Ecofriendly management of Rhizoctonia leaf blight of Amaranthus
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ABSTRACT

The present study was undertaken to assess the effect of biocontrol agents and indigenous organic formulations on disease incidence, disease severity of Rhizoctonia leaf blight and yield of Amaranthus. In in vitro studies, Trichoderma harzianum has completely overgrown the Rhizoctonia solani with maximum inhibition of 49.56% compared to Pseudomonas fluorescens with the inhibition of 28.30%. In indigenous organic formulations, turmeric powder and baking soda combination inhibited the maximum growth of the pathogen by 64.40%. Under field studies on disease suppression and plant yield promotion, foliar spray of Pseudomonas fluorescens (2%) recorded the lowest disease index of 51.66% with maximum yield of 22875.00 kg/ha.

Keywords: Rhizoctonia solani, Rhizoctonia leaf blight, Disease severity, Yield

I. INTRODUCTION

Amaranthus is the most important leafy vegetable consumed and cultivated in Southern India. Amaranthus (Amaranthus tricolor L.), widely known as ‘poor man’s spinach’ is one of the cheapest, most accepted and commercially cultivated leafy vegetable in Kerala. Among the different diseases affecting amaranth, leaf blight disease, caused by Rhizoctonia solani Kuhn, is the most devastating, especially during the monsoon period. The disease is characterised by light cream coloured spots on the foliage which rapidly spread causing extensive damage leading to economic losses. On the under surface of the infected leaves, white powdery masses of basidiospores of the teleomorph of the causative fungus Thanatephorus cucumeris (Frank) Donk are clearly visible[17] (Nayar et al., 1996). Susceptibility of popular cultivars and humid conditions in Kerala make the disease a serious constraint in amaranth cultivation. The pathogen infects more than 90% of plants in the field and causes considerable economic loss owing to reduced marketability of the produce. Although chemical control of the disease through the use of fungicides can lessen the severity of this aerial blight disease (Gokulapalan et al. 1999), application of chemicals on a regular basis causes serious health hazards.

Rhizoctonia solani Kühn (teleomorph: Thanatephorus cucumeris [A.B. Frank) Donk.]) is a soilborne fungus that causes disease on many economically important crop plants worldwide. The pathogen overwinters as soil-borne sclerotia and mycelium in plant debris these constitute the primary inoculum. Control of the pathogen is difficult because of its ecological behavior, extreme broad host range and the high survival rate of sclerotia under various environmental conditions (Anderson 1982 and Ogoshi, A. 1987).

In order to avoid pesticide residues in agricultural products, alternative methods for the management of pests and diseases using non-hazardous, eco-friendly agents have to be explored. Among biocontrol agents, Trichoderma harzianum and Pseudomonas fluorescens are more reliable and ecologically, as well as economically, sustainable. Plant derivatives possessing pesticidal properties are gaining worldwide importance as indigenous organic formulations. Recognizing the potentiality of organic agriculture and the importance of leaf blight disease of amaranth in Kerala, in vitro and field experiment was carried out to study the the effect of biocontrol agents and
indigenous organic formulations on disease incidence, yield and disease severity of Rhizoctonia leaf blight of Amaranthus.

II. MATERIALS AND METHODS

A. In vitro evaluation of biocontrol agents and indigenous organic formulations against Rhizoctonia solani

Isolate of the pathogen causing Rhizoctonia leaf blight in amaranthus was collected from instructional farm, College of Agriculture, Vellayani. The pathogen brought under pure culture after isolation and Koch’s postulate were proved for the confirmation of casual agent and was used for the further in vitro studies. Modified dual plate method by Skidmore and Dickinson (1976) was followed to test the effect of these antagonists on R. solani. Five mm diameter agar blocks of seven day old actively growing mycelial growth of R. solani and the KAU isolate of Trichoderma harzianum placed five cm apart on PDA in a petri dish and incubated at room temperature (25±2°C) with three replications. The bacterial antagonist tested for antagonism to R. solani by dual culture technique (Utkhede and Rahe, 1983). The PDA medium was melted and poured into sterile petri plates. After solidification, The KAU isolate of Pseudomonas fluorescens was streaked 2.5 cm away on the both sides and then culture bits of 5 mm size of the pathogen was placed at the centre of each petri plate. Plates inoculated with R. solani alone served as control. Colony diameter of the fungus was measured and percentage inhibition was calculated using the formula,

\[
\text{Percentage Inhibition} = \left(1 - \frac{T}{C}\right) \times 100
\]

Where, \(C\) = colony diameter (cm) of the control
\(T\) = colony diameter (cm) of the test plate.

Fish amino acid was tested at 2.5%, 5% and 10% concentrations under in vitro study after passing through bacterial filters. As per the quantities mentioned below, three ratios of turmeric powder- baking soda combination were added to 200 ml PDA taken in 250 ml conical flasks before filtering through a bacterial filter.

<table>
<thead>
<tr>
<th>Ratios of turmeric powder- baking soda</th>
<th>Quantity of turmeric-powder in 200 ml PDA</th>
<th>Quantity of baking soda in 200 ml PDA</th>
</tr>
</thead>
<tbody>
<tr>
<td>6:4</td>
<td>0.480g</td>
<td>0.320g</td>
</tr>
<tr>
<td>8:2</td>
<td>0.640g</td>
<td>0.160g</td>
</tr>
<tr>
<td>10:1</td>
<td>0.727g</td>
<td>0.073g</td>
</tr>
</tbody>
</table>

Media containing the different concentrations of turmeric powder and baking soda combination and fish amino acid were poured into sterile Petri dishes after passing through bacterial filters. From the edges of seven day old mycelial growth of R. solani five mm discs were cut out with a cork borer and then placed in the centre of each petri plate with three replications for each concentration. Plates containing PDA with R. solani inoculated at the centre served as control. Observation on the mycelial growth of the pathogen was recorded.

B. Field studies on disease suppression and yield improvement

The field study has been conducted at Coconut Research Station, Balaramapuram as an intercrop in 53 year old coconut plantation. The experimental site was incorporated with required quantity of organic manure in soil. Fertilizer application and other cultural operations have been done as per Package of Practices Recommendations of crops, Kerala Agricultural University and crop was irrigated by using hose pipe. The details of experiment are as follows; Design - RBD.
Treatments – 5, Replication - 4, Variety - Arun, Plot size - 2 x 2 m. Treatments: T1-Pseudomonas fluorescens talc formulated product of KAU @ 2%, T2-Trichoderma harzianum talc formulated product of KAU @ 2%, T3- Fish aminoacid – 5%, T4-Turmeric powder-baking soda combination 10:1 and T5-Absolute control. The seeds were broadcasted in nursery beds, 21 days old seedlings were transplanted into each plot by maintaining population of 100 plants per plot.

After transplanting, number of days taken for the first appearance of the disease in each plot was observed and recorded and also per cent incidence of disease under natural epiphytotic condition was recorded as number of plants diseased/ total number of plants assessed*100. Disease severity from each treatment was rated at 10 days interval on ten randomly selected leaves (three from top, four from middle and three from bottom) from each of ten randomly selected plants. Each leaf was scored using a 0-9 scale (KAU, 1996), where 0=no disease; 1=1 to 10% infected leaf area; 3=10 to 25% infected leaf area; 5= >25 to 50% infected leaf area; 7= >50-75% infected leaf area; 9= > 75% leaf area infected. In addition, % disease severity or Percentage Disease Index (PDI) was calculated using the formula suggested by Wheeler (1969),

\[
(PDI) = \frac{\text{Sum of grades of each leaf}}{\text{Number of leaves assessed}} \times \frac{100}{\text{Maximum grade used or 9}}
\]

The fresh yield of each plot was recorded after the harvest in terms of kg/ha.

Data were analyzed by using WASP 2.0 program. The significant difference, if any, among the means were compared by Duncan's Multiple Range Test (DMRT)

III. RESULTS AND DISCUSSION

In the present in vitro studies, Trichoderma harzianum has completely overgrown the pathogen with maximum inhibition of 49.56% compared to Pseudomonas fluorescens with the inhibition of 28.30%. In vitro studies of Monga (1993) and Naeimi et al. (2010) Showed that T. harzianum was excellent bicontrol agent against R. solani. Gandhi and Kumar (2006) reported that T. harzianum significantly inhibited the growth of R. solani upto 60% under in vitro conditions.

Antagonism of Pseudomonas fluorescens was tested against R. solani. P. fluorescens showed in vitro antagonism of 28.3%. Dantre et al., (2003) Reported that fifteen strains of P. fluorescens shown to inhibit the mycelial growth of R. solani under in vitro condition. Effectiveness of P. fluorescens is related with inherent quality to produce antibiotics, hydrogen cyanide and siderophores, which are involved in suppression of plant root pathogens (Kloeper et al, 1980; O'Sullivan and O'Gara, 1992) and their ability to compete with indigenous microflora, may explain their ability to colonize the rhizosphere (Weller. 1988; Mazzola and Cook, 1991).

In indigenous organic formulations, turmeric powder and baking soda combination inhibited the maximum growth of the pathogen by 64.40%. Turmeric powder and baking soda combination inhibited the maximum growth of the pathogen by 64.40% and fish amino acid recorded less suppression of the pathogen by 29.00%. According to Dhanya et al., (2000), three levels of turmeric powder – baking soda 0.05, 0.10 and 0.15 per cent were tested against Xanthomonas axonopodis pv dieffenbachiae causing bacterial blight of anthurium and inhibition to a lesser extent was observed.

Field studies on disease suppression and plant growth promotion was studied. The number of days taken for first symptom appearance of Rhizoctonia leaf blight in amaranthus plots ranged from 13 to 14 days after transplanting. Foliar application of Trichoderma harzianum (2%) recorded the minimum disease incidence of 41.92% where as in indigenous organic formulations foliar spray of fish amino acid (5%) recorded the minimum disease incidence of 43.23%. According to Chet et al.,(1982) use of T.harzianum in solarized field infested with R. solani has been shown to improve disease control while delaying the buildup of inoculum. Trichoderma could have a stimulatory effect on plant growth as a result of modification of soil conditions (Naseby et al., 2000). Das and Hazarika
(2000) claimed that *T. harzianum* was found to be more effective than *T. viride* in reducing sheath infection of rice caused by *R. solani* and increase in yield. Matloob et al., (2013) concluded that the biocontrol agent *T. harzianum* decreased root rot disease incidence and increased plant resistance against infection with *R. solani* and improve plant growth and yield.

**Table 1. Effect of biocontrol agents and indigenous organic formulations on appearance of disease, disease incidence and disease severity of amaranthus.**

<table>
<thead>
<tr>
<th>Trt No.</th>
<th>Treatments</th>
<th>FAD</th>
<th>Percent disease incidence</th>
<th>Per cent disease index</th>
<th>PDS</th>
<th>Yield (kg/ha)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>30DAT</td>
<td>40 DAT</td>
<td>50 DAT</td>
<td>60 DAT</td>
</tr>
<tr>
<td>1</td>
<td><em>Pseudomonas fluorescens</em> (2%)</td>
<td>14.00</td>
<td>43.78</td>
<td>30.77 (33.68)</td>
<td>41.6 (40.17)</td>
<td>51.66 (45.95)</td>
</tr>
<tr>
<td>2</td>
<td><em>Trichoderma harzianum</em> (2%)</td>
<td>13.00</td>
<td>41.92</td>
<td>33.05 (34.91)</td>
<td>45.87 (42.64)</td>
<td>55.06 (47.91)</td>
</tr>
<tr>
<td>3</td>
<td>Fish amino acid (5%)</td>
<td>13.33</td>
<td>43.23</td>
<td>37.67 (37.82)</td>
<td>62.84 (52.45)</td>
<td>63.60 (52.89)</td>
</tr>
<tr>
<td>4</td>
<td>Turmeric powder and baking soda combination (10:1)</td>
<td>13.33</td>
<td>45.57</td>
<td>40.62 (39.59)</td>
<td>63.07 (52.58)</td>
<td>66.55 (54.60)</td>
</tr>
<tr>
<td>5</td>
<td>Absolute control</td>
<td>13.33</td>
<td>49.95</td>
<td>47.94 (43.74)</td>
<td>63.75 (52.99)</td>
<td>70.85 (57.34)</td>
</tr>
</tbody>
</table>

Mean of three replications*, values in the parenthesis are arc sin transformed. Treatments with same alphabets in the superscript, do not differ significantly (FAD - No. of days for first appearance of disease, PDS-Percentage disease suppression.)

Foliar spray of *P. fluorescens* recorded the lowest disease index of 57.21% with maximum yield of 22875.00 kg/ha which was on par with foliar application of *T. harzianum* with the disease index of 59.81%. According to Sudhakar et al., (2013) efficacy of *P. fluorescens* strain in improving drought tolerant traits and higher biometric traits such as plant height, shelling per cent, pod yield and harvest index over control as well as 2% urea spray under stress imposed field conditions. De Freitas and Germida (1991) reported that *Psudomonas fluorescens* strains have been found to suppress a wide range of plant diseases caused by microbial pathogens including foliar diseases caused by fungi such as *Gaeumannomyces graminis*, *Pythium spp.* and *R. solani* in green house as well as in field trials.

Among the indigenous organic formulations, foliar spray of turmeric baking soda combination has shown 6.35% disease suppression over control which was followed by fish amino acid with 4.20% disease suppression over the control. Smitha (2000) conducted several pot culture experiments and found that soil application followed by foliar spray with one per cent suspension of the talc based formulated product of antagonist was very effective in reducing the intensity of foliar blight caused by *R. solani* and was selected as the mode of delivery in the field. The use of turmeric-baking soda combination for the management of soil borne disease in rice has been reported by Gangopadhyay (1998). The spraying of fish aminoacid at weekly interval could reduce leaf spot disease and leaf feeder attack in amaranthus in farmers field (KAU, 2014). Foliar application of turmeric and sodium carboneate mixture in the ratio 5:1 significantly suppressed the leaf blight pathogen of amaranthus by 68.38% over untreated control and also resulted in higher yield compared to untreated control (Sheela et al., 2015).

**IV. ACKNOWLEDGMENT**

This research is a part from the first authors M.Sc (Ag.) work. The authors would like to thank the authorities of Kerala Agricultural University for their financial support and would like to
acknowledge the co-operation of Department of Plant Pathology, College of Agriculture, Vellayani for the successful completion of research work.

BIBLIOGRAPHY