

Original Research Article

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**Management of Fruit Rot Disease of Arecanut (*Areca catechu* L.)
caused by (*Phytophthora meadii* Mc Rae.)**

B. Gangadhara Naik¹, H.P. Maheshwarappa², Gowdra Nagamma¹ and S. Latha^{3*}

¹Department of Plant Pathology, ³Department of Entomology,
University of Agricultural and Horticultural Sciences, Shivamogga, Karnataka, India

²Department of Agronomy, Indian Council of Agricultural Research,
Kasaragod, Kerala, India

*Corresponding author

ABSTRACT

Keywords

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Fruit rot or Mahali disease of Arecanut (*Areca catechu* L.) is a serious threat from countries in those regions which receives heavy rainfall during *kharif* season specially, malnad, hilly and coastal regions of Karnataka. Arecanut grown in these regions are highly prone to the occurrence of the fruit rot disease resulting in severe loss in yield Therefore, present investigations were carried out during 2015-17, at three different locations viz., Koluru, Manchale and Melige of Shivamogga districts of Karnataka, India. Field trials were conducted under All India Coordinated Research Project (AICRP) on Palms to identify suitable ecofriendly management measures. Among the treatments tested, Bordeaux mixture (BM. 1 %) was found most effective followed by Fenamidone 10% +Mancozeb 50 % (W/W) (@ 0.3 % spray +Adhesive. However, among the biocontrol agents tested, microbial consortia containing *Trichoderma harzianum* (IMI304056), *Pseudomonas fluorescens* (NCB19046) and *Bacillus megatarium* (NCTC9848) was found to be most effective in reducing the disease incidence as well as enhancing the development of new roots, increase in number of leaves and yield per palms indicating the merits of using bio agents.

Introduction

Arecanut (*Areca catechu*) is an important commercial crop cultivated in many parts of the world with an area of world 846 (000 ha), production of 1.21 mt and productivity 1.43 (tons/ha). In India 466.2 (000 ha), production of 7.30 mt with a productivity 1.56 tones/ha. Whereas, Karnataka arecanut is 227.8 (000 ha) with cultivated in an area of production of 435.8 (000 tons) and productivity of 1.91

tones/ha (NHB data, 2017). Farmer of hilly and coastal regions is findings lot of difficulties in arecanut cultivation as the crop suffers from various pest and diseases. Among the diseases, fruit rot (Koleroga) of arecanut caused by *Phytophthora meadii* and has been considered as a major threat in successful cultivation of Arecanut causing a yield loss of upto 90 % (Sarma *et al.*, 2002). This disease occurs in severe form in those areas which receives heavy rainfall (Coleman,

1910). Koleroga infection results in dropping of both immature and mature nuts. Infected nuts show the presence of brownish water soaked lesions near the stem end and falls down. After a few days, infected fallen nuts exhibit the presence of white felt like mycelial mat covering the entire nut. Such mycelia mat not only consists of mycelia but also lots of sporangia and chlamydospores. The disease under congenial environmental condition *viz.*, high relative humidity (90 to 100 %) and low temperature (20 °C to 22 °C) results in heavy yield loss (Lokesh *et al.*, 2014; Ramesh *et al.*, 2014; Hegde and Chowdappa, 2015).

In the light of present day constraints with the use of chemical pesticides in plant disease management as well as farmers orientation towards non chemical management the biological control management is found to be best alternate option as it is ecofriendly and cost effective. Under biological control of plant diseases, various antagonistic organisms have been identified, which fight against the pathogens by different mechanisms *viz.*, competition, lysis, antibiosis, siderophore production and hyper parasitism (Elad *et al.*, 1982). Syed Sajeed Ali and Vidhale (2013) stated that, fluorescent pigments produced by *Pseudomonas* are sequester Fe³⁺ ions (Ferric) and are termed as siderophores, which act as inhibitors for the growth of some phytopathogenic bacteria and fungi. Biochemical studies conducted by Chatterjee *et al.*, (1996) showed that efficient strains of *Pseudomonas fluorescens* produces an antibiotic phenazine-1-carboxylic acid (PCA) responsible for hindering the growth of plant pathogenic bacteria.

In this research paper, we would like to introduce the role of bioagents in the management of fruit rot of arecanut, caused by *Phytophthora meadii* and comparisons with the Bordeaux mixture and chemical Sectin In this regard, efforts are being made to develop

efficient biocontrol agents to manage the disease.

Thus, the present study was aimed at finding out an effective long lasting management practices using biocontrol agents and fungicides for the management of fruit rot disease of arecanut.

Materials and Methods

An experiment was conducted at 3 areca gardens *viz.*, Melige and Koluru (Thirthahalli Tq.) and Manchale (Sagar Tq.) of Shivamogga district in Karnataka during *Kharif* 2015-16, 2016-17 and 2017-18. The trial was designed by following Randomized Complete Block Design (RCBD) with seven treatments and three replications. For each replication 16 plants were maintained and observations were recorded from four plants at the centre of the replicated area.

Treatment details:

T1: *Trichoderma harzianum* (IMI 304056) @ 20 ml (105×10⁻⁷ cfu/ml stock soln/L water)+soil application of microbial consortia (IMI 304056) @ 50 g + 1 kg FYM/palm.

T2: *Pseudomonas fluorescens* (NCBI9046) @ 20 ml (105×10⁻⁷ cfu/ml stock soln/L water)+soil application of microbial consortia (NCBI9046) @ 50 g+ *Trichoderma harzianum* (IMI 304056) 50 g + 1 kg FYM/palm.

T3: *Bacillus megatarium* (NCTC9848) @ 20 ml (105×10⁻⁷ cfu/ml stock soln/L water) +soil application of microbial consortia (Shivamogga isolate) @ 50 g+ *Trichoderma harzianum* (IMI 304056) 50 g + 1 kg FYM/palm. T4: *Trichoderma harzianum* (IMI 304056) @ 20 ml (105×10⁻⁷ cfu/ml stock soln/L water+ *Pseudomonas fluorescens* (NCBI9046))@ 20 ml (105×10⁻⁷ cfu/ml stock

soln/L water+ *Bacillus megatarium* (NCTC9848) @ 20 ml (105×10⁻⁷ cfu/ml stock soln/L water+ soil application of microbial consortia @ 100 g/plant along with 1 kg of FYM/decomposed compost to the soil.

T5: Fenamidone 10 % + Mancozeb 50 % (W/W) (Sectin) (@ 0.3 % spray) + Adhesive

T6: Application of Bordeaux mixture @ 1 % + Adhesive

T7: Control

*Microbial consortia: *Trichoderma harzianum* (50 g) *Pseudomonas fluorescens* and *Bacillus megatarium* @ 25 g each.

Spray schedule (T1 to T4)

Ist application – Between 15-25th of April

IInd application – 30 days after first spray / application

IIIrd application – 30 days second spray / application

Spray schedule (T5 to T6)

Prior to onset of monsoon the per cent disease incidence was calculated for all the locations and pooled analysis of data is presented in table the formula given below.

Per cent disease incidence =

$$\frac{\text{Number of infected plants}}{\text{Total number of plants observed}} \times 100$$

Bordeaux mixture 1 % was prepared as per the procedure given by (Ramesh, *et al.*, 2014).

Preparation of spray solution for biocontrol agents

The pure cultures of biocontrol agents *viz.*, *Trichoderma harzianum* (IMI 304056),

Pseudomonas fluorescens (NCBI9046) and *Bacillus megatarium* (NCTC9848) obtained from the department of microbiology UAHS, Shivamogga were multiplied in potato dextrose broth (*T. harzianum*) and nutrient broth (*Pseudomonas fluorescens* and *Bacillus megatarium*) and the spray solution was prepared by adding required volume of the stock solution in known volume of water to get desired concentration.

10 ml of stock solution is suspended in 90 ml of sterile water and stirred the content to get 1:10 dilution (10⁻¹), and then the serial dilution was made upto 10⁻⁷ for bacterial and 10⁻⁵ for *Trichoderma*. The stock solution thus prepared, later diluted with required quantity of water and utilized for spraying as per the technical programme. However, the fungicidal spray solution was prepared by adding required quantity of fungicides in known volume of water.

Results and Discussion

The field studies were carried out to assess the importance of biocontrol agents in the management of fruit rot disease in comparison to the chemicals *viz.*, Bordeaux mixture and chemical sectin (Fenamidone + Mancozeb).

Towards the end, of the field trials, we had compared the bioagents load in our research plot and conventionally grown arecanut field. Imposition of bioagents had shown a strong anti-fungal activity (with specific reference to fruit rot).

The results obtained thus, revealed that, application of biocontrol agents significantly reduced occurrence in the affected garden by the way of reducing the nut fallen and increase in the root growth and yield during three years of treatments and were explained below (Fig. 1).

The least number of nuts fallen was recorded with treatment T₆ in all the three experimental plots viz., Manchale, Koluru and Melige with 8.40, 9.33 and 8.67 followed by treatment T₅ and T₄ respectively. However, among the biocontrol based treatments, T₄ (Microbial consortia) resulted in least number of nuts fallen and differed significantly compared to treatment T₁, T₂ and T₃ respectively (Table 1).

Infected nuts fallen/plant revealed that, the treatment T₆ effectively reduced the disease incidence in all the three experimental plots with 2.72, 3.11, 2.89 Manchale, Koluru and Melige respectively followed by T₅ [Fenamidone 10 % + Mancozeb 50 % (W/W) (Sectin) (@ 0.3 % spray) + Adhesive]. However, among the biocontrol agents tested, treatment T₄ (*Trichoderma harzianum* (IMI304056) @ 20 ml (105×10⁻⁷ cfu/ml stock soln/L water + *Pseudomonas fluorescens* (NCIB9046) @ 20 ml (105×10⁻⁷ cfu/ml stock soln/L water + *Bacillus megatarium* (NCTC9848) @ 20 ml (105×10⁻⁷ cfu/ml stock soln/L water + soil application of microbial consortia @ 100 g/plant along with 1 kg of FYM/decomposed compost to the soil per year) was found effective in reducing the disease incidence significantly reduced number of nuts fallen in treatment T₁ T₂ and T₃ (Table 2).

The data on per cent disease incidence due to koleroga at three locations during all the three years of experiment is and is presented in Table 3. The data revealed that, among the treatments tested, treatment T₆ (BM. 1 %) was found to be most effective in reducing the disease incidence (5.64, 7.75 and 7.29) followed by T₅ [Fenamidone 10 % + Mancozeb 50 % (W/W) (Sectin) (@ 0.3 % spray) + Adhesive] and were on par with each other and differed significantly compared to all other treatments tested even though it was reduced the disease we cannot recommend to

average farmers because its cost more. However, among the biocontrol agents tested, treatment T₄ was found superior compared to other treatments tested with T₁ to T₃ with per cent reduction in disease incidence it is ecofriendly helps in plant growth promotion (Table 3).

Mean pooled data on per cent incidence of koleroga disease revealed that among the treatments tested, treatment T₆ (BM. 1 %) was found to be the most effective (4.88, 6.98 and 5.85) in reducing the disease incidence during all the three years of observations, followed by treatment T₅ [Fenamidone 10 % + Mancozeb 50 % (W/W) (Sectin) (@ 0.3 % spray) + Adhesive]. However, among the biocontrol agents tested, treatment T₄ was found effective in reducing the disease incidence (Table 4).

The biocontrol agents also effective in reducing the nut fallen, increase in number of root as well as population of bioagents multiplied in the soil. May be because of involving in mycoparasitism, antibiotic and competition for nutrients also induce the defense responses in plants. *Pseudomonas fluorescens* also used for soil borne pathogens it will help in root colonization. The antifungal metabolite 2,4 diacetyl phloroglucinol, HCN production has role of disease suppression and producing antibiotic compound pyrrolnitrin which inhibit growth of pathogen play major role.

Bordeaux mixture also effective in controlling the fungus may be Cu²⁺, these ions affect enzyme in the fungal spore in such a way as to prevent spore germination. And sectin is alternative chemical for this disease its also controlling the disease fenamidone inhibits mitochondrial respiration by blocking electron transport at ubihydroquinone; cytochrome -C- oxidoreductase (Complex III) and mancozeb inhibit respiration.

The biocontrol agents are the best management for controlling koleroga of arecanut. There is no risk of resistance. It will help to boosting the roots as well as soil fertility.

Population dynamics of biocontrol agents in koleroga experimental gardens revealed that, all the three biocontrol agents viz., *Trichoderma harzianum* (IMI 304056),

Pseudomonas fluorescens (NCBI9046) and *Bacillus subtilis* (NCTC9848) were survived in the crown region during entire year. However, population density of these biocontrol agents was varied from 0.2 to 2.4 CFU's BIT (Before Imposition of Treatment), whereas, population was increased thereafter and ranged from 0.2 to 2.9 CFU's AIT (After Imposition of Treatment).

Table.1 Average number of fallen nuts/plant in experimental gardens

Treatment	Manchale	Koluru	Melige
T1: <i>Trichoderma harzianum</i> (IMI 304056) @ 20 ml (105×10^{-7} cfu/ml stock soln/L water)+soil application of microbial consortia (Shivamogga isolate) @ 50 g + <i>Trichoderma harzianum</i> (Shivamogga isolate) 50 g + 1 kg FYM/palm.	14.33 (3.92)*	14.67 (3.96)	14.00 (3.87)
T2: <i>Pseudomonas fluorescens</i> (NCBI9046) @ 20 ml (105×10^{-7} cfu/ml stock soln/L water)+soil application of microbial consortia (Shivamogga isolate) @ 50 g+ <i>Trichoderma harzianum</i> (Shivamogga isolate) 50 g + 1 kg FYM/palm.	14.33 (3.92)	14.33 (3.92)	13.67 (3.83)
T3: <i>Bacillus megatarium</i> (NCTC9848) @ 20 ml (105×10^{-7} cfu/ml stock soln/L water) +soil application of microbial consortia (Shivamogga isolate) @ 50 g+ <i>Trichoderma harzianum</i> (Shivamogga isolate) 50 g + 1 kg FYM/palm.	14.00 (3.87)	13.67 (3.83)	13.00 (3.74)
T4: <i>Trichoderma harzianum</i> (IMI 304056) @ 20 ml(105×10^{-7} cfu/ml stock soln/L water+ <i>Pseudomonas fluorescens</i> (NCBI9046))@ 20 ml (105×10^{-7} cfu/ml stock soln/L water+ <i>Bacillus megatarium</i> (NCTC9848) @ 20 ml (105×10^{-7} cfu/ml stock soln/L water+ soil application of microbial consortia @ 100 g/plant along with 1 kg of FYM/decomposed compost to the soil per year.	10.50 (3.69)	12.67 (3.70)	11.00 (3.46)
T5: Fenamidone 10 %+Mancozeb 50 % (W/W) (Sectin) (@ 0.3 % spray) +Adhesive	9.20 (3.56)	11.33 (3.51)	10.00 (3.31)
T6: Application of Bordeaux mixture@ 1 % +Adhesive	8.40 (3.41)	9.33 (3.21)	8.67 (3.10)
T7: Control	31.67 (5.71)	31.00 (5.66)	34.00 (5.91)
SE m±	0.08	0.06	0.09
CD at 5 %	0.25	0.17	0.28

*Figures in parenthesis are square root transformed values

Table.2 Average number of infected nut fallen/plant in experimental gardens

Treatment	Manchale	Kuluru	Melige
T1	4.78 (2.40)	4.89 (2.43)	4.67 (2.34)
T2	4.78 (2.40)	4.78 (2.40)	4.56 (2.36)
T3	4.67 (2.38)	4.56 (2.36)	4.33 (2.31)
T4	3.86 (2.28)	4.22 (2.28)	3.67 (2.16)
T5	3.10 (2.21)	3.78 (2.19)	3.34 (2.08)
T6	2.72 (2.13)	3.11 (2.03)	2.89 (1.97)
T7	14.56 (3.39)	17.12 (4.26)	14.83 (4.01)
SE m±	0.04	0.03	0.05
CD at 5 %	0.13	0.11	0.15

*Figures in parenthesis are square root transformed values

Table.3 Mean per cent incidence of koleroga disease in experimental plots (2017)

Treatments	Melige	Kolur	Manchale	Mean
T1	19.04	17.40	19.69	18.71
T2	16.49	14.98	18.05	16.51
T3	14.69	14.43	17.28	15.47
T4	9.21	13.20	13.99	13.13
T5	6.48	8.87	9.98	9.11
T6	5.64	7.75	7.29	7.56
T7	25.94	22.99	21.96	23.63
SE m±	0.28	0.27	0.49	0.72
CD @ 5%	0.86	0.85	1.52	2.21

Table.4 Mean pooled per cent incidence of koleroga disease in experimental arecanut gardens (2015-17)

Treatment	Cumulative mean per cent disease incidence (2015-17)		
	Manchale	Koluru	Melige
T1	15.13	14.61	15.61
T2	14.60	14.37	15.92
T3	13.62	13.62	14.38
T4	10.45	11.91	9.79
T5	5.98	7.12	6.79
T6	4.88	6.98	5.85
T7	19.82	22.94	23.93
SE m±	0.15	0.33	0.16
CD at 5 %	0.48	0.11	0.50

Table.5 Microbial population in the crown and rhizosphere soil of koleroga experimental Gardens

Places	Mean microbial population											
	Crown region						Rhizosphere soil					
	BIT			AIT			BIT			AIT		
	Tr.	Ps.	B	Tr.	Ps.	B	Tr.	Ps.	B	Tr.	Ps.	B
Koluru	0.4	0.2	2.2	0.8	0.2	2.9	2.2	2.9	3.4	3.5	3.6	4.3
Melige	0.3	0.2	1.6	0.6	0.4	2.7	2.6	2.8	2.2	3.7	3.9	4.0
Manchale	0.4	0.3	2.4	0.6	0.3	2.8	1.1	1.9	2.1	4.3	3.3	5.1

Tr.: *Trichoderma* B: *Bacillus* Ps: *Pseudomonas*

BIT: Before imposition of treatments cfu: Colony forming units, AIT: One year after imposition of treatments.
Population: (Fungus cfu x 10³ g⁻¹ dry soil) (Bacteria cfu x 10⁵ g⁻¹ dry soil)

Table.6 Effect of biocontrol agents on root growth of areca plants

Places	Mean no. of healthy primary roots/palm in experimental garden up to the depth of 30 cm		Mean no. of healthy roots/palm in farmers practice	
	Before imposition of treatments	After imposition of treatments	Initial number*	After one year*
Koluru	244.50	275.25	248.00	251.50
Melige	205.75	254.50	213.10	224.50
Manchale	252.30	289.65	235.25	245.75

*Observations were taken simultaneously along with experimental garden

Table.7 Cost economics of treatments in the management of koleroga disease of Arecanut

Sl. No.	Treatments	Yield (t/ha)	Cost of plant protection (Rs/ha)	Total cost of production (Rs/ha)	Gross returns (Rs/ha)	Net returns (Rs/ha)	C:B
1.	T1	1.25	17,750.00	87,750.00	3,60,487.50	2,72,737.50	1:4.11
2.	T2	1.25	17,750.00	87,750.00	3,60,487.50	2,72,737.50	1:4.11
3.	T3	1.37	17,750.00	87,750.00	3,96,537.50	3,08,787.50	1:4.52
4.	T4	1.50	23,750.00	93,750.00	4,32,585.00	3,38,835.00	1:4.61
5.	T5	1.62	13,125.00	83,125.00	4,68,635.00	3,85,510.00	1:5.64
6.	T6	1.75	4,500.00	74,500.00	5,04,682.50	4,30,182.50	1:6.77
7.	T7	0.90	-	70,000.00	2,59,551.00	2,18,390.00	1:3.71
S.Em ±		0.01	-	-	-	-	-
CD @ 5 %		0.03	-	-	-	-	-

Gross returns = Yield × Market price (Rs.2883.9 /- t/ha), Net returns = Gross returns – Total cost

Fig.1



Fallen nuts due to koleroga



***Phytophthora meadii* mycelia mat on nuts**

In rhizosphere soil, population density was increased during the assessment period. Among the biocontrol agents isolated per gram of dry soil *Bacillus* sp. recorded the

highest colony forming unit per gram of dry soil followed by *Trichoderma* sp. in the entire three experimental gardens (Table 5).

Application of biocontrol agents to the soil, induced the production of new roots by increasing the root density after imposition of treatments (275.25, 254.50 and 289.65 roots/palm) in all the koleroga experimental gardens compared to farmers field after imposition of treatments (251.50, 224.50 and 245.75 roots/palm) where no biocontrol agents was applied (Table 6).

As the evident from table 7, at the end of three years increased the roots. The highest C:B ratio was obtained in T6 (Bordeaux mixture 1 %) 1:6.77 Rs/ acre. Followed by T5 1:5.64Rs/ acre and T4 1:4.61 Rs/ acre. However lowest C: B ratio was recorded in control 1:3.71 Rs/ acre respectively.

These above results are in conformity with the findings of Sastry (1982), who reported that Bordeaux mixture (1 %), copper oxychloride and metalaxyl were found effective in inhibiting the growth and sporulation of *Phytophthora capsici* and *P. meadii*. Similarity Fruit rot of arecanut being season bound, it is the previous seasons inoculum in the form of latent infection within the dried bunches and canopy which serves as initial inoculums. The secondary spread is by means of sporangia which are produced abundantly on the infected fruits. The minimum incidence of fruit rot in Bordeaux mixture and Metalaxyl MZ treated plots may be due to reduced number of secondary inoculums due to antispore activity of this fungicide (Chowdappa *et al.*, 2002; Anandraj and Sarswathy, 1986), there by restricting the rapid secondary spread of the disease. The similar findings are in conformity with Jeeva *et al.*, (2015) over the years various chemicals were screened for the management of the *Phytophthora* diseases in arecanut. In a muttilocal trial on management of fruit rot disease using different fungicides revealed that Boredeaux mixture 1 per cent spray still holds good in controlling fruit rot disease

(Narayanaswamy *et al.*, 2017; Chowdappa *et al.*, 2000). Application of conventional Bordeaux mixture at 1 per cent has significantly reduced number of fallen nuts due to fruit rot of arecanut at 6 locations (0.91) and increased the nut yield (green nut 85.2 t/ha and dry nut 3.0 t/ha) followed by one per cent of stabilized Bordeaux mixture and Metalaxyl MZ. Maximum disease incidence and affected fallen nuts was recorded in untreated check (Narayanaswamy *et al.*, 2017).

Even though Bordeaux mixture 1 % with pH 7 is very effective against fruit rot disease, if it is properly prepared and applied.

However, application of microbial consortia containing *T. harzianum*, *P. fluorescens* and *B. subtilis* organisms applied to the crown region as well as soil application will gave lot of influence in managing the disease as there organisms survives by producing endospores (*Bacillus megatarium*) and chlamyospores (*T. harzianum*) which will survives during adverse climatic condition and can suppress the pathogen which is survives in soils as well as in the crown region by producing structures like chlamyospores and dormant mycelia.

In conclusion, present investigation showed that the application of Bordeaux mixture (1%) twice (for normal rainfall) as thrice for continued rainfall to the bunches of arecanut during pre monsoon and 25 days after first or second application drastically reduce the incidence of Koleroga with increase in the yield. Among the biocontrol agents combination of (*Trichoderma harzianum* (IMI304056) @ 20 ml (105×10⁻⁷ cfu/ml stock soln/L water+ *Pseudomonas fluorescens* (NCIB9046) @ 20 ml (105×10⁻⁷ cfu/ml stock soln/L water+ *Bacillus megatarium* (NCTC9848) @ 20 ml (105×10⁻⁷ cfu/ml stock soln/L water+ soil application of microbial consortia @ 100 g/plant along with 1 kg of FYM/decomposed compost to the soil per

year) was found effective in reducing the disease incidence and also enhancing the root growth.

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