Screening of Napier Grass (*Pennisetum purpureum* Schumac.) Clones for Stunt Disease in Western Kenya

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Abstract
Napier stunt disease (NSD) is a threat to Napier grass farming in the smallholder dairy industry in east and central Africa. NSD in Kenya is caused by *Candidatus* Phytoplasma oryzae (Ns-phytoplasma) belonging to the 16SrXI group; vectored by a leaf hopper *Maiestas banda*. The objective of the study was to determine the incidence (proportion of diseased leaves) and severity (proportion of leaf area diseased [PLAD]) of stunt disease in Napier clones by screening in Bungoma, Kakamega and Busia Counties in Western Kenya. The Napier clones were germplasm collections maintained at KALRO Kitale and Muguga nurseries and in farmers' fields. Data were collected from twenty farmers' open fields and eighteen Napier clones were planted in a replicate per farm. A total of 360 (n) observations were done within the Counties in 2013. Five cuts were done and each cut was done every eight weeks at each farm. Low stunt disease severity with mean PLAD of 0.084 (n=126) and low disease incidence with mean of 0.008 was observed in Kakamega county. This showed that there were few new infections in the farmers sites. Low severity with mean PLAD of 0.002 (n=126) and high stunt disease incidence with mean of 0.047 was observed in Bungoma county. Low severity was attributed to soils high in humus. Higher severity with mean PLAD of 0.408 (n=108) and high incidence with mean of 0.101 was observed in Busia county with songor and the farmer clones having high disease Incidence. The severity and incidence were attributed to a high exchange rate of the planting materials within Busia County and across border exchange. The Napier clones with the lowest incidences and severity indices after five cuts can be multiplied using either clonal or micro-propagation methods to supply to farmers.

Introduction

Napier or Elephant grass (*Pennisetum purpureum* Schumach) (Poaceae) is a major fodder crop feed grown by small scale dairy farmers for cut-and-carry in intensive and semi-intensive livestock production system in Kenya and other East African countries (Staal *et al*., 1999 ; Muyekho *et. al*., 2003; Kabirizi *et al.* 2007). Farmers in Eastern Africa use Napier grass to protect soil erosion, trap Maize/Sorghum stem borers and control striga weed through the Push-Pull technology (Midega *et al*. 2012; Khan, *et. al* .2014). Presently, a disease known as Napier grass stunt disease (NSD) caused by a phytoplasma of group 16SrXI, '*Candidatus* Phytoplasma *oryzae'*; the rice yellow dwarf phytoplasma vectored by a leaf hopper *Maiestas banda* (Jones *et al.* 2004, 2007; Nielsen, *et. al*., 2007) and through infected planting materials is a threat to production of Napier grass in Western Kenya (Muyekho *et. al*., 2006). In Eastern Africa many smallholder Dairy farmers have reported overall loss in biomass of up to 100% of their crop to NSD and a significant reduction in milk output that has led to decline in household incomes (Khan *et al*. 2012). The disease symptoms are small chlorotic leaves, proliferation of tillers, and shortening of internodes to the extent that clumps appear very stunted, ultimately resulting in death of the plants (Ajanga 2005;).The current mitigation strategies that include weeding, rouging infected plants, applying organic and inorganic fertilizer, and planting disease free Napier grass cuttings have not been effective in controlling NSD (Orodho, 2006). Breeding of resistant clones may constitute the only defence against NSD that may wipe out Napier grass clones with a narrow genetic base (Farrell et. al. 2004). The collection of germplasm of *Pennisetum purpureum* and its *Pennisetum glaucum* hybrids from African countries and USA, planted at the International Livestock Research Institute (ILRI) in Ethiopia and found at the Kenya Agriculture and Livestock Research Organization (KALRO) Nurseries at Muguga and Kitale Research centers and farmers' local clones, offered the most dependable source of Napier grass materials. The objective of this study was to identify Napier grass clones with resistance by screening for the incidence and severity to NSD in Western Kenya where the disease is prevalent.

Methods and Study Site

The experiments were planted in 2013 in three counties in Western Kenya namely, Bungoma, Kakamega and Busia bordering Uganda. Experiments were planted by 20 farmers groups selected across the Counties with the help of the Agriculture extension County officers. The replicates were six in Bungoma, seven in Kakamega and seven in Busia Counties laid out in open fields. The lead farmer in each of the groups planted eighteen Napier clones (L14 2, Farmer's clone, Okame. T112, ILRI 16815. T 20. KK1, Bana. T 60. T 41. ILRI 16805. KK3. ILRI 16802. Songor. KK2. T 105. T 89, South Africa and Alupe) in their farm in a Randomized complete block design (RCBD) assisted by other members of the group. The plot size was 7m x 3m each with 28 plants spaced at 1 x $1m^2$. Fertilizer (Triple super phosphate –T.S. P) at 60kg $P_20_5ha^{-1}$ was applied at planting and 80kg N ha^{-1} of Calcium Ammonium Nitrate – C.A.N) was applied at top-dressing the first six weeks after planting and after every cut. Harvesting (cut back) was done at an interval of 8 weeks. Disease assessments were performed monthly on 28 Napier plants per plot in each farm. Incidence (I) which is the extent of manifestation or spread of a disease in a population. was assessed on the youngest fully expanded leaves per plant as the proportion of leaf area diseased using a diagrammatic scale with 0.05 steps (0, 0.05, 0.10, 0.15…1.00). Severity (S) was based on visual scoring on the magnitude of the spread of the disease within the affected stools based on percentage spread and calculated as the mean severity (over all 28 plants) using a scale of 0-100% (Ajanga, 2005). Data for all clones, farmers sites and time of score were pooled and variables tested for normality before carrying out a repeated measures Analysis of Variance (ANOVA) in GenStat (12th Edition) Statistical Software (GenStat, 2010). Where significant differences were observed, clone means were separated using Fishers Protected Least Significant Difference (LSD) at 1% level of significance (Table 1). Graphical presentation was done using Excel.

Results

In this study, Sites, Clones and time did have a significant effect on NSD incidence and severity at P = 0.001 (Table 1). The farmers clone and Songor had high NSD incidence manifestation and NSD severity as compared with clones ILRI1681, L14 and South Africa across all the sites. The disease incidence and severity increased over time for all clones as is shown in (Figures 1 and 2) below. Low stunt disease severity with mean PLAD of 0.084 (n=126) and low disease incidence with mean of 0.008 was observed in Kakamega county. This showed that there were few new infections in the farmers sites. Low severity with mean PLAD of 0.001611 (n=126) and high stunt disease incidence with mean of 0.047 was observed in Bungoma county. Low severity was attributed to soils high in humus. Higher severity with mean PLAD of 0.408 (n=108) and high incidence with mean of 0.101 was observed in Busia county. This indicates that new infections in the county is high. This could be attributed to a high exchange rate of the planting materials within the county and more so cross border exchange. The farmer clones and songor had high NSD incidence and NSD severity across the counties and over the time unlike in other clones where infections increased but at a slow rate (Figures 1 and 2). The clones not diseased like ILRI16815, T20, L14 and South Africa could be used as preliminary candidates to create breeding populations and there after selection.

Table 1. Pooled data score for Incidence and Severity of different Napier Clones across the Counties

Napier Clones	**Disease Incidence**	**Disease Severity%**
Songor	0.152[a]	44.500 [a]
KK3	0.045[b]	9.500 [cbd]
Farmer Clone	0.044[b]	24.000 [b]
Bana	0.040[cb]	10.000[cbd]
Okame	0.034[cbd]	17.632[cb]
KK1	0.020[cbd]	9.500 [cbd]
T60	0.020[cbd]	10.000 [cbd]
KK2	0.020[cbd]	11.500 [cbd]
T41	0.020[cbd]	12.750 [cbd]
ILRI1680	0.011[cbd]	11.000 [cbd']

Alupe	0.011^{cbd}	7.500 ^{cd}
T105	0.007^{cbd}	6.000 ^{cd}
T89	0.003^{cd}	10.000 ^{cbd}
T112	0.003^{cd}	7.000 ^{cd}
ILRI1681	0.000^{d}	0.000 ^{d}
T20	0.000^{d}	0.000 ^{d}
L14	0.000^{d}	0.000 ^{d}
South Africa	0.000^{d}	0.000 ^{d}
Means	**0.0033**	**10.60**
LSD (P<.0001)	**0.0382**	**15.245**

* Means with same letter within the column are significantly different by Least Significant Difference Test

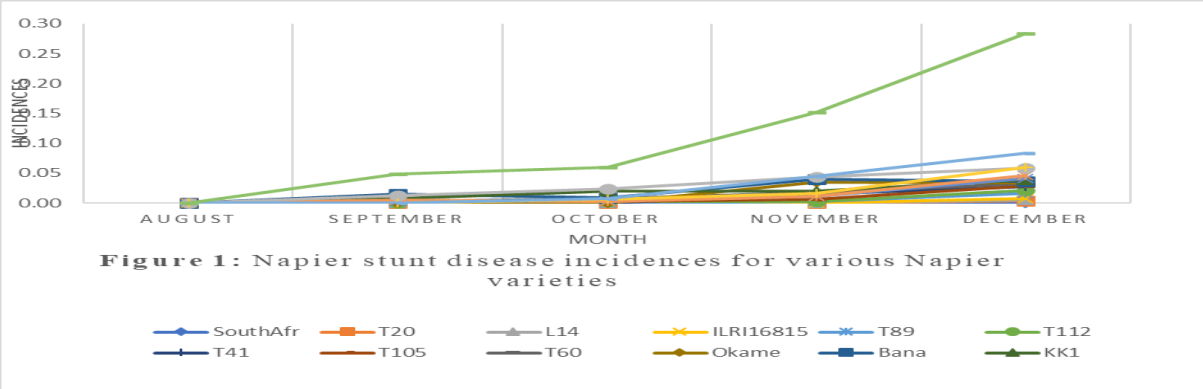

Figure 1: Napier stunt disease incidences for various Napier varieties

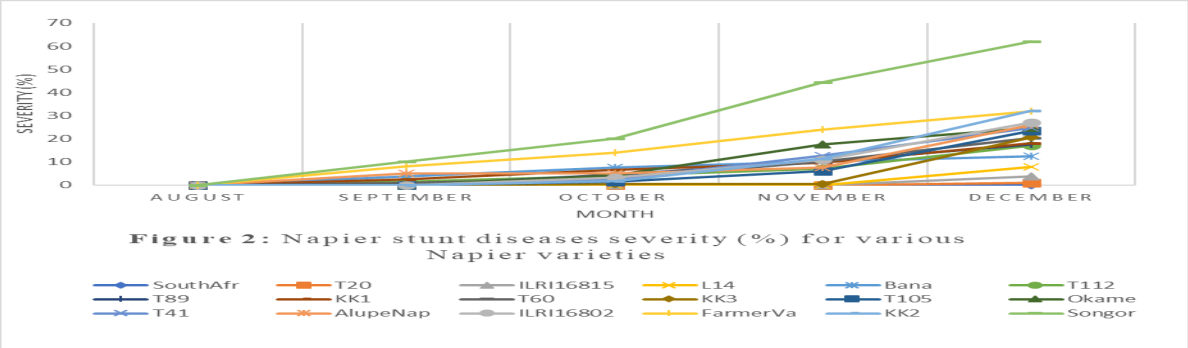

Figure 2: Napier stunt diseases severity (%) for various Napier varieties

Discussion

Although all Napier grass clones succumbed to NSD infection, ILRI16815, T20, L14 and South Africa were not affected across the sites. Kawube *et, al,* 2014 also observed similar trends of NSD infections on Napier clones except for clone ILRI 16837. The use of clean planting Napier grass cuttings of a clone with moderate reaction to NSD combined in an integrated manner with measures that prevent the insect vectors infesting the grass, would provide an interim strategy for managing NSD, especially in low NSD pressure areas. The Napier clones that had the lowest incidences and severity indices after five cuts like ILRI1681, T20, L14 and South Africa can be multiplied using either clonal or micro propagation methods to supply to farmers or be recommended for utilization in breeding programs.

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