Project number M106/10								
Project title		The role	e of	soil	microbiolog	jy in	maize	production
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Final abstract

Soil microbiology in local agriculture has not been extensively studied especially with regards to maize production. Microorganisms are responsible for numerous processes in the soil. Most of the species in soil are microorganisms, such as bacteria, fungi and protozoans, which are the chemical engineers of the soil, responsible for the decomposition of plant organic matter into nutrients readily available for plants, animals and humans. Any change in agronomic practices will reflect in the soil's biological status. With this in mind the aim of the study was to investigate the role of soil microbiology in maize production. Various soil types from the national maize cultivar trials were studied over a five year period and subjected to selected chemical and microbiological analyses such as heterotrophic plate counts, actinomycete and filamentous fungal counts. Soil samples were also subjected to microbial soil enzyme analyses i.e. β-glucosidase, and acid phosphatase activities. Whole community profiling were also studied through Biolog Ecoplate analyses. The results showed that in general, microbiological properties such as microbial β-glucosidase and acid phosphatase activities are linked to a specific locality in this case soil type. Correlations could not be found between cultivar and biological properties. Whole community profiling showed no distinct pattern in carbon assimilation amongst localities and cultivars. Results generated provided a sound baseline of soil microbiological data relevant to local maize production.

Keywords: soil microorganisms, agricultural practices, maize production

INTRODUCTION

This project was initiated during 2006/07 season in order to address the lack of knowledge of soil microbiology research under South African conditions. Yields in crop production systems especially with regard to crop rotation, could not be sufficiently explained by physical and chemical interactions (parameters) alone. In this regard, microbial interactions probably play a crucial and contributing role (Arias et al., 2005). Maize production at farm level is currently under severe economic pressure and subject to a high level of inherent risk. Soil health, moreover, soil microbiology as an integral part of our agricultural production systems and a key factor in quality crop production, has been implicated as a possible underlying cause (Girvan et al., 2003). Healthy soils maintain a diverse community of soil organisms that help to control plant diseases, insect and weed pests, form beneficial symbiotic associations with plant roots (e.g. nitrogen fixing bacteria and mychorrhizal fungi), recycle plant nutrients and improve soil structure (e.g. filamentous fungi) with positive repercussions for water-and nutrient availability (Arias et al., 2005). Any improvement in the efficiency of biological farming production technologies might contribute to reduce maize production costs (R t⁻¹). Soils of the maize producing areas in South Africa, especially those under monoculture maize, are likely to be unbalanced with limited microbial activity. Depletion of carbon is one of the measurable symptoms of the poor state of these soils. Another indicator of poor soil conditions is the frequently-encountered yield improvement of maize when grown in rotation with certain crops that are known for their positive effect on soil biology. An improvement in the microbiology of the soil will in all likelihood, also improve efficiency of production and thereby, reduce the production cost of maize.

If the importance of microbial activity in maize production is known and if it could be manipulated through addition of ameliorants, adaptation of production practices or by other means, it might be possible to improve the efficiency and economy of maize production at farm level. Improved soil microbial activity is likely also to contribute to resource conservation, which is not measurable in monetary terms but could lead to long-term vast improvements. Nitrogen, which is an expensive nutrient, can if not utilised by maize, turn into a pollutant of ground water (potentially a growing problem in future). Soil microbes can act both as a sink as well as a source of nitrogen and if managed well, might be able to reduce leaching of nitrates but release it again during active crop growth, thus reducing nitrogen fertiliser demand.

It is well known that soil microbial organisms such as bacteria (actinomycetes and *Pseudomonas* spp.) and filamentous fungi as well as mychorrhizal fungi can vary considerably in population composition and their relative abundance play an important role in the soil as a substrate for plant growth (Kennedy & Smith, 1995). These microbes form an integral part of our agricultural production systems. Crops planted in soil with a well-balanced and thriving microbial component utilise soil nutrients and water more efficiently. Until recently this aspect of soil science received very little attention, if any, in local maize production and the importance of microbial activity in the improvement of maize production is unknown.

Microorganisms are responsible for numerous processes that occur in soils and can be viewed as the main source of enzymes in soils (Acosta-Martínez *et al.*, 2007). Soil enzymes serve as mediators and catalysts of biochemical processes that are important in soil functions such as nutrient mineralisation and cycling, formation of soil organic matter as well as decomposition of xenobiotics. Therefore knowledge of soil enzyme activities provides information regarding degradation potential of a soil. Any change in soil management and land use is also reflected in soil enzyme activities and changes in soil quality could be anticipated. Various studies have shown that enzyme activity is sensitive to soil change due to tillage, cropping systems and land use (Acosta-Martínez & Tabatabai, 2001; Kandeler *et al.*, 1999; Klose *et al.*, 1999).

A comprehensive assessment of soil microbial community characteristics is one way in which to address the incomplete picture of soil status that other approaches provide. If changes in microbial community function and structure in an ecosystem are monitored, it can serve as an indicator on how microbial diversity in different soil types affects maize production under various agronomic practices.

Assessment of microbial soil characteristics represents a promising approach for a thorough characterisation of soil and for the detection of factors that affect soil quality (Hartmann *et al.*, 2005). A valuable approach to monitor biological status is the use of functional profiles, since it relates to the actual or potential activities or organisms that ultimately result in ecosystem dynamics. In this context, assays of enzymatic activities and community level physiological profiles (CLPP's) are often applied to determine functional diversity of microbial

communities. Soil enzymatic assays have been suggested as potential indicators of soil quality due to their essential role in soil biology, ease of measurement and rapid response to changes in soil management (Bandick & Dick, 1999). They also integrate information from the microbial status and the physico-chemical conditions of the soil (Garcia *et al.*, 1993). An example of a CLPP method is the Biolog system, specifically the Biolog EcoPlate, which was created purposely for community analysis and microbial ecological studies. The Biolog EcoPlate contains 31 of the most useful carbon sources for soil community analysis. Communities of organisms will give a characteristic reaction pattern called a metabolic fingerprint. These fingerprint reaction patterns rapidly and easily provide a vast amount of information from a single Biolog MicroPlate.

With this in mind the aim of the project was to determine how agronomic practices such as crop rotation, cultivation and nitrogen fertilisation affect the soil microbial activity and composition in some typical maize-producing soils.

MATERIALS & METHODS

Trials

Two aspects were investigated with the current project i.e. the effect that crop rotation and cultivation practices would have on soil microbiology as well as the effect that locality and cultivar would have on soil microbial parameters

Cultivation practices and crop rotation effect on soil microbial parameters

During the 2006/07 and 2007/08 seasons a trial at Bloekomspruit was studied that quantified the rotational effect of soybean, dry bean and sunflower on maize, combined with conventional and reduced cultivation and nitrogen application under conditions of relatively high yield potential. For the 2006/07 season a trial at Bothaville was also studied comprising of treatments that consisted of two crop systems (maize in rotation with soybean and monocropped maize), three cultivation intensities (ripping, ripping every second season and no-till) and two nitrogen fertilisation procedures (conventional and delta yield). In the crop rotation trial at Vaalharts winter as well as summer crops were grown and only monitored for the 2006/07 season. Treatments were maize planted every summer or every second summer preceded by canola, wheat or barley (two-year system).

Cultivar and locality effect on soil microbial parameters

From 2008 (season 2) onwards studies were conducted on selected national maize trials *viz*. Bethlehem, Bloekomspruit, Coligny, Tweebuffels, Ventersdorp, Vierfontein and Wesselsbron. Soil from various cultivars was selected for this study as reflected in Table 1.

Cultivar 1	Cultivar 2	Cultivar 3
PAN6611	PAN6616	DKC 80-12B
PAN6611	PAN6616	DKC 80-12B
PAN6616	DKC 80-12B	DKC 78-15B
PAN6616	DKC 80-12B	DKC 78-15B
PAN6Q-308B	DKC 80-12B	DKC 78-15B
	Cultivar 1 PAN6611 PAN6611 PAN6616 PAN6616 PAN6Q-308B	Cultivar 1 Cultivar 2 PAN6611 PAN6616 PAN6611 PAN6616 PAN6616 DKC 80-12B PAN6616 DKC 80-12B PAN6Q-308B DKC 80-12B

Table1	Selected	maize	cultivars	studied
Iable I	Selected	maize	cultivals	Sludieu

Laboratory analyses

Soil was collected from various localities that form part of the national maize cultivar trials since 2008. Five soil samples were taken per replicate from inter-row surface soil (0 to 15 cm) during the flowering stage of the crop. These soil samples were pooled from all the replicates per locality. Samples were then air-dried for 48hr at room temperature (22°C) and subjected to chemical as well as biological analyses.

Conventional counts

A serial dilution series (10⁻¹ to 10⁻⁶) of soil samples was prepared for every sample using distilled water. Viable cell counts were obtained using the aseptic spread plate technique. Different microbial growth media designed to be selective for heterotrophic microbes; actinomycetes and filamentous fungi were used in the microbial analyses. These microbial populations were subjected to the physiological ability of microbes to grow on each of the selective media. General heterotrophic plate counts were done on nutrient agar (NA), (Biolab, Midrand and South Africa)

Actinomycetes were enumerated on Actinomycete isolation agar (Sigma-Aldrich, South Africa). To obtain filamentous fungal counts, malt extract agar (MEA;Biolab, Midrand, South Africa) was used supplemented with 30ppm chloramphenicol and 50 ppm streptomycin.

Enzyme assays

Soil enzymes regulate ecosystem functioning and in particular play a key role in nutrient cycling. The microbial activities of β -glucosidase, and acid phosphatase were determined using 1g of air-dried soil and incubated for 1h (37 °C) with the appropriate substrate for each enzyme at their respective optimal pH values (Tabatabai, 1994). In the case of urease 5g of air-dried soil was used. Methods used are summarised in Table 2. These selected enzymes have been implicated in the carbon, nitrogen and phosphorous soil cycles.

EC number ^a	Recommended	Assay conditions ^c	Optimum pH
	name ^b	[Substrate]	
3.1.3.2	Acid phosphatase	<i>p</i> -Nitrophenyl	6.5
		phosphate [25mM]	
3.2.1.21	β-glucosidase	<i>p</i> -Nitrophenyl- β-	6.0
		glucopyranoside	
		[25mM]	
3.5.1.5	Urease	Urea [80mM]	Non-buffered
a EC numbe	r denotes enzvme class		

Table 2 The methods used to determine enzyme activity in soils.

^b Methods according to Tabatabai (1994 and 1982)

^c Values in parentheses are substrate concentrations under the respective assay conditions. The product of reactions for glucosidase and phosphatase is *p*-Nitrophenol=PN

Determination of the Functional Diversity

The functional diversity of the microbial communities within each of the consolidated soil samples was determined according to the procedure as described by Buyer & Drinkwater (1997) using commercially available Biolog[®] microtiter Ecoplates (Biolog[®] Inc., Hayward, USA). The Biolog Ecoplate system provides a representation of metabolic potential also known as phenotyping of the bacterial component in the soil.

Biolog Ecoplate Preparations

Soil solutions were prepared by shaking approximately 10 g of soil (wet weight) in 90 ml sterile deionised water for 50 minutes on a rotary shaker at 250 rpm. The suspension was allowed to settle for a period of 2 h to remove larger soil particles, where after the supernatant was diluted with sterile deionised water to a final dilution of 1:10. The soil suspension was poured into a reagent reservoir and the Biolog[®] microtiter plates were aseptically inoculated with 150 μ l aliquots of each sample using a multi-channel pipettor. Community level physiological profiles (CLPP) analyses of each sample were performed in triplicate. After an initial reading (time = 0), the Biolog[®] microtiter plates were incubated at 25°C. The tetrazolium violet reduction in each well was spectrophotometrically quantified at

590 nm for a period of 4 days using a Biotek Elx800 microtiter plate reader (Analytical Diagnostics Products, South Africa).

Statistical Analyses

Parametric and non-parametric statistical analyses were performed on all data obtained using STATISTICA 5 (StatSoft, Inc ©) and Statgraphics. The data was tested for normality using the Shapiro-Wilk's test. In the case of data being normally distributed (parametric) a breakdown and one-way ANOVA was performed and the Tukey's honest significant difference (HSD) test was used to determine statistical significance between the various samples. In the case of non-parametric data, non-parametric data analysis was performed and the Kruskal-Wallis ANOVA and Median test was used to determine statistically significant difference between samples. In all cases, there were no differences between the results obtained from the respective normality analyses. As a result, only the parametric analyses of the relevant statistics are discussed.

For Biolog data two multivariable methods were used. PCA and cluster analysis (AHM) using Ward's binary method were employed to establish clusters. These techniques were performed using XLSTAT.

RESULTS

Chemical analyses were performed on all collected maize cultivar soil samples during this study (Table 3). These properties together with biological data were subjected to statistical analyses.

2008										
Locality	рН	NO3	NH4	PBray1	к	Са	Mg	Na	Zn	%С
Bethlehem	3.99	9.25	6	91	115	238	50	3	3.12	0.51
Bloekomspruit	5.46	0.25	0.6	29	70	300	100	0	2.16	0.43
Hartbeesfontein	4.95	6.25	0	42	120	298	85	0	3.92	0.58
Vaalharts	5.44	9	0.5	43	148	485	140	38	4.16	0.75
Vierfontein	5.44	3.25	0.15	19	53	148	43	0	2.08	0.22
Wesselsbron	6.09	4.65	0.5	25	98	208	90	0	2.88	0.31
2009										
Bethlehem	5.06	1.4	0.5	43	85	655	113	5	2.8	0.5101
Bloekomspruit	4.64	1.15	1	33	78	213	78	3	2.76	0.421
Coligny	6.44	1.25	0.5	38	110	365	73	3	4.88	0.3978
Vaalharts	5.66	1	0.9	43	105	383	110	23	2.64	0.359
Vierfontein	6.84	2	0.75	43	78	285	120	3	6.24	0.4171
Wesselsbron	5.79	2	0.9	26	90	190	85	0	4.12	0.421
2010										
Bethlehem	5.51	5	0.65	75	108	753	123	15	6.16	0.65
Bloekomspruit	4.49	0.5	0.85	33	85	208	75	15	2.28	0.4
Coligny	5.42	0.4	0.85	39	40	258	83	13	3.72	0.34
Hartbeesfontein	5.2	0.65	0.5	46	25	178	63	5	4.56	0.41
Nampo	4.38	0.4	0.35	46	40	138	50	10	4.36	0.36
Vaalharts	5.55	1.15	1.1	42	98	553	208	35	3.64	0.49
Wesselsbron	4.06	0.5	0.6	32	35	65	23	13	2	0.27
2011										
Betlehem	5.4	9.25	12.6	86	120	678	118	20	4.4	0.36
Bloekomspruit	4.78	1	0.75	67	100	245	78	5	4.48	0.43
Coligny	5.78	1.75	0.35	47	73	273	88	3	4.4	0.35
Ventersdorp	4.18	4.4	0.35	48	100	303	98	18	4.4	0.73
Vierfontein	5.17	0.25	0.75	62	43	205	55	3	3.44	0.4
Wesselsbron	4.19	0.75	0.75	37	43	60	23	10	3.36	0.29

Table 3 Chemical properties of selected cultivar trials

2012										
Locality	рН	NO3	NH4	PBray1	К	Ca	Mg	Na	Zn	%C
Betlehem	4.62	5.75	23.9	60	88	593	98	10	4.16	0.66
Bloekomspruit	4.53	1.5	1.5	28	85	233	65	3	2.48	0.49
Coligny	6.18	0.9	0.5	37	73	320	98	3	4.36	0.35
Tweebuffels	6.23	7.5	0.902	101	128	480	95	5	7.8	0.42
Ventersdorp	3.98	2.9	0.75	48	43	180	58	3	2	0.29
Vierfontein	5.74	1.25	0.65	25	53	230	68	3	3.52	0.42
Wesselsbron	5.41	15.15	2.65	35	118	215	80	5	3.2	0.32

Season 1

During 2006/07 season the first series of soil samples on maize in crop rotation trials were collected for microbiological analysis at three different localities, viz. Bloekomspruit, Bothaville and Vaalharts. All soil samples were taken during the active growing phase of the crop. Various microbiological methods i.e. plate counts, community level physiological profiling and soil enzyme assays have been used in studying microbial activity within these agricultural systems at above-mentioned localities. Although the 2006/07 season was a very dry season suggesting that microbial activity might have been suppressed, the results at Bloekomspruit revealed that soils from maize grown after sunflower had higher bacterial activity than that of maize after soybean and monoculture maize. Low nitrogen treatments also revealed that fungal activity was higher in soils from monoculture maize rotation system than soils from maize grown after sunflower. Also, microbial phosphatase activity was higher in soils from maize and soybean than that of sunflower (Fig 1). At Vaalharts, the soils of maize in the two year crop rotation system after wheat showed a higher microbial phosphatase activity than that of maize after canola in the one year production system (Fig 2). Microbial carbon assimilation patterns in soils of maize after barley and the two year system maize after canola were higher than that of soils from maize after wheat. Actinomycete activity was also significantly higher in the two-year system soils from maize after canola than that of soils of maize after wheat. At Nampopark (Bothaville) actinomycete activity in soils with a higher nitrogen treatment level were significantly influenced by the rotation system. A similar finding also revealed that fungal activity in soils with zero nitrogen treatments were significantly affected by the rotation system.



Fig. 1 Phosphatase activity in soils (0-15cm depth) sampled from different crop rotation systems at Bloekomspruit (2006/7). LSD(0.05)=12.57



Fig. 2 Phosphatase activity in soils (0-15cm depth) sampled from different crop rotation systems at Vaalharts (2006/07). LSD(0.05)=7.77

Season 2

During the 2007/08 production season the second series of soil samples from maize fields in crop rotation trial at Bothaville were collected for microbiological analysis. The trial at Bothaville consisted of two cropping systems (maize in rotation with soybean and monocropped maize), three cultivation intensities (ripping, ripping every second season and no-till). Soils from maize cultivar trials at Bethlehem, Bloekomspruit, Hartbeesfontein, Vierfontein, and Wesselsbron were also collected. All soil samples were taken during the active growing phase of the crop. Various microbiological methods i.e. plate counts; community level physiological profiling and soil enzyme assays have been used in studying microbial activity within these agricultural systems at above-mentioned localities. The trial at Bothaville (Nampopark) indicated that crop rotation had an effect on phosphatase activity in soils. The cultivar trials revealed that bacterial numbers were significantly affected by cultivars and among localities. These trials showed a relationship exists between phosphatase activity and P, Ca and Mg content of the soil. Furthermore, a strong relationship was found to exist between glucosidase activity and Na as well as organic C content among the cultivar trials.

Source	Sum of squares	Df	Mean square	F-ratio	P-value
Main effects					
Cultivar	1.94909E13	2	9.74544E12	5.0	0.0111
Locality	4.16668E13	5	8.33335E12	4.27	0.0030
Rep	1.1274E12	2	5.63702E11	0.29	0.7504
Residual	8.58096E13	44	1.95022E12		
Total corrected	1.48095E	53			

Table 4 Effect of cultivar and locality on heterotrophic bacteria

All F-ratios are based on the residual mean square error.

Season 3

During the 2008/09 season a second series (first series-2007/08season) of soil sampling was performed from selected fields from the national maize cultivar trials at Bethlehem, Bloekomspruit, Coligny, Vaalharts, Vierfontein and Wesselsbron. All the collected soil samples were subjected to chemical and microbiological analyses. The latter included

determination of bacterium and fungal counts on nutrient agar and malt-extract agar media, respectively, as well as enumeration of actinomycetes. Microbial enzyme activities were determined by extracting soil enzymes such as ß-glucosidase and phosphatase. These enzymes have been implicated in soil nutrient cycles that serve as soil health indicators. The results obtained showed that glucosidase activity was significantly affected by locality (Table 3 and 4). Results also showed relationship between phosphatase activity and P, Ca and Mg content of the soil for the 20007/08 season, whereas in the following season a relationship existed between both glucosidase and phosphatase activity, Na and content NO₃ (Correlation Coefficient = 0.519, $R^2 = 26.96$ percent) in the soil.

-						
Source	Sum	of	Df	Mean	F-ratio	P-value
	squares	5		square		
Main						
effects						
Cultivar	796.751		2	398.375	2.49	0.0974
Locality	5577.66		5	1115.53	6.98	0.0001
Interaction	3633.44		10	363.344	2.27	0.0357
(CxL)						
Residual	5597.16		35	159.919		
Total	15657.7		52			
corrected						

Table 3 Effect of cultivar and locality on glucosidase activity OR Effect of cultivars on ßglucosidase over localities

All F-ratios are based on the residual mean square error.

Source	Sum of squares	Df	Mean square	F-ratio	P-value
Main effects					
Cultivar	414.361	2	398.375	207.18	0.6904
Locality	76387.5	6	1115.53	12731.2	0.0000
Rep Interaction (CxL)s	613.493 4936.34	2 12	306.746 411.361	0.55 0.74	0.5791 0.7023
Residual	22157.8	40	553.946		
Total corrected	104509.0	62			

Table 4 Effect of cultivar and locality on glucosidase activity

All F-ratios are based on the residual mean square error.

Season 4

During the 2009/10 season a third series (first series-2007/08season) of soil sampling was performed from selected fields from the national maize cultivar trials at Bethlehem, Bloekomspruit, Coligny, Hartbeesfontein, Vaalharts, Vierfontein and Wesselsbron. All the collected soil samples were subjected to chemical and microbiological analyses. The latter included determination of bacterium and fungal counts on nutrient agar and malt-extract agar media, respectively, as well as enumeration of actinomycetes. Microbial enzyme activities were determined by extracting soil enzymes such as ß-glucosidase and phosphatase. These enzymes have been implicated in soil nutrient cycles that serve as soil health indicators. As compared to the 2008/09 season both ß-glucosidase and phosphatase activities were significantly affected by locality. Actinomycete, bacterial and fungal populations were also not significantly affected by maize cultivars or locality. The results also showed a relationship between both ß-glucosidase activity and organic carbon soil content (Correlation Coefficient = 0.513, R^2 = 26.34 percent) (Fig 3). ß-glucosidase activity also displayed a correlation with P and Ca. Phosphatase activity also correlated with soil pH (Correlation Coefficient = -0.419, $R^2 = 17.64$ percent) (Fig 4), nitrate (Fig 5) and K (Correlation Coefficient = 0.419, $R^2 =$ 17.64 percent) (Fig 6) as compared to the 2008/09 season where a strong relationship existed between phosphatase activity and sodium and nitrate.



Fig 3 Regression plot of ß-glucosidase activity and percentage organic carbon in soil for 2009/10 season.



Fig 4 Regression plot of acid phosphatase activity and soil pH for 2009/10 season.



Fig 5 Regression plot of acid phosphatase activity and nitrate in soil for 2009/10 season.



Fig 6 Regression plot of acid phosphatase activity and soil potassium for 2009/10 season.

Season 5

During the 2010/11 season a fourth series (first series-2007/08 season) of soil sampling was performed from selected fields from the national maize cultivar trials at Bethlehem, Bloekomspruit, Coligny, Ventersdorp, Vierfontein and Wesselsbron. All the collected soil samples were subjected to chemical and microbiological analyses. The latter included determination of bacterium and fungal counts on nutrient agar and malt-extract agar media, respectively, as well as enumeration of actinomycetes. Microbial enzyme activities were determined by extracting soil enzymes such as ß-glucosidase phosphatase and urease. These enzymes have been implicated in soil nutrient cycles that serve as soil health indicators. Once again compared to the 2009/10 season only ß-glucosidase levels were significantly affected by locality. Actinomycete, bacterial and fungal populations were also not significantly affected by maize cultivars or locality. The results also showed a relationship between both ß-glucosidase activity and ammonium as well as nitrate soil content. Similar to the preceding season ß-glucosidase activity also displayed a correlation with soil phosphorus and calcium. For the 2010/11 season urease activity also correlated with soil potassium (percentage) as compared to the 2009/10 season where a strong relationship existed between phosphatase activity and soil potassium.

Season 6

For the 2011/12 season a final series of soil sampling was performed from selected fields from the national maize cultivar trials at Bethlehem, Bloekomspruit, Coligny, Tweebuffels Ventersdorp, Vierfontein and Wesselsbron. The collected soil samples were subjected to chemical and microbiological analyses. Compared to the 2011/12 season all microbiological parameters *viz*. Actinomyctes, heterotrophic bacteria and filamentous fungi were significantly affected by locality. The same applied for all soil enzymes tested i.e. ß-glucosidase, phosphatase and urease activities. No correlations could be established between microbiological and chemical properties tested. Whole community metabolic potential was also performed through Biolog Ecoplate analysis. Biolog Ecoplate data revealed that three clusters of carbon assimilation were present (Fig 7 and 8). However, these groupings could not be distinguished amongst locality or cultivar.



Fig 7 Dendrogram of community level physiological profile (CLPP) patterns of soil microbial communities amongst cultivar and localities.



Fig 8 Principle component analysis (PCA) showing carbon assimilation patterns of soil microbial communities amongst cultivar and locality.

DISCUSSION

For first two seasons cultivation practices and the crop rotational effect in maize production on soil microbial parameters were studied at Bloekomspruit, Bothaville (Nampopark) and Vaalharts respectively. The results showed that in general bacterial numbers were higher in maize soils after rotation with sunflower. These activities suggest that possible correlations could be drawn from yield and bacterial data in order to establish if the particular system favours high or low yields. Fungal activity was not significantly affected. This could be due to conventional tillage practices that disturb the fungal hyphal network. Phosphatase activities were also higher for maize soils after rotation with soybean compared to maize after canola and wheat at the Vaalharts trial. For instance, research has shown that legumes secrete more phosphatase enzymes than cereals (Yadav and Tarafdar, 2001). This may probably be due to a higher requirement of P by legumes in the symbiotic nitrogen fixation process as compared to cereals. In their studies, Li *et al.* (2004) reported that chickpea roots were also able to secrete greater amounts of acid phosphatase than maize.

The levels of actinomycetes were also higher in two-year system soils from maize after canola than that of soils of maize after wheat. Actinomycetes are particularly effective at breaking down tough substances like cellulose (which makes up the cell walls of plants) and chitin (which makes up the cell walls of fungi) even under harsh conditions, such as high soil pH. Actinomycetes form associations with some non-leguminous plants and can fix nitrogen, which is then available to both the host and other plants in the near vicinity. Some soil nitrogen is unusable by plants until bacteria convert it to forms that can be easily assimilated.

For the cultivar trials levels of actinomycetes, bacteria, and fungi were not significantly different amongst cultivar treatments but differed significantly in the last season of monitoring among localities. A possible reason could be that conventional tillage practices was employed implying that if there was yield differences it could be ascribed to the soil type. This was further supported by the significant levels of glucosidase activities among various localities. This enzyme is commonly found in soil and is involved in catalysing the hydrolysis and biodegradation of glucosides in plant debris with glucose as final product. It is sensitive to changes in pH and soil management practices (Makoi & Ndakidemi, 2008).

The complex interactions within the soil ecosystem is further demonstrated through whole community metabolic profiling in that microorganisms showed no clear carbon assimilation pattern among cultivar or locality. For future studies canonical variance analysis (CVA) might provide a better understanding to the relevant role players that are active in the soil ecosystem.

CONCLUSION

The study highlighted that microbial activities among localities are significantly different amongst different maize production fields and that soil determinants such as climate and soil properties could probably explain these differences. Due to variation in soil properties no group of microbes correlate with a particular soil chemical property. In most case there is a strong association between microbiological properties and locality. Thus it can be implied that the soil type in all likelihood is the factor responsible for different responses in microbial activity.

Our results indicated that tillage practices and cropping rotations can impact soil microbial populations and community composition. Interestingly, cropping sequence had a larger impact on bacterial community composition and relative levels of soil enzymes than did the tillage treatment. However, this study only analysed samples from one time point, only during the active growing phase of the crop. Future studies should also consider analysing samples over an extended period of time in order to gain a better understanding of microbial dynamics in response to the different crops in the rotation system and in different soil types. It is possible that additional sequencing of the bacterial community as well as fungal and archaeal sequencing would reveal further differences. Future work should attempt to also characterise soils using additional sequencing approaches (e.g., metagenomic sequencing) and/or functional gene assays.

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