Seed Dormancy and Germination of *Vaccinium arctostaphylos* L.

Sedaghathoor Shahram
Department of Horticultural Science, Islamic Azad University, Rasht Branch, Rasht, Iran

**Abstract:** Seed morphology and micrographs of seed surface and shape of *Vaccinium arctostaphylos* L. prepared using Scanning Electron Microscope (SEM) studied in this experiment. To evaluation of Qare-Qat seed dormancy and assessment of the best conditions for seed germination, the experiment carried out using factorial design with three factors in three replications in RCBID. Factor A was seed chilling period in four levels (15-90 days), Factor B was chilling conditions in two levels (wet and dry chilling) and factor C was germination conditions in two level too (germination in absolute dark and germination in light and darkness alternation). Results showed that the seeds of ripened fruits were not similar size and they are seen from minute to tiny size. Electron micrographs showed that Qare-Qat seeds are ovate or elliptical. Germination conditions test showed that seeds after expire of dormancy period could only germinate in light and darkness alternation and they could not germinate in absolute darkness. Therefore, Qare-Qat seeds had positive photoblastic reaction. Seed dormancy testing revealed that dry chilling of seeds for 15 to 90 days removes Qare-Qat seeds dormancy, however the best result obtained by 90 days dry chilling.

**Key words:** *Vaccinium arctostaphylos* L., Qare-Qat, seed dormancy, photoblastic

**INTRODUCTION**

Seed dormancy is defined as the failure of an intact viable seed to complete germination under favorable conditions (Bewley, 1997) and is controlled by several environmental factors such as light and temperature. Dormancy and germination are determined by the co-action of the growth potential of the embryo and the restraints imposed by the tissues surrounding it. The substantial influence of environmental effects on the expression of germination characteristics and the involvement of many genes make dormancy a typical quantitative trait (Koomneef et al., 2002).

Iranian Vaccinium or Qare-Qat (*Vaccinium arctostaphylos* L.) is a member of Ericaceae (Azadbaikh, 1999). It grows in Guilan province, Kalar Duash and Khangh Ardebel in north part of Iran. Qare-Qat grows as shrub or woody bush form up to 2.5 m height in *Pusus* forest. Hermafroditic and complete flowers of Qare-Qat appear in raceme inflorescence with pinkish red or white petals in June (Sedaghathoor et al., 2006). Many-seeded berries are produced on current or one-year-old shoots on the lateral buds according to Westwood (1993) that reported this situation for other *Vaccinium* bearing. Blueberries are close domestic relatives of Qare-Qat that are produced in Europe and America.

Based on Eck (1988) and Hartmann et al. (1997), none pre-treatment need for blueberries seed germination, but cranberry seed germination improved by three months seed chilling. Thakur and Rathore (1991) suggested stratification of blueberries seeds at 3-4°C for one month. Germination is further increased by the provision of alternate temperature and light (Thakur and Rathore, 1991). Some reports suggested three months chilling period to remove *Vaccinium arctostaphylos* seeds dormancy (Anonymous, 2000). According to Giba et al. (1995), some of *Vaccinium* species need light for germinating and germination percentage in complete darkness is less than 0.5%. The aim of this study was evaluation of the effects of chilling and light treatment on Qare-Qat seed germination.

**MATERIALS AND METHODS**

Qare-Qat berries were collected from main natural habitat in Iran (Talesh-Ardebil highlands). All seeds isolated from fruits and counted. According to Griffin and Blazich (2002), fruits are poured in water for seed extracting and then isolated by pressing for short time. Seeds counted after isolating from debris. All seeds isolated from fruits and counted. Furthermore, seed weight, seed to fruit weight ratio and seed size was evaluated.

**Corresponding Author:** Sedaghathoor Shahram, Department of Horticultural Science, Islamic Azad University of Rasht, Rasht, Iran
Since, seed traits and differences between the seed of different species used in recognition and registering new records, therefore, we prepared surface and shape seed micrographs using LEı model Scanning Electron Microscope (SEM). Qare-Qat seeds coated by thin layer gold for 4 min and then coated sample pasted on the holder and placed in electron microscope case (Shariatdokht and Madjd, 2000).

To study chilling requirement and dormancy period of Qare-Qat seeds and assessment of the best conditions for seed artificial chilling and germination, the experiment carried out using factorial design with three factors in three replications in RCBD. Separate lots of seed were subjected to the following treatments:

- **Factor A**: Days number for seed chilling (chilling period) in four levels at 4°C (including \( a_1 = 15, a_2 = 30, a_3 = 60 \) and \( a_4 = 90 \) days).
- **Factor B**: Chilling conditions of seeds in two levels (including \( b_1 = \) wet condition and \( b_2 = \) dry seed or dry chilling)
- **Factor C**: Germination conditions in two level (\( c_1 = \) germination in absolute dark and \( c_2 = \) germination in light and darkness alternation)

Twenty seeds studied per treatment and germination percent and rate assessed. Absolute darkness treatment prepared in laboratory incubator with 20-25°C and light-darkness alternation established in botanic laboratory using environment common light and dark. Seeds were germinated under continuous light and in darkness in glass Petri dishes on Whatman No. 1 filter paper moistened with distilled water. Seeds located at forecasting germination conditions for 60 days and traits studied. Temperature during the germination period was maintained at approximately 25°C. Three replications of seeds immediately after seed extracting (without chilling) studied as a control (out of test). We defined germination as the emergence and development of the radicle to the point where a portion of the seed was lifted from the germination medium. Daily records were kept of germination.

**RESULTS AND DISCUSSION**

The investigation of specimens showed that Qare-Qat, Siaghileh, Siahdar or Qareghileh are local and Persian names of just species of Vaccinium LEV. arctostaphylos L. in Iran. Based on fruit related data, average seed number per Qare-Qat berry was 45. Numerous seeds in fruits typically lead little fruit flesh imagination. But, comparing of seed/fruit ratio showed that 6% of fruit weigh allocated to seeds. Average weight of fruit and seed are 0.3 and 270 miligrams respectively. The seeds of ripened fruits were not similar size and are seen at minute to tiny size. Large diameter of seeds were times bigger than small one. The seeds were bright yellow to brown in color.

Seed micrographs obtained using Scanning Electron Microscope (SEM) showed that seeds were ovate elliptical in shape. The seed surface had cells with distinct walls (Fig. 1). There are tiny apertures on the Qare-Qat seed surface (Fig. 2). Saeidi-Mehrvarz et al. (2001) used seed electron micrographs of Veronica genus for appointment of taxons limits and distinguished six of type seeds. During dormancy period test, no seeds did germinate in absolute darkness and indicated that Qare-Qat seeds germinated only in the light/darkness alternation after chilling expiring. Thus, the seeds have positively photoblastic reaction. Seed germination exposed to light was reported for Vaccinium vitis-idaea (Thakur and Rathore, 1991). According to Giba et al. (1995), some of other Vaccinium species need light for germinating and germinated seed percentage in complete darkness is less than 0.5%. Since, the seeds did not germinate in dark conditions, thus germination in darkness factor i.e., \( c_2 \), removed from statistical analysis and it was only analyzed light/dark treatments data. Therefore, statistical design analyzed as factorial with two factors (A and B factors). No seed germinated 15-20 days after transferring of seeds from refrigerator to germination condition except seeds under \( a_2 b_1 \) treatment (i.e., 30 days dry chilling). This treatment led to germinate 10% of seeds after twenty days. Data obtained 30, 40, 50 and 60 days after seeds transferring to germination condition were analyzed (Table 1).

Table 1 showed ANOVA of germinated seed numbers at 30, 40, 50 and 60 days after transferring to germination environment. Based on results (Table 1), there is significant difference between chilling days number (Factor A), chilling type (factor B) and their interaction treatments at \( p = 1\% \) or \( p = 5\% \). One month after seeds transferring, two treatments including \( a_2 b_1 \), i.e., 15 days dry chilling of seeds and \( a_2 b_2 \), i.e., 90 days dry chilling of seeds were the best interaction effect with 25 and 20% germination respectively (Fig. 3).

The experiment revealed that seed chilling without watering (dry chilling of seeds) was been better than wet chilling. Sixty days after seed transferring to germination conditions, 30 days chilling \( (a_1) \) showed the best result. However, treatment 90 days dry chilling of seeds \( (a_1 b_3) \) caused increasing germination up to 55%. While, the least germination of seeds resulted under \( a_1 b_1 \) treatment (90 days wet chilling) with 10% germination (Fig. 3).
Fig. 1: Elliptical shape and distinct walls of seed surface of Qare-Qat

Fig. 2: Micrographs of seeds obtained by SEM, tiny apertures on the seed surface
Table 1: ANOVA of treatments effect on germinated seed numbers (30, 40, 50 and 60 days after transferring to germination condition)

<table>
<thead>
<tr>
<th>Variables</th>
<th>Degree of freedom</th>
<th>30 days</th>
<th>40 days</th>
<th>50 days</th>
<th>60 days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Replication</td>
<td>2</td>
<td>0.67**</td>
<td>4.63**</td>
<td>2.67**</td>
<td>8.79**</td>
</tr>
<tr>
<td>Factor A (Chilling Days)</td>
<td>3</td>
<td>3.83**</td>
<td>7.71**</td>
<td>11.38**</td>
<td>3.44*</td>
</tr>
<tr>
<td>Factor B (Chilling Condition)</td>
<td>1</td>
<td>60.17**</td>
<td>126.04**</td>
<td>145.04**</td>
<td>170.67**</td>
</tr>
<tr>
<td>Interaction AB</td>
<td>3</td>
<td>1.83*</td>
<td>11.71**</td>
<td>16.04**</td>
<td>22.11**</td>
</tr>
<tr>
<td>Error</td>
<td>14</td>
<td>0.38</td>
<td>1.44</td>
<td>1.1</td>
<td>0.74</td>
</tr>
<tr>
<td>CV%</td>
<td>-</td>
<td>32.20%</td>
<td>35.49%</td>
<td>24.82%</td>
<td>12.04%</td>
</tr>
</tbody>
</table>

ns: non-significant difference, * and **: significant at 5 and 1% level respectively

Fig. 3: Germination percentage of Qare-Qat seeds under treatments, 30-60 days after transferring of seeds to germination conditions. (Chilling period: a = 15, a = 30, a = 60 and a = 90 days, chilling type: b_1 = wet chilling and b_2 = dry chilling)

According to Nikolaeva (1977), there are three types of physiological dormancy: nondeep, intermediate and deep. Nondeep physiological dormancy is broken by relatively short (1-8 week) periods of warm or cold stratification, depending on the species. Intermediate physiological dormancy is broken by long (8-14 week) periods of cold stratification. Deep physiological dormancy is broken by long periods of cold stratification. (Baskin and Baskin, 1998). Since Qare-Qat seeds had 15-90 days chilling requirements to break dormancy, therefore, these seeds belong to second type of physiological dormancy i.e., intermediate.

In conclusion, test of Qare-Qat seeds chilling indicated that seed chilling in refrigerator at dry environment (without watering) is better than wet conditions (with watering) and 15 to 90 days chilling period could removed these seed dormancy, although, the best result obtained under 90 days dry chilling. It seems that wet existence beside seeds, not only does not remove dormancy, but also causes to reduction of germination after chilling period. Consequently, Qare-Qat seeds do not germinate in absolute dark. Based on Gorbunov and Kuznetsov (1995) studies on high bush blueberry seeds, 1-3 months chilling cause to remove of seeds dormancy. Phytochromes interfere in germinating of Vaccinium myrtillus seeds and red light causes germination inducing in this species, while far-red wavelength decreases seed germination (Giba et al., 1995). Sidorovich et al. (1991) perceived that high bush blueberry seeds are unable to germinate in absolute dark. This circumstance observed in Qare-Qat too.

It has proved that low bush blueberry seeds germinate, when those plant immediately extracting from fruit. However, seeds germination increased by dry chilling (3-5 degree centigrade) for 90 days (Griffin and Blazich, 2002). Some reports suggested 90 days chilling period to remove Vaccinium arctostaphylos seeds dormancy and it is said that Vaccinium arctostaphylos seed germinates if it has cultivated immediately after extracting from fruit (Anonymous, 2000). However, no seeds of Qare-Qat did germinate after extracting from fruits in our experiment. Finally, based on our results, Vaccinium arctostaphylos seeds have positively photoblastic reaction and do not germinate immediately after extracting from fruits.

According to Butkiewicz and Butkiewicz (1989), light improves fresh seed germination of some cultivars of V. corymbosum, but it has negative effect on stratificated seeds. Light is an essential component of any successful dormancy-breaking regime for seeds of Vaccinium spp. (Aalders and Hall, 1979; Austin and Cundiff, 1978; Devlin and Kareznarzycyk, 1977), but high light intensities may reduce germination (Austin and Cundiff, 1978). The second essential component is an alternating temperature regime (Hellman and Moore, 1983).

ACKNOWLEDGMENTS

Thanks to Islamic Azad University, Rasht Branch for financial assistance.

REFERENCES


