Gamma ray-induced mutagenesis and its impact on per cent disease index in Tuberose cv. 'Hyderabad Single''

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Abstract

Tuberose (*Polianthes tuberosa* Linn.) is one of the most important flower crops grown under the tropical and subtropical conditions in India. The crop is affected by leaf spot disease caused by *Alternaria polianthi*, which significantly impacts its growth and flower yield. The disease is prevalent in both single and double varieties, especially in the regions with high rainfall and humidity. A study was conducted with gamma ray irradiated bulbs using 44 variants of mutant population with one untreated bulb (Control). Among the variants of mutant population studied in the VM_2 generation, mutant M_{333} exhibited significantly highest per cent disease index (susceptible) compared to untreated control. No incidence of disease was noticed with mutants *viz.*, M_{473C}, M_{178} and M_{21}, hence these were characterized as immune mutants to the leaf spot disease caused by *Alternaria polianthi*.

Keywords: Tuberose, Alternaria, Percent Disease Index (PDI), Immune, Resistant, Susceptible

Introduction

Tuberose (*Polianthes tuberosa Linn.*) is an important ornamental flower crop in tropical and subtropical regions (Biswas *et al.*, 2002) [1] and commercially cultivated in India for its cut flowers, loose flowers and its highly valued essential oil (Mishra *et al.*, 2008) [10]. However, this crop is susceptible to various fungal and viral diseases too, impacting the plant's growth and flower yield. Among these, leaf spot incited by *Alternaria polianthi* is a significant fungal disease (Mariappan *et al.*, 1977) [9]. The disease manifests as brown specks on leaf tips or margins, developing into round to oval spots (10-30 cm long, 4-5 cm diameter) that can coalesce, causing blight symptoms. *Alternaria polianthi* leaf spot in tuberose was first reported in Coimbatore, Tamil Nadu (Mariappan *et al.*, 1977) [9] and later from the same state (Muthukumar *et al.*, 2007) [12]. The disease is prevalent in both single and double-type tuberose cultivars grown under high rainfall and humid conditions. Further, it has been observed to induce certain mutations in the plant's DNA, leading to development of new plant varieties with improved resistance to pests and diseases (Lourdes *et al.*, 2022) [8]. The X-ray and gamma ray irradiations are another form of ionizing radiation that has been studied for its effectiveness in plant disease resistance.

Use of gamma ray irradiation is a physical method for controlling the plant diseases. It does not require administration of any chemicals. Gamma ray irradiation has been extensively researched for its potential to manage the plant diseases caused by fungi, bacteria, viruses and nematodes. Use of gamma ray irradiation has been shown to be effective in controlling various fungal diseases in other crops such as gerbera (Ghani and Sharma, 2019 and Serrano-Fuentes *et al.*, 2022) [3, 15]. Numerous studies have proven that this technique is effective in combating plant diseases. For instance, gamma ray irradiation has been found to be effective in controlling the powdery mildew, gray mold and downy mildew on cucumbers (Faris *et al.*, 2016) [2].

Gamma ray irradiation can be used to control the biotic factors by damaging their DNA and preventing them from reproducing (Hong *et al.*, 2022) [4]. Gamma ray irradiation offers numerous advantages including the fact that it is a non-chemical method, which implies no harmful residue on plants. Therefore, this technique has been considered an ecological method of controlling the plant diseases. Thus, as of the available information gamma ray irradiation has its potential to reduce the disease incidence, hence an attempt was made to investigate the effect of gamma ray irradiation on tuberose bulbs for screening of disease resistance.

Material and Methods

The present investigation was conducted in a Completely Randomized Design with two replications by using 44 variants of mutant population with untreated bulbs of cv. 'Hyderabad Single' as control, totaling to 45 treatments. Second generation gamma irradiated bulbs of tuberose were planted in poly bags possessing dimensions of 14 x 16 inch with 10 kg soil capacity. Poly bags were filled with potting medium consisting of red earth, FYM and cocopeat in the ratio of 3:1:1. Recommended dose of NPK fertilizers were applied as nitrogen in the form of urea @ 500 g, phosphorous in the form of single super phosphate @ 1570 g and potash in the form of muriate of potash @ 410 g for all the poly bags as per the recommendations of 'Udyana Pantala Saagu – Samagra

Yajamanyam' published by Dr. Y.S.R. Horticultural University, Venkatramannagudem in four equal doses starting at the time of planting of bulbs and later at every three months interval up to 90 days of the crop stage (N:P:K @ 200:80:200 kg/ha).

Leaf samples were neatly cleaned using tissue paper and were collected in the morning hours between 8:00 - 9:00 AM. Cleaned leaves were carried to laboratory and with the use of a scalpel the infected portion was scrapped and the collected powder was placed on the slide and observed under compound microscope for the presence of disease-causing microorganisms.

The observations on disease intensity were recorded using 0-5 scale (Sharma, 1986) [17] during the month of November when the disease expression was found maximum and the per cent disease index (PDI) was calculated by using the following formula which was developed by Wheeler (1969) [18].

$$\text{PDI} = \frac{\varepsilon \ of \ infected \ leaves \ on \ the \ plant}{Number \ of \ leaves \ observed \times Maximum \ Disease \ Score} \times 100$$

Scale to measure degree of resistance against leaf spot disease of tuberose

Disease Severity	Category	Reaction
<5	0	Immune
5-10	I	Resistant
11-20	II	Moderately Resistant
21-40	III	Moderately Susceptible
41-60	IV	Susceptible
>61	V	Highly Susceptible

Source: Sharma (1986) [17] and Pathak *et al.* (1986) [14]

Further, the mutants were placed on different categories of resistance and susceptibility on the basis of method explained by Pathak *et al.* (1986) [14].

Results and Discussion

Per cent disease index was analyzed for their means and the data were furnished in Table 1 and illustrated in Plate 1. The data were found non-significant with regard to per cent disease index as some of the variants in the mutant population have shown immune to *Alternaria* leaf spot. However, among the variants of mutant population, variant M_{21}, M_{178} and M_{473C} were found immune to *Alternaria* leaf spot disease, whereas variant M_{32} was found resistant to leaf spot disease. Most of the variants were found moderately resistant to leaf spot disease. None of the variants were found either susceptible or highly susceptible to leaf spot disease.

Among the variants of mutant population studied in the VM_2 generation, mutant M_{333} exhibited significantly highest per cent disease index (susceptible) compared to untreated control. No incidence of disease was noticed with mutants *viz.,* M_{473C}, M_{178} and M_{21}, hence these variants were characterized as immune mutants to leaf spot disease caused by *Alternaria polianthi*.

Kumar *et al.* (2012) [7] observed similar kind of results with regard to disease resistance when chrysanthemum was treated with gamma ray irradiation. Similarly, Mohanraj *et al.* (1996) [11] screened 700 somaclonal variants against the red rot disease in sugarcane and out of these 120 somaclonal variants exhibited moderately resistant reaction in the field trials. Javed *et al.,* (2003) [6] obtained 35 and 205 variants after *in-vitro* screening in two different varieties of sugarcane and out of these only 10 and 57 were found to be resistant in both the varieties respectively after field trials. *In-vitro* selection and final screening of red rot resistant lines of sugarcane after field trials has also been reported by Naik and Vedamurthy (1997) [13]; Javed *et al.,* (2001) [5] and Shahid *et al.,* (2003) [16].

Table 1: Effect of γ-ray irradiation on Percent Disease Index of VM_2 generation mutants of tuberose cv. 'Hyderabad Single'

Treatments	PDI (%)	Category	Reaction
M_{21}	0 (0)	0	Immune
M_{32}	2.37 (8.76)	I	Resistant
M_{33}	7.29 (15.62)	II	Moderately Resistant
M_{34}	6.07 (14.21)	II	Moderately Resistant
M_{37}	6.06 (14.14)	II	Moderately Resistant
M_{44}	5.67 (13.77)	II	Moderately Resistant
M_{50}	11.15 (18.93)	II	Moderately resistant
M_{54}	4.78 (12.55)	II	Moderately resistant
M_{63}	5.16 (13.12)	II	Moderately resistant
M_{81}	7.86 (16.26)	II	Moderately resistant
M_{116}	20.78 (25.48)	III	Moderately susceptible
M_{119}	10.98 (19.02)	II	Moderately resistant
M_{142C}	9.38 (17.69)	II	Moderately resistant
M_{146}	6.88 (15.15)	II	Moderately resistant
M_{162}	13.25 (19.93)	III	Moderately susceptible
M_{164}	9.75 (17.19)	II	Moderately resistant
M_{178}	0 (0)	0	Immune
M_{183}	8.43 (16.85)	II	Moderately resistant
M_{189C}	8.65 (17.1)	II	Moderately resistant
M_{202}	13.93 (21.78)	III	Moderately susceptible
M_{221C}	20.67 (25.85)	III	Moderately susceptible
M_{226C}	6.9 (15.12)	II	Moderately resistant
M_{264C}	4.05 (11.12)	II	Moderately resistant
M_{292C}	7.16 (15.11)	II	Moderately resistant
M_{301C}	14.67 (21.38)	III	Moderately susceptible
M_{333}	40.13 (36.95)	III	Moderately susceptible
M_{339C}	23.95 (27.9)	III	Moderately susceptible
M_{342}	11.56 (19.5)	II	Moderately resistant
M_{351}	4.44 (8.66)	II	Moderately resistant
M_{354C}	18.94 (24.69)	III	Moderately susceptible
M_{369}	8.42 (16.33)	II	Moderately resistant
M_{389C}	10.49 (17.43)	II	Moderately resistant
M_{408C}	8.16 (16.29)	II	Moderately resistant
M_{409C}	7.74 (16.14)	II	Moderately resistant
M_{410}	8.07 (16.46)	II	Moderately resistant
M_{440C}	14.58 (19.0)	III	Moderately susceptible
M_{456C}	27.56 (25.97)	III	Moderately susceptible
M_{462}	9.58 (15.34)	II	Moderately resistant
M_{463}	16.54 (18.94)	III	Moderately susceptible
M_{468C}	9.07 (16.49)	II	Moderately resistant
M_{473C}	0 (0)	0	Immune
M_{496C}	14.58 (21.97)	III	Moderately susceptible
M_{498}	14.33 (21.84)	III	Moderately susceptible
M_{465}	8.33 (15.97)	II	Moderately resistant
Untreated	10.06 (17.5)	II	Moderately resistant
CD @ 5%	NS	--	--
SEm±	7.475	--	--

Parenthesis represents the arc sign transformed values

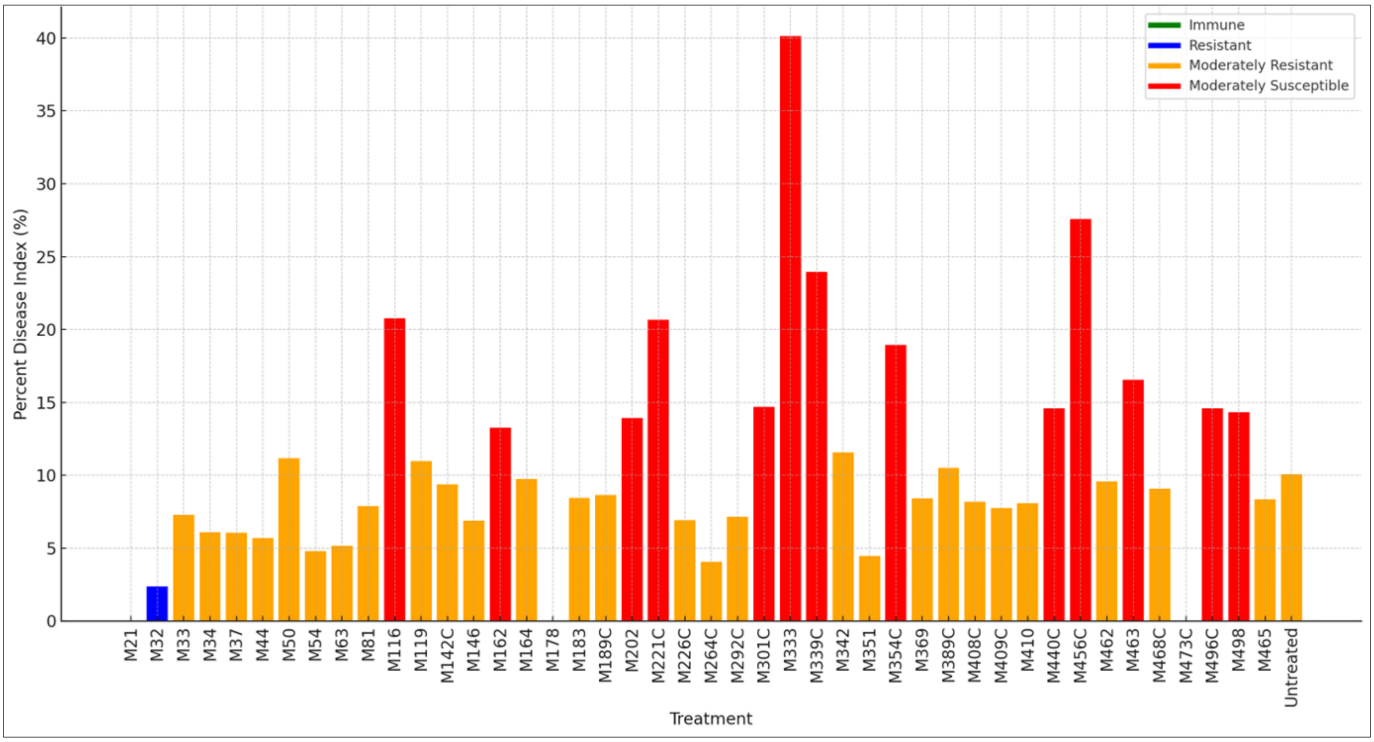
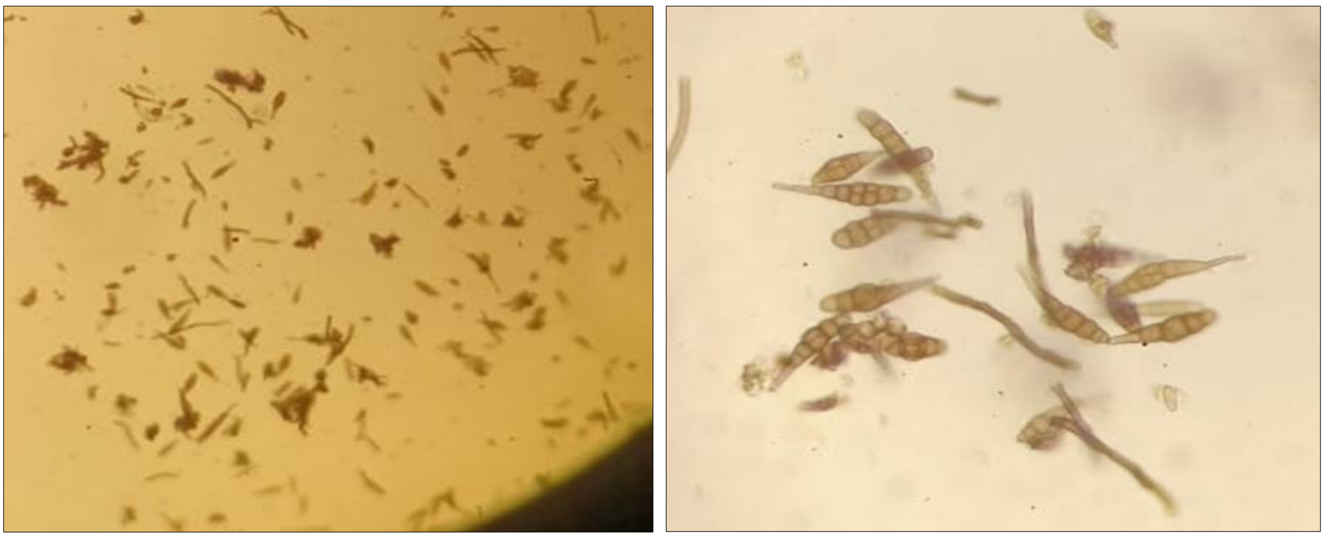


Fig 1: Disease Resistance Categorization based on PDI in VM2 Tuberose Mutants (Hyderabad Single)

Alternaria leaf spot observed on tuberose leaves of cv. 'Hyderabad Single'



Isolated Conidia of Alternaria

Fig 2: Observation of *Alternaria* leaf spot and isolated conidia on the γ-ray irradiated mutants of tuberose cv. 'Hyderabad Single'

Conclusion

Among the variants of mutant population evaluated in the VM_2 generation, mutant M_{333} recorded significantly highest per cent disease index (susceptible) compared to untreated control. No incidence of disease was noticed with the mutants *viz.,* M_{473C}, M_{178} and M_{21}. These results indicated that when tuberose bulbs of cv. 'Hyderabad Single' were exposed to gamma ray irradiation, they exerted certain amount of disease resistance to *Alternaria* leaf spot in some of the variants in the mutant population. Knowing and isolating these mutants helps in creating new genotypes which can create a revolution in the tuberose breeding.

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