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Effect of High Temperatures on Seed Germination of *Solanum aculeastrum*

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Abstract: *Solanum aculeastrum* is a medicinal plant that showed low seed germination under laboratory conditions. The objective of this study was to establish whether germination could be improved by the exposure of its seeds to high temperatures such as those registered on surface soils during natural fires. Our results indicated that temperatures of 100 and 120°C applied to green mature seeds of *S. aculeastrum*, for 45 to 60 min may break their innate dormancy, thus stimulating their subsequent germination to more than 85%. Germination in dry seeds, however, showed very low germination when subjected to high temperatures. Where the effect of smoke was tested on the germination of the green mature and dry seeds of *S. aculeastrum*, no stimulation of germination was observed. The ecological implications of these observations are discussed.

Key words: Fire, high temperature, *S. aculeastrum,* seed germination, smoke, Solanaceae

INTRODUCTION

High temperatures, especially when generated by fire, are an ecological factor which plays an important role in the evolution of ecosystems (Trabaud, 1980). It may affect plant regrowth potential and seed germination (Gibson *et al*., 1990; Canadell *et al*., 1991). Several authors (Auld, 1986; Tarrega *et al*., 1992; Bradstock *et al*., 1992; Harrington and Driver, 1995) observed that after fire, the seed germination rates of many species increased. In Africa, bush fire is a wide-spread feature of the dry savannas and contributes largely to changes in the composition of vegetation communities (Gillon, 1983; Greerling, 1985; Monnier, 1990; Kozlowski, 2000). A number of reasons have been given for the enhancement of seed germination by high temperatures caused by fire. Among these are a reduction of inhibitory substances in soil and litter (Keeley *et al*., 1985; Gonzalez-Rabanal and Casal, 1995), chemical stimulation from charred wood (Keeley *et al*., 1985), fracturing of hard seed coats, stimulation of seed embryos, desiccation of seed coats and release of ethylene and ammonia in the plant-derived smoke (Keeley, 1987; Brown, 1993; Brits *et al*., 1993; Baskin and Baskin, 1998).

Solanum aculeastrum Dunal, a member of the family Solanaceae, is frequently used in herbal medicine. It is a multi-branched shrub, 1-5 m high, heavily armed with large prickles and is wide-spread in South Africa. Local healers use the extremely bitter fruits for the treatment of various diseases in humans and domestic animals (Hutchings *et al.,* 1996). Both mature and immature berries contain the poisonous alkaloid, α-solanine (Hutchings *et al*., 1996). Other bioactive compounds that have been isolated from this plant include solaculine A (Wanyonyi *et al*., 2002) from the root bark and solamargine, beta-solamarine, solasonine and solasodine from the fruits (Drewes and Van Staden, 1995; Wanyonyi *et al.,* 2002). The fresh and boiled berries are used as a cure for jigger wounds and gonorrhoea respectively (Agnew and Agnew, 1994). Traditional healers of the Eastern Cape Province in South Africa also use the plant for the treatment of neoplasm, particularly breast cancer.

S. aculeastrum is increasingly being exploited in South Africa due to its widespread medicinal use. Thus, there is an urgent need to conserve this species. Reinforcement of wild plant populations using individuals raised *exsitu* is considered a valid means of reducing the risk of extinction of overexploited plant populations (Bowes, 1999). If the future demand for this plant is to be met, it is imperative that this species be domesticated and commercially cultivated. Techniques for efficient low-cost cultivation practices are determined, to a large extent, by the germinability of the seeds. However, each species has peculiar requirements for seed germination as a result of adaptive radiation into patchy and changing environments (Schütz and Milberg, 1997). Many species respond well to sterile *in vitro* conditions, including a nutrient rich medium and also phytohormone supply (Fay, 1992; Pence, 1999). However, for some species, these techniques are not necessary for successful seed germination and would waste resources if

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used. Appropriate techniques must be selected based on the requirements of each species (Fay, 1994; Benson *et al*., 2000).

Requirements for seed germination in *S. aculeastrum* have not been investigated. This study was designed to evaluate the *exsitu* requirements for optimal seed germination in *S. aculeastrum*. Specifically, the aim of this project was to investigate the effect of high temperature and smoke on the germination of its seeds under controlled environmental conditions.

MATERIALS AND METHODS

Seed collection: Two types of berries of *S. aculeastrum* were collected from plants naturally growing in the wild at Kayalethu village in the Eastern Cape Province of South Africa (latitudes 30°00'-34°15'S and longitudes 22°45'-30°15'E), the green looking mature berries and the yellow, dry-looking mature fruits. For the purpose of this report, they are referred to as green mature seeds and yellow dry seeds respectively. The plant was identified at the Botany Department, University of Fort Hare and a voucher specimen (Vedic Med 2005/16) was prepared and deposited in the Griffen Herbarium of the University. The seeds were removed from both groups of berries and used immediately for the germination experiments. This work was carried at the Botany Department, University of Fort Hare, during February, 2005 to April, 2005.

Treatment with high temperature: Three replicates of 50 seeds each were exposed to 60°C for 30, 45 or 60 min, respectively. Prior to the treatment, the seeds were placed in Petri dishes containing two discs of Whatman No. 1 filter paper. The filter papers were moistened with distilled water to ensure adequate moisture for seed germination. The petri dishes containing the seeds were then left and observed on the side benches at room temperature. Seeds were considered to have germinated when the radicle had grown 2 mm (Come, 1970; De Villalobos *et al*., 2002). The number of the germinated seeds were recorded every 24 h over 21 days (De Villalobos *et al*., 2002). Same treatments were repeated at 80°C, 100°C and 120°C.

Treatment with smoke: Instant smoke (Baxter *et al.*, 1994; Sparg *et al*., 2005) was obtained from Kirstenbosch, National Botanical Garden, Cape Town, South Africa. Each smoke-impregnated paper was soaked in 50 mL water for 5 min to allow the diffusion of smoke into the water. Seeds were then placed in the smoke-impregnated water for 12, 24 and 48 h, before they were transferred into Petri dishes, containing two discs of Whatman No. 1 filter paper. The filter papers were moistened as needed with distilled water to ensure adequate moisture for seed germination. Three replicates of 50 seeds each were used for each treatment. Seeds were considered to have germinated when the radicle had grown 2 mm. The number of the germinated seeds were recorded every 24 h over 21 days. Untreated seeds were used as control.

Germination percentage was calculated as the proportion of germinated seeds within a replicate. Significance differences within the means of the treatments and the controls were calculated using the LSD statistical test (Steel and Torrie, 1960).

RESULTS AND DISCUSSION

Significant differences ($p<0.05$) in percentage germination was observed between temperatures and the period of exposure of the seeds of *S. aculeastrum* (Fig. 1). The germination of green mature seeds was significantly higher than the germination of dry seeds when exposed to 100°C and 120°C for 30, 45 and 60 min. In all temperature treatments, the highest germination was recorded in green mature seeds exposed to 100° C for 45 min and 120° C for 45 and 60 min, the germination being one hundred percent (Fig. 1). Higher germination percentages of green mature seeds were also observed when they were exposed to 100°C for 30 and 60 min (81.33 and 85.33%, respectively). Dry seeds showed very minor responses in germination to the increased temperatures except when exposed to 120°C for 60 min (26% compared to the control). There are significant differences between germination percentages of control and green mature seeds at all temperatures (Fig. 1). It was also noted that there was a difference between the germination percentages of the green mature and dry seeds control treatments (3.2 and 0%, respectively), which might indicate that the seeds become dormant on ripening.

Our results indicate generally that high temperatures applied for a few minutes to green mature and yellow, dry seeds of *S. aculseastrum* may break their innate dormancy, allowing their subsequent germination (Fig. 1). The optimum temperatures needed for breaking the innate seed dormancy in *Acacia suaveolens* were 60-80°C for a range of exposure times of 1-360 min. Seeds exposed to 100°C for less than 2 h also broke dormancy (De Villalobos *et al*., 2002). At temperatures above 100°C, only short exposure times led to successful germination, while longer exposure caused seed death (Auld, 1986). Auld and O'Connel (1991) found that for 35 species of Leguminosae under study, seed dormancy was broken in over half the species by exposure to temperatures over

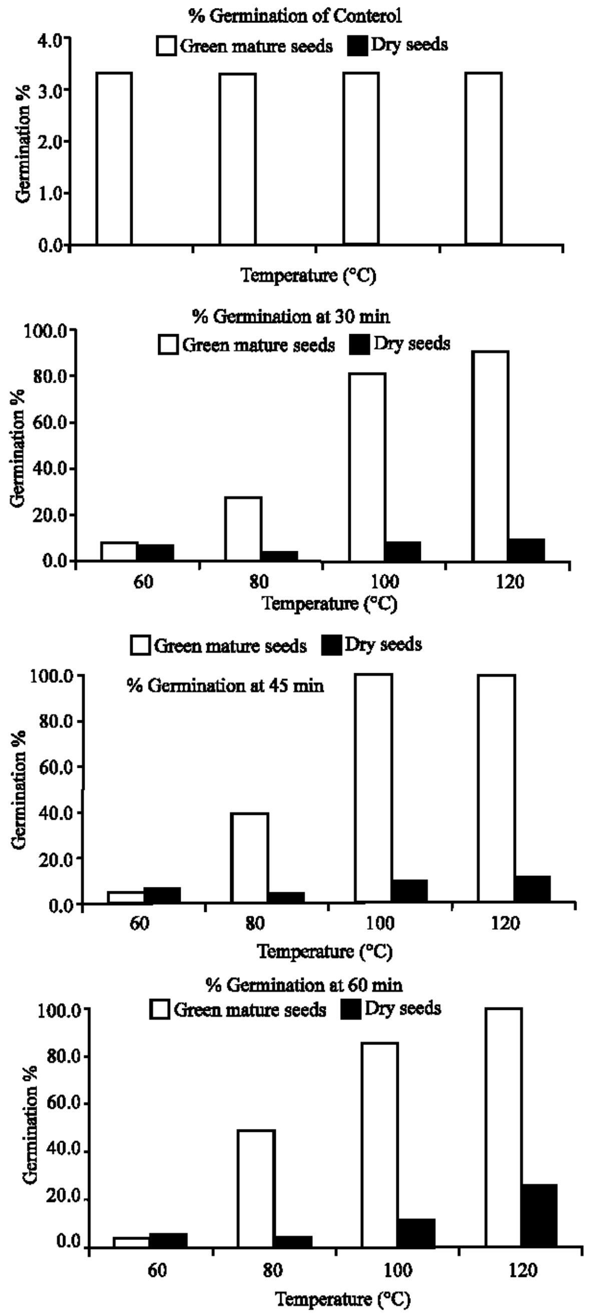

Fig. 1: Effects of the temperature and exposure time on seed germination of *S. aculeastrum*

60°C, whereas dormancy was broken in all those species with exposures above 80°C. Tarrega *et al.* (1992) reported that moderate heat treatments (70°C and 100°C) significantly increased the germination rate of *C. scoparius* and *G. florida* seeds. Where the effect of smoke was tested on the germination of the green mature and yellow-dry seeds of *S. aculeastrum*, no stimulation of germination was observed.

In generally, our laboratory results demonstrated that high temperatures might stimulate the germination of *S. aculeastrum* seeds. This might encourage, under favorable environmental conditions, the post-fire establishment of seedlings. However, plant response to fire depends on the interaction of several environmental and biotic factors. Therefore, more detailed studies on this subject should be conducted in the future.

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