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OVERCOMING SEED DORMANCY OF *Robinia pseudoacacia* L. AND *Ceratonia siliqua* L. SPECIES USING DIFFERENT PRETREATMENTS IN MALTA FOREST NURSERY – DUHOK

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ABSTRACT

Seeds of the legume species are characterized by physical hard seed coat dormancy. Several pre-sowing treatments were tested of these species to enhancing seed germination. This study was carried out in the Malta Forest Nursery to increasing the germination percentage of (*Robinia pseudoacacia)* and (*Ceratonia siliqua*) seeds. The experiment was laid out in randomized complete design with four replications. Both species, seeds were soaked in hot water at (80 C°) for 30, 60 and 90 min, and mechanical scarification; the second trait seeds treated with factorial experiment which were seeds of both two species soaked in sulfuric acid for 30, 60 and 90 min, and then immersed in tap water for 24, 48 and 72h. The results showed that the seeds of *Robinia pseudoacacia* species treated with hot water for 60 min were increased germination percentage (63.75%), and the same result obtained of *Ceratonia siliqua* seeds treated with hot water for 60 min (61%). In the factorial experiment, the seeds of *Robinia pseudoacacia* species were treated with Sulfuric acid scarification at 90 min, then immersed with tap water for 24h were the best result of germination percentage (42.5%). In *Ceratonia siliqu* seeds were treated with sulfuric acid at 60 min and soaking with tap water for 24h increased the seed germination percentage (65%).

KEYWORD: legume species, dormancy, sulfuric acid, mechanical scarification.

INTRODUCTION

Black locust (*Robinia pseudoacacia)* and Carob (*Ceratonia siliqua*) both are woody legume species where enhanced physical seed dormancy and their persistent soil profile (Richardson and Kluge 2008). These trees have been grown in the most countries of the Mediterranean region and has accordingly become one of the most important woody plant invaders and naturalized in Asia and Australia as well as in the Western and Central part of North America (Batlle and Tous, 1997; Holle et al. 2006).

These species are mainly propagated vegetatively in nature with high adaptability and nitrogen-fixing character (Ortiz et al. 1995 and Rice et al. 2004). The legume seeds have been prolonged by antimicrobial proteins accumulated in the seed tissues making them resistant to most pathogens (Talas-Ogras et al. 2005).

These species are economically important trees and they can be used in many tree-planting purposes: charcoal, wood production, soil erosion control, land reclamation and ornamental trees (Batlle and Tous, 1997). Both species were used in Mediterranean countries and they recommended for afforestation of degraded coastal areas threatened by soil erosion and desertification and often used in ornamental plantings and to regenerate disturbed sites in the parks and Agroforestry in nutrient deficient (Batlle and Tous, 1997).

Fruits and seeds in both of *R. pseudoacacia and C. siliqua* are low reproductive yield; most ovules fail to develop within the mutliseeded fruits of both species (Susko, 2006).

Seed germination is influenced by the environmental factors including temperature, water, oxygen, light and the chemicals (Tlig et al. 2008). The water content of the seed is a critical

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factor to trigger cell elongation and initiate radical emergence (Baskin and Baskin, 2014). Rising of temperature from (40 up to 45°C) may be decreased germination percentage of carob species, but the optimum temperature is within (25-30°C) for germinability of carob seeds (Lamlom and Abdalrasol, 2016).

The two tree species (Black locust and carob) have different germination percentage in the nursery for planting purposes. They are useful for stands or forests on medium and poor quality sites. They are utilized for the production of fuel wood, fodder, poles and props, as well as soil erosion control and environmental improvement (Zeleznik and Skousen 1996).

The applications of pre-sowing methods, including acid and mechanical scarification are exogenous treatments have been used to breaking hard seed coat and improve seed germination (Bonner et al., 1994**;** Masaka and Yamada, 2009; Lamlom and Abdalrasol, 2016).

The last decades have been an increasing demand for woody legume species, plantations of these species in forestry expanded to using a much broader range of species, so that a wider set of sites is exploited or restored for economic and conservation purposes (Weber 2003).

Both tree species of Fabaceae family have hard and impermeable seed coats caused physically dormancy (Zoghi et al., 2011).

This study aimed to determine the best treatment for breaking dormancy and increasing seed germination of both legume species.

MATERIALS AND METHODS

The experiment was carried out in the field of Malta forest nursery- Duhok, General Directorate of Forestry and Rangeland (Duhok Province).

Fruits (Pods) containing mature seeds of Carob and Black locust were harvested after mature brown seeds in autumn (10 November, 2015). After the collection, seeds were extracted and cleaned manually and placed in paper bags, then stored dry under laboratory conditions at about 25ºC until the start of the experiments 20-March 2016. Seeds of both species were dipped into the water and floating seeds were removed.

Two experiments were conducted; the first one was carried out of both two species; by using with the following pre-sowing treatments:

- Control (untreated seeds).
- Mechanical scarification was performed manually with sand-paper.
- Soaking in Hot water at (80 C^{o}) for 30 min., 60 min and 90 min.

The second experiment was carried out factorial experiment with seeds of both two species soaking in Sulfuric acid (98%) for 30 min., 60 min., 90 min. and then immersion in tap water for 24, 48 and 72h (3*3 interaction).

In hot water, seeds were placed in a cloth bag and then dipped into hot water (80 C^{o}), after that immersion seed were allowed to cool water to laboratory temperature. After Sulfuric acid (98%) scarification for each duration, seeds were washed several times with water to remove acid.

The seeds were kept in pots (container) containing soft soil after treatments. The two experiments of both species were designed according to randomize complete design (CRD) with four replications 25 seeds were used for each replicate.

Before analysis the percentages were transformed to arcsine values. All data were analyzed using ANOVA model (SAS software, 2001), and the Duncan test was used for the comparison of the means between the different treatments.

RESULTS AND DISCUSSION

- The First Experiment:

The results of the effect of treatments had significant differences at 0.01 levels (Table 1). Means of seed germination percentage of *Robinia pseudoacacia* from the Duncan test showed that in test one was various significant among them. The highest germination percent (63.75%) was shown in hot water treatment for 60 min, while the lowest value was untreated seeds (control) (20.63%) (Table 2).This same result was reported with Bagas, et al. (2010) by using acid treatment to improve the germination percentage of black locust seeds; also results found by Saikou, et al.(2008) that treating *Acacia Senegal* with hot water for 10 – 40 minutes gave the best results. Pipinis, et al. (2011), were using hot water and Sulfuric acid on breaking seed coat of Tamarind and Acacia seeds which were increased seed germination clearly.

On the other hand, Gebre and Keram (2004) are used hot water treatment for increasing seed germination of *Cercis siliquastrum*, and Nasiriand Eisvand (2001) with *Albizia julibrissin* species.

The seed of Gleditsia triacanthos L. for soaking in hot water at 90 C° for 20 minutes was the best method for breaking seed dormancy (Ahmed, 2015).

Soften hard seed coat of *Robinia pseudoacacia* L. and improving seed germination which treated with hot water (Mitra, et al. 2013), and the same effect on *Gleditsia triacanthos* (Basbag, et al. 2010 and Vasichkina, et al. 2014; Poşta and Camen, 2015).

The seeds of *Ceratonia siliqua* species in comparison means of Duncan test; the highest germination was soaking in (60 mints) of hot water (61%), whereas the lowest value (11%) germination percentage was observed with seeds were mechanically scarified (Table 2). Similarly, these results were observed with Perez-Garcia (2009) who used different pre-sowing to improving the seed germination of carob and found the highest germination in Carbo (Tsakaldimi & Ganatsas (2001), Nasiri and Eisavand (2001) on *Albizia julibrissin* and *Ceratonia siliqua.*

Table (1): Variance of analysis of the germination percentage of *Robinia pseudoacacia* and *Ceratonia siliqua* seeds under different single treatments.

S.O.V.	d.f.	*Robinia pseudoacacia*			*Ceratonia siliqua*		
		Sum of squares	Mean of squares	F	Sum of squares	Mean of squares	F
Treatments	4	1663.53	415.88	10.41**	2246.89	561.72	21.46**
Error	15	539.26	39.95		392.55	26.17	
Total	19	2202.79			2639.44		

Table (2): The effect of various pre-sowing treatments on the germination percentage of *Robinia pseudoacacia* and *Ceratonia siliqua* seeds.

Single Treatments	*Robinia pseudoacacia*	*Ceratonia siliqua*
Control	20.63d	28c
Mechanical scarification	26.25c	11d
Soaking in Hot water (30 min)	33.75b	36b
Soaking in Hot water (60 min)	63.75a	61a
Soaking in Hot water (90 min)	30bc	40b

- The Second experiments (the interaction treatments):

The results of data analysis (Table 3) showed that the effect of Sulfuric acid treatment on seed germination percentage of both species of *Robinia pseudoacacia* and *Ceratonia siliqua* seeds was significant at a probability of 0.01. While the effect of soaking seeds in durations tap water is non-significant. Seeds of both species were soaked in sulfuric acid and then immersion in tap water for different duration showed highly significant differences in the probability of 0.01 (Table 3). Seeds of *Robinia pseudoacacia* in 90 min of Sulfuric acid treatment gave highest germination percentage (30%) compared with 30 min of sulfuric acid (12.5%) because the interaction of treatments may be decreasing germination percentage (Table 4) compared with the effect of the single factor, because these species just external dormancy causes for killing embryo of the seed. Seeds of *Robinia pseudoacacia* species were immersed of tap water for 24, 48 and 72h were non-significant differences (20.83%, 20% and 22.5%) respectively (Table 4). The highest seed germination (42.5%) was observed when seeds treated with sulfuric acid at 90 min and soaked with one day of water (Table, 4). From the same table and same species, the lowest percentage of germination (10%) was found in both interaction of 60 min in sulfuric acid with one day water and 30 min with one day tap water.

When the seeds of *Ceratonia siliqua* species treated in sulfuric acid at 60 min gave highest germination percentage (47%) compared with 90 min and 30 min (43.33% and 28%) sequentially (Table 4). While seeds of this species treated with tap water for one, two and three days were non significant differences (40%, 41.33% and 37%) sequentially (Table 4).

From the same table (4), the highest seed germination (65%) was found when seeds treated with sulfuric acid at 60 min and soaking in water for one day. While the lowest percentage of germination (22%) was found in both interaction of 30 min in sulfuric acid with three days water (Table 4).These results are agreeing with other researchers, such as (Goor and Barney 1968; Karschon 1960), where the seeds of *Ceratonia siliqua* impermeability seed coat were soaked in sulfuric acid for 1 hour and then in tap water for 24 hours have been increasing seed germination. In addition, Saim and Derya (2014) were recommended the best treatment for overcoming seed dormancy of *Ceratonia siliqua* seeds was by immersing in sulfuric acid for 30 minutes, and then soaking in water for 2 days. Merou, et al. (2011) concluded that the broken seed dormancy and increasing seed germination of *Albizia julibrissin* seeds were soaking in sulfuric acid for 2 hours and immersion in 48h of tap water. Similarly, Piotto and Di Noi (2003) have shown that the Carob seeds had been dipped in sulfuric acid and then washed in water, which was increasing seed germination. The most effective method for increasing seed germination of *Ceratonia siliqua* was showed in sulfuric acid for 30 minutes and immersing seeds in hot water for 60 minutes (Mansour 2013; Zaen El Deen, et al. 2014).

Table (3): Variance analysis of germination percentage of *Robinia pseudoacacia* and *Ceratonia siliqua* seeds under different interaction treatments.

S.O.V.	d.f.	*Robinia pseudoacacia*			*Ceratonia siliqua*		
		Sum of squares	Mean of squares	F	Sum of squares	Mean of squares	F
H_2SO_4	2	875.46	437.73	33.73**	922.23	461.11	16.33**
Water/hours	2	20.02	10.01	0.77	47.95	23.97	0.85
H_2SO_4 X Water	4	1723.06	430.76	33.19**	1208.31	302.07	10.70**
Error	27	350.39	12.977		762.24	28.23	

Table (4): The effect of interaction pre-sowing treatments on the germination percentage (%) of *Robinia pseudoacacia* and *Ceratonia siliqua* seeds.

H_2SO_4	*Robinia pseudoacacia* WATER			Effect of H_2SO_4
	24h	48h	72	
30 min	10c	15c	12.5c	12.5c
60 min	10c	12.5c	40ab	20.83b
90 min	42.5a	32.5b	15c	30a
Effect of water	20.83a	20a	22.5a	

H_2SO_4	*Ceratonia siliqua* WATER			Effect of H_2SO_4
	24h	48h	72h	
30 min	24d	38bc	22d	28b
60 min	65a	37bc	39bc	47a
90 min	31cd	49b	50b	43.33a
Effect of water	40a	41.33a	37a	

CONCLUSION

Pre-sowing treatments of hot water at 60 min. are increasing the maximum number of germination percentage for both species. In factorial interactions, both long and short duration of sulfuric acid (30 or 90 min) decreases the germination percentage of *Ceratonia siliqua*, while, the long duration of sulfuric acid increase seed germination of *Robinia pseudoacacia*. In addition, the moderate duration interaction of both factors we obtain a moderate germination percentage.

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زالبون لسەر دەمێ خاڤبونا توڤێ دارێين روبينيا و سيراتونيا بكارئينانا سەرەدەرێن بەروەختێن جورا و جور ل نەمامگەها مالتايێ.

پوختە

توڤێ پرانيا جورێن خيزانا باقلوكا يێ ژتوڤەكێ رەق يێ دورمانسى چەندين سەرەدەرين بەرى توڤكرنى هاتينە ئەنجامدان لسەر ڤان جوران ژبو باشكرنا شينبونا توڤى. ئەڤ ڤەكولينە ل نەمامگەها مالتايێ ژبو زێدەكرنا رێژا شينبونا توڤێ روبينيا و سيراتونيا هاتنە كرن و تاقيكرن ژبو شروڤەكرنا ئەنجاما برێكا شێوازى **CRD** يێ شروڤەكرنا ئەنجامدان ودوباركرنا چارجارا بو سەر دەرئينانا شينبونا توڤێ. شێوازێن بەر بەلاڤ فاكتەرى تاك وەكو سەرەدەريكرنا ب ئاڤا گەرم بو ماوێ **60** خولەكا بويە ئەگەرێ زێدەكرنا شينبونا توڤێ روبينيا برێژێن **(63.75%)**, و هە روەها بو توڤێن سيراتونيا ب رێژبيا **61%** بدەست ڤە هات. وهەمان ئەنجام بو هەردوو جورێن توڤان ب شێوازێ فاكتوريال بدەست ڤە هاتن. ژبو توڤێن روبينيا يێن سەرەدەريا ئەسيدێ سولفوريك دگەل هێلان بو ماوێ **90** خولەكان ب سەرەدەرى ب ئاڤا حەنەفيێ بو ماوى **24** دەمژمێرا باشترين ئەنجامێن شينبونا توڤى ب رێژا **42.5%** هاتە توماركرن. توڤێن سيراتونيا ئەوێن ب ئەسيدێ سولفريك سەرەدەرى هاتبونە كرن بو ماوێ **60** خولەكان و ئەوێن ب ئاڤا حەنەفيێ بو ماوێ **24** دەمژمێرا ب شيوەيەكێ بەرچاڤ رێژا شينبونى باشترلێ بو.

التغلب على كسر طور السكون لبذور اشجار الروبينيا و الكاروب باستخدام المعاملات الاولية المختلفة في مشتل غابات مالطا

الخلاصة

ان بذور معظم انواع البقوليات تتصف بطور السكون الخارجي بغلاف بذري صلب. ولتحسين انبات هذه الانواع تتعرض الى العديد من المعاملات الاولية لكسر سكون هذه الانواع. حيث اجريت هذه الدراسة في مشتل غابات مالطا لزيادة نسبة انبات بذور (الروبينيا) و (الكاروب). صممت التجربة باستخدام تصميم القطاعات العشوائية الكاملة لمختلف المعاملات وباربع مكررات. وتم استخدام المعاملات التقليدية حيث اعطت معاملة البذور الروبينيا بالماء الحار لمدة **60** **(63.75%)**، وكذلك بالنسبة الى بذور الكاروب التي عوملت بنفس المعاملة **(61%)**. ونفس النتائج لبذور كلا النوعين التي عوملت بعوامل متداخلة، حيث وجد ان افضل نسبة انبات **(42.8%)** لبذور الروبينيا التي غمرت بحامض الكبريتيك المركز لمدة **90** دقيقة وابقائها في الماء العادي لمدة **24** ساعة. بينما بذور الكاروب التي غمرت بحامض الكبريتيك لمدة **60** دقيقة وبقائها في الماء العادي لمدة **24** ساعة حصلت على افضل نسبة انبات **(65%)**. التجربة تشير الى امكانية الاستفادة من النقع بالماء الحار والحامض لكسر طور السكون في الروبينيا والكاروب على التوالي.