

IMPACT OF VARIOUS SCARIFICATION METHODS AND OSMOTIC STRESS ON *Gleditsia triacanthos* seeds

Mohamed TOUMI^{1,2}, Selma BARRIS^{1,3}

1. Department of Natural and Life Sciences, Faculty of Sciences, University of Algiers 1, Benyoussef Benkhedda, 2, Rue Didouche Mourad 16000, Algiers, Algeria.

2. Laboratory of Plant Pathology and Molecular Biology, National School of Agricultural Sciences, Rue Hassan Badi, El Harrach, Algiers, Algeria

3. Laboratory of Biology and Physiology of Organisms, Faculty of Biological Sciences, University of Science and Technology, Houari Boumediene, BP 32 El Aaliya, 16111, Bab-el-Zouar, Algeria, Algeria

INTRODUCTION & AIM

Desertification is one of the most worrying environmental issues of the 21st century. It is a real problem for arid and semi-arid environments, where rainfall is variable and plants are subject to periods of drought of varying length. In Algeria, almost 500,000 hectares of land in steppe areas are undergoing desertification, and more than 7 million hectares are directly threatened by the same process (MATE, 2002). The surface area of degraded rangelands stood at 7.5 million hectares in 1995 and this degradation, attributed in particular to stressful environmental conditions, wind erosion, land clearance and overgrazing, is having increasingly harmful ecological and economic effects. Improving the plant cover of the degraded forest massifs of the Saharan Atlas, by planting diversified species adapted to these areas, will help to combat desertification more effectively (Bouhata and Bensekhria, 2021).

Gleditsia triacanthos L. is a nitrogen-fixing species in the legume family. It is a fast-growing tree that can develop on degraded soils because of its highly efficient root system, which can spread over a radius of 15 metres around the trunk on dry land (Bross, 2000). Resistant to many environmental stresses (low and high temperatures, drought, atmospheric pollutants), *G. triacanthos* has great potential for restoring vegetation and regenerating ecosystems.

To grow this tree on a large scale, you need to have a good command of germination conditions and plant rearing. Germination itself is regulated by the genotypic characteristics of the seed, but also by environmental factors, in particular the availability of water in the soil. These factors are thought to play an important role in optimising germination capacity (Li et al., 2019). *Gleditsia triacanthos* L seeds are coated with a hard integument that acts as a protective layer but also slows down the entry of water, preventing the seed from soaking. Pre-treatment of the seeds is therefore necessary to obtain good imbibition and a better germination rate (Nedjraoui and Bédrani, (2008).

The objectives of the present work are, on the one hand, to determine the various types of effective pretreatments for improving the germinative capacity of *G. triacanthos* seeds that are within the reach of forestry institutions and, on the other hand, to study the germinative capacity of these seeds under the effect of osmotic stress induced by different concentrations of PEG₆₀₀₀ in order to understand the ability of the seeds to germinate under conditions of water shortage and possibly to determine the tolerance threshold of these seeds to this constraint in nature.

METHOD

➤ Scarification methods.

The integuments of legume seeds, especially those of *Acacia ehrenbergiana*, are often rigid and impede water infiltration into the seed. Consequently, a scarification is essential to ensure optimal germination. Several types of scarifications are carried out to remove the impermeability of the seed coat and allow the metabolic activities necessary for germination to resume.

➤ Application of osmotic stress.

Following mechanical scarification, the seeds are immersed in beakers containing distilled water as a control, along with varying concentrations of PEG₆₀₀₀ (0%, 5%, 10%, 15%, 20%, 25%, and 30%) to induce osmotic stress. The seed counting is conducted at 8-hour intervals over a period of 15 days as part of the experiment. Each point is denoted by the mean of 20 seeds, and 3 replicates are conducted for each point.

RESULTS & DISCUSSION

1. Effect of scarification on the germination of *G. triacanthos* seeds

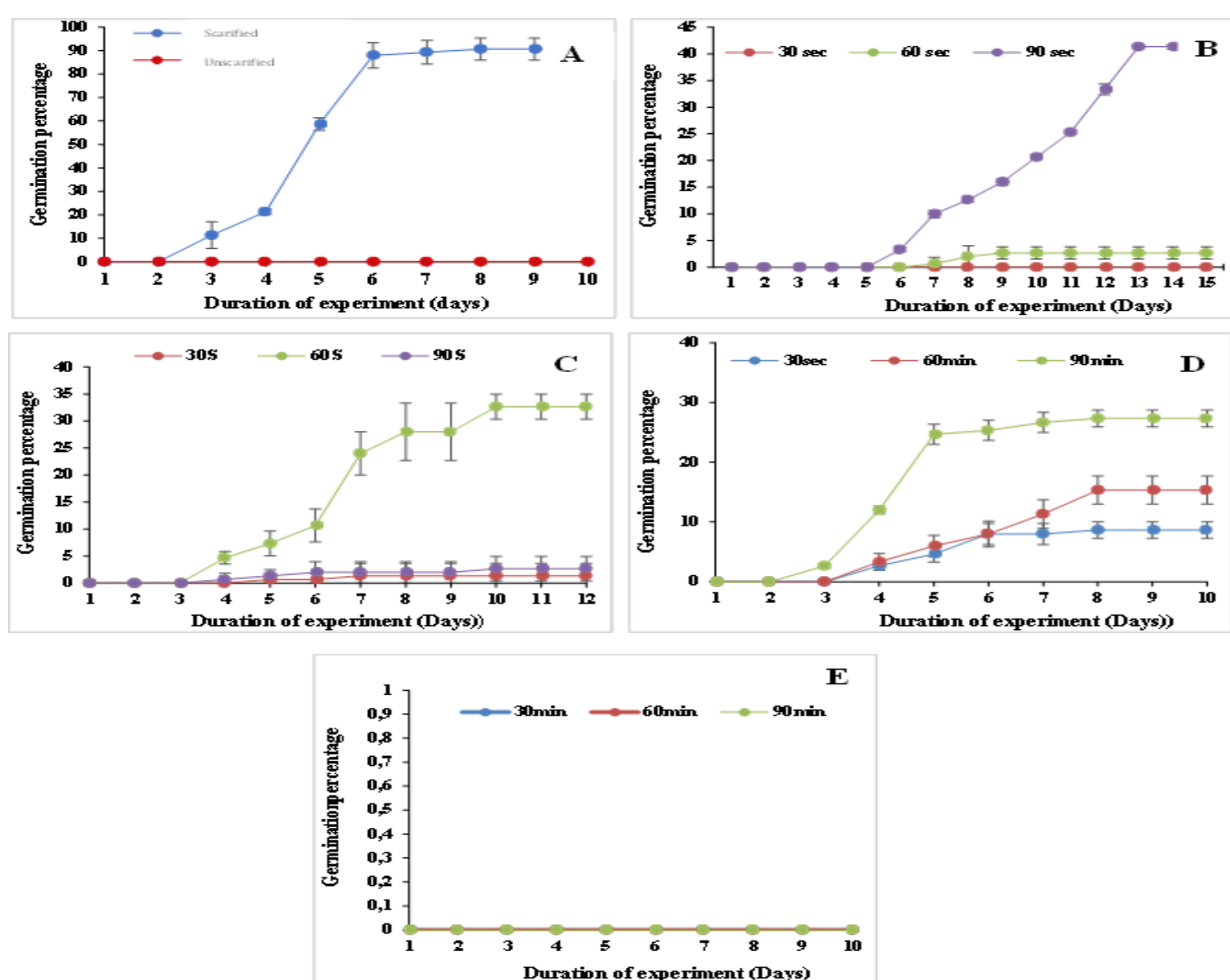


Figure 1 : Percentage of germination of *Gleditsia triacanthos* L. seeds as a function of time (days): manual scarification (A), microwave scarification (360 Watts) (B), microwave scarification (600 Watts) (C), scarification with sulphuric acid (H₂SO₄) (D) and scarification with boiling water (E).

A lag time of at least 24 hours was observed for the various pre-treatments. This time is needed to initiate the metabolic process required for germination.

The best germination rates were observed in seeds that had been scarified by hand, by sulphuric acid (90 minutes), by microwave scarification at 360 watts for 90 minutes and by microwave scarification at 600 watts for 60 minutes. On the other hand, scarification with boiling water does not allow seeds to germinate (Figure 1).

An optimal germination rate close to 100% was reached after 8 days of experimentation in seeds that had undergone manual scarification (Figure 1 A), which represents the best germination rate of the treated seeds. For the other pre-treatments, the germination rate varied according to the duration and intensity of the pre-treatment, but did not exceed 45% (Figure 1).

The scarification results obtained for this study are in agreement with those of Toumi et al. (2017) on *Robinia pseudoacacia* seeds microwave pre-treatment, considered to be the most suitable method for scarification pre-treatment of *R. pseudoacacia*. L. However, treatment of *G. triacanthos* seeds with boiling water appears to be ineffective in eliminating tegumental inhibition. boiling water did not give any germination for our species this would probably be due to the long exposure time which would have destroyed the seed embryo, soaking in hot water might have given better results as stated by Neffati and al, (1996,) in their study of *M. arborea* seeds, which were also very sensitive to chemical treatment with sulphuric acid. After 4 minutes in the presence of H₂SO₄, the seed embryo was probably damaged by the acid, which greatly reduced the germination percentage

2. Effect of osmotic stress on the germination kinetics of *Gleditsia triacanthos* L. seeds.

Seeds soaked in a 20% PEG₆₀₀₀ solution do not germinate. This concentration completely inhibits germination. For PEG₆₀₀₀ concentrations of 0%, 5%, 10% and 15%, the seeds show sigmoid germination kinetics, with a latent phase, an exponential phase and a plateau (Figure 2).

The latency time increased from 24 h to 48 h for 5% and 10% PEG respectively. seeds begin to germinate after 6 days when the PEG concentration is 15%.

A maximum germination rate of 90.33± 4.66% was observed in the control seeds after 8 days of experimentation. A 22.66% drop in the maximum germination rate was observed with 5% PEG. At 10% PEG, only 26±12.16 % of total germination was obtained. the maximum germination rate at 15% is insignificant, not exceeding 3% (Figure 2).

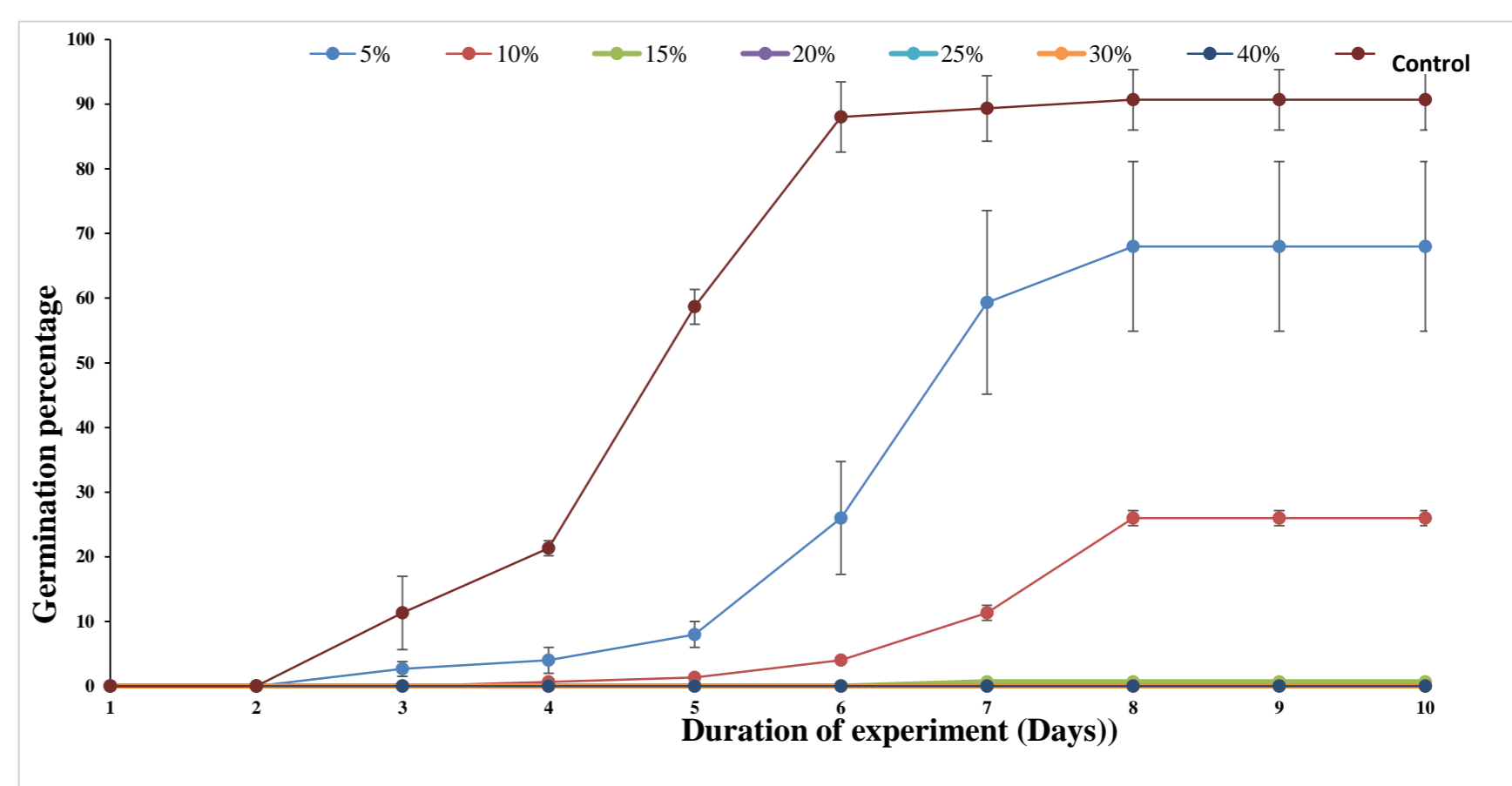


Figure 2. Evolution of the germination rate of *Gleditsia triacanthos* L. seeds subjected to pre-treatment with different concentrations of PEG₆₀₀₀ as a function of experiment days.

The osmotic stress, which was induced by PEG₆₀₀₀. This highly hydrophilic polymer retains a large quantity of free water in a given solution, thereby reducing the osmotic pressure of the external environment.

The presence of PEG₆₀₀₀ in the imbibition solutions considerably reduced the germination rate of *G. triacanthos* seeds. This reduction could be due to a hydration deficit in the seeds following a high osmotic potential resulting in an inhibition of the degradation of reserves which leads to the radicle leaving the integuments and consequently a delay in seed germination (Zhang and al., 2011). An alteration in the enzymes and hormones found in the seed following the lack of water could also explain this reduction in germination

CONCLUSION

For *Gleditsia triacanthos* seeds, the best scarification methods are manual scarification and microwave scarification at 360 watts for 90 seconds. These methods facilitate the penetration of water and oxygen into the seed

The addition of PEG₆₀₀₀ reduce the quantity of free water in the imbibition solutions, which limits seed imbibition, thus reducing the germination percentage of *G. triacanthos* seeds.

This reduction was greater at 15% PEG. The germination capacity of *Gleditsia triacanthos* is more affected by osmotic stress than those given in the literature for dryland species such as *Acacia raddiana* (Abbasdokhta et al, 2014) and *Robinia pseudoacacia* (Toumi et al, 2017)

FUTURE WORK / REFERENCES

- Abbasdokhta, H., Gholamia, A., 2014. Asgharia, H. Desert19 27-34.
Brosse, J., 2000. Larousse des Arbres et Arbustes. Paris, Larousse pp 351.
Bouhata, R., Bensekhria, A., 2021. Adaptation of MEDALUS method for the analysis depicting desertification in Oued Labiod valley (Eastern Algeria). Arab. J. Geosci. 14, 365. <https://doi.org/10.1007/s12517-021-06679-2>
Li B, Wang J, Yao L, Meng Y, Ma X, Si E, Ren P, Yang K, Shang X, Wang H (2019). Halophyte Halogeton glomeratus, a promising candidate for phytoremediation of heavy metal-contaminated saline soils. Plant Soil 442:323–331
MATE, Ministère de l'Aménagement du Territoire et de l'Environnement, 2002. Rapport annuel du Plan National d'Actions pour l'Environnement et le Développement Durable (PNAE-DD) p 140.
Nedjraoui, D., & Bédrani, S. (2008). La désertification dans les steppes algériennes: causes, impacts et actions de lutte. Vertig O, 8(1), 1-15. <https://doi.org/10.4000/vertigo.5375>
Neffati, M., Behaeghe, T., Akrim, N., et Floc'h, L. Viabilité des semences de quelques espèces pastorales steppiques tunisiennes en rapport avec les conditions de leur conservation. Ecologia mediterranea, (1996). 22 (1), 39-50.
Silvertown, J. Seed ecology, dormancy and germination: a modern synthesis. Am. J. Bot86(6) (1999) 903-905.
Toumi, M., Barris, S., Seghiri, M., Cheriguene, H., & Aid, F. (2017). Effet de plusieurs méthodes de scarification et du stress osmotique sur la germination des graines de Robinia pseudoacacia L. Complexus Rendus Biologies, 340(5), 264-270. <https://doi.org/10.1016/j.crv.2017.02.002>