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Research Article

In Vivo Bio-efficacy and Phytotoxicity of Propiconazole 13.9 (15% w/v) + Difenoconazole 13.9 % w/w (15% w/v) 300 EC on Soybean against leaf spots and rust diseases

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Abstract

Soybean (*Glycine max.* L. Merril) belongs to family Leguminaceae is designated as miracle bean established its potential as an industrially vital and viable oilseed crop in many areas of India. leaf spots caused by *Alternaria*, *Cercospora* and *Helminthosporium* and rust of soybean has become a serious problem for successful cultivation of soybean. Therefore, a field experiment was carried out to know the efficacy of Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) against leaf spots and rust of soybean during 2017-18 and 2018-19, at College of Horticulture, Hiriyur. Experimental results revealed that all the treatments significantly reduced the leaf spots and rust disease severity over untreated control. Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL/lt is most effective in management of foliar diseases (leaf spots and rust) of soybean which is followed by the same fungicide @ 1 mL/lt when compared to the other treatments. Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 12 mL/lt was found to be optimum dosage for management of foliar diseases and harvest of maximum yield.

Keywords: Sunflower, nutrient management, soybean, rust diseases

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Introduction

Soybean [*Glycine max* (L.) Merr.] is the leading oilseed crop produced and consumed in the world. According to Hymowitz *et al* [1] as of 2013, soybean crop was grown in more than 70 countries with an annual production rate of 268 million metric tons (mmt). Top eight leading producers of soybean are United States (31 %), Brazil (31 %), Argentina (19 %), China (5 %), India (4 %), Paraguay (3 %) and Canada (2 %). As of December 2015, USDA projection of World Soybean Production 2015/2016 is 320.11 mmt it is an increase of 1.11 mmt or a 0.35% compared with 2014 [2].

The Asian soybean rust (*Phakopsora pachyrhizi* Syd) is the economically important disease not only in the Sub continent but also rest of the soybean growing regions of the world. *Phakopsora pachyrhizi* which cause rust of soybean has been known to drastically reduce yields in Asia. In areas where the pathogen occurs in most virulent form yield losses up to 80% have been reported. Initially, the pathogen was confined to eastern hemisphere before it had appeared in epiphytotic form in Hawaii region in 1994. At present the pathogen has been reported from different continents such as Africa, Asia, Australia, South America and Hawaii. The rapid spread of *Phakopsora pachyrhizi* and potential for severe yield losses makes this, the most destructive foliar disease of soybean. Soybean rust have a major impact on both total soybean production and production costs in the India. In India, the disease was first reported on soybean in 1951. Two *Phakopsora* species are known to cause soybean rust. The more aggressive species is *Phakopsora pachyrhizi*, which is also known as the Asian soybean rust. *P. meibomiae*, the less virulent species, has only been found in limited areas in the Western hemisphere of the world, and it is not known to cause severe yield losses in soybean, Hemachandra [3].

Most research on management strategies has been focused on the use of fungicides and host plant resistance. Some cultural practices have been recommended that minimize the incidence of rust. The recommendations were differed, but were based upon avoiding the conditions that promote disease development or were practices that optimized overall yields. The soybean growers of the subcontinent are seriously facing the severe infestation of rust disease in the last few years with a yield loss ranging from 30-100 %. Lack of resistant cultivars at present for Asian soybean rust and continuous application of fungicides has further aggravated the concerns regarding over usage of pesticides and pesticide resistance. Most of the new generation fungicides are highly specific and single site in mode of action. Thus, a novel fungicide with novel mode of action needs to be identified and evaluated under field conditions. Our objective was to determine the efficacy of different doses of newer generation fungicidal formulations of Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) to develop a management module for leaf spots (*Alternaria*, *Cercospora* and *Helminthosporium*) and rust of soybean.

Methodology

The experiment was laid out with randomized block design (RBD). The treatment fungicides were sprayed to the soybean plot at the beginning of the disease appearance. Spray schedule was repeated at 14 days interval. The observation of leaf spots and rust were recorded using 0-5 scale at before and after each spray. Observations are taken at 0 and 14 days after each application.

The per cent disease index (PDI) was calculated by the formula:

$$PDI= \frac{\text{Sum of all individual disease ratings}}{\text{Total number of leaves observed}} \times \frac{100}{\text{Max. grade in scale}}$$

Treatment details for Phytotoxicity studies

Sl. No	Treatment	g. a.i/ha	Formulation (mL/lt of water)
1	Untreated check		
2	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	180 (90 + 90)	1.2
3	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	360 (180 + 180)	2.4

Observations recorded

- Per cent Disease Index of leaf spot and rust.
- Phytotoxicity on foliage

The field experiment on bio efficacy and phytotoxicity of Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) against foliar diseases of soybean was carried out during 2017-2018 and 2018-19 at College of Horticulture, Hiriyur, UAHS, Shivamogga. The spray schedule was initiated soon after the disease appearance. The research results of 2017-2018 and 2018-19 revealed that the foliar diseases like leaf spot and rust were presented here under.

Experimental Results and Discussion

During the first spray during 2017-2018 there was not much significant differences among the treatments imposed with respect to the reduction of foliar diseases like leaf spots and rust. However the plot sprayed with Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL/lt has recorded lowest Per cent Disease Index (PDI) of leaf spot (6.88) which is on par with the Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1 mL/lt (7.20) and significantly superior over the rest of the treatments. Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL/lt and 1 ml/lt recorded 81.34 and 80.47 per cent disease reduction over check, respectively and were at par with each other (**Table 1**).

The less disease severity of rust 3.20 has been observed in treatment treated with Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL/lt which is on par with the Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1 mL/lt (3.36). Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL/lt and 1 ml/ha recorded 82.94 and 82.08 per cent disease reduction over check, respectively and were at par with each other. The treatment treated with Propiconazole 25 EC @ 1 mL/lt (4.48) Pyraclostrobin 133 g/l + Epoxiconazole 50 g/lit SE @ 1.5 ml/lt (5.40), Hexaconazole 5% EC @ 1 mL/lt (5.12) and untreated check (18.76) were inferior over the Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 and 1 mL/lt (**Table 2**).

During the first application during 2018-19 there was not much significant difference among the treatments imposed with respect to the reduction of foliar diseases like leaf spots and rust. However the plot sprayed with Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL/lt has recorded lowest terminal Per cent Disease Index (PDI) of leaf spot (6.12) which is on par with the Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1 mL/lt (6.80) and significantly superior over the rest of the treatments. Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL/lt and 1 mL/lt recorded 85.56 and 83.96 per cent disease reduction over check, respectively and were at par with each other (**Table 3**).

Table 1 Bio efficacy of Propiconazole 13.9 (15% w/v) + Difenoconazole 13.9 % w/w (15% w/v) (300 EC) against leaf spots of soybean (2017-18)

Sl. No	Treatment	Dosage mL/lt	PTO	15 DAA1	15 DAA2	% disease reduction
1	Untreated check	--	2.24	10.96	36.88	--
2	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	0.8	1.96	5.80	12.20	66.91
3	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	1	1.88	3.12	7.20	80.47
4	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	1.2	1.60	2.92	6.88	81.34
5	Propiconazole 25 EC	1	1.96	4.40	9.60	73.96
6	Difenoconazole 25% EC (Score 25 EC)	0.72	2.00	6.80	13.88	62.36
7	Pyraclostrobin 133 g/l + Epoxiconazole 50 g/l SE	1.5	1.60	4.48	9.44	74.40
8	Hexaconazole 5% EC	1	2.12	5.76	11.24	69.52
SEm±			NS	0.55	0.62	
CD (0.05%)			NS	1.67	1.87	
CV			NS	12.70	10.66	

* PTO- Pretreatment observation, DAA1- Days after 1st application, DAA2- Days after 2nd application

Table 2 Bio efficacy of Propiconazole 13.9 (15% w/v) + Difenoconazole 13.9 % w/w (15% w/v) (300 EC) against rust of soybean (2017-18)

Sl. No	Treatment	Dosage mL/lt	PTO	14 DAA1	14 DAA2	% disease reduction
1	Untreated check	--	0.48	7.88	18.76	--
2	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	0.8	0.52	2.12	5.88	68.65
3	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	1	0.24	1.46	3.36	82.08
4	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	1.2	0.20	1.12	3.20	82.94
5	Propiconazole 25 EC	1	0.12	2.40	4.48	76.11
6	Difenoconazole 25% EC (Score 25 EC)	0.72	0.40	3.50	9.92	47.12
7	Pyraclostrobin 133 g/l + Epoxiconazole 50 g/l SE	1.5	0.20	3.10	5.40	71.21
8	Hexaconazole 5% EC	1	0.44	2.60	5.12	72.70
SEm±			NS	0.20	0.12	
CD (0.05%)			NS	0.62	0.38	
CV			NS	16.08	4.17	

* PTO- Pretreatment observation, DAA1- Days after 1st application, DAA2- Days after 2nd application

Table 3 Bio efficacy of Propiconazole 13.9 (15% w/v) + Difenoconazole 13.9 % w/w (15% w/v) (300 EC) against leaf spots of soybean (2018-19)

Sl. No	Treatment	Dosage mL/lt	PTO	14 DAA1	14 DAA2	% disease reduction
1	Untreated check	--	1.24	14.66	42.40	--
2	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	0.8	1.33	6.36	14.36	66.13
3	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	1	1.22	2.84	6.80	83.96
4	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	1.2	1.46	2.48	6.12	85.56
5	Propiconazole 25 EC	1	1.26	3.90	10.24	75.84
6	Difenoconazole 25% EC (Score 25 EC)	0.72	1.36	7.76	15.75	62.85
7	Pyraclostrobin 133 g/l + Epoxiconazole 50 g/l SE	1.5	1.60	3.96	9.48	77.64
8	Hexaconazole 5% EC	1	1.82	5.12	12.20	71.22
SEm±		NS		0.12	0.24	
CD (0.05%)		NS		0.38	0.71	
CV		NS		4.70	3.72	

* PTO- Pretreatment observation, DAA1- Days after 1st application, DAA2- Days after 2nd application

The less terminal percent disease index of rust 4.50 has been observed in treatment treated with Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL/lt which is on par with the Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC)@ 1 mL/lt (4.80) and Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL/lt and 1 mL/lt recorded 81.85 and 80.64 per cent disease reduction over check, respectively and were at par with each other. Hexaconazole 5% EC @ 1 mL/lt (5.88) is at par with the Pyraclostrobin 133 g/l +

Epoxiconazole 50 g/lit SE @ 1.5 ml/lt (6.12), Propiconazole 25 EC @ 1 mL/lt (6.20) and Difenoconazole 25% EC @ 0.72 mL/lt (12.66) and inferior over the Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC @ 0.8 mL/lt (7.12) (**Table 4**).

Yield

During 2017-18 higher yield was recorded in the Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL/lt (14.88 q/ha) followed by Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1 mL/lt (14.12 q/ha) (**Table 5**).

Table 4 Bio efficacy of Propiconazole 13.9 (15% w/v) + Difenoconazole 13.9 % w/w (15% w/v) (300 EC) against rust of soybean (2018-19)

Sl. No	Treatment	Dosage mL/lt	PTO	14 DAA1	14 DAA2	% disease reduction
1	Untreated check	--	0.60	8.48	24.80	--
2	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	0.8	0.32	3.96	7.12	71.29
3	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	1	0.36	1.10	4.80	80.64
4	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	1.2	0.34	0.90	4.50	81.85
5	Propiconazole 25 EC	1	0.30	1.90	6.20	75.00
6	Difenoconazole 25% EC (Score 25 EC)	0.72	0.36	4.68	12.66	48.95
7	Pyraclostrobin 133 g/l + Epoxiconazole 50 g/l SE	1.5	0.46	3.60	6.12	75.32
8	Hexaconazole 5% EC	1	0.36	3.36	5.88	76.29
SEm±			NS	0.18	0.15	
CD (0.05%)			NS	0.54	0.47	
CV			NS	12.13	4.37	

* PTO- Pretreatment observation, DAA1- Days after 1st application, DAA2- Days after 2nd application

Table 5 Effect of Propiconazole 13.9 (15% w/v) + Difenoconazole 13.9 % w/w (15% w/v) (300 EC) on Yield of soybean (2017-18 and 2018-19)

Sl. No	Treatment	Dosage mL/lt	Yield (q/ha)	
			2017-18	2018-19
1	Untreated check	8.86	9.30	8.86
2	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	11.62	12.46	11.62
3	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	13.90	14.12	13.90
4	Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	14.36	14.88	14.36
5	Propiconazole 25 EC	12.88	13.68	12.88
6	Difenoconazole 25% EC (Score 25 EC)	9.92	10.72	9.92
7	Pyraclostrobin 133 g/l + Epoxiconazole 50 g/l SE	11.46	12.86	11.46
8	Hexaconazole 5% EC	12.48	13.24	12.48
SEm±			0.25	0.32
CD (0.05%)			0.76	0.97
CV			4.68	6.51

During 2018-19 higher yield was recorded in the Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL/lt (14.36 q/ha) followed by Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1 mL/lt (13.90 q/ha) (Table 5). The observation is on par with results agreed by Sunilkumar Shirasangi *et al*. [4], who reported maximum yield was recorded in Propiconazole (12.22 q/ha), followed by Difenoconazole (11.33 q/ha) and Hexaconazole (11.11q/ha). Least yield was recorded in control (9.11 q/ha), while the remaining treatments were at par with each other; these differences observed in the efficacy among tested fungicides depend on their fungicidal action. These fungicides showed considerable reduction in leaf spot and rust percent disease incidence. These results are in agreement with the reports of Hemachandra [3], who reported that four sprays of Propiconazole (0.1%) at seven days interval resulted lowest disease severity (20.5%) of pea rust caused by *U. fabae* with highest grain yield (1037.50 kg/ha) followed by Hexaconazole (0.1%) and the results are supported by the work of Ashwani *et al*. [5]. Efficiency of triazoles (difenoconazole, epoxiconazole, tebuconazole) and their combination with benzimidazoles (carbendazim-flutriafol and carbendazim-flusilazole) in pea rust management was also demonstrated [6, 7]). Triazoles are sterol synthesis inhibitors and many of them have good action against rust diseases [8]. In absence of accessibility of resistant varieties, fungicides application can be a suitable short term strategy for rust disease management in chickpea. Thus, new generation fungicides and/or combination of new and old generation fungicides provide a promising approach for the management of leaf spot and rust disease in soybean.

Phytotoxicity

There were no phytotoxicity symptoms like epinasty, hyponasty, vein clearing, yellowing, necrosis, leaf margin burning, rossetting and wilting were observed in different concentrations of Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) (**Table 6**).

Table 6 Impact of Propiconazole 13.9 (15% w/v) + Difenoconazole 13.9 % w/w (15% w/v) (300 EC) on soybean (mean of two sprays) (2017-18 and 2018-19)

Treat ments	Dose (g or mL/lt)	Score values on																																			
		0 DAA						1 DAA						3 DAA						5 DAA						7 DAA						10 DAA					
		A	B	C	D	E	F	A	B	C	D	E	F	A	B	C	D	F	A	B	C	D	E	F	A	B	C	D	E	F	A	B	C	D	E	F	
Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	1.2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC	2.4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
Untreated check	--	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	

A: Leaf injury on tips and leaf surface; B: Wilting; C: Leaf vein clearing; D: Necrosis; E: Epinasty; F: Hyponasty; DAA: Days after Application.

Conclusions

Based on the experimental results it has been found that, Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL/lt is most effective in management of foliar diseases (leaf spots and rust) of soybean which is followed by the same fungicide @ 1 mL/lt when compared to the other treatments. Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL/lt was found to be optimum dosage for management of foliar diseases and harvest of maximum yield. Propiconazole 13.9 + Difenoconazole 13.9 % w/w EC (300 EC) @ 1.2 mL as well as 2.4 mL/lt was found to be non-phytotoxic and was safe to soybean crop.

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