Int.J.Curr.Microbiol.App.Sci (2020) *9*(10): 1803-1808

International Journal of Current Microbiology and Applied Sciences
ISSN: 2319-7706 **Volume 9 Number 10 (2020)**
Journal homepage: http://www.ijcmas.com

Original Research Article https://doi.org/10.20546/ijcmas.2020.910.219

Effect of Pre-sowing Seed Treatments on *Aristolochia tagala* Cham: A Threatened Medicinal Plant of South India

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Keywords

Aristolochia tagala, Seed germination, Water soaking, Thiourea

Article Info

Accepted:
18 September 2020
Available Online:
10 October 2020

A B S T R A C T

An experiment was conducted to standardize the seed germination technique in *Aristolochia tagala* Cham. at the Division of Floriculture and medicinal crops, ICAR-Indian Institute of Horticulture, Bengaluru during 2019-20. Fresh seeds of *A. tagala*were collected from the Field Gene Bank of RET medicinal plants, IIHR Bengaluru and different treatments such as GA_3, KNO_3, Thiourea and water soaking was imposed on it. The maximum germination percent (70.57%) was obtained from treatment with thiourea (2.0 %), which was on par (67.33%) with water soaking treatment. The maximum shoot length (15.13 cm), root length (6.40 cm), plant height (21.53 cm), fresh weight of the seedling (726.33 mg/plant), dry weight of seedling (72.90 mg/plant) and seedling vigour index I (1433.52) was recorded in water soaking for 30 minutes.

Introduction

Aristolochia tagala Cham. is a threatened medicinal plant of Aristolochiaceae family. It is generally called "Nallaisvara", "Nallayisvari" or "Nallaeeshvara" in Telugu, "Doddaeeshwariballi" or "Gattadaeeshvari" in Kannada, "Aadutheendapalai" in Tamil and "Valia arayan" in Malayalam. It is a sun-loving climber found in forests and open lowland thickets, climbing over bushes and trees. It is distributed throughout tropical and subtropical countries (Murugan *et al.*, 2006.). It is found to grow at 1000 to 1400 m above sea level elevation and occurs along the Western Ghats in India (Nayar *et al.*, 1976). This plant is found to have a significant role in the tribal medicinal system in its natural habitat. It is mainly used to treat snake bites and other poisonous bites. Apart from this, roots are also used to treat bone fracture, malaria, indigestion, toothache, rheumatism and various dermatological conditions by Kani tribes, Thiruvananthapuram and Tirunelveli hills (Ravikumar and Ved, 2000). Due to indiscriminate use of its roots and

deforestation, its population in reducing in its natural habitat. The threatened status was assigned to this species under the red data list of south Indian medicinal plants (Sarvalingam and Rajendran, 2016). Hence the conservation of this species is crucial.

One way in conservation is to standardize the propagation technique for its commercial cultivation and *ex-situ* conservation. But in *A. tagala*, seed germination is low due to the presence of insufficient endosperm, which leads to low seed viability (Biswas *et al.*, 2007). Therefore, the present study aims to improve seed germination and the effect of different pre-soaking seed treatment on seed germination and seedling growth in *Aristolochia tagala* Cham.

Materials and Methods

Experimental site

The experiment was carried out at the ICAR-Indian Institute of Horticultural Research (ICAR-IIHR), Hesaraghatta Lake Post, Bengaluru. It is located in the eastern dry zone of Karnataka state at$13^{O}58^{l}$ North latitude and 78^{O}East longitude. It is situated at an altitude of 890 m above mean sea level (MSL).

Seed collection

The fresh seeds of *Aristolochia tagala Cham.* were collected freshly from the plant maintained in the FGB of RET medicinal plants at ICAR-Indian Institute of Horticultural Research (IIHR), Bengaluru. The seeds were harvested in October 2019.

Experimental description

The experiment was carried out with eleven different pre-sowing treatments consisting of three replications having twenty-five seeds each. The pre-treatments include GA_3 at 100, 150, 200 ppm, KNO_3 at 1%, 1.5%, 2.0%, Thiourea at 1%, 1.5%, 2.0% and water soaking along with control. Apart from control, the seeds were soaked in the above treatments for 30 minutes and then air-dried. The pre-treated seeds sown in the port rays filled with cocopeat. The experiment was carried out in a poly house. The observations for parameters like germination percentage, rate of germination, days have taken to initiate germination, number of leaves, seedling height, fresh weight of shoot and root, dry weight of shoot and root were recorded.

Statistical analysis

The experimental data were analyzed using Completely Randomized Design (CRD) with three replications. The data were analyzed statistically as per the method suggested by Panse and Sukhatme (1967).

Results and Discussion

Effect of pre-sowing treatments on seed germination of *Aristolochia tagala* Cham

The data of seed germination as an influence by pre-sowing treatments in *A. tagala* Cham. is presented in Table 1. All the treatments were significantly superior over the control. The seed treated with thiourea (2.0 %) recorded the highest germination percent (70.57%), which was on par (67.33%) with water soaking treatment. The minimum time for complete germination (39.67 days)was taken by thiourea (2.0 %) and was followed by water soaking. The rate of germination (0.93) was maximum in thiourea (2.0 %). This may be due to a strong neutralizing effect of thiourea on inhibitor in the seed or increase in cytokinin activity by thiourea to overcome the seed coat inhibiting effect (Hore and Sin, 1994). A similar result was

also obtained in *Hippophae salicifolia* seeds which showed the highest germination rate (76%–83%) when treated with thiourea (100 mM) (Airi *et al.*, 2009). Khan (2015) reported that the seed germination rate (76% ± 0.6%) of *Boswellia serrata* Roxb. improved by thiourea 1 % for 30 minutes. Nandi *et al.*, (2000) obtained a higher germination rate and percentage in *Aconitum heterophyllum* Wall. and *A. balfourii Stap f.* by thiourea treatment.

Table.1 Effect of pre-sowing treatments on seed germination of *Aristolochia tagala* Cham

Treatment	Days taken for complete germination	Germination percent	Rate of germination
T_1 - Control	51.78	46.24	0.56
T_2 - GA_3 100 ppm	44.67	51.19	0.64
T_3 - GA_3 150 ppm	44.11	52.38	0.64
T_4 - GA_3 200 ppm	41.89	62.10	0.65
T_5 - KNO_3 1%	44.22	52.38	0.63
T_6 - KNO_3 1.5%	44.11	53.57	0.65
T_7 - KNO_3 2%	43.55	54.76	0.67
T_8 - Thiourea 1%	44.44	52.57	0.75
T_9 - Thiourea 1.5%	44.00	57.90	0.77
T_{10} -Thiourea 2%	39.67	70.57	0.93
T_{11} – Water soaking	41.33	67.33	0.78
Mean	43.98	56.45	0.70
S. Em ±	0.548	1.412	0.02
C D @ 5 %	1.617	4.167	0.06

Table.2 Effect of pre-sowing treatments on seedling parameters of *Aristolochia tagala* Cham. at 75 days after sowing

Treatment	Shoot length (cm)	Root length (cm)	Seedling height (cm)
T_1 - Control	7.68	3.53	11.21
T_2 - GA_3 100 ppm	8.37	3.87	12.24
T_3 - GA_3 150 ppm	10.15	4.82	14.97
T_4 - GA_3 200 ppm	12.33	5.23	17.56
T_5 - KNO_3 1%	8.47	4.10	12.57
T_6 - KNO_3 1.5%	10.53	5.07	15.60
T_7 - KNO_3 2%	11.50	5.20	16.70
T_8 - Thiourea 1%	10.30	4.32	14.62
T_9 - Thiourea 1.5%	10.93	5.15	16.08
T_{10} -Thiourea 2%	13.50	5.72	19.22
T_{11} – Water soaking	15.13	6.40	21.53
Mean	10.81	4.85	15.66
S. Em ±	0.239	0.091	0.276
C D @ 5 %	0.705	0.269	0.815

Table.3 Effect of pre-sowing treatments on the fresh and dry weight of shoot and root of *Aristolochia tagala* Cham. at 75 days after sowing

Treatment	Fresh weight of seedling (mg/plant)	Dry weight of seedling (mg/plant)	Seedling vigour index I
T_1 - Control	191.33	22.97	518.86
T_2 - GA_3 100 ppm	218.00	36.57	626.07
T_3 - GA_3 150 ppm	277.00	38.80	783.33
T_4 - GA_3 200 ppm	317.33	40.10	1090.64
T_5 - KNO_3 1%	253.33	38.10	658.33
T_6 - KNO_3 1.5%	291.67	45.43	836.55
T_7 - KNO_3 2%	425.67	53.33	916.37
T_8 - Thiourea 1%	367.33	45.33	768.50
T_9 - Thiourea 1.5%	385.67	50.57	931.21
T_{10} -Thiourea 2%	532.67	60.07	1356.68
T_{11} – Water soaking	726.33	72.90	1433.52
Mean	362.39	45.83	901.82
S. Em ±	7.74	0.90	31.332
C D @ 5 %	22.64	2.63	92.487

Effect of pre-sowing treatments on seedling parameters of *Aristolochia tagala* Cham

All growth parameters influenced by pre-sowing seed treatments of *A. tagala* are presented in Tables 2 and 3. There was a significant influence of the treatments on the various seedling parameters. Among the different treatment, the maximum shoot length (15.13 cm), root length (6.40 cm), plant height (21.53 cm), fresh weight of the seedling (726.33 mg/plant), dry weight of seedling (72.90 mg/plant) and seedling vigour index I (1433.52) was recorded in the treatment water soaking for 30 minutes. In general, water soaking of seeds at room temperature helps to soften the seed coat, remove inhibitors, and reduce the time required for germination and increase the germination percentage. The water imbibition by seeds increases the fresh weight of the seeds and water uptake drives the emergence of radicle and enhances germination (Yashaswini, 2009). In *Aristolochia tagala*, the seed's embryo size is small and at the time of seed maturity they are not developed fully (Baskin and Baskin, 1998). The embryo development in *A. tagala* occurs after water absorption (Biswas *et al.,* 2007). Due to which water soaking helped in improving seed germination in *A. tagala* Cham. The effect of water soaking on other growth parameters may be due to its germination stimulating effect. The results recorded by Krishnan *et al.,* (2019) showed that increased germination per cent of 52% in soil within 32 days in *A. tagala* was obtained in seeds soaked in warm water 50^0C for 5min.Debi and Prakash (2015) reported that maximum germination percentage (100%) and mean germination time were obtained in the seeds treated with normal water (24 hours) in *Oroxylum indicum.* Similar results were also obtained in *Pterocarpus santalinus* (Naidu and Mastan, 2001), *Rauvolfia serpentina* (Gupta, 2003), *Gloriosa superba* (Anandhi *et al.,* 2016) and *Terminalia bellirica* (Vikas, 2018).

In conclusion among different pre-sowing treatments, water soaking for 30 minutes was the best treatment with respect to seed germination and other growth parameter and the next best treatment was thiourea 2%. Though the germination percent was highest in thiourea (2.0 %), it was on par with the water soaking. Other growth parameters such as seedling height, root length, shoot length, dry weight, fresh weight and seedling vigour index I were maximum in water soaking. The result of the present investigation will help large scale seedling production conservation of this threatened species.

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How to cite this article:

Rajani Bhat, G. Raviraja Shetty, P. E. Rajasekharan, D. A. Pooja, M. Ganapathi and Sadhashiv Nadukeri. 2020. Effect of Pre-sowing Seed Treatments on *Aristolochia tagala* Cham: A Threatened Medicinal Plant of South India. *Int.J.Curr.Microbiol.App.Sci.* 9(10): 1803-1808. doi: https://doi.org/10.20546/ijcmas.2020.910.219