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Germination Studies in *Rhanterium epapposum* Oliv

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Abstract: Conservation of native plants are possible by their utilization in urban landscape, as indigenous plant species have evolved and adapted to the local harsh climatic conditions over the years and they are more likely to function adequately under the local climate as opposed to exotic plants. As Kuwait's native plants are threatened in their natural habitat and have begun disappearing at an alarming rate, their use in landscape projects will help in conserving biodiversity and heritage. Dormancy is widespread among desert shrubs, with only a few species having non-dormant seeds. Effective dormancy breaking treatments are important to develop mass propagation techniques in native plants like *Rhanterium epapposum*. In this germination study, effects of dry heat (50°C) and/or various concentrations (500, 750 and 1000 ppm) of gibberellic acid (GA_3) on seed germination of *Rhanterium epapposum* were assessed. Treating the seeds with 750 ppm of GA_3 was found to be effective for *Rhanterium epapposum* with 71% of germination. The next best treatment combination (52%) was exposure to dry heat (50°C) for ten days followed by a treatment with 500 ppm GA_3 for 24 hrs.

Key words: Urban landscape · Greenery development · Dormancy · Germination study · Conservation of native plants

INTRODUCTION

Kuwait has a typical desert climate with extreme temperatures, minimal and variable rainfall, intense sunshine and frequently occurring dust storms. The climate is characterized by extremely hot summers, with daytime temperature exceeding 50°C and winter, cooler and at times wet, with temperature sometimes falling below 4°C [1]. The rainy season extends from October to May. The mean annual rainfall is 113mm [2]. The total conventional freshwater resources available in Kuwait are six million cubic meter per year, while the total water demand has exceeded 350 million m^3/ yr in 2000 [3]. Native soil in Kuwait is mostly sandy in texture with high infiltration rate and is calcareous in nature [3].

Perennial vegetation cover is sparse, usually less than 10% [4]. Although introduced ornamental plants are necessary in urban and sub-urban landscape for diversity, they rely on high levels of nutrients and water. Conservation of native plants are possible by their utilization in urban landscape, as indigenous plant species have evolved and adapted to the local harsh climatic conditions over the years and they are more likely to function adequately under the local climate as opposed to exotic plants. As Kuwait's native plants are threatened in their natural habitat and have begun disappearing at an alarming rate, their use in landscape projects will help in conserving biodiversity and heritage [5]. Shrubs, in particular, are significant and have good potential for urban landscape application [6]. Unfortunately, efficient propagation and establishment techniques that are crucial for both conservation of native plants and large-scale use in landscape programs are currently unavailable.

Accordingly, 'arfaj' (*Rhanterium epapposum* Oliv, Family: Asteraceae) was selected for its aesthetic appeal, flower color and potential to adapt to urban landscape conditions. It is the national flower of Kuwait. It is a C_3 desert shrub that can form monotonous stands covering vast areas of north-eastern Arabia [4]. It grows as a perennial woody shrub approximately 80 cm high with many stems branching out from the base. The leaves are small and narrow and in late spring, it is covered with straw-yellow flowers about 1cm wide. It flowers from April to May and produces numerous fruits (Fig. 1) which forms in late spring and falls off the branches after maturity. It accumulates under the shrub and remains dormant until

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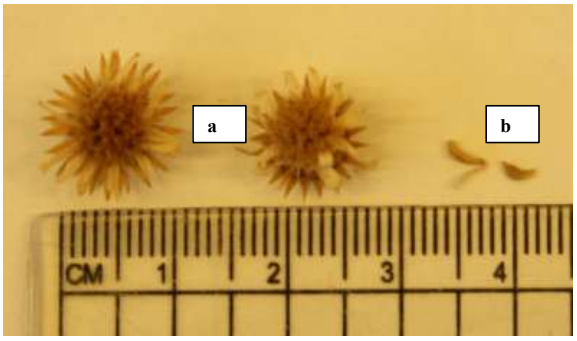

Fig. 1: *Rhanterium epapposum* (a) capituli; (b) seeds

favorable conditions for germination prevail. Each fruit (capitulum) contains about 6-8 seeds that are transported by wind or water. The total area covered by this plant is about 2% of Kuwait. It is considered one of the main desert forage plants for camels and sheep [4].

Germination is a critical phase in the life cycle of a plant, particularly in arid regions where seeds can be confronted by severe environmental constraints such as high temperatures, drought and high levels of salinity as a result of fluctuating soil moisture [4]. Baskin and Baskin [7] Stated that dormancy is widespread among desert shrubs, with only a few species having non-dormant seeds. In Kuwait and adjacent areas, *Rhanterium epapposum* is found mainly in deep sandy soils [8]. The capitulum is the unit of dispersal for *Rhanterium epapposum*, either being blown short-distances by the wind or transported over long distances by zoochory [4]. For germination to take place, the capituli must be positioned with their basal part on the ground, in an upright position [9]. He also demonstrated that dormancy over a number of years is a typical feature of the seeds. Often, several seeds germinate at once in a capitulum and it appears that mortality of seedlings due to intra-specific competition may not be the norm [9]. The regeneration of *Rhanterium epapposum* from seed has not been observed, although this must be the natural method of propagation [8]. Seed germination under simulated environmental conditions ranged from 0-4.69% [10]. It is difficult to propagate and establish this species by vegetative means. It can be propagated by tissue culture method [11] but, the production cost will be higher than the seedling method. In view of the above facts, studies were conducted to determine the effects of pre-treatments on germination of *Rhanterium epapposum* seeds.

MATERIALS AND METHODS

Seeds: Seeds used in this study were obtained from seed bank of Kuwait Institute for Scientific Research (KISR) in November 2007. These seeds were collected from Kuwait desert in 2004. Prior to their use in the experiment, seed viability was determined using Triphenyl Tetrazolium Chloride (TTC) Test [12]. In this test, viable embryos are stained pink due to the reduction of 2, 3, 5- TTC by reparative activity in the cell. For this, 100 seeds (four replicates of 25 seeds each) were soaked in distilled water overnight, excised to expose the embryos, placed in a petri dish, soaked in 0.1% TTC solution, covered the petri dish with aluminum foil and kept for 24 hours at 25±1°C. Following the treatment, seeds were washed thoroughly with distilled water to remove excess stain and examined under the microscope.

Germination Studies: Seeds stored at room temperature were kept in an oven at 50°C for 10 days or 20 days and treated with various concentrations of gibberellic acid (GA_3) for 24 hours. Treatments included exposure to dry heat at 50^0C for 10 or 20 days and/ or pre-sowing soaking in GA_3 solution (0, 500, 750 or 1,000 ppm) for 24 hours. There were twelve treatments, which were replicated five times in a completely randomized design. Each replication contained 20 seeds. The control seeds were not subjected to either heat or GA_3 treatment. The total germination was calculated when no more seeds germinated. Pre-treated seeds were sown in agricultural soil medium in Petri dishes and maintained in growth chamber at 18°C with 60% humidity.

The data were analyzed using R analysis procedure of Analysis of Variance (ANOVA) and Dunkan's Multiple Range Test to ascertain the significant differences among treatments [13, 14].

RESULTS AND DISCUSSION

Viability of seeds used in these studies was found to be 88%. The highest germination of (Table 1) 71% was obtained from seeds that were treated with 750 ppm GA_3 for 24 hours (Fig. 2). The next best treatment combination (52%) was the ten days of dry heat (50°C) exposure followed by a treatment with 500 ppm GA_3 for 24 hrs. When the seeds were subjected to 10 days heat treatment, increase in the concentration of GA_3 had a negative effect on the germination. Exposure of seeds to dry heat alone

Table 1: Effect of Dry Heat (50°C) Exposure and GA_3 on Germination of *Rhanterium epapposum* Seeds

	Germination (%) [i]			
	GA_3 Treatment (ppm)			
Heat Treatment	0	500	750	1000
RT [ii]	50±10.75 de [iii]	21±3.28 abc	71±5.36 e	44±2.60 cd
50°C 10 Days	27±5.39 abcd	52±5.39 de	38±3.89 bcd	27±6.71 abcd
50°C 20 Days	7±2.28 a	17±9.11 ab	28±10.99 abc	22±9.93 abc

[i] Seeds with 2 mm or longer radicle or shoot are considered as germinated.

ii RT- Room Temperature (25°C)

[iii] The means followed by the same letter are not statistically different at $p \leq 0.001$

Fig. 2: *Rhanterium epapposum* germination

for 10 or 20 days also did not improve the germination. Untreated seeds resulted in 50% of germination. Extended duration (20 days) of dry heat exposure diminished the germination. The improvement in germination by pre-treatment was significant at $P \leq 0.001$ level.

The germination percentage of 71% in the present study is a clear improvement over the previous reports where only 4.69% [10] germination was recorded under simulated environmental conditions. The fact that, pre-treatment with GA_3 increased the germination percentage suggests that the dormancy in this species may be due to physiological factors. GA_3 treatment alters hormonal balance in favor of promoters to trigger germination process and shorten time needed for germination [15]. Results of this study are important as desert rehabilitation and biodiversity conservation is crucial to prevent the extinction of valuable native plants in Kuwait [2, 5].

CONCLUSIONS

Effective dormancy breaking treatments are important to develop mass propagation techniques in native plants like *Rhanterium epapposum* which exhibit dormancy to adapt to the harsh environmental conditions in the desert. Pre-treating the seeds with 750 ppm GA_3 was effective in breaking seeds dormancy and in significantly improving the germination in *Rhanterium epapposum*.

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