Screening of chemical fungicides in control of *Rhizoctonia solani* causing aerial or web blight of soybean

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ABSTRACT

Foliar/web blight of soybean is one of the major fungal diseases. A pot experiment was conducted at the college of agriculture in C.S.A. U. A & T. Kanpur (U.P) during the Kharif season of 2011-2013 to study the Aerial blight of soybean caused by *Rhizoctonia solani* Kuhn. The efficacy of eight different fungicides, *viz.*, Roko, Mancozeb, Indofil Z-78, Acrobat, Vitavax, Taquat, Matco & Antrocol (i.e. application @ 0.2 per cent were tested *in vitro* and *in vivo* conditions were against *Rhizoctonia solani*, the causal organism of aerial blight of soybean. Results revealed that the under *in vitro* conditions among all the fungicides, Vitavax was found significantly highly effective inhibiting complete radial growth of fungus, i.e. application @ 0.2 per cent. Next in order Roko and Indofil Z-78 were found effective with 97.63 per cent and 97.37 per cent inhibition at @ 0.2 per cent. This was closely followed by Mancozeb 96.72 per cent. However, Antrocol fungicide was found least effective with 36.39 per cent.

Key words : Rhizoctonia solani, Fungicides, Aerial blight and Soybean

Introduction

Foliar/web blight of soybean caused by *Rhizoctonia* solani Kuhn is a severe disease and causes heavy losses in soybean production (Anwar *et al.*, 1995). Soybean [*Glycine max* (L.) Merill] is a leguminous oilseed crop having worldwide adaptation. It is known as "Golden bean" or "Miracle crop", as it is the richest source of protein (40%) and oils (20%). Its protein has a good balance of amino acids except methionine and its oil is rich in unsaturated fatty acids. Soybean belongs to the *fabaceae* or *leguminaceae* (legume) family. Seeds from these varieties were used primarily as pulses by the local population and the green and dried vegetative parts

were used as forage for cattle (Saxena, 1976). The native of soybean is eastern Asia. Soybean was introduced to India during 1880. This is economically important pulse crop suffers from a number of diseases which is caused by fungi, bacteria, viruses, nematodes and other physiological disorders also disturbs the normal physiological process of the plants. Among the various disease, the incidence of aerial blight of Soybean disease has been observed with varying degrees of disease in Kanpur districts of Uttar Pradesh in 2011-13. Soybean [*Glycine max* (*L.*) Merril] plants are infected by the pathogen at any stage of development, which causes very rapid defoliation and frequent crop failure (Wrather *et al.*, 2001). Fenille *et al.* (2002) have reported 31-60% yield

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The symptoms of aerial blight of soybean caused by R. solani, as leaf and pod spots, leaf blight, defoliation, stem and petiole lesions, cob web like mycelium and sclerotia developed over infected leaves were described by Atkins and Lewis (1954). The *R*. solani produces sclerotia as survival structure which is brown to black composed of clusters of melanin encrusted, thick walled cells, formed by repeated branching from short, thick, lateral hyphae, when produced on plant parts, it is difficult to separate the sclerotia from their surrounding embeded sclerotia. Temperature is more considerable parameter for their growth and development along with sclerotia production. Under certain conditions young plants were found heavily affected with the disease leading to premature death of plants and substantial losses in yield. The disease attacks all the aerial part of the plants i.e. leaves, stem, petioles and pods when the disease occurred in severe form. The leaves and host plant has given blighted appearance. The mycelium has found web like appearance on the leaves so that disease has been named as web blight of Soybean. It also inhibited root elongation causing seedling root rot, yellowing and shredding of cotyledons and leaves in soybean.

Materials and Methods

Isolation and purification of the pathogen

Survey and collection of sample showing blight symptoms from soybean growing areas of central regions mainly in Kanpur were done. The isolates (Student Farm C.S.A .Uni. of Ag. & Tech. Kanpur, Nawabganj, Bilhaur, Bithoor and Chaubeypur) of *R.solani*, were isolated on PDA and purified through hyphal tip/single sclerotial method (Rangaswami and Mahadevan, 2004). In cultural studies; mycelial discs of 5 mm diameter from 3 days old cultures of each isolates were transferred into the center of sterilized different culture media and plates were incubated for 5 days at 28±1°C. The basic cultural characteristics such as colony diameter, colour and growth pattern were studied. The colony colour was determined with help of Munsell's soil colour chart (Munsell, 1954). Based on mycelial pigmentation, the cultures were assigned in different groups as dark brownish, dark white and dirty brown. Colony growth pattern was recorded by visual observation according to growth of hyphae. Colony diameter growth rate was recorded on the six different media i.e. five synthetic media (Asthana and Hawkers, Czapek's agar, Sabourand's medium, Oat meal medium and Richard's agar) and one semi synthetic medium (PDA) after 48 hrs of inoculation at 28±1°C with the help of scale. The isolates were classified into slow; splash, fast, thin and fluffy growth. Growth was measured of the each isolate with three replications. The number, weight, colour, texture (smooth and rough) and patterns of sclerotia formed were recorded.

Identification of pathogen

The pathogen produced the characteristics symptoms on the affected parts of the soybean plants. It was identified on the basis of its morphological and cultural characters and pathogenic behaviour towards the host.

Pathogenicity test of the pathogen

Pathogenicity test of the isolate obtained from the affected soybean root and other parts of the host. For pathogenicity test surface sterilized (0.1% Hgcl₂) seed of soybean variety "Gaurav" were sown in the 9 inch pots filled with sterilized (autoclaved) soil. Four week old plants were inoculated with mycelial suspension of the tested fungus grown on Potato Dextrose Broth medium in 250 ml conical flasks. Such inoculated plants were kept in net house and were irrigated as and when required. Proper humidity for development of the disease was maintained by using the bell jar with cotton. Three replication of the inoculated plants along with an uninoculated control were maintain in net house, these inoculated and uninoculated plants both were regularly observed for the development of the disease. The symptoms of the disease observed in the natural condition. Observation was also recorded on the time taker for the development of the disease after inoculation. From the diseased plant in pots the pathogen was isolated on Potato Dextrose Agar

 Table 1. The observation of sclerotial development in Petri plates

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No. of sclerotia/Petri-plate	Grade	Symbol
0	Nil	-
15 or below	Poor	+
16-30	Fair	++
31-50	Good	+++
51 and above	Excellent	++++

medium and was compared with the original culture of the pathogen with which the plants were inoculated thus, Koch's postulates rules were proved.

Production of perfect stage in culture medium

The most vigorously growing isolate of the test fungus indicates to produce perfect stage in the laboratory by applying the method proposed by Stretton *et al.*, 1964. The test isolate was grown on Potato Dextrose Agar medium in Petri-plate for 7 days. The loads were then removed and the culture were covered to 1 cm. depth with steamed soil and watered 4 times a day to keep Petri plates moist and then incubate at room temperature 26-28°C.

The following eight fungicides were evaluated against the pathogen under laboratory conditions to screen out the best fungicides depending upon their inhibitory effect on the growth of the fungus (Rhizoctonia solani). The different fungicides were screened for their efficacy against the pathogen by "Food poison techniques" described by Schmitz (1930) in which required quantity of each fungicide was thoroughly mixed with 100 ml well sterilized potato dextrose agar medium contained in 150 ml flasks. Now this medium mixed with fungicides was poured in Petri-plates and allowed to solidify. Each treatment was replicated three times. One set of control was also kept in which the medium was not mixed with fungicides. Equal pieces of the fungal growth, cut by the cork borer were inoculated in Mechanism of interaction was observed and the data were expressed as percent inhibition of growth by the following formula (Bliss, 1934).

Percent inhibition (P.I.) = $\frac{\text{Growth in control (mm)} - \text{Growth in treated plates (mm)}}{\text{Growth in control (mm)}} \times 100$

Results and Discussion

Pathogenic behaviour of Rhizoctonia solani

Method used to test the pathogenicity of this fungus is described under 'Materials and Methods' and the result obtained are given in Table 3.

The results in Table 3 can be concluded that injured leaves were found to be more susceptible than uninjured. In all leaves typical symptoms appeared after 24 hrs of inoculation with mycelial suspension, the symptoms appeared as small circular to irregular water soaked spots on the leaves. These spots later become change into tan a colour and enlarge on the leaves; the typical symptoms were also produced on stems and pods. The symptoms developed on leaves after 8 days of inoculation. The pathogen was isolated from inoculated plant; the

S.No.	Fungicides	Active ingredients	Dose percent
1	Indofil Z-78	Zineb 75% wp	0.2
2	Roko	Thiophanote methyl 70% wp	0.2
3	Vitavax	5,6-dihydro-2-methyl-N-phenyl-1,4-oxathiin-3-carboxanlido	0.2
4	Mancozeb	[[1,2-ethanediylbis-[carbamodithioato]](2-)]manganese, mixture	
		with [[1,2-ethanediylbis-[carbamodithioato]]-(2-)] zinc	0.2
5	Acrobat	Dimethomorph 50% wp	0.2
6	Taquat	Capton 70% +Hexaconazole 5%wp	0.2
7	Matco	Metalaxyl 8% + Mancozeb 64% wp	0.2
8	Antrocol	Propineb 70% ai	0.2
9	Control	-	

Table 2. Screening of fungicides against the pathogen in vitro

Table 3. Showing pathogenic behaviour of *Rhizoctonia solani* on soybean plants.

S. No.	Treatment	No. of leaf inoculated	No. of leaf infected	Infection in percentage
1.	Injured	20	20	100
2.	Un Injured	20	15	75
3.	Sprayed with sterilized water	20	00	00



T-1	=	VITAVAX
T-2	=	ROKO
T-3	=	INDOFIL Z-78
T-4	=	MANCOZEB
T-5	=	ACROBAT

T-6	=	TAQUQT
T-7	=	MATCO
T-8	=	ANTROCOI
T-9	=	CONTROL

organism thus isolated resembled the original pathogen in morphological and cultural characteristics and was able to produce typical symptoms of aerial blight on soybean.

Screening of fungicides against the *Rhizoctonia* solani in-vitro:

Eight fungicides were tested against the pathogen under laboratory conditions. The screening of the best and effective fungicides were done on the basis of the inhibitory effect of the fungicides on the



Fig. 2. Efficacy of different fungicides against *Rhizoctonia* solani

growth of the fungus by the Agar plate method after 7 days of incubation at 27 ± 1 °C (Fig. 1). The average diameter of the fungal colonies was noted in the poured plates containing different fungicides as reported in Table 4.

It is evident from the results of Table 4 and its corresponding histogram that out of 8 fungicides @ 0.2% tested in laboratory, the Vitavax completely inhibited the growth of fungus. Roko, Indofil Z-78 and Mancozeb were found effective with 97.63 percent, 97.37 percent, 96.72 percent inhibited the growth of the fungus. Other fungicides which were also found least effective to check the growth of fungus were Acrobat 92.47 percent, Taquat 62.36 percent, Matco 47.31 percent and Antrocol 36.39 percent. The rate of inhibition of fungal growth was very less in the case of Matco 47.31 percent and Antrocol 36.39 percent.

Conclusion

In present investigation efficacy of eight fungicides were tested *in vitro* against *R. Solani* which Vitavax

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S. No.	Fungicides	Dose%	Average diameter of fungal growth (mm)	Percent Inhibition
1.	Vitavax	0.2	0.00	100.00
2.	Roko	0.2	2.10	97.63
3.	Indofil Z-78	0.2	2.33	97.37
4.	Mancozeb	0.2	2.90	96.72
5.	Acrobat	0.2	6.66	92.47
6.	Taquat	0.2	33.33	62.36
7.	Matco	0.2	46.66	47.31
8.	Antrocol	0.2	56.33	36.39
9.	Control		88.56	
	CD 5%		1.7388	

Table 4. Inhibitory effect of fungicides on the growth of *Rhizoctonia solani in vitro* incubated at 27 ± 1 °C after 10 days

was found the effective to inhibit the growth of fungus completely. Among fungicides Roko, Indofil Z-78, Mancozeb and Acrobat were checked the growth of fungus effectively. Matco and Antrocol fungicide partially effective, which show all the fungicides proved better as compare to control.

In vitro efficacy of various fungicides against *Rhizocotonia solani* was reported by Chaudhary and Sharma (1988), Sharma and Tripathi (2001), Rai *et al.* (2007), Shah and Rehaman (2005), Meena and Chattopadhyay (2002) and Anjana Ray Kumar (2008) which is accordance with the present findings.

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