



## The Effects of Some Dormancy Breaking Treatments and Temperature on Seed Vigor of Gum Tragacanth (*Astragalus gummifer* Labill.)

Gülüm GÜREL<sup>1</sup>, Bilal KESKİN<sup>\*2</sup>, Süleyman TEMEL<sup>3</sup>

<sup>1</sup> Department of Field Crops, Postgraduate Education Institute, Iğdır University, Iğdir, Turkey

<sup>2,3</sup> Department of Field Crops, Faculty of Agricultural, Iğdır University, Iğdir, Turkey

<sup>1</sup><https://orcid.org/0000-0003-0120-3902>, <sup>2</sup><https://orcid.org/0000-0001-6826-9768>, <sup>3</sup><https://orcid.org/0000-0001-9334-8601>

\*Corresponding author e-mail: bilalkeskin66@yahoo.com

### Article Info

Received: 22.11.2022

Accepted: 11.05.2022

Online published: 15.06.2022

DOI: 10.29133/yyutbd.1026792

### Keywords

*Astragalus gummifer*,  
Dormancy breaking,  
Germination,  
Seed dormancy,  
Temperature

**Abstract:** This research was carried out to determine the effects of germination temperature and 12 dormancy breaking applications on the germination of the seeds of the gum tragacanth (*Astragalus gummifer* Labill.) bush. The research was carried out in the Field Crops Department laboratory, Iğdır University Faculty of Agriculture, in 2019. Gum tragacanth seeds were germinated for 28 days in the dark at constant temperatures of 10, 15, 20, and 25 °C and variable temperatures of 20/10 °C, 20/15 °C, 25/10 °C, and 25/15 °C. As a result of the research, the highest total germination rate was determined at 10.7% at 25/10 °C and 25/15 °C temperatures. It was determined that there was 89.3% dormancy in gum tragacanth seeds. Then, 12 dormancy breaking methods (matrix priming, hydro priming, gibberellic acid (GA<sub>3</sub>, potassium nitrate, cold, moist stratification, warm moist stratification, warm+cold moist stratification, cold+warm moist stratification, cold water, hot water, mechanical scarification, and chemical scarification) were applied. After dormancy breaking applications were made, the seeds were germinated again at 25/15 °C. At the end of the study, it was revealed that the highest total germination percentage with 50.7% was obtained from the application of hot water for 2 minutes. On the other hand, it was determined that matrix priming, hydro priming, gibberellic acid, potassium nitrate, cold, moist stratification, warm moist stratification, cold+warm moist stratification, mechanical scarification, and chemical scarification applications did not have any effect on removing the dormancy status of gum tragacanth seeds.

**To Cite:** Gürel, G, Keskin, B, Temel, S, T. 2022. The Effects of Some Dormancy Breaking Treatments and Temperature on Seed Vigor of Gum Tragacanth (*Astragalus gummifer* Labill.) *Yuzuncu Yil University Journal of Agricultural Sciences*, 32(2): 266-279. DOI: <https://doi.org/10.29133/yyutbd.1026792>

**Footnote:** This study was produced from Gülüm GÜREL's Master's thesis.

## 1. Introduction

The genus *Astragalus*, which includes annual and perennial herbaceous and shrubby plants and belongs to the leguminous family, includes 3000 species. *Astragalus* species are spread over a wide area such as Europe, Asia, and North America, with 463 species and 41% (210 species) of these species endemic in Turkey (Dinç et al., 2013; Erkul and Aytac, 2013). *Astragalus* species are widely used as a feed source for animals, as a raw material for medicine, dye, textile industry, as a source of nectar for bees, and also in the control of erosion areas (Demir and Keskin, 2016; Keskin and Temel, 2019; Bagheri

et al., 2015; Budge et al., 2012; Lee et al., 2007; Abd Kadir et al., 2013). *Astragalus brachycalyx*, *Astragalus gummifer*, *Astragalus kurdicus*, and *Astragalus microcephalus* species from which gum called tragacanth is extracted are widely found in Turkey, Iran, Caucasus, and Afghanistan regions, and this gum obtained is used in pharmacy, paint and weaving industries (Khan and Abourashed, 2010).

Gum tragacanth (*Astragalus gummifer* Labill.) is a perennial herb that can grow up to 60 cm. It is piled rooted, and its stem and shoots are thorny. Its flowers are white and pollinated by bees. When their stems are cracked and scratched, they ooze a gum called tragacanta (tragacanth gum). It is resistant to sandy, acidic, and arid areas (Keskin and Temel, 2019). Gum tragacanth spends the winter period in a dormant state and forms new shoots and leaves with the onset of temperatures, and the newly formed fresh shoots and leaves maintain their greenery throughout the summer. Flowering continues between May and October, and fruit formation continues for a long period, from June to November (Keskin and Temel, 2019).

Gum tragacanth, which is used in many areas, grows in natural areas, and these plants are not cultivated and grown in the field. Therefore, these plants cannot be controlled, and their generation may be extinct. The germination rate is expected to be high in cultivated seeds. On the other hand, non-cultured plants are more likely to have hard seeds and dormancy. It is also difficult to obtain seeds in these plants. Knowing the optimum germination temperatures of the seeds and the level of dormancy characteristics, if any, and which methods and methods are effective on breaking dormancy in seeds in order to spread the plants of high importance to wider areas will contribute to the dissemination of this plant to wider areas. Dormancy is defined as the inability of seeds to germinate due to internal (water and gas impermeability of the seed coat, chemical substances in the seeds) and external (temperature, oxygen, light) factors even though the environmental conditions are suitable. The practices required to break dormancy in seeds vary from species to species and even between seeds of the same species from different origins. In order to break dormancy, stratification, soaking in water, using growth regulators, washing, drying, heat and light treatment, mechanical scarification, chemical scarification, and combinations of one or more of these are used (Obalı, 2009; Agrawal and Dadlani, 1995; Erken and Kaleci, 2010; Moradi et al., 2016; Lagha et al., 2001; Tuncer, 2019; Maesaroh and Demirbağ, 2020).

In the literature review on the germination status and dormancy breaking methods of the seeds of gum tragacanth, it was seen that no study was conducted. Therefore, this study was planned to determine the temperature at which gum tragacanth showed high germination and which methods methods would be appropriate for breaking the dormancy in seeds.

## 2. Materials and Methods

### 2.1. Plant material

The study was carried out in the laboratory of the Department of Field Crops, Faculty of Agriculture, Iğdır University, in 2019. The seeds of gum tragacanth were cut in October-November and dried in the shade after being brought to the laboratory. The dried shoots were crushed with a plastic board on the bottom of the sieve, and the seeds were removed from the shoots. Damaged and broken seeds in the seeds were removed. The 1000 grain weight of the seeds was determined as 3.7 g. Seeds were stored in airtight packages at 5 °C until used. Seed viability and dormancy applications were carried out according to ISTA rules (ISTA, 2017).

### 2.2. Germination test

The seeds were germinated in the dark at constant temperatures of 10, 15, 20, and 25 °C and variable temperatures of 20/10 °C, 20/15 °C, 25/10 °C, and 25/15 °C. 3x25 seeds were used in each application. The seeds to be germinated were kept in 5% sodium hypochlorite for 60 seconds and then left to dry for 30 seconds on germination papers for half an hour. After the seeds were placed on the germination paper in a 120x20 mm glass petri dish, they were left on the germination papers with 25 seeds in each glass petri dish and then covered with a second germination paper. Germination papers were wetted with water prepared by adding 0.2 g of pomarsol to 1 liter of distilled water as starting water to prevent fungus growth. After the establishment of the experiment, daily counts were made for 28 days, and rootlets germinating above 2 mm were accepted as germinated (ISTA, 2017). At the end of the 28th day, normal, abnormal, and dead seeds were counted.

## 2.3. Dormancy breaking applications

12 dormancy breaking methods were applied to gum tragacanth milkvetch based on ISTA (2017) rules. After the dormancy breaking methods were applied, the seeds that showed the highest total and normal germination at the end of the germination tests were subjected to germination tests for 28 days at 25/10 °C and 25/15°C. Since the highest total and normal germination in all dormancy breaking applications occurred at 25/15 °C, statistical analyzes were made based on data at only 25/15 °C temperature.

### 2.3.1. Matrix priming

Seed: vermiculite: water; The seeds and vermiculite were placed in gauze nets in a 2: 1: 3 ratio and placed in opaque containers with water. The dishes were kept in the dark for 24, 36, and 48 hours at 15 °C and then dried to their initial weight at 25 °C.

### 2.3.2. Hydro priming

The seeds to be used in hydro priming application were kept in water at 20 °C for 5 hours, and then surface drying was applied to the seeds. After the surface drying process, the seeds in the tulle pouch were left on the wire tray in the aging pots filled with water so that they do not come into contact with water. The mouth of the aging containers was covered with a cling film in an airtight manner, and the lids of the containers were tightly closed. Seeds kept in aging containers at 20 °C for 60, 72, and 96 hours were dried at room temperature until they reached their initial weight.

### 2.3.3. Gibberellic acid

The seeds were kept in the dark for 24 hours in 250, 500, and 1.000 ppm GA<sub>3</sub> solutions so that they were completely submerged, and then surface drying was applied to the seeds.

### 2.3.4. Potassium nitrate

The seeds were kept in the dark for 6 hours in 2% and 4% KNO<sub>3</sub> solutions so that they were completely submerged, and then surface drying was applied to the seeds.

### 2.3.5. Cold moist stratification

After the seeds were placed between completely saturated coarse filter papers, they were placed in tulle pouch, and the applications were kept at 5 °C for 3 and 4 weeks.

### 2.3.6. Warm moist stratification

The seeds were kept for 1 and 2 weeks at 20 °C between completely saturated coarse filter papers and surface drying was applied to the seeds.

### 2.3.7. Warm+cold moist stratification

The seeds were kept between completely saturated coarse filter papers at 20 °C for 1 and 2 weeks, then kept at room temperature for 24 hours and then kept at 5 °C for 3 and 4 weeks. Afterwards, surface drying was applied to the seeds.

### 2.3.8. Cold+warm moist stratification

Seeds were kept between completely saturated coarse filter papers at 5 °C for 3 and 4 weeks, then 24 hours at room temperature, and then at 20 °C for 1 and 2 weeks. Then, surface drying was done on the seeds.

### 2.3.9. Coldwater

The seeds were kept in tulle pouch for 1, 2, and 4 weeks at 5 °C, completely submerged in water, and then surface drying was performed on the seeds.

### 2.3.10. Hot water

The seeds were kept in tulle pouch for 2 and 4 minutes in boiling (100 °C) water so that they were completely submerged in the water. Then the surface drying process was carried out.

### 2.3.11. Mechanical scarification (Sanding)

Seeds were abraded in a shaking device for 5, 10, and 15 minutes in size 10 sandpaper.

### 2.3.12. Chemical scarification (Sulfuric acid)

Seeds were kept in 96% H<sub>2</sub>SO<sub>4</sub> for 10, 20, and 30 seconds. After the application, the seeds were washed with pure water and left to dry on blotting paper.

## 2.4. Comparison of dormancy breaking applications

In the germination tests, the germination rate control data obtained at 25/15 °C temperature were accepted, and a comparison was made with the dormancy breaking application, which obtained the highest germination rate after each dormancy breaking method was applied.

## 2.5. Statistical analysis

The analysis of variance was carried out according to the randomized plots experimental design according to the JMP 5.0.1 package program of the research data. The mean of the important factors is grouped according to the least significance difference (LSD).

## 3. Results and Discussion

### 3.1. Germination rates of *Astragalus gummifer* at different temperatures

There were significant changes in the total and normal germination rates of gum tragacanth milkvetch seeds germinated at different temperatures. Total germination rates were 2.7%, 8.0%, 8.0%, 8.0%, 5.3%, 8.0%, 10.7% and 10.7%. at 10, 15, 20, 25, 20/10, 20/15, 25/10 and 25/15 °C temperatures, respectively. On the other hand, normal germination rates were 0.0%, 4.0%, 5.3%, 8.0%, 5.3%, 8.0%, 9.3% and 9.3%, respectively. The highest total and normal germination rates were found at 25/10 °C and 25/15 °C temperature values. There was no significant change in abnormal germination rates at different temperatures. Although the germination rate of *Astragalus gummifer* seeds increased slightly due to the increase in temperature, it was observed that the seeds showed dormancy at a significant rate (89.3%). It is an expected situation to show dormancy in uncultured plants. *Astragalus gummifer* seeds were not found to germinate normally at 10 °C, the lowest temperature used in the study, while there was some germination due to the increase in temperature, but this germination rate remained at 9.3% (Figure 1).

It was determined that there was no significant change in the germination rate between different temperature values without pretreatment, and the highest germination rate was 10% in *Astragalus adsurgens*, 3.17% in *Astragalus maritimus*, 4% in *Astragalus arpilobus* and *Astragalus bibullatus*, and the species showed significant dormancy. (Kondo and Takeuchi, 2004; Bacchetta et al., 2011; Albrecht and Penagos, 2012; Long et al., 2012). On the other hand, it was determined that *Astragalus adsurgens* germinated at different temperature values at 30 °C, *Astragalus gines-lopezii* at 15/25°C and *Astragalus membranaceus* at 10 °C (Jaganathan et al., 2019; Zhou et al. al., 2012; Schnadelbach et al., 2016). Generally, *Astragalus* seeds appear to show high dormancy. *Salsola kali* subsp. *ruthenica* seeds at 10 and 25 °C (Obalı, 2009), *Alhagi pseudodalhagi* seeds at 25 °C (Moradi et al., 2016). As can be seen in previous research, the germination temperatures of the seeds vary according to the plant genus and species.

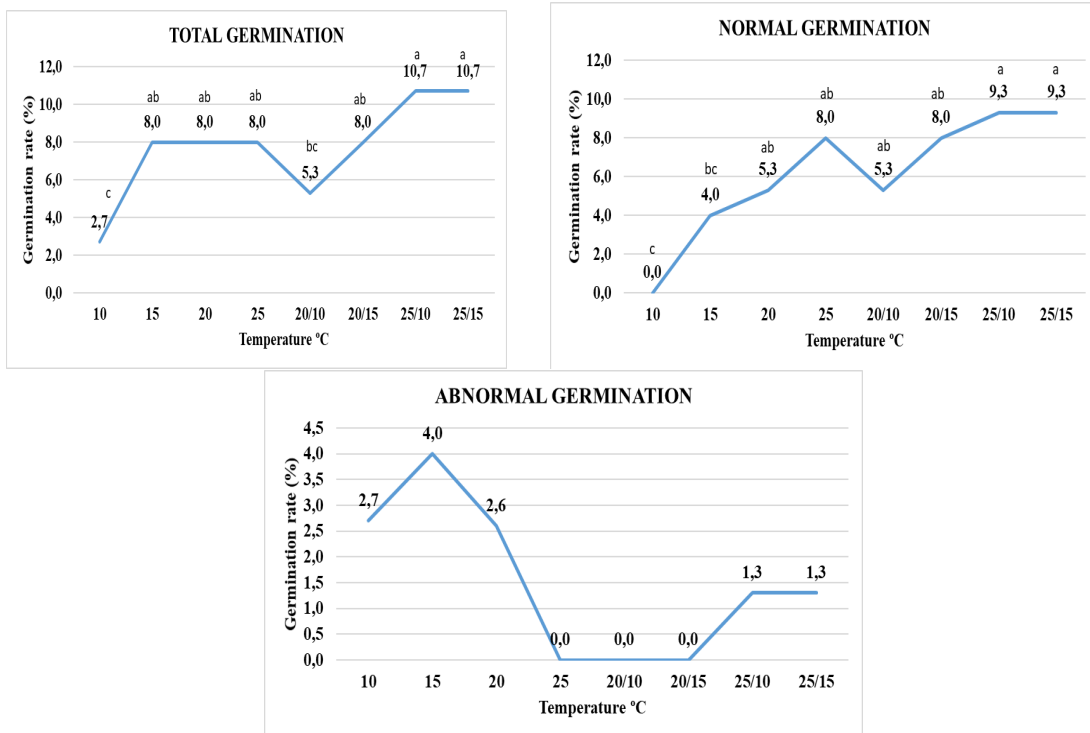


Figure 1. Total, normal and abnormal germination rates of *Astragalus gummifer* seeds at different temperatures (Total germination LSDs: 3.0\*\*, Normal germination LSDs: 4.7\*\*, Abnormal germination LSDs: 4.7ns), \*\* $P < 0.01$  are significant within the probability limits, ns is insignificant.

### 3.2. Germination rates of *Astragalus gummifer* in dormancy breaking applications

#### 3.2.1. Matrix priming

While matrix priming application at different times had a significant effect on total seed germination and normal seed germination rates of gum tragacanth milkvetch, it was observed that it did not have a significant effect on abnormal seed germination rate. When matrix priming was applied for 24, 36, 48 hours, total seed germination rates in gum tragacanth milkvetch seeds were 8.0%, 22.7%, and 8.0%, respectively. The highest total seed germination rate was 22.7% in the matrix priming application for 36 hours. The lowest total germination rate of 8.0% was observed in the seeds applied 24 and 48 hours of matrix priming. Normal germination rates were determined as 20.0% in gum tragacanth milkvetch seeds, which were applied matrix priming for 36 hours (Table 1).

In the literature studies, it was seen that matrix priming application was not tested in *Astragalus* species. However, studies on different plant species (*Allium cepa*, *Abelmoschus esculentus* and *Allium ampeloprasum*) determined that matrix priming increased the seed germination rate compared to control and was a successful method in breaking dormancy (Özden et al., 2018a; Pandita et al., 2010; Ozden et al., 2018b). In the present study, it was found that matrix priming application increased the germination rate in *Astragalus gummifer* seeds and revealed that it was an effective method in breaking dormancy, which also supports previous studies.

#### 3.2.2. Hydro priming

The effect of different hydro priming applications on seed germination rates of gum tragacanth milkvetch was not significant. With the application of hydro priming for 48, 72, and 96 hours, total seed germination rates of gum tragacanth milkvetch were 10.7%, 9.3%, and 2.7%, while normal germination rates were 10.7%, 6.7%, and 2.7%, and abnormal germination rates were 0.0%, 2.7%, and 0.0%, respectively.

In previous studies, no hydro priming application was found to break the dormancy in the seeds of *Astragalus* species. However, they determined hydro priming of *Nigella sativa* (Tajbakhsh et al.,

2014), *Allium cepa* (Özden et al., 2018a), and *Lactuca sativa* (Rao et al., 1987) seeds increased the germination rate compared to the control and was a successful method in breaking dormancy.

### 3.2.3. Gibberellic acid

Gibberellic acid application at different concentrations had a significant effect on the total germination and abnormal germination rate of gum tragacanth, but the effect on the normal germination rate was not significant (Table 1).

Table 1. The effects of some dormancy breaking practices on seed viability of *Astragalus gummifer*

Applications	Application levels	Total germination (%)	Normal germination (%)	Abnormal germination (%)
<b>Matrix Priming</b>	24 hour	8.0 b	8.0 b	0.0
	36 hour	22.7 a	20.0 a	2.7
	48 hour	8.0 b	8.0 b	0.0
<b>LSD value and significant</b>		6.05**	0.00**	6.05 ns
<b>Hydro priming</b>	48 hour	10.7	10.7	0.0
	72 hour	9.3	6.7	2.7
	96 hour	2.7	2.7	0.0
<b>LSD value and significant</b>		ns	ns	ns
<b>Gibberellic acid</b>	250 ppm	6.7 b	6.7	0.0 b
	500 ppm	10.7 a	6.7	4.0 a
	1000 ppm	10.7 a	9.3	1.3 ab
<b>LSD value and significant</b>		3.83**	ns	3.02*
<b>Potassium nitrate</b>	%2	10.7	9.3	1.3
	%4	10.7	9.3	1.3
<b>LSD value and significant</b>		ns	ns	ns
<b>Cold moist stratification</b>	3 week	24.0 a	21.3 a	2.7
	4 week	4.0 b	2.7 b	1.3
<b>LSD value and significant</b>		9.94**	5.74**	ns
<b>Warm moist stratification</b>	1 week	9.3	8.0	1.3
	2 week	14.7	12.0	2.7
<b>LSD value and significant</b>		ns	ns	ns
<b>Warm + cold moist stratification</b>	1 week+3 week	14.7 b	12.0 bc	2.7
	1 week+4 week	24.0 a	17.3 a	6.7
	2 week+3 week	12.0 b	9.3 c	2.7
	2 week+4 week	16.0 ab	16.0 ab	0.0
<b>LSD value and significant</b>		9.23**	4.80*	ns
<b>Cold + warm moist stratification</b>	3 week+1 week	10.7	10.7	0.0
	3 week+2 week	14.7	12.0	2.7
	4 week+1 week	14.7	13.3	1.3
	4 week+2 week	9.3	6.7	2.7
<b>LSD value and significant</b>		ns	ns	ns
<b>Cold water</b>	1 week	13.3 b	8.0	5.3
	2 week	20.0 a	13.3	6.7
	4 week	14.7 b	12.0	2.7
<b>LSD value and significant</b>		3.70**	ns	ns
<b>Hot water</b>	2 minute	50.7	38.7	12.0 b
	4 minute	46.7	20.0	26.7 a
<b>LSD value and significant</b>		ns	ns	5.74**
<b>Mechanical scarification</b>	5 minute	20.0 a	10.7	9.3 a
	10 minute	9.3 b	9.3	0.0 b
	15 minute	9.3 b	9.3	0.0 b
<b>LSD value and significant</b>		6.05**	ns	3.02**
<b>Chemical scarification</b>	10 second	6.7 b	6.7	0.0
	20 second	10.7 a	10.7	0.0
	30 second	9.3 ab	8.0	1.3
<b>LSD value and significant</b>		3.02*	ns	ns

\*P < 0.05 significant at probability limits, \*\*P < 0.01 significant at probability limits, ns not significant.

Total germination rates of gum tragacanth seeds were 6.7%, 10.7%, and 10.7% in 250, 500, and 1000 ppm applications of gibberellic acid. While the highest total germination rate was obtained in 500 and 1000 ppm gibberellic acid application, the lowest total germination rate was found in 250 ppm

application. Gibberellic acid application at different concentrations did not have an effect on increasing the germination rates obtained without any dormancy breaking the application. Although the application of gibberellic acid in breaking dormancy is a common application, the application of gibberellic acid in gum tragacanth seeds did not make a significant contribution to the increase in germination rate. In studies on some *Astragalus* species, it has been revealed that gibberellic acid application is not effective in breaking dormancy (İkram et al., 2014). On the other hand, it is an effective method in breaking dormancy (Zhou et al., 2012; Keshtkar et al., 2008). On the other hand, it was determined that the application of gibberellic acid to *Centaurea tchihatcheffii*, *Capparis ovata*, *Arbutus andrachne*, *Thlaspi lilacinum*, *Draba brunifolia*, and *Vitis vinifera* seeds increased the germination rate of seeds and was an effective method in breaking dormancy (Okay and Günöz, 2009; Gökçöl and Duman, 2018). ; Onursal and Gözlekçi, 2007; KIRMIZI, 2017; Akkurt et al., 2013). However, in studies conducted on *Achillea gypsicola*, *Saponaria halophila*, and *Chamaecytisus pygmaeus* species, it was determined that gibberellic acid application was not effective in increasing the seed germination rate (Çolak, 2011; Erken et al., 2014; Açıkgöz and Kara, 2019).

#### 3.2.4. Potassium nitrate

There was no significant difference between 2% and 4% potassium nitrate application on total germination, normal germination, and abnormal germination rate of gum tragacanth. Total germination, normal germination, and abnormal germination rates were 10.7%, 9.3%, and 1.3%, respectively, in both potassium nitrate applications (Table 1). In previous studies, it was determined that potassium nitrate application was not effective in increasing the germination rate of some *Astragalus* seeds (İkram et al., 2014), while it increased the germination rate in some species (Zhou et al., 2012). In the current study, it was determined that potassium nitrate application was not effective in increasing the germination rate of gum tragacanth seeds.

#### 3.2.5. Cold moist stratification

Changes in total germination and normal germination rates were observed in gum tragacanth seeds, which were subjected to cold moist stratification at different times. The total germination rates in the seeds germinated after the 3rd week and 4th week of cold moist stratification application were 24.0% and 4.0%, respectively, while the normal germination rates were 21.3% and 2.7% (Table 1). Considering these values, total germination and normal germination rates were higher in seeds that were cold moist stratification for 3 weeks. It has been determined that there will be significant decreases in germination rates of gum tragacanth seeds in case of longer cold stratification (4 weeks).

In a study conducted by Cavieres and Almedia (2018), although cold stratification increased the germination of *Astragalus looseri* seeds by 9% compared to the control, this increase was found to be statistically insignificant. Germination rates were determined as 7.7% in control application and 16.7% in cold stratification. Long et al. (2012) applied *Astragalus arpilobus* seeds in wet sand at 4 °C for 4, 8, 12, and 16 weeks. Cold stratification did not increase the germination rate of seeds, and they determined that it was not an effective method for breaking dormancy. Jones et al. (2016) applied cold stratification to *Astragalus filipes* seeds for 3 weeks at 4 °C. Cold stratification increased the germination rate of seeds, and it was determined to be an effective method for breaking dormancy. Isavand et al. (2005) applied cold stratification to *Astragalus siliquosus* seeds. Cold stratification increased the germination rate of seeds, and it was determined to be an effective method for breaking dormancy.

Studies have shown that cold folding has a positive effect on breaking dormancy. In the current study, it has been seen that cold stratification is an effective method in breaking dormancy and encourages germination, which supports previous studies.

#### 3.2.6. Warm moist stratification

There was no significant difference in seed germination rates between 1 and 2 weeks of warm stratification of seeds of gum tragacanth. The total germination rates of seeds, which were germinated after 1 and 2 weeks of warm stratification, varied between 9.3% and 14.7%, normal germination rates between 8.0% and 12.0%, and abnormal germination rates between 1.3% and 2.7%, respectively (Table 1). In a study, It was determined that the application of warm stratification to *Salsola kali* subsp. *ruthenica*. Seeds decreased the germination rate compared to the control and had no effect on the breaking of dormancy (Obalı, 2009).

### 3.2.7. Warm+cold moist stratification

It was observed that the effect of warm+cold stratification at different times on gum tragacanth seeds on total germination rate and the normal germination rate was significant, but the effect on abnormal germination was insignificant (Table 1). In 1 week warm+3 weeks cold, 1 week warm+4 weeks cold, 2 weeks warm+3 weeks cold, and 2 week warm+4 week cold stratification, total germination rates were 14.7%, 24.0%, 12.0% and 16.0%, normal germination rates were determined as 12.0%, 17.3%, 9.3%, and 16.0%, respectively. The highest total germination and normal germination rates were obtained in 1 week warm+4 weeks cold stratification (Table 1). Previously, no study has been found in which warm+cold stratification has been tested in seeds of *Astragalus* species. However, it was determined that 3 week warm stratification + 12 week cold stratification application of *Fraxinus ornus* seeds had a significant effect on the germination rate (Tilki, 2005). On the other hand, it was determined that the application of warm+cold stratification to *Flueggea anatolica* seeds had no effect on breaking dormancy (Avşar and Ok 2009).

### 3.2.8. Cold+warm moist stratification

It was observed that cold+warm stratification application of gum tragacanth seeds at different times did not have a significant effect on total, normal and abnormal seed germination rates (Table 1). In the current study, as a result of 3 weeks cold+1 week warm, 3 weeks cold+2 weeks warm, 4 weeks cold+1 week warm, and 4 weeks cold+2 weeks warm stratification, total seed germination rates were 10.7%, 14.7%, 14.7%, and 9.3%, normal seed germination rates were 10.7%, 12.0%, 13.3%, and 6.7%, respectively.

### 3.2.9. Cold water

While significant differences were observed in the total germination rate of gum tragacanth seeds by soaking in cold water for 1, 2, and 4 weeks, no significant differences were observed in normal and abnormal germination rates. Total germination rates were found as 13.3%, 20.0%, and 14.7% in cold water soaking for 1, 2, and 4 weeks, respectively. In applications, the highest total germination rate was obtained with 20.0% in cold water soaking for 2 weeks (Table 1). In a study conducted on the subject, it was determined that keeping *Astragalus adscendens* and *Astragalus podolobus* seeds in cold water at 4 °C for 10 days increased the germination rate of seeds and was an effective method in breaking dormancy (Tavili et al., 2014). In another study, when *Salsola kali* subsp. *ruthenica* seeds were washed in running water for 24, 48 and 72 hours, germination rates of 87.33%, 86.67%, and 84.67% were obtained, respectively. In the control, a germination of 84% was achieved. Therefore, it was determined that the germination rate decreased with the increase in the soaking time of the seeds (Obalı, 2009).

### 3.2.10. Hot water

There were significant changes in the total, normal and abnormal seed germination rates of gum tragacanth seeds in different times (2 and 4 minutes) soaking in boiling water. Total germination rates of gum tragacanth seeds kept for 2 and 4 minutes in hot water were 50.7% and 46.7%, normal germination rates were 38.7% and 20.0%, and abnormal germination rates were 12.0% and 26.7%, respectively (Table 1). On the other hand, significant differences were observed in abnormal germination rates in 2 different periods of soaking in hot water. The highest total and normal germination rates were obtained in the application of soaking gum tragacanth seeds in boiling water for 2 minutes. It was determined that there was a decrease in the total germination and normal germination rates of the seeds that were kept in boiling water for a longer time and an increase in the abnormal germination rate at the same time. It is estimated that hot water application has a significant effect on the permeability of the seed coat and contributes to the increase in germination rate.

In many studies, the seeds of *Astragalus* species were soaked in hot water. *Astragalus maritimus* and *Astragalus verrucosus* seeds in 100 °C hot water (Bacchetta et al., 2011), *Astragalus adscendens* and *Astragalus podolobus* seeds in hot water for 5 minutes (Tavili et al., 2014), *Astragalus hamosus* seeds in 60, 70, 80, 90 and 100 °C hot water for 5, 10, 15 and 20 minutes (Patane and Gresta, 2006), *Astragalus cyclophyllon* seeds in 60, 80 and 100 °C hot water for 5 and 10 minutes (Keshtkar et al., 2008), *Astragalus arpilobus* seeds in 70, 80, 90 and 100 °C hot water (Long et al., 2012) and *Astragalus podolobus* seeds in boiling water for 1 minute (Agh et al., 2017) applied soaking and determined that soaking in hot water is an effective method for breaking dormancy. On the other hand, it has been



determined that soaking in hot water for seeds of *Astragalus filipes*, *Astragalus gines-lopezii*, and *Astragalus cicer* is not an effective method for breaking dormancy (Kildisheva et al., 2018; Schnadelbach et al., 2016; Statwick, 2016).

### 3.2.11. Mechanical scarification

Mechanical scarification of gum tragacanth seeds for 5, 10, and 15 minutes caused significant changes in total germination and abnormal germination rates. On the other hand, it was determined that mechanical scarification application for different durations did not cause significant changes in the normal germination rate (Table 1). As a result of mechanical scarification application of gum tragacanth seeds for 5, 10, and 15 minutes, total germination rates were 20.0%, 9.3%, and 9.3%, while abnormal germination rates were determined as 9.3%, 0.0%, and 0.0%, respectively. It was observed that mechanical scarification caused an increase in abnormal germination rate in the application for 5 minutes, but there was no significant change in normal germination.

Mechanical scarification has been applied to seeds of many *Astragalus* species. Mechanical scarification increased the germination rate of the seeds compared to the control in *Astragalus peckii*, *Astragalus penduliflorus*, *Astragalus filipes*, *Astragalus siliquosus*, *Astragalus hamosus*, *Astragalus cicer*, *Astragalus fridae*, *Astragalus tribuloides*, *Astragalus bibullatus*, and *Astragalus contortuplicatus* seeds and determined that mechanical scarification is an effective method for breaking dormancy (Miklas et al., 1987; Isavand et al., 2005; Eisevand et al., 2006; Fateh et al., 2006; Patane and Gresta, 2006; Arbabian et al., 2009; Albrecht et al., 2012; Molnár et al., 2015; Pearson, 2015; Jones et al., 2016; Schnadelbach et al., 2016; Siles et al., 2016; Statwick, 2016; Kildisheva et al., 2018; Dziurka et al., 2019).

### 3.2.12. Chemical scarification (Sulfuric acid)

It was observed that keeping gum tragacanth seeds in 96% sulfuric acid for 10, 20, and 30 seconds caused a difference in total germination rates, while it did not cause a significant change in normal germination and abnormal germination rates. The total germination rates in seeds kept for 10, 20 and 30 seconds in chemical scarification were found to be 6.7%, 10.7%, and 9.3%, respectively. The highest total germination rate was observed in the seeds that applied chemical scarification for 20 seconds (Table 1). When the seeds of gum tragacanth were kept in sulfuric acid for 10 and 20 seconds, no abnormal germination was observed. However, abnormal seed germination was observed at the rate of 1.3% when kept for 30 seconds.

As chemical scarification, sulfuric acid applications at different concentrations and durations have been applied to many *Astragalus* species. It was determined that the application of sulfuric acid to *Astragalus cicer* and *Astragalus hamosus* seeds did not increase the germination rate of the seeds compared to the control, and it was not an effective method for breaking dormancy (Siles et al., 2016; Statwick, 2016). On the other hand, the application of sulfuric acid to the seeds of *Astragalus vulnerariae*, *Astragalus adsurgens*, *Astragalus penduliflorus*, *Astragalus maritimus*, *Astragalus verrucosus*, *Astragalus adscendens*, *Astragalus podolobus*, *Astragalus siliquosus*, *Astragalus hamosus*, *Astragalus lehmannianus*, *Astragalus cyclophyllon*, *Astragalus armatus*, *Astragalus sinicus*, *Astragalus cicer*, and *Astragalus arpilobus* determined that it is an effective method in breaking dormancy (Miklas et al., 1987; Kondo and Takeuchi, 2004; Isavand et al., 2005; Eisevand et al., 2006; Patane and Gresta, 2006; Keshtkar et al., 2008; Kim et al., 2008; Bacchetta et al., 2011; Long et al., 2012; Abudurehman et al., 2014; Tavili et al., 2014; Dilaver et al., 2017; Kheloufi et al., 2018; Dziurka et al., 2019).

## 3.3. Comparison of dormancy breaking applications

Each dormancy breaking application with the highest total germination rate was compared with the germination rates obtained in the control (25/15°C) application. Seed germination rates of gum tragacanth are given in Figure 2 according to the statistical analysis made to compare dormancy breaking practices with each other.

It was determined that the differences between different dormancy breaking treatments on total germination rate, normal germination rate, and abnormal germination rate of gum tragacanth were statistically significant. The highest total germination and normal germination rates of 50.7% and 38.7%, respectively, were obtained from gum tragacanth seeds kept in hot water for 2 minutes. Gum tragacanth

seeds kept in hot water for 2 minutes had 40% more total germination and 29.4% more normal germination than the control treatment.

Compared with the control application, it was determined that hydro priming, gibberellic acid, potassium nitrate, warm moist stratification, cold + warm moist stratification, and chemical scarification applications did not cause a significant change in the total germination rates of gum tragacanth seeds. On the other hand, it was determined that matrix priming increased the total germination rate of seeds by 12%, cold moist stratification 13.3%, warm + cold moist stratification 13.3%, cold water soaking 9.3%, and mechanical abrasion 9.3%, compared to the control application.

Compared with the control application, it was determined that hydro priming, gibberellic acid, potassium nitrate, warm moist stratification, cold+warm moist stratification, cold water soaking, mechanical scarification, and chemical scarification did not cause any increase in the normal germination rate of gum tragacanth seeds. On the other hand, matrix priming, which increased the total germination rate of gum tragacanth seeds slightly compared to the control application, caused an increase in normal germination rate as 10.7%, cold moist stratification 12.0%, and warm + cold moist stratification 8.0%.

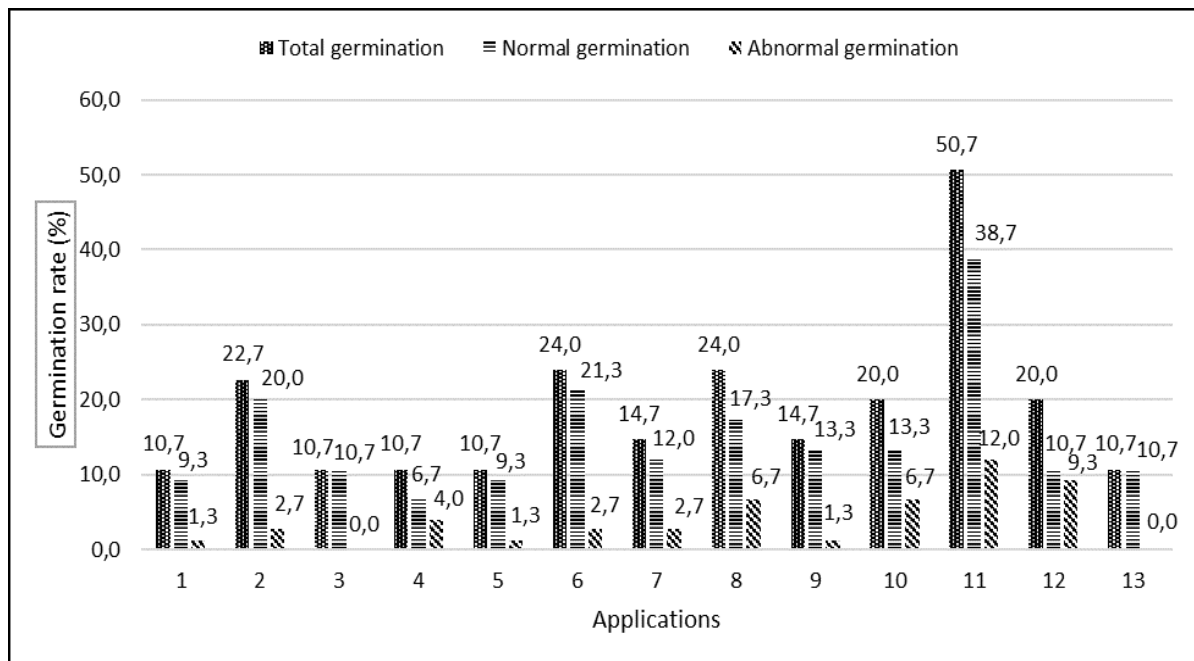


Figure 2. Effects of different dormancy breaking treatments on germination of *Astragalus gummifer* seeds (at 25/15°C temperature degrees). Treatment= 1: Control, 2: Matrix priming (36 hours), 3: Hydro priming (48 hours), 4: Gibberellic acid (500 ppm), 5: Potassium nitrate (%2), 6: Cold moist stratification (3 weeks), 7: Warm moist stratification (2 weeks), 8: Warm + cold moist stratification (1 week+4 week), 9: Cold + warm moist stratification (4 week+1 weeks), 10: Cold water (2 weeks), 11: Hot water (2 minutes), 12: Mechanical scarification (5 minutes), 13: Chemical scarification (20 seconds). (Total germination LSDs: 7.1\*\*, Normal germination LSDs: 6.0\*\*, Abnormal germination LSDs: 4.6\*\*), \*\*P < 0.01 is significant in the range of probability.

Compared with the control application, matrix priming, hydro priming, gibberellic acid, potassium nitrate, cold moist stratification, warm moist stratification, cold+warm moist stratification, and chemical scarification applications did not affect the abnormal germination rate of gum tragacanth seeds. Compared to the control application, it was determined that there was 5.4% more abnormal germination rate in the warm+cold moist stratification application, 5.4% in the cold water soaking, and 10.7% in the hot water soaking.

**Conclusion**

In the current study, the most suitable germination temperatures were found at 25/10°C and 25/15°C values for the seeds of the gum tragacanth bush. The highest dormancy breaking application was observed in gum tragacanth seeds kept in hot water for 2 minutes. According to the control application, matrix priming, cold moist stratification, warm moist stratification, warm+cold moist

stratification, cold+warm moist stratification, cold water soaking, hot water soaking, and mechanical scarification applications caused a slight increase in germination rates by breaking the dormancy state of the gum tragacanth seeds. On the other hand, it was determined that hydro priming, gibberellic acid, potassium nitrate, and chemical scarification applications did not have significant effects on breaking the dormancy of gum tragacanth seeds.

### Acknowledgement

This research was carried out with the support of Iğdir University Scientific Research Projects Coordination Unit as a project numbered 2019-FBE-L07.

### References

- Abd Kadir, S.L., Yaakob, H., & Mohamed Zulkifli, R. (2013). Potential anti-dengue medicinal plants: A review. *Journal of Natural Medicines*, 67, 677-689.
- Abudurehman, B., Liu, H., Zhand, D., & Guan, K. (2014). Identification of physical dormancy and dormancy release patterns in several species (Fabaceae) of the cold desert, north-west China. *Seed Science Research*, 24, 133-145.
- Açıkgöz, M.A., & Kara, Ş.M. (2019). Effect of Various Pretreatments on Germination of Turkish Endemic *Achillea gypsicola* Hub.-Mor. Species under *In Vivo* and *In Vitro* Conditions. *Journal of the Institute of Science and Technology*, 9(4), 2321-2329.
- Agh, K., Esmaceli, M.M., Hossinimoghaddam, H., & Mostafalo, H. (2017). The effect of different treatments on seed dormancy and germination of *Astragalus podolobus*. *Nova Biologica Reperta*, 4, 147-154.
- Agrawal, P.K., & Dadlani, M. (1995). Techniques in seed science and technology. Second Edition. South Asian Publishers Limited, India
- Akkurt, M., Keskin, N., Shidfar, M., & Çakır, A. (2013). Effects of some treatments prior to stratification on germination in Kalecik Karası (*Vitis vinifera* L.) seeds. *Journal of the Institute of Science and Technology*, 3(4), 9-13.
- Albrecht, M.A., & Penagos, Z.J.C. (2012). Seed germination ecology of three imperiled plants of rock outcrops in the southeastern United States. *Journal of the Torrey Botanical Society*, 139(1), 86-95.
- Arbabian, S., Moghanloo, M., & Majd, A. (2009). Seed dormancy breakage methods in the endangered species *Astragalus fridae* Rech. *The Quarterly Journal of Biological Sciences*, 2(4), 45-50.
- Avşar, D.M., & Ok, T. (2009). Some fruit and seed characteristics of Kadıncık shrub (*Flueggea anatolica* Gemici) population in the Tarsus region of Mersin. *Faculty of Forestry Journal Süleyman Demirel University*, A (1), 1-7.
- Bacchetta, G., Fenu, G., Mattana, E., & Pontecorvo, C. (2011). Ecological remarks on *Astragalus maritimus* and *A. verrucosus*, two threatened exclusive endemic species of Sardinia. *Acta Botanica Gallica*, 158(1), 79-91.
- Bagheri, S.M., Keyhani, L., Heydari, M., & Dashti, R.M.H. (2015). Antinociceptive activity of *Astragalus gummifer* gum (gum tragacanth) through the adrenergic system: A in vivo study in mice. *Journal of Ayurveda and Integrative Medicine*, 6(1), 19-23.
- Budge, C., Carryer, J., & Boddy, J. (2012). Learning from people with chronic pain: Messages for primary care practitioners. *Journal of Primary Health Care*, 4(4), 306-312.
- Cavieres, L.A., & Almedia, A.S. (2018). Assessing the importance of cold-stratification for seed germination in alpine plant species of the high-andes of central chile. *Perspectives in Plant Ecology, Evolution and Systematics*, 30, 125-131.
- Çolak, Ö.F. (2011). Investigation of dormancy breaking methods in *Saponaria halophila* Hedge & Hub. - Mor. Seeds. *Selçuk University Institute of Science*, Master's thesis.
- Demir, U., & Keskin, B. (2016). Determination of changes occurring annual nutritional contents of gum tragacanth (*Astragalus gummifer* L.). *Journal of the Institute Science and Technology*, 6(4), 121-125.

- Dilaver, Z., Mirzapour, M., & Kendir, H. (2017). Breaking seed dormancy and micropropagation of perennial vulneraria milkvetch (*Astragalus vulnerariae* DC.). *Acta Scientiarum Polonorum Hortorum Cultus*, 16(4), 79-88.
- Dinç, M., Aytaç, Z., & Doğu, S. (2013). A new species of *Astragalus* (Fabaceae) from Turkey. *Turkish Journal of Botany*, 37, 841-846.
- Dziurka, K., Skrzypek, E., & Dubert, F. (2019). Breaking seed dormancy of *Astragalus penduliflorus* Lam. *Polish Botanical Society*, 88(1), 3617.
- Eisvand, H.R., Arefi, H.M., & Afshari, R.T. (2006). Effects of various treatments on breaking seed dormancy of *Astragalus siliquosus*. *Seed Science and Technology*, 34, 747-752.
- Erken, S., & Kaleci, N. (2010). Determination of germination characteristics of yellow gentian (*Gentiana lutea* subsp. *symphyandra*) seeds under controlled conditions. *Bahçe*, 39 (2), 17-26.
- Erken, K., Erken, S., Gülbağ, F., & Özzambak, M.E. (2014). The effects of different sowing time and pre-treatments on seedling emergence in *Chamaecytisus pygmaeus* (Wild.) Rothm. *Turkey 5th Seed Congress with International Participation*, Diyarbakır, pp: 615-619.
- Erkul, S.K., & Aytaç, Z. (2013). *Astragalus yukselii* (Leguminosae), a new species from Turkey. *Turkish Journal of Botany*, 37, 836-840.
- Fateh, E., Majnoun, H.N., Madah, A.H., & Sharifzadeh, F. (2006). Seed dormancy methods breakage in *Astragalus tribuloides*. *Iranian Journal of Rangelands and Forests Plant Breeding and Genetic Research*, 13(4), 345-360.
- Gökçöl, A., & Duman, İ. (2018). Determination of the effects of different seed treatments to improve the germination of Caper seeds. *Journal of Agriculture Faculty of Ege University*, 55(4), 433-440.
- Isavand, H.R., Madah, A.H., & Tavakol, A.R. (2005). Study of dormancy breakage and germination in seeds of *Astragalus siliquosus*. *Iranian Journal of Rangelands and Forests Plant Breeding and Genetic Research*, 13, 67-84.
- ISTA, 2017. International seed testing association. international rules for seed testing, Zurich, Switzerland. *Journal of Ethnopharmacology*, 96, 71-77.
- İkram, R.M., Tanveer, A., Ata, Z., & Saqib, M. (2014). Dormancy studies on *Euphorbia dracunculoides* and *Astragalus* spp.: major weeds of arid areas. *Planta Daninha*, 32(4), 747-753.
- Jaganathan, G.K., Li, J., Biddick, M., Han, K., Song, D., Yang, Y., Han, Y., & Liu, B. (2019). Mechanisms underpinning the onset of seed coat impermeability and dormancy-break in *Astragalus adsurgens*. *Scientific Reports*, 9(9695), 1-10.
- Jones, T.A., Johnson, D.A., Bushman, B.S., Connors, K.J., & Smith, R.C. (2016). Seed dormancy mechanisms in Basalt Milkvetch and Western Prairie Clover. *Rangeland Ecology and Management*, 69, 117-122.
- Keshtkar, A.R., Keshtkar, H.R., Razavi, S.M., & Dalfardi, S. (2008). Methods to break seed dormancy of *Astragalus cyclophyllon*. *African Journal of Biotechnology*, 7(21), 3874-3877.
- Keskin, B., & Temel, S. (2019). Changes in monthly macro-mineral content of the gum tragacanth (*Astragalus gummifer* L.) for the nutrition of ruminants. *Umteb 6. International Vocational and Technical Sciences Congress*, 11-12 April 2019, Iğdır-Türkiye. pp: 254-260, ISBN: 978-605-7875-36-5.
- Khan, I.A., Abourashed, E.A. (2010). Leung's encyclopedia of common natural ingredients used in food, drugs, and cosmetics Third Edition. John Wiley & Sons, Inc., Hoboken, New Jersey, ISBN 978-0-471-46743-4.
- Kheloufi, A., Mansouri, L.M., Bouafia, B., Khamari, Y., Kheloufi, H., & Bouguern, Y. (2018). Morphological characteristics and seed germination improvement of two ecotypes of *Astragalus armatus* Willd. subsp. *armatus* in Algeria. *Cercetări Agronomice în Moldova*, 51(4), 96-107.
- Kırmızı, S. (2017). Dormancy and germination requirements of five species from Brassicaceae. *Journal of the Institute of Science and Technology*, 7(2): 21-29.
- Kildisheva, O.A., Erickson, T.E., Merritt, D.J., Madsen, M.D., Dixon, K.W., Vargas, J., Amarteifio, R., & Kramer, A.T. (2018). Do abrasion- or temperature-based techniques more effectively relieve physical dormancy in seeds of cold desert perennials? *Rangeland Ecology and Management*, 71(3), 318-322.
- Kim, S.Y., Oh, S.H., Hwang, W.H., Kim, S.M., Choi, K.J., & Kang, H.W. (2008). Physical dormancy in seeds of Chinese milk vetch (*Astragalus sinicus* L.) from Korea. *Korean Journal of Crop Science*, 53, 421-426.

- Kondo, T., & Takeuchi, S. (2004). Breaking seed dormancy and growth after germination of *Astragalus adsurgens* (Leguminosae), a rare species in Hokkaido. *Journal of the Japanese Society of Revegetation Technology*, 29(4), 495-502.
- Lagha, G.M., Hekmatshear, H., & Ferahvash, F. (2001). The correlation of temperature and salinity variations on seed germination of two *Atriplex* species of Azarbaijan. *Journal of Agricultural Sciences*, 6(4), 33-46.
- Lee, K.C., Chiu, T.T., & Lam, T.H. (2007). The role of fear avoidance beliefs in patients with neck pain: Relationships with current and future disability and work capacity. *Clinical Rehabilitation*, 21, 812-821.
- Long, Y., Tan, D.Y., Baskin, C.C., & Baskin, J.M. (2012). Seed dormancy and germination characteristics of *Astragalus arpilobus* (Fabaceae, subfamily Papilionoideae), a central Asian desert annual ephemeral. *South African Journal of Botany*, 83, 68-77.
- Maesaroh, S., & Demirbağ, N.Ş. (2020). Pretreatment Effect on Amelioration of Seed Germination of Zollinger's Indigo (*Indigofera zollingeriana* Miq.). *Yüzüncü Yıl Üniversitesi Tarım Bilimleri Dergisi*, 30(1), 1-8.
- Miklas, P.N., Townsend, C.E., & Ladd, S.L. (1987). Seed coat anatomy and the scarification of Cicer Milkvech seed. *Crop Science*, 27, 766-772.
- Molnár, V.A., Sonkoly, J., Lovas-Kiss, Á., Fekete, R., Takács, A., Somlyay, L., & Török, P. (2015). Seed of the threatened annual legume, *Astragalus contortuplicatus*, can survive over 130 years of dry storage. *Preslia*, 87, 319-328.
- Moradi, A., Ghanbari, A., Rashad Mohassel, M.H., & Izadi Darbandi, E. (2016). Investigations on the cardinal temperatures for germination of *Alhaji pseudalhagi*. *Journal of Plant Protection*, 29(2), 283-290.
- Obalı, A. (2009). Researches on the germination biology of Russian Thistle (*Salsola kali* subsp. *ruthenica* (Iljin) Soo.) seeds. *Selcuk University Institute of Science*, Master Thesis, Konya.
- Okay, Y., & Günöz, A. (2009). The effects of some applications on seed germination of endemic *Centaurea tchihatcheffii* Fisch. et Mey. in Gölbaşı province. *Tarım Bilimleri Dergisi*, 15(2), 119-126.
- Onursal, C.E., & Gözlekçi, Ş. (2007). The effects of some pre-sowing treatments on seed germination percentage and duration of sandal wood (*Arbutus andrachne* L.) trees. *Mediterranean Agricultural Sciences*, 20(2), 211-218.
- Özden, E., Kapçak, D., Memiş, N., & Demir, İ. (2018a). The effects of priming altered in relation to initial seed quality and sowing environment in onion cultivars. *The Turkey 6. Seed Congress with Internatinal Participation*, 10-13 September 2018, Niğde
- Özden, E., Ermis, S., Sahin, O., Taskin, M.B., & Demir, I. (2018b). Solid matrix priming treatment with O<sub>2</sub> enhanced quality of leek seed lots. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*, 46(2), 371-375.
- Pandita, V.K., Ananad, A., Nagarajan, S., Seth, R., & Sinha, S.N. (2010). Solid matrix priming improves seed emergence and crop performance in okra. *Seed Science and Technology*, 38(3), 665-674.
- Patane, C., & Gresta, F. (2006). Germination of *Astragalus hamosus* and *Medicago orbicularis* as affected by seed-coat dormancy breaking techniques. *Journal of Arid Environments*, 67, 165-173.
- Pearson, T. (2015). *Astragalus peckii* population establishment: developing protocols for seed germination, seedling cultivation, and planting. Honors Scholar thesis, Oregon State University.
- Rao, N.K., Roberts, E.H., & Ellis, R.H. (1987). The influence of pre and post-storage hydration treatments on chromosomal aberrations, seedling abnormalities, and viability of lettuce seeds. *Annals of Botany*, 60, 97-108.
- Schnadelbach, A., Barbosa, L.V., Ruiz, C., Pita, J.M., & Garcia, F.P. (2016). Dormancy breaking and germination of *Adenocarpus desertorum*, *Astragalus gines-lopezii* and *Hippocrepis grosii* (Fabaceae) seeds, three threatened endemic Spanish species. *Seed Science and Technology*, 44: 1-14.
- Siles, G., Torres, J.A., Valenzuela, L.R., & Fuentes, A.G. (2016). Germination trials of annual autochthonous leguminous species of interest for planting as herbaceous cover in olive groves. *Agriculture, Ecosystems and Environment*, 217, 119-127.

- Statwick, J.M. (2016). Germination pretreatments to break hard-seed dormancy in *Astragalus cicer* L. (Fabaceae). *Peer J.*, 4, e2621.
- Tajbakhsh, M., Ghiyasi, M., Amirnia, R., Danesh, Y.R., & Najafi, S. (2014). Studing the effects of drying and non-drying of hydroprimed of *Nigella sativa* seed's germination. *Turkey 5<sup>th</sup> Seed Congress with International Participation* Diyarbakır, pp: 478-481.
- Tavili, A., Mirdashtvan, M., Alijani, R., Yousefi, M., & Zare, S. (2014). Effect of different treatments on improving seed germination characteristics of *Astragalus adscendens* and *Astragalus podolobus*. *Journal of Rangeland Science*, 4(2), 110-116.
- Tilki, F. (2005). The effects of stratification, storage and temperature on the germination of *Fraxinus ornus* L. seeds. *Anadolu University Journal of Science and Technology*, 6(2), 191-195.
- Tuncer, B. (2019). İn Vitro Kültür Koşullarında Yabani Ravent (*Rheum ribes* L.) Tohumlarında Çimlenme ve Çıkış. *Yüzüncü Yıl Üniversitesi Tarım Bilimleri Dergisi*, 29(3),425-432.
- Zhou, J., Kulkarni, M.G., Huang, L.Q., Guo, L.P., & Staden, J.V. (2012). Effects of temperature, light, nutrients and smoke-water on seed germination and seedling growth of *Astragalus membranaceus*, *Panax notoginseng* and *Magnolia officinalis* — Highly traded Chinese medicinal plants. *South African Journal of Botany*, 79, 62-70.