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Mango Fruit Anthracnose and the Effects on Mango Yield and Market Values in Southwestern Nigeria

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ABSTRACT

This study investigated the etiology of mango fruit anthracnose, its effect on yield and market values in humid Southern Nigeria. The result of the investigation revealed that 96 out of 231 fungal isolates recovered from symptomatic mango parts were *Colletotrichum gloeosporioides* based on their whitish orange colony, hyaline; single-celled and cylindrical appearance and pathogenicity test. In addition, 60% of mango trees surveyed were infected with anthracnose and over 34% of fruits produced on those trees were found severely infected. Trees treated with fungicide during fruiting retained mean fruit of 57.65 (38.41% above mean in control) while untreated trees retained least mean fruits of 18.35 (55.94% lower than mean in control). A significant reduction in the price of mango was found associated with anthracnose-infected fruits. From the result of the investigation, it was evident that *Colletotrichum gloeosporioides* was responsible for anthracnose disease in mango and was prevalent in all the study areas. Furthermore, the result revealed that the disease was the cause of mango yield loss and of rendering marketable fruits worthless in Southwestern, Nigeria.

Key words: Mango anthracnose, *Colletotrichum gloeosporioides*, effect on yield, market values

INTRODUCTION

Mango (*Mangifera indica* L.) is among the fruit plants rich in vitamin C that is grown throughout the humid region of Southern Nigeria. The fruit though eaten as dessert in Nigeria rank first among other tropical fruits like citrus and pineapple fruits. It is an important fruit crop in most tropical regions of the world and most eaten in the developed countries (Diedhiou *et al*., 2007; Crane and Campbell, 1994). The dietary contribution of mango fruits in the diet of most people in the tropics rank above that of citrus fruits. It has been reported that the fruits can be processed into dry mango, mango pickle, mango jelly, or can be eaten cooked (Crane *et al*., 2006; Match Maker Associates, 2008) while the fat contained in the seed can be processed into flour or as a substitute to cocoa butter. When unripe, the fruit is tart, crispy and somewhat dry just like the green apples (Wijeratnam *et al*., 2008).

In Nigeria, mango production is limited by both man-caused factors (unmanned orchards) and natural factors (disease pathogens) despite the large expanse of land dedicated to its production. Fresh mango fruits from the country hardly make it to the international market due to anthracnose

infection. The unmanned orchards predispose the crop to severe anthracnose infection especially in the years when rains are heavy during the fruiting season. Fruits produced from trees in those orchards are commonly found associated with anthracnose-infected fruits that rot so quickly soon after harvest. Anthracnose has been reported in many parts of the world where the climate is suitable for mango production to be the most important field and postharvest disease of the crop (Sangeetha and Rawal, 2009; Chowdhury *et al*., 2008; Ploetz and Prakash, 1997). In Nigeria, not much had been done on anthracnose disease of mango in the area of research, whereas, the country had been in the forefront in the research on anthracnose disease of yam (Amusa, 2010). Okigbo and Osuinde (2003) only reported the role of *Colletotrichum gloeosporioides* in fungal leaf spots of mango in Nigeria while the first report of this disease on mango fruits in Southwestern Nigeria was only recently, reported by Onyeani *et al*. (2012). The disease has presently become a menace to both farmers and home gardeners in the Southwestern region of Nigeria making mango production no longer attractive in the area.

This present study was initiated to further investigate the etiology of mango fruit anthracnose, its effect on yield and market values in mango-growing areas of the humid forest region of Southwestern, Nigeria. The study identified the fungus responsible for fruit anthracnose in mango, its effect on yield as well as on market value of fresh mango fruits in Southwest, Nigeria.

MATERIALS AND METHODS

Survey of fruit anthracnose of mango in Ayetoro, Ibadan, Ogbomosho and Lagos in the humid forest zone of Southern Nigeria was carried out in 2008, 2009 and 2010.

Fungi pathogens associated with mango fruit anthracnose: Samples of symptomatic leaves, panicles and fruits of mango were, collected from mango orchards and home gardens in Ayetoro, Ibadan, Ogbomosho and Lagos and taken to the laboratory. At the laboratory, the infected mango parts were, cut into small pieces. For infected mango fruits, a portion of peeled epicarp and flesh were removed at the point of progression of the disease symptom; cut into small pieces and then soaked in 70% ethanol solution for 3 min, later, soaked into 1% Sodium hypochlorite (NaOCl) for another 3 min, then rinsed in two changes of sterile distill water. The parts were, dropped on sterile paper towels, allowed to dry before plating them onto Potato Dextrose Agar (PDA) and incubated for 5 days at room temperature. Isolated colonies were, sub-cultured into fresh plates until pure cultures were obtained. Pure cultures obtained were identified by visual examinations and viewing under stereo and compound electronic microscopes. They were then described and classified based on conidia and colony morphology as described by Dugan (2006) and Mordue (1971). Isolation was carried out in both the Nigeria Agricultural Quarantine Service and in International Institute of Tropical Agriculture, plant pathology laboratories.

Pathogenicity tests: Six healthy freshly harvested green matured mango fruits were surface sterilized by swabbing with 70% ethanol and 1% NaOCl solution. The fruits were inoculated with spore suspension of *Colletotrichum gloeosporioides* prepared following the procedures of Sivakumar *et al*. (1997). Inoculation of fruits were done following Sun *et al*. (2008) wound inoculation procedures. The fruits were then sealed in moist plastic bags and incubated for 5 days in a moist chamber after which observations on the development of anthracnose infection were made. Isolation and re-isolation of pathogens from fruits that showed symptom of anthracnose after 5 days of fruits incubation was carried out following Koch's postulates for proof of pathogenicity as described by Schumann and D'Arcy (2006).

Assessment of effect of anthracnose infection on the yield of mango: Two mango tree branches randomly selected at the upper canopy were sprayed with spore suspension of *Colletotrichum gloeosporioides* using hand sprayer. Another one was sprayed with sterile distilled water as a control following Okigbo and Osuinde (2003) method slightly modified. All branches were then covered with sterile polyethylene bags for 24 h. After 24 h, one branch out of the two inoculated with *Colletotrichum gloeosporioides* spore suspension was treated with Ferbam (Ferric dimethyldithiocarbamate) at 4.3 kg ha^{-1} applied as a dilute spray at 400 L ha^{-1} and thereafter maintained at fourteen days intervals until three weeks to harvesting while the other was left untreated. The experiments consisted of 5 trees with each tree representing a replicate. Fruits retained at harvest were counted and anthracnose infection was assessed as a product of the difference in number of fruits retained in treatment from control using the following formula:

$$\text{Infection effect (\%)} = \frac{\text{No. of fruits set in control branch-No. of fruits set in treatment branch}}{\text{No. of fruits set in control branch}} \times 100$$

Market survey: Market survey of anthracnose infected mango fruits were conducted in the year 2008, 2009 and 2010 during mango seasons, respectively. The prices of both anthracnose infected mango fruits and non-infected ones were obtained from 4 major markets located in Ojo in Ibadan, Ogbomosho fruit market, Ile-Epo in Lagos and Oja Oba in Ayetoro.

Statistical analysis: Data collected were, subjected to one-way Analysis of Variance (One-way ANOVA) using Statistical Package for Social Sciences (SPSS) 14.0 version. Post Hoc tests were, conducted using Duncan Multiple Range test to separate means of treatments into homogeneous subsets at 5% level of significance.

RESULTS

Symptoms of anthracnose disease: The first observable phenomenon on the field was unmanned mango orchards with most trees found infected with anthracnose and fruits produced on those trees severely infected (Fig. 1). On immature fruits, symptom was not noticeable but on green-matured fruits, the symptom observed was dark brown to black spots of about 0.1 mm but as the fruits ripen, the spots develops into very dark sunken lesions containing spores. On some fruits, black irregular lesions of varying sizes were found scattered all over the fruits' surface. Within few days, these irregular lesions in some cases coalesced to give large spots which eventually cause the rotting of the fruits. Also, on some fruits, the symptom was tear-stain black lesions that ran from the stem-end of the fruit to the basal end.

On leaves, observed symptoms were small dark brown spots that coalesce to form irregular lesions. The centers of old lesions dry up and fall out giving the leaf a perforated or tattered appearance. In most cases, symptoms were observed only on leaf edges while in some cases the mid-rib of the leaf was also affected.

On panicles, symptom begins as minute dark spots usually beginning from the attachment end of the panicle to the tree stem progressing upward to the tip resulting in shriveled, blighted flowers and small fruits. In some panicles, all blossoms were completely dried up resulting in no fruit setting.

Fig. 1(a-e): Anthracnose symptoms on (a) Single lesion on ripen fruit (b) Lesions all over fruit surface (c) Tear-stain pattern (d) Leaves and (e) Early infection on a panicle

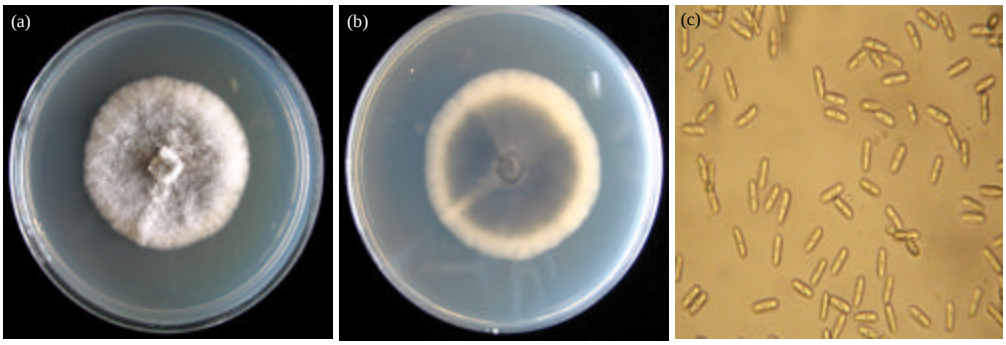

Fig. 2(a-c): *Colletotrichum gloeosporioides*, the causal agent of mango fruit anthracnose disease (a) Top view of colony in a petri dish (b) Reverse view and (c) Microscopic view

Isolation and identification of anthracnose pathogen: Inoculation of solidified potato dextrose agar with small cut pieces of lesions from the symptomatic mango parts and incubation at temperature that fluctuated between 28-30°C for 5 days produced mixed fungal growth, which was later sub-cultured to obtain pure cultures. Some of the pure cultures obtained, on potato dextrose agar, had colonies that were whitish to dark grey with thick to sparse lawns of aerial mycelium when viewed from the top of Petri dishes while the revised view appeared greenish to orange or dark brown centre bordered by creamy surrounding. When viewed under the microscope, conidia were hyaline; single celled and cylindrical with obtuse ends. The fungus was, identified to be *Colletotrichum gloeosporioides* (Fig. 2). Other fungi isolates recovered were *Alternaria alternata*,

Table 1: Fungi isolates recovered from anthracnose-infected mango fruits

Fungus recovered	No. of Isolates	Isolation (%)
Alternaria alternata	13	5.63^{e}
Alternaria tenuissima	2	0.87^{j}
Aspergillus flavus	11	4.76^{f}
Aspergillus fumigatus	4	1.73^{i}
Aspergillus nidulans	9	3.90^{g}
Aspergillus niger	25	10.82^{c}
Bipolaris hawaiiensis	13	5.63^{e}
Botryodiplodia theobromae	28	12.12^{b}
Cochliobolus nodulosus	1	0.43^{k}
Colletotrichum gloeosporioides	96	41.55^{a}
Curvularia lunata	8	3.46^{h}
Entyloma spp.	2	0.87^{j}
Fusarium dimerum	2	0.87^{j}
Fusarium longipes	16	6.93^{d}
Fusarium verticillioides	1	0.43^{k}

Means with same letter are not significantly different at 5% probability by Duncan's multiple range test (Data analysis 2010)

Table 2: Effect of anthracnose on mango yield in Southwestern Nigeria

Treatment	Mean fruit yield/treatment	Difference from control (%)
Untreated with fungicide	18.70^{c}	-55.94
Treated with fungicide	57.65^{a}	38.41
Control	41.65^{b}	-

Means with same letter are not significantly different at 5% probability by Duncan's multiple range test, results are mean of five replicates

Alternaria tenuissima, *Aspergillus flavus*, *Aspergillus fumigatus*, *Aspergillus nidulans*, *Aspergillus niger*, *Bipolaris hawaiiensis*, *Botryodiplodia theobromae*, *Cochliobolus nodulosus*, *Curvularia lunata*, *Entyloma* species, *Fusarium dimerum*, *Fusarium longipes*, *Fusarium verticillioides* (Table 1).

Pathogenicity tests: Pathogenicity tests carried out separately for the three highest occurring pathogens (*Colletotrichum gloeosporioides*, *Botryodiplodia theobromae* and *Aspergillus niger*) isolated from symptomatic mango fruits showed that only *Colletotrichum gloeosporioides* reproduced anthracnose disease symptom typical of those observed on healthy mango fruits. Following Koch's postulate, cultures re-isolated from the inoculated fruits were similar to those of the original isolates used for the inoculations.

Effect of anthracnose on the yield of mango in different mango-growing areas: There were significant differences in fruit yield among treated and untreated mango trees in the different mango-growing areas (Table 2). Trees treated with Ferbam (ferric dimethyldithiocarbamate) fortnightly from fruiting to maturity retained more fruits with mean fruit retention record of 57.65 fruits about 38.41% yield increase over control mean yield record of 41.65 fruits. Untreated trees retained least fruit mean of 18.35 fruits about 55.94% yield reduction when compared with control.

Table 3: The effect of anthracnose on the market prices of mango fruits in four major markets in Southwestern Nigeria

	Infected fruit/market prices (Naira/5 medium size fruits)			Non-infected fruits/market prices (Naira/5 medium size fruits)		
Market location	2008	2009	2010	2008	2009	2010
Ojo	10	15	20	50	100	100
Ogbomosho	5	10	15	50	75	100
Ile-epo	10	15	20	75	100	150
Oja-Oba	10	10	15	50	65	100

Infected mango fruits are sold in 'tens' while non-infected fruits are sold in 'fives' per Naira price (Price survey 2008, 2009 and 2010)

Effect of anthracnose on market value of mango fruits: Market survey carried out in four major markets in the study areas, revealed that anthracnose-infected fruits attracted low prices (Table 3). A significant reduction in the price of mango was found associated with anthracnose-infected fruits in all the major markets surveyed.

DISCUSSION

Based on the results of this study initiated to determine the etiology of mango fruit anthracnose, its effects on yield and fruits' acceptability, 231 isolates comprising of 15 different fungal species from nine fungi genera (*Alternaria*, *Aspergillus*, *Bipolaris*, *Botryodiplodia*, *Cochliobolus*, *Colletotrichum*, *Curvularia*, *Entyloma* and *Fusarium*) were recovered from diseased mango. Out of these 15 species, the three most frequently occurring species included *Colletotrichum gloeosporioides*, *Botryodiplodia theobromae* and *Aspergillus niger*. Several workers including Rawal (1998) and Sangchote (1991) implicated these three fungal species in earlier reports, to be the fungi responsible for postharvest diseases of mango associated with fruit rotting during ripening.

This result confirms the observation of Johnson *et al.* (1991), Sangchote (1997), Johnson (1992), Johnson and Sangchote (1994) and Lonsdale (1993) that soft brown rot is the most economically important postharvest decay of mango fruits in many countries where anthracnose is well controlled. Prabakar *et al.* (2005) reported that *Botryodiplodia theobromae* causes varying percentages of mango fruit losses ranging from 20-50%. In this study, field survey of fruit rot showed that soft-brown rot of mango was widely encountered. This result confirms Flores (2009) report on stem-end or soft-brown rot as a major problem limiting storage and shelf life of mango fruits.

Pathogenicity test conducted with the three most frequently recovered fungi pathogens from symptomatic mango parts indicated that only *Colletotrichum gloeosporioides* reproduced symptoms typical of the original anthracnose symptom observed on symptomatic mango parts. This result supports the reports of several workers implicating *Colletotrichum gloeosporioides* as the causal agent of anthracnose of mango (Than *et al.*, 2008; Wharton and Dieguez-Uribeondo, 2004; Sangeetha and Rawal, 2009; Jayasinghe and Fernando, 2009). On the other hand, *Botryodiplodia theobromae* and *Aspergillus niger* showed rotting symptoms typical of soft brown rot and black mould rot on wounded fruits, respectively. This observation agrees with Rawal (1998) and Sangchote (1991) who implicated the two fungi as causal agents of stem-end or soft brown rot and *Aspergillus* rot (black mould) of mango fruits, respectively.

The assessment of the effect of anthracnose on yield conducted indicated that trees treated with fungicides had higher mean fruit yield than untreated trees. The implication of these results is that, mango trees with anthracnose infection before fruiting are most likely to produce fewer fruits except treated with fungicides. Furthermore, the result was in concordance with the results of Diedhiou *et al.* (2007), Hunsberger (2008) and Sangeetha and Rawal (2009) that implicated anthracnose disease in the reduction and/or failure of fruit setting in mango.

Market survey also revealed that anthracnose-infected fruits attracted low prices in all the major markets surveyed in the study areas. Severe anthracnose-infected fruits were no longer attractive and acceptable in the local markets let alone international market.

CONCLUSIONS

Based on the findings of this study, mango grows abundantly in every part of Southwestern zone of Nigeria. However, the orchards are not regularly attended to; given room to orchards over-grown with weeds. In addition, mango fruiting in the zone usually coincides with rainy seasons and high humid conditions associated with high temperatures, which have been reported by other workers to favours infection and colonization of the crop by fungal pathogens and in turn predisposes mango production to serious fungal attack. Marketable fruits produced in those orchards are rendered worthless while its market value as a means of livelihood to the peasant's farmers is affected. Farmers therefore, have greater responsibilities in keeping the humidity level of the orchards as low as possible especially during the fruiting period. If orchards are free of weeds, there could be greater aeration within the orchards, which will in-turn, reduce temperature and increase sunlight penetration, which could reduce the relative humidity within the orchards.

There was a positive correlation between soft-brown rot and acceptable fruits suggesting that, apart from fruit anthracnose, other diseases affect the marketability of mango fruits. This further suggests that a holistic approach is required in controlling mango fungal diseases both on the field and in storage rather than targeting one disease pathogen.

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