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Seed germination and seedling growth as influenced by seed treatment in sarpagandha (*Rauvolfia serpentina* Benth.)

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Abstract
The experiment was conducted at Kittur Rani Channamma College of Horticulture, Arabhavi from October to December, 2016 to know the effect of seed treatment on germination and seedling growth in sarpagandha (*Rauvolfia serpentina* Benth.). The experiment was laid out in completely randomized design with three replications and seven treatments *viz*., Water soaking, GA_3 (1000 mg l^{-1}), H_2SO_4, KNO_3 (1%) + HNO_3 (1%), Cow urine, Cow dung slurry treatment and control. Among these, GA_3 treatment showed the significant effects for seed germination and seedling parameters. Overnight soaking in GA_3 resulted in highest speed of germination (2.47), germination percentage (54.67 %), length of seedling (19.61 cm) and vigour index (1072.31). The control (no seed treatment) showed the lowest speed of germination (0.30), germination percentage (14.33 %) and vigour index (199.38). The length of seedling was lowest in cow urine treatment (13.56 cm).

Keywords: Sarpagandha, *Rauvolfia serpentina,* Seed germination, Seed treatment

Introduction
Rauvolfia serpentina (L.) Benth. Ex. Kurz., commonly known as sarpagandha is an indigenous medicinal herb of Indian sub-continent, mentioned in ancient Indian medicinal literature, 3000 years back. Sarpagandha commonly known as serpent wood is having different vernacular names *viz*., Sarpagandha in Sanskrit, Chandrabhaga, Chandrika and Chota-Chand in Hindi, Shivanabhiballi, Sarpagandhi, Garuda paataala and Paataalagandhi in Kannada and Serpentine root, Serpentine wood and Indian snake root in English (Anon., 1956) [1]. It is widely distributed in the foothills of Himalayan range up to an elevation of 1400 m and in the humid tropics of India, Nepal, Myanmar, Thailand, Bangladesh, Indonesia, Cambodia, Philippines and Sri Lanka. In India, it is naturally distributed in Assam in the lower hills of Gangetic plains, Eastern and Western Ghats, in some parts of central India. It is cultivated in small scattered area in Uttar Pradesh and Uttaranchal. A climate with temperature range of 10 to 38°C is well suited for the growth of sarpagandha plant. Best areas for its growth are those which combine high rainfall (1500-4000 mm) with proper drainage and gentle slopes.

Sarpagandha belongs to Apocynaceae family and *Rauvolfia serpentina* is the chief, commercial source of important alkaloids. The roots of this plant have being used in Ayurveda, Unani, Homeopathy and Siddha systems of medicines for the treatment of high blood pressure, insomnia, cardiac diseases and a number of mental problems such as psychic disorders, mental retardation, epilepsy, agitation and neurotic disorders, asthma, hypochondriasis, certain forms of insanity, acute stomach ache and painful delivery. Juice of leaves is used as a remedy for removal of opacities of cornea. Apart from traditional use in health care and culture it has been increasingly used in pharmaceutical industries in the country as well as abroad. 'Serpasil' tablet for high blood pressure is prepared from roots. About 30 alkaloids are known to exist in this plant and the total alkaloid content ranges from 1.7 to 3 per cent of dried roots. The most important alkaloids are reserpine, serpentine, ajmaline, ajmalicine, rauvolfinine, rescinnamine and deserpidine.

The continuous exploitation of sarpagandha from forests without taking proper care of its regeneration has resulted in decline in wild populations, rendered it to a vulnerable and

threatened state, particularly in India (Farooqi and Sreeramu, 2001) [2]. Indiscriminate harvesting, loss of habitat, human and bio-interference, over-exploitation, *etc.* pose further serious threat to its wild resources. It has been categorized as an endangered species based on the IUCN (International Union for Conservation of Nature and natural resources) Red Data Book and critically endangered in CAMP, 2001 report (Bhattarai *et al.,* 2002) [3]. Sarpagandha is banned for export except when accompanied by certificate of its cultivation. The NMPB has prioritized 32 and Planning commission had enlisted 24 medicinal plant species for research and development to meet the desired level of the medicinal plant sector and both these lists include sarpagandha.

It has become necessary to develop proper agro-technology for its domestication and cultivation on scientific lines. Availability of good quality planting material is essential for commercial cultivation. The seed propagated crop is known to give good quality material and higher yield but, seed germination is very poor in sarpagandha. It is reported to vary from 5 to 30 per cent even when only heavy seeds (only about 10 % of normal seed lot) are chosen for sowing. Though the seeds appear to be perfectly normal externally, the seeds exhibit very poor germination (Farooqi and Sreeramu, 2001) [2]. In this regard, an investigation was carried out with an objective to improve seed germination in sarpagandha.

Material and methods

The study was conducted at Kittur Rani Channamma College of Horticulture, Arabhavi from October to December, 2016.The experiment was laid out in completely randomized design with seven treatments, replicated thrice. The treatments include water soaking, GA_3 @ 1000 mg l^{-1}, conc. H_2SO_4, KNO_3 (1%) + HNO_3 (1%), Cow urine, Cow dung slurry and control. For the treatments exceptH_2SO_4 and control, overnight soaking of seeds was followed whereas, in H_2SO_4 treatment, seeds were soaked for one minute and in control, the seeds were sown without any treatment. The seeds were collected from the wild stand of sarpagandha plants found in the evergreen forests of Sirsi, Uttara Kannada (Dist.), Karnataka. The fruits collected from scattered plants found in the lower storey of evergreen forests were depulped manually to extract the seeds. Extracted seeds were washed in clean water and dried in shade. These seeds were subjected to floating test by immersing in water. The seeds which sink in water were selected for germination test. Freshly collected heavy seeds that sink in water were used for sowing within a week time. After imposing treatment, the seeds (hundred seeds per treatment) were sown in protrays filled with coco peat and watered frequently based on necessity. The protrays were maintained in the indoor (ambient) condition in the laboratory. Observations were recorded on different parameters with respect to germination and seedling growth. The speed of germination was calculated by counting the germinated seeds every day from the first day and the cumulative index was made by the formula, Speed of germination (N) = $n_1/1 + n_2/2 + \ldots\ldots\ldots + n_x/x$. Where, $n_1 \ldots n_x$ are the number of seeds germinated on day 1 to day x (Agrawal, 1995) [4]. Germination percentage was calculated by counting the number of germinated seeds out of hundred seeds sown. The per cent data was subjected to arc sin transformation for further analysis. The days to reach four leaf stage was recorded by counting the number of days from the day of first germination till the day when seedling attains four leaves. Length of seedlings was recorded at four leaf stage. Seedling vigour index was calculated by multiplying the germination percentage by length of seedling and expressed as whole number. For all the parameters observed, the percent deviation of the treatment over control was calculated using the formula,

$$\text{Per cent deviation} = \frac{\text{Mean value} - \text{value in control}}{\text{Value in control}} \times 100$$

The data recorded was subjected to statistical analysis using the Fischer's method of analysis of variance with 1 per cent level of significance for 'F' and 't' tests.

Results and discussion

The results obtained from the experiment are discussed hereunder (Table 1). The speed of germination was significantly higher in GA_3 treatment (2.47) followed by cow dung slurry (1.03) and KNO_3 + HNO_3 treatment (0.91) whereas, the lowest was observed in control (0.30). The seed treatment with GA_3 increased the speed of germination by 715.38 per cent as compared to control. Improved speed of germination in GA_3 might be due to increased ratio of GA: ABA in the seeds by exogenous application of GA_3 which could have overcome the inhibitory effect of ABA present in seeds and inhibition of mRNA synthesis which might have been accelerated by gibberellins (Bewley and Black, 1994) [5]. With respect to germination percentage, data revealed the maximum germination in GA_3 treatment (54.67 %) which was significantly higher than rest of the treatments (Fig. 1). The lowest germination percentage was observed in control (14.33 %). Seed treatment with GA_3 recorded 281.40 per cent more germination over control. The highest percentage of germination observed in GA_3 might be due to efficient utilization of limited food reserve present in the seeds by early induction of α - amylase activity. Similar results have been reported by Bhuyar *et al.* (2000) [6] and Ponkumar *et al.* (2008) [7] in *Rauvolfia serpentina,* Mithra and Ghosh (2004) [8] in aswagandha. Whereas, Paul *et al*. (2008) [9] reported that, none of the chemical or acid seed treatments improved germination percentage significantly in sarpagandha. The germination percentages of treated *Rauvolfia tetraphylla* seeds were improved to 52.70 per cent (KNO_3) and 56.66 per cent (GA_3) as compared to 31.26 per cent in untreated seeds (Hussain and Jha, 2014) [10].

The number of days taken for attaining four leaves in germinated seedling was significantly lower in GA_3 (50.33) as compared to rest of the treatments. However, significantly higher number of days to attain four leaves was observed in control (104.67). Seed treatment with GA_3 lead to 51.91 per cent reduction in number of days taken to reach four leaf stage. As it is universally known that, GA_3 helps in the cell elongation; the exogenous application of GA_3 might have enhanced the elongation of cells in a better way. It might have helped in the faster growth of shoot and root as a result, the seedlings might have reached the four leaf stage earlier in seeds treated with GA_3. The length of seedling was significantly highest in GA_3 treatment (19.61 cm) followed by KNO_3 + HNO_3 treatment (17.92 cm) and cow dung slurry treatment (17.01 cm). The lowest seedling length was recorded in cow urine treatment (13.56 cm). There was 41.18 per cent increment of seedling length in GA_3 treatment over the control. The results are in accordance with Ponkumar *et al.* (2008) [7] and Muneshwar (2015) [11] in sarpagandha and Velmurugan *et al*. (2003) [12] in ashwagandha. The significantly higher vigour index (Fig. 1) was recorded in GA_3

(1072.31) compared to the lowest in control (199.38). The vigour index in seed treatment with GA_3 recorded 437.82 per cent increase over control. The increased vigour index might be attributed to increased length of seedlings and germination percentage in seeds due to GA_3 treatment.

Table 1: Effect of seed treatment on germination and seedling growth parameters in sarpagandha

Treatment	Speed of Germination		Germination (%)		Days to 4- leaf stage		Length of seedling		Vigour index	
	Mean	Per cent deviation*	Mean	Per cent deviation*	Mean	Per cent deviation* (Earliness)	Mean	Per cent deviation*	Mean	Per cent deviation*
T_1	0.43	42.86	21.00 (27.15)	46.51	101.00	3.50	14.12	1.63	296.28	48.60
T_2	2.47	715.38	54.67 (47.68)	281.40	50.33	51.91	19.61	41.18	1072.31	437.82
T_3	0.79	159.34	23.00 (28.55)	60.47	67.00	35.99	14.30	2.98	329.02	65.02
T_4	0.91	201.10	34.67 (36.06)	141.86	77.00	26.43	17.92	29.04	621.65	211.79
T_5	0.53	75.82	20.33 (26.64)	41.86	93.33	10.83	13.56	-2.38	276.02	38.44
T_6	1.03	240.66	39.33 (38.84)	174.42	83.67	20.06	17.09	23.04	672.65	237.37
T_7	0.30	-	14.33 (22.16)	-	104.67	-	13.89	-	199.38	-
S.Em. ±	0.03	-	0.86	-	1.80	-	0.26	-	17.93	-
C.D. @ 1%	0.11	-	3.64	-	7.59	-	1.11	-	75.50	-
CV (%)	5.07	-	5.05	-	3.79	-	2.90	-	6.27	-

*Per cent deviation of respective treatment over the control; Figures in parenthesis are arc sin transformed values

T_1: Water soaking
T_2: GA_3 (1000 mg l^{-1})
T_3: Conc. H_2SO_4
T_4: KNO_3 + HNO_3 (1% each)
T_5: Cow urine treatment
T_6: Cow dung treatment
T_7: Control

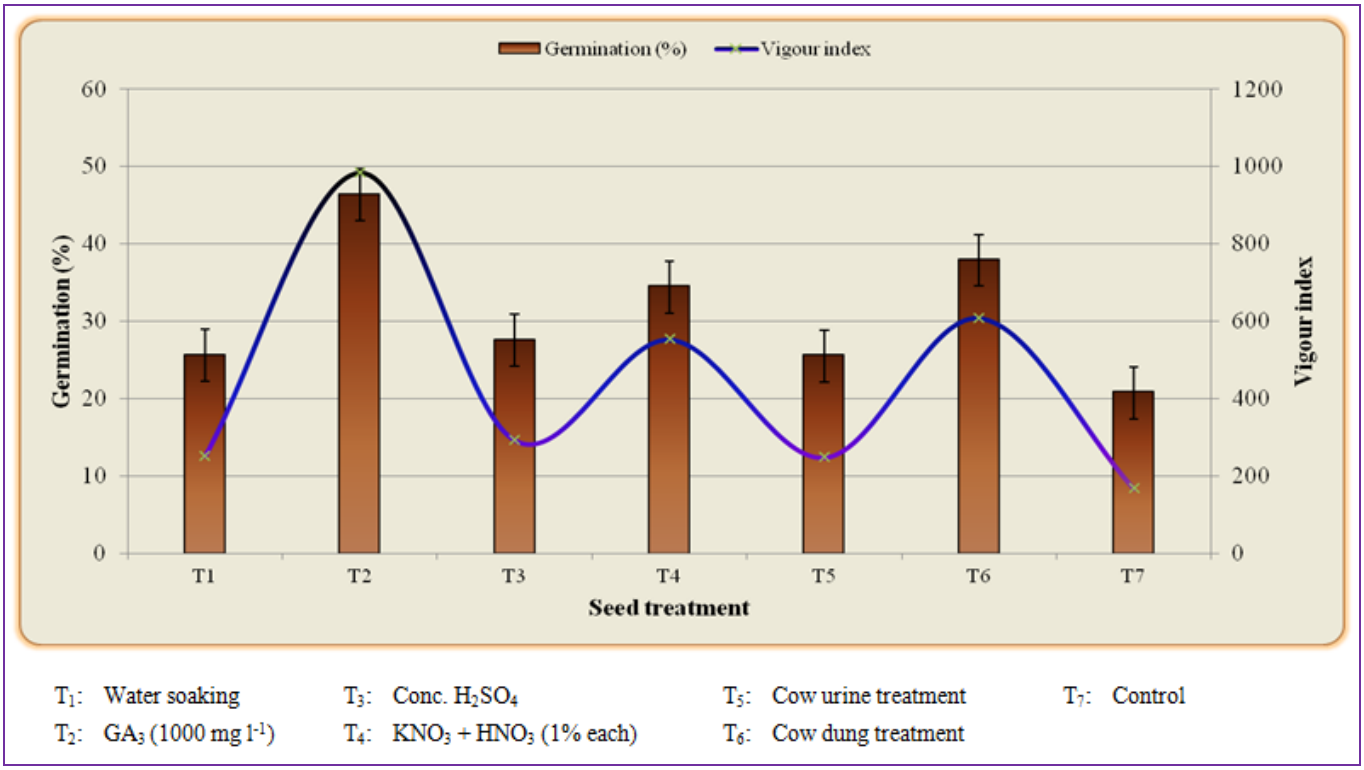

Fig 1: germination and vigour index as influenced by seed treatment in sarpagandha.

Conclusion

Among the different seed treatments tested in sarpagandha, GA_3 (1000 mg l^{-1}) treatment was found to be the best, recording highest values for speed of germination, germination percentage, length of seedling and vigour index. The GA_3 treatment resulted in 715.38 per cent increased speed of germination, 281.40 per cent increased germination, faster growth of seedlings which lead to 41.18 per cent increased seedling length and 437.82 per cent increase in vigour index over the control.

References

1. Anonymous. The Wealth of India- Raw Materials. Council of Scientific and Industrial Research, New Delhi. 1956; 4:139-140.
2. Farooqi AA, Sreeramu BS. Cultivation of medicinal and

aromatic crops. Universities press (India) Limited. 2001, 234-241.
3. Bhattarai NK, Tandon V, Ved DK. Highlights and outcomes of the conservation assessment and management plan (CAMP) workshop. In: Proceedings, Regional Workshop on Sharing Local and National Experience in Conservation of Medicinal and Aromatic Plants in South Asia, Pokhara, Nepal, 2001, 46-53.
4. Agrawal RL. Seed technology. Oxford and IBH publishing company, 1995, 587.
5. Bewley JD, Black M. Control of the mobilization of stored reserves, seeds physiology of development and germination. *Plenum Press*, New York, 1994, 346- 351.
6. Bhuyar S, Wankhade SG, Paturde JT, Khode PP. Seed germination studies in sarpagandha (*Rauvolfia serpentina* Benth). Res. Crops 2000; 1(2):189-191.
7. Ponkumar P, Padma M, Rajkumar M, Madulety TY. Effect of chemicals and plant growth substances on breaking of seed dormancy in sarpagandha (*Rauvolfia serpentina* L.). J Res. ANGRAU. 2008; 36(1):54-56.
8. Ghosh DK, Nath A, Bandopadhyay A. Effect of substitutions of chemical nitrogen fertilizer with organics on growth and yield of sarpagandha grown under coconut platntation. In: *Proceedings of Int. Symp. Minor Fruits and Med. Pl.*, December 19-22, Kalyani, West Bengal, 2009, 207-210.
9. Paul D, Paul NK, Basu PK. Seed germination response of *Rauvolfia serpentina* Benth. To certain physical and chemical treatments. *J Bio-Sci*. 2008; 16:129-131.
10. Hussain A, Jha DK. Seed germination improvement in two threatened medicinal plants. Curr. Agric. Res. J. 2014; 2(2):131-136.
11. Muneshwar BR. Standardization of seed germination testing procedure in sarpagandha (*Rauvolfia serpentina* Benth.). *M. Sc. (Agri.) Thesis*, Mahatma Phule Krishi Vidyapeeth, Rahuri, Maharashtra, 2015.
12. Velmurugan S, Vadivel E, Paramaguru P. Studies on seed germination in Ashwagandha (*Withania somnifera*). In: Proc. Nat. Sem. New. Prespect. Sps. Med. Arom. Pl., 2003, 78.