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RESEARCH ARTICLE

Efficacy of herbicides' combinations in managing weeds and on crop productivity and soil microbial safety in sugarcane fields of Kenya

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ABSTRACT

The efficacy of herbicides combinations in managing annual weed flora and to assess their effect on soil microbes in sugarcane (*Saccharum officinarum L.*) were evaluated during 2018-19 and 2019-20 cropping seasons at the Kenya Agricultural and Livestock Research Organization in Kisumu, Kenya. The sugarcane (variety KEN 83-737) field with natural weed infestation was used for experimentation in a randomized complete block design (RCBD) with four replications. The tested treatments included: post-emergence applications (PoE) of metribuzin 960g/ha; diuron + hexazinone + sulfometuron-methyl 603 + 170 + 14.5 g/ha; trifloxysulfuron-sodium + ametryn 1097 + 27.8 g/ha; diuron + hexazinone 1170 + 330 g/ha; untreated/weedy check, and hand hoeing twice at 30 and 45 days after sugarcane planting (DAP). The weed density, sugarcane tiller numbers, cane height, millable stalks, and cane yield were significantly different (p< 0.05) across the treatments. All herbicides and the hand hoeing twice proved effective for weed control, resulting in higher sugarcane yields. The combination of diuron, hexazinone, and sulfometuron-methyl resulted in the best weed control, albeit with slight phytotoxicity. The herbicides exhibited varying levels of efficacy in weed control, phytotoxic effects on sugarcane, and impacts on microbial composition and cane yield. Diuron + hexazinone 1170 + 330 g/ha PoE recorded the highest net returns amongst the tested treatments.

Keywords: Diuron + hexazinone, Diuron + hexazinone + sulfometuron-methyl, Metribuzin, Phytotoxicity, Sugarcane, trifloxysulfuron-sodium + ametryn, Weed management

INTRODUCTION

Sugarcane (Saccharum officinarum L.) is a globally important agro-industrial crop (Singh et al. 2015). It is a main source of sugar and bio-energy, accounting for over 70% of the raw table sugar supply in the world. In Kenya, it ranks as one of the top six commercial crops alongside tea, cut flowers, vegetables, coffee, and maize. Sugarcane plays a key socio-economic role in the country. It is used as raw material in sugar and ethanol production, the burning of bagasse as an electricity source, and as animal feed, among other uses (Castro et al. 2019).

Weeds affect yields, quality, harvesting, and sugarcane processing, resulting in huge yield losses (Castro *et al.* 2019, Mandal *et al.* 2020, Patel *et al.* 2024). Weeds in sugarcane production are more problematic than in other crops because sugarcane is planted with relatively wider spacing, and the crop has a relatively slow growth at the initial stages with

30 days to germinate and 60 to 75 days to develop a full canopy (Anusha and Rana 2016). The loss to sugarcane due to weed competition, combined with the cost of weed management, runs into millions (Barceló and Cruz 2015). The losses could be due to competition or indirectly caused by reduced quality, increased costs during operations, such as harvesting and land preparation, or may harbour insect pests and diseases (Rono *et al.* 2015). Weeds are heavy feeders and extract a high amount of nutrients from the soil, while others, such as the morning glory (*Ipomoea purpurea*), twine around the cane stalks, bending and damaging their tops, resulting in a 20-25% loss (Rono *et al.* 2015).

The use of herbicides to control weeds in sugarcane has been recommended as an alternative to hoeing due to their efficacy and as a cheaper alternative (Castro *et al.* 2019). Both pre- and postemergence herbicides have been recommended for weed control in sugarcane farming in Kenya. However, these herbicides control specific weed species and may affect non-target soil microorganisms as well as have phytotoxic effects on the sugarcane crop. To be more effective, herbicide mixtures that have both additive and synergistic

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effects are available. However, herbicides' efficacy, their effect on non-target microorganisms and arthropods, and their levels of phytotoxicity on sugarcane are major concerns. Thus, the current study was conducted to evaluate the efficacy of various herbicide combinations in managing weeds and increasing sugarcane yield, as well as to assess their phytotoxicity on sugarcane and their effect on soil microbes.

MATERIALS AND METHODS

A two-year field study was conducted during the long rain cropping seasons of 2018-19 and 2019-20 at the Kenya Agricultural and Livestock Research Organization- Sugar Research Institute (KALRO-SRI) in Kibos, Kisumu County, Kenya. The site is situated at an elevation of 1,250 m above sea level (0°21'01.0"S 34°49'17.0"E), with a mean annual temperature of 19.7°C and average annual precipitation of 1,900 mm. The average soil pH of the field was 5.3, with organic carbon of 1.24%, and nitrogen of 0.15%

The experiment was laid out in a randomized complete block design (RCBD) with four replications. The experiment consisted of six treatments, *viz.* post-emergence application (PoE) of metribuzin 960 g/ha, diuron + hexazinone + sulfometuron-methyl 603 + 170 + 14.5 g/ha, trifloxysulfuron-sodium + ametryn 1097 + 27.8 g/ha, diuron + hexazinone 1170 + 330 g/ha, untreated/ weedy check, and hand hoeing twice at 30 days and 45 days after sugarcane planting (DAP).

Clean seed cane material of variety KEN 83-737, acquired from the KALRO-SRI farm at Kibos was planted into 5-meter-long furrows spaced 1.2 metres apart. The setts were planted end-to-end in the furrows, thus translating to a seed rate of 7 t/ha. Diammonium phosphate (DAP) (18:46:0) fertilizer was basally applied at the time of planting at the rate of 100 kg/ha, and topdressing was done at 5 months of age using Calcium Ammonium Nitrate (CAN) at 100 kg/ha.

The herbicides were applied using a handoperated Jacto HD 550 knapsack sprayer, calibrated to deliver 400 L/ ha of water with an effective spray swath of 2m, and fitted with a flat fan nozzle. Spraying was done at 30 DAP (when weeds were at the four to six-leaf stage).

Weed counts were established just before spraying and at 7, 14, and 30 days after herbicide application (DAA) to establish the efficacy. Weed species counts were done in four randomly placed 0.25 m² quadrats. Weeds were grouped into three

categories, *i.e.* broad-leaved, grasses or sedges, and weed counts are expressed as weed density (no./m²).

In the 2019/2020 testing year, soil samples (0-15 cm depth) from each experimental plot were randomly collected using a trowel and later mixed thoroughly to make a composite representative sample for fungal and bacterial populations enumeration. The samples were collected before spraying, at 7, 14, 21, and 60 DAA, and during harvest. A sub-sample of approximately 150 grams per sample was placed in a freezer at 4°C until microbial analysis (bacteria and fungi) was conducted. Enumeration of microbes was done on agar plates following the serial dilution technique and pour plate method (Koch et al. 2014). The bacteria were analyzed in nutrient agar, whereas fungi were analyzed on Rose Bengal agar media with streptomycin (Singh et al. 2017) and expressed as colony-forming units/gram (cfu/g).

At harvest, the number of millable stalks in the net plot (2 inner rows) per treatment was counted and expressed as numbers per ha by extrapolation. Cane yield per treatment was determined by weighing all millable stalks per plot using a salter scale and extrapolating to kg/ha. The % yield change was calculated by comparing the yield per treatment to the yield of the weedy check.

Five stalks were randomly selected per plot, and the height of each stalk (cm) from the ground to the dewlap leaf was measured using a tape measure. On the same stalks, the number of internodes per stalk was determined by counting, and total sugars (brix) were measured using a handheld refractometer to estimate the effect on sucrose content at harvest.

The phytotoxic effects of the herbicide treatments were assessed through visual observation of symptoms, including chlorosis (yellowing), stunting, leaf scorching, and epinasty. Evaluations were conducted at appropriate intervals using a visual rating scale ranging from 0 to 100%, where 0% indicated no visible phytotoxic symptoms, and 100% represented complete plant death (Castro *et al.* 2019). The net returns were calculated by subtracting the varying costs of production from the gross returns (average yield in the two seasons and prevailing price per ton).

The data on the weed density was transformed into Log (2 + value) before analysis. The data was then subjected to analysis of variance by t-test at 5% probability using the Statistical Analysis Software (SAS Version 9.4). The means were then compared and separated using Fisher's least significant difference (LSD) at p=0.05.

RESULTS AND DISCUSSION

Effect on weeds

The major weed flora observed in the experimenta field were: grasses; *Panicum* spp., *Setaria* spp., *Rhottboelia exaltata*, and *Digitaria* spp. constituting 12%, 8%, 5%, and 2%, respectively. Of the total weeds recorded; broad-leaved weeds (BLWs) were: *Ageratum conyzoides* (15%), *Bidens pilosa* (21%), *Comellina benghalenis* (7%), *Euphoria hirta* (5%), *Galinsoga parviflora* (3%), *Amaranthus* spp. (2%) and *Datura stramonium* (2%). *Cyperus esculentus* was the only sedge with 23% relative density. Broad-leaved weeds constituted over 46% of the total weed density. Similar dominance of broad-leaved weeds as the most predominant in sugarcane was reported earlier (Rasker 2004).

The treatments had a significant (p=0.05) effect on the BLWs, grasses and sedges in the three years (Table 1). Hand hoeing had the best weed control (100%) in the three seasons. The highest BLW density was recorded in the weedy check. Among the herbicides, a combination of diuron + hexazinone + sulfometuron-methyl 603 + 170 + 330 g/ha PoE caused the best control of broad-leaved weeds. On the other hand, the diuron + hexazinone + sulfometuron-methyl 603 + 170 + 14.5 g/ha PoE, trifloxysulfuron-sodium + ametryn 1097.3 + 27.8 g/ ha PoE and diuron + hexazinone applied 1170 + 330 g/ ha PoE, significantly reduced the grassy weed flora. Only hoeing gave appreciable control of sedges. Herbicide mixtures have been known to perform better than single-molecule herbicides, though sometimes expensive (Barceló and Cruz 2015). In a trial in Egypt, post-emergence herbicides containing triclopyr and clomazone, and hand hoeing at 30 and

45 DAP had a significant effect on weeds in comparison to the untreated control (Mohamed and Marzouk 2021).

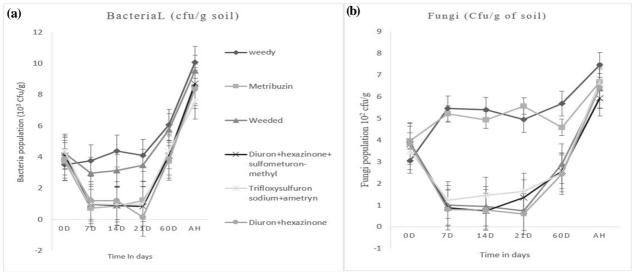
Bacterial population: Different herbicide treatments had no significant effect on the bacterial populations in the soil as reported earlier by Singh *et al.* (2017). However, the weedy check and the plot with hoeing twice treatment had higher bacterial colony-forming units than the herbicide-treated plots. From the 21st day after treatment, all the bacterial populations increased to peaks of 7.5 to 9.52 x10³cfu/g of soil from diuron + hexazinone and the weedy check, respectively (**Figure 1a**).

Fungal population: Fungal counts drastically declined within the first seven days after treatment, and picked up gradually from the fourteenth day. The fungi in the hoeing twice and weedy check treatments were significantly different (P=0.05) from the herbicide-treated plots from the 7th to 60th day after treatment but was not different at harvesting (Figure **1b).** This concurs with findings by Singh *et al*. (2017) who reported a decline in the fungal population in India after the use of halosulfuron + metribuzin at different doses. The interaction between herbicides-cultivars and season influenced rhizospheric soil variables in Brazil's sugarcane (Faria et al. 2018). Microorganisms were stressed (low respiratory levels) when diuron was used at high concentrations, but this did not happen when lower levels of diuron mixed with hexazinone were used (Faria et al. 2018). According to Da Silva et al. (2014), sugarcane varieties vary in their capacity to associate with soil microorganisms, leading to varied responses of the microbes to the herbicides.

 $\label{thm:conditional} \textbf{Table 1. Efficacy of different weed management treatments on density (no./m^2) of broad-leaved weeds (BLWs), grasses and the sedge in the two seasons}$

Treatment	Rate (kg or l/ha	Weed density (log2 + value/m²)							
	formulated	BL	Ws	Gra	sses	Sedge			
	product)	2018-19	2019-20	2018-19	2019-20	2018-19	2019-20		
Metribuzin 960 g/ha	2.0	0.9(5.8)	0.8(4.8)	0.6(2.3)	0.5(1.2)	1.9(74.2)	1.5(30.2)		
Diuron + hexazinone + sulfometuron- methyl 603 + 170 + 14.5 g/ha	1.0	0.4(0.6)	0.4(0.5)	0.4(0.5)	0.4(0.5)	1.8(68)	1.2(14.0)		
Trifloxysulfuron-sodium + ametryn 1097.3 + 27.8 g/ha	1.5	0.6(2.3)	0.7(3.1)	0.5(1.5)	0.4(1.5)	1.7(50.3)	1.4(32.0)		
Diuron + hexazinone1 170 + 330 g/ha	2.5	0.6(2.0)	0.6(1.9)	0.5(1.5)	0.4(0.5)	1.9(80.2)	1.3(21.2)		
Weedy		2.1(130.5)	0.9(6.3)	0.6(2.4)	0.6(2.3)	2.1(110.9)	1.6(40.6)		
Hand hoeing twice		0.3(0.0)	0.3(0.0)	0.3(0.0)	0.3(0.0)	0.3(0.0)	0.3(0.0)		
Cv (%)		30.8	40.9	28.3	26.5	9.7	22.9		
LSD (p=0.05)		0.4	0.4	0.2	0.2	0.2	0.4		

LSD: least significant difference at the 5% level of significance, CV: Coefficient of variation. The weed density was log transformed (Log2 + value)



D: days, AH: at harvest

Figure 1. Effect of herbicide treatments on (a) bacteria and (b) fungi populations before and at different times after treatment application in the 2019/2020 season

Table 2. Effect of tested weed management treatments on sugarcane quality (brix), growth parameters, yield and net return

Treatment	Brix		No. of Stalks ('000/ha)		Height (cm)		No. of Internodes		Cane yield (t/ha)			Net
	2018- 19	2019- 20	2018- 19	2019- 20	2018- 19	2019- 20	2018- 19	2019- 20	2018- 19	2019 -20	% change	returns (USD/ha)
Metribuzin 960 g/ha	21.6	20.4	89.3	164.5	199.4	277.4	28.5	26.3	55.4	147	12	2419
Diuron + hexazinone + sulfometuron- methyl 603 + 170 + 14.5 g/ha	20.9	20.4	100.0	149.3	210.6	279.3	32.8	26	53.9	146	11	2394
Trifloxysulfuron-sodium + ametryn 1097.3 + 27.8 g/ha	21.2	20.9	131.0	160.3	216.4	286.3	31.0	28.8	60.4	143	13	2451
Diuron + hexazinone 1170 + 330 g/ha	21.8	20.1	110.0	175.3	215.0	288.6	30.8	26	65.9	159	25	2737
Weedy	20.6	20.1	104.0	155.5	200.2	261.6	30.3	27.0	51.0	129	0	2261
Hand hoeing twice	21.6	20.4	103.0	168.3	205.3	258.2	31.3	25.8	57.0	163	22	2442
Cv (%)	3.2	1.7	18.5	16.6	8.7	7.4	7.2	7.1	21.9	18.1	-	-
LSD(p=0.05)	1.0	0.5	29.6	40.6	27.1	30.7	3.3	2.8	8.9	42.6	-	-

LSD: least significant difference at the 5% level of significance, CV: Coefficient of variation

Sugarcane growth parameters and yield

The sugarcane yield attributes varied significantly (p=0.05) across the various treatments, except for the millable stalks in 2019-20 and the cane height during both years of study (**Table 2**). The weeds had a quality and quantity yield loss on sugarcane. Weedy check recorded the lowest cane yields of 51 and 128 t/ha in the 2018-19 and 2019-20 seasons, respectively. The unrestricted growth of weeds in sugarcane at the early stages caused yield losses of up to 22%. Similarly, the lowest brix (total sugars) was recorded in the weedy check.

All the treatments had an increase in yield when compared to the weedy check. In the first season, the highest yield was recorded with diuron + hexazinone, whereas in the second season, the highest yields was with hand hoeing twice. Diuron + hexazinone treatment had the highest percent yield increase of

25%, followed by hand hoeing twice with a 22% yield increase. The effect of weeds on sugarcane yields was attributed to competition for moisture, nutrients, and light during growth (Barceló and Cruz 2015, Anusha and Rana 2016). The higher yields observed are attributed to decreased weed biomass, leading to improved plant growth and sugarcane yields (Singh *et al.* 2015, Ali *et al.* 2018). The highest net returns (US \$ 2737/ha or 410,600 shillings/ha) were recorded with diuron + hexazinone 1170 + 330 g/ha PoE, while the least (US \$ 2261/ha or 339,166 shillings/ha) was recorded with weedy check.

Herbicide phytotoxicity on sugarcane

There were no major phytotoxic effects in terms of scorching, necrosis, hyponasty, or epinasty due to tested herbicides. However, moderate to slight chlorosis and stunting were noted with diuron + hexazinone + sulfometuron-methyl and diuron +

hexazinone at 7 DAA, which recovered fully by the 60th day after application (data not presented in this paper). A similar response was observed with diuron + hexazinone treatment in Brazil, but the effect varied across varieties (Castro et al. 2019). Cultivars exhibited differential susceptibility to varying doses of ametryn + trifloxysulfuron-sodium. Cultivar RB855113 had the highest phytotoxicity 28 days after herbicide application (Ferreira et al. 2005; Da Silva et al. 2014). Trifloxysulfuron-sodium was more tolerated by most varieties as compared to ametryn and its combinations. However, higher doses of the trifloxysulfuron-sodium enhanced the growth of sugarcane (Da Silva et al. 2014). A similar effect to that from diuron + hexazinone + sulfometuronmethyl was reported in sugarcane fields treated with sulfometuron-methyl (Assis et al. 2018).

It is concluded that the highest sugarcane yield and net returns were recorded under the diuron + hexazinone 1170 + 330 g/ha PoE and hence can be recommended for managing weeds and realizing higher productivity of sugarcane in Kenya.

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