

SEPTORIA LEAF-SPOT DISEASE OF BANANA
***Mycosphaerella eumusae*: A NEW RECORD FOR SRI LANKA**

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ABSTRACT

Two leaf spot diseases of banana caused by *Mycosphaerella musicola* Leach, (Yellow Sigatoka) *Mycosphaerella fijiensis* Morelett, (Black Sigatoka), are already prevalent in Sri Lanka. In year 2000, a previously undescribed leaf spot disease of bananas was discovered. Its causal agent was identified as *Mycosphaerella eumusae* (anamorph *Septoria eumusae*), which resembles *M. musicola* and *M. fijiensis* in the teleomorph stage. It was named *S. eumusae* on its anamorph stage. This disease has been named Septoria leaf spot or Sigatoka like disease. This is the first record of Septoria leaf disease in Sri Lanka. The first symptoms of Septoria leaf spot are brown streaks, which distinguish it from Sigatoka disease. The latter produce yellow or light green specks initially, which later develop into diamond shaped or black streak-like lesions. In Septoria leaf spot, the mature lesions are more or less oval or irregular. The three fungi can be differentiated on the basis of the characteristics of their conidia, conidiophores and from the fruiting bodies. Conidia differ in the number of septa and size. Conidia of Septoria have 0- 5 septa and are shorter and broader in comparison with the conidia of the *Mycosphaerella* spp. have 8- 10 septa and are longer and thinner. Septoria produce pycnidia and perithicia as fruiting bodies while the *Mycosphaerella* spp. produce sporodochia and perithicia. At present Septoria leaf spot is a disease of minor importance affecting banana in Sri Lanka.

KEY WORDS: Banana, Leaf Spot Disease, *Mycosphaerella* spp, Septoria Disease, Sigatoka Disease.

INTRODUCTION

Banana (*Musa* spp.) is affected by leaf spot diseases caused by species of *Mycosphaerella*. Yellow Sigatoka (YS) caused by *M. musicola* Leach was first recorded from Java in 1902 (Zimmermann, 1902). Another leaf spot disease called black leaf streak (BLS) or black Sigatoka caused by *M. fijiensis* Morulet was recorded from the Sigatoka District, Fiji Island in 1963. Yellow sigatoka was recorded from Sri Lanka in 1919 (Stover, 1972).

In the Latin American region, BLS is regarded as the most economically important disease of banana. It is very similar to YS but is more virulent and affects a wide range of banana genotypes (Jones, 1999). It develops more rapidly than YS and causes more severe defoliation. Destruction of banana leaves results in premature ripening of fruit and reduced yields. Since it was first recorded in the South Pacific region, it has been detected in most of the major banana growing countries. This disease was identified in Sri Lanka in 1995 (Personal communication Dr. D.R. Jones, International Institute of Banana and Plantain France).

Another unrecorded leaf spot disease of banana caused by *M. eumusae* was recorded from Sri Lanka (Carlier *et al.*, 1999). This disease resembles *M. musicola* and *M. fijiensis* in the telemorph stage and named *S. eumusae* on its anamorph stage. This disease was named Septoria leaf spot or Sigatoka-like disease (Carlier *et al.*, 2000). A survey was initiated with collection of samples from different varieties of bananas and different regions to determine the distribution of Septoria leaf spot disease in the country, and to differentiate the three diseases and their causal agents.

MATERIALS AND METHODS

Sample collection

More than 100 samples were collected from different agroclimatic regions and 64 of them were studied for Sigatoka and the new Septoria diseases. Leaf tissues with advanced symptoms of leaf spots were cut into squares of 10 cm and dried on newspapers and kept for further use.

Clearing of tissue and microscopic observations

Lesions on diseased leaf specimens were cleared in 10% KOH over night and washed 10 times in water. Fungal fruiting bodies associated with the lesions were observed under stereomicroscope for morphological characters were studied. To observe conidia, cleared tissues were stained for one minute in 0.5% cotton blue: lactic acid glycerol (1:1) followed by washing in water. The morphological characters of fruiting bodies associated with these lesions were described and the fungi were identified.

Ascospore discharge and culturing

Diseased banana leaves with necrosis were dried at room temperature for 48 h, were taken and soaked in distilled water for 15 minutes. Sections of about one mm were taken and planted on the underside of the lids of petridishes containing water agar. Ascospores were discharged overnight on to the agar surface and single ascospore was transferred on to fresh PDA medium in the next morning. The cultures were incubated at 25⁰ C for about three months until an ash grey mycelium developed. These cultures were scraped out with a scalpel and secured on slides for microscopic observation. As a shortcut method, lesions were crushed on slides with drops of distilled water and ascospores were observed directly under the microscope. Microscopic photographs of the spores were obtained at the same time.

RESULTS AND DISCUSSION

Symptoms of the whole plant

Sigatoka and Septoria do not kill plants immediately, but crop losses increases gradually with the age of plants. The disease results in a reduction of

the quality and quantity of fruit (Stover, 1983). Fruits from infected plants ripen prematurely and do not fill properly.

Symptoms of leaf spots

Observations reported here were made in year 2000 on banana fields in Gannoruwa, and Wagolle and from many other locations around Central Province (Table 1). Most of the samples were from Cavendish (*Anamalu*, *Ambon*, *Bimkehel*) Mysore (*Embul*) and from Pome (*Pooalu*) variety.

Yellow Sigatoka (YS) cause by *M. musicola*

The earliest symptom of YS is a green narrow speck about 1 mm on the upper leaf surface. The speck develops into a streak, several mm long and 1 mm or less width running parallel to the veins. The streak elongates and expands laterally to become a diamond shape and turns rusty red (Figure 1). A water soaked halo forms around the lesion when the leaf is turgid. This tissue quickly turns brown and young spot is formed. The dark brown center of the spot later turns darker brown forming a well defined ring around the mature spot which remains distinct even after the leaf tissue has died.

Table 1. Incidences of Yellow Sigatoka, Black Sigatoka, and Septoria

Disase	Variety (genomic group)					Disease incidence (%)
	Mysore <i>Embul</i> AAB	Pome <i>Pooalu</i> AAB	<i>Gross michel</i> <i>Ambon</i> AAA	<i>Giant cavendish</i> <i>Anamalu</i> AAA	<i>Dwarf Cavendish</i> <i>Bimkehel</i> AAA	
YS	13	00	08	10	03	27%
BS	32	04	06	16	02	45%
SD	04	03	05	04	02	13%
Others	08	02	02	07	01	15%
Total	57	09	21	37	08	

Samples collected from mid country wet, intermediate zones, low country intermediate zone.
YS – Yellow Sigatoka, BS – Black Sigatoka, SD – Septoria Disease.

Black Sigatoka (BLS) causes by *M. fijiensis*

Symptoms of BLS (Figure 2) are first visible to the naked eye as faint minute yellow specks. These specks turn rusty brown in color and are often most abundant near margin of the left side of the leaf particularly towards the tip. The first streak stage starts with elongating the initial specks becoming slightly wider to form a characteristic narrow reddish brown streak parallel to the leaf venation. Then these streaks gradually turns dark brown or almost black. The streak is now clearly visible on the upper surface of the leaf. The streaks become longer when conditions are favorable. Then the lesions in this stage coalesce and cause leaf necrosis.

The first spot stage initiate by broadening the streak and becomes more or less diamond shape and develop a light brown water soaked boarder around the spot. This spot is very much similar to YS first spot stage. Then

gradually the dark brown or black central area of the spot becomes slightly depressed and the water soaked boarder becomes more pronounced due to darkening. At this stage a slight yellowing of the leaf tissue immediately surrounding the water soaked boarder may occur. However in YS the diamond shape lesion stage shows a dark yellow margin where it is absent in BLS. Individual spots, the center of the spot dries out becoming light gray and further depressed. A well-defined black boarder usually surrounds the spot. The gray spot with dark boarder remains visible after the leaf dried and died.

Septoria leaf spot or Sigatoka like disease cause by *M. eumusae*

Mature spots of Septoria leaf spot on a young leaf variety Embul (AAB) in Sri Lanka is given in Figure 3. The first obvious symptom is a brown streak, which expands, in to a large spot and darkens. The spot later develops in to more or less rounded or irregular shape lesions with dark brown boarder, as the center turns grey. When the infection density is high, lesions coalesce as an early stage of development and large areas of leaf tissue become necrotic. When the leaf dries and died still the grey spot remains visible as in BLS.

Fruit body description

Sporodochia, conidiophores, and conida differences: In some observation of lesions, a mass of tightly packed mycelium could be seen and it developed in the substomatal air chamber and the conidiophores grew out from it. This is the sporodochia, and the conidiophores extend through the stomatal pore to the outside. Sporodochia of YS can be seen through stereomicroscope especially under side of the leaf and densely spread on the upper side of the leaf. Conidiophores produced from the dark brown or black stromata, which aliened the sporodochia, are pale to very pale brown, paler towards the apex and were straight or slightly curved. Without conidial scars at the apex, conidia are borne terminally and singly on the conidiophores. Conidia are pale to very pale brown, smooth, straight or variously curved and 0- 8 septet (Figures 4,5, 6,7, 8 and 9).

In Black Sigatoka conidiophores first develop in initial brown specks or early brown streak stage on the lower surface of the leaf. They emerge singly or in small groups from stomata. Sporodochia or stromata are absent here. Conidiophores are pale to medium brown becoming slightly paler towards the tip and mere longer than in *M. musicola*. They are straight or bent, with basal swelling and 0- 5 septets. One or more scars are present near the tip of the conidiophores. Conidia are formed singly at the apex and later becoming lateral as conidiophores develops. (Up to 4 mature conidia may be attached to single conidiophore. Conidia are 1-10 septet). Conidia of *M. fijiensis* are on average longer than those of *M. musicala* and are distinguishable because of a thickened basal hylum. Conidiophores of *M.*

fjiencis posses conidial scars, which are absent from conidiophores of *M. musicola*. Conidia of BLS is much more larger than that of YS.

In Septoria leaf spot disease, which is caused by *S. eumusae* (anamorph stage) the conidiophores were non-septet and shorter than the other two. Conidia produced from the apex of the conidiophore and were hyaline, 0- 5 septet and much broader than the other two. In Septoria two distinct types of fruiting structures found. In anamorph stage it producers Pycnidia which were more prevalent in lesions on the upper surface. Pycnidia were immersed, flask shaped and ostiolated when young, but often acervular like when mature.

Ascospores and sexual fruiting bodies of *Mycosphaerella* spp. of bananas

The sexual stage (telomorph stage) of *S. eumusae* was identified as *Mycosphaerella eumusae* and was indistinguishable from that of *M. musicola* and *M. fjiensis*. In anamorph stage in all these three *Mycosphaerella* spp. produced Perithecia to produce ascospores The ascospores are two celled.

Varietal Susceptibility

In this study the varieties *Embul* (AAB-Mysore group), *Pooalu* (AAB-Pome group), *Anamalu*, *Ambon* and *Bimkehel* (AAA-Cavendish sub group) were checked for the incidents of Septoria disease as well as for the YS and BLS. All the varieties were susceptible to Septoria disease while some varieties for the other two diseases. Sometimes a mixture of above three diseases could be observed. According to the data gathered, in variety *Embul* BLS is the major problem when compared with the disease incidents of Septoria.

CONCLUSIONS

From this study it was confirmed the presence of Septoria disease among the varieties *Embul* (Mysore), *Pooalu* (Pome), *Anamalu* (Giant Cavendish), *Ambon* (Gros michel), *Bimkehel* (Dwarf Cavendish), in agroclimetic regions of Mid country wet, Mid country Intermediate, Low country wet and Low country Intermediate zones of Sri Lanka. According to the survey, it was found that the public was unaware of Septorial disease. Therefore, it is essential to educate them to distinguish the three related leaf spot diseases, using the macroscopic and microscopic characters of fruit bodies, conidiophores, and conidia of the three related *Mycosphaerella* spp. A generalized statement cannot be made on the disease distribution, as the disease samples were not represented the whole banana population in the country. Therefore, much research in all aspect of the biology of *M. eumusae* is needed in Sri Lanka, including Molecular biology, Host pathogen interaction, Population structure analysis, selection of resistant clones and rational chemical control.

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LEAF SPOT DISEASES OF BANANA



Figure 1



Figure 2



Figure 3



Figure 4



Figure 5



Figure 6



Figure 7

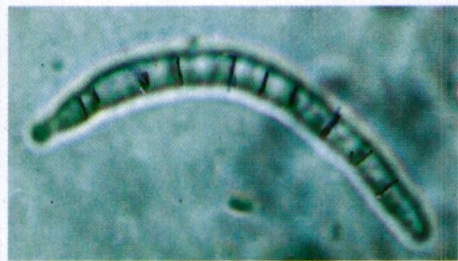


Figure 8



Figure 9

- Figure 1. Symptoms of Yellow Sigatoka
 Figure 2. Symptoms of Black Sigatoka
 Figure 3. Symptoms of Septoria
 Figure 4, 5, 6. Conidiospores of *M. musicola*,
M. fijiensis, *M. eumusae*
 Figure 7, 8, 9. Spores of *M. musicola* (YS),
M. fijiensis (BLS), *Septoria*
eumusae