

In Vitro Evaluation of Different Fungicides against Collar Rot Caused By *Sclerotium Rolfsii* in Yard Long Bean (*Vigna Ungiculata*)

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Abstract

Yard long bean (*Vigna unguiculata*) is one of the most essential and widely cultivated common vegetable crops grown in Sri Lanka. Damping off of seedlings threatens yard long bean production by killing a large number of seedlings and as the infection is usually not lethal it reduces the plant growth and the quality and quantity of the yield. This is a common issue in yard long bean nurseries which leads to patches in nursery beds and the affected plants are generally found in scattered areas. It has the potential to cause severe loss in yard long bean production. This disease is commonly caused by *Sclerotium rolfsii*, and is widely distributed throughout the world. This study will be undertaken to investigate the efficacy of six recommended fungicides and one fungicide which is not recommended i.e. Captan 80% WDG for controlling collar rot. The experiment was arranged in a Completely Randomized Design (CRD) with eight treatments randomized in five replicates. These fungicides were evaluated at recommended concentrations on the growth of *Sclerotium rolfsii* in Potato dextrose agar (PDA) medium using poisoned food technique, in vitro. The result shows that the effect of Mancozeb 75% WP, Captan 50% WP, Captan 80%

WDG and Homai WP has been highly effective in suppressing radial expansion as well as percent inhibition of the fungus at recommended concentrations compared with Thiram 80% WP. Carbendazim 50% WP and Topsin 70%WP did not control the disease. Based on study findings, we can recommend Captan 80% WDG and assess the proper fungicide concentrations to reduce probable toxicities due to excess application and reduce the wastage of money in order to improve profit.

Keywords: Yard long bean, Collar rot, *Sclerotium rolfsii*, Fungicides.

Introduction

Yard long bean (*Vigna unguiculata*) belongs to the family fabaceae is a widely cultivated vegetable crop throughout the subtropical and tropical countries including Sri Lanka. Immature yard long bean pods are one of the very low calorie vegetables containing large quantities of fiber, protein, vitamin C & A. They also contain minerals such as iron, copper, manganese, calcium and magnesium [1].

Collar rot disease caused by *Sclerotium rolfsii* is a serious threat to yard long bean that may cause serious crop loss at

seedling stage under favorable environmental conditions (Personal communication with extension officers). Diseases caused due to *S. rolfsii* requires warm climates, occurs more frequently at high moistures and high temperatures. *S. rolfsii* control has met with very limited success. This may be due to the prolific growth, extensive host range of the pathogen and having the ability to produce large number of sclerotia that may persist in the soil for several years [2]. Collar rot is initially manifested in the collar region of the plants right from the seedling stage. It begins as brownish – black lesions at soil level near collar region girdling the base of the stem resulting in yellowing and drooping of leaves and rotting of roots. White mycelial growth often studded with small sclerotia is characteristically seen on the affected regions [3]. The fungus spreads by mycelial contact with healthy plants and as sclerotia in soil. In order to regulate this damage from early stage of the cropping season usage of fungicides is one of the methods.

Unfortunately, very limited research has been carried out regarding the bio efficacy testing of fungicide against of yard long bean collar rot in Sri Lanka. Therefore, this research is designed to evaluate the in vitro efficacy of different fungicides against collar rot, caused by *Sclerotium rolfsii*, of yard long bean.

Materials and Methods

The test pathogen *sclerotium rolfsii* was isolate from the infected yard long bean (variety Sena) plants having white luxurious mycelial growth and abundant sclerotia bodies of the fungus at the collar region and collected from the research field at HORDI, Gannoruwa during the crop season of 2020/21 *Maha*. The specimens were brought to the laboratory

and critically examined and studied for the symptoms of the disease and isolation of the pathogen. The part of collar region showing typical symptoms of the disease was cut into small pieces of 1-2mm size including the advancing margins of infection with the use of sterile scalpel. The segments were surface disinfected in 70% ethanol solution for 2 min and rinsed in three changes of sterile water. The segments were separately dried in between sheets of sterile filter paper and plated (3 pieces per plate) on fresh potato dextrose agar (PDA) medium impregnated with streptomycin, and culture on autoclaved culture medium in a laminar flow cabinet. Culture plates were incubated for 7 days at 28°C. Culturing tools were sterilized at 160°C for two hours. Pure culture was obtained by sub-culturing three times. Pure cultures of the final isolates were maintained on PDA slants in McCartney bottles and kept in the refrigerator until required. Koch's postulates were proved by artificial inoculation of yard long bean plants with the pathogen.

The efficacy of non-systemic fungicides and systemic fungicides against *sclerotium rolfsii* was assessed by poisoned food technique [4] and using potato dextrose agar (PDA) as basal culture media. The required quantities of each fungicide were calculated and thoroughly mixed with autoclaved and cooled (40-45°C) PDA in conical flasks separately to obtain desired concentrations. The flask containing the poisoned medium was well shaken to facilitate a uniform mixture of fungicides and 20ml of the poisoned medium was poured into 9cm diameter sterile Petri plates and keep for solidifying. The mycelial disc of 5 mm diameter of seven days old culture of *Sclerotium rolfsii* was cut with the help of a sterile cork borer. Each disc was transferred aseptically to the center of each Petri plate. The PDA plates

without fungicide were also inoculated and maintained as control and incubated at 28 ± 2 °C. Five replications per treatment were maintained. The observation on colony growth and sclerotia formation were recorded periodically until the Petri plate of control treatment was fully covered with mycelial growth [5].

T = Growth of mycelium in treatment (cm).

Data Analysis

The data were analyzed by using the Analysis of Variance (ANOVA) procedure of statistical analysis system (SAS) 9.1. Duncan's Multiple Range Test (DMRT) was performed to compare the differences among treatment means at $p=0.05$.

Table i. Different Fungicides applied to the treatments

Treatment number	Fungicides	Mode of action	Formulation	Dosage
T1	Thiophanate methyl 50% WP + Thiram 30% WP (Homai)	Systemic	80% WP	14g/10L
T2	Thiram	Contact	80% WP	14g/10L
T3	Mancozeb	Contact	75% WG	20g/10L
T4	Captan (New formulation)	Contact	80% WDG	7.5g/10L
T5	Captan	Contact	50% WP	12g/10L
T6	Carbendazim	Systemic	50% WP	12g/10L
T7	Control			

Results and Discussion

Control rot of yard long bean caused by *Sclerotium rolfsii* a common disease in Sri Lanka. It causes seedling death at early stage resulting in very poor plant stand which ultimately produces very low yield. The fungus can attack the crop during any time from seedling to flowering stage. In vitro evaluation of fungicides provides useful preliminary information regarding its efficacy against a pathogen within the shortest period of time and therefor, serve as guide for further field testing. In the present study six fungicides including both systemic and non-systemic fungicides were tested.

The results presented in Table ii and it revealed that the treatment of fungicides, homai WP, mancozeb 75% WG, captan 80% WDG and captan 50% WP treatments were not significantly different. Thiram 80% WP was less inhibitory than either homai WP, mancozeb 75% WG, captan 80% WDG and captan 50% WP treatment. Among the six fungicides tested, minimum percent inhibition of growth *Sclerotium rolfsii* was observed in Carbendazim 50% WG treated plate.

Data Collection

The efficiency of various fungicides was expressed as percent inhibition of mycelia growth over control that was calculated by using the formula given by Vincent [6]. The percent values were converted to log transformations, the data were analyzed statistically.

$$I = \frac{C - T}{C} \times 100$$

Where,

I = Per cent inhibition

C= Growth of mycelium in control (cm).

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Followed by homai WP (97.32%), mancozeb 75% WG (84.88%), captan 80% WDG (99.10%), captan 50% WP (97.77%), Thiram 80% WP (98.21%) and Carbendazim 50% WG (0.00%) showed mean colony diameter 0.24 cm, 1.36 cm, 0.08 cm, 0.20 cm, 0.16 cm and 9.00 cm, respectively. From the results it was concluded that, the fungicides homai WP,

mancozeb 75% WG, captan 80% WDG and captan 50% were effective for inhibiting the growth of *Sclerotium rolfsii* as compared to other fungicides tested.

Table ii. Effect of fungicides on the percentage reduction in growth of *S.rolsfii*.

Treatment	Radial growth in (cm) *
T1- Homai WP	97.32 (1.99) ^a
T2- Thiram 80% WP	84.88 (1.93) ^b
T3- Mancozeb 75% WG	99.10 (2.00) ^a
T4- Captan 80% WDG	97.77 (1.99) ^a
T5- Captan 50% WP	98.21 (1.99) ^a
T6- Carbendazim 50% WP	0.00 (0.00) ^c
T7- Control	0.00 (0.00) ^c
CV %(Coefficient of variation)	3.2

*Figures in parentheses are log transformed values.

Different superscript letters indicate significant differences among treatments, according to the least significant difference (P =0.05)

Table iii. Efficacy of fungicides against the *S.rolsfii* disease development.

Treatment	Radial growth in cm *
T1- Homai WP	0.24 ^c (0.08) ^c
T2- Thiram 80% WP	1.36 ^b (0.36) ^b
T3- Mancozeb 75% WG	0.08 ^c (0.03) ^c
T4- Captan 80% WDG	0.20 ^c (0.07) ^c
T5- Captan 50% WP	0.16 ^c (0.07) ^c
T6- Carbendazim 50% WP	9.00 ^a (1.00) ^a
T7- Control	9.00 ^a (1.00) ^a
CV %(Coefficient of variation)	6.8

* Means of five replications.

For each column, means followed by the same letter are not significantly different according to Duncan's multiple range test ($P \leq 0.05$).

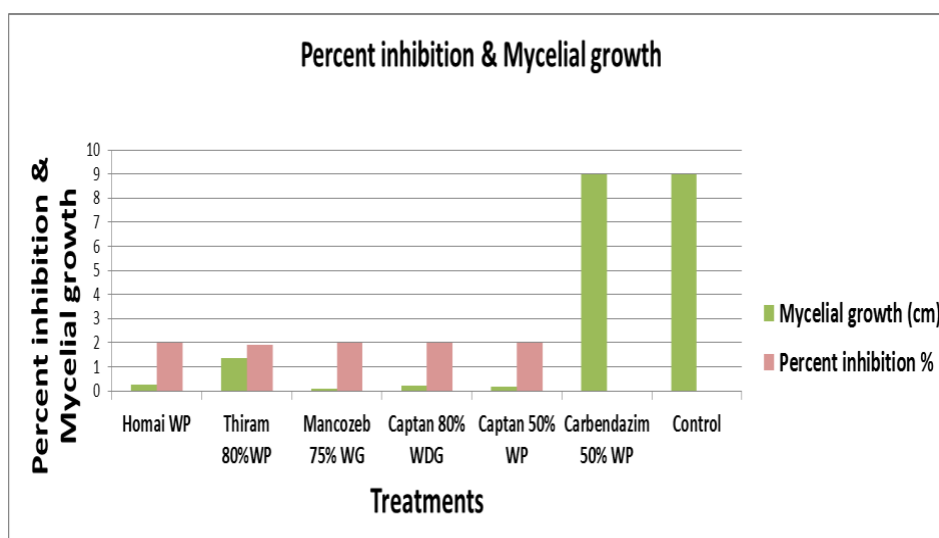


Figure i. Efficacy of different fungicides against *S. rolsfii*



Figure ii. Growth of *S. rolsfii* isolate on PDA amended with fungicides.

Conclusion

Collar rot is a worldwide disease of economic importance in vegetables.

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Different chemical fungicides are commercially available in market to control this disease. Chemical control is one of the indispensable approaches of integrated disease management. However, there is lack of effective chemicals with suitable concentration against *Sclerotium rolsfii* causing collar rot disease. Therefore, in this study one new fungicide formulation and recommended fungicides has been tested under in vitro condition. Major causal agent of yard long bean collar rot disease in Kandy district is *Sclerotium rolsfii*. The in vitro efficacy of fungicides has shown that Mancozeb 75% WG, Captan 80% WDG, Captan 50% WP and Homai WP are effective against *Sclerotium rolsfii* pathogen and it is recommended to be used in the treatment of collar rot disease.

Therefore, present studies reveal that some of the new chemicals i.e. Captan 80% WDG for effective in controlling *Sclerotium rolsfii* causing collar rot disease. However, further evaluations of effective fungicides are needed in this field for better recommendation for the management of collar rot disease.

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