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Assessment and identification of diseases on sugar cane accessions in the germplasm of national cereals research institute, Badeggi, Nigeria

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ABSTRACT

A study was carried out at the National Cereals Research Institute, Badeggi (lat. 9º045'N; long 6⁰07'E at an altitude of 70.57m above sea level) for the assessment and identification of diseases on some sugar cane accessions in the National germplasm maintained at the institute and research plots of scientists in 2013. The cane fields were surveyed using random field checking technique. Ten leaf samples with mixed infections were cut from the national germplasm, the CFC-WASDP multiplication field, the introductions from Brazil and experimental plots of researchers. The cut samples were taken to the microbiology unit of Central Services Laboratory of the institute for isolation and characterization. The samples were placed in running tap water to remove sand and debris. They were then soaked in distilled water with addition of sodium hypochlorite for 3-5 minutes and were rinsed 3 to 5 times with distilled water. With a sterilized surgical blade, samples were cut from the junction of lesions and healthy regions and plated on Potato dextrose agar (PDA) prepared in a ratio of 200:20:15 g/litre at a PH of 5.6±0.2. and incubated at 30-35ºC for 2-5 days. Germinating spores were sub cultured and brought to pure culture and then mounted in glass slides with lactophenol blue solution at a magnification range of ×10 - ×100. Four of the isolated pathogens were identified to species level based on the characteristic colonies produced on PDA and spore morphology in the case of whip smut which was not cultured as it does not grow on axenic media with reference to relevant texts. Pathogens were identified at different percentage infection rates from both local and exotic cane accessions. Results showed that one was of bacterial origin (*Xanthomonas axonopodis*), and four of fungal origin namely *Sporisorium scitamineum* Syd., *Colletotricum falcatum*, *Paraphaeosphaeria michotii* and *Puccinia melanocephala*.

KEYWORDS

Pathogens;
Infection;
Leaf lesions;
Sugarcane.

INTRODUCTION

The sustainable and consistent production of sugar cane by sugar estates and smallholder farmers and other chains of producers in Nigeria has in the recent past been adversely hampered by biotic constraints particularly diseases[19, 98].

Important sugar cane diseases in Nigeria include

Smut, red rot, leave blast, Sugarcane mosaic, ratoon stunting disease, sugarcane wilt disease among others[14, 20]. However, of all these diseases, smut is the most important on sugar cane in Nigeria owing to its destructiveness. Smut can be classified into four known forms but whip smut *Sporisorium scitamineum* Sydow [M. Piepenbr., M. Stoll & Oberw. 2002 (Syn: *Ustilago scitamiea* H. & P.Sydow)] is the most widespread and is one of the diseases that led to the discontinued cultivation of some high yielding sugar cane varieties in Nigeria[9].

Whip smut is a serious disease of sugarcane and reaches epidemic proportions where susceptible cultivars are grown[16]. Whip smut as the name implies is easily recognized by a 'whip-like' structure that consists of a central core of host tissue with a thin layer of black spores of fungal pathogen[21]. Smut also causes quantitative and qualitative loss to sugarcane growers worldwide[22].

Field surveys of cane fields across some sections of the country involving Sokoto, Kebbi, Kwara, Niger, Benue, Plateau, Kaduna and Katsina have revealed the menace posed by smut to cane farmers in these states[20]. Smut terminates the growth of tillers at 2-3 months of their life when infected setts are planted by turning them into 'smut whips'. Infected stalks quickly elongate and thin out before whips appear. Smut infected canes have low sucrose content; low yield and poor juice quality.

Red rot lowers sucrose content of infected milling canes, increases processing cost due to juice impurity from infected canes by the fungus. Red rot also greatly reduces germination of infected planting setts. Red rot affects both plant and ratoon crops showing that it is persistent in the soil even after removal of affected clumps have been done. In recent surveys of some states in the country, the disease was observed in Katsina, Benue, Plateau, Adamawa and FCT, Abuja[20]. The present effective management for the disease is the use of resistant varieties in places like India[4] and in Nigeria it is not an economical disease at present but that is not to say proactive management strategies cannot be developed for its curtailment.

The isolation of leaf blast from most of the sugar cane accessions and varieties in the present study is not surprising as the disease has been reported to occur in many areas including Niger state. Blast has been observed on cane at Ibadan, Badeggi and Numan and on cane farms in Niger, Kwara, Sokoto, Katsina, Borno and the Federal Capital Territory (FCT) Abuja in Nigeria[20].

On the contrary, sugar cane rust and gumming diseases though reported to occur in the country,[14] have not been observed at Badeggi close to three decades now. Their isolation from the leaves of sugar cane multiplication plots of canes recently introduced suggests that they were brought with the canes. This is because, the introduction of sugarcane varieties in collaboration with National Sugar Development Council (NSDC) and other international organizations, and in spite of the watchful eyes of plant Quarantine Services, pathogen introduction could and is always possible. Thus, the identification of new or indigent pathogens on sugar cane forms the basis for effective disease management methods.

However, at present only, palliative control measures such as chemical dips of planting setts and many other cultural control practices are employed to reduce disease effect[21]. Some integrated practices have shown to also help reduce disease effect in sugar cane[5]. Comstock, (2014) reported that hot water treatments for 52^{o}C for either 20, 30, or 45 minutes vary in the elimination of *S. scitamineum* and are 90, 95 or 100% effective respectively. Or red rot, use of the biological agent –*Trichoderma* sp- is currently in vougue[10, 7].

The present study consisted of a survey of sugar cane germplasm of some prospective local and exotic cane accessions and varieties in the field in order to access their natural adaptation or susceptibility to diseases. The identification of these diseases from the new introductions of sugar cane at Badeggi poses serious challenges and calls for effective management measures.

MATERIALS AND METHODS

The sugarcane research field of National Cereals Research Institute, (NCRI), Badeggi was surveyed using random field checking. Ten leaf samples with mixed infections were cut from the national germplasm, the CFC-WASDP multiplication field, the introductions from Brasil and experimental plots

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of researchers. The cut samples were taken to the Central Services Laboratory of the institute (microbiology) for isolation and characterization.

The samples were placed in running tap water to remove and wash away sand and debris. The leaf samples were then soaked in distilled water with the addition of sodium hypochlorite for 3-5 minutes. Samples were then rinsed for 3 to 5 times with distilled water. With a sterilized surgical blade, samples were cut from the lesion regions in small bits of 0.5cm at the junction of disease and healthy areas.

Alpha laboratory reagent PDA (potato dextrose agar) was prepared in a ratio of 200:20:15 g/litre at a PH of 5.6±0.2. Thirty nine grammes (39 g) of the PDA was then dispensed into 1000mls of distilled water and heated to dissolve after which it was autoclaved for 15 minutes, at 15 psi at 121°C. Leaf samples were then plated in the already poured plates and incubated at 30-35°C for 2-5 days.

Further sub culturing of the samples was done in PDA to obtain pure cultures of the growing strains. The strains were then mounted in glass slides with lactophenol blue solution (Diagonistica Merck D-61 Darmstadt) at a magnification range of ×10 ×100. The pathogens isolated were identified up to species level based on characteristic colonies produced on the PDA and with references from Watanabe (2002).

Smut, (*Sporisorium scitamineum Syd.*) which does not grow in exenic media was identified by collecting fresh whips and dislodging the teliospores aseptically and viewing under the microscope as described by Nasr (1977).

RESULTS AND DISCUSSION

Five pathogens, one of bacterial origin (*Xanthomonas axonopodis*), which causes gumming disease and four of fungal origin namely: red rot, (*Colletotricum falcatum*), leaf blast (*Paraphaeosphaeria michotii*) and rust (*Puccinia melanocephala*) and whip smut (*S. Scitamineum*) were identified.

Results presented in TABLE 1 shows that whip smut was isolated from 9 out of the ten randomly surveyed cane varieties in the West African Sugar cane Development Project (WASDP), giving a 90% field incidence of the disease. The other pathogens recorded incidences of 80, 70, 60 and 60% respectively for *C. falcatum, P. Michotii, P.melanoceophala* and *X. axonopodis* respectively.

The results from lesions and whips collected from the sugar cane collections from Brazil shown in TABLE 2 indicate a decreased incidence of the five pathogens associated with sugar cane at Badeggi. Whip smut recorded 80% incidence, red rot 70%, leaf blast 60%, while rust and gumming diseases recorded 50% incidences each.

TABLE 3 shows the disease incidence of the five pathogens on isolated and identified from the leaves of some tissue culture canes using Agar plate method and smut whips collection. Whip smut had the high-

TABLE 1 : Pathogenic organisms isolated and identified from the leaves of some selected sugarcane varieties at Badeggi, Nigeria

s/no		Pathogens				
	CFC varieties	S.scitamineum	C.falcatum	P. michotii	P. melanocephala	X. axonopodis
1	B80689	+	-	+	-	-
2	B85266	+	+	-	+	+
3	D8687	+	+	+	+	+
4	BT87164	+	-	-	+	+
5	Co91017	+	+	-	+	+
6	Co94012	+	+	+	-	+
7	KnH80412	+	+	+	-	-
8	Kn93063	+	+	+	+	-
9	MI672/90	+	+	+	-	-
10	M2238/89	-	+	+	+	+
Percentage		90	80	70	60	60

+ = Present; - = Absent

TABLE 2 : Pathogenic organisms isolated and identified from the leaves of some selected sugarcane varieties at Badeggi, Nigeria

s/no		Pathogens				
	Gernplasm varieties	S.scitamineum	C.falcatum	P. michotii	P. melanocephala	X. axonopodis
1	KNB	+	+	-	+	-
2	DB75159	+	-	+	-	+
3	BR0009	+	-	+	-	+
4	BR971007	+	+	-	+	-
5	BBZ951034	-	+	-	+	-
6	B881104	+	-	-	+	-
7	KNB9218	-	+	+	-	+
8	B00270	+	+	+	-	+
9	B881602	+	+	+	-	+
10	BBZ921101	+	+	+	+	-
	Percentage	80	70	60	50	50

+ = Present; - = Absent

est incidence of 60%, while red rot, leaf blast and leaf rust recorded 40% incidence on the tissue culture canes got from Brazil and gumming disease recorded only 20% incidence on the varieties.

The aim of the study was to assess and identify the diseases in some selected prospective sugar cane accessions in the germplasm at NCRI, Badeggi Niger state Nigeria, having spotted different lesions on sugar cane and as the crop is known to be vulnerable to a number of diseases[14]. Sugarcane diseases are more prominent with tropical weather conditions than subtropical though all regions have their peculiar disease infection history.

Singh (2006), reported that diseases caused by fungi, bacteria, viruses and mycoplasmas-like organisms cause considerable damage to sugar cane. Of all the diseases assessed, five pathogens, one of bacterial origin (*Xanthomonas axonopodis*), which causes gumming disease and four of fungal origin namely: smut, (*Sporisorium scitamineum* Syd.), red rot, (*C. falcatum*), leaf blast (*P.michotii*) and rust (*P. melanocephala*) were identified.

Red rot disease caused by the fungus *C. falcatum* incites to a large extent the production constraints experienced for sugar cane and sugar industry in the country. Wada (2003) reported that the pathogen causes symptoms of tissue discolouration, infects the cane stalk, inversion of sucrose due to production of pathogen induced invertases and drying up of the cane. It was found that *C. falcatum* affected 8 out of the 10 assessed CFC cane varieties causing about 80 percent infection.

Infection by this disease is not noticed at the early stages of the growth phase of sugar cane, as it is usually observed when the rains recede and sucrose accumulation begins[12]. Management options with the use of biological agents are becoming popular. Lal et al., (2014) and Olufolaji et al) reported the use of *Trichoderma* spp in the management of red rot and other diseases. While Lal *et al*., (2014) obtained 20-30% protection from *Trichoderma* against red rot, Olufolaji *et al*., (2014) reported that the *Trichoderma* sp used in their study had *in vitro* antagonistic activity against the pathogen by showing strong inhibition effect on its mycelia growth with respect to the various methods of inoculation used. Thus there is great promise in exploring the use of this biological agent in the management of the identified diseases in the present study, especially red rot and whip smut.

The isolation of leaf blast from most of the sugar cane accessions and varieties in the present study is not surprising as the disease has been reported to occur in many areas including Niger state. Blast has been observed on cane at Ibadan, Badeggi and Numan and on cane farms in Niger, Kwara, Sokoto, Katsina, Borno and the Federal Capital Territory (FCT) Abuja in Nigeria[20].

The destructive effect of whip smut (*S, scitamineum)* on sugar cane in Nigeria is well known. Wada and Anaso, (2013) reported the existence of nine races of the disease in the country show-

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TABLE 3 : Pathogenic organisms isolated and identified from the leaves of some selected sugarcane varieties at Badeggi, Nigeria

s/no		Pathogens				
	CFC varieties	S.scitamineum	C.falcatum	P. michotii	P. melanocephala	X. axonopodis
1	RB86-3129	+	-	+	+	+
2	RB82-5211	+	+	-	+	-
3	RB86-3129	-	+	-	-	-
4	RB86-7512	+	-	+	-	-
5	RB94-2291	-	-	-	-	-
Percentage		60	40	40	40	20

+ = Present; - = Absent

ing that many hitherto resistant cultivars can be succumb to any of the nine virulent races and break down, Whip smut is accounted for significant tonnage loss and reduced juice quality and can devastate large areas cultivated with susceptible varieties[1].

Indi *et al*., (2012) also reported the destructive effect of *S. scitamineum* on sugar cane that the pathogen incites considerable losses in yield and quality of infected sugar cane. In their study, the authors showed tat quality parameters like sucrose, brix and purity were adversely affected in smutted canes [6]. Comstock (2014) also elucidated the effect of whip smut on cane and concluded that its management was best with hot water treatment at different temperatures and times resulting in 90, 95 and 100% of its elimination respectively. Tushari *et al*., (2014) reported the use of soil bacteria as bio agent for the management of *S. scitamineum*. This aspect of the use of bio agent for whip smut management will form a novel strategy for the disease in future studies.

On the contrary, sugar cane rust and gumming diseases though reported to occur in the country,[14] have not been observed at Badeggi close to three decades now. Their isolation from the leaves of sugar cane multiplication plots of canes recently introduced suggests that they were brought with the canes. This is because, the introduction of sugarcane varieties in collaboration with National Sugar Development Council (NSDC) and other international organizations, and in spite of the watchful eyes of plant Quarantine Services, pathogen introduction could and is always possible. This view is further supported by similar symptoms of the diseases observed on the evaluation sites of the CFC /WASDP sugar cane varieties at Ikenne, Tsaragi, Nigeria and Zounoula in Cote d'Ivoire in July, 2014. Thus, the identification of new or indigent pathogens on sugar cane could form the basis for effective disease management strategies in order to restrict and eradicate potentially dangerous pathogens from spreading beyond manageable areas.

Biological agents are being explored elsewhere for the management of *S. Scitamineum* and *C. falcatum.* Lal *et al*., (2014) evaluated *Trichoderma* strains against major sugar cane diseases like red rot, smut; root rot, wilt and sett rot which recorded significant protection against all the diseases on soil application and/ or sett treatment. The exploration of biological agents for the management of the identified diseases is thus the priority of strategic research in the near future. This depends on the collaboration and active participation of stakeholders in Nigerian sugar industry in supporting the development of the technology. Singh (2014) advocated the use of biological management of sugar cane diseases particularly red rot and wilt of sugar cane and based their successes on three or four criteria. The first that because sugar cane is a commercial crop of high economic value, any investment in biological management will be suitably rewarded in terms of higher productivity. Secondly, that cultivation of sugar cane in large and continuous areas enables easy application of biological techniques. Thirdly, that continuous presence of sugar cane in the same fields over extended periods by way of ratoons or as the result of monocropping renders the biological agents self-sustainable and without any interruption. Lastly, sugar cane crop management is very effectively carried out by efficient agencies such as development organizations, sugar mill management among others,

providing adequate infrastructure to mass produce the biological management agents, monitor thir applications and obtain regular feedback. This advantaged management strategy in developed sugar cane and sugar industries can also be adopted by sugar cane pathologists in Nigeria when backed by a responsive stakeholder like emerging sugar mills and other users of sugar and sugar cane bye-products. As stated by Singh (2014), the practice of biological management is an economically viable strategy option which will certainly assist cane growers in improving the nutritional quality of their produce as well as their profits to alleviate poverty.

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