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Phytophthora black pod disease in Ekiti state of Nigeria: Species differentation and pathogenic variation

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ABSTRACT

Incidence and pathogenic variation among *Phytopthora* species was studied in some selected cocoa plantations in Ekiti State of Nigeria. Infected cocoa pods were collected from five selected plantations each at Ilu-Omoba, Aisegba, Ikere, Ijero, Ise/Emure and Aramoko and brought to the laboratory. Identification of fungal isolates were based on colony morphology, slide culture and pathogenicity tests. Isolates from Ilu-Omoba, Aisegba, Ikere-Ekiti and Ise/Emure produced white cotton-wool like colonies on potato dextrose agar, with spherical sporangia and narrow pedicel. Two isolates were obtained from Ise/Emure. These show characteristics similar to those obtained from Ilu-Omoba, Aisegba, Ikere-Ekiti axis and Aramoko axis. Isolates from Aramoko-Ekiti and Ijero produced sparse aerial mycelium with stellate appearance, and broad sporangia pedicel. All the isolate however produced chlamydospores. Healthy cocoa pods inoculated with isolates showed the characteristic *Phytophthora* pod rot symptoms of water soaked lesions, which gradually increases in size and turned black. Pathogenic variations were observed with the two groups of isolates. Isolates from Ilu-Omoba, Aisegba, Ikere-Ekiti and Ise/Emure were more virulent than those from Aramoko and Ijero. The colony morphology, cultural characteristic and pathogenicity tests observed show that the two groups of isolates belong to *Phytophthora megakarya* and *P. palmivora. P. megakarya* was more prevalent in Ilu-Omoba, Aisegba, Ikere, and Ise/Emure while *P. palmivora* was limited to Aramoko and Ijero-Ekiti. The same fungus *P. megakarya* was also more virulent than *P.palmivora.*

Key words: Incidence, Pathogenic variations, *Phytophthora* species and Cocoa plantations.

INTRODUCTION

Black pod disease is one of the major diseases limiting the yield of cocoa in Nigeria. Different species of *Phytophthora* are known to cause this disease (Brasier and Griffin, 1979). Two species, *Phytophthora palmivora* and *P. megakarya* have been attributed to the black pod disease in Nigeria (Opeke and Gorenz, 1974). Though *P. palmivora* is reported worldwide as a causative agent of black pod disease of cocoa, however, *P. megakarya* is the major pathogen reported from Nigeria, Equatorial Guinea and Ghana (Brasier and Griffin, 1979).

Various reports have been published on the estimates of worldwide losses resulting from black pod disease (Padwick, 1956; Mederios, 1977; Ward *et. al.* 1981; de Figueiredo and Lellis, 1982). In Nigeria, the estimate ranges from 30-60%. of total cocoa produced (Ward *et. al.,* 1981). Though *P. megakarya* is more virulent than *P. palmivora,* however infection varies according to both location and climate (Luterboacher, 1993). Since the two species are present in Nigeria, species differentiation as well as climate and geographical location could play a considerable role in the epidemiology of black pod disease from different parts of the country.

Ekiti State is a medium cocoa growing area in Nigeria, however there are few reports in literature concerning the epidemiology of black pod disease of cocoa in the state. Arowolo and Fagbohun (2001) reported the occurrence and distribution of *Phytophthora canker* on cocoa trees as well as surveying the occurrence of *Phytophthora* spp in the soil of some cocoa plantations in Ekiti State.

Therefore, the present study reported here was carried out to identify the type of *Phytophthora* species associated with black pod/ pod rot disease of coca and their pathogenicity in cocoa plantations in Ekiti State.

MATERIAL AND METHODS

Naturally infected cocoa pods were obtained from five plantations each at Ilu-Omoba, Aramoko, Ikere, Aisegba, Ise/Emure and Ijero-Ekiti.They were kept in separate sterile polythene bags and brought to the laboratory.

Isolation of pathogen

The infected pods were sterilized using 70% ethanol. The lesion from each infected pod was cut out using sterilized knife. Pieces of the samples from each plantation were inoculated into plates of potato dextrose agar(PDA). The plates were incubated at 28ºC for five days. Pure cultures were obtained by sub-culturing the isolates on pre sterilized PDA medium and stored at 4^{0}C.

Identification of isolates

Slide culture

One square centimeter of PDA was cut from a plate and placed on a sterile glass slide using sterile needles. Each slide received different isolates. The slides were placed on glass rods in Petri dishes containing 20% glycerol in water until adequate growth occurred. (Zoberi 1967). The sporing surface of the pure culture of each isolate was teased out into a drop of alcohol and the preparation was observed under the microscope for fruiting bodies and mycelia branching. To study the colony morphology of each isolate, the method of Brasier and Griffin (1979) was used. After five days in total darkness, the isolate were observed against a dark background.

Pathogenicity test

Mature healthy green cocoa pods of different sizes and ages were harvested from each farm and labelled. Five cocoa pods were inoculated with seven-day old culture of each isolate and placed in moisture sterilized polythene bags. They were incubated at 28^{0}C for seven days. Observations were carried out daily on the inoculated pods for one week and infection rating was given a rating of between (+) and (+5).(+) being no infection and +5,a completely rotten pod)(Firman and Vernon, 1970).

RESULTS AND DISCUSSION

Isolates from Ilu-Omoba, Aisegba, and Ise/ Emure on PDA produced a white cotton-wool like aerial mycelium over the entire medium. The sporangia were spherical to fairly ovate, round base with narrow pedicel. Brasier and Griffin (1979) described isolates of *Phytophthora spp* with similar colony morphology and of sporangia types as the ones identified above as *P.megakarya.*

Isolates from Ikere produced colonies similar to that of Ilu-Omoba, Aisegba and Ise/Emure however, they produced sporangia with broader pedicel. This could be regarded as a variant of the Ilu-Omoba, Aisegba and Ise/Emure isolates. The isolates from Aramoko produced whitish colonies with stellate appearance and sharply defined edges. Aerial mycelium was sparse. The sporangia has longer pedicel. These characteristic were the features described for isolates of *P. palmivora* by Turner and Wharton (1960), and Brasier and Griffin (1979). Table 2 is a summary of pathogenicity tests carried out with the isolates. There were variation in the rate of infection from Ilu-Omoba, Ikere and Ise/ Emure on inoculated cocoa pods produced lesions with a more diffuse leading edge and considerable more fungal growth on the surface which extend with a few millimeters of the lesion edge. These characteristics when compared with Brasier's isolates confirmed the fungus to be of *P. megakarya*. *P.palmivora* does not usually produce the white fungal bloon at the earlier stage of infection as *P. megakarya* isolates (Okaisabor 1965 and Brasier *et. al.,* 1981). From the observation reported on pod infection, it is obvious that the isolates from Ilu-Omoba, Aisegba and Ikere are more virulent than Aramoko isolates. It has been reported that

Table 1: Distribution and identification of isolates from five towns In Ekiti state

Towns	1	2	3	4	5
Aramoko	a	a	a	a	A
Ijero	a	a	a	a	A
Aisegba	b	b	b	b	B
Ikere	b	b	b	b	B
Ilu-Omoba	b	b	b	b	B
Ise/Emure	ab	ab	ab	ab	Ab

a =Phytophthora palmivora
b = *P. megakarya*

P. megakarya is more virulent in infected pods than *P. palmivora(*Maddison and Griffin 1981*).*This further confirmed the identification of Ilu-Omoba, Aisegba and Ikere isolates as *P. megakarya* and the Aramoko and Ise/Emure as *P. palmivvora*.

In Nigeria, *P. palmivora* is the predominant species, while *P. palmivora* occured to a certain extent. In this study, both species are present in Ekiti-State, though P*.palmivora* is not as widely distributed as *P. megakarya.* Further more Ilu-Omoba, Aisegba, Ikere and Ise/Emure axis represents the zone where major cocoa production is carried out in the state and where *P. megakarya* is the predominant type.

Table 2: Result of pathogenicity test of various isolates from selected farms/towns in Ekiti state

Towns	1	2	3	4	5
Aramoko	+3a	+2a	+3a	+3a	+3a
Ijero	+3a	+3a	+3a	+3a	+3a
Aisegba	+5b	+3b	+5b	+4b	+4b
Ikere	+5b	+4b	+5b	+5b	+5b
Ilu-Omoba	+ 5b	+4b	+4b	+5b	+4b
Ise/Emure	+ 4ab	+ 4ab	+ 4ab	+ 4ab	+ 4ab

Disease Rating

+	No infection	+ +	Slight infection
+ + +	Moderate infection	+ + + +	Severe infection
+ + + + +	Very severe infection		
Isolates	a = *Phytophthora palmivora*	b = *P. megakarya*	

Though considerable difference in epidemiology of black pod disease is known to exist within *Phytophthora* species (Madisson and Griffin, 1981), however, infection varies according to climate and location. It is expected that the zone where *P.megakarya* predominates will be more heavily affected than those of *P.palmivora*. Though no concrete evidence has been presented on the epidemiology of black pod disease in state,but areas of highest annual rainfall are known to suffer the greatest losses from the black pod disease (Butler, 1981;Wood, 1974)

In the classification of Opeke and Gorenz (1974), the state falls under the category of medium loss area. In a study yet to be published, the two species sometime appear together in the same location in Ekiti.The variation observed in the rate of pathogenicity of the two groups of isolates in the state could play a major factor in the epidemiology of black pod disease in Ekiti State. Hence, there is need to carry out an epidemiological survey of the areas to determine the level and severity of the disease to facilitate the development of appropriate control measures.

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