

GERMINATION IMPROVEMENT OF *REPTONIA BUXIFOLIA* (FALC) A. DC. WITH VARIOUS PHYSICO-CHEMICAL FACTORS

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Abstract

Reptonia buxifolia (Falc) A.DC., shows very poor regeneration in subtropical regions of Pakistan. The present study was undertaken to improve its germination under laboratory condition. Non-treated seeds exhibited 26% germination at 20 and 25 °C and 54% at 30 °C at pH 5–7. Seeds treated either alone with warm water, mechanical scarification, or coupled with soaking in water, KNO₃, and IAA gave over 90% germination at 20–30 °C. A 60–80% germination was achieved with acid scarification, IAA, KNO₃, sodium hypochlorite and acid scarification coupled with IAA at various temperatures tried. Large sized and surface sown seeds had better germinability at field capacity and in water-logged situation. Storage upto 12 months did not affect the germinability.

Introduction

Reptonia buxifolia (Falc) A.DC, is an important component of *Acacia-Reptonia*, *Acacia-Olea* or *Reptonia-Olea* forests in Pakistan. It is a source of fire-wood, fruit, animal fodder and other domestic utility. Biotic factors have almost eliminated this tree in the Attock-Nizampur valley (Ilahi and Hussain, 1987) and Khyber Hills (Naqvi, 1974). Although such deformed trees shed fruits, yet hardly one can see any seedling around them. It appears that there is some problem with the germination of this tree which might be responsible for its poor regeneration. The germination requirement of different species varies. Deforestation hampers the regeneration and self-perpetuation of the susceptible plants. The ecological study helps to identify suitable conditions for enhancing the germination. The germination of *Acacia* (Shaikh, 1986; Qadir and Lodhi, 1971), *Capparis* (Qaiser and Qadir, 1971), Juniper (Shaikh, 1982), Pines (Ilahi and Perveen, 1982), *Erianthus* (Ilahi et al., 1987) and many other economically important species (Ilahi and Hussain 1987) has been reported. The present study, therefore, reports methods for prompt and promoted germination of *Reptonia buxifolia* under laboratory condition. The findings will help in afforestation/reforestation of the degraded *Reptonia*-type forests in Pakistan.

Materials and Methods

Seeds of *Reptonia buxifolia* collected/purchased from Attock-Nizampur area were dried, fleshy epicarps removed and stored at room temperature. The glassware was sterilized at 170 °C

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for 4 hours and seeds were sterilized with alcohol, mercuric chloride or chlorax. Ten seeds with 5 replications were placed on 4-folds of whatman No. 1 filter papers in a petri dish and topped with two-folds of filter papers. The dishes were kept at optimum moisture level and incubated at the desired temperature. The emergence of radicle was taken as an index of germination. The radicle growth was recorded to the nearest millimeter. This procedure was followed throughout this investigation.

Results

1. *Effect of Different Temperatures*

Seeds were incubated at 15, 20, 25, 30, 35 °C and alternating temperatures of 20/25, 20/50, 20/35, 25/30, 25/35 and 15/50 °C. The observations were taken after every 48 hours upto 30 days.

There was no germination at 15, 35 °C and at alternating temperatures. The germination was 26% at 20 °C and 30 °C and 48% at 25 °C on the 16th and subsequent days. The radicle growth was better at 25 °C than at other temperatures tried.

2. *Effect of Cold, Warm (60 °C) and Boiling Water*

Seeds, soaked in water at room temperature (25 °C) for 1,2,4,6 and 8 hours and tried for germination at 20, 25 and 30 °C, gave almost equal germination to that of control after 20 days. The seeds soaked in warm water for 5, 10 and 15 minutes exhibited 86, 88 and 94% germination respectively at 20, 25 and 30 °C (Table 1). Seeds treated with boiling water for 5, 10 and 15 minutes exhibited 42, 20 and 24% germination respectively at 20 °C. There was no germination at 25 and 30 °C.

3. *Effect of Acid Scarification*

Seeds were scarified with concentrated H₂SO₄ for 2, 5 and 10 minutes and incubated at 20, 25 and 30 °C. Two-and 5-minutes scarified seeds exhibited 64 and 68% germination respectively at 20 °C and at 25 °C (Table 2). At 30 °C the germination percentage decreased. The growth of radicle was also better in the acid treated seeds.

4. *Effect of Mechanical Scarification alone and coupled with soaking in Water*

Mechanically scarified seeds exhibited 90–92% germination at 20, 25 and 30 °C showing early and better percent germination and growth than the control (Table 3). Scarified seeds were soaked in water for 5, 10 and 15 minutes and incubated as before.

The treated seeds had 90–94% germination at 20–30 °C with augmented germination percentage and rate (Table 3).

5. *Effect of potassium nitrate alone and coupled with Mechanical Scarification*

Seeds were grown in 0.00, 0.25, 0.50, 0.75 and 1.00% solutions of KNO_3 at 25 °C. The germination was respectively 78 and 60% in 0.25 and 0.50% treatments. It decreased at higher concentrations. The radicle growth remained unaffected (Table 4). Mechanically scarified seeds grown in KNO_3 as above exhibited 94, 96, 72 and 80% germination respectively in 0.25, 0.50, 0.75 and 1.0% treatment (Table 4).

6. *Effect of Hormones alone and coupled with Acid or Mechanical Scarification*

Seeds were soaked in 0.25, 0.50, and 1.0 ppm IAA and GA_3 solutions for 5 minutes and grown at 25 °C. Seeds treated with 0.25 and 0.50 ppm IAA respectively exhibited 78 and 60% germination against 52% in the control (Table 5) with improved radicle growth. Five minutes acid scarified seeds were treated with IAA and GA_3 as above and tried for germination at 25 °C. The germination was 44, 82 and 78% respectively in control, 0.50 and 0.25 ppm IAA treated seeds (Table 5). The radicle growth was better in 0.25 ppm treatment. Mechanically scarified seeds treated with IAA and GA_3 and incubated at 25 °C, gave 94, 92, 88 and 48% germination in 0.50, 0.25, 1.00 and control treatments respectively (Table 5) with better radicle growth. GA_3 remained ineffective in all the cases.

7. *Effect of Sodium hypochlorite, pH and Seed Size*

Seeds grown in 0.25, 0.50, 0.75% sodium hypochlorite gave 80% germination at 25 °C compared to 52% in the control. The germination decreased at higher concentration.

Seeds exhibited 56 and 44% germination and better growth at pH 7 and 5 respectively on the 15th day. There was no germination at pH 2, 3, 9 and 11. The germination was respectively 64 and 44% in large and small sized seeds at 25 °C.

8. *Effect of Sowing Depths and Storage*

Seeds were grown in sandy loam soil in pots at 2.5, 5, 7.5, 10 and 15 cm depths during March in the open environmental condition. The germination was 58, 50, 46 and 42% in 2.5, 5, 7.5, 10 and 15 cm deeply sown seeds respectively. The germination decreased and got delayed with an increase in sowing depth. The seeds germinated during 3rd week at 15 cm depth. Seeds stored for zero, 2, 4, 8 and 12 months exhibited 50–56% germination showing no significant effect of storage.

9. *Other treatments*

There was either no or retarded germination when seeds were treated alone with acetone, hydrogen peroxide, NaCl, $CaCl_2$, $MgCl_2$, Na_2SO_4 , Sacch's, Hoagland's and Knop's nutrient solutions and thiourea, or in combination of different physico-chemical factors.

Discussion

Reptonia seeds are generally deposited around the parent plants and are dispersed by man and animals. They germinated at 20 to 30 °C only because high temperatures usually retard germination (Hussain and Nasrin, 1985; Hussain *et al.*, 1984; Hussain *et al.*, 1980). The hard testa of *Reptonia* retarded the imbibition of water, emergence of embryo and availability of gases as mechanical and acid scarification increased the germination to over 90%. Scarification is similar to the natural wear and tear of testa in the soil due to the interaction of environmental factors. Seeds of *Datura* (Hussain *et al.*, 1980), *Peganum* (Hussain and Nasrin, 1985), and *Hyoscyamus* (Hussain *et al.*, 1984) gave over 90% germination with scarification or removal of testa. Scarified seeds are also rescued from waxy and oily coatings. Shaikh (1986) reported promoted germination of *Acacia* by acid scarification and same is true for *Reptonia*. Soaking the seeds in water augmented germination to over 90%. Due to the imbibition of water, softening of testa and removal of inhibitors from seed coat (Mubarak and Hussain 1978; Evenari, 1949; Ilahi *et al.*, 1987). Scarified seeds treated with either water, IAA, KNO₃ or sodium hypochlorite further accelerated the germination due to better interaction. *Reptonia* might do better in nitrogen and potassium enriched habitat as KNO₃ promoted the germination. Ilahi and Perveen (1982) reported IAA to promote germination of pine seeds and we observed the same for *Reptonia*. GA₃ and acetone failed to increase germination of *Reptonia* which agrees with Ilahi *et al.* (1987). The seeds lying in the top soil are subjected to a variety of environmental factors which hasten the germination. Although surface sown seeds exhibit prompt germination but will be susceptible to desiccation in the following dry season compared to seeds emerging from deeper layers owing to deep root system and stout seedlings. *Reptonia* seeds retain viability and germinability for long duration as opposed to *Capparis* seeds (Qaiser and Qadir, 1971; Ilahi and Hussain, 1987). *Reptonia* exhibited higher germination with stout seedlings in soil at field capacity and in water-logged condition. The water-logged condition simply triggers the germination and by itself may not be favourable for the seedling establishment. The better germination exhibited by large seeds might be due to healthy embryo containing sufficient reserve food.

Heavy deforestation and over grazing caused isolation of trees, low organic matter and increased soil erosion making the *Reptonia* habitat unfavourable for its regeneration. The findings suggest that during afforestation/reforestation of such degraded sites seeds be pretreated with IAA, KNO₃, water or sodium hypochlorite subsequent to scarification and tried for germination in spring and monsoon seasons. Soil fertilization will further improve survival and vigour of the seedlings. Seed broadcast will not be much helpful as *Reptonia* seedlings from surface might fail to establish. The seedlings flourish in a microhabitat created by the parent and dominant tree layer which is almost non-existent in Attock-Nizampur valley and other such degraded sites (Ilahi and Hussain, 1987; Naqvi, 1974). This can be accomplished by sowing the seeds in and around the thickets of spiny, obnoxious and non-palatable plants. Small alluvial depressions might also favour seedling establishment. The findings suggest that *Reptonia* seeds have sufficient viability and germinability. The poor regeneration is due first to the selling of fruits by local inhabitants outside its ecological range and elimination of mature trees; secondly due to the altered habitat condition resulting from deforestation and overgrazing. The afforestation and reforestation of *Reptonia* cannot be successful unless a complete protection is accord-

ed to it. Man has changed the natural habitats and associated forests for his immediate benefits at the cost of renewable bioresource.

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TABLE 1

Effect of Warm Water (at 60 °C) on the Germination and Growth of *Reptonia buxifolia*.

Days	Treatment Time (Minutes)			
	Control	5	10	15
a. 20 °C				
8	18	—	6	6
12	22	36	62	66
16	26	74	74	76
20	26	78	84	86
b. 25 °C				
8	16	2	10	18
12	46	26	38	44
16	48	72	68	78
20	50	88	88	86
c. 30 °C				
8	—	20	24	20
12	18	80	42	40
16	26	86	48	76
20	26	94	90	92
Radicle Growth (mm)				
a.	8	64	64	66
b.	26	66	64	69
c.	13	74	72	80

TABLE 2
Effect of Acid Scarification on the Germination and Radicle Growth of *Reptonia buxifolia*

Days	20 °C						25 °C						30 °C					
	Control		5		10		Control		5		10		Control		5		10	
	Soaking Time (Minutes)						Soaking Time (Minutes)						Soaking Time (Minutes)					
12	18	50	34	28	38	50	26	58	44	40	48	50	26	58	44	40	48	50
16	26	58	44	40	48	54	26	64	54	56	66	26	64	54	56	66	26	64
20	26	64	54	56	50	66	26	64	54	56	66	26	64	54	56	66	26	64
Radicle Growth (mm)																		
8	18	20	20	20	26	25	18	20	20	20	26	25	18	20	20	20	26	25
							12	14	14	14	18	14	12	16	16	16	21	11

TABLE 3
Effect of Mechanical Scarification alone (MS) and combined with Soaking water (MSW) on the Germination and Radicle Growth.

		20 °C						25 °C						30 °C							
Days	Control	MS			MSW			Control	MS			MSW			Control	MS			MSW		
		5	10	15	5	10	15		5	10	15	5	10	15		5	10	15	5	10	15
8	—	68	20	30	36	—	48	32	38	—	40	48	38	70	—	40	48	38	70		
12	20	72	22	34	44	36	78	36	42	26	6	78	56	42	76	6	78	56	42	76	
16	26	90	90	86	96	50	92	88	92	88	26	90	90	94	26	90	90	90	94		
	12	53	74	69	67	39	68	65	57	61	15	67	52	65	15	67	52	68	65		

Radicle Growth (mm)

TABLE 4

Effect of KNO_3 alone and in combination with Mechanical Scarification on the Germination and Radicle Growth of *Reptonia buxifolia* at 25 °C

Day	KNO ₃ concentration (%) alone				KNO ₃ concentration (%) + Mechanical Scarification			
	Control	0.25	0.50	1.0	0.25	0.50	0.75	1.00
8	38	52	48	42	66	78	42	48
12	50	66	48	50	78	90	64	66
18	56	78	60	50	94	96	72	80
Radicle Growth (mm)								
	54	56	45	19	66	70	66	60

TABLE 5

Effect of IAA and GA₃ on the Germination and Radicle Growth of *Reptonia buxifolia* at 25 °C

Concentration (ppm)		IAA			GA ₃		
Day	Control	0.25	0.50	1.00	0.25	0.50	1.00
A. IAA and GA ₃ alone							
5	10	16	16	10	10	12	10
10	16	36	34	32	16	20	18
15	48	52	66	48	32	36	26
20	52	60	78	50	38	38	34
B. IAA and GA ₃ + Acid Scarification							
5	10	30	32	18	10	18	12
10	14	66	60	44	12	40	46
15	44	78	82	54	44	48	48
C. IAA and GA ₃ + Mechanical Scarification							
5	10	28	26	22	20	22	10
10	14	68	72	70	28	46	56
15	48	92	94	88	48	56	60
Radicle Growth (mm)							
A.	45	54	60	48	39	37	18
B.	45	69	58	50	41	49	39
C.	50	59	63	60	47	54	41

REFERENCES

- Evenari, M. 1949. Germination inhibitors. Bot. Rev., 15: 153–194.
- Hussain. F., B. Mubarak and H.H. Naqvi. 1980. Germination studies on *Datura innoxia* M. and *D. fastuosa*. Jour. Sci. Technol., 4:79–81.
- Hussain. F., G. Khanzada and H.H. Naqvi. 1984. Preliminary study on the germination of *Hyoscyamus niger* L. and *H. insanus* Stock. under laboratory condition. Jour. Sci. & Technol., 8:4–6.
- Hussain. F. and R. Nasrin. 1984. Germination study on the seeds of *Peganum harmala* L. Pak. J. Agric. Res., 6:113–118.
- Ilahi. I. and R. Perveen. 1982. The effect of stratification and chemical treatments on the germination of different pine species. Jour. Sci. & Technol., 6:179–187.
- Ilahi. I. N. Siddiqi and F. Hussain. 1987. Effects of physico-chemical factors on the germination and seedling growth of *Erianthus griffithii* (Munro) KH. f. Sarhad J. Agric., 3:349–364.
- Ilahi. I. and F. Hussain. 1987. Ecophysiological Approaches for the improvement of vegetation of Attock–Nizampur valley. Final Tech. Rep. Botany Deptt., Peshawar University.
- Mubarak, B. and F. Hussain. 1978. Biochemical inhibition exhibited by *Datura innoxia* M. seeds. Pak. J. Bot., 10:149–156.
- Naqvi. H.H. 1974. Some observations on the vegetation of Peshawar and Khyber Pass area. Biologia, 20:152–162
- Qadir, S.A. and N. Lodhi. 1971. Germination behaviour of seeds of some common shrubs. Jour. Sci. Univ. Karachi 1:84–87.
- Qaiser. M. and S.A. Qadir. 1971. A contribution to the autecology of *Capparis decidua* (Forsk) Edgew. Pak. J. Bot., 3:37–60.
- Sheikh, M.I. 1986. Afforestation of arid and semi-arid areas in Pakistan. F.A.O. Field Document 13, FO. DP/PAK/78/036 Pakistan Forest Institute, Peshawar.