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In Vitro Establishment and Clonal Propagation of Sebri Pear Cultivar

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ABSTRACT

Two experiments were performed for *in vitro* establishment, proliferation and shoot growth in axillary bud explants of Sebri pear cultivar. In the first experiment, the effect of different concentrations of IBA(Indole-3-butyric acid) on explants' establishment was evaluated. In a second experiment, the influence of several combinations of PGRs (Plant growth regulators)on shoot proliferation and shoot growth was investigated. IBA at 0 and 0.1 mg L^{-1} concentration led to successful bud establishment. At higher IBA concentrations, callus was induced, but fewer explants were successfully initiated. BAP increased shoot proliferation, while TDZ(Phenyl-N'-(1, 2, 3-thiadiazol-5-yl) urea) did not show any effect on shoot proliferation. BAP, at 3 mg L^{-1} was more effective than 2 mg L^{-1} on the number of proliferated shoots. Maximum shoot length was obtained for the medium containing BAP(6-benzylaminopurine) (2.0 and 3.0 mg L^{-1})+IBA (0.1 mg L^{-1}) and BAP (2.0 and 3.0 mg L^{-1})+IBA (0.1 mg L^{-1})+GA_3 (0.5 mg L^{-1}). IBA in BAP combinations induced lateral bud swelling, while GA_3 (Gibberelic acid) inhibited it. Maximum leaf number was obtained for MS medium with 2.0 mg L^{-1} TDZ and 0.5 mg L^{-1} GA_3. Moreover, medium supplemented with 2.0 mg L^{-1} BAP+IBA (0.1 mg L^{-1})+GA_3 (0.5 mg L^{-1}) and 2.0 mg L^{-1} TDZ+GA_3 (0.5 mg L^{-1}) produced maximum shoot length. Vegetative growth habit varied with different combinations and BAP concentrations, being the highest in BAP (2.0 mg L^{-1})+IBA (0.1 mg L^{-1}) treatment. The combination of BAP at 2 mg L^{-1} and 0.5 mg L^{-1} of GA_3 is finally recommended for a proliferation of Sebri pear cultivar.

Keywords: Establishment, *In vitro, Pyrus communis*, *Pyrus pyrifolia*, Shoot proliferation.

INTRODUCTION

Sebri (Rosaceae) is among some of the most important Iranian local pear cultivars widely grown throughout the country. Some authors believe Sebri cultivar to belong to *P. communis* L. (Safarpour, 2008; Erfani *et al.,* 2012) but there are reports statingthat stated this cultivar belongs to *Pyrus Pyrifolia* (Burm.) Nak. (Syn. *Pyrus setorina* Rehd.) (Amiri, 2002; Rahemi and Baghbani, 2002; Davarynejad and Davarynejad, 2007; Zafari nia *et al.,* 2010). This cultivar is of some unique characteristics. Fruits are enriched with an excellent eating quality. They are very juicy, sweet, solid firm with a high stone cell content (Amiri, 2002). Propagation of Sebri cultivar through common vegetative methods is cumbersome. The successful chance of either grafting or budding on rootstock is low because of the incompatibility with many rootstocks. Also, propagation by means of either cuttings or through layering is difficult because of the problematic rooting (Amiri, 2002; Davarynejad and Davarynejad, 2007; Davarynejad *el al.,* 2008; Vatandoost *et al.,* 2011). The graft incompatibility problems may

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be avoided by production of self-rooted trees. Potentials of pear *in vitro* culture have been previously studied (Shibli *et al.,* 1997; Rouzban et al., 2002; Kadota and Niimi, 2003).

Bud or shoot tip cultures have commonly been employed in micropropagation of woody plants (McCown and McCown, 1987). In pear, it was reported *in vitro* shoot proliferation can be affected by type and concentration of Cytokinin in the medium (Kadota and Niimi, 2003). Supplementation of growth regulators to the medium could compensate for the probable deficiencies of their endogenous contents that originate from donor plants (Grattapaglia and Machado, 1998). Two major groups of Cytokinins are synthetically used in tissue culture including such phenylurea derivatives, as 1-phenyle-3-(-1, 2, 3-Thidiazol-5-yl) urea (Thidiazuron, TDZ), and adenine derivatives, which may occur naturally, e.g. 6-benzylaminopurine (BAP) (Nandi *et al.,* 1989). TDZ and BAP induced shoot proliferation on pear (Kadota and Niimi, 2003). Many studies on pear micropropagation have focused on medium type (Abdollahi et al., 2006; Bell et al., 2009) and Cytokinins (Kadota and Niimi, 2003). Therefore, the present study was undertaken to develop an *in vitro* propagation protocol for Sebri pear cultivar grown in Iran as for future use, use in research programs, and as well for an evaluation of the effect of different PGR combinations on *in vitro* establishment of Sebri pear cultivar.

MATERIALS AND METHODS

Plant Material and Media Preparation

Shoots of Sebri cultivar grown in *in vivo* conditions were used as a source of plant material. Current season shoots were taken during late winter from 20 year old trees of pear cultivars grown in the Collection Garden of Agriculture and Natural Resources Research Center of Khorasan Razavi, Mashhad, Iran (36º 13' N and 59º 38' E, 3291 ft Elevation). Two experiments were conducted, in each, explants being cultured in glass vials (100 ml) containing 25 ml of medium with the vials being capped with aluminum foil caps. Agar (8 g L^{-1}) was added to the medium after pH being adjusted to 5.7. The medium was autoclaved at 0.12 MPa pressure for 20 minutes and at 121°C. The glassware and utensils were sterilized for 45 minutes while TDZ and Gibberelic acid (GA_3) being sterilized through filter sterilization. The cultures were grown at 24±1°C in a photoperiodicity of 16-h and at light intensity of 40 µmol $m^{-2}s^{-1}$ supplied through white fluorescent tubes.

The First Experiment

Dormant buds of current season shoots collected in late winter were made use of as primary explants. The explants [vegetative bud, medium in size, (4×2.5 mm)] were placed under running tap water for 30 minutes. Surface sterilization of the samples was conducted using 70% ethanol for 30 seconds followed by being washed for 10 minutes with 1% sodium hypochlorite (NaOCl) supplemented with Tween-20 (0.2%). The explants were then rinsed with sterile distilled water three times for 15 minutes. Following a removal of all the bud scales, the buds were cultured on MS solid medium (Murashige and Skoog, 1962) supplemented with 30 g L^{-1} sucrose and four concentrations of IBA (0, 0.1, 0.3 and 0.5 mg L^{-1}). Percentage of establishment (explants producing 3-4 leaves), callus production and browned explants were recorded after 1 month past of cultivation.

The Second Experiment

In vitro shoots initiated from Sebri pear cultivar dormant bud cultures that had been maintained on MS medium containing 0.1 mg L^{-1} IBA for 4 weeks were made use of for shoot proliferation. Shoots were transferred to MS-based media supplemented with different combinations of BAP or TDZ, IBA and GA_3 (Table 1). Thirty g L^{-1} sucrose and 8 g L^{-1} agar

were added to the media. The number of Proliferated Shoots (PSH), Proliferated Shoots Lengths (PSHL), Proliferated Shoot Extension (PSHE; PSH×PSHL), the number of Activated lateral Buds (AB), the number of Developed Leaf production (DL), Main Stem Elongation (MSE), and Vegetative

Growth (VG) were recorded after 4 weeks past of cultivation.

Statistical Analysis

The experiments were performed in 5 replications each replicate comprised of 5 vials, each vial containing two explants. Completely randomized designs were chosen for the experiments. Data were analyzed through SAS PROC GLM (SAS Institute Inc., 1989). Mean values were compared according to Duncan's Multiple Range Test (DMRT) at 1% probability. Percentage data were transformed into *arcsin* before being analyzed.

RESULTS AND DISCUSSION

The First Experiment

The percentage of establishment and callus production were significantly affected by IBA concentration ($P \leq 0.01$). With no IBA and with a lower IBA concentration (0.1 mg L^{-1}) samples led to more successful establishments of culture (73.57 and 82.2, respectively) while high IBA concentrations (0.3 and 0.5 mg L^{-1}) leading to the most callus induction (32.6 and 37.6, respectively). The callus in these concentrations was moist, of white-transparent mass and apical meristem becoming inactive after 1-2 leaf growth and tip of buds gradually becoming brown (after 2-3 months). These explants remained within the same status for a long period of time (4-6 months). The percentage of browned explants was not affected by IBA concentration (Table 2).

Increasing IBA concentration caused decrease in bud establishment percentage (Figure 1). Maximum bud establishment was obtained at 0 and 0.1 mg L^{-1} whilst the lowest being resulted at 0.3 and 0.5 mg L^{-1}. Callus production was higher when IBA at 0.5 mg L^{-1} was used. IBA did not cause browning of the culture. IBA at either 0 or 0.1 mg L^{-1} produced higher calluses. At these concentrations a high percentage of bud explants was successfully established and the percentages of explants that produced calluses were at their lowest

Table 1. Plant growth regulators combination used for shoot growth and proliferation of Sebri pear cultivar.

Combinations	PGRs concentrations (mg L^{-1}) [a]			
	BAP [b]	TDZ [c]	IBA [d]	GA_3 [e]
2BAP	2	-	-	-
2BAP+IBA	2	-	0.1	-
2BAP+GA_3	2	-	-	0.5
2BAP+IBA+GA_3	2	-	0.1	0.5
3BAP	3	-	-	-
3BAP+IBA	3	-	0.1	-
3BAP+GA_3	3	-	-	0.5
3BAP+IBA+GA_3	3	-	0.1	0.5
1TDZ	-	1	-	-
1TDZ+IBA	-	1	0.1	-
1TDZ+GA_3	-	1	-	0.5
1TDZ+IBA+GA_3	-	1	0.1	0.5
2TDZ	-	2	-	-

[a] Plant growth regulator; [b] 6-benzylaminopurine, [c] Phenyl-N'-(1, 2, 3-thiadiazol-5-yl) urea, [d] Indole-3-butyric acid, [e] Gibberelic acid.

Table 2. ANOVA [a] of different IBA concentration effects on the percentages of establishment, callus production and browned explants in Sebri cultivar of pear.

		Mean Square		
Source	*df* [b]	Establishment	Callus production	Browned explants
IBA concentration [c]	3	603.922 **	695.616 **	16.928 ns
Error	16	36.067	72.612	50.784
Corrected Total	19			
2TDZ [d] +IBA	-	2	0.1	-
2TDZ+ GA_3 [e]	-	2	-	0.5
2TDZ+IBA+GA_3	-	2	0.1	0.5

** Significant at 0.01 and ns: Not significant, [a] Analysis of variance, [b] degrees of freedom, [c] Indole-3-butyric acid, [d] Phenyl-N'-(1, 2, 3-thiadiazol-5-yl) urea, [e] Gibberelic acid.

levels.

The Second Experiment

Propagation and shoot growth of Sebri pear cultivar was significantly affected by plant growth regulator combinations ($P \leq 0.01$) (Table 3).

Proliferated Shoots, Shoots Length, Shoot Extension and Activated Lateral Buds

BAP induced shoot proliferation, while TDZ did not affect this phenomenon. On the other hand, TDZ had no stimulatory effect on proliferation. This is in agreement with the previous findings by Kadota and Niimi (2003) on pear, using BAP. The highest number of proliferated shoots (2.6-3.6) was obtained at 3 mg L^{-1} BAP concentration. Shibli *et al.* (1997) and Improved pear proliferation was reported with combination of cytokinin and auxin (Baviera *et al*., 1989; Nosrati *et al*., 2009).

Higher BAP concentrations led to lower proliferated shoot lengths. This inverse relation has been obtained in other studies (Rouzban *et al.,* 2002; Shibli *et al.,* 1997; Dolcet-Sanjuan *et al.,* 1990). Maximum shoot length was observed to be either produced in medium supplemented with BAP and IBA or on medium supplemented with BAP, IBA and GA_3. It has been reported that a combination of BAP (0.5 mg L^{-1}) and IBA (0.09 mg L^{-1}) is suitable for good shoot length in *Pyrus calleryana*, cv. Bradford (Stimart and Harbage, 1989). Also, the combination of

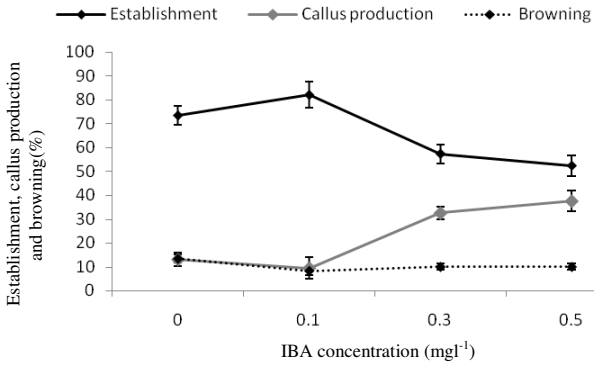

Figure 1. Effect of different IBA concentrations on bud establishment, callus production and browning of Sebri pear cultivar after 1 month past of growth periods.

Table 3. ANOVA of the effect of different combinations of plant growth regulators on shoot growth and proliferation characteristics.

		Mean Square						
Source	*df*	PSH [a]	PSHL [b]	PSHE [c]	AB [d]	DL [e]	MSE [f]	VG [g]
Treatments	15	10.02**	61.49**	399.98**	22.88**	38.33**	278.35**	808.75**
Error	64	0.49	9.07	45.49	1.11	1.16	24.34	67.16
Corrected Total	79							

**: Significant at 0.01.

[a] The number of Proliferated SHoots; [b] Proliferated SHoots Length; [c] Proliferated SHoot Extension; [d] The number of Activated lateral Buds; [e] The number of Developed Leaf production; [f] Main Stem Elongation, [g] Vegetative Growth.

BAP + NAA + GA_3 was recommended for 'Seckel' pear shoot elongation (Singha, 1980). The different stages of shoot formation in medium containing 3BAP+IBA have been illustrated in Figure 2.

In all combinations, high BAP concentration led to a higher propagation rate except in BAP+GA_3 combination where propagation remained almost intact. Proliferated shoot extension was the highest (28.79 mm) in BAP at 3 mg L^{-1}+IBA and the lowest (3.40 mm) in the case of BAP at 2 mg L^{-1}.

The presence of IBA (0.1 mg L^{-1}) and BAP in the medium activated and inflated the lateral buds on main stem, while GA_3 inhibited it. Inhibiting effect of GA_3 was seen in BAP+IBA+GA_3, too (Figure 3). Bud activation and swelling was not observed with a supplementation of TDZ (Table 4).

Developed Leaves, Main Stem Elongation and Vegetative Growth

The presence of TDZ in medium increased leaf number. A maximum number of leaves (9.0) was produced in medium supplemented with TDZ (2 mg L^{-1})+GA_3 (0.5 mg L^{-1}). Medium supplemented with

Table 4. The effect of plant growth regulator combinations on DL, PSH, PSHL, MSE, PSHE, AB and VG of Sebri pear cultivar (*in vitro* shoot growth and proliferation).

Treatments	PSH[a]	PSHL[b] (mm)	PSHE[c] (mm)	AB[d]	DL[e]	MSE[f] (mm)	VG[g] (mm)
2BAP	0.4 b*	3.4 ab	3.40 cd	0.6 b	0.0 g	12.60 bcd	16.00 cdefg
2BAP+IBA	1.0 b	9.01 a	9.01 bcd	5.0 a	0.6 g	18.02 abc	27.03 bc
2BAP+GA_3	2.6 a	5.11 ab	13.34 bc	0.0 b	0.0 g	13.00 bcd	26.34 bc
2BAP+IBA+GA_3	0.6 b	8.28 a	8.28 bcd	1.8 b	0.8 gf	25.50 a	33.78 ab
3BAP	3.6 a	4.22 ab	19.03 ab	1.0 b	0.0 g	12.25 bcd	31.27 abc
3BAP+IBA	3.4 a	8.18 a	28.79 a	6.0 a	0.0 g	6.00 ed	42.80 a
3BAP+GA_3	3.0 a	4.38 ab	13.55 bc	0.0 b	0.0 g	17.50 abc	31.05 abc
3BAP+IBA+GA_3	2.6 a	6.7 a	16.30 b	5.2 a	0.0 g	14.00 bcd	22.30 bcde
1TDZ	0.0 b	0.0 b	0.00 d	0.0 b	2.0 efg	2.08 e	2.08 g
1TDZ+IBA	0.0 b	0.0 b	0.00 d	0.0 b	2.6 def	1.20 e	1.20 g
1TDZ+GA_3	0.0 b	0.0 b	0.00 d	0.0 b	3.8 cde	4.90 ed	4.90 fg
1TDZ+IBA+GA_3	0.0 b	0.0 b	0.00 d	0.0 b	3.4 de	5.10 ed	5.10 fg
2TDZ	0.0 b	0.0 b	0.00 d	0.0 b	5.6 bc	9.00 cde	9.00 efg
2TDZ+IBA	0.0 b	0.0 b	0.00 d	0.0 b	6.2 b	10.00 cde	10.00 defg
2TDZ+GA_3	0.0 b	0.0 b	0.00 d	0.0 b	9.0 a	24.60 a	24.60 bcd
2TDZ+IBA+GA_3	0.0 b	0.0 b	0.00 d	0.0 b	4.2 cd	19.80 ab	19.80 bcdef

* Means followed by the same letter in columns are not significantly different as by Duncan's test at 0.01 probability level.

[a] The number of Proliferated SHoots; [b] Proliferated SHoots Length; [c] Proliferated SHoot Extension; [d] The number of Activated lateral Buds; [e] The number of Developed Leaf production; [f] Main Stem Elongation, [g] Vegetative Growth.

Figure 2. Different stages of shoot formation, Sebri pear cultivar in medium containing 3BAP+IBA (a-h). The pictures were taken, using digital microscope model Dino-Lite.

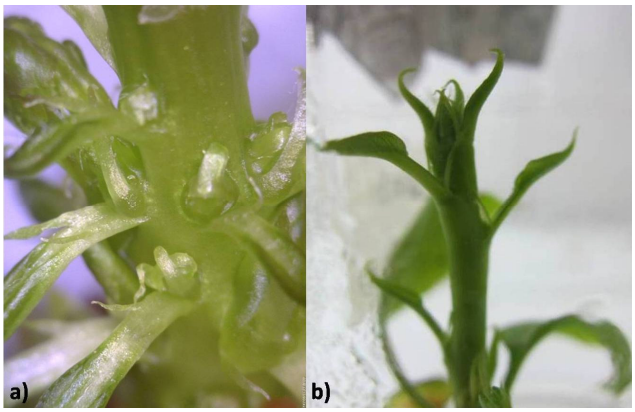

Figure 3. The activated lateral bud in medium containing 2BAP+IBA (a) and the non swelling in the lateral bud in medium supplemented with 2BAP+GA_3 (b), Sebri pear cultivar.

BAP produced only undeveloped leaves. These leaves were developed in 8-12 weeks after the start of the cultures. The highest main stem elongation in BAP combinations was obtained in BAP (2 mg L^{-1})+IBA (0.1 mg L^{-1})+(0.5 mg L^{-1}) GA_3. This is in agreement with the results reported by Machado *et al.* (2011) who reported the highest height in BAP+IBA+GA_3 combination. In TDZ combinations, maximum main stem elongation showed in TDZ (2 mg L^{-1})+(0.5 mg L^{-1}) GA_3. The highest vegetative growth (42.80 mm) was obtained in medium supplemented with BAP (3 mg L^{-1})+IBA and the lowest observed in TDZ (2 mg L^{-1})+IBA and TDZ (2 mg L^{-1}) (Table 4). In BAP (2 mg L^{-1}), BAP (2 mg L^{-1})+IBA (0.1 mg L^{-1}) and as well in BAP (2 mg L^{-1})+IBA (0.1 mg L^{-1})+GA_3 (0.5 mg L^{-1}) combinations, the main stem elongation occurred as more than the proliferated shoot extension, while in BAP (3 mg L^{-1}), BAP (3 mg L^{-1})+IBA (0.1 mg L^{-1}) and in BAP (3 mg L^{-1})+IBA (0.1 mg L^{-1})+GA_3 (0.5 mg L^{-1}) proliferated shoot extension was pronounced more than the main stem elongation. In BAP+GA_3 combination for both concentrations of BAP, the vegetative growth was almost equally divided between the main stem elongation and proliferated shoot extension (Figure 4). Plants derived from this method of propagation were morphologically uniform. Further studies are needed to increase the levels of proliferation of Sebri pear cultivar using either other growth regulators or other chemical supplements. The procedures developed in this study can be used as the basis for a

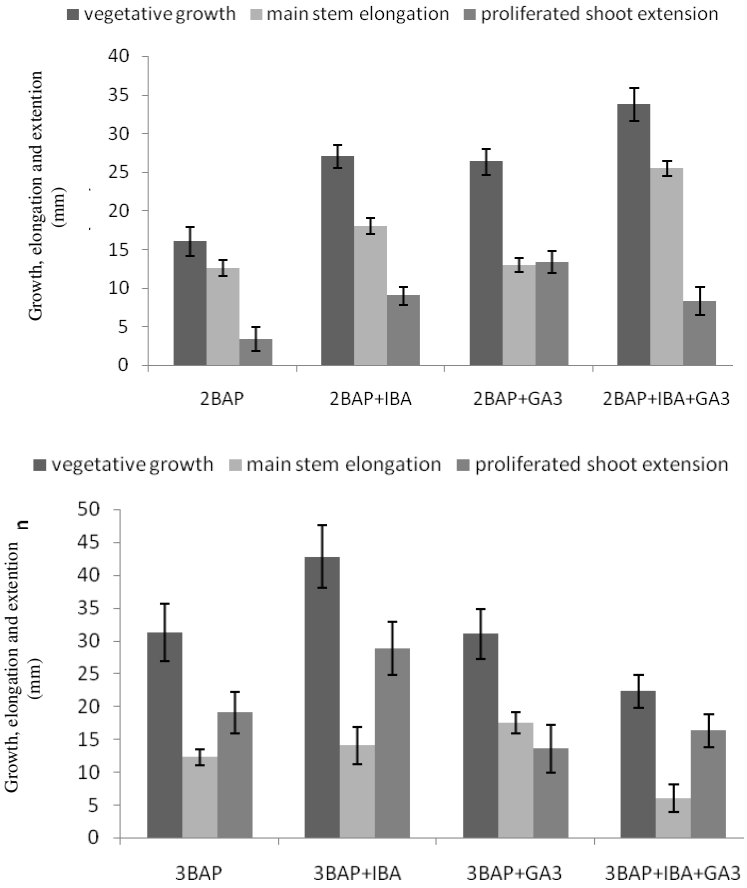

Figure 4. Effect of different plant growth regulator combinations on vegetative growth, main stem elongation, proliferated and shoot extension in Sebri cultivar of pear after 1 month past of growth period. BAP at 2 mg L^{-1} (upper figure) and 3 mg L^{-1}(lower figure). Error bars indicate standard error.

conservation of this valuable cultivar.

REFERENCES

1. Abdollahi, H., Muleo, R. and Rugini, E. 2006. Optimisation of Regeneration and Maintenance of Morphogenic Callus in Pear (*Pyrus communis* L.) by Simple and Double Regeneration Techniques. *Scientia Horticulture*, **108:** 352–358.
2. Amiri, M. E. 2002. Mass Propagation of Unique Variety of Pear (*Pyrus pyrifolia* (Burm) Nak. cv. Sebri by Shoot Tip Culture *In vitro*. *Acta Hort*., **587:** 555-561.
3. Baviera, J. A., García, J. L. and Ibarra, M. (1989) Commercial *In vitro* Micropropagation of Pear cv Conference. *Acta Hort.*, **256:** 63-68.
4. Bell, R. L., Srinivasan, C. and Lomberk, D. 2009. Effect of Nutrient Media on Axillary Shoot Proliferation and Preconditioning for Adventitious Shoot Regeneration of Pears. *In Vitro Cell. Dev. Biol.,* **45:** 708-714.
5. Davarynejad, G. H. and Davarynejad, E. 2007. Comparative Performance of Graft Incompatibility in Pear/Quince (*Pyrus comunis*/*Cydonia oblanga*) Combination. *Acta Hort*., **732:** 221-227.
6. Davarynejad, G. H., Shahriari, F. and Hassanpour, H. 2008. Identification of Graft Incompatibility of Pear Cultivars on Quince Rootstock by Using Isozymes Banding Pattern and Starch. *Asian J. Plant Sci.,* **7(1):** 109-112.
7. Dolcet-Sanjuan, R., Mok, D. W. S. and Mok, M. C. 1990. Micropropagation of Pyrus and Cydonia and Their Responses to Fe-limiting Conditions. *Plant Cell. Tiss. Organ Cult.*, **21**: 191-199.
8. Dwivedi, S. K. and Bist, L. D. 1999. *In vitro* Propagation of Low-chill Pear cv. Gola. *Indian J. Hort.,* **56:** 189-193.
9. Erfani, J., Ebadi, A., Abdollahi and H., Fatahi, R. 2012. Genetic Diversity of Some Pear Cultivars and Genotypes Using Simple Sequence Repeat (SSR) Markers. Plant Mol. Biol. Rep., 30: 1065–1072.
10. Grattapaglia, D., Machado, M. A. 1998. Micropropagação. In: "*Cultura de Tecidos e Transformação Genética de Plantas*", (Eds.): Torres, A. C., Caldas, L. S. and Buso, J. A.. EMBRAPA, Brasília, **1:** 508.
11. Kadota, M. and Niimi, Y. 2003. Effects of Cytokinin Types and Their Concentrations on Shoot Proliferation and Hyperhydricity in *In vitro* Pear Cultivar Shoots. *Plant Cell, Tiss. Organ Cult.,* **72:** 261–265.
12. Machado, M. P., Silva, A. L. L. and Biasi, L. A. 2011. Effect of Plant Growth Regulators on in Vitro Regeneration of *Lavandula denata* L. Shoot Tips. *J. Biotech. Biodiv*. **3**: 28-31
13. McCown, D. D. and McCown, B. H. 1987. North American hardwoods. 3. Case Histories: Gymnosperms, Angiosperms and palms. In: "*Cell and Tissue Culture in Forestry*", (Eds.): Bonga, J. M and Durzan, D. J.. Martinus Nijhoff Publishers, Dordrecht, PP. 247-260.
14. Murashige, T. and Skoog, F. 1962. A Revised Medium for rapid growth and bioassays with tobacco tissue cultures. *Physiol. Plant*., **15:** 473– 497.
15. Nandi, S. K., Letham, D. S., Palni, L. M. S., Wong, O. C. and Summons, R. E. 1989. 6-Benzylaminopurine and Its Glycosides as Naturally Occurring Cytokinins. *Plant Sci.* **61:** 189–196.
16. Nosrati, S. Z., Zamani, Z. and Babalar, M. 2009. Micropropagation of Four Cultivars (Dargazi, Natanzi, Shahmiveh and Williams) of Pear (*Pyrus communis* L.). *Iran. J. Horti. Sci.,* **40(2)**: 83-91.
17. Rahemi, M., Baghbani, R. 2002. Effects of Harvesting Time and Storage Period on Poststorage Ripening of Esfahan 'Sebri' Pear (*Pyrus serotina* Rehd. cv. 'Sebri'). *Acta Hort. (ISHS),* **587:** 519-523.
18. Rouzban, M. R., Arzani, K. and Moieni, A. 2002. Study on *In vitro* Propagation of Some Asian Pear (*Pyrus serotina* Rehd.) Cultivars. *Seed Plant Improv. J.*, **18:** 348-361.
19. Safarpour, M., Bahari, M., Tabatabaei, E. B. S. and Abdelahi, H. 2008. Determination Old Genetic Diversity in Pear (Pyrus spp.) Using Microsatellite Markers. Iran. J. Horti. Sci. Tech., **9(2)**: 113-128.
20. SAS Institute Inc. 1989. *SAS/STAT User's Guide, Version 6.* 4th Edition, Volume 2. SAS Institute, Inc., Cary.
21. Shibli, R. A., Ajlouni, M. M., Jaradat, A., Aljanabi, S. and Shatnawi, M. 1997. Micropropagation in Wild Pear (*Pyrus syricca*). *Scientia Hort.,* **68**: 237-242.
22. Singha, S. 1980. *In vitro* Propagation of 'Seckel' Pear. In: *Proceedings of the Conference on 418 Nursery Production of Fruit Plants through Tissue Culture:*

Application and Feasibility, April 21-22, P. 7.
23. Stimart, D. P. and Harbage, J. F. 1989. *In vitro* Shoot Proliferation of *Pyrus calleryana* from Vegetative Bud. *HortSci.*, **24**: 298-299.
24. Vatandoost J, S., Davarynejad, G. H., Tehranifar, A. and Kaveh, S. 2011. Effect of Auxin Treatments and Cutting Position on Rooting of Pear Cultivars (Sebri , Shekari and Natanz) Cuttings. *J. Horti. Sci.*, **25(1):** 38-44.
25. Zafari nia, H., Arzani, K. and Ghasemi, A. A. 2010. An Evaluation of Carbohydrates and Nutrient Content of Some Young Asian Pear (*Pyrus serotina* Rehd.) Cultivars on European Pear (*Pyrus communis* L.) Seedling Rootstocks under Isfahan Environmental Conditions. *Iran. J. Horti. Sci.,* **41(3):** 209-221.

استقرار و تکثیر همگروهی گلابی سبری در شرایط درون شیشه ای

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چکیده

دو آزمایش برای بررسی استقرار، پرآوری و رشد شاخساره از جوانه های جانبی گلابی سبری انجام گرفت. در آزمایش اول، غلظت های مختلف IBA (۰، ۰/۱، ۰/۳ و ۰/۵ میلی گرم بر لیتر) بر استقرار ریزنمونه بررسی شد. در آزمایش دوم تاثیر ترکیبات مختلف هورمونی بر پرآوری و رشد شاخساره مورد ارزیابی قرار گرفت. در غلظت های پایین IBA (۰ و ۰/۱ میلی گرم بر لیتر) ریزنمونه ها بیشتر استقرار یافتند در صورتیکه در غلظت های بالاتر کالوس بیشتری تولید شد و ریزنمونه های کمتری استقرار یافتند. پرآوری شاخساره در تمام ترکیبات حاوی BAP دیده شد در حالیکه TDZ هیچ تاثیری بر پرآوری شاخساره نداشت. غلظت ۳ میلی گرم BAP تعداد شاخساره پرآوری شده بیشتری را نسبت به غلظت ۲ میلی گرم بر لیتر القا کرد. طول شاخه های پرآوری شده در ترکیبات BAP (2, 3 mg L^{-1}) + IBA (0.1 mg L^{-1}) و BAP (2, 3 mg L^{-1}) + IBA (0.1 mg L^{-1}) + GA_3 (0.5 mg L^{-1}) بالاترین بود. غلظت های بالاتر منجر به توسعه بالاتر BAP شاخه های پرآوری شده گردید. حضور IBA در ترکیبات دارای BAP باعث تورم جوانه های جانبی شد در حالیکه GA_3 از آن ممانعت کرد. بیشترین برگ های توسعه یافته در ترکیب TDZ (2 mg L^{-1}) + GA_3 (0.5 mg L^{-1}) تولید شد. در ترکیبات BAP (2 mgL^{-1}) + IBA (0.1 mg L^{-1}) + GA_3 و TDZ (2 mgL^{-1}) + GA_3 (0.5 mg L^{-1}) بالاترین میزان طویل شدن ساقه اصلی بدست آمد. عادت رشد رویشی با توجه به غلظت BAP و ترکیب تنظیم کننده رشد متفاوت بود و بالاترین مقدار آن در ترکیب BAP (2 mgL^{-1}) + IBA (0.1 mg L^{-1}) مشاهده شد. ترکیب هورمونی BAP در غلظت ۲ میلی‌گرم بر لیتر به همراه ۰/۵ میلی‌گرم بر لیتر GA_3 برای پرآوری شاخساره تیمار مناسبی معرفی می‌گردد.