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EFFECT OF PRE-TREATMENTS FOR ENHANCING THE GERMINATION OF *ADANSONIA DIGITATA* L. AND *COCHLOSPERMUM RELIGIOSUM* L.

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ABSTRACT

The present study deals with breaking the seed dormancy by various physical and chemical agents and establishment of seedlings of *Adansonia digitata* and *Cochlospermum religiosum*. In *A. digitata*, pre treatment with concentrated sulphuric acid for 12 hrs exhibited 68% germination which was further enhanced up to 90.67% by post treatment soaking in 0.1M glucose solution. *C. religiosum* exhibited 32% germination with pre treatment of sulphuric acid for 25 min and with post treatment soaking in 0.1M glucose solution showed 54.67% germination. Seedling survival was also found to be more in the combination treatment of sulphuric acid and 0.1M glucose solution (*A. digitata*- 81.33% and *C. religiosum*- 53.33%) than pure sulphuric acid treatment.

Key words: Adansonia, *Cochlospermum,* Germination, Survival rate, Agro-forestry, Seed dormancy.

Introduction

Adansonia digitata L. (Family-Bombacaceae) and *Cochlospermum religiosum* L. (Family-Cochlospermaceae) are multipurpose resourceful tree species, different parts of which are used as food, medicine, fuelwood, fibre, etc. Both the trees are considered as good agroforestry crops as almost all plant parts like leaves, bark, fruit pulp, fruit shell, seed, roots, flowers are commercially used. Medicinal properties of *A. digitata* and *C. religiosum* are exploited in treatments of various diseases like cough, dysentery, fever, chronic bronchial asthma, diarrhoea, dermatitis, etc. (Kirtikar and Basu, 1933) and cough, dysentery, diarrhoea, pharyngitis, gonorrhoea, syphilis, trachoma, etc. respectively (Warrier *et al.*, 1993). The fruit pulp of *A. digitata* is rich source of vitamin C which is approximately six times more than that of an orange (Vertuani *et al.*, 2002); while *C. religiosum* is planted near temples in India for its bright yellow flowers which are in high demand to be used as offerings to God and also for aesthetics (Swaminathan and Kochhar, 2003). Additionally gum and floss of this plant are commercially utilized.

Adansonia digitata and *C. religiosum* thrive well in hot and dry conditions of India and are best choice for waste and marginal lands. However, the status of distribution of both these tree species all over India appears to be sparse despite favourable conditions for their growth and propagation. Over exploitation for commercial purposes by village people without concern for their propagation and cultivation might have led to fast decline in their population. Attempts to grow these plants indicated that high degree of seed dormancy is exhibited in both the plants probably as a way of seed protection from inconsistent weather. Members of Bombacaceae and Cochlospermaceae show physical dormancy. In *A. digitata* the physical dormancy is attributed partly to the hard testa and partly to the pulp (Esenowo, 1991). *C. religiosum* shows physical dormancy due to hard seed coat (Baskin *et al.*, 2000).

In view of their importance in various industries like pharmaceutical, nutraceutical, cosmetics, dyeing, agro-forestry, etc., it is imperative to find methods of breaking seed dormancy and to establish feasible propagation technique for successful regeneration and large scale cultivation. Therefore, the objective of the study was to assess the effects of pre and post soaking seed treatments and physical factors like temperature, light, storage time and soil depth on seed germination and to achieve faster seedling establishment by different fertilizer treatments.

Material and Methods

Seeds were collected from dry mature fruits of *A. digitata* and *C. religiosum* plants growing in Nagpur and Kondhali region respectively. A mixture of sand, coco peat and vermicompost in 1:1:1 ratio was used for germination experiments. The seedlings were established in poly bags containing above mentioned germinating mixture and soil in 1:1 ratio.

For each treatment 25 seeds were soaked for 3 days (*A. digitata*) and 2 days (*C. religiosum*) including the time of treatment and sown in trays. The trays were kept

Germination of seeds of *Adansonia digitata* can be enhanced upto 90.67% by pre-treatment with conc. Sulphuric acid for 12 hrs followed by post treatment with 0.1 M glucose solution.

in daylight with ambient temperature 32±2°C and watering at 2 days interval for germination. Data for triplicate treatments was pooled after one month from the date of sowing.

Pre- treatments

To break the physical dormancy of seeds induced by hard seed coat, seeds were pretreated with concentrated sulphuric acid (*A. digitata*: 1, 3, 6, 12, 18, 24, 48 hrs; *C. religiosum*: 5, 10, 15, 20, 25, 30 min) and treatment was given by keeping the seeds in hot water at 60°C (*A. digitata:* 3, 6, 12, 18, 24, 48, 72 hrs; *C. religiosum*: 1, 3, 6, 12, 18, 24, 48 hrs). Mechanical scarification of the seeds was carried out by rubbing out water impervious brown cover with sand paper. Control was maintained without any pre-treatment.

Post treatments

Seeds treated with sulphuric acid for 12 hrs (*A. digitata*) and 25 min (*C. religiosum*) were further subjected to the following post treatments.

Chemical factors

Seeds were further soaked in Gibberellic acid-Loba Cheme (0.005%, 0.001% and 0.002%), market samples of Sodium chloride (0.1M, 0.5M, 1M and 2M) and Glucose (0.1M, 0.5M, 1M and 2M) for 3 days (*A. digitata*) and for 2 days (*C. religiosum*).

Physical factors

The pretreated seeds were independently subjected to different depths of sowing (2, 4 and 6cm) and temperatures (25±2°C and 32±2°C) and continuous light and dark conditions. To check the effect of storage duration on seed viability, seeds were categorized as harvested freshly (0yr), one year back (1yr old) and two years back (2yrs old) and sown in trays.

Fertilizer treatments

Eight days old established seedlings were irrigated twice by different fertilizer solutions like Urea (N 48%), Diammonium phosphate-DAP (N:P, 20:20) and NPK (N:P:K, 19:19:19) after eight days interval. For this total 0.7gm/ bag dose of each fertilizer for *A. digitata* and 0.3gm/ bag for *C. religiosum* was split in two applications. Data was recorded one month after treatment.

Results and Discussion

A one-way ANOVA was performed to compare the significance of different treatments by Graph Pad Prism Software. The significance level was tested using Post-hoc comparisons by Tukey's test.

Seed dormancy poses serious problems in propagation and cultivation of *A. digitata* and *C. religiosum*. Barrier effect on the germination could be due to physical or chemical characteristics of the seed coat and its impermeability to water, gases or solutes. Sulphuric acid has been used effectively in various plants (Aboutalebi *et al.*, 2012 and Gokturk *et al.*, 2012) to break hard seed coat induced dormancy. However, it exhibits great variations in its effect. In *A. digitata* and *C. religiosum*, concentrated sulphuric acid proved better means of breaking dormancy with 68% at $P<0.05$ level [$F_{(15,32)}=43.63$] and 32% germination at $P<0.05$ level [$F_{(14,30)}=10.93$] as against 0% and 1.33% in control respectively. However, the optimum time duration of sulphuric acid treatment in *A. digitata* was found to be much greater (12 hrs) than in *C. religiosum* (25 min). It can be contributed to very hard seed coat in *A. digitata*. Sulphuric acid is reported to exhibit great variation in its ability to break seed dormancy in *A. digitata* (Esenowo, 1991 and Danthu *et al., 1995*). No germination in control seeds (0%) was found to be in complete contradiction

Table 1 : Effect of different post treatments on the germination and survival rate of seeds of *Adansonia digitata*.

Post treatments		*Adansonia digitata*	
		Germination (%)	Survival rate (%)
Gibberellic acid	0.005 %	8±0.58	4±0.76
	0.01%	10.67±1.01	4±0.58
	0.02 %	33.33±1.18	30.67±1.26
NaCl	0.1 M	60±1.24	26.67±0.71
	0.5 M	65.33±1.08	60±1.29
	1 M	66.67±0.88	34.67±0.92
	2 M	72±0.76	14.67±0.62
Glucose	0.1 M	90.67±0.71	81.33±0.44
	0.5 M	90.67±0.62	78.67±0.71
	1 M	88±0.82	74.67±1.08
	2 M	86.67±0.71	65.33±0.71
Control	12hr H_2SO_4	68±0.82	33.33±1.18

Note: All means are significantly different at $P < 0.05$ having $F_{(11,24)}=23.25$ and $F_{(11,24)}=19.08$ for Germination and survival rate respectively.

Table 2 : Effect of different post treatments on the germination and survival rate of seeds of *Cochlospermum religiosum*.

Post treatments		*Cochlospermum religiosum*	
		Germination (%)	Survival rate (%)
Gibberellic acid	0.005 %	49.33±0.44	48±0.00
	0.01%	45.33±0.71	44±0.58
	0.02 %	38.67±0.44	36±0.58
NaCl	0.1 M	42.67±0.88	38.67±0.83
	0.5 M	26.67±1.01	26.67±1.01
	1 M	13.33±0.88	13.33±0.88
	2 M	8±0.58	8±0.58
Glucose	0.1 M	54.67±1.01	53.33±1.04
	0.5 M	52±0.00	52±0.00
	1 M	45.33±0.88	45.33±0.88
	2 M	14.66±1.08	5.33±0.71
Control	25 min H_2SO_4	32±1.10	32±1.10

Note: All means are significantly different at $P < 0.05$ having $F_{(11,24)}=9.13$ and $F_{(11,24)}=12.07$ for Germination and survival rate respectively.

with the result (57%) of Assogbadjo *et al*. (2010). Seeds of *A. gregorii* showed 100% germination when treated with sulphuric acid for 24 hrs (Turner and Dixon, 2009). This differential response might be due to extreme variations in edaphic as well as climatic factors to which the plants are exposed, time of fruit and seed maturation, etc. There are no reports on germination studies of *C. religiosum*, the present study being the first attempt to increase its germination rate.

Among different post treatments with Gibberellic acid, NaCl and Glucose all the concentrations of glucose significantly enhanced the germination percentage (maximum 90.67%) in *A. digitata* at P<0.05 level [$F_{(11,24)}=23.25$] (Table-1). In *C. religiosum*, enhancement was found in all concentrations of Gibberellic acid and lower concentrations of NaCl and Glucose which is significant at P<0.05 level [$F_{(11,24)}=9.13$] (Table-2). Post treatments not only induced higher germination but dramatic increase in survival rate of seedlings was also observed in *A. digitata* and *C. religiosum*. Maximum response (81.33% in *A. digitata* and 53.33% in *C. religiosum*) was obtained in 0.1M Glucose solution significant at P<0.05 level. Enhancement in germination capacity by post treatments with sugar and NaCl is in confirmation with the reports in *Asphodelus aestivus* by Ozturk and Pirdal (1986).

Seed viability is a key factor for propagation success. Prolonged post harvest storage many a times brings about certain biochemical changes making the seeds inviable (Igeleke and Omorusi, 2007). Germination process is greatly affected by the microclimatic and soil conditions like aeration, oxygen content or light and temperature. These factors vary as we go deep down in the soil (Glucu *et al*., 2010). Both *A. digitata* and *C. religiosum* exhibited better germination and seedling growth when sown 2cm deep and kept at 32±2°C temperature significant at P<0.05 level. Similar results were obtained in *Adansonia digitata* by Chia *et al.* (2008). Presence of light had a positive effect on germination and survival percentage.

In both the plants survival rate was found to be very low. Among many factors availability of primary nutrients like N, P and K promotes establishment and subsequent seedling growth. One month old seedlings of *A. digitata* irrigated with urea (source of N), DAP (source of N and P) and NPK 19:19:19 (source of N, P and K) exhibited good survival and better growth. However, in *C. religiosum* even low concentrations of fertilizers had adverse effect on the seedlings. This can be attributed to the high sensitivity of small and delicate seedlings and it is suggested that fertilizer application should be postpone for next one to two months in *C. religiosum*. These investigations have led to the establishment of an effective nursery protocol for mass propagation of otherwise difficult to propagate commercially important plants.

***एडनसोनिया डिजीटाटा* एल तथा *कोहिल्कोपर्सम रेलीजियोसम* के अंकुरण को बढ़ाने के लिए पूर्व उपचारों का प्रभाव**

आर.एन. गहाने तथा के.के. कोगजी

सारांश

वर्तमान अध्ययन का उद्देश्य विभिन्न भौतिक और रासायनिक उपायों से *एडनसोनिया डिजीटाटा* तथा *कोहिलोस्पमर्म रेलीजियोसम* के बीजो की सुप्तावस्था को तोड़ना तथा पौधों की स्थापना करना है। *ए. डिजीटाटा* में 12 घंटों तक सान्द्रित सलफ्यूरिक अम्ल से पूर्व उपचार करने पर 68% अंकुरण पाया गया जो उपचार के उपरांत 01 एम ग्लूकोज के घोल में डुबाकर रखने पर 90.67% तक बढ़ गया। *सी. रेलीजियोसम* 25 मिनट तक सल्फ्यूरिक

अम्ल से पूर्व और उपचारित करने पर 32% अंकुरण प्राप्त हुआ जो उपचार के पश्चात 0.1 एम ग्लूकोज के घोल में डुबाने पर 54.67% पाया गया है जो सल्फ्यूरिक अम्ल है 0.1 एम ग्लूकोज घोल के संयुक्त उपचार से (*ए. डिजीटाटा* –81.33% तथा *सी. रेलीजियोसम* 53.33%) शुद्ध सल्फ्यूरिक अम्ल की तुलना में अधिक था।

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