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INFLUENCE OF GA3 AND CHARCOAL ON IN VITRO SEED GERMINATION OF TERMINALIA ARJUNA

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ABSTRACT

Terminalia arjuna is a deciduous tree belongs to the family combretaceae. The leaves of this tree is fed to tasar silk worm Anthereae mylitta to rear tasar silk, one of the traditional silks of India. The bark is used in Ayurveda from ancient days. The tree multiplied only via seeds collected from forest. Seed germination in Terminalia arjuna is very difficult because of hard seed coat, high heterozygosity, seed dormancy and high phenoloic compounds. To conquer this for conservation of this valuable tree a protocol is developed for increasing the rate of in vitro seed germination. The seeds soaked in GA-3 (100 ppm) solution for 24h and inoculated in MS medium by supplemented with charcoal (100 mg/lit) resulted high percentage of seed germination.

Keywords: Charcoal, In vitro, Terminalia arjuna, Tasar silk

Introduction

Terminalia arjuna is a deciduous tree belongs to the family combretaceae. It is commonly known as arjuna. The arjuna is about 20-25 meters tall; usually has a buttressed trunk and forms a wide canopy at the crown, from which branches drop downwards. It has oblong, conical leaves which are green on the top and brown below; smooth grey bark, it has pale yellow flowers which appear between March and June; its glabrous, 2.5-5 cm fibrous woody fruit, divided into five wings, appears between September and November. The leaves of this tree is fed to tasar silk worm Anthereae mylitta to rear tasar silk, one of the traditional silks of India. The bark is used in Ayurveda from ancient days. Now the bark powder of arjuna is used in allopathic also for curing cardiac diseases. The tree multiplied only via seeds collected from forest. Seed germination in Terminalia arjuna is very difficult because of hard seed coat, high heterozygosity, seed dormancy and high phenoloic compounds.

Objective of the present study is to standardize the protocol for *in vitro* seed germination of *Terminalia arjuna* by using GA_3 and Charcoal.

Materials and Methods

Seed germination in *Terminalia arjuna* is very difficult because of hard seed coat, high heterozygosity, seed dormancy and high phenolic compounds. To overcome these problems suitable method was devised to increase the rate of seed germination with better survival rate. Healthy dry seeds of *Terminalia arjuna* were collected from Tirumala hills of Chittor district and Thimmapur of Warangal District. The dry seeds were soaked in water for two days. On the third day the seeds are soaked in GA_3 solution (50-250 ppm) for 12 hrs, 24 hrs, 36 hrs, 48 hrs and 60 hrs to observe the effect of GA_3 on seed germination. These seeds were washed with running tap water for 30 min then treated with Bavastin (0.2%) for 15 min and with streptomycin (0.1%) for 15 min to prevent the bacterial contamination. These seeds were treated with savlon for 5 min and washed thoroughly with distilled water for 2-3 times. Again these seeds were surface sterilized with 70% ethanol for 1 min and $HgCl_2$ (0.1%) for 8 min and washed with autoclaved distilled water for 4-5 times. These seeds were dried under laminar air flow, inoculated on MS medium by adding charcoal (0.5-2.5 mg/lit).

Control batch was maintained without treatment with GA_3 and without adding Charcoal into the medium for comparison of percentage of seed germination.

Results and Discussion

Terminalia arjuna seeds are very hard and the percentage of seed germination is low. In the present study for enhancement of *in vitro* seed germination seeds soaked in GA_3 solution and addition of charcoal resulted better than untreated seeds. Highest percentage (86.66± 1.5) of seed germination was observed at seeds soaked in GA_3 solution (100ppm) for 24 hours by adding 1 mg/l charcoal to the medium and lowest percentage (68.00± 2.00) of seed germination was observed at GA_3 solution(50 ppm) for 12 hours by adding 0.5 mg/l charcoal. In the present study the seeds soaked in plain water started germination after 7-12 days, where as the seeds treated with GA_3 started germination after 5-7 days. GA_3 at 100 ppm for 24 hrs was very effective in initiation and completion of germination process within a short duration than that of untreated seeds.

Previously considerable work was conducted on *in vitro* seed germination of *Terminalia arjuna* by incorporating charcoal and GA_3 in order to promote seed germination by Ramesh *et al* (2001). Treating seeds with GA_3 enhance early seed germination and increase percentage of seed

germination. Adding of charcoal into the medium will inhibit the release of phenolic compounds from the seed. It results in controlling contamination and initiates the seed germination rapidly. Treatment with GA_3 (100 ppm/24 hrs) and charcoal 1 gm/lit results high germination percentage in *Terminalia arjuna*.

Enhancement of seed germination by Pre treatment of seeds with GA_3 on invitro seed germination of Bobgunnia madagascariensis was also reported by Blackson L.K et al 2011.

Effect of GA-3 and Charcoal on in vitro seed germination of Carica papaya was also reported by Ravishankar chauhan et al 2014.

When a seed separates from a plant, has a little primary dormancy that not only prevents the quick seed germination, but adjusts time conditions and place of germination. Charcoal is to remove its dormancy to propagate this worth species. Results of this study indicate that breaking ash seeds dormancy *in vitro* is possible. One way to overcome this dormancy is using special hormones like GA_3 and medium supplements. Many researchers aim that using GA_3 is suitable for breaking dormancy. In this case some researchers reported that GA_3 cause removing dormancy and increasing in germination value.

The use of *in vitro* approaches for the propagation, conservation and genetic improvement of tropical tree germplasm has improved slightly in the last 10 years. Tropical timber trees are long lived and require many years to reach reproductive maturity. Once harvested, many more years are required for natural regeneration to occur or for pure stands cultivated in plantations to reach harvestable market size. In addition to providing timber for a vast array of wood products, tropical tree species are invaluable biological resources and provide unique environmental protection, such as reducing soil erosion and compaction, increasing soil fertility, absorbing pollutants, releasing oxygen, sustaining water sheds and effecting climatic change on a global scale. Apart from meeting the demands for high quality wood, tropical trees provide medicinal, insecticidal, industrial and many unknown commercial products. For these and many more reasons, we must make a concerted effort to conserve this valuable tree. Conservation through *in vitro* propagation technologies will have a global economic and ecological impact on sustaining tropical forest tree biodiversity.

These *in vitro* raised rooted plantlets are directly transferred to vermiculate containing root trainers and kept under green house for hardening before transferred to field.

Influence of GA_3 and charcoal on in vitro seed germination of Terminalia arjuna

S. No	No. of seeds	GA_3 conc (ppm)	GA_3 treatment(h)	Charcoal (mg/lit)	Germination (%)	F-value	P-value
1	50	50	12	0.5	68.0 ± 2.0	76.633***	0.000
2	50	100	24	1.0	86.66 ± 1.5		
3	50	150	36	1.5	73.66 ± 1.5		
4	50	200	48	2.0	70.0± 2.0		
5	50	250	60	2.5	63.66± 1.5		
Total					72.4± 8.2		

Conclusion

In *Terminalia arjuna* seeds soaked in GA_3 solution (100 ppm) for 24 hours by addition of activated charcoal (1 mg/l) enhanced *in vitro* seed germination. Thse findings may be helpful for further research.

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