

LOW INPUT TECHNOLOGY FOR COMMERCIAL CULTIVATION OF *GLADIOLUS*

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Commercial floriculture is a profitable agro industry in many developed countries. Many growers in India and other developing countries are also taking up large-scale cultivation of floricultural crops as a viable alternative for the traditional cash and field crops. However, many of the imported varieties require a heavy input of fertilizers and irrigation resulting in an increase in cost of cultivation and reduction in the margin of profit. In recent years vesicular arbuscular fungi (VAM fungi) and phosphate solubilizing microbes (PSM) have become important biotechnological tools and are being employed to reduce the input of fertilizers and irrigation. A number of VAM fungi and PSM were evaluated for their efficacy in improving the performance of *Gladiolus*. A package of specific VAM- *Glomus fasciculatum* and PSM- *Aspergillus niger* was developed for less expensive cultivation of this important floricultural crop.

Keywords: *Aspergillus niger*; Floriculture; *Gladiolus*; *Glomus fasciculatum*; Phosphate solubilizing microbes; VAM technology.

Introduction

Floriculture is now emerging as an important venture in the world. Flowers easily suppress almost all agricultural crops in respect of the economic potential. The international floriculture trade is estimated to be the order of billions of dollar. Cut flowers account for 60% of this turn over. Over the last decade there has been sharp increase in the demand for the flowers all over the world. The important floricultural crops in the international cut flower trade are rose, carnation, chrysanthemum, gladiolus, tulip, anthurium, lilies, gerbera, orchids, iris and lisanthus etc. Out of these, *Gladiolus* occupies a prime position in floriculture industry. However, its fertilizer and water requirements are very high, increasing the cost of production in multiples. Because of expensive cultivation, it is difficult for the growers to maintain the quality of the flowers within their limited resources. To catch up the export market, there is a need for scientific approach, well planned and well executed. Development of any technology, which may curtail the cost of cultivation of such varieties, will go a long way to help the farmers.

During last four decades, much attention has been paid to VAM fungi as a tool for improving the growth of agricultural and horticultural plants. The plant growth is stimulated because of increased uptake of phosphorus as well as zinc, copper, sulphur, potassium, calcium¹ and improved water economy². On the other hand, phosphate

solubilizing microorganisms have been shown to be successful in enhancing the performance of plants by solubilizing the bound phosphates and making them available to the plants³.

The present work has been planned to investigate the synergistic effect of VAM, *Glomus fasciculatum* and PSM, *Aspergillus niger* to improve the performance of *Gladiolus* and to curtail external input of fertilizer.

Materials and Methods

The experiment was planned to test the potentiality of selected VAM and PSF in curtailing the dose of phosphorus fertilizers. Selected inoculants of VAM and PSF were evaluated for the efficacy in improving the quality and number of flowers and in increasing keeping quality and duration of flowering by employing three different doses of phosphorus fertilizers (Super phosphate, full dose: 180 kg per hectare; half dose: 90 kg per hectare; quarter dose: 45 kg per hectare and no fertilizer for *Gladiolus*).

Following treatments were planned:

Gladiolus cv. Aldebaran

1. Control (Full dose of P fertilizer);
2. Full dose of P fertilizer + selected VAM;
3. Full dose of P fertilizer + selected PSF;
4. Full dose of P fertilizer + selected VAM + selected PSF;
5. Half dose of P fertilizer + selected VAM + selected PSF;
6. Quarter dose of P fertilizer + selected VAM + selected

PSF; 7. No fertilizer + selected VAM + selected PSF.
Selected VAM - *Glomus fasciculatum*; Selected PSF - *Aspergillus niger*.

Usual nursery practice was followed for raising the crops. Earthen pots were filled with pot mixture having soil, FYM and leaf mould in a proportion of 1:2:2. Pot mixture was sterilized before use by spraying 2% formaline. Then it was covered with alkanthine sheet for about a week.

Corms of *Gladiolus* were procured from the nursery of NBRI, Lucknow, India aseptically in sterilized polythene bags. Cuttings of equal size were planted in earthen pots at rate of one cutting per plant. Before planting them the inoculum of VAM fungi and PSF were supplemented to the pot mixture. For inoculating PSF, 20 ml of PSF inoculum, conidial suspension containing 9×10^6 conidia / ml prepared from PSF culture raised on liquid potato dextrose medium was mixed thoroughly in lower half portion of pot mixture in each pot. For adding the VAM inoculum the upper layer of pot mixture up to a depth of 5 cm was removed and mycorrhizal inoculum consisting of infected root pieces and spores (250 spore / pot) collected from the pot culture was spread as a layer over the surface. The soil removed earlier was replaced in the pot. In order to ensure the development of mycorrhization, the corms were planted in such a way that the inoculum layer came in contact with them. Super phosphate was added to the pot mixture in pots to achieve full (60 g / pot), half (30 g / pot), and quarter (15 g / pot) dose of fertilizer. 40g /pot nitrogen in the form of ammonium nitrate, 35g / pot potassium in the form of potash (120 kg / hectare and 105 kg / hectare respectively) were added in pot mixture for *Gladiolus*. Pots of *Gladiolus* were irrigated appropriately at the interval of 1 or 2 days according to soil moisture. Doses of potash and urea (10 g + 5 g per pot) were added to the plant fortnightly until budding was initiated. Watering followed it.

Seven replicates were taken for every treatment and the pots of treatment and control series were maintained under green house condition.

Plants from treatment as well as control series were uprooted at vegetative pre-flowering and flowering stages. Samples of root with adhering soil were collected and processed for determining the mycorrhizal intensity in roots by the method of Philips and Hayman⁴, population of VAM spores by the wet sieving and decanting method given by the Gerdemann and Nicolson⁵ using the sieves of different sizes (500 μ m, 210 μ m, 150 μ m, 90 μ m and 60 μ m) and PSF in rhizosphere by using serial dilution plate technique given by Timonin⁶ on the solid Pikovaskaya's medium, girth of plants, number of branches, length, fresh

and dry weight of roots and shoots were recorded. Number of buds, number of flowers, duration of flowering, keeping quality of flowers and diameter as well as fresh and dry weight of flowers was also recorded.

Statistical Analysis - The experiments were performed in pot condition and for each set 7 replicates were taken. Significant differences were determined by Randomized block and Latin square statistical method calculating critical difference at 5% level⁷.

Results and Discussion

Test of the potentiality of selected VAM and PSF for Gladiolus- Selected VAM inoculant (*Glomus fasciculatum*) and selected PSF inoculant (*Aspergillus niger*) were evaluated for their efficacy in improving the performance of *Gladiolus* in combination with different doses of phosphorus fertilizer (super phosphate, full dose, half dose, quarter dose and no fertilizer). Mycorrhizal status, growth of shoots and roots, period of flowering and budding, number of buds and flowers, fresh and dry weight of flowers, keeping quality of flowers were recorded.

Mycorrhizal status - The mycorrhizal infection in the roots and spore population in the rhizosphere of *Gladiolus* raised in field soil provided with selected VAM and PSF inoculants and supplemented with different doses of phosphorus fertilizer, at different stages of growth is presented in Table 1. Both VAM as well as PSF inoculants individually as well as in combination improved the mycorrhizal intensity in roots and VAM spore population in the rhizosphere provided with full dose of P fertilizer. However, the improvement varied with the treatment as well as the stages of the plant growth. Maximum improvement in mycorrhizal intensity (average in the stages) was recorded when plants were provided with VAM inoculant alone.

Improvement in the mycorrhizal status was also observed even when the dose of P fertilizer was curtailed. However, maximum improvement was recorded when the plants were not given fertilizer.

Population of PSF - Population of PSF in the rhizosphere soil of *Gladiolus* raised in field soil inoculated with selected VAM and PSF inoculants and supplemented with different doses of phosphorus fertilizer, at different stages of growth is presented in Table 2. When full dose of fertilizer was provided to the crop, the effect on the PSF population in the rhizosphere varied with the inoculants and stages of plant growth. Both VAM as well as PSF inoculants individually as well as in combination improved the PSF population in the rhizosphere soil. Maximum population (average of 3 stages) of PSF in rhizosphere was recorded when both the inoculants were used in combination.

Table 1. Mycorrhizal intensity in the roots and spore population in the rhizosphere soil of *Gladiolus* raised in soil supplemented with VAM and /or PSF inoculants and provided with different doses of P fertilizer at different stages of growth under pot conditions.

Treatments	Mycorrhizal Intensity (% Root bits infected)				Spore Population (Number/20g air dry soil)			
	Stages of plant growth				Stages of plant growth			
	Veg.	Pre- fl	Fl	Av	Veg	Pre-fl	Fl	Av
Control	21	32	38	30	52	78	85	72
Full dose + VAM	35	44	47	42	65	87	102	85
Full dose + PSF	25	37	45	36	61	75	82	73
Full dose + VAM + PSF	29	38	49	39	68	82	95	82
Half dose + VAM + PSF	31	46	52	43	72	95	107	91
Quarter dose + VAM + PSF	38	49	58	48	85	98	110	98
Nil dose + VAM + PSF	41	52	58	50	92	105	112	103

(Veg: Vegetative, Pre-fl: Pre flowering, Fl: Flowering, Av: Average)

VAM: *Glomus fasciculatum*

PSF: *Aspergillus niger*

Minimum difference required for significance (C.D.) at 5% level:

Mycorrhizal intensity: 1.124

Spore population : 1.426

Control: The plants of control series were provided with full dose of fertilizer but no VAM and / or PSF inoculants.

Table 2. Population of phosphate solubilizing fungi in the rhizosphere soil of *Gladiolus* raised in soil supplemented with VAM and /or PSF inoculants and provided with different doses of P fertilizer at different stages of growth under pot conditions. (1×10^5 /g oven dry soil).

Treatments	Stages of Plant Growth			
	Veg.	Pre- fl	Fl	Av
Control	0.68	1.25	0.85	0.92
Full dose + VAM	0.55	1.85	0.75	1.05
Full dose + PSF	6.25	7.83	6.22	6.76
Full dose + VAM + PSF	17.82	21.32	17.32	18.82
Half dose + VAM + PSF	9.87	13.23	8.34	10.48
Quarter dose + VAM + PSF	7.34	8.15	6.25	7.24
Nil dose + VAM + PSF	5.24	6.89	5.12	5.75

(Veg: Vegetative, Pre-fl: Pre flowering, Fl: Flowering, Av: Average)

VAM: *Glomus fasciculatum*

PSF: *Aspergillus niger*

Minimum difference required for significance (C.D.) at 5% level:

PSF population: 3.128

Control: The plants of control series were provided with full dose of fertilizer but no VAM and / or PSF inoculants.

Table 3. Height and fresh/ dry weight of shoot of *Gladiolus* raised in soil supplemented with VAM and /or PSF inoculants and provided with different doses of P fertilizer at different stages of growth under pot conditions.

Treatments	Height of Plant (cm)	Fresh Weight of Shoot (g/Plant)	Dry Weight of Shoot (g/Plant)
Control	82.3	58.110	11.020
Full dose + VAM	95.4	67.725	14.101
Full dose + PSF	89.1	65.112	13.916
Full dose + VAM + PSF	110.2	87.056	16.202
Half dose + VAM + PSF	105.3	85.108	15.025
Quarter dose + VAM + PSF	83.8	60.105	12.156
Nil dose + VAM + PSF	63.0	42.129	8.256

VAM: *Glomus fasciculatum*PSF: *Aspergillus niger*

Minimum difference required for significance (C.D.) at 5% level:

Height of plant : 0.7074

Fresh weight of shoot : 0.6568

Dry weight of shoot : 0.6359

Control: The plants of control series were provided with full dose of fertilizer but no VAM and / or PSF inoculants.

Table 4. Length and fresh/ dry weight of root of *Gladiolus* raised in soil supplemented with VAM and /or PSF inoculants and provided with different doses of P fertilizer at flowering stage under pot conditions.

Treatments	Root Length (cm)	Fresh Weight of Root (g / plant)	Dry Weight of Root (g / plant)
Control	28.3	12.320	1.242
Full dose + VAM	30.7	16.191	1.570
Full dose + PSF	31.3	15.051	1.533
Full dose + VAM + PSF	35.6	20.302	2.125
Half dose + VAM + PSF	33.6	18.160	1.686
Quarter dose + VAM + PSF	29.4	13.323	1.502
Nil dose + VAM + PSF	20.8	9.765	0.883

VAM: *Glomus fasciculatum*PSF: *Aspergillus niger*

Minimum difference required for significance (C.D.) at 5% level:

Length of root : 1.6390

Fresh weight of root : 0.808

Dry weight of root : 0.4891

Control: The plants of control series were provided with full dose of fertilizer but no VAM and / or PSF inoculants.

Table 5. Time of emergence of spike/initiation of flowering number of buds/flower per spike in *Gladiolus* raised in soil supplemented with VAM and /or PSF inoculants and provided with different doses of P fertilizer under pot conditions.

Treatments	Period of emergence of spike	Initiation of flowering	Buds/spike	Flowers/spike
Control	93 rd day	105 th day	10	10
Full dose + VAM	85 th day	99 th day	11	11
Full dose + PSF	90 th day	100 th day	11	11
Full dose + VAM + PSF	78 th day	91 st day	14	14
Half dose + VAM + PSF	80 th day	91 st day	14	14
Quarter dose + VAM + PSF	92 nd day	105 th day	10	10
Nil dose + VAM + PSF	110 th day	125 th day	8	8

VAM: *Glomus fasciculatum*

PSF: *Aspergillus niger*

Minimum difference required for significance (C.D.) at 5% level:

Period of emergence of spike: 2.332

Initiation of flowering : 2.326

Number of buds : 1.995

Number of flower : 1.129

Control: The plants of control series were provided with full dose of fertilizer but no VAM and / or PSF inoculants.

Table 6. Length of spike, fresh/dry weight and keeping quality of flowers in *Gladiolus* raised in soil supplemented with VAM and /or PSF inoculants and provided with different doses of P fertilizer under pot conditions.

Treatments	Length of spike (cm)	Weight of Flowers/Plant		Keeping Quality of Flowers	
		Fresh weight (g)	Dry weight (g)	On plants (Days)	Detached (Days)
Control	56.2	12.50	4.50	12	6
Full dose + VAM	60.3	16.50	5.28	14	7
Full dose + PSF	58.7	16.05	5.50	13	6
Full dose + VAM + PSF	65.8	20.10	7.70	18	8
Half dose + VAM + PSF	63.7	19.68	7.28	16	8
Quarter dose + VAM + PSF	54.4	13.09	4.89	11	5
Nil dose + VAM + PSF	49.5	8.00	2.56	8	5

VAM: *Glomus fasciculatum*

PSF: *Aspergillus niger*

Minimum difference required for significance (C.D.) at 5% level:

Length of spike : 1.847

Fresh weight of flowers : 1.235

Dry weight of flowers : 0.168

Keeping quality of flowers on plants: 1.548

Keeping quality of flowers detached: 1.637

Control: The plants of control series were provided with full dose of fertilizer but no VAM and / or PSF inoculants.

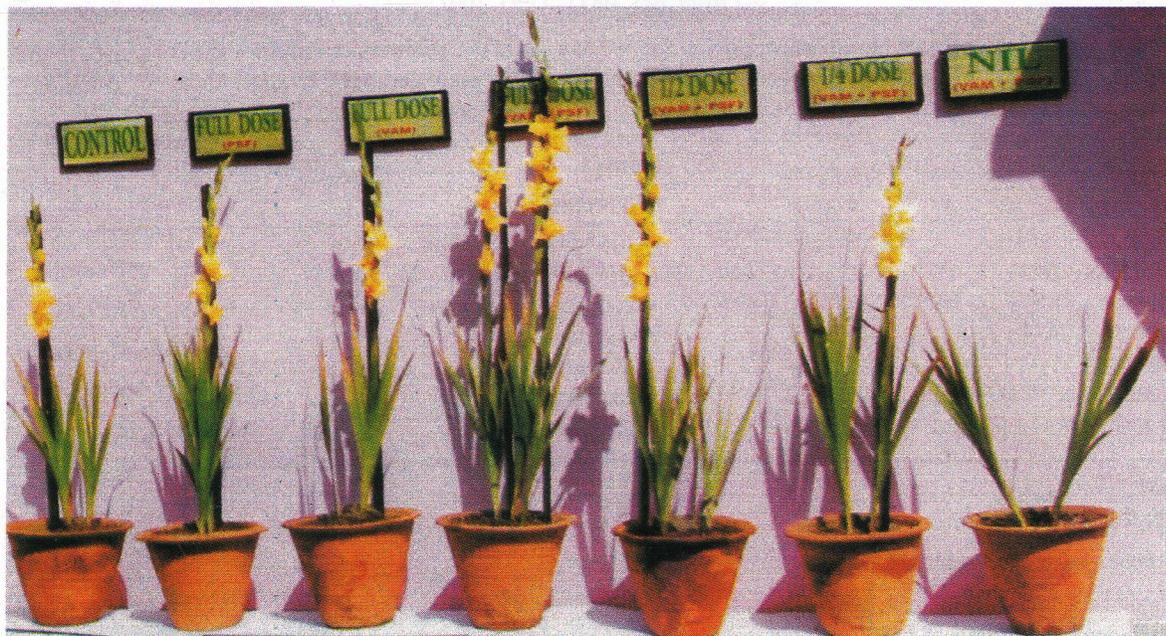


Fig.1. Performance of *Gladiolus* cv. Aldebaran (Flowering Stage) raised in unsterilized soil and inoculants individually and in combination and provided with full or curtailed doses of phosphatic fertilizer.

A combination of both the inoculants improved the PSF population in the rhizosphere soil of *Gladiolus* even at reduced doses of P fertilizer. However, the improvement varied with doses of fertilizer and stages of plant growth. Maximum improvement in the population (average of three stages) was recorded when the dose of fertilizer was curtailed to half.

Introduction of *G. fasciculatum* improved the mycorrhizal status of *Gladiolus*. This was a clear indication of its superiority as a colonizer over indigenous VAM fungi present in soil. Likewise, introduction of *A. niger* improved its population in the rhizosphere soil. This was a clear indication that it could establish in the field soil.

Potentiality of VAM inoculants in improving the mycorrhizal association and performance of diverse crops in field soil having not only indigenous VAM fungi but also other microbes has been reported by many workers⁴⁰. The present findings are in conformity with these reports and show that the selected VAM inoculant could sustain the competition of indigenous VAM fungi and other microbes in field soil and was able to maintain its potentiality.

Growth of shoots - The height of plants as well as fresh and dry weight of the shoots of *Gladiolus* raised in field soil provided with selected VAM and PSF inoculants and supplemented with different doses of phosphorus fertilizer,

at different stages of growth is presented in Table 3. Both VAM as well as PSF inoculants individually as well as in combination improved the height, fresh weight and dry weight of the shoots in *Gladiolus* when they were provided with full dose of P fertilizer. However, the improvement varied with the treatments as well as stages of plant growth. Maximum improvement in the height as well as fresh and dry weight of the shoots was shown when plants were provided with both inoculants.

A combination of both the inoculants improved the height of the plants as well as fresh and dry weight of the shoots of *Gladiolus* even when dose of P fertilizer was curtailed to half. However, when the plants were provided with quarter dose or no fertilizer was provided to them, there was either only slight improvement or a decrease in these parameter.

Growth of roots- The length as well fresh and dry weight of the roots of *Gladiolus* raised in field soil provided with selected VAM and PSF inoculants and supplemented with different doses of phosphorus fertilizer at different stages of growth are presented in Table 4. Both VAM as well as PSF inoculants individually as well as in combination improved the length, fresh weight and dry weight of the roots in *Gladiolus* provided with full dose of fertilizer. However, the maximum improvement in the length as well as fresh and dry weights of the roots was shown when

plants were provided with both the inoculants.

A combination of both the inoculants improved the length as well as fresh and dry weights of the roots of *Gladiolus* even when the dose of P fertilizer was curtailed to half. However, when the plants were provided with quarter dose or no fertilizer was provided to them, there was either only slight improvement or a decrease in these parameters.

Budding and flowering - Period of emergence of spike and initiation of flowering, number of buds / spike, number of flowers / spike, fresh and dry weights of the flowers per plant and keeping quality of flowers in *Gladiolus* raised in field soil provided with selected VAM and PSF inoculants and supplemented with different doses of phosphorus fertilizer, are presented in Tables 5 & 6. Both VAM and PSF inoculants individually as well as in combination caused preponement of emergence of spike and initiation of flowering in *Gladiolus* provided with full dose of P fertilizer. However, the period of preponement varied with treatments. Maximum preponement was shown when plants were provided with both the inoculants in combination.

A combination of both the inoculants preponed the emergence of spike and flowering even when the dose of P fertilizer was curtailed to half. However, when the plants were given quarter dose or no fertilizer was given to them, there was either very little preponement or the emergence of spike and initiation of flowering was delayed. Both VAM as well as PSF inoculants individually as well as in combination increased the number of buds and flowers per spike in *Gladiolus* provided with full dose of P fertilizer. However, the magnitude of increase varied with the treatments. Maximum increase was shown when plants were provided with both inoculants.

A combination of both the inoculants caused an increase in the number of buds and flowers per spike even when the dose of P fertilizer was curtailed to the half. However, the dose was reduced to quarter or no fertilizer was given to the plants, the number of buds and flower per spike was either decreased or there was no change.

Both VAM as well as PSF inoculants individually as well as in combination caused an increase in the length of spike, fresh and dry weights of the flowers per plant and keeping quality of flowers in *Gladiolus* provided with full dose of P fertilizer. However, magnitude of increase varied with treatments. Maximum increase was shown in above parameter when the plants were provided with both the inoculants.

A combination of both the inoculant caused an increase in the length of spike, fresh and dry weights of

the flowers per plant and keeping quality of flowers even when the dose of P fertilizer was curtailed to half (Fig. 1). However, when the plants were given quarter dose of fertilizer or no fertilizer was given to them, there was either only slight improvement in these parameters or there was an unfavorable effect.

VAM inoculation has been shown to favour the vegetative growth and flowering in a number of floricultural crops¹³⁻¹⁵. This supports the present findings and is a clear indication that VAM fungi through their multiple benefits may be employed also for improving the performance of floricultural crops.

There are many reports which show that introduction of phosphate solubilizing microorganism in soil improve the performance of crops^{16,17}. Similar observation was recorded with *Gladiolus* in the present study. The beneficial effect of phosphate solubilizing microorganism has been attributed to their potentiality to ensure better phosphate nutrition to the plants. The organic acids produced by them act on the phosphorus present in the soil in unavailable forms.

Improvement in growth and performance of *Gladiolus* by a combination of selected VAM and PSF in the present study is in the conformity with similar reports by many other investigators^{18,19}. It has been suggested that combination of both the inoculants ensures not only increased supply of available phosphorus to the plants but also its better absorption by them. Better performance of the plants is because of sustainable availability of Phosphorus to the plants²⁰.

A critical analysis of the present findings clearly indicate that with selected VAM and PSF inoculants, the performance of both the crops with half dose of P fertilizer was as good as the full dose of fertilizer. This gives a clear conclusion that 50% phosphate fertilizer recommended for the cultivation of *Gladiolus cv. Aldebaran* can be saved by inoculating them with selected VAM and PSF inoculants.

There are several reports, which indicate the importance of VAM fungi in curtailing the dose of P fertilizer²¹⁻²⁵. In China aster, use of VAM inoculants gave a saving of 25% of recommended dose of P fertilizer²⁶. These reports support the present findings.

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