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UTILIZATION OF ANTAGONISTS FOR CONTROLLING SEED-BORNE PATHOGENS OF BOTTLEGOURD

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ABSTRACT :

The present investigation has been undertaken in the Marathwada region of Maharashtra during the period from December 2014 to December 2016 to determine the prevalence of fungi in the selected vegetable seeds. Eleven fungal genera of Bottlegourd (Lageneria siceraria) viz.Aspergillus spp., Alternaria spp., Fusarium spp., Curvularia spp., Macrophomina spp., Dreshrella spp., Mucor spp., Rhizopus spp., Rhizoctonia spp., Penicillium spp. and Chaetonium spp. were found to be associated on standard blotter paper method and Agar plate method. Whereas, Aspergillus spp., Fusarium spp. and Alternaria spp. were found to be most dominant. Antagonists like Trichoderma harzianum, Trichoderma viride, Bacillus subtilis and Pseudomonas fluorescens used for seed treatment. All the antagonists were found to be successfully inhibits the growth of seedborne fungi and promotes seedling growth and vigor index.

Keywords: *Leaf extract, seed-borne fungi, vegetable seed*

INTRODUCTION

Vegetables comprises a major portion of human diet and plays significant role in human nutrition, especially as a source as phyto-nutriceuticals: vitamins (A, B1, B6, B9, C and E), minerals, dietary fibers and phytochemicals (Dias, 2012). Worldwide production of vegetables tremendously gone up during last two decades and the value of global trade in vegetables now exceeds that of cereals. This sector has emerged as an economically rewarding option for diversification in agriculture and is playing an increasingly important role in the country's nutritional security, poverty alleviation and employment generation. India is the second largest producer of vegetables in the world, next to China. Major vegetables grown in India includes Bitter gourd, Bottle gourd, Brinjal, Tomato, Potato, Cabbage, Capsicum, Carrot, Cauliflower, Onion, Ladies finger, Parwal, Pumpkin, Peas, etc. (Horticultural statistics at a glance, 2016). Cultivated *Lageneria siceraria* is commonly known as white flowered Bottle gourd. Bottle gourd has always been regarded as one of the healthiest veggies. It is a vegetable high on water and is a rich source of vitamin C, K and Calcium. It helps in maintaining healthy heart and brings down bad cholesterol levels. Bottle gourd is susceptible to diseases like fusarium wilt, seedling rot, Anthoceros, etc. As, seed is the most vital input in crop production, it should be of high quality and pathogen free. Crop suffers from various types of diseases caused by fungi which are mostly seed borne in nature. Seed provides natural substrate for the growth of associated fungi. Fungi form the largest group among the pathogenic microorganisms causing seed damage, seed-rot diseases at later stages of crop growth till maturity. Application of fungicides is almost always found to be effective but their non-target environmental impact and development of pathogen resistance is more harmful, which let us to think about alternative methods for their control. Biological control is thus being considered as an alternative or supplement to reduce the use of synthetic chemicals in agriculture (Compant et al., 2005) Biological seed treatment is usually very specialised and uses very specific micro-organisms (antagonists)

that attacks or interfere with targeted pathogens. Biological control is one of the viable, eco-friendly propositions, which can substantially minimise the diseases (Ashwini and Giri, 2014).

Considering these facts, during present investigation, the seed samples of bottle gourd investigated for seed health, isolation of seed moulds as per ISTA, 1996 by Standard Blotter paper and Agar plate method. Eleven fungal genera of Bottlegourd (*Lageneria siceraria*) *viz.Aspergillus* spp*., Alternaria* spp*., Fusarium* spp*., Curvularia* spp., *Macrophomina* spp., *Dreshrella* spp., *Mucor* spp*., Rhizopus* spp*., Rhizoctonia* spp*., Penicillium* spp*.* and *Chaetonium* spp*.* were found to be associated on standard blotter paper method and Agar plate method. Whereas, *Aspergillus* spp*., Fusarium* spp*.* and *Alternaria* spp*.* were found to be most dominant. In second part of investigation, integrated seed health management of seed borne fungi responsible for seed and seedling rot, blight, wilt and dumping off diseases were carried out by seed treatment method with selected antagonists. All antagonists were found to be most effective against seed borne fungi and also enhanced seed germination, seedling health and vigour index were recorded in Bottle gourd.

II. RESEARCH METHODOLOGY

2.1Collection of seed samples (Cultivars)

The method described by Neergaard (1973) has been adopted for the collection of seed samples. Accordingly, two random samples of following seeds (250 gm each) were collected from local farmers and market places of *Marathwada* region of Maharashtra state of India.

Bottle gourd (*Lagenaria siceraria* Mol.)

2.1.1 Nisco

2.1.2 Bottle gourd local

During the course of studies, seed samples were separately collected and stored in plastic containers without any treatment of fungicide/insecticide at laboratory conditions.

2.2Detection of Seed Mycoflora

The seed mycoflora was isolated by using different methods such as Standard blotter paper method and Agar plate method as recommended by International Seed Testing Association ISTA (1966), De Tempe (1953), Neergaard (1973) and Agrawal (1976).

Observations were recorded in percent incidence of seed borne fungi associated with unsterilized seeds. Fungi appeared on seed were isolated in pure culture for identification and for further study. Two different methods of isolation techniques were used for assessment of seed mycoflora.

2.2.1Standard blotter paper method

A pair of sterile blotter papers of 8.5 cm diameter were soaked in sterile distilled water and were placed in pre-sterilized petriplates of 9 cm. diameter. Ten seeds of test sample per petriplate were then placed equidistance on moist blotter. The plates were incubated at $28^0 \pm 2^0$C under diurnal conditions.

2.2.2 Agar plate method

Pre-sterilized petriplates were poured with 15 mL of autoclaved Potato Dextrose Agar (PDA). On cooling the medium, ten seeds per plate of the sample to be studied were equidistantly placed aseptically. Incubation and other details of the study were same as described for blotter test method. The various moulds appeared on seeds in blotter paper and Agar plates were isolated and maintained on PDA/ GNA slants.

2.2.3 identification of fungi

Detailed examination of fungal characters was done by compound microscope after seventh day of incubation periodand identification was confirmed with the help of identification Manual (Mathur and Kongsdal, 2003) and pectoral atlas of soil and seed fungi. (Watanabe, 2002).

The percentage incidence of mycoflora observed in seed samples were calculated by using following formulas.

$$\text{Percentage incidence of fungus} = \frac{\text{No. of seeds containing particular fungus}}{\text{Total no. of seeds}} X \ 100$$

$$\text{Percentage incidence of seed mycoflora} = \frac{\text{No. of seed infected by fungi}}{\text{Total no. of seeds}} X \ 100$$

2.3Bio-control of seed borne pathogens by using antagonists:

Vegetable seeds were treated with fungal and bacterial antagonists. The seeds were soaked in fungal spores and bacterial suspension for 15 minutes. The treated seeds were incubated for 7-8 days. The treated seeds were grown and incidence of fungi, rate of germination, vigour index and seedling health were studied by using blotter paper method as described by International Seed Testing Association (ISTA, 1976)

Seeds were treated with antagonistic fungi and bacteria. The antagonistic fungi like ***Trichoderma* species** were used. The antagonistic bacteria used were ***Bacillussubtilis*** and ***Pseudomonas fluorescens.***

2.3.1 Seed treatment with *Trichoderma* species:

200 g of seeds were coated with 100mL aqueous spore suspension of ***Trichoderma species*** (8×10^9 spores/mL) by adding 1 mL of 0.5% carboxyl methyl cellulose (CMC) as sticker and 20 g of Bentonite powder as filler for seed dressing. Treated seeds were incubated for 7-8 days. The percent incidence of fungi, seed germination and vigour index were observed in seed samples.

2.3.2 Seed treatment with *Bacterial* species:

The method of Weller and Cook (1983) was followed for seed bacterization. ***Pseudomonas fluorescens*** and ***Bacillussubtilis*** were separately grown in succinate broth for 24 hours at 28 ± 1^0C under shaking condition and finally centrifuged at 7000 rpm for 15 minutes at 4^0C. The supernatant was discarded and pellets were washed with SDW and resuspended to obtain a population density of 10^7 cfu/mL. This suspension was mixed with 1.0% carboxyl methyl cellulose (CMC). Seeds were allowed to air dry overnight under aseptic condition after coating with CMC slurry of bacterial culture. Care was taken to avoid clumping of seeds. Seeds coated with slurry of CMC (without bacteria) served as control. The seeds were incubated on sterile blotter paper. The percent mycoflora, seed germination and vigour index were observed in seed samples.

For observations and results, following formulas were used:

1) $\text{Percentage incidence of fungus} = \frac{\text{No. of seeds containing particular fungus}}{\text{Total Seeds}} X \ 100$

2) $\text{Percentage incidence of seed mycoflora} = \frac{\text{No. of seed infected by fungi}}{\text{Total no. of Seeds}} X \ 100$

3) $\text{Percentage germination} = \frac{\text{No. of seeds germinated}}{\text{Total no. of Seeds}} X \ 100$

4) Vigour Index = Percentage germination X Length of seedling

Data Analysis

Data was analysed by one-way analysis variance (ANNOVA) and LSD was calculated at P=0.05 for significance. The analysis was performed with Microsoft excel software.

III. EXPERIMENTAL RESULTS AND DISCUSSION

3.1 Fungi associated with Local seeds of Bottle gourd

The results shown in Table 1, Plate I and II indicates that Bottle gourd seed collected from farmers were associated with twelve fungi such as Aspergillus flavus, Aspergillus niger, Aspergillus fumigatus, Alternaria alternata, Fusarium oxysporum, Fusarium moniliforme, Curvularia clavata, Rhizoctonia spp., Rhizopus stolonifer, Chaetonium funicola, Penicillium notatum and Dreshrella spp. Ten mycoflora on blotter paper method and Twelve on Agar plate method were isolated.

On blotter paper method Fusarium oxysporum (33.3%) showed maximum incidence followed by Aspergillus flavus (30.0%) and Alternaria alternata (23.3%). Along with these, Aspergillus niger (20.0%), Aspergillus fumigatus (13.3%), Curvularia clavata (10.0%), Rhizopus stolonifer (10.0%) were recorded lesser amount while Rhizoctonia spp. (6.7%) and Chaetonium funicola (6.7%) were recorded in least.

On agar plate method Fusarium oxysporum displayed maximum percent of incidence (36.7%) followed by Aspergillus flavus (33.3%), Alternaria alternata (30%) and Aspergillus niger (30.0%). Incidence of Aspergillus fumigatus (13.3%), Fusarium moniliforme (16.7%), Curvularia clavata (13.3%), Rhizoctonia spp. (13.3%), Rhizopus stolonifer (13.3%), Chaetonium funicola (10.0%), Penicillium notatum (10.0%) and Drechslera spp. (6.7%) were recorded in varying amount.

Fusarium oxysporum showed highest percent incidence under blotter paper and agar plate methods (33.30% and 36.70% respectively) followed by Aspergillus flavus (30.0 and

33.3 % respectively) and Alternaria alternata (23.3 and 30.0 % respectively). On an average, the percent fungal incidence was 13.88 under blotter paper method, while 18.88% under agar plate method. The difference among them was statistically non-significant (t=1.20). In all other fungi the percent incidence was significantly less.

Table 1: Fungi associated with seeds of Bottle gourd (Local Seeds)

Sr. No.	Name of the Fungi	Percentage of incidence	
		Standard Blotter	Nutrient Agar
1	*Aspergillus flavus*	30	33.3
2	*Aspergillus niger*	20	30
3	*Aspergillus fumigatus*	13.3	13.3
4	*Alternaria alternata*	23.3	30
5	*Fusarium oxysporum*	33.3	36.7
6	*Fusarium moniliforme*	13.3	16.7
7	*Curvularia clavata*	10	13.3
8	*Rhizoctonia* spp.	6.7	13.3
9	*Rhizopus stolonifer*	10	13.3
10	*Chaetonium funicola*	6.7	10
11	*Penicillium notatum*	0	10
12	*Drechslera* spp.	0	6.7
Statistical Analysis			

	Mean	13.8833	18.8833
	SD	10.7972	10.4801
	CV	77.7710	55.4992
	SE	3.1169	3.0253
	CD 5%	6.8571	6.6558
	CD 1%	9.6935	9.4088
	t	1.1981	

3.2 Fungi associated with Nisco seeds of Bottle gourd

Incidence of seed borne mycoflora associated with bottle Gourd seeds has been given in Table no. 2, plate III and IV. Total twelve fungi were isolated from seeds viz. *Aspergillus flavus, Aspergillus niger, Fusarium oxysporum, Fusarium moniliforme, Alternaria alternata, Alternaria tenuis, Rhizoctonia solani, Rhizopus stolonifer, Mucor mucedo, Penicillium* spp.*,*

Dreshrella spp. and *Macrophomina* spp. whereas, last three fungal species were not recorded in Blotter paper plate.

In case of blotter paper method, seeds showed maximum incidence of Fusarium oxysporum with 36.7% followed by *Aspergillus flavus* (33.3%) and *Alternaria alternata* (33.3%); while, Other fungi viz. *Aspergillus niger* (16.7%), *Alternaria tenuis* (16.7%), *Rhizoctonia solani* (20.0%), *Fusarium moniliforme* (13.3%), *Rhizopus stolonifer* and *Mucor mucedo* (10.0% each) showed lesser incidence.

PLATE I – Fungi associated with Bottle gourd seeds (Local) on blotter paper

PLATE II – Fungi associated with Bottle gourd seeds (Local) on Agar plate

PLATE I – Fungi associated with Bottle gourd seeds (Local) on blotter paper

PLATE II – Fungi associated with Bottle gourd seeds (Local) on Agar plate

PLATE III – Fungi associated with Bottle gourd seeds (Nisco) on blotter paper

PLATE IV – Fungi associated with Bottle gourd seeds (Nisco) on Agar plate

In glucose agar plate, maximum percent incidence was recorded by *Aspergillus flavus* (43.3%) followed by *Fusarium oxysporum* and *Alternaria alternata* (36.7% each). Along with these, *Rhizoctonia solani* (23.3%), *Aspergillus niger* (20.0%), *Fusarium moniliforme* (16.7%), *Rhizopus stolonifer* (16.7%), *Alternaria tenuis* (13.3%), *Mucor mucedo* (10.0%), *Penicillium* spp. (10.0%), *Drechslera* spp. (6.7%) and *Macrophomina* spp. (6.7%) showed varying degree of incidences.

On an average the percentage fungal incidence was 15.83% under the influence of standard blotter method, which was increased to 20.28% in agar plate method. However, the increase in percentage fungal incidence was statistically non-significant (t=0.89). *Fusarium oxysporum* showed maximum incidence under standard blotter (36.7%), while it was maximum (43.3%) with *Aspergillus flavus* under agar plate method. Wide variation in fungal incidence (C.V.= 82.81) was observed under blotter method, as compared to that observed on agar plate method (C.V.=60.89).

Table 2.: Fungi associated with seeds of Bottle gourd (Nisco Seeds)

Sr. No.	Name of the Fungi	Percentage of incidence	
		Standard Blotter	Nutrient Agar
1	*Aspergillus flavus*	33.3	43.3
2	*Aspergillus niger*	16.7	20
3	*Fusarium oxysporum*	36.7	36.7
4	*Fusarium moniliforme*	13.3	16.7
5	*Alternaria alternata*	33.3	36.7
6	*Alternaria tenuis*	16.7	13.3
7	*Rhizoctonia solani*	20	23.3
8	*Rhizopus stolonifer*	10	16.6
9	*Mucor mucedo*	10	13.3
10	*Penicillium notatum*	0	10
11	*Drechslera spp.*	6.7	6.7
12	*Macrophomina spp.*	0	6.7
Statistical Analysis			
	Mean	15.8333	20.2750
	SD	13.1114	12.3460
	CV	82.8090	60.8928
	SE	3.7849	3.5640
	CD 5%	8.3269	7.8408
	CD 1%	11.7712	11.0840
	t	0.8892	

3.3 Effect of antagonists on Bottle gourd seeds

3.3.1 Effect of antagonists on seed mycoflora incidence, germination and vigour index:

Treatment of Bottle gourd seed with antagonists caused significant reduction in seed borne fungi with increased germination and vigour index which is depicted in Table 3 and Fig. 1. *Aspergillus flavus, Alternaria alternata* and *Fusarium oxysporum* was the most dominant fungal pathogen observed on seeds. Their prevalence was significantly reduced due to treatment of seeds with antagonists over the control. The minimum incidence of these fungi was recorded with treatment of *Trichoderma harzianum* (10.0%, 3.3% and 6.7%)on

Aspergillus flavus, Alternaria alternata and *Fusarium oxysporum* followed by *Trichoderma viride* (13.3%, 10.0% and 10.0%), *Bacillus subtilis* (13.3%, 10.0% and 10.0%), and *Pseudomonas fluorescens* (16.7%, 10.0% and 10.0%).

Reduced incidence of seed mycoflora favoured more germination rate with increased seedling length and vigour index in treated seeds. The maximum germination (80.0%) was recorded with *Trichoderma harzianum* and minimum germination (40.0%) recorded on seeds without treatment (control seeds). The rate of germination in *Trichoderma* treated seeds were found statistically identical. While treatment of *Bacillus subtilis* and *Pseudomonas fluorescens* recorded 70.0% and 60.0% respectively. The highest seedling length of 7.2 cm. and vigour index 552.24 was recorded in seeds treated with *Trichoderma viride* followed *Trichoderma harzianum* with 6.8 cm. seedling length and 544 vigour index.

Table 3.: Effect of antagonists on seed mycoflora, seed germination and vigour index of Bottle gourd.

Sr. No.	Antagonists	Seed mycoflora (%)			Seed germination (%)	Seedling length (cm.)	Vigour index
		A. flavus	*A. alternata*	*F. oxysporum*			
1	Control	33.3	23.3	30	40	5.2	208
2	*T. harzianum*	10	3.3	6.7	80	6.8	544
3	*T. viride*	13.3	10	10	76.7	7.2	552.24
4	*P. fluorescens*	16.7	10	10	60	6.8	408
5	*B. subtilis*	13.3	10	10	70	6.9	483
	Mean				13.3400	65.3400	6.5800
	SD				8.4275	14.3966	0.7054
	CV				63.1745	22.0334	10.7205
	SE				3.7689	6.4384	0.3155
	CD 5 %				10.4775	17.8986	0.8770
	CD 1 %				17.3369	29.6165	1.4512

Table 3a: Analysis of variance (ANOVA)

Source	df	SS	MSS	F	S/NS
Antagonists	4	884.2093	221.0523	69.0285	S
Mycoflora	2	93.2013	46.6007	14.5521	S
Error	8	25.6187	3.2023	-	-
Total	14	1003.0293	-	-	-
SE	1.4611	-	-	-	-
CD 5%	3.3021	-	-	-	-
CD 1 %	4.7487	-	-	-	-

3.3.2 Effect of antagonists on seed germination, germination failure and seedling health:

Results presented in Table 4 and Fig. 2 revealed that seed treatment with selected bioagents *viz.Trichoderma harzianum, Trichoderma viride, Pseudomonas fluorescence* and *Bacillus subtilis* show enhanced seed germination and seedling health with decreased germination failure over the control in Bottle gourd. The minimum germination (40.0%) was recorded in control while maximum (80.0%) germination observed with seed treatment of *Trichoderma harzianum* treated seeds, *Trichoderma viride* treated seeds gave better performance with 91.75% increased rate. Application of bacterial antagonists seeds also gave better results by increased germination rate over untreated seeds. Bacterial as well as fungal antagonists enhance rate of germination with decreased germination failure. Lowest germination failure recorded in treatment of *Trichoderma harzianum* and *Trichoderma viride* (20.0% and 25.3%) which was

statically identical; followed by bacterial antagonists. All treatment showed improved seedling health with highest 87.5% healthy and 12.5% infected seedling in *Trichoderma harzianum* treated seeds; followed by, *Trichoderma viride* seeds with 82.6% healthy and 17.4% infected seedling formation. Treated seed of bacterial antagonists *viz.Bacillus subtilis* and *Pseudomonas fluorescence* also recorded 76.21% and 72.22% healthy and 23.79% 27.78% infected seedling in Bottle gourd respectively.

Table 4: Effect of antagonists on seed germination, seedling health and germination failure of Bottle gourd.

Sr. No.	Leaf Extract	Seed germination (%)	Seed germination % over control (+)	Germination failure (%)	Healthy seedlings (%)	Infected seedlings (%)
1	Control	40	0	60	58.33	41.67
2	*T. harzianum*	80	100	20	87.5	12.5
3	*T. viride*	76.7	91.75	23.3	82.6	17.4
4	*P. fluorescens*	60	50	40	72.22	27.78
5	*B. subtilis*	70	75	30	76.21	23.79
	Mean	65.3400	63.3500	34.6600	75.3720	24.6280
	SD	14.3966	35.9915	14.3966	10.0040	10.0040
	CV	22.0334	56.8138	41.5367	13.2728	40.6204
	SE	6.4384	16.0959	6.4384	4.4739	4.4739
	CD 5 %	17.8986	44.7466	17.8986	12.4375	12.4375
	CD 1 %	29.6165	74.0411	29.6165	20.5800	20.5800

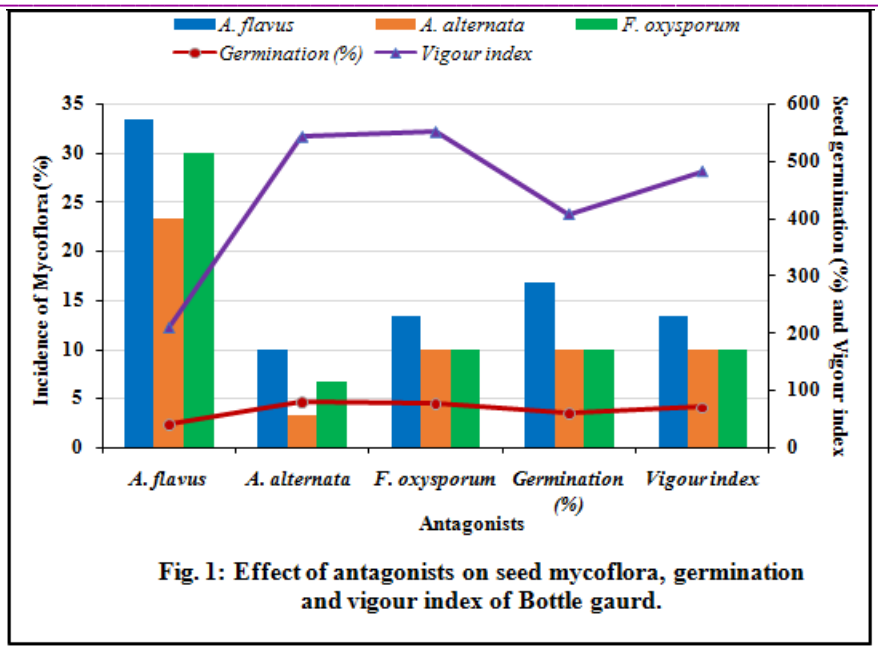

Fig. 1: Effect of antagonists on seed mycoflora, germination and vigour index of Bottle gaurd.

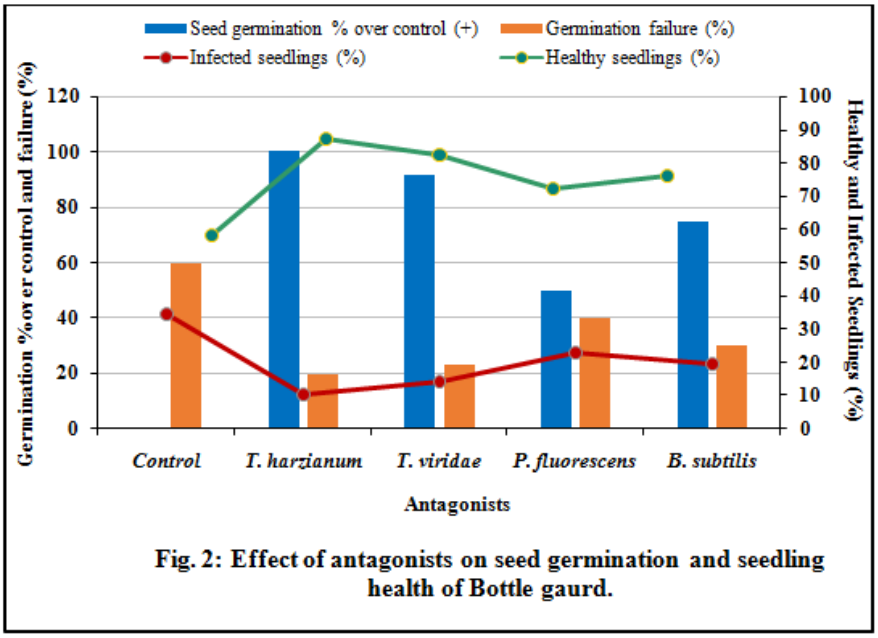

Fig. 2: Effect of antagonists on seed germination and seedling health of Bottle gaurd.

IV. DISCUSSION

Bottle gourd seeds recorded eleven different fungal genera *viz.Aspergillus* spp., *Alternaria* spp., *Fusarium* spp., *Curvularia* spp., *Rhizoctonia* spp., *Rhizopus* spp., *Chaetonium* spp., *Penicillium* spp., *Drechslera* spp. and *Macrophomina* spp. A considerable number of seed borne fungal pathogen with high incidence belonging to genera *Aspergillus, Alternaria* and *Fusarium* spp. (Table 1.3 and 1.4). Agar plate method yielded quantitatively and qualitatively more number of fungi from seeds. Agar plate method was found to be more sensitive than the blotter paper assay. Similar results were obtained by Dervis *et. al.* (2010). Whereas, blotter paper assay yielded maximum number of fungal species in Bottle gourd by Sultana and Ghaffar (2009). Avinash and Rai (2013) concluded that the blotter paper method was the most suitable technique for the isolation and detection of fungal pathogens. They isolated eight fungi from Bottle gourd and Ridge gourd seeds. They reported that the fungi present in seed coat as well as plumule decreases seed germination, seedling vigour and increases seed deterioration.

Bioagents like *Trichoderma harzianum, T. viride, Bacillus subtilis* and *Pseudomonas fluorescens* were used for seed treatment of vegetables. Antagonist micro-organisms kills pathogenic fungi and also improves the germination rate.Preparation of live micro-organism (Bacteria and Fungi) utilized for improving plant growth and crop productivity are generally referred to as microbial inoculants. These microbial inoculants when applied to seeds or plants, enhances the growth of plant or reduces the damage from seed and soil borne pathogens. (Neeraj and verma, 2010; Starovic *et al*., 2013).

Seed treatment with selected bioagents reduces the fungal incidence compared to untreated seeds. Such seed treatment also significantly increases the seed germination, seedling length and vigour index of selected vegetable seeds. This study also revealed that, statistically significant increase in percentage of healthy seedling were recorded after treatment.

The growth parameters and vigour index were recorded high in *Trichoderma harzianum* treatment with enhanced seed germination, upto 80.0 % in selected seeds, seedling vigour index of544 with increased healthy seedling. *Trichoderma viride* seed treatment showed germination of 76.7%, while bacterial antagonist also gave good result with respect to increased seed germination, seedling length, vigour, seedling health and inhibition of mycoflora. This study revealed that seed treatment with *Trichoderma* spp. and bacterial antagonist have potential to reduce the seedborne and disease causing pathogens. Identical results were recorded by Alwathnani *et al.,* 2012; Shanthi and Vittal, 2013; Fayadh and Aledani, 2011.

Trichoderma spp. and bacterial antagonist are also known to provide plants with useful molecule such as glucose oxidase and growth stimulating compounds like auxins, gibberllins and solubilies phosphorus in the soil, which also promotes plant growth and this could be the reason for the higher vigour and percentage germination, as with increased vigour, plants show high resistance to pathogens. (Jegathambigai *et al*., 2009; Neeraj and Verma, 2010; Starovic *et al.,* 2013; Makhlouf and Abdeen, 2015)

Bacillus and *Pseudomonas* have great diverse nature including antibiotic production, nitrogen fixation, degradation of cellulose, starch, pectin and protein. They show good plant growth promoting activities along with biological control of various fungal diseases involving various mechanisms such as antibiosis and lysis. (Shobha and Kumudini, 2012; Pratibha and Siddalingeswara, 2013) Production of antifungal compounds and siderophores is a primary mechanism of disease causing pathogen suppression by *Bacillus* spp. and *Pseudomonas fluorescence.* (Weller, 2007; Abou-Zeid, *et al.,* 2009; Kanti *et al*., 2013).

V. CONCLUSION

The incidence of major mycoflora found statistically similar in company seeds as well as seeds collected from farmers as tested by Standard blotter paper and Agar plate method. The detection of seedborne pathogenic fungi and seed diseases is an important aspect of integral disease management. Thus, the benefits of using fungi and bacteria as mycofungicides and biofertilizers includes decreasing occurrence of plant diseases by inhibiting the growth of pathogens, suppressing the amount of inocula of pathogens, increasing in uptake of nutrient from the soil and atmosphere and producing bioactive compounds, hormones and enzymes, which stimulate plant growth.

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