Occurrence of Seed borne Fungal pathogens in popular cultivars of Green gram (*Phaseolus aureus* Roxb.)

D. H. Tandel, A. N. Sabalpara, R.C.Patel, V.R. Patel

***Abstract*— Seven genera of fungi which included Macrophomina sp. (4 to 20%), Curvularia sp. (4-12%), Aspergillus sp.(12-28%), Alternaria sp. (4-20%), Rhizopus sp. (0-8%), Colletotrichum sp. (4-8%) and Fusarium sp. (4-12%) were detected in moist blotter and PDA methods under unsterilized and sterilized seed lots variously in all varieties tested. Rhizopus sp., Colletotrichum sp. and Fusarium sp. were recorded in few varieties. Macrophomina sp. was present as externally as well as internally seed borne.**

***Index Terms*— Mycoflora, Association, Seed, Greengram, PDA, Moist blotter.**

I. INTRODUCTION

Green gram is one of the important pulse crops grown extensively in all parts of Gujarat during all the seasons especially more during kharif and summer. Disease is one of the major constraint in economic crop production as they inflict heavy losses. Like other crops, greengram is also attacked by many diseases during seed germination to seed production and maturity. Over 35 fungal pathogens, few viral, bacterial pathogens and nematode species are known to attack greengram resulting into substantial yield losses (Agrawal, 1989). The crop is affected by many important fungal disease viz., anthracnose (Colletotrichum lindemuthianum), Cercospora leaf spot , Alternaria leaf spot (Alternaria alternata),dry root rot/charcoal rot/ ashy stem blight/stem canker, leaf blight, pod blight (Macrophomina phaseolina), Fusarium root rot, Fusarium wilt (Fusarium oxysporum), powdery mildew (Erysiphe polygoni); rust (Uromyces phaseoli); bacterial diseases viz., bacterial blight (Xanthomonas); virus disease viz., yellow mosaic virus disease and nematode disease viz., root-knot nematode (Meloidogyne incognita), M. javanica, M. arenaria (Agrawal, 1989). Patel (2003) recorded the presence of different fungi viz., Aspergillus sp., Alternaria sp., Rhizoctonia sp., Macrophomina sp., Curvularia sp., Fusarium sp., and Rhizopus sp., by moist blotter method and PDA method with and without surface sterilization in greengram seeds. By considering the seriousness of the problem the present investigation was carried out to know the association and occurrence of Seed borne fungal pathogens in popular cultivars of Green gram (Phaseolus aureus Roxb.)

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II. MATERIAL AND METHODS

Mycoflora associated with the seeds of green gram were detected by using moist blotter method and PDA method. The seeds of seven green gram varieties were collected from Pulse Research Station, N.A.U., Navsari,Gujarat. Three hundred seeds were taken at random from each variety. From this, one hundred seventy five seeds of seven cultivars were plated in plastic petriplates (100 mm diameter) having three layers of moistened blotters made by dipping in sterile water and holding the blotters to drain out excess water. Other one hundred seventy five seeds of seven cultivars were plated in PDA petriplates. In each plate, 25 seeds were placed equidistantly without any pre-treatment. Each treatment replicated three times.

Simultaneously, for determining the internal seed borne nature of fungi (i.e. deep seated in tissue), 175 seeds of 7 cultivars were plated on PDA and 175 seeds of 7 cultivars were plated on moist blotter in similar way after giving pre-treatment i.e. surface sterilization with mercuric chloride 0.1 per cent for two minutes followed by three subsequent washings with sterile distilled water. The plates were incubated at room temperature. After eight days of incubation, the fungi developed on seeds were recorded and identified by colony character as well as by microscopic examination. The fungi detected were brought into pure culture by picking up a bit of mycelial growth aseptically and transferring it on a PDA slant.

III. RESULTS AND DISCUSSION

The green gram seeds of different seven varieties were tested for the presence of mycoflora by moist blotter method and PDA method with and without surface sterilization (pre-treatment) and after eight days of incubation, the fungal counts were recorded. The result presented in tables indicated that mycoflora of the seed of different varieties were much higher.

Moist blotter method

A study on nature of mycoflora of greengram seeds, using the moist blotter method showed that in all the seven varieties, fungi like Aspergillus sp. (16-28%), Macrophomina sp. (12-20%) and Alternaria sp. (8-20%) were most frequently associated whereas Fusarium sp. (4-12%), Curvularia sp. (4-12%),Colletotrichum sp. (4-8%), Rhizopus sp. (0-8%) and non sporulating unidentified fungi with aseptate (0-8%) and septate (0-12%) mycelia were less frequently detected in unsterilized seed lots. The seeds of BARC TM 98-50 were

totally free from the infection of Rhizopus sp. while BARC TM 97-25 and BARC TM 96-2 were free from the infection of fungi with aseptate mycelium.

In sterilized seed lots, fungi like Aspergillus sp. (12-20%), Alternaria sp. (4-12%), Macrophomina sp. (4-8%), Fusarium sp. (4-8%) (except GM-3 variety), Curvularia sp. (4-8%), Colletotrichum sp. (4-8%) (except BARC TM 97-52 and BARC TM 97-55 varieties), unidentified fungi with septate mycelium (4-8%) (except BARC TM 97-52 and BARC TM 98-50 varieties) and with aseptate mycelium (0-4%) (except GM-3, BARC TM 97-52, BARC TM 97-25 and BARC TM 96-2 varieties) were detected in the varieties tested while Rhizopus sp. (0-4%) was recorded only in BARC TM 97-52 and CO-4 varieties.

PDA method

In PDA method, fungi like, Aspergillus sp. (16-24%), Macrophomina sp. (8-20%) and Alternaria sp. (8-16%) were most frequently associated whereas Fusarium sp. (8-12%), Curvularia sp. (4-12%), Colleteotrichum sp. (4-8%), Rhizopus sp. (0-4%) (except GM-3 variety) and non-sporulating fungi with septate mycelia (4-12%) and aseptate mycelia (0-4%) except BARC TM 97-52 and GM-3 varieties were less frequently detected in all the seven greengram varieties in unsterilized seed lots. In sterilized seed lots, fungi like, Aspergillus sp. (8-16%), Alternaria sp. (4-16%), Macrophomina sp. (8-12%), Fusarium sp. (4-8%), Curvularia sp. (4-8%), Colletotrichum sp. (4-8%) and unidentified fungi with septate mycelium (4-8%) except BARC TM 97-55 variety were detected while Rhizopus sp. found in BARC TM 97-52 (4%), BARC TM 97-55 (4%) and BARC TM 97-25 (4%0 and unidentified fungi with aseptate mycelium found in BARC TM 97-25 (4%) and BARC TM 98-50 (4%) only.

Among the internal seed borne fungi, Aspergillus sp., Alternaria sp., Macrophomina sp. and Curvularia sp. were more common.This results suggests that it can be effectively and economically controlled by producing disease free seeds and with proper seed treatment. It is also suggested that during seed production and certification the presence of the pathogen should also be considered as one of the criteria.

The results are supported by the findings of various workers on the seeds of greengram. Alternaria spp., Colletotrichum spp. and Fusarium spp. (Agrawal et al., 1971) , Aspergillus spp. and Curvularia spp. (Raut and Ahire, 1988) and Rhizopus spp. (Sahu and Jena, 1997) reported earlier by Patel (2003) recorded the presence of different fungi viz., Aspergillus sp., Alternaria sp., Rhizoctonia sp., Macrophomina sp., Curvularia sp., Fusarium sp., and Rhizopus sp., by moist blotter method and PDA method with and without surface sterilization in greengram seeds.

IV. CONCLUSION

In all varieties of green gram four genera of fungi, which included Curvularia sp., Macrophomina sp., Aspergillus sp. and Alternaria sp. were detected in both the methods under sterilized and unsterilized seed lots tested. Rhizopus sp. was not detected in GM-3 variety under sterilized as well as unsterilized seed lots, while under sterilized seed lots, it was not detected in C0-4, BARC TM-9850 and BARC TM-2 varieties in PDA method. Rhizopus sp. was not detected in BARC TM-9850 variety under unsterilized seed lot while it was detected in BARC TM-9752 and CO-4 varieties under sterilized seed lot in moist blotter method. Colletotrichum sp. was detected in all varieties under unsterilized seed lot in both the methods and it was not detected in BARC TM 9752 and BARC TM-9755 varieties in moist blotter method under sterilized seed lot. Fusarium sp. was detected in all the varieties except GM-3 under sterilized seed lot in moist blotter method.

Table: Percentage association of fungi with unsterilized and surface sterilized seed lot of green gram (Moist blotter method)

Sr. No.	Name of fungi/Variety	BARC TM-97-52		GM-3		BARC TM-97-55		CO-4		BARC TM-97-25		BARC TM-9850		BARC TM-96-2	
		US	S	US	S	US	S	US	S	US	S	US	S	US	S
1	*Curvularia* sp.	8	4	4	4	8	4	8	4	8	4	12	8	8	8
2	*Macrophomina* sp.	12	4	20	8	12	4	12	8	12	8	12	8	16	8
3	*Aspergillus* sp.	24	16	28	16	20	12	24	12	16	12	20	20	20	16
4	*Alternaria* sp.	8	4	12	8	16	8	8	4	12	8	16	12	20	12
5	*Rhizopus* sp.	8	4	4	0	8	0	4	4	4	0	0	0	4	0
6	*Colletotrichum* sp.	4	0	4	4	8	0	8	4	8	8	8	4	4	4
7	*Fusarium* sp.	8	4	4	0	12	8	8	4	8	4	4	4	8	4
8	Unidentified fungi with (a) Aseptate Mycelium	8	0	4	0	4	4	4	4	0	0	4	4	0	0
	(b) Septate Mycelium	4	0	8	4	8	4	12	8	8	4	8	0	4	4
9	No mycoflora	16	64	12	56	4	56	12	48	24	52	16	40	16	44

US=Unsterilized

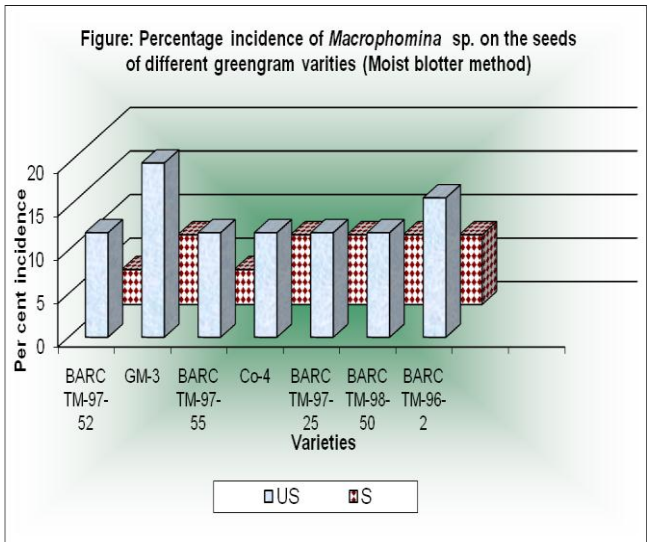
S=Sterilized

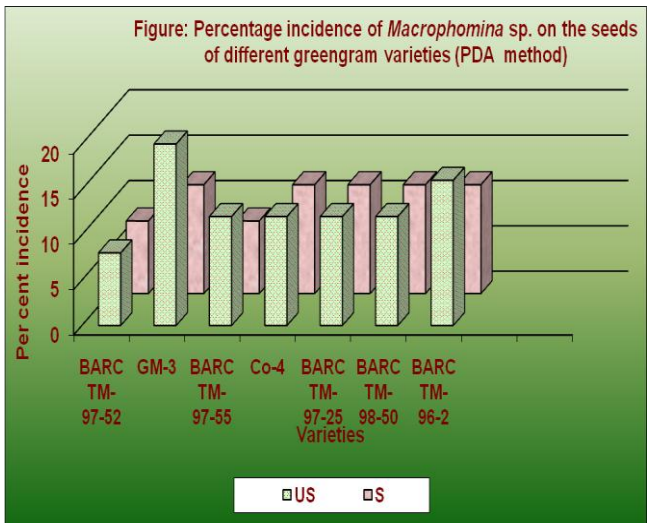
Table: Percentage association of fungi with unsterilized and surface sterilized seed lot of green gram (PDA method)

Sr. No.	Name of fungi/Variety	BARC TM-97-52		GM-3		BARC TM-97-55		CO-4		BARC TM-97-25		BARC TM-9850		BARC TM-96-2	
		US	S	US	S	US	S	US	S	US	S	US	S	US	S
1	*Curvularia* sp.	4	4	8	8	8	4	8	8	8	8	12	4	8	4
2	*Macrophomina* sp.	8	8	20	12	12	8	12	12	12	12	12	12	16	12
3	*Aspergillus* sp.	16	12	24	12	20	8	20	12	16	16	16	12	16	12
4	*Alternaria* sp.	16	12	12	12	16	12	8	4	16	12	16	16	16	12
5	*Rhizopus* sp.	4	4	0	0	4	4	4	0	4	4	4	0	4	0
6	*Colletotrichum* sp.	4	4	4	4	8	4	8	8	8	4	4	4	8	4
7	*Fusarium* sp.	8	8	8	4	12	4	12	8	8	8	8	4	12	4
8	Unidentified fungi with (a) Aseptate Mycelium	0	0	0	0	4	0	4	0	4	4	4	4	4	0
	(b) Septate Mycelium	4	4	8	8	8	0	12	4	8	4	8	4	8	4
9	No mycoflora	36	44	16	40	8	56	12	44	16	28	16	40	8	48

US=Unsterilized

S=Sterilized





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