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Seed germination studies in Amaltas

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Abstract
The present study entitled "Seed Germination Studies in Amaltas (*Cassia fistula*)" was conducted during the year 2024-25 at the Centre of Excellence for Citrus, Horticulture Section, College of Agriculture, Nagpur. The primary objectives were to evaluate the effect of different seed treatments on germination and to identify the most effective treatment for enhancing seed germination in *Cassia fistula*. The experiment was designed in a Completely Randomized Design (CRD) with ten seed treatments, including acids such as concentrated HCl, H_2SO_4, and HNO_3 applied for varying durations (2.5, 5, and 7.5 minutes), along with a control, and each treatment was replicated thrice. Observations were recorded for germination and growth parameters. Among all treatments, seeds treated with conc. H_2SO_4 for 7.5 minutes (T_7) showed significantly superior results, including the earliest germination (2.00 days) and the highest germination percentage (98.67%). T_7 also exhibited maximum values for growth parameters (plant height, shoot length, number of leaves per seedling, stem girth, leaf length, fresh weight, dry weight and survival percentage). Based on the findings, conc. H_2SO_4 treatment for 7.5 minutes is recommended as the most effective pre-sowing treatment for improving germination and early seedling growth in *Cassia fistula*.

Keywords: Direct organogenesis, BAP, TDZ, Kinetin, MS media, germplasm conservation

Introduction
Ornamental trees are primarily cultivated for their aesthetic value, enhancing landscapes with their flowers, foliage, form, bark, and fragrance. In addition to beautifying surroundings, they contribute to environmental improvement by absorbing pollutants and releasing oxygen. With growing interest in urban landscaping, ornamental horticulture has become a vital economic sector. Among ornamental species, *Cassia fistula* (golden shower tree or Amaltas) is notable for its vibrant yellow blooms, drought tolerance, and low maintenance needs (Arora, 1988) [4]. Native to the Indian subcontinent, it is widely distributed across tropical Asia, Africa, South America, and Australia.

Beyond its ornamental appeal, *C. fistula* holds therapeutic value in traditional medicine systems such as Ayurveda, Unani, and Siddha. Various plant parts exhibit antibacterial, antifungal, antioxidant, antitumor, and hypolipidemic properties. Its pulp acts as a natural laxative, and leaf extracts possess mosquito larvicidal activity (Mehdi *et al.*, 2011) [13], making it suitable for roadside and institutional plantations. However, seed dormancy due to a hard, impermeable seed coat hampers germination (Cavanagh, 1980) [8]. Pre-sowing treatments with concentrated HCl, H_2SO_4, and HNO_3 have shown efficacy in breaking dormancy and enhancing seed performance. This study evaluates the effects of such treatments on germination and seedling growth in *C. fistula*, aiming to improve propagation of this ecologically and medicinally valuable species, while addressing practical challenges in nursery practices and seed handling.

Material and Methods
The experiment was conducted during autumn of 2024 to study the effect of different seed treatments in germination of Amaltas. The experiment was laid out in a Completely Randomized Design (CRD) with ten treatments and three replications, using crates as experimental units. Fifty seeds of *Cassia fistula* were sown per crate, total 30 crates. Treatments included control (T_1-water soaking for 24 hours), seed treatment conc. HCl for 2.5 min (T_2), conc. HCl for 5 min (T_3), conc. HCl for 7.5 min (T_4), conc. H_2SO_4 for 2.5

min (T_5), conc. H_2SO_4 for 5 min (T_6), conc. H_2SO_4 for 7.5 min (T_7), conc. HNO3 for 2.5 min (T_8), conc. HNO3 for 5 min (T_9), conc. HNO3 for 7.5 min (T_{10}). After seed treatment the seeds were washed with cold tap water for several times to remove the residual of acids. Seeds were collected from healthy mature trees within the college premises in June-July 2024 and sown in September 2024 in a soil: sand: FYM (2:1:1) medium. Crates were irrigated daily until germination, followed by twice-weekly irrigation and regular weeding. Meteorological data was recorded from the Department of Agronomy, Nagpur.

Observations were recorded from five randomly selected seedlings per treatment for germination (days to germination, percentage), growth (height, shoot length, stem girth, leaf number, leaf length, leaf area, fresh and dry weight and survival percentage. Germination percentage was calculated using: (Germinated seeds/Total seeds sown) × 100. Leaf area was measured using a digital leaf area meter. Dry weight was taken after oven-drying at 60 °C for 24 hours. Survival percentage was computed as: (Alive seedlings/Germinated seedlings) × 100. Statistical analysis was done as per Panse and Sukhatme (1967) [16] with SE(m)±and CD at 5% significance, and appropriate graphical representations were used for clarity.

Results and Discussion

Germination parameters

The data in respect of the germination parameters of Amaltas as influenced by different seed treatment was presented in Table 1.

Days to germination (days)

The treatment T_7-conc. H_2SO_4 for 7.5 minutes recorded significantly minimum days to germination (2.00 days) as compared o other treatments, and it was followed by the treatment T_6-conc. H_2SO_4 for 5 minutes (3.16 days) and T_5-conc. H_2SO_4 for 2.5 minutes (4.14 days). Whereas, maximum days to germination (24.33 days) was recorded in control (T_1). In hard-coated seeds, acid scarification efficiently breaks dormancy by removing physical barriers. Similar results had been reported by Babalola *et al.* (2014) [5] in *Cassia fistula* and Negi *et al.* (2015) [14] in *Ribes alpestre*.

Germination percentage (%)

The treatment T_7-conc. H_2SO_4 for 7.5 minutes had the highest germination percentage (98.67%) which was significantly superior over all the treatments under study. Whereas, T_1-control treatment recorded minimum germination percentage (4.67%). Sulfuric acid breaks physical dormancy, leading to more uniform and higher germination. These results are in agreement with findings by Gupta *et al.* (1997) [10] in *Glycyrrhiza glabra* and Bhuse *et al.* (2001) [6] in *Cassia angustifolia*.

Growth parameter

The data in respect of the growth parameters of Amaltas as influenced by different seed treatment was presented in Table 2.

Plant height (cm)

At the stage of 90 days after sowing, the treatment T_7-conc. H_2SO_4 for 7.5 minutes had recorded significantly maximum plant height (19.98 cm) which was found at par with the treatment T_6-conc. H_2SO_4 for 5 minutes (18.99 cm) and it was followed by the treatment T_5-conc. H_2SO_4 for 2.5 minutes (17.89 cm). Whereas, minimum plant height (9.42 cm) was recorded under T_1-control treatment.

At the stage of 120 days after sowing, the treatment T_7-conc. H_2SO_4 for 7.5 minutes had recorded significantly maximum plant height (25.38 cm) which was found at par with the treatment T_6-conc. H_2SO_4 for 5 minutes (23.96 cm) and it was followed by the treatment T_5-conc. H_2SO_4 for 2.5 minutes (22.28 cm). Whereas, minimum plant height (10.46 cm) was recorded under T_1-control treatment. Improved physiological activity and longer growing duration contribute to increased height. Similar observations were by Bichi *et al.* (2012) [7] in *Delonix regia* and Agbogid *et al.* (2007) [1] in *Dacryodes edulis*.

Shoot length (cm)

At the stage of 90 days after sowing, the treatment T_7-conc. H_2SO_4 for 7.5 minutes had recorded significantly maximum shoot length (15.87 cm) which was found at par with the treatment T_6-conc. H_2SO_4 for 5 minutes (15.17 cm). Whereas minimum shoot length (7.96 cm) was recorded under T_1-control treatment.

At the stage of 120 days after sowing, the treatment T_7-conc. H_2SO_4 for 7.5 minutes had recorded significantly maximum shoot length (18.68 cm) which was found at par with the treatment T_6-conc. H_2SO_4 for 5 minutes (17.84 cm). Whereas minimum shoot length (8.73 cm) was recorded under T_1-control treatment.

Acid scarification helps in establishing a stronger shoot-root axis. The present results are in agreement with the findings of Al Menaie *et al.* (2010) [2] in *Cassia fistula* and *Cassia nodosa*, Tadros *et al.* (2011) [17] in *Leucaena leucocephala*.

Stem girth (cm)

At the stage of 120 days after sowing, the treatment T_7-conc. H_2SO_4 for 7.5 minutes had recorded significantly maximum stem girth (0.36 cm) which was followed by the treatment T_6-conc. H_2SO_4 for 5 minutes (0.34 cm) and T_5-conc. H2 SO_4 for 2.5 minutes (0.32 cm). Whereas minimum stem girth (0.11 cm) was recorded under T_1-control treatment. Sulfuric acid (H_2SO_4) seed treatment increases stem girth in plants by enhancing early germination, improving nutrient uptake, and stimulating hormonal activity that promotes cell division and thickening. Olatunji *et al.* (2012) [15] reported the similar results in respect of stem diameter in *Acacia auriculiformis*.

Number of leaves per seedling

At the stage of 90 days after sowing, the treatment T_7-conc. H_2SO_4 for 7.5 minutes had recorded significantly maximum number of leaves (18.12) which was followed by the treatment T_6-conc. H_2SO_4 for 5 minutes (16.88) and T_5-conc. H_2SO_4 for 2.5 minutes (16.15). Whereas, minimum number of leaves (8.62) was recorded under T_1-control treatment.

At the stage of 120 days after sowing, the treatment T_7-conc. H_2SO_4 for 7.5 minutes had recorded significantly maximum number of leaves (23.88) which was followed by the treatment T_6-conc. H_2SO_4 for 5 minutes (21.77) and T_5-conc. H_2SO_4 for 2.5 minutes (20.34). Whereas, minimum number of leaves (10.13) was recorded under T_1-control treatment.

The increase in leaf production can be linked to the accelerated shoot growth and better nutrient uptake

facilitated by a strong root system in sulfuric acid-treated seeds. Similar increases in leaf number were observed by Mabundza *et al.* (2010) [12] in *Tamarindus indica*.

Length of leaf (cm)

At the stage of 120 days after sowing, the treatment T_7-conc. H_2SO_4 for 7.5 minutes had recorded significantly maximum length of leaf (10.98 cm) which was followed by the treatment T_6-conc. H_2SO_4 for 5 minutes (10.23 cm) and T_5-conc. H_2SO_4 for 2.5 minutes (9.97 cm). Whereas minimum length of leaf (3.65 cm) was recorded under T_1-control treatment. Acid scarification boosts early root activity, enhancing the uptake of water and nutrients necessary for leaf expansion. The result gotten in this study is similar to the findings of Usman and Asan (2017) [18] in *A. digitata*.

Leaf area (cm^2)

At the stage of 120 days after sowing, the treatment T_7-conc. H_2SO_4 for 7.5 minutes had recorded significantly maximum leaf area (22.03 cm^2) which was found statistically at par with the treatment T_6-conc. H_2SO_4 for 5 minutes (21.92 cm^2) and T_5-conc. H_2SO_4 for 2.5 minutes (21.45 cm^2). Whereas minimum leaf area (10.03 cm^2) was recorded under T_1-control treatment. This increased leaf area might due to the cumulative effect of improved leaf number and length, which results from better root-shoot coordination in acid-treated seedlings. Similar findings were reported by Amira *et al.* (2013) [3] in *Cassia fistula*.

Fresh weight of seedling (g)

At the stage of 120 days after sowing, the treatment T_7-conc. H_2SO_4 for 7.5 minutes had recorded significantly maximum fresh weight of seedling (8.33 g) which was found statistically at par with the treatment T_6-conc. H_2SO_4 for 5 minutes (7.96 g) and it was followed by the treatment T_5-conc. H_2SO_4 for 2.5 minutes (7.24 g). Whereas minimum fresh weight of seedling (3.73 g) was recorded under T_1-control treatment. This might be explained by the better water absorption and storage capacity of vigorously growing seedlings derived from acid-scarified seeds. Similar increases in fresh weight were observed by Fallah *et al.* (2014) [9] in *Canna indica*.

Dry weight of seedling (g)

At the stage of 120 days after sowing, the treatment T_7-conc. H_2SO_4 for 7.5 minutes had recorded significantly maximum dry weight of seedling (3.45 g) which was followed by the treatment T_6-conc. H_2SO_4 for 5 minutes (3.22 g) and T_5-conc. H_2SO_4 for 2.5 minutes (2.93 g). Whereas minimum dry weight of seedling (0.62 g) was recorded under T_1-control treatment. This could be attributed to the efficient utilization of photosynthates and nutrient reserves in seedlings with better shoot and root systems from H_2SO_4-treated seeds. A higher dry weight reflects structural tissue buildup and metabolic maturity. Similar results were found by Muhammad *et al.* (2017) in date palm.

Survival percentage (%)

At the stage of 120 days after sowing, the treatment T_7-conc. H_2SO_4 for 7.5 minutes had recorded significantly maximum survival percentage (89.96%) which was found statistically at par with the treatment T_6-conc. H_2SO_4 for 5 minutes (87.45%). Whereas minimum survival percentage (44.44%) was recorded under T_1-control treatment. This phenomenon could be linked to improved structural strength and physiological balance in sulfuric acid-treated seedlings, enabling better post-transplant survival. These findings are in agreement with Karaguzel *et al.* (2004) [11] in *Lupinus varius*.

Table 1: Effect of different seed treatments on germination parameters in Amaltas (*Cassia fistula*)

Treatments	Germination percentage (%)	Days to germination
T_1	4.67	24.33
T_2	13.47	14.67
T_3	15.33	13.67
T_4	17.33	11.33
T_5	68.00	4.14
T_6	92.67	3.16
T_7	98.67	2.00
T_8	16.67	13.33
T_9	18.33	10.33
T_{10}	20.67	9.67
'F' test	Sig.	Sig.
SE (m)±	1.34	0.33
CD at 5%	3.96	0.98

Table 2: Effect of different seed treatments on growth parameters in Amaltas (*Cassia fistula*)

Treatments	Plant Height (cm)		Shoot length (cm)		Number of leaves		Stem girth (cm) at 120 DAS	Leaf length (cm) at 120 DAS	Leaf Area (cm^2) at 120	Fresh weight of seedling (g) at 120 DAS	Dry weight of seedling (g) at 120 DAS	Survival percentage (%)
	90 DAS	120 DAS	90 DAS	120 DAS	90 DAS	120 DAS						
T_1	9.42	10.46	7.96	8.73	8.62	10.13	0.11	3.65	10.03	3.73	0.62	44.44
T_2	11.73	13.33	9.62	10.53	10.23	13.04	0.17	5.14	16.28	5.21	1.03	57.22
T_3	11.97	13.81	9.71	10.88	10.84	13.67	0.18	5.50	16.46	5.66	1.34	61.67
T_4	13.84	16.02	11.53	12.78	13.62	17.31	0.21	6.27	18.36	6.19	1.54	65.84
T_5	17.89	22.28	14.23	16.64	16.15	20.34	0.32	9.97	21.45	7.24	2.93	74.58
T_6	18.99	23.96	15.17	17.84	16.88	21.77	0.34	10.23	21.92	7.96	3.22	87.45
T_7	19.98	25.38	15.87	18.68	18.12	23.88	0.36	10.98	22.03	8.33	3.45	89.96
T_8	13.39	15.44	11.02	12.24	13.08	15.94	0.19	5.74	17.13	5.83	1.41	64.93
T_9	15.28	17.83	12.02	13.65	14.59	17.92	0.22	6.63	18.78	6.56	2.59	67.96
T_{10}	15.67	18.94	12.18	13.96	15.13	18.86	0.26	6.95	19.57	6.87	2.73	68.33
'F' test	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.
SE (m)±	0.40	0.48	0.32	0.36	0.31	0.47	0.007	0.19	0.48	0.17	0.06	1.60
CD at 5%	1.18	1.42	0.95	1.08	0.94	1.38	0.01	0.58	1.43	0.50	0.17	4.73

Conclusion

From the above investigation, it is concluded that seed treatment with concentrated sulfuric acid (H_2SO_4) for 7.5 minutes (T_7) proved to be the most effective among all treatments. This treatment resulted in significantly earlier germination and the highest germination percentage. It also exhibited superior performance in various growth parameters, including plant height, shoot length, stem girth, number of leaves per seedling, leaf length, leaf area, fresh and dry weight of seedlings, and survival percentage. Therefore, concentrated H_2SO_4 treatment for 7.5 minutes (T_7) can be recommended as the optimum seed pre-treatment for enhancing seed germination and seedling growth in Amaltas (*Cassia fistula*).

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