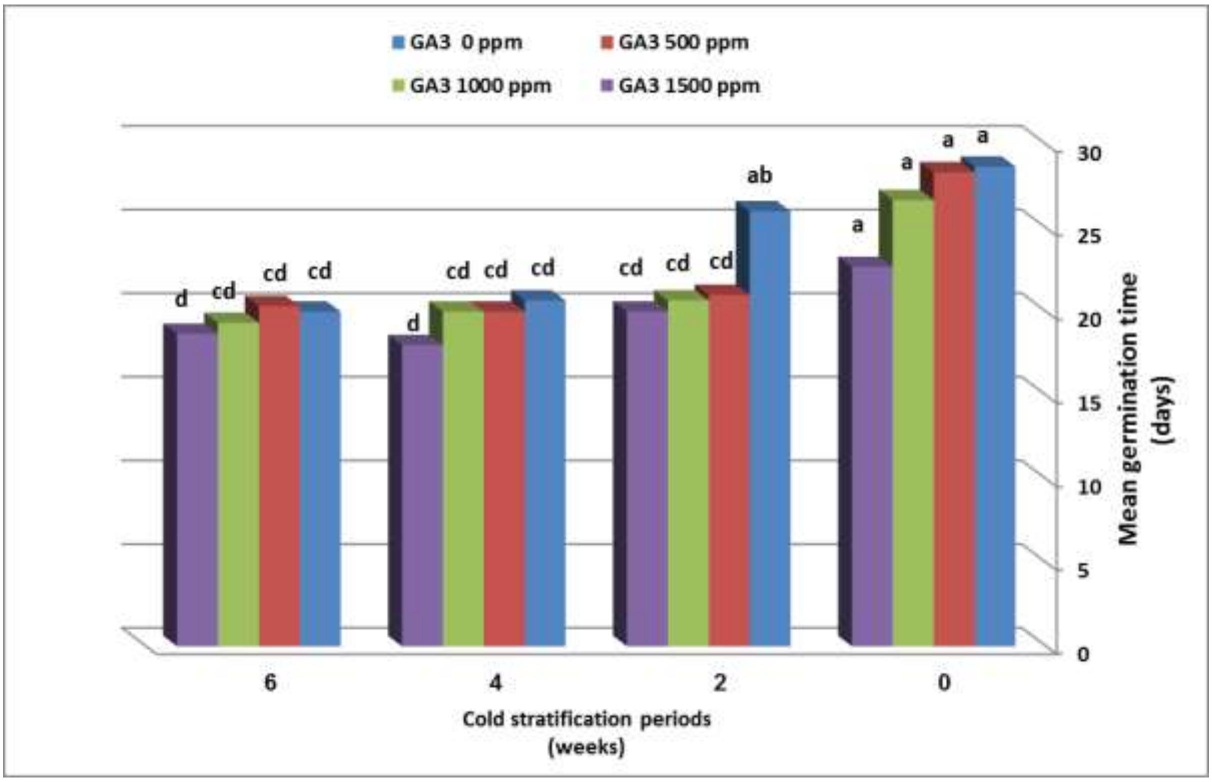

Figure 2. Interaction effects of cold stratification periods and gibberellic acid concentrations on mean germination time of bitter almond seeds.

3.3. Seedling growth parameters

Table 2 revealed that there were significant differences in stem length (cm) between different GA_3 concentrations and different periods of cold stratification than the control. The highest mean value of seedling stem length (35 cm) was obtained from the application of GA_3 at 1500 ppm with CS at (6 weeks) followed by stratified periods for (4 and 2) weeks, respectively at the same concentrations while the minimum values of stem length (15.33 cm) was obtained by 0 CS + 0 GA_3. Applying GA_3 at 1500 ppm was more effective than 1000, and 500 ppm on the stem length and that is true on every CS period.

As for stem diameter Table 3, it has tack the same trend of stem length. Data revealed that the maximum value of stem diameter (4.78 mm) was obtained from the CS treatment for (6 weeks) with GA_3 at 1500 ppm concentration, however, the control was recorded the minimum values (3.87 mm) in this respect. There was no significant difference between the other treatments. It can be concluded that there is a significant correlation between gibberellin concentration and both stem length, root length, and diameter of the seedling. These results agree with Rahemi and Baninasab (2000); Mobli and Baninasab (2008) who mentioned that GA_3 application during and after CS significantly increased the length and trunk diameter.

Table 2 showed an increase in root length by all treatments than the control with slight significant differences between the root length in the two CS periods (2 and 4 weeks) at the different concentrations of GA_3. The longest root (50.11cm) was recorded in the application GA_3 at 1500 ppm with CS at (6 weeks). While, the shortest roots (20.44 cm) was recorded by 0 CS + 0 GA_3.

Number of leaves/seedlings was significantly affected as a result of the interaction between GA_3 concentrations and cold stratification periods (Table 2). The biggest number of leaves was recorded by CS for (4 weeks) with GA_3 treatment at 1500 ppm, while the lowest number was recorded at 0 CS + 0 GA_3. CS at (6 weeks) was less than (4 weeks) CS at the same concentration in this respect; this may be due to negative responses accumulating too long up to a certain point. Moreover, there was no significant difference between applications of GA_3 at concentrations of 500, 1000 and 1500 ppm when seeds stratified for (6 weeks).

The above results may be due to the good and enhanced effect of the treatments on the length and diameter of the stem. These results are roughly in line with those achieved by Thanaa et al. (2020) who found that the large leaf area is due to the good parameters of seedling growth (stem length and diameter). Rahemi et al. (2011) Demonstrate that growth capacity can be determined by stem dimensions.

Table 2. Interaction effects of cold stratification and gibberellic acid concentrations on stem length, stem diameter, root length and leaves number of bitter almond seedlings.

Parameters	GA$_3$ con. (ppm)	Cold stratification periods (weeks)				LSD 0.05
		0	2	4	6	
Stem length (cm)	0	15.33j	17.00ij	18.00i	19.13hi	2.44
	500	18.00i	22.67fg	24.33ef	26.00de	
	1000	21.00gh	25.67de	25.67de	30.00bc	
	1500	23.67ef	28.02cd	31.33b	35.00a	
Stem diameter (mm)	0	3.87b	3.67b	4.00ab	4.00ab	0.91
	500	4.18ab	4.24ab	4.00ab	4.20ab	
	1000	4.19ab	4.27ab	4.17ab	4.30ab	
	1500	4.34ab	4.37ab	4.37ab	4.78a	
Root length (cm)	0	20.44k	24.33j	29.47g	33.00e	1.44
	500	25.00ij	26.00hi	31.00f	36.00d	
	1000	27.37h	29.00g	40.33c	44.37b	
	1500	29.67fg	32.47e	43.04b	50.11a	
No. of leaves/seedling	0	8.00f	8.33f	10.00def	11.00cde	2.16
	500	8.67f	9.00ef	11.33cd	13.67ab	
	1000	9.00ef	11.00cde	13.00abc	13.67ab	
	1500	10.00def	12.00bcd	15.00a	14.00ab	

The number of lateral shoots and roots were clearly affected by the combined treatments between stratification by long periods and GA$_3$ applications until a certain point more or less. It is remarked that there were significant differences between (4 and 6 weeks) of stratified almond seeds this respect at the same concentration. A high level of GA$_3$ concentration recorded a high number of lateral shoots and roots (Table 3). Dhupper (2013) reported that the use of GA$_3$ contributed to increasing seed germination and obtained a uniform seedling size in the nurseries.

The fresh and dry weight of almond seeding was significantly affected by the interaction between stratification periods and the application of GA$_3$ than the control (0) CS (Table 3). The biggest weight was found by the high level of GA$_3$ at 1500 ppm by the two long periods (4 and 6 weeks) respectively. These results agreed with those obtained by Mobli and Baninasab (2008) who mentioned that the application of most plant growth regulators to break seed dormancy significantly increased the fresh and dry weight of subsequent seedlings.

Statistical analysis shows the same trend in seedling vigor index and chlorophyll content index (Fig. 3 & 4). It was observed that significantly increased by increasing cold stratification periods with the level of GA$_3$ compared to the control treatment 0 CS + 0 GA$_3$ which recorded the less effect in this respect.

Table 3. Interaction effects of cold stratification and gibberellic acid concentrations on number of lateral (shoots & roots) and (fresh & dry) weight of bitter almond seedlings.

Parameters	GA_3 con. (ppm)	Cold stratification periods (weeks)				LSD 0.05
		0	**2**	**4**	**6**	
No. of lateral shoots	**0**	1.00g	1.00g	2.00efg	3.33bcd	**1.08**
	500	1.33g	1.67fg	3.00cde	3.33bcd	
	1000	1.33g	2.67def	4.00abc	4.33ab	
	1500	1.67fg	3.00cde	4.67a	5.00a	
No. of lateral roots	**0**	1.00d	2.33d	5.00c	8.00b	**1.64**
	500	2.00d	4.00c	8.00b	9.00ab	
	1000	1.67d	5.00c	9.00ab	9.00ab	
	1500	1.00d	5.00c	10.00a	10.00a	
Seedling fresh weight (g)	**0**	2.93f	5.00de	5.00de	5.00de	**1.02**
	500	3.20f	5.33de	6.00cd	5.33de	
	1000	4.83e	6.00cd	7.00bc	8.00b	
	1500	5.50de	6.00cd	9.33a	7.57b	
Seedling dry weight (g)	**0**	2.43g	3.33e-g	3.67d-g	4.33c-f	**1.69**
	500	2.83fg	4.33c-f	4.00c-g	4.33c-f	
	1000	3.83fg	5.33a-d	5.67a-c	6.67ab	
	1500	4.00c-g	5.00b-e	6.71a	7.00a	

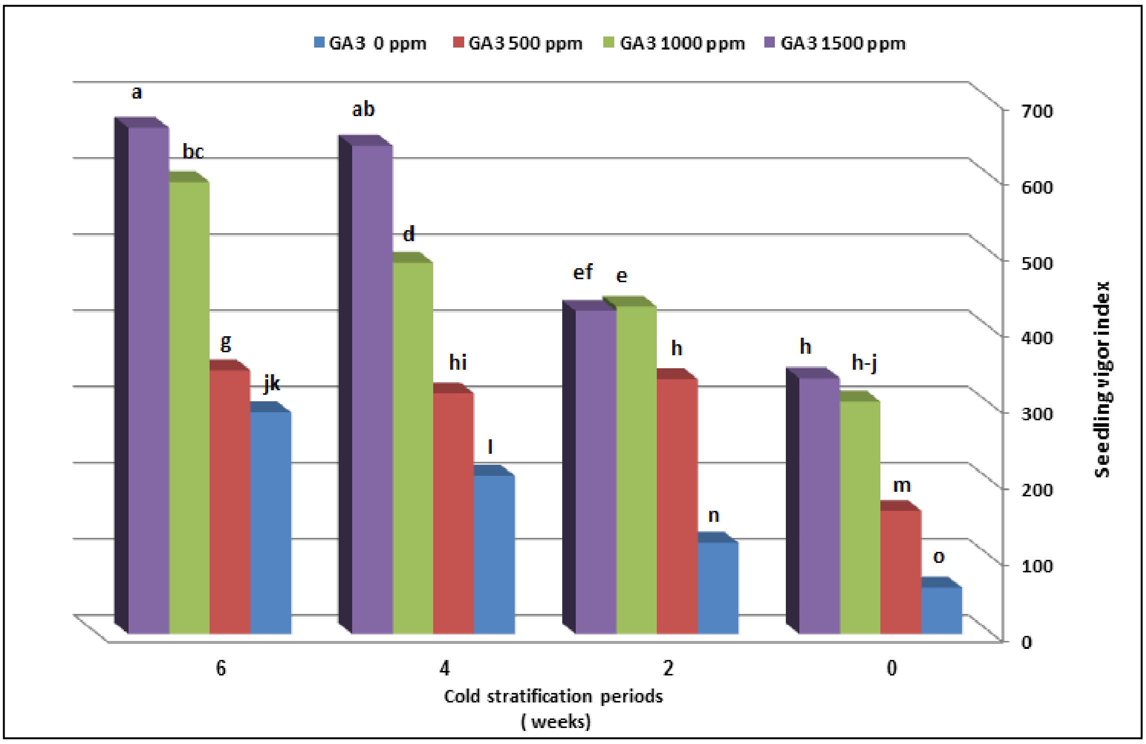

Figure 3. Interaction effects of cold stratification periods and gibberellic acid concentrations on vigor index of bitter almond seeding.

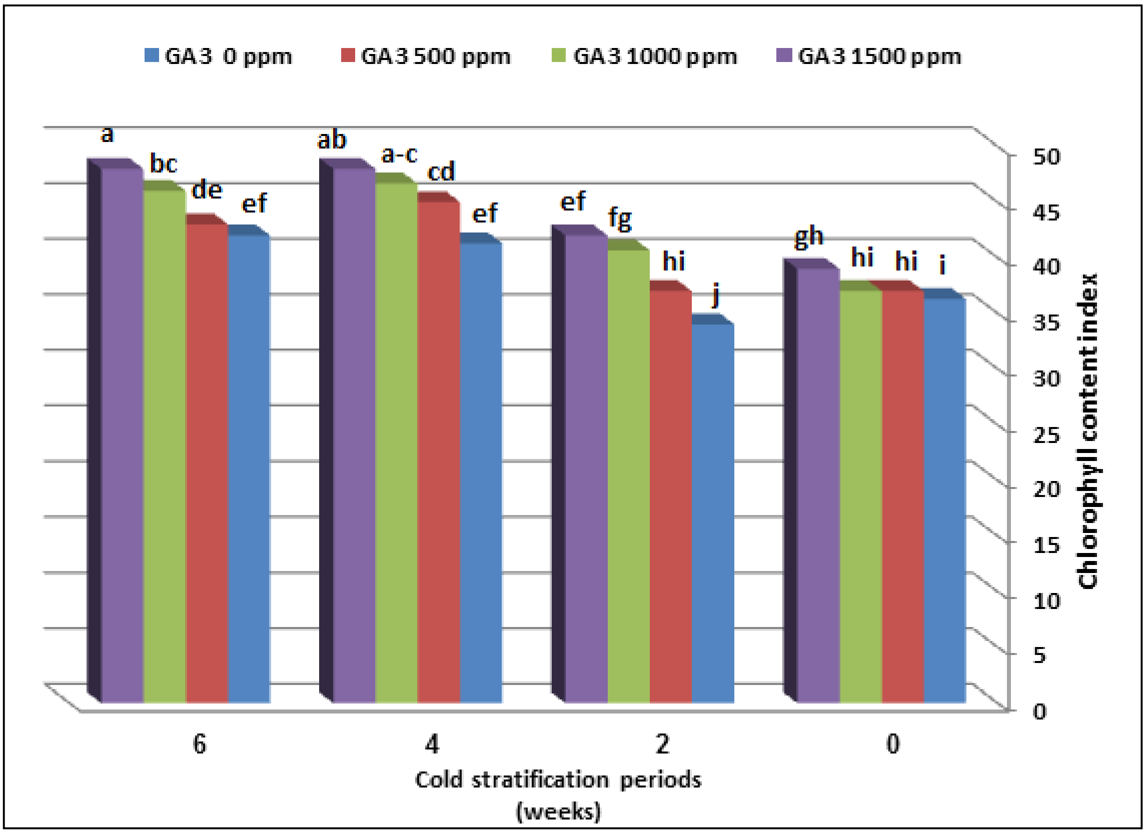

Figure 4. Interaction effects of cold stratification periods and gibberellic acid concentrations on chlorophyll content index of bitter almond seeding.

4. Conclusion

In conclusion, it was found that the combined treatments of cold stratification for 4 weeks followed by soaking in GA_3 1500 ppm solution is effective in breaking dormancy of bitter almond seeds thus, can be recommended to enhance the seed germination process and improving growth characteristics of the produced seedlings.

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Author's Contribution

MSAR and NEK designed this work. TSMM and MAS designed and conducted the field experiment treatments and following up the seeds germination and growth of almond seedlings. MAS and TSMM coordinated the data collection for analysis and performed data statistical analysis and wrote the manuscript. MSAR reviewed the manuscript. All authors read and approved the final version.

References

1. Abou Rayya, M. S., Thanaa, Sh. M., Eisa, R. A. and Nabila, E. K. (2018). Effect of water soaking periods on germination percentage and growth characteristics of "Ashoury" pistachio (*Pistacia Vera* L.) seedlings. BIOSCIENCE RESEARCH, 15(4):4274-4278.
2. Abou Rayya, M. S., Nabila, E. K., Malaka, A. S. and Thanaa, Sh. M. Mahmoud (2021). Effect of cold stratification and GA_3 on deshelled seeds germination and seedlings growth of Bitter almond. Middle East Journal of Agriculture Research, 10 (4):1173-1181. DOI: 10.36632/mejar/2021.10.4.80
3. Abu-Qaoud, H. (2007). Effect of scarification, gibberellic acid and stratification on seed germination of three Pistacia species. An-Najah University Journal for Research-Natural Sciences, 21:1-11.
4. Acar, I., Yasar, H. and Ercisli, S. (2017). Effects of dormancy-breaking treatments on seed germination and seedling growth of Pistacia khinjuk Stocks using as rootstock for pistachio trees. Journal of Applied Botany and Food Quality, 90:191-196.DOI:10.5073/JABFQ.2017.090.024.
5. Al-Imam, N. M. A. and Qrunfleh, M. M. (2007). Physiological study of the stratification effect on the dynamic changes in endogenous abscisic acid in bitter almond seeds (Prunus amygdalus L.) and its relation to seed germination. Mesopotamia Journal of Agriculture, 35(4):10-18 DOI: 10.33899/magrj.2007.26518
6. Amooaghaie, R. (2010). Effect of gibberellin and stratification on stimulating seed germination and subsequent seedling growth, of "Japanese medlar". Journal of Biology, 23: 299-308.
7. Aygun, A., Erdogan, V. and Bozkurt, E. (2009). Effect of some pretreatments on seed germination of Turkish Hazel (*Corylus colurna* L.). Acta horticulturae, 845: 203-206. DOI: 10.17660/ActaHortic.2009.845.27
8. Dhupper, R. (2013). Effect of gibberellic acid on seed germination and seedling growth behaviour in three desert tree species. The Journal of Biological Chemistry research. 30(1): 227-232.
9. Esmailpour, A. and Van Damme, P. (2016). Evaluation of seed soaking times on germination percentage, germination rate and growth characteristics of pistachio seedlings. Acta Horticulturae, 1109:107-112.DOI: 10.17660/ActaHortic.2016.1109.17
10. Ghayyad, M., Kurbysa, M., Napolsy, G. (2010). Effect of endocarp removal, gibberelline, stratification and sulfuric acid on germination of Mahaleb (*Prunus mahaleb* L.) seeds. Am-Eur J Agric Environ Sci., 9 (2):163-168.
11. Gomez, K. A. and Gomez, A. A. (1984). Statistical procedures for agricultural research. 2nd Ed New York Wiley XVI pp. 680.
12. Hartman, H.T. and Kester, D.E. (1983) Plant propagation principles and practices. 4th Ed. Prentice Hall, Englewood Cliffs, NJ.
13. Hartman, H.T., Kester, D.E., Davies, F.T. and Geneve, R.L. (2002). Plant propagation principles and practices. 7th edition prentice Hall, Upper Saddle River , New Jersey 07458.
14. Hartmann, H. T., Kester, D. E. and Davies, F. T. (1997). Plant Propagation, Principles and Practices. Prentice Hall Internacional Inc Upper Saddle River NJ.
15. Hassan, F.A. and Fetouh, M.I. (2014). Seed germination criteria and seedling characteristics of *Magnolia grandiflora* L. trees after cold stratification treatments. International Journal of Current Microbiology and Applied Sciences, 3(3): 235-241.
16. Hilhorst, H.W.M. and Karssen, C.M. (1992) Seed dormancy and germination: the role of abscisic acid and gibberellins and the importance of hormone mutants. Plant Growth Regulation.11: 225- 238.
17. Ikramullah, A.R., Rahman, J., Jehangir, M. and Llyas, M. (2017). Effect of soaking durations and sowing dates on the germination and seedlings growth of peach stones. Int. J. Agri and Env. Res., 3(1): 106 - 111.
18. Isfendiyaroglu, M., Özeker, E. (2001). The relations between phenolic compounds and seed dormancy in Pistacia spp. XI GREMPA Seminar on Pistachios and Almonds. Cahiers Options Méditerranéennes 56: 227-232.
19. Martínez-Gómez, P. and Dicenta, F. (2001). Mechanisms of dormancy in seeds of peach (*Prunus persica* L.) cv. GF305. Scientia Horticulturae, 91: 51-58.
20. Matilla, A.J. and Matilla-Vazquez, M.A. (2008). Involvement of ethylene in seed physiology. Plant Science, 175: 87-97.

21. Mobli, M. and Baninasab, B. (2008). Effects of plant growth regulators on growth and carbohydrate accumulation in shoots and roots of two almond rootstock seedlings. Fruits, 63: 363–370 DOI: 10.1051/fruits:2008032
22. Mohammadi, H., M., Imani, A. and Ebadi A. (2014). Post – germination changes in hormones, enzymatic activities and biochemical compounds of different domesticated almond varieties in response to stratification. International Journal of Biosciences, 4 (10): 298 – 308.
23. Page, A.L., Miller, R.H. and Keeny, D.R. (1982). Methods of Soil Analysis. Part 2: Chemical and Biological Properties, American Society of Agronomy Inc., Madison, Wisconsin, USA.
24. Pipinis, E., Milios, E. and Smiris, P. (2011). Effect of sulphuric acid scarification, cold moist stratification and gibberellic acid on germination of *Paliurus spinachristi* Mill. Seeds. Forestry Ideas., 17(1): 45-52.
25. Pipinis, E., Milios, E., Kiamos, N., Mavrokordopoulou, O., Smiris, P. (2012). Effects of stratification and pretreatment with gibberellic acid on seed germination of two Carpinus species. Seed Science and Technology, 40: 21-31.
26. Rahemi, A., Taghavi, T., Fatahi, R., Ebadi, A., Hassani, D., Chaparro, J. and Gradziel, T. (2011). Seed germination and seedling establishment of some wild almond species. African Journal of Biotechnology, 10 (40):7780-7786.
27. Rahemi, M. and Baninasab, B. (2000). Effect of gibberellic acid on seedling growth in two wild species of pistachio, J. Hortic. Sci. Biotech., 75:336–339.
28. Rajjou, L., Duval, M., Gallardo, K., Catusse, J., Bally, J., Job, C., and Job, D. (2012). Seed germination and vigor. Annu Rev Plant Biol 63 (1): 507–533. http://dx.doi.org/10.1146/annurev-arplant-042811-105550.
29. Sedaghat, S. and Rahemi, M. (2011). Effect of pre-soaking seeds in polyamines on seed germination and seedling growth of *Pistacia vera* L. cv. Ghazvini. International Journal of Nuts and Related Sciences, 2(3): 7-14.
30. Suja, N. ,Qureshi, M.S. and Javaid W. (2016). Effect of stratification and gibberellic acid on seed germination and seedling growth of walnut under Kashmir valley conditions. Indian Journal of Horticulture, 73(2):171-176. DOI: 10.5958/0974-0112.2016.00042.6
31. Tewari, B., A. Tewari, Shah, S., Pande, N. and Singh, R.P. (2011). Physical attributes as indicator of seed maturity and germination enhanced in Himalayan Wild cherry (*Prunus cerasoides* D. Don). New Forests 41:139-146. DOI: 10.1007/s11056-010-9216-x
32. Thanaa, Sh. M. Mahmoud, Abou Rayya, M.S., Nabila, E.K. and Malaka, A. Saleh (2020). Effect of removal endocarp and sowing dates on seeds germination and seedling growth of bitter almond rootstock. Plant Archives, 20 (2): 8347-8352.
33. Yamauchi, Y., Ogawa, M., Kuwahara, A., Hanada, A., Kamiya, Y., Yamaguchi, S. (2004). Activation of gibberellin biosynthesis and response pathways by low temperature during imbibition of Arabidopsis thaliana seeds. Plant Cell. 16 (2):367-78. doi: 10.1105/tpc.018143.