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Choose a Planting Substrate and Fertilization Method to Achieve Optimal Growth of *Araucaria excelsa*

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Abstract

Due to the lack of scientific knowledge for the use of substrate and best fertilizing method in the growth of *Araucaria excelsa*, it is crucially important to find the most appropriate planting substrate and fertilizing method. The effect of conventional fertilizing method and the application of Fe nanofertilizer were studied on the growth of *A. excelsa* in which six fertilizing methods and three substrates were examined in a factorial experiment based on a Randomized Complete Design with three replications. The seedlings of *A. excelsa* were transferred to 4-L pots after the selection of substrate type and were placed in greenhouse at day/night temperature of 21-23/17-19°C. Some growth indices were recorded including plant height, lateral shoot length, stem diameter, root length, shoot and root fresh weight, shoot and root dry weight, total fresh and dry weight, and the concentrations of some nutrients in leaves. It was found that the substrate A_3 (25% cocopeat + 25% vermicompost + 25% light soil + 25% perlite) was the most appropriate substrate for this plant in terms of most estimated traits (lateral shoot length, root fresh weight, root dry weight, total dry weight, and shoot P and Fe content). Among fertilizing compounds, the treatment B3 (Ferrilene + NPK (20-20-20) + diammonium phosphate + potassium sulfate + ammonium sulfate) was found to be superior for some traits like lateral shoot length, shoot fresh weight, total fresh weight, shoot dry weight and total dry weight. It was revealed that the treatment A_3B_2 (25% cocopeat + 25% vermicompost + 25% light soil + 25% perlite) + diammonium phosphate, potassium sulfate and ammonium sulfate was the most appropriate treatment in terms of shoot and root fresh weight, total fresh weight and total dry weight. So, it is recommended to apply this treatment in the production of *Araucaria excelsa.*

Keywords: *Araucaria excelsa*, Fe nanofertilizer, Growt medium, Vermicompost.

INTRODUCTION

Given the importance of ornamental plants, their nutrient management plays a critical role in enhancing their production and quality. When soil is used as the growing substrate for pot plants, plants may encounter with severe, adverse physical problems due to the possible problems of the soil. So, growers prefer to use substrates with the features of soilless substrate. The beginning of the use of soilless substrates dates back to the use of organic substrates, especially peat, in 1960 (Shi *et al.*, 2002). Planting substrates may be prepared with different materials for optimum physical and nutritional characteristics. But, organic matters for substrate improvement are expensive and difficult-to-prepare (Dibenedetto *et al*., 2004). Organic matters are an important element of soil fertility. The main resources of organic matters are manure, plant residue, sewage and compost of urban waste which are growingly interested due to the importance of organic agriculture and reducing the environmental problems through sustainable agriculture (Sihag and Singh, 1997).

In a study on the effect of vermicompost (100 or 200 g pot^{-1}) and NPK fertilizer ratios (6-12-12) on growth and yield of roses, Senthilkumar *et al*. (2004) found that vermicompost and NPK significantly improved the growth, yield and quality of rose flowers as compared to control and that the improvement was greater when vermicompost and NPK fertilizers were mixed. Chamani *et al.* (2008) studied the impact of vermicompost on the growth and flower-ing of *Petunia hybrida* 'Dream Neon Rose' in greenhouse conditions using 0, 20, 40 and 60% ratio of vermicompost and reported that vermicompost had positive, significant influence on the number of flower, leaf growth, and branch fresh and dry weights as compared to plants grown in vermicompost-lacking soil. Also, they stated that the highest yield was obtained from the treatment of 20% vermicompost, the highest Fe and Zn amounts were obtained from the treatment of 60% vermicompost and the lowest amounts were obtained from control. Vleeschauwer *et al.* (1980) applied a mixture of 50% urban waste compost + 50% tree bark or peat as the substrate and found that it had significant effect on the growth of Cordyline as compared to pure peat as control.

In a study in Alexandria, Egypt in 2006 and 2007, six treatments including four macronutrients and six micronutrients were applied as foliar to carnation cv. "Red Sim" to study their effect on growth and flowering. It was found that such flowering parameters as diameter and fresh and dry weight of the flowers were increased and number of days to flowering was decreased. Also, stem length, diameter and fresh and dry weight and leaf number and fresh and dry weight were increased significantly (El-Naggar, 2009). Sajid *et al*. (2009) examined the effect of foliar application of plant growth regulators and nutrients on lily flowers in which 19 mg L^{-1} KNO_3, 16.5 mg L^{-1} NH_4NO_3, 1.7 mg L^{-1} KH_2PO_4, 4.4 mg L^{-1} $CaCl_2$ and 3.7 mg L^{-1} $MgSO_4$ were sprayed. Also, gibberellic acid (GA3) at the rate of 20 mg L^{-1} was applied. It was revealed that the application of nutrients increased stem length by 25% as compared to control and that the indices were improved by 33% when nutrients and GA_3 were applied.

In an examination of the effect of N, P and K fertilizers on marigold, Zhiping *et al*. (2008) stated that K increased leaf yield and plant dry weight as well as plant growth and longevity. In an evaluation of the impact of different planting media including rice husk, coco peat, perlite, and a mixture of perlite and vermiculite (1:1) and NPK rates including two NPK concentrations (25:7:7) of 1.75 and 2.5 g L^{-1} on chrysanthemum, the best quality of cut flowers were obtained from rice husk substrate sprayed with 2.5 g L^{-1} NPK (Budiarto *et al.*, 2006).

The main method for propagating ornamental Norfolk Island pine is seed planting. But, the seeds produced in the climatic conditions of Iran have low viability. So, a few agribusinesses that are officially certified by Seed and Plant Production Institute for the import of seeds and plant organs import the seeds of Norfolk Island pine and, recently, its newly emerged seedlings. Unfortunately, a lot of these seedlings are eliminated from production cycle or are ready to be sold with delay due to the lack of technical knowledge about the best type and mixture of substrate and fertilizing method which imposes extra ex-penses. The present study aims at proposing the best substrate and fertilizing method in order to accelerate vegetative growth of Norfolk Island pines and reduce their losses by which crop quality and marketability can be increased and production costs can be reduced.

MATERIALS AND METHODS

The present study was carried out as a factorial experiment on the basis of a Random-ized Complete Design with two factors of planting substrate and fertilizing method. The for-mer in-cluded three substrates and the latter was composed of six fertilizing methods. In total, there were 18 treatments with three replications. The study was carried out in the laboratory of Rasht Branch of Islamic Azad University and a greenhouse in Chaboksar in 2014. The study location had been disinfected from all diseases, pests and weeds.

The seedlings of Norfolk Island pine (*Araucaria excelsa*) were purchased from a commer-cial greenhouse in Tonekabon and then, were immediately handled to the greenhouse in Chaboksar. All seedlings were applied with copper oxychloride (1:1000) to kill all possible pathogens like Phytophthora. The seedlings with the same and uniform height and covering crown were used in the study. Table 1 shows the properties of the studied substrates and

When the substrates were ready, it was plants' turn to be established for which 144 seedlings were transferred to 3-liter pots. The soil around roots was slowly cleaned and then, the roots were soaked in pre-prepared Roral TS (1:1000) fungicide to remove all soil-borne fungi around the roots. At the next step, the seedlings were planted in substrates so that the crown was at the same level with substrate surface, and the pots were labeled. The pots were stored in the roofed green-house with day/night temperature range of 21-23/17-19°C accord-ing the preplanned plan.

NPK (20-20-20) produced by Mil-Tar Agriculture Company in Turkey were produced from the market in 10-kg packages. This powdery fertilizer is composed of macro-elements including 20% N + 20% P + 20% K as well as micro-elements. It is homogenous and uniform, easily dis-solves in water and has low EC. It, also, has a suitable quantity of micronutrients. Fe-enriched nanofertilizer 10% was the production of Sepehr Parmis Technological Firm which was used to remedy Fe deficiency in addition to balancing plants' nutrition. Ferrilene fertilizer was produced in Spain. It has 6% absorbable Fe with amendments. Also, it has 3% N and 5% S. It is produced as microgranules which are water solvable. The EDDHA of this fertilizer is stable and available to plants in whole growth period even in the most alkaline soils because of containing a high ratio of O-ortho isomer.

Measurement of plant growth indices

Plant height was measured from the crown to the tip of terminal shoot by a digital cal-iper (in cm) six times in 15-day intervals from the planting of seedlings in pot until harvesting. The number of lateral shoots, the number of branches and inter-branch spacing were measured by a digital caliper (in mm) and the distance of the first branch to the lowest leaf was also measured.

After the harvesting of the plants, their leaves were detached and shoot fresh weight was weighed by a digital scale (in g). To measure root fresh weight by the digital scale, the soil was

Table 1. Chemical properties of the studied substrates

Substrates	N (%)	P (mg/kg)	K (mg/kg)	M (mg/kg)	Zn (mg/kg)	Fe (mg/kg)	EC (1:5 ratio) (dS/m)	pH	CEC (Cmol(+)/kg)	OM (%)	Naex (Cmol(+)/kg)
		Substrates									
A1	2.10	64.0	300	24.80	12.60	59.60	1.09	5.3	36.4	48.40	24.0
A2	0.23	23.9	1400	6.88	1.76	9.62	0.43	6.2	24.8	3.93	1.3
A3	0.22	2.2	2450	7.76	22.73	9.72	0.32	5.0	9.6	2.28	0.9

A_1: 100% cocopeat
A_2: 25% cocopeat + 25% peanut shell compost + 25% light soil (loam sandy) + 25% perlite
A_3: 25% cocopeat + 25% vermicompost + 25% light soil (loam sandy) + 25% perlite

Table 2. Fertilizing treatments used in the study

Treatment	Symbol	Type of applied fertilizer	Specification and application method
B1	FC	Ferrilene and NPK (20-20-20)	Foliar application of a mixture of two fertilizers, both with the ratio of 2:1000, at three stages on Oct. 24, Nov. 7 and Nov. 21, 2014
B2	NPK	Diammonium phosphate, ammonium sulfate, and potassium sulfate	Diammonium phosphate 0.5 g/pot applied as base added to pots on Oct. 24, 2014. Potassium sulfate applied at two stages as foliar and ammonium sulfate 2:1000 applied at four stages in 2-week intervals on Oct. 24, Nov. 7, Nov. 21 and Dec. 5, 2014.
B3	FC and NPK	Ferrilene and NPK (20-20-20) and diammonium phosphate, ammonium sulfate and potassium sulfate	Foliar application of Ferrilene + NPK (20-20-20) with 2:1000 ratio at 2-week intervals (three stages) + NPK treatment (diammonium phosphate 0.5 g/pot (once at the beginning of fertilization) + potassium sulfate 2:1000 at 2 stages + ammonium sulfate 2:1000 at two four stages (2-week intervals)) on Oct. 24, Nov. 7, Nov. 21 and Dec. 5, 2014
B4	FeN	Nano enriched fertilizer Fe 10%	Nano enriched fertilizer Fe 10% 2:1000 at two stages at 2-week intervals on Oct. 24 and Nov. 7, 2014
B5	FeN	Nano enriched fertilizer Fe 10%	Nano enriched fertilizer Fe 10% 2:1000 at four stages at 2-week intervals on Oct. 24, Nov. 7, Nov. 21 and Dec. 5, 2014
B6		Nano enriched fertilizer Fe 10% + diammonium phosphate + ammonium sulfate + potassium sulfate	Nano enriched fertilizer Fe 10% 2:1000 at two stages (Oct. 24 and Nov. 7, 2014) + NPK (diammonium phosphate 0.5 g/pot, potassium sulfate 2:1000 at two stages, ammonium sulfate 2:1000 at four stages) on Oct. 24, Nov. 7, Nov. 21 and Dec. 5, 2014

evacuated from the pots, the roots were cleaned from the soil, and then they were detached from the crown by scissors. For dry weight measurement, all plants of each treat-ment were placed in paper packages labeled with the specifications of the relevant treatment. Then, the samples were oven-dried at 75°C for 48 hours. Next, root and shoot dry weights were separately estimated by a digital scale (in g). Plant total fresh weight was considered as the sum of total shoot and root fresh weights and plant total dry weight as the sum of total shoot and root dry weights.

Chemical specifications of substrates and plant leaves

The pH and EC of substrates were measured by pH-meter and EC-meter, respectively in an extract of 1:5 ratio of substrate to water (Manteghi, 1986). The substrates and plant leaves were extracted (Ehyaei and Behbahanizadeh, 1993). Five grams of substrates and plant leaves were selected and poured into 250-ml Erlenmeyer. Then, 25 ml AB-DTPA and 25 ml distilled water were added, and the samples were shaken for one hour. Afterwards, they were poured into 50 ml centrifuge tube and were centrifuged for 10 minutes to separate the liquid and solid parts. Then, they were filtered. The N content of substrate and plant leaf samples was estimated by Kjeldahl method, the P content by spectrophotometry, the K content by photometry film method, and the Fe, Zn and Mn contents by atomic absorption method (Imami, 1995).

The data were analyzed by SPSS and SAS software packages and the means were compared by Least Significant Difference (LSD) test.

RESULTS AND DISCUSSION

Analysis of variance

According to the results of analysis of variance (Tables 3, 4 and 5), the interaction be-tween substrate and fertilizing method was statistically significant for root fresh weight, root dry weight

Table 3. Analysis of variance that is related to the effect of treatments on some growth indices

S.o.V	df	Means of squares					
		Plant height	Longest branch length	Branch number	Stem diameter	Shoot fresh weight	Root fresh weight
Substrate	2	32.667^ns	4.741^ns	0.019^ns	0.056ns	309.894*	192.600**
Fertilization	5	159.722**	40.696**	0.152^ns	0.817**	2144.681**	205.184**
Substrate × fertilization	10	2.956^ns	1.585^ns	0.041^ns	0.285ns	226.232**	81.566*
Error	36	11.778	3.370	0.093	0.186	65.845	28.944
Total	54	-	-	-	-	-	-
C.V. (%)		54.39	61.41	26.77	71.94	40.44	28.12

* and ** show significance at the 1 and 5% probability levels, respectively. ns shows non-significance at the 5% level

Table 4. Analysis of variance that is related to the effect of treatments on some growth indices

S.o.V	df	Means of squares				
		Total fresh weight	Root length	Root dry weight	Shoot dry weight	Total dry weight
Substrate	2	977.635**	200.685**	31.812*	74.979**	156.930**
Fertilization	5	3629.152**	41.352**	42.756**	214.143**	438.740**
Substrate × fertilization	10	478.439**	33.302**	15.297*	30.939**	67.836**
Error	36	150.296	8.407	5.996	7.654	21.038
Total	54	-	-	-	-	-
C.V. (%)		33.74	16.06	40.89	39.27	36.85

* and ** show significance at the 1 and 5% probability levels, respectively. ns shows non-significance at the 5% level

Table 5. Analysis of variance for the effect of treatments on P, K, Fe, Zn and Mn

S.o.V	df	Means of squares				
		P	K	Fe	Zn	Mn
Substrate	2	868.320^ns	16438.542**	2.251^ns	0.038^ns	5.242**
Fertilization	5	14451.591**	4078.542**	73.070**	0.505**	2.150**
Substrate × fertilization	10	2308.217^ns	378.542^ns	9.798*	0.493**	1.862**
Error	36	1243.494	521.875	4.694	0.093	0.618
Total	54	-	-	-	-	-
C.V. (%)		18.78	13.03	34.51	47.34	100.6

* and ** show significance at the 1 and 5% probability levels, respectively. ns shows non-significance at the 5% level

and Fe content in shoot at the 5% level and for shoot fresh weight, total plant fresh weight, root length, shoot dry weight, plant total dry weight, and Zn and Mn content in shoot at the 1% level. The interaction between the treatments was not significant for the number of branches, plant height, the length of the longest shoot, stem diameter, and P and K contents in shoot (Table 2, 3 and 4).

Substrate × fertilizing method interaction and plant growth

Shoot fresh weight: Means comparison revealed that plants grown in A_3B_2 produced the highest shoot fresh weight (74.400 g). The lowest one was produced by plants grown in A1B4 (foliar spray of nano enriched fertilizer Fe 10%, 2:1000, at two stages at 2-week inter-vals in 100% cocopeat) (Table 6). Highest shoot fresh weight under A_3B_2 treatment was likely to be related to the physical and chemical properties of substrate facilitating water absorption by plants. In general, peat as growth substrate induces plant growth and development because of its porosity and the retention of water and nutrients.

Table 6. Means comparison of interaction of substrate and fertilizing method for growth indices

Treatment	Shoot fresh weight (g)	Root fresh weight (g)	Total fresh weight (g)	Root length (cm)	Root dry weight (g)	Shoot dry weight (g)	Total dry weight (g)
A_1B_1	31.700 ^{fgh}	21.533 ^{d}	53.233 $^{e-h}$	34.500 ^{abc}	6.233 ^{c}	10.333 ^{ef}	16.567 ^{efg}
A_1B_2	43.500 $^{c-j}$	29.333 $^{a-d}$	72.833 $^{c-g}$	37.000 ^{a}	9.100 ^{bc}	14.033 $^{c-f}$	23.133 $^{c-g}$
A_1B_3	55.933 ^{abc}	22.400 ^{d}	78.333 $^{b-f}$	30.500 $^{a-g}$	6.800 ^{c}	18.100 $^{a-d}$	24.900 $^{b-f}$
A_1B_4	22.300 ^{h}	18.333 ^{d}	40.667 ^{h}	35.667 ^{ab}	5.700 ^{c}	7.300 ^{f}	13.000 ^{g}
A_1B_5	26.100 ^{gh}	22.000 ^{d}	48.100 ^{fgh}	32.000 $^{a-f}$	5.733 ^{c}	9.200 ^{f}	14.933 ^{fg}
A_1B_6	49.433 $^{b-f}$	31.400 $^{a-d}$	80.833 $^{b-e}$	34.000 $^{a-d}$	10.633 ^{abc}	16.833 $^{b-e}$	27.467 $^{b-e}$
A_2B_1	32.000 ^{fgh}	27.133 ^{bcd}	59.133 $^{e-h}$	37.000 ^{a}	8.333 ^{c}	12.300 $^{d-f}$	20.633 $^{d-g}$
A_2B_2	53.000 $^{b-e}$	28.600 $^{a-d}$	81.600 $^{b-e}$	26.000 ^{efg}	8.700 ^{c}	18.700 $^{a-d}$	27.400 $^{b-e}$
A_2B_3	68.433 ^{ab}	38.133 ^{abc}	106.567 ^{ab}	30.500 $^{a-g}$	9.733 ^{abc}	24.600 ^{a}	34.333 ^{abc}
A_2B_4	33.833 ^{efgh}	31.000 $^{a-d}$	64.833 $^{d-h}$	27.000 $^{d-g}$	8.700 ^{c}	13.033 $^{d-f}$	21.733 $^{d-g}$
A_2B_5	24.400 ^{gh}	21.633 ^{d}	46.033 ^{gh}	24.500 ^{g}	4.800 ^{c}	8.600 ^{f}	13.400 ^{fg}
A_2B_6	62.933 ^{abc}	30.000 $^{a-d}$	92.933 $^{a-d}$	28.000 $^{c-g}$	9.333 ^{abc}	22.600 ^{ab}	31.933 $^{a-d}$
A_3B_1	30.200 ^{fgh}	24.433 ^{cd}	54.633 ^{efh}	27.500 $^{c-g}$	6.900 ^{c}	9.433 ^{f}	16.333 ^{efg}
A_3B_2	74.400 ^{a}	41.733 ^{a}	116.133 ^{a}	28.000 $^{c-g}$	14.900 ^{ab}	24.400 ^{a}	39.300 ^{a}
A_3B_3	61.200 ^{abc}	38.500 ^{ab}	99.700 ^{abc}	32.500 $^{a-e}$	15.267 ^{a}	20.133 ^{abc}	35.400 ^{ab}
A_3B_4	36.100 $^{d-h}$	29.133 $^{a-d}$	65.233 $^{d-h}$	29.500 $^{b-g}$	8.833 ^{c}	13.633 $^{c-f}$	22.467 ^{g}
A_3B_5	27.133 ^{gh}	20.633 ^{d}	47.767 ^{fgh}	23.500 ^{g}	5.400 ^{c}	11.100 ^{ef}	16.500 ^{efg}
A_3B_6	40.000 $^{d-h}$	26.600 ^{bcd}	66.600 $^{d-h}$	25.000 $^{f-g}$	8.600 ^{c}	13.200 ^{def}	21.800 $^{d-g}$

* and ** show significance at the 1 and 5% probability levels, respectively. ns shows non-significance at the 5% level

Aslam Khan and Ahmad (2002) studied the effect of various NPK doses on the growth and flowering of gladiolus and concluded that optimum rates of P and K and high rate of N resulted in higher vegetative growth indices including height, leaf number, leaf length and spike length. In a study on the impact of various substrates and fertilizing methods on growth and yield of common daisy, Mohamadi Torkashvand *et al.* (2014) found that soil application of fertilizer increased leaf number, plant height, flower diameter, flowering branch length, flower number, flower durability, shoot fresh weight and shoot dry weight as compared to its foliar application and no-fertilization. Zadehbagheri *et al*. (2011) stated that simultaneous use of N, P and K improved leaf width of Narcissus. Niedziela *et al*. (2008) stated that lower N level reduced the vegetative growth of leaves due to the decrease in the capacities of amino acids. The highest fresh weight under treatment A3 (25% cocopeat + 25% vermicompost + 25% light (sand, loamy) soil + 25% perlite) can be related to good physical properties of perlite and physicochemical properties of cocopeat, especially its high water and nutrients retention. Peanut shell compost, also, increases N level and plant growth. In a study on *Dracaena marginata* L., Mohammadi Torkashvand *et al.* (2014) found that the highest N level was related to the substrate of 45% peanut shell compost. P level was decreased at higher compost levels. It seems that as more compost is decomposed at higher rates, the activity of microorganisms transitionally results in the conversion of mineral P to organic P. Consequently, P becomes unavailable to plants. The quantity of K in peanut shell compost is 50 times greater than that in peat. So, as more peanut shell compost is applied to substrate, K level increases proportionally. Under the treatment of 100% peanut shell compost, plant growth was significantly decreased because of higher porosity and lower water retention capacity (Mohammadi Torkashvand *et al*., 2014). Omidi *et al*. (2013) studied the influence of peanut shell compost on physical properties of soil and the yield of violet in free space. They revealed that higher compost level significantly affected stem length and shoot fresh and dry weight as compared to control. In total, peat is a substrate with wide range of application in the retention of water and nutrients due to its porosity which, in turn, affects plant growth and development. However, the effect of higher rates of peat can be associated with its higher C/N ratio, so that when it is added to substrate, microbial activities for decomposing organic matter increases and microorganisms receive more share of substrate N than the plants. Then, plants are exposed to N deficiency which, in turn, results in the loss of plant growth (Campbell, 2008). Similar results were reported by Senthilkumar *et al.* (2004) for roses and by Nikfarjam (2013) and Misaghi (2013) for *Aruacaria excelsa*. In a study on the effect of K nanofertilizer on wheat, Tavan *et al*. (2014) found that shoot fresh weight was not influenced by the applied treatments and that no significant difference was observed between control and other treatments.

Root fresh weight: According to the results of means comparison (Table 6), plants grown under A_3B_2 had the highest root fresh weight (41.733 g). The lowest one (18.333 g) was observed in plants grown under A1B4 (foliar spray of nano enriched fertilizer Fe 10%, 2:1000, at two stages at 2-week intervals in 100% cocopeat). It appears that root fresh weight under A_3B_2 is affected by substrate physical and chemical properties and also P and K amounts in NPK which increases water uptake to plant root. Senthilkumar *et al.* (2004) reported similar findings for roses. This finding is also consistent with Mohammadipour (2012) results in the study on the application of Fe nanofertilizer on the growth *Spathiphyllum* in the sense that did not find significant difference in root fresh weight be-tween the application of nano iron chelate fertilizer and other Fe fertilizers. The better growth of the plants in vermicompost, cocopeat and perlite can be related to suitable condi-tions including appropriate porosity and ventilation. Similar results were reported by Mahboob Khomami (2011) for *Dieffenbachia* and by Nikfarjam (2013) in a study on the ef-fect of various substrates on the growth of *Aruacaria excelsa.* In an experiment on the impact of substrate on the growth of *Aruacaria excelsa*, Misaghi (2013) found vermicompost to be the best treatment for root fresh weight which, seemingly, was associated with the optimum ventilation and light weight of this substrate as well as its very high water retention property and high content of nutrients. Also, good physical properties of perlite and physicochemical properties of cocopeat, especially its high water and nutrients retention capacity, can be the reason for higher fresh weight under the treatment of A_3 (25% cocopeat + 25% vermicompost + 25% light (sand, loamy) soil + 25% perlite) in the present study. Similar findings were re-ported by Snagwan *et al*. (2010) for marigold, Arshad (2009) for vegetative and flowering characteristics of petunia, and Bachman and Metzger (2008) for tomato and parsley. In an experiment on wheat growth factors as affected by K nanofertilizer, Tavan *et al*. (2014) concluded that K nanofertilizer reduced root fresh weight in all treatments as compared to control. The results are in agreement with Banijamali (2004) on the effect of sources of K and micronutrients on quantitative and qualitative yield of chrysanthemum and with Aslam Khan (2002) on the effect of different NPK levels on the growth and flowering of gladiolus.

Total fresh weight: As results showed, the highest total fresh weight of 116.133 g was obtained under A_3B_2 and the lowest one (40.667 g) under A1B4 (Table 6). It can be said that total fresh weight under A_3B_2 was influenced by physical and chemical properties, appropriate ventilation and porosity of substrate and better, faster uptake of nutrients in foliar application which resulted in wider leaf area, higher photosynthesis and the mobilization of these substances towards root development. In a study on the effect of various substrates and fertilizing methods on growth and yield of common daisy, Mohammadi Torkashvand *et al.* (2014) concluded that the mixture of tea waste compost, urban waste compost and Azolla compost (each with the ratio of one-third) and foliar application brought about the best root growth and the highest root fresh weight. On the other hand, when ammonium is used as fertilizer, this type of nitrogen should be detoxified immediately after uptake due to high uptake rate of ammonium. Then, more organic N compounds would be synthesized which can enhance the shoot growth and flower and fruit formation (Claussen and Lenz, 1995). Gholam nezhad *et al*. (2012) showed for *Capsicum annuum* L. that the highest root and shoot fresh and dry weight, seedling diameter, leaf area rate and seedling height were obtained from the substrate of vermicompost:cocopeat (1:3). In a study on the effect of fertilizing method and substrate on Aruacaria excelsa, Nikfarjam (2013) obtained the highest total fresh weight from the substrate of 25% cocopeat + 25% vermicompost + 25% light soil + 25% perlite.

The results are in agreement with El-Naggar (2009) about the use of materials containing various microelements to achieve optimum growth of *Dianthus caryophyllus* L., Banijamali (2004) about the effect of the source of K and microelements on quantitative and qualitative traits of chrysanthemum, and Aslam Khan (2002) about the effect of different NPK levels on growth and flowering of gladiolus.

Root length: It was revealed that the highest root length was produced by plants grown under A_1B_2 and A_2B_1 conditions and the lowest root length of 23.500 cm was exhibit-ed by plants grown under A_3B_5 treatment. It is suggested that different patterns for the uptake of minerals by different plants may be the likely reason for the varied response of plants to different vermicompost concentrations (Samiran *et al.*, 2010). In a study on the effects of substrate on vegetative and reproductive traits of strawberries, Tehrani (2013) found that cocopeat gave better results than other substrates. So, it could be said that the physical attributes of the substrate had significant impact on the root growth and development of strawberries. Indeed, it could be concluded that how a substrate is prepared and what physical and chemical attributes may influence the root growth and nutrients uptake by strawberries affecting shoot growth and yield. Mohammadi *et al.* (2014) studied the impact of different ratios of vermicompost in substrate on the seedling growth of *Physalis peruviana* L. and concluded that the best root length was obtained under cocopeat-perlite substrate. These results are in agreement with our findings. It seems that the increase in water uptake by roots is influenced by substrate physical and chemical properties as well as P and K content of NPK fertilizer and micronutrients. In a study on rose, Senthilkumar *et al.* (2004) found similar findings. Higher root length in A_1 and A_2 substrates and foliar application method can be associated with appropriate ventilation and porosity of these substrates and foliar application method, which results in better and faster uptake of nutrients, higher leaf area and higher photosynthesis and the mobilization of photosynthates to root development (Mohammadi Torkashvand *et al.,* 2014). In a study on the effect of Fe nanofertilizer on the growth of *Spathiphyllum*, Mohammadipour (2012) concluded that root length was decreased by the treatment of Fe nanofertilizer as compared to control. Peyvandi *et al.* (2011) reported similar results for nanofertilizers.

Root dry weight: Results showed that plants grown in A_3B_3 produced the highest root dry weight of 15.267 g and those grown in A_2B_5 produced the lowest root dry weight of 4.8 g (Table 6). Substrate A_3 was a mixture of four different substrates in equal ratios (cocopeat, vermicompost, light soil and perlite). Each substrate on its own has the properties needed for creating a growing medium for the plants. Their mixture can provide a better growing medium supplying the main requirements including water, nutrients and oxygen for root respiration and being a harbor for the roots to improve root system and root dry weight. The physical properties of the substrate, such as high porosity, ventilation, drainage, high water retention capacity and microbial activity, may improve root system. Gayasinghe *et al.* (2010) used cattle manure compost (CMC) and synthetic aggregates (SA) instead of peat in the production of marigold and found that 40% SA + 60% CMC enhanced plant height, flower number per plant, stem fresh and dry weight, root length, and root fresh and dry weight. In a study on the influence of different substrates and fertilizing treatments on the growth of *Aruacaria excelsa*, Nikfarjam (2013) obtained the highest root dry weight from 25% cocopeat + 25% vermicompost + 25% light soil + 25% perlite. Similar results were reported by Senthilkumar *et al.* (2004) for rose and Sainz *et al.* (1998) for clover and cucumber. In a study on the growth and yield response of marigold to potting medium containing vermicom-posts produced from different wastes at different mixture ratios of 10, 20, 30 and 40%, Snagwan *et al.* (2010) concluded that the appropriate ratio application of vermicompost resulted in higher bud number, flower number, shoot weight, root weight and plant height, which is in agreement with our study.

Shoot dry weight: According to means comparison, plants grown in A_2B_3 produced the highest shoot dry weight of 24.600 g and those grown in A_1B_4 (nano enriched fertilizer Fe 10%, 2:1000, at two stages at 2-week intervals in 100% cocopeat) produced the lowest one of 7.300 g

(Table 6). One important index for substrate evaluation is shoot dry weight. Davidson *et al*. (1994) reported that composts with C/N ratio of less than 20 are ideal for plant production. C/N ratio of 13.4 in substrates containing peanut shell compost is appropriate for the growth of ornamental plants. However, ratios of higher than 30 may cause problems for plant growth (Verdonck and Gabriels, 1992). In a study on the effect of peanut shell compost as a substrate on the growth of *Dracaena marginata* L., Mohammadi Torkashvand *et al.* (2014) concluded that peanut shell compost had improving effect on such growth traits as height, leaf number and shoot dry weight through reducing C/N ratio, creating suitable pH and supplying more nutrients. It seems that dry weight is influenced by physical and chemical properties of substrate as well as P and K content of NPK and micronutrients which result in improved performance of roots in water uptake. This result is in agreement with the results of Zhiping *et al*. (2008)'s study on the effect of N, P and K fertilization on English marigold, El-Naggar (2009)'s study on the use of materials containing various micronutrients for optimum growth of pink, and Banijamali (2004)'s study on the effect of fertilization with K and micronutrients on quantitative and qualitative yield of chrysanthemum. Substrate physical and chemical properties had decisive role in the activity of plant growth hormones like humic acid and growth regulators like auxin, gibberellin and cytokinin. Nikfarjam (2013), also, obtained the highest shoot dry weight from the treatment of Ferrilene and Kristalon in the substrate containing 25% cocopeat + 25% vermicompost + 25% light soil + 25% perlite. In a study on the application of Fe nanofertilizer on the growth of *Spathiphyllum*, Mohammadipour (2012) obtained similar results for root dry weight. Sajedi and Ardakani (2008) reported for forage corn that micronutrient fertilization was more effective on metabolic activities and increased plant dry weight indirectly via increasing plant growth rate, absorption area, leaf duration and photosynthesis and that it was less effective on growth physiological indices. Senthilkumar *et al*. (2004) reported similar results for rose.

Total dry weight: As is evident in Table 6, plants grown in A_3B_2 had the highest total dry weight of 39.300 g and those grown in A1B5 had the lowest one of 9.200 g. It appears that plant total dry weight is influenced by substrate physical and chemical properties and P and K contents of NPK and micronutrients which improves root capability in water uptake. Since the substrate A_3 contained vermicompost, cocopeat, loam-sandy soil and perlite in equal ratios, it had the optimum physical and chemical conditions for the improvement of plant growth. This improvement was caused by the activity of plant growth quasi-hormones like humic acid and growth regulators like auxin, gibberellin and cytokinin. Similar results have been reported by Chamani *et al*. (2008) for *Petunia hybrida* 'Dream Neon Rose', Sainz *et al*. (1998) for clover and cucumber, Arshad (2009) for petunia, and Senthilkumar *et al*. (2004) for rose. In a study on growth and yield response of marigold to potting media containing vermicompost from different sources with vermicompost mixture ratios of 10, 20, 30 and 40%, Snagwan *et al.* (2010) concluded that the application of vermicompost with appropriate ratio increased bud number, flower number, shoot weight, root weight and plant height, which is in agreement with our study. Nikfarjam (2013) studied the effect of substrate and fertilizing method on Aruacaria excelsa and concluded that the highest total dry weight was obtained from the treatment of Ferrilene and Kristalon to the substrate containing 25% co-copeat + 25% vermicompost + 25% light soil + 25% perlite. The results of the present study confirm Mohammadi Torkashvand *et al.* (2014)'s report about the effect of peanut shell com-post as substrate on the growth of *Dracaena marginata* L. and Aslam Khan (2002)'s report about the effect of different NPK levels on growth and flowering of gladiolus.

Substrate × fertilizing method interaction and nutrients concentration of leaf

Iron concentration: Table 7 shows the interaction between substrate and fertilizing method for leaf Fe content. As means comparison indicated, the treatment A_3B_5 had the high-est Fe content of 15.38 mg kg^{-1} and the lowest one (4.63 mg kg^{-1}) was observed in plants grown in A_3B_2. Fe plays several functions in plant physiology. Since Fe is an intermediate element with more than one ox-

Table 7. Interaction of substrate and fertilizing method for leaf nutrient contents

Treatment	Fe (mg/kg)	Zn (mg/kg)	Mn (mg/kg)
A_1B_1	11.5000 ad	21.533 d	53.233 eh
A_1B_2	8.4833 bf	29.333 ad	72.833 cg
A_1B_3	5.5233 ef	22.400 d	78.333 bf
A_1B_4	10.3667 ae	18.333 d	40.667 h
A_1B_5	12.6700 ac	22.000 d	48.100 fh
A_1B_6	10.5967 ae	31.400 ad	80.833 be
A_2B_1	11.0333 ae	27.133 bd	59.133 eh
A_2B_2	8.6400 bf	28.600 ad	81.600 be
A_2B_3	7.1467 cf	38.133 ac	106.567 ab
A_2B_4	7.0733 cf	31.000 ad	64.833 dh
A_2B_5	13.5967 ab	21.633 d	46.033 gh
A_2B_6	11.4300 ad	30.000 ad	92.933 ad
A_3B_1	10.2133 ae	24.433 cd	54.633 eh
A_3B_2	4.6300 f	41.733 a	116.133 a
A_3B_3	6.8600 df	38.500 ab	99.700 ac
A_3B_4	11.1200 ad	29.133 ad	65.233 dh
A_3B_5	15.3800 a	20.633 d	47.767 fh
A_3B_6	14.4967 a	26.600 bd	66.600 dh

*Values in each column that are followed by the same letter are not significantly different by LSD test

idation state, it can be electron donor or receiver depending on oxidative potential of the reactors. It plays an important role in all respiratory enzymes. It is also involved in photosynthetic reactions (Bindra, 1983). Fe is observed in cytochrome enzyme which is a hematine and protein compound and a prerequisite for oxidative and reductive respiration and reactions in plant. In chlorophyll buildup, first heme compounds, whose molecules are very similar to chlorophyll molecules, are constructed. The only difference is that Fe rather than Mn is in its central nucleus and thus, Fe deficiency results in plant's severe chlorosis (Salardini, 2005). Total Fe content is not a good indicator of Fe nutrition status in all plants because total Fe content of some plants with chlorosis is as great as or even greater than healthy plants. It seems that it is better to consider active Fe in these cases (Katyal and Sharma, 1980). Active Fe, i.e. Fe^{+2}, is a part of plant total Fe which is in-volved in chlorophyll synthesis.

The uptake of an element by a plant is supplied with two factors: element concentra-tion in plant and plant's shoot dry weight. Higher plant dry weight results in higher Fe uptake. Fe nanofertilizer has a natural foundation composed of organic and mineral matters. Nano Fe chelate fertilizer is a reliable fertilizer source for Fe release in rice-growing conditions because of its high stability and controlling capacity under waterlogged conditions. Owing to its Fe and Zn content, this nanofertilizer can enrich the crop naturally (Khalaj *et al.*, 2009). In a study on the effect of nano iron oxide and conventional iron oxide particles on Fe, Zn, Cu and Mn concentration in wheat plants, Mazaherinia *et al*. (2010) reported that the increase in nano iron oxide powder vs. conventional one led to a significant increase in Fe concentration in plant, which could be associated with the properties of nano particles. In the present study, the highest Fe content was observed in foliar application of Fe nanofertilizer which is in agreement with Mohammadipour (2012)'s study on the effect of Fe fertilizers on the growth of *Spathiphyllum*. It was concluded that the foliar application of Fe nanofertilizer produced the second highest leaf Fe content after $Fe_2 (SO4)_3$, showing significant differences with other treatments. Fe nanofertilizer increased leaf Fe content by 2.5 times as compared to control. Mortvedt *et al*. (1982) concluded that foliar application of Fe is the most effective method for correcting Fe deficiency in all crops as compared to soil application. In a study on the effect of substrate containing vermicompost on nutrient concentrations in spinach, Sheikhi and Ronagi (2013) concluded that vermicompost enhanced Fe concentration in spinach shoot. In addition to influencing soil chemical characteristics like pH and nutrient supply, organic mat-ter positively affects soil physical properties improving soil ventilation and penetration. As organic matter dissolves, chelate organic matters are produced too which facilitates Fe uptake (Mohammadi Torkashvand, 2000). In a study on the effect of foliar application of $Fe_2(SO_4)_3$ in two forms – conventional and nanoparticles – on the growth of sunflower cultivars under salinity stress, Torabian and Zahedi (2013) revealed that the two studied forms of $Fe_2(SO_4)_3$ left similar impacts on shoot Fe concentration. However, total Fe content of the plant is not an appropriate indicator of the status of Fe nutrition because some plants suffering from chlo-rosis have the same or even higher amount of total Fe as healthy plants. The critical level of Fe in leaves is about 50 mg kg^{-1} dry matter (Gheibi and Malakouti, 1999).

Zinc concentration: As means comparison showed (Table 7), A_1B_4 (nano enriched fertilizer Fe 10%, 2:1000, at two stages at 2-week intervals in 100% cocopeat) resulted in the highest Zn concentration of 1.7966 mg kg^{-1} and A1B6 in the lowest one (0.21 mg kg^{-1}). Zn deficiency is the second most prevailing symptom of deficiency in flowers and ornamental plants after Fe deficiency. Zn is closely related to auxin level in plant and is observed in some enzymes involved in plant metabolism. Higher Zn uptake may be associated with better plant growth and higher uptake of Zn by plants. In a study on the effect of Fe nanofertilizer on growth, Mohammadipour (2012) obtained the highest Zn content from the foliar application of Fe nanofertilizer. The effect of Fe nanofertilizer was significant on increasing Zn content of shoot in this study too. Saberi et al. (2013) examined the effect of different substrates on the uptake of macro and microelements by cherry tomatoes and recorded the highest Zn up-take in cocopeat treatment. In a study on the effects of peanut shell compost as substrate on the growth of *Dracaena marginata* L., Mohammadi Torkashvand *et al*. (2014) concluded that the highest Zn concentration was obtained from the treatment of 60% peanut shell compost. Sheikhi and Ronagi (2013) studied the effect of vermicompost application on nutrient concentration and yield of spinach and stated that vermicompost decreased mean Zn concentration as compared to control. Hosseinpour *et al*. (2015) studied the effect of vermicompost on micronutrient concentrations in soil and lettuce and found that vermicompost significantly affected Fe and Zn concentrations in lettuce shoot. Fe had an antagonistic relationship with Zn reducing its uptake. As is evident, Fe reduced Zn uptake. High rates of Fe application via fertilization by Fe chelates can result in higher Fe uptake disturbing nutritional balance and creating severe deficiencies of Cu, Mn and Zn in plants (Mousavi and Ronaghi, 2007).

Manganese concentration: Means comparison revealed that A_1B_5 (nano enriched fertilizer Fe 10%, 2:1000, at two stages at 2-week intervals in 100% cocopeat) resulted in the highest Mn concentration of 3.14 mg kg^{-1} (Table 7). The lowest Mn concentration of 0.17 mg kg^{-1} was related to plants growth in A_3B_2. Mn plays an important role in respiration and N metabolism in plants. Furthermore, it is known as the activator of some enzymes and is known to be involved in oxidation and reduction reactions in photosynthesis. Mn is directly or indirectly involved in chloroplast formation and since chlorosis is the general symptom of Mn deficiency, it is believed that Mn plays a role in chlorophyll formation or destruction (Moez Ardalan and Savaghebi Firouzabadi, 1997). In a study on the effect of vermicompost application on nutrient concentrations and yield of spinach, Sheikhi and Ronagi (2013) found that vermicompost increased mean Fe concentration as compared to control but decreased the concentration of Zn, Mn and Cu. Organic matters increase microbial exudation by inducing microbial activity in soil. These substances hinder the precipitation of Mn in insolvable form by Mn chelating resulting in higher Mn absorbability in soil. In a study on the effect of Fe nanofertilizer on *Spathiphyllum*, Mohammadipour (2012) found that leaves had the highest Mn content when treated with nanofertilizer with significant differences with other treatments including foliar application. In this study, foliar treatment of Fe except for nanofertilizer resulted in lower Mn content as compared to control. Having Mn element in its structure, Fe nanofertilizer resulted in the highest Mn content in plants.

CONCLUSION

Results show that among studied substrates, A_3 (25% cocopeat + 25% vermicompost + 25% light soil + 25% perlite) was the most appropriate one for the rooted seedlings of the studied plant in terms of most estimated traits (lateral branch length, root fresh weight, root dry weight, plant total dry weight, and shoot P and Fe content). Pure cocopeat substrate (A_1) enhanced root length, stem diameter and shoot Mn and K content since it can maintain mois-ture for gradual consumption by roots. Since this substrate has high ventilation, root volume develops a lot given the fact that adequate amount of moisture is available to it. Since co-copeat is not an enriched source of nutrients needed for vegetative growth, it reduces lateral branch length, root, shoot and total fresh weight, and root, shoot and total dry weight. Since the seedlings used in the present study were in early growth stages, cocopeat had favorable effects in the first 30 days. But as plants grew, substrate containing organic matter was more favorable to supply the required nutrients. In terms of the interaction between fertilizing method and substrate for root length, A_1B_2 (NPK fertilizers including diammonium phos-phate 0.5 g pot^{-1}, potassium sulfate 2:1000 at two stages and ammonium sulfate 2:100 + 100% cocopeat) was found to be the best treatment.

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