

# Germination Methods and Characteristics of Endemic *Centaurea olympica* DC. Koch Grown in Cultural Conditions

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The most suitable generative production method was selected in this work by applying different treatments to endemic *Centaurea olympica* seeds. The development performance and adaptation ability of the species in natural and cultural conditions (in the natural population, in pots and in the garden) were determined, with the goal to protect the species under *ex situ* conservation. The growth performance of the plant was monitored, and its ornamental plant potential was evaluated. The obtained data were compared with the data from the natural population. All morphological and phenological observations showed that the species can easily adapt to cultural conditions. For 2 months in the summer period, it is a lilac-flowered herbaceous plant that can be preferred for gardens with increasing number of shoots and flowers in parallel with its development. Among the different pretreatments applied to the seeds of the species, the highest germination percentage (73.5%) and germination speed ( $T_{50}=8$  days) was found with 600 ppm GA<sub>3</sub> treatment for 24 h after cold-wet stratification at 4 °C for 3 months. The appropriate germination temperature was determined as 20 °C.

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## INTRODUCTION

The genus *Centaurea* is of special importance in terms of species diversity. According to Uysal *et al.* (2022), *Centaurea* includes approximately 790 taxa. Turkey is the main diversity center of this genus; *Centaurea* is represented by 194 taxa in Turkey (Güner *et al.* 2012). Uysal *et al.* (2022) stated that the number has reached 220 with the recently added records, where 133 of these taxa are endemic, and the endemism proportion of the genus for Turkey is 60%. The distribution in some species of the genus *Centaurea* is significantly restricted and is represented by a single population. The risk of extinction of species is very high as a result of damage to the natural habitat and fragmentation of the population. Due to the effects of dormancy of the seeds and environmental conditions, dispersal, reproduction and regeneration are limited (Yankova-Tsvetkova 2018). Thus, it is necessary to conduct research on seed production of the endemic species belonging to the genus *Centaurea*.

One of these endemic species is *C. olympica*. Noroozi (2020) and Uysal *et al.* (2022) defined *C. olympica* as an endemic species that grows naturally in western and northwestern Anatolia, especially in Uludag and its surroundings. Kaynak (2014) lists the

species as endangered (NT, Near Threatened) species, which is currently not endangered, but a candidate for VU (Vulnerable), EN (Endangered), or CR (Critically Endangered) in the near future. Because the natural habitats are faced with many risks such as intense construction, human circulation, uncontrolled seed collection, drought and drought-related fires, endemic species that do not fall into any risk category at the moment should also be protected. In addition, the natural distribution areas of *C. olympica* are under the pressure of anthropogenic effects. It is an endemic species of our region. Therefore, *C. olympica* was chosen as the subject of study. *C. olympica* species is found in a population close to the highway in the region. Besides, there is a risk that the population will be exposed to construction machinery and human circulation in the future. In fact, the population of another endemic species in a nearby location was destroyed due to the road widening work. This species was chosen because of these developments and examples. This manuscript covered the endemic species belonging to Yalova province.

*C. olympica* is a 2-year herbaceous plant. Flowering period is July -August. It is an endemic species that reproduces by seed. From April to May, young shoots begin to form on the plant. With the development of these shoots, the plant forms a very branched structure. Lilac-colored blooms develop. In late September, the plant becomes flowerless. After flowering, the seeds mature in the seed pods. Researchers conducting this study have worked with botanists on other projects with some endemic plants, including *C. olympica*. For this reason, herbarium samples taken within the scope of these projects are available and their definitions have been made.

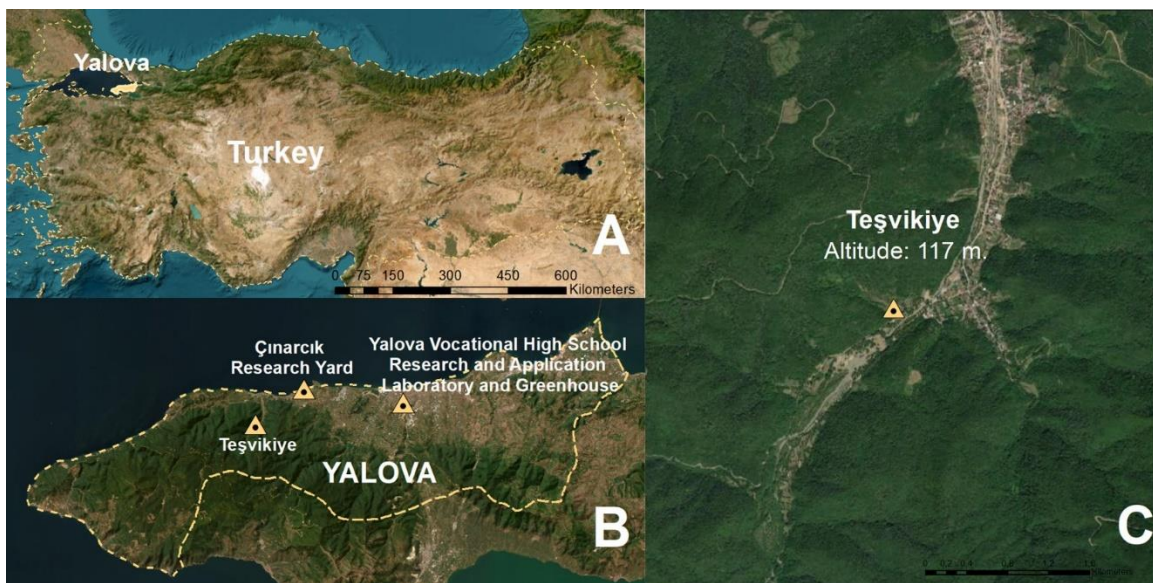
The goals of this study were as follows: (1) to determine the most suitable germination method by making different pre-treatments for seed germination of the endemic *C. olympica* taxon, (2) to evaluate the aesthetic, morphological, and phenological characteristics of the specimens, to evaluate its development performance in natural and cultural conditions, and (3) to reveal its potential for use as an ornamental plant and to put it under ex situ conservation.

## EXPERIMENTAL

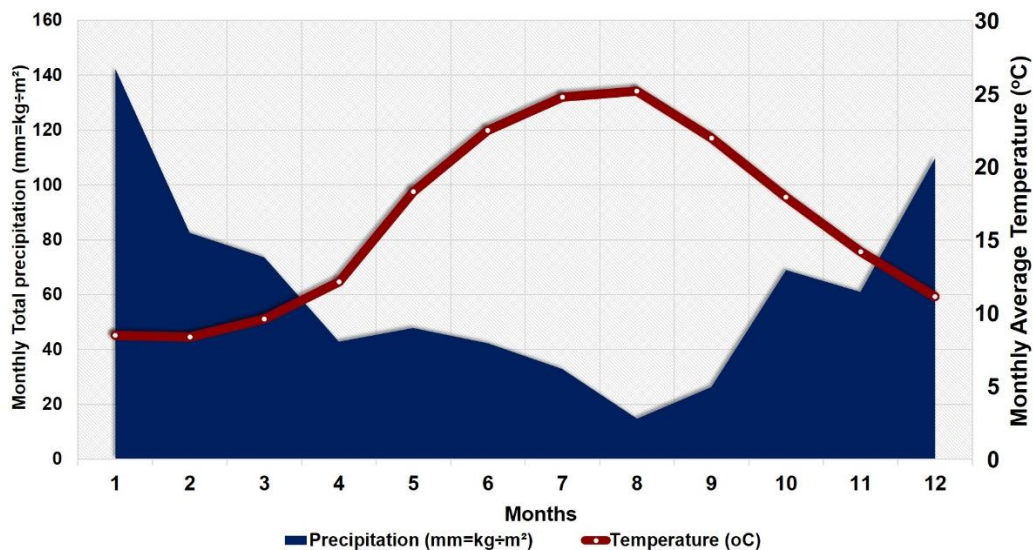
### Materials

*Centaurea olympica* DC. K. Koch seeds were collected on 02.08.2019, from the location between 3<sup>rd</sup> and 4<sup>th</sup> kilometers on the Yalova province Çınarcık district, Teşvikiye village-Delmece plateau road, whose coordinates was 40°36'50.63" N (latitude), 29°4'31.87" E (longitude). The number of individuals from which the seeds were collected was not taken into account, and the randomly selected collection was made within the population until the sufficient number was reached. Seeds were taken from multiple plants, not from a single plant. In order not to harm the plant population, a limited number of seeds were collected to be used in the study, and the number of repetitions was limited in order not to risk the existence of plants in the natural population. The natural population of the species is located on arid and rocky slopes in this region (Fig. 1). The study was carried out in Yalova between 2019 and 2021, under the climatic conditions of Yalova province (Fig. 2, Turkish State Meteorological Service, 2022). Data was collected from Two different fields were used. Seed germination studies and seedling growth processes were carried out in Yalova University Yalova Vocational School Research Laboratory and greenhouses (Fig. 1). To make measurements and observations of the plants after they are planted in the soil and in pots and to determine their adaptation abilities, the research garden

established in the Çınarcık Headquarters of the Nature Conservation and National Parks Yalova Branch Directorate was used (Fig. 1). In addition, measurements and observations were made in the area where the natural population is located. Measurements made in the natural population were made on randomly selected plants of different ages and different sizes. 1 and 2 year old plants were observed in plants planted in the soil under cultural conditions, and measurements and observations were made on 1-year plants in potted plants. The soil characteristics of the natural population area and the research garden (Çınarcık) are given in Table 1. Soil samples were taken in accordance with the technique for soil analysis as reported (Jackson 1962; Hızalan and Unal 1966). The prepared soil sample was analyzed in Yalova Atatürk Horticultural Central Research Institute Soil Analysis Laboratory. Knowing such data is important for comparing the development of plants grown under natural conditions with those grown under culture conditions.



**Fig. 1.** Geographical location of the population and study areas, Yalova Province Turkey Location (A), Location of the study areas in Yalova (B), Location of the natural population (C)



**Fig. 2.** Yalova province average climate data for 2019 to 2021

**Table 1.** Analysis of Soil in the Natural Area and in the Research Garden

Locations	% CaCO <sub>3</sub>	pH (1:2,5)	E.C. (1:2,5)	O.M. (%)	P (ppm)	K (ppm)	% Sand	% Silt	% Clay
Teşvikiye	0.78	6.3	1713	9.41	80	440	53.97	32.07	13.96
Research Garden Soil	4.11	7.63	547	2.69	20	190	47	17	36

## Methods

### *Morphological, phenological, and biological characteristics of seeds*

The collected seeds were cleaned and dried in a room with an average day/night temperature of 20 to 21/13 to 14° C and air flow with a fan. After approximately 10 days of drying, the seed morphology (seed size, 1000 kernel weight, number of seeds per g) was analyzed. For measurements and weighing, 200 seeds consisting of 50 seeds with 4 replications were randomly selected, and the measurements were made on these seeds. The seed phenology (maturation date, capsule cracking date, and seed shedding date) was recorded throughout the vegetation periods.

### *Seed viability test*

The seeds were divided into 3 groups containing 20 seeds, and each group was counted as one replicate. Seeds were kept in water at room temperature (20 to 22 °C) for 24 h. Then, 1/3 of the seeds were cut and kept in a 1% solution of tetrazolium (2,3,5 Triphenyl tetrazolium chloride) (Merck, Darmstadt, Germany) for 24 h at 30 °C. At the end of the period, the shells of the seeds were peeled off. Physical observations were made about the staining extents of the seeds. If the seed was completely stained, it was considered viable; if the seed had very little coloration or if some parts of the seed remained unpainted, it was considered semi-viable; and if there was no coloration, the seed was considered dead (Moore 1985; Peters 2000).

### *Germination studies*

To determine the optimum temperature for germination of *C. olympica* seeds, the seeds were subjected to wet stratification at 4 °C for 2 months. Germination experiments were carried out under different temperature conditions (15, 20, and 25 °C). In germination treatments, 12/12 light regime and 70% humidity were used. The light source had an intensity of 3400 lumens.

After determining the suitable temperature for germination as 20 °C, the second part of the germination study was started by application of the treatments: cold-wet stratification, different doses of GA<sub>3</sub> (200, 400, 600 ppm) treatments or their combinations. In cold-wet stratification, sterilized seeds were kept in moistened perlite at 4 °C for 3 months. In GA<sub>3</sub> treatments, seeds were kept in 200, 400, or 600 ppm GA<sub>3</sub> solutions for 24 h. All treatments of this study were carried out at 20±0.5 °C, in 12/12 light regime, at 70% humidity, with a light intensity of 3400 lumens. Different treatments and concentrations were tried to see which treatment provides the best germination speed and germination percentage.

A programable plant growth chamber (SWGC-450, Daihan Scientific, Seoul, Korea) was used for germination. All Petri dishes (100 × 20 mm glass) and blotting papers used during the germination tests were sterilized by keeping them at 100 °C for 30 min before the processes started. The sterilization process of the seeds was carried out by keeping the seeds in 70% ethanol (Soltek, Turkey) for 1 min and keeping them in a 20%

solution containing 5.25% sodium hypochlorite (BRTR Kimya, İzmir, Turkey) for 10 min. After sterilization, seeds were rinsed in distilled water. After placing two layers of moistened blotting paper on the bottom of the Petri dishes, seeds were planted on the papers. Petri dishes were capped and wrapped with parafilm. To prevent disease, 2.5 mL/L commercial fungicide was applied to each Petri dish with an active ingredient of 25 g/L Fludioxonil + 10 g/L Metalaxyl-M (Maxim XL 035 FS, Syngenta, Gaillon/France). The results of the germination tests were evaluated considering the total percentage of germinated seeds (FGP, final germination percentage). Seeds were considered to be germinated when the radicle came out of the shell by 2 mm. Counting processes in germination tests were made every two days, and germination monitoring continued for 60 days (ISTA 2013). Seed germination percentage and speed were calculated.

#### *Determination of aesthetic, morphological, and phenological characteristics in natural and cultural conditions*

To determine their morphological and phenological characteristics under culture conditions, the plants were planted in the soil field and pots in the research garden. The aesthetic, morphological, and phenological characteristics of the species were observed throughout the development period in the pot (1), in the soil (2), and natural population. The traits were compared with the data from the natural population. Between April and September, the natural population and plants in the research garden were monitored, and the records of observations and measurements were kept during periodic visits in these areas. Random plant selection was made for observations and records. For this purpose, three groups consisting of 10 plants, which were determined completely randomly, were formed; a total of 30 plants were studied. The following traits were observed:

- Number of main shoots: The number of main shoots emerging from the first 5 cm on the stem of the plant (pieces)
- Stem length to flowering part: Length from ground level to the point where the first flower appears (cm)
- Number of shoots in the flowering part: The number of side shoots with flowers on the main shoots (pieces)
- Flower width: The distance between the two petal tips when the flower is in full bloom (mm)
- Flower size: Distance from the bottom of the calyx to the tip of the petal at the very end of the petals when the flower is in full bloom (mm)
- First bloom date: The date the first flower appeared in the population
- Last flower death date: The date when the last flower wilted in the population

Plants to be grown in soil and pots were obtained by growing the seedlings formed after germination tests. Seedlings removed from Petri dishes at the beginning of January were planted in 104 compartment viols filled with peat. At the beginning of February, seedlings grown in the greenhouse without heating (day/night 14/9 °C, 70% relative humidity) were planted in pots (16x13,5 cm) filled with soil belonging to the research garden, the contents of which are given in Table 1. Plants growing in pots in the greenhouse were taken outdoors in mid-March when they had 7 to 8 real leaves. In the middle of April, when they had 10 to 12 real leaves, they were transferred to the Çınarcık research garden (Table 2) and the ones to be monitored in the soil area were planted in the soil (Table 1). All observations of the plants that will grow in pots and soil under culture conditions were made in this area during the vegetation periods.

**Table 2.** Stages of Development of Seedlings

Applied Process	Date
Planting the germinating seedlings in 104 viols (unheated greenhouse and 70% humidity) and day/night temperature 14/9 °C)	Early January
Transferring the developing plants to 16x13.5 cm pots	Early February
Removing plants that form 7 to 8 true leaves to the outside	15.3.2020
Transferring 10 to 12 true leaf-forming plants to the Çınarcık research garden to monitor their development in pots and soil	15.4.2020

Plants were grown for the establishment of an *ex situ* conservation garden. These plants were planted in the *ex situ* conservation garden at the beginning of April. No herbal supplement was made to the natural population.

#### *Trial design and data analysis*

Germination experiments were planned according to the Randomized Block Experimental Design, with 4 replications (4 Petri) and 50 seeds in each replication. The data obtained from the research were evaluated with the IBM SPSS Statistics Base 22.0 (IBM Corp., Armonk, NY, USA) statistical prog. Multidirectional analysis of variance was performed on the data, and the results that were found to be statistically different were grouped with Duncan multiple comparison test. The percentage of germination (%) was determined after transformation with arc-sin data transformation. In order not to harm the plant population, enough seeds were collected to be used in the study. The number of repetitions was limited in order to protect the plants in the natural population.

## RESULTS AND DISCUSSION

### Determination of Morphological, Phenological and Biological Characteristics of Seeds

The seeds are yellowish beige, the seed coat is shiny, slightly tapering at the tip of the base. At the top of the seed, there are hard hair-like structures (pappus) arranged in the form of brushes. These structures act as propellers and play an important role in transporting seeds over long distances (Fig. 3).

The measurements, counts, and weighing of the seeds of the *C. olympica* species and the viability test results are given in Table 3. The seeds were approximately 4.48 x 1.71 mm in size, 1000 kernel weight was 3.2 g, and there were an average of 311.7 seeds per 1 g. Considering *C. olympica* seed structure, Eroğlu *et al.* (2014) and Tel *et al.* (2019) stated that the seed size of *C. hermannii* is 4 to 5 mm. Yücel (2022) stated the dimensions of *C. hermannii* seeds as 2.37x5.27 mm. *C. olympica* is smaller than *C. hermannii* in terms of seed size.



**Fig. 3.** General view of *Centaurea olympica* seeds

From the seeds collected at the populations around Yalova Teşvikiye village in 2019, the net viability rate was 76.7%. However, this rate can vary greatly according to the years and the time of collection of the seeds. The seed viability rate was low in *C. olympica* in the year of the study. The high rate of semi-viable seeds indicates that there might be insect damage, physical effects, or problems with the maturation of the seed. The non-viable seed rate was within normal limits.

**Table 3.** Measurements and Viability of *Centaurea olympica* Seeds

Seed Size (mm)	Seed Width (mm)	1000 kernel weight (g)	Number of seeds per 1 g	Non-viable seed (%)	Semi-viable (%)	Viable Seed (%)
4.18±0.50	1.71±0.36	3.2078±0.02	311.74±1.70	6.66±0.36	16.67±0.58	76.67±1.47

Different results have been reported for the viability studies of *Centaurea* species. For example; Yücel (2022) determined the viability rate for *C. hermannii* seeds as 72.22%, while Atasagun and Aksoy (2018) determined 82% for *C. amaena* seeds, Yankova-Tsvetkova *et al.* (2018) found this rate to be 17.5% for *C. achtarovii*, Özel and Khawar (2006) found 85% for *C. tchihatcheffii*, and Emek and Erdağ (2012) reported 80% viability for *Rhaponticoides mykalea* species. All these reports revealed that the viability rates can vary significantly according to species, years, and environmental conditions. Similarly, Gough and Basu (2020) stated that the quality of the starting seed, the moisture level in the soil, the temperature values it is exposed to, the duration of light, the mechanical damage that may occur before and during the harvest, the rains that fall just before the harvest, the wrong harvesting time may affect viability of the seed. Therefore, any change that may occur in these factors may cause changes in the seed viability rate.

Data on the phenological observations of *C. olympica* seeds under different ecological conditions are given in Table 4. The, fruit ripening, capsule cracking, and seed pouring stages occur simultaneously in natural populations and plants under cultural conditions. The fourth week of July and the first week of August appear to be the most suitable time for seed collection in *C. olympica*.

**Table 4.** Observations of *C. olympica* under Natural and Cultured Conditions

Growing Conditions		Ripening Date	Capsule Cracking Date	Seed Spill Date
Natural habitat		3rd week of July	1st week of August	2nd week of August
Culture medium– Pot		3rd week of July	1st week of August	2nd week of August
Culture medium - Soil	1 year	3rd week of July	1st week of August	2nd week of August
	2 years	4th week of July	2nd week of August	3rd week of August

### Germination Studies

#### *Determination of optimum germination temperature*

The optimum germination temperature was 20 °C, with a germination rate of 65.8% (Table 5). When the effects of different temperature treatments on the germination of *C. olympica* seeds were evaluated, it was determined that a constant temperature of 20 °C was the optimum temperature value and gave the best results in terms of average germination percentage (65.8%). This result is in correlation with study by Abbasian *et al.* (2016), who stated that the highest germination was obtained at 20 °C with *C. balsamita*. Similarly, Pitcairn *et al.* (2002) reported the optimum temperature required for germination of *C. calcitrapa* was 20 °C. Albert *et al.* (2002) found that the highest germination percentage of *C. pinnata* seeds was obtained at temperatures of 15 to 20 °C. On the other hand, Turkoglu *et al.* (2009) suggested that the appropriate germination temperature for 3 different *Centaurea* species (*C. balsamita*, *C. virgata*, *C. iberica*) was 15 °C. Zare *et al.* (2020) reported that they obtained the highest germination percentage at 25 °C for *C. bruguierana*. Clements *et al.* (2010), on the other hand, found that optimum germination occurred at variable temperature values of 5/25 and 10/25°C among the many temperature values they tried for *C. diffusa* seeds.

**Table 5.** Effect of Temperature on *C. olympica* Germination

Treatments	Average Germination (%)± SE
2 months cold-wet stratification at 4 °C and germination at 15 °C	55.75 ± 1.60 b*
2 months cold-wet stratification at 4 °C and germination at 20 °C	65.75 ± 0.57 a
2 months cold-wet stratification at 4 °C and germination at 25 °C	52.75 ± 1.65 b
	p≤0.05

\* There is no statistically significant difference between values expressed with the same letter in the same column.

#### *Determination of the effects of GA<sub>3</sub> concentration, cold-wet stratification and their combinations on the germination of C. olympica seeds.*

The effects of pretreatments consisting of 3 months cold-wet stratification, different GA<sub>3</sub> concentrations and their combinations on the germination of *C. olympica* seeds were evaluated. The combination of soaking in 600 ppm GA<sub>3</sub> solution for 24 h after 3 months of cold-wet stratification (73.50%) was more effective than other treatments (Table 6). In terms of germination speed (*T*<sub>50</sub>), there was a statistical difference between the treatments, and the best results were obtained by soaking seeds in 400 ppm and 600 ppm GA<sub>3</sub> solutions after cold-wet stratification at 4 °C for 3 months (Table 5). Thus, GA<sub>3</sub> treatments after cold-wet stratification increase the germination speed (*T*<sub>50</sub>) of *C. olympica* seeds. This



result is similar to the results obtained by Okay *et al.* (2011) and Okay and Demir (2021), who suggested the stratification for 120 to 150 days and 100 ppm GA<sub>3</sub> treatment before planting to increase the germination rate of *C. tchihatcheffii*.

**Table 6.** Effect of Cold-wet Stratification and GA<sub>3</sub> on *C. olympica* Germination

Treatments	Germination (%) ± STD	Germination Speed (days) ± STD
Control	64.00 ± 1.28 b*	28.00 ± 0.50 d
200 ppm GA <sub>3</sub> treatment for 24 h	65.00 ± 0.77 b	14.00 ± 2.69 bc
400 ppm GA <sub>3</sub> treatment for 24 h	67.50 ± 0.92 b	12.00 ± 3.11 b
600 ppm GA <sub>3</sub> treatment for 24 h	67.00 ± 1.23 b	14.00 ± 2.83 b
3 months cold (4 °C) – wet stratification	52.50 ± 0.98 c	17.00 ± 0.50 c
3 months cold (4 °C) – wet stratification + 200 ppm GA <sub>3</sub> treatment for 24 h	62.50 ± 1.65 b	10.00 ± 1.37 ab
3 months cold (4 °C) – wet stratification + 200 ppm GA <sub>3</sub> treatment for 24 h	69.50 ± 1.07 ab	8.00 ± 0.86 a
3 months cold (4 °C) – wet stratification + 200 ppm GA <sub>3</sub> treatment for 24 h	73.50 ± 2.09 a	8.00 ± 1.08 a
Stratification x GA <sub>3</sub> = ns**	p ≤ .001	p ≤ .001

\* There is no statistically significant difference between values expressed with the same letter in the same column; \*\* not significant (p ≤ .05)

**Table 7.** Effects of GA<sub>3</sub> Treatments on Germination Percentage and Germination Speed (*T*<sub>50</sub>) of *C. olympica* Seeds as a Single Factor

Treatments	Average Germination (%) ± STD	Average Germination Speed (days) ± STD
Control	58.25 ± 2.91 b*	22.50 ± 2.10 b*
200 ppm GA <sub>3</sub> treatment for 24 h	63.25 ± 1.73 ab	12.00 ± 1.44 a
400 ppm GA <sub>3</sub> treatment for 24 h	68.50 ± 1.84 a	10.00 ± 1.65 a
600 ppm GA <sub>3</sub> treatment for 24 h	69.75 ± 4.40 a	11.00 ± 1.60 a
	p ≤ .037	p ≤ .000

\* There is no statistically significant difference between values expressed with the same letter in the same column.

Compared with the control group, all GA<sub>3</sub> concentrations were statistically effective in increasing the germination percentage (Table 7). In terms of germination speed (*T*<sub>50</sub>), all concentrations were effective compared with the control, but there was no statistical difference between GA<sub>3</sub> concentrations (Table 7). This result is also similar to Nolan (1989), who stated that GA<sub>3</sub> is a strong stimulant for germination of *C. diffusa* and *C. maculosa*.

When the effect of cold-wet stratification was evaluated as a single factor, it was not statistically effective in terms of germination percentage. In terms of germination speed (*T*<sub>50</sub>), cold-wet stratification was effective. Stratification provided an average (*T*<sub>50</sub>) 6-day earliness in seed germination (Table 8). In parallel to this study, Luna *et al.* (2008) also stated that for *C. ornata* and *C. pinae* species, stratification did not have a significant effect in terms of germination percentage. However, when the germination speed (*T*<sub>50</sub>) was taken

into account, the wet stratification as a seed pretreatment increased the germination speed compared to the control. The effect of cold-wet stratification processes varies according to the *Centaurea* species. For example, Eddleman and Romo (1988) stated that the cold-wet stratification applied to *C. maculosa* seeds shortened the germination period and increased the total germination. Nolan (1989) stated that some dormant seeds of *C. diffusa* and *C. maculosa* species, which were cold-treated at 3 °C, germinated at 25 °C, while Aghilian *et al.* (2014) stated that there was no effect of cold-treatment for *C. cyanus* seeds.

**Table 8.** Effect of Cold-wet Stratification on Germination Percentage and Germination Speed ( $T_{50}$ ) of *C. olympica* Seeds

Treatments	Average Germination (%) $\pm$ STD	Average Germination Speed (days) $\pm$ STD
Control	65.38 $\pm$ 1.23**	17.00 $\pm$ 1.95 b*
Cold-wet stratification	64.50 $\pm$ 3.04	10.75 $\pm$ 1.00 a
	p $\leq$ .791	p $\leq$ .008

\* There is no statistically significant difference between values expressed with the same letter in the same column; \*\*not significant (p $\leq$ .05)

The reactions of the seeds of *Centaurea* species to different treatments can result in many very different outcomes. There are great differences in the germination characteristics of plant species depending on genetic and environmental factors and even the environments used for germination testing (Elias *et al.* 2012; Baskin and Baskin 2014). The percentage of germination in seeds can be affected even from the side of the plant where the seed was collected (Nielsen 1988; Copeland and McDonald 2001). Another reason for obtaining different results in similar studies may be the conditions during the waiting period between the harvest and germination studies (Probert 2000). As a result, germination is a very complex physiological process and can be affected by many factors (Gresta *et al.* 2010).

### Determination of Aesthetic, Morphological and Phenological Characteristics in Natural and Cultural Conditions

The results of the morphological and phenological characteristics of *C. olympica* in the natural population and cultural conditions are given in Table 9. When the results of *C. olympica*'s adaptability to cultural conditions and potential ornamental plant characteristics are evaluated, the species does not lose its characteristics under culture conditions. An increase in the number of main branches and sub-branches has been observed over the years, especially for plants grown in soil. The number of shoots per plant was 6.21 in the natural area in perennial plant, while the number of shoots was 8.66 for 1-year plant and 10.6 for 2-year plant in cultural conditions. Moreover, in plants grown in soil, a significant increase was observed in the number of side shoots attached to the main shoots bearing flowers, and this number increased to 72.3. It is possible to explain these significant increases observed in the parameters monitored as the years progressed, with the careful maintenance conditions in the cultural environment.

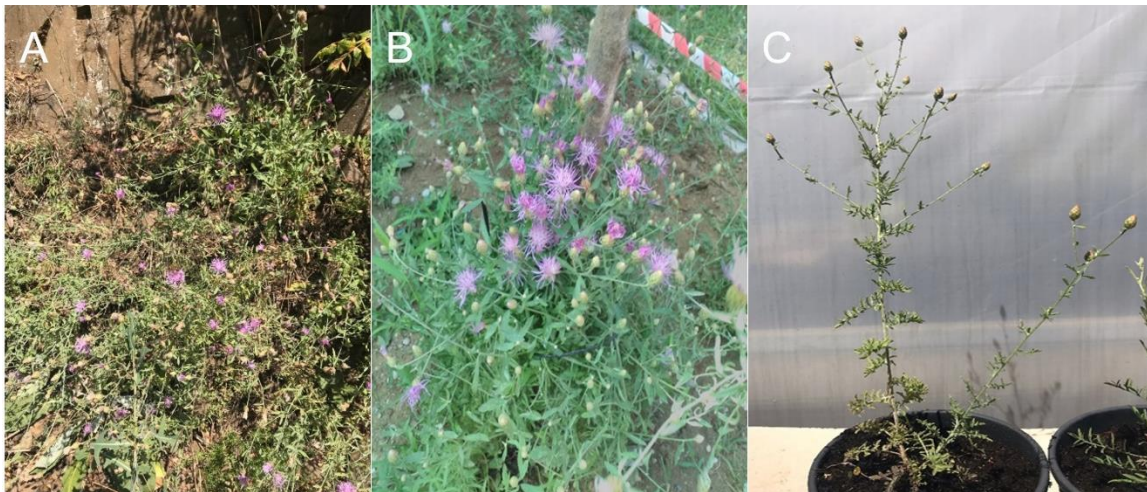
Considering the stem length up to the flowering part of the plant, the length was 26.82 cm in the natural habitat, where 30.46 cm in the soil at the end of the 1<sup>st</sup> year and 22.66 cm in the 2<sup>nd</sup> year under cultural conditions.

**Table 9.** Characteristics of *C. olympica* in Natural and Cultured Populations

Growing Conditions	Number of Main Shoots (pieces) $\pm$ STD	Stem Length Up To Flowery Part (cm) $\pm$ STD	Number of Shoots in Flowering Part (pieces) $\pm$ STD	Flower size (mm) $\pm$ STD	Flower width (mm) $\pm$ SE	Number of flowers (pieces) $\pm$ SE	First Bloom Date	Last Flower Death Date
Natural population (in perennial plant)	6.21 $\pm$ 0.55	26.82 $\pm$ 0.55	20.41 $\pm$ 0.43	18.00 $\pm$ 0.47	27.14 $\pm$ 0.59	124.40 $\pm$ 20.45	3.07.2020	4.09.2020
Cultural conditions / 1 <sup>st</sup> year in pot	5.33 $\pm$ 0.45	24.18 $\pm$ 1.19	9.31 $\pm$ 0.79	17.11 $\pm$ 0.64	26.71 $\pm$ 0.77	19.70 $\pm$ 2.53	28.06.2020	27.08.2020
Cultural conditions / 1 <sup>st</sup> year in soil	8.66 $\pm$ 0.54	30.46 $\pm$ 1.27	28.46 $\pm$ 0.90	18.57 $\pm$ 1.20	29.70 $\pm$ 1.16	146.90 $\pm$ 11.62	5.07.2020	8.09.2020
Cultural conditions 2 <sup>nd</sup> year in soil	10.6 $\pm$ 1.18	22.673 $\pm$ 1.77	72.3 $\pm$ 3.02	18.514 $\pm$ 1.34	27.561 $\pm$ 1.20	315.90 $\pm$ 23.63	10.07.2021	11.09.2021

These results can be explained by the fact that as the development of the plant and its adaptation to the soil increase, it can form flowering shoots from a closer distance to the soil surface. This is a positive development for the use as an ornamental plant. While the number of shoots in the flowering part was 20.41 in the natural habitat, this feature was determined as 28.46 in the 1<sup>st</sup> year and 72.3 in the 2<sup>nd</sup> year in cultural conditions in the soil. This remarkable increase in the number of shoots in the flowering part naturally led to an increase in the number of buds. Especially in the 2<sup>nd</sup> development year, a considerable increase in the number of flowers was observed in the plants grown in the soil (315.9 units). There was no significant change in the size (height and width) of the flowers of the species over the years, compared to the natural area, in the culture conditions (pot and soil area). The flowering period of *C. olympica* was determined as approximately 2 months during July-August in all three growing media. The measurement and observations of this part of the study show that the plant can easily adapt to the culture environment under regular maintenance conditions.

According to the information obtained from the study, *C. olympica* tends to expand to the sides during development. Because it has a very branched structure, a large number of purple-pink flowers (19.70-315.90 pieces) can form on the shoots under culture conditions, which is a special feature for the plant (Table 9, Fig. 4).



**Fig. 4.** General view of *C. olympica* plants A) Natural population B) Culture medium/garden C) Culture medium/pot

## CONCLUSIONS

1. *Centaurea olympica* easily adapted to cultural conditions without changing or losing its characteristics associated with its natural environment. The species has no special demands during its growing, needs less cultural processes, and provides numerous lilac-colored flowers that bloom continuously for 2 months in dry land conditions during the summer period, which makes it a candidate as an ornamental plant in landscape design and projects.

2. The appropriate temperature for the germination of *C. olympica* seeds was 20 °C. Among the different pre-treatments applied at this temperature, it was determined that the best results in terms of germination percentage and germination speed were achieved by cold-wet stratification at 4 °C for 3 months and then soaking in 600 ppm GA<sub>3</sub> for 24 h. It has also been shown that the cold-wet stratification together with GA<sub>3</sub> soaking can shorten the germination time considerably.

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