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EFFECT OF GROWTH STIMULANTS ON SEED GERMINATION AND MORPHO-PHYSIOLOGICAL ATTRIBUTES IN PUNGAM (*PONGAMIA PINNATA*)

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VENKATESH, A., VANANGAMUDI, MALLIKA, VANANGAMUDI, K., PARTHIBAN, K. T., RAVICHANDRAN, V. & VINAYA RAI, R. S. 2000. Effect of growth stimulants on seed germination and morpho-physiological attributes in pungam (*Pongamia pinnata*). An investigation was carried out to study the effect of growth stimulants on seed germination and morpho-physiological attributes of seedlings in pungam (*Pongamia pinnata*). The growth regulators and chemicals used in this study were IAA (indole-3-acetic acid) (100 ppm), IBA (indole-3-butyric acid) (100 ppm), GA_3 (gibberellic acid) (100 ppm), KN (kinetin) (100 ppm), NAA (α-napthalene acetic acid) (100 ppm), ethrel (100 ppm), KCl (1%), KNO_3 (1%), $ZnSO_4$ (1%), KH_2PO_4 (1%), $FeSO_4$ (1%) and $MgSO_4$ (1%). The seeds were soaked in the respective solutions for 12 h. Water-soaked seeds served as control. Seeds soaked in IBA (100 ppm) gave high germination. IBA also enhanced the root length. GA_3 (100 ppm) enhanced the shoot length, total leaf area, total dry weight and total chlorophyll content more than other treatments. The values for most of the above parameters recorded for IBA (100 ppm) were comparable with those of GA_3 (100 ppm). KH_2PO_4 (1%) induced a greater soluble protein content than the water-soaked seeds and other growth promoting chemicals.

Key words: *Pongamia pinnata* - growth stimulants - germination - leaf area - dry weight - chlorophyll - soluble protein

VENKATESH, A., VANANGAMUDI, MALLIKA, VANANGAMUDI, K., PARTHIBAN, K. T., RAVICHANDRAN, V. & VINAYA RAI, R. S. 2000. Kesan perangsang pertumbuhan terhadap sifat-sifat percambahan biji benih dan morfo-fisiologi anak benih pungam (*Pongamia pinnata*). Penyiasatan dijalankan untuk mengkaji kesan perangsang pertumbuhan terhadap sifat-sifat percambahan dan morfo-fisiologi anak benih pungam (*Pongamia pinnata*). Penentu pertumbuhan dan bahan kimia yang digunakan dalam kajian ini ialah IAA (indole-3-asid asetik) (100 ppm), IBA (indole-3-asid butirik) (100 ppm), GA_3 (asid giberelik) (100 ppm), KN (kinetin) (100 ppm), NAA (a - asid asetik naftalena) (100 ppm), etrel (100 ppm), KCl (1%), KNO_3 (1%), $ZnSO_4$ (1%), KH_2PO_4 (1%), $FeSO_4$ (1%) dan $MgSO_4$ (1%). Biji benih direndam di dalam larutan masing-masing selama 12 jam. Biji benih yang direndam di dalam air bertindak sebagai kawalan. Biji benih yang direndam di dalam IBA (100 ppm) memberikan percambahan yang tinggi. Ia juga menambahkan panjang akar. GA_3 (100 ppm) menambahkan panjang pucuk, jumlah luas daun, jumlah berat kering dan jumlah klorofil lebih daripada rawatan-rawatan yang lain. Nilai bagi kebanyakan ciri-ciri di atas yang dicatatkan bagi IBA (100 ppm) adalah setanding dengan ciri-ciri bagi GA_3 (100 ppm). KH_2PO_4 (1%) mengaruhkan kandungan protein terlarut yang lebih banyak berbanding dengan biji benih yang direndam di dalam air dan lain-lain bahan kimia yang menggalakkan pertumbuhan.

Introduction

Pungam [*Pongamia pinnata* (L.) Pierre] is extensively planted for afforestation of watersheds in the drier parts of India (Anonymous 1969). It is fairly drought resistant, moderately frost hardy and highly tolerant to salinity. The amount and distribution of rainfall play a paramount role in the success or failure of an afforestation programme. As the rainfall received is not uniform over the years with a good amount of total annual rainfall received in some years as intensive storms followed by prolonged drought spells (Chitale 1994), pungam is regarded as a good choice. Supply of good quality planting stock material is also one of the deciding factors. A considerable amount of initial costs are involved in the production and supply of quality planting stock. The establishment and survival of the planting stock in the field are mainly dependent on the production of good quality planting stock, which is again controlled by several internal and external factors.

Application of growth regulators has attracted much attention in recent years, thanks to their effect on the growth and development of plants. In forestry, the application of growth regulators and nutrients has been extensively used for enhancing the growth and development of seedlings under nursery conditions (Bhatnagar & Singh 1981). They have a major role in enhancing shoot and root growth (Street 1966) and internal differentiation including initiation of cambial activity, xylem differentiation and annual ring formation (Wareing *et al.* 1964).

Studies on the effect of growth stimulants on germination and the production of elite seedlings are, however, scanty for all tropical tree species in general and for pungam in particular. As growth regulators may help to improve planting stock, experiments were performed to explore the effect of growth stimulants on pungam seedlings.

Materials and methods

Seeds of pungam were collected from Pommidi Range of Salem district in Tamil Nadu, cleaned and prepared for the experiment. Seeds were soaked for 12 h in 100 ppm of IAA (indole-3-acetic acid), IBA (indole-3-butyric acid), GA_3 (gibberellic acid), KN (kinetin), NAA (α-napthalene acetic acid), ethrel and 1% of KCl, KNO_3, $ZnSO_4$, KH_2PO_4, $FeSO_4$ and $MgSO_4$. Water-soaked seeds served as control. The chemicals (99% purity) were obtained from E. Merck (India) Limited, Chennai, India.

The presoaked seeds were shade dried for 30 min and sown in 15 × 25 cm polybags, filled with nursery soil mixture consisting of red soil, sand and farm-yard manure (FYM) (2:1:1). The experiment was set up in a completely randomised design and replicated twice. Four hundred seeds were used per replication.

The number of seeds germinated was counted and the germination was expressed in percentage. At 30, 60 and 90 days after sowing, ten seedlings in each treatment were selected at random and observed for shoot and root lengths, total seedling dry weight, total leaf area (LI-COR model LI-3000 leaf area meter), total

chlorophyll content (Yoshida *et al.* 1971), soluble protein (Lowry *et al.* 1951), transpiration rate and diffusive resistance (LI-1600 Steady State Porometer, LI-COR Lincoln, Nebraska, USA). The vigour index (VI) was derived by the following formula (Abdul-Baki & Anderson 1973),

VI = percentage germination × seedling length (cm), where seedling length is the sum of root and shoot lengths

The results were subjected to analysis of variance and tested for significance (p=0.05) as per Panse and Sukhatme (1967). Percentage values were transformed into arcsin values prior to statistical analysis.

Results and discussion

Compared to water-soaked seeds, enhanced germination was evident only in the following treatments, IBA, GA_3, KN, NAA and KH_2PO_4, which showed similar responses. Inhibition of germination was associated with ethrel, KNO_3, $ZnSO_4$, $FeSO_4$ and $MgSO_4$. Increased germination following application of 100 ppm IBA has been reported in *Picea smithiana* (Singh 1990), *Cassia fistula* and *Bauhinia purpurea* (Gopikumar *et al.* 1991). Growth regulators like IBA and GA_3 probably antagonise the effect of growth inhibitory substances and also enhance the rate of metabolism during germination (Verma & Tandon 1988). Auxins are critical in germination because of their effect on cell elongation in *Thyrsostachys siamensis* and *Dendrocalamus strictus* (Richa & Sharma 1994). The promoting effect of auxins on germination may be attributed to their indirect effect through change in the membrane permeability, solubilisation of carbohydrates through synthesis of different enzymes responsible for promotive effects and production of some precursors needed for germination. The auxins may exert their primary effect on the cell wall and change subcellular proton concentration (Jann & Aman 1977). The augmentation effect of 1% KH_2PO_4 on germination has earlier been reported in neem (Kumaran *et al.* 1996).

Seedlings produced from seeds soaked in GA_3 (100 ppm) showed an increase in shoot length by 50.6% over the wated-soaked control at 150 days after sowing (Table 1). Increased shoot length was also discernible due to NAA and ethrel. Mehrotra and Dadwal (1978) in teak and Pal *et al.* (1988) in *Dalbergia sissoo* reported that the application of nutrients had an additive effect on GA_3 which boosted seedling growth of nursery stock. Several research reports are available to endorse the enhancement of shoot by GA_3 application (Verma & Tandon 1988, Singh *et al.* 1995). Application of auxins was found to enhance the seedling length in *Cassia obtusifolia* (Singh & Murthy 1987a), *C. fistula* (Singh & Murthy 1987b), *Enterolobium cyclocarpum* (Brahman 1995) and *Albizia lebbeck* (Palani *et al.* 1996). Root length did not vary due to application of any of the treatments. Except for KN, $ZnSO_4$ and $FeSO_4$, all other treatments increased total leaf area but the magnitude of the increase was maximum with GA_3 (143%).

Table 1. Influence of growth stimulants on germination, shoot length and root length of pungam seedlings

Growth stimulant	Germination (%)	Shoot length (cm) Days after sowing 30	90	150	Root length (cm) Days after sowing 30	90	150
IAA, 100 ppm	51.6 (46.0)	21.5	23.1	33.7	36.25	44.50	49.16
IBA, 100 ppm	66.7 (54.8)	27.5	29.5	38.5	34.50	42.65	58.66
GA_3, 100 ppm	61.1 (51.4)	35.0	39.6	50.0	29.50	31.35	44.17
KN, 100 ppm	61.1 (51.4)	19.8	20.6	22.2	29.00	38.60	37.17
NAA, 100 ppm	61.0 (51.4)	20.0	21.3	43.5	15.00	24.50	40.67
Ethrel, 100 ppm	35.0 (36.3)	20.0	21.8	40.2	32.50	37.30	48.50
KCl, 1%	58.3 (49.8)	25.0	27.0	37.7	21.00	39.90	53.83
KNO_3, 1%	36.9 (37.4)	25.5	28.5	31.5	27.00	35.42	45.33
$ZnSO_4$, 1%	38.3 (38.2)	20.0	21.7	28.8	29.00	35.05	43.99
KH_2PO_4, 1%	63.3 (52.8)	22.6	26.6	38.0	26.00	37.10	52.33
$FeSO_4$, 1%	29.2 (32.7)	17.5	20.7	38.0	30.50	32.80	46.50
$MgSO_4$, 1%	43.3 (41.2)	18.6	21.0	38.3	26.50	39.70	50.00
Dry seed	45.0 (42.1)	14.0	17.0	35.3	19.00	32.95	37.80
Water soaked (control)	53.3 (46.9)	16.5	18.5	33.2	23.00	28.90	44.66
SEd	(1.67)		2.96			7.20	
CD(p=0.05)	(3.43)		5.97			ns	

SEd - standard error deviation
CD - critical difference
ns - not significant

(Figures in parentheses indicate arcsin values).

Relative to the water-soaked control, total dry weight was more in the IAA, IBA, GA_3, NAA, KCl and KH_2PO_4 treatments (Table 2). Several researchers have demonstrated the positive effect of growth regulators along these lines (Unnikrishnan & Rajeeve 1990, Kumaran *et al.* 1994, Palani *et al.* 1996). The treatment with GA_3 hastened the process of leaf differentiation which was not an actual increase in the number of leaves (Krishnamoorthy 1981). GA_3 (100 ppm) recorded higher chlorophyll content than other treatments (Table 2). Additional supply of nutrients and GA_3 stimulated the production of phytochrome which ultimately resulted in extension of leaf surface area (Prasad & Mohammad 1987) and synthesis of chloroplasts. GA_3 enhances mobilisation of reserve starch (Nanda & Dhindsa 1967) and chlorophyll concentration in the leaves, thereby indirectly increasing the rate of photosynthesis and net assimilation rate (Misra *et al.* 1968).

The soluble protein content was maximum in the IBA and NAA treatments (Table 3). The results are concordant with the findings for many agricultural crops. The enhancement of reserve starch and chlorophyll content is also responsible for increased soluble protein content (Sale & Campkell 1968). Transpiration rate and diffusive resistance did not differ due to the treatments.

From a holistic perspective, IBA (100 ppm) is recommended for maximising both germination and vigour in pungam.

Table 2. Influence of growth stimulants on total leaf area, total dry weight and total chlorophyll content of pungam seedlings

Growth stimulant	Total leaf area (cm^2 $plant^{-1}$) Days after sowing			Total dry weight (g $plant^{-1}$) Days after sowing			Total chlorophyll content (mg g^{-1}) Days after sowing		
	30	90	150	30	90	150	30	90	150
IAA 100 ppm	45	141	376	0.79	2.16	7.99	1.62	1.89	2.02
IBA 100 ppm	63	142	409	0.91	2.25	8.99	1.90	1.98	2.10
GA_3 100 ppm	73	158	496	1.17	2.03	9.58	2.10	2.47	2.59
KN 100 ppm	81	160	211	1.03	1.83	3.11	1.12	1.53	1.76
NAA 100 ppm	60	142	415	0.81	2.19	9.24	0.90	1.12	1.44
Ethrel 100 ppm	93	103	323	1.26	1.50	6.14	0.65	0.95	1.18
KCl 1%	91	100	327	1.04	1.69	7.29	1.07	1.38	1.62
KNO_3 1%	80	124	315	0.99	1.59	6.50	1.05	1.13	1.36
$ZnSO_4$ 1%	60	75	247	0.66	1.01	5.52	1.58	1.74	1.90
KH_2PO_4 1%	116	127	358	1.19	1.46	7.91	1.71	2.00	2.11
$FeSO_4$ 1%	65	145	213	0.79	1.93	5.79	0.43	0.82	1.09
$MgSO_4$ 1%	111	135	289	1.29	1.87	6.43	1.01	1.28	1.48
Dry seed	48	96	184	0.74	1.27	5.09	0.58	0.84	1.11
Water soaked (control)	56	87	204	0.78	1.28	5.86	0.55	0.91	1.15
SEd		29.7			0.458			0.064	
CD(p=0.05)		59.9			0.925			0.129	

SEd - standard error deviation
CD - critical difference

Table 3. Influence of growth stimulants on soluble protein, transpiration rate and diffusive resistance of pungam seedlings

Growth stimulant	Soluble protein (mg g^{-1}) Days after sowing			Transpiration rate (µg cm^{-1} s^{-1}) Days after sowing			Diffusive resistance (s cm^{-1}) Days after sowing		
	30	90	150	30	90	150	30	90	150
IAA 100 ppm	0.92	1.42	2.57	13.9	13.3	11.4	1.55	1.88	2.27
IBA 100 ppm	0.83	1.57	3.44	9.7	8.1	7.2	2.86	3.07	3.72
GA 100 ppm	0.77	1.25	2.92	18.1	16.2	14.2	0.83	1.06	1.39
KN 100 ppm	0.93	1.42	2.87	16.1	15.1	13.9	1.18	1.32	1.50
NAA 100 ppm	0.58	1.53	3.53	20.6	19.6	17.8	0.43	0.71	0.89
Ethrel 100 ppm	0.60	1.07	2.72	18.1	18.2	17.0	0.76	0.95	1.31
KCl 1%	1.02	1.47	2.40	13.6	13.0	12.9	1.55	1.90	2.07
KNO_3 1%	1.07	1.58	2.42	16.6	17.7	16.9	0.94	1.14	1.54
$ZnSO_4$ 1%	0.77	1.28	2.50	11.6	10.2	9.0	1.81	2.32	2.38
KH_2PO_4 1%	1.92	2.25	2.35	10.5	9.9	8.9	2.38	2.49	2.66
$FeSO_4$ 1%	1.23	1.80	2.00	13.1	11.6	10.2	1.59	1.72	2.32
$MgSO_4$ 1%	0.55	1.05	2.28	19.6	18.1	16.5	0.67	0.79	1.00
Dry seed	0.78	1.22	1.85	15.8	14.9	12.1	1.25	1.39	1.55
Water soaked (control)	0.55	1.07	1.65	16.4	15.3	13.3	0.93	0.97	1.28
SEd		0.127			1.15			0.186	
CD(p = 0.05)		0.257			ns			ns	

SEd - standard error deviation
CD - critical difference
ns - not significant

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