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RESPONSE OF BIO-INOCULANTS TO EARLY SEEDLING GROWTH IN ASH GOURD AND RIDGE GOURD

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A B S T R A C T

The present study was carried out to evaluate the response of different bio-agents/ bio-fungicides and growth regulators on seed germination and early seedling growth of Ash gourd (*Benincasahispida)* and Ridge gourd (*Luffaacutangula*). Gibberellicacid(GA3)and NAA(Napthoxy Acetic Acid) were used as growth regulator and *Trichodermaviride*, *Pseudomonas fluorescens* and *Trichodermaharzianum*like bio-pesticides or bio-fungicides were used as bio-agents. Eleven treatments were used for the present investigation. Higher germination was observed in Treatment-5, Treatment-6, Treatment-10 and Treatment-11 in contrast to other treatments like chemicals as well as control in case of Ridge gourd and Treatment 7, Treatment 3 and Treatment-11 was found significantly better than all other treatments in case of Ash gourd. Generally root and shoot length increased with the advancement of growth stages. *Pseudomonas fluorescens* (Treatment-4) treated seeds showed higher number of secondary root in comparison to all other treatments. The shoot length and root length in both the cases (ash gourd and ridge gourd) were highly influenced by the bio-inoculants and chemicals but influence of bio-agents were found better than the chemicals .Similarly the seedling weight at 96hours after sowing and at 144 hours after sowing (in both the cases of Ash gourd and Ridge gourd) was reported higher when the seeds were treated with bio-inoculants which reflected the efficacy of the bio-inoculants compared to others.

Keywords: Ash gourd, Ridge gourd, Bio-agents, Early seedling vigor.

INTRODUCTION

The present study was carried out to evaluate the response of different bio-agents/bio-pesticides / bio-fungicides and growth regulators on seed germination and early seedling growth of ash gourd and ridge gourd and to find out the best treatment combination in terms of seedling characters such as seed germination, biomass and phytomass yield under controlled conditions. Gibberellic acid (GA3) and NAA were used as growth regulator and *Trichodermaviride*, *Pseudomonas fluorescens* and *Trichodermaharzianum*like bio-pesticides were used as bio-agents and blitox was used as seed treating chemical. Bio-pesticides are certain types of pesticides derived from such natural materials as animals, plants, bacteria, and certain minerals. Microbial pesticides consist of a microorganism (e.g. bacterium, fungus, virus or protozoan) as the active ingredient. Microbial pesticides can control many different kinds of pests, although each separate active ingredient is relatively specific for its target pest[s]. For example, there are fungi that control certain weeds, and other fungi that kill specific insects or pathogens.

Bio fungicides are the beneficial/ specialized fungi and to attack and control plant pathogens and the diseases they cause. So what are these "specialized fungi" that can attack and control plant pathogens? They are microorganisms that are part of the normal microbiologicalenvironment of most "healthy" soils.

Bio-inoculants are living organisms containing strains of specific bacteria, fungi, or algae which enhance plant uptake of phosphorus and zinc or provide physical barriers against pathogens or: stimulate plant growth or decompose organic residues.

In Our present study, eleven treatments were used for investigation the role of the bio-inoculants on seed germination and early seedling growth on ridge gourd and ash gourd. In the case of seed germination, the

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percentage of seed germination was higher in bio agents treated seeds than in control. Similarly, higher values of shoot and root lengths and fresh and dry weights of ash gourd and ridge gourd were recorded when the seeds treated with bio-inoculants or with growth regulator or chemical like Blitox than the untreated plants. Treatment-4 (*Pseudomonas fluorescens*) treated seeds showed higher number of secondary root in comparison to all other treatments. The better early seedling growth in case of bio-agents treated seeds may be due to the secretion of plant growth hormones by microorganisms inoculated. Generally root and shoot length increased with the advancement of growth stages. The shoot length (3.5cm ±0.8 cm), root length(7.5cm ±0.8 cm), seedling weight in 96hours after sowing (0.28g ±0.8 g), seedling weight in 120 hours after sowing (0.45g ±0.8 g),seedling weight in 144 hours after sowing (0.75g ±0.8 g),shoot weight in 144 hours(0.58g ±0.8 g) and root weight in 144 hours (0.38g ±0.8 g) was reported higher in Treatment-7 and Treatment-3 treatment followed by Treatment -11 & Treatment-10 and Treatment-9 respectively. The overall studies indicated that the growth of Bio-agent treated seedlings excelled over the untreated ones which may due to growth hormonal effect. The observations of the present study is that *T. viride, P. fluorescens*and *T. harzianum* and their combinations have significant role on seed germination and early seedling growth and all the bio-agents either in combination or individually proved to be a booster as a bio-fertilizers/bio-stimulant and reported at par with the growth regulators in relation to seed germination and early seedling growth in case of cucurbits.

MATERIALS AND METHODS

The seeds were sterilized with 2% mercuric chloride solution before treatment. After sterilization, the seeds were washed well with sterile distilled water. Twenty five seeds were selected from each type and dressed well with the water. A control set up was also made by following the same conditions except the addition of bio-inoculants. Ten seedlings were selected at random from each trough and the following observations were made on the 2^{nd}, 4^{th} and 6^{th} day of planting. The seedlings were uprooted gently without causing any damage to the root and shoot systems. The shoot and root lengths were measured with a metric scale. The shoot and root fresh weights were determined using an electronic balance.

After every 12 hours the data on seed germination was recorded upto72 hours. Total number of germinated seeds were counted in all the treatments, at an interval of 12hours after soaking and recorded as emergence count / Petri plate. The data on shoot and root length was recorded at 48, 96 and 144 hours after soaking from 10 randomly selected seedlings. For growth study, height of ten randomly selected seedlings from each treatment and each replication was measured with a meter scale from the ground level to the tip of the shoot, (shoot length) and mean height was calculated from each treatment. The seedling weight was recorded in an Electronic balance in 96 hours and 144 hours after soaking.

Present study was done using following material and method:

Treatments:

Treatment-1(T-1): *Trichodermaviride : Strain -I -20g/ lit*(2%v/v)

Treatment -2 (T-2): *Trichodermaviride : Strain -II -20g/ lit* (2%v/v)

Treatment -3 (T-3) :*Trichodermaharzianum-20g/ lit*; (2%v/v)

Treatment -4 (T-4) :*Pseudomonas fluorescens--20g/ lit*(2%v/v)

Treatment -5 (T-5) :T.viride-Str-1(5.0g/lit) (0.5%v/v) +P.*fluorescens(5.0g/lit)* (0.5%v/v)

Treatment -6 (T-6) : T.viride-Str-2(5.0g/lit) (0.5%v/v) +P.*fluorescens(5.0g/lit)* (0.5%v/v)

Treatment -7 (T-7) : Control :Double Distilled water*)*

Treatment-8 (T-8) : T.*harzianum* (5.0g/lit) (0.5%v/v) +P.*fluorescens(5.0g/lit)* (0.5%v/v)

Treatment -9 (T-9) : Blitox (2.0g/lit) (0.2%v/v)

Treatment -10 (T-10): Seed plus (Gibberellic acid -10%)-2.0g/lit

Treatment -11 (T-11) :Sudhagerminaid (Growth regulator)-1.0g/lit (0.1%v/v)

(Population density of the bio-inoculant- 2 X 109 (c.f.u./gram),

Microbial adjuvant - 2%,

Microbial media residue inert ingredient - 95-97%)

The experiment was conducted in Petri plate. 50numbers of seeds were placed on the filter paper within the Petri plates. 5 ml of liquid solution of each treatment were applied on the filter paper on every 24 hours intervals.

RESULTS AND DISCUSSION

The results indicated that the growth of *Trichodermaviride, Trichodermaharzianum, Pseudomonas fluorescens* or their combinationtreated seedlings excelled over the untreated ones. Germination

Counts: The germination percentage was influenced by different treatments (Table-1). Result showed that the maximum number of seedling emergence was reported in T-1(94%) and T-8 treatment (96%), which contains bio-agents (T.viride-Str-1 -10.g/lit &5g/lit) which was at par with T-13 (Blitox 2.0g/lit) & T-17 (Sudhagerminaid Growth regulator-1.0g/lit). In all the treatments the germination percentage was significantly higher than the control .The results indicates that the bio-agents like *T.viride, T. harzianum and P. fluorescens* have similar type of influence on seed germination like growth regulator and chemicals. The explanation of the present study is that *T. viride*, *P. fluorescens* and *T. harzianum* and their combinations have significant role on seed germination and early seedling growth and all the bio-agents either in combination or individually proved to be a booster as a bio-fertilizers/bio-stimulant and reported at par with the growth regulators in relation to seed germination and early seedling growth.

Table No. 1. Response of Bio-inoculants to ASH GOURD.

Treatments	96 HOURS				144 HOURS			
	Germination %	Seedling wt (g)	Shoot length (cm)	Root length (cm)	Germination %	Seedling wt (g)	Shoot length (cm)	Root length (cm)
T1: T.v (str.-I) (20g/lit)	40.0D	2.29 CD	3.1CD	5.1D	45.0D	4.49 BC	5.0D	9.6DE
T2:T.v (str.-II) (20g/lit)	35.0E	2.14D	3.5B	5.6BC	40.0EF	4.31 DE	5.5 C	9.9D
T3 :T.h (20g/lit)	50.0B	2.88A	2.9D	5.9AB	54.0AB	4.89A	5.9AB	10.9AB
T4: P.f (20g/lit)	45.0C	2.12DE	2.7EF	4.2E	50.0BC	3.91F	4.9D	11.5A
T5 : T.v (str.-I)+P.f(10=10g/lit)	45.0C	2.54B	3.8A	4.9D	46.0CD	4.62B	5.8BC	8.9F
T6: T.v (str.-II)+P.f (10=10g/lit)	35.0E	2.57B	3.2C	3.9E	36.0 F	4.67 AB	6.2 A	9.2EF
T7: T.h (str.-I)+P.f(10=10g/lit)	55.0A	2.12 DE	3.6AB	5.9AB	57.0A	4.15E	5.6 BC	10.9B
T8: Control	15F	1.65F	2.3E	5.7BC	21.0G	4.26E	4.0E	6.8H
T9: Blitox (2g/lit)	40D	2.31 CD	3.2C	2.5F	43.0DE	3.65G	5.6BC	8.7F
T10:Seed plus (2g/lit)	46C	2.4BC	2.9D	5.5C	39.0EF	4.45BC	4.9 D	9.2EF
T11: Sudhagerminaid (1g/lit)	50B	2.57B	3.6AB	6.2A	54.0 AB	4.67 AB	5.6 BC	10.5BC
Ems	5.70	0.015	0.016	0.048	6.07	0.02	0.03	0.143
L.S.D	3.97	0.2037	0.210	0.364	4.09	0.235	0.311	0.628

The seed germination studies revealed that the percentage germination of seeds were highest in *Trichodermaharzianu*combination *with Pseudomonas fluorescens* (5.0g/l +5.0g/l) followed by *Trichodermaharzianu(10.0g/lit)&*Sudhagerminaid (Growth regulator)-1.0g/lit , Seed plus (Gibberellic acid -10%)-2.0g/lit and Blitox (2.0g/lit) respectively. The fungal growth was best checked when the seed treated

blitox(no infection) followed by *Pseudomonas fluorescens*(<5% infection*)*, *P.fluorescens combined with Trichodermaharzianu* (<10% infection) respectively. The inoculation of the bio-fungicide in both the species showed a considerable increase in the seed germination than the other under same experimental conditions. The reason for this may be due to the tremendous pressure developed inside the seeds, which is responsible for breaking of the seed coat quickly (Sifton, 1959). This pressure may be induced by phytohormones especially auxin, indole acetic acid (IAA), cytokinin and gibberellic acid (GA) like substances secreted by *Trichodermaharzianu* or P*fluorescens or Trichoderm*. *viride* .The findings shows close proximity withOkon, 1985 and1986 in case of a bio-fartilizer, *Azospirillum*in case of rice.

The observations made on 2nd, 4th and 6th days of sowing revealed that bio-inoculums treated seeds had higher early seedling development than the control. The seedlings from these particular bio-inoculants treated seeds had longer shoot and root lengths than the untreated ones. From our experimental findings it may be concluded that the seed dressing by these bio-inoculants induces the production of plant growth promoting substances and leads to the increase of shoot and root length (Table 2).

Table No. 2. Response of Bio-inoculants to RIDGE GOURD

Treatments	96 HOURS				144 HOURS			
	Germination %	Seedling wt (g)	Shoot length (cm)	Root length (cm)	Germination %	Seedling wt (g)	Shoot length (cm)	Root length (cm)
T1: T.v (str.-I) (20g/lit)	25.0C	3.44C	3.70C	3.90DE	28.0E	4.54B	6.10C	6.20D
T2:T.v (str.-II) (20g/lit)	27.0C	3.25 CD	3.40D	3.60EF	30.0DE	4.35BC	5.20EF	5.40F
T3 :T.h (20g/lit)	30.0C	4.20 AB	3.40D	5.35B	33.0CD	5.60A	6.10C	5.90F
T4: P.f (20g/lit)	25.0C	4.60A	2.60F	3.60EF	28.0E	5.60A	5.20EF	5.30G
T5 : T.v (str.-I)+P.f(10=10g/lit)	40.0B	3.10CDE	3.30DE	3.10FG	43.0B	4.20 BCD	5.30E	7.80C
T6: T.v (str.-II)+P.f(10=10g/lit)	45.0AB	2.60F	3.40D	3.60EF	35.0C	3.80D	5.60D	6.20D
T7: T.h (str.-I)+P.f(10=10g/lit)	30.0C	3.93B	4.20A	5.40B	33.0CD	4.20 BCD	6.40B	7.80C
T8: Control	15.0D	2.60 F	2.20G	3.00FG	18.0F	3.90CD	4.00G	3.80H
T9: Blitox (2g/lit)	28.0C	2.80 EF	3.30DE	4.60C	33.0CD	4.10 BCD	6.50AB	6.10DE
T10:Seed plus (2g/lit)	40.0B	3.30CD	3.20E	6.70A	45.0B	4.50B	6.70A	9.10A
T11: Sudhagerminaid (1g/lit)	50.0A	2.90 DEF	3.90B	4.30CD	55.0A	4.10 BCD	5.70D	8.10B
Ems	19.60	0.07	0.011	0.013	8.85	0.100	0.019	0.018
L.S.D	7.36	0.44	0.166	0.608	4.95	0.525	0.229	0.2231

Secretion of plant growth hormones by *Azospirillum* was reported in several cereals and grasses (Balasubramaniam and Kumar, 1987),(Bashan and Holgain, 1995). This also reflects a specific capability of

the host plant to attract the bacteria and modify the rhizosphere and/or to respond to some bacterial activity and benefit from it (Bottini et al., 1989). The fresh and dry weights of root and shoot system of ridge gourd and ash gourd were also found to be increased to a considerable extent in *Trichodermaharzianu*combination *with Pseudomonas fluorescens* (5.0g/l +5.0g/l) followed by *Trichodermaharzianu(10.0g/lit)* treated seedlings. (Table 1 and 2). T-4 (*Pseudomonas fluorescens*) treated seeds showed higher number of secondary root (8.5 - 15.8) in comparison to all other treatments. This may be due to the formation and development of numerous root branching, root hairs and primary and secondary lateral roots which increases the nutrient uptake capacity of roots (Gopalswamy and Vidhyasekaran, 1988; Hartmann et al., 1983).

This effect on the root system as well as more root colonization and root proliferation are probably due to the growth hormones secreted by the bacteria or fungi. The increased nitrogen uptake from the soil might have correspondingly increased the biomass to some extent. The changes in root functions due to *Azospirillu* treatment in different wheat cultivars were also reported (Kapulnik et al., 1981). These growth enhancing effects are of interest because of their potential significance for yield increases in agronomic systems in which the use of fertilizers is the limiting factors for their development (Sarig et al., 1984). Net shoot, root and seedling weight was higher in *bio-inoculants* treated seedlings than in control. It may be due to the absorption of nutrients from the growing media and stimulate the metabolism of photosynthesis. Photosynthetic activity plays an important role in the increase of leaf area leading to more biomass accumulation. The plants like *Digitariadetcumbens*, *Panicum maximum* and *Pennisetumamericanum* were subjected to *Azospirillum*inoculation and observed that the photosynthetic rate and dry matter contents were increased to a limited extent (Smith et al., 1976; Sarig et al., 1984;).

The increased chlorophyll content could be correlated with the high level of photosynthesis this might be due to uptake of more nitrogen from the growing media, and for these activities the working bacteria or fungi or artificial growth inoculants have found to be a great importance. The experimental findings may be due to increasing level of protein content which may be due to the presence of kinetin which promotes the amino acid content which in turn helps in active protein synthesis (Tien et al., 1979). Similarly, the findings may be due to increasing level of sugar content in the leaves might also be due to active role of bio/artificial inoculants in sugar metabolism (Watanabe et al., 1981).

CONCLUSSION

From the above study, it was observed that among the cucurbits species like ash gourd and ridge gourd tested in response to *Trichodermaharzianu, T.viride either alone or* combined *with Pseudomonas fluorescens* inoculation, high response was observed in case of seed germination, early shoot and root length, early seedling weight which may be due to high phytomass and biomass accumulation. From the experimental findings it may be concluded that above parameters may be influenced by bio-antagonists under study . The beneficial effect of *Trichodermaharzianu ,T.viride and Pseudomonas fluorescens*varies itself which depend upon the plant species, microbial strains, and species X microbial strain interaction.

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