




Effects of long-term sisal residue returning on soil physiochemistry, microbial community, and sisal yield

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ABSTRACT: Returning agricultural waste/by-products to the field is an effective management measure to improve soil fertility and maintain crop productivity in agroecosystems. However, we still have limited understanding of the complex response of sisal (*Agave sisalana* Perrine) growth, soil nutrients and microbial communities to the long-term sisal residue return to the ecological system. This study aimed to investigate the comprehensive effects of returning sisal residue to the field on sisal yield, soil nutrient, and microbial community. Based on a four-year field experiment, the results showed that returning sisal residue to the field increased the annual yield of sisal by 30.77–33.36%. Compared with control group, sisal residue inputs significantly improved soil total nitrogen, available phosphorus, and available potassium by 46.59, 3.02, and 1.21%, respectively. Under sisal residue input condition, the activities of soil catalase, urease, acid phosphatase, and sucrase also significantly increased by 2.01, 17.20, 17.40 and 16.60%, respectively. A higher ratio of bacteria/fungi was observed in the soil amended with sisal residue. The microbial diversity analysis showed that the α -diversity of the bacterial community increased, while the α -diversity of the fungal community decreased with the sisal residue treatment. However, sisal residue return had a higher impact on the α -diversity of fungi. This study provides evidence that returning sisal residue to the field affects sisal productivity by regulating nutrient cycling and soil microbial community. Moreover, the study suggests that microbial α -diversity has significant influence on sisal yield.

Key words: *Agave sisalana* Perrine; sisal residue returning; soil fertility; microbial community; yield.

INTRODUCTION

Agaves, belonging to the Asparagaceae family, are a group of perennial monocotyledonous plants in semi-arid to arid environments. The genus *Agave* contains more than 210 species, of which about 70% are economically valuable (Bermúdez-Bazán et al. 2021). As a tropical fiber crop, sisal is widely grown in America, Africa, Asia, Oceania, the Pacific Ocean, some islands of the Indian Ocean, on tropical and subtropical regions between 30° north and south latitude (Klimova et al. 2023).

Since its introduction to China in 1901, sisal has undergone testing and demonstration and has become an important fiber crop in tropical regions. Currently, sisal is mainly distributed in southern China, such as Guangxi, followed by Guangdong, and Yunnan, while regions like Sichuan, Hainan, and other provinces are also actively introducing sisal. The main product of sisal is the hard fiber extracted from the leaves. This fiber is white in color, tough in texture, elastic, strong in tension, resistant to friction, and not easy to break, and because it contains less glue, it is not slippery. Because of the prementioned characteristics of sisal, its fiber can be used to make ropes for ships and fishing boats, tires of airplanes and automobiles, cores of steel ropes for drilling and cranes, conveyor belts of machines, protective nets and other products,



which can be also woven into sacks, carpets, hats, and other daily necessities and used in plastic pressed hard boards as building materials, so they have great economic value and are widely used in national defense, fishery, forest industry, and other sectors (Damiao Xavier et al. 2018).

Sisal residue is a by-product of sisal fresh leaves after extracting the fiber. Sisal leaves only contain 3–5% hard fiber, and the remaining 95–97% are by-products. After the sisal leaf is scraped to obtain fiber, the remaining leaf cuticle, epidermal layer, palisade tissue, and spongy tissue are called sisal residue. A large quantity of sisal residues is produced annually in China. The average annual sisal residue yield in the south of China is 2.1 m. With the continuous development of sisal fiber production, the comprehensive utilization of its by-products after blade processing has expanded. It is a carrier of material, energy and nutrients, and a precious natural resource (Wang, Y. C. et al. 2014, Oliveira do Carmo et al. 2021). Sisal residues are rich in nutrients such as carbon, nitrogen, phosphorus, and potassium.

In modern agricultural production, returning crop residue to the field is one of the techniques to improve the ecological environment of sisal fields. The decomposing of crop residue after returning to the field plays an important role in improving soil fertility and promoting crop growth (Witt et al. 2012, Chen et al. 2022a, 2022b). The interaction of straw return and potassium fertilizer increased maize lodging resistance and yield (Liu et al. 2023). Long-term straw returning increased soil available K and slowly available K under rice and wheat cultivation (Zhang et al. 2021). Crop straw return can effectively increase the soil aggregate structure and improve soil enzyme activity by increasing the secretion of soil enzymes and affecting the soil microbial community (Hu et al. 2021, Miao et al. 2021).

Soil microorganisms are the driving force for the transformation and circulation of organic matter and nutrients in the straw returning ecosystem. They participate in the decomposition of organic matter and the formation of humus, and regulate various biochemical processes, such as energy and nutrient cycling, in the soil (Mwafulirwa et al. 2021, Wu et al. 2021). Crop residues incorporated into the soil affect different soil microorganisms (Chen et al. 2021).

Research on the effect of sisal residue returning on soil fertility and crop yield is very limited. At present, it has been reported that the return of sisal residue to the field is a crucial yield-increasing measure for sisal cultivation (Yang et al. 2017). Nevertheless, there are few reports on the overall impact of the sisal residue return on crop productivity, soil nutrient, soil enzyme activity and microorganisms. We hypothesized that sisal residue returning could increase crop yield by changing soil physicochemical properties and microbial community structure. To examine the actual effects, we designed a four-year field experiment consisting of two treatments of sisal residue returning under chemical fertilizer application in south of China. The aims of the present study were to investigate the changes of sisal yield, soil fertility, enzyme activity, and soil microorganisms after returning sisal residue to the field, to reveal how it may affect the sisal yield, and to provide a reference for the rational use of sisal residues.

MATERIALS AND METHODS

Experimental site

The sisal residue returning experiment was carried out at Shanxu state-owned farm of Guangxi Agricultural Reclamation, in Fusui county (south China, 22°53'N latitude, 107°20'E longitude; 115 m above sea level). The experimental site is characterized by subtropical monsoon climate with average annual temperature of 21.3–22.8°C. The lowest and highest temperature recorded over the years is -0.6 and 39.5°C, respectively. The average annual precipitation ranges from 1,050 to 1,300 mm. The total annual radiation is 108.4 kcal·cm⁻²; the average annual sunshine duration is 1,693 h, and the frost-free period lasts up to 346 days. The soil of the experimental site was classified as lateritic red earth derived from arenaceous shale. At the depth of 0–20 cm, the basic soil properties prior to the experiment were as follows: 7.26 pH, 2.45% soil organic matter, 0.13% total N, 5.14 mg·kg⁻¹ available phosphorus (P), and 82.14 mg·kg⁻¹ available potassium (K). Before 2011, the experimental field was used for interplanting spring corn with summer soybean. An annual amount of NPK (15-15-15) fertilizer (Sinochem, China) of 375 kg·ha⁻¹ was applied to the field as the base fertilizer before spring corn was grown.

Experimental design

The sisal cultivar H.11648 was cultivated in the field in November 2011. Sisal seedlings with a similar size (about 2.5 kg) were selected and transplanted in wide-narrow rows, which is a planting pattern of double rows. The planting specification was $(3.8+1.0) \times 1$ m (wide-narrow row distance and plant spacing). The planting density was 4,200 plants per hectare. In each plot (14.4×10.4 m), 60 sisal seedlings were planted with three double rows. All experimental fields received uniform water and fertilizer management from 2011 to 2014. In December 2014, the perennial sisal in the experimental field reached the harvest standard of 100 growing leaves. Since 2015, the same sisal plants were annually harvested for fiber extraction every January, which continues until 10 years later, when the sisal lifespan ends. Mature leaves of sisal were harvested manually by cutting with a sharp specialized sickle at the leaf base during mid-January coinciding with the inactive growth of sisal at low environment temperatures. Whole leaves with an angle greater than 45° below the central leaf were collected to measure and calculate the annual fresh leaf yield. Fifty leaves were left behind in the plant to maintain photosynthesis after harvest.

Sisal residue was obtained as a by-product. The organic matter content of the sisal residue was 21.30%, the total nitrogen content was 0.42%, the P_2O_5 content was 0.14%, the K_2O content was 0.52%, and the carbon–nitrogen ratio was 50.71:1. The sisal residue returning experiment was conducted from January 2015 to January 2019. The experimental setup followed a completely random block design with four replications. The area for each plot was 150 m².

The experimental treatments conducted once a year was divided into two groups. The treatments were based on the planting on sisal that received only chemical fertilizer as control (Ctrl) and sisal residue mulching plus chemical fertilizer (Sr). The same treatment plots were maintained for the duration of the four-year experiment. The NPK (12-20-7) fertilizers (Sinochem, China) were broadcast over the soil at rates of 500 kg·ha⁻¹ after annual sisal leaf harvest. Sisal residue was directly returned to the field and mulched on the ground surface at 36 t·ha⁻¹. The sisal residues were spread manually on top of the soil as a mulch. Other field management procedures were the same as in traditional sisal cultivation.

Every year, sisal residue was returned to the field following leaf yield assessment and soil sampling conducted in January. Except for the border row plants, 10 normal-grown plants were randomly selected from each plot at the leaf harvest stage.

Soil sample collection and analysis

After sisal leaf harvest and before the experimental treatments, soil samples were taken from the control and the sisal residue returning plots in January 2016, 2017, and 2018. Surface soil subsamples (0–40-cm depth) were collected from each plot with a corner of 40-mm internal diameter and 50-mm height using a diagonal sampling approach (Tian et al. 2023). Five subsamples collected from the same plot were pooled to form a composite sample, which served as one biological replicate. Every treatment consisted of four replicates. Four composite soil samples were obtained for each treatment, forming a total of eight soil samples. All samples were divided into two parts and kept at 4°C.

The soil was air-dried, homogenized and sieve (< 2 mm) to determine its physical and chemical properties. The methods for determining soil nutrients and enzyme activities are as follows: the soil pH was measured at soil/water ratio of 1/2.5 (w/v) (Lu 2000); the total nitrogen was determined by Kjeldahl method (Abrams et al. 2014); the available phosphorus and the available K were determined as described by Gong et al. (2018) and Meng et al. (2014), respectively.

In order to understand soil nutrient cycling dynamics, we determined the activities of sucrase, urease, catalase, and acid phosphate activities. The soil urease activity was measured using the indophenol colorimetry method (Ji et al. 2014); the catalase activity was determined by ultraviolet absorption method (Yang et al. 2011); the activity of sucrase was determined by the 3, 5-dinitrosalicylic acid colorimetry method (Ji et al. 2014), while the acid phosphatase activity was determined by the sodium p-nitrophenyl phosphate method (Du et al. 2018). Assessment of soil enzyme activities was conducted during each year of the experiment.



High-throughput sequencing

To investigate the changes of soil microbial community structure, we performed 16S rRNA and internal transcribed spacer (ITS) sequencing on soil samples. High-throughput sequencing was conducted to analyze the microbial community structure of soil sampled in 2018 (the third year after the initial residue treatment). Microbial DNA was extracted from the soil samples using TIANamp Soil DNA Kit (Tiangen, Beijing, China) according to the manufacturer protocols. A SmartSpec™ Plus spectrophotometer (Bio-Rad, United States of America) was used to evaluate the DNA quality based on the absorbance ratios of 260/280 nm and 260/230 nm. The extracted DNA was stored at -20°C before being used.

The 16S ribosomal DNA (rDNA) V3-V4 region and ITS region of the ribosomal RNA (rRNA) gene were amplified by polymerase chain reaction (PCR). PCR reactions were performed in triplicate using a 50- μ L mixture containing 5 μ L of 10 \times KOD Buffer, 5 μ L of 2.5 mM dNTPs, 1.5 μ L of each primer (5 μ M), 1 μ L of KOD Polymerase, and 100 ng of template DNA.

The universal primers 341 F (5'-CCTACGGGNGGCWGCAG-3') and 806 R (5'-GGACTACHVGGGTATCTAAT-3') were used to amplify the V3-V4 region of the 16S rRNA gene in each sample for the evaluation of bacterial abundance of 16S rRNA gene (Nossa et al. 2010), while ITS3_KYO2F(5'-GATGAAGAACGYAGYRAA-3') and IST4-R(5'-TCCTCCGCTTATTGATATGC-3') were used to amplify fungal ITS2 gene (Turenne et al. 1999).

Thermal cycling was performed as follows: initial denaturation at 95°C for 2 min, followed by 30 cycles of 10 s at 98°C, annealing for 30 s at 62°C (for 16S) or 52°C (for ITS), and extension at 68°C for 30 s. The final extension was conducted at 68°C for 10 min, followed by a holding step at 4°C.

The PCR amplicons were combined in equimolar ratios, and sequencing was conducted by Genedenovo (Guangzhou, China) on an Illumina HiSeq2500 platform with separate sequencing runs for the 16S and ITS rRNA gene amplicon pools. The sequencing data were processed using the UPARSE pipeline (http://drive5.com/usearch/manual/uparse_pipeline.html). The raw sequences were subjected to quality control. The singleton and chimeric sequences were removed after dereplication, and the remaining sequences were categorized into operational taxonomic units (OTU) with 97% similarity and then assigned taxonomy using the Silva (<https://www.arb-silva.de/>) and the UNITE databases (<https://unite.ut.ee/>) for the 16S and ITS rRNA genes, respectively.

Data analysis

The relative abundance of each microbial species type was expressed as a percentage. The α -diversity of species was determined by calculating the Shannon, Chao, and Simpson indices of a single sampling site (Wang, Y. K. et al. 2014), while the β -diversity of species was analyzed using PCoA based on FastUnifrac (Hamady et al. 2010). The functional analysis of the microbiome was performed using PICRUST software (Langille et al. 2013). The differences between the two groups (control and sisal residue returning groups) were analyzed by single factor analysis of variance (ANOVA) and minimum significant difference (LSD) test. The Mantel's test and Spearman's correlation were used to calculate the correlation coefficient. All statistical analysis was conducted using the Vegan package (v.2.4-1) in R software (version 3.3.2).

RESULTS

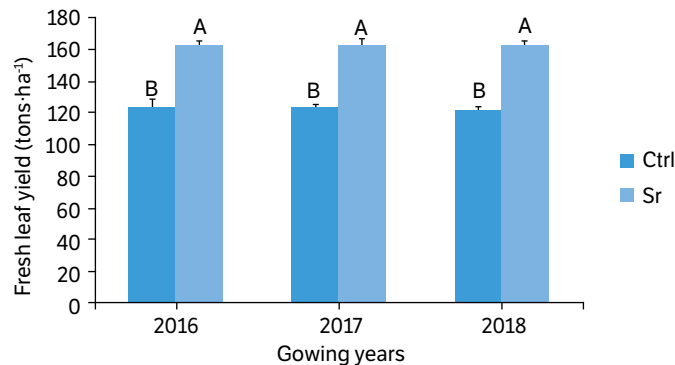
Effect of sisal residue return on sisal yield

The sisal leaf width and thickness showed a significant difference when treated with sisal residue in comparison to the control group (Table 1). The variance analysis found that year and year \times treatment interaction had no significant differences in sisal growth traits. Compared to the control group, the annual yield of sisal following sisal residue treatment showed a large significant difference and increased by 30.77, 31.97, and 33.36%, respectively, between 2016 and 2018 (Fig. 1). This indicates that returning sisal residue to the field can increase sisal yields.

Table 1. Average annual effect of returning sisal residue to the soil on leaf size, 2016–2018*.

Treatment	Leaf length	Leaf width	Leaf thickness
Sr	133.1 ± 2.28a	13.14 ± 0.24a	0.214 ± 0.02a
Ctrl	132.8 ± 1.48a	13.42 ± 0.06b	0.231 ± 0.01b

Sr: annual sisal residue mulch application, plus chemical fertilizer application; Ctrl: control, chemical fertilizer application alone; *different lowercase letters in each column indicate significant difference at 0.05 level according to minimum significant difference test. Data indicates mean ± standard deviation.

**Figure 1.** The mean annual yield of sisal in the control (Ctrl) and the sisal residue returning treatment (Sr). Different capital letters in the same column indicate significant difference at 0.01 level between the treatments in the same year. Vertical bars correspond to standard error.

Effects of sisal on soil nutrients and enzyme activities

In order to evaluate soil microbial activity and reflect the function of soil nutrient cycling, the activity of catalase, sucrase, urease, and phosphatase activity was detected. The results showed that, compared with the control group, returning sisal residue to the field increased the content of available phosphorus, total nitrogen, and available potassium by 3.02, 46.59, and 1.21%, respectively, over the three-year period. The soil pH showed a statistically significant increase under sisal residue returning condition (Table 2). Returning sisal residue to the field significantly increased catalase, urease, and acid phosphatase enzyme activities, which were 2.01, 17.20 and 17.40% ($p < 0.05$) higher than the control group, respectively. In the same way, the sisal residue significantly improved the activity of sucrase enzyme by up to 16.60% (Table 2).

Table 2. Effects of long-term sisal residue returning on soil biochemical properties¹.

Treatment	Year	pH	Available P mg·kg ⁻¹	Available K mg·kg ⁻¹	Total N %	Sucrase mg·d ⁻¹ ·g ⁻¹	Urease μg·d ⁻¹ ·g ⁻¹	Catalase μmol·d ⁻¹ ·g ⁻¹	Acid phosphatase μmol·d ⁻¹ ·g ⁻¹
Ctrl	2016	7.15c	28.51c	145.56b	0.12b	25.21b	999.73c	54.81d	16.86c
	2017	7.13c	28.98bc	147.22ab	0.12b	25.89b	972.24c	56.63ab	16.65c
	2018	7.25b	29.01bc	147.24ab	0.12b	25.95b	1,013.27bc	55.80c	17.17c
Sr	2016	7.53a	29.30abc	148.63a	0.17a	29.19ab	1,117.49ab	57.21a	19.15b
	2017	7.52a	30.14a	148.55a	0.17a	26.97b	1,228.78a	56.14bc	20.43a
	2018	7.56a	29.67ab	148.17a	0.18a	33.69a	1,152.46a	57.2521a	19.92ab
F-value	Treatment (T)	363.53**	11.17**	11.96**	751.97**	10.15**	34.70**	33.47**	204.98**
	Year (Y)	6.68*	2.20ns	0.86ns	0.20ns	2.35ns	0.70ns	2.51ns	3.02ns
	Y×T	1.46ns	0.32ns	1.63ns	2.16ns	2.08ns	2.20ns	19.15**	4.67*

Sr: annual sisal residue mulch application, plus chemical fertilizer application; Ctrl: control, chemical fertilizer application alone; ¹same lowercase letters in each column indicate no significant difference according to minimum significant difference test. Data indicates mean ± standard deviation; *significant difference at the 0.05-probability levels; **significant difference at the 0.01-probability levels; ns: not significant.



Effect of sisal residue return on soil microorganisms

After quality filtering, 4,930 OTUs were identified using 16S rRNA sequencing, and 1,533 OTUs were identified using ITS sequencing. While the fungal abundance in the sisal residue returning group showed a decreasing trend compared to the control group, the bacterial abundance in both the control and sisal residue returning groups was consistently higher than the fungal abundance. We observed that the sisal residue return was associated with a higher bacteria/fungi ratio (Fig. 2). The sequencing data showed that the sisal residue return caused significant changes in the bacterial and fungus communities. Specifically, Bacteroidetes, Acidobacteria, Chloroflexi, Proteobacteria, Actinobacteria, and several other phyla were more abundant in the control group, while Firmicutes, Verrucomicrobia, Gemmatimonadetes, Planctomycetes, Nitrospirae, and various other phyla were more prevalent in the sisal residue returning group (Fig. 3a). For the soil fungi, the most abundant fungal phyla in the soil treated with sisal residue were Ascomycota, Chlorophyta, Basidiomycota, and Anthophyta. Among them, Ascomycota was the most dominant phylum, with a relative abundance of 76.15–77.60%; whereas Basidiomycota and Anthophyta showed a higher relative abundance in the control group (Fig. 3b).

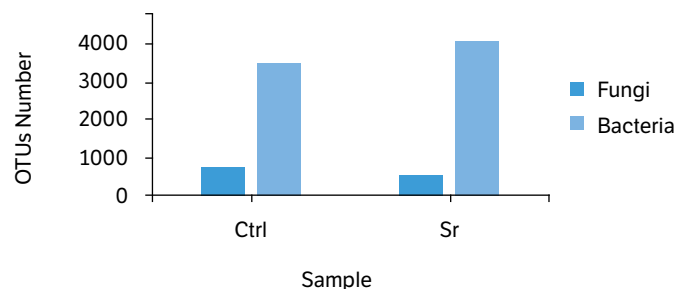


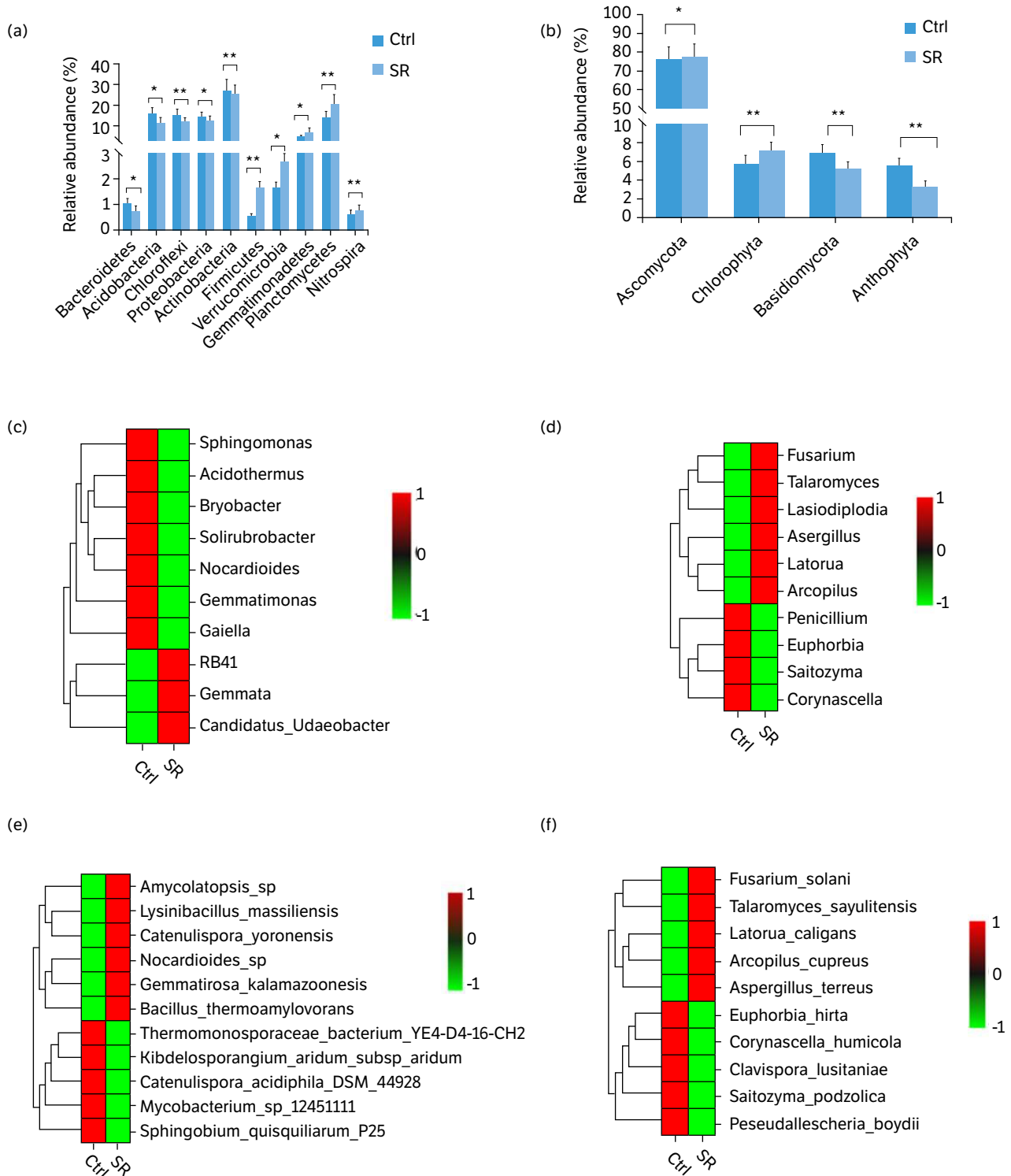
Figure 2. Soil bacteria and fungi operational taxonomic units number in control (Ctrl) and sisal residue returning group (Sr) in 2018. All data from high-throughput sequencing were collected in 2018.

Ten of the most abundant genera of soil bacteria were identified and classified (Fig. 3c). The relative abundance of RB41, *Gemmata*, and *Candidatus Udaeobacter* was significantly higher in the sisal residue returning group compared to the control. The classification of soil fungi genera revealed that *Fusarium*, *Talaromyces*, *Aspergillus*, *Lasiodiplodia*, and *Latorua* were more dominant in the sisal residue returning group (Fig. 3d).

Additionally, we also studied soil bacteria and fungi at the species level and found that, among the soil bacteria, the relative abundance of *Amycolatopsis* sp., *Nocardioides* sp., *Bacillus thermoamylovorans*, *Gemmatirosa kalamazoonesis*, *Lysinibacillus massiliensis*, and *Catenulispora yoronensis* in the sisal residue returning group was significantly higher than the control (Fig. 3e). For soil fungi, *Fusarium solani*, *Talaromyces sayulitensis*, *Latorua caligans*, and *Arcopilus cupreus* had higher relative abundances in the sisal residue returning group (Fig. 3f).

Effect of sisal residue return on microbial diversity

The α -diversity indices of soil bacterial and fungal community were calculated, as shown in Table 3. The α -diversity of bacterial communities in the field treated with sisal residue was significantly higher than that of the control group. For the fungal community, returning sisal residue to the field resulted in a lower soil microbial α -diversity (Table 3).



* $p < 0.05$; ** $p < 0.01$.

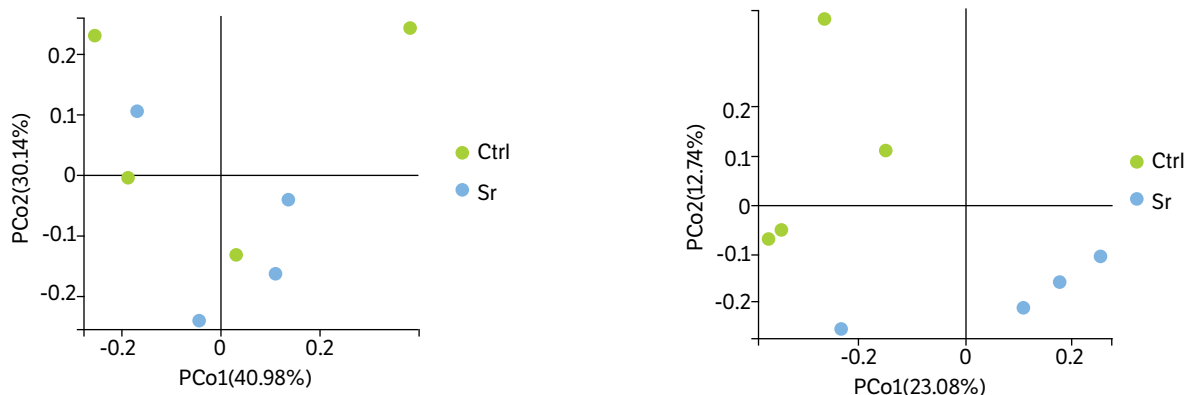
Figure 3. Changes of microbial composition after returning sisal residue to the field in 2018. Bar graph of relative abundance statistics of (a) bacteria phylum and (b) fungi phylum. Heat map of relative abundance statistics of (c) bacteria genus, (d) fungi genus, (e) bacterial species and (f) fungal species. All data from high-throughput sequencing were collected in 2018.

Table 3. α -diversity indices analysis of microbial community in 2018 conducted three years after initial and subsequent annual sisal residue applications.

Treatment	Bacteria				Fungi			
	Diversity index		Richness index		Diversity index		Richness index	
	Shannon	Simpson	Sobs	Chao	Shannon	Simpson	Sobs	Chao
Ctrl	9.53 ± 0.42	0.996 ± 0	3,521.00 ± 563.87	4,149.89 ± 698.31	6.14 ± 0.62	0.95 ± 0.04	700.75 ± 78.21	845.53 ± 79.05
Sr	9.90 ± 0.13	0.997 ± 0	4,078.50 ± 380.01	4,790.84 ± 422.07	5.76 ± 0.74	0.95 ± 0.03	508.75 ± 97.33	677.92 ± 96.95

*Data are mean ± standard deviation, n = 4; Sr: annual sisal residue mulch application, plus chemical fertilizer application; Ctrl: control, chemical fertilizer application alone.

In the resulting PCoA scatter plot, the greater distance between points indicated a higher dissimilarity in the microbial communities. Conversely, microbial communities with similar compositions were clustered together. At the genus level, the bacterial community showed no noticeable change before and after the sisal residue treatment (Fig. 4a), whereas significant change was observed for the fungal microbiome (Fig. 4b). The microbial analysis shows that the return of sisal residue to the field has a higher impact on the β -diversity of fungi.

**Figure 4.** β -diversity analysis of the microbial community in 2018 conducted three years after initial and subsequent annual sisal residue applications. (a) Bacterial community; (b) fungal community.

Relationship between microbial α -diversity and sisal productivity

Certain differences in environmental factors affect the structure and diversity of microbial communities in different habitats. To investigate how microbial changes respond to environmental factors, the Spearman's rank correlations between the environmental factors of soil and the diversity of bacteria and fungi were analyzed. The activity of the soil enzymes (sucrose, catalase, acid phosphatase) was found to be positively correlated with bacterial abundance, but not with fungal abundance (Table 4).

Table 4. Spearman's correlation analysis of environmental factors and soil microbial diversity index.

Environmental factors	Bacteria				Fungi			
	Shannon	Simpson	Chao	Ace	Shannon	Simpson	Chao	Ace
Available P	1.00**	0.83*	0.94**	0.71	-0.60	-0.31	-0.83*	-0.83*
Sucrase	0.83*	0.83*	0.66	0.37	-0.66	-0.43	-0.66	-0.66
Urease	0.60	0.77	0.37	-0.09	-0.77	-0.71	-0.77	-0.77

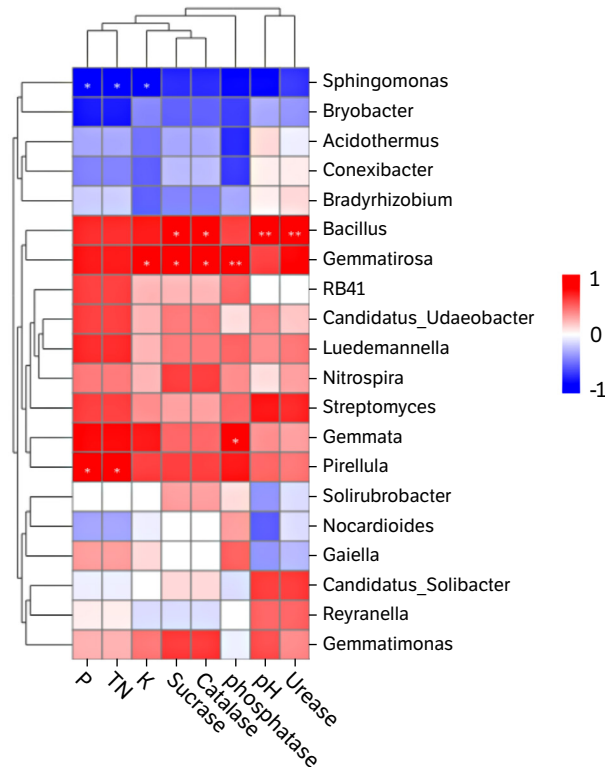
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Table 4. Continuation...

Environmental factors	Bacteria				Fungi			
	Shannon	Simpson	Chao	Ace	Shannon	Simpson	Chao	Ace
Catalase	0.83*	0.83*	0.66	0.37	-0.66	-0.43	-0.66	-0.66
Acid phosphatase	0.77	0.94**	0.60	0.26	-0.71	-0.54	-0.60	-0.60
pH	0.77	0.60	0.60	0.26	-0.486	-0.31	-0.94**	-0.94**
Total N	1.00**	0.83*	0.94**	0.71	-0.6	-0.31	-0.83*	-0.83*
Available K	0.89*	0.71	0.71	0.49	-0.37	-0.09	-0.71	-0.71

* $p < 0.05$: different significance at the 0.05-probability level; ** $p < 0.01$: different significance at the 0.01 probability level.

The genus *Sphingomonas* is one of the probiotic microorganisms characterized with degradation of aromatic and xenobiotic compounds, promotion of nutrient recycling and resistance to multiple pathogens (Liu, Z. et al. 2021). It has been reported that several *Sphingomonas* strains with the characteristics of dehydrogenation and nitrogen fixation play a vital role in maintaining the nitrogen balance of soil (Liu, H. et al. 2021). In this study, the relative abundance of *Sphingomonas* bacteria was significantly negatively correlated with soil available P, total N, and available K ($p < 0.05$) (Fig. 5).



* $p < 0.05$; ** $p < 0.01$; TN: total N; P: available P; K: available K.

Figure 5. Correlation heatmap of soil properties and relative bacterial abundances of dominant genus. The color intensity indicates the correlation between soil property and relative abundance of each genus.

Functional analysis of bacterial community

In the current study, a functional analysis of the bacterial community revealed that the microbiota related to transcription, cell motility, and biosynthetic pathways of secondary metabolites were more abundant in the sisal residue returning group compared to the control (Fig. 6).

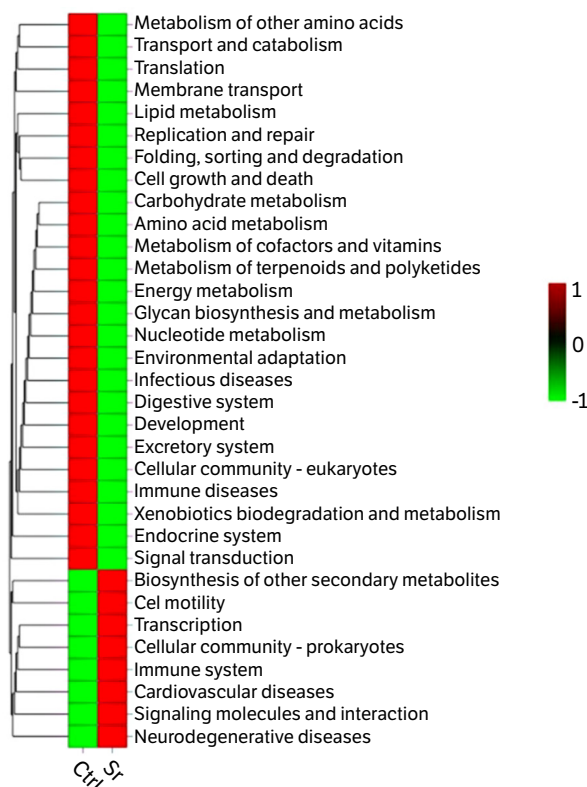


Figure 6. Function analysis of bacterial community in soils receiving annual crop residue applications from sisal.

DISCUSSION

The current results suggest that leaf width and thickness are the main factors that influence fresh leaf weight and yield. Previous studies have shown that returning straw to the field can improve soil fertility and crop yield (Liu et al. 2023, Wen et al. 2019). Various studies have also reported improved enzyme activities in response to crop residue return. The improved soil environment can provide nutrient elements, resulting in increased sisal production (Ji et al. 2014, Huang et al. 2016). In this study, sisal residue return significantly increased the level of N, and soluble P and K in the soil, which was comparable to the findings of Yamoah et al. (2002) and Yang et al. (2017). The significant correlation between the content of NPK and the relative abundance of microorganism suggest that the return of sisal residue to the field can affect soil microbial community structure. The sisal residue treatment also significantly increased the activities of soil sucrase, catalase, and acid phosphatase, which suggests that the secretion of soil enzymes was more active.

The present research indicates that the return of sisal residue to the field affects soil microbial activity and improves soil fertility. Several studies have reported similar findings on increased yields in response to straw return in other crops (Wen et al. 2019, Yang et al. 2018). Similarly, soil quality and sisal growth were positively affected by the deep-buried return of sisal stem waste (Tan et al. 2019). However, the present study is the first report incorporating microbial analysis after sisal residue applications.

Returning of sisal residue to the field significantly affected the composition of soil microorganisms. In the current research, we observed that the relative abundance of soil bacteria increased, while the relative abundance of soil fungi showed a downward trend after the sisal residue return. Additionally, a high bacteria/fungi ratio also indicated bacteria as the dominant microorganisms in the soil. Fungi usually prefer a more stable ecosystem (Tolkkinen et al. 2015), and adding sisal residue back to the field causes significant changes in the soil environment, which affects the ecosystem's stability.

Regarding the impact of sisal residue on the composition of microbial communities, we observed that different microbial taxa exhibited different behaviors. Firmicutes were relatively more prevalent in the sisal residue returning group. Firmicutes are beneficial bacteria involved in the decomposition of cellulose-containing materials, which is in agreement with the carbon-rich nature of decomposed straw (Pu et al. 2020, Zhu et al. 2014). The control group showed a higher relative abundance of Acidobacteria. This observation aligns with previous research demonstrating that Acidobacteria is typically dominant in soil environments with low nutrient content (Yang et al. 2019).

Nitrospirae not only oxidizes nitrite, but may also oxidize ammonia (Daims et al. 2015), which ultimately affects crop productivity by stimulating the nitrogen cycle in the soil (Longa et al. 2017). In the present study, sisal residue return resulted in significant increases in relative abundances of Nitrospirae. The result is similar to previous research demonstrating a higher relative abundance of Nitrospirae in the soil treated with maize straw return (Chen et al. 2017). A consistent correlation between taxonomic classification and the microbial communities was observed, such as the presence of Planctomycetes (phylum), *Gemmata* (genus) and *G. kalamazoonesis* (species) in the bacterial community; and of Ascomycotina (phylum), *Fusarium* (genus), and *F. solani* (species) in the fungal community. The soil microbial analysis indicates that *G. kalamazoonesis* and *F. solani* may be primary indicators of microorganisms following the reintroduction of sisal residue into the field.

Fungi adapt slowly to change in their ecological environment. In the current study, returning sisal residue to the field increased soil nitrogen, phosphorus, and potassium levels, which may have contributed to fungal β -diversity increases (Chen et al. 2020). Additionally, following a functional prediction of the microbial community differentiation, it was found that the biological pathway of secondary metabolites synthesis was more dominant in the sisal residue returning group compared to the control group. Secondary metabolites have broad-spectrum activities against a variety of pathogens (Maddox et al. 2010). However, the potential impact of secondary metabolites released from sisal residues on disease suppression needs further verification as our study did not evaluate the effect of residues on disease incidence in sisal.

The diversity of the soil bacteria plays a vital role in maintaining agroecosystem stability and improving crop resistance, growth, and yield (Bossio et al. 1998). As a result, in the current study, a high bacterial/fungal ratio may have or likely contributed positively to sisal production after sisal residue return. The increase of soil microbial diversity not only inhibits soil-borne diseases, but also improves the supply capacity of soil nitrogen (Weidner et al. 2015), which may partially explain the observed relationship between the increase of bacterial α -diversity and sisal yield after returning sisal residue to the field.

CONCLUSION

In summary, we found that returning sisal residue to the field affects sisal productivity by possibly regulating the soil microbial community and nutrient turnover, and that the increase in sisal productivity may be primarily determined by microbial α -diversity. Further research is required to confirm our initial observation from this four-year study. The findings presented herein support the practice of returning sisal residues as a means of improving crop productivity and soil fertility.

CONFLICT OF INTEREST

Nothing to declare.

AUTHORS' CONTRIBUTION

Conceptualization: Jin, G. and Chen, T.; **Methodology:** Chen, T.; **Investigation:** Huang, X., Wu, M., Huang, C., Qin, X., Jiang, Y. and Peng, X.; **Data curation:** Zhong, J. and Chen, L.; **Writing – Original Draft:** Jin, G. and Chen, T.; **Writing – Review and Editing:** Jin, G. and Huang, X.; **Funding Acquisition:** Chen, T.; **Supervision:** Jin, G. and Chen, T.



DATA AVAILABILITY STATEMENT

The data used to support the findings of this study are available from the corresponding author upon request.

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