UTILIZING BIOTECHNOLOGY TOOLS FOR SILECTING SALT TOLERANT ROOTSTOCK OF PEACH HASSAWI

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ABSTRACT

This study was carried out with an aim of evaluation of performance of Hassawi peach rootstocks and testing their variance towards salinity stress thus to find out a quick effective modern technique for their reculture and re-propagation again in the region. Eight different concentrations of sodium chloride (NaCl) that represent progressive gradual levels of salinity were used to investigate tolerability of the explants towards variant levels of salinity stress. Thus to evaluate the performance of these Hassawi peach rootstocks (peach, plum [ghwj], and almond). Date and results declared the general superiority of bitter almond to the other rootstocks, as it vitally survived and tolerated salinity to 3500 ppm NaCl, followed by peach that tolerated salinity to 2500 ppm, and eventually ghwj that just survived with maximum tolerance of 2000 ppm of NaCl. The study recommends to count on bitter almond as a salinity tolerant rootstock followed by peach for grafting Hassawi peach rootstocks for its regeneration again in Al-Ahsa oasis, and to exclude plum rootstock because of its low salinity tolerance besides its very low harmonization with peach that negatively affect grafting process.

Keywords: abiotic stress, deterioration, extinction, grafting, *In vitro*, tissue culture.

استخدام تطبيقات التقنية الحيوية لانتخاب أصول وراثية مقاومة للملوحة للخوخ الحساوي

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المستخلص

اجريت هذه الدراسة بهدف تقييم أصول الخوخ الحساوية واختبار التباين لمدى استجابتها للملوحة، وإيجاد تقنية حديثة سريعة وفعالة لإعادة نموها. استعملت ثماني تراكيز مختلفة من كلوريد الصوديوم (NaCl) تمثل مستويات متدرجة لتحديد مدى قدرة النبيتات على التحمل لأي مستوى من الملوحة، ومن ثم تقييم إداء هذه الأصول (الخوخ، البرقوق [المعروف محلياً باسم الغوج]، اللوز المر) المتواجدة بالمنطقة الشرقية للمملكة وقدرتها على التعايش تحت هذه الظروف. أوضحت نتائج القراءات تفوق أصل اللوز المر عن باقي الأصول الأخرى حيث ظل بحيويته ومتعايش تحت ظروف عالية من الاجهاد الملحي 3500 جزء في المليون (ppm)، تلاه أصل الخوخ فظل متحملاً حتى (2500 ppm)، وأخيراً يأتي أصل البرقوق (الغوج) فكان أقصى قدرة له على التحمل والتعايش تحت ظروف الاجهاد الملحي عند تركيز 2000 ppm. توصى الدراسة بالاعتماد على اللوز المر كأصل متحمل للملوحة يليه الخوخ البذري يطعم عليه الخوخ الحساوي بهدف إكثاره وإعادة انتشاره بواحة الأحساء مرة أخرى. وعدم الاعتماد على أصل الغوج بسبب ضعف قدرته على التحمل للإجهاد الملحي. بالإضافة إلى أن درجة التوافق بينه وبين الخوخ ضعيفة جداً مما يقلل من نسبة نجاح عملية التطعيم.

الكلمات المفتاحية: الاجهاد غير الحيوي، الانقراض، التدهور، التطعيم، زراعة الانسجة، مختبرياً.

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INTRODUCTION

Al-Ahsa oasis is one of the most important agricultural regions in Saudi Arabia because of its characteristic variant natural water resources represented by many wells and springs (14). But because of the misuse of these resources and the unsafe exhaustion of groundwater, water level has decreased that led to draught of more than 30 springs and wells in it (5). In addition to, the environmental changes that predominantly took place in the region like raise of temperature and humidity has also led to increasing the salinity stress and drought spread of some diseases and insects and impairment of most of farms and distinction of previously famous for plants like peach, plum and almond (25). Salinity is generally one of the most important agricultural problems that obstructs farmer. It is the main determinant factor of plants growth in the deserts (25% of earth) specially if associated with high temperature or low water content, as salt concentration increases in the soil because of few rains and water loos through evaporation and transpiration (22). Despites the low aquatic need of peach as it can be cultivated at desserts depending on rains water (25), it is sensitive towards salinity as it can't be cultured on land salt, or alkaline, or waterlogged (33). After the initial study of the current situation (circumstances), it turns out the distinction of Hassawi peach rootstocks and its in capability of surviving with the environmental changes that took place at Al-Ahsa like salinity stress. This study concluded three approaches; Frist: Enumerate Hassawi peach rootstocks in the region, Second: *In vitro* re-culture of them, and third: Study the effect of salinity stress on them by adding gradual concentrations of NaCl. The irrigation and drainage authority of Al-Ahsa region has classified the area into ten agricultural sectors according to salinity and water content (availability) of each sector. Generally, the total salinity of Al-Ahsa oasis ranges from 1.2 – 9.7 gm/l. besides, that the sources of irrigation water has been restricted to wells water treated sewage and agricultural drainage (4). Al-Omran region represents the 10th sector according the classification as its total salinity ranges from 1000- 3744 ppm i.e 2.3- 6.1 mmoh/cm = 29-68 MPa (13, 4). In addition, that peach can handle soil salinity to 1100 ppm and water salinity to 800 ppm (33). This study is aiming to find out a scientific practical solution to overcome salinity induced Hassawi peach rootstocks extinction by using tissue culture technique for *in vitro* regeneration and propagation of these rootstocks. It is designed to evaluate their salinity tolerance and surviving ability to match the predominant environmental conditions of the region and subsequently to count on the superior one as a rootstock for peach grafting and recultured on Al-Ahsa oasis.

MATERIALS AND METHODS

Explant sterilization and culture: Fresh 20-30 cm shoots were collected from studied trees under study (peach, plum [ghog] and almond) grown at local farm of Al-Omran region Al-Ahsa during spring March. After removing the branches and leaves and getting the side buds and the shoots tips primary sterilization was directly done on the explants according to (30), by applying 30% Clorox for 30 min then 70% ethyl alcohol for 3 min. All sterilized explants were dissolved in an antioxidant solution at 0.1 g/l of both citric and ascorbic acids and transferred into culture cabinets. Side buds and shoot tips were cut at 1-2 ml long and cultured on primary medium, then incubated in growth room at temperature of 25±2 °C and light intensity of 2000 lux for illumination period of 8:16 hr.

Culture medium composition

culture media recommended by **(30)** shown at Table 1 from their ex-study concerning *in vitro* Hassawi peach propagation were used in this study with putting into consideration renewing media every one month to transfer explants on it.

Table 1. Culture medium constitution for Hassawi peach regeneration protocol *in vitro*

growing stages	Composition medium (mg l^{-1})		
	Salts Strength (MS)	Growth regulators	Organic Constituent's
1. Starting	Full MS [1]	0.1 IBA[2] + 2 BAP[3]	100 Myo-Inositol + 100 Gibberellin + 30000 Suc. + 7000 Agar
2. Multiplication	Full MS	0.1 IBA + 2 BAP	
3. Rooting	Half MS	0.5 IBA	

1- MS = (19) & 2- IBA = Indole-3 -Butyric Acid & 3- BAP = 6-Benzylaminopurine

Salinity stress impact
The study was designed that the three rootstocks (peach, plum, and almond) are subjected to variant levels of salinity stress by adding gradual variant concentrations of NaCl to the nutrient medium shown at Table 2. Proline amino acid was also added to the nutrient media in a concentration of 0.1 gm/l to improve the efficacy and the salinity tolerance of the explants.

Table 2. Gradient concentrations of NaCl to induce salinity stress on peach rootstocks.

Salt stress	Code	Concentrations of Sodium Chloride (NaCl)							
	Concentration	N 1	N 2	N 3	N 4	N 5	N 6	N 7	N 8
	ppm*	0.0	1000	1500	2000	2500	3000	3500	4000
	g/l	0.0	1.00	1.50	2.00	2.50	3.00	3.50	4.00

***ppm = 0.03 Mpa = 1 dsm = 1 mmhos/cm = 640 ppm.**

Data showing salinity tolerance parameters of *in vitro* cultured explants were recoded as each treatment was represented by five replications and each replication has three tubes. Parameters showing salinity tolerance were as the following:

1. No. of survived plantlets.
2. No. of shoots
3. Shoot length (cm)
4. No. of leaves (fresh leaflets)

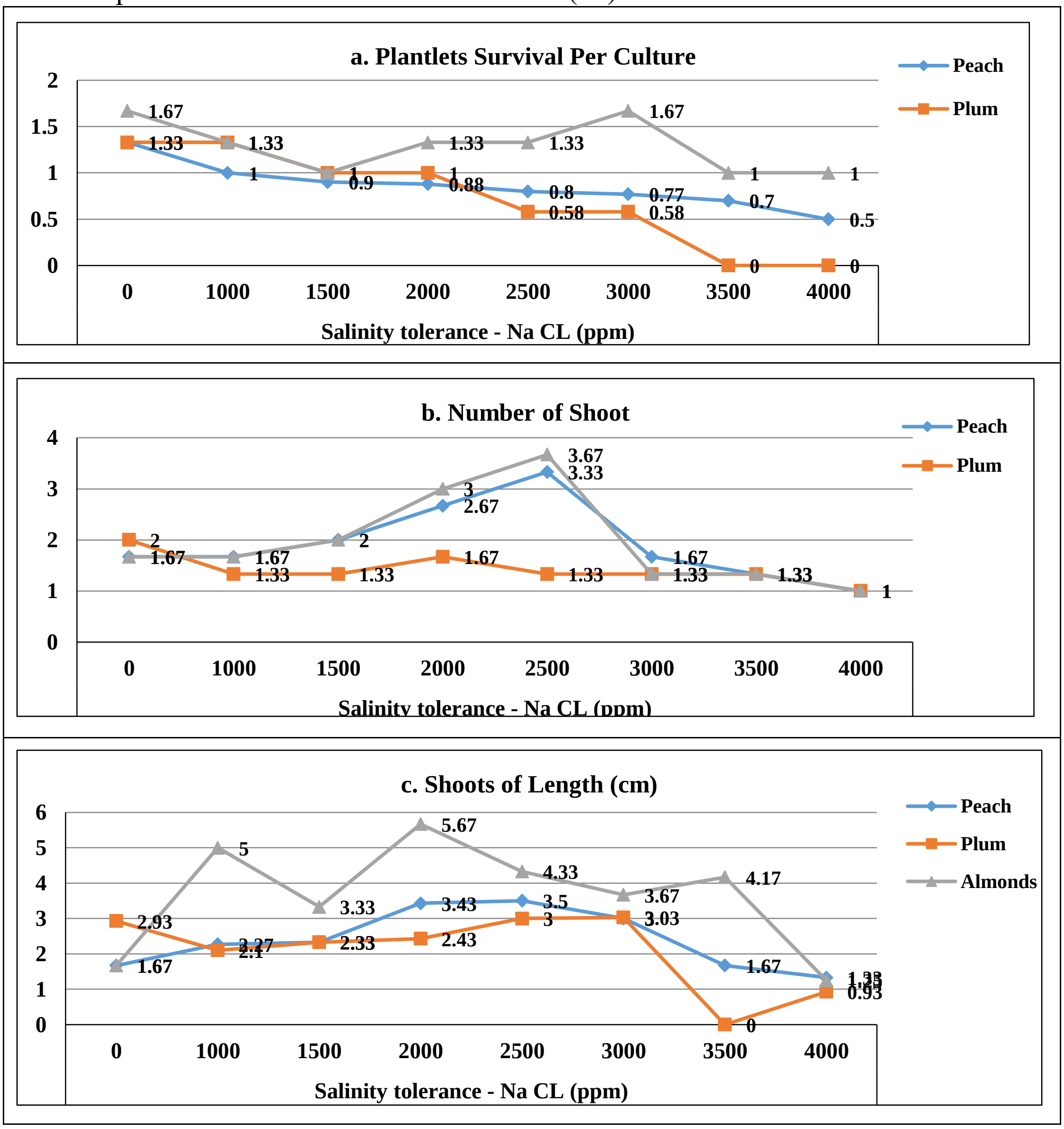
Rooting stag: All the regenerated plantlets of the three peach rootstocks (peach, plum, and almond) were transferred to be cultured on rooting medium that is half strengthened MS medium in addition to adding IBA in a concentration of 1.00 mg/l and other components illustrated in Table 1.

Acclimatization stage
All the three rootstocks plantlets that formed roots were subjected to unified acclimatization process. As the well rooted, plantlets were removed from the culture vessels. Then thoroughly washed by water to get rid of the residual traces of the nutrient medium, then immersed in fungicidal solution (Bnlat) with a concentration of 3 g/l. Then rewashed to eliminate the fungicidal traces, then planted in 8 cm pots filled with artificial soil consisting of equal volumes of peat moss, sand, and vermiculite 1:1:1 **(**30**)**. The potted plantlets were covered with bags to maintain humidity around the plantlets inside the green house for one month. After one week the bags were gradually opened to expose, the plantlets to ambient conditions till the bags were completely removed after one month. The acclimatized plantlets were recultured on bigger pots to start fertilization and gradual irrigation for three months before being planted in the field.

Statistical analysis: The experiment was designed according to randomized complete design according to rootstocks, treatments and replications. Data and measurements recorded by using Excel for treatments and replications then data were statically analyzed based on analysis of variance using significance difference of 5% using (M stat-c) statistical analysis program according to (11).

RESULTS AND DISCUSSION

Figure 1 & 2 show the interactive effect between variant peach rootstocks and the concentrations of NaCl used to induce salinity stress on the *in vitro* cultured explants. The results showed significant differences between rootstocks and NaCl concentrations. The results also showed the superiority of bitter almond rootstock to the other rootstocks concerning salinity tolerance all over studied parameters. Also, as shown at figure (1-a) bitter almond achieved the highest means of survived plantlets (1.67) at 3000 ppm followed by peach and plum by (1.00) but at 2000 & 1500 ppm, respectively. Data also declared that high salinity con concentration (3500-4000 ppm) negatively affected plum plantlets leading to its damage which indicates the lower salinity tolerance of plum rootstock than almond and peach as being the most negatively affected rootstock by high salinity stress. This may by attributed to its cultivar according to (23). Concerning leaves Figure (1-b), bitter almond achieved the best results followed by peach and plum as it achieved the highest mean of No. of shoots (3.67) followed by peach by (3.33) but at NaCl concentration of 2500 ppm, and finally plum with the lowest mean of (1.67) but with a lower NaCl concentration of 2000 ppm. While, the lowest mean of No. of shoots (1.00) was with all cultivars at high concentration of NaCl 4000 ppm that indicates the severe decrease in growth and multiplication average under high concentration of NaCl (10, 17), as growth decreases by increasing NaCl concentration in leaves (3). In a like manner, concerning shoot lengths Figure (1-c) bitter almond proved its superiority to the other rootstocks by achieving the highest mean of length (5.70 cm) at 2000 ppm, then plum and peach by 3.43 & 2.43 cm, respectively at the same concentration. Furthermore, it was obviously more salinity tolerant till 3500 ppm as it attained higher lengths with a mean of 4.17 cm. While plum and peach couldn't tolerate or survive at that level of salinity and suffered from dwarfism by length mean of (1.67 cm) at the same salinity level. Likewise, data from Figure (1-d) showed that bitter almond was the best concerning No. of fresh leaflets under the same treatments with a mean of 32.67 at 2500 ppm, followed by peach then plum with means of 30.33 and 21.33, under the same level of salinity stress. Generally, data showed significant interactive effect between the gradual concentrations of salinity on all studied rootstocks. Ghog was the least salinity stress tolerant levels, which led to leaves burning as a clear indication of its low salt tolerability (29). The variance of salt tolerance degrees by different plants is attributed to the plant type and cultivar (23). As well, the good shoot growth and resistant brown spots and burns on leaves of almond under high salinity stress are indications of its high salinity tolerance (29). The low growth and development rates are due to NaCl effect on water content of plantlets and to the resultant high osmotic pressure, which negatively affect the No. and size of plantlets and lowers cells fission and elongation, rates (16).



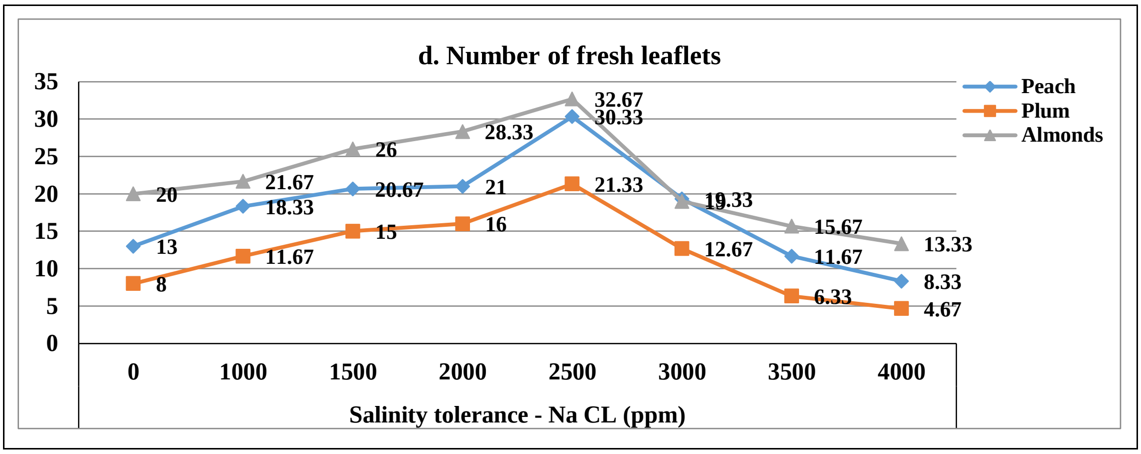


Figure 1. the interactive effect between NaCl concentrations that induce salinity stress on Hassawi peach rootstocks under study; a- No. of survived plantlets, b- No. of shoots, c- Shoot length (cm), and d- No. of fresh leaflets.

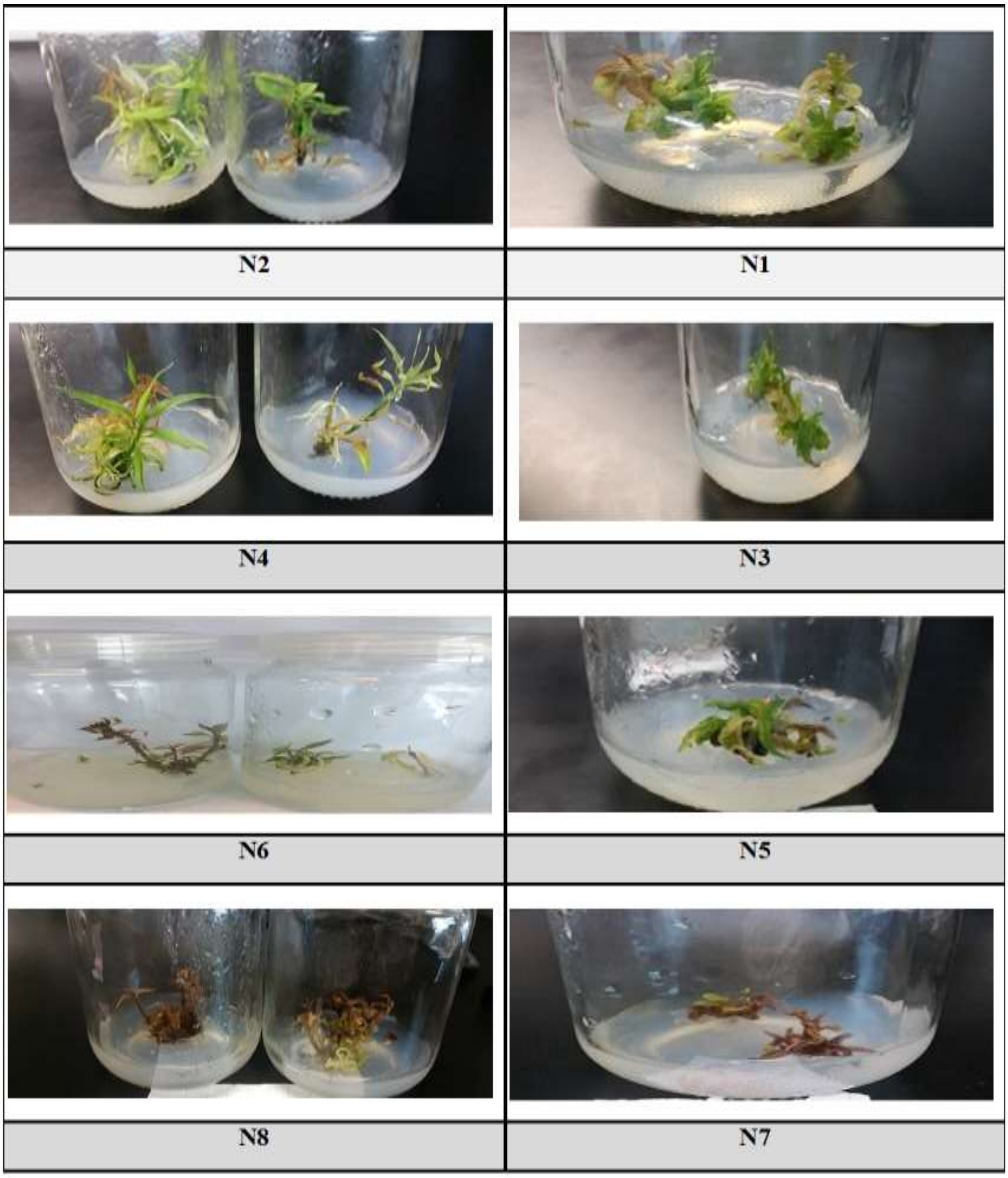


Figure 2. The gradual effect of NaCl to induce salinity stress on *in vitro* Hassawi peach rootstocks.

On another side, it has been noticed that rooting started after three weeks from culture on rooting medium that is confirmed by (26), for all the three peach rootstocks as shown on Table 3, recording 85.77 % rooting percent. Butter almond achieved the highest mean of roots No. (6.70), and highest mean of root length (7.30 cm). As well, the maximum achieved plantlets elongation mean (7.80 cm) followed by peach with the following means; No. of roots (5.40), root length (6.80 cm), and plantlets elongation (7.60 cm). Eventually, plum with the following means; No. of roots (4.10), root length (5.30 cm), and plantlets elongation (5.90 cm). It has been noticed that the half strengthened MS medium (2.2 gm/l) enhanced rooting of deciduous plants as peach, almond and plum by stimulating the plants to look for nutrients through rooting that agreed with the ex-studies (21, 2, 1, 32, 30). The former studies also confirmed that ½ MS medium achieved best results of rooting concerning number, lengths and plantlets elongation during *in vitro* peach rooting stage (12, 30). An artificial soil has been used for plantlets acclimatization that is consisted of peat moss, vermiculite, sand with equal ratios, growth was normal, and there were no plant abnormalities or deformities in plants cultured on green house for 60 days. This mixture proved to be efficient for acclimatization process of many deciduous plants like peach (30), almond (34, 27) and plum (15).

Table 3. Number, lengths and elongation of *in vitro* peach rootstocks cultured plantlets during rooting stage the success percentage of acclimatization stage in green house

Parameters / Rootstocks	No. roots (means)	Root lengths- cm (means)	Elongation of plantlets - cm (means)	Success percentage of acclimatization (%)
Almonds	**6.70**	**7.30**	**7.80**	**91.20**
Peach	**5.40**	**6.80**	**7.60**	**88.70**
Plum (Ghog)	**4.10**	**5.30**	**5.90**	**77.40**

From all of the above, all the results of this study agree with the previous studies concerning that almond rootstock was more salinity tolerant than plum and peach that are known of their sensitivity towards salinity that doesn't exceed 1100 ppm (33). Salinity stress is considered one of the important obstacles that generally hinders agriculture because of its high impact on most of components inside plant cells like reduction of nuclear acids, carbohydrates and proteins (16, 17). Also, severe reduction of growth and multiplication ratio (10), besides impairing absorption of some nutritive elements from the medium as phosphorous, magnesium, ferrous, manganese, capper, and K^+/Na^+ ratio. It also, increases other components (elements) like proline, potassium, sodium, calcium and zinc a compound resulted from metabolism of fatty acids inside moldy rotting tissues (28). In addition, to enhancing activation of peroxidase enzyme that contains cupper which results in oxidation and browning of plant tissues (31). Thence, proline as an amino acid was added to the nutrient medium as recommended by (30) for prophylaxis to increase the efficacy and durability of plantlets towards salinity stress (17). It was added with the optimum concentration 0.1gm/l as recommended by the previous (23, 17). As proline is an antioxidant and it helps adjusting the osmotic pressure and pH of cells cytoplasm (8), thus it can protect the cellular protein and enzyme from being metabolized and subsequently helps cell survive (16, 24). Generally, proline reduce the harmful effects of ammonium and other poisonous components resulted from salinity stress. As well, it retains carbon and nitrogen necessary for growth, in addition to elevation of NaCl levels inside plants, which promotes water absorption that reduces the negative effects of salinity stress (8). Proline is also, considered as one of the most defensive proteins that enhance the tolerability of plant towards salinity like amides and fluoridone (6, 20). It is also, characterized by its adaptive characters to hard conditions and by being a good growth activator that result in delaying aging of plants (10). It is generally, known that MS medium is characterized by its calcium and potassium content that reduces the NaCl negative effects on the explant because of potassium accumulation inside its cells, that enhances growth and salinity tolerance besides achieving balance between Na, K & Ca that helps adjusting pH of the nutrient medium (7). Besides that presence of myoinositol in association with proline in the nutrient medium increases, salinity tolerance as proline moves to leaves and roots which sustains growth rate under stress (24). Tissue culture technique can be used to overcome the bothering problems and it could be used to evaluate the salinity tolerance variance between the variant peach rootstocks and to reculture it after getting rid of the problem (20). Evaluation and selection of rootstocks is carried out by gradual salinity induction to mimic nature as salinity stress increases gradually till reaching the harmful levels at the end. It is necessary for plants to have morphological or physiological mechanisms to survive under salinity stress hard conditions (3, 6). Great attention should be paid to overcome Hassawi peach rootstock extinction due to many different reasons including rare rains or low water resources or invalid resources because their high content of salt due to their combination with agricultural drainages as (5). They recommended to pay a greet attention for continuous washing of salts in case of irrigation with this type of water in Al-Ahsa specially for salinity sensitive plants or to cultivate a substituent salinity tolerant ones. Researchers exert continuous efforts to overcome salinity problem to get the best use of saline water and soil for agriculture of economically important salinity tolerant plants. Variant methods are used to increase salinity tolerance that can be classified into:

1) physiological methods: by using botanical growth regulators like abscisic acid (ABA) that close stomata and reduce stomatic delivery to the shoots, or by adding indol acetic acid (IAA) that on contrarily open stomata and reduce water loss of plants (18), or by adding kinetin or espermen that elevate salinity resistance of plants (6).

2) Biological methods: by adding salinity resistive bacteria to the soil like Halo tolerance bacteria that lives in seas and saline soils (9).

3) Biotechnological methods: by using tissue culture technique or genetic engineering to produce new salinity tolerant cloning or by transplanting salinity resistant genes from a resistant plant to a sensitive one.

The study recommends to count on bitter almond as a rootstock on which Hassawi peach can be grafted for its recultured and respread on Al-Ahsa region followed by peach rootstock. The study also excluded plum as a rootstock because of its inability to survive under high levels of salinity besides its very weak adhesion to peach which extremtly limits success of the process. Also recommended to support biotechnological researches like genetic engineering, genes transplantation and micro grafting concerning conserving genetic rootstocks of Kingdom Saudi Arabia (KSA) in general and Al-Ahsa in particular.

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REFERENCES

1. Ahmad, T.A., Ch. H. Ur-Rahman, M.S. Ahmed and M.H. Laghari. 2003. Effect of culture media and growth regulators on micro-propagation of peach rootstock GF 677. Pak. J. Bot., 35(3): 331- 338

2. Ainsely, P.J., G.G. Collins and M. Sedgley. 2001. *In vitro* rooting of almond (*Prunus dulcis* Mill.). *In Vitro* Cell. Dev. Biol. Plant, 37(6): 778-785

3. Alarcón, J.J., M.J., Sánchez-Blanco, M.C., Bolarin and A, Torrecillas. 1994. Growth and osmotic adjustment of two tomato cultivars during and after saline stress. Plant Soil. 166: 75-82

4. Al-Dakheel, Y.Y., 2011. Assessing NDVI spatial pattern as related to irrigation and soil salinity management in Al-Hasa oasis, Saudi Arabia. J. Indian Soc. Remote Sens, 39: 171-180, [DOI: 10.1007/s12524-010-0057-z].

5. Al-Gossaibi, A.M and A.M., Almadini. 2000. The assessment of irrigation water quality and its agricultural uses at Al-Hasa oasis, KSA. Scientific Journal of King Faisal Univ., 1 (1): 87-102

6. Al-Ouda, A., R., Saleh and R.A., Ali. 2006. Evaluation the response of some local barley (*Hordeum vulgare L.*) varieties to PEG

induced osmotic stress at early growth stage. Damascus University Journal of Agricultural Sciences. 22 (1): 15- 33

7. Al-Shorafa, W., A., Mahadeen and K., Al-Absi. 2014. Evaluation for salt stress tolerance in two strawberry cultivars. American J. Agricult. and Biolog. Sci., 9 (3): 334-341

8. Berkowitz, G.A. 1998. Water and salt stress. A. S. Raghavendra, ed. Photosynthesis A comprehensive treatise. Cambridge University Press, New York., 226-237

9. Dieter, H. and S., Helmut. 2007. Osmose sing and osmosignaling, Academic Press. 579, ISBN: 0-12-373921-7

10. Dixe, P.J., V., Myclysugut and A., PlunKett. 1986. Salt stress: resistance mechanisms and *in vivo* selection procedures in plant tissue culture and its agricultural applications (Witners, L.A and Alderson, P.O., Ed). Butterworths, London. 469-478

11. Duncan, B.D. 1955. Multiple ranges and multiple F test. Biometrics. 11: 1-42

12. Fotopoulos, S. and E.T. Sotiropoulos. 2005. *In vitro* rooting of PR 204/84 rootstock (*Prunus persica* x P. *amygdalus*) as influenced by mineral concentration of the culture medium and exposure to darkness for a period. Agron. Res., 3: 3-8

13. HARC, Hofuf Agricultural Research Centre. 1979. Water Resources of the Al-Hasa Oasis. A Report on the Work of the Leichtweiss-Institute Research Team, Technical University Braunschweig Prepared for Hofuf Agricultural Research Centre, KSA, Publication No. 38

14. Hussain, Z. 1982. Problems of irrigation agriculture in Al-Hasa, Saudi Arabia. Agricultural Water Management. 5: 359-374

15. Jain, N. and S.B. Babbar. 2000. Recurrent production of plants of black plum, *Syzygium cuminii* (L.) Skeels, a myrtaceous fruit tree, from *in vitro* cultured seedling explants. Plant Cell Rep., 19(5): 519-524

16. Levitt, J. 1980. Response of plants to environmental stresses. 1, chilling, freezing and high temperature stresses. Academic Press, New York: 275-282

17. Mohamed, K.H. 2007. Effect of proline treatment on salinity tolerance of *Ziziphus spp* seedling *cv*. TUFFAHI. Basra Journal of Science (B). 25 (2): 102-89

18. Mulholland, B.J., I.B., Taylor, A.C., Jackson and A.J., Thompson. 2003. Can ABA mediate responses of salinity stressed tomato? Environmental and Experimental Botany. 50: 17-28, ISSN: 0098 8472

19. Murashige, T. and F., Skoog. 1962. A revised medium for rapid growth and bioassay with tobacco tissue cultures. Physiologic plant arum. 15: 473-497

20. Obaid, H. and S., Haddad. 2011. Evaluation responses of seeds germination in some varieties of cucumber (*Cucumis sativa*) to drought stress. Damascus University Journal of Agricultural Sciences. 27 (1): 97-114

21. Perez-Tornero, O., J.M. Lopez, J. Egea and L. Burgos. 2000. Effect of basal media and growth regulators on the *in vitro* propagation of apricot (*Prunus armenical* L,) *cv*. Canino. J. Hortic. Sci. Biotech., 75(3): 283- 286

22. Rains, D.W. 1972. Salt transport by plants in relation to salinity. Annu. Rev. Plant Physiol. 23: 367-388

23. Ramoliya, P.J. and A.N., Pandey. 2003. Soil salinity and water status affect growth of *Phoenix dactylifera* seedlings. N. Z. J. Crop Hortic. Sci. 4: 345-353

24. Reza, S., R., Heidari, S., Zare and A., Norastehnia. 2006. Antioxidant response of two salt-stressed Barley varieties in the presence or absence of exogenous proline. Gen. Appl. Plant Physiology, 32: 233-251

25. Rogers, P. and P., Lydon. 1994. Water in the Arab World: perspectives and prognoses. The division of applied sciences: Harvard University Cambridge Mass. 267-316.

26. Ruzic, D., P. Rosati and G. Marino. 1984. The effect of growth regulators in the micro-propagation of peach x almond hybrid GF (677). Riv. Della Ortof. Frutic. Ital., 68: 413-422

27. Saeed, W.T., 1998. *In vitro* propagation of two almonds (*Prunus dulcis* Mill) *cv*. Bull. Fac. Agric. Cairo Univ., 49: 563-574

28. Saleh, B. 2011. Effect of salt stress (NaCl) on biomass and K^+/Na^+ ratio in cotton. J. Stress Physiology & Biochemistry, 7 (4): 5-14, ISSN: 1997-0838

29. Shannon, M.C. and L.E., Francois. 1978. Salt tolerance of three muskmelon cultivars. J. Am. Soc. Hort. Sci., 103: 127-130

30. Shehata, W.F. and J.M., Al-Khayri. 2013. Conservation of endangered hassawi peach

(*Prunus persica* L.) through micropropagation. Journal of Biological Sciences. 13 (2): 75-81. ISSN: 1727-3048. DOI: 10.3923/ jbs

31. Shehata. W. F. 2014. The effect of explants types' difference of some Hassawi date palm cultivars on *in vitro* physiological problems and obstacles occurrence. Scientific Journal of King Faisal Univ., 15 (1): 25-43

32. Touqeer, A., H.Ur., Rahman, C.M.S. Ahmed and M.H. Laghari. 2003. Effect of culture media and growth regulators on micropropagation of peach rootstock GF 677. Pakistan Journal of Botany. 35(3): 331- 338

33. Wang, Y. 1985. Peach growing and germplasm in China. Acta Hortic. 173: 51-55

34. Zaied, N.S., 1997. Studies on the Vegetative Propagation of Stone Fruit Trees. Ph.D. Thesis, Department of Horticulture Faculty of Agriculture, Moshtohor, Zagazig University, Egypt.