Seed priming with chitosan alleviates salinity stress by improving germination and early growth parameters in common vetch (*Vicia sativa*)

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Abstract. Soil salinity is a serious environmental threat with varied effects on several aspects of growth and production of plants. Recently, however, the utilization of chitosan in agriculture has increased as it enhances the resistance of crop plants to different stresses factors such as salinity. In this experiment, the effects of chitosan seed-priming on alleviation of salinity stress was investigated in common vetch (*Vicia sativa* L.) using a completely randomized design with six replications. Chitosan pretreatment was applied at three levels (2, 4, and 8 g/l) and salinity of sodium chloride (NaCl) was applied at four levels (50, 100 and 200, and 300 mM). Different variables have been measured, including germination percentage, hypocotyl length, radical length,
hypocotyl dry weight and radical dry weight. Salinity is found to affect the germination percentage of vetch by decreasing germination with increasing in NaCl concentration (50, 100 and 200, and 300 mM). However, the results showed an increase in germination percentage upon priming seeds with different levels of chitosan (2, 4, and 8 g/l) even with increasing the concentration NaCl salinity. Additionally, all salinity levels caused a significant reduction in vetch hypocotyl and radical dry weight and resulted in a decrease in their length when compared with the water control. However, chitosan at all levels of concentration improved all of these growth parameters (hypocotyl length, radical length, hypocotyl dry weight and radical dry weight) irrespective of salinity level indicating a successful role of chitosan in alleviating salinity stress. In general, these results may suggest that in saline soil seed pretreatment in chitosan could be utilized successfully for alleviating the effect of salinity stress on germination and on some growth parameters of common vetch.

1. Introduction

Soil salinity is one of the most serious environmental threat of agriculture, which increasingly exacerbate in different countries worldwide by human activities. Salinity causes a decrease in water potential of a plant and leads to the accumulation of Na\(^+\) and Cl\(^-\) ions that eventually limit germination and seedling establishment and reduce the growth of plants [1,2]. To deal with salinity, plants have evolved varied physiological and structural strategies that increase the utilization of water and the elimination of toxic ions from the cell [3]. Moreover, it is well documented that salinity stress raises free radicals and reactive oxygen species (ROS), damaging macromolecules and cell integrity [4,5]. Several studies indicated that salinity develops a series of morphological and physiological changes through reduced germination percentage and affect the early growth parameters, such as hypocotyl and radical length and biomass by increased the accumulation of salt in leaves and to decreased water use efficiency as well as reducing seed viability and resource allocation for germination [6]. One option is currently being in an increased use to alleviate the effect of salinity stress on different plants is by application of biopolymers-based chemical compounds such as chitosan [7]. Chitosan is a linear oligosaccharide composed of a beta-(1-4)-linked N-acetyl-2-amino-2-deoxy-D-glucose (acetylated) and 2-amino-2-deoxy-D-glucose [8]. Chitosan is obtained from chitin, which is a structural component of insect and crustacean cuticle and exoskeleton, the cell wall of fungi, and some algae [9]. Chitosan is a naturally occurring, inexpensive, and environmentally safe biopolymer with diverse beneficial biological properties, such as biodegradability, biocompatibility, and non-allergenicity [10]. Chitosan has been demonstrated to enhance varied biological responses in plants, dependent on its concentration and on plant species. Additionally, chitosan has been proved to increase defence responses of the plant to different abiotic and biotic stresses [11]. For example, chitosan has shown to able to act as germination elicitor of Oryza sativa L. [12], and to positively increase seed germination and early seedling growth of wheat (Triticum aestivum L.) [13]. In addition, nanoparticles-based chitosan was able to improve the expression of pathogenesis-related proteins, thus increasing the resistance of tomato to F. andiyazi pathogen [14].

Common vetch (Vicia sativa L.) is an economically important self-pollinating annual forage legume. Common vetch has gained valuable attention in different countries throughout the world as fodder, a cover crop, hay, green manure, and also for silage production [15]. As a legume crop, it establishes successfully in mixtures with varied cereal crops and has the excellent ability for nitrogen fixation [16]. The main goal of this study was to determine the effect of seed priming with chitosan on common vetch under different salinity stress.

2. Materials and methods

Seven different chitosan concentrations, ranging from 0 to 6 g/L (pH 6.1), were used to prime seeds. Six replicates of 60 vetch seeds were used for each treatment in this experiment. Every treatment included wetting the seeds with 12 ml of chitosan solution in petri dishes. The seeds were exposed to 25°C for 48 hours in the dark. Once the priming was complete, the seeds were allowed to completely dry. 50, 100, 200, and 300 mM NaCl were added to the seeded vetch seeds, and the four levels of salinity were labelled.
"control," "50," "100," "200," and "300" mM. For this experiment, various variables have been measured, including the rate of germination, the length of the hypocotyl, the length of the radical, the weight of the hypocotyl, and the weight of the radical.

The MSTAT software was used for analysis of variance and LSD mean separation (Michigan State University, East Lansing, MI).

3. Results and discussion

We found that germination percentage of common vetch are declined significantly with increasing NaCl indicating that salinity of different levels may affect plant growth by reducing most likely seed viability, plant growth and establishment (Table 1). This result agrees with other studies that suggested that salinity reduces seed germination by delaying germination rate and or seed viability [11]. The lowest germination percentage was found at the NaCl concentration of 300 mM of NaCl, due to a reduction in water absorption into the seeds as they soak and swell. In addition, our results show significant increases in germination percentage, regardless of salinity concentration (Table 1).

Table 1. Germination percentage of common vetch (Vicia sativa) under the combined effect of different concentrations of salinity of NaCl and chitosan.

| Chitosan concentration (g/l) | 0    | 50   | 100  | 200  | 300  | LSD (p=0.05) |
|-----------------------------|------|------|------|------|------|--------------|
| 0                           | 88   | 81   | 73   | 73   | 58   | 1.7          |
| 2                           | 90   | 84   | 81   | 75   | 59   | 2.1          |
| 4                           | 93   | 88   | 83   | 77   | 62   | 1.5          |
| 8                           | 90   | 87   | 86   | 75   | 59   | 1.6          |
| LSD (p=0.05)                | 1.7  | 1.6  | 1.9  | 1.6  | 1.6  |              |

In fact, the results revealed that all the three concentrations of chitosan (2, 4 and 8 g/l) significantly enhanced germination percentage under all salinity levels of concentration (50, 100, 200, 300) suggesting that chitosan have the potential of alleviating the stressful condition of salinity [3,14]. Additional findings have proven that the highest germination percentage was reached when vetch seed was treated with 8 g/l chitosan in accordance with other studies which have shown that chitosan coating can increase germination percentage [7]. Generally, the results on germination effect of vetch may be demonstrated that the effects of chitosan are possibly attributed to its ability in enhancing germination rate, increasing seed viability and improving the energy allocation for germination. Additionally, germination enhancement by chitosan could be attributed to the fact that chitosan is an ideal film-forming property. Furthermore, as for germination, seeds priming with different concentrations of chitosan resulted in an increase in both hypocotyl and radicle length and also in their dry weight compared to control (Table 2, 3, 4, 5).

Table 2. The effects of different combined concentrations of salinity of NaCl and chitosan on radical length (cm) of common vetch (Vicia sativa).

| Chitosan concentration (g/l) | 0    | 50   | 100  | 200  | 300  | LSD (p=0.05) |
|-----------------------------|------|------|------|------|------|--------------|
| 0                           | 2.3  | 2.0  | 1.7  | 1.5  | 1.0  | 0.70         |
| 2                           | 2.5  | 2.2  | 2.0  | 1.5  | 1.0  | 0.19         |
| 4                           | 2.5  | 2.4  | 2.2  | 1.8  | 1.1  | 0.15         |
| 8                           | 2.4  | 2.4  | 2.2  | 1.7  | 1.1  | 1.70         |
| LSD (p=0.05)                | 0.15 | 0.14 | 0.15 | 0.15 | 1.60 |              |
Table 3. Influence of seed priming with chitosan and salinity levels on hypocotyl length (cm) of common vetch (*Vicia sativa*).

| Chitosan concentration (g/l) | 0   | 50  | 100 | 200 | 300 | LSD (p=0.05) |
|-----------------------------|-----|-----|-----|-----|-----|---------------|
| 0                           | 3.1 | 2.7 | 2.3 | 2.0 | 1.5 | 1.90          |
| 2                           | 3.3 | 2.9 | 2.5 | 2.0 | 1.6 | 0.14          |
| 4                           | 3.9 | 2.9 | 2.6 | 2.2 | 1.6 | 0.14          |
| 8                           | 3.9 | 2.8 | 2.6 | 2.3 | 1.6 | 0.16          |
| LSD (p=0.05)                | 0.15| 0.14| 0.18| 0.16| 0.16|               |

Table 4. Influence of seed priming with chitosan and salinity levels on radical dry weight (g) of common vetch (*Vicia sativa*).

| Chitosan concentration (g/l) | 0  | 50  | 100 | 200 | 300 | LSD (p=0.05) |
|-----------------------------|----|-----|-----|-----|-----|---------------|
| 0                           | 0.77| 0.62| 0.47| 0.39| 0.32| 0.017         |
| 2                           | 0.80| 0.64| 0.50| 0.39| 0.33| 0.017         |
| 4                           | 0.82| 0.64| 0.52| 0.40| 0.33| 0.016         |
| 8                           | 0.82| 0.64| 0.52| 0.40| 0.34| 0.016         |
| LSD (p=0.05)                | 0.02| 0.016| 0.015| 0.016| 0.016|               |

Table 5. Influence of seed priming with chitosan and salinity levels on hypocotyl dry weight (g) of common vetch (*Vicia sativa*).

| Chitosan concentration (g/l) | 0    | 50   | 100  | 200  | 300  | LSD (p=0.05) |
|-----------------------------|------|------|------|------|------|---------------|
| 0                           | 0.93 | 0.66 | 0.83 | 0.73 | 0.63 | 0.015         |
| 2                           | 0.95 | 0.91 | 0.85 | 0.73 | 0.63 | 0.017         |
| 4                           | 1.00 | 0.93 | 0.88 | 0.75 | 0.64 | 0.018         |
| 8                           | 1.10 | 0.92 | 0.88 | 0.74 | 0.65 | 0.016         |
| LSD (p=0.05)                | 0.016| 0.019| 0.014| 0.018| 0.018|               |

These results agree with several studies on the effect of chitosan on different plant species. Additionally, these results confirm the findings of those of Chandrkrachang [15] who found that the application of chitosan could increase the growth parameters of cucumber, chilli, pumpkin, and cabbage. The exact mechanism of the effect of chitosan on plant growth is currently not well documented. Yet, the effect of chitosan we found on common vetch could be attributed to it is able in the induction of plant defence enzymes, increasing the synthesis of secondary metabolites, such as polyphenolics, lignin, flavonoids, and phytoalexins as observed in many plant species treated with this oligopolymer substance [17]. Droughts can happen where very dry soils and reduced plant cover may decrease precipitation in a dry environment [18-22]. Plants and their production is limited by environmental factors such as salinity and drought in many parts of the world [23-30]. The lack of water in dryland farming is the only significant obstacle to crop production[25-28]. Many of these studies have found that low productivity is primarily associated with dryland agriculture management practices [31-70]. Chitosan or chitin is the most common natural polysaccharide present in the cell walls of fungi, crabs, shrimp, insect exoskeletons,
parasitic nematodes, and intestinal linings[56-58]. Many scientists have stated that chitin can improve plant growth and suppress plant disease.

4. Conclusions
Soil salinity is a significant environmental problem that affects various aspects of plant growth and development. The use of chitosan in agriculture was, however, recently increased by increasing the resistance of crop plants to various stress factors, such as salinity. In general, these results may indicate that chitosan could successfully be used to mitigate the effect of salinity stress on germination and certain growth parameters of common vetch in saline soil seed pretreatment.

References
[1] Yasmine F, Rahman M A, Hasan M M, Amirul Alam M D, Haque M S, Ismail M R, Rafii M Y 2019 Morphophysiological and yield attributes of groundnut varieties under different salinity stress conditions Legum. Res. 42 684–87
[2] Kiani P A, Rasouli F, Rabbi B, Boland Z R, Yong M, Chen Z H, Zhou M, Shabala S 2020 Stomatal traits as a determinant of superior salinity tolerance in wild barley J. Plant Physiol. 245: 1–9
[3] Ahmed I M, Cao F, Zhang M, Chen X, Zhang G, Wu F 2013 Difference in yield and physiological features in response to drought and salinity combined stress during anthesis in Tibetan wild and cultivated barleys Plos One. 8 e77869
[4] Chaves M M, Flexas J, Pinheiro C 2009 Photosynthesis under drought and salt stress: regulation mechanisms from whole plant to cell Ann. Bot. 103 551–60
[5] Ramana G V, Padhy S P, Chaitanya K V 2012 Differential responses of four soybean (Glycine max L.) cultivars to salinity stress Legum. Res. 35 185–93
[6] Shabala S, Munns R 2012 Salinity stress: physiological constraints and adaptive mechanisms Plant Stress Physiol 1 59–93
[7] Sen S K, Chouhan D, Das D, Ghosh R, Mandal P 2010 Improvisation of salinity stress response in mung bean through solid matrix priming with normal and nano-sized chitosan Int. J. Biol. Macromol. 145 108–23
[8] Ifuku S 2014 Chitin and chitosan nanofibers: Preparation and chemical modifications Molecules 19 18367–80
[9] Kaur S, Dhillon G S 2014 The versatile biopolymer chitosan: Potential sources, evaluation of extraction methods and applications Crit. Rev. Microbiol. 40 155–75
[10] Malerba M and Cerana R 2016 Chitosan effects on plant systems Int. J. Mol. Sci. 17 1–15
[11] Demir M and Aril I 2003 Effects of different soil salinity levels on germination and seedling growth of safflower Turkish J. Agric. 27 221–7
[12] Ghasemnezhad M, Nezhad M A, Geraldo S 2011 Changes in postharvest quality of loquat (Eriobotrya japonica) fruits influenced by chitosan. Hortic. Environ. Biotechnol 52 40–5
[13] Ghasemnezhad M, Shiri M A, Sanavi M 2010 Effect of chitosan coatings on some quality indices of apricot (Prunus armeniaca L.) during cold storage Casp. J. Environ. Sci. 8 25–33
[14] Khan W M, Prithiviraj B, Smith D L 2002 Effect of foliar application of chitin and chitosan oligosaccharides on photosynthesis of maize and soybean Photosynthetica. 40 621–4
[15] Cakmakci S and Acikgoz E 2010 Components of seed and straw yield in common vetch (Vicia sativa L Plant Breeding. 113 71–4
[16] Firincioglu H K, Erbektas E, Dogruyol L, Mutlu Z, Karakurt E 2009 Phenotypic variation of autumn and spring-sown vetch (Vicia sativa ssp.) populations in central Turkey Spanish J. Agric. Res. 7 596–606
[17] Badawya M E I and Rabea E I 2009 Potential of the biopolymer chitosan with different molecular weights to control postharvest gray mold of tomato fruit Postharvest Biol. Technol. 51 110–17
[18] Turk M A, Hameed K M, Aqel A M And Tawaha A M 2003 Nutritional status of durum wheat grown in soil supplemented with olive mills by products Agrochimica. 47 209–19
[19] Tawaha A M and Turk M A 2002 Lentil (Lens culinaris Medic.) productivity as influenced by rate and method of phosphate placement in a Mediterranean environment Acta Agron. Hungarica. 50 197–201

[20] Turk A M and Tawaha A M 2002 Impact of seeding rate, seeding date, rate and method of phosphorus application in faba (Vicia faba L. Minor) in the absence of moisture stress. Biotechnol. Agron. Soc. Environ. 6 171–8

[21] Turk A M and Tawaha A M 2002 Response of winter wheat to applied N with or without ethrel spray under irrigation planted in semi-arid environments Asian J. Plant Sci. 1 464–6

[22] Al-Tawaha A M, Seguin P, Smith D L And Bonnell B 2007 Effects of irrigation on isoflavone concentrations of soybean grown in southwestern Québec J. Agron. Crop Sci. 193 238–46

[23] Al-Rifaee M K and Al-Tawaha A M 2005 Doubling chickpea yield by shifting from spring to winter sowing using ascochyta blight resistant lines under typical mediterranean climate Biosci. Res. 2 80–5

[24] AL-Jamali A F, Turk M A and Tawaha A M 2002 Effect of Ethephon spraying at three developmental stages of barley planted in arid and semi-arid Mediterranean locations. J. Agron. Crop Sci. 188 254–9

[25] Turk A M and Tawaha A M 2002 Seed germination and seedling growth of two barley cultivars under moisture stress Res. Crop. 3 467–72

[26] Turk M A, Tawaha A M, Nikus O and Rifaee M 2003 Response of six-row barley to seeding rate with or without ethrel spray in the absence of moisture stress Int. J. Agric. Biol. 4 416–8

[27] Othman Y, Al-Karaki G, Al-Tawaha A R, and Al-Horani A 2006 Variation in germination and ion uptake in barley genotypes under salinity conditions World J. Agric. Sci. 2 11–5.

[28] Turk M A, Tawaha A R M and Lee K D 2004 Seed germination and seedling growth of three lentil cultivars under moisture stress Asian J. Plant Sci. 3 394–7

[29] Musallam I W, Al-Karaki G, Ereifej K and Tawaha A M 2004 Chemical composition of faba bean genotypes under rainfed and irrigation conditions Int. J. Agric. Biol. 6 359–62

[30] Turk A M and Tawaha A M 2002 Onion (Allium cepa L.) as influenced by rate and method of phosphorus placement Crop Research. 23 105–7

[31] Turk M A and Tawaha A M 2001 Common vetch (Vicia sativa L.) productivity as influenced by rate and method of phosphate fertilization in a mediterranean environment Agric. Mediterr. 131 108–11

[32] Tawaha A M and Turk M A 2001 Effects of dates and rates of sowing on yield and yield components of Narbon vetch under semi-arid condition Acta Agron. Hungarica. 49 103–5.

[33] Sulpanjani A, Yang M S, Tawaha A R M And Lee K D 2005 Effect of Magnesium application on yield, mineral contents and active components of Chrysanthemum coronarium L. under Hydroponics Conditions Biosci. Res. 2 73–9

[34] Lee K D, Sulpanjani, Tawaha A M and Min Yang S 2005 Effect of Phosphorus application on yield, mineral contents and active components of Chrysanthemum coronarium L. Biosci. Res. 2 118–24

[35] Lee K D, Turk M A and Tawaha A M 2005 Nitrogen fixation in rice based farming system. Biosci. Res. 2 130–8

[36] Nikus O, Nigussie M, and Al Tawaha A M 2005 Agronomic performance of maize varieties under irrigation In Awash Valley, Ethiopia Biosci. Res. 2 26–30

[37] Abera T, Feyisa D, Yusuf H, Nikus O, Al-Tawaha A M 2005 Grain yield of maize as affected by biogas slurry and N-P fertilizer rate at Bako, Western Oromiya, Ethiopia Biosci. Res. 2 31–8

[38] Supanjani, Tawaha A M, Min Yang M S, and Lee K D 2005 Role of calcium in yield and medicinal quality of Chrysanthemum coronarium L. J. Agron. 4 188–92

[39] Supanjani, Tawaha A M, Yang M S, Lee Y D 2005 Calcium effects on yield, mineral uptake an terpene components of hydroponic chrysanthemum coronarium L Res. J. Agric. Biol. Sci. 1 146–51
[40] Nikus O, Abebe G, Takele A, Harrun H, Chanyalew S, Al-Tawaha A M, and Mesfin T 2005 Yield response of tef (Eragrostis tef (Zucc.) Trotter) to NP fertilization in the semi arid zones of the central rift valley in Ethiopia Eur. J. Sci. Res. 4 49–60
[41] Lee K D, Tawaha A R M and Supanjani 2005 Antioxidant status stomatal resistance and mineral composition of hot pepper under salinity and boron stress Biosci. Res. 2 148–54
[42] Tawaha A M, Turk M A, Lee K D, Supanjani, Nikus O, Al-Rifaee M, Sen R 2005 Awnless barley response to Crop Management under Jordanian Environ. Biosci. Res. 2 125–9
[43] Abebe G, Assefa T, Harrun H, Mesfine T, and Al-Tawaha A M 2005 participatory selection of drought tolerant maize varieties using mother and baby methodology: A case study in the semi arid zones of the central rift valley of ethiopia World J. Agric. Sci. 1 22–7
[44] Assefa T, Abebe G, Fininsa C, Tesso B and Al-Tawaha A M 2005 Participatory Bean Breeding with Women and Small Holder Farmers in Eastern Ethiopia World J. Agric. Sci. 1 28–35
[45] Yang M S, Tawaha A M, and Lee Y D 2005 Effects of ammonium concentration on the yield, mineral content and active terpene components of Chrysanthemum coronarium L. in a hydroponic system Res. J. Agric. Biol. Sci. 1 170–5
[46] Tawaha A R M, Turk M A, and Lee K D 2005 Adaptation of chickpea to cultural practices in a Mediterranean type environment. Res. J. Agric. Biol. Sci. 1 152–7.
[47] Mesfine T, Abebe G, and Al-Tawaha A M 2005 Effect of reduced tillage and crop residue ground cover on yield and water use efficiency of sorghum (Sorghum bicolor (L.) Moench) under semi-arid conditions of Ethiopia World J. Agric. Sci. 1 152–60
[48] Abebe G, Sahile G, and Al-Tawaha A M 2005 Evaluation of potential trap crops on Orobanche soil seed bank and tomato yield in the central rift valley of Ethiopia World J. Agric. Sci. 1 148–51
[49] Abebe G, Sahile G, and Al-Tawaha, A.M. 2005. Effect of soil solarization on Orobanche soil seed bank and tomato yield in the central rift valley of Ethiopia World J. Agric. Sci. 1 143–7.
[50] Abebe G, Hattar B and Al-Tawaha A M 2005 Nutrient availability as affected by manure application in cowpea (Vigna unguiculata (L.) Walp on calacarious soils J. Agric. Soc. Sci. 1 1–6
[51] Zheng W J, Tawaha A M and Lee K D 2005 In situ hybridization analysis of mcMT1 gene expression and physiological mechanisms of Cu-Tolerant in (Festuca rubra cv Merlin) Biosci. Res. 1 21–6
[52] Al-Tawaha A M, Turk M A, Lee K D, Zheng W Z, Ababneh M, Abebe G and Musallam I W 2005 Impact of fertilizer and herbicide application on performance of ten barley genotypes grown in Northeastern part of Jordan Int. J. Agric. Biol. 7 162–6
[53] Zaitoun S T, Al Ghzawi A, Shannag H K and Al-Tawaha A M 2006 Comparative study on the pollination of strawberry by bumble bees and honeybees under plastic house conditions in Jordan valley. J. Food, Agric. Environ. 2 237–40
[54] Assaf T A, Hameed K M, Turk M A and Tawaha A M 2006 Effect of soil amendment with olive mill by-products under soil solarization on growth and productivity of faba bean and their symbiosis with mycorrhizal fungi World J. Agric. Sci. 2 21–8
[55] Turk M A, Al-Jamali A F and Tawaha A M 2002 Effect of seeding rate and ethrel spray on the morphology and the yield traits of irrigated faba bean (Vicia faba (L) major) Crop Res 23 305–7
[56] Al-Tawaha A M, Seguin P, Smith D L and Beaulieu C 2006. Foliar application of elicitors alters isoflavone concentrations and other seed characteristics of field-grown soybean. Can. J. Plant Sci. 86 677–84
[57] Seguin P, Bodo R and Al-Tawaha A M 2007 Soybean isoflavones: Factors affecting concentrations in seeds Advances in Medicinal Plant Research ed S N Acharya and J E Thomas (Kerala: Research Signpost) pp 65–81
[58] Al-Tawaha A R M and Al-Ghzawi A L A 2013 Effect of chitosan coating on seed germination and salt tolerance of lentil (Lens culinaris L.) Res. Crop. 14 489–91.
[59] Al-Tawaha A M and Ababneh F 2012 Effects of site and exogenous application of yeast extract on the growth and chemical composition of soybean International Conference on Agricultural, Environment and Biological Sciences pp 52–4.
[60] Turk A M and Tawaha A M 2002 Response of six-row barley to seeding rate and weed control methods under moisture stress Agric. Medit. 132 208–14
[61] Turk A M and Tawaha A M 2002 Irrigated winter barley response to seeding rates and weed control methods under mediterranean environments Bulg. J. Agric. Sci. 8 175–80
[62] Turk A M and Tawaha A M 2002 Effect of sowing rates and weed control methods on winter wheat under Mediterranean environment Pakistan J. Agron. 1 25–7
[63] Turk A M Tawaha A M 2002 Inhibitory effects of aqueous extracts Black mustard (Brassica nigra L.) on germination and growth of wheat Pakistan J. Biol. Sci. 5 278–80
[64] Turk A M and Tawaha A M 2002 Effect of dates of sowing and seed size on yield and yield components of local faba bean under semi-arid condition Legum. Res. 25 301–2
[65] Turk M A and Tawaha A M 2001 Faba bean (Vicia faba L.) response to seeding rate seeding date, rate and method of phosphorus application in Mediterranean type environments. Bulg. J. Agric. Sci. 7 615–21
[66] Turk A M and Tawaha A M 2002 Response of winter wheat to seeding rate with or without Ethrel spray under irrigation Bulg. J. Agric. Sci. 8 37–42
[67] Turk A M and Tawaha A M 2002 Effect of variable sowing ratios and sowing rates of bitter vetch on the herbage yield of barley-bitter vetch mixed cropping Asian J. Plant Sci. 1 467–9
[68] Tawaha A M and Turk M A 2002 Effect of dates and rates of sowing on yield and yield components of Lentil (Lens culinaris Medik.) under semi-arid conditions. Pakistan J. Biol. Sci. 5 531–2
[69] Tawaha AM, Turk MA and Maghaireh G A 2001 Morphological and yield traits of awnless barley as affected by date and rate of sowing under Mediterranean condition Res. Crop. 22 311–3
[70] Al-Rifaee M K, Al-Yassin A, Haddad N N and Al-Tawaha A M 2007 Evaluation of chickpea breeding lines by examining their responses to sowing date at two mediterranean climatic locations Am. J. Sustain. Agric. 1 19–2