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Cultivar Resistance of Sugarcane and Effects of Heat Application on Nematodes in Kenya

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ABSTRACT

This study was carried out with the aim of screening selected sugarcane germplasm for their response to nematodes and evaluating the potential of soil heat treatment in the management of the nematodes. Seven sugarcane cultivars namely CO421, CO617, CO945, EAK70-97, KEN83-737, KEN82-808 and KEN82-216 were evaluated against N14 as the reference germplasm for resistance against plant-parasitic nematodes in the greenhouse. Data on nematode numbers, shoot and root weight was recorded at 0, 60 and 120 Days after Planting (DAP). Nematodes were extracted from the sugarcane rhizosphere using the Modified Baermann funnel technique. Sugarcane cultivar KEN83-737 was rated as highly resistant while moderately resistant cultivars included CO421, CO617, CO945, EAK70-97 and KEN82-216. Cultivar N14 was confirmed to be susceptible to nematodes. Nematodes from four genera namely *Pratylenchus*, *Scutellonema*, *Helicotylenchus* and *Hoplolaimus* were significantly responsive to the sugarcane cultivars. Among the nematodes associated with sugarcane, lesion nematodes (*Pratylenchus* sp.) were dominant in all the cultivars averaging 188/200 cm^3 soil, while the lance nematode (*Hoplolaimus* sp.) had the lowest numbers (92/200 cm^3 soil). Soil heat treatment resulted in increased sugarcane root biomass. In addition, heat treating the soil resulted in a 27 and 30% increase in sugarcane root and shoot lengths, respectively. Similarly, compared to the controls, 52 and 25% increase in root and shoot weights were recorded in sugarcane grown in heat treated soil, respectively. This study has shown that sugarcane cultivar resistance is a viable management strategy for nematodes and further demonstrated the potential of soil heat treatment in the sustainable management of nematodes in sugarcane.

Key words: Pratylenchus, Scutellonema, reaction, thermo-treatment, varieties, susceptible

INTRODUCTION

A long history of association has existed between nematodes and sugarcane (*Saccharum officinarum*). Plant parasitic nematodes have been identified as one of the most important causes of the decline in sugarcane production worldwide (Cadet and Spaull, 2003). According to Shoko and Zhou (2009) annual sugarcane yield losses attributed to nematodes were 0.2% in Australia, 3% in Peru, >5% in South Africa, 6% in USA, 11% in Cote d'Ivoire and 14% in Burkina Faso. A survey conducted in the major sugarcane growing regions in Kenya revealed great nematode diversity, especially those considered to be highly pathogenic sugarcane (Chirchir *et al.*, 2008). To date, a record 96 species of nematodes have been associated with sugarcane (Quireshi *et al.*, 2002). Globally, nematodes in the genera *Pratylenchus*, *Helicotylenchus*, *Meloidogyne* and *Scutellonema* cause serious damage to sugarcane (Shoko and Zhou, 2009). While, nematode infestation in a variety of crops results in root damage, reduced water and nutrient uptake and stunted growth (Gaidashova *et al.*, 2008), yield losses by nematodes in sugarcane are largely due to reduction in the number of stalks and decreased length of stalks (Berry *et al.*, 2007). Management of plant-parasitic nematodes would potentially result in considerable yield increases (Chirchir *et al.*, 2008). Presently, nematode management in sugarcane is mainly through rotation, replanting, mulching as well as nematicide application (Cadet and Spaull, 2003). Application of nematicides, which has been the backbone of nematodes management in sugarcane crop, has been shown to increase stalk population and stalk length by 46 and 35%, respectively (Quireshi *et al.*, 2002).

Breeding of sugarcane cultivars with resistance to nematodes may be the most practical option for sustainable nematode management (Klink and Matthews, 2009). Nematode tolerant cultivars have been shown to reduce root damage by 32% (Cadet and Spaull, 2003). However, since sugarcane is often attacked by a diverse community of plant parasitic nematodes, breeding for combined resistance is a challenging task. Nevertheless, previous studies have identified multiple resistances to *M. javanica*, *P. zeae* and tolerance to *P. brachyurus* in a Brazilian cultivar SP70-1143 (Tew *et al.*, 2005). Similarly, cultivar CP70-321 is tolerant to several plant parasitic nematodes including species of *Criconemoides, Paratrichodorus* and *Tylenchorhynchus* (Koenning *et al.*, 1999). Resistance and/or tolerance to species of *Meloidogyne* and *Pratylenchus* have been identified in the sugarcane cultivar collections of several countries (Berry *et al.*, 2007). In India, Rashid and Singh (2002) showed that both resistant and susceptible cultivars respond to pathogenic invasion with qualitative and quantitative modifications in peroxidase and acid phosphatase activity in roots and in leaves.

While there is a remote chance of finding cultivars that are resistant to a wide spectrum of plant-feeding nematodes (Cadet and Spaull, 2003), the selection of resistant and/or tolerant cultivars that withstand nematode damage is a viable option. This is particularly important in Kenya where new sugarcane cultivars have been recently released to the local farmers by the Kenya Sugar Research Foundation (KESREF). Non-specific neurotoxins with poor environmental and worker-safety profiles associated with nematicides such as organophosphates and carbamates has led to restriction/withdrawal of such from the market (McCarter, 2008). Therefore, there is potential in improving the soil biological efficiency by the substitution of chemical nematicides with soil solarization (Stapleton *et al.*, 2002). Soil solarization minimizes adverse effects to non-target organisms and further enhances soil recolonization by beneficial microorganisms (Ruiz *et al.*, 2003). Combined with host resistance, soil solarization may provide great potential as an ecologically sound practice to be integrated in the management of nematodes in sugarcane. The objective of this study was therefore, to screen sugarcane cultivars in Kenya for their reaction to and to evaluate the potential of soil solarization in the management plant parasitic nematodes in sugarcane.

MATERIALS AND METHODS

This study was carried out in the period between June, 2007 and June 2008 in a glasshouse at the University of Nairobi, Kabete campus. Seven varieties representing 70% of the total hectarage under sugarcane were screened for their reaction to nematodes. These included foreign introductions namely CO421, CO617, CO945 and locally-bred recently-released varieties;

EAK70-97, KEN83-737, KEN82-808 and KEN82-216 with N14 was as the reference cultivar. All seed-cane was sourced from Kenya Sugar Research Foundation (KESREF), Kibos, Kisumu.

Natural nematode infested soil was collected from sugarcane rhizosphere and separated into two batches of which one was subjected to heat treatment at 60°C for 30 min. All soil was mixed with sand at a ratio of 2:1 and potted in 15 cm-diameter polythene sleeves and 20 g of N:P:K fertilizer (17:17:17) added. A single budded-cane seed sett treated with Antracol WP70 (a.i. Probineb) to control fungal infections was planted in each pot and the experiment arranged in a completely randomized design with three replications in a glasshouse. Top dressing was done using 20 g of Urea (46 %N) 40 days after planting. Data on nematode numbers and shoot length were collected at 0, 60 and 120 Days after Planting (DAP). Nematodes were extracted from 200 cm^3 soil using the modified Baermann funnel technique described by Hooper *et al.* (2005). Nematodes from each sample were fixed using rapid Seinhorst technique and thereafter mounted on Cobb-type aluminum double cover glass that allow for examination from either side (Siddiqi, 2000). Nematode identification was based on morphological characteristics and pictorial keys using high power microscope (Hunt *et al.*, 2005). At the end of experiment, 120 DAP, data on root length and root weight were collected.

Statistical analysis: Nematode numbers and their effects on shoot length, root length and weight were subjected to analysis of variance (ANOVA) using Genstat computer software package (Lawes Agricultural Trust Rothamsted Experimental Station 2006, version 9). Nematode numbers were transformed to Log(X+1) before analysis. When ANOVA indicated significant differences among treatments, means, were separated using the Fisher' protected LSD test at 5% probability level.

RESULTS

Fourteen genera of plant parasitic nematodes occurring in significantly varying ($p \le 0.05$) proportions were recovered from soil collected from sugarcane rhizosphere (Fig. 1). The most dominant nematodes in the sugarcane rhizosphere belonged to the genera *Pratylenchus* (17.7%). Nematodes of the genera *Scutellonema* (11.5%), *Helicotylenchus* (10.6%) *Paratylenchus* (9.6%) *Tylenchus* (8.8%), *Hoplolaimus* (8.2%) and *Rotylenchus* (7.5%) showed moderate occurrence compared to *Tylenchorynchus*, *Trichodorus*, *Belonolaimus*, *Aphelenchoides*, *Meloiodgyne*, *Xiphinema* and *Ditylenchus* whose occurrence was lowest.

Significant variations ($p \le 0.05$) were observed in the response of sugarcane cultivars to nematode infestation (Table 1). Compared to the standard cultivar N14, all the sugarcane cultivars evaluated supported had significantly less nematode population densities. Among the sugarcane varieties, KEN83-737 supported significantly the lowest nematode numbers belonging to the

Table 1: Reaction of sugarcane varieties to plant parasitic nematodes

	Nematode numbers/200 cm^3 soil								
Nematode genera	N14	CO945	CO421	KEN 82-808	KEN 82-216	EAK 70-79	CO617	KEN 83-737	Overall mean
Helicotylenchus	17a*	17a	14ab	12abc	17a	11bc	14ab	8c	13b
Hoplolaimus	23a	12bc	11bc	8c	8c	9bc	13b	8c	10b
Scutellonema	20a	15bc	18ab	15bc	13cd	15bc	13cd	10d	14b
Pratylenchus	31a	23b	21b	25ab	20b	22b	22b	24b	21a

*Means followed different letter(s) along rows are significantly different genera *Pratylenchus*, *Scutellonema*, *Helicotylenchus* and *Hoplolaimus*. As expected, cultivar N14 supported had the highest number of plant parasitic nematodes. Among these nematodes, those belonging to the genera *Pratylenchus* were, again, the most dominant compared to *Hoplolaimus*, *Helicotylenchus* and *Scutellonema* which recorded the lowest numbers.

Significant differences ($p \leq 0.05$) in the numbers of plant parasitic nematodes were observed when sugarcane was planted in heat treated soil. Treating the soil with heat higher led to higher sugarcane root biomass was observed (Fig. 2). When soil was heated treated, a 27 and 30% increase were recorded in sugarcane root and shoot lengths, respectively (Fig. 3). Similarly, compared to the controls, a 52 and 25% increase in root and shoot weight, was recorded in sugarcane grown in heat treated soil (Fig. 4).

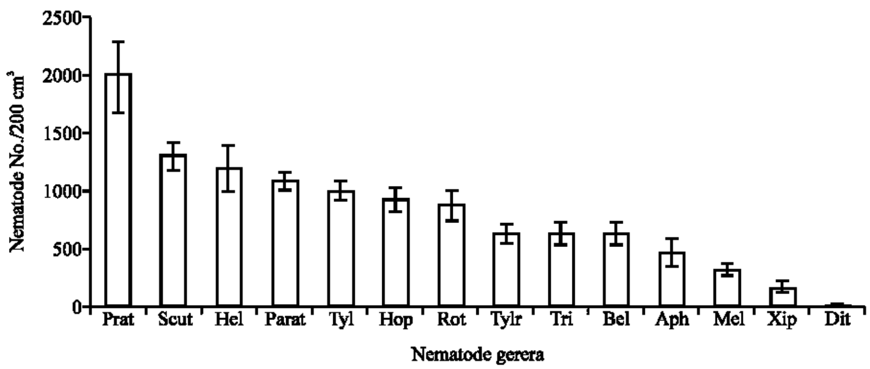

Fig. 1: Nematode genera and numbers recovered from soil before planting sugarcane. Prat-*Pratylenchus*, Scut-*Scutellonema*, Hel-*Helicotylenchus*, Parat-*Paratylenchus*, Tyl-*Tylenchus*, Hop-*Hoplolaimus*, Rot-*Rotylenchus*, Tylr-*Tylenchorhynchus*, Bel-*Belonolaimus*, Aph-*Aphelenchoides*, Mel-*Meloidogyne*, Xip-*Xiphinema* and Dit-*Ditylenchus*

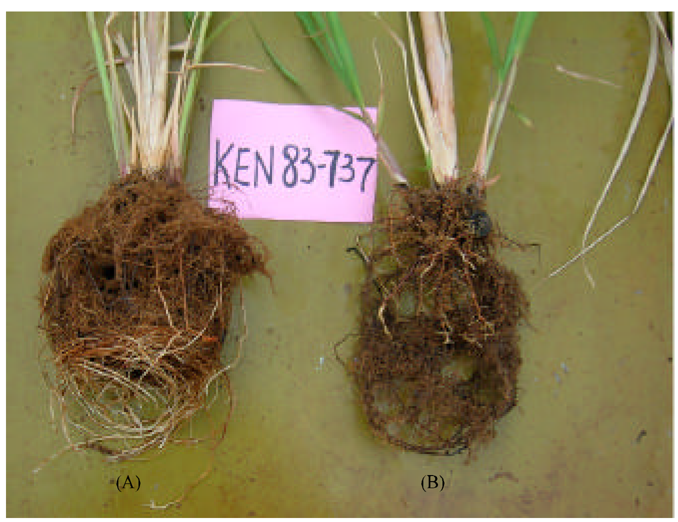

Fig. 2: (A) Root biomass of sugarcane grown in heat treated soils (B) and reduction of sugarcane root biomass by parasitic nematodes in sugarcane grown in non-heat treated soil

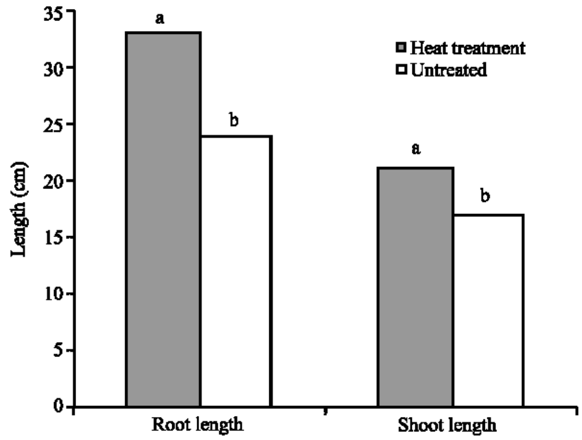

Fig. 3: Effect of soil heat treatment on the root and shoot lengths of sugarcane. Bars headed by different letter(s) are significantly ($p \le 0.05$) different by least significant difference test

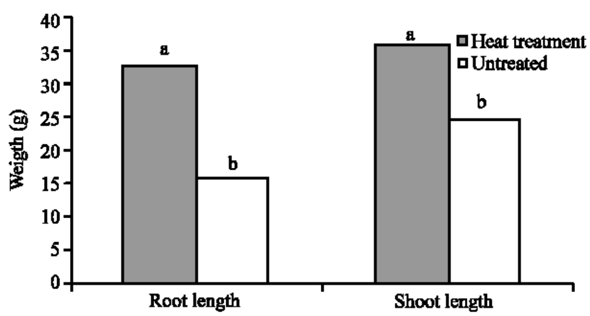

Fig. 4: Effect of soil heat treatment on the root and shoot weight of sugarcane. Bars headed by the same letter(s) are not significantly ($p \le 0.05$) different by least significant difference test

DISCUSSION

Nematode resistance and susceptibility are collectively terms used when a host plant suppresses (resistance) or allows (susceptibility) nematode development and reproduction (De Waele and Elsen, 2007). In this regard, root damage due to nematodes and their population densities, were used to establish the status of host resistance of selected sugarcane cultivars to nematodes. In this study, differences in root damage and reduction in stalk length were an indication of the varying levels of host resistance among the sugarcane varieties. Host resistance has been reported to be a viable option in recent studies on tomato (Adebayo and Salawu, 2010). Several authors in different countries have reported varying levels of resistance or tolerance when sugarcane cultivars were subjected to various species of plant parasitic nematodes (Koenning *et al.*, 1999; Cadet and Spaull, 2003; Tew *et al.*, 2005; Shoko and Zhou, 2009). While environment in itself may have contributed to the variations in root biomass (Rajab *et al.*, 1999), nematode population densities and reduced root and shoot weight were the most significant in this study in showing differences in host reaction by sugarcane varieties to nematodes.

Among the sugarcane cultivars evaluated, KEN83-737 was classified as resistant since it supported the least nematode population densities and recorded the highest root biomass among all the cultivars. Present findings of sugarcane cultivar resistance to plant parasitic nematodes have been corroborated by Koenning *et al.* (1999), Cadet and Spaull (2003) and Tew *et al.* (2005). However, Chaves *et al.* (2009) reported that all sugarcane varieties assayed were good hosts to *M. incognita*, allowing free nematode reproduction which subsequently resulted in higher reproductive factors. With the exception of N14 which was classified as susceptible, all the other cultivars were rated as moderately resistant. While reports relating to nematode host status of KEN83-737 are scanty, our results confirm the classification of N14, which is a recent introduction from South Africa as a nematode susceptible cultivar (Shoko and Zhou, 2009). Cultivar KEN83-737 is among the recent new introductions by the Government of Kenya whose main attributes include early maturity, high sucrose content and high cane yield.

De Waele and Elsen (2007) have advanced the concept of 'multispecies nematode populations as the commonest nematode species associated with crops generally known. Our findings revealed sugarcane multispecies nematode populations with a specific nematode profile constituting of *Pratylenchus, Scutellonema, Helicotylenchus* and *Hoplolaimus* in the Kenyan soils. Nematode species profiles for some crops, including sugarcane, may vary between regions as evidenced in studies by Quareshi *et al.* (2001) and Cadet and Spaull (2005). Evidence is available on the influence of different sugarcane varieties on *Pratylenchus* which is clearly confirmed in the current study (Tew *et al.*, 2005). However, the influence of varieties on *Scutellonema, Helicotylenchus* and *Hoplolaimus* is seemingly a new finding of this study as little has been reported about them.

Heat treatment of soil led to reduction of plant parasitic nematodes resulting in increase in growth. Soil heat treatment has been employed successfully in vegetable production systems in reducing pathogenic microbes (Megueni *et al.*, 2006). In this study, it was evident that soil heat treatment inactivated nematode parasites of sugarcane. In addition, the higher biomass associated with the root and shoots of sugarcane grown in heat treated soil are reported by Braga *et al.* (2001). Conventional heat treatment of soil at 60°C for a minimum period of 1 h has been shown to have an added benefit of high recolonization by soil microorganisms that are antagonistic to subsequently invading nematodes, pathogens, or weed propagules. The inactivation of the nematodes tested in these experiments using heavily-infested soil is a clear indication that heat treatment can manage phytoparasitic nematodes from containerized soils without the risk or expense of chemical or steam treatments (Stapleton *et al.,* 2002). As such, soil heat treatment may be used in establishment of new sugarcane orchards especially where steam generation may be technically or economically unfeasible and safety precautions for use of highly toxic fumigant chemicals may be lacking. Moreover, studies have shown that free-living nematodes were better able to tolerate the 46°C heating treatment than phytoparasites (Ruiz *et al.*, 2003).

In conclusion this study has demonstrated that sugarcane cultivars have variable host resistance status to nematodes belonging to the genera *Pratylenchus*, *Scutellonema*, *Helicotylenchus* and *Hoplolaimus*. With the exception of nematode susceptible cultivar N14, all the sugarcane cultivars offered varying levels of resistance to nematodes.

In view of the progress made in understanding nematode cultivar resistance, this study has helped classify sugarcane cultivars in Kenya according to their host response to plant parasitic nematodes.

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