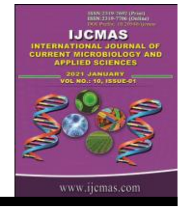
Int.J.Curr.Microbiol.App.Sci (2021) *10*(01): 420-425

International Journal of Current Microbiology and Applied Sciences
***ISSN: 2319-7706* Volume 10 Number 01 (2021)**
Journal homepage: http://www.ijcmas.com



Original Research Article https://doi.org/10.20546/ijcmas.2021.1001.051

Assessment of Biocontrol Agents and Botanicals against White Rot of Onion Caused by *Sclerotium rolfsii* under Field Condition in Manipur

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A B S T R A C T

Keywords

Management, White rot, Onion, Disease intensity, Yield

Article Info

Accepted:
04 December 2020
Available Online:
10 January 2021

Field trial was carried out to investigate the efficacy of *Trichoderma harzianum, Tricoderma viride,* garlic (*Allium sativum* L.), turmeric (*Cucurma longa* L.) and sweet flag (*Acorus calamus* L.) in comparison to efficacy of carbendazim (50 WP) for the management of white rot of onion (*Allium cepa* L.) caused by *Sclerotium rolfsii* in Manipur. All the treatments were found to reduce percent disease incidence. Carbendazim was found to be most effective resulting in lowest disease incidence of 10.51% and recorded highest disease control of 79.41%. Highest bulb yield (19.12 t/ha) was also recorded from carbendazim treatment. *Trichoderma harzianum* was next to carbendazim recording second lowest disease incidence of 15.50% and second highest disease control of 69.60%, giving a bulb yield of 17.79t/ha.

Introduction

Onion (*Allium cepa* L.) is one of the important bulbous vegetable crops under the family *Amaryllidaceae*. It is used not only as vegetable and spice but also as medicinal plants. The center of origin of onion is Central Asia (Vavilov, 1951). Onion is rich in antioxidant enzymes including superoxide dismutase, catalase, peroxidise and glutathione peroxidase (Stajner and Varga, 2003). It also contains antimicrobial phenolic compounds such as procatechuic acid, catechol, *p*-coumaric, ferulic acid, quercetin and kaemferol (Santas *et al.,* 2010). Onion is rich in energy value and mineral composition having high amount of moisture (82.77%), carbohydrate (14.772%), total sugar (2.32%), vitamin C (5.7mg), calcium (25.7mg), phosphorus (30.3mg), potassium (129mg) having potential to impart numerous health benefits (Bhattacharjee *et al.,* 2013).

There are several diseases which decreases onion productivity in Manipur. White rot of onion caused by *Sclerotium rolfsii* is one of the important diseases which decrease onion productivity to a considerable extent. Disease development of white rot increases as the root system develops and the mycelial growth spreads upwards from the roots to the stem plate, the bulb, and onto the leaves aboveground (Amin *et al.,* 2014). Infected bulb is characterised by the presence of white fluffy mycelium and after the bulb is infected, the leaves starts yellowing from the tips downwards followed subsequently by wilting and death of the aerial parts (Walker, 1924).

The pathogen produces abundant sclerotia which can survive in a wide range of soil pH and temperatures (Kator *et al.,* 2015). Sclerotia also remain viable in the soil for more than a year (Marcuzzo and Schuller, 2014). Due to the long viability period of sclerotia, crop rotation as well as soil solarisation does not come under effective management practices (Entwistle, 1990). Moreover, *Sclerotium rolfsii* also have wide host range (El-Nagar *et al.,* 2013). Therefore, crop rotation is not efficient enough in managing the disease. Removal and destruction of affected plants is also not practical and efficient in field conditions (Towsend and Willets, 1954). The use of fungicides for managing white rot is also not convenient due to degradation of environment, non target effects on unrelated organisms, development of resistance in pathogens, residue problem and health hazards. Therefore, the use of eco-friendly biocontrol agents and botanicals are must in managing white rot of onion. Thus, the present study was conducted to assess the efficacy of biocontrol agents namely, *Trichoderma harzianum* and *Trichoderma viride* and botanicals namely, garlic (*Allium sativum* L.), turmeric (*Cucurma longa* L.) and sweet flag (*Acorus calamus* L.) which are already known for its anti-fungal properties in management of white rot of onion caused by *Sclerotium rolfsii* in Manipur as it was essential to confirm the efficacy of the above mentioned biocontrol agents and botanicals under natural environmental field condition.

Materials and Methods

The research study was conducted at the Department of Plant Pathology, College of Agriculture, Central Agricultural University, Imphal during 2015-2016.

Collection and maintenance of biocontrol agents

Biocontrol agents namely, *Trichoderma harzianum* and *T. viride* were collected from the Department of Plant Pathology, College of Agriculture, Imphal. Both potato dextrose agar (PDA) and potato dextrose broth (PDB) were prepared and sterilized at 121°C at 15lbs for 20 minutes. PDA was poured into sterilized petriplates. 5mm disc of both the biocontrol agents were then inoculated and maintain on both PDA and PDB throughout the research period.

Preparation of biocontrol agents formulation

5mm disc of both *Trichoderma* species were inoculated on sterilized 100 ml of potato dextrose broth (PDB) and were incubated at 28 ± 1^0C for 15 days. Mycelial filtrates were harvested by filtering the broth through Whatman No.42 filter paper and were allowed to dry for 24 hours. The weight of mycelial filtrates was recorded. Mycelial filtrates were then grinded. Talc powder was mixed with 5% carboxy methyl cellulase (CMC) of its weight. This mixture was then blended with the grounded mycelial filtrates of both *Trichoderma* species at the ratio of 1:2 (Jeyarajan *et al.,* 1994).

Preparation of plant extracts

Locally available botanicals namely, clove of garlic (*Allium sativum* L.) and rhizomes of both turmeric (*Cucurma longa* L.) and sweet flag (*Acorus calamus* L.) were collected and washed thoroughly. The plant parts to be used were grinded independently with sterile water at the ratio of 1:1 (w/v) and the plant extracts were filtered through muslin cloth. The resultant plant extracts were considered as 100% concentration.

Evaluation of biocontrol agents and plant extracts against white rot of onion

The biocontrol agents and aqueous plant extracts were evaluated under field conditions in comparison with a fungicide namely, carbendazim (50WP). The experiment was laid out during rabi season in Randomized Block Design. Each treatment was replicated thrice. Nasik Red N-53 variety was used for the research study. Seeds were sown in nursery beds. Seedlings were uprooted 45 days after sowing and were transplanted in a plot size measuring 2m x 1.5m with spacing of 10m x 10 m. A single plot accommodates 266 plants. Seedlings dipping and sprayings were adopted for evaluation. For seedlings dipping, onion seedlings were dipped in suspension of each treatment of both *Trichoderma* species (10g/lit), garlic (3ml/lit), turmeric (10ml/lit), sweet flag (10ml/lit), and carbendazim (1g/lit) for 20 minutes. For plots that serve as control, seedlings were transplanted without dipping. Spraying of both *Trichoderma* species (10g/lit), garlic (3ml/lit), turmeric(10ml/lit), sweet flag (10ml/lit), and carbendazim (1g/lit) at the base of the plants were given one month after transplanting followed by two subsequent sprayings at 30 days interval. For plots that serve as control, spraying was not given. Percent disease intensity was recorded for each treatment at April during harvesting since the initial infection are constricted to the underground plant parts and also both the aerial and undergrounds plant parts are affected.

Percent disease intensity was calculated by using the following formula.

Percent disease incidence =

$$\frac{\text{Total number of diseased plants}}{\text{Total number of healthy plants}} \text{ X } 100$$

Percent disease control was calculated by using the following formula.

$$\text{Percent disease control} = \frac{C-T}{C} \text{ X } 100$$

where, C= Disease incidence in control
T= Disease incidence in treatment

Bulb yield was also recorded for each treatment and converted to tonne per hectare.

Percent increase in bulb yield over control was calculated by using the formula.

$$\text{Percent increase in bulb yield} = \frac{T-C}{T} \text{ X } 100$$

where, T= Bulb yield in treatment
C= Bulb yield in control

Results and Discussion

All the treatments reduced percent disease incidence compared to control. Treatment with carbendazim recorded lowest disease incidence of 10.51%. Among the two biocontrol agents, *Trichoderma harzianum* was found to be more effective than *T. viride* recording second lowest disease incidence of 15.50%. *T. viride* gave the disease incidence of 19.50%. Among the plant extracts, garlic was found to be most effective resulting in disease incidence of 20.00%, followed successively by turmeric and sweet flag which recorded the disease incidence of 26.00% and 28.00% respectively. Treatment with carbendazim gave highest disease control of 79.41%, followed successively by *T. harzianum* which gave second highest disease control of 69.60%. Application of garlic gave a disease control of 60.78%. Treatment with carbendazim gave highest bulb yield of 19.12 t/ha. Treatment with *T. harzianum and T. viride* gave a bulb yield of 17.79 t/ha and 17.56 t/ha respectively. Among the plant extracts, garlic gave more bulb yield of 17.49 t/ha compared turmeric and sweet flag which gave a bulb yield of 16.66 t/ha and 15.89 t/ha respectively. Highest disease incidence of 51.00% and lowest bulb yield of 13.04% was recorded in control (Table 1 and 2).

Table.1 Effect of biocontrol agents, plant extracts and carbendazim on disease incidence of white rot of onion under field condition

Sl. No.	Treatment (Seedlings dipping + sprayings)	Percent disease incidence*	Percent disease control
1	*Trichoderma harzianum*	15.50	69.60
2	*Trichoderma viride*	19.50	61.76
3	Garlic	20.00	60.78
4	Turmeric	26.00	49.01
5	Sweet flag	28.00	45.09
6	Carbendazim	10.50	79.41
7	Control	51.00	-
	S.E. (d)	0.60	-
	CD (5%)	1.32	-

*Mean of three replications

Table.2 Effect of biocontrol agents, plant extracts and carbendazim on bulb yield of onion

Sl. No.	Treatment (Seedlings dipping + sprayings)	Bulb yield (t/ha)*	Percent increase in bulb yield over control
1	*Trichoderma harzianum*	17.79	26.70
2	*Trichoderma viride*	17.56	25.74
3	Garlic	17.49	25.44
4	Turmeric	16.66	21.72
5	Sweet flag	15.89	17.93
6	Carbendazim	19.12	31.79
7	Control	13.04	-
	S.E. (d)	0.39	-
	CD (5%)	0.85	-

*Mean of three replications

The present findings are in agreement with findings of Vanitha and Suresh (2002) who reported that seed treatment with carbendazim resulted in lowest disease incidence (10.83%) of collar rot of brinjal caused by *Sclerotium rolfsii* compared to control (39.30%). The results also corroborates to the findings of Siddique *et al.,* (2016) who reported that spraying of carbendazim at the base of plants gave lowest disease incidence (7.10%) of foot and root rot disease of eggplant caused by *S. rolfsii* and also gave highest yield (18.07 t/ha). He also stated that *Trichoderma harzianum* showed promising effect against brinjal foot and root rot disease recording a disease incidence of 31.93% and produced 11.58 t/ha compared to control which recorded a highest disease incidence of 64.90% and lowest yield of 4.86 t/ha of Elad *et al.,* (1980) reported that *T. harzianum* was an effective biocontrol agent for protecting a number of crop plants from damage by *S. Rolfsii*. Jegathambigai *et al.*, (2010) stated that *T. viride* Tv1 isolate greatly reduced the disease incidence of collar rot of *Zamioculcas zamiifolia* by 75.54%. Chet *et al* (2007) also reported that *T. viride* can be used as biocontrol agent reducing the effect of pathogens on the plant, increased the resistance in plant and also stimulate plant growth by enhancing uptake of water in plant. The present findings are also in accordance with the findings of Gupta *et al.*, (2012) who reported that garlic was effective in managing collar rot of chickpea caused by *S. rolfsii* giving a good disease control of 76.6%. Similarly, Wongkaew and Sinsiri (2014) reported that crude extract of turmeric rhizomes was effective against *S. Rolfsii* and other fungal plant pathogens and also stated that turmeric extract provided an alternative regime for the control of the fungal plant diseases and a promising appreciable choice for a replacement of chemical fungicides. Subedi *et al.*, (2015) observed disease index of 35.33% and disease control of 28.69% of *Stemphylium* blight of lentil in plots treated with sweet flag (8% w/v). He also reported that crop yield of 987.39 kg/ha was obtained from plots treated with sweet flag.

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How to cite this article:

Rimamay Konjengbam, Rajkumari Tombisana Devi and Naorem Iboton Singh. 2021. Assessment of Biocontrol Agents and Botanicals against White Rot of Onion Caused by *Sclerotium rolfsii* under Field Condition in Manipur. *Int.J.Curr.Microbiol.App.Sci.* 10(01): 420-425. doi: https://doi.org/10.20546/ijcmas.2021.1001.051