World Applied Sciences Journal 5 (1): 105-110, 2008
ISSN 1818-4952

Studying Some Compounds Existing in Pistachio Fruits and the Effect of Pollen Grains of Different Male Genotypes on the Changes in Their Quantity

[1]*Hossein Afshari,* [2]*Ali Tajabadipour,* [3]*Mehdi Mohamadi Moghadam,*
[2]*Hossein Hokmabadi and* [1]*Ghanbar Laee*

[1]Department of Horticulture, Islamic Azad University, Damghan Branch, Damghan , Iran
[2]Pistachio Research Institute, P.O. Box: 77175-435, Rafsanjan, Iran
[3]Pistachio Research Institute Damehan Station, Damghan , Iran

Abstract: To study the effects of xenia, methaxenia, lipid and fatty acid rates and elements on pistachio, 4 male genotypes from among all male genotypes provided by Rafsanjan Pistachio Research Institute were selected. The flowering period of the chosen genotypes was the same as 3 female cultivars of Kalleghoochi, Ohadi, Ahmadaghaii. By using completely randomized blocks design we conducted various experiments during the years 2006 and 2007. The highest percentage of fat was found in the kernels of Ahmadaghaii (52.92%) and the lowest in Kalleghoochi (45.48%). From among different factors and their interacting effects, only the effect of pollen and cultivar type on the fat content of the fruit was significant only at 5% level. Thus R27 and N16 pollen produced the highest fat content in the kernels of Ahmadaghaii (58.7 and 57.7%). The most common simple unsaturated fatty acid found in the kernels of all cultivars was oleic acid and the most important compound unsaturated fatty acid in fruit kernels was linoleic acid. Pollen type was only effective on the amount of meristoleic and palmitoleic fatty acids. The amount of macro and micro elements measured in kernels and hulls of 3 pistachio cultivars had a significant difference and the interactive effect of pollen and cultivar type affected the content of nitrogen, phosphorus, potassium, iron and boron in fruit kernels at 1% level.

Key words: Ahmadaghaii · Fatty acid · Kalleghoochii · Methaxenia · Ohadi · Xenia

INTRODUCTION

Pistachio is a very significant and critical commercial tree in Iran. Pistachio kernel is rich with fats, fatty acids and elements such as calcium, magnesium, potassium and vitamins like B2, B1, B, A and B6 [1]. Garcia *et al.* [2] studied the kernels of some pistachio cultivars and concluded that the amount of protein found in the samples was 25-31% and total fats were 40-53%. Agar *et al.* [3] conducted research on the effects of environmental elements on the fat and fatty acid content of pistachio cultivars in Turkey. They concluded that the fatty acid compounds of one cultivar differ in various climatic conditions [3]. Compound unsaturated fatty acids are quite useful in human diets for they are effective in cell preservation and blood pressure control. Choosing pistachios with high oleic acid and lower linoleic acid will bring higher value to the diet, they are also more preservable. Compound unsaturated fatty acids are more sensitive to oxidative changes due to their extra compound bounds [1]. In a study conducted in Iran, Shokraii and Hosseini [4] compared the contents of Ohadi cultivar of Kerman Province with other cultivars of Iran. The general minerals of this cultivar were like others except for higher calcium, sodium and magnesium found in it which was not reported before. Comparison of organic materials brought great similarities yet fatty acid compounds found in pistachio fat were quite diverse. Although amino acids were similar to some extent, argenine, lysine and cytosine levels were found to be higher than other studies. Roozban *et al.* [5] studied the amount of fat and fatty acid contents of 4 Iranian cultivars in Qazvin city. The quality of pistachio is affected by fatty acids most of which are oleic and linoleic acids. Extraction of fat was done by n-hexane and Soxhlet method and fatty acid contents of the fat were determined by gas chromatography. Based on obtained results the best cultivar for Qazvin climate in terms of nutrition and preservation was the Qazvini cultivar [5]. Garcia *et al.* [6] added that there was a contrasting relationship between linoleic and oleic acid in pistachio, which is observed in other fruits as well. Therefore, if a cultivar has high

Corresponding Author: Hossein Afshari, Department of Horticulture, Islamic Azad University, Damghan Branch, Damghan , Iran

amounts of linoleic acid its oleic acid content will be low vice versa. The goal of the present study is to determine the amount of macro and micro elements, fats and fatty acids in the fruits of three pistachio cultivars and to study the effects of different male pollen genotypes in their quantities.

MATERIALS AND METHODS

This study was conducted in 2006 and 2007 on three commercial female pistachio cultivars including Kalleghoochi, Ohadi and Ahmadaghaii with 4 pollen seeds of R27, R28, N2 and N16 genotypes. Eight similar trees of each cultivar were selected, marked during sprout swelling phase and then from 6 of 7 branches each having 3 sprouts were isolated in bags. Branches having male flower grapes were separated when the flowers were not fully open. They were then taken into the lab and the branch ends were put inside water containers. Once the Anther opened the male flowers of different male genotypes were shaken on white cloths inside 4 separate rooms; then pollen seeds were passed through fine sieves. Hands and instruments were washed by 70% alcohol. Pollen were pollinated only once (flowers opened 70%) by separate brushes. Seven marked branches were used for this task: 4 branches were used for pollination with R27, R28, N2 and N16 genotypes; one branch was used for pollination with a combination of 4 pollen types; one branch was used for free pollination (control) and the last branch remained without pollination. At harvest time 10 fruits of each branch were selected and used for quantitative and qualitative tests.

Soxhlet Method for Measuring Fats of Pistachio Kernels: For determination of oil content, 50 g kernels of each sample were ground and extracted in Soxhlet apparatus with petroleum ether for 4 hr. The residue was ground to a fine powder and extracted once more overnight. The extracts were combined and solvent was removed by Biichi vacuum distillation apparatus at low temperatures. The remainder was dried at 103 + 2 C for 20 min, cooled and weighed [2,7,8].

Extraction and Preparation of Fatty Acids: The moisture contents of the kernels were determined using a volumetric water determination apparatus. All the yields were calculated on a dry weight basis. Kernel oils were obtained in petroleum ether for 6 h by Soxhlet extraction. The solvent was evaporated under reduced pressure using a rotary evaporator at 40°C and the residue refluxed with 0.5 N sodium hydroxide solution in methanol (5 ml) for 10 min. After adding 5 ml of 14% BF3-MeOH solution by a pipette through the condenser, the mixture was boiled for 2 min. Then 5 ml of heptane was added through the condenser and boiled one more minute. After cooling the mixture, 5 mL of saturated NaCl solution was added and the flask was rotated very gently and the required methyl esters were extracted with heptane (2´5 ml); then the organic layer was separated using Pasteur pipettes and dried over anhydrous Na_2SO_4. The fatty acid methyl esters were recovered after solvent evaporation [9].

Gas Chromatography/Mass Spectrometry (GC/MS): The fatty acid composition of the kernel oils was determined by GC/MS. A Shimadzu GCMS-QP5050A system with a CPSil 5CB column (25 m´ 0.25 mm film thickness) was used with helium as a carrier gas. The GC oven temperature was kept at 60°C and programmed to 260°C at a rate of 5°C/min, then kept constant at 260°C for 40 min. Split flow was adjusted at 50 ml/min. The injector temperature was at 250°C. MS were taken at 70 eV. Mass range was between *m/z* 30 to 425. Identification of individual components was achived using library search software from The Wiley/NBS Registry Mass Spectral Data and in-house "BASER Library of Fatty Acid Constituents [10, 7].

Determining Abundant and Rate Elements in Pistachio Kernels and Green Hulls: In order to determine abundant and rate elements, nitrogen content were determined using micro-Kjeldhal digestion method, the phosphorus was analyzed by flame spectrophotometer method and potassium by the flame photometric method. The calcium and magnesium were determined by titrimetric method [11]. The experimental design for the chemical analyses was a Split – split - plot. in completely randomized blocks design with three replications and the results were analyzed based on Duncan group by SAS software.

RESULTS AND DISCUSSION

-Studying the Effect of Pollen Type of Different Male Genotypes on the Rate of Formation of Lipid and Fatty Acids in the Kernels of 3 Pistachio Cultivars in 2006 and 2007: Based on data obtained from the variance analysis of tale, factors such as experiment year, pollen type, interaction of year and pollen type and interaction of year, variety and pollen type did not have a significant effect on the lipid percent at 5% level. Yet factors such as pollen type at 1% level and interaction of variety and pollen type at 5% level had significant effects on the lipid percent of pistachio kernels. Maximum lipid percent in pistachio

Table 1: Studying the Fatty Acid Content in Kernels of 3 Cultivars of Pistachio

Fatty Acid				
Cultivar	Linoleic Acid	Palmitic Acid	Palmitoleic Acid	Arachidic Acid
Ahmadaghaii	35b	11.8a	1.1a	2b
Ohadi	45a	12.3a	0c	6a
Kalleghoochi	41ab	B7.4	5b	2b

The figures in the columns have been compared together. Numbers having similar letters lack a significance difference. α=5%

Table 2: Studying the Effect of Pollen Seeds of Different Male Genotypes on the Lipid Percent in Kernels of 3 Different Varieties

Cultivar	N16	N2	R28	R27	Control
Ahmadaghaii	57.7a	Bcd50.2	47.5de	58.7a	47ef
Ohadi	48.4cde	52b	52b	47ef	46.4g
Kalleghoochi	46.4g	49h	46.9fg	46.3g	51.1bc

The figures in the columns have been compared together. Numbers having similar letters lack a significance difference. α=5%

kernels was observed in Ahmadaghaii (52.92%) and its minimum was observed in kernels of Kalleghoochi (45.48%). Seeds of R27 and N16 genotype produced the highest lipid rate in the kernels of Ahmadaghaii (58.7% and 57.7%) (Table 1). But the minimum lipid percent in the kernels of tested fruits were in Kalleghoochi cultivars pollinated by N2 genotype pollens (39%) (Table2). Based on Table 1 the percent of fatty acids, linoleic acid, palmitoleic, palmitic acid and arachidic acid in the kernels of three cultivars tested had a significant statistical difference at 5% level. The type of pollen used in pollination at 5% level caused a change in meristoleic and palmitoleic fatty acids. The interactions of pollen type and variety on meristoleic and palmitoleic fatty acids were significant at a 5% level. During both research years the lowest percent of lipid in pistachio kernels was observed in Kalleghoochi. Agar *et al.* [1] found the same in their studies on the lipid content of various cultivars. The effect of pollen type on the fruit lipid content accords with the research activities of AK and Kaska [12]. They studied pollen seeds of different cultivars and demonstrated that the domestic pistachio variety caused the highest rate of lipid content in kernels of pollinated fruits. Though Agar *et al.* [3] believed fruit lipid content is not affected by environmental conditions and is controlled genetically; there is evidence of the effect of temperature on the fruit lipid content. Letchworth *et al.* [13] demonstrated that the pollen seeds of corn species having high lipid contens, cause an increased lipid percent in the products of such pollens. Also Lambert *et al.* [14] used the pollens of hybrid corn varieties having high lipid contents and produced seeds with high lipid and protein rates in which starch content and seed size decreased. Existence of different volatile substances, isoprenoids, lipids, fatty acids, benzoids, etc. in the pollen walls of different plants may be one of the reasons behind the effect of pollen seeds on the lipid content of the fruits [15].

The highest rate of fatty acid in pistachio kernels was found in Kalleghoochi and Ahmadaghaii varieties was as follows: oleic acid (40.8 and 40.5%), linoleic acid (40.6 and 34.9%), palmitic acid (11.8 and 7.3%), stearic acid (1.7 and 2.2%), meristoleic acid (1 and 1.1%), Eicosonoic acid (0.7 and 0.9%), palmitoleic acid (0.5 and 0.7%), arachidic acid (0.20 and 0.24%). Yet in Ohadi cultivar the highest fatty acid found in kernels was linoleic acid (46.45%) and then oleic acid (30.8%), palmitic acid (12.3%), meristic acid (5.3%), stearic acid (2.7%), meristoleic acid (1.1%) icosonoic acid (0.8%) and arachidic acid (0.56%). Unlike the previous two cultivars no palmitoleic acid was found in the lipids produced from Ohadi fruits. The most common simple unsaturated fatty acid of all kernels was oleic acid and the most important compound unsaturated fatty acid of all fruit kernels of 3 cultivars being tested was linoleic acid, which accorded with the results of many other studies [5,16,17,3]. Unsaturated fatty acids of pistachio kernels enhance its nutritional qualities yet increases the self-oxidant feature of this cultivar as well [5] such that kernels having higher amounts of oleic acid and lower amounts of linoleic acid have higher nutritional values and may be kept for longer periods [6].

In the present study the highest amount of linoleic acid was observed in the kernels of Ohadi cultivar (46.4%). Roozban *et al.* [5] found the highest amount of linoleic acid in the kernels of Ohadi and Kalleghoochi cultivars. The highest rate of saturated fatty acid determined for all cultivars was palmitic acid, meristic acid and stearic acid which accorded with the studies of some other researchers [4, 8, 17]. In his studies on tropical trees, Wallace [18] demonstrated that pollen type was capable of change the quality of fruits of macadamia and Citrus [7]. Also Weingarhner *et al.* [19] reported the effects of corn pollen seed on the quality of produced fruits. Agar *et al.* [1] showed that the total of unsaturated fatty acids was always above 80%, which is probably because the total of saturated fatty acids is always low (12%). The reason behind the effect of pollen seed on the percent of fatty acids in fruit kernels, like the effect on the percentage of lipids, may be the fatty acid compounds found in the pollen seed walls [20]. Therefore pollen seeds

Table 3: Studying the Rate of Macro and Micro Elements inGreen Hulls and Fruits of 3 Pistachio Cultivars

Cultivar		B (ppm)	P (%)	K (%)	Ca (%)	Fe (ppm)	Zn (ppm)	N (%)	Mg (%)
Ahmadaghaii									
Kernel	Ohadi	48c	35c	1c	4b	43b	-	-	19b
	Kalleghoochi	50b	47a	1.6a	6a	43b	-	-	24a
	Ahmadaghaii	53a	45a	1.2b	3b	49a	-	-	24a
Hull	Ohadi	340a	2b	7.2a	1.6a	343a	14.6a	3.4a	-
	Kalleghoochi	245b	24a	7.2a	7c	250b	13.2b	2.8b	-
		241b	2b	6.5b	7c	244b	13.7b	3.4a	-

The figures in the columns related to kernel and hull have been compared separately. Numbers having similar letters lack a significance difference. α=5%

may directly influence the biosynthesis of lipids to cause changes in lipids and fatty acids. However, it has been shown that environmental conditions influence lipid percentage. Satil *et al.* [7] demonstrated that temperatures above 25ºC cause a reduction in the amount of palmitic acid and some other fatty acids in pistachio kernels. Thus lower temperatures in some regions are a cause of increase in the amount of some saturated fatty acids in kernels of pistachio varieties [9]. Also an enzyme named oleic desatorase has been recognized responsible for the change in the formulations of saturated fatty acids of different pistachio varieties [3]. This may well be another reason behind the effect of pollen seeds on the changes in the rates of pistachio kernel compounds.

-Studying the Effect of Pollen Type from Different Male Genotypes on Macro and Micro Elements in the Hull and Kernel of Pistachio: According to the data obtained from variance analysis elements such as bore and zinc in the hulls of three cultivars being studied in the 2 consecutive years had significant statistical differences at 1% level, while iron was different at 5%. Elements such as phosphorus, potassium, calcium, boron, iron and zinc each had a significant different in the hulls of various cultivars at 1% level. The interaction of year and variety on the elements of boron and zinc was significant at 1% and that of year, variety and pollen type on bore was significant at 5% level.

The amount of bore found in the hulls of three various cultivars was 289.7 ppm in the second year and 272.6 ppm in the first. Iron was 53.24 ppm and 50.33 ppm in the second and first years, yet the amount of zinc was higher in the first year (14.46 ppm) than the second (13.53 ppm).

However, according to Table 3 the highest amount of phosphorus (0.24%) was found in Kalleghoochi as compared to Ahmadaghaii (0.20%) and Ohadi (0.20%) varieties. Kalleghoochi and Ohadi varieties had the highest amount of potassium in their hulls (7.25 and 7.23%) as compared to Ohadi (6.5%). The calcium content of the hulls of Ahmadaghaii (1.74%) was higher than Kalleghoochi (0.73%) and Ohadi (0.69%). Highest boron and zinc content was found in hulls of Ahmadaghaii (351.6 and 14.78 ppm), while Kalleghoochi (249.96 and 13.46 ppm) and Ohadi (242 and 13.74 ppm) had lower amounts. Iron was found more in Kalleghoochi hulls (58 ppm) than Ohadi (49.1 ppm) and Ahmadaghaii (48.26 ppm). In connection with the interactions of experiment year and pistachio cultivar the highest amount of boron was found in the hulls of Ahmadaghaii for the first and second years (353 ppm) and the lowest bore content was seen in the first year of the experiment in Kalleghoochi cultivar. The highest amount of zinc was found in green hulls of Ahmadaghaii in the first year (15.6 ppm) and its lowest rate was seen in green hulls of Kalleghoochi fruits (12.9 ppm).

The interactions of variety, pollen type and experiment year slightly influenced the bore element of green hulls in such a way that the highest bore content in the first and second years of experiment (358 and 359 ppm) was produced by R28 pollen seeds applied on Ahmadaghaii, while the lowest was produced by R27 pollen seeds applied in the first year on the green hulls of Kalleghoochi fruits (141.33 ppm).

According to Table 4 elements such as nitrogen, phosphorus, potassium, calcium, magnesium, iron, boron had a significant statistical difference among the tree pistachio cultivars at a 1% level.

Different pollen seeds being tested could affect the elemental contents (phosphorus, potassium, iron, boron) of the fruits of 3 cultivars being studied at a 1% level. The interaction of pollen type and cultivar on elements such as nitrogen, phosphorus, potassium, iron and bore was significant at 1% level. Based on Table 3 the highest rate of nitrogen was seen in kernels of Ohadi (3.34%) and Ahmadaghaii (3.32%) while the lowest rate was seen in Kalleghoochi.

Table 4: Studying the Effect of Pollen Seeds of Different Male Genotypes on the Elements of Pistachio Kernels

Element					
Pollen	B (ppm)	Fe (ppm)	Ca (%)	K (%)	P (%)
Control	50bc	48a	36b	1.15c	45a
R27	52b	47a	52a	1.4a	43b
R28	54a	37b	52a	1.3b	43b
N2	48c	40b	51a	1.18b	42b
N16	51bc	46a	50a	1.18b	42b

The figures in the columns have been compared together. Numbers having similar letters lack a significance difference. α=5%

Elements such as phosphorus, potassium and calcium are more abundant in kernels of Kalleghoochi pistachio and the kernels of Ohadi (0.46, 1.21 and 0.40%) and Ahmadaghaii (0.35, 0.98, 0.44%) have lower amounts of such elements. The highest rate of magnesium and iron were found in the kernels of Ohadi (0.24%, 49 ppm), then Ahmadaghaii (0.19%, 42.40 ppm) and finally Kalleghoochi (0.24%, 42.20 ppm) (Table 3). The highest amount of boron was seen in the kernels of Ohadi (53.86 ppm), then Kalleghoochi (51 ppm) and then Ahmadaghaii (47.86 ppm). Fruits produced by free pollination (control) had the highest phosphor content in the kernels (0.45%) as compared to other fruits produced through pollination with other pollen seeds (Table 3).

Pollen seeds of R27 male genotypes produced fruits with maximum potassium in kernels (1.42%) while fruits produced by free pollination had minimum potassium content in their kernels (1.12%) (Table4). Kernels of fruits produced by R28, N2, R27, N16 male genotypes had higher calcium content in comparison to kernels of control fruits (0.36%) (Table4). Different pollen of had no significant effect on the nitrogen content of the kernels of various pistachio.

Kernels of fruits pollinated with N16, control and R27 pollen had higher iron content (48.44, 47.66 and 46.77 ppm) in comparison to kernels of fruits pollinated by N2 (41.33 ppm) and R28 (38.66 ppm) (Table 4). Maximum boron content was observed in the kernels of fruits pollinated with R28 (53.66 ppm) and the lowest boron content was seen in the kernels of fruits pollinated with N2 (42.22 ppm). Interactions of pollen type and 3 varieties of pistachio could influence the elements of nitrogen, phosphorus, potassium, iron and boron in fruit kernels. That pollen seeds had no effect on elements in the green hulls of pistachio varieties may be due to the fact that pistachio green hulls mostly inherit their properties from their female base. The elemental contents of the three pistachio cultivars used in our study accord with those reported by Kamangar *et al.* [21] and Caruso *et al.* [22]. They reported that accumulation of elements in kernels happens during the embryo growth period [22, 21]. According to previous experiments of our study (data not presented here) the rate of elements such as iron (N16, R27) and bore (R28) in pollen seeds shows a positive relationship with the rates of elements detected in the kernels of fruits produced by the same pollen seeds, yet this relationship is not seen in the rate of other elements found in the pollen seeds and fruit kernels. As few studies have been conducted on the role of pollen seeds of different cultivars on the elements found in produced fruits, the need for further research is quite obvious.

REFERENCES

1. Agar, I.T., S. Kafkas and N. Kaska, 1998. Lipid characteristics of Turkish and Iranian pistachio kernels. Acta Horticulturae, 470: 378-384.
2. Erturk, U., C. Mert and A. Soylu, 2006. Chemical composition of fruits of some important chestnut cultivars. Braz. Arch. Biol., 49: 2.
3. Agar. I.T., S. Kaska. and N. Kafkas, 1995. Characterization of lipids in pistacia species in Turkey. Acta Horticulturae, 419: 417-422.
4. Shokraii, H. and E. Hosseini, 1997. Chemical composition of the pistachio nuts of Kerman, Iran. Journal of Food Science, 42(1): 244-245.
5. Roozban, M.R., N. Mohamadi and K. Vahdati, 2005. Fat content and fatty acid composition of four Iranian pistachio varieties grown in Iran. IV international symposium on pistachios and almonds, Tehran, Iran.
6. Garcia, J.M., I.T. Agar and J. Streif, 1994. Lipid characteristics of kernels from different hazelnut cultivars. J. Agricul. and Forestry, 18: 199-202.
7. Taheri, M. and A.R. Talaie, 2001. The effect of chemical spray on the qualitative and quantitative characteristics of "Zard" olive fruits. Acta Horticulturae, 564. No. 57.
8. Yesim, O., 2002. The comparison of some pistachio cultivars regarding their fat, fatty acids and protein content. Gartenbauwissenschaft, 67(3): 107-113.
9. Satil, F., A. Azcan. and K.H.C. Baser, 2003. Fatty acid composition of pistachio nuts in Turkey. J. Chem. Natural Compounds, 39(4): 322-324.
10. Pearson, T.C., D.C. Slaughter and H.E. Studer, 1996. Hull adhesion characteristics of early-split and normal pistachio nuts. J. Appl. Engineering in Agricul., 12(2): 219-221.

11. Tekin, H., F. Akkok., C. Kuru and G. Genc, 1995. Determination of nutrient content of *Pistacia vera L*. and assessment of the most suitable leaf collection time. Acta Horticulturae, 419: 64-69.
12. Ak, B.E. and N. Kaska, 1998. Determination of viability and germination rates of pistacia spp. Pollen kept for artificial pollination. Acta Horticulturae, 470: 300-306.
13. Letchworth, M.B. and R.J. Lambert, 1998. Pollen parent effects on oil, protein and starch concentration in maize kernels. Plant physiology and biochemistry, 38(2): 363-367.
14. Lambert, R.G. and D.E. Alexander, 1998. A High oil pollinator enhancement of kernel oil and effects on grain yields of maize hybrids. American Society of Agronomy, 90: 211-215.
15. Anttonen, M., Ornan and B. Sener, 2002. Composition of the seed fats of *Pistacia vera L* . of different origins respect to fatty acids. Int. J. Food Sci. Technol., 37(3): 331.
16. Kuchukoner, E. and B. Yurt, 2003. Some chemical characteristics of *Pistacia vera* varieties produced in turkey. European Food Res. Technol., 217: 308-310.
17. Garcia, J.M., I.T. Agar and J. Streif, 1992. Fat content and fatty acid composition in individual seeds of pistachio varieties grown in turkey. Gartenbauwissenschaft. 57(3): 130-133.
18. Wallace, H.M., 2003. Genetic and environmental control of quality in subtropical fruit and nut crops. XXVIth International horticulture symposium on citrus and other subtropical and tropical fruit crops, pp: 1120-1140.
19. Weingarthner, U., O. Kaeser, M. Long and P. Stamp, 2002. Combining cytoplasmic male sterility and xenia increase grain yield of maize hybrids. Crop Science, 42: 1848-1856.
20. Edlund, A., R. Swanson and D. Preuss, 2004. Pollen and stigma structure: the role of diversity in pollination. J. Plant Cell., 16: 84-97.
21. Kamangar, T., F. Farrohi and M. Mehran, 1975. Characteristics of pistachio kernel fats from Iranian cultivars. J. Am. Fat Chemist's Soci., 53(12): 512-513.
22. Caruso, T., F. Sottile and A. Motisi, 1995. Growth analysis and mineral content in pistachio (*Pistacia vera* L) infructescence and its components. Acta Horticulturae, 419: 103-108.