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EFFECT OF FOLIAR SPRAY OF GIBBERELLIC ACID ON THE RHIZOSPHERE AND RHIZOPLANE MYCOFLORA OF *VIGNA SINENSIS* L. SEVI EX. HASSK.

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The effect of foliar spray of different concentrations of gibberellic acid on the rhizosphere mycoflora was studied at pre-flowering, flowering and fruiting stages. Foliar spray of different concentrations of gibberellic acid reduced the number of fungi/g of dry soil at each concentration and each stage. Fungi number also reduced in rhizosphere and rhizoplane at each stage. Maximum inhibition was noted at 200 ppm concentration.

Keywords : Foliar application, Gibberellic acid, Rhizosphere, Rhizoplane mycoflora.

Introduction

Foliar application of different chemicals and their influence on the rhizosphere microflora begin with the work of Halleck and Cochrane[1], who showed reduction in the bacterial population in the rhizosphere. The effect of growth regulators particularly gibberellic acid (GA) has been studied by a number of workers[2-6] on rhizosphere and they were of the view that quantitative change occurs in the rhizosphere microflora. Deb and Bora[7] reported that foliar spray of NPK. fertilizer influenced the rhizosphere mycoflora of pea plants.

Materials and Methods

The solution of GA was prepared in three concentrations (50, 100 and 200 ppm) in distilled water and plants were sprayed with a home spray automizer. Three pots each with three plants were treated with each concentration of solution. Control pots were sprayed with distilled water only. First spray was done when the seedlings were 15 days old. Three sprays were made in all for two regular days, at all the three stages viz., pre-flowering, flowering and fruiting stages. Soil surface was covered with sterilized cotton to avoid falling of drops direct on the soil. Samplings were done 10 days after second spray.

In each sampling, three plants (one each from the three pots) were dugout with the help of sterilized trowel and adhering soil of root system was removed by gentle tapping. The roots were cut off and kept in conical flask containing 100 ml of sterilized distilled water. Five petridishes were inoculated with 1 ml aliquot and 15 ml of sterilized and cooled Czapek's medium.

The petridishes were incubated for 5 to 6 days at 25 °C. The weight of rhizosphere soil was determined by evaporating the rhizosphere suspension in conical flask placing in an oven at 105 °C for 24 hours. From this, the number of fungi/g dry weight of soil was calculated. The number of fungal species and number of fungi/g dry weight of soil were recorded for the control and treated plants after each sampling. All the data were analysed statistically.

Results and Discussion

In the present investigation, the effect of foliar application viz., 50, 100 and 200 ppm of gibberellic acid on rhizosphere and rhizoplane mycoflora has been studied at different stages of plant growth. The variation in the number of fungi/g of dry soil was found to be statistically significant. All the three concentrations of G.A. viz., 50, 100 and 200 ppm inhibited the number of fungi/g of dry soil at each sampling. There was a regular pattern of decrease in the total number of rhizosphere fungi. Maximum decrease in the rhizosphere fungi/g of dry soil was noted at 200 ppm concentration (Table 1).

At the pre flowering stage the percentage occurrence of *Paecilomyces varioti*, *Aspergillus niger*, *A. flavus*, *A. terreus* and *Verticillium glaucum* increased in the rhizosphere where as percentage occurrence of *P. fusisporus* decreased. *Cladosporium herbarum* was only confined to the rhizosphere of control plant. Yellow sterile mycelium was only present in 50 and 100 ppm treated plants whereas it disappeared in 200 ppm treated plants. The percentage occurrence of *P. citrinum* increased and *Fusarium udum* decreased in treated plants (Table 2).

The percentage occurrence of *Paecilomyces fusisporus*, *Curvularia lunata*, *Alternaria humicola*, Black sterile mycelium and White sterile mycelium increased in the rhizosphere of treated plants at flowering stage whereas percentage occurrence of *Rhizopus nigricans*, *Aspergillus niger* and *F. udum* increased on the rhizoplane

Table 1. Average number of fungi/g of dry soil (in thousand) in the rhizosphere (R) and the number of species isolated from the rhizosphere and rhizoplane (RP) of the test plant sprayed with different concentrations of gibberellic acid.

Stage of plant	Number of spraying	Conc. of gibberellic acid	Gibberellic acid			Control		
			No. of fungi/g of dry soil (X1000) R	No. of species in R	No. of species on RP.	No. of fungi/g of dry soll (X1000) R	No. of species in R	No. of species on RP.
Pre flowering stage	1	50 ppm	137.9	16	6	143.8	19	6
		100 ppm	121.9	14	5	143.8	19	6
		200 ppm	103.1	13	5	143.8	19	6
Flowering stage	2	50 ppm	127.6	16	4	130.0	17	5
		100 ppm	103.6	13	3	130.0	17	5
		200 ppm	101.0	12	3	130.0	17	5
Fruiting stage	3	50 ppm	129.3	16	4	139.0	18	5
		100 ppm	121.9	15	4	139.0	18	5
		200 ppm	110.8	10	3	139.0	18	5

of treated plants (Table 3).

At fruiting stage the percentage occurrence of *A. flavus*, *A. terreus*, *Stachybotris atra* and *Nigrospora spherica* increased whereas the per centage occurrence of *F. nivale*, decreased in the rhizosphere of treated plants. The percentage occurrence of *A. niger*, *A. flavus* and White sterile mycelium increased and percentage occurrence of *F. udum* decreased on the rhizoplane of treated plants. *A. sydowi* and *Myrothecium roridum* only confined to control plants (Table 4).

Chromatographic analysis of the root extract of separately treated plants with gibberellic acid was also done for the presence of amino acids and sugars at pre-flowering, flowering and fruiting stages. It was found that there was no change in quality of amino acids (Table 5).

The effect of foliar application of hormones and antibiotic substances on rhizosphere mycoflora has been studied by many workers. Venkata Ram[8] reported a decrease in fungal numbers in the rhizosphere of tea after foliar spray of urea. Kandaswamy and Rangaswamy[9] observed a decrease in rhizosphere microflora of *Sorgham* varieties sprayed with urea. Ramachandra Reddy[10-11] working with the rice seedling, reported a decrease in rhizosphere effect in case of plants treated with hormones, antibiotics and urea. Sethunathan[12-13] observed a decrease in rhizosphere microflora in *Cajanus cajan* (Mill. Sp.) sprayed with G.A. and melic hydrazide (MH). Singh[14] studied the effect of foliar application of hormones on rhizosphere and rhizoplane mycoflora and reported that rhizosphere and rhizoplane mycoflora are enhanced due to spray. Mahamood *et al.*[6] studied the effect of foliar spray of GA_3, MH_3 and 2-4 D on rhizosphere mycoflora and found that all the three chemicals stimulated the mycoflora in the rhizosphere. Dublish[5] studied the effect of foliar application of antibiotics, growth hormones and urea on rhizosphere mycoflora of *A. esculentus* and reported that the effect of both the hormones on rhizosphere mycoflora were statistically insignificant.

Agnihotri[15] reported that the microflora in the rhizosphere is increased due to an increase in the exudation of amino acid and glutamine, glucose and fructose and decrease in organic acids. Balasubramanian and Rangaswamy[16] observed the decrease in the number of mycoflora in the change in the root exudates pattern of amino acids sugars. Halleck and Cochrane[1] reported that the change in the number of rhizosphere microflora is due to the direct translocation of chemicals to root and exudates in soil. The present findings are also in accordance with the observation of Balasubramanian and Rangaswamy[16] and Halleck and Cochrane[1].

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Table 2. Effect of foliar spray of gibberellic acid on the percentage occurrence of fungi isolated from the rhizosphere and rhizoplane at pre-flowering stage.

No.	Fungi	Rhizosphere				Rhizoplane			
		C	50 ppm	100 ppm	200 ppm	C	50 ppm	100 ppm	200 ppm
1.	*Rhizopus nigricans*	5	-	-	7	11	13	6	-
2.	*Mucor luteus*	3	1	-	5	-	-	-	-
3.	*Cephalosporium coremioides*	5	-	5	-	-	-	-	-
4.	*Paecilomyces varioti*	6	7	11	11	-	-	-	-
5.	*Paecilomyces fusisporus*	9	7	8	5	-	-	-	-
6.	*Trichoderma lignorum*	5	-	-	5	11	7	12	-
7.	*Aspergillus niger*	5	10	9	10	22	13	29	7
8.	*Aspergillus flavus*	7	13	6	30	-	-	-	60
9.	*Aspergillus luchuensis*	-	3	-	-	-	-	-	-
10.	*Aspergillus terreus*	8	7	18	5	-	-	-	-
11.	*Aspergillus nidulans*	6	18	17	1	-	-	-	-
12.	*Penicillium citrinum*	6	3	2	-	17	47	35	-
13.	*Verticillium glaucum*	3	-	6	7	-	-	-	-
14.	*Humicola grisea*	2	3	-	-	-	-	-	-
15.	*Cladosporium herbarum*	9	-	-	-	-	-	-	-
16.	*Alternaria humicola*	7	3	7	4	-	-	-	7
17.	*Fusarium udum*	3	2	-	-	22	7	-	13
18.	*Myrothecium roridum*	3	4	3	5	-	-	-	-
19.	Black sterile mycelium	3	3	1	-	-	-	-	-
20.	White sterile mycelium	5	14	5	5	17	13	18	13
21.	Yellow sterile mycelium	-	2	2	-	-	-	-	-
	Total no. of species isolated	19	16	14	13	6	6	5	5

C = Control

Table 3. Effect of foliar spray of gibberellic acid on the percentage occurrence of fungi isolated from the rhizosphere and rhizoplane at flowering stage.

No.	Fungi	Rhizosphere				Rhizoplane			
		C	50 ppm	100 ppm	200 ppm	C	50 ppm	100 ppm	200 ppm
1.	*Rhizopus nigricans*	-	-	-	-	27	36	31	-
2.	*Cunninghamella*	7	6	12	5	-	-	-	-
3.	*Cephalosporium coremioides*	5	2	10	-	-	-	-	-
4.	*Paecilomyces fusisporus*	4	6	5	7	-	-	-	-
5.	*Aspergillu niger*	12	10	15	12	20	36	38	31
6.	*Trichoderma flavus*	9	8	-	5	-	-	-	-
7.	*Aspergillus sydowi*	-	6	3	-	-	-	-	-
8.	*Aspergillus terreus*	6	6	5	-	-	-	-	-
9.	*Penicillium citrinum*	5	-	-	9	13	21	-	-
10.	*Stachybotris atra*	3	3	5	4	-	-	-	-
11.	*Cladosporium herbarum*	5	6	7	-	-	-	-	-
12.	*Curvularia lunata*	5	6	10	12	-	-	-	-
13.	*Curularia tetrametra*	5	5	-	-	-	-	-	-
14.	*Alternaria humicola*	6	9	10	11	-	-	-	-
15.	*Fusorium uchum*	7	5	-	9	20	7	31	46
16.	*Fusarium Roseum*	3	-	2	5	-	-	-	-
17.	*Myrothecium roridum*	6	8	2	-	-	-	-	-
18.	Black sterile mycelium	3	7	-	7	-	-	-	-
19.	White sterile mycclium	9	7	14	14	20	-	-	23
	Total *no species isolated*	17	16	13	12	5	4	3	3

C= Control

Table 4. Effect of foliar spray of gibberellic acid on the percentage occurrence of fungi isolated from the rhizosphere and rhizoplane at fruiting stage.

No.	Fungi	Rhizosphere				Rhizoplane			
		C	50 ppm	100 ppm	200 ppm	C	50 ppm	100 ppm	200 ppm
1.	*Mucor mucedo*	8	7	3	16	-	-	-	-
2.	*Cunninghamella echinulata*	2	-	6	-	-	-	-	-
3.	*Neocosmospora*	2	5	6	9	-	-	-	-
4.	*Paecilomyces fusisporus*	6	5	6	6	-	-	-	-
5.	*Aspergillus niger*	10	11	10	-	25	35	34	50
6.	*Aspergillus flavus*	10	10	18	15	20	22	26	-
7.	*Aspergillus sydowi*	4	-	-	-	-	-	-	-
8.	*Aspergillus terreus*	3	5	12	9	-	-	-	-
9.	*Penicillum citrinum*	-	5	6	-	13	-	20	-
10.	*Verticillium glaucum*	6	-	-	7	-	-	-	-
11.	*Stachybotris atra*	2	2	3	7	-	-	-	-
12.	*Nitrospora sphaerica*	6	5	6	9	-	-	-	-
13.	*Curvularia lunata*	6	5	-	-	-	-	-	-
14.	*Curvularia tetramera*	6	10	-	-	-	-	-	-
15.	*Alternaria humicola*	4	5	6	-	-	-	-	-
16.	*Fusorium udum*	-	5	6	7	34	28	-	25
17.	*Fusorium nivale*	7	5	6	-	-	-	-	-
18.	*Myrothecium roseum*	6	5	3	5	-	-	-	-
19.	*Myrothecium roridum*	6	-	-	-	-	-	-	-
20.	White Sterile mycelium	6	10	3	10	8	15	20	25
	Total no. of species isolated	18	16	15	10	5	4	4	3

C=Control

Table 5. Effect of foliar spray of different concentrations of gibberellic acid in the presence of amino-acids in the root extract at pre-flowering, flowering and fruiting stages of plant growth.

No.	Known amino-acids	Different concentrations of GA											
		Control			50 ppm			100 ppm			200 ppm		
		PF	F	Fr	PF	F	Fr	PF	F	Fr	PF	F	Fr
1.	DL-Alanine	-	-	-	-	-	-	-	-	-	-	-	-
2.	L. Cystine	-	-	-	-	-	-	-	-	-	-	-	-
3.	DL-Tryptophan	-	-	-	-	-	-	-	-	-	-	-	-
4.	L. Tyrosine	+	+	+	+	+	+	+	+	+	+	+	+
5.	Glycine	+	+	+	+	+	+	+	+	+	+	+	+
6.	DL. Iso Leucine	+	+	+	+	+	+	+	+	+	+	+	+
7.	DL B. Phenyl alanine	+	+	+	+	+	+	+	+	+	+	+	+
8.	DL. Serine	-	-	-	-	-	-	-	-	-	-	-	-
9.	L. Ornithine mono HCl	+	-	-	+	-	-	+	-	-	+	-	-
10.	L. Glutamic acid	-	-	-	-	-	-	-	-	-	-	-	-
11.	L. Proline	-	-	-	-	-	-	-	-	-	-	-	-
12.	DL. Methionine	-	-	-	-	-	-	-	-	-	-	-	-
13.	DL. Thrionine	+	+	+	+	+	+	+	+	+	+	+	+
14.	L. Arginine mono HCl	+	+	-	+	+	-	+	+	-	+	+	-
15.	DL. Valine	-	-	-	-	-	-	-	-	-	-	-	-
16.	L. Hydroxy proline	-	-	-	-	-	-	-	-	-	-	-	-
17.	DL. 2 Amino Butyric Acid	+	+	+	+	+	+	+	+	+	+	+	+
18.	DL. Aspartic Acid	-	-	-	-	-	-	-	-	-	-	-	-
19.	L. Leucine	-	-	-	-	-	-	-	-	-	-	-	-
20.	L. Cystine HCl	-	-	-	-	-	-	-	-	-	-	-	-
21.	DL. Alanine	-	-	-	-	-	-	-	-	-	-	-	-
22.	L. Histidine mono HCl	-	-	-	-	-	-	-	-	-	-	-	-
	Total	8	7	6	8	7	6	8	7	6	8	7	6

+ = Present PF = Pre - flowering stage Fr = Fruiting stage
- = Absent F = Flowering stage

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