REFEREED PAPER

SOIL MICROBIAL BIOMASS IN SUGARCANE CROPPING SYSTEMS OF SOYBEANS, SUNN HEMP AND VELVET BEANS DURING FALLOWING FOR CONTROL OF RATOON STUNTING DISEASE

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Abstract

Most growers in Zimbabwe consider leaving sugarcane fields fallow for 90 days to control Ratoon Stunting Disease (RSD) to be wasteful and uneconomic. A long-term study was established in 2009 to determine the potential benefits of growing legumes during the 90 day RSD fallow period.Five sugarcane cropping systems(SCSs) tested were: (i) sugarcane monoculture (SM), (ii) 90 days of rsd fallow followed by sugarcane (90DFS), (iii) soybeans followed by sugarcane (SBS), (iv) sunn hemp green crop followed by sugarcane (SHGCS), and (v) velvet beans green crop followed by sugarcane (VBGCS). This report is on quantitative changes of soil microbial biomass among the five SCSs and on soil organic matter (SOM).

Soil cores were collected at 0-30 cm and 31-60 cm depths.Initial soil samples were collected before planting any crops.Thereafter, soils were sampled when legumes flowered and were ploughed under,and again 24 to 48 hours after the first irrigation following the harvest of sugarcane plant crops.Soil samples were analysed for colony forming units (CFUs) using the dilution and plating methods.

Legume crops improved soil microbial densities. SBS, SHGCS, and VBGCS increased CFUs in the top 0-30 cm soil after harvesting sugarcane plant crops by 6.7, 40.1 and 67.4% respectively, compared to crops following 90 day fallows. There were significantly more bacterial than fungal CFUs, and the CFUs were more abundant at 0-30 than at 31-60 cm soil depths.The increased CFUsat the harvest of sugarcane plant crops were not sustained in the subsequent ratoon crop.

*Keywords:*fallow, legume, microbial biomass, soil, sugarcane

Introduction

In Zimbabwe, sugarcane (*Saccharumofficinarum*L.) is grown under irrigated conditions and the crop is ready for harvesting after 14 months in the plant crop, and 12 months in ratoon crops. Ratoon Stunting Disease (RSD) is the most important disease in reducing cane yields worldwide (Lentini and Comstock, 1998). *Leifsoniaxyli* subsp. *xyli*, the bacterium that causes RSD, only survives in live roots and cane stalks. RSD is spread through planting diseased seedcane, infected cane cutting knives and mechanical harvesting, as well as by movement of people and equipment within and between cane fields (Hoy and Flynn, 2001; McFarlane,

2002). Sugarcane monoculture production systems exacerbate the disease situation (Hoy and Flynn, 2001) and also resulted in yield decline (Garside and Bell, 2007).

RSD is a common and economically important disease in most sugarcane producing countries (Bailey and Tough, 1992) including Zimbabwe (Zvoutete, 2004). The bacterium colonises the vascular bundles of the sugarcane plant, causing stunting in the growth of cane (Rozeff, 1999). During 2006, theRSD infection rate was 18.6% in sugarcane variety N14 and 0.25% in CP72-2086 in Zimbabwe (Anon, 2006). TheRSD infection rate had declined to 16.6% in variety N14 by 2009.There were no statistics for variety CP72-2086 because it has almost been phased out of commercial production (Anon, 2009). Variety N14 is very susceptible to RSD, whereas variety CP72-2086 is resistant. Despite this susceptibility, sugarcane growers still prefer N14 because it has a greater yield potential in Zimbabwe.

Sugarcane growers worldwide control RSD by practising one or a combination of good sanitation, treating seedcane setts with hot water at 50°C for two hours, or dipping of cane knives in, and spraying implements with, chemicals such as sodium hypochlorite (Jeyes Fluid) (McFarlane, 2002). These methods are not very effective without complete destruction of sugarcane stools during ploughing, and leaving fields free of sugarcane volunteers for a certain length of time. The RSD pathogen remains live in cane volunteers and subsequently infects new crops. As a result, growers of sugarcane worldwide leave cane fields fallow for variable periods after plough-out. The practice of leaving cane lands fallow starves the bacteria and breaks its life cycle.

In Zimbabwe, most growers consider fallowing of cane fields for 90 days to control RSD wasteful and uneconomic. Small-scale cane growers are concerned that their individual lands are too small to leave some to fallow for 90 days.In contrast, the estates, which ploughout approximately 10-12% of their land annually, argue against practising RSD fallowing because this results in too much land being left idle for too long. Thus both small-scale cane growers and estates often avoid the 90 day fallow because of the opportunity cost of time for cane growth.Moreover, there is no income accruing to the cane grower during the fallow periods.

Several low input break crops such as cowpeas, peanuts, sunn hemp, soybeans, oats, lupins and pastures planted during the RSD fallows in Australia, South Africa, Colombia, Mauritius, Papua New Guinea and Zimbabwe improved the productive capacity of soils (Anon, 2000; Pankhurst *et al*., 2000, 2003; Olivier, 2004). The break crops helped to improve soil fertility, reduced harmful insects and diseases, improved sugarcane yields and also provided income to the sugarcane growers (Pankhurst *etal*., 2000, 2003; Nixon and Simmonds, 2004).In Australia, breaking sugarcane monoculture with another crop species improved sugarcane yields by 20-30% in the plant crop, due primarily to improvement in soil health (Pankhurst, *et al*., 2000; Garside and Bell, 2007).

It is commonly accepted that legumes have considerable potential to contribute to soil fertility and sustainable crop yields.Legumes improve soil microbial biomass, which is the living portion of soil organic matter, excluding plant roots and soil animals larger than 5×10^{-3} μm^3 (Jenkinson and Ladd, 1981). Microbial biomassis the centre of the majority of biological activity in the soil.

The general objectives of this multidisciplinary study were to quantify the changes in soil nutrient elements, microbes and nematodes, and to assess the economic benefits of practicing

RSD fallow, or planting soybeans (*Glycine max*), sunnhemp (*Crotalaria juncea* L.), and velvet beans (*Mucunapruriens*) break crops during the RSD fallows in sugarcane cropping systems.

The objectives of the plant pathology component of the study was to quantify changes in soil microbial biomass as indicators of soil health resulting from planting legumes as break crops during RSD fallows.

Materials and Methods

Site of experiment

The study wasconducted inField Z4, Zebra Block,at the Zimbabwe Sugar Association Experiment Station (ZSAES), which is 430 m above sea level, latitude 21°02'13.18"S, and longitude 31°36'56"E. The soils are sandy clay loams with an average of 20.6% clay and 0.74% organic matter. Average annual rainfall is 625 mm per annum, falling predominantly during the hot summer months of October to March. Potential evaporation is approximately 1800 mm/annum.Absolute maximum air temperatures rangefrom 34-36°C in summer, and the absolute minimum air temperature is about 11°C in winter. Although Field Z4 had been previously cropped with sugarcane, it had been fallow for a year at the time of the experiment.

Land preparation and soil sampling

The field wascross-ripped at right angles and then ploughed to approximately 25 cm using a disc plough.The field was divided into four equal blocks, each approximately 4500 m^2, and each block marked into 20 plots of 225 m^2. An earth irrigation canal was built running east-west and dividing Field Z4 into two equal portions to facilitate the application of water.Two replicates were on either side of the canal.Each plot was30 m long and 7.5 m wide.

Furrows to plant soybeans, sunn hemp and velvet beans were set at 0.75, 1.0 and 0.45 m, respectively.Fallow plots were not ridged until planting was due, but were kept free of weeds and volunteer cane for 90 days.

Dutch augers were used to collect soil samples from individual plots before planting of any crops and again immediately after harvesting of each crop, but prior to planting the next crop. Samples were collected from five positions within a plot to depths of 0-30 and 31-60 cm. A 1.0 m buffer was left around each plot.

The soil samples were analysed at the ZSAES Chemistry and Soils Laboratory using standard soil analysis methods. The pH of the soil samples was analysed using the $CaCl_2$ method, and conductivity was determined using an inoLab Conductivity Meter. Available nitrogen was extracted using acidified 1 M KCl followed by distillation on a micro Kjeldahl before titration. Organic matter was determined as organic C using a Leco CN analyser and converted to organic matter by multiplying organic C by 1.72.

Planting and management of legume crops

Soybeans variety Storm, sunn hemp (variety unknown), and velvet beans (variety unknown) were planted by hand at depths of 0.05 m using a seed rate of 80 kg/ha. The soybean seeds were inoculated with 200 g rhizobium/50 kg seed before planting. No fertilisers were applied to the legume crops. Amixture of Commando (63 mL) and Lasso (286 mL) in 15 L water was sprayed at planting to control weeds.

Sunn hemp and velvet bean plants were ploughed under at the flowering stage (when 50% of the plants had flowered) using a disc plough. The plots that had soybeans were also ploughed immediately after harvesting the soybean crop. Soybean was harvested when the pods were dry but before they shattered. The stems were cut at ground level and left to dry in the fields. The stems and the pods were taken out of the field for threshing. Only the root system and some leaves that senesced and dropped were incorporated into the soil during land preparation.

Clods and plant material were disced with a disc harrow.Thereafter, ridges 1.5 m apart and about 0.25 m deep were opened using a ridgerin preparation for the planting of sugarcane.

Experiment design and treatments during the first cropping year
The 20 treatments were arranged in a randomised complete block design replicated four times.The treatment details are provided in Table 1.

Planting and management of sugarcane crops
In each case, sugarcane varieties N14 and CP72-2086 were planted immediately after completing land preparation. Cane knives used in preparing seedcanesetts were disinfected against RSD by dipping in sodium hypochlorite. Seedcanesetts were dipped in Bayfidan to reduce the transmission of fungal disease pathogens. Three-eyed seedcanesetts were planted, double-stick, along each furrow at a depth of 200 mm and covered with 50 mm of soil.

Phosphorus was applied by hand as single superphosphate (SSP, 8% P) at a rate of 50 kg P/ha just before planting. The SSP was incorporated into the soil manually using hoes. Nitrogen was applied as ammonium nitrate (34.5% N) at four and eight weeks after emergence of cane at rates defined in Table 1. Potassium, as muriate of potash (MOP, 50% K) was applied twice at the same times as nitrogen and at a rate of 50 kg K/ha. Ammonium nitrate and MOP were banded on either side of the cane line. A mixture of Lasso (295 mL) and Atrazine (295 mL) in 15 L of water was applied to control weeds in sugarcane. Irrigation was scheduled using evaporation data and was applied at 50% depletion of total available water (TAW) in the 0-70 cm depth.

Sampling and determination of soil microbial biomass
The **s**ampling method was similar to that for nutrient determination. Soil cores were collected using Dutch augers from 0-30 and 31-60 cm, or at 0-10 and 11-20 cm depths. Initial soil samples were collected before planting any crops. Thereafter, soils were sampled (i) after ploughing in of green legume crops but prior to planting sugarcane, and (ii) 24 to 48 hours after the first post-harvest irrigation. The soil samples were sieved through a 2 mm sieve and stored at 4°C before analysis. Samples were analysed for microbial biomass at the Tobacco Research Board's Kutsaga Research Station using the dilution and plating method (Martens, 1995). Diluted solutions were cultured in nutrient agar to determine the number of bacterial colonies, and in malt extract agar to determine colonies of fungi and moulds. Plates with nutrient agar were incubated at 37°C for 12-24 hours before counting the bacteria, while those with malt extract agar were incubated at 25°C for five days before counts were done. Bacterial colonies and counts of fungi/moulds provided the total colony forming units (CFUs) which indicate a soil's microbial biomass.

Table 1.Twenty treatment combinations of sugarcane, legumes and applied nitrogen rates, which were tested in the five cropping sequences during the first year of cropping.

1	Monoculture sugarcane variety N14 planted on 6 May 2009 and fertilised with 60 kg N/ha
2	Monoculture sugarcane variety N14 planted on 6 May 2009 and fertilised with 120 kg N/ha
3	Monoculture sugarcane variety CP72-2086 planted on 6 May 2009 and fertilised with 60 kg N/ha
4	Monoculture sugarcane variety CP72-2086 planted on 6 May 2009 and fertilised with 120 kg N/ha
5	Sugarcane variety N14 planted on 25 August 2009 after 90 days RSD fallow and fertilised with 60 kg N/ha
6	Sugarcane variety N14 planted on 25 August 2009 after 90 days RSD fallow and fertilised with 120 kg N/ha
7	Sugarcane variety CP72-2086 planted on 25 August 2009 after 90 days RSD fallow and fertilised with 60 kg N/ha
8	Sugarcane variety CP72-2086 planted on 25 August 2009 after 90 days RSD fallow and fertilised with 120 kg N/ha
9	Soybeans sown on 6 May 2009, harvested dry and sugarcane variety N14 planted on 6 October 2009 and fertilised with 60 kg N/ha
10	Soybeans sown on 6 May 2009, harvested dry, and sugarcane variety N14 planted on 6 October 2009 and fertilised with 120 kg N/ha
11	Soybeans sown on 6 May 2009, harvested dry and sugarcane variety CP72-2086 planted on 6 October 2009 and fertilised with 60 kg N/ha
12	Soybeans sown on 6 May 2009, harvested dry, and sugarcane variety CP72-2086 planted on 6 October 2009 and fertilised with 120 kg N/ha
13	Sunn hemp sown on 6 May 2009, ploughed under at flowering stage and sugarcane variety N14 planted on 14 July 2009 and fertilised with 60 kg N/ha
14	Sunn hemp sown on 6 May 2009, ploughed under at flowering stage and sugarcane variety N14 planted on 14 July 2009 and fertilised with 120 kg N/ha
15	Sunn hemp sown on 6 May 2009, ploughed under at flowering stage and sugarcane variety CP72-2086 planted on 14 July 2009 and fertilised with 60 kg N/ha
16	Sunn hemp sown on 6 May 2009, ploughed under at flowering stage and sugarcane variety CP72-2086 planted on 14 July 2009 and fertilised with 120 kg N/ha
17	Velvet beans sown on 6 May 2009, ploughed under at flowering stage and sugarcane variety N14 planted on 23 September 2009 and fertilised with 60 kg N/ha
18	Velvet beans sown on 6 May 2009, ploughed under at flowering stage and sugarcane variety N14 planted on 23 September 2009 and fertilised with 120 kg N/ha
19	Velvet beans sown on 6 May 2009, ploughed under at flowering stage and sugarcane variety CP72-2086 planted on 23 September 2009 and fertilised with 60 kg N/ha
20	Velvet beans sown on 6 May 2009, ploughed under at flowering stage and sugarcane variety CP72-2086 planted on 23 September 2009 and fertilised with 120 kg N/ha

Sunn hemp, velvet beans and soybean dry weights

All sunn hemp and velvet bean plants from a predetermined 5 m^2 position were cut at ground level for the determination of plant biomass. The plants were harvested when 50% of the plant stand was flowering.The date and age when 50% of the plants flowered were recorded. In the case of soybeans, ten plants selected at random were harvested by cutting them at ground level when the pods were dry. The plants were partitioned into leaves+petioles, pods and stems.The plant material was dried in an oven at 80°C until constant weights were attained.The dry weights were recorded.

Analyses of RSD bacterium in sugarcane stalks
Assessments of RSD infections were done on ten month old sugarcane crops by randomly sampling ten stalks from each plot and analysing juice from each stalk for RSD bacteria using phase-contrast microscopy.

Statistical analyses
Analyses of variance (ANOVA) of the plant and soil data weredone using SAS Version 7 (SAS Institute Inc. Cary, NC, USA).

Results

The results reported in this paper were for organic matter and microbial biomass (CFUs) before and after growing legume crops,up to and including the harvest of the subsequent sugarcane plant crops. The results for microbial biomass were not separated to account for differences in sugarcane varieties and rates of applied nitrogen.

Changes in soil organic matter content
The soils in Field Z4 had generally very low soil organic matter (SOM) during tests done before growing legumes (Table 2). The SOM was generally greater in the upper 0-30 cm soil depth than 31-60 cm. After ploughing under of the green legumes, average SOM across soil depths increased by 15% in former sunn hemp plots, and by 62% in former velvet bean plots. At the same time, average SOM in sugarcane monocultureplots increased by 42.2% compared to the SOMat planting of sugarcane (Table 2).There were no increases in SOM in plots left fallow for 90 days and among plots planted to soybeans, where above ground plant biomass was not incorporated into the soil.

SOM increased markedly across all the treatments during cropping (Table 2). Variety and nutrition did not significantly influence the accumulation of SOM. Levels of SOM were greater in plots previously planted to legumes and least in the 90 day fallow plots. Contrary to observations soon after harvesting soybeans, %SOM increased significantly in plots that had soybeans.

Table 2.Percentage soil organic matter (SOM) before planting, after incorporating legumes, and after harvesting sugarcane plant and first ratoon crops.

Treatment	Before planting		After legumes		Plant crop sugarcane		1st ratoon sugarcane	
	0-30 cm	31-60 cm	0-30 cm	31-60 cm	0-30 cm	31-60 cm	0-30 cm	31-60 cm
Monoculture	0.62a	0.66	0.97a	0.84a	1.26a	0.99a	1.16a	0.93a
Sunnhemp	0.81b	0.72	0.92a	0.84a	1.31a	1.25b	0.92a	0.84a
Fallow	0.73a	0.56	0.67b	0.62b	1.04b	0.84c	0.61b	0.67b
Velvet bean	0.66a	0.55	0.92a	1.04a	1.38a	1.28b	0.92a	1.04a
Soybean	0.87b	0.75	0.72b	0.81a	1.33a	1.29b	0.89a	0.98a
Means	**0.74**	**0.65**	**0.84**	**0.83**	**1.26**	**1.13**	**0.90**	**0.89**
LSD	**0.07**	**ns**	**0.18**	**0.17**	**0.20**	**0.13**	**0.24**	**0.21**

Means followed by the same letter within a column are not significantly different at $p<0.05$
ns = not significant at $p<0.05$.

Changes in pH, conductivity and soil N

Soil pH averaged 6.25 and soil N was 7 ppm before planting crops into Field Z4. The various treatments had no apparent influence on pH (Table 3). In general, legumes did not result in extra residual soil N after harvesting sugarcane plant crops. However, sunn hemp and velvet bean plots had higher soil N than plots which had sugarcane after 90 day fallows and soybeans (Table 3). The apparent spike in N levels in the sugarcane monoculture plots could not be explained.

Table 3. Soil pH, conductivity and N after harvesting the plant and first ratoon crops of sugarcane.

Treatment	After plant sugarcane			After 1R sugarcane		
	pH $CaCl_2$	Cond. µS/cm	N ppm	pH $CaCl_2$	Cond. µS/cm	N ppm
SM	6.36	55	*	6.64	41a	21a
SHGCS	7.18	45	8	6.44	39a	20a
90DFS	7.19	40	5	7.19	40a	5b
VBGCS	7.94	88	8	7.08	90b	15a
SBS	6.39	66	8	6.80	44a	6b
Means	**7.01**	**59**	**7.3**	**6.83**	**51**	**11.4**
LSD	**ns**	**ns**	**ns**	**ns**	**23**	**8**

Means followed by the same letter within a column are not significantly different at $p<0.05$.
ns = not significant at $p<0.05$ Cond. = Conductivity *= data not available

Biomass of legume crops at flowering

Sunn hemp, velvet beans and soybeans were ploughed under on 9 July, 14 September and 17 September 2009, respectively. This was 64, 131 and 134 days respectively, after sowing on 6 May 2009. The legume crops produced different quantities of above-ground biomass. Sunn hemp, velvet beans and soybeans accumulated 18.5, 14.6 and 3.4 t/ha DM, respectively.

Total colony forming units (CFUs) in the soil

Sunn hemp, velvet beans and soybeans all improved soil microbial densities (Table 4a). When sugarcane plant crops were harvested during September and November 2010, CFUs in the topsoil had increased by 6.7, 40.1 and 67.4% in plots that initially had soybeans, sunhemp and velvet beans respectively, compared to those that were left fallow (Table 4b). The topsoil profile had significantly more CFUs than the 31-60 cmprofiles. There were no differences in CFUs between 0-10 and 11-20 cm depths (Table 4b). Bacteria were more abundant than fungi (data not shown). Sunn hemp had higher CFUs in the lower soil profiles than the other treatments, whereas velvet beans had the highest in the topsoil after ploughing under the legumes. The increased CFUs were not sustained in subsequent ratoon crops that did not have relay legumes (data not shown).

Microbial densities in the 31-60 cm soil profile improved after cropping compared to pre-planting (Tables 4a and 4b).

Table 4(a). Mean colony forming units (CFUs) per gram soil before planting, and 26 days after ploughing under green sunn hemp and velvet beans, and after harvesting dry soybeans. The legume crops were sown on 6 May 2009; sunn hemp was ploughed under on 9 July, velvet beans on 14 September and soybeans on 17 September 2009.

Treatment	Pre-planting CFUs/g x 1000		After legumes CFUs/g x 1000	
	0-30 cm	31-60 cm	0-30 cm	31-60 cm
Sugarcane monoculture	19.0	8.2	38.6a	18.2a
Sunn hemp break crop	17.4	6.6	37.4a	20.1a
90 day RSD fallow	26.4	12.1	26.7b	8.3c
Velvet beans break crop	26.8	9.5	44.7a	16.2a
Soybeans break crop	24.2	12.8	28.5b	14.3b
Means	**22.8**	**9.8**	**35.2**	**15.4**
LSD	**ns**	**ns**	**8.6**	**5.6**

Means followed by the same letter within a column are not significantly different at $p<0.05$.
ns = not significant at $p<0.05$.

Table 4(b). Mean colony forming units (CFUs) per gram soil 160 days after ploughing under sunn hemp, velvet beans and soybeans, and four days after harvesting sugarcane plant crops. Sunn hemp was ploughed under on 9 July, velvet beans on 14 September and soybeans on 17 September 2009. The respective plant crops were harvested on 15 September (ex-sunn hemp plots), 24 November 2010 (ex-velvet bean and soybean plots).Monoculture crops were harvested on 20 July and crops following fallows on 3 November, 2010.

Treatment	160 days after legumes CFUs/g x 1000		4 days after harvesting cane CFUs/g x 10000	
	0-30 cm	31-60 cm	0-10 cm	11-20 cm
Sugarcane monoculture	27.6	21.1	33.4a	31.8a
Sunn hemp break crop	32.6	19.0	58.9b	56.3b
90 day RSD fallow	28.9	22.4	43.2a	38.8a
Velvet beans break crop	33.9	21.5	44.1a	36.2a
Soybeans break crop	28.3	19.3	44.4a	32.9a
Means	**30.3**	**20.7**	**42.2**	**39.2**
LSD	**ns**	**ns**	**12.3**	**11.6**

Means followed by the same letter within a column are not significantly different at $p<0.05$.
ns = not significant at $p<0.05$.

RSD results in the plant crops

All sugarcane plant crops of the susceptible N14 and resistant CP72-2086 were free of the RSD bacteria. This was expected because the planting materials used in the trial were free of bacteria.

Discussion

Field Z4 had an average of 0.74% organic matter (OM) in the topsoil and 0.65% at31-60 cm before any crops were grown,indicating low OM content. The field had been cropped to sugarcane for over 20 years, and burning of cane prior to harvesting was routinely practised. Frequent tillage, periods of bare ground, and removal of crop residues were reported to contribute to reductions in soil organic matter (SOM) (Jenkinson and Ladd, 1981; Follett and Schimel, 1989).Soils in sugarcane growing areas of Zimbabwe are generally known to have

low OM (LT Mupondi, personal communication[1]). Soils analysed at ZSAES between 2009 and 2013 had on average 1.55% OM compared to >3% considered ideal (LT Mupondi, personal communication[2]).Low OM negatively affects soil aggregate stability, cation exchange capacity, the rate of nutrient release by mineralisation, and low water-holding capacity, all of which affect plant growth (Jenkinson and Ladd, 1981).

Soybeans, sunn hemp and velvet beans all improved SOM content,with greater benefits derived from growing velvet beans and sunnhemp (Table 2). The legume crops, which were sown in May, were not planted during their recommended growing seasons for best productivity. Soybean, normally grown in summer, was most affected by being grown off-season. At flowering sunn hemp had accumulated the most dry matter(18.5 t/ha) followed by velvet beans (14.6 t/ha) and soybeans (3.4 t/ha). Velvet beans and sunn hemp grow vigorously in tropical and subtropical areas and produce huge amounts of biomass (Abdul-Baki *et al*, 2001; Baijkya *et al*, 2005; Whitbread *et al*, 2004). The increase in SOM content resulted from the legume biomass that was incorporated into the soil.Velvet beans and sunn hemp were ploughed under as green manure whereas soybeans were harvested when the pods were dry, and most of the crop residue was not returned to the field.Sugarcane grown as a monoculture crop improved SOM probably from dead tillers and leaves. Although the legumes increased SOM, average SOM content was still understandably low because this was done over a single season. SOM content declined during the first ratoon crop (Table 2). The lowest SOM was recorded in the fallow treatment, where the 0-30cm profile averaged 0.61% and the 31-60cm profile averaged0.67%. SOM content for the other treatments ranged from 0.84-1.16%, with no significant differences between the soil depths. Burning of sugarcane crops prior to harvesting would reduce the gains in SOM content. Relay cropping of legumes would be expected to further improve SOM.

Soil microbial biomassis defined as the part of OM in the soil that constitutes living microorganisms smaller than 5-10 μm^3 and consists mostly of bacteria and fungi (Jenkinson and Ladd, 1981).Soils in Field Z4 had variable densities of CFUsafter land preparation but, before planting any crops, the top 30 cm soil profile averaged 22.76 x 10^3CFUs/g compared to 9.84 x 10^3CFUs/g of soil in the subsequent 30 cm layer. Microbial biomass increased to 35.18 x 10^3CFUs/g (0-30 cm) and 15.42 x 10^3CFUs/g (31-60 cm) immediately after ploughing under the legume crops, and to 30.3 x 10^3CFUs/g and 20.7 x 10^3CFUs/g of soil 150 days thereafter. Four days after harvesting the sugarcane plant crops,soil microbial densities peaked at 422.3 x 10^3CFUs/g and 392.4 x 10^3CFUs/gin the respective soil depths. Although the topsoils generally had more CFUs per unit weight of soil than the deeper profiles, differences were smaller than those reported by Follett and Schimel (1989), Gonzalez-Quinones *et al*. (2011) and Ley *et al*.(2001)possibly because decomposed OM leached to the 31-60 cm depths of the sandy clay loam soils in Field Z4. Martens (1995) noted that about half the microbial biomass was located in the surface 10 cm of a soil profile and generally, up to 5% of the total organic carbon in the soil was in the microbial biomass (Murphy *et al.*, 1998). The size and distribution of microbial biomass was also influenced by soil properties such as pH, clay content, and the availability of organic carbon (Olsen and Bakken, 1987; Martens, 1995).In the present case, the distribution of microbial biomass could have been affected by ploughing and discing to incorporate the legumes and to prepare the field for planting of sugarcane.Moreover, microbial biomasses after harvesting sugarcane plant crops may have been influenced by differences in sampling dates.Microbial biomass

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readings are not stable numbers; they fluctuate depending on environmental conditions preceding and on the day of sampling (Martens, 1995; Gonzalez-Quinones *et al.*, 2011). Storage conditions of sampled soils also change the biomass counts. Soil samples in this experiment were stored at 4°C for 4-10 days before transportation to the Tobacco Research Board's Kutsaga Research Station for culturing and assessment.

In the experiment, microbial biomass trends were similar to those of OM. Velvet beans and sunn hemp sustained the highest biomass, while soybeans had lower densities of culturable microbes. Fallowed areas, which had the least OM, also had the lowest microbial biomass.Management of crop residues influences microbial biomass as they are one of the primary forms of organic carbon and nutrients used by microorganisms. Retaining crop residues instead of burning would provide a practical way of increasing microbial biomass in soil by increasing the amount of available organic carbon (Dalal and Mayer, 1987). The type of crops in a rotation also affected microbial biomass. Residues of legume crops are likely to increase microbial biomass because of their greater N content (Whitbread*et al.*, 2004).

Conclusion

The experiment demonstrated the potential benefits of growing velvet beans, sunn hemp or soybeans as break crops during RSD fallows in sugarcane cropping systems.The legumes improved both soil OM content and soil microbial biomass. Based on these results, it is recommended that growers include velvet beansor sunn hemp into their cropping system during the RSD fallow.

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