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Full Length Research Paper

Association of maize streak virus disease and its vectors (Homoptera: Cicadelidae) with soil macronutrients and altitudes in Kenya

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Maize (*Zea mays* L.) is a staple food for more than 100 million people in Africa. Its production is constraint by maize streak virus (MSV) disease. This study investigated the interrelationships between altitudes and levels of soil macronutrients with MSV disease and its vectors. The influence of nitrogen, phosphorus and potassium levels on MSV and on populations of *Cicadulina spp.* (Homoptera: Cicadellidae) were investigated at six geographically separated maize producing regions in Kenya. Comparisons of numbers of sampled *Cicadulina spp* showed that areas at lower than 1550 m above sea level had significantly more leafhoppers than those at higher altitudes ($p < 0.05$, SNK). Soil analysis showed that nitrogen was low (0.06 - 0.12%), phosphorus (13.5 - 37.3 ppm) and potassium adequate (86 - 707 ppm). The yield loss due to MSV expected by the farmers in Oyani was similar to that in Mwea (>40%) and higher than that in Bahari, Muhoroni, Kimilili and Githunguri (< 30%) and it correlated positively with numbers of *Cicadulina mbila*. A significant positive correlation was detected between potassium with *C. mbila.* This study shows that soil nutrient levels influence MSV vector (*Cicadulina spp*) and that this relationship is critical during the design of crop protection packages.

Key words: Agro-ecology, *Cicadulina*, nitrogen, phosphorus, potassium and elevation.

INTRODUCTION

Maize (*Zea mays* L.) is a principal food and cash crop for over 100 million people in Africa (Bosque-Perez, 2000). It is mainly produced by small-scale farmers for food and income generation (Ajanga and Hillocks, 2000; Mati, 2000), although, its production is constrained by maize streak virus (MSV) disease (Kyetere et al., 1999; Bosque-Perez, 2000; Alegbejo et al., 2002). Several MSV management strategies like host plant resistance, chemical and cultural control have been tried (Bosque-Pérez, 2000; Rensburg, 2001), however, sporadic outbreaks of the disease has continued to occur with significant yield losses (Bosque-Perez et al., 1998; Atiri et al., 2000).

Most of the current MSV disease management initiatives have assumed that the causative viral pathogen is responsible for the significant reductions in productivity of maize, in Africa (Bosque-Perez, 2000; Willment et al., 2001; Alegbejo et al., 2002). Consequent studies have tended to focus on understanding the genomic variability of the streak virus, improvement of genetic traits of the crop against the virus and an understanding of the mechanisms of MSV transmission (Wambugu and Wafula, 2000; Martin et al., 2001; Danson et al., 2006). On the contrary, Altieri and Nicholls (2005) stated that an imbalance in a plants' ecosystem could be responsible for triggering insect and disease organisms to become constraints to crop production.

This study examined the interrelationships within an insect-disease-plant system with the aim of identifying synergisms between the main biological components of a maize agro-ecosystem. The specific objectives were to assess the interrelationships between altitudes and levels of soil macronutrients with MSV vectors and disease prevalence. This paper reports the results of the activities carried out in farm surveys in six ecologically distinct

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Table 1. Description of the study areas.

Region	Average altitude (m a.s.l)	Annual mean Temperature (^{o}C)	Annual average rainfall (mm)
Bahari	80	25.9	1000
Mwea	1350	21.9	1100
Muhoroni	1380	23.8	1150
Oyani	1550	20.0	1550
Kimilili	1680	18.6	1700
Githunguri	1850	16.8	1500

Source: Kenya Department of Meteorology (averages of data taken over five years 2000 - 2005).

maize producing regions in Kenya. Information on the interrelationships between the components of a maize agro-system is useful for designing packages for the management of MSV disease.

MATERIALS AND METHODS

Study area

Quantitative surveys were conducted using checklists during the months of April to May (long rain season), in 2005 and 2006, in six diverse maize growing regions of Kenya (3° 55'S - 0° 47'N, 34° 43'E - 39° 44'E). A team of researchers and agricultural extension agents surveyed maize fields in Kimilili, Oyani and Muhoroni in the western part of the country, Githunguri and Mwea in the central part, and Bahari at the coast of the Indian Ocean. Regions were predetermined on the basis of difference in altitude and soil type. Thirty farms were visited per region to obtain a representative sample of the different agro-ecological areas (Table 1).

Estimation of yield loss due to MSV

Interviews were conducted using a checklist on every seventh farm along a predetermined rural path in each region. The farmers identified the maize varieties that they grew and estimated the annual yield lose due to MSV that they incurred in the last five years.

Leafhopper and soil sampling

A criterion of selecting farms for leafhopper sampling was the presence of young maize (3 to 6 weeks old plants) and/or adjacent grass patches. Leafhoppers (*Cicadulina spp*) were collected by suddenly dropping a trapping cage (1 m^2) on host plants (maize or grass) on a farm where interview was done. The trapping cage used, was a modification from the description by Okoth and Dabrowski (1987). Moreover, ten trappings were carried out per region (5 on maize and 5 on the prevalent natural grass species). The collected leafhoppers (approximately 210 insects per region), were preserved in 70% alcohol and taken for taxonomic identification. The sampled host plants were maize H614, H513, Pioneer variety, and open- pollinated local varieties (all are susceptible to MSV disease); grasses: *Eragrotis hispida* K. Schum., *Digitaria nuda* Schumach and *Sporobolus scitulus* W. D. Clayton (Clayton et al., 2002).

A hand-auger sampler was used to obtain five soil samples (0.5 kg each) to a depth of 0.2 m (That is the tillage depth) in a V-pattern covering each farm on four selected farms per region, among those that were sampled for leafhoppers. The farms were selected to cover the cross section of each sampling region. Soil samples from each farm were mixed thoroughly in a clean plastic container, out of which a sub- sample of the mixture (about half a kilogram of soil), was labeled and taken to the laboratory. These samples were oven-dried (at 40°C for three days) to prevent alteration of the nutrient concentrations by soil microorganisms and subjected to soil analysis (N, P and K) according to Okalebo et al. (2002).

Data analysis

All data were analyzed using the SPSS (2001) statistical analysis program (release 11.5 for Windows). *Cicadulina spp* and *Cicadulina mbila* abundance were calculated by multiplying the numbers of leafhoppers in each 1 m^2 (size of sampling cage) by ten samplings per region and compared with soil nutrient values per region using ANOVA (proc GLM). Pearson's correlation coefficients were calculated between soil nutrient status, vector populations and perceived yield loss due to MSV. Multiple regression analysis and stepwise procedure was carried out to determine the contribution of each of the nutrients in explaining the fluctuations of the *C. mbila* populations and perceived yield loss due to MSV.

RESULTS

Leafhopper population in relation to altitude

The leafhopper species identified from the different regions were *Cicadulina chinai* Ghauri, *Cicadulina bipunctata* (Melchar), *Cicadulina storeyi* China and *C. mbila* (Naudé) (Table 2). The later was present in all the six regions. In addition, the high altitude regions recorded low numbers of leafhoppers. Regions with elevations of below 1550 m a.s.l (Bahari, Mwea, Muhoroni and Oyani respectively), recorded significantly higher *Cicadulina spp.* numbers than those at high altitudes (Kimilili: 1680 and Githunguri: 1850 m a.s.l) (p = 0.021, SNK). However, for *C. mbila* alone, the numbers were significantly higher at altitudes of below 1380 m a.s.l (p = 0.006, SNK) (Table 2).

The mean number of *Cicadulina* individuals recorded from ten trapping cages was 15.0 in Bahari (of which 39% were *C. mbila*), 15.1 (40%) in Mwea, 13.6 (43%) in Muhoroni, 16.5 (18%) in Oyani, 7.8 (23%) in Kimilili and 6.6 (35%) in Githunguri regions (Table 3). However, the from ten trapping cages was 15.0 in Bahari (of which 39% were *C. mbila*), 15.1 (40%) in Mwea, 13.6 (43%) in

Table 2. The number of leafhopper species sampled at different altitudes of Kenya.

Altitude (m a.s.l)	*Cicadulina* species present	Number of other *Cicadulina spp.* sampled	Number of *C. mbila* sampled alone
80	*C. mbila, C. bipunctata, C. storey*	150 a	59 a
1350	*C. mbila, C. chinai*, *C. storeyi*	151 a	60 a
1380	*C. mbila, C. bipunctata, C. storeyi, C. chinai*	136 a	58 a
1550	*C. mbila, C. bipunctata, C. storeyi, C. chinai*	165 a	29 b
1680	*C. mbila*	76 b	18 b
1850	*C. mbila, C. storeyi, C. chinai*	66 b	23 b

Means in columns followed by the same letter do not differ significantly ($p < 0.05$) according to SNK test.

Table 3. The number of *Cicadulina* species sampled from six selected regions of Kenya

	Total number of different species of *Cicadulina* present	Number of all other species of *Cicadulina* apart from *C. mbila*	Number of *C. mbila* alone
Bahari	209 a	15.0± 3.1 a	5.9± 1.7 a
Mwea	211 a	15.1± 2.4 a	6.0± 1.1 a
Muhoroni	194 a	13.6± 2.1 a	5.8± 1.1 a
Oyani	194 a	16.5± 3.6 a	2.9± 0.8 b
Kimilili	96 b	7.8± 1.0 b	1.8± 0.3 b
Githunguri	86 5b	6.6± 1.4 b	2.3± 0.6 b

Means in columns followed by the same letter do not differ significantly ($p < 0.05$) according to SNK test. Sampling regions are ordered with increasing altitude.

Table 4. Soil nutrient levels (n = 24) in six major maize production regions in Kenya.

Region	Nitrogen (%)	Phosphorus (ppm)	Potassium (ppm)
Bahari	0.12 ± 0.01 b	27.8 ± 4.9 a	137 ± 9.8 b
Mwea	0.23 ± 0.01 a	16.0 ± 4.2 a	412 ± 132.8 ab
Muhoroni	0.23 ± 0.04 a	13.5 ± 0.7 a	707± 182.5 a
Oyani	0.25 ± 0.01 a	37.3 ± 7.4 a	168 ± 9.8 b
Kimilili	0.24 ± 0.01 a	18.5 ± 3.3 a	86± 22.4 b
Githunguri	0.29 ± 0.01 a	24.8 ± 8.4 a	407± 63.2 ab

Legend: N; Low, 0.06-0.12; moderate, 0.13 - 0.25; high, >0.26. P; Low, < 30; Moderate, 31 - 45; High, > 46. K; Low, 1 - 50; Moderate, 51 - 100; High, 101 - 175; Very high, >301. Mean values in columns followed by the same letter do not differ significantly ($p > 0.05$) according to SNK test. Sampling regions are ordered with increasing altitude.

Muhoroni, 16.5 (18%) in Oyani, 7.8 (23%) in Kimilili and 6.6 (35%) in Githunguri regions (Table 3). However, the total number of trapped *Cicadulina spp.* decreased with increasing altitude (Table 2). Nevertheless, the percent number of *C. mbila* and altitude were not correlated ($r = -0.393$; $p = 0.441$).

Soil analysis

The results showed that N and P levels were inadequate in all regions. All the sampled regions had low levels of nitrogen (< 0.3%); with Bahari, significantly recording the lowest N levels (Table 4) . Conversely, the levels of P did not differ among regions (Table 4). However, the levels of K differed significantly in regions; Muhoroni region recorded the highest levels of K.

Perceived yield loss due to MSV

The maize yield losses due to MSV in Oyani (46%) and Mwea (43%) were significantly different from the losses in Muhoroni (25%), Bahari (28%), Kimilili (28%) and Githunguri (29%) ($p = 0.002$) (Figure 1).

Table 5. Correlation matrix for *Cicadulina spp, C. mbila,* farmers' perception on MSV yields loss (PERCMSV), nitrogen, phosphorus and potassium in six sites in Kenya.

	***Cicadulina spp.* (n = 60)**	***C. mbila* (n = 60)**	**PERCMSV (n = 60)**	**Nitrogen (n = 24)**	**Phosphorus (n =24)**	**Potassium (n =24)**
Cicadulina spp	X					
C. mbila	0.646**	X				
PERCMSV	0.154	0.295*	X			
Nitrogen	-0.414*	-0.365	-0.187	X		
Phosphorus	0.039	-0.224	-0.071	-0.046	X	
Potassium	0.172	0.655**	0.337	0.002	-0.339	X

Asterisks indicate significance levels of correlation coefficients: *= p 0.05; **= p 0.01.

Correlation of perceived yield loss, *C. mbila* numbers and soil nutrient levels

Perceived yield loss due to MSV correlated significantly to the number of *C. mbila* (r = 0.295, p = 0.022) (Table 5). Moreover, K correlated significantly to the number of *C. mbila* (r = 0.655, p = 0.01). In general, fields that exhibited low K levels (Oyani and Bahari) had the highest numbers of *C. mbila*. Conversely, a negative correlation existed between the levels of N and species of *Cicadulina* (r = -0.414, p = 0.044), *C. mbila* (r = -0.365, p = 0.079) (Table 5).

Stepwise regression showed that N and P as separate elements were not significantly correlated to the number of *C. mbila*. However, cumulatively, all nutrient variables significantly influenced *C. mbila* numbers (F = 56.107, p = 0.001) and the perception of farmers on yield loss due to MSV (F = 27.594, p = 0.001) (Table 6) . Some nutrient variables had positive or negative partial contributions to the overall influences on either perceived yield loss or *C. mbila* numbers (Table 6). On the other hand, *C. mbila* numbers significantly correlated positively with yield loss due to MSV.

DISCUSSION

The occurrence of *C. mbila* in all the six agro-ecological regions is consistent with studies that reported its presence in all the maize growing regions (Downham et al., 1997; Bosque-Pérez, 2000; Alegbejo et al., 2002). Other leafhoppers species identified included *Cicadulina chinai, Cicadulina bipunctata* and *Cicadulina storeyi,* confirming the occurrence of the first three species in regions of Kenya (Wambugu and Wafula, 2000). This study is the first to confirm *C. bipunctata* individuals in the coastal region and *C. chinai* in the central and eastern regions of the country, indicating that these species have a wider range of distribution than previously reported (Okoth et al., 1987; Odhiambo et al., 2000; Wambugu and Wafula, 2000). Further studies on the biology of the two species and an assessment of the factors that influence their efficiency to transmit MSV are proposed.

Leafhoppers (*Cicadulina spp*.) were in abundance at lower altitudes compared to those above 1550 meters, above sea level, except for the number of *C. mbila* that did not correlate with altitude. The proportion of *C. mbila* out of all the other MSV vector species was consistently high in all the regions, which is consistent with reports by Mesfin et al. (1995), Bosque-Pérez (2000), Smith et al. (2000) and Lett et al. (2002). The authors propose further studies to determine the probable presence of biotypes in populations of *C. mbila*, which could explain the ability of this species to adapt to the different altitudes.

The classes of soils in the six regions were oxisols (found in Bahari and Kimilili regions), ultisols (Githunguri, Kimilili and Muhoroni) and alfisols: Muhoroni (Almanac Characterization Tool v3.0, 2001). The soil analysis showed that the levels of nitrogen and phosphorus elements in the six regions were inadequate for crop growth, except for potassium. These results are consistent with the characterization of African oxisols, ultisols and alfisols as soil types with low total and available phosphorus content but adequate potassium (Sanchez and Uehara, 1980; Kwabiah et al., 2003).

The results of this study indicate the existence of a positive relationship between potassium element and the population of *C. mbila* at a regional scale. In addition, a positive relationship existed between *C. mbila* and the yield loss attributed to MSV, which is consistent with a previous study on the coincidence of MSV with numbers of *Cicadulina spp.* (Alegbejo and Banwo, 2005). Failure to detect correlation between *C. mbila* and the effects of MSV disease in a study by Welz et al. (1998), could be due to the presence of region-specific strains of the MSV, that show difference in disease aggressiveness. Potassium indirectly affects vector population numbers by modifying the magnitude of plant protein synthesis (Myers and Gratton, 2006). It is therefore probable that the levels of nutritional constituents in host plant tissues could indirectly influence the population dynamics of leafhoppers and thus affect transmission of MSV disease. A previous study had attributed the ability of *C. mbila* to transmit MSV on the vectors' capacity to select a plant with the appropriate nutrient constituents (Alegbejo et al., 2002). Additionally, the nutritive quality of host plants

Table 6. Results of stepwise regression analysis using *Cicadulina spp* (C), *C. mbila* counts (*mbila*), perceived yield loss due to MSV by farmers ($MSV_{Percep.}$) and soil nutrient values (N = nitrogen; P = phosphorus; K = potassium).

Parameter	Model		Partial R	Model R^2	F	p
		Cicadulina spp.	0.646	0.417	41.548	0.001**
		MSV_{Percep}	0.295	0.087	5.547	0.022*
C. mbila	*mbila* = - 2.275+0.355C+0.057MSV-4.561N-0.028+0.006K	Nitrogen	-0.365	0.133	3.386	0.079
		Phosphorus	-0.224	0.050	1.163	0.292
		Potassium	0.655	0.429	16.496	0.001**
		Total Model			56.107	0.001**
		Cicadulina spp.	0.154	0.024	1..407	0.240
		C. mbila	0.295	0.087	5.547	0.022*
Perceived yield loss due to MSV	MSV_{Percep} = 38.260-3.775C +10.799m +23.622N+0.334P-0.055K	Nitrogen	-0.187	0.035	0.800	0.381
		Phosphorus	-0.071	0.005	0.111	0.743
		Potassium	0.337	0.113	2.814	0.108
		Total Model			27.594	0.001**

Complete models considered for *C. mbila* and MSV including all variables; thereafter, variables that did not show any significant cumulative effect were considered alone. Partial *R* provides each variable's contribution, while R^2 is the proportion of explained variation: * Significant ($p < 0.05$).

plants may be related to the developmental performance of leafhoppers and therefore indirectly influence vector population outbreaks.

Farmers in regions with low levels of phosphorus experienced a high percentage of yield loss due to MSV. It is probable that the low levels of soil phosphorus influenced the prevalence of MSV disease in the studied regions. Chaboussou (2004) reported that phosphorus acts against viral diseases by promoting plant maturity, thus restricting the pathological effects of the virus. The findings of this study indicate that host nutrition may play an important role in the assessment of yield losses attributable to the MSV disease under Kenyan environments. The authors propose further empirical studies to determine the appropriate levels of phosphorus to lessen incidence and severity of MSV. In recent past, farmers have been encouraged to apply chemical fertilizers for increased maize yields (Sanchez, 2002; Yardim and Edwards, 2003; Okori et al., 2004). However, if this trend continues, the soil macronutrients will have an unpredictable impact on MSV disease and its vectors. Assessment of this impact will depend on further research on crop loss under various nutritional regimes. In summary, the findings of this study highlight the significance of webs of associations between insect / disease pests and conditions of soil nutrient balance. Similar observations indicate that the acceptance and colonization of plants by insect pests is dependent on the type of soil and the fertilizers applied (Altieri and Nicholls, 2003; 2005). There is need to explore the potential of combining the current MSV control methods with proven rates of soil macronutrients, as packages of integra- ted pest management (IPM) for MSV disease. In conclusion, this study confirms that the pathological manifestation of MSV is associated to the different leafhopper vector species, altitudes and levels of soil macronutrients.

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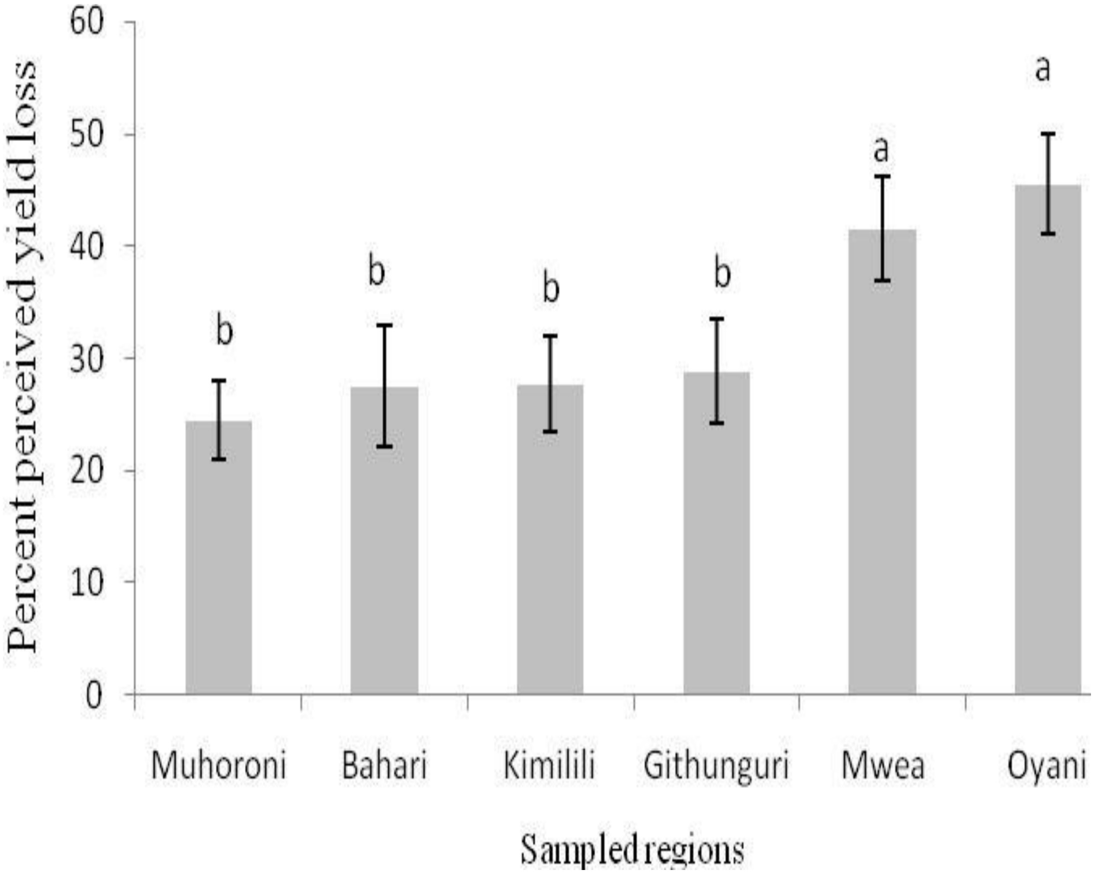

Figure 1. Mean percent of yield loss expected by farmers due to maize streak disease with standard error at 95% confidence limits, in six maize growing regions in Kenya in 2005 and 2006. Different letters on top of bars indicate that mean values are significantly different at p 0.01 as determined by SNK test (n = 30).

extension agents and farmers in all the surveyed areas.

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