Abstract: Vesicular arbuscular mycorrhizal fungi form symbiotic association with roots of most terrestrial plants including many agricultural crops. These are known to occur globally in a broad range of dissimilar environments from arctic to tropic and occupy a wide range of ecological niches. The role of VAM fungi in the improvement of crop plants is well documented. VAM fungi are known to improve the nutrient status of the plants, increase growth and development, protect plants against pathogens and confer resistance to drought and salinity. Sustainable agriculture has become a serious issue due to the energy crisis and environmental issues. Therefore, agricultural production is aimed to produce maximum output with minimum input. During agricultural production it is necessary to use organic fertilizers rather than chemical fertilizers. In the present study, authors assessed the efficacy of inoculants of three species of VAM fungi singly and in combination, on growth of tomato.

Keywords: Pathogen, Resistance, Symbiotic, Tomato, Vesicular arbuscular mycorrhiza (VAM).

INTRODUCTION
Agriculture is the most important and the critical sector of the economy of any country which is nowadays badly influenced by indiscriminate anthropogenic activities (Prakash and Verma, 2022). The increased use of chemical fertilizers in agriculture has increased the food grain production and helped the country in achieving self-sufficiency in food grains but also produced many harmful effects like environmental problems including water supply contamination, shortage in quality of agricultural products, decrease in the amount of soil fertility. The VAM is used to benefit microorganisms in various purposes, such as mining, agriculture, biotechnology, environmental science and so on. These are required for the establishment of ecological balance and maintenance of biological diversity. The conservation and maintenance of rich biodiversity are essential for the survival of the entire biotic world including plants, animals and humans with ecosystem sustainability (Ashok, 2018). Sustainable development is an utmost need of today across the globe that must be achieved with environmental ethics (Verma, 2019).
The vesicular arbuscular mycorrhiza benefits the plants by improving the supply of nutrients, especially phosphorus and other minerals such as, Zn, Cu, S, K, and Ca (Cooper and Tinker, 1978) and the plant supplies the fungus with photosynthetic sugars (Verma and Schuepp, 1995). Pure cultures of single species of VAM fungus are being assessed for the appreciable plant growth and crop production. VAM fungi increased biomass production of sustainable agricultural crops. Multiple inoculation of the plants with VAM fungi has often yielded increased biomass production, but less emphasis has been paid to exploit their practical utilization (Hepper et al., 1987; Sieverding, 1988; Kumar, 1990). In the present study, three species of VAM fungi, i.e., Glomus constrictum Trappe, Glomus mosseae, and Glomus fasciculatum have been assessed, singly and in combination, for their efficacy as potential VAM inoculants for tomato.

MATERIALS AND METHODS
Three species of VAM fungi were screened for their efficacy. Pure culture of all the three VAM species which were maintained on buffel grass (Cenchrus ciliaris L.) in the 12 inches pots containing sands were used for inoculation. Steam-sterilized sandy loam soil sand mix (1:1 volume/volume) substrate was dried and potted in 1 kg capacity clay pots. Sixty grams of single or mixed dry soil inoculum containing 400-450 spores/50 g soil was mixed in the top 6 cm of the soil of each treatment pot. Control plants received 60 g of soil containing non-mycorrhizal root pieces of buffel grass. Preliminary studies with the standardization of the dose of soil inoculum showed that this quantity of mycorrhizal inoculum was sufficient for the production of a reasonable amount of infection. The amount of VAM inoculum for each treatment was so adjusted that equal quantity of soil inoculum could be added to each pot. The different doses of soil inoculum used for each treatment were as follows:

- Single Endophyte - 60 g,
- Double Endophyte - 30 + 30 g, and
- Triple Endophyte - 20 + 20 + 20 g.

Each treatment was replicated seven times, and the following treatments were included in the study:

- Soil without inoculation (control),
- Soil inoculated with Glomus constrictum (60 g),
- Soil inoculated with Glomus mosseae (60 g),
- Soil inoculated with Glomus fasciculatum (60 g),
- Soil inoculated with Glomus constrictum (30 g) and inoculum of Glomus mosseae (30 g),
- Soil inoculated with Glomus constrictum (30 g) and inoculum of Glomus fasciculatum,
- Soil inoculated with Glomus mosseae (30 g) and inoculum of Glomus fasciculatum (30 g) and
- Soil inoculated with Glomus constrictum (20 g) Glomus mosseae (20 g) and inoculum of Glomus fasciculatum (20 g).

Five surface-sterilized (10% sodium hypochlorite for three minutes) seeds of tomato (Lycopersicum esculentum L. cv. Pusa Ruby) were sown in each pot above the soil inoculum. Seedlings were thinned to one seedling/pot seven days after germination, pots were maintained under greenhouse conditions. Plants were uprooted periodically and per cent colonization of the roots was assessed by methods of Phillips and Hayman (1970). The number of VAM fungal spores were extracted from the rhizosphere soil by wet sieving and decanting method (Gerdemann and Nicolson, 1963) and spore count was recorded. Biomass production was recorded in the term of shoot and root fresh weight and shoot and root dry weight, phosphorus content in the shoots and roots in term of g/mg tissue (Anderson and Ingram, 1989). The data were statistically analysed (Panse and Sukhatme, 1985).

RESULTS AND DISCUSSION
All the three VAM fungi were tested for their efficacy on tomato singly and in various combinations. All the treatments showed improvement in the mycorrhizal colonization, number of chlamydospores recovered from the rhizosphere soil, plant biomass, and phosphorus contents of the shoots and roots. However, all the three VAM fungi tested behaved differently in different combinations and differ from one species to another species.
Maximum growth in plants and spore count was observed by the inoculation of *Glomus mosseae* followed by *Glomus constrictum* and *Glomus fasciculatum* respectively. In combination, the maximum increase was observed in the plants which received *Glomus mosseae* and *Glomus constrictum* followed by *Glomus constrictum* and *Glomus fasciculatum* respectively. However, highest increase was observed in the shoot biomass, mycorrhizal colonization in P content in the shoots and roots when the plants were inoculated with all the three VAM fungi. The result is shown in table 1.

| Treatment          | Shoot Fresh wt. (g) | Root fresh wt. (g) | Shoot dry wt. (g) | Root dry wt. (g) | Phosphorus g/mg | Colonization (%) | No. of chlamydospores recovered from 20g soil |
|--------------------|---------------------|--------------------|-------------------|------------------|-----------------|------------------|---------------------------------------------|
| Control            | 39.20               | 3.2                | 3.32              | 0.20             | 1.05            | 0.25             | 10                                          | 32                                          |
| *G. constrictum* (Gc) | 62.5               | 5.0                | 5.85              | 0.68             | 1.23            | 0.17             | 86                                          | 170                                         |
| *G. mosseae* (Gm)  | 53.3                | 4.9                | 4.85              | 0.62             | 1.12            | 0.30             | 80                                          | 196                                         |
| *G. fasciculatum* (Gf) | 41.5               | 4.6                | 3.82              | 0.31             | 1.10            | 0.27             | 75                                          | 141                                         |
| Gc + Gm            | 69.5                | 9.5                | 9.42              | 1.63             | 1.70            | 0.69             | 67                                          | 117                                         |
| Gc + Gf            | 65.5                | 8.0                | 8.00              | 1.27             | 1.62            | 0.55             | 70                                          | 115                                         |
| Gm + Gf            | 63.0                | 6.2                | 6.75              | 0.90             | 1.45            | 0.70             | 61                                          | 102                                         |
| Gc + Gm + Gf       | 74.3                | 10.8               | 12.00             | 2.00             | 1.95            | 0.90             | 90                                          | 95                                          |

These enhancements in the plant growth parameters, mycorrhizal colonization, and biomass production may be reflected in higher yield of tomato due to the multiple inoculation of VAM and this deserves due attention and further emphasis to confirm the suitability of multiple inoculum as compared to single inoculum for better crop production. Mycorrhizal symbiosis is an attractive process in agriculture and forest management to enhance crop and wood production in the sense of sustainable agriculture and restoring soil fertility (Verma, 1995).

Many attempts have been made, over the years, to determine the influence of mixed VAM fungal species on root colonization and sporulation. There are numerous reports of increased plant growth from inoculation with non-indigenous VAM species even into non-sterile soil containing indigenous species. The competitive ability of *Glomus tenue* was demonstrated by Powell and Daniel (1978), who observed an increase in plant growth when *Glomus tenue* was added to host pot cultures already colonized by other VAM fungi. Further evidence of competition between VAM fungal species came from Ross and Ruttencutter (1977) who found that less colonization occurred in *Glomus macrocarpum* inoculated peanuts and soyabeans as compared to hosts inoculated with *Gigaspora gigantea* and *Glomus macrocarpum* together.

**CONCLUSION**

The enhancements in the plant growth parameters, mycorrhizal colonization, and biomass production may be reflected in higher yield of tomato due to the multiple inoculation of VAM and this deserves due attention and further emphasis to confirm the suitability of multiple inoculum as compared to single inoculum for better crop production.

**CONFLICT OF INTEREST**

Authors have no conflict of interest.

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