# Integrated Management of Sigatoka Leaf Spot Disease of Banana Caused by *Mycosphaerella musicola* R. Leach ex J.L. Mulder

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#### **DEPARTMENT OF PLANT PATHOLOGY**

## **COLLEGE OF AGRICULTURE**

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# Integrated Management of Sigatoka Leaf Spot Disease of Banana Caused by *Mycosphaerella musicola* R. Leach ex J.L. Mulder

by

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# (2012-11-179)

## THESIS

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### **DEPARTMENT OF PLANT PATHOLOGY**

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KERALA, INDIA

2015

#### **DECLARATION**

I, hereby declare that this thesis entitled "Integrated Management of Sigatoka Leaf Spot Disease of Banana Caused by *Mycosphaerella musicola* R. Leach ex J.L. Mulder" is a bonafide record of research work done by me during the course of research and the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society.

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#### <u>CERTIFICATE</u>

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EXTERNAL EXAMINER

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#### LIST OF ABBREVIATIONS

cm	-Centimeter
cm <sup>2</sup>	-Centimeter square
CD (0.05)	-Critical difference at 5per cent level
cv.	-Cultivar
et al.	-And others
Fig.	-Figure
g	-Gram
mg	-Milligram
ha <sup>-1</sup>	-Per hectare
i.e.	-That is
K	-Potassium
kg	-Kilogram
kg ha <sup>-1</sup>	-Kilogram per hectare
g l <sup>-1</sup>	-Gram per litre
DAP	-Days after planting
m	-Meter
cm	-Centimeter
m <sup>-2</sup>	-Per meter square
mg	-Milligram
mg g <sup>-1</sup>	-Milligram per gram
μg <sup>-1</sup>	- Microgram per gram
BCA	-Bio Control Agent
NPK	-Nitrogen, Phosphorous and Potassium
В	-Boron
Mg	-Magnesium

.

Zn	-Zinc
FeSO <sub>4</sub>	-Iron sulphate
S	-Sulphur
N	-Normal (unit of Normality)
М	-Molar (unit of Molarity)
NaOH	-Sodium Hydroxide
WP	-Wettable powder
EC	-Emulsifying Concentrate
NS	-Not significant
nm	-Nanometer
rpm	-Revolutions per minute
ppm	-Parts per million
Р	-Phosphorous
P <sup>H</sup>	-Negative logarithm of hydrogen ion concentration
PGPR	-Plant Growth Promoting Rhizobaoteria
var.	-Variety
cv.	-Cultivar
Т	-Treatment
chl.	-Chlorophyll
Plant <sup>-1</sup>	-Per plant
RBD	-Randomized block design
Rs.ha <sup>-1</sup>	-Rupees per hectare
BCR	- Benefit Cost Ratio
sp.	-Species
t	-tonnes
viz.,	-Namely
201	

#### LIST OF SYMBOLS

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- @ -At the rate of
- <sup>0</sup> -Degree
- <sup>0</sup>C -Degree Celsius
- % -Per cent
- ′ -Or

.

# **INTRODUCTION**

#### INTRODUCTION

Banana (*Musa* spp.), can be considered as the most popular fruit crop being commercially grown in many tropical and subtropical countries for its utilization as dessert and as staple food. It is a large monocotyledonous herb that originated in South East Asia. Being the fourth most important food after rice, wheat and maize products, it tolls up as a major fruit crop of India. In exports, it ranks fourth among all agricultural commodities. The crop having wide distribution in the country accounts for 27.01 million tonnes from an area of 0.765 million hectare contributing nearly 22.15 percent of the global production (FAO, 2012).

Despite the fact that crop is grown in diverse agro climatic conditions of the country, its production is continuously being hampered by a range of diseases inflicting yield losses of staggering dimension both in quantity as well as quality aspects. Of the many fungal diseases occurring on banana, Sigatoka leaf spot incited by *Mycosphaerella musicola* R. Leach ex J.L. Mulder also called as yellow Sigatoka (Plate 1) is considered as a serious threat to world banana production (Mourichon and Fullerton, 1990; Selvarajan *et al.*, 2000). When leaf spot become severe, it reduces the yield drastically. Burt *et al.* (1997) reported more than 50% of economic losses due to this disease across the world. The disease not only affects the banana leaves, but also bunch weight and fruit quality. Further Marin *et al.* (2003) opined that the disease induces significant reduction in yield, apart from premature ripening of fruits which can occur in the field and during transport and storage (Marin and Romero, 1992). Thus this disease has assumed serious dimension during recent days.

Intensive farming practices that warrant high yield and quality require extensive use of chemical fertilizers and pesticides, which are not only costly but also pollute the environment and is against the interest of environment friendly, sustainable agricultural practices (Harish *et al.*, 2008a). Due to imbalanced nutrition, the disease incidence and thus economic loss is increasing in India.



Plate 1. Disease severity of yellow Sigatoka in Nendran

Preventive management by balanced nutrition is least understood and curative management of diseases using costly fungicides is becoming a problem. Exclusive fungicide oriented disease control has resulted in residue accumulation which is harmful to environment and consumers. This has also resulted in new resistant land races of pathogens.

Improved nutrition can boost the host defense mechanism which in turn results in disease reduction through direct inhibition of fungal activity. Micronutrient disorders of Zn, Mn, B, Cu and Fe are widespread in India (Anonymous, 1995) and correction of these disorders has resulted in resistance to diseases. Among the important nutrients required by the banana crop, only the NPK fertilizers are used on a large scale and the farmers have ample knowledge about their application. Quite often, the secondary and micronutrients remain neglected. Consequently, the banana crop succumbs to several diseases which in turn affect the yield of the crop adversely. The same scenario exists in Kerala also, where banana is a predominant crop and Sigatoka leaf spot is very severe disease especially during the rainy season (Suharban, 1977; Selvarajan *et al.*, 2000).

Integrated management of the disease is the only strategic, economical and environmental pollution free option. For achieving integrated management, we have to supplement secondary and micronutrients along with NPK for attaining the balanced nutrition. In addition, biocontrol agents are also applied as foliar sprays in addition to seed treatments for optimum control of the disease. While existing control measures are having many lacunae, biological control using *Pseudomonas fluorescens* in a system of integrated control measures can provide effective and sustainable management in order to avoid the consequences of exclusive and extensive use of chemicals. Recently, more emphasis has been laid on supplementing various nutrients with bio-protectants, which is better than either alone (Janisiewics and Bors, 1995; Kavino *et al.*, 2008). Hence the field study "Integrated management of Sigatoka leaf spot disease of banana caused by

*M. musicola* R. Leach ex J.L. Mulder" was undertaken with the following objectives.

- 1. To study the effect of new generation fungicides (both contact and systemic fungicides) on growth, incidence of Sigatoka leaf spot disease and yield of banana in coconut gardens.
- 2. To investigate the effect of nutrients both, secondary (Magnesium) and micronutrients (Zinc and Boron).
- 3. To evaluate the efficacy of *Pseudomonas fluorescens* (PGPR mix II) as a biocontrol agent.

# **REVIEW OF LITERATURE**

#### **2. REVIEW OF LITERATURE**

Sigatoka leaf spot is one of the serious fungal disease which is caused by two ascomycetous fungi *viz.*, *M. musicola* that causes yellow Sigatoka disease and *M. fijiensis* Morlet causing black Sigatoka or black leaf streak disease. Both diseases destroy large proportion of leaf tissue resulting in drastic reduction of yield losses of about 30% (Mourichon *et al.*, 1997).

#### 2.1 YELLOW SIGATOKA DISEASE

Yellow Sigatoka, caused by *M. musicola*, was first identified by the anamorph stage, originally named as *Cercospora musae* (Zimm.) Deighton in Java (Zimmermann, 1902). *M. musicola* was first reported from the Sigatoka Valley, Fiji in 1912 (Leach, 1941), where it caused an epidemic. Banana leaf spot caused by *Cercospora musae* associated with banana leaf spot in Madagascar (Bouriquet and Bataille, 1958). The disease was named as Sigatoka disease or simply Sigatoka, after an outbreak of the disease in the Sigatoka valley on the island of Viti Levu in Fiji in 1912 and the spread of Sigatoka disease around the world to all banana growing regions is well documented by Meredith (1970).

The disease was then detected in banana plantations of Australia 1924 and in Surinam and Trinidad in 1934 and, by 1937; it had reached most parts of the Caribbean, Central America, Colombia and Venezuela. After a short time, the disease had spread to the neighbouring countries of Brazil (1944), Peru (1946) and Ecuador (1952).

*M. fijiensis* has been reported from sites where *M. musicola* was formerly present, suggesting a gradual displacement of *M. musicola* to higher altitudes (inter-Andean valley populations). Therefore, the dynamics of population structure of both pathogens were in some way interdependent and most likely to be influenced by parameters common to both.

Sigatoka disease had been reported in most of the banana growing areas of the world, since 1962, making it one of the important epidemic plant diseases (Stover, 1980). Yellow Sigatoka has the widest distribution though black Sigatoka is rapidly replacing it in many tropical coastal regions (Carlier *et al.*, 2000; Jones, 2000). The disease caused by *M. musicola* known as Sigatoka disease and more recently as yellow Sigatoka to distinguish it from black Sigatoka. The causal organism may have arisen in South East Asia / Australian region (Jones, 2000).

Hayden *et al.* (2003) examined the population differential in the Sigatoka pathogen (*M. musicola*) on global scale and observed that yellow Sigatoka disease originated in South East Asia from where it was disseminated between Africa, Latin America and Caribbean region. Hayden *et al.* (2003) concluded that intercontinental spread via airborne spores from Australia, as proposed by Stover (1962) is improbable and that instead gene flow between continents is likely to have occurred from movement of infected plant material. *Mycosphaerella* spp. that infect banana induce necrosis of leaf tissue leading to a reduction in the photosynthetic capacity of the plant and also cause physiological changes that contributes to premature ripening of the fruits (Marin *et al.*, 2003). Hayden *et al.* (2005) reported that Sigatoka disease caused by *M. musicola* is endemic to Australia.

Leach (1946) reported that number of hands, fingers as well as size of fingers was reduced and finger are not fully filled and the bunches remained small and stunted and the fruits became soft in bunch showing the tendency of premature ripening in Sigatoka affected plants.

#### 2.1.1 Incidence and Intensity of Sigatoka Disease

With regard to economic impact of Sigatoka disease, heavy losses were met with in 1956 on account of banana leaf spot caused by *C. musae* (Anon. 1957). Between 1937 and 1941, production in Mexico was halved as a direct result of Sigatoka disease. In Honduras, production declined to less than one third of the pre - disease level (Meredith, 1970). Perfoura *et al.* (1993) reported 50 - 100 per

cent loss in banana production in Ghana due to Sigatoka disease. Economic losses of more than 50 per cent due to yellow Sigatoka were reported worldwide (Burt *et al.*, 1997).

With regard to Sigatoka disease status in India, Rangaswami and Kolandaiswamy (1962) reported the occurrence of Sigatoka disease caused by *C. musae* (Zimm.) in the Annamalai areas of Madras State. In India, Sigatoka disease was first reported from Assam in 1962 (Meredith, 1970).

Suharban (1977) recorded the incidence of Sigatoka along with other leaf spot diseases in the banana orchard of Instructional Farm of College of Agriculture, Vellayani in Thiruvananthapuram district.

A severe incidence of the disease was recorded from Gujarat during 1976 to 1982, causing drying and defoliation of leaves and premature ripening of fruits in bunches (Vala, 1996). A preliminary survey conducted in peninsular India confirmed the occurrence of yellow Sigatoka in addition to black Sigatoka (Selvarajan *et al.*, 2000). The recorded disease severity indices are 14.4 and 49.64 in Nendran and Robusta cultivars respectively for black Sigatoka, in Thrissur district.

The leaf spot diseases are particularly threatening in tropical areas where banana is a staple food (Crous *et al.*, 2002).

In an experiment on estimation of yield loss due to Sigatoka disease caused by *M.musicola*, plants in the control plot which did not receive any chemical spray, recorded the maximum Percentage Disease Index (PDI) of 41.38 per cent (Thammaiah *et al.*, 2005)

Sahlen and Soemargono (2011) reported that severity level of black Sigatoka increased from 15 per cent to 62.31 per cent in Northern Sumatra.

#### 2.1.2 Symptomatology and Disease Development

Various accounts of symptoms of Sigatoka were given previously by Campbell (1926), Simmonds (1933), Wardlaw (1939), Leach (1941,1946) and Brun (1958) in which they described the presence of a profusion of small discrete spots on the lamina of older leaves with areas of scorched or brown leaf tissues where spots were closely grouped together. It was particularly noticed that the two youngest leaves or heart leaf of an actively growing plant were usually free from evident spotting and also from minute specks.

Warldlaw (1941) observed that elliptic spots usually appear on the fourth to sixth leaf. On lower leaves of younger plants, larger and broader spots are formed.

Symptom development from specks through streaks to spot has been developed in to six stages (Leach, 1946; Klein, 1960; Brun, 1963; Stover, 1972) have compared these stages.

#### 2.1.3 Etiology

*M. musicola*, one of the most severe fungal pathogens of plantain and banana, is the major cause of economic losses in commercial plantations and in numerous smallholdings.

The Sigatoka leaf spot disease is caused by the ascomycetes fungus *M*. *musicola* R. Leach ex J.L. Mulder and is present in all continents (Jones, 2003).

The morphological characters of anamorph stage of the three species of *Mycosphaerella* is reported by Crous and Mourichon (2002) and Churchill (2011).

The anamorph of *M. musicola*, *Pseudocercospora musae*, lacks the thickened cell walls and the condiophores produce dense fascicles (sporodochia) on a dark brown or black stroma (Stover, 1972).

Stover and Fulton (1966) reported that ascospores of *M. musicola* from the air are deposited mostly on the lower surface of the apical third of the furled heart leaf, resulting in infection streaks along the margin of the leaf lamina.

The disease cycle begins with spore germination *i.e.*, when conidia or ascospores germinate on leaf surface. Germination is highly dependent on moisture and temperature; stomatal penetration by germ tubes which occurs when temperatures are above  $20^{\circ}$ C for 2 - 3 days and moisture is at near 100% relative humidity. Both the conidia and ascospores play roles in the spread of the disease and cause successful infection and a perfect correlation found in between the incidence with humidity (Stover, 1980). After germination, both species undergo a period of epiphytic growth. In *M. musicola*, this is 2 - 3 days for ascospore germ tubes (Brun, 1963) and 4 - 6 days for conidial germ tubes (Jones, 2000).

Subsequently, the germ tube produces an appressorium over a stomatal pore through which the fungus directs a fine, infection hypha. Once in the palisade layer, the hypha proliferates, growing into the air chambers between the veins (Stahel, 1937). At this stage, the first symptoms become apparent as small yellow specks (*M. musicola*) or red-brown specks (*M. fijiensis*). The transition period is the time taken for symptom development from streaks to spots. Brun (1963) observed lengthening transition periods for *M. musicola* when temperatures decreased. Shorter transition periods were observed for both *M. musicola* (Leach, 1946) and *M. fijiensis* (Four'e, 1987) when inoculum levels increased.

Conidia of *M. musicola* are produced within a sporodochium at the first brown spot, stage 4 of the disease cycle (Brun, 1958). Sporodochia are produced on both leaf surfaces within the substomatal air chambers, and the conidiophores grow out through the stomatal pore. Conidial production in *M. musicola* can occur over a wide range of temperatures, provided water is present as dew or rainwater (Stahel, 1937). Perithecium production is strongly linked to 'wet season' conditions of high rainfall and temperatures greater than 21° C, and is thus only observed for short periods of the year (Stover, 1968). Spermagonia and perithecia of *M. musicola* are produced in the final disease stages, when mature lesions start to coalesce and leaf tissue dries out. The production of both spore types is dependent upon moisture and temperature.

# 2.2 INFLUENCE OF WEATHER PARAMETERS ON SIGATOKA DISEASE OF BANANA CAUSED BY *Mycosphaerella musicola*

Climatic differences between different areas can also significantly affect symptom expression of the Sigatoka diseases (Vakili, 1968; Shephered, 1990).

Plant diseases are one of the important factors which have direct impact on global agricultural productivity and climate change - the biggest threat will further aggravate the situation. Local weather conditions such as rain, temperature, sunshine and wind in combination with locally adapted plant varieties, cropping systems and soil conditions can maximize food production as long as plant diseases can be controlled. Increase in temperature with sufficient soil moisture may increase evapotranspiration resulting in humid microclimate in crops and may lead to incidence of diseases favored under these conditions (Mcelrone *et al.*, 2005).

The speed of development of fungus is closely related to temperature (Ganry, 1973, 1975).

Stover (1968) reported that the production of perithecia and sporodochia of *M. musicola* in Honduras was maximum in July - October and declining in November – December regardless of rainfall, after the temperature dropped below 21°C. Extent of perithecia formation was found to increase two to three weeks after a wet spell lasting atleast two days and were found to be more abundant in mass spotted leaf tissue than in scattered spots. They were more frequent on the upper surface of single spots and there was no consistent difference in ascospore discharge from upper and lower surfaces of mass infected leaves. When the leaves were thoroughly wetted, ascospore discharge commenced in 10 minutes, 85 per cent being discharged in two hour.

The release of *M. musicola* conidia from the sporodochia requires the presence of water as rain or dew or may be achieved by buffeting in high winds (Leach, 1946).

*M. musicola* and *M. fijiensis* differ in their requirements for moisture during germination: *M. musicola* requires a film of free water for conidial germination (Leach, 1946; Stahel, 1937) and 95% relative humidity for ascospore germination (Brun, 1963). Conidia are produced continuously throughout the rainy season and disseminated through a film of free water, resulting from either rain water or dew dripping on the nearby healthy leaves which aid to faster spread of pathogen (Simmonds, 1966). Conidia are water dispersed, washed downwards to infect younger plants underneath, or splash-dispersed onto younger leaves above (Stover and Fulton, 1966).

Stover (1968) correlated perithecium formation with seasonal rainfall and minimum temperature, declining from December or January to low levels in March and April.

Both species require rain or free water for the ejection of ascospores from perithecia (Stover, 1980). Conidia of *M. fijiensis* have also been detected in airspora (Burt *et al.*, 1997). Ascospores are wind disseminated and can be carried upwards on air currents to younger leaves or dispersed over long distances by laterally moving air currents (Leach, 1946; Brun, 1963; Four'e, 1987).

Further, Elangovan *et al.* (1990) reported severe incidence of Sigatoka disease in high rainfall areas of Tamil Nadu. These findings were in agreement with Jacome *et al.* (1991) who stated that conidia become more important and only means of disease spread during dry periods. Also due to existence of less conducive climatic conditions the disease development could have been delayed by such dry conditions.

In case of both yellow Sigatoka and black Sigatoka, the critical weather factors determining severity are RH and temperature (Wardlaw, 1961; Foure, 1994).

According to Stover (1980), infection was enhanced at temperature above 20°C while dry weather and temperature below 20°C slowed less disease development (Jacome and Schuh, 1992).

# 2.3 EFFECT OF FUNGICIDES ON SIGATOKA LEAF SPOT DISEASE, BIOCHEMICAL AND YIELD PARAMETERS

Chemical control of the Sigatoka leaf spots has evolved considerably since the mid 1930's. Bordeaux mixture, the first effective fungicide, had to be applied as a high volume spray and large pipeline systems were installed at great expense to deliver the chemical in plantations. Later, petroleum oil applied as a low volume spray from aircraft proved effective and costs were reduced, especially when combined with forecasting systems. Protectant fungicides, such as dithiocarbomates, and systemic fungicides, such as benomyl, improved the standard of control. However, control measures, which included labour intensive cultural practices, such as pruning old diseased leaves, were an expense that was borne by growers. In 1990 in Queensland, where Sigatoka disease is still the dominant leaf spot disease, control measures were estimated as 14 % of total production costs.

Pont (1958) applied copper oxychloride under high pressure at high volume as a protectant against *C. musae* (*M. musicola*) and the same with wettable sulphur were found to be equally efficient. Moreover copper fungicides were found to be generally better than organic fungicides. Regular spraying of the heart leaves, especially during late summer and autumn months with about four week's interval between each application brought the disease under control. Calpouzos *et al.*, (1959) studied the action of oil in the control of *M. musicola* on banana leaves and a complete control achieved by chemicals containing up to 10-25% aromatic compounds.

Klein (1961) applied Dithane M 22, Tribasic copper sulphate, Microgel, COCS, Texaco 522, and ESSO oil from air and Bordeaux mixture from the ground, timing of application being based on streak counts of *M. musicola*. He also observed control of Cercospora leaf spot of banana with oil sprays based on the disease cycle.

Rhodes (1961) reported control of Cercospora leaf spot of banana with oil sprays based on the disease cycle.

Calpouzos *et al.*, (1961) found the relation of petroleum oil composition to phytotoxicity and control of Sigatoka disease on banana leaves.

Tollenaar (1961) studied the efficacy of copper and oil in the control of Sigatoka banana leaf spot and found that the use of oil had a negative effect on growth, production, fruit weight and production of new suckers.

Kranz (1966) conducted field tests to control Sigatoka disease of banana. Low volume sprays of copper oxychloride, copper oxide, maneb, zineb and thiram- ziram urbacide in water were found to be inadequate against the disease while fentin acetate, though nearly as effective as mineral oil proved to be phytotoxic. Mineral oil (alone or as 40:50 emulsion with water) gave complete protection when sprayed at 10 day intervals. The same effect was achieved by spraying with a 50 per cent solution of copper oxychloride at a concentration of  $30 \text{ g l}^{-1}$  of oil (or 40:50 oil water emulsion) at 15 day intervals, with a reduction in labour and quantity of oil. By applying the treatment at an appropriate stage of symptom development, the number of sprays needed per year could be reduced.

Vonprahil (1967) reported that antracol gave results as good as those with Maneb against infection of *M. musicola*. In trials with oil sprays against

ascospore, antracol was found to be better than other products tested and was less phytotoxic. No sticker was necessary in the antracol:oil mixtures and the fungicidal film withstood several days of heavy rain.

Brodrick and Kuhne (1971) reported that Sigatoka in banana could be profitably controlled. Effective control of *M. musicola* and 7.1 per cent increase in banana yields were obtained with semi - concentrated sprays of Mancozeb and effectiveness was maintained 4 months after each application.

Melin and Tezenasdumontcel (1975) gave results of aerial application of different fungicides against Cercospora disease of banana. In field trials, derosal, folcidine and bavistin at 150 g a.i ha <sup>-1</sup> and tecto flow at 200 g ha <sup>-1</sup> were found to give satisfactory control of *M.musicola*.

Melin *et al.*, (1976) worked on the comparative action of Imazalil with other fungicides on Cercospora infection of banana in Cameroon. In aerial spraying, Imazalil at 300 g a.i. ha <sup>-1</sup> in 10 litre oil gave good control. The product was suggested to be a good replacement for benzimidazoles in the event of development of resistant strains of the fungus or to prevent this contingency.

The first triazole fungicide, propiconazole (Tilt), was introduced for use on banana in 1984. It had high post- infection activity and this new fungicide allowed producers to reduce the number of applications in a plantation from as many as 45 to fewer than 20 applications per year. Propiconazole and other sterol demethylation inhibitors (DMIs) now comprise the largest group of systemic fungicides that are used on banana.

West (1983) reported that propiconazole (Tilt) gave good control of leaf spot caused by *M.musicola*. Eswaramurthy *et al.*, (1988) reported that application of carbendazim and aureofungin had brought about the reduction of banana wilt and Sigatoka leaf spot in banana.

Beginning in the late 1950's petroleum oils began to be used in Sigatoka control programs. Oils also assist the penetration of the leaf by systemic fungicides, reduce conidium germination, germ tube growth and appressorium formation, and increase the pathogen's incubation period. Finally, they retard the growth and development of the pathogen within the host leaf. The introduction of benomyl and subsequent systemic heralded a new era of Sigatoka control (Stover, 1990). Unlike the protectants, systemic inhibited the development of symptoms after they first appeared.

Despite the large number of triazoles that have been tested against black Sigatoka [e.g., bitertanol (Baycor), cyproconazole (Alto), difenoconazole (Score), flusilazole (Punch), hexaconazole (Anvil), metconazole (Caramba), and tebuconazole (Folicur)], propiconazole remains the most frequently used fungicide of this group (Romero and Sutton, 1997).

Chemical control with fungicides such as triazoles, benzimidazoles and strobilurines can be very efficient at suppressing the development of these diseases, especially with the use of aerial application and forecasting strategies (Ganry *et al.*, 2008).

Chemical control by spraying fungicides on a preventive or curative basis is the only acceptable strategy adopted in the management of this disease (Duvert *et al.*, 2002).

Continuous use of the systemic fungicides is reported to increase the risk of development of resistance to these fungicides in both *Mycosphaerella musicola* and *Mycosphaerella fijiensis* (Mourichon *et al.*, 1997 and Duvert *et al.*, 2002). The toxicant produced by application of systemic fungicides inhibit protein synthesis by binding to the larger ribosomal subunits including change in the enzyme system, ceasing ATP and NADP formation (Siddiqui, 1997) leading to decreased disease incidence with increase in yield.

Ramsey *et al.* (1987) reported the efficacy of propiconazole 50 g a.i ha<sup>-1</sup> for controlling the premature ripening of banana due to Sigatoka, the bunch weight was increased compared with untreated plants.

Management practices like the use of resistant cultivars, cultural methods and quarantine methods have been recommended to control Sigatoka leaf spot disease in banana, but the most reliable tool has been the use of chemical products, mainly site-specific fungicides (Jones, 2000; Gasparotto *et al.*, 2006).

In Karnataka at Munavalli and Ramadurga two sprays of propiconazole 0.05% effectively contolled the disease and recorded the lowest percent disease index (Tammaiah *et al.*, 2008).

Patel (2009) concluded that propiconazole, hexaconazole, tridemorph and carbendazim remained significantly at par with each other and recorded least leaf spots followed by tebuconazole and companion (combination of mancozeb and carbendazim).

# 2.4 EFFECT OF SECONDARY AND MICRO NUTRIENTS ON SIGATOKA LEAF SPOT, PHYSIOLOGY AND BIOMETRIC PARAMETERS

According to Huber (1980), plant nutrition has a big effect on the plant's susceptibility to disease and helps to mobilize disease resistance. Micronutrients play an important role in plant metabolism by affecting the phenolics and lignin content and also membrane stability (Graham and Webb, 1991). The effect of micronutrients on reducing the severity of diseases can be attributed to the involvement in physiology and biochemistry of the plant (Marschner, 1995) and influencing the disease resistance indirectly.

It is important to manage nutrient availability through fertilizers or change the soil environment to influence nutrient availability, and in that way to control plant disease in an integrated pest management system (Graham and Webb, 1991; Huber and Graham, 1999). Nutrients can affect disease resistance or tolerance. Disease resistance of the host is its ability to limit the penetration, development and reproduction of the invading pathogens (Graham and Webb, 1991). On the other hand, tolerance of the host is measured in terms of its ability to maintain its own growth or yield in spite of the infection. Nutrients are important for growth and development of plants and also microorganisms and they are important factors in disease control (Agrios, 2005).

The micronutrients (Fe, Mn, Zn, Cu, B, Mo and Cl) are essential but the plant requires them in minute amounts (Fritz, 1985).

Banana is always referred to as a gross feeder and requires large amounts of nitrogen and potassium followed by phosphorus, calcium and magnesium to maintain high yields (Robinson, 1996; Abdullah *et al.*, 1999).

The micronutrients boron, copper, iron, manganese, molybdenum and zinc act as activators of many plant functions and major functions of micronutrients in banana are listed below. Boron is necessary for translocation and promotes fruit maturity. Copper catalyzes several plant processes like photosynthesis, development of reproductive stage, indirect role in chlorophyll production, increases sugar content, intensifies color and improves flavor of the fruits on ripening. Iron promotes formation of chlorophyll pigments, acts as an oxygen carrier and reactions involving cell division and growth. Zinc aids in regulating plant growth hormones and enzyme system, necessary for chlorophyll production, carbohydrate and starch formation. Manganese functions as a part of certain enzyme systems and aids in chlorophyll synthesis. Molybdenum required to form the enzyme "nitrate reductase" which reduces nitrates to ammonium and needed for converting inorganic phosphates to organic forms in plants.

Micronutrients have considerable influence on resistance or tolerance mechanisms to pathogens in plants (Krauss, 1999). The use of fertilizers produces a more direct means of using nutrients to reduce the severity of many diseases and together with cultural practices can affect the control of diseases (Atkinson and McKinlay, 1997; Obron *et al.*, 2003).

Plants are deficient when some of these essential nutrients are not supplied or undersupplied while oversupply results in toxicity problems (Stevens *et al.*, 2002). The location of nutrient deficiency symptoms depends on the mobility of the nutrient. Therefore some macronutrients (N, P, K, Mg) become deficient in the older leaves while other elements (Ca, Zn, B, Mn) become deficient in new leaves when supply is insufficient (Mengel, 2002). Different soil conditions influence availability such as pH, texture, moisture, temperature, as well as mineral solubility, and soil microbial activity.

Plants with balanced fertilization are less susceptible to diseases while plants with unbalanced fertilization become more susceptible to diseases (Henn, 2004).

Stresses associated with mineral deficiencies, water supply (excess or shortage) and heavy defoliation have a very clear impact on fruit growth rate (Chillet *et al.*, 2006).

Nutrients are involved in different processes like enzyme activation, and metabolic regulation but also in structural components (Moreno *et al.*, 2003; Daurob and Snyder, 2007).

#### 2.4.2.1 Boron (B)

B deficient plants are more susceptible to powdery mildew and the fungus also spreads more rapidly over the plant. It was also observed that in B- deficient wheat plants, the disease severity was several fold higher than that in B- sufficient plants, with the fungus spreading more rapidly than in B-sufficient plants (Schutte, 1967).

Boron deficiency is the most widespread micronutrient deficiency in the world (Brown *et al.*, 2002).

It is a structural component because it promotes the stability and rigidity of the cell wall. B is involved in the lignin synthesis, cell wall crosslinking of pectin polymers (Romheld and Marschner, 1991; Blevins and Lukaszewski, 1994; Brown *et al.*, 2002; García-Hernández and Cassab-López, 2005) and the integrity of the plasma membrane (Brown *et al.*, 2002; Dordas and Brown, 2005).

Boron reduces diseases caused by *Plasmodiophora brassicae* in crucifers, *Fusarium solani* in bean, *Verticillium albo-atrum* in tomato and cotton, tobacco mosaic virus in bean, tomato yellow leaf curl virus in tomato, *G.graminis* (Sacc.) (Graham and Webb, 1991) and *Blumeria graminis* in wheat (Marschner, 1995).

Nutrients can affect the physiology and biochemistry and especially the integrity of the cell walls, membrane leakage and the chemical composition of the host, e.g., the concentration of phenolics can be affected by B deficiency. The role of Boron in reducing the severity of many diseases because of the function that B has on cell wall structure, cell membrane permeability and its role in metabolism of phenolics or lignin (Blevins and Lukaszewski, 1998; Brown *et al.*, 2002).

Boric acid (1% - 5%) was shown to be a good inhibitor of ascospore germination under *in vitro* conditions and a potential alternative for benlate. Similar results were found under field conditions against ascospore germination of the pathogen *Eutypa lata* which causes the dieback disease of grapevines (Rolshausen and Gubler, 2005).

Boron has role in lignification and as a structural component it seems that it is involved in forming a barrier against pathogen invasion, though a profound understanding of the physiological and biochemical mechanisms in plant defense are still lacking (Stangoulis *et al.*, 2007).

#### 2.4.2.2 Zinc (Zn)

The micronutrients influence disease resistance indirectly, as nutrient deficient plants not only exhibit an impaired defense response, but often may also become more suitable host plant for feeding as many metabolites such as reducing sugars and amino acids leak outside the plant cell. For example, plants suffering from a Zn deficiency showed increased disease severity after infection by *Oidium* spp. (Bolle- Jones and Hilton, 1956).

Zinc is essential for carbohydrate formation and an enzyme activator involved in protein, hormone, RNA and DNA synthesis and growth regulation (Marschner, 1995; Graham and McDonald, 2001).

Leakage of sugars onto the surface of leaves increases the severity of infections of powdery mildew. Up to an extend, Zn applications can reduce the severity of such pathogens. In most cases, the application of Zn reduced disease severity, which could be because of the toxic effect on the pathogen directly and not through metabolic changes. Wheat plants receiving Zn via soil applications exhibited a reduced infection by *Fusarium graminearum* and root-rot diseases caused by *Gaeumannomyces graminis* (Graham and Webb, 1991).

Numerous reports confirmed the role of Zn in relation to plant diseases. In wheat high concentrations of Zn decreased *Rhizoctonia* root rot (Thongbai *et al.*, 1993).

Zn plays an important role in protein and starch synthesis, and therefore a low zinc concentration induces accumulation of amino acids and reducing sugars in plant tissue (Marschner, 1995).

Application of Zn to the soil reduced infections by *F. graminearum* and root rot diseases, e.g., caused by *G.graminis* (Sacc.) in wheat (Graham and Webb 1991; Grewal *et al.*, 1996).

In upland rice, the occurrence of panicle blast (*Pyricularia grisea*) was inversely related to the Zn content in the plant (Filippi and Prabhu, 1998). The Zn content is inversely associated with citrus blight also (Tucker *et al.*, 1984; Derrick and Timmer, 2000).

As an activator of Cu/Zn-SOD, Zn protects the cell membrane against oxidative damage through the detoxification of superoxide radicals (Cakmak, 2000; Mengel and Kirkby, 2000). The application of Zn (4 kg ha<sup>-1</sup>) or Mo (0.1 kg ha<sup>-1</sup>) increased the biomass production, LAI, CGR and yield attributes helped to maintain higher LAI towards the later stages of groundnut.

In *Medicago truncatula*, Zn did not directly inhibit *R. solani* but stimulated root development partly offsetting fungal damage (Streeter *et al.*, 2001). However, in the same crop and in rotation with pasture, Zn reduced two other diseases: root rot disease and common leaf spot disease caused by *P. megasperma* f. sp. *medicaginis*, and *Pseudopeziza medicaginis* respectively (Grewal, 2001).

High Zn concentrations in the soil affect the growth and metabolism of beneficial microorganisms, with consequences on plant health and biological control (Knight *et al.*, 1997; Moffet *et al.*, 2003).

*Macrophomina phaseolina, Fusarium solani* and *Rhizoctonia solani* in tomato were reduced by increasing Zn concentrations in the soil (Siddiqui *et al.*, 2002).

Zn sulphate was effective in reducing potato late blight caused by *Phytophthora infestans* and *Phytophthora* root rot of avocado (Baider and Cohen, 2003).

#### 2.4.2.3 Magnesium (Mg)

Its role in plant resistance remains unclear and there are some reports linking it to plant diseases caused by *P. brassicae* in crucifers (Myers and Campbell, 1985).

In wheat, Mg deficiency predisposes it to the take-all disease caused by *G*. *graminis* (Huber, 1989).

Magnesium is essential for chlorophyll and enzyme activation for photosynthesis (Shaul, 2002).

#### 2.4.2.4 Manganese (Mn)

Manganese containing superoxide dismutase catalyzes the disproportion of superoxide free radicals ( $O_2^-$ ) termed in many biologic oxidations during disease incidence and appears to play a vital role in protecting cells against deleterious effects of this radical (Fridovich, 1974).

Manganese appears to be involved in at least two steps in the biosynthetic pathways leading to lignin, its deficiency can increase susceptibility to pathogen invasion (Graham, 1983).

Wheat plants in Mn deficient soils were more susceptible to *G. graminis* (causal organism of take-all disease) and Mn sulphate reversed that situation by detoxifying the saprophytic survival of the fungus (Brennan, 1992).

Rengel *et al.* (1994) reported that Mn fertilization does not significantly influence the rate of phenolics and lignin accumulation but reduced depth of radial penetration by hyphae of *Gaemannomyces graminis* var. *tritici* in wheat.

A significant reduction in common scab development an increase in tuber yield was achieved in potato following soil application of MnSO<sub>4</sub> (Keinath and Loria, 1996).

Bag and Sinha (1997) reported that MnSO<sub>4</sub> had inhibitory effects on *Sclerotium rolfsii* disease by increasing total phenols and peroxidase activity.

Manganese controls lignin and suberin biosynthesis (Vidhyasekaran, 1997; Ehara *et al.*, 2000). Both the lignin and suberin are important biochemical barriers to fungal pathogen invasion (Kolattukudy *et al.*, 1994; Biggs and Rioux, 1994; Hammerschmidt and Nicholson, 2000; Viddhyasekaran, 2004), since they are phenolic polymers resistant to enzymatic degradation (Agrios, 2005).

Mn provides resistance against powdery mildew and take-all disease (Krauss, 1999).

Manganese is involved in enzyme activation, in chlorophyll formation, redox processes, and RNA and DNA synthesis. It is involved as a co-factor in many reactions (Porro, 2002).

Infection of cowpea by *Rhizoctonia solani* and *R.bataticola* was reduced after applications of Manganese sulphate. This was associated with increased polyphenol oxidase, peroxidase and total phenols (Kalim *et al.*, 2003). Manganese effectively reduced disease severity in case of take all patch disease on creeping bent grass (Heckman *et al.*, 2003).

Manganese soil applications reduce *Fusarium* spp. infections in cotton and *Sclerotinia sclerotiorum* in squash (Graham and Webb, 1991; Agrios, 2005).

2.4.2.5 Iron

Patel *et al.*, (1993) also found increase in leaf chlorophyll content of groundnut with one per cent ferrous sulphate and two per cent ferric citrate.

Application of FeSO<sub>4</sub> showed a significant increase in seed yield in soybean (Bhanavase *et al.*, 1994).

On calcareous soils of western Rajasthan, Kumawat *et al* (2006) noted that soil application of 25 kg FeSO<sub>4</sub> ha<sup>-1</sup> significantly increased iron concentration in green leaves of mung bean as compared to control, further N,P,K and S uptake by grain and straw also increased compared to control.

Sahu *et al* (2008) reported that application of  $FeSO_4$  @ 2 kg ha<sup>-1</sup> along with biofertilizer inoculation gave the highest grain yield (1473 kg ha<sup>-1</sup>) and straw yield (1423 kg ha<sup>-1</sup>) as compared to control in chickpea.

#### 2.4.2.6 Copper (Cu)

Copper is involved in chlorophyll synthesis. This metal is a component of plastocyanin, peroxidases, multi Cu-protein and several oxidases (Sandmann and Boger, 1980).

Cu is involved in the lignin biosynthesis of the cell walls (Marschner, 1995).

Harker *et al.*, (1990) reported that  $CuCl_2$  was an abiotic elicitor in the induction of chalcone synthetase, an enzyme necessary for the biosynthesis of different flavonoids, involved in plant disease resistance.

Physiological processes influencing disease resistance or susceptibility are not well understood but Cu is a regulator of various enzyme systems linked to plant defence and the production of antimicrobial compounds (Graham and Grahan, 1991; Lebeda *et al.*, 2001).

Many common diseases and disorders are associated with Cu. Copper amine oxidase was essential in the signaling molecules of  $H_2O_2$  (hydrogen peroxide) production in wound tissue of chickpea cv. Sultano infected by *Ascochyta rabiei* (Rea *et al.*, 2002).

Excess Cu causes inhibition of plant growth and problems in cellular processes like electron transport (Yruela, 2005).

Leaf rust (*Puccinia triticina* sp. *tritica*), tan spot (*Pyrenophora tritica-repentis*), and Fusarium head blight (*F. graminearum*) incidence and disease severity in wheat were reduced with Cu treatments (Franzen *et al.*, 2008).

### 2.4.4 Combined Effect of Secondary and Micro Nutrients in Disease Management, Physiological Response and Biometric Parameters

Chandel *et al.*, (1989) reported that the application of micronutrients increased the number of branches in soybean.

Zn applications and *P. fluorescens* together contributed to controlling *Fusarium* crown in wheat and tomatoes. A similar positive influence of Zn content in the soil and *P. fluorescens* 2-79 improved in wheat the biocontrol against *G. graminis* var. *tritici* (Thongbai *et al.*, 1993; Duffy and Defago, 1997; Ownley *et al.*, 2003).

Shoot dry weight of groundnut increased with combined application of iron, zinc and manganese in the sulphate form (Moussa *et al.*, 1996).

Soil amendments with Zn reduced the severity of maize smut, caused by *Ustilago maydis*, by over 10% and, in combination with N fertilizers, induced severity by over 20% (Kostandi *et al.*, 1997).

Systemic acquired resistance (SAR) may be involved in the suppression of plant disease by micronutrients. Reduction in disease severity has been reported in other crops after a single foliar application of H<sub>3</sub>BO<sub>3</sub>, CuSO<sub>4</sub>, MnCl<sub>2</sub> or KMnO<sub>4</sub>, which provided systemic protection against powdery mildew in cucumber plants. The application of nutrients such as Mn, Cu and B can exchange and therefore release Ca<sup>2+</sup> cations from cell walls, which interact with salicylic acid and activate systemic acquired resistance mechanisms (Reuveni *et al.*, 1997a, Reuveni and Reuveni, 1998).

The concentration of Zn and Cu were high in resistant cultivars as compared to susceptible ones. Similarly, Filippi and Prabhu (1998) noticed the low panicle blast severities of improved cultivars of rice were associated with high K and Zn and low N, P and Mg tissue concentrations.

The application of B, Mn and Zn separately increased the resistance of plants by providing systemic protection to tan spot (Dordas and Simoglou, 2006).

# 2.5 EFFECT OF PGPR ON DISEASE MANAGEMENT, PHYSIOLOGY AND BIOMETRIC PARAMETERS

An uninterrupted growth in terms of height and girth is an important character, which helps to judge the plant vigor (Simmonds, 1966).

The accumulation of phenolic compounds may be due to excess production of  $H_2O_2$  in infected plants through increased respiration or due to the activation of the hexose- monophosphate shunt pathway, acetate pathway and release of bound phenolic compounds by hydrolytic enzymes (Goodman *et al.*, 1967).

*Pseudomonas flourescens* are the most effective rhizosphere bacteria, because in addition to disease control, they exert beneficial effect on plant growth promotion (Kloepper *et al.*, 1980).

Banana roots are adventitious and proliferate horizontally in the topsoil and they cannot acquire nutrients and water from the deeper soil profile like other fruit crops. This undeveloped root system limits the large-scale production of bananas under adverse tropical conditions, where the root systems are crucial for plant support, nutrient and water acquisition and production of plant growth regulators by rhizobacteria (de Langhe *et al.*, 1983; Stover and Simmonds, 1987; Kapulnik, 1991; Price, 1995).

Biofertilizers, microbial inoculants that can promote plant growth and productivity is internationally accepted as an alternative source of N-fertilizer. It is environmental friendly and can be used to ensure a sustainable banana production. Nitrogen fixation and growth promotion by plant growth promoting bacteria are important criteria for an effective biofertilizer. Inoculation of associative and freeliving nitrogen fixing bacteria have been shown to produce beneficial effects on plant growth, thus they are termed as PGPR (Kloepper *et al.*, 1980; Bashan and Holguin, 1998).

*P. cepaciae* isolated from conidia of *Bipolaris maydis* obtained from infected corn leaves successfully controlled foliar diseases like Cercospora leaf spot of peanut and Alternaria leaf spot of tobacco (Blakeman and Fokkema, 1982).

In a banana bunch, the number of hands is an important yield component and is normally decided at the time of fruit bud initiation and differentiation which is generally influenced by better nutrition (Obeifuna, 1984).

Maintaining the physiological characters at a higher level is essential to increase biomass and banana yield. Higher chlorophyll content observed and it could be related to higher activities of iron containing enzymes such as catalase and peroxidase which are enhanced during the application of PGPR. The role of these enzymes in chlorophyll synthesis was an established factor (Singh, 1988).

*Pseudomonas* sp. and *Burkholderia* sp. Showed strong antifungal activity to plant pathogens including *R. solani* and this ability was correlated with the production of pyrrolnitrin by the bacteria (Hammer *et al.*, 1997).

Plants may also be exposed to elevated mineral concentrations applied to improve the beneficial activity of biological control strains of *P. fluorescens* (Duffy and Defago, 1997).

Significant increases in crop yields following application of PGPR have been documented under diverse field conditions (Bashan, 1998).

The plant growth promoting effects of PGPR are mainly derived from morphological and physiological changes in roots of inoculated plant (Okon *et al.*, 1988; Sarig *et al.*, 1988). The inoculation process could stimulate the root growth in bananas occurred almost in all dimensions namely production of primary and secondary roots, longer roots (42 %) and greater volume (48 %) and mass (43 %). Inoculation stimulated the reproductive growth as shown by early flowering (3 weeks) and increased bunch yield (51%). The earliness of flowering is attributed

to early development of plants with efficient nutrient and water uptake. The study reflected an additional 3 weeks saving in the maturation period of bananas. This indicated that PGPR stimulated plants for early reproductive development through enhanced nutrient uptake.

Similar results have been also demonstrated by Tiwary *et al.*, (1998), who inoculated of banana suckers with Azospirillum twice (sucker + soil inoculation). The result showed that there was an increase in plant height, leaf size in plants receiving 50% dose of nitrogen. Azospirillum inoculated plants produced higher number of hands/ bunch, compensated 50% of the recommended dose of nitrogen and the number of hands/bunch obtained was at par with double inoculation. Inoculation with Azospirillum produced maximum yield of banana (69.15 t ha<sup>-1</sup>).

Biofertilizers containing beneficial microorganisms are known to improve plant growth in many ways when compared to synthetic fertilizers by enhancing plant nutrient availability and thus help to sustain the eco-friendly environment and soil productivity (O'Connell, 1992).

In the past 40 years, greenhouse and field inoculation studies with PGPR have shown that these rhizobacteria are able to promote yield of agriculturally important crops grown under different soil and climatic conditions (Okon and Labandera- Gonzales, 1994).

To fulfill the plant demand for nutritional attributes it is essential to apply those elements in the soil, which mostly comes from inorganic sources. The increased use of chemical fertilizers is undesirable. Biofertilizers and biocontrol agents containing beneficial microorganisms are known to improve plant growth in many ways when compared to synthetic fertilizers by enhancing plant nutrient availability and thus help to sustain the ecofriendly environment and soil productivity apart from induction of disease resistance. The PGPR is a group of rhizosphere colonizing bacteria, which produce substances that increase the growth of plants and/or protect them against pathogens (Glick, 1995; Harish *et al.*, 2009a).

Rabindran and Vidhyasekaran (1996) assessed the effective dose of a peat based formulation of *P. fluorescens* for seed, root, soil and foliar application.

Plant growth- promoting rhizobacteria (PGPR) are the major root colonizers and most of the effective colonizers are from species of *Pseudomonas, Bacillus, Serratia* etc. The strains of *P. fluorescens* are known to survive both in rhizosphere and phyllosphere (Krishnamurthy and Gnanamanickam, 1998).

PGPR inoculation of plants resulted in increased rate of photosynthesis (Mia *et al.*, 2000).

Inoculation of oil seed (*Salicornia bigelovii* Torr.) with PGPR has been reported to increase plant biomass, palmitic acid, total N and protein content. The PGPR inoculation also increased P content through Phosphorus solubilization (Bashan *et al.*, 2000).

Shamsuddin *et al.* (2000) found increased amounts of P and K uptake in banana plants inoculated with PGPR.

The application of PGPR isolate increased yield in tomato (Murphy *et al.*, 2000), mango (Vivekananthan *et al.*, 2004) and in tea (Saravanakumar *et al.*, 2007) under field conditions.

Inoculation of rhizobacteria viz., P. fluorescens, Azotobacter cryoococcum and A. brasilense alone and in combination with root symbionts, Rhizobium sp. And Glomus mossae improved plant growth and reduced gall formation on chick pea - Cicer arietinum (Siddiqui and Mahmood, 2001). Successful management using the rhizobacterial isolate, *P. flourescense*, Pf1 reported, economically important plant diseases such as sheath blight of rice (Nandakumar *et al.*, 2001), anthracnose in chillies (Bharathi *et al.*, 2004).

Recently, PGPR strain UPMB 10 (*Bacillus sphaericus*), isolated from oil palm, observed to produce beneficial effects on plantation crops namely oil palm (Amir, 2001), coconut and banana (Mia *et al.*, 2005, 2007).

In the year 1995, Vidhyasekaran and Muthamilan developed the talc- based bioformulation of *P. fluorescens* strain Pf1 against root rot disease of chick pea. PGPR application results in successful management of various plant diseases *viz.*, red rot of sugarcane (Viswanathan and Samiyappan, 2001), sheath blight of rice (Radjacommare *et al.*, 2002), anthracnose of chillies (Bharathi *et al.*, 2004), anthracnose of mango (Vivekananthan *et al.*, 2004) and dry root rot disease on mung bean plants (Saravankumar *et al.*, 2007) using talc based bioformulation of strain Pf1.

It was found that application of mixtures of rhizobacterial and endophytic bacterial isolates to banana at the time of planting and during 3<sup>rd</sup>, 5<sup>th</sup> and 7<sup>th</sup> month after planting was effective in reducing banana bunchy top virus (BBTV) under field conditions, accounting for 52.4% reduction over control. PGPR treatments may induce resistance to the virus in the form of no symptoms, reduced symptoms and in some cases no detectable accumulation of virus (Murphy *et al.*, 2003). The reduction in BBTV disease incidence was probably due to the action of induced resistance in the host.

ISR elicited by PGPR has shown promise in managing a wide spectrum of plant pathogens, including virus in several plant species under green-house and field environments (Murphy *et al.*, 2003; Kavino *et al.*, 2008).

The efficacy of PGPR against fungal and bacterial diseases has been reported by various groups (Kloepper *et al.*, 2004).

Plant growth promoting rhizobacteria have recently been shown to induce systemic resistance (ISR) against fungi, bacteria and viruses, as well as to enhance plant growth. Combined application of arbuscular mycorrhizal fungi and PGPR highly benefits banana plants and therefore, could be considered during the acclimatization stage of micropropagated banana. Micropropagated banana plantlets from the 'Grand Naine' cultivar were inoculated with mycorrhiza and rhizobacteria either alone or combined and the results showed that combined inoculated plants showed growth parameters *i.e.*, total fresh weight, aerial dry weight, shoot length and leaf area, significantly higher than non-treated control bananas. Leaf mineral content (N, P and K) was also significantly increased by following the combined application of the same microorganisms in banana. The inoculation of PGPR showed better growth and seedling health and consequently increased the seedling survival rates (Rodriguez-Romero, 2005).

Biological control of plant pathogens is gaining momentum since chemicals raise environmental issues. Intensive farming practices that warrant high yield and quality require extensive use of chemical fertilizers and pesticides, which are not only costly but also pollute the environment. Recently there has been a resurgence of interest in environmentally friendly, sustainable agricultural practices (Esitken *et al.*, 2005).

PGPR inoculation study in bananas showed a significant amount of nitrogen  $(N_2)$  fixation by  $N_{15}$  isotopic dilution technique with 45 days old banana seedlings conclusively indicated that rhizobacterial inoculation with 33% fertilizer – N (50 ppm) could fix 8.85 to 9.69 mg N per plant. This rate of  $N_2$  fixation was further increased (10.26- 10.85 mg per plant ; 12.4- 12.5% Ndfa) with lesser inorganic- N supply, 13% fertilizer- N (20 ppm) due to a synergistic effect between the rhizobacteria and fertilizer- N (Mia *et al.*, 2007).

Enhanced PPO (poly phenol oxidases) activities against disease and insect pests have been reported in several beneficial plant-microbe interactions (Rajendran *et al.*, 2007).

Kavino *et al.* (2008) demonstrated that application of *Pseudomonas fluorescens* strain CHAO+ chitin bio-formulations significantly reduced the BBTV incidence in hill banana under greenhouse and field condition.

Among the various PGPRs identified, *P. fluorescens* is one of the most extensively studied rhizobacteria, because of its growth promoting activity and antagonistic action against plant pathogens (Kavino *et al.*, 2007; Saravanakumar and Samiyappan, 2007).

Several *Pseudomonas* strains were demonstrated to protect plants against many fungal, bacterial and viral diseases (Saravanakumar *et al.*, 2009).

Harish *et al.*, (2009a) observed that polyphenol oxidase (PPO) and peroxidase (PO) activities were greater in the plants treated with mixtures of rhizobacteria and endophytic bacteria and challenged with viruliferous aphids, compared to control plants. *P. fluorescens*, Pf1 along with the endophytic bacterial isolate significantly increased the yield compared to other treatments and the untreated control.

*P. fluorescens* strain CHAO + chitin bio-formulations of *Pseudomonas* strains improved the plant height, girth, LAI, bunch weight, number of hands and chlorophyll content when compared to untreated plants (Kavino *et al.*, 2010).

Banana, an important fruit crop, requires high amounts of chemical fertilizers for commercial cultivation, which is costly and can be hazardous to the environment, when used excessively. Plant growth promoting rhizobacteria (PGPR) could be used for growth promotion, nutrient uptake and some time as an alternative source of N-fertilizer of non-leguminous crops. Recently, research on PGPR for crop improvements are gaining prominence and thousands of research works have been published so far. However, use of this noble technique in banana production system is limited. Nevertheless, reports from various experimental findings suggested that PGPR strains could successfully formed colonies on the root surface of bananas, where more bacterial cells were found in the root hair

proliferation zone. Application of PGPR alone could not produce significant benefits that require minimal or reduced levels of fertilizer-N consequently could produce a synergistic effect on root growth and development. The inoculation also increased the N yield and fixed  $N_2$  in association with banana roots subsequently increased the yield, improved the physical attributes of fruit quality and initiated early flowering. PGPR are effective as a bioenhancer and biofertilizer for banana cultivation. For consistent and precise results extensive field experiments of bananas inoculated with PGPR strains should be continued. (Mia *et al.*, 2010)

#### **2.5.3 Effect of PGPR in Combination Treatments**

The bacterial biocontrol agents improve plant growth by suppressing either major or minor pathogens of plants (Defago *et al.*, 1990).

The inoculation process could stimulate the root growth and development (Mia *et al.*, 1998), which occurred almost in all dimensions namely production of primary and secondary roots, longer roots and greater volume and mass (43%).

PGPR play an important role in suppression of soil-borne plant pathogens, and improving nutrient availability (Biswas *et al.*, 2000; Saravanakumar and Samiyappan, 2007).

Plant growth promoting rhizobacteria have recently shown to induce systemic resistance against fungi, bacteria and viruses, as well as to enhance plant growth. Combined application of arbuscular mycorrhizal fungi and PGPR highly benefits banana plants and therefore, could be considered during the acclimatization stage of micropropagated banana (Rodriguez-Romero, 2005).

## **MATERIALS AND METHODS**

#### **3. MATERIALS AND METHODS**

The present study entitled "Integrated management of Sigatoka leaf spot disease of banana caused by *Mycosphaerella musicola* R. Leach ex J.L. Mulder", was conducted during the period 2012-2014 at College of Agriculture, Vellayani, Thiruvananthapuram and field experiment on integrated disease management was carried out at Coconut Research Station, Balaramapuram.

Intercropping system was adopted in a selected area of 0.34 ha, 50 year old coconut plantation with banana as intercrop.Disease free suckers of cultivar Nendran (AAB) of uniform age and size were used for planting. The experiment was carried out from March 2013 - December 2013. The field trial was conducted in a disease prone area of Sigatoka leaf spot disease. Details of the experiment are given below.

The experiment was laid out in RBD with 13 set of treatments with 3 replications. The layout of the experimental field is given in Fig.1 and general view of the experimental site is given as plate 2. The chemical properties of the soil are given in Appendix I.

Design : RBD Treatments : 13 Replication : 3 Plot size : 8 plants / replication Variety : Nendran (AAB) Spacing : 2 m x 2 m Number of Plants in field : 312

Plants were numbered in each of the replication including control plot in order to avoid the border effect. After three months of planting, the treatments were applied twice at the middle of June and at the end of July with an interval of 45 days. Foliar application of the treatments was given at the rate of 200 ml spray

R III

Т 13	Т 9	Т 7
Т 5	Т 3	Т 3
Т 2	Т 5	Т 9
T 11	T 12	Т 5
T 1	Т б	Т 10
Т 7	T 11	Т 8
Т б	T 2	T 11
Т 3	T 8	T 4
Т 4	Т 10	Т 13
T 12	T 4	Т 2
Т 8	Τ7	Т 12
T 10	T 1	T 1
Т 9	T 13	Т б

Fig.1. Layout plan of the experiment

Ν



Plate 2. General view of experimental plot

fluid per plant. At both times, all the treatments were given independently. *i.e*, all the secondary and micro nutrients were applied two days prior to the application of fungicides and bio control agent.

The following three different components were given as treatments *viz.*, Nutrients, Fungicides and BCA. Nutrients included Micronol, (Micronutrient mix of Zn - 7.06%, Mn - 4.20%, Fe - 2.60%, Cu - 2.00%, B - 0.60% and Mo - 0.05%). Magnesium (given as Magnesium sulphate @ 2 g  $l^{-1}$ ), Zinc (given as Zinc sulphate @ 3 g  $l^{-1}$ ) and Boron (given as Boric acid @ 2 g  $l^{-1}$ ).

The fungicides included two systemic new generation fungicides *viz.*, tebuconazole (Folicur 25 EC @ 0.1%), propiconazole (Tilt 25 EC @ 0.1%) and one contact fungicide copper hydroxide (Kocide 77% WP @ 0.25%).

PGPR Mix II @ 20 g  $l^{-1}$  was used to check out the efficacy of biocontrol agent in management of Sigatoka disease. The spray with Micronol was carried out two days prior to application of fungicide and the details of treatments are given in Table 1.

The parameters studied were grouped into four main categories namely disease factors, weather factors, biochemical or physiological responses and growth and yield characteristics. The data generated out of the field experiment and the subsequent chemical analysis was analyzed statistically using RBD for interpretation of results. Statistical techniques like analysis of variance applicable for RBD and correlation studies with weather parameters were employed to bring out the effect of secondary and micro nutrients, PGPR Mix II and various fungicides on growth and yield characters of the crop.

The materials used and methods followed for recording each factors are described below.

Sl. no.	Treatments
1	Micronol (2 g $l^{-1}$ ) plus tebuconazole 25 EC (0.1 %)
2	Micronol (2 g l <sup>-1</sup> ) plus copper hydroxide 77 % WP (0.25 %)
3	Micronol (2 g l <sup>-1</sup> ) plus propiconazole 25 EC (0.1 %)
4	Micronol (2 g $l^{-1}$ ) plus PGPR mix II (20 g $l^{-1}$ )
5	Mg $(2 g l^{-1}) + Zn (3 g l^{-1}) + B (2 g l^{-1})$ plus tebuconazole 25 EC $(0.1 \%)$
6	Mg $(2 g l^{-1}) + Zn (3 g l^{-1}) + B (2 g l^{-1})$ plus copper hydroxide 77% WP (0.25%)
7	Mg $(2 g l^{-1}) + Zn (3 g l^{-1}) + B (2 g l^{-1})$ plus propiconazole 25 EC $(0.1 \%)$
8	Mg $(2 g l^{-1}) + Zn (3 g l^{-1}) + B (2 g l^{-1})$ plus PGPR mix II $(20 g l^{-1})$
9	Tebuconazole 25 EC @ 0.1 %
10	Copper hydroxide 77 % WP @ 0.25 %
11	Propiconazole 25 EC @ 0.1 %
12	PGPR mix II (20 g $l^{-1}$ )
13	Absolute control (untreated check)

#### **3.1 DISEASE FACTORS**

#### **3.1.1 Disease Incidence**

The total number of leaves and number of infected leaves in each observation plant were separately counted and recorded to find out disease incidence. Monthly observations were recorded including one pretreatment observation (June) and six post treatment observations *viz.*, July, August, September, October, November and December 2013. The disease incidence was calculated using the following formula.

Disease incidence (%) = <u>Number of leaves infected X</u> 100 Total no: of leaves

#### **3.1.2 Disease Intensity (Disease Index)**

To find out the effect of treatments on the disease, disease intensity was calculated. For that, observation plants were selected from each replication excluding the border plants in order to avoid the border effect. Observations were taken at monthly intervals till December, 2013. The total observations included one pretreatment observation (June) and the rest were post treatment observations *viz.*, July, August, September, October, November and December.

A 0-6 scale was followed for scoring the Sigatoka leaf spot symptoms and the schematic representation of Gauhl's modification (1993) of the Stover's severity scale (1970) (Carlier *et al.*, 2002) is shown in Plate 3. The description of each grade is also shown in Table 2. The total number of leaves was scored as per the scale based on the area of infection.

Using the score values the extent of infection was estimated based on the proportion of the area affected by the leaf spot infection in relation to the total leaf area and calculated the disease index using the Gauhl's formula (1993) modified from Stover's Disease Severity Scale.

 Table 2. Score chart for yellow Sigatoka disease (Carlier et al., 2002)

Score	Description				
0	No symptoms				
1	Not more than 1% of leaf area affected				
2	Less than 5% of the leaf area affected				
3	From 6 to 15% of the leaf area affected				
4	From 16 to 33% of the leaf area affected				
5	From 34 to 50% of the leaf area affected				
6	More than 50% of the leaf area affected				

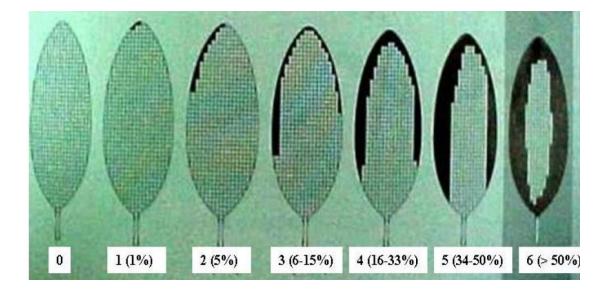


Plate 3. Gauhl's modification (1993) of Stover's scale (Carlier et al., 2002)

#### **3.2 BIOCHEMICAL ANALYSIS**

Biochemical changes in banana plants infected with *Mycosphaerella musicola* was studied with respect to the content of total phenol, ortho di-hydroxy phenol (OD phenol), chlorophyll, leaf nutrient and protein at different stages of crop growth.

#### **3.2.1 Estimation of Total Phenol**

The total phenol was estimated using leaf samples collected during active growth period  $(4^{th}, 5^{th} \text{ and } 6^{th} \text{ months})$ . It was estimated by following the procedure described by Bray and Thorpe (1954).

One gram of leaf sample was homogenized in 10 ml of 80 per cent ethanol. The homogenate was then centrifuged at 10,000 rpm for 20 min, supernatant was saved and residue was extracted with five times the volume of 80 per cent ethanol and centrifuged as above. The supernatant was saved and evaporated to dryness in a boiling water bath. The residue was dissolved in five ml distilled water. An aliquot of 0.2 ml was pipetted out and made up to three ml with distilled water. Folin-Ciocalteau reagent (0.5 ml) was added and two ml of 20% sodium carbonate solution was added to each tube after three minutes. This was mixed thoroughly and kept in boiling water for one minute. The reaction mixture was cooled and absorbance was measured at 650 nm against a reagent blank. Standard curve was prepared using different concentrations of catechol and expressed in catechol equivalents as milligram per gram leaf tissue on fresh weight basis.

#### 3.2.2 Estimation of ortho-dihydroxy phenol (OD- Phenol)

The ortho-dihydroxy phenol content was estimated using leaf samples during active growth period ( $4^{th}$ ,  $5^{th}$  and  $6^{th}$  months). It was estimated following the procedure described by Johnson and Schaal (1957).

To one ml of ethanol extract taken in a boiling tube, 1 ml of Arnow's reagent (Appendix II), 10 ml of distilled water and two ml of 1N NaOH were added. Soon after the addition of alkali, pink colour appeared. The color was diluted to 25 ml and read the transmittance at 522 nm in a spectrophotometer. A reagent blank was maintained with one ml of distilled water. A standard curve was prepared with catechol from which the unknowns were calculated.

#### 3.2.3 Estimation of Chlorophyll

Chlorophyll was estimated by Arnon's method (1949).

One gram of leaf sample collected at bunch emergence stage was finely cut and ground in a mortar with 20 ml of 80 per cent acetone. The homogenate was centrifuged at 5000 rpm for five minutes and the supernatant was transferred to a 100 ml volumetric flask. The above procedure was continued till the residue became colorless. The final volume in volumetric flask was made up to 100 ml with 80 percent acetone. Absorbance of the solution at 645 and 663 nm was read in a spectrophotometer against the solvent (80% acetone) as blank. The chlorophyll content was calculated using the following equations and expressed as milligrams chlorophyll per gram leaf tissue on fresh weight basis.

Chlorophyll a = 12.7 (A<sub>663</sub>) - 2.69 (A<sub>645</sub>) x  $\frac{V}{1000 \text{ x W}}$ Chlorophyll b = 22.9 (A<sub>645</sub>) - 4.68 (A<sub>663</sub>) x  $\frac{V}{1000 \text{ x W}}$ Total chlorophyll = 20.2 (A<sub>645</sub>) + 8.02 (A<sub>663</sub>) x  $\frac{V}{1000 \text{ x W}}$ 

Where

A= absorbance at specific wavelengths

V= final volume of chlorophyll extract in 80% acetone

W= fresh weight of tissue extracted.

#### **3.2.4 Estimation of Protein**

During the late vegetative stage, the protein content estimated using leaf samples. Total soluble protein content was estimated as per the procedure described by Bradford (1976).

One gram of leaf sample was homogenized in 10 ml, 0.1 M sodium acetate buffer ( $P^{H}$  4.7) (Appendix III) and centrifuged at 5000 rpm for 15 minutes at 4°C. The supernatant was saved for the estimation of soluble protein. The reaction mixture consisted of 0.5 ml enzyme extract, 0.5 ml distilled water and 5 ml of diluted (5 times) dye solution. The absorbance was read at 595 nm in a spectrophotometer against reagent blank. Bovine serum albumin was used as the protein standard. The protein content was expressed as milligram albumin equivalent of soluble protein per gram on fresh weight basis.

#### 3.2.5 Estimation of Plant Secondary and Micro Nutrients

One observation plant from each of the replication was selected for destructive sampling at bunch emergence stage to check out the leaf nutrient content. Mainly micronutrients such as Boron, Zinc and secondary nutrient such as Magnesium were chemically analyzed.

Petiole, the distal half portion were collected at bunch emergence stage, analyzed for their nutrient contents. For indexing the secondary nutrient, Magnesium and micronutrients such as Boron and Zinc the petiole of the third leaf was used (Sumam George, 1994). The sample leaves were cleaned, and dried in a hot air oven to constant weight at 70°C. The samples were powdered using plant sample grinding mill and were kept in brown paper cover for further analysis. The methods adopted for the chemical analysis are showed in Table 3.

Table 3. Standard	analytical n	nethods follo	wed in pla	ant analysis

Estimated character	Method adopted	Reference	
Magnesium (secondary nutrient)	Nitric – Perchloric acid digestion (9:4) and versanate titration	Piper (1967)	
Boron (micro nutrient)	Nitric – Perchloric acid digestion (9:4) and colorimetry	Bingham (1982)	
Zinc (micro nutrient)	Nitric – Perchloric acid digestion (9:4) and atomic absorption spectro photometry	Jackson (1973)	

#### **3.3 BIOMETRIC OBSERVATIONS**

#### **3.3.1 Growth Characteristics**

Growth characteristics on observation plants in each plot were recorded. Average of observation plants was worked out and presented. The following growth characters of the crop were recorded as detailed below.

#### 3.3.1.1 Height of the Plant (m)

Height of the plant was measured from the base of the stem at the soil level to the axil of the youngest unopened leaf during the harvest stage. Mean height was arrived at and recorded in meter.

#### 3.3.1.2 Girth of the Plant (cm)

Girth of the plant at 20 cm height above the soil level was measured using a flexible measuring tape during the harvest stage. Mean girth was arrived at and recorded in centimeter.

#### 3.3.1.3 Number of Leaves per Plant

The total number of leaves including both green and senescent ones was counted and the average number was recorded.

Monthly observations of number of leaves were recorded for a period of seven months during the crop season. Observations include one pre treatment observation and six post treatment observations. The first observation (June), made as pre treatment observation followed by the post treatment observations *viz*; July, August, September, October, November and December 2013.

#### 3.3.1.4 Number of Functional Leaves per Plant

The number of green leaves capable of photosynthesis was counted and whose average was recorded. Monthly observations of number of leaves were recorded for a period of seven months during the crop season. Observations include one pre treatment observation and six post treatment observations. The first observation (June), was the pre treatment observation. The following were the post treatment observations *viz*; July, August, September, October, November and December 2013.

#### **3.3.2 Yield Characteristics**

At harvest stage, observations like time of bunch maturity, bunch weight, number of hands, number of fingers per bunch, number of suckers were recorded.

#### 3.3.2.1 Time of Bunch Emergence

Mean number of days from planting to the stage of bunch emergence was calculated and recorded to check out whether the treatments had any influence on the same.

#### 3.3.2.2 Time of Bunch Maturity

Mean number of days from planting to the stage of disappearance of angles in fruit was calculated and recorded. The disappearance of angles followed by rounding of the fruit angles was taken as the indication of maturity (Stover and Simmonds, 1987).

#### 3.3.2.3 Yield per Plant (kg)

The bunch weight including the portion of peduncle to the first scar of observation plants was recorded and its average was expressed in kilograms.

#### 3.3.2.4 Number of Hands per Bunch

The number of hands in a bunch of banana observation plants from each replication and the absolute control were counted and whose average was recorded.

#### 3.3.2.5 Number of Fingers per Bunch

The total number of fingers in a bunch of banana observation plants from each of the treatment and absolute control was noticed and the average was recorded.

#### 3.3.2.6 Number of Suckers

The number of suckers produced for banana plants in each of the replication was counted and the mean data recorded.

#### **3.4 WEATHER PARAMETERS STUDIES**

3.4.1 Correlation Studies on the Influence of Weather Parameters on the Intensity and Incidence of Sigatoka Disease of Banana Caused by *Mycosphaerella musicola* 

Weather parameters such as maximum and minimum temperature, maximum and minimum relative humidity, rainfall, evaporation, sunshine hours prevailing in the field at the time of field trials over a period recorded and were statistically correlated with disease intensity (disease index) and disease incidence so as to study the influence of meteorological factors on the development of Sigatoka leaf spot disease of banana caused by *Mycosphaerella musicola*.

#### **3.5 ECONOMIC ANALYSIS**

The economic analysis of the crop in terms of net income and BCR was worked out as follows.

Net income (Rs. ha<sup>-1</sup>) = Gross income (Rs. ha<sup>-1</sup>) - Cost of cultivation (Rs. ha<sup>-1</sup>) BCR = Gross income (Rs. ha<sup>-1</sup>)  $\div$  Cost of cultivation (Rs. ha<sup>-1</sup>)

### RESULTS

#### 4. RESULTS

The present study on the 'Integrated management of Sigatoka leaf spot disease of banana caused by *M. musicola* R. Leach ex J.L. Mulder was conducted during the period 2012-2014 at the Department of Plant Pathology, College of Agriculture, Vellayani, Thiruvananthapuram, Kerala and at the experimental field at Coconut Research Station, Balaramapuram. The results obtained from the laboratory and field experiment are summarized below.

#### **4.1 DISEASE FACTORS**

The effect of the treatments on the disease intensity and incidence of yellow Sigatoka of Nendran banana was assessed under field condition.

## 4.1.1 Influence of Treatments on the Incidence of Banana Sigatoka Leaf Spot Disease Caused by *M. musicola*

The results of investigations on the incidence of Sigatoka leaf spot disease in CRS, Balaramapuram are detailed below:

Monthly observations of 13 treatments on the disease incidence of Sigatoka disease were recorded for a period of seven months. The first observation (June), made as pre treatment observation and the post treatment observations were made during July, August, September, October, November and December 2013 and the results are presented in Table 4.

During the pre treatment observation (June), there was no significant variation among the experimental plants with respect to disease incidence.

During the first post treatment observation, there was a significant difference in the incidence of disease in treatments when compared with the untreated plants.

Table 4. Effect of treatments on disease incidence of banana Sigatoka disease caused by Mycosphaerella musicola

Treatment		Disease incidnce (%)							
	Treatment	June	July	Aug	Sept	Oct	Nov	Dec	
T1	Micronol @ 2 g l <sup>-1</sup> plus tebuconazole @ 0.1%	44.61 (6.75)	31.21 (5.68) <sup>a</sup>	52.33 (7.30)	49.08 (7.08) <sup>abc</sup>	46.90 (6.92)	23.62 (4.96) <sup>a</sup>	53.04 (7.35)	
T2	Micronol @ 2 g l <sup>-1</sup> plus copper hydroxide @ 0.25%	42.52 (6.60)	44.16 (6.72) <sup>c</sup>	53.02 (7.35)	56.24 (7.57) <sup>d</sup>	57.02 (7.62)	48.93 (7.07) <sup>bcd</sup>	41.50 (6.52)	
T3	Micronol @ 2 g l <sup>-1</sup> plus propiconazole @ 0.1%	43.49 (6.67)	32.85 (5.82) <sup>ab</sup>	53.37 (7.37)	49.36 (7.10) abcd	46.47 (6.89)	47.10 (6.94) <sup>bcd</sup>	44.83 (6.77)	
T4	Micronol @ 2 g l <sup>-1</sup> plus PGPR mix II @ 20 g l <sup>-1</sup>	47.12 (6.94)	38.59 (6.29) <sup>abc</sup>	50.35 ( 7.17)	52.54 (7.32) <sup>bcd</sup>	50.89 (7.20)	51.42 (7.24) <sup>cd</sup>	50.07 (7.15)	
Т5	Mg + Zn + B plus tebuconazole @ $0.1\%$	40.34 (6.43)	36.00 (6.08) <sup>abc</sup>	53.64 (7.39)	49.98 (7.14) abcd	46.85 (6.92)	51.40 (7.24) <sup>cd</sup>	50.62 (7.25)	
T6	Mg + Zn + B plus copper hydroxide @ $0.25\%$	42.16 (6.57)	34.56 (5.96) <sup>ab</sup>	50.55 (7.18)	52.87 (7.34) <sup>bcd</sup>	49.70 (7.12)	51.37 (7.24) <sup>cd</sup>	49.86 (7.13)	
Τ7	Mg + Zn + B plus propiconazole @ 0.1%	42.52 (6.60)	38.08 (6.25) <sup>abc</sup>	51.01 (7.21)	45.41 (6.81) <sup>a</sup>	44.64 (6.76)	46.94 (6.92) <sup>bc</sup>	46.07 (6.86)	
Т8	Mg + Zn + B plus PGPR mix II @ 20 g l <sup>-1</sup>	41.25 (6.50)	37.06 (6.17) <sup>abc</sup>	52.88 (7.34)	55.89 (7.54) <sup>cd</sup>	53.21 (7.36)	54.38 (7.44) <sup>d</sup>	52.70 (7.33)	
Т9	Tebuconazole @ 0.1%	41.25 (6.50)	41.00 (6.48) <sup>bc</sup>	47.34 (6.95)	47.54 (6.97) <sup>ab</sup>	50.69 (7.19)	48.25 (7.02) bcd	44.72 (6.76)	
T10	Copper hydroxide @ 0.25%	37.28 ( 6.19)	33.92 (5.91) <sup>ab</sup>	51.23 (7.23)	52.14 (7.29) abcd	49.11 (7.08)	49.13 (7.08) bcd	41.09 (6.49)	
T11	Propiconazole @ 0.1%	43.71 (6.69)	37.83 (6.23) <sup>abc</sup>	52.62 (7.32)	53.17 (7.36) <sup>bcd</sup>	55.93 (7.55)	49.21 (7.09) bcd	42.96 (6.63)	
T12	PGPR mix II @ 20 g l <sup>-1</sup>	40.92 (6.48)	38.22 (6.26) <sup>abc</sup>	55.32 (7.50)	56.30 (7.57) <sup>d</sup>	55.77 (7.53)	45.04 (6.79) <sup>bc</sup>	44.92 (6.78)	
T13	Control	44.61 (6.75)	62.35 (7.96) <sup>d</sup>	63.23 (8.01)	64.00 (8.06) <sup>e</sup>	65.27 (8.14)	76.15 (8.78) <sup>e</sup>	85.55 (9.30)	
	SE (±)	-	0.24	0.20	0.16	0.56	0.17	5.89	
	CD (0.05)	NS	(0.70)	NS	(0.48)	NS	(0.50)	NS	

\*Mean of three replications \*\*Transformation used is square root transformation NS : Non significant

\*\*\*Figures followed by the same letter do not differ significantly according to two way ANOVA at P=0.05

The lowest incidence was found in treatment T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (31.21%) followed by T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (32.85 %), T10 - copper hydroxide @ 0.25% (33.92%), T6 - Mg + Zn + B plus copper hydroxide @ 0.25% (34.56%), T5 - Mg + Zn + B plus tebuconazole @ 0.1% (36.00%), T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> (37.06 %), T11 - propiconazole @ 0.1 % (37.83%), T7 - Mg + Zn + B plus propiconazole @ 0.1% (38.08%) , T12 - PGPR mix II @ 20 g l<sup>-1</sup> (38.22%) and T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> (38.59%), which were statistically on par and significantly superior over control. The highest disease incidence was observed in absolute control (62.35%).

During the second post treatment observation (August), where the treatments recorded a slight variation in disease incidence. The lowest disease incidence was found in T4 - Micronol @ 2 g  $l^{-1}$  plus PGPR mix II @ 20 g  $l^{-1}$  (50.35%) whereas the highest in absolute control (63.23%).

During the third post treatment observation (September), it was found that there existed a significant difference in the disease incidence between the treatments. The treatment, T7 - Mg + Zn + B plus propiconazole @ 0.1% (45.41 %) recorded the lowest disease incidence followed by T9 - tebuconazole @ 0.1% (47.54 %), T1- Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (49.08%), T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (49.08%), T5 - Mg + Zn + B plus tebuconazole @ 0.1% (49.98%) and T10 - copper hydroxide @ 0.25% (52.14%). All these treatments were statistically on par with each other. The remaining treatments are also found to be better than absolute control and the treatments are *viz.*, T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> (55.89%), T2 - Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25% (52.87%) and T12 - PGPR mix II @ 20 g l<sup>-1</sup> (56.30%). The highest disease incidence (64.00%) was noticed in absolute control.

During the fourth post treatment observation (October), it was revealed that there was no significant difference between any of the treatments. However, lowest disease incidence observed in T7 - Mg + Zn + B plus propiconazole @ 0.1% (44.64%) whereas the highest incidence was noticed in T13 - absolute control (65.27%).

The fifth post treatment observation (November) showed that there exist a significant difference between the treatments. All the treatments performed significantly superior than control (T13) having the highest disease incidence (76.15%). The lowest disease incidence was recorded by T1, Micronol (a) 2 g  $l^{-1}$ plus tebuconazole (a) 0.1% having a disease incidence of 23.62%, which was significantly superior from the remaining treatments. The following are the next best treatments viz., T12 - PGPR mix II (a) 20 g  $l^{-1}$  (45.04%), T7 - Mg + Zn + B @ 0.1% (46.94%), T3 - Micronol @ 2 g l<sup>-1</sup> plus plus propiconazole propiconazole @ 0.1% (47.10%), T9 - tebuconazole @ 0.1% (48.25%), T10 copper hydroxide @ 0.25% (49.13%) and T11 - propiconazole @ 0.1% (49.21%). All these treatments were statistically on par with each other. The performance of remaining treatments are in the following order viz., T6 - Mg + Zn + B plus copper hydroxide (a) 0.25% (51.37%), T5 - Mg + Zn + B plus tebuconazole (a) 0.1% (51.40%), T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> (51.42%) and T8 - Mg + Zn + B plus PGPR mix II ( $\hat{a}$ , 20 g l<sup>-1</sup> (54.38%). All the treatments were found to be better than absolute control having the highest disease incidence (76.15%).

In the last post treatment observation (December), it was found that there exists no significant difference between treatments. However, the lowest disease incidence was observed in the treatment, T10 - copper hydroxide @ 0.25% (41.09%) and a highest disease incidence was observed in control plot (85.55%).

With respect to disease incidence, among the 13 treatments; treatment T1 (Micronol @ 2 g  $l^{-1}$  plus tebuconazole @ 0.1%) showed good disease suppression during three months of observation. Followed by, T3 (Micronol @ 2 g  $l^{-1}$  plus

propiconazole (@ 0.1%), T5 (Mg + Zn + B plus tebuconazole (@ 0.1%) and T10 (copper hydroxide (@ 0.25%).

The result analysis revealed that the foliar application of treatments, tebuconazole in combination with either micronol (T1) or Mg + Zn + B (T5), Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (T3) and copper hydroxide @ 0.25% (T10) recorded the minimum disease incidence during the crop period.

# 4.1.2 Influence of Treatments on the Intensity of Banana Sigatoka Leaf Spot Disease Caused by *Mycosphaerella musicola*

The results of the observations on the effect of 13 treatments on reduction of disease intensity are detailed below.

Six post treatment observations on the effect of treatments on reduction of disease intensity were observed at monthly interval, starting from July to December 2013. The results are presented in Table 5.

During the pre treatment observation (June), there was no significant difference among the experimental plants with respect to disease intensity.

From the first post treatment observation (July), it was seen that there was a significant difference in the intensity of disease with respect to the treatments. The treatment T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1%, recorded the lowest disease intensity (20.82) and it was significantly superior to the rest of the treatments. The next best treatments were T5 - Mg + Zn + B plus tebuconazole @ 0.1% (20.87), T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (21.09), T6 - Mg + Zn + B plus copper hydroxide @ 0.25% (22.13), T10 - copper hydroxide @ 0.25% (22.65), T11 - propiconazole @ 0.1% (23.54), T7 - Mg + Zn + B plus propiconazole @ 0.1% (24.36), T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> (25.33), T2 - Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25% (26.81), T9 - tebuconazole @ 0.1% (27.31) and T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> (27.35) and these treatments were found to be statistically on par with each other and performed better than control (49.96). The highest disease

	Treatment	Disease intensity							
		June	July	Aug	Sept	Oct	Nov	Dec	
T1	Micronol @ 2 g l <sup>-1</sup> plus tebuconazole @ 0.1%	22.98 (4.90)	21.09 (4.70) <sup>a</sup>	38.45 (6.28) <sup>ab</sup>	39.05 (6.33) <sup>ab</sup>	37.00 (6.16) <sup>ab</sup>	34.85 (5.99) <sup>a</sup>	33.36 (5.87) <sup>a</sup>	
T2	Micronol @ 2 g l <sup>-1</sup> plus copper hydroxide @ 0.25%	31.67(5.72)	26.81 (5.27) <sup>ab</sup>	45.33 (6.81) °	44.40 (6.74) <sup>b</sup>	45.68 (6.82) <sup>d</sup>	36.46 (6.12) <sup>a</sup>	27.21 (5.31) <sup>a</sup>	
T3	Micronol @ 2 g l <sup>-1</sup> plus propiconazole @ 0.1%	20.87 (4.68)	20.82 (4.67) <sup>a</sup>	42.74 (6.61) <sup>bc</sup>	36.39 (6.12) <sup>a</sup>	37.13 (6.17) <sup>ab</sup>	34.23 (5.94) <sup>a</sup>	30.79 (5.64) <sup>a</sup>	
T4	Micronol @ 2 g l <sup>-1</sup> plus PGPR mix II @ 20 g l <sup>-1</sup>	29.72 (5.54)	25.33 (5.13) <sup>ab</sup>	42.74 (6.61) <sup>bc</sup>	41.49 (6.52) <sup>ab</sup>	40.56 (6.45) °	35.72 (6.06) <sup>a</sup>	29.81 (5.55) <sup>a</sup>	
T5	Mg + Zn + B plus tebuconazole @ $0.1\%$	24.10 (5.01)	20.87 (4.68) <sup>a</sup>	40.22 (6.42) <sup>abc</sup>	38.66 (6.30) <sup>ab</sup>	39.65 (6.38) <sup>abc</sup>	35.93 (6.08) <sup>a</sup>	33.00 (5.83) <sup>a</sup>	
T6	Mg + Zn + B plus copper hydroxide @ 0.25%	25.36 (5.13)	22.13 (4.81) <sup>a</sup>	40.91 (6.47) <sup>abc</sup>	41.67 (6.53) <sup>ab</sup>	39.33 (6.35) <sup>abc</sup>	35.43 (6.04) <sup>a</sup>	29.17 (5.49) <sup>a</sup>	
Τ7	Mg + Zn + B plus propiconazole @ 0.1%	29.06 (5.48)	24.36 (5.04) <sup>ab</sup>	43.98 (6.71) <sup>bc</sup>	36.62 (6.13) <sup>a</sup>	36.74 (6.14) <sup>a</sup>	33.02 (5.83) <sup>a</sup>	28.13 (5.40) <sup>a</sup>	
T8	Mg + Zn + B plus PGPRmix II @ 20 g l <sup>-1</sup>	29.29 (5.50)	27.35 (5.32) <sup>ab</sup>	43.60 (6.68) <sup>bc</sup>	42.18 (6.57) <sup>ab</sup>	40.11 (6.41) <sup>bc</sup>	38.16 (6.26) <sup>a</sup>	33.90 (5.91) <sup>a</sup>	
Т9	Tebuconazole @ 0.1%	27.69 (5.36)	27.31 (5.32) <sup>ab</sup>	37.08 (6.17) <sup>a</sup>	38.70 (6.30) <sup>ab</sup>	38.31 (6.27) <sup>abc</sup>	32.85 (5.82) <sup>a</sup>	25.92 (5.19) <sup>a</sup>	
T10	Copper hydroxide @ 0.25%	27.73 (5.36 )	22.65 (4.86) <sup>ab</sup>	41.33 (6.51) <sup>abc</sup>	40.21 (6.42) <sup>ab</sup>	39.18 (6.34) <sup>abc</sup>	35.38 (6.03) <sup>a</sup>	33.83 (5.90) <sup>a</sup>	
T11	Propiconazole @ 0.1%	29.52 (5.52)	23.54 (4.95) <sup>ab</sup>	42.15 (6.57) <sup>abc</sup>	39.37 (6.35) <sup>ab</sup>	38.99 (6.32) <sup>abc</sup>	35.53 (6.04) <sup>a</sup>	29.10 (5.49) <sup>a</sup>	
T12	PGPR mix II @ 20 g l <sup>-1</sup>	33.68 (5.89)	32.65 (5.80) <sup>b</sup>	45.36 (6.81) <sup>c</sup>	45.04 (6.79) <sup>b</sup>	44.41 (6.74) <sup>d</sup>	39.47 (6.36) <sup>a</sup>	36.31 (6.11) <sup>a</sup>	
T13	Control	32.64 (5.89)	49.96 (7.14) °	53.35 (7.37) <sup>d</sup>	53.22 (7.36) °	52.68 (7.33) <sup>e</sup>	65.38 (8.15) <sup>b</sup>	76.42 (8.80) <sup>b</sup>	
	SE (±)	-	0.33	0.15	0.18	0.18	0.21	0.50	
	CD (0.05)	NS	(0.95)	(0.43)	(0.53)	(0.25)	(0.62)	(1.45)	

Table 5. Effect of treatments on disease intensity of banana Sigatoka disease caused by Mycosphaerella musicola

\*Mean of three replications \*\*Transformation used is square root transformation NS : Non significant

\*\*\*Figures followed by the same letter do not differ significantly according to two way ANOVA at P=0.05

intensity was observed in absolute control (49.96) and it showed significant variation from the rest of the treatments.

In the second post treatment observation (August), it was found that there was a significant difference in the intensity of disease with respect to the treatments. The best treatment was found to be fungicide alone treatment, T9 tebuconazole (a) 0.1 % recorded the minimum disease intensity (37.08) followed by T1 - Micronol @ 2 g  $l^{-1}$  plus tebuconazole @ 0.1% (38.45), T6 - Mg + Zn + B plus copper hydroxide (a 0.1% (40.91), T10 - copper hydroxide (a 0.25% (41.33) and T11 - propiconazole (a) 0.1% (42.15) and these treatments remained significantly at par with each other. The performance of remaining treatments are in the order T3 - Micronol (a) 2 g  $l^{-1}$  plus propiconazole (a) 0.1% and T4 -Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> with disease intensity 42.74%, T8 - Mg + Zn + B plus PGPR mix II @ 20 g  $l^{-1}$  (43.60), T7 - Mg + Zn + B plus propiconazole (a) 0.1% (43.98), T2 - Micronol (a) 2 g  $l^{-1}$  plus copper hydroxide (a) 0.25% (45.33) and T12 - PGPR mix II (a) 20 g  $l^{-1}$  (45.36) and these treatments were significantly superior over control having the highest disease intensity (53.35) in combating the Sigatoka leaf spot disease.

In the third post treatment observation (September), it was noticed that there was a significant difference between the treatments imposed. The lowest disease intensity was found in the treatment, T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (36.39) followed by T7 - Mg + Zn + B plus propiconazole @ 0.1% (36.62), T5 - Mg + Zn + B plus tebuconazole @ 0.1% (38.66%), T9 - tebuconazole @ 0.1% (38.70), T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (39.05), T11 - propiconazole @ 0.1% (39.37), T10 - copper hydroxide @ 0.25% (40.21), T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> (41.49), T6 - Mg + Zn + B plus copper hydroxide @ 0.25% (41.67) and T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> (42.18). All the treatments performed better and significantly superior when compared to untreated check, which recorded the highest disease intensity (53.22).

From the fourth post treatment observation (October), it was found that there was a significant difference between treatments imposed. The lowest disease intensity was found in T7 - Mg + Zn + B plus propiconazole @ 0.1% (36.74) followed by T1 - Micronol @ 2 g  $\Gamma^1$  plus tebuconazole @ 0.1% having (37.00), T3 - Micronol @ 2 g  $\Gamma^1$  plus propiconazole @ 0.1% (37.13), T9 - tebuconazole @ 0.1% (38.31) and T11 - propiconazole @ 0.1% (38.99), T10 - copper hydroxide @ 0.25% (39.18), T6 - Mg + Zn + B plus copper hydroxide @ 0.25% (39.33) and T5 - Mg + Zn + B plus tebuconazole @ 0.1% (39.65). All these treatments were on par with each other. The highest disease intensity was noticed in absolute control (52.68), which was significantly different from all the treatments. The remaining treatments found to be better than control are T8 - Mg + Zn + B plus PGPR mix II @ 20 g  $\Gamma^1$  (40.11) which was on par with T4 - Micronol @ 2 g  $\Gamma^1$  plus PGPR mix II @ 20 g  $\Gamma^1$  (40.56) followed by T12 - PGPR mix II @ 20 g  $\Gamma^1$  plus copper hydroxide @ 0.25% (45.68).

The fifth post treatment observation (November) revealed that there was a significant difference in the disease intensity with respect to treatments. The disease intensity was lowest in T9 - tebuconazole @ 0.1% (32.85) which was followed by the treatments *viz.*, T7 - Mg + Zn +B plus propiconazole @ 0.1% (33.02), T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (34.23), T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (34.85), T10 - copper hydroxide @ 0.25% (35.38), T6 - Mg + Zn + B plus copper hydroxide @ 0.25% (35.43), T11 - propiconazole @ 0.1% (35.53), T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> (35.72), T5 - Mg + Zn + B plus tebuconazole @ 0.1% (35.93), T2 - Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25% (36.46), T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> (38.16) and T12 - PGPR mix II @ 20 g l<sup>-1</sup> (39.47). All the treatments were statistically on par with each other and they performed better than absolute control (65.38) which recorded the highest disease intensity.

During the last post treatment observation (December), it was found that there was a significant difference among the treatments. T9 - tebuconazole @

0.1% (25.92) recorded the lowest disease intensity followed by T2 - Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25% (27.21), T7 - Mg + Zn + B plus propiconazole @ 0.1% (28.13), T11 - propiconazole @ 0.1% (29.10), T6 - Mg + Zn + B plus copper hydroxide @ 0.25% (29.17), T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> (29.81), T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (30.79), T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (33.36), T5 - Mg + Zn + B plus tebuconazole @ 0.1% (33.00), T10 - copper hydroxide @ 0.25% (33.83), T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> (36.31). All the treatments were statistically on par with each other and they performed better than absolute control (76.42) which was recorded the highest disease intensity during the entire experiment.

Among the 13 treatments, maximum disease suppression was observed with T1 (Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1%), T3 (Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1%), T5 (Mg + Zn + B plus tebuconazole @ 0.1%), T6 (Mg + Zn + B plus copper hydroxide @ 0.25%), T7 (Mg + Zn + B plus propiconazole @ 0.1%), T9 (tebuconazole @ 0.1%), T10 (copper hydroxide @ 0.25%) and T11 (propiconazole @ 0.1%).

Hence, the analysis of the results indicated that the application of fungicides tebuconazole, propiconazole and copper hydroxide alone or in combination with Micronol or Mg + Zn + B had significant effect in reducing the disease intensity of Sigatoka leaf spot in banana.

#### **4.2 BIOCHEMICAL ANALYSIS**

Biochemical changes in banana plants infected with *Mycosphaerella musicola* were studied with respect to the total phenol, ortho di-hydroxy phenol (OD phenol), chlorophyll, Leaf nutrient and protein at different stages of growth.

# 4.2.1 Estimation of Phenol

Plant tissues respond to injury with the production of chemical substances like phenolics and the products of their oxidation. The accumulation of phenolics in banana leaves in both treated and untreated samples was studied. The estimation was done during the active growth periods *viz.*, 4<sup>th</sup>, 5<sup>th</sup> and 6<sup>th</sup> month after planting. The results revealed that there was a significant increase in phenol content during July *i.e.*, after the first spraying of treatments and the results are presented in Table 6.

During the fourth month (June), it was found that the treatments were not significantly different. The highest phenol content of 1725.00  $\mu$ g g<sup>-1</sup> was observed in T5, Mg + Zn + B plus tebuconazole @ 0.1% followed by T7, Mg + Zn + B plus propiconazole @ 0.1% (1710.00  $\mu$ g g<sup>-1</sup>) whereas, the lowest phenol content was found (600  $\mu$ g g<sup>-1</sup>) in case of T12, PGPR @ 20 g l<sup>-1</sup>.

In fifth month (July), a significant difference in phenol content was observed between the treatments. Among them, the treatment T11, propiconazole (@~0.1% showed the maximum phenol content (3705.17 µg g<sup>-1</sup>) followed by T7, Mg + Zn + B plus propiconazole (@~0.1% (2430.00 µg g<sup>-1</sup>) and T5, Mg + Zn + B plus tebuconazole (@~0.1% (2082.60 µg g<sup>-1</sup>) which was on par with each other. The lowest phenol content (555.00 µg g<sup>-1</sup>) was noticed in case of T12, PGPR (@~20 g l<sup>-1</sup>.

In the sixth month (August), it was found that even though there was no significant difference, the highest phenol content was noticed in case of T7, Mg + Zn + B plus propiconazole @ 0.1% (2066.54  $\mu$ g g<sup>-1</sup>) followed by T5, Mg + Zn + B plus tebuconazole @ 0.1% (1554.00  $\mu$ g g<sup>-1</sup>) whereas, lowest found in T12, PGPR mix II @ 20 g l<sup>-1</sup> (509.93  $\mu$ g g<sup>-1</sup>).

# 4.2.2 Estimation of Ortho di-hydroxyphenol

Defense related OD-phenol activity in banana leaves was recorded at different months viz; 4<sup>th</sup> 5<sup>th</sup>, and 6<sup>th</sup>. All the treatments imposed were not differing significantly in all the months.

During the fourth month, the highest OD-phenol content recorded was 64.83  $\mu g g^{-1}$  (T7, Mg + Zn + B plus propiconazole @ 0.1%) followed by T3, Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (50.00  $\mu g g^{-1}$ ) and the lowest was noticed in T13, absolute control (30  $\mu g g^{-1}$ ).

# Table 6. Effect of treatments on phenol content of banana infected with Sigatoka disease caused by Mycosphaerella musicola

		Phenol content * (µg g <sup>-1</sup> )					
	Treatment	June (4 <sup>th</sup> month)	July (5 <sup>th</sup> month)	August (6 <sup>th</sup> month)			
T1	Micronol @ 2 g l <sup>-1</sup> plus tebuconazole @ 0.1%	1216.67	1448.33 <sup>cd</sup>	1133.33			
T2	Micronol @ 2 g l <sup>-1</sup> plus copper hydroxide @ 0.25%	1099.18	1101.67 <sup>de</sup>	831.72			
T3	Micronol @ 2 g l <sup>-1</sup> plus propiconazole @ 0.1%	743.33	874.33 <sup>ef</sup>	776.17			
T4	Micronol @ 2 g $l^{-1}$ plus PGPR mix II @ 20 g $l^{-1}$	1177.50	852.00 <sup>ef</sup>	1272.33			
T5	Mg + Zn + B plus tebuconazole @ $0.1\%$	1725.00	2082.60 <sup>b</sup>	1554.00			
Т6	Mg + Zn + B plus copper hydroxide @ 0.25%	958.33	1129.29 <sup>de</sup>	889.83			
Τ7	Mg + Zn + B plus propiconazole @ 0.1%	1710.00	2430.00 <sup>b</sup>	2066.54			
T8	Mg + Zn + B plus PGPR mix II @ 20 g $l^{-1}$	800.00	968.00 <sup>e</sup>	949.00			
Т9	Tebuconazole @ 0.1%	808.33	1692.33 °	853.33			
T10	Copper hydroxide @ 0.25%	900.83	980.38 <sup>e</sup>	1271.59			
T11	Propiconazole @ 0.1%	1184.17	3705.17 <sup>a</sup>	1111.94			
T12	PGPR mix II @ 20 g l <sup>-1</sup>	600.00	555.00 <sup>f</sup>	509.93			
T13	Control	1050.83	1349.08 <sup>cd</sup>	1316.39			
	SE (±)	368.18	124.33	359.38			
	CD (0.05)	NS	(362.91)	NS			

\*Mean of three replications

\*\*\*Figures followed by the same letter do not differ significantly according to two way ANOVA at P=0.05 NS : Non significant

During the fifth month, the highest OD-phenol content (30.83  $\mu$ g g<sup>-1</sup>) was observed in T11, propiconazole @ 0.1% followed by T7, Mg + Zn + B plus propiconazole @ 0.1% (30.17  $\mu$ g g<sup>-1</sup>). The lowest OD-phenol content (16.83  $\mu$ g g<sup>-1</sup>) was observed in absolute control.

During the sixth month, the highest OD-phenol content was observed in T5, Mg + Zn + B plus tebuconazole @ 0.1% (65.00  $\mu$ g g<sup>-1</sup>) followed by T11, propiconazole @ 0.1% (59.33  $\mu$ g g<sup>-1</sup>). The lowest OD-phenol content observed in absolute control (40.00  $\mu$ g g<sup>-1</sup>) (Table 7).

## 4.2.3 Estimation of Chlorophyll content

The result revealed that there was no significant difference between treatments. However, all the treatments recorded superior values over absolute control which was recorded the lowest chlorophyll content (1.2  $\mu$ g g<sup>-1</sup>). The most favorable effect was for T5, Mg + Zn + B plus tebuconazole @ 0.1% (2.14  $\mu$ g g<sup>-1</sup>) followed by T8, Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> (2.12  $\mu$ g g<sup>-1</sup>). (Table 8).

# 4.2.4 Estimation of Protein Content

During the late vegetative stage, the total soluble protein content in banana leaves was estimated and the results are presented in Table 9.

Though there was no significant variation among treatments, the highest protein content (12250  $\mu$ g g<sup>-1</sup>) was noticed in absolute control whereas, the lowest was observed in case of T1, Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (11156.67  $\mu$ g g<sup>-1</sup>).

# 4.2.5 Estimation of Plant Secondary and Micro Nutrients

One observation plant from each of the replication was identified for destructive sampling at bunch emergence stage to estimate the leaf nutrient content. The micronutrients such as Boron, Zinc and secondary nutrient such as Magnesium were chemically analyzed. The distal half portion of petiole was collected at bunch emergence stage and analyzed for their nutrient contents. (Table 10).

 Table 7. Effect of treatments on OD - phenol content of banana infected

 with Sigatoka disease caused by Mycosphaerella musicola

		OD - Pher	<b>OD</b> - Phenol content * (µg g <sup>-1</sup> )				
	Treatment	June (4 <sup>th</sup> month)	July (5 <sup>th</sup> month)	August (6 <sup>th</sup> month)			
T1	Micronol @ 2 g l <sup>-1</sup> plus tebuconazole @ $0.1\%$	35.58	20.33	41.00			
T2	Micronol @ 2 g l <sup>-1</sup> plus copper hydroxide @ $0.25\%$	41.83	27.00	51.00			
Т3	Micronol @ 2 g l <sup>-1</sup> plus propiconazole @ $0.1\%$	43.58	30.00	56.67			
T4	Micronol @ 2 g l <sup>-1</sup> plus PGPR mix II @ 20 g l <sup>-1</sup>	32.83	23.67	45.00			
T5	Mg + Zn + B plus tebuconazole @ 0.1%	47.92	27.83	59.33			
T6	Mg + Zn + B plus copper hydroxide @ $0.25\%$	38.92	23.67	54.17			
T7	Mg + Zn + B plus propiconazole ( $a$ 0.1%)	50.00	30.17	56.67			
T8	Mg + Zn + B plus PGPR mix II @ 20 g $l^{-1}$	41.00	23.67	40.50			
Т9	Tebuconazole @ 0.1%	30.83	21.67	33.33			
T10	Copper hydroxide @ 0.25%	38.92	20.67	47.83			
T11	Propiconazole @ 0.1%	64.83	30.83	65.00			
T12	PGPR mix II @ 20 g l <sup>-1</sup>	35.83	28.33	43.33			
T13	Control	30.00	16.83	40.00			
	SE (±)	13.66	7.00	11.28			
	CD (0.05)	NS	NS	NS			

\*Mean of three replications NS : Non significant

	Treatment	Chlorophyll content * (µg g <sup>-1</sup> )
T1	Micronol @ 2 g $l^{-1}$ plus tebuconazole @ 0.1%	1.79
T2	Micronol @ 2 g l <sup>-1</sup> plus copper hydroxide @ $0.25\%$	1.26
Т3	Micronol @ 2 g l <sup>-1</sup> plus propiconazole @ $0.1\%$	1.62
T4	Micronol @ 2 g l <sup>-1</sup> plus PGPR mix II @ 20 g l <sup>-1</sup>	1.77
T5	Mg + Zn + B plus tebuconazole @ $0.1\%$	2.14
Т6	Mg + Zn + B plus copper hydroxide @ $0.25\%$	1.64
T7	Mg + Zn + B plus propiconazole @ $0.1\%$	1.72
T8	Mg + Zn + B plus PGPR mix II ( $@$ 20 g l <sup>-1</sup>	2.12
Т9	Tebuconazole @ 0.1%	1.71
T10	Copper hydroxide @ 0.25%	1.35
T11	Propiconazole @ 0.1%	1.66
T12	PGPR mix II (a) 20 g $l^{-1}$	1.72
T13	Control	1.20
	SE (±)	0.25
	CD (0.05)	NS

 Table 8. Effect of treatments on chlorophyll content of banana infected

 with Sigatoka disease caused by Mycosphaerella musicola

\*Mean of three replications

\*\*Observations taken at bunch emergence stage

NS : Non significant

# Table 9. Effect of treatments on protein content of Sigatoka disease of banana caused by Mycosphaerella musicola

	Treatment	Protein content*(µg g <sup>-1</sup> )
T1	Micronol @ 2 g $l^{-1}$ plus tebuconazole @ 0.1%	11156.67
T2	Micronol @ 2 g l <sup>-1</sup> plus copper hydroxide @ 0.25%	12243.05
Т3	Micronol @ 2 g l <sup>-1</sup> plus propiconazole @ 0.1%	11180.00
T4	Micronol @ 2 g l <sup>-1</sup> plus PGPR mix II @ 20 g l <sup>-1</sup>	12246.67
Т5	Mg + Zn + B plus tebuconazole @ $0.1\%$	11183.69
Т6	Mg + Zn + B plus copper hydroxide @ $0.25\%$	12236.00
Τ7	Mg + Zn + B plus propiconazole @ 0.1%	12240.00
Т8	Mg + Zn + B plus PGPR mix II ( $a$ ) 20 g l <sup>-1</sup>	12242.00
Т9	Tebuconazole @ 0.1%	12215.00
T10	Copper hydroxide @ 0.25%	12233.33
T11	Propiconazole @ 0.1%	11190.00
T12	PGPR mix II @ 20 g l <sup>-1</sup>	12211.73
T13	Control	12250.00
	SE (±)	39.07
	CD (0.05)	NS

\* Mean of three replications

NS : Non significant

		Leaf nutrient content				
	Treatment	Magnesium content (%)	Boron content (ppm)	Zinc content (ppm)		
T1	Micronol @ 2 g l <sup>-1</sup> plus tebuconazole @ 0.1%	0.25 <sup>d</sup>	15.50 <sup>h</sup>	35.34 <sup>ab</sup>		
T2	Micronol @ 2 g l <sup>-1</sup> plus copper hydroxide @ 0.25%	0.19 <sup>f</sup>	22.67 <sup>e</sup>	32.89 <sup>ab</sup>		
T3	Micronol @ 2 g l <sup>-1</sup> plus propiconazole @ 0.1%	0.19 <sup>f</sup>	21.70 <sup>f</sup>	35.58 <sup>ab</sup>		
T4	Micronol @ 2 g l <sup>-1</sup> plus PGPR mix II @ 20 g l <sup>-1</sup>	0.29 °	23.60 <sup>d</sup>	36.23 <sup>a</sup>		
T5	Mg + Zn + B plus tebuconazole (a) $0.1\%$	0.34 <sup>a</sup>	25.50 <sup>b</sup>	26.30 bcd		
T6	Mg + Zn + B plus copper hydroxide @ 0.25%	0.31 <sup>b</sup>	24.43 °	29.07 abc		
T7	Mg + Zn + B plus propiconazole @ 0.1%	0.31 <sup>b</sup>	24.00 <sup>cd</sup>	27.17 <sup>abcd</sup>		
T8	Mg + Zn + B plus PGPR mix II @ 20 g l <sup>-1</sup>	0.35 <sup>a</sup>	72.77 <sup>a</sup>	30.43 abc		
T9	Tebuconazole @ 0.1%	0.18 <sup>f</sup>	14.70 <sup>i</sup>	20.87 <sup>cd</sup>		
T10	Copper hydroxide @ 0.25%	0.14 <sup>g</sup>	13.50 <sup>j</sup>	18.40 <sup>d</sup>		
T11	Propiconazole @ 0.1%	0.09 <sup>h</sup>	14.67 <sup>i</sup>	26.10 bcd		
T12	PGPR mix II @ 20 g l <sup>-1</sup>	0.22 °	19.00 <sup>g</sup>	27.57 <sup>abcd</sup>		
T13	Control	0.08 <sup>h</sup>	12.20 <sup>k</sup>	18.37 <sup>d</sup>		
	SE (±)	0.01	2.49	3.32		
	CD (0.05)	(0.04)	(7.27)	(9.68)		

Table 10. Effect of treatments on leaf nutrient content of banana Sigatoka disease caused by *Mycosphaerella musicola* 

\*Mean of three replications

\*\*Observations taken at bunch emergence stage

\*\*\*Figures followed by the same letter do not differ significantly according to two way ANOVA at P=0.05

The plant samples were analyzed for micro nutrient, boron to find out the influence of Boron content on Sigatoka leaf spot disease of banana. It was observed there was a significant difference between the treatments with respect to boron content.

Among treatments, the boron content varied within a range from 13.50 ppm to 72.77 ppm. The highest boron content (72.77 ppm) was obtained in case of T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> and the lowest (12.20 ppm) was for T13 - absolute control. The performance of treatments are in the order *viz.*, T5 - Mg + Zn + B plus tebuconazole @ 0.1% (25.50 ppm), T6 - Mg + Zn + B plus copper hydroxide @ 0.25% (24.43 ppm) , T7 - Mg + Zn + B plus propiconazole @ 0.1% (24.00 ppm) , T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> (23.60 ppm) , T2 - Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25% (22.67 ppm), T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (21.70 ppm) , T12 - PGPR mix II @ 20 g l<sup>-1</sup> (19.00 ppm), T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (14.70 ppm), T11 - propiconazole @ 0.1% (14.67 ppm) and T10 - copper hydroxide @ 0.25% (13.50 ppm). The plant samples were estimated for zinc content to check out the influence of zinc content on Sigatoka leaf spot disease of banana. It was found that, there was a significant variation among the treatments.

The treatment, T4 - Micronol @ 2 g I<sup>-1</sup> plus PGPR mix II @ 20 g I<sup>-1</sup> recorded the highest zinc content (36.23 ppm) followed by T3 - Micronol @ 2 g I<sup>-1</sup> plus propiconazole @ 0.1% (35.58 ppm), T1 - Micronol @ 2 g I<sup>-1</sup> plus tebuconazole @ 0.1% (35.34 ), T2 - Micronol @ 2 g I<sup>-1</sup> plus copper hydroxide @ 0.25% (32.89 ppm), T8 - Mg + Zn + B plus PGPR mix II @ 20 g I<sup>-1</sup> (30.43 ppm), T6 - Mg + Zn + B plus copper hydroxide @ 0.25% (29.07 ppm), T12 - PGPR mix II @ 20 g I<sup>-1</sup> (27.57 ppm), T7 - Mg + Zn + B plus propiconazole @ 0.1% (27.17 ppm), and these treatments were on par with each other. The remaining treatments are in the following order *viz.*, T5 - Mg + Zn + B plus tebuconazole @ 0.1% (26.30 ppm), T11 - propiconazole @ 0.1% (26.10 ppm), T9 - tebuconazole @ 0.1% (20.87 ppm) and T10 - copper hydroxide @ 0.25% (18.40 ppm). The lowest zinc content (18.37 ppm) was noticed in T13, untreated check.

The leaf samples were used to estimate the secondary nutrient content, magnesium to find out the influence of Magnesium content on the intensity of Sigatoka leaf spot disease of banana. It was found that there exists a significant difference between the treatments imposed with respect to the Mgcontent.

The maximum magnesium content (0.35%) was noticed in T8 - Mg + Zn + B plus PGPR mix II @ 20 g  $\Gamma^1$  which was on par with T5 - Mg + Zn + B plus tebuconazole @ 0.1% (0.34%), followed by T6 - Mg + Zn + B plus copper hydroxide @ 0.25% and T7 - Mg + Zn + B plus propiconazole @ 0.1% with each of 0.31%, T4 - Micronol @ 2 g  $\Gamma^1$  plus PGPR mix II @ 20 g  $\Gamma^1$  (0.29%), T1 -Micronol plus tebuconazole @ 0.1% (0.25%), T12 - PGPR mix II @ 20 g  $\Gamma^1$ (0.22%), T2 - Micronol @ 2 g  $\Gamma^1$  plus copper hydroxide @ 0.25% and T3 -Micronol @ 2 g  $\Gamma^1$  plus propiconazole @ 0.1% with each of 0.19%, T10 - copper hydroxide @ 0.25% (0.19%), T3 - Micronol @ 2 g  $\Gamma^1$  plus propiconazole @ 0.1% (0.19%), T9 - tebuconazole @ 0.1% (0.18%), T10 - copper hydroxide @ 0.25% (0.14%) and T11 - propiconazole @ 0.% (0.09%). All the treatments performed better than absolute control having Magnesium content of 0.08%.

## 4.3 BIOMETRIC OBSERVATIONS

# 4.3.1 Effect of Treatments on Growth Characters

Plant growth characteristics *viz.*, height of the plant, girth of the plant, number of leaves, number of functional leaves etc recorded were statistically analyzed and the results are presented hereunder.

# 4.3.1.1 Height of the Plant (m)

Average height of the banana plants recorded from each of the replication at the time of harvest and the results are given in Table 11. The perusal of the data revealed that the different treatments had any significant difference on plant height. Though not significantly different, the maximum plant height (2.5 m) was

 Table 11. Effect of treatments on plant height of banana plants with Sigatoka

 disease caused by Mycosphaerella musicola

	Treatment	Plant height * (m)
T1	Micronol @ 2 g l <sup>-1</sup> plus tebuconazole @ 0.1%	2.37
T2	Micronol @ 2 g l <sup>-1</sup> plus copper hydroxide @ 0.25%	1.96
Т3	Micronol @ 2 g l <sup>-1</sup> plus propiconazole @ 0.1%	2.11
T4	Micronol @ 2 g l <sup>-1</sup> plus PGPR mix II @ 20 g l <sup>-1</sup>	2.36
Т5	Mg + Zn + B plus tebuconazole @ $0.1\%$	2.50
Т6	Mg + Zn + B plus copper hydroxide ( $@ 0.25\%$	1.84
Τ7	Mg + Zn + B plus propiconazole @ $0.1\%$	2.16
T8	Mg + Zn + B plus PGPR mix II @ 20 g l <sup>-1</sup>	2.42
Т9	Tebuconazole @ 0.1%	2.26
T10	Copper hydroxide @ 0.25%	2.01
T11	Propiconazole @ 0.1%	2.08
T12	PGPR mix II @ 20 g l <sup>-1</sup>	2.19
T13	Control	1.68
	SE (±)	0.17
	CD (0.05)	NS

\*Mean of three replications

NS : Non significant

recorded in case of Mg + Zn + B plus tebuconazole @ 0.1% and the lowest was recorded by absolute check (1.68 m). All the treatments recorded better plant height than control.

## 4.3.1.2 Girth of the Plant (cm)

Average girth of the banana plants recorded from each of the replication at the time of harvest and the results are given in Table 12. The perusal of the data revealed that the different treatments did not cause significant variation in plant girth. However, the highest value was 54.67 cm obtained in case of Mg + Zn + B plus tebuconazole @ 0.1% and the lowest value(45.67 cm) recorded by absolute control.

#### 4.3.1.3 Number of Leaves

The observations on effect of different treatments on the total number of leaves were recorded. Observations included one pre treatment observation and six post treatment observations.

The first pre treatment observation was taken during June. The following are the post treatment observations *viz*; July, August, September, October, November and December 2013 were carried out and the results are presented in Table 13.

During the pre treatment observation (June), there was no significant difference among the experimental plants with respect to number of leaves.

The first post treatment observation (July) implies that the treatments did not showed significant difference between them. The maximum number of leaves (13.33) was noticed in T7 - Mg + Zn + B plus propiconazole @ 0.1% and the minimum was found in untreated check (10.67).

During the second post treatment observation (August) also, none of the treatments varied significantly. T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (14.00) and the minimum was recorded by T13, untreated check (11.67).

# Table 12. Effect of treatments on plant girth of banana plants with Sigatoka disease caused by Mycosphaerella musicola

	Treatment	Plant girth * (cm)
T1	Micronol @ 2 g $l^{-1}$ plus tebuconazole @ 0.1%	48.33
T2	Micronol @ 2 g l <sup>-1</sup> plus copper hydroxide @ $0.25\%$	46.33
Т3	Micronol @ 2 g l <sup>-1</sup> plus propiconazole @ $0.1\%$	47.67
T4	Micronol @ 2 g l <sup>-1</sup> plus PGPR mix II @ 20 g l <sup>-1</sup>	49.00
Т5	Mg + Zn + B plus tebuconazole @ 0.1%	54.67
Т6	Mg + Zn + B plus copper hydroxide @ $0.25\%$	47.00
Т7	Mg + Zn + B plus propiconazole @ 0.1%	49.67
Т8	Mg + Zn + B plus PGPR mix II @ 20 g $l^{-1}$	52.33
Т9	Tebuconazole @ 0.1%	49.67
T10	Copper hydroxide @ 0.25%	49.00
T11	Propiconazole @ 0.1%	49.00
T12	PGPR mix II @ 20 g l <sup>-1</sup>	50.00
T13	Control	45.67
	SE (±)	2.58
	CD (0.05)	NS

\*Mean of three replications

NS : Non significant

	Treatment	Number of leaves*							
			July	Aug	Sept	Oct	Nov	Dec	
T1	Micronol @ 2 g l <sup>-1</sup> plus tebuconazole @ 0.1%	12.33	11.33	13.00	15.33 <sup>a</sup>	14.67 abc	13 .00 <sup>ab</sup>	10.33 <sup>cd</sup>	
T2	Micronol @ 2 g l <sup>-1</sup> plus copper hydroxide @ 0.25%	10.67	12.67	12.67	13.33 <sup>de</sup>	13.00 °	12.00 de	10.33 <sup>cd</sup>	
T3	Micronol @ 2 g l <sup>-1</sup> plus propiconazole @ 0.1%	12.67	12.33	14.00	13.00 °	13.67 <sup>de</sup>	13.33 <sup>a</sup>	10.33 <sup>cd</sup>	
T4	Micronol @ 2 g l <sup>-1</sup> plus PGPR mix II @ 20 g l <sup>-1</sup>	11.67	12.00	12.33	15.00 <sup>ab</sup>	15.33 ª	13 .00 <sup>ab</sup>	11.33 <sup>a</sup>	
T5	Mg + Zn + B plus tebuconazole @ $0.1\%$	10.67	11.00	13.67	14.00 °	14.33 bc	11.67 °	10.00 de	
T6	Mg + Zn + B plus copper hydroxide @ $0.25\%$	10.67	11.67	13.00	13.67 <sup>cd</sup>	14.33 bc	12.00 de	10.00 de	
T7	Mg + Zn + B plus propiconazole @ $0.1\%$	11.33	13.33	13.67	13.33 <sup>de</sup>	14.00 <sup>cd</sup>	11.33 <sup>f</sup>	9.00 <sup>f</sup>	
T8	Mg + Zn + B plus PGPR mix II @ 20 g $\Gamma^1$	10.67	11.00	13.33	15.00 <sup>ab</sup>	14.67 abc	12.33 <sup>cd</sup>	11.00 <sup>ab</sup>	
Т9	Tebuconazole @ 0.1%	11.33	11.67	12.67	14.67 <sup>b</sup>	15.00 <sup>ab</sup>	12.33 <sup>cd</sup>	9.67 °	
T10	Copper hydroxide @ 0.25%	10.33	11.67	13.33	13.33 <sup>de</sup>	13.00 <sup>e</sup>	12.33 <sup>cd</sup>	10.33 <sup>cd</sup>	
T11	Propiconazole @ 0.1%	12.33	13.00	12.33	12.67 <sup>f</sup>	12.67 °	12.00 de	10.67 bc	
T12	PGPR mix II @ 20 g l <sup>-1</sup>	11.33	11.00	13.33	15.00 <sup>ab</sup>	14.33 bc	12.67 bc	10.33 <sup>cd</sup>	
T13	Control	10.67	10.67	11.67	12.00 <sup>g</sup>	11.33 <sup>f</sup>	9.00 <sup>f</sup>	6.00 <sup>g</sup>	
<b> </b>	SE (±)	0.6	0.72	0.96	0.64	0.71	0.39	0.54	
	CD (0.05)	NS	NS	NS	(1.86)	(2.06)	(1.15)	(1.59)	

\*Mean of three replications

\*\*\*Figures followed by the same letter do not differ significantly according to two way ANOVA at P=0.05 NS : Non significant

In the third post treatment observation taken during the month September, there was a significant difference with respect to the number of leaves. T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (15.33) found to be the best treatment for the number of leaves which was on par with T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup>, T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> and T12 - PGPR mix II @ 20 g l<sup>-1</sup> with a value of 15. The performance of remaining treatments are in the following order *viz.*, T9 - tebuconazole @ 0.1% (14.67), T5 - Mg + Zn + B plus tebuconazole @ 0.1% (14.00), T6 - Mg + Zn + B plus copper hydroxide @ 0.25% (13.67), T2 - Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25% with a value of 13.33, T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (12.67). All treatments performed better than absolute control, having the minimum number of leaves (12.00).

During the fourth post treatment observation (October), the experimental results revealed that, T4 - Micronol @ 2 g  $\Gamma^1$  plus PGPR mix II @ 20 g  $\Gamma^1$  (15.33) recorded as superior among the treatments. The lowest value observed in the absolute control (11.33). All the treatments were significantly different from control. The performance of the remaining treatments are in the following order *viz.*, T9 - tebuconazole @ 0.1% (15.00), T1 - Micronol @ 2 g  $\Gamma^1$  plus tebuconazole @ 0.1% and T8 - Mg + Zn + B plus PGPR mix II @ 20 g  $\Gamma^1$  with a value of 14.67, T5 - Mg + Zn + B plus tebuconazole @ 0.1%, T6 - Mg + Zn + B plus copper hydroxide @ 0.25% and T12 - PGPR mix II @ 20 g  $\Gamma^1$  with each of 14.33, T2 - Micronol @ 2 g  $\Gamma^1$  plus copper hydroxide @ 0.25% with each of 13.00 and T11 - propiconazole @ 0.1% (12.67).

During the fifth post treatment observation (November), it was found that the treatments had a significant difference. Among them, T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (13.33) had maximum number of leaves which was on par with T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% and T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> with each of 13.00 and T1 - Micronol @ 2

g l<sup>-1</sup> plus tebuconazole @ 0.1 % (13.00). The lowest was found in absolute control (9.00). All the treatments were found to be better than control *viz.*, T12 - PGPR mix II @ 20 g l<sup>-1</sup> (12.67), T8 - Mg + Zn + B plus PGPR @ 20 g l<sup>-1</sup>, T9 - tebuconazole @ 0.1% and T10 - copper hydroxide @ 0.25% with each of 12.33, T2 - Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25%, T6 - Mg + Zn + B plus copper hydroxide @ 0.1% with a value of 12.00, T5 - Mg + Zn + B plus tebuconazole @ 0.1% (11.67) and T7 - Mg + Zn + B plus propiconazole @ 0.1% (11.33).

During the last observation (December), T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> litre recorded the maximum number of leaves (11.33) and which was on par with T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> (11.00) followed by T11 - propiconazole @ 0.1% (10.67), T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1%, T2 - Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25%, T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1%, T10 - copper hydroxide @ 0.25% and PGPR mix II @ 20 g l<sup>-1</sup> with a value of 10.33, T5 - Mg + Zn + B plus tebuconazole @ 0.1% and T6 - Mg + Zn + B plus copper hydroxide @ 0.25% with each of 10.00, T9 - tebuconazole @ 0.1% (9.67) and T7 - Mg + Zn + B plus propiconazole @ 0.1% (9.00). All the treatments were significantly superior to control which recorded for minimum number of leaves (6.00).

The overall performance of the treatments indicated that the treatments T1 (Micronol @ 2 g  $l^{-1}$  plus tebuconazole @ 0.1%), T3 (Micronol @ 2 g  $l^{-1}$  plus propiconazole @ 0.1%), T4 (Micronol @ 2 g  $l^{-1}$  plus PGPR @ 20 g  $l^{-1}$ ), T9 (tebuconazole @ 0.1%) and T12 (PGPR @ 20 g  $l^{-1}$ ) were found to be the best and recorded the maximum number of leaves during September, October, November and December.

#### 4.3.1.4 Number of Functional Leaves

The observations on effect of different treatments on the total number of functional leaves were recorded. Observations included one pre treatment observation and six post treatment observations. The first pre treatment observation was taken during June. The following are the post treatment observations *viz*; July, August, September, October, November and December 2013 and the results are presented in Table 14.

Different treatments didn't cause significant variation in number of functional leaves except the last two observations.

During the pre treatment observation (June), there was no significant difference among the experimental plants with respect to number of functional leaves.

In the first post treatment observation (July), the number of functional leaves was observed to vary significantly among treatments. T5 - Mg + Zn + B plus tebuconazole @ 0.1% recorded as best treatment with maximum number of functional leaves (11.33), followed by T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (11.30), T2 - Micronol @ 2 g l<sup>-1</sup> of water plus copper hydroxide @ 0.25%, T6 - Mg + Zn + B plus copper hydroxide @ 0.25% and T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> with each of 11.00, T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% and T11 - propiconazole @ 0.1%, T9 - tebuconazole @ 0.1%, T10 - copper hydroxide @ 0.25% and T12 - PGPR mix II @ 20 g l<sup>-1</sup> with each of 10.00, T4 - Micronol @ 2 g l<sup>-1</sup> of water plus PGPR mix II @ 20 g l<sup>-1</sup> of water plus PGPR mix II @ 20 g l<sup>-1</sup> of water plus PGPR mix II @ 20 g l<sup>-1</sup> with each of 10.00, T4 - Micronol @ 2 g l<sup>-1</sup> of water plus PGPR mix II @ 20 g l<sup>-1</sup> with each of 10.00, T4 - Micronol @ 2 g l<sup>-1</sup> of water plus PGPR mix II @ 20 g l<sup>-1</sup> with each of 10.00, T4 - Micronol @ 2 g l<sup>-1</sup> of water plus PGPR mix II @ 20 g l<sup>-1</sup> (9.67). However, all the treatments were significantly superior to the absolute control, which recorded minimum number of functional leaves (7.00).

In the second observation (August), the treatments didn't show significant variation in number of functional leaves. However, T1 - Micronol @ 2 g  $l^{-1}$  plus tebuconazole @ 0.1% and T8 - Mg + Zn + B plus PGPR mix II @ 20 g  $l^{-1}$  recorded the maximum number of functional leaves (11.67). The minimum number of functional leaves (9.33) was noticed in case of control. However, all the treatments performed better than absolute control.

	Treatment	Number of functional leaves*							
	reachent	June	July	Aug	Sept	Oct	Nov	Dec	
T1	Micronol @ 2 g l <sup>-1</sup> plus tebuconazole @ 0.1%	11.66	10.67 <sub>ab</sub>	11.67	13.33	12.00	11.00 <sub>ab</sub>	8.33 <sup>a</sup>	
T2	Micronol @ 2 g l <sup>-1</sup> plus copper hydroxide @ 0.25%	10.33	11.00 <sup>a</sup>	10.33	10.67	11.67	10.67 <sup>b</sup>	8.33 <sup>a</sup>	
Т3	Micronol @ 2 g $l^{-1}$ plus propiconazole @ 0.1%	12.33	11.30 <sup>a</sup>	11.33	11.33	11.33	10.33 bc	8.67 <sup>a</sup>	
T4	Micronol @ 2 g $l^{-1}$ plus PGPR mix II @ 20 g $l^{-1}$	10.33	9.67 <sup>b</sup>	10.33	9.67	10.00	9.33 <sup>d</sup>	9.00 <sup>a</sup>	
T5	Mg + Zn + B plus tebuconazole @ $0.1\%$	10.33	11.33 ª	11.00	12.33	13.00	10.67 <sup>b</sup>	9.00 <sup>a</sup>	
Т6	Mg + Zn + B plus copper hydroxide @ 0.25%	10.67	11.00 <sup>a</sup>	10.00	10.33	11.00	10.67 <sup>b</sup>	8.33 <sup>a</sup>	
Τ7	Mg + Zn + B plus propiconazole @ 0.1%	10.67	10.00 <sup>b</sup>	10.67	11.67	11.67	9.33 <sup>d</sup>	7.67 <sup>b</sup>	
Т8	Mg + Zn + B plus PGPR mix II @ 20 g l <sup>-1</sup>	10.33	11.00 <sup>a</sup>	11.67	11.33	11.00	11.33 <sup>a</sup>	9.00 <sup>a</sup>	
Т9	Tebuconazole @ 0.1%	10.33	10.00 <sup>b</sup>	11.00	12.33	12.33	10.33 bc	8.33 ª	
T10	Copper hydroxide @ 0.25%	10.00	10.00 <sup>b</sup>	11.00	11.00	11.33	11.00 <sub>ab</sub>	8.33 ª	
T11	Propiconazole @ 0.1%	10.67	10.67 <sub>ab</sub>	10.67	11.33	10.67	10.00 °	8.33 <sup>a</sup>	
T12	PGPR mix II @ 20 g l <sup>-1</sup>	10.33	10.00 <sup>b</sup>	10.00	10.67	10.67	11.00 <sub>ab</sub>	8.67 <sup>a</sup>	
T13	Control	10.00	7.00 °	9.33	9.30	9.33	7.33 °	4.33 °	
	SE (±)	0.64	0.71	0.63	0.82	0.76	0.65	0.73	
	CD (0.05)	NS	(2.07)	NS	NS	NS	(1.9)	(2.14)	

#### Table 14. Effect of treatments on number of functional leaves of banana

\*Mean of three replications

\*\*\*Figures followed by the same letter do not differ significantly according to two way ANOVA at P=0.05

NS : Non significant

In the third observation (September) also it was observed that, the treatments had no significant variation between them, with maximum value of 12.33 by combination treatment, T5 - Mg + Zn + B plus tebuconazole (@ 0.1% and fungicide alone treatment, T9 - tebuconazole (@ 0.1%. The lowest value obtained by control (9.30). All the treatments were performed better than absolute control.

During the fourth observation (October), it was observed that the treatments had no significant difference between them. T5 - Mg + Zn + B plus tebuconazole @ 0.1% produced maximum number of functional leaves (13.00) and minimum (9.33) was found in case of control.

Regarding the fifth observation (November), a significant difference could be noticed among the treatments. The maximum number of functional leaves recorded with T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> (11.33) which was found to be on par with T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1%, T10 - copper hydroxide @ 0.25% and T12 - PGPR mix II @ 20 g l<sup>-1</sup> with a value of 11.00, T2 - Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25%, T5 - Mg + Zn + B plus tebuconazole @ 0.1% and T6 - Mg + Zn + B plus copper hydroxide @ 0.25% with each of 10.67, T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% and T9 - tebuconazole @ 0.1% with 10.33, T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> and T7 - Mg + Zn + B plus propiconazole @ 0.1% with 9.33. The lowest number of functional leaves was noticed in T13, absolute control (7.33) and all the treatments were significantly superior over the same.

During the last observation (December), there could be noticed a significant difference among them. The maximum number of functional leaves was recorded in T5 - Mg + Zn + B plus tebuconazole @ 0.1% and T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> having a value 9.00. Except T7 - Mg + Zn + B plus propiconazole @ 0.1%, all the remaining treatments were on par *viz.*, T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1%, T2 - Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25%, T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1%, T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR @ 20 g l<sup>-1</sup>, T6 - Mg + Zn + B plus copper hydroxide @ 0.25%, T9 - tebuconazole @ 0.1%, T10 - copper hydroxide

@ 0.25%, T11 - propiconazole @ 0.1% and T12 - PGPR @ 20 g l<sup>-1</sup>. The number of functional leaves recorded in case of T7 - Mg + Zn + B plus propiconazole @ 0.1% was 7.67. The lowest number of functional leaves was found in absolute control (4.33).

The concise analysis of the results indicated that the treatments T1 (Micronol @ 2 g l<sup>-1</sup> plus tebuconazole (0.1%) and T8 (Mg + Zn + B plus PGPR @ 20 g l<sup>-1</sup>) were found to be the best and recorded the maximum number of functional leaves during July, November and December.

#### 4.3.2 Effect of Treatments on Yield Parameters

#### 4.3.2.1 Effect of Treatments on Time of Bunch Emergence

The comparison of the average time taken for bunch emergence from the respective treatments showed that there was no significant difference between any of the treatments. However, the shortest period or number of days taken for bunch emergence was observed in the combination of treatment T5, Mg + Zn + B plus tebuconazole @ 0.1% (218.33) whereas the longest time period for bunch emergence was observed in absolute control (267). The results are presented in Table 15.

# 4.3.2.2 Effect of Treatments on Time of Bunch Maturity

The number of days up to harvest was recorded to check whether the treatments had any influence on the same.

While the average time for bunch maturity of each treatments were compared, it was found that shortest number of days or period for bunch maturity was observed in the combination treatment T5 - Mg + Zn + B plus tebuconazole (@~0.1% (297.55) which was statistically on par with all other treatments. Maximum number of days for bunch maturity was observed in absolute control (358.01). (Table 15).

Table 15. Effect of treatments on yield charecters in the Sigatoka leaf spot disease infected banana plants cv. Nendran

	Treatment	Yield per plant * (kg)	No: of hands *	Fingers per bunch (no:) *	Suckers (no:) *	Time of bunch emergence (days)*	Time of bunch maturity (days) *
T1	Micronol @ 2 g l <sup>-1</sup> plus tebuconazole @ 0.1%	10.98 abc	5.67	48.33	4.33	229.67	311.67
T2	Micronol @ 2 g l <sup>-1</sup> plus copper hydroxide @ 0.25%	9.77 bc	5.33	43.00	4.33	235.00	319.51
Т3	Micronol @ 2 g l <sup>-1</sup> plus propiconazole @ 0.1%	10.56 abc	5.67	47.67	4.00	235.00	319.51
T4	Micronol @ 2 g l <sup>-1</sup> plus PGPR mix II @ 20 g l <sup>-1</sup>	10.59 <sub>abc</sub>	5.67	43.33	3.67	225.67	306.16
T5	Mg + Zn + B plus tebuconazole @ 0.1%	12.63 a	5.67	52.00	5.33	218.33	297.55
Т6	Mg + Zn + B plus copper hydroxide @ 0.25%	9.42 bcd	5.67	43.67	4.67	241.33	327.42
Τ7	Mg + Zn + B plus propiconazole @ 0.1%	11.03 abc	5.67	49.00	4.67	228.00	309.73
Т8	Mg + Zn + B plus PGPR mix II @ 20 g l <sup>-1</sup>	11.85 <sub>ab</sub>	5.67	50.67	5.33	218.67	298.05
Т9	Tebuconazole @ 0.1%	9.97 bc	5.33	46.33	4.67	225.33	305.68
T10	Copper hydroxide @ 0.25%	9.27 cd	5.00	41.33	3.67	231.00	313.86
T11	Propiconazole @ 0.1%	11.08 abc	5.33	49.33	4.67	222.67	302.73
T12	PGPR mix II @ 20 g l <sup>-1</sup>	10.78 abc	5.67	48.33	4.33	234.67	317.67
T13	Absolute control	6.99 <sup>d</sup>	5	41.00	3.33	267.00	358.01
	SE (±)	0.86	0.272	3.15	0.768	10.77	15.55
	CD (0.05)	(2.51)	NS	NS	NS	NS	NS

\*Mean of three replications

\*\*\*Figures followed by the same letter do not differ significantly according to two way ANOVA at P=0.05

NS : Non significant

# 4.3.2.3 Effect of Treatments on Yield of Banana

The average fruit yield plant <sup>-1</sup> is presented in Table 15. The results revealed that, all the treatments were significantly superior over control in order to increase the yield. The mean bunch weight ranged from 12.63 kg to 6.99 kg per plant.

The maximum bunch weight (12.63 kg) was obtained from the combination treatment T5, Mg + Zn + B plus tebuconazole @ 0.1% (Plate.4) followed by combination treatment T8, Mg + Zn + B plus PGPR @ 20 g l<sup>-1</sup> (11.85 kg) (Plate.5) which were on par and superior than the other six treatments such as T11 - propiconazole @ 0.1% (11.08 kg), T7 - Mg + Zn + B plus propiconazole @ 0.1% (11.03 kg), T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (10.98 kg), T12 - PGPR mix II @ 20 g l<sup>-1</sup> alone (10.78 kg), T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> (10.59 kg) and T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (10.56 kg) and was followed by T9 - tebuconazole @ 0.1% (9.97 kg), T2 - Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25% (9.77 kg), T6 - Mg + Zn + B plus copper hydroxide @ 0.25% (9.27 kg). However, all the treatments were superior over control. The lowest bunch weight (6.99 kg) was recorded in control (Plate.6).

# 4.3.2.4 Effect of Treatments on Number of Hands per Bunch

On comparison of the maximum number of hands per bunch representing each treatment, it was found that there was no significant difference between treatments. (Table 15).

The minimum number of hands per bunch was obtained in untreated check (5). The maximum number of hands (5.67) was found in eight treatments *viz.*, T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole (0.1%), T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole (0.1%), T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR @ 20 g l<sup>-1</sup>, T5 - Mg + Zn + B plus tebuconazole (0.1%) , T7 - Mg + Zn + B plus propiconazole (0.1%) , T7 - Mg + Zn + B plus propiconazole (0.1%) and T12 - PGPR mix II @ 20 g l<sup>-1</sup>.



Plate 4: Produce obtained from Mg+Zn+B plus Tebuconazole



Plate 5: Produce obtained from Mg+Zn+B plus PGPR Mix II



Plate 6: Produce obtained from the control plot

## 4.3.2.5 Effect of Treatments on Number of Fingers per Bunch

Comparison of average number of fingers per bunch of each treatments from the statistical analysis it was found that there were no significant difference between treatments. However, the most favorable effect was found in, T5 - Mg + Zn + B plus tebuconazole (0.1%) (52.00) followed by T8 - Mg + Zn + B plus PGPR mix II @ 20 g  $1^{-1}$  (50.67). The lowest number of fingers was found in untreated check (41.00). (Table 15).

#### 4.3.2.6 Effect of Treatments on Number of Suckers

The experimental results on number of suckers (Table 15) revealed no significant influence on the sucker production. However, the maximum number of sucker was observed in T5 - Mg + Zn + B plus tebuconazole @ 0.1% (5.33) and T8 - Mg + Zn + B plus PGPR mix II @ 20 g  $l^{-1}$  (5.33) in comparison with the T13, absolute control (3.33).

# 4.4 WEATHER PARAMETERS STUDIES

4.4.1 Correlation Studies on the Influence of Weather Parameters on the Intensity and Incidence of Sigatoka Disease of Banana Caused by *Mycosphaerella musicola* 

The parameters such as maximum and minimum temperature, maximum and minimum relative humidity, rainfall, evaporation, sunshine hours prevailing in the field at the time of field trials were correlated with disease intensity (disease index) and disease incidence so as to study the influence of meteorological factors on the development of Sigatoka leaf spot disease caused by *Mycosphaerella musicola* (yellow Sigatoka) of banana.

The statistical analysis of correlation studies revealed that, the disease was influenced by almost all the factors of weather *viz.*, Temperature (minimum), Relative Humidity (maximum and minimum), Rainfall and Evaporation. Both the

disease intensity and incidence showed a significant correlation with all the above factors.

It was found that, the minimum temperature, minimum relative humidity and evaporation showed a negative correlation with the disease intensity and disease incidence whereas, maximum relative humidity and rainfall showed a significant positive correlation with respect to disease intensity and disease incidence at 0.05% level of significance. The results are showed in Table16.

At 0.05% level (0.404), the minimum temperature was negatively (-0.643) correlated with disease intensity as well as disease incidence (-0.527). Statistically, the influence of minimum relative humidity was -0.506 with respect to disease intensity and it was -0.554 with respect to disease incidence at 0.05% level of significance. Also, evaporation showed the inverse relation with disease intensity (-0.578) and with disease incidence (-0.500).

There was a positive correlation of maximum relative humidity (0.679) and rainfall (0.549) with disease intensity at 0.05% level (0.404) of significance. Also, the same parameters are positively correlated with disease incidence with values of 0.610 and 0.542 respectively. Therefore it can be inferred that high relative humidity and rainfall contributes to increases in disease incidence and disease intensity.

# 4.5 ECONOMIC ANALYSIS

The economics of cultivation was worked out in terms of input cost, labour charges and treatment cost and is given in appendix IV and V.

The economic analysis was worked out in terms of both net income and BCR and is presented in Table 17.

The results revealed that the lowest cost of cultivation (4,76,378 Rs.ha<sup>-1</sup>) was for T13 (Control) followed by T9- tebuconazole @ 0.1% (5,10,290 Rs.ha<sup>-1</sup>), T11- propiconazole @ 0.1% (5,10,618 Rs.ha<sup>-1</sup>), T10- copper hydroxide @ 0.25% (5,10,868 Rs.ha<sup>-1</sup>), T6 - Mg + Zn + B plus copper hydroxide @ 0.25% (5,12,340

Table 16. Correlation studies on the influence of various weather parameters on the intensity and incidence of banana Sigatoka disease caused by *Mycosphaerella musicola* 

Parameters	Max Temp	Min Temp.	max. RH	min. RH	Rainfall (mm)	Evaporation (mm)	Sunshine hours
Disease intensity	-0.143	-0.643*	0.679*	-0.506	0.549*	-0.578*	-0.304
Disease incidence	0.0379	-0.527*	0.610*	- 0.554*	0.542*	-0.500*	-0.216

\*Correlation coefficient at 0.05 % level= 0.404

#### Table 17. Economic analysis

Treatments	Cost of cultivation	Income (Rs. ha <sup>-1</sup> )	Net profit	B:C ratio
	(Rs.ha <sup>-1</sup> )	(Fruits : Rs. 30 / Kg	$(Rs.ha^{-1})$	
		Suckers : Rs. 7/ sucker)		
T1	512990 /-	Fruits : 823500 /-	380510 /-	1.74
		Suckers : 70000 /-		
		Total : 893500 /-		
T2	513030 /-	Fruits : 732750 /-	289720 /-	1.56
		Suckers : 70000 /-		
		Total : 802750 /-		
T3	518960 /-	Fruits : 792000/-	343040 /-	1.66
		Suckers : 70000/-		
		Total : 862000 /-		
T4	512880 /-	Fruits : 794250 /-	333870 /-	1.65
		Sucker : 52500 /-		
		Total : 846750 /-		
T5	947250 /-	Fruits : 947250 /-	519260 /-	2.01
		Sucker : 87500 /-		
		Total : 1034750 /-		
T6	512340 /-	Fruits : 706500 /-	264160 /-	1.52
		Sucker : 70000 /-		
		Total : 776500 /-		
Τ7	512590 /-	Fruits : 827250 /-	384660/-	1.75
		Sucker : 70000 /-		
		Total : 897250 /-		
T8	518896 /-	Fruits : 888750 /-	457354 /-	1.88
		Sucker : 87500/-		
		Total : 976250/-		
Т9	510290 /-	Fruits : 747750 /-	307460 /-	1.60
		Sucker : 70000/-		
		Total : 817750/-		
T10	510868 /-	Fruits : 695250/-	236882 /-	1.46
-		Sucker : 52500/-		
		Total : 747750/-		
T11	510618 /-	Fruits : 831000/-	390382 /-	1.76
		Sucker : 70000/-		
		Total : 901000/-		
T12	515924 /-	Fruits : 808500/-	362576 /-	1.70
		Sucker : 70000/-		
		Total : 878500/-		
T12	47(270 /	E 1 524250/	100272 /	1.01
T13	476378 /-	Fruits : 524250/-	100372 /-	1.21
		Sucker : 52500/-		
		Total : 576750/-		
1				

Rs.ha<sup>-1</sup>), T7 - Mg + Zn + B plus propiconazole @ 0.1% (5,12,590 Rs.ha<sup>-1</sup>), T4 - Micronol @ 2 g l<sup>-1</sup> of water plus PGPR mix II @ 20 g l<sup>-1</sup> (5,12,880 Rs.ha<sup>-1</sup>), T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (5,12,990 Rs.ha<sup>-1</sup>), T2 -Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25% (5,13,030 Rs.ha<sup>-1</sup>), T5 - Mg + Zn + B plus tebuconazole @ 0.1% (5,15,490 Rs.ha<sup>-1</sup>), T12 - PGPR mix II @ 20 g l<sup>-1</sup> (5,15,924 Rs.ha<sup>-1</sup>), T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> (5,18,960 Rs.ha<sup>-1</sup>), T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (5,18,960 Rs.ha<sup>-1</sup>).

Highest income was obtained for T5 - Mg + Zn + B plus tebuconazole @ 0.1% with 10,34,750 /- Rs.ha<sup>-1</sup> followed by T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> (9,76,250 /- Rs. ha<sup>-1</sup>), T11- propiconazole @ 0.1% (9,01,000 /- Rs. ha<sup>-1</sup>), T7 - Mg + Zn + B plus propiconazole @ 0.1% (8,97,250 /- Rs.ha<sup>-1</sup>), T1 (8,93,500 /- Rs.ha<sup>-1</sup>), T12 - PGPR mix II @ 20 g l<sup>-1</sup> (8,78,500 /- Rs.ha<sup>-1</sup>), T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (8,62,000 /- Rs.ha<sup>-1</sup>), T4 - Micronol @ 2 g l<sup>-1</sup> of water plus PGPR mix II @ 20 g l<sup>-1</sup> (8,46,750 /- Rs.ha<sup>-1</sup>), T9 - tebuconazole @ 0.1% (8,17,750 /- Rs.ha<sup>-1</sup>), T2 - Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25% (8,02,750 /- Rs.ha<sup>-1</sup>), T6 - Mg + Zn + B plus copper hydroxide @ 0.25% (7,76,500 /- Rs.ha<sup>-1</sup>), T10 - copper hydroxide @ 0.25% (7,47,750 /- Rs.ha<sup>-1</sup>) and the lowest was obtained in T13-control (5,76,750 /- Rs.ha<sup>-1</sup>).

# DISCUSSION

#### **5. DISCUSSION**

Sigatoka leaf spot disease caused by *Mycosphaerella musicola* is a limiting factor for banana production in India and other worldwide countries. It has considerable social and agricultural economic impacts due to lowering of crop yields. It was reported that more than 50 per cent of economic losses occurs due to this disease across the world. Fungicides have still become the major tool in mitigating development and reducing effect of the diseases. An experimental trial was conducted during the year 2013 (March-December) at Coconut Research Station, Balaramapuram, Trivandrum district, Kerala in Nendran banana. In this study, emphasis had been laid on application of certain micronutrients, secondary nutrient with biocontrol agent/ plant protection chemicals to combat the disease in banana.

# **5.1 DISEASE FACTORS**

#### 5.1.1 Disease incidence

The field trials were conducted to check the efficacy of fungicides (systemic and contact), bio control agent (PGPR mix II) through their alone treatments and combination treatments on Sigatoka leaf spot in banana, cv. Nendran. The experimental results with respect to disease incidence revealed a significant variation due to application of fungicides/ *Pseudomonas fluorescens* with or without secondary and micro nutrients. The minimum disease incidence was recorded by foliar application of tebuconazole in combination with either Micronol or Mg + Zn +B. All the combination treatments of fungicides or PGPR along with either Micronol or Mg + Zn +B was also effective in combating the disease incidence during certain crop period. This exhibited the role of secondary and micronutrients.

The promising organic treatments when compared to control (Fig.2) are in the following order *viz.*, copper hydroxide @ 0.25% (T10), Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25% (T2), Mg + Zn + B plus copper hydroxide @

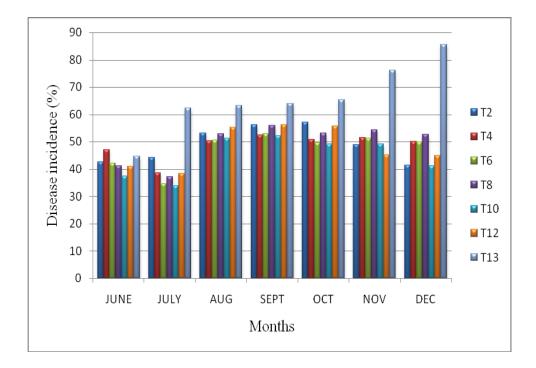


Fig. 2. Effect of organic treatments on disease incidence (%)

T2- Micronol @ 2g l<sup>-1</sup> plus copper hydroxide @ 0.25% T4- Micronol @ 2g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> T6- Mg + Zn +B plus copper hydroxide @ 0.25%, T8- Mg + Zn +B plus PGPR mix II @ 20 g l<sup>-1</sup> T10- Copper hydroxide @ 0.25% T12- PGPR mix II @ 20g l<sup>-1</sup> T13- Control 0.25% (T6), Micronol @ 2 g  $l^{-1}$  plus PGPR mix II @ 20 g  $l^{-1}$  (T4), Mg + Zn +B plus PGPR mix II @ 20 g  $l^{-1}$  (T6), PGPR mix II @ 20 g  $l^{-1}$  (T12).

The promising inorganic treatments when compared to control (Fig.3) are in following order *viz.*, Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (T1), Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (T3), Mg + Zn +B plus tebuconazole @ 0.1% (T5), Mg + Zn +B plus propiconazole @ 0.1% (T7), tebuconazole @ 0.1% (T9) and propiconazole @ 0.% (T11).

In 2008, Ganry *et al.*, reported similar results with aerial application of azole fungicides such as triazoles, benzimidazoles and strobilurins in suppressing the development of Sigatoka leaf spot disease in banana. Similar results by copper fungicides were reported by Tollenaar (1961), when he studied the efficacy of copper and oil in the control of Sigatoka leaf spot disease.

#### 5.1.2 Disease intensity

With regard to Sigatoka leaf spot intensity of banana cv. Nendran, the foliar application of tebuconazole, propiconazole and copper hydroxide alone or in combination with Micronol (micronutrient mixture) or Mg + Zn + B was highly effective in reducing the disease intensity. Hence, the results of the study indicated that, foliar spray of new generation systemic fungicides (tebuconazole or propiconazole) or the contact fungicide (copper hydroxide) gave better suppression of Sigatoka disease.

Management practices like the use of resistant cultivars, cultuaral methods and quarantine measures have been recommended to control Sigatoka disease, but the most reliable tool has been the use of chemical products, mainly site specific fungicides (Jones, 2000; Marin *et al.*, 2003; Gasparotto *et al.*, 2005). The fact that disease control depends upon fungicide application is in conformity with the findings of FRAC (2010), as different groups of fungicides such as demethylation inhibitors (DMI), Amine fungicides, Qo inhibitors (QoI), Anilinopyrimidines (AP), benzimidazoles (BCM), succinate dehydrogenase inhibitors (SDHI)

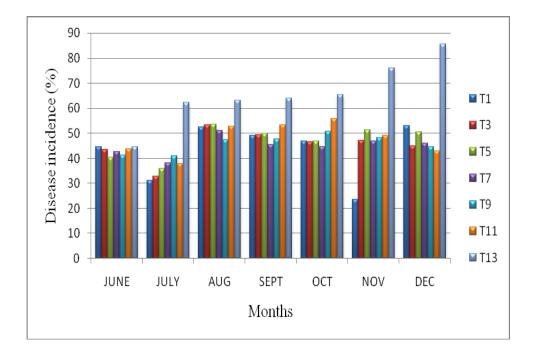


Fig. 3. Effect of inorganic treatments on disease incidence (%)

T1- Micronol @ 2 g  $1^{-1}$  plus tebuconazole @ 0.1% T3- Micronol @ 2 g  $1^{-1}$  plus propiconazole @ 0.1% T5- Mg + Zn + B plus tebuconazole @ 0.1%, T7- Mg + Zn + B plus propiconazole @ 0.1% T9- Tebuconazole @ 0.1% T11- Propiconazole @ 0.1% T13- Control fungicides and guanidines are used to control Sigatoka disease in banana plantations. Unlike the protectants, systemic fungicides inhibited the development of symptoms after they first appeared. The efficacy of systemic fungicides in Sigatoka disease management has been reported by Stover (1990).

Continuous use of the systemic fungicides is reported to increase the risk of development of resistance to these fungicides in both *Mycosphaerella musicola and Mycosphaerella fijiensis* (Mourichon *et al.*, 1997 and Duvert *et al.*, 2002). Hence, Copper hydroxide can be also included in spray schedule in order to avoid resistance development due to the continuous use of systemic chemicals. Similar findings have been reported by Mourichon *et al.*, 1997 and Duvert *et al.*, 2002.

In the present study, it is clear that, there was a gradual increase in both the disease intensity and incidence in absolute control. In all the observations, with respect to disease incidence, it ranged from 62.35 to 85.55% and 49.96 to 76.42 % in case of disease intensity. Selvarajan *et al.* (2000) recorded disease severity indices as 14.4 and 49.64 in Nendran and Robusta cultivars respectively for black Sigatoka in Thrissur district.

The promising organic treatments when compared to control (Fig.4) are Mg + Zn + B plus copper hydroxide @ 0.25% (T6), copper hydroxide @ 0.25% (T10), Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25% (T2) , Mg + Zn + B plus PGPR @ 20 g l<sup>-1</sup> (T8), Micronol @ 2 g l<sup>-1</sup> plus PGPR @ 20 g l<sup>-1</sup> (T4) and PGPR @ 20 g l<sup>-1</sup> (T12).

The promising inorganic treatments when compared to control (Fig.5) are Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (T1), Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (T3), Mg + Zn + B plus tebuconazole @ 0.1% (T5), Mg + Zn + B plus propiconazole @ 0.1% (T7), Tebuconazole @ 0.1% (T9) and propiconazole @ 0.1% (T11).

The development of Panama disease on banana caused by *Fusarium* oxysporum sp. cubensis is supposedly related to nutritional imbalances especially

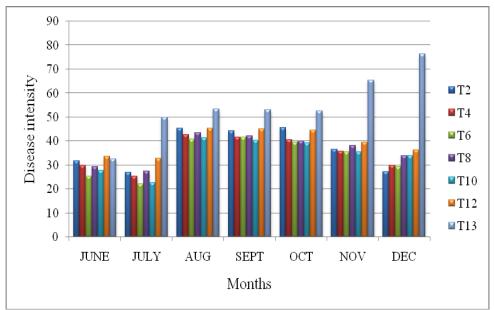


Fig.4. Effect of organic treatments on disease intensity

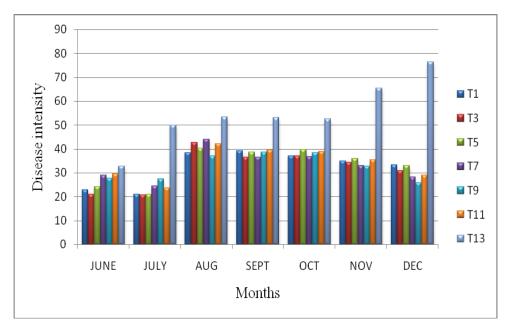


Fig.5. Effect of inorganic treatments on disease intensity

**T1**- Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1%, **T2**- Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25%, **T3**- Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1%, **T4**- Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup>, **T5**- Mg + Zn + B plus tebuconazole @ 0.1%, **T6**-Mg + Zn + B plus copper hydroxide @ 0.25%, **T7**- Mg + Zn + B plus propiconazole @ 0.1%, **T8**- Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup>, **T9**- Tebuconazole @ 0.1%, **T10**- Copper hydroxide @ 0.25%, **T11**- propiconazole @ 0.1%, **T12**- PGPR mix II @ 20 g l<sup>-1</sup>, **T13**- Control Zinc, Calcium and Magnesium (Borges, 1983). Fertilizer reduced leaf thickness but micronutrients tended to increase leaf thickness. It was also proved that micronutrients B, Cu, Mn and Zn had a direct inhibitory effect on fungal growth and development and Cu and Zn resulted in significant inhibition of *Mycosphaerella fijiensis* Morelet.

The results of this study reveal that foliar application of new generation systemic fungicides such as tebuconazole, propiconazole and the contact fungicide copper hydroxide can be included in the spray schedule for the management of Sigatoka leaf spot disease to avoid the development of resistance as reported by Mourichon *et al.*,1997 and Duvert *et al.*, 2002). In Karnataka at Munavalli and Ramadurga, two sprays of propiconazole 0.05% effectively controlled the disease and recorded the lowest percent disease index (Tammaiah *et al.*, 2008). Patel (2009) concluded that propiconazole, hexaconazole, tridemorph and carbendazim remained significantly at par with each other and recorded least leaf spots followed by tebuconazole and companion (combination of mancozeb and carbendazim).

The efficacy of *P. fluorescens* against fungal and bacterial diseases has been well documented by various groups. Disease reduction due to application of PGPR could be attributed to the direct / indirect action of biocontrol agent that might have boost the host defense mechanism by various means of its activity. In 2005, Rodriguez- Romero reported the similar findings in case of banana, *viz.*, combined application of arbuscular mycorrhizal fungi and PGPR highly benefits banana plants by induced systemic resistance against fungi, bacteria and viruses, as well as to enhance plant growth and therefore, it could be considered during the acclimatization stage of micropropagated banana. Saravanakumar and Samiyappan (2007) also reported about the effectiveness of PGPR against early and late leaf spot in groundnut.

In this field study also, it was observed that application of secondary/ micronutrients with plant protection chemicals/ PGPR has resulted in the decreased disease incidence and severity during most of the crop period.

### **5.2 BIOCHEMICAL ANALYSIS**

#### 5.2.1 Phenol and OD-phenol

Total phenol and OD-phenol (ortho dihydroxy phenol) content in banana leaves during fourth, fifth and sixth month old plants were estimated and the results revealed that there is a significant variation in phenol content among the treatments during one of the active growth period (fifth month). The phenol content was higher in the propiconazole applied plants which clearly indicates its efficacy and activation of defense mechanism. This was followed by the combination treatment of the same with micronutrients Mg + Zn + B which is also having the role in phenolic compound production. Results of the present study agree with findings of Graham and Webb, 1991 and Marschner in 1995 who opined that boron reduces diseases caused by *Plasmodiophora brassicae* in crucifers, *Fusarium solani* in bean, *Verticillium albo-atrum* in tomato and cotton, tobacco mosaic virus in bean, tomato yellow leaf curl virus in tomato, *G.graminis* (Sacc.) (Graham and Webb, 1991) and *Blumeria graminis* in wheat (Marschner, 1995).

### 5.2.2 Chlorophyll

The total chlorophyll content in banana leaves was estimated and the results exhibited no significant variation due to different treatment application. Effect of treatments on chlorophyll content is given in Fig.6. Even though it lacks significance, higher value was showed by the treatment T5 - Mg + Zn + B plus Tebuconazole @ 0.1%. This was followed by treatment T8, Mg + Zn + B plus PGPR @ 20 g  $\Gamma^1$  which is a combination treatment of PGPR along with micronutrients. Both the treatments given in combination with magnesium (Shaul, 2002). Magnesium is essential for chlorophyll and enzyme activation for photosynthesis. Leaf spots can coalesce and lead to premature death of large areas

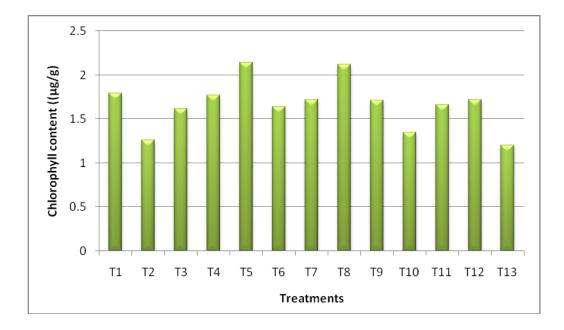


Fig. 6. Effect of treatments on chlorophyll content

T1- Micronol @ 2g l<sup>-1</sup> plus tebuconazole @ 0.1% T2- Micronol @ 2g l<sup>-1</sup> plus copper hydroxide @ 0.25% T3- Micronol @ 2g l<sup>-1</sup> plus propiconazole @ 0.1% T4- Micronol @ 2g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> T5- Mg + Zn + B plus tebuconazole @ 0.1% T6- Mg + Zn + B plus copper hydroxide @ 0.25% T7- Mg + Zn + B plus propiconazole @ 0.1% T8- Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> T9- Tebuconazole @ 0.1% T10- Copper hydroxide @ 0.25% T11- propiconazole @ 0.1% T12- PGPR mix II @ 20 g l<sup>-1</sup> T13- Control of leaf tissue reducing the photosynthetic capabilities of the plant. The disease can also disturb the physiology of the fruits, resulting in premature or uneven ripening (Jones, 1999).

The two treatments (T5 and T8) was followed by T1 (Micronol @ 2 g  $\Gamma^1$  plus Tebuconazole @ 0.1%) and T4 (Micronol @ 2 g  $\Gamma^1$  plus PGPR @ 20 g  $\Gamma^1$ ) both containing manganese. Similar results were reported by Chatterjee, 1989; Kavino *et al.*, 2010. It was observed an improved plant height, girth, LAI, bunch weight, number of hands and chlorophyll content when compared to untreated plants. Maintaining the physiological characters at a higher level is essential to increase biomass and banana yield. Higher chlorophyll content observed could be related to higher activities of iron containing enzymes such as catalase and peroxidase which are enhanced during the application of PGPR. The role of these enzymes in chlorophyll synthesis is an established factor (Singh, 1988). Manganese is directly or indirectly involved in chloroplast formation. Excess manganese caused reduction in chlorophyll concentration. A poor correlation between chlorophyll content and the reflectance indices was also found after *Mycosphaerella* leaf infection in young eucalyptus (Pietrzykowski *et al.*, 2006).

Among all the treatments, Mg + Zn + B plus Tebuconazole @ 0.1% (T5) followed by Mg + Zn + B plus PGPR @ 20 g l<sup>-1</sup> (T8) recorded the maximum percentage increase in chlorophyll content over control (Fig.7) and these were the first two treatments which showed the maximum percentage increase in number of functional leaves also. With respect to this attribute also the growth promotion of PGPR is visible.

# 5.2.3 Protein

Results of protein content in banana leaves indicated no significant variation due to treatment application. Eventhough it lacks significance, the lowest value was showed by T1 - Micronol @ 2 g  $l^{-1}$  plus tebuconazole @ 0.1% followed by T3 - Micronol @ 2 g  $l^{-1}$  plus propiconazole @ 0.1%. The use of systemic fungicides caused a significant decrease in the total protein content of wheat

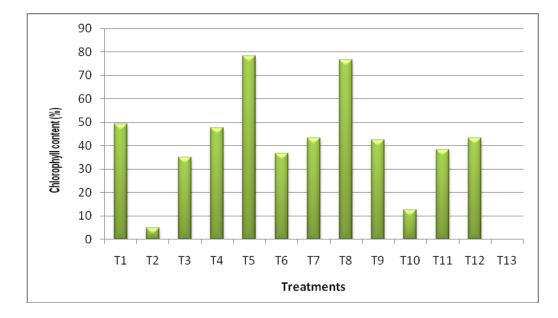


Fig. 7. Percentage increase in chlorophyll content over control

T1- Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% T2- Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25% T3- Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% T4- Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> T5- Mg + Zn + B plus tebuconazole @ 0.1% T6- Mg + Zn + B plus copper hydroxide @ 0.25% T7- Mg + Zn + B plus propiconazole @ 0.1% T8- Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> T9- Tebuconazole @ 0.1% T10- Copper hydroxide @ 0.25% T11- propiconazole @ 0.1% T12- PGPR mix II @ 20 g l<sup>-1</sup> T13- Control cultivars. It has been suggested that the toxicant produced by the application of systemic fungicides inhibits protein synthesis by binding to the larger ribosomal subunits including change in the enzyme system, ceasing ATP and NADP formation (Person *et al.*, 1975; Siddiqui, 1997) leading to decreased disease incidence with increase in yield.

In all the alone treatments, which lacks the micronutrient, zinc, the protein content was comparatively low and Marschner (1995) has reported the same result. Zinc is essential for carbohydrate formation and an enzyme activator involved in protein, hormone, RNA and DNA synthesis and growth regulation (Marschner, 1995; Graham and McDonald, 2001).

# 5.2.3 Plant Secondary and Micro Nutrients content

Micronutrients have considerable influence on the resistance or tolerance mechanisms to pathogens in plants (Krauss, 1999).

Jimenez in 2008 investigated the role of micronutrients on the reduction of disease and enhanced plant growth in greenhouse and field conditions. The response of the fungus to some nutrients was investigated especially those which have had an effect on plant defence in other crops. B, Cu, Mn and Zn delayed the fungus development in their presence but after removal from the medium only Cu had a lasting inhibitory effect on the fungus.

Boron is involved in forming a barrier against pathogen invasion. Boric acid application was shown to be a good inhibitor of ascospore germination under in vitro and field conditions and this is in conformity with the following findings.

Boron in combination with fungicide increase resistance to *Cercospora* spp. as it increased the biosynthesis and oxidation of phenolic compounds relating to changes in the cell wall lignifications (Ruiz *et al.*, 1998. and Rodrigues *et al.*, 2004). Boric acid (1% - 5%) was shown to be a good inhibitor of ascospore germination under *in vitro* conditions and a potential alternative for benlate. Similar results were found under field conditions against ascospore germination of

the pathogen *Eutypa lata* which causes the dieback disease of grapevines (Rolshausen and Gubler, 2005). The effectiveness of boric acid, zinc sulphate, magnesium sulphate were reported by several other groups also (Graham and Webb, 1991, Whiley *et al.*, 1991; Labeda *et al.*, 2001; Baider and Cohen, 2003).

Zinc applications and *P. fluorescens* together contributed in controlling Fusarium disease in wheat and tomatoes. Numerous reports confirmed the role of zinc in relation to plant diseases (Siddique *et al.*, 2002; Baider and Cohen, 2003).

Copper has a large after effect, confirming the usefulness of Cu containing fungicides for Sigatoka control in the past. Copper is a regulator of various enzyme systems linked to plant defense and the production of antimicrobial compounds (Lebeda *et al.*, 2001).

The leaf nutrient content mainly magnesium, boron and zinc at bunch emergence stage was estimated and revealed that there is a significant difference due to the treatment application. The highest magnesium was noticed in T8 - Mg + Zn + B plus PGPR @ 20 g l<sup>-1</sup>. This clearly indicates the increase in uptake of minerals due to the presence of PGPR followed by T5 - Mg + Zn + B plus Tebuconazole (0.1%). All the four combination treatments contain magnesium showed comparatively higher magnesium content. As reported by Shaul (2002), Magnesium is essential for chlorophyll, enzyme activation and indispensible for photosynthesis.

In the present study, the highest boron content was noticed in combination treatment, T8 - Mg + Zn + B plus PGPR @ 20 g  $l^{-1}$  which contain both the zinc and PGPR. Similar findings were reported earlier by many groups. PGPR play an important role in suppression of soil-borne plant pathogens, and improving nutrient availability (Biswas *et al.*, 2000; Saravanakumar and Samiyappan, 2007).

Regarding the studies of zinc content, the treatment T4 - Micronol @ 2 g  $l^{-1}$  plus PGPR @ 20 g  $l^{-1}$  recorded the maximum content of zinc. This may be due to

two factors namely, Micronol and PGPR. Micronol is a rich source of zinc and PGPR enhance the uptake of minerals as mentioned in many reports.

# **5.3 BIOMETRIC OBSERVATIONS**

# 5.3.1 Effect of Treatments on Plant Growth Characterestics 5.3.1.1 Height and Girth of the Plant

With regard to plant height and girth, there was no significant variation due to treatment application. The maximum height and girth was obtained by T5 - Mg + Zn + B plus Tebuconazole (0.1%) followed by T8 - Mg + Zn + B plus PGPR (a) 20 g l<sup>-1</sup> respectively. Hence, it is clearly expressed the efficacy of fungicide and the growth enhancement property of PGPR. *Pseudomonas flourescens* are the most effective rhizosphere bacteria. Because in addition to disease control, they exert beneficial effect on plant growth promotion (Kloepper *et al.*, 1980). Similar results were reported by many other workers *viz.*, Harish *et al.*, 2009a; Kavino *et al.*, 2010.

#### 5.3.1.2 Number of Leaves and Functional Leaves

Application of fungicides or PGPR with or without micro/ secondary nutrients showed a significant variation in the number of leaves and functional leaves in the later crop stage. During the last post treatment observation, the maximum number of leaves and functional leaves were noticed in treatments with PGPR alone and combination treatments of PGPR along with Micronol or Mg + Zn + B. This indicates the growth enhancement property of PGPR (Fig.8).

The treatments, T5 - Mg + Zn + B plus tebuconazole @ 0.1% (inorganic), T8 - Mg + Zn + B plus PGPR @ 20 g  $1^{-1}$  (organic) and T4 - Micronol @ 2 g  $1^{-1}$ plus PGPR @ 20 g  $1^{-1}$  (organic) recorded the maximum percentage increase in number of functional leaves over control (Fig.9). T5 was one of the best treatments that rated as best for disease incidence and intensity reduction. Figure 9 also revealed that all the organic and inorganic treatments showed a good

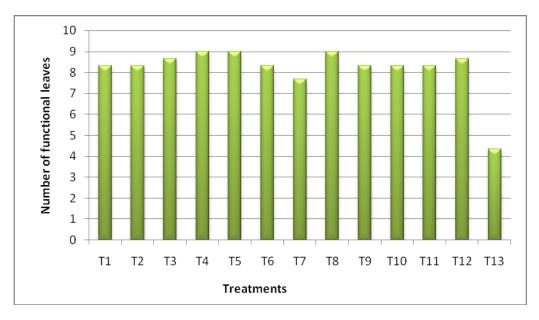


Fig.8. Effect of treatments on number of functional leaves (At the month of harvest)

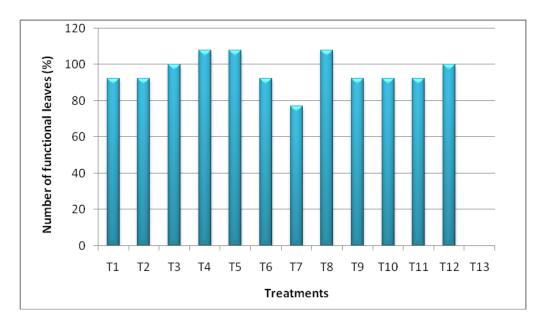


Fig.9. Percentage increase in number of functional leaves over control

**T1-** Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1%, **T2-** Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25%, **T3-** Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1%, **T4-** Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup>, **T5-** Mg + Zn + B plus tebuconazole @ 0.1%, **T6-** Mg + Zn + B plus copper hydroxide @ 0.25%, **T7-** Mg + Zn + B plus propiconazole @ 0.1%, **T8-** Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup>, **T9-** Tebuconazole @ 0.1%, **T10-** Copper hydroxide @ 0.25%, **T11-** propiconazole @ 0.1%, **T12-** PGPR mix II @ 20 g l<sup>-1</sup>, **T13-** Control

percentage increase over control in which, the combination and alone treatments of PGPR scored the highest.

This is in conformity with the findings of Okon *et al* (1988) and Sarig *et al* (1988) in the case of banana under field conditions.

#### 5.3.2 Effect of Treatments on Yield Parameters

A significant difference in yield was observed due to treatment application. The highest yield was noticed in T5 - Mg + Zn + B plus tebuconazole @ 0.1% followed by T8 - Mg + Zn + B plus PGPR @ 20 g l<sup>-1</sup>. Significant increases in crop yields following application of PGPR have been documented under diverse field conditions (Bashan, 1998). Harish *et al.*, (2009) observed that polyphenol oxidase (PPO) and peroxidase (PO) activities were greater in the plants treated with mixtures of rhizobacteria and endophytic bacteria and challenged with viruliferous aphids, compared to control plants. *P. fluorescens*, Pf1 along with the endophytic bacterial isolate significantly increased the yield compared to other treatments and the untreated control. In 2010, Kavino *et al.*, reported about the enhanced bunch weight, plant height, girth, LAI, number of hands and chlorophyll content when plants treated with *P. fluorescens* strain CHAO and chitin bioformulations of *Pseudomonas* strains. Many other workers reported similar results *viz.*, Okon *et al.* (1988), Sarig *et al.* (1988), Harish *et al.* (2009).

With respect to number of hands, the eight combination treatments showed higher values over control. In 1984, Obeifuna reported about the same results in the same crop. In a banana bunch, the number of hands is an important yield component and is normally decided at the time of fruit bud initiation and differentiation which is generally influenced by better nutrition.

The best organic and inorganic treatments which produced higher yield are given in Fig.10 and Fig.11 respectively. The promising organic treatments are in the following order *viz.*, Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> (T8), PGPR mix II @ 20 g l<sup>-1</sup> (T12), Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> (T4),

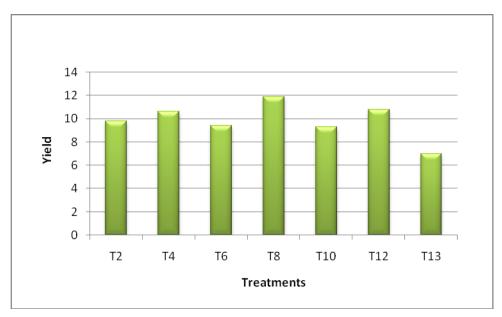


Fig. 10. Effect of organic treatments on yield

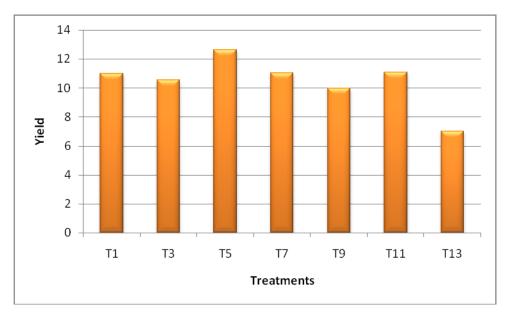


Fig. 11. Effect of inorganic treatments on yield

**T1-** Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1%, **T2-** Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25%, **T3-** Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1%, **T4-** Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup>, **T5-** Mg + Zn + B plus tebuconazole @ 0.1%, **T6-** Mg + Zn + B plus copper hydroxide @ 0.25%, **T7-** Mg + Zn + B plus propiconazole @ 0.1%, **T8-** Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup>, **T9-** Tebuconazole @ 0.1%, **T10-** Copper hydroxide @ 0.25%, **T11-** propiconazole @ 0.1%, **T12-** PGPR mix II @ 20 g l<sup>-1</sup>, **T13-** Control

Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25% (T2), Mg + Zn + B plus copper hydroxide @ 0.25% (T6) and Copper hydroxide @ 0.25% (T10). The promising inorganic treatments are in the following order *viz.*, Mg + Zn + B plus tebuconazole @ 0.1% (T5), Propiconazole @ 0.1% (T11), Mg + Zn + B plus propiconazole @ 0.1% (T7), Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (T1), Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1% (T3) and tebuconazole @ 0.1% (T9).

#### **5.4 WEATHER PARAMETERS STUDIES**

Influence of weather parameters on the Sigatoka leaf spot disease of banana was studied and the disease was influenced by almost all the factors of weather *viz.*, atmospheric temperature, relative humidity, rainfall and evaporation. Both the disease intensity and incidence showed a significant correlation with all the above factors of weather. Similar results have been recorded by Stover (1968); Simmonds (1939); Brun (1963); Stahel (1937); Leach (1946); Elangovan *et al* (1990) and Perez *et al* (2000a; 2000b).

The climatic variables that positively influenced the Sigatoka disease progression in banana were rainfall, relative humidity and leaf wetness.

#### 5.5 ECONOMIC ANALYSIS STUDIES

The economics of cultivation was worked out in terms of input cost, labor charges and treatment cost. The economic analysis was also worked out in terms of both net income and BCR and is presented in Table 17. It showed higher net income and BCR in all the treatments compared to control as in the case of yield.

The effect of treatments on B: C ratio (Fig.12) reveals that, all the organic and inorganic treatments recorded a higher B: C ratio when compared to control (T13). The highest B:C ratio was recorded by Mg + Zn + B plus tebuconazole @ 0.1% (T5) which was the treatment showed the highest percentage increase in number of functional leaves, chlorophyll content and the highest yield also. This

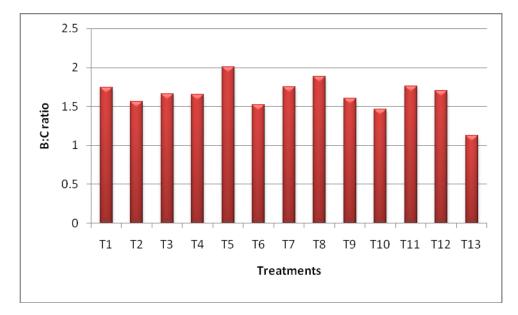


Fig. 12. Effect of treatments on B:C ratio

**T1-** Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1%, **T2-** Micronol @ 2 g l<sup>-1</sup> plus copper hydroxide @ 0.25%, **T3-** Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1%, **T4-** Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup>, **T5-** Mg + Zn + B plus tebuconazole @ 0.1%, **T6-** Mg + Zn + B plus copper hydroxide @ 0.25%, **T7-** Mg + Zn + B plus propiconazole @ 0.1%, **T8-** Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup>, **T9-** Tebuconazole @ 0.1%, **T10-** Copper hydroxide @ 0.25%, **T11-** propiconazole @ 0.1%, **T12-** PGPR mix II @ 20 g l<sup>-1</sup>, **T13-** Control

inorganic treatment is followed by T8 - Mg + Zn + B plus PGPR mix II @ 20 g  $l^{-1}$  which recorded for the second best yield.

The least cost of cultivation  $(5,10,290 \text{ Rs.ha}^{-1})$  was found in case of tebuconazole @ 0.1% (T9, inorganic treatment) and the highest net profit of 5,19,260 Rs.ha<sup>-1</sup> and BCR of 2.01 were obtained in case of inorganic treatment, Mg + Zn + B plus tebuconazole @ 0.1%. The results indicated that, this is the best treatment for banana (cv. Nendran) production.

Organic treatments were also found to be profitable as revealed from the net income obtained ranging from Rs.7,47,750 to 9,76,250 ha<sup>-1</sup>.

The analysis of the results indicated that, among 13 treatments, foliar application of tebuconazole (0.1%) in combination with either Micronol (2 g l<sup>-1</sup>) or Mg (2 g l<sup>-1</sup>) + Zn (3 g l<sup>-1</sup>) + B (2 g l<sup>-1</sup>) followed by Micronol @ 2 g l<sup>-1</sup>plus propiconazole (0.1%) and copper hydroxide (0.25%) recorded minimum disease incidence during the study. Hence, the ideal inorganic treatments selected are *viz.*, tebuconazole and propiconazole in combination with Micronol (T1 and T3), Mg + Zn + B plus tebuconazole (T5) and copper hydroxide (T10) rated as the best organic treatment with respect to disease incidence control.

With respect to disease intensity, the analysis of the results indicated that the application of new generation fungicides (tebuconazole, propiconazole and copper hydroxide) alone or in combination with Micronol or Mg + Zn + B was effective in reducing the intensity of Sigatoka leaf spot disease. The best organic treatment to reduce intensity of the disease was found to be the application of copper hydroxide alone or in combination with Micronol / Mg + Zn + B. The application of new generation systemic fungicides alone or in combination can be rated as the ideal inorganic treatments.

The suppression of the disease by the application of these inorganic and organic treatments is due to fungal growth inhibition, activation of defense related mechanisms *viz.*, increase in phenol production and thereby enhanced plant growth and yield.

The economic analysis also revealed that the best inorganic treatment is application of Mg + Zn + B plus tebuconazole (0.1%) which recorded the highest yield, net income and benefit cost ratio which is followed by Mg + Zn + B plus PGPR (20 g  $l^{-1}$ ) under organic treatments.

For the integrated management of banana Sigatoka leaf spot disease (caused by *M. musicola* R. Leach ex J.L. Mulder) in humid tropical zone of Kerala, the best treatment was found to be; Mg (2 g  $l^{-1}$ ) + Zn (3 g  $l^{-1}$ ) +B (2 g  $l^{-1}$ ) plus tebuconazole (0.1%) with respect to maximum disease suppression, disease intensity reduction and highest yield.

# SUMMARY

#### 6. SUMMARY

Effect of treatments on the incidence of banana Sigatoka leaf spot disease caused by *M. musicola* revealed that among the 13 treatments, minimum disease incidence was observed with T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole (0.1%), T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole (0.%), T5 - Mg + Zn + B plus tebuconazole (0.1%) and T10 - copper hydroxide (0.25%).

All the treatments except absolute control (T13) were statistically onpar with each other. Among the 13 treatments, maximum disease suppression was observed with T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1%, T3- Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1%, T5 - Mg + Zn + B plus tebuconazole @ 0.1%, T6 - Mg + Zn +B plus copper hydroxide @ 0.25, T7 - Mg + Zn + B plus propiconazole @ 0.1%, T9 - tebuconazole @ 0.1%, T10 - copper hydroxide @ 0.25% and T11 - propiconazole @ 0.1%. The application of fungicides tebuconazole, propiconazole and copper hydroxide alone or in combination with Micronol or Mg + Zn + B was effective in reducing the disease intensity of Sigatoka leaf spot in banana.

The pathophysiological studies revealed about defense related phenol and OD- phenol, chlorophyll content, protein content and leaf nutrient content etc.

With respect to the biochemical changes, a significant difference in phenol and OD- phenol content was observed in five months old crop. Among them, the treatment T11- propiconazole (a) 0.1% showed the maximum phenol content (3705.17 µg g<sup>-1</sup>) followed by T7 - Mg + Zn + B plus propiconazole (a) 0.1% (2430.00 µg g<sup>-1</sup>) and T5 - Mg + Zn + B plus tebuconazole (a) 0.1% (2082.60 µg g<sup>-1</sup>) which was on par with each other. The lowest phenol content (555.00 µg g<sup>-1</sup>) was noticed in case of T12, PGPR mix II (a) 20 g l<sup>-1</sup>.

The defense related OD - phenol content was highest in T7 - Mg + Zn + B plus propiconazole @ 0.1% (64.83  $\mu$ g g<sup>-1</sup>), T11 - propiconazole @ 0.1% (30.83  $\mu$ g g<sup>-1</sup>) and T11 - propiconazole @ 0.1% (65.00  $\mu$ g g<sup>-1</sup>) during the three months respectively.

With regard to chlorophyll content of banana leaf, all the treatments recorded superior values over absolute control (1.2  $\mu$ g g<sup>-1</sup>). The most favorable effect was for T5 - Mg + Zn + B plus tebuconazole @ 0.1% (2.14  $\mu$ g g<sup>-1</sup>) followed by T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> (2.12  $\mu$ g g<sup>-1</sup>).

The study revealed that there was a significant difference between the treatments with respect to magnesium, boron and zinc content in leaves.

Magnesium content, was maximum in T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup>which was on par with T5 - Mg + Zn + B plus tebuconazole @ 0.1% (0.34%). In case of zinc, the highest content (36.23 ppm) was recorded with T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> and the highest boron content (727.67 ppm) was obtained in case of T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup>. In all the cases, the lowest was found in absolute control.

The results of growth characteristics namely, plant height and girth revealed that, there was no significant variation among treatments. However, all the treatments were recorded higher value than control. The highest plant height (2.5m) and girth (54.67 cm) was recorded in case of T5 - Mg + Zn + B plus tebuconazole @ 0.1% and the lowest was recorded by absolute check (T13) in both the cases.

The other growth characters like number of leaves and number of functional leaves were also studied. With respect to number of leaves, the results indicated that there was an increase in treated plants. The overall performance of the treatments indicated that the treatments *viz.*, T1 (Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1%), T3 (Micronol @ 2 g l<sup>-1</sup> plus propiconazole @ 0.1%), T4 (Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup>), T9 (tebuconazole @ 0.1%) and T12 (PGPR mix II @ 20 g l<sup>-1</sup>) were found to be the best and recorded the maximum number of leaves during the maturity stages of crop.

With respect to the number of functional leaves, the concise analysis of the results indicated that the treatments *viz.*, T1 (Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1%) and T8 (Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup>) were found to be

the best and recorded the maximum number of functional leaves during the post treatment months *viz.*, July, November and December. The lowest number of functional leaves was noticed in absolute control in all the post treatment observations.

The comparison of the average time taken for bunch emergence from the respective treatments showed that there was no significant difference between any of the treatments. However, the shortest number of days taken for bunch emergence was observed in the combination treatment T5 - Mg + Zn + B plus tebuconazole @ 0.1% (218.33). While the average time for bunch maturity of each treatments were compared, it was found that shortest number of days or period for bunch maturity was observed in the combination treatment T5 - Mg + Zn + B plus tebuconazole @ 0.1% (297.55) which was statistically on par with all other treatments. The longest time period for bunch emergence (267) and bunch maturity (358.01) was observed in absolute control.

The results of the study on effect of treatments on yield expressed that, the maximum bunch weight (12.63 kg) was obtained from the combination treatment T5 - Mg + Zn + B plus tebuconazole @ 0.1% which was on par with T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup> (11.85 kg), T11 - propiconazole @ 0.1% (11.08 kg), T7 - Mg + Zn + B plus propiconazole @ 0.1 % (11.03 kg), T1- Micronol @ 2 g l<sup>-1</sup> plus tebuconazole @ 0.1% (10.98 kg), T12 - PGPR mix II @ 20 g l<sup>-1</sup> alone (10.78 kg), T4 - Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup> plus PGPR mix PGPR

The other yield parameters such as number of hands per bunch, number of fingers per bunch and number of suckers didn't differ significantly between treatments even though all the treatments were recorded higher values than the absolute control.

The maximum number of hands (5.67) was found in eight treatments *viz.*, T1 - Micronol @ 2 g l<sup>-1</sup> plus tebuconazole (0.1%), T3 - Micronol @ 2 g l<sup>-1</sup> plus propiconazole (0.1 %), T4- Micronol @ 2 g l<sup>-1</sup> plus PGPR mix II @ 20 g l<sup>-1</sup>, T5 - Mg + Zn + B plus tebuconazole (0.1%), T7 - Mg + Zn + B plus propiconazole @ 0.1%, T8 - Mg + Zn + B plus PGPR mix II @ 20 g l<sup>-1</sup>, T11 - propiconazole @ 0.1% and T12 - PGPR mix II @ 20 g l<sup>-1</sup>.

With respect to the number of fingers per bunch, the most favorable effect was found in, T5 - Mg + Zn + B plus tebuconazole (0.1%) (52.00) followed by T8 - Mg + Zn + B plus PGPR mix II @ 20 g  $l^{-1}$  (50.67).

Regarding with the number of suckers, the maximum was observed in two treatments *viz.*, T5 - Mg + Zn + B plus tebuconazole @ 0.1% (5.33) and T8 - Mg + Zn + B plus PGPR mix II @ 20 g  $1^{-1}$  (5.33).

The correlation studies on the influence of weather parameters on the intensity and incidence indicated that maximum relative humidity and rainfall showed a significant positive correlation with respect to disease intensity and disease incidence at 0.05% level of significance. At 0.05% level (0.404), the minimum temperature was negatively (-0.643) correlated with disease intensity as well as disease incidence (-0.527). Statistically, the influence of minimum relative humidity was -0.506 with respect to disease intensity and it was -0.554 with respect to disease incidence at 0.05% level of significance. Also, evaporation showed the inverse relation with disease intensity (-0.578) and with disease incidence (-0.500).

There was a positive correlation of maximum relative humidity (0.679) and rainfall (0.549) with disease intensity at 0.05% level (0.404) of significance. Also, the same parameters are positively correlated with disease incidence with values of 0.610 and 0.542 respectively. Therefore it can be intended that high relative humidity and rainfall contributes to increase in disease incidence and disease intensity.

The economic analysis also revealed that the best inorganic treatment is application of Mg + Zn + B plus tebuconazole (0.1%) which recorded the highest

yield, net income and benefit cost ratio which is followed by Mg + Zn + B plus PGPR (20 g l<sup>-1</sup>) under organic treatments.

For the integrated management of banana Sigatoka leaf spot disease (caused by *M. musicola* R.),the foliar application of Mg  $(2 \text{ g } \text{ I}^{-1}) + \text{Zn} (3 \text{ g } \text{ I}^{-1}) + \text{B} (2 \text{ g } \text{ I}^{-1})$  plus tebuconazole (0.1%) at fourth and fifth month after planting performed best with respect to disease suppression and enhanced yield in humid tropical southern zone of Kerala.

# REFERENCES

#### 7. REFERENCES

- Abdullah, M.Y., Hassan, N.M., Mahmood, Z. and Talib, Z. 1999. Trend in foliar nutrient concentrations and contents and its implication on leaf area index development and yield in banana cultivar 'berangan'. In: Zakaria, W., T.M.M. Mahmud, D., Siti Khalijah, M. F., Aini, N. and Marziah, M. (eds.), *Proc. First Natl Banana Seminar*, pp : 95-105.
- Agrios, N.G. 2005. Plant Pathology (5th Ed.). Elsevier, Amsterdam, 635p.
- Amir, H.G., Shamsuddin, Z.H., Halimi, M.S., Ramlan, M.F. and Marziah, M. 2001. Effects of *Azospirillum* inoculation on N<sub>2</sub> fixation and growth of oil palm plantlets at nursery stage. *J. Oil Palm Res.* 13 : 42-49.
- Anon. 1995. Musa news. Infomusa 4(2): 26-30.
- Anon. 1957. Control of banana leaf spot. Report on trials with oil and other fungicides in Jamaica. Fernhurf, Research Station, England, 19 : 55-57.
- Arnon, D.I. 1949. Copper enzymes in isolated chloroplasts. Polyphenol oxidase in *Beta vulgaris*. *Pl. Physiol*. 24 : 1-15.
- Atkinson, D. and McKinlay, R.G. 1997. Crop protection and its integration within sustainable farming systems. *Agr. Ecosyst. Environ.* 64 : 87-93.
- Bag, K. and Sinha, C. 1997. Proc. Indian Acad. Sci. (Chem. Sci.). 109p.
- Bal, J.S., Bajwa, G.S. and Singh, S.N. 1993. Effect of ethepon application at turning stage on ripening and quality of Umran ber. *Punjab Hort.* 33 : 84-87.
- Bashan, Y. 1998. Inoculants of plant growth promoting rhizobacteria for use in agriculture. *Biotechnol. Adv.*, 16 : 729-770.
- Bashan, Y. and Holguin, G. 1998. Proposal for the division of plant growth promoting rhizobacteria into two classifications: biocontrol- PGPB (plant growth promoting bacteria) and PGPB. *Soil Biol. Biochem.* 30 : 1225-1228.

- Bashan, Y., Monero, M. and Troyo, E. 2000. Growth promotion of seawaterirrigated oilseed halophyte *Salicornia bigelovii* inoculated with mangrove rhizosphere bacteria and halotolerant *Azospirillum* spp. *Biol. Fertil. Soils*, 32 : 265-272.
- Baider, A. and Cohen, Y. 2003. Synergistic interaction between BABA and mancozeb in controlling *Phytophthora infestans* in potato and tomato and *Pseudoperonospora cubensis* in cucumber. *Phytoparasitica*. 31(4): 399-409.
- Bhanavase, D.B., Jadhav, B.R., Kshirsagar, C.R. and Patil, P.L. 1994. Studies on chlorophyll, nodulation, N- fixation, soybean yield and their correlation as influenced by micronutrients. *Madras Agric. J.* 81 : 325-328.
- Bharathi, R., Vivekananthan, R., Harish, S., Ramanathan, A. and Samiyappan,
   R. 2004. Rhizobacteria –based bio-formulations for the management of
   fruit rot infection in chillies. *Crop prot.* 23 : 835-843.
- Biggs, A.R., El-Kholi, M.M. and El-Neshawy, S.M. 1994. Effect of calcium salts on growth, pectic enzyme activity, and colonization of peach twigs by *Leucostoma persoonii*. *Plant Dis.* 78: 886-890.
- Bingham, F.T. 1982. Boron. In: Methods of soil analysis Part (2<sup>nd</sup>Ed.) A.L. Page. WI: Am. Society of Agronomy. Madison, 438p.
- Biswas, J.C., Ladha, J.K. and Dazzo, F.B. 2000. Rhizobia inoculation improves nutrient uptake and growth of lowland rice. *Soil Sci. Soc. Am. J.* 64 : 1644-1650.
- Biswas, J.C., Ladha, J.K. and Dazzo, F.B. 2000b. Rhizobial inoculation influences seedling vigour and yield of rice. *Agron. J.* 92 : 880-886.
- Blakeman, J.P. and Fokkema, N.J., 1982. Potential for biological control of plant diseases on the phylloplane. *Ann. Rev. Phytopathol*, 20 : 167-192.

- Blevins, D. and Lukaszewski, K. 1994. Proposed physiologic functions of boron in plants pertinent to animal and human metabolism. *Environ. Health Perspectives*. 102 (7): 31-33.
- Blevins, D.G. and Lukaszewski, K.M. 1998. Boron in plant structure and function. *Annu. Rev. Plant Phys.* 49 : 481-500.
- Bolle-Jones, E.W. and Hilton, R.N. 1956. Zinc-Deficiency of *Hevea* brasiliensis as a predisposing factor to Oidium infection. Nat. (Lond.). 177: 619-620.
- Bouriquet, G. and Bataille, J. 1958. Two parasitic fungi on banana in Madagascar: 'Cercospora musae' and 'Alternaria musae' nov.spp. Fruits d'outremer.13 : 47-55.
- Borges, A., Trujillo, I., Gutierrez, F. and Angulo, D. 1983. Influencia de los desequilibrios nutritivos P-Zn y K-Mg del suelo, en alteración de los mecanismos de resistencia de la platanera (Cavendish banana) al Mal de Panamá. *Fruits*. 38 : 755-758.
- Bradford, M. M. 1976. A rapid and sensitive method for qualification of microgram quantities of protein utilizing the principle of protein dye binding. *Anal. biochem.* 72 : 248-254.
- Bray, H.G. and Thorpe, W.V. 1954. Analysis of phenolic compounds of interest in metabolism. *Methods of Biochem. Anal.* 4 : 27-52.
- Brennan, R.F. 1992. The role of manganese and nitrogen nutrition in the susceptibility of wheat plants to take-all in Western Australia. *Nutrient Cycling in Agroecosystems*. 31(1): 35-41.
- Brodrick, H.T. and Kunhe, F.A. 1971. Sigatoka in banana can be profitably controlled. *F. mg. S. Afr.* 47(7) : 22-23.
- Brown, P.H., Bellaloui, N., Wimmer, M.A., Bassil, E.S., Ruiz, J., Hu, H., Pfeffer, H., Dannel, F. and Römheld, V. 2002. Boron in plant biology. *Plant Biol.* 4 (2) : 205-223.
- Brun, J. 1958. Etude sur l'action des fongicides huileux dans la luttre contre la cercoporiose. Note prélinimaire. *Fruits D'outre Mer.* 13 : 3-14.

- Brun, J. 1963. La cercosporiose du bananier en Guinée. Etude de la phase ascosporée de Mycosphaerella musicola Leach. Thèse de doctoratès science. Orsay, Paris, France.
- Burt, J. A., Rutter, J. and Gonzalez, H. 1997. Short distance wind dispersal of the fungal pathogens causing Sigatoka diseases of banana and plaintain, *Pl. Pathol.* 40 : 451-458.
- Cakmak, I. 2000. Possible roles of zinc in protecting plant cells from damage by reactive oxygen species. *New Phytologist*. 146 : 185-205.
- Calpouzos, L., Carmen, M., Rivera, M., Colberg, C. and Theis, T. 1959. Studies on the action of oil in the control of *Mycosphaerella musicola* on banana leaves. *Phytopathol.* 49 (3) : 119-122.
- Calpouzos, L., Defel, N.E., Colberg, C. and Theis, T. 1961. Relation of petroleum oil composition to phytotoxicity and Sigatoka disease control in Banana leaves. *Phytopathol.* 51 (5): 317-321.
- Campbell, J.G.C. 1926. 'Banana disease.'Report by the government Mycologoist on a visit to Queensland to investigate. Council paper No. 32, Lesilative Council, Fiji.
- Carlier, J., De Waele, D. and Escalant, J.V. 2002. Global evaluation of *Musa* germplasm for resistance to Fusarium wilt, Mycosphaerella leaf spot diseases, and nematodes- depth evaluation. In: Vézina, A. and Picq, C. (eds), Technical Guidelines 6, INIBAP.
- Carlier, J., Zapater, M. F., Lapeyre, F., Jones, D. R. and Mourichon, X. 2000.
  Septoria leaf spot of banana: a newly discovered disease caused by *Mycosphaerella eumusae* (anamorph *Septoria eumusae*). *Phytopathol.* 90 : 884–890.
- Cerkauskas, R.F. and Sinclair, J.B. 1980. Use of paraquat to aid detection of fungi in soybean tissues. *Phytopathol.* 70 : 1036-1038.
- Chillet, M., Abadie, C., Hubert, O., Chilin-Charles, Y. and de Lapeyre de Bellaire, L. 2009. Sigatoka disease reduces the greenlife of bananas. *Crop Prot.* 28 : 41-45.

- Chillet, M., Hubert, O., Rives, M.J. and de Lapeyre de Bellaire, L. 2006. Effects of the physiological age of bananas on their susceptibility to wound anthracnose due to *Colletotrichum musae*. *Plant Dis.* 90 : 1181-1185.
- Churchill, A.C.L. 2011. *Mycosphaerella fijiensis*, the black leaf streak pathogen of banana: progress towards understanding pathogen biology and detection, disease development, and the challenges of control. *Mol. Plant Pathol.* 12 : 307-328.
- Cowling, W.A., Wood, P. McR. and Brown, A.G.P. 1984. Use of paraquat, diquat herbicide for the detection of *Phomopsis feptostromiJormis* infection in lupins. *Aust. plant pathol.* 13 : 45-46.
- Crous, P.W. 2002. Taxonomy and pathology of *Cylindrocladium (Calonectria)* and allied genera. APS Press, St. Paul, Minnesota, U.S.A.
- Crous, P.W., Groenewald, J.Z. and Hill, C.F. 2002. *Cylindrocladium pseudonaviculatum* sp. nov. from New Zealand, and new *Cylindrocladium* records from Vietnam. *Sydowia*.54 : 23-33.
- Daniel, M. and Purkayastha, R.P. 1995. *Handbook of Phytoalexin Metabolism and Action*. Marcel Dekker, New York. 615p.
- Daurob, S.H. and Snyder, G.H. 2007. Chemistry of plant nutrients in soil, In:
  Datnoff, L.E., Elmer, W.H., and Huber, D.M. (eds), *Mineral Nutrition* and Plant Disease. APS Press – The Am. Phytopathological. Society, St. Paul, Minnesota, U.S.A. de la température. Fruits. 28 : 7-8.
- De Langhe, E.R., Swennen, R., and Wilson, G.F. 1983. Aspects hormoaux rejectionage des bananiers plantains. *Fruits*, 38 : 318-325.
- De'fago, G., Haas, D., Berling, C.H., Burger, U., Keel, C., Voisard, C., Wirthner, P. and Wuthrich, B. 1990. Suppression of black root rot of tobacco and other root diseases by strains of *Pseudomonas fluorescens*: potential applications and mechanisms. In: Hornby, D. (eds), *Biol. control of soil-borne plant pathogens*. CAB International, Wallingford, pp. 93-108.

- Derrick, K.S. and Timmer, L.W. 2000. Citrus blight and other diseases of recalcitrant etiology. *Annu. Rev. Phytopathol.* 38 : 181-201.
- Dordas, C. and Brown, P.H. 2005. Boron deficiency affects cell viability, phenolic leakage and oxidative burst in rose cell cultures. *Plant and Soil.* 268 (1): 293-301.
- Dordas, C. And Simoglou, K. 2006. Effect of foliar applied boron, manganese and zinc on tan spot in winter durum wheat. *Crop Prot.* 25 : 657–663.
- Duffy, B.K. and Defago, G. 1997. Zinc improves biocontrol of fusarium crown and root rot of tomato by *Pseudomonas fluorescens* and represses the production of pathogen metabolites inhibitory to bacterial antibiotic biosynthesis. *Phytopathol.* 87(12) : 1250-1257.
- Duvert, P., Milling, R. and Quesada, R. G. 2002. Monitoring the sensitivity of *Mycosphaerella fijiensis*, to pyremathanil. In: Acrobat., Memorias. xv reunion. Realizada en cartogena de Indas, Colombia, 27 de October al -02 novembrer, 2002, Medellen (COL), pp.153-157.
- Ehara, K., Iiyoshiand, Y. and Nishida, T. 2000. Polyethylene degradation by manganese peroxidase in the absence of hydrogen peroxide. *J. of Wood Sci.* 46(2) : 180-183.
- Elangovan, R., Mohan, S., Arumugam, R. and Jeyarajan, R. 1990. A survey report on the incidence of major diseases of banana in Tamilnadu. *South Indian Hortic.* 38 : 339-340
- Esitken, A., Karliag, H., Ercisli, S., Turan.M., Sahin, F. 2003. The effect of spraying a growth promoting bacterium on the yield, growth and nutrient element composition of leaves of apricot (*Prunus armeniaca* L.cv. Hacihaliloglu). *Aust. J. Agric.Res.* 54 : 377-380.
- Eswaramurthy, S., Muthusamy, M., Muthusamy, S., Jayasekar, R.R. and Natarajan, S. 1988. *Effect of Bavistin, Aureofungin applicayion on panama wilt and sigatoka leaf spot of banana*. Hindusthan Antibiotic Bulletin, 30 (1&2) : 25-26.

- FAO [Food and Agriculture Organisation]. 2012. Annu. Report 2012. Rome, Italy,1p.
- Figueiredo, M.V.B., Seldin, L., Araujo, F.F., Mariano, R.L.R. 2010. Plant growth promoting rhizobacteria: fundamentals and applications. In: Maheshwari, D.K. (ed.) *Plant growth and health promoting bacteria*. Microbiology monographs 18, Springer, Berlin, pp.21-43.
- Filippi, M. C. and Prabhu, A. S. 1998. Relationship between panicle blast severity and mineral nutrient content of plant tissue in upland rice. J. *Plant Nutr.* 21(8) : 1577-1578.
- Foure, E. 1987. Proc.Varietal reactions of bananas and plantains to black leaf streak disease. In: Persley, G. J. and De Langhe, E. A. (eds), *Proc. Banana and Plantain Breeding Strategies. ACIAR* Proc. No. 21. *ACIAR*, Canberra, pp. 110-113.
- Fouré, E. 1994. Leaf spot diseases of banana and plantain caused by *Mycosphaerella musicola* and *M. fijiensis*. In: Jones, D.R. (ed.), The Improvement and Testing of *Musa*: A Global Partnership INIBAP, Montpellier, France, pp. 37-46.
- Fouré, É. and Ganry, J. 2008. A biological forecasting system to control Black Leaf Streak Disease of bananas and plantains, *Fruits*. 63(5): 311-317.
- Franzen, D.W., McMullen, M.V. and Mosset, D.S. 2008. Spring wheat and durum yield and disease responses to copper fertilization of mineral soils. *Agron. J.* 100 : 371-375.
- Freiberg, S.R. 1955. Effect of growth regulators on ripening, split peel, reducing sugars and diastatic activity of bananas. *Bot. Gaz.* 117 : 377-392.
- Fridovich, I. 1974. Adv. Enzymol. 41,35.
- Fritz, A. 1985. Micronutrient supply: Growing problem in the cultivation of high value crops in arid zones. *Acta Horticulturae*. 158 : 293-306.
- Ganry, J. 1973. Etude du développement du système foliaire des bananiers en function de la température. *Fruits*. 28 : 7-8.

- Ganry, J. et Meyer, J.P. 1975. Recherche d'une loi d'action de la température sur la croissance des Fruits des bananiers. *Fruits*, 30 (6).
- Ganry, J., Lapeyre de Bellaire, L. and Mourichon, X. 2008. A biological forecasting system to control Sigatoka leaf spot of bananas and plantains. *Fruits*, 63 : 381-387.
- Garcia de Salamone, I. E., Hynes, R.K. and Nelson, L.M. 2001. Cytokinin production by plant growth promoting rhizobacteria and selected mutants. *Can. J. of Microbiol.* 47(5): 404 411.
- Garcia-Hernandez, E. and Cassab Lopez, G.I. 2005. Structural cell wall proteins from five pollen species and their relationship with boron. *Brazilian J. of Plant Physiol.* 17(4): 375 378.
- Gasparotto, L., Pereira, J.C.R., Urben, A.F., Hanada, R.E. and Pereira, M.C.N.
  2005. *Heliconia psittacorum:* hospedeira de *Mycosphaerella fijiensis*, agente causal da sigatoka-negra da bananeira. *Fitopatologia Brasileira*.
  30: 423 425.
- Getter, C. D., Ballou, T. G. and Koons, C. B. 1985. Effects of dispersed oil on mangroves: synthesis of a seven year study. J. Mar Poll Bull. 16 : 318 -324.
- Glick, B.R, Cheng, Z., Czarny, J. and Duan, J. 2007. Promotion of plant growth by ACC deaminase- producing soil bacteria. *Euro. J. of Plant Biol.* 119 : 329 - 339.
- Glick, B.R, Patten, C.L., Holguin, G. and Penrose, D.M. 1999. Biochamical and genetic mechanisms used by plant growth promoting bacteria. Imperial College Press, London.
- Goodman, R.N., Kiraly, E. and Zaitlin, M. 1967. The biochemistry and physiology of infectious plant diseases. D. Van Nostrand, Princeton, NJ.

- Graham, W.A. and McDonald, G.K. 2001. Effects of zinc on photosynthesis and yield of wheat under heat stress. In: *Proc. of the 10th Australian Agron.Conference*.
- Graham, D.R. 1983. Effects of nutrients stress on susceptibility of plants to disease with particular reference to the trace elements. *Adv. Bot. Res.* 10 : 221-276.
- Graham, D.R. and Webb, M.J. 1991. Micronutrients and disease resistance and tolerance in plants. In: Mortvedt, J.J., Cox, F.R., Shuman L.M., and Welch, R.M. (eds.), *Micronutrients in Agriculture* (2<sup>nd</sup> Ed.). Soil Science Society of America Inc., Madison, Wisconsin, USA, pp. 329 370.
- Graham, M.Y. and Graham, T.L. 1991. Rapid accumulation of anionic peroxidases and phenolic polymers in soybean cotyledon tissues following treatment with *Phytophthora megasperma* f. sp. *Glycinea* wall glucan. *Plant Physiol*. 97 : 1445-1455.
- Graham, R.D. and Webb, M.J. 1991. Micronutrients and disease resistance and tolerance in plants, In: Mortvedt, J.J., Cox, F.R., Shuman, L.E., and Welch, R.M. (eds), *Micronutrients in Agriculture*. Soil Science Society of America, pp. 329-370.
- Grewal H.S., Graham R.D. and Rengel Z. 1996. Genotypic variation in zinc efficiency and resistance to crown rot disease (*Fusarium graminearum* Schw. Group 1) in wheat. *Plant Soil*. 186 : 219-226.
- Grewal, S.H. 2001. Zinc influences nodulation, disease severity, leaf drop and herbage yield of alfalfa cultivars. *Plant and Soil*. 234 (1) : 47-59.
- Hammer, P. E., Hill, D. S., Lam, S. T., van Pee, K. H. and Ligon, J. M. 1997. Four genes from *Pseudomonas fluorescens* that encode the biosynthesis of pyrrolnitrin. *Appl. Environ. Microbiol.* 63 : 2147-2154.
- Hammerschmidt, R. and Nicholson, R.L. 2000. A survey of plant defense responses to pathogens. In: Agrawal, A.A., Tuzun, S., Bent, E. (eds), *Induced plant defenses against pathogens and herbivores*. APS, Minneapolis, USA, 390p.

- Hanada, R.E., Gaspatotto, L. and Pereira, J.C.R. 2002. Esporula

  éco de Mycosphaerella fijiensis em diferentes meios de cultura. Fitopatologia Brasileira. 27(2): 170-173.
- Hannam, R.D. R.J. and Uren, N.C. 1988. Role of Manganese in diseases resistance in manganese. *Heliconia psittacorum*: hospedeira de *Mycosphaerella fijiensis*, agente causal da.
- Harish , S., Kavino, M., Kumar, N., Balasubramanian, P. and Samiyappan, R. 2009a. Induction of defense- related proteins by mixtures of plant growth promoting endophytic bacteria against Banana bunchy top virus. *Biol. Control* 51(1): 16-25.
- Harish, S., Kavino, M., Kumar, N., Saravanakumar, D., Sooriananthasunadaram, K. and Samiyappan, R., 2008a. Biohardening with plant growth promoting rhizosphere and endophytic bacteria induces systemic resistance against Banana bunchy top virus. *Appl. Soil Ecol.* 39 : 187-200.
- Harish, S., Saravanakumar, D., Radjacommare, R., Ebenezar, E. G. and Seetharaman, K. 2008b. Use of plant extracts and biocontrol agents for the management of brown spot disease in rice. *Biol. control.* 53 : 555-567.
- Harker C.L., Ellis T.H. and Coen E.S. 1990. Identification and genetic regulation of the Chalcone synthase multigene family in pea. *The Plant Cell.* 2(3): 185-194.
- Hayden, H. L., Carlier, J. and Aitken, E. A. B. 2003. Genetic structure of *Mycosphaerella fijiensis* populations from Australia, Papua New Guinea and the Pacific Islands. *Pl. Pathol.* 52 :703-712.
- Hayden, H. L., Carlier, J. and Aitken, E. A. B. 2005. The genetic structure of Australian populations of *Mycosphaerella musicola* suggests restricted gene flow at the continental scale. *Phytopathol.* 95 : 489-498.

- Heckman J.R., Clarke B.B. and Murphy J.A. 2003. Optimizing Manganese Fertilization for the Suppression of Take-All Patch Disease on Creeping Bentgrass. *Crop Sci.* 43 : 1395-1398.
- Hedge, J.E. and Hofrieter, B.T. 1962. In : Methods in carbohydrate chemistry, Whistler, R.L. and BeMiller, J.N. (eds), Academic Press, New York, Vol. 17.
- Henn, A. 2004. The plant doctor: plant disease and fertilization . Mississippi State University Extension Service.
- Huber D.M. and Graham R.D. 1999. The role of nutrition in crop resistance and tolerance to disease. In: Rengel Z. (ed.), *Mineral Nutrition of Crops Fundamental Mechanisms and Implications*. Food Product Press, New York, pp. 205-226.
- Huber, D.M. 1980. The role of mineral nutrition in defense. In: Horsfall, J.G. and Cowling, E.B. (eds.), Plant Disease: An Advanced Treatise. Academic Press, New York, USA. pp. 381-406.
- Huber, D.M. 1989. The role of nutrition in the take-all disease of wheat and other small grains, p. 46-74. In: Engelhard A.W. (ed.), Soilborne Plant Pathogens. Management of disease with macro and microelements. *Am. Phytopathol. Society*, St. Paul, Minnesota.
- Huber, D.M. and Wilhelm, N.S. 1988. The role of manganese in resistance to plant diseases. *Developments in Plant and Soil Sci.* 33 : 155-173.
- Hulme, A.C. 1970. *The Biochemistry of fruits and their products. Vol. I.* Academic press, London.
- Jacome, L. H., Schuh, W. and Stevenson, R. E. 1991. Effect of temperature and relative humidity on germination and germ tube development of *Mycosphaerella fijiensis var. difformis. Phytopathol.* 81 : 1480-1485.
- Jacome, L.H., and Schuh, W. 1992. Effects of leaf wetness duration and temperature on development of black Sigatoka disease on banana infected by *Mycosphaerella fijiensis* var. *difformis*. *Phytopathol.* 82 : 515-520.

- Jaizme- Vega, M.C., Rodriguez- Romero, A.S. and Guerra, M.S.P. 2004. Potential use of rhizobacteria from the *Bacillus* genus to stimulate the plant growth of micropropagated bananas. *Fruits*, 59 : 83-90.
- Janisiewics, W.J. and Bors, B.1995. Development of microbial community of bacterial and yeast antagonists to control wound invading postharvest pathogens of fruit. *Appl. Environ. Microbiol.* 61: 3261-3267.
- Jiménez, M., 2008. EFFECT OF THE NUTRITIONAL STATUS OF BANANA (MUSA SPP.) ON LEAF DISEASE INFESTATION BY MYCOSPHAERELLA FIJIENSIS MORELET IN ECUADOR. PhD. Thesis. Katholieke Universiteit Leuven. Ecuador.
- Johnson, G. and Schaal, L.A. 1957. Chlorogenic acid and other orthodihydroxy phenols in scab resistant Russet Burbank and scab susceptible Trirumph potato tubers of different maturities. *Phytopathol.* 47:253-255.
- Jones, D. R. 2000. Diseases of Banana, Abacá and Enset. CABI Publishing, New York.
- Jones, D.R.2003. The distribution and importance of the *Mycosphaerella* leaf spot diseases of banana. In: Jacome, L., Lepoivre, P., Marin, D., Ortiz, R., Romero, R. and Escalant, J.V. (eds), '*Mycosphaerella* leaf spot diseases of bananas: present status and outlook. *Proc. of the 2nd International Workshop on Mycosphaerella leaf spot diseases*, 20-23 May 2002, San Jos'e, Costa Rica, International Plant Genetic Resources Institute, Rome, 317p.
- Kapulnik, Y. 1991. Non- symbiotic nitrogen fixing microorganisms. In: Waisel, Y., Eshel, A. and Kafkafi, U. (eds), *Plant Roots; The Hidden Half*, Marcel Dekker Inc., New York, pp.703-716.
- Kavino, M., Harish, S., Kumar, N., Saravanakumar, D. and Samiappan, R. 2010. Effect of chitinolytic PGPR on growth, yield and physiological

attributes of banana (*Musa* spp.) under field conditions. *Appl. Soil Ecol.* 45 : 71-77.

- Kavino, M., Harish, S., Kumar, N., Saravanakumar, D. and Samiyappan, R. 2008. Induction of systemic resistance in banana (*Musa* spp.) against bunchy top virus (BBTV) by combining chitin with root- colonizing *Pseudomonas fluorescens* strain CHAO. *Eur. J. Plant Pathol.* 120 : 353-362.
- Kavino, M., Harish, S., Kumar, N., Saravanakumar, D., Damodaran, T., Soorianathasundaram, K. and Samiyappan, R. 2007. Rhizosphere and endophytic bacteria for induction of systemic resistance of banana plantlets against bunchy top virus. *Soil Biol. Biochem*.39 : 1087-1098.
- Keinath, P.A. and Loria, R. 1996. Management of common scab of potato with plant nutrients. In: Engelhard W.A. (ed.), *Management of Diseases with Macro- and Microelements*. APS, Minneapolis, USA, pp. 152-166.
- Klein, H. H. 1960. Control of *Cercospora* leaf spot of bananas with application of oil sprays based on the disease cycle. *Phytopathol.* 50 : 488-490.
- Klein, H.H. 1961. Effects of fungicides, oil and fungicide-oil-water emulsions on development of Cercospora leaf spot of bananas in the field. *Phytopathol.* 51: 294-297.
- Kloepper, J.W., Leong, J., Teintz, M. and Schroth, M.N. 1980. Enhanced plant growth by siderophores produced by plant growth promoting rhizobacteria. *Nat.* 286 : 885-886.
- Kloepper, J.W., Ryu, C.M. and Zhang, S. 2004. Induced systemic resistance and promotion of plant growth by *Bacillus* spp. *Phytopathol*. 94:1259– 1266.
- Knight, B.P., McGrath, S.P. and Chaudri, A.M. 1997. Biomass carbon measurements and substrate utilization patterns of microbial population from soils amended with cadmiun, copper and zinc. *Appl. Environ. Microbiol.* 63(1): 39-43.

- Kolattukudy, E.P., Kämper, J., Kämper, U., González-Candelas L. and Guo,
   W. 1994. Fungus-induced degradation and reinforcement of defensive barriers of plants. In: Petrini O., Guellete G.B. (eds.), *Host Wall Alterations by Parasitic Fungi*. APS, Minneapolis, USA, pp. 67-79.
- Kostandi, S.F., Soliman, M.F., and Ghaly, A.A. 1997. Smut disease and yield performance in corn (*Zea mays* L.) as influenced by nitrapyrin, urea and zinc applications in coarse-textured soils. *J. of Agronomy and Crop Sci.* 179(4) : 219-226.
- Kranz, J. 1966. Field tests to control banana Sigatoka disease (*M. musicola*) in Guinea. *Phytopathol. Z.* 53(4): 335-348.
- Krauss, A. 1999. Balanced nutrition and biotic stress. In: IFA . Agricultural Conference on Managing Plant Nutrition.
- Krishnamurthy, K. and Gnanamanickam, S.S. 1997. Biological control of sheath blight of rice. Induction of systemic resistance in rice plant associated *Pseudomonas spp. Curr. Sci.*, 72 : 331-334.
- Krishnamurthy, K. and Gnanamanickam, S.S. 1998. Biological control of rice blast by *Pseudomonas fluorescens* strain *Pf* 7-14: Evaluation of a marker gene and formulations. *Biol. Contr.* 13 : 158-165.
- Krishnamurty, K and Gnanamanickam, S.S. 1997. Biocontrol of sheath blight of rice: Induction of systemic resistance in rice plant associated with *Psuodomonas* spp. *Curr. Sci.* 72 : 331-334.
- Kunawat, R.N., Rathore, P.S. and Pareek, N. 2006. Response of mung bean to sulphur and iron nutrition grown on calcareous soil of Western Rajasthan. *Indian J. Pulse Res.* 19 : 228-230.
- Leach, R. 1941. Banana leaf spot *Mycosphaerella musicola*, the perfect stage of *Cercospora musae* Zimm. *Trop. Agric.* 18 : 91-95.
- Leach, R. 1946. Banana leaf spot (*M. musicola*) on the gros Michel variety in Jamaica. Investigations on the etiology of the disease and the principles of control by spraying, Bull., govt printer, Kingston.
- Leach, R. 1965. Banana leaf spot. Outlook on Agric. 3(5): 203-208.

- Lebeda, A., Luhova, L., Sedlarova, M. and Jancova, D. 2001. The role of enzymes in plant-fungal pathogens interactions. *Zeitschrift für Pflanzenkrankheiten und Pflanzenschutz*. 108(1): 89-111.
- Loesecke, V.H.W. 1950. Bananas. (2<sup>nd</sup> Revised Ed.) Interscience, New York, pp. 67-118.
- Marin, D.H. and Romero, R.A. 1992.El combate de la Sigatoka negra. Bulletin NO.4, Department de Investigations,Corporation Bananera Nacional Costa Rica.
- Marin, D.H., Romero, R.A., Guzman, M. and Suton, T.B .2003. Black Sigatoka: An increasing threat to banana cultivation. *Plant Dis.* 87(3) : 208-222.
- Marschner, H. 1995. Mineral nutrition of higher plants. Academic Press, London, UK.
- Martin, J. and Grossmann, F. 1972. Uber die Hemmung pektischer und Zellulolytischer Enzyme.
- Melin, P. and Tezenasdumonteel, H. 1975. Results of aerial application of different fungicides against Cercospora disease of banana. *Fruits d' outremer*, 29 (3) : 179-180.
- Melin, P.G., Plaud, Tezenasdumonteel, H. and Laville, E. 1976. Comparative action of imazalil on cercospora infection of banana in cameroon. *Fruits*, 20(5): 301-306.
- Mengel, K. 2002. Alternative or complementary role of foliar supply in mineral nutrition. *Acta Horticulturae*.
- Mengel, K. and Kirby, E. 2000. Principios de la nutrición vegetal. International Potash Institute.
- Meredith, D. S. 1970. Banana leaf spot disease (Sigatoka) caused by Mycosphaerella musicola Leach.Commonwealth Mycological Institute, Kew, Surrey, England.

- Mia, M. A. B., Shamsuddin, Z.H. and Mahmood, M. 2010. Use of plant growth promoting bacteria in bacteria in banana: a new insight for sustainable banana production. *Int. J. Agric. Biol.*, 12 : 459-467.
- Mia, M. A. B., Shamsuddin, Z.H., Zakaria, W. and Marziah, M. 1998. Root stimulation and nutrient uptake of banana inoculated with *Azospirillum brasilense* and grown under hydroponic condition. In: Zakaria, W., Mahmud, T.M.M., S.D. Khalijah, M.F. Nor Aini and M. Marziah (eds.), *Proc. First National Banana Seminar*, Genting, Pahang, Malaysia, pp. 122-133.
- Mia, M. A. B., Shamsuddin, Z.H., Zakaria, W. and Marziah, M. 2000. Growth and physiological attributes of hydroponically-grown bananas inoculated with plant growth promoting rhizobacteria. *Trans. Malaysian Soc. Plant Physiol.* 9 : 324-327.
- Mia, M. A. B., Shamsuddin, Z.H., Zakaria, W. and Marziah, M. 2005. Highyielding and quality banana production through plant growth promoting rhizobacterial inoculation. *Fruits*, 60 : 179-185.
- Mia, M. A. B., Shamsuddin, Z.H., Zakaria, W. and Marziah, M. 2007. Associative nitrogen fixation by *Azospirillum and Bacillus* spp. In bananas. *Infomusa*. 16 :11-15.
- Mitchell, J.W. and Marth, P.C. 1944. Effect of 2,4 dichlorophenoxyacetic acid on the ripening of detached fruits. *Bot. Gaz.* 106 : 199-207.
- Moffet, B.F., Nicholson, F.A., Uwakwe, N.C., Chambers, B.J., Harris J.A. and Hill T.C. 2003. Zinc contamination decreases the bacterial diversity of agricultural soil. FEMS *Microbiol. Ecol.* 43 (1) : 13-19.
- Moreno, D.A., Villora, G. and Romero, L. 2003. Variations in fruit micronutrient contents associated with fertilization of cucumber with macronutrients. *Scientia Horticulturae*. 97 (2) : 121-127.
- Mourichon, X., Carlier, J. and Fouré, E. 1997. Sigatoka leaf spot diseases. *Musa* disease. Fact sheet no. 8, Montpellier, France, INIBAP.

- Mourichon, X. and Fullerton, R. A. 1990: Grographical distribution of the species *Mycosphaerella musicola* Leach. (*Cercospora musae*) and *M. fijiensis* Morelet (*C.fijiensis*), respectively agents of Sigatoka Disease and Black Leaf Streak Disease in bananas and plantains. *Fruits*, 45 : 213-218.
- Mourichon, X. and Zapater, M. F. 1990. Obtention *in vitro* du stade *Mycosphaerella fijiensis*, forme parfaite de *Cercospora fijiensis*. *Fruits*, 45 : 553-557.
- Moussa, B. I. M., Dahdoh, M. S. A. and Shehata, H.M. 1996. Interaction effect of some micronutrients on yield, elemental composition and oil content of peanut. Communications in Soil Science and Plant Analysis. 27 : 1995–2004.
- Murphy, C.T., McCarroll, A., Bargmann, I., Fraser, A., Kamath, R., Ahringer, J., Kenyon, H.L. and Yon, C. 2003. Genes that act downstream of DAF-16 to influence the life span of Caenorhabditis elegans. *Nat.* 424 : 277-283.
- Murphy, J.F., Zehnder, G.W., Schuster, D.J., Sikora, E.J., Polston, J.E. and Kloepper, J.W. 2000. Plant growth promoting rhizobacterial mediated protection in tomato against Tomato mottle virus. *Plant Dis.* 84 : 779-784.
- Myers, D. and Campbell, R.N. 1985. Lime and the control of club root of crucifers: effects of pH, calcium, magnesium, and their interactions. *Phytopathol.* 75: 670-673.
- Nandakumar, R., Babu, S., Viswanathan, R., Raghuchander, T. and Samiyappan, R. 2001. induction of systemic resistance in rice sheath blight disease by plant growth promoting rhizobacteria. *Soil Biol. Biochem.* 33 : 603- 612.
- Northover, J. and Cerkauskas, R.F. 1994. Detection and significance of symptomless latent infection of *Monilinia fructicola* in plums. *Canadian* 1. *Plant Pathol.* 16 : 30-36.

- O,Connell, P.F. 1992. Sustainable agriculture- a valid alternative. *Outlook Agric*.21 : 5-12.
- Obeifuna, E. 1984. Effect of potassium application during floral initiation stage of plantains. *Ferti. News.* 5 : 315-319.
- Oborn, I., Edwards, A.C., Witter, E., Oenema, O., Ivarsson, K., Withers, P.J.A., Nilsson, S.I. and Richert Stinzing A. 2003. Element balances as a toll for sustainable nutrient management: a critical appraisal of their merits and limitations within an agronomic and environmental context. *Eur. J. Agron.* 20 : 211-225.
- Okon, Y. 1985. *Azospirillum* as a potential inoculant for agriculture. *Trends Biotechnol.*, 3 : 223-228.
- Okon, Y. and Labandera-Gonzales, C.A. 1994. Agronomic applications of *Azospirillum:* an evaluation of 20 years world wide field inoculation. Soil *Biol.Biochem.*, 26 : 1591-1601.
- Okon, Y., Kapulnik, Y. and Sarig, S. 1988. Field inoculation studies with Azospirillum in Israel. In: Suba Rao, N.S. (ed.), Biological Nitrogen Fixation, Recent Developments, Oxford and IBH, New Delhi, India, 175p.
- Ownley, B., Duffy B. and Weller, D. 2003. Identification and manipulation of soil properties to improve the biological control performance of phenazine-producing *Pseudomonas fluorescens*. *Appl. and Environ*. *Microbiol.* 69 (6) : 3333-3343.
- Palaniswami, A. 1978. STUDIES ON FRUIT ROT DISEASE OF MANGO AND BANANA CAUSED BY BOTRYODIPLOIDA THEOBROMAE. Ph.D Thesis, Tamil Nadu Agric. Univ. Coimbatore, 117p.
- Patel, A.B., Patel, B.I. and Katrodia, J.S. 1993. Extension of storage life of guava (*Psidium guajava* L.) fruits. *Indian Food Packer*. 14 : 5-7.
- Patel., P.R. 2009. Chemical control leaf spot (*Mycosphaerella musicola*) of Kerala. Int. J. Protec. 2 (1): 98-100.

- Periz, L., Mauri, F., Hernandez, A., Abreu, E. and Porras, A. 2000a. Epidemiologia de la Sigatoka negra (*Mycosphaerella fijiensis* Morelet) en Cuba. I. Pronostico bioclimatico de los tetramientos contra la enfermedad en platanos (*Musa* spp. AAA). *Revista Mexicana de fitopatologia*. 18(1): 27-36.
- Periz, L., Hernandez, A. and Porras, A. 2000b. Epidemiologia de la Sigatoka negra (*Mycosphaerella fijiensis* Morelet) en Cuba. II. Pronostico bioclimatico de los tetramientos en bananos (*Musa accuminata* AAA). *Revista Mexicana de fitopatologia*. 18(1) : 15-26.
- Pefoura, M. A., Blizoua-Bi, P., Kobenan,K. and Kone D. 1993. Survey on the special distribution of banana and plantain in Cote d' Ivoire. *Cahiers Agric.* 5 : 181-184.
- Person, C.D., Sambroski, J. and Forysth, F.R. 1957. The effect of BZI on detached wheat leaves. *Can. J. Bot.* 180 : 1294-1295.
- Pieterse, C.M.J., Van Wees, S.C.M, Hoffl., Van Pelt, J.A.E. and Van Loon, L.C. 1996. Systemic resistance in Arabidopsis induced by biocontrol bacteria is independent of salicylic acid accumulation and pathogenesisrelated gene expression. *Plant Cell*. 8 : 1225-1237.
- Pietrzykowski, E.A., Stone, C., Pinkard, E.A., and Mohammed, C.L. 2006. Effects of Mycosphaerella leaf disease on spectral reflectance properties of juvenile *Eucalyptus globulus* foliage. *Forest Pathol.* 36 : 1-15.
- Pont, W. 1958. Progress in banana leaf spot control in north Queensland. *Qld. agric. J.* 83 (6) : 317-326.
- Pont, W. 1963. Control of Banana leaf spot and speckle. *Qld. agric. J.* 88 (7) : 423-425.
- Porro, D., Comai, M., Dorigoni, A., Stefanini, M. and Ceschini, A. 2002. Manganese foliar application to prevent leaf drop. In: Tagliavini, M., Toselli, M., Bertschinger L., Brown, P., Neilsen, D., and Thalheimer,

M. (eds), *Foliar Nutrition and Perennial Fruits Plants*. International Symposium, 1-31, November, 2002. ISHS, *Acta Horticulturae*, 594p.

- Price, N. 1995. The origin and development of banana and plantain cultivation, In: Gowen, S. (eds), *Bananas and Plantains*. Chapman & Hall, London, UK, pp. 1-14.
- Prusky, D., Kobiler, I., Fishman, J.J., Sims, S., Midland, S.L. and Keen, N.T. 1991. Identification of an antifungal compound in unripe avocado fruits and its possible involvement in the quiescent infections of *Colletotrichum gloeosporioides*. *Phytopathol.* 132 : 319-327.
- Pscheidt, J.W. and Pearson, R.C, (1989b). Time of infection and control of *Phomopsis* fruit rot of grapes. *Plant Dis.* 73 : 829-833.
- Pscheidt, J.W. and Pearson, R.C. 1989a. Effect of grapevine training systems of pruning practices on occurrence of *Phomopsis* cane and leaf spot. *Plant Dis.* 73 : 825-828.
- Rabindran, R. and Vidhyasekaran, P. 1996. Development of a formulation of Pseudomonas fluorescens PfALR2 for management of rice sheath blight. *Crop Prot.* 15 :715-721.
- Radjacommare, R., Nandakumar, R., Kandan, A., Suresh, S., Bharathi, M., Raguchander, T. and Samiyappan, R. 2002. *Pseudomonas fluorescens* based bio-formulation for the management of sheath blight disease and leaf folder insect in rice. *Crop Prot.* 21 : 671-677.
- Rajendran, L., Samiyappan, R., Raguchander, T. and Saravanakumar, D. 2007.Endophytic bacteria mediate plant resistance against cotton bollworm.*J. Plant Interact.* 2 : 1-10.
- Ramsey, M. D., Daniells, J. W. and Anderson, V. J. 1990. Effects of sigatoka leaf spot (*Mycosphaerella musicola* Leach) on fruit yield, field ripening and greenlife of bananas in North Queensland. *Sci. Hortic.* 41 : 305-313.

- Ramsey, M. D., Daniells, J. W., Anderson, V. J. 1990. Effects of sigatoka leaf spot (*Mycosphaerella musicola* Leach) on fruit yield, field ripening and greenlife of bananas in North Queensland. *Sci. Horticul.* 41 : 305-313.
- Ramsey, M.D., Vandrey, L.L. and Schipke, L.G. 1987. Evaluation of systemic and protectant fungicides for the control of Sigatoka leaf spot (*Mycosphaerella musicola* Leach) of banana in North Queensland. *Aust. J. of Exp. Agric.* 27(6) : 919-923.
- Rangaswami, G and Kolandaiswamy.1962. Studies on relative susceptibility of banana varieties to three leaf spot diseases. *J. Annamalai Univ.* 23 : 127-134.
- Rea, G., Metoui, O., Infantino, A., Federico, R. and Angelini, R. 2002. Copper amine oxidase expression in defense responses to wounding and *Ascochyta rabiei* invasion. *Plant Physiol.* 128(3): 865-875.
- Rengel, Z., Graham, R.D. and Pedler, J.F. 1994. Time-course of biosynthesis of phenolics and lignin in roots of wheat genotypes differing in manganese efficiency and resistance to take-all fungus. *Ann. of Bot.*74 : 471-477.
- Rengel, Z., Gutteridge, R., Hirsch, P., and Hornby, D. 1996. Plant genotype, micronutrient fertilization and take-all infection influence bacterial populations in the rhizosphere of wheat. *Plant and Soil*. 183(2) : 269-277.
- Rengel, Z.1994. Effects of Al, rare earth elements, and other metals on net Ca<sup>2+</sup> uptake by *Amaranthus* protoplasts. *J. Plant Physiol.* 143 : 47-51.
- Restrepo, J. 1998. Abonos orgánicos fermentados: experiencias de agricultores de Centroamérica y Brasil. Corporación Educativa para el Desarrollo Costarricense. San José, Costa Rica.

- Restrepo, J. 2000. Agricultura Orgánica: una teoría y una practica. Abonos orgánicos fermentados experiencias de agricultores en Centroamérica y Brasil. Cali, Colombia.
- Reuveni, M., Agapov, V. and Reuveni, R. 1997a. A foliar spray of micronutrient solutions induces local and systemic protection against powdery mildew (*Sphaerotheca fuliginea*) in cucumber plants. *Eur. J. Plant Pathol.* 103 : 581–588.
- Reuveni, R. and Reuveni, M. 1997. Foliar-fertilizer therapy a concept in integrated pest management. *Crop Prot.* 17(2) : 111-118.
- Reuveni, R. and Reuveni, M. 1998. Foliar-fertilizer therapy a concept in integrated pest management. *Crop Prot.* 17 : 111-118.
- Rhodes, P.L. 1961. Oil spraying to control Banana leaf spot disease in Fiji. *Agric. J. Fiji*, 30 (1) : 29-32.
- Robinson, J.C. 1996. Bananas and Plantains. Crop Production Science in Horticulture 5, CAB International, Wallingford, UK, 238p.
- Rodrigues, F.A., McNally, D.J., Datnoff, L.E., Jones, J.B., Labbé, C., Benhamou, N., Menzies, J.G. and Bélanger, R.R. 2004. Silicon enhances the accumulation of diterpenoid phytoalexins in rice: a potential mechanism for blast resistance. *Phytopathol.* 94 (2) : 177-183.
- Rodriguez- Romero, A.S., Guerra, M.S.P. and Jaizme- Vega, M.C. 2005. Effect of arbuscular micorhizal fungi and rhizobacteria on banana growth and nutrition. *Agron. Sustain. Devel.*, 25 : 395-399.
- Rolshausen, P.E. and Gubler, W.D. 2005. Use of boron for the control of Eutypa dieback of grapevines. *Plant Dis.* 89 : 734-738.
- Romero, C.R. 1986. Impacto de Sigatoka negra y roya del cafeto en actividad platanera nacional. Revista de la Asociación Bananera Nacional (ASBANA), San José, Costa Rica 12 : 7-10.

- Romero, R.A. and Sutton, T.B. 1997. Reaction of four *Musa* genotypes at three temperature to isolates of *Mycosphaerella fijiensis* from different geographical regions. *Plant Dis.* 81(10) : 1139-1142.
- Romero, R.A. and Sutton, T.B. 1997. Sensitivity of *Mycosphaerella fijiensis*, causal agent of black sigatoka of banana, to propiconazole. *Phytopayhol.* 87(1) : 96-100.
- Römheld, V. and Marschner, H. 1991. Function of micronutrients in plants, In: Mortvedt J.J., Cox F.R., Shuman L.M., and Welch R.M. (eds), *Micronutrients in Agriculture*. Soil Science Society of America, Inc., Madison, WI, USA, pp. 297-328.
- Ruiz, J.M., Bretones, G., Baghour, M., Ragala, L., Belakbir, A., and Romero,
  L. 1998. Relationship between boron and phenolic metabolism in tobacco leaves. *Phytochem.* 48(2): 269-272.
- Sahlen. and Soemargono, A. 2011. Distribution and incidence of leaf diseases of banana in several banana producing centres in North Sumatra, West Sumatra, Bengkulu and West Java. *Agrivita*, 33 : 174-188.
- Sahu, S., Lider, R.S. and Singh, P.K. 2008. Effect of micronutrients and biofertilizers on growth, yield and nutrient uptake by chick pea (*Cicer aeritinum* L.) in vertisols of Madhyapradesh. Adv. Plant Sci. 21 : 501-503.
- Sandmann, G. and Boger, P. 1980. Copper-mediated lipid peroxidation processes in photosynthetic membranes'. *Plant Physiol*. 66 : 797-800.
- Saravanakumar, D. and Samiyappan, R. 2007. ACC deaminase from *Pseudomonas fluorescens* mediated resistance in groundnut (*Arachis hypogea*) plants. J. Appl. Microbiol. 102 (5): 1283-1292.
- Saravanakumar, D., Vijayakumar, C., Kumar, N. and Samiyappan, R. 2007. PGPR-induced defense responses in the tea plant against blister blight disease. *Crop Prot.* 26 : 556–565.

- Saravanakumar, D., Lavanya, N., Muthumeena, K., Raghuchander, T. and Samiyappan, R. 2009. Fluorescent pseudomonad mixtures mediate disease resistance in rice plants against sheath rot (*Sarocladium oryzae*) disease. *Biocontrol.* 54 (2) : 273-286.
- Sarig, S., Blum, A. and Okon, Y. 1988. Improvement of water status and yield of field- grown grain sorghum (Sorghum bicolor) by inoculation with Azospirillum brasilense. J. Agric. Sci., 110-271.
- Schutte, K.H. 1967. The Influence of boron and copper deficiency upon infection by *Erysiphe graminis* DC the powdery mildew in wheat var. Kenya. *Plant Soil.* 27 : 450–452.
- Selvarajan, S., Uma, S. and Sathiamoorthy, S. 2000. Etiology and survey of banana leaf spot diseases in India. In: Molina, A.B., Roa, V.N. and Maghuyop, M.A.G. (eds), *Advancing banana and Plantain R&D in Asia* and the Pacific. 10 : 94-115.
- Shamsuddin, Z.H., Amir, H.G., Mia, M.A.B., Premalatha, P., Halimi, M.S., Khor, S.K., Marziah, M., Arif, A.B. and Ooi, T.C. 2000. Commercial production of biofertilizer and bioenhancer using Azospirillum and Bacillus spp. for improved growth of oil palm seedlings and bananas. *Proc. Bio Thailand*. 129p.
- Shaul, O. 2002. Magnesium transport and function in plants: the tip of the iceberg. *Bio.Metals.* 15(3) : 307-321.
- Shepherd, K. 1990. Genetic improvement of bananas in Brazil: aspects related to resistance to the genus *Mycosphaerella*. In: Fullerton, R.A. and Stover, R.H. (eds), Sigatoka leaf spot diseases of bananas. Montpellier, France, INIBAP.
- Siddiqui, I.A., Shaukat, S.S., and Hamid, M. 2002. Role of zinc in Rhizobacteriamediated suppression of root infecting fungi and root-knot nematode. *J. of Phytopathol.* 150 (10) : 569-575.

- Siddiqui, Z.A. and Mahmood, I. 2001. Effect of rhizobacteria and root symbionts on the reproduction of *Meloidogyne javanica* and growth of chickpea. *Biores. Technol.*, 79 : 41-45.
- Siddiqui, Z.S. 1997. Effect of systemic fungicides on total protein, carbohydrate and phenolic contents of *Solanum melongena* and *Avena sativa*. *Appl. Ent. Phytopathol.* 64 : 17-22.
- Siddiqui, Z.S., Ahmed, S. and Gulzar, S. 1997. Effect of topsin-M (Methylthiophenate) and Bayleton (Triademifon) on seedling growth, biomass, nodulation and phenolic content of *Sesbania sesban*.
- Simmonds, J. H. 1933. 'Banana Leaf Spot' dep. Agric. and stock Div. of Entomology and Path., Qld, Pamphlet, no. 6, also in Qld Agric.J. 39 : 21-40.
- Simmonds, J. H. 1939. Influence of seasonal conditions on the development of Cercospora leaf spot of banana with special reference to the control programme. *Qld. Agric.J.* 52 : 633-647.
- Simmonds, N.W. and Shepherd, K. 1955. The taxonomy and origins of the cultivated bananas. *J. Linnean Soc. Lond. Bot.* 55 : 302-312.
- Simmonds, J.H. 1963. Studies in the latent phase of *Colletotrichum* species causing ripe rots of tropical fruits. *Qld J. Agric. Sci.* 20 : 373-424.
- Simmonds, N.W. 1966. Planting and management in. In: Banana (2<sup>nd</sup> Ed.) Longman Group Limited. Lond. and Newyork, pp.156-240.
- Singh, H.G. 1988. Sulphur management in fine textured calcareous soils. In: Proc.TSI- FAI Symp. Sulphur in Indian Agriculture, 1988. 1-9March, New Delhi, S111/2-8.
- Stahel, G. 1937. Banana leaf spot. 'Cercospora musae.' Trop. Agric. 14: 56-60.
- Stangoulis, J., Graham, R., Young, E.C., Peter, K., Ann, C., Collins, H., Zhang, Q. and Huynh, B. 2007. The mechanism of boron tolerance for maintenance of root growth in barley (*Hordeum vulgare* L.).

- Stevens, G., Motavalli, P., Scharf, P., Nathan, M. and Dunn, D. 2002. Crop nutrient deficiencies and toxicities. University of Missouri-Extension, Columbia.
- Stover, R.H. 1962. Intercontinental spread of banana leaf spot (Mycosphaerella musicola Leach). Trop. Agric. Trin. 29: 327-338.
- Stover, R. H. 1968. Leaf spot of banana caused by *Mycosphaerella musicola*: Perithecia and Sporodochia production in different climate. *Trop. Agric*. 45 : 1-12.
- Stover, R. H. 1972. 'Banana, Plantain and Abacá Diseases.' Commonwealth Mycological Institute (CAB): Kew, England.
- Stover, R. H. 1980. Sigatoka leaf spot of bananas and plantains. *Plant Dis.* 64 : 750-756.
- Stover, R. H. 1990. Sigatoka leaf spots: Thirty years of changing control strategies: 1959-1989. In: Fullerton, R. A. and Stover, R. H. (eds), *Sigatoka Leaf Spot Diseases of Banana*. INIBAP. Montpellier, France. pp.66-74.
- Stover, R. H. and Simmonds, N. W. 1987. Bananas. (3<sup>rd</sup> Ed.) Longman Scientific and Technical, Essex, England, UK. 468p.
- Stover, R.H. 1976. Distribution and cultural characteristics of the pathogen causing banana leaf spot. *Trop. Agric.* 53 : 111-114.
- Stover, R.H. 1980. Sigatoka leaf spots of banana and plantains. *Plant Dis.* 64 : 750-755.
- Stover, R.H. 1983. Effect of temperature on ascospore germ tube growth of Mycosphaerella musicola and Mycosphaerella fijiensis var. difformis. Fruits. 38(9) : 625-628.
- Stover, R.H. and Fulton, R.H. 1966. Leaf spot of Banana caused by Mycosphaerella musicola: Perthecia and Sporodochia production in different climate. *Trop. Agric. Trin.* 45 (1): 1-12.

- Streeter, T.C., Rengel, Z., Neate, S.M. and Graham, R.D. 2001. Zinc fertilisation increases tolerance to *Rhizoctonia solani* (AG 8) in *Medicago truncatula*. *Plant and Soil*, 228(2): 233-242.
- Suharban, M. 1977. Studies on the leaf spot and post-harvest diseases of banana and their control. M.Sc (Ag.) thesis. Kerala Agricultural University, Thrissur. 76p.
- Sumam George. 1994. STANDARDISATION OF PLANT PART AS AN INDEX OF POTASSIUM STATUS IN BANANA. MUSA (AAB GROUP) NENDRAN. Ph.D Thesis. Kerala Agricultural University. Kerala, India.
- Thammaiah, N. 2003. STUDIES ON THE EPIDEMIOLOGY AND MANAGEMENT OF SIGATOKA LEAF SPOT OF BANANA. Ph.D Thesis, Univ. Agric. Sci., Dharward, Karnataka, India.
- Thammaiah, N., Kanamadi, V.C. and Shirol, A. M. 2008. Management of sigatoka leaf spot disease (*Mycosphaerella musicola*) in banana at different locations in Belgaum district of Karnataka, India. *Int. J. of Agric. Sci.* 4 : 57-58.
- Thongbai, P., Graham, R.D., Neate, S.N. and Webb, M.J. 1993. Interaction between zinc nutritional status of cereals and *Rhizoctonia* root rot severity. *J. Plant and Soil.* 153(2) : 215-222.
- Tiwary, D.K., Hasan, M.A. and Chattopadhyay, P.K. 1998. Studies on the effect of inoculation with Azotobacter and Azospirillum on growth, yield and quality of banana. *Indian Agric.*, 42 : 235-240.
- Tollenaar, D. 1961. Effects of copper and oil in the control of Sigakota banana leaf spot. Netherlands. *J. agric. Sci.* 8 (4) : 253-260.
- Tucker, D.P., Lee, R.F., Timmer, L.W., Albrigo, L.G. and Brlansky, R.H. 1984. Experimental Transmission of Citrus Blight. *Plant Dis.* 68 : 979-980.
- Vakili, N.G. 1968. Response of Musa acuminata species and edible cultivars to infection by *Mycosphaerella musicola*. *Tropic*. *Agric*. 45 : 13-22.
- Vala, D.G., 1996. Fungal and bacterial diseases of banana. In: Singh, S.J. (ed.), advances in diseases of fruit crops in India. Kalyani Publishers, Ludhiana, pp. 37-46.

Verhoeff, K. 1974. Latent Infection by fungi. Ann. Rev. Phytopathol. 12: 99-109.

- Vidhyasekaran, P. 1997. Fungal pathogenesis in plants and crops. In: Dekker, M. (ed.), *Molecular Biology and Host Defense Mechanisms*, New York, USA, 568p.
- Vidhyasekaran, P. 2004. Concise Encyclopaedia of Plant Pathology. Food Products Press, The Haworth Reference Press, 619p.
- Viswanathan, R. 1999. INDUCTION OF SYSTEMIC RESISTANCE AGAINST RED ROT DISEASE IN SUGARCANE BY PLANT GROWTH PROMOTING RHIZOBACTERIA. Ph.D. Thesis. Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India, 175p.
- Viswanathan, R. and Samiyappan, R. 1999. Induction of systemic resistance by plant growth promoting rhizobacteria against red rot disease caused by *Colletotrichum falcatum went* in sugarcane. *Proc. Sugar Technol. Assoc. India*, 61 : 24-39.
- Viswanathan, R. and Samiyappan, R. 2001. Antifungal activity of chitinase produced by some fluorescent pseudomonads against *Colletotrichum falcatum* Went causing red rot disease in sugarcane. *Microbiol. Res.* 155 : 309-314.
- Vivekananthan, R., Ravi, M., Saravanakumar, d., Kumar, N., Prakasam, V. and Samiyappan, R., 2004. Microbially induced defense related proteins against post harvest anthracnose infection in mango. *Crop Prot.* 23 : 1061-1067.
- Vonprahil, H. 1967. Results of trials for controlling Sigatoka disease of Bananas with anthracol in Columbia. P. fl. Schutz. Nachr. Bayer. 20(2): 617-622.
- Wardlaw, C.W. 1938. 'Banana disease. Diseases of the banana in Haiti, with special reference to a condition described as "plant failure". *Trop. Agric.* 15 : 276-282.
- Wardlaw, C.W. 1939. Cercospora Leaf Spot Disease of bananas. Nat. 14(4):11.
- Wardlaw, C.W. 1941. 'Banana diseases 14. The cultivation and diseases of the banana in Central america. *Trop. Agric. Nature*, Lond.18 : 157-163.

- Wardlaw, C.W. 1961. Leaf spot (Sigatoka disease). In: Wardlaw CW. (ed.) Banana Diseases: Including Plantains and Abaca. Edingburgh, Longman, pp. 314-341.
- West, M. 1983. New systemic fungicide highely effective on spot and speckle. Banana Bulletin, 47 : 4-6.
- Whiley, A.W., Pegg, K.G., Saranah, J.B. and Langdon, P.W. 1991. Correction of zinc and boron deficiencies and control of phytophthora root rot of avocardo by trunk injection. *Aust. J. of Exp. Agric.* 31(4) : 575-578.
- Witting, H.P., Johnson, K.B. and Pscheidt, J.W. 1991. Potential of resident epiphytic fungi for biological control of brown rot blossom blight in stone fruits. *Phytopathol.* 81 : 11-53.
- Yruela, I. 2005. Copper in plants. Brazilian J. of Plant Physiol. 17(1): 145-146.
- Zapata, P., Rodriguez, G., Cuesta, T., Armijo, C., Abuchar, D. and Tabora, P. 1999. Feasibility of organic 'Gros Michel' banana production in small farming systems. In: Rosales, F., Tripon, S., and Cerna, J. (eds), Org. /environ. friendly banana production. Montpellier, France, INIBAP.
- Zimmermann, A. 1902. Uber einige tropischer. Kitturpftanjen beobuchsete pilze. *Cent. Bukt. Abt.* 8 : 219p.

# APPENDIX

# APPENDIX I

# Chemical properties of the soil of experimental site: CRS, Balaramapuram

Sl.No:	Parameter				
1	P <sup>H</sup>	05.40			
2	EC (µS)	50.81			
3	OC (%)	00.22			
4	$Av.P(kg ha^{-1})$	29.10			
5	Av.K ( kg ha <sup>-1</sup> )	123.2			
6	Ex.Ca (ppm)	215.00			
7	Ex.Mg (ppm)	30.00			
8	Av.S (ppm)	17.10			
9	Fe (ppm)	17.71			
10	Cu (ppm)	02.10			
11	Mn (ppm)	51.48			
12	Zn (ppm)	01.45			
13	B (ppm)	00.23			

#### **APPENDIX II**

#### Arnow's reagent

To prepare Arnow's reagent (100 ml), 10 g of sodium molybdate and 10 g of sodium nitrite was dissolved in 100 ml of distilled water.

#### **APPENDIX III**

# 0.1M Sodium acetate buffer (P<sup>H</sup> 4.7)

Stock solutions

A : 0.2 M solution of acetic acid (11.55 ml in 1000 ml)

B:0.2~M solution of sodium acetate (16.4 g of  $C_2H_3O_2$  or 27.2 g of  $C_2H_3O_2$  .  $3H_2O~in~1000~ml)$ 

#### Preparation of stock dye solution for estimation of protien

100 mg of Coomassie brilliant blue G – 250 was dissolved in 50 ml of 95% ethanol and 100 ml of concentrated orthophosphoric acid was added. The volume was made upto 200 ml with water and kept at 4°C. The working dye was prepared just before use by diluting the stock solution to five times with water.

# **APPENDIX IV**

# Weather data of experimental site: CRS, Balaramapuram

Standard	Temperature Relative		/e		Number	Rainy	Sunshine	
week			humidity(%)		Rainfall	of days	days	hours
	Max.	Min.	Ι	II	(mm)	having		
						rain		
1	30.6	23.4	95.4	72	17.5	2	1	8.8
2	30.0	22.6	96.4	74.6	24	1	1	8.5
3	30.1	20.8	96.0	75.1	0	0	0	9.4
4	30.5	21.3	96.1	73.6	0	0	0	9.4
5	30.4	20.8	94.3	75.4	0	0	0	9.3
6	31.2	22.9	93.3	74.3	2.5	2	1	9.2
7	32	23	92.4	75.7	11	3	2	9.3
8	31.4	21.8	89.9	74.9	0	0	0	9.3
9	32	21.4	91.3	67.4	0	0	0	9.5
10	32.1	24.3	94.7	80.6	7	3	3	9.3
11	32.3	23.9	93.4	81.3	34	1	1	9.3
12	32.3	23.7	91.4	75.4	0	0	0	9.8
13	32.6	25.3	92.6	76.3	31	1	1	9.9
14	32.9	26.0	92.7	77.0	0	0	0	9.9
15	32.8	25.6	89.9	71.4	1.5	1	0	9.7
16	33.2	25.1	84.8	76.0	0	0	0	10.2
17	33.3	25.0	87.0	72.7	20.3	2	2	9.6
18	32.7	25.8	90.6	81.7	3.6	2	1	9.2
19	32.0	26.1	90.7	80.9	11.5	3	2	9.1
20	32.4	25.7	90.6	76.4	5.2	1	1	10.0
21	32.1	24.2	91.7	84.6	7.2	5	4	9.0
22	30.1	22.3	95	87.7	33.2	4	4	8.3

23	29.2	22.8	93.6	83.3	15.0	6	3	8.7
24	29.1	23.2	95.1	89.3	20.2	7	7	7.0
25	28.3	22.5	95.4	86.1	23.6	6	6	7.6
26	29.9	23.3	90	82.3	8.6	4	3	9.3
27	29.3	23.4	93.9	85.1	6.7	5	4	9.0
28	28.5	23	93.7	79.6	10.1	6	4	8.4
29	28.3	23.5	94	87.9	10.1	5	5	8.1
30	29.4	21.9	92.3	88.0	11.6	5	4	9.0
31	29.0	21.6	93.1	87.0	23.2	4	4	8.4
32	28.8	23.9	96.7	82.7	3.9	2	1	9.4
33	28.6	23.7	93.3	78.4	1.6	2	0	9.41
34	29.8	24.0	92.7	78.7	1.5	2	0	9.9
35	30.2	24.4	86.6	80.1	2.4	1	0	9.3
36	28.8	23.7	97	86.7	20.1	7	7	7.9
37	28.7	23.4	98.6	84.0	6.2	6	3	8.21
38	28.8	24.3	96.3	85.4	7.3	5	3	8.37
39	30.2	24.0	93.7	85.1	2.3	1	0	10.2
40	30.5	22.6	94	74.1	6.7	2	2	9.74
41	30.6	23.3	91.4	75.4	5.7	4	2	9.52
42	30.7	23.7	92.1	79.9	16.5	4	3	8.5
43	30.7	23.0	95.0	70.9	18.1	3	3	9.1
44	30.7	23.6	93.9	80.1	10.3	6	4	7.8
45	30.9	23.7	97.0	76.9	1.6	2	0	8.8
46	30.3	23.4	97.7	78.3	138.8	3	3	7.8
47	30.6	23.7	97.3	78.1	58.1	4	2	8.0
48	30.8	23.0	97.3	75.9	49.8	3	2	8.5
49	30.9	22.8	98.6	69.9	1.4	1	0	7.8
50	30.3	22.6	96.7	69.6	26	1	1	8.4
51	31.2	21.7	97.7	72.0	47	1	1	9.2
52	31	20.2	96.6	59.1	0	0	0	9.2
1	1							

# **APPENDIX V**

Input cost				
Particulars	Quantity	Rate	Amount (Rs)	
Planting material	2500 numbers	Rs. 10/ sucker	25,000/-	
Urea	897.44 kg	g 10/ kg 8974		
Raj-phos	1375.56 kg	10/ kg	13755/-	
MOP	1498.40 kg	18/ kg	2647/-	
FYM	25tons	380/ ton	9,500/-	
Neemcake	2500 kg	10/kg	25,000/-	
PGPR mix II	48.08 kg 60		2884/-	
Roger	1000 ml 9		950/-	
takes 2500 stakes		4.80/stake	12000/-	
		Total	1,00,710/-	
Labour charges				
For cultural operations	648 numbers 588/ labour		3,81,024/-	
For treatment application	For treatment application 38 numbers		22,344/-	
For Harvesting	10 numbers	588/ labour	5,880/-	
		Total	4,09,248/-	

# Input cost and Labour charges

### **APPENDIX VI**

#### **Treatment cost**

Treatment cost (for two applications)	Amount (Rupees) ha <sup>-1</sup>
T1 - Micronol @ 2g/litre plus tebuconazole @ 0.1%	1890/-
T2- Micronol @ 2g/litre plus copper hydroxide @ 0.25%	2680/-
T3- Micronol @ 2g/litre plus propiconazole @ 0.1%	1490/-
T4- Micronol @ 2g/litre of water plus PGPR mix II @ 20g/litre	5849/-
T5- Mg + Zn +B plus tebuconazole @ $0.1\%$	2146/-
T6- Mg + Zn +B plus copper hydroxide @ $0.25\%$	2936/-
T7- Mg + Zn +B plus propiconazole @ 0.1%	1746/-
T8- Mg + Zn +B plus PGPR mix II @ 20g/litre	6105/-
T9- tebuconazole @ 0.1%	1810/-
T10- copper hydroxide @ 0.25%	2600/-
T11- propiconazole @ 0.1%	1410/-
T12- PGPR mix II @ 20g/litre	5769/-
T13- Control	0/-
Total	36,431/-

# Integrated Management of Sigatoka Leaf Spot Disease of Banana Caused by *Mycosphaerella musicola* R. Leach ex J.L. Mulder

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#### Abstract of the

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#### ABSTRACT

Banana (*Musa* spp.), being the fourth most important food after rice, wheat and maize products, it tolls up as a major fruit crop of India. Among the various diseases of this crop, Sigatoka leaf spot disease is a serious factor limiting the productivity of the same. It causes losses by reducing the functional leaf surface of plant, which results in small bananas that fail to ripen and may fall. Hence a field study entitled "Integrated management of Sigatoka leaf spot disease of banana caused by *Mycosphaerella musicola* R. Leach ex J.L. Mulder" was conducted at Coconut Research Station, Balaramapuram with an objective to study the effect of *Pseudomonas fluorescens*, the secondary nutrient magnesium, the micronutrients, zinc and boron and new generation fungicides on growth, incidence of Sigatoka leaf spot disease and yield of banana as intercrop in coconut gardens.

The parameters studied were grouped into four main categories namely disease factors, weather factors, biochemical or physiological responses and growth and yield characteristics.

Studies were conducted to assess the effect of treatments on disease intensity and incidence revealed that, there was a reduction in disease factors during all months with respect to treatments in contrast with the control plots. In both the case of disease incidence and disease intensity all the treatments were significantly superior over control.

With respect to the disease incidence studies, it is clear that fungicide alone or combination treatments of the same along with Micronol or Mg + Zn + B is highly effective to lower the disease incidence. Disease intensity studies indicated that, foliar spray of new generation systemic fungicides such as tebuconazole, propiconazole or the contact fungicide copper hydroxide gave better disease suppression of yellow Sigatoka disease.

The pathophysiological studies of chlorophyll content revealed that there was an increase in chlorophyll content in all the treatments. Mainly the

combination treatments of fungicides with Mg + Zn + B and Bio control agent (PGPR mix II). There was an increase in magnesium content, boron content and zinc content in all combination treatments when compared to the alone treatments and from the untreated plants. But in case of protein content analysis, there was a decrease in total protein content in treated plants, mainly in the plants treated with systemic fungicides. The activity of defense related phenol and OD - phenol study exhibited that, there was an increased phenol content in treated plats.

Effect of treatments on plant height and plant girth indicated that all treatments resulted in higher height and girth observed in control at harvest stage.

Study of the other growth parameters like number of leaves and number of functional leaves also resulted in the same conclusion that a 1.5 to 2 fold per cent increase in number of functional leaves at harvest stage where as 0.5 fold per cent increase in number of leaves in all the treated plants when compared to the absolute control.

The study of yield per plant revealed that, 0.5 to 1 fold per cent increase in bunch weight. The other yield parameter studies revealed that the total number of hands, fingers per bunch and number of suckers were more than in untreated plots whereas, the number of days took for bunch emergence and bunch maturity was comparatively lower in case of treatment with maximum bunch yield, T5, Mg + Zn + B plus tebuconazole @ 0.1%. The maximum number of days took for bunch emergence and bunch maturity was in absolute control. Hence the results of integrated management for banana Sigatoka disease with respect to disease management, yield attributing characteristics were effective in the field trial that conducted.

The economic analysis also revealed that the best inorganic treatment is application of Mg + Zn + B plus tebuconazole (0.1%) which recorded the highest yield, net income and benefit cost ratio which is followed by Mg + Zn + B plus PGPR (20 g  $l^{-1}$ ) under organic treatments.

For the integrated management of banana Sigatoka leaf spot disease (caused by *M. musicola* R. Leach ex J.L. Mulder) in humid tropical southern zone of Kerala, the best treatment was found to be; Mg  $(2 \text{ g } 1^{-1}) + \text{Zn} (3 \text{ g } 1^{-1}) + \text{B} (2 \text{ g } 1^{-1})$  plus tebuconazole (0.1%) with aspect of maximum disease suppression, disease intensity reduction and highest yield.

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