

STUDIES ON THE CONTROL OF COLLAR ROT DISEASE CAUSED BY *RHIZOCTONIA SOLANI* IN TEAK (*TECTONA GRANDIS*) BY SEED TREATMENT

K. R. Ramesh*

Forest College and Research Institute, Tamil Nadu Agricultural University, Mettupalayam - 641 301
Tamil Nadu, India. E-mail: ramesh20@bycos.com

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RAMESH, K. R. 2002. Studies on the control of collar rot disease caused by *Rhizoctonia solani* in teak (*Tectona grandis*) by seed treatment. A survey which was carried out in the teak nurseries of Coimbatore Forest Division revealed that seedlings of teak (*Tectona grandis*) are susceptible to collar rot disease caused by *Rhizoctonia solani*, anamorph of *Thantephorous cucumeris*. Seed treatment with fungicides Emisan-6, Indofil M-45 or Bavistin reduced the collar rot of teak seedlings. Biological control agent *Trichoderma viride* MNT-7 reduced the collar rot disease even at a low concentration of 5 ml spore suspension kg⁻¹ of seed. The results suggest that these control measures can be used for successful raising of teak seedling in nurseries.

Key words: *Tectona grandis* - collar rot - *R. solani* - seed treatment - biological and chemical control

RAMESH, K. R. 2002. Kajian terhadap kawalan penyakit reput kolar yang disebabkan oleh *Rhizoctonia solani* pada pokok jati (*Tectona grandis*) secara rawatan biji benih. Kajian yang dijalankan di tapak semaian jati di Coimbatore Forest Division menunjukkan bahawa anak benih jati (*Tectona grandis*) adalah rentan terhadap penyakit reput kolar yang disebabkan oleh *Rhizoctonia solani*, anamorf *Thantephorous cucumeris*. Rawatan biji benih dengan racun kulat Emisan-6, Indofil M-45 atau Bavistin mengurangkan penyakit reput kolar pada anak benih jati. Agen kawalan biologi *Trichoderma viride* MNT-7 mengurangkan penyakit reput kolar walaupun pada kepekatan yang rendah iaitu 5 ml spora terampai bagi setiap sekilogram biji benih. Keputusan mengesyorkan bahawa cara kawalan ini dapat digunakan bagi kejayaan pembesaran anak benih jati di tapak semaian.

Introduction

Teak (*Tectona grandis*) is a valued all-purpose timber and grows over large natural areas in Southeast Asia (Kaosa-ard 1981). It is also grown as an exotic species in several tropical countries in Asia, Africa as well as Central and South America (White 1991). Today, it is considered to be one of the most promising tropical plantation species (Keogh 1996) and the area covered with teak plantations, which exceeds 82 000 ha, increases annually. For this purpose 21 million seedlings are

*Present address: National Bureau of Plant Genetic Resources, Regional Station, Indian Council of Agriculture Research, Phagli, Shimla, Himachal Pradesh Pin: 171004, India

raised annually (Bapat & Phulari 1995). Chemicals are not commonly used for the control of nursery diseases in forestry because they cause ecological imbalances and environmental pollution. The search for biological control methods of plant diseases are increasing. Few data are available on biological control of collar rot of teak (Sharma & Sankaran 1987). Higher incidence of collar rot caused by *Rhizoctonia solani*, anamorph of *Thantephorus cucumeris*, was observed in forest nurseries in Coimbatore district of Tamil Nadu. The mortality rate of seedlings ranged from 20 to 100% from October 1996 to October 1997 (Ramesh 2000a). This investigation was undertaken to study the effects of chemically- and biologically-treated seed in controlling rot disease.

Materials and methods

Five nurseries, Thakampatty Nursery, Lingapuram Nursery, Kallar Nursery, Mettupalayam Forest College Nursery and Coimbatore Agricultural University Nursery, were selected for the study. All the above nurseries fall under Coimbatore district which lies between 10° 10' and 11° 30' N latitude and 76° 40' and 77° 30' E longitude. Due to the presence of the Western Ghats and Anamalais hills the district benefits from the south-west monsoon. The climatic condition is humid to subhumid with 170–300 cm rainfall per annum. The soils are acidic, loamy sand with high percentage of organic carbon. The physico-chemical properties of the nursery soils are given in Table 1. The altitude ranges from 300–410 m above mean sea level. Survey was carried out every month for the collar rot disease incidence from October 1996 to October 1997.

Table 1 Physico-chemical properties of nursery soils

Characteristics	Thakampatty	Lingapuram	Nursery Kallar	Mettupalayam	Coimbatore
pH	4.7	4.6	2.5	4.4	4.7
Organic C (%)	0.38	1.62	3.96	1.83	1.57
Total N (%)	0.15	0.18	0.35	0.29	0.26
Available P ($\mu\text{g g}^{-1}$)	9.00	4.50	5.00	1.50	2.50
Available K ($\mu\text{g g}^{-1}$)	70.00	120.0	125.0	140.0	140.0
CEC (me 100 g^{-1} soil)	13.8	12.7	33.3	16.4	21.3
Maximum WHC (%)	34.0	56.1	59.4	74.1	67.4
Field capacity (%)	11.7	22.1	25.7	37.8	31.3
Sand (%)	85.0	60.0	66.3	75.0	55.0
Silt (%)	12.5	31.3	26.2	8.7	23.7
Clay (%)	2.5	8.7	7.5	16.3	21.3

Fungal isolates

For the pathogenicity tests, teak seedlings, showing symptoms of collar rot, were collected from the above five nursery areas. The pathogens were isolated

by using the tissue segment method (Rangaswamy 1972). The isolates were maintained on potato dextrose agar (PDA). They were then purified by the single hyphal tip method (Rangaswamy 1972). Two of the isolates obtained as mentioned above and one isolate obtained from the laboratory culture were used for the pathogenicity test. These isolates were designated as A, B and C respectively. These isolates were multiplied on sand-maize medium (Muthuswamy 1972). Sand and ground maize seeds were mixed at a ratio of 19:1 (w/w) and moistened. In the laboratory these were filled in conical flasks (500 ml) and autoclaved at a pressure of 1.8 kg cm⁻² for two hours. After sterilisation, the medium was inoculated separately with each one of the three isolates A, B and C and incubated at room temperature (28 ± 2 °C) for seven days.

Plant inoculation

Rhizoctonia solani, prepared by the above procedure, was then inoculated separately into sterilised nursery soil of Coimbatore nursery, contained in polyethelene bags at a ratio of 1:20 (w/w). Teak seeds were sown in these inoculated polyethylene bags at the rate of one seed per bag. These treatments were replicated four times and all bags were placed in a green house. Disease incidence was then calculated as a percentage of diseased plants 45 days after sowing. From this, highly virulent culture of the pathogen (between A, B and C) which caused the highest mortality was selected for further studies.

Biocontrol agent

Fungal antagonist, *Trichoderma viride* gray MNT-7 (MNT-Mutant Nitrosoguanidane Trichoderma), and a bacterial antagonist, *Pseudomonas fluorescens*, were used to test against *R. solani*. Cultures of both biocontrol agents were obtained from the Department of Plant Pathology, Tamil Nadu Agricultural University, Coimbatore. *Trichoderma viride* MNT-7 was maintained on PDA. *Pseudomonas fluorescens* was maintained on King's B solid medium (King *et al.* 1954).

Fungal inoculation

Spores of *T. viride* MNT-7 were harvested in sterile water from the culture multiplied on PDA medium 14 days after the incubation period. The spores were suspended in sterile distilled water, blended and filtered through a muslin cloth. The suspension containing conidia was centrifuged at 3000 rpm for 10 minutes. The supernatant was discarded and the conidial pellet was re-suspended in sterile distilled water. The process was repeated twice and then the conidia were suspended in 10 ml of 0.1% carboxy methyl cellulose solution. The spore concentration was adjusted to 4.5 × 10⁹–5.8 × 10⁹ conidia per ml using a haemocytometer. The teak seeds were treated first with a suspension of 5, 10 and 15 ml of *T. viride* MNT-7 before being inoculated with the pathogen.

Talc-based formulation of *P. fluorescens*, obtained from the Department of Plant Pathology, Tamil Nadu Agricultural University, Coimbatore, was used to treat the seeds at the rate of 5, 10 and 15 g kg⁻¹ of seeds. They were then inoculated with the pathogen, dried in the shade for 12 hours and sown in the polyethylene bags containing sterilised nursery soil at the rate of one seed per bag. Seeds inoculated with the pathogen alone were used as control and the whole experiment was conducted in a green house having optimum level of humidity.

Chemical seed treatment

Common seed dressing fungicides, namely, Kavach (chlorothalonil), Emisan-6 (methoxyethyl mercury chloride), Indofil M-45 (mancozeb), Fytolan (copper oxychloride) and Bavistin (carbendazim) at the rate of 2, 3, 4 and 5 g as well as neem oil and Calixin (Tridemorph), each 2, 3, 4 and 5 ml, were used to treat 1 kg of seeds. The seed treatment was done 24 hours before sowing. The treated seeds were dressed with the above fungicides as w/w and full coverage of the seed was ensured. Treated and untreated (control) seeds were sown in the soil inoculated with the pathogen at the rate of one seed for each polyethylene bag. Each treatment was replicated 10 times and regular observations were made after seed germination. Watering was conducted from time to time as recommended by Singh (1982). Seedling mortality count was carried out 45 days after sowing.

Statistical analysis

The data collected from various experiments were analysed statistically adopting the methods described by Panse and Sukhatme (1967). Wherever necessary the data were transformed into angular (arcsin percentage) values and square root transformation before carrying out statistical analysis. Critical difference values were calculated at five per cent probability level and the treatment mean values of the experiments were compared using Duncan's multiple range test (DMRT) (Gomez & Gomez 1984).

Results and discussion

Collar rot caused by *R. solani* was observed in all the five nurseries surveyed. The results are tabulated in Tables 2 and 3. Isolation from the diseased stem also yielded *R. solani*, which was confirmed by pathogenicity test. The disease incidence, which ranged from 20 to 100%, was highest in July, August and September in the five nurseries.

Table 2 shows that Emisan-6, Indofil M-45 and Bavistin at all treatment levels completely inhibited the disease incidence. Kavach also inhibited the occurrence of the disease and only 2 to 8% collar rot was noted at the various concentrations of the fungicide. On the other hand, Fytolan, Calixin and neem oil only managed to partially inhibit the disease incidence, giving a percentage of 92 to 97%. In the control treatment, 99% of the seedlings exhibited the collar rot disease.

Table 2 Effect of fungicides and neem oil seed treatment on the incidence of collar rot of teak

Treatment	Disease incidence (%)*			
	5 g ml ⁻¹	4 g ml ⁻¹	3 g ml ⁻¹	2 g ml ⁻¹
Kavach	2 ^c	2 ^c	5 ^d	8 ^b
Emisan-6	0 ^c	0 ^c	0 ^c	0 ^c
Indofil M-45	0 ^c	0 ^c	0 ^c	0 ^c
Fytolan	92 ^b	93 ^b	94 ^c	96 ^a
Bavistin	0 ^c	0 ^c	0 ^c	0 ^c
Calixin	92 ^b	93 ^b	93 ^c	95 ^a
Neem oil	94 ^b	94 ^b	96 ^b	97 ^a
Control	99 ^a	99 ^a	99 ^a	99 ^a

*Means in a column with the same letter are not significant as per DMRT ($p < 0.05$)
Mean of three replications

Table 3 Effect of seed treatment with antagonist on the control of collar rot incidence of teak

Antagonist	Seedling mortality (%)*		
	15 ml g ⁻¹ kg ⁻¹ seeds	10 ml g ⁻¹ kg ⁻¹ seeds	5 ml g ⁻¹ kg ⁻¹ seeds
<i>Trichoderma viride</i> MNT-7	2.0 ^c	5.0 ^c	7.0 ^c
<i>Pseudomonas fluorescens</i>	93.0 ^b	94 ^b	94.0 ^b
Control	97.0 ^a	97.0 ^a	97.0 ^a

*Means in a column with the same letter are not significant as per DMRT ($p < 0.05$)

In an investigation using poisoned food technique, it was reported that Kavach, Emisan-6, Indofil M-45 and Bavistin showed very effective control of the pathogen *R. solani* (93.75% over control), which caused collar rot in teak at all the concentrations tested, namely, 0.1, 0.05 and 0.025% (Ramesh 2000b). However, there were no significant differences between Fytolan, Calixin, neem oil and control in inhibiting the pathogen. These results are in agreement with Lisker and Meiri (1992), Ahmed *et al.* (1994) and Shukla (1995). Mehrotra (1990) reported that *R. solani* caused top flagging in *Pinus kesia* and leaf web blight in *Michelia champaca* and *Pongamia pinnata* seedlings. The author further found that, using poisoned food technique, 0.2% Dithane M-45 and 0.1% Bavistin inhibited the radial growth of *R. solani*.

T. viride MNT-7 significantly reduced seedling mortality (2 to 7%) (Table 3). Minimum seedling mortality was recorded when the seeds were treated with 15 ml g⁻¹ kg⁻¹ seeds. Similar results had been reported by Ehteshamul-Haque *et al.* (1994) in okra, sunflower, soya bean and mung bean seeds. Ehteshamul-Haque and Ghaffar (1991) reported that seed dressing of mustard with *T. viride* controlled *R. solani* infections. Similarly, *R. solani* damping-off was controlled by *T. viride* seed

treatment at 5 ml kg⁻¹ of seed in *Eucalyptus camaldulensis* (Kumar & Marimuthu 1994). In contrast, *P. fluorescens* slightly checked the disease and up to 94% of the seedlings were infested with the collar rot disease. However, sheath blight in rice caused by *R. solani* was controlled through seed treatment with *P. fluorescens* (5 g kg⁻¹ of seeds) (Rabindran & Vidyasekaran 1996).

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