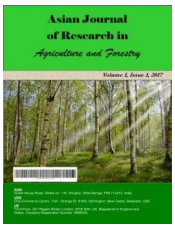

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Breaking Secondary Dormancy in Sesbania (*Sesbania sesban)* Seeds

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Authors' contributions

This work was carried out in collaboration between both authors. Author KKL designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author FNWM managed the analyses of the study and managed the literature searches. Both authors read and approved the final manuscript.

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ABSTRACT

Aims: To investigate the efficacy of various dormancy breaking methods on Sesbania seeds.
Study Design: Complete Randomized Design with 5 treatments and 4 replicates of 100 seeds each for the treatments.
Place and Duration of Study: Department of Seed, Crop and Horticultural Science and duration of the study was 2 months.
Methodology: Sesbania seeds were subjected to the following methods; mechanical scarification, soaking in hot water (67°C) for 15 minutes, soaking in concentrated H_2SO_4 for 15 seconds and leaching. A control experiment where the seeds were not subjected to any treatment was also prepared. The seeds were then set up in Petri dishes up lined with 3 moist filter papers substratum for each method. The Petri dishes were then put in a growth chamber set at a temperature of 24°C and 70% relative humidity. Seeds that germinated every day were noted.
Results: Dormancy breaking methods differed significantly from the control regarding the number of seeds that germinated at P≤0.05. Leaching method (77.8%) hot water (69.5%) Mechanical scarification (62.3%) and acid scarification (64.5%) were higher than for the control (51%).

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Conclusion: Leaching was shown to be the best method to alleviate dormancy in the Sesbania seeds. Soaking in hot water, mechanical and acid scarifications also significantly increase germination.

Keywords: Dormancy; Sesbania sesban; leaching; germination.

1. INTRODUCTION

Sesbania is an important legume crop which is grown by most large scale and small scale farmers as a forage crop. The crop is high biomass producing crop having a high protein and fiber making it an important crop in livestock diet. The crop is also used as human food (leaf and seeds), green manure, source of fuel, fences and has medicinal value in India [1]. Sesbania is a fast-growing, perennial legume tree, reaching a height of up to 8m. It has a shallow root system and its stems may reach 12 cm in diameter. The leaves are pinnately compound with 6 to 27 pairs of leaflets. Inflorescences are 30 cm racemes bearing 2 to 20 yellow flowers with purple or brown streaks. Fruits are linear or slightly curved pods up to 30 cm long. The pods of the crop contain 10 to 50 seeds [2].

The crop is grown by many farmers in Kenya but with limited success in its establishment on the farm due to both biotic and a biotic factors hindering its production. Among the main constraints is seed dormancy. Dormany refers to a state in which seeds are prevented from germinating even under environmental conditions normally favorable for germination. This results in poor crop stand that leaves gaps in the field. Dormancy breaking methods therefore enhance germination by removing the restrictive factors inside and outside of the seed [3]. Sesbania seeds are believed to have seed coat imposed dormancy hence increasing the period taken for the seed germination which takes 14 to 24 days [4]. Dormancy in Sesbania seed is also caused by presence of waxy substance on the seed coat of Sesbania seed. This type of dormancy is known as physical dormancy [5].

Seed coat-imposed dormancy is caused by a continuous layer of tightly packed palisade cells which is a major barrier to water uptake and gaseous exchange. The seed coat may act as a mechanical restraint to growth of the embryo or as a barrier to photoreception [6]. The seed coat can also be a source of germination inhibitors or prevent germination inhibitors from escaping from inner seed tissue.

Various seed pre-treatment have been used in dormancy alleviation in seeds. Soaking the seeds in water softens the seed coat and allows the emergence of the embryo [3]. Soaking seeds in hot water provides a suitable temperature range for enzymes that catalyse reactions that break down the seed coat. Breakdown of the seed coat allows the emergence of the embryo, water and gas uptake [7]. Leaching washes away germination inhibitors and hence allows germination to occur. Mechanical and acid scarification physically destroys seed coat hence allowing absorption of water and gases for germination to occur [8]. This study seeks to investigate which seed pretreatments can effectively break dormancy in Sesbania seeds.

2. MATERIALS AND METHODS

The experiments were conducted in the University of Eldoret, School of Agriculture and Biotechnology Seed Science laboratory in May 2017. Freshly harvested Sesbania seeds were obtained from VI Arboretum Kitale, Kenya and used in this study.

2.1 Dormancy Breaking Seed Treatments

The seeds were subjected to mechanical scarification, acid hot water and leaching treatments. The seeds were replicated four times in each treatment. 100 seeds were used in each replicate [9]. The seeds for all the treatments were sterilized using 75% alcohol to remove any pathogen and insect pest. The seeds were placed on a clean container with distilled water to wash off all the traces of alcohol. For mechanical scarification treatment 400 seeds were rubbed using sand paper for 20 revolutions. For the acid treatment 400 seeds were placed in 90% sulphuric acid for 15 seconds. These seed were then washed with plenty of distilled water. For the hot water treatment 400 seeds were placed in 67°C hot water for 15 minutes [10]. For leaching treatment 400 seeds subjected to running water for 7 hours, then soaked for 6 hours [11]. The control setup seeds were not subjected any treatment. Seeds from all the treatment and control setup

were then sown on 3 moist filter papers placed in petridishes. Since the seeds used in each treatment were 400 four petridishes were used for each treatment. All the petri dishes were then placed in a growth chamber set at 24°C and 70% relative humidity as recommended [9]. The substratum was kept moist by addition of small amount of water every 2 days. Number of seeds that germinated was counted and recorded daily. Data on number of seeds that germinated for each treatment was subjected to Analysis of variance (ANOVA) to find out which methods differed significantly from the control. The percent germination values for the methods were separated using DMRT.

3. RESULTS AND DISCUSSION

All of the treatments significantly differed from the control at P≤0.05. The most effective method of breaking dormancy was leaching followed by hot water, acid treatment and finally scarification using sand paper (Table 1). Leaching washes away enzyme inhibitors found on the seed coat or aleurone layer (under the seed coat) allowing germination to occur [11,12]. This implies that dormancy in this species is to some extent caused by germination inhibitors attached to the seed coat. After leaching the seeds were soaked for 6 hours. This improved germination as water softens the seed coat hence allowing the embryo to emerge [3]. Leaching requires a lot of water hence the water used should be recycled to avoid wastage of water. In one study dormancy in Sesbania seeds was broken by subjecting the seeds to soaking in hot water (80°C) for 8 minutes then selecting the ungerminated seeds and scarifying them mechanically [13]. This led to almost 100% germination. This implies that that use of 2 methods concurrently may result in high germination.

Hot water also improved germination which implies that inactive enzymes present in the seeds were activated and enabled germination to take place. Water also softened the seed coat hence allowing the embryo to emerge. However, it has been noted that the hot water method have both the negative and positive effects on the seeds [14]. The positive effect is that when used well and with a compatible seeds, it result in a high germination. The negative result is that there is a high risk of losing the seed especially when the water is too hot. In the dormancy alleviation on the *Sesbania sesban* seeds, there was a high germination percentage due to the action of the hot water [10]. The other problem with hot water treatment was that there were a high number of abnormal seedlings. This might be as a result of denaturing of the enzymes on the seed endosperm or due to the destruction of the important germination hormones in the seeds [3].

Seed coat-imposed dormancy was also present because acid and sand paper scarification resulted in improved germination. Other authors have found mechanical and acid scarification to quite effective in breaking dormancy in the Fabaceae family that Sesbania sp. belongs [15,16]. The relatively low (%) in sand paper scarification in the present study may be due to the degree of intensity during the rubbing of the seeds. The relatively low (%) also in acid scarification in the present study may be due to the short duration of 15 seconds which the seeds were soaked. Other authors have soaked for 15 minutes or 40 minutes [15].

Table 1. Means of the germination percentage of *Sesbania sesban* seeds treated with leaching, hot water, sand paper and acid treatment

Treatments	Percentage germination	Duncan multiple range test (DMRT)
Sand paper	62.25	b
Hot water treatment	69.50	d
Leaching	77.75	e
Acid treatment	64.50	c
Control	51	a
Mean	65	
Probability	<.001	
S.E.D	0.964	
S.E	1.363	
%CV	2.1	

Means having the same letters are not significantly different at P≤ 0.05 (DMRT)

4. CONCLUSION

Seed dormancy in Sesbania was broken by leaching, hot water, mechanical and acid scarification. This implies that dormancy in Sesbania was due to a combination of factors which were hard seed coat, presence of enzyme inhibitors and inactive enzymes.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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