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Effects of Rootstock on Nutrient Acquisition by Leaf, Kernel and Quality of Pistachio (*Pistacia vera* L.)

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Abstract: A research was conducted at Pistachio Research Institute in Rafsanjan, Iran, to evaluate the effects of *P. vera* L. 'Badami', *P. vera* L. 'Sarakhs' and *P. mutica* F and M. 'Beneh' rootstocks on mineral content of leaves and kernels of 'Owhadi', 'Kalleh-ghuchi' and 'Ahamd-aghaii' pistachio scion cultivars. Twenty year old trees of these combinations were used during study over 2003 and 2004. Leaves of cultivars grafted on 'Beneh' rootstock had higher K, P, Zn and lower Mg and Na content than other rootstocks. Pistachio cultivars growing on 'Badami' rootstock had higher Ca and lower Zn content in the leaves than other rootstocks. 'Ahmad-aghaii' cultivar grafted on 'Sarakhs' rootstock had higher Cu content than those on other rootstocks. Cultivars growing on 'Sarakhs' rootstock had higher Fe and Cu content than other rootstocks. The kernels of cultivars grafted on 'Sarakhs' rootstock had higher K, P, Mg, Cu, Fe and Zn than other rootstocks. The 'Sarakhs' rootstock gave the highest and 'Beneh' rootstock the lowest protein content in kernels. Type of rootstock had no significant effect on crude fat content of kernel. Crude fat content of the kernels of 'Ahmad-aghaii' was higher than other cultivars. It was concluded that, rootstock selection can be a useful management tool in poor soils.

Key words: Pistachio % rootstock % cultivar % nutrient acquisition % kernel quality

INTRODUCTION

Rootstock selection is one of the most important decisions in orchard management. In Kerman province and other pistachio growing areas in Iran, pistachio cultivars are grown on pistachio seedling rootstocks of different cultivars such as 'Badami Zarand' and 'Ghazvini'. Three wild *Pistacia* species, *P. vera* 'Sarakhs', *P. Khinjuk* Stock and *P. mutica* F and M 'Beneh' are grown in Iran. Native *P. vera* forests are located in north eastern part of Iran particularly in Sarakhs region. This native *P. vera* is the origin of cultivated pistachio trees in Iran [1]. *P. mutica* is a wild species indigenous to Iran, growing with almond, oak and other forest trees common to most Alpine regions [2, 3]. *P. khinjuk* is another wild species indigenous to Iran, growing along with *P. mutica* in Alpine area, low altitudes and warmer areas as well [4]. Native pistachio trees can tolerate temperatures between-20 to 45 °C. The main roots grow vertically through the soil 2 to 6 m or more in depth which allow the plant to absorb water deep in the soil during the periods of drought [1]. Native pistachio cultivar 'Sarakhs' is susceptible to *Phytophthora* spp. [5]. *P. mutica* has been selected as a stock resistant to root-knot nematodes [1] and salinity stress [6]. *P. mutica* and *P. khinjuk* are more susceptible to all *Phytophthora* species than *P. vera* cultivars [1].

Pistachio rootstocks differ significantly in their ability to take up nutrients from the soil. Trees grafted on 'Atlantica' rootstock are less likely to show B, Ca or Zn deficiency than other rootstocks with *P. integerrima* parentage [7]. In another study, pistachio 'Bianca' budded onto eight *in vitro*-propagated clonal rootstocks (*P. atlantica* and *P. integerrima*) and one seedling rootstock (*P. terebinthus*). Results showed that there were no particular significant differences in the foliar concentrations of N, P, Fe, Mn and Zn, whereas there were significant differences among rootstocks for Mg and K. Clone 3 of *P. atlantica* was the most efficient in using Mg and *P. terebinthus* in using K [8].

Chemical composition of pistachio nuts may vary depending upon cultivar, rootstock and maturity at harvest and moisture content. The composition of pistachio kernels of various Iranian cultivars from different response was studied. The amount of constituent in 100g kernel were within the following ranges, oil 55.2-60.5% ; protein 15.0-21.2%; carbohydrate 14.9-17.7%; Na 4.0 mg; K 1048-1142 mg; Ca 120-150 mg;

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P 494-514.5 mg; Fe 5.8-11.4 mg; Cu1.0-1.4 mg and Mg 157.5-165.0 mg [9]. In another study, pistachio nuts were collected from large cites in southern parts of turkey. They found 'Owhadi' and 'Halebi' cultivars had the highest K content. 'Halebi' had the highest Mg content. The highest Na was determined in 'Uzun' cultivar. There were significant differences among pistachio nuts regarding fatty acid contents [10].

The use of a given rootstock based on the chemical composition of kernel in pistachio is rare and has not been reported.

The purpose of the present study was to evaluate the influence of rootstock on nutrient acquisition by leaf and kernel and quality of kernel of pistachio cultivars.

MATERIALS AND METHODS

The experiment was conducted in 2003 and 2004 at the Pistachio Research Institute, in a sandy loam, located in Rafsanjan, Iran. The physical and chemical characteristics of the soil were clay 12%, silt 15% and sand 73%, pH 8.1, electrical conductivity (Ec_e) 5.5 ds m^{-1}, N 1-0.2% P_2O_5 9ppm and K_2O 604 ppm. The 20 to 25 year old commercial cultivars, 'Kalleh-ghuchi' 'Owhadi' and 'Ahmad-aghai' were grafted on a variety of rootstock cultivars. Three rootstock were used including: *P. Vera* L. 'Badami', *P. vera* L. 'Sarakhs' and *P. mutica* F and M 'Beneh'. Trees were trained with an open-center system and distance of trees were 4×7 m. A split plot experiment was used in a randomized complete block design with 3 replications. The 3 commercial cultivars (scion) were assigned to main plot and the 3 rootstocks were assigned to subplot within each main plot.

Minerals determination: In both years of study, from six trees of each cultivar per rootstock, samples of 10 leaflets from the mid-section of current year shoots and 400 mature fruits were collected. Leaflets were washed with mild detergent and then rinsed with distilled water, dried in a forced air drying oven at 70°C to constant weight. Leaf tissue was then ground to pass a 40 mesh screen. One g of dried ground leaf sample dry ash at 550°C for 5 h. The ash was then dissolved in 5 ml of 20% HCl. These samples were analyzed for P, K, Mg Ca, Fe, Zn and Cu by atomic absorption spectrophotometer [11].

The pericarp of fruits were removed and kernels were dried in a forced air oven at 70°C for 24 h. Mineral contents of kernels were determined as described for leaf tissues.

Kernel quality: For determination of protein content, nitrogen was determined in 0.5 g dried kernels using the micro-Kjeldahl technique as described by Tandon [11]. The protein was calculated by using the factor of N×6.25.

Samples of dried kernel used for crude fat determination, were ground to a fine powder and the crude fats extracted with ether in a Soxhlet-type extractor. The percentage of ether-extractable fat was determined according to Horwitz [12].

Data analysis: Combined data of two years (2003 and 2004) for mineral contents of leaf and kernel, percentage of kernel protein and crude fat were analyzed using SAS statistical software. Treatment means were compared using least significant differences (LSD, $p = 0.05$). Excel software was used for regression analysis.

RESULTS

Mineral contents of leaf and kernel: The utilization of several different combinations of scion/rootstock allow us to evaluate the nutrient efficiency of the various rootstocks. In the following, rootstocks responses are discussed in relation to each nutrient.

Phosphorous (P): Significant differences in P content of scion leaves were observed (Table 1). Cultivars grafted on 'Beneh' had higher P content than other rootstocks. 'Beneh' was the most effective rootstock in uptaking P in comparison with other rootstocks and it was significantly different at $p = 0.05$ (Table 1). Though P deficiency has not been identified in the cultivars, but P concentration in the leaves of 'Kalleh-ghuchi' cultivar was lower than other cultivars.

The Kernels of scion cultivars grafted on 'Sarakhs' rootstock had higher P content than other rootstocks (Table 2). 'Owhadi' fruits had higher kernel P concentration than other cultivars (Table 2).

Potassium (K): Cultivars grafted on 'Beneh' rootstock had higher K concentration in their leaves than other rootstocks (Table 1). The effect of rootstocks on uptaking K were highly significant at $p = 0.05$. 'Beneh' was the most effective rootstock followed by 'Badami' and 'Sarakhs' (Table 2). 'Kalleh-ghuchi' cultivar growing on three rootstocks had lower but 'Ahamd-aghiai' growing on these rootstocks showed higher K content in leaves (Table 1).

The kernels of cultivars grafted on 'Sarakhs' rootstock had significantly higher K content than other

Table 1: Effects of rootstock on micro and macro nutrients of leaves of three cultivars (Combined data of 2 years 2003 and 2004)

Rootstock	Zn	Fe	Cu	Na	K	P	Mg	Ca
‘Ahmad-aghaii’(Scion)								
‘Badami’	6.56c*	97.86b	7.20b	0.059b	2.13b	0.153b	1.52a	5.44a
‘Sarakhs’	8.32b	128.58a	11.90a	0.083a	1.93b	0.161b	1.09b	4.03b
‘Beneh’	11.49a	118.52ab	8.95b	0.031c	2.69a	0.191a	0.64c	4.47ab
‘Kalleh-ghouchi’(Scion)								
‘Badami’	8.92a	118.75a	5.63a	0.040b	1.74b	0.136b	1.72a	5.62a
‘Sarakhs’	8.28a	133.64a	7.19a	0.064a	1.28c	0.138b	1.25b	3.95b
‘Beneh’	8.95a	89.75b	4.71a	0.016c	2.20a	0.171a	0.83c	3.55b
‘Owhadi’(Scion)								
‘Badami’	6.56b	89.17a	4.23a	0.062a	1.72b	0.153b	1.18a	4.88a
‘Sarakhs’	7.61b	101.93a	4.22a	0.069a	1.51b	0.146b	1.09ab	3.15b
‘Beneh’	10.62a	89.16a	4.22a	0.008b	2.06a	0.193a	0.90b	3.39b

*Mean separation in each column by LSD at 5% level, Micro nutrient: mg/1000 g leaf, Macro nutrient: g/100 g leaf

Table 2: Effects of rootstock on micro and macro nutrients of kernels of three cultivars (Combined data of 2 years 2003 and 2004)

Rootstock	Zn	Fe	Cu	K	P	Mg	Ca
‘Ahmad-aghaii’(Scion)							
‘Badami’	10.79b*	7.29b	3.43b	0.54b	0.11c	0.25b	0.67b
‘Sarakhs’	22.91a	17.07a	7.98a	1.05a	0.38a	0.73a	1.31a
‘Beneh’	15.49ab	9.54b	4.34b	0.66b	0.22b	0.34b	0.39b
‘Kalleh-ghouchi’(Scion)							
‘Badami’	12.19ab	10.44ab	4.73b	0.71b	0.17b	0.32b	0.59b
‘Sarakhs’	20.02a	14.00a	7.41a	1.11a	0.32a	0.70a	1.28a
‘Beneh’	8.34b	7.29b	2.49c	0.70b	0.14b	0.25b	0.67b
‘Owhadi’(Scion)							
‘Badami’	12.33b	8.34b	3.01b	0.70b	0.15b	0.24c	0.81b
‘Sarakhs’	35.82a	25.89a	9.97a	1.33a	0.46a	0.82a	1.27a
‘Beneh’	14.64b	9.04b	4.14b	0.71b	0.20b	0.45b	0.64b

*Mean separation in each column by LSD at 5% level, Micro nutrient: mg/1000 g kernel, Macro nutrient: g/100 g kernel

rootstocks (Table 2), Therefore, rootstock had significant effect on kernel K content.

Magnesium (Mg): Significant differences in Mg concentration of scion leaves were observed (Table 1). Cultivars growing on ‘Beneh’ had lower Mg content than other growing on ‘Sarakhs’ and ‘Badami’ rootstocks. Cultivars growing on ‘Badami’ had higher Mg content than other rootstocks and it was significant at p = 0.05 (Table 1). The rootstock ‘Beneh’ had the lowest and the rootstock ‘Badami’ had the highest Mg uptake in this study (Table 1).

There was significant effect of rootstock on concentration of kernel. ‘Sarakhs’ rootstock significantly increased concentration of Mg in the kernels of scion cultivars (Table 2).

Calcium (Ca): Significant differences in Ca content of scion leaves were observed (Table 1). Pistachio cultivars growing on ‘Badami’ rootstock had higher Ca content in leaves than other rootstocks (Table 1). The rootstock ‘Badami’ took up more Ca than other rootstocks and this effect was significant at p = 0.05 (Table 1).

The concentration of Ca in kernel significantly was affected by rootstock. The rootstock ‘Sarakhs’ significantly increased Ca content of kernels of cultivars than other rootstocks (Table 2).

Sodium (Na): Cultivars growing on ‘Beneh’ rootstock had lower Na content in leaves than other rootstocks and this effect was highly significant at p = 0.05 (Table 1). Leaves of ‘Kalleh-ghuchi’ cultivar in comparison with other cultivars had lower Na content (Table 1).

Table 3: Effects of rootstock on protein % and fat % of kernel of three pistachio cultivars. (Combined data of 2 years 2003 and 2004)

	'Ahmad-aghaii'(Scion)		'Kalleh-ghouchi'(Scion)		'Owhadi'(Scion)	
Rootstock	% protein	% fat	% protein	% fat	% protein	% fat
'Badami'	19.50b	63.91a	21.85ab	56.25a	21.41b	55.02b
'Sarakhs'	27.51a	64.16a	26.63a	58.81a	28.74a	62.06a
'Beneh'	20.06b	64.24a	19.97b	58.01a	18.30b	59.76ab

*Mean separation in each column by LSD at 5% level

Copper (Cu): our data showed that 'Ahmad-aghai' cultivar had higher Cu content than other cultivars. Leaves of this cultivar are growing on 'Sarakhs' rootstock had higher Cu content than other rootstocks. The rootstock 'Badami' took up lower Cu than other rootstocks (Table 1).

The rootstock 'Sarakhs' significantly increased Cu content of kernels. Kernels of 'Owhadi' cultivar was grafted on 'Sarakhs' rootstock took up more Cu than other rootstocks (Table 2).

Iron (Fe): Leaves of 'Ahmad-aghai' and 'Kaleh-ghuchi' had lower Fe content than 'Owhadi' cultivar (Table 1). Cultivars growing on 'Sarakhs' rootstock had higher Fe content than other rootstocks (Table 1).

The rootstock 'Sarakhs' was found to be efficient in increasing Fe content of kernels of cultivars (Table 2). Kernels of 'Owhadi' cultivar is growing on 'Sarakhs' rootstock had higher Fe content than other cultivars.

Zinc (Zn): Our data showed that Zn content of leaves of cultivars of 'Ahamd-aghaii' and 'Owhadi' growing on 'Beneh' rootstock significantly was higher than other rootstocks (Table 1). Leaves of cultivars grafted on 'Badami' rootstock had lower Zn concentration than other rootstocks.

The kernels of cultivars growing on 'Sarakhs' rootstock had higher Zn content than other rootstocks and this effect was significant at $p = 0.05$ (Table 2). Kernels of 'Owhadi' cultivar was grafted on 'Sarakhs' rootstock had the highest Zn content. Cultivars growing on 'Badami' rootstock had the lowest Zn content in kernels (Table 2).

Protein and crude fat: The present study showed that type of pistachio rootstock has significant effect on protein and crude fat content of kernels. The rootstock 'Sarakhs' gave the highest and 'Beneh' rootstock the lowest protein content in kernels (Table 3). Crude fat of the kernels was not significantly affected by rootstock (Table 3). However, the kernels of 'Owhadi' cultivar grafted on 'Sarakhs' in comparison with 'Badami' rootstock had significantly higher crude fat content. Crude fat of kernels was between 63.91 to 64.22%, 56.25 to 58.81% and 55 to 62% in 'Ahmad-aghai', 'Kelleh-ghuchi' and 'Owhadi', respectively (Table 3).

DISCUSSION

According to the extensive pistachio growing in Iran, soil salinity is a serious problem and deficiencies of micronutrients are widespread. Soils in which pistachio is grown is characterized by high pH, carbonate content and low organic matter. In these soils, deficiencies of Zn, Fe and Cu, can become severe, resulting in the development of delayed bud break, impaired growth and meristematic regions and significant yield loss [13]. Because of high complexing capacity of the leaf waxes in pistachio, the crop is notoriously inefficient at the absorption of foliar applied micronutrients [14]. Research on multiple scion-rootstock combinations has shown that the effect of salinity on growth and yield is usually determined more by rootstock than by the scion [15, 16]. Like citrus rootstocks, pistachio rootstock species differ greatly in their water relations and their ability to transport mineral nutrients [17]. The importance of the rootstock in regulating plant water relation and transport was discussed by Sinclair [18], who noted that trees on rough lemon and *P. trifoliate* had higher transpiration rates than those on 'Cleopatra' mandarin and sour orange rootstock. Therefore, water relations and mineral nutrients uptake and transport could also be altered in rootstock × scion combinations, in both normal and saline condition [17]. The rootstock 'Sarakhs' was found the most effective in terms of enhancing leaf concentration for the Cu and Fe nutrients. The nutritional efficiency of this rootstock was constant during this study for all scion cultivars.

Specific mechanism may exist for Cu and Fe uptake similar to those reported by Zhang *et al*. [19]. Idem and Gezerel [20] reported that *P. vera* L. cv Siirt and *P. Khinjuk* seedling took up more Fe and Cu than other rootstocks. They found also more micronutrients Fe and

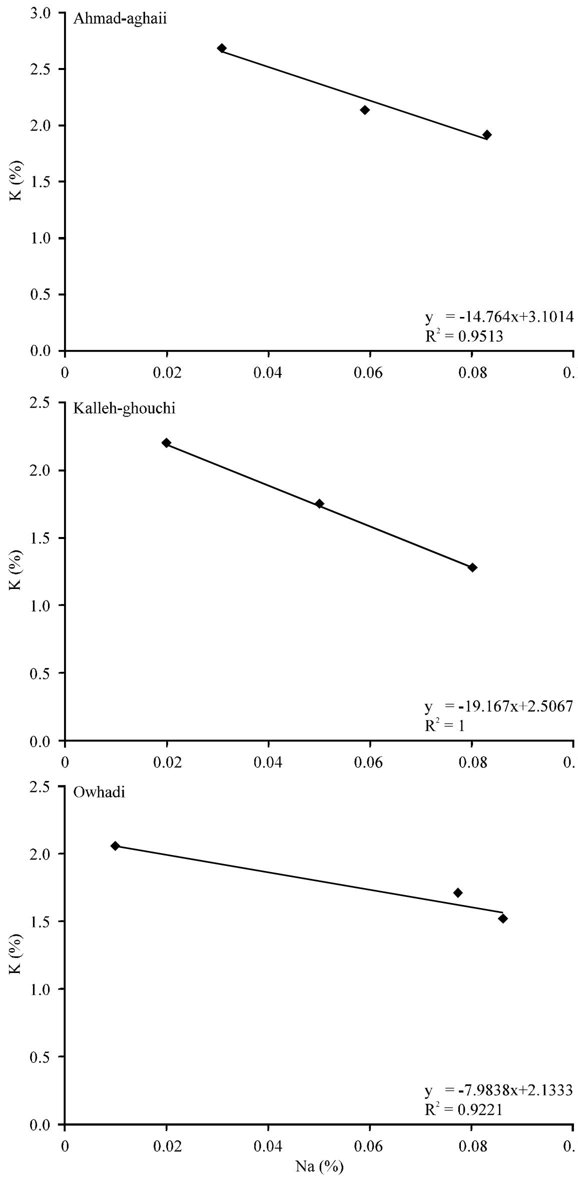

Fig. 1: Relationship between K and Na% in leaves of pistachio cultivars on three rootstocks

Cu in stems than leaves. The threshold Zn level of pistachio leaves for deficiency appears to be 7-25 ppm in mid summer [21]. In this study the Zn concentration in leaves of 3 cultivars grafted on 'Beneh' rootstock was between 9-11 ppm. The concentration of Cu in leaves of 3 cultivars grafted on 3 rootstocks was not below the threshold level, therefore, no deficiencies of Zn and Cu were observed in this study [21]. The rootstock 'Beneh' was found to be a very efficient rootstock for alkaline and dry soil [6]. The low concentration of Na in the leaves of cultivars on 'Beneh' rootstock suggests that the mechanism of Na exclusion by rootstock is similar to that of *Poncirus trifoliata* (a citrus relative) which restricts Na transport to shoots by retaining it in the main root and basal stem [22]. We did not harvest basal stem separately from the remaining stem and hence it is difficult to assess whether there was any significant accumulation of Na in the basal stem of cultivars on different rootstocks. Walker *et al.* [23] showed that *P. vera* appears to be a better Na excluder than *P. atlantica*, which in turn appears to be better than *P. terebinthus*. The leaf Zn content of the cultivars grafted on it was high. The rootstock 'Beneh' was found to be efficient in absorption lower Na and higher K than other rootstocks. Also, the correlation between K and Na was highly significant ('Ahmad-aghai' r = 0.97, 'Kalleh-ghuchii' r = 1, 'Owhadi' r = 0.96), (Fig. 1). Walker *et al.* [23] suggest that rates of uptake and root to shoot transport of K were at most only marginally different between the species of *P atlantica* and *P. vera*. On the other hand, the ratio of Na:K in leaves and stems was markedly different between these rootstocks. Therefore the deficiencies of Fe, Zn and Cu were not observed in this study [21]. Precise critical nutrient values for pistachio have not been established in Iran, but according to the mineral elements ranges in pistachio leaves exhibiting normal growth [21], the P and Mg levels in the leaves of cultivars grafted on 3 rootstocks were below the threshold level. As the pH of the soil was 8.1, the level of Ca in the leaves of cultivars was higher than the threshold level.

Pistachio nut is rich in protein, fat and minerals and is an excellent source of P, K, Mg, Ca and Fe [7]. There are a few reports dealing with chemical composition in relation to cultivar and stage of maturation. However, no detailed study has so far been reported about the effect of rootstock on chemical composition of the pistachio. These results confirm that rootstocks can have an important effect on kernel quality of pistachio. Mineral contents of pistachio kernels were highly affected by 'Sarakhs' rootstock. This rootstock significantly increased K, P, Mg, Ca, Zn, Fe and Cu contents of pistachio kernels. The present study also showed that 'Ahmad-aghai' can produce kernels with higher macro and micro nutrients content. The present study also indicated that cultivar and rootstock selection is important in pistachio production. Chloride accumulation in shoots is rootstock-dependent [24]. Salinity can affect on root and leaf K, Ca and Mg concentrations. Reductions in these nutrients in roots and leaves with increasing salinity have been observed in *Citrus* [25]. Trees with one or two grafts (rootstock/scion) have higher Cl exclusion than non-grafted trees and can improve Ca, K and Mg absorption. High root and leaf Ca and K concentrations ameliorate the negative effect of salinity on root, shoot and fruit growth [26]. Crude fat of pistachio is the main constituents of pistachio, generally exceeding 55% in Iranian origin (based of dry weight), [9]. In the present study, it was found that type of rootstocks increased crude fat of kernel up to 64% in kernel. 'Sarakhs' had better performance in increasing crude fat in Kernel. Our data are in agreement with finding on almond [27]. Protein content ranges form 15 to 19% in Iranian pistachio origin [9]. Our data showed that protein content in kernel was between 19 to 27%. The kernel of 'Owhadi' grafted on 'Sarakhs' increased protein content up to 28.74%. Pistachio kernels with the highest oil content were found to be the highest in protein too. This data is not in agreement with those reported by Schirra [27] on almond.

CONCLUSIONS

These data show that type of rootstock has an important effect on mineral content of leaves and kernels in pistachio. Positive effects of rootstocks were found in increasing crude fat and protein contents of kernels.

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