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Full Length Research Paper

Determining the chilling requirements of four Pistachio cultivars in Semnan province (Iran)

Hossein Afshari[1], Ali Tajabadipour[2], Hossein Hokmabadi[2] and Mehdi Mohamadi moghadam[2]

[1]1Horticulture Department, Islamic Azad University, Damghan Branch, Iran.
[2] Scientific board of Iran's Pistachio Research Institute, P. O. Box: 77175-435, Rafsanjan Iran

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Most deciduous trees enter dormancy in respect to the shortening day length in fall; dormant buds require a period of chilling to break this rest. The present research was carried out under laboratory conditions in order to determine the chilling requirements of 4 local cultivars of Damghan city. In this experiment some traits of flower bud breaking of mentioned cultivars in different chilling times (500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, and 1400 h) were studied. So an experiment as factorial in completely randomized design established in two levels of cultivar and chilling. Results indicated that the chilling requirements of Akbari cultivar were higher than others. This was due to genetic differences among cultivars. Due to the direct effect of the chilling amount during blooming period from the very entering time of the buds to the greenhouse a measure known as the Chilling Requirement Index (CRI) was used. This index seemed to calculate the chilling effects more accurately. Chilling requirements were estimated to be 1100 h for Khanjari, Shahpasand, and Abasali cultivars and 1200 h for Akbari cultivar respectively. However, chilling more than the above amount was found to be effective on reduction of the bud blooming time.

Key words: Abasali cultivar, Akbari cultivar, chilling requirement, Khanjari cultivar, Pistachio, Shahpasand cultivar.

INTRODUCTION

Like other trees of moderate regions, pistachio trees require cold weather in their annual cycle so that the buds bloom naturally afterwards and suitable conditions are prepared for their growth (Erez, 2000; Couvillon, 1995). The minimum time for chilling a given cultivar in one season that enables its natural growth is called “chilling requirement” (Erez, 2000; Javanshah et al., 2006). Trees go into dormancy or rest mode during the time they receive chilling, and this mechanism serves to protect the buds against the winter cold. Buds that have entered dormancy mode do not wake up due to unusual heats of the winter (George et al., 2002; Crane and Iwakiri, 1981).

In many studies the efficient temperature for meeting the chilling requirements has been reported to range from 0 to 7ºC. Temperatures lower than 0ºC or higher than 7°C are not effective in this respect (Javanshah et al., 2006; Yazzetti et al., 2001). In models such as Utah (Richardson et al., 1974) temperatures as high as 12.5°C have also been recognized effective in meeting the chilling requirements. Daily temperatures above 15.5°C may even have negative impacts on the accumulated chilling set (Erez, 2000; Couvillon, 1995). The hours in which a plant receives chilling are not necessarily continuous; in other words, these hours accumulate and there is no need for them to be consecutive (Esmaeilzadeh et al., 2006). The Utah model was created by Richardson et al. (1974) for peaches planted on lands with very cold winters, but it has been developed a lot since then. This model shows the amount of effective cold a plant has received in cold regions without any mistake, yet its accuracy suffers a bit for warmer regions (Myers et al., 1996). Chilling requirements are different among various pistachio cultivars (Kuden et al., 1995). The chilling requirements of various pistachio cultivars have been reported to range between 450 and 1100 h at a temperature of 0 to 7°C (Crane and Iwakiri, 1981). Determining the chilling requirements of pistachio trees is very hard due to the final dominance. This is because the final dominance usually causes the decline of side buds (Kuden

*Corresponding author.Email: afshari2000ir@yahoo.com.

et al., 1995; Thakur and Rathore, 1991; Crane and Takeda, 1979). Knowing the chilling requirements of various pistachio cultivars and the temperatures of each region enables us to recommend the suitable cultivar for each region. Regions having mild winters are not able to provide sufficient chilling so farmers should use cultivars with lower chilling requirements. Also given the results of research conducted on meeting the chilling requirements of fruit trees of moderate regions (Finetto, 1997; Erez, 1995), particularly pistachio trees (Beede, 1997; Kuden et al., 1995) one can use some chemicals to reduce the losses in case the required chilling is not provided, and to do this one should already know the chilling requirement of various cultivars. Finetto (1997) studied some apple cultivars with medium chilling requirements planted in 1985. The experiment was run in the winter of 4 years, every time 30 one-year branches with at least 3 flower bud and 30 spore branches were cut on weekly basis and kept with their bases in water at a temperature of 20 - 21°C. Simultaneously, end and side buds were measured and the growth percent of the sprouts up to the final stage as well as the growing degree hours (GDH) were calculated. The beginning of the buds' awakening and end of inactivity in each branch and spore were compared and the ratio between the CU (chilling unit) and the CH (chilling hours) and GDH were determined by the said factors. Though there was a significant correlation between the opening of end and side buds with CU, CH, and GDH indices, the best method for estimating the time of ending the rest period was an accurate measurement of the chilling amount (Finetto, 1997).

Another study has been conducted to determine the chilling requirements of six cultivars of Arkansas Blackberry in which 10 slips each having 12 lumps were gathered from each cultivar and exposed with a mist system to 100 - 1000 h of chilling with 100 h intervals in a temperature below 7.2°C. There was a significant difference among various cultivars that indicated that mutual effects of chilling requirement time and cultivar. This shows that the different awakening of the buds is due to the different chilling requirements of various cultivars (Yazzetti et al., 2001; Richardson et al., 1974). Javanshah et al. (2006) studied the chilling requirements of 3 pistachio cultivars in Rafsanjan. Kaleghoochi cultivar had the lowest chilling requirement (600 h) and Akbari cultivar had the highest (1200 h). The goal of this study is to determine the chilling requirements of 4 commercial pistachio cultivars in Damghan region.

MATERIALS AND METHODS

This study was conducted by Islamic Azad University, Damghan Branch in 2006 on 4 cultivars of commercial pistachio in Damghan region (Shahpasand, Khanjari, Abasali, and Akbari) in order to determine their chilling requirements. Therefore a fully random factorial experiment was run with the two factors of slip chilling at levels (400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, and 1400 h) and cultivar at 4 levels (Shahpasand, Khanjari, Abasali, and Akbari).

In order to determine the chilling requirements of the above cultivars, slips of 50 - 60 cm length were cut from the trees after their dormancy in November. Treatments of this section included various chilling hours. Three runs were considered for each treatment and each test unit contained 5 slips. The slips of the said cultivars were sprayed with Benomile fungicide solution in order to prevent fungus growth; they were first put inside a wet cloth and then raped in rubber covers to prevent evaporation, and then transferred to a refrigerator to be chilled at a temperature of 4 - 5°C. Taking the slips out of the refrigerator was done according to the above chilling times. After that the slips were kept in greenhouse at a temperature of 20 - 25°C and in containers the water of which was changed every four days. With 10 day intervals new cuts were made with a length of 0.5 cm at the end of the slips in order to cut the vessels that were blocked of rotting. Of course in the primary plan it was arranged that such cuts be made with 4 day intervals, but due to the fact that no symptoms of tissue rot or destruction were observed the period was prolonged to 10 days. When entering the greenhouse, the number of grown and born buds on the slips was counted and afterwards the number of newborn buds was counted on daily basis. The signal for opening of the buds was taken as the time when the first leaves were seen out of the bud tip. This was done until no more bud opened. The rate of opening of the newborn buds was calculated based on the chilling requirement index (CRI) as detailed here:

Equation (1): CRI = (number of opened buds×100) / (total number of buds× number of germination days)

The chilling requirement index (CRI) was considered for estimating the speeds of bud germination in various treatments, and at the end the most suitable chilling requirement for each cultivar was determineed through studying the interactions of the treatments.

RESULTS

Based on the data obtained from variance analysis and Table 1 no significant difference was observed in terms bud reduction of pistachio. Low chilling hours caused maximum reduction and chilling hours above 900 caused minimum reduction in the buds (Table 1). But the effects of chilling hours and the reduction of buds make us conclude that Pistachio had higher reduction rates with lower chilling, in such a way that slips of Abasali cultivar faced a 30% reduction with 500 h of chilling. Khanjari and Shahpasand cultivars showed the same results with 700 h of chilling. However, 30% of the buds of Akbari slips were reduced even with 800 h of chilling.

Bud abscission

Studying the effects of factors such as cultivar, chilling hours, and their mutual effects on the abscission of the buds indicated that only the chilling hours had a significant influence at a 5% level. Maximum abscission of the buds was seen with 500, 600, and 700 h at temperatures of 2 - 3°C and minimum abscission was observed with 800 and higher chilling hours (Table 1).

Bud swelling

The swelling feature of the buds alone showed no significant difference among the 4 cultivars. Slips chilled for

Table 1. Studying the average effect of chilling hours on some features of Pistachio branches.

Chilling hours	Bud reduction (%)	Bud falling (%)	Bud swelling (%)	Bud opening (%)	Cluster % in flowering phase	Cluster % in full flowering phase	Cluster % in end flowering phase	Total number of flowers
500	28 a	8 a	0 c	0 c	0 b	0 c	0 c	0 c
600	23 ab	7.5 a	2 c	0 c	0 b	0 c	0 c	0 c
700	27 a	5.2 ab	2 c	0 c	0 b	0 c	0 c	0 c
800	25 ab	1.6 b	10 ab	5 c	0 b	0 c	0 c	0 c
900	21 b	0.5 b	13 ab	7 ab	0 b	0 c	0 c	0 c
1000	11 c	0.2 bc	17 a	11 a	4 ab	0 c	0 c	36 a
1100	4 d	0 c	14 ab	8 a	6 a	2 b	5 a	25 ab
1200	3 d	0 c	13 ab	8 a	7 a	2 b	2 b	25 ab
1300	3 d	0 c	14 ab	7 ab	4 ab	6 b	2.5 b	43 a
1400	3 d	0 c	12 ab	7 ab	4 ab	6 b	2.5 b	40 a

The numbers of each column have been compared separately. Numbers with similar letters have no significant different (α = 0.05).

Table 2. Studying the average effect of chilling hours and cultivar on the swelling of the buds of Pistachio branches.

Chilling hours (h)	Cultivars			
	Akbari	Abasali	Khanjari	Shahpasand
500	0 e	0 e	0 e	0 e
600	0 e	0 e	1 e	0 e
700	0 e	0 e	0 e	1 e
800	7 d	6 d	7 d	7 d
900	7 d	8 d	5 d	8 d
1000	11 c	30 a	21 b	8 d
1100	12 c	13 c	15 c	10 cd
1200	14 c	13 c	15 c	13 c
1300	12 c	13 c	15 c	14 c
1400	13 c	14 c	12 c	13 c

The numbers of each column have been compared separately. Numbers with similar letters have no significant different (α = 0.05).

more than 800 h at a temperature of 2 - 3°C had higher numbers of swollen buds (Table 1). By examining the mutual effects of cultivar and chilling hours on the swelling feature of the buds it was shown that Abasali and Khanjari cultivars had maximum swollen buds with 1000 h of chilling at 2 - 7°C (respectively 30 and 21%). Other cultivars had higher swelling rates with chilling hours of above 1000, as compared to lower chilling hours (Tables 1 and 2).

Bud break

The index to bud break was taken to be their green tips. In the slips of cultivars treated with 500 to 700 h of 0 - 7°C chilling no bud opened. From among all buds of the studied cultivars, nearly all of those receiving 800 to 1400 h of chilling had green tips. Nevertheless, 1000 h of chilling had more opened buds (11%) in comparison to other chilling hours (Table 1).

Flower cluster percent in the first phase of flower opening

The percent of opened flower clusters in the first phase of flower opening had no significant difference among the cultivars of our study. Yet the chilling hours had a significant effect on the percent of flower clusters at a 5% level in such a way that maximum flower cluster percent was observed in the first phase of opening of buds receiving 1000 - 1400 h of chilling (Table 1).

Flower cluster percent in full flower phase

Like previous treatments the rate of opened flower clusters in the full flower phase of all slips of chilled cultivars had no significant difference, but chilling hours of 1300 and 1400 and then 1100 and 1200 at a temperature of 0 - 7°C had maximum effect on the opening of the flowers in the full flower phase of the slips (Table 1) (CV = 26.4).

Flower cluster percent at the flowering end phase

Maximum percent of opened flower clusters in the flowering end phase were observed in chilling treatments of 1100 h at a temperature of 0 - 7°C (5%) (Table 1) (CV = 29.2). Chilling treatments of 1200, 1300, and 1400 were next. Opening of the flower clusters showed no significant difference among the studied cultivars.

Length and width of opened flower clusters

Based on the data gained from variance analysis it was observed that no significant difference existed between the length and width of the opened flower clusters among cultivars or due to chilling treatments. Nevertheless, the length of opened flower clusters on the slips of Akbari (11

Table 3. Comparison of CRI in 4 Pistachio cultivars.

Chilling hours (h)	Cultivars			
	Akbari	Abasali	Khanjari	Shahpasand
500	0 e	0 e	0 d	0 e
600	0 e	0 e	0 d	0 e
700	0 e	0 e	0 d	0 e
800	0.11 d	0.21 d	0.14 cd	0.12 d
900	0.70 cd	0.78 cd	0.28 cd	0.55 cd
1000	1.11 c	1.25 c	0.91 c	0.94 c
1100	1.61 c	2.91 b	1.93 b	1.81 b
1200	2.11 b	3.11 ab	1.97 b	2.41 ab
1300	2.95 b	3.15 ab	2.46 ab	2.87 ab
1400	3.87 a	3.91 a	2.84 a	3.15 a

The numbers of each column have been compared separately. Numbers with similar letters have no significant different (α = 0.05).

mm) were more than Abasali (6 mm), Khanjari (5 mm) and Shahpasand (4 mm) cultivars.

Total number of flowers in the clusters

The total number of flowers in the clusters showed no significant statistical difference, yet chilling treatments of 1000 to 1400 h are the ones that created the highest number of flowers in the clusters (Table 1).

Chilling Requirement Index (CRI)

The buds of each cultivar began to open after receiving a certain amount of chilling and the amount of chilling had an inverse relation with the duration of bud germination. Given the results obtained from Table 3 it was obvious that the chilling requirement of Akbari cultivar (1200 h) was higher than Shahpasand (1100), Abasali (1100), and Khanjari (1100) cultivars (Figure 1 and 2). Also it was shown that the amount of chilling provided had a direct effect on the time of opening of the buds. Yet with higher chilling hours the reaction of each cultivar to enhanced ambient temperature was slightly different. Nevertheless, according to the rate of opening of the buds most of the cultivars had the highest rate of bud opening first with 1000 h then with 1100 and 1200 h of chilling.

DISCUSSION

Due to insufficient chilling, all cultivars receiving lower chilling treatments (400, 500, 600 h at 0 - 7°C) experienced increased reduction and falling of the buds while lower swelling and opening in comparison to other treatments. The buds of the 4 cultivars did not demonstrate considerable difference in the experiment for 1000 h of chilling. Nevertheless, the 1200 h treatment showed to some extent the increased opening of Akbari cultivar in comparison to the other two. This may be due to the genetic features of this cultivar that make it react better than other cultivar due to the elimination of growth inhibitors and provision of sufficient chilling requirements. In all cultivars the buds opened within 8 - 12 days with 1300 and 1400 h of chilling and this was the cause of large CRI for them. The rate of opening of the buds (Table 2) shows nearly similar results in all cultivars with the CRI (Table 3). In both tables the reaction of the plant to chilling has occurred in nearly similar hours. Opening of the buds with 1000 h of chilling indicates that the CRI is 1100 for Khanjari, Shahpasand, and Abasali and 1200 for Akbari cultivars. In fact with 1000 h of chilling the number of opened buds was quite acceptable, yet this opening took a long time to happen. The results obtained from this study accord in many ways with those obtained by Esmeilzadeh et al. (2006). In the orchard environment the prolongation of the bud opening period exposes them to high spring temperatures and the plant is challenged by many problems. Meanwhile according to annual studies, Akbari pistachio trees have shown symptoms of insufficient chilling in winters with about 1000 h of cold weather. But in 1200 h of chilling weather the time required for opening of the buds decreases considerably and naturally there are less problems afterward. The CRI showed a significant difference between 1000 and 1200 h for Akbari cultivar (Table 3). Likewise for Khanjari, Shahpasand, and Abasali treatments of 400 to 1000 h are, while treatments of 1100 h had a significantly higher CRI (CRI Table) which indicated that such amount of chilling was sufficient for Khanjari, Shahpasand, and Abasali cultivars. Increasing of the CRI in chilling hours above 1200 was due to the shortening of the bud opening period. It was the same for other cultivars. What can be seen from the results of the experiment is that some commercial cultivars of pistachio trees have high chilling requirements. Given the higher winter temperatures of the recent years the chilling requirements of some cultivars of pistachio trees, which like Akbari cultivar need 1200 h of chilling, are not met in some regions and this cultivar has faced many problems in such lands.

Despite numerous reports on the determination of the chilling requirements of various trees and this fact that unsatisfied chilling requirements cause delay in opening of the buds, unfortunately none of the conducted studies and research efforts have focused on this correlation and it is possible that effective chilling requirements are not fully met. For instance, a cultivar such as Ohadi has a low chilling requirement of 600 h and all of its flower buds open during a long 45 day period; but if the chilling is increased to 800 h all buds will open in a very shorter period (15 days).

Therefore in this study the chilling requirement index (CRI) was taken as the index for chilling requirement and shows the relation between the number of opened buds and the time needed for opening of the buds (Equation 1). Since the CRI is a measure of the percent of bud opening in time, it is obvious that the larger it is the shorter the time required for the opening of the buds will be. If the chilling requirement of a tree is met the plant will stay in dorm-

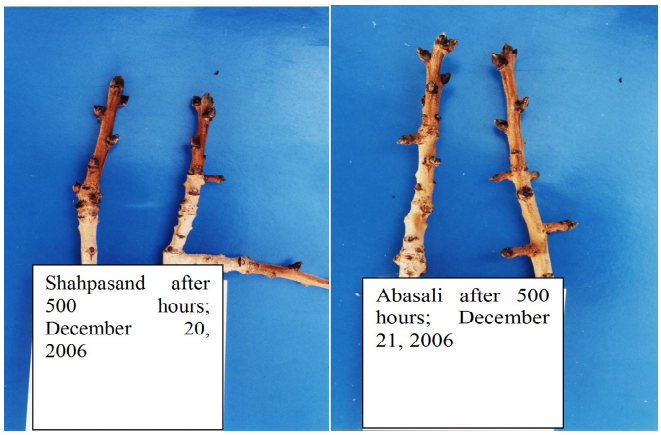

Figure 1. Right Image: (1) Slips of Abasali cultivar after 500 h; Left Image: (2) Slips of Shahpasand cultivar after 500 h, both cultivars did not showed any bed breaking in after 500 hchilling.

Figure 2. Right Image: (3) Slips of Shahpasand cultivar after 800 h; Left Image: (4) Slips of Shahpasand cultivar after 1100 h.

ancy state until temperature is once more suitable for its growth (Westwood, 1993). In trees which have received sufficient or extra chilling, the buds will open in a shorter time. In other words, there is a reverse relation between the bud opening time and the met chilling requirements. So the role of the CRI which was used in this study is quite clear.

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