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STUDIES ON THE EFFECT OF CERTAIN CHEMICALS AND BIO-REGULATORS ON GERMINATION AND SEEDLING GROWTH IN GLORY LILY (*GLORIOSA SUPERBA* L.)

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Abstract

Glory lily (*Gloriosa superba* L.) is one of the important medicinal plants which has attained commercial importance in Tamil Nadu in recent times. The medicinal value of glory lily is commendable which is attributed to the presence of alkaloids, chiefly „Colchicine and „Gloriosine. The drug has so far been obtained from collection from wild sources but of late, the indiscriminate exploitation of tubers has affected its supply badly. This has necessiated establishment of commercial plantations to meet the demand of pharmaceutical industries. Being a newly domesticated crop, the appropriate production technologies for this medicinal plant have not yet been standardized on a scientific footing. Among the technologies of crop production, the method of propagation assumes greater importance due to its marked effect on growth and yield. With this background, studies were undertaken to standardize the propagation methods in glory lily to find out the suitable growth regulators/ chemicals to improve seed germination and seedling growth. The study was conducted at the Department of Horticulture, Faculty of Agriculture, Annamalai University. The results revealed that Gibberellic acid and ethrel treatments were found to accelerate the seed germination. GA_3 @ 200 ppm resulted in early germination (23.35 days) as compared to dry seeds (Control) which took 57.76 days. The percentage of germination was the highest with 56.55 % in GA_3 @ 200 ppm as against the least in the control. Gibberellic acid @ 200 ppm followed by ethrel @ 200 ppm produced maximum number of roots. Thiourea @ 1000 ppm followed by GA_3 @ 100 ppm were promising in improving the seedling height. Thiourea @ 1000 ppm followed by GA_3 @ 100-200 ppm were promising in increasing the leaf number. The fresh and dry weight of shoot were maximum at GA_3 @ 200 ppm, whereas the fresh and dry weight of roots were higher in cytokinin treatment (BA @ 50 ppm) followed by ethrel @ 200 ppm.

Key words : Glory lily, seed germination, GA_3, Ethrel, BA.

Introduction

India is endowed with a rich wealth of medicinal plants, which deserve special attention for extensive cultivation on a commercial scale. Among others, the Glory lily (*Gloriosa superba* L.) has attracted large scale cultivation in recent times. It belongs to the family Liliaceae, being differently called as “Kanvali Kizhangu” or “Karthigai Kizhangu” or “Kalappai Kizhangu” or “Kanthazh” in local vernacular (Tamil). large glabrous, herbaceous branching climber which arises from a perennial, fleshy tuberous rhizome. The petals darken in colour with age finally changing from scarlet to crimson. The medicinal value of glory lily is commendable which is attributed to the presence of alkaloids, chiefly, Colchicine and Gloriosine. Colchicine extracted from the tubers and seeds are medically used in the treatment of „gout and rheumatism. It is also used by breeders to induce polyploids in crop plants. It has been reported that the seeds contain comparatively a higher content of total alkaloids (0.81 per cent) and colchicine (0.60 per cent) as against the tubers have with 0.57 and 0.05 per cent respectively on dry weight basis, which indicates the greater commercial value of seeds. The drug has so far been obtained from collection from wild sources but of late, the indiscriminate exploitation of tubers has affected its supply badly. This has necessiated establishment of commercial plantations to meet the demand of pharmaceutical industries. Being a newly domesticated crop, the appropriate production technologies for this medicinal plant have not yet been standardized on a scientific footing. Seeds and tubers are being used for propagation of Glory lily. Propagation through tubers are

preferred as the seedlings take a longer time to come to flowering and yield. Seed propagation gets its importance as an exclusive method to multiply newer types collected during exploratory trips or in breeding programmes. It has been observed by farmers and breeders that wide variation in germination of seeds are experienced. In order to find out means to improve seed germination and seedling growth. Studies were undertaken with different chemicals and bio-regulators on germination and seedling growth of Glory lily.

Materials and Methods

The experiment was conducted in the Orchard field unit of Department of Horticulture, Annamalai University as a pot culture in a Completely Randomized Design (CRD) with 14 treatment combinations of different chemicals and growth regulators, *viz.,* dry seed (control), water soaking, soaking in GA_3 @ (100, 200, 300 ppm), BA (10, 20, 50 ppm), Ethrel (100, 200, 300 ppm) and Thiourea (1000, 2000, 3000 ppm).

The required quantity of gibberellin (GA_3) was dissolved in 95 per cent ethyl alcohol and diluted with distilled water to make a stock solution of 500 ppm. Further dilutions were made according to requirements using distilled water. A few drops of Teepol were added as a wetting agent.

The weighed quantity of cytokinin (Benzyl Adenine) at 2, 4 and 10 mg were dissolved separately in a few drops of 1 N KOH. The volume was made upto 200 ml with distilled water. A few drops of Teepol were added as a wetting agent.

The required quantity of ethrel (40 %) was measured and dissolved in distilled water and a 2 per cent stock solution was prepared. Subsequently, dilutions were made from this stock solution using distilled water to get the required concentrations.

Two hundred milligrams of thiourea were dissolved in 200 ml of distilled water to get 1000 ppm concentration. Similarly, 400 and 600 mg of thiourea were dissolved in 200 ml of distilled water separately to get solutions of 2000 and 3000 ppm concentration. A few drops of Teepol were added as a wetting agent.

Fifty seeds each were soaked with different growth regulator and thiourea solution as per the treatment schedule for 24 hours. They were then drained off the solution and sown immediately.

In control, the well filled dry seeds were sown directly in pots at 50 seeds per treatment per replication.

For water soaking treatment, fifty seeds were pre-soaked in distilled water for a period of 24 hours, shade dried for a short time and sown immediately.

The treated and un-treated seeds were sown @ 25 nos for each treatment/ replication in ½ size pots (15´ 15 cm size) filled up with germination mixture containing river sand and red earth at equal proportions. The pots were kept in greenhouse for germination with regular watering. The seedlings at one leaf stage were transplanted to full size pots (30´ 30 cm size) at one seedling per pot. These pots were filled with pot mixture comprising garden soil and FYM at equal proportions. A total of six seedlings (six pots) were kept for each treatment/ replication.

Observations were recorded on days taken for germination, germination percentage, days taken for appearance of first pair of leaves after germination, distance of the first pair of leaves from the base, number of roots, length of root, plant height, number of leaves, fresh weight of shoot, dry weight of shoot, fresh weight of roots and dry weight of roots.

The data were statistically analysed as applicable to Completely Randomized Design (Panse and Sukhatme, 1978). Wherever the results were found significant, Critical Differences (CD) were computed at 5 per cent level of probability to draw statistical conclusions.

Results and Discussion

The results on the effect of different growth regulators and chemicals on seedling growth attributes of glory lily are discussed hereunder.

Seed propagation becomes greatly relevant in Glory lily. wherever new types are to be multiplied after their collection during exploration. Besides, seed propagation is the only method in any hybridization and polyploidy breeding programmes. The scarcity of seed tubers for planting and their high cost have assumed as limiting factors. In this background, investigations were undertaken to study the effect of gibberellic acid, kinetin, ethrel and thiourea on seed germination of glory lily and the results obtained are discussed hereunder.

Speed of germination and germination percentage

The study revealed that GA_3 and ethrel treatments were found to accelerate the seed germination and its percentage. Especially, GA_3 @ 200 ppm showed early germination in 23.35 days, whereas the untreated seeds took 57.76 days. Similar results were obtained as regard to germination percentage, wherein seeds pre-treated with GA_3 @ 200 ppm showed highest germination percentage (56.55) followed by ethrel @ 100 and 200 ppm (48.50 %) as compared to the least (16.28 %) in the control (Table 1).

Diaz and Martin (1971) have reported that gibberellins can stimulate the germination of seeds whose dormancy is imposed by incomplete embryo development or presence of germination inhibitors and factors relating to physiological competence of the embryo axis. Ovcharov (1977) reported that dormancy in the freshly harvested seeds may be ascribed to the presence of large quantities of inhibitors and relatively, nil or lesser amount of growth promoters, particularly gibberellins and cytokinins and he further suggested that the dormancy can be broken in seeds by activating GA synthesis endogenously or by supplying them exogenously.

Table 1: Effect of pre-germination seed treatment on days taken for germination and percentage of germination in glory lily.

Tr. No.	Treatment details	Days taken for Germination	Germination percentage
T_1	Dry seed	57.76	16.28
T_2	water soaked	42.95	27.44
T_3	GA_3 - 100 ppm	23.80	45.23
T_4	GA_3 - 200 ppm	23.35	56.55
T_5	GA_3 - 300 ppm	24.84	54.24
T_6	BA - 10 ppm	44.79	24.80
T_7	BA - 20 ppm	45.39	23.24
T_8	BA - 50 ppm	39.15	30.21
T_9	Ethrel - 100 ppm	25.78	50.11
T_{10}	Ethrel - 200 ppm	27.80	48.31
T_{11}	Ethrel - 300 ppm	35.24	33.21
T_{12}	Thiourea - 1000 ppm	31.40	41.20
T_{13}	Thiourea - 2000 ppm	33.50	36.18
T_{14}	Thiourea - 3000 ppm	47.43	22.00
	SED	1.38	0.26
	CD (p=0.05)	2.98	0.56

Table 2: Effect of pre-germination seed treatment on number of roots and length of root in glory lily.

Tr. No.	Treatment details	Number of roots	Length of roots
T_1	Dry seed	2.09	6.55
T_2	water soaked	2.51	6.88
T_3	GA_3 - 100 ppm	4.29	8.62
T_4	GA_3 - 200 ppm	6.59	8.99
T_5	GA_3 - 300 ppm	3.75	9.17
T_6	BA - 10 ppm	2.47	7.11
T_7	BA - 20 ppm	2.70	6.34
T_8	BA - 50 ppm	2.21	4.37
T_9	Ethrel - 100 ppm	4.56	8.26
T_{10}	Ethrel - 200 ppm	5.49	7.37
T_{11}	Ethrel - 300 ppm	3.13	5.77
T_{12}	Thiourea - 1000 ppm	3.70	7.11
T_{13}	Thiourea - 2000 ppm	5.37	7.97
T_{14}	Thiourea - 3000 ppm	5.11	8.93
	SED	0.091	0.057
	CD (p=0.05)	0.196	0.123

Obviously, the exogenous treatment of seeds with GA_3 in the present study might have favourably acted to induce speedy germination.

In glory lily, Suparna (1991), Supari *et al*. (1993) and Raina *et al*. (2000) have reported early germination with high germination percentage due to GA_3 treatments which are in agreement with the results of the present study.

Ethylene in addition to its influence on fruit ripening, bud dormancy, leaf abscission and other growth processes is also known to stimulate seed germination of many species.

In recent times, a number of workers have reported that ethylene can break dormancy in seeds of a very large number of species.

In the present study, ethrel was also found promising next to GA_3 in accelerating the germination with higher percentage.

Chauhan (1978) in *Solanum khassianum*, Barman and Sarma (1985) in *Camellia sinensis*, Shekafandeh and Shaybany (1986) in Pistacia, Saini and Spencer (1987) in *Chenopodium album* have also reported the favourable effect of ethrel on seed germination which corroborates the results of the present study.

Earliness in production of first pair of leaves and their distance from the base

The plants under GA_3 seed treatment (200 ppm) followed by ethrel @ 100-200 ppm showed early production of first pair of leaves comparatively at a higher distance from the base of the plant as compared to control and water soaked treatments. This is obvious as gibberellins are well known for their action to induce shoot elongation. Utada and Suzuki (1974) in *Lilium auratum* and Bhujbal (1975) in *Phyllanthus emblica* observed good shoot development with GA_3 treatment. Shah and Gupta (1979) observed early seedling emergence with GA_3 @ 60 ppm as seed treatment in *Atropa belladonna*.

Root growth parameters

In the present study, GA_3 @ 200-300 ppm were found to influence the root growth in terms of higher number of lengthy roots. Besides GA_3, both the traits were also found to be influenced by thiourea (2000-3000 ppm) (Table 2). A number of researchers have reported the beneficial effects of GA_3 and thiourea on the root growth

Table 3: Effect of pre-germination seed treatment on plan height (cm) in glory lily.

Tr. No.	Treatment details	Plant height (cm) (Days after transplanting)					
		0 days	15 DAP	30 DAP	45 DAP	60 DAP	Per cent Increase*
T_1	Dry seed	4.10	4.78	5.13	5.40	5.60	36.59
T_2	water soaked	4.30	5.16	5.36	5.60	5.90	37.21
T_3	GA_3 - 100 ppm	5.01	6.01	6.37	6.76	7.58	51.29
T_4	GA_3 - 200 ppm	4.91	5.81	6.21	6.65	7.21	46.84
T_5	GA_3 - 300 ppm	4.59	5.49	5.87	6.14	6.50	41.61
T_6	BA - 10 ppm	4.39	5.19	5.46	5.86	6.07	38.26
T_7	BA - 20 ppm	4.33	5.03	5.39	5.79	6.09	40.66
T_8	BA - 50 ppm	4.20	4.87	5.16	5.56	5.86	39.52
T_9	Ethrel - 100 ppm	4.52	5.42	5.77	6.07	6.27	38.70
T_{10}	Ethrel - 200 ppm	4.70	5.60	5.70	6.06	6.18	31.49
T_{11}	Ethrel - 300 ppm	4.63	5.13	5.44	5.84	6.46	39.50
T_{12}	Thiourea - 1000 ppm	5.12	6.02	6.62	6.94	7.82	52.73
T_{13}	Thiourea - 2000 ppm	4.87	5.27	6.11	6.72	7.20	47.81
T_{14}	Thiourea - 3000 ppm	4.77	5.67	5.78	6.11	6.70	40.41
	SED	0.230	0.245	0.064	0.049	0.026	
	CD (p=0.05)	0.495	0.530	0.138	0.106	0.056	

* Per cent increase from initial to final day of observation.

parameters. Suparna (1991) has reported the profound effect of GA_3 (300 ppm) on root growth of the seedling in glory lily which is in line with the findings of the present study. Krishnan and Kulasekaran (1984) have also highlighted the role of thiourea in enhancing the root parameters in *Zyzyphus rotundifolia*.

Plant height

In the present study, significant increase in plant height from initial to final stage was observed in GA_3 (51.29 %) and thiourea (52.73 %) treated seeds, when compared to least increase (36.59 %) in untreated control (Table 3).

This increase might have been due to the early germination and better seedling vigour caused by GA_3 and thiourea treatments. Besides, better development of root system due to GA_3 and thiourea treatments might have resulted in efficient uptake of water and minerals resulting in a better seedling growth (Krishnan and Kulasekaran, 1984).

The beneficial effect of thiourea in enhancing the seedling growth has also been reported in glory lily (Suparna, 1991), which corroborates the results of the present study.

Number of leaves

In the present study, thiourea (1000 ppm) followed by GA_3 (100-200 ppm) were found to exert favourable effect on leaf production. This may be attributed to higher absorption and utilization of nutrients due to vigorous nature of seedlings with a good root system.

Krishnan and Kulasekaran (1984) reported higher leaf production in *Zizyphus rotundifolia* due to GA_3 @ 200 ppm treatment. Ziang (1982) has also reported higher

Table 4: Effect of pre-germination seed treatment on fresh and dry weight of shoot (g) in glory lily.

Tr. No.	Treatment details	Fresh weight of shoot (g)	Dry weight of shoot (g)
T_1	Dry seed	2.24	0.83
T_2	water soaked	2.55	0.94
T_3	GA_3 - 100 ppm	2.87	1.23
T_4	GA_3 - 200 ppm	3.37	1.45
T_5	GA_3 - 300 ppm	3.28	1.41
T6	BA - 10 ppm	2.73	1.03
T_7	BA - 20 ppm	2.71	1.03
T_8	BA - 50 ppm	2.83	1.22
T_9	Ethrel - 100 ppm	3.10	1.24
T_{10}	Ethrel - 200 ppm	2.95	1.18
T_{11}	Ethrel - 300 ppm	2.87	1.15
T_{12}	Thiourea - 1000 ppm	2.75	1.16
T_{13}	Thiourea - 2000 ppm	3.16	1.33
T_{14}	Thiourea - 3000 ppm	3.27	1.37
	SED	0.196	0.023
	CD (p=0.05)	0.424	0.050

Table 5: Effect of pre-germination seed treatment on fresh and dry weight of roots (g) in glory lily.

Tr. No.	Treatment details	Fresh weight of shoot (g)	Dry weight of shoot (g)
T_1	Dry seed	0.53	0.21
T_2	water soaked	1.56	0.61
T_3	GA_3 - 100 ppm	2.11	0.87
T_4	GA_3 - 200 ppm	2.08	0.87
T_5	GA_3 - 300 ppm	2.09	0.88
T6	BA - 10 ppm	2.10	0.97
T_7	BA - 20 ppm	2.18	1.00
T_8	BA - 50 ppm	2.37	1.09
T_9	Ethrel - 100 ppm	2.13	0.95
T_{10}	Ethrel - 200 ppm	2.16	0.95
T_{11}	Ethrel - 300 ppm	2.10	0.92
T_{12}	Thiourea - 1000 ppm	2.10	0.79
T_{13}	Thiourea - 2000 ppm	2.09	0.84
T_{14}	Thiourea - 3000 ppm	2.09	0.84
	SED	0.023	0.019
	CD (p=0.05)	0.050	0.042

leaf number due to GA_3 treatment (500 ppm) in *Coptis chinensis*. Suparna (1991) in glory lily has also reported a similar beneficial effect of thiourea in leaf production which is line with the findings of the present study.

Shoot and root weight

In the present study, fresh and dry weight of shoot was found to be maximum in GA_3 @ 200 ppm followed by thiourea @ 3000 ppm (Table 4). This response may be attributed to higher shoot length and leaf number brought about by early germination and vigorous growth of the seedlings due to GA_3 and/ or thiourea treatments leading to higher accumulation of dry matter.

The root fresh and dry weight were maximum in BA (Kinetin) @ 50 ppm, whereas GA_3 @ 200 ppm ranked next in recording higher root fresh and dry weight (Table 5).

Effect of BA (Kinetin) on tuber formation is well supported by the findings of Ginzburg and (1973) in gladiolus where kinetin induced formation of cormels, whereas GA_3 inhibited it. As GA_3 influences shoot growth predominantly, its effect on root formation was not well expressed in the present study.

To sum up, the present study revealed that pretreatment of seeds with Gibberellic acid and Ethrel were found to increase the seed germination, early production of first pair of leaves, and maximum number of lengthier roots. Thiourea and gibberellic acid treatments were found to enhance the seedling height with increased the leaf number. The fresh and dry weight of shoot were maximum with GA_3 treatments. The fresh and dry weight of roots were higher with cytokinin treatment (BA @ 50 ppm) followed by ethrel @ 200 ppm.

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