

*Full Length Research Paper*

# Characterization of physiologic races of sugarcane smut (*Ustilago scitaminea*) in Kenya

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Sugarcane smut disease caused by the fungus *Ustilago scitaminea* significantly reduces the yield and quality of sugarcane. The disease was first reported in Kenya in 1958, in Nyanza and Coastal provinces and currently occurs in all major sugarcane growing areas of Kenya. Planting resistant varieties is the main smut control measure in Kenya. Planting smut resistant varieties was made compulsory in Kenya in 1963. However, varieties previously confirmed resistant are now susceptible. Reports indicate that some varieties are resistant to smut in one zone and susceptible in another. An attempt was thus made to identify physiologic races of sugarcane smut in Kenya. Sugarcane smut teliospores were collected from the major sugarcane growing zones of Western Kenya in South Nyanza, Nyando, Mumias, Busia, Nzoia and west Kenya. A set of 11 sugarcane cultivars which had previously shown differential response to smut in Kenya and elsewhere were each artificially inoculated with a mixture of smut spores from each zone by dipping in a suspension of smut spores that contained  $5 \times 10^6$  teliospores per ml. Susceptibility of the cultivars to smut was measured by recording the number of smut whips that appeared within 6 to 7 months after planting. The reaction of the cultivars to smut from the various zones varied from resistance to susceptible. Three cultivars were seen in smut reaction in two tests. The results suggested existence of smut races in Kenya.

**Key words:** Kenya, sugarcane smut, *Ustilago scitaminea*.

## INTRODUCTION

Sugarcane smut was first reported in Natal, South Africa in 1877. Although, smut was first reported in 1958, it was suspected to have been present in Kenya since 1956 (Robinson, 1959). The causal organism for sugarcane smut is a fungus *Ustilago scitaminea* Sydow. Several races of *U. scitaminea* have been reported by Lee-Lovick (1978). The disease occurs wherever sugarcane is grown in Kenya and is spread by windblown spores, infested seed-cane and infested soil. A recent yield loss assessment trial in Kibos, Kenya established yield losses of 21 to 38% in plant cane in commercial cultivars under field conditions (Nzioki et al., 2006). The trial is currently ongoing and more losses are expected in successive ratoons crops. Current smut control measures are hot water treatment of seedcane, rouging out diseased plants, planting resistant or tolerant cultivars, decreasing number of ratoons for susceptible cultivars and fungicides

(Agnhotri, 1983; Ferreira and Comstock, 1989). Because host resistance is the most cost effective favourable control, many resistant varieties have been developed. Recent reports show that when available resistance is short lived, it could be attributed to pathogen genetic variability. A recent survey carried out in western Kenya showed that some varieties susceptible to smut in one location were immune or resistant in other locations (KESREF, 2002) indicating genetic diversity of smut races. Moreover, the sugarcane varieties in Kenya are polyploids of several *Saccharum* species whereby genetic resistance for smut do not follow gene-for-gene pattern. Varietal differences in susceptibility to different smut isolates have been reported (Comstock and Heinz, 1977; Gillaspie et al., 1983; Grisham, 2001). Smut rating and ranking of the cultivars can vary significantly from year to year since host reaction to smut is dependent on the environment and probably races of the pathogen present (Lee-Lovick, 1978).

Smut races have been documented in many sugarcane growing areas of the world. Races of *U. scitaminea* have

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**Table 1.** Reactions of sugarcane cultivars to smut from four zones in 2004 at Kibos.

Cultivar	Smut source (% smut infection)			
	Nyando	S. Nyanza	Mumias and Busia	Nzoia and W. Kenya
B52-107	6.0	0.0	0.0	0.0
CO1148	5.0	0.0	0.0	0.0
CP29-116	1.0	0.0	17.3	0.0
NCO376	16.0	17.0	29.0	18.0
CO421	13.0	13.4	23.0	14.2
F134	15.0	0.0	0.0	13.2
H50-7209	14.1	15.0	17.2	15.9
EAK 7097	0.0	0.0	0.0	0.0
CO945	18.8	14.9	18.3	13.9

been described in Hawaii (Comstock and Heinz, 1977), Taiwan (Hsieh and Lee, 1978; Leu, 1978), Brazil (DaSilva and Sanguino, 1978; Toffano, 1976) and Pakistan (Muhammaed and Kausar, 1962). Six races were differentiated on five sugarcane cultivars among a collection of teliospores from Argentina, Florida, Hawaii, Taiwan and Zimbabwe (Gillaspie et al., 1983). Recently, Schenck (2003) reported a new race of *U. scitaminea* in Hawaii using 10 varieties (H65-7052, H77-4643, H78-3567, H78-4153, H83-7061, H87-4319, H87-5794, H88-2953 and H90-7492). He found that varieties H58-7750 and H83-7061 which were previously resistant to the old races were now susceptible to the new race. Grishan (2001) conducted an international project to identify races of *U. scitaminea* at fourteen locations in ten countries using a set of 11 differential cultivars (B52-107, CO421, CO1148, CP29-116, CP63-588, F134, H50-7209, H73-6110, M31/45, N55-805 and NCO310). He obtained strong evidence for distinct races in Taiwan.

In Kenya, cultivar CO 421, which was previously resistant to smut is now susceptible and is gradually being withdrawn from commercial cultivation in Mumias, South Nyanza and Nyando. Similarly, cultivar NCO376 was withdrawn from commercial cultivation in 1990's due to its susceptibility to smut. Some smut whips are now being observed in cultivars EAK7097, CO1148 and N14 which were previously resistant. These observations indicate that cultivars CO421, NCO376, N14, EAK7097 and CO1148 can be used as differential hosts for distinguishing smut races in Kenya. The contemporary population of smut in Kenya has not been characterized for genetic diversity to quantify physiologic races. The objective of this study was to identify physiologic races of sugarcane smut (*U. scitaminea*) in Kenya.

## MATERIALS AND METHODS

Two field trials were established at KESREF Hqs, Kibos at an altitude of 1184 m.a.s.l and situated at latitude 0°, 34° and longitude 04°S/48°E at lower midland zone 2. Kibos has a sub-humid climate characterized by high day temperatures, cool nights and bimodal rainfall pattern. Mean annual rainfall is 1464 mm while mean daily

temperature is 23°C. The long rains start in March and end in June while the short rains start in September and end in November. Average temperatures, day lengths, evaporation and radiation vary very little through out the year (Table 1). The first trial was planted during long rains (May 2004) and the second one during the same season in June 2006.

## Collection of smut populations

Dry smut spores were collected from commercial and pre-released varieties in the four major sugarcane growing zones of western Kenya [South Nyanza; Mumias and Busia; Nyando (Chemelil, Muhoroni, Miwani and Kibos) and Nzoia; and West Kenya] one month prior to planting each trial. The collection was carried out from nucleus estates, farmers' fields and at KESREF Kibos from commercial varieties (CO421, CO945, CO617, CO1148, CB3822, EAK7097 and EAK 33-335) and pre-released (KEN82-472 and D8484) varieties. Smut spores from each zone were bulked to form a composite mixture and stored in size 5 paper envelopes under dry conditions in the laboratory.

## Cultivars

Eleven cultivars were used in this study. They included B52-107, CO 421, CP29-116, F134, H50-7209, M31/45, NCO376, CO1148, CO945, N14 and EAK7097. Cultivars B52-107, Co 421, CP29-116, F134, H50-7209 and M31/45 were chosen because they had previously been used as differential hosts in other countries (Grishan, 2001; Gillaspie et al., 1983). In addition, cultivar CO421 was chosen because of its commercial importance in western Kenya and because its resistance to smut has breakdown over time. Cultivar NCO376 was selected because of its susceptibility to smut in all cane growing areas. Cultivars CO1148, CO945, N14 and EAK7097 were chosen because they are resistant to smut and are commercially important to the sugar industry in Kenya. Seed-cane was obtained from breeders seed maintained at KESREF. The seed-cane age of cultivars planted in 2004 varied from 18 - 24 months whereas, that of 2006 was uniform (eight months). Three budded setts of each cultivar were subjected to hot water treatment at 52°C for 20 min (Fauconnier, 1993) in a water bath, cooled and stored in polythene sacks.

## Smut inoculation

In 2004, 73 budded setts of each cultivar were inoculated with smut populations from all sugarcane growing zones in western

**Table 2.** Reactions of sugarcane to smut from four zones in 2006 at Kibos.

Cultivar	Smut source (% smut infection)			
	Nyando	S. Nyanza	Mumias and Busia	Nzoia and W. Kenya
B52-107	0.0	0.0	0.0	0.0
CO1148	13.1	0.0	0.0	0.0
CP29-116	14.0	0.0	0.0	0.0
NCO376	0.0	0.0	0.0	0.0
CO421	26.9	13.4	16.7	33.0
F134	16.0	14.4	13.5	14.2
H50-7209	0.0	0.0	0.0	0.0
M31/45	0.0	0.0	61.4	16.7
N14	15.1	59.7	0.0	3.4
EAK 7097	0.0	0.0	0.0	0.0
CO945	18.8	14.9	18.3	13.9

**Table 3.** Smut rating by % plant infection.

Description	Rating	Plant infection (%)
Highly resistant	1	0 - 3
Resistant	2	4 - 6
Resistant	3	7 - 9
Resistant	4	10 - 12
Moderately susceptible	5	13 - 25
Susceptible	6	26 - 35
Highly susceptible	7	36 - 50
Highly susceptible	8	51 - 65
Highly susceptible	9	66 - 100

Kenya (Nyando, S. Nyanza, Mumias-Busia, Nzoia and W. Kenya) to form a 7 x 4 cultivar by smut population combination. The dipping method of inoculation was used by dipping setts for 30 min in suspension of smut spores containing about  $5 \times 10^6$  teliospores per ml. The respective setts were incubated for 12 h in polyether sacks and then planted in the field (Nasr, 1977; Ferreira et al., 1980).

In 2006, 11 three budded setts of each cultivar were inoculated with smut populations from all sugarcane growing zones of western Kenya (Nyando, S. Nyanza, Mumias-Busia, Nzoia and W. Kenya) to form a 11 x 4 cultivar by smut population combinations. Since smut inoculum from S. Nyanza was not abundant, the bud- paste method of inoculation was used. This was done by brushing a thick paste of smut teliospores on the buds. The cultivars were immediately planted in the field after inoculation.

#### Experimental design

2 m long single row plots were planted in 2004 without replication due to shortage of seed-cane. In 2006, the test cultivars were planted in a randomized complete block design and replicated three times. The test cultivars were placed in the main block and smut populations from each sugarcane growing zone in the sub-plots. Each test cultivar-smut population combination consisted of 6 setts, 2.5 m long.

#### Recording smut incidence

The trials were monitored for appearance of first smut whips. The

smut whips appearance were observed and recorded monthly from the emergence of the first whips until peak whip formation, about seven to nine months after planting, depending on cultivar. Thereafter, plants infected with smut were recorded, cut and removed from the field until the trial was completed. Smut description, rating and infection were done as explained by Agnihotri (1983) (Table 2). 2006 planted trial, data on smut incidence was collected from the three replicates. The average of the three replicate trial represented cultivar reaction to smut. Cultivars were considered resistant when percentage plant infection was 0 - 12%, while those showing greater than 13% plant infection were considered susceptible (Table 3).

## RESULTS

### 2004 planted trial

All the cultivars inoculated with smut from Nyando zone were susceptible to smut. However, B52-107 and CO 1148 were resistant to smut from S. Nyanza, Mumias-Busia and Nzoia-W. Three sugarcane cultivars (NCO 376, CO 421 and H50-209) were susceptible to smut populations from all sugarcane growing zones. CP29-116 was resistant to smut obtained from S. Nyanza and Nzoia-W. Kenya, while F134 was resistant to smut from S. Nyanza and Mumias-Busia (Tables 1 and 3).

### 2006 planted trial

Results from cultivars planted in 2006 were variable from those in 2004. Cultivars F134, Co 421 and Co 1148 were susceptible to smut from Nyando whereas, B52-107, CP 29-116, NCO 376 and H50-7205 were resistant to all smuts. New cultivars (N14, and CO 945) evaluated in 2006 field trials were susceptible to smut from Nyando zone population with the exception of M31/45 and EAK 7097. Cultivar M31/45 was susceptible to smut population from Mumias-Busia and Nzoia-W. Kenya but resistant to smuts from Nyando and S. Nyanza. EAK 7097

7097 was resistant to smut from the four zones. N14 was susceptible to smut from Nyando and S. Nyanza. Cultivars F134, CO 421 and CO 945 were susceptible to smut from all the four zones (Tables 2 and 3).

## DISCUSSION

With the exception of cultivars CO 421 and CO 1148, the reactions to smut of the other cultivars were variable in years 2004 and 2006 plantings, respectively. This was probably due to variations in smut population races from each zone over the two year period, cultivar-environment interaction, differences in seed cane age and in inoculation methods used during each planting.

Results from this study show the existence of smut races in Kenya. Evidence indicates the presence of 11 races, that is, races A and B defined in Hawaii (Comstock and Heinz, 1977), two races defined in Brazil (da Silva et al., 1978; Toffano, 1976), two races defined in Taiwan (Hsieh et al., 1978; Leu et al., 1976) and five races defined in Pakistan (Muhammad et al., 1962). In South Africa, cultivar H50-7209 is susceptible to smut but resistant in Taiwan. This suggests existence of different smut races in South Africa and Taiwan. F134 is susceptible to strain 2 but resistant to strain 1 of smut in Taiwan (Leu et al., 1972). The 2004 results indicate that probably, both races 1 and 2 were present whereas, the 2006 inoculum consisted of only race 1. Cultivar M31/45 has been observed to be resistant to smut in many locations (Grishan, 2001). In this study, it showed resistance and susceptible reactions suggesting that it is a potential differential host in Kenya.

In Kenya, commercial cultivation of NCO 376 was discontinued due to its susceptibility to smut. During the 2004 trial, this variety was susceptible to smut races from all the four zones and was almost wiped out by this disease. In 2006 trial, NCO 376 was resistant to all smut populations suggesting that the smut races tested in 2004 were different from those used in 2006. There is evidence indicating existence of smut races in Kenya. Variability in smut reactions of cultivars used in this study indicate that the cultivars can be used as differential hosts for differentiating smut races in Kenya.

## Conclusion

Our preliminary results suggest existence of smut races in Kenya. There was variability in seed cane age and in inoculation protocol over the two years of testing in the trials. In order to obtain conclusive results, our cultivars and testing protocols need to be standardized. Plans are currently underway to repeat the trial using dip-in and bud paste inoculation methods with few cultivars of the same age.

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## REFERENCES

- Agnhotri VP (1983). Diseases of sugarcane. Oxford and IBH Publishing Co. pp. 65-86.
- Da Silva WM, Sanguino A (1978). Evaluating reaction of American cane varieties to *Ustilago scitaminea* in Brazil. Sugarcane Pathologists' Newsletter, 21: 10-11.
- Comstock JC, Heinz DJ (1977). A new race of culmicolous smut of sugarcane in Hawaii. Sugarcane Pathologists' Newsletter, 19: 24-25.
- Fauconnier R (1993). The Tropical Agriculturist: Sugar Cane. 3<sup>rd</sup> Edition, Published by Macmillan Press Ltd, London and Basingstoke
- Ferreira SA, Comstock JC, WU KK (1980). Evaluating sugarcane for smut resistance. Proc. Int. Soc. Sugarcane Technol., 17: 1463-1476.
- Ferreira SA, Comstock JC (1989). In: Diseases of sugarcane. Major Diseases. C. Ricaud, B.T. Egan, A.G. Gillespie Jr. and C.G. Hughes (Eds.), Amsterdam, The Netherlands, pp. 211-229.
- Gillaspie AG, Mock RG, Dean JL (1983). RNA from the yeast transposable element TY1 has both ends in the direct repeats, a structure similar to retrovirus RNA. Proceedings National Academy of Sciences. USA., 80: 2432-2436.
- Gillaspie AG, Mock RG, Dean JL (1983). Differentiation of *Ustilago scitaminea* isolates in greenhouse tests. Plant disease, 67: 373-375.
- Grishan MP (2001). An international project on genetic variability within sugarcane smut. Proc. ISSCT 24: 459-461.
- Hsieh WH, Lee CS (1978). Compatibility and pathogenicity of two races of *Ustilago scitaminea* Sydow in Taiwan. Taiwan Sugar, 25: 46-48.
- Lee-Lovick GL (1958). Smut of Sugarcane-*Ustilago scitaminea*, Rev. Plant Pathol., 57: 181-188.
- Leu LS, Teng WS (1972). Pathogenic strains of *Ustilago scitaminea* Sydow. Sugarcane Patholo. Newsl., 8: 12-13.
- Leu LS, Wang ZZ, Tzean SS (1976). Cooperative disease resistance trial for foreign sugarcane varieties in Taiwan. Taiwan Sugar Expt. Station Reports 72: 31-39.
- Muhammad S, Kausar AG (1962). Preliminary studies on the genetics of sugarcane smut, *Ustilago scitaminea* Sydow. Biologia, Lahore, 8: 65-74.
- Nasr IM (1977). Standardization of inoculation techniques for sugarcane smut disease. Sugarcane Pathologist' Newsletter, 18: 2-5.
- Robinson RA (1959). Sugarcane smut. East Afr. Agricul. For. J., 24: 240-343.
- Sanguino A, Tokeshi H (1976). Vacuum and *Ustilago scitaminea* changing bud development of sugarcane cuttings. Brasil Acucareiro, 87: 30-35.
- Schenck S (2003). New race of sugarcane smut on Maui. Hawaii Agriculture Research Center Pathology Report.
- Toffano WB (1976). Estudos sobre polaridade sexual e possíveis raças fisiológicas de *Ustilago scitaminea* Sd., no estado de Sao Paulo. Arq. Inst. Bio. Sao Paulo, (43 ¾): 65-79.