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Occurrence of storage rot disease of kolanut across the kola growing belt of Nigeria

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ABSTRACT: Studies were carried out at the Cocoa Research Institute, Ibadan, Nigeria to determine the frequency of occurrence of storage rot disease caused principally by *Botrodiplodia theobromae* and *Fusarium pallidoroseum*on the two edible *Cola* species, *Cola nitida* and *Cola acuminata*. Several samples of healthy and diseased kolanuts were randomly collected from fourteen locations across the kola growing belt of Nigeria which is predominantly rain forest. Samples were surface dis-infected and placed on acidified potato dextrose agar at 25°C for 7 – 10 days.

Botryodiplodia theobromae, Fusarium pallidoroseum Aspergillus, Penicillium, Mucor and *Chlamydomyces* spp were the common fungal genera isolated from both *C. nitida* and *C. acuminata* nuts. The mean frequency of occurrence of *B. theobromae* and *F. pallidoroseum* were 0.47 and 0.34 respectively on *C. nitida* while it was 0.20 and 0.34 respectively on *C. acuminata*. Storage rot disease was found to be present in all the locations where samples of kolanut were collected. Similarly, the mean frequency of occurrence of *B. theobrome* and *F. pallidoroseum* were 0.69 and 0.53 respectively on kola pods when compared with the occurrence of other fungal genera. Whereas *Peacilomyces* and *Mucor* spp were isolated on *C. nitida* nut they were not isolated from *C. acuminata* nut. This present study confirmed the wide spread of the kolanut storage rot disease across the rain forest zone of Nigeria.

Key Words: Kolanut (*Cola* sp.); Storage rot disease; *Botrodiplodia theobromae; Fusarium pallidoroseumon*.

Introduction

Cola popularly called kolanut is a genus of the family Sterculiaceae in the order Malvales. It is indigenous to the tropical rain forest of West Africa, West Indies, brazil and Java (Rusell, 1955). *Cola nitida* and *Cola acuminata* are the two edible Cola species. However, *C. nitida* has secure an important economic crop and kola of commerce in both intra and international trade.

Nigeria is the world largest producer of kolanuts. Trade of kola started as early as the nineteenth century and has since increased tremendously. Outside, West Africa, cultivation and trade in kolanut has been established in countries like Cameroon, Mauritus, Sri-Lanka and Malaysia (Oladokun, 1982). The geographical and chronological spread in trade and use of kola has inevitably created a high demand for kola far in excess of its production. The nut is harvested once a year, and if they must remain in the market throughout the year, they must be stored and preserved in such a way to maintain its freshness and quality.

Storage rot disease caused by *Botryodiplodia theobromae* and *Fusarium pallidoroseum* is generally the most serious post-harvest problem of kolanut which farmers and kola traders seek to solve. These fungi

can also initiate latent infections on nuts in the field when harvest is delayed and then cause rot during storage (Agbeniyi and Fawole, 1998). Infected nuts decay rapidly and though there is little or no secondary infection, the surrounding nuts are covered with masses of fungal spores. This contamination of healthy nuts with spores from rotted nuts is termed spoilage and is often a greater economic problem than diseased nut in fresh market production areas. Kola traders often control spoilage by removing diseased nuts at intervals during the storage period (Agbeniyi and Fawole, 1998). Our preliminary investigation had shown that kolanut can easily succumb to rot during storage (Agbeniyi et al., 2000). The high moisture content of 54 – 60% at which nuts are stored, necessary predisposed them to rot. The present studies were therefore designed to determine the frequency of occurrence of *B. theobromae* and *F. pallidoroseum* throughout the kola belt of Nigeria and to compare their incidence with other fungi found on *C. nitida and* C. acuminata.

Materials and Methods

Random samples of healthy and infected kolanuts *Cola nitida* and *C. acuminata* were collected from Ibadan, Ife, Ogunmakin, Sagamu, Ishara, Sabogida-ora, Owena, Garage Olode, Osogbo, Okenne, Ayangba, Otukpo, Onitsha and Ikom. The areas represented the major kola growing belt of Nigeria (Fig. 1) which is predominantly rain forest. Kolanut samples were cut into several 4mm pieces and surface – sterilized by immersion in 1% sodium hypochlorite solution for 60s followed by two rinses in sterile distilled water and blotted dry with sterile whatman filter papers Grade 41 Ashless. They were then plated on potato dextrose agar (PDA) (DIFCO, Detroit, U.S.A.) amended with 70% lactid acid to eliminate bacteria in 9cm diameter petridishes.. The plates were incubated at 25°C and the incidence of suspected pathogens were recorded at seven-ten days depending on when growth could be observed. Similarly, the number of fungal colonies were sub-cultured to obtain pure cultures for the identification of the fungi. The frequency of occurrence for each fungal genus were expressed as described by Britton et al (1993).

The frequency of occurrence of fungal isolates were also determined on pod, testa and nuts of *C. nitida* obtained from kola orchards at CRIN Headquarters, Ibadan. The fungi were identified by microscopic examination of culture, by reference to mammals and literatures especially Banett and Hunter (1972), Samson and Pit (1989), Singh et al (1991), nelson et al (1983) and Gerlach and Niremberg (1982). Data were statistically analysed using SAS (NC, U.S.A.).

Results and Discussion

Several fungi were isolated from kolanuts obtained throughout the kola growing belt of Nigeria (Table 1). The occurrence of the storage rot fungi was found to be present across the kola growing belt of Nigeria (Fig. 1)

Some of these fungi could not be identified either because they did not sporulate or failed to grow on PDA. The frequency of occurrence of the most common fungal genera isolated on both *C. nitida* and *C. acuminata* is shown in Table 2.

Botryodiplodia, Aspergillus, Penicillum and *Mucor* were frequently isolated on *C. nitida* throughout the kola growing belt of Nigeria. *Paecilomyces, Chlamydomyces* and *Curvularia* spp were not encountered on *C. acuminata*. The frequency of occurrence of *Botryodiplodia* and *Fusarium* species were 0.34 and 0.44 respectively on *C. nitida* while it was 0.20 and 0.34 on *C. acuminata*.

The isolations of major storage rot fungi on both species of *Cola* suggest the susceptibility of kolanut to infection during the storage period. Thus appropriate control measures on either of the two species of *Cola* should be applicable to both for the control of storage rot disease.

The present study established the presence of storage rot disease across the kola growing belt of Nigeria which is predominantly rain forest (Fig. 1). Thus, while the storage rot disease may not be apparent at location of production, the occurrence of storage rot disease is possible at other distant locations. The incidence of several other post-harvest diseases on crops such as potatoes, carrots, apple and store-fruit similarly followed this trend (Punja and Gaye, 1993).

Table 1: Occurrence of the most common fungal species associated with kolanut across the kola growing belt of Nigeria.

Sample site	State	Fungi Isolated*											
		Bt	**Fm**	**Fp**	**Cu**	**An**	**Afu**	**Pn**	**Fe**	**Mue**	**Afl**	**Pv**	**Ch**
Ibadan	Oyo	+	+	+	+	+	-	+	-	+	-	+	+
Ife	Osun	-	+	-	+	+	+	-	-	-	-	-	-
Ogunmaki	Ogun	+	+	+	+	+	-	-	-	-	-	+	-
Sagamu	Ogun	+	+	+	+	+	+	+	+	+	+	+	+
Ishara	Ogun	+	+	+	+	+	+	+	+	+	+	+	+
Sabogida-Ora	Edo	-	-	+	-	+	+	-	-	-	-	-	-
Owena	Ondo	+	+	-	-	-	-	-	-	-	-	-	-
Garage Olode	Osun	+	-	+	+	+	-	+	-	-	+	-	-
Osogbo	Osun	+	+	+	+	+	+	+	-	-	-	-	-
Okenne	Kogi	-	+	-	-	-	+	+	-	-	+		-
Ayangba	Kogi	-	-	-	+	-	-	-	+	-	-	+	-
Otukpo	Benue	+	-	+	+	-	-	-	+	-	-	-	-
Onitsha	Anambra	+	-	-	+	-	-	-	-	-	-	-	-
Ikom	Cross River	+	+	-	-	+	-	-	-	+		+	-

*Bt – *Botrodiplodia theobromae*; Fm. – *Fusarium moniliforme*; Fp – *Fusarium pallidoroseum*; Cu – *Curvularia* spp.; An – *Aspergillus niger*; Afu – *Aspergillus fumigatus*; Pn – *Penicillum* spp.; Fe – *Fusarium cavispermum*; Muc – *Mucor* spp.; Afl – *Aspergillus flavus*; Pv – *Paecilomycesvariotii*; Ch – *Chlamydomyces* spp.
** + = Present; - = Absent.

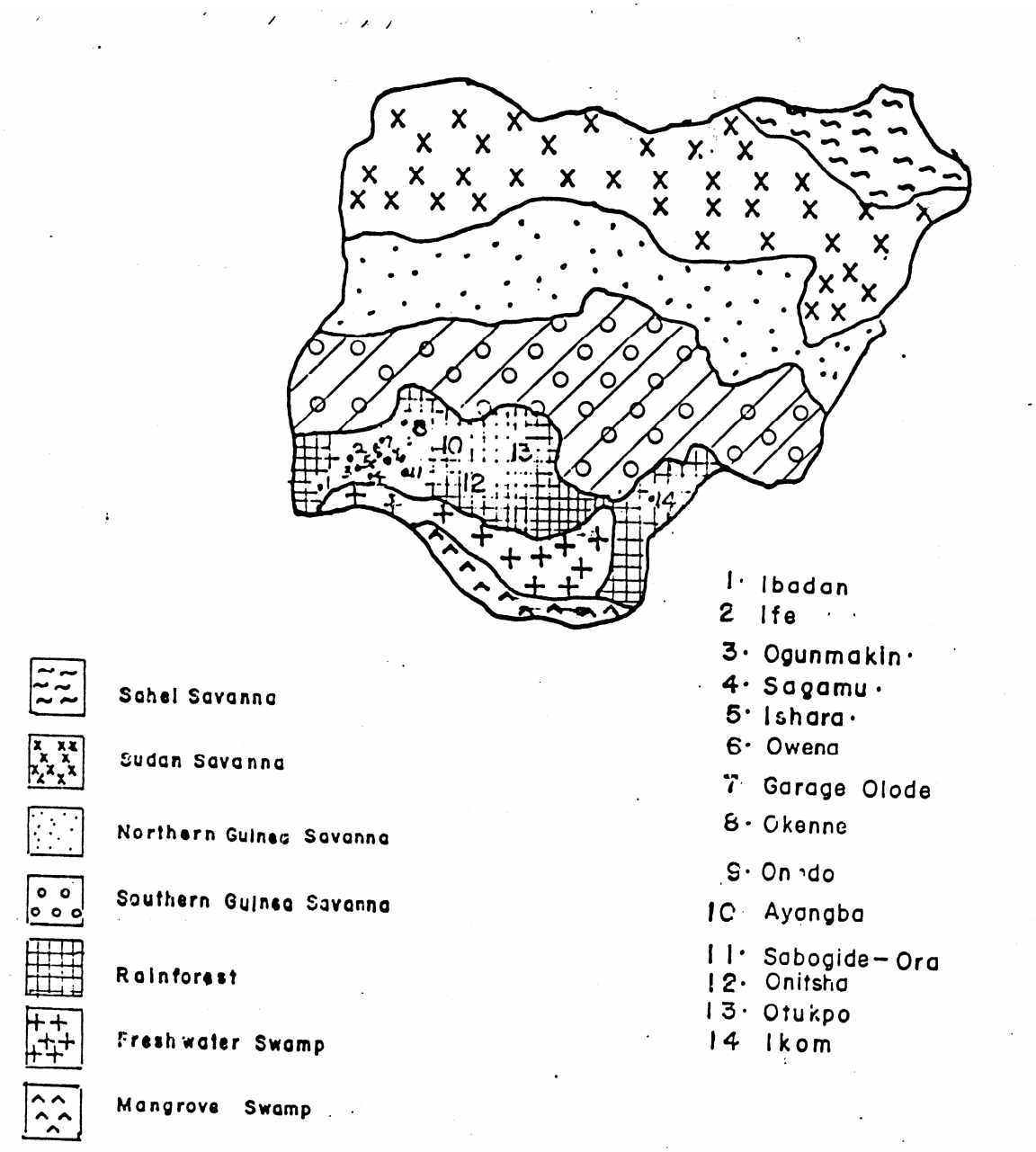

Fig. 1: Map of Nigeria showing sampling sites of Cola nitida and the agro-ecological zones.

The isolations of *B. theobromae* and *F. pallidoroseum* from kola pods and testa, suggest that the inocula of these fungi were transmitted from testa to the nuts through wounds that were created during skinning. In the absence of wounding, *B. theobromae* is incapable of penetrating the nut tissues (Olunloyo, 1979). It is probable that the pathogen could not easily degrade the intact cells of kolanut, or that they need to establish first on injured cells which provide exposed nutrients before they could colonise healthy tissue (Odebode, 1990).

Throughout the kola producing areas, many commercial production farms cropped with kola and other tree crops, mainly cocoa are naturally infested with inoculum of *B. theobromae* and *Fusarium* spp. The wet weather of the area (Fig. 1) is well – suited for kolanut production but also favours the growth and survival of *B. theobromae* and *Fusarium* spp.

A number of workers had found *B. theobromae* to be a common and a widespread fungus in the tropical areas where it is well-known as a wound parasite, for example, Chona (1933) cited the fungus as the cause of pseudostem rot and finger-tip rot of banana fruits in Punjab, India. It occurred as a seed-borne fungus in rubber and cotton (Noble et al, 1958). A partial list of the recorded hosts included mango (Charles, 1960), peanut (Wilson, 1947), coffee and castor bean (Rilley, 1960). The present study had shown the occurrence of *B. theobromae* and *F. pallidoroseum* on both *C. nitida* and *C. acuminata.* The high moisture content of 54-64% at which kolanut are stored necessarily predispose the nut to invasion by storage pathogen.

Throughout the kola growing belt of Nigeria traditional method of processing kolanuts were still being employed. Subsequently, additional inocula were usually encountered on kolanut during the routine processing stages (Agbeniyi et al., 2000). This observation partially explained why different fungi were encountered at each location across the kola growing belt. Thus appropriate control measures aim at minimizing invasion of kolanuts by storage rot fungi is recommended.

From the result presented in Table 3, there was no significant difference (P = 0.05) in the frequency of occurrence of *Penicillum, Paecilomyces* and *Mucor* spp. on both testa and nuts of *C. nitida.* However, while *Chlamydomyces* sp was not isolated on nut, it was isolated on pods and testa (Table 3). Similarly, *Paecilomyces variotii* was found on *C. nitida* whereas it was not found on *C. acuminata*. The results from this study clearly indicate that kolanuts were susceptible to fungal infection. *B. theobromae, Fusarium, Aspergillus* and *Penecillum* spp. were well distributed across the rain forest zone ofNigeria.

Table 2: The frequency of occurrence of the most common fungal genera on *Cola nitida* and *Colaacuminata.*

Genus	**Frequency of occurrence**	
	C. nitida	***C. acuminata***
Fusarium	0.47a	0.20b
Botryodiplodia	0.34b	0.34a
Penicillum	0.21c	0.07d
Aspergillus	0.23c	0.22b
Curvularia	0.05e	0.00d
Chlamydomyces	0.03ef	0.00d
Paecilomyces	0.02f	0.00d
Mucor	0.09d	0.03c

Data were normalised by log (x + 1).
Means not followed by the same letter in the same column are significantly different (P = 0.05) according to Duncan's multiple ranged test.

Table 3: The frequency of occurrence of the most common fungal genera obtainedfrom *C. nitida* collected at CRIN Ibadan Headquarters.

Genus	Pod	Testa	Nut
Botryodiplodia	0.69a	0.4a	0.5a
Fusarium	0.53b	0.3b	0.4bc
Aspergillus	0.32c	0.3b	0.3c
Penicillum	0.28d	0.2bc	0.1d
Curvularia	0.10e	0.2c	0.1d
Paecilomyces	0.00f	0.1cd	0.1d
Mucor	0.00f	0.1c	0.1d
Chlamydomyces	0.1e	0.01cd	0.0e

Data were normalised by log (x + 1).
Means not followed by the same letter in the same column are significantly different (P=0.05) according to Duncan's multiple ranged test.

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