SUGARCANE STRAW REMOVAL AND IRRIGATION SYSTEM SHAPE ARBUSCULAR MYCORRHIZAL FUNGI AND RHIZOSPHERE MICROORGANISMS IN THE CERRADO

por

Thaynara da Silva Barbosa

Dissertação apresentada ao Programa de Pós-Graduação em Bioenergia e Grãos do Instituto Federal de Educação, Ciência e Tecnologia Goiano – Campus Rio Verde, como parte dos requisitos para obtenção do grau de Mestre em Bioenergia e Grãos.

RIO VERDE - GO
agosto – 2026

SUGARCANE STRAW REMOVAL AND IRRIGATION SYSTEM SHAPE ARBUSCULAR MYCORRHIZAL FUNGI AND RHIZOSPHERE MICROORGANISMS IN THE CERRADO

por

Thaynara da Silva Barbosa

Comitê de Orientação:

Aurélio Rubio Neto, Prof. Dr. – IF Goiano

RIO VERDE - GO

agosto – 2026

FICHA CATALOGRÁFICA

Silva Barbosa, Thaynara
B238s SUGARCANE STRAW REMOVAL AND IRRIGATION SYSTEM SHAPE ARBUSCULAR MYCORRHIZAL FUNGI AND RHIZOSPHERE MICROORGANISMS IN THE CERRADO / Thaynara Silva Barbosa. Rio Verde 2026.

38f. il.

Orientador: Prof. Dr. Aurélio Rubio Neto.
Coorientador: Prof. Dr. Jadson Belém de Moura.
Dissertação (Mestre) - Instituto Federal Goiano, curso de 0233154 - Mestrado Profissional em Bioenergia e Grãos - Integral (Campus Rio Verde).
I. Título.

PÁGINA DE APROVAÇÃO

SERVIÇO PÚBLICO FEDERAL
MINISTÉRIO DA EDUCAÇÃO
SECRETARIA DE EDUCAÇÃO PROFISSIONAL E TECNOLÓGICA
INSTITUTO FEDERAL DE EDUCAÇÃO, CIÊNCIA E TECNOLOGIA GOIANO

Documentos 29/2026 - SREPG/CMPR/CPG-RV/DPGPI-RV/CMPRV/IFGOIANO

SUGARCANE STRAW REMOVAL AND IRRIGATION SYSTEM SHAPE ARBUSCULAR MYCORRHIZAL FUNGI AND RHIZOSPHERE MICROORGANISMS IN THE CERRADO

Autora: Thaynara da Silva Barbosa
Orientador: Aurélio Rubio Neto

TITULAÇÃO: Mestre em Bioenergia e Grãos - Área de Concentração Agroenergia

APROVADA em 28 de agosto de 2026.

Assinado eletronicamente
Prof. Dr. Jadson Belém de Moura
Avaliador externo - Centro Universitário Evangélico de Goianésia

Assinado eletronicamente
Prof. Dr. Pablo Diego Silva Cabral
Avaliador interno - IF Goiano Campus Rio Verde

Assinado eletronicamente
Prof. Dr. Wagner Gonçalves Vieira Junior
Avaliador externo - Centro Universitário Evangélico de Goianésia

Assinado eletronicamente
Profa. Dra. Silvia Sanielle Costa de Oliveira
Avaliadora interna - IF Goiano Campus Rio Verde

Assinado eletronicamente
Prof. Dr. Aurélio Rubio Neto
Presidente da Banca - IF Goiano Campus Rio Verde

Documento assinado eletronicamente por:

- **Aurelio Rubio Neto**, **PROFESSOR ENS BASICO TECN TECNOLOGICO** , em 05/10/2026 12:04:56.
- **Wagner Goncalves Vieira Junior**, **PROF ENS BAS TEC TECNOLOGICO-SUBSTITUTO**, em 05/10/2026 14:17:32.
- **Jadson Belem de Moura**, **Jadson Belem de Moura - Professor Avaliador de Banca - Instituto Federal Goiano - Campus Rio Verde (10651417000500)**, em 05/10/2026 14:26:19.
- **Silvia Sanielle Costa de Oliveira**, **PROFESSOR ENS BASICO TECN TECNOLOGICO** , em 05/10/2026 15:01:33.
- **Pablo Diego Silva Cabral**, **PROFESSOR ENS BASICO TECN TECNOLOGICO** , em 05/10/2026 15:35:33.

Este documento foi emitido pelo SUAP em 27/08/2026. Para comprovar sua autenticidade, faça a leitura do QRCode ao lado ou acesse https://suap.ifgoiano.edu.br/autenticar-documento/ e forneça os dados abaixo:

Código Verificador: 853204
Código de Autenticação: 67a3256936

INSTITUTO FEDERAL GOIANO
Campus Rio Verde
Rodovia Sul Goiana, Km 01, Zona Rural, 01, Zona Rural, RIO VERDE / GO, CEP 75901-970
(64) 3624-1000

TERMO DE CIÊNCIA E DE AUTORIZAÇÃO

INSTITUTO FEDERAL
Goiano

Repositório Institucional do IF Goiano - RIIF Goiano
Sistema Integrado de Bibliotecas

TERMO DE CIÊNCIA E DE AUTORIZAÇÃO
PARA DISPONIBILIZAR PRODUÇÕES TÉCNICO-CIENTÍFICAS NO REPOSITÓRIO INSTITUCIONAL DO IF GOIANO

Com base no disposto na Lei Federal nº 9.610, de 19 de fevereiro de 1998, AUTORIZO o Instituto Federal de Educação, Ciência e Tecnologia Goiano a disponibilizar gratuitamente o documento em formato digital no Repositório Institucional do IF Goiano (RIIF Goiano), sem ressarcimento de direitos autorais, conforme permissão assinada abaixo, para fins de leitura, download e impressão, a título de divulgação da produção técnico-científica no IF Goiano.

IDENTIFICAÇÃO DA PRODUÇÃO TÉCNICO-CIENTÍFICA

☐ ■ Artigo científico ☐ Capítulo de livro ☐ Livro
■ Tese (doutorado) ☐ Dissertação (mestrado) ☐ Monografia (especialização) ☐ TCC (graduação)
☐ ☐ Trabalho apresentado em evento
☐ Produto técnico e educacional - Tipo:

Nome completo do autor:

Thaynara da Silva Barbosa	Matrícula: 2024202331540021

Título do trabalho:

Sugarcane Straw Removal and Irrigation System Shape Arbuscular Mycorrhizal Fungi and Rhizosphere Microorganisms in the Cerrado

Assinatura do autor e/ou detentor dos direitos autorais

RESTRIÇÕES DE ACESSO AO DOCUMENTO

Documento confidencial: ■ Não ☐ Sim, justifique:

Informe a data que poderá ser disponibilizado no RIIF Goiano: 23/10/2026

O documento está sujeito a registro de patente? Sim ☐ Não ■
O documento pode vir a ser publicado como livro? Sim ☐ Não ■

DECLARAÇÃO DE DISTRIBUIÇÃO NÃO-EXCLU

O(a) referido(a) autor(a) declara:

Que o documento é seu trabalho original, detém os direitos autorais da produção técnico-científica e não infringe os direitos de qualquer outra pessoa ou entidade;

Que obteve autorização de quaisquer materiais inclusos no documento do qual não detém os direitos de autoria, para conceder ao Instituto Federal de Educação, Ciência e Tecnologia Goiano os direitos requeridos e que este material cujos direitos autorais são de terceiros, estão claramente identificados e reconhecidos no texto ou conteúdo do documento entregue;

Que cumpriu quaisquer obrigações exigidas por contrato ou acordo, caso o documento entregue seja baseado em trabalho financiado ou apoiado por outra instituição que não o Instituto Federal de Educação, Ciência e Tecnologia Goiano.

Goianésia (Local), 21 / 10 / 2026 (Data)

Ciente e de acordo:

Assinatura do(a) orientador(a)

gov.br Documento assinado digitalmente
AURELIO RUBIO NETO
Data: 22/10/2026 14:08:00-0300
Verifique em https://validar.iti.gov.br

gov.br Documento assinado digitalmente
THAYNARA DA SILVA BARBOSA
Data: 22/10/2026 13:34:26-0300
Verifique em https://validar.iti.gov.br

SUGARCANE STRAW REMOVAL AND IRRIGATION SYSTEM SHAPE ARBUSCULAR MYCORRHIZAL FUNGI AND RHIZOSPHERE MICROORGANISMS IN THE CERRADO

por

Thaynara da Silva Barbosa

Orientador : _____
Aurélio Rubio Neto, Prof. Dr. – IF Goiano

Coorientador : _____
Jadson Belém de Moura, Prof. Dr. – UNIEGO

Examinadores: _____
Wagner Gonçalves Vieira Junior, Dr. – UNIEGO

_____
Pablo Diego Silva Cabral , Dr. – IF Goiano

_____
Silvia Sanielle C. Oliveira – IF Goiano

Rio Verde, 28 de agosto de 2026.

AGRADECIMENTOS

Agradeço primeiramente a Deus, por guiar meus passos e conduzir nesta jornada, proporcionando saúde, paz e sabedoria. A Ele toda a glória.

Ao meu orientador, Prof. Dr. Aurélio Rubio Neto, pelo incentivo, paciência, orientação, amizade e compartilhamento de conhecimentos.

Ao meu coorientador, Prof. Dr. Jadson Belém de Moura, sou imensamente grata por sua contribuição, você foi fundamental para o meu aperfeiçoamento acadêmico e profissional.

A minha família, obrigada pelo apoio incondicional e constante incentivo. Em especial à minha mãe Telma, pai Gilberto e aos meus irmãos. Vocês são a base que sustenta minha vida e a fonte de motivação para seguir em frente.

Ao Instituto Federal Goiano-Campus Rio Verde, pela disponibilização de recursos essenciais para realização deste projeto.

Ao Centro Universitário Evangélico de Goianésia - UNIEGO, em especial ao Prof. Dr. Jadson, pela disponibilização dos recursos necessários a execução deste mestrado, incluindo o laboratório de microbiologia do grupo de pesquisa SEDMO. A todos os alunos da instituição, que tiveram envolvidos na execução das atividades de campo e laboratoriais e contribuíram para a condução deste trabalho.

A FAPEG e ao CEBIO (Cento de Excelência em Bioinsumos) na pessoa do coordenador regional da URB de Rio Verde, Dr. Edson Luiz Souchie, pela disponibilidade e pela concessão da bolsa de estudos.

A empresa Jalles, pela disponibilização da área experimental e pelo fornecimento do material utilizado na condução deste projeto.

A todos que, diretamente ou indiretamente, contribuíram para a realização deste mestrado, meus sinceros agradecimentos.

BIOGRAFIA DO AUTOR

Thaynara da Silva Barbosa, 27 anos, natural de Anápolis - Goiás, filha de Telma Francisca da Silva e Gilberto Gomes Barbosa. Graduada em agronomia pelo Centro Universitário Evangélico de Goianésia – UNIEGO.

Durante sua formação acadêmica, integrou, desde o início da graduação, o Grupo de Pesquisa Solos, Ecologia e Dinâmica da Matéria Orgânica - SEDMO, no qual desenvolveu atividades relacionadas à pesquisa científica, com ênfase em estudos voltados à ciência do solo, microbiologia agrícola e interações entre microrganismos e plantas. A participação no grupo de pesquisa contribuiu para sua formação científica e para o desenvolvimento de habilidades relacionadas à condução de experimentos, processamento de amostras, análise e interpretação de resultados.

Atualmente, exerce a função de técnica de campo na cadeia produtiva de olericultura do Serviço Nacional de Aprendizagem Rural – SENAR Goiás, atuando na prestação de assistência técnica e gerencial aos produtores rurais. Suas atividades profissionais envolvem o acompanhamento e planejamento da produção, manejo e conservação do solo, recomendação de práticas de adubação, manejo fitossanitário, planejamento das atividades agrícolas e orientação quanto à adoção de tecnologias e práticas de produção.

Sua trajetória acadêmica e profissional tem sido pautada pela integração entre conhecimento científico e aplicação prática no setor agropecuário, com atuação voltada ao aprimoramento dos sistemas de produção e à adoção de práticas agrícolas tecnicamente adequadas e sustentáveis.

SUMÁRIO

ÍNDICE DE TABELAS *11*
ÍNDICE DE FIGURAS *12*
LISTA DE SÍMBOLOS, SIGLAS, ABREVIAÇÕES E UNIDADES *13*
1. ABSTRACT *14*
2. RESUMO *13*
3. INTRODUCTION *16*
4. MATERIALS AND METHODS *18*
4.1 Study site and experimental design *17*
4.2 Mycorrhizal characterization *17*
4.3 Molecular quantification *18*
4.4 Soil chemical analysis *19*
4.5 Statistical analysis *19*
5. RESULTS *21*
5.1 Mycorrhizal responses *21*
5.2 Molecular target..s *24*
5.3 Pathogens and the balance of the community *26*
6. DISCUSSION *28*
7.CONCLUSIONS *31*
8. STATEMENTS AND DECLARATIONS *32*
8.1 Conflict of interest. *32*

ÍNDICE DE TABELAS

Página

Table 1 - Two-way analysis of variance for the arbuscular mycorrhizal fungal variables under three straw removal times and three irrigation systems in commercial sugarcane fields of the Cerrado. 22

Table 2 - Diversity indices of the arbuscular mycorrhizal fungal community under three straw removal times. 22

Table 3 - Molecular targets quantified by real-time PCR under three straw removal times and three irrigation systems. 25

ÍNDICE DE FIGURAS

Página

Figure 1 - Response of the arbuscular mycorrhizal fungal community to sugarcane straw removal time in commercial fields of the Cerrado. 23

Figure 2 - Composition of the arbuscular mycorrhizal fungal community under three straw removal times 24

Figure 3 - Principal coordinates analysis on the Bray-Curtis distance for a the community of arbuscular mycorrhizal fungal families and b the community of molecular targets, with samples coloured by straw removal time 25

Figure 4 - Abundance of six beneficial taxa quantified by real-time PCR as a function of straw removal time within each irrigation system. 26

Figure 5 - Soil-borne pathogens and the balance of the rhizosphere community. 27

LISTA DE SÍMBOLOS, SIGLAS, ABREVIAÇÕES E UNIDADES

Símbolo/sigla	Significado	Unidade
AMF	Arbuscular mycorrhizal fungi	-
DNA	Deoxyribonucleic acid	-
PCR	Polymerase chain reaction	-
qPCR	Quantitative polymerase chain reaction	-
CEC	Cation exchange capacity	cmolc kg^{-1}
V	Base saturation	%
P	Phosphorus	mg dm^{-3}
K	Potassium	mg dm^{-3}
Ca	Calcium	cmolc dm^{-3}
Mg	Magnesium	cmolc dm^{-3}

1. ABSTRACT

Removing sugarcane straw for second-generation bioenergy may compromise soil biological quality, but the effect is rarely evaluated together with the irrigation system that farms already operate. We assessed a 3 x 3 factorial combining straw removal time, that is no removal, one year and two consecutive years, with reel, centre pivot and rainfed systems in commercial fields of the Brazilian Cerrado. Arbuscular mycorrhizal fungi were characterized morphologically and six beneficial taxa and three soil-borne pathogens were quantified by real-time PCR. Mycorrhizal responses depended on straw removal alone, with no effect of irrigation and no interaction. Spore density and family richness fell to a minimum after one year and recovered by the second year to values indistinguishable from undisturbed soil, whereas root colonization did not change. Evenness declined only after two removals, as Acaulosporaceae rose disproportionately and came to dominate the community. Molecular targets behaved differently: most were governed by interactions between straw management and irrigation, so that a single removal effect cannot be stated for them. Pseudomonas fluorescens was the exception, disappearing wherever straw was removed regardless of system. The ratio between beneficial organisms and pathogens declined after two removals across all systems. Sporulation, colonization and spore-derived richness therefore diverge, and none of them alone reports soil biological quality reliably.

Keywords: Arbuscular mycorrhizal fungi, Bioenergy, Plant growth-promoting bacteria, Quantitative PCR, Soil-borne pathogens, Straw management.

2. RESUMO

A remoção da palha de cana-de-açúcar para a produção de bioenergia de segunda geração representa uma oportunidade econômica significativa, porém seus efeitos sobre a sustentabilidade biológica do solo permanecem incertos, especialmente no bioma Cerrado. Este estudo avaliou o impacto de diferentes tempos de remoção da palha (sem remoção, 1 ano e 2 anos consecutivos) sobre as comunidades de fungos micorrízicos arbusculares (FMA), microrganismos benéficos (bactérias promotoras de crescimento e fungos antagonistas) e atributos químicos do solo. A metodologia envolveu amostragem de solo na rizosfera, quantificação de esporos, avaliação da colonização radicular, identificação morfológica de FMA e análise molecular (qPCR) para quantificação de *Bacillus*, *Pseudomonas*, *Azospirillum brasilense* e *Trichoderma*. Os resultados indicaram que a remoção da palha por dois anos consecutivos aumentou significativamente a densidade de esporos de FMA, sugerindo uma resposta de sobrevivência ao estresse ambiental, enquanto a riqueza e diversidade de gêneros micorrízicos foram maiores na área sem remoção. A população de microrganismos benéficos apresentou picos na remoção de 1 ano, declinando sob remoção prolongada. Correlações positivas fortes foram observadas entre a densidade de esporos e os teores de matéria orgânica, cálcio e capacidade de troca catiônica do solo. Conclui-se que a manutenção da palha é essencial para sustentar a diversidade e o equilíbrio funcional do microbioma do solo, e que a remoção contínua compromete a qualidade biológica e a resiliência do sistema agrícola no Cerrado.

Palavras-chave: Bioenergia, Fungos *Micorrízicos Arbusculares*, Bactérias promotoras de crescimento vegetal, Sustentabilidade agrícola, Qualidade do solo.

3. INTRODUCTION

The global transition to renewable energy has placed sugarcane in a strategic position, not only for first-generation ethanol obtained from the juice but also for the residual biomass that feeds second-generation bioenergy (Galdos et al. 2017; Formann et al. 2020). Straw was historically burned to facilitate manual harvesting, with severe environmental costs. Mechanized harvesting made it standard practice to leave the residue on the soil surface, which conserves moisture, suppresses weeds, cycles nutrients and builds organic matter (Sousa Junior et al. 2018; Morais et al. 2020). Recent syntheses across cropping systems confirm that residue return raises nutrient availability and organic carbon stocks, with a magnitude that varies with soil and climate (Ninkuu et al. 2025).

Interest in this lignocellulosic biomass for cogeneration and cellulosic ethanol has led producers to collect part or all of the straw (Castioni et al. 2019). Excessive removal raises concerns for highly weathered tropical soils such as those of the Brazilian Cerrado, which depend on a continuous supply of organic matter to sustain their physical, chemical and biological functions (Cherubin et al. 2021; Pimentel et al. 2024). The trade-off is not restricted to the energy balance; it reaches the resource base that sustains the soil biota.

Soil organic matter is the substrate, the energy source and the physical habitat of most soil microorganisms, and its supply governs how long a population can persist between residue inputs. Labile fractions supply the carbon that sustains microbial growth, enzyme production and nutrient mineralization, while stabilized fractions contribute to aggregate formation, water retention and thermal buffering of the surface layer (Gmach et al. 2021; Primo et al. 2024). When residue input is interrupted, the first consequence is not necessarily a decline in total microbial biomass but a change in which functional groups can be sustained, since taxa with high carbon demand and low tolerance to desiccation are filtered out first (Ninkuu et al. 2025; Wang et al. 2023). In the Cerrado this dependence is sharper, because low natural fertility, high mineralization rates and a marked dry season leave little margin for compensation. Straw management therefore acts indirectly and cumulatively, through carbon availability, thermal amplitude and moisture retention, rather than through a direct effect on the organisms (Wang et al. 2023; Ninkuu et al. 2025).

Those same three pathways are modified by irrigation. Supplemental water raises and stabilizes soil moisture, lowers thermal amplitude and accelerates residue decomposition, and the magnitude of each effect differs between a centre pivot, a reel system and rainfed production. Any consequence of straw removal that operates through the microclimate of the

surface layer may therefore be attenuated or amplified by the water regime, which makes the two factors difficult to interpret in isolation. Field studies of straw removal in Brazil have nonetheless been conducted almost exclusively under rainfed conditions, and the interaction has not been described.

Among the organisms that respond most readily to changes in the edaphic environment are the arbuscular mycorrhizal fungi (AMF), which form mutualistic symbioses with most terrestrial plants including sugarcane (Moura et al. 2017; Moura and Cabral 2019). AMF extend the volume of soil explored by roots, improve water and nutrient uptake, particularly of phosphorus, and can confer tolerance to biotic and abiotic stresses (Azevedo et al. 2014; Juntahum et al. 2020). Their three descriptors, root colonization, spore density and taxonomic richness, do not necessarily vary in the same direction, which makes it advisable to report and interpret them separately.

Other rhizosphere organisms contribute to plant health through biological nitrogen fixation, phosphate solubilization, phytohormone production and suppression of root pathogens (Senkovs et al. 2021; Yao et al. 2023; Kong and Liu 2022; Gobbi and Mattos 2024; Fanai et al. 2024). Their populations depend on labile carbon and on the microclimate at the soil surface, both directly affected by the presence of straw (Wang et al. 2023; de Alcantara Neto et al. 2025). Because the agronomic consequence of a shift in the rhizosphere depends on the balance between organisms that benefit the crop and those that damage it, soil-borne pathogens were quantified alongside the beneficial taxa. The taxa selected here are functionally relevant to sugarcane, responsive to changes in carbon supply, and quantifiable with taxon-specific primers.

This study evaluated how the time of consecutive straw removal and the irrigation system, acting together, affect the AMF community, the abundance of specific beneficial microorganisms and pathogens, and the balance between them, in commercial sugarcane fields of the Cerrado. We hypothesized that straw removal alters the mycorrhizal community independently of the water regime, because the symbiosis depends on the host rather than on residue-derived carbon, while free-living organisms respond jointly to the two factors, because they depend directly on the carbon and moisture conditions that irrigation modifies.

4. MATERIALS AND METHODS

4.1 Study site and experimental design

The study was conducted in commercial sugarcane fields in Goianesia, state of Goias, Brazil (-15.213168S, -48.988389W). The climate is Aw, tropical with a dry winter, according to the Koppen classification (Alvares et al. 2014), with a mean annual temperature of 24.4 degrees Celsius and mean annual rainfall of 1442 mm concentrated between October and March. The soil is an Oxisol (Santos et al. 2018), very clayey, with 800 g kg-1 of clay in the 0-20 cm layer. All areas were planted with cultivar IAC91-1099 in the fourth ratoon cycle and received 100 kg ha-1 of N, 30 kg ha-1 of P2O5 and 120 kg ha-1 of K_2O.

A 3 x 3 factorial arrangement in a completely randomized design was used. The first factor was straw removal time, with three levels: no removal, in which the straw from mechanized harvesting was left entirely on the surface; removal for one year, collected after the harvest immediately preceding sampling; and removal for two years, collected in the two preceding seasons. The second factor was the irrigation system, with three levels: reel, centre pivot and rainfed. Sampling took place in May 2024. Rhizosphere soil and roots were collected at 0-20 cm, and each experimental unit was a composite sample of 15 simple subsamples taken along the planting row. The morphological characterization of AMF used four replicates per cell, giving 36 experimental units, and the molecular quantification used three replicates per cell, giving 27 units.

4.2 Mycorrhizal characterization

AMF spores were extracted by wet sieving and decanting (Gerdemann and Nicolson 1963) followed by centrifugation in a 50% sucrose gradient, quantified under a stereomicroscope and expressed per 50 cm^3 of soil. Identification was performed on permanent slides mounted in polyvinyl alcohol lactoglycerol and in the same medium with Melzer reagent. Family-level assignment was based on spore size, colour, number, thickness and ornamentation of the wall layers and the characteristics of the subtending hypha, following Schenck and Perez (1990) and the taxonomic framework of Oehl et al. (2011). Root colonization was assessed after clearing in 10% KOH and staining with methyl blue (Phillips and Hayman 1970), by the gridline intersect method (Giovannetti and Mosse 1980).

4.3 Molecular quantification

Total genomic DNA was extracted from 0.25 g of moist soil with the DNeasy PowerSoil Pro Kit (Qiagen, Hilden, Germany). Concentration and purity were determined on a NanoDrop 2000c spectrophotometer (Thermo Fisher Scientific), with A260/A280 ratios between 1.8 and 2.0, and integrity was confirmed on a 1% agarose gel. Six beneficial taxa and three soil-borne pathogens were quantified. The 16S rRNA gene was targeted for *Bacillus subtilis* and *Bacillus amyloliquefaciens* (Fraiture et al. 2022) and for *Pseudomonas fluorescens* (Bach et al. 2002), gyrB for *Bacillus megaterium*, synonym *Priestia megaterium* (Kaminsky and Bell 2022), 16S rRNA and nifH for *Azospirillum brasilense* (Stets et al. 2015), and the translation elongation factor 1-alpha gene for Trichoderma spp. (Gerin et al. 2018). Primer sequences, amplicon sizes and annealing temperatures are given in Online Resource 1. Reactions were run on a CFX96 Touch system (Bio-Rad Laboratories) in 20 microlitres containing 10 microlitres of iQ SYBR Green Supermix 2X, 0.8 microlitres of each primer at 10 micromolar and 2 microlitres of template DNA at 10 nanograms per microlitre. Cycling comprised activation at 95 degrees Celsius for 3 minutes and 40 cycles of 95 degrees Celsius for 15 seconds, 60 degrees Celsius for 30 seconds and 72 degrees Celsius for 30 seconds, followed by a dissociation curve from 65 to 95 degrees Celsius. Standard curves were built from tenfold dilutions of plasmids carrying the target fragments, from 10 to the third through 10 to the eighth copies, with efficiencies between 92 and 105% and coefficients of determination of at least 0.99. Specificity was confirmed by melting curve analysis and no-template controls were run in every plate. Each sample was analysed in three technical replicates, whose means were used in the analyses.

4.4 Soil chemical analysis

Phosphorus, potassium, calcium and magnesium were extracted with ion exchange resin, exchangeable aluminium with 1 mol L-1 KCl, and potential acidity with SMP buffer at pH 7.5. Organic matter was determined by oxidation with potassium dichromate in sulphuric acid, following the modified Walkley-Black colorimetric method. Cation exchange capacity and base saturation were calculated from these values.

4.5 Statistical analysis

Community descriptors comprised family richness, the Shannon index, the Simpson index and Pielou evenness. Normality of the residuals and homogeneity of variances were tested

for every response with the Shapiro-Wilk and Levene tests, and the outcomes are reported in Online Resource 2. Variables that met both assumptions were submitted to two-way analysis of variance with straw removal, irrigation system and their interaction, followed by the Tukey test at 5% probability, with omega squared reported as effect size. Variables that violated either assumption were analysed after logarithmic transformation, log (x + 1), and the means are presented on the original scale. Several molecular targets were absent from entire cells of the design, which drives the within-cell variance to zero and makes the F distribution an unreliable reference. For all molecular targets the probabilities were therefore obtained by unrestricted permutation of the observations, with 4999 permutations on the sums of squares, and the F ratio is reported only for targets in which no cell was empty. Detection frequency is reported for every target. The beneficial-to-pathogen ratio was calculated for each experimental unit as the sum of copy numbers of the six beneficial targets divided by the sum of copy numbers of the three soil-borne pathogen targets. Where an interaction was significant, simple effects of straw removal were tested within each irrigation system. Community composition was compared by permutational multivariate analysis of variance on the Bray-Curtis distance with 999 permutations, and ordinated by principal coordinates analysis. Associations between biological and chemical variables were assessed with the Spearman rank correlation on the nine cell means, since soil chemistry was determined once per cell. Analyses were performed in Python 3.10.12 with pandas 2.0.3, SciPy 1.11.1, scikit-learn 1.3.0, scikit-bio 0.5.8, statsmodels 0.14.0, Matplotlib 3.7.2 and seaborn 0.12.2. A large language model was used for language editing and for checking the reproducibility of the analyses; all results were verified against the raw data by the authors, who take full responsibility for the content.

5. RESULTS

5.1 Mycorrhizal responses

The mycorrhizal variables responded to straw removal alone. Neither the irrigation system nor the interaction was significant for any of them (Table 1), so the effects below are described across the three water regimes. Root colonization did not differ among removal times and ranged from 50.3 to 63.2% ($F = 2.44$; $P = 0.106$). Spore density did ($F = 10.48$; $P < 0.001$), but not in a monotonic way: the minimum occurred after one year of removal, 23.7 spores per 50 cm3, which differed from both the undisturbed soil, 58.4, and the soil under two removals, 135.7, while these two did not differ from each other. Family richness followed the same shape, with the lowest value after one year ($F = 4.29$; $P = 0.024$). Pielou evenness declined only after two removals, from 0.83 in undisturbed soil to 0.72 ($F = 5.21$; $P = 0.012$), whereas the Shannon and Simpson indices did not differ (Table 2). Because evenness is the ratio between the Shannon index and the natural logarithm of richness, the stability of the Shannon index alongside a rising richness is arithmetically equivalent to the observed decline in evenness (Fig. 1).

Table 1. Two-way analysis of variance for the arbuscular mycorrhizal fungal variables under three straw removal times and three irrigation systems in commercial sugarcane fields of the Cerrado.

Variable	No removal	One year	Two years	CV (%)	F removal	P removal	Ω2	P irrigation	P interaction
Root colonization (%)	62.2 +/- 15.5 a	50.3 +/- 17.6 a	63.2 +/- 11.1 a	27.0	2.44	0.106 ns	0.081	0.802 ns	0.713 ns
Spore density (spores 50 cm-3)	58.4 +/- 41.7 a	23.7 +/- 12.6 b	135.7 +/- 112.7 a	20.5	10.48	<0.001	0.323	0.263 ns	0.173 ns
Family richness (S)	4.75 +/- 1.14 a	4.25 +/- 1.14 b	5.83 +/- 1.53 a	27.4	4.29	0.024	0.168	0.900 ns	0.677 ns
Acaulosporaceae (spores 50 cm-3)	24.8 +/- 17.0 b	10.3 +/- 5.3 b	65.0 +/- 62.1 a	23.0	12.73	<0.001	0.366	0.162 ns	0.179 ns
Glomeraceae (spores 50 cm-3)	16.1 +/- 11.7 a	8.5 +/- 4.3 b	37.5 +/- 36.0 a	30.9	5.01	0.014	0.170	0.252 ns	0.218 ns
Gigasporaceae (spores 50 cm-3)	5.7 +/- 6.0 a	1.4 +/- 1.6 b	11.7 +/- 9.1 a	61.7	9.31	<0.001	0.334	0.957 ns	0.551 ns

Values are means followed by the standard deviation, and different letters in the same row indicate differences by the Tukey test at 5% probability. CV coefficient of variation of the experiment, omega2 effect size of the straw removal factor, ns not significant

Table 2. Diversity indices of the arbuscular mycorrhizal fungal community under three straw removal times.

Index	No removal	One year	Two years	F removal	P removal	omega2
Shannon (H')	1.26 +/- 0.21 a	1.13 +/- 0.23 a	1.24 +/- 0.17 a	1.28	0.294 ns	0.017
Simpson (D)	0.66 +/- 0.07 a	0.62 +/- 0.08 a	0.63 +/- 0.08 a	0.70	0.504 ns	0.000
Pielou evenness (J)	0.83 +/- 0.06 a	0.81 +/- 0.07 a	0.72 +/- 0.10 b	5.21	0.012	0.208
Family richness (S)	4.75 +/- 1.14 a	4.25 +/- 1.14 b	5.83 +/- 1.53 a	4.29	0.024	0.168

Values are means followed by the standard deviation and different letters in the same row indicate differences by the Tukey test at 5% probability. Neither the irrigation system nor the interaction was significant for any index. ns not significant

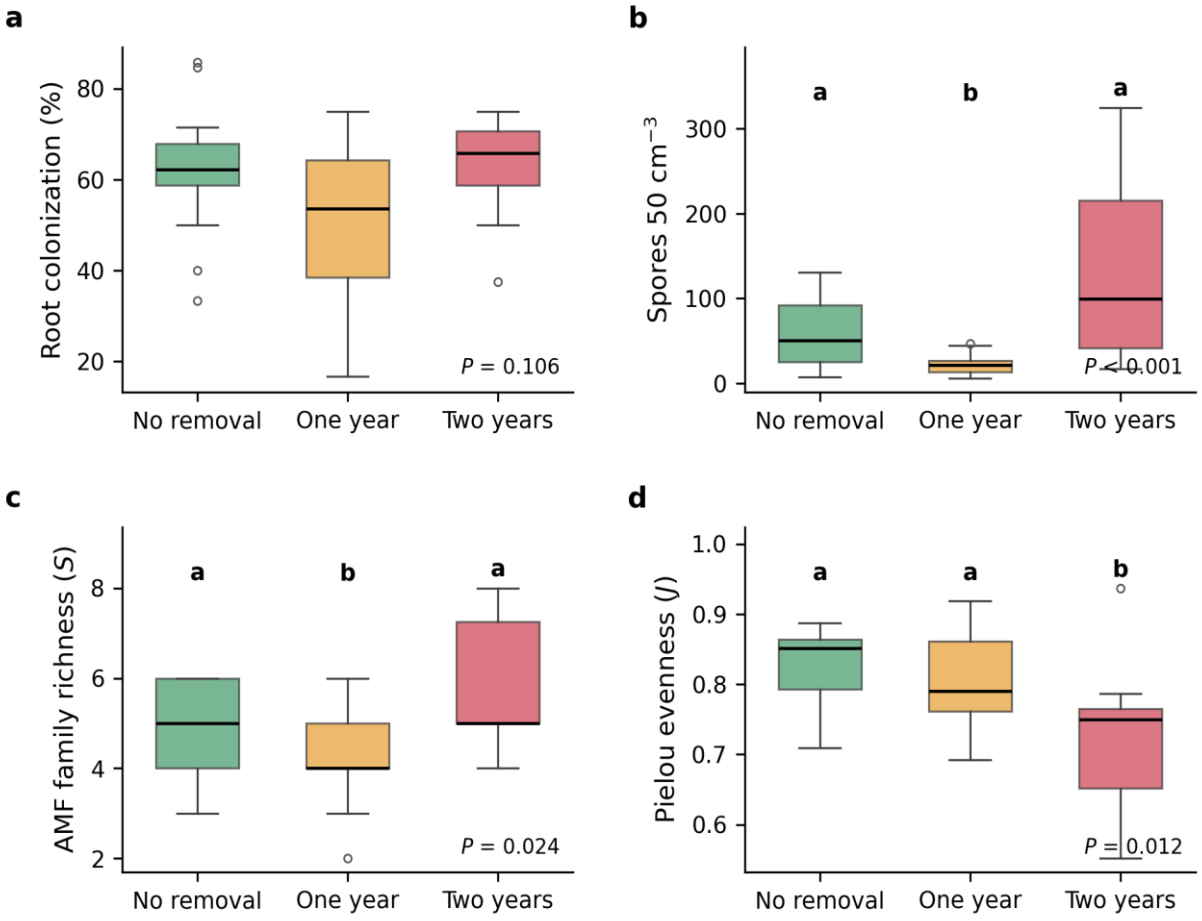

Fig. 1. Response of the arbuscular mycorrhizal fungal community to sugarcane straw removal time in commercial fields of the Cerrado. a root colonization, b spore density, c family richness, d Pielou evenness. Boxes span the interquartile range, the horizontal line marks the median, whiskers extend to 1.5 times the interquartile range and circles are values outside that interval. Different letters indicate differences by the Tukey test at 5% probability. The irrigation system and the interaction were not significant for any of these variables

Glomeraceae and *Acaulosporaceae* predominated in every treatment (Fig. 2a). At family level the same dip after one year appeared in *Acaulosporaceae* (F = 12.73; P < 0.001),

Glomeraceae (F = 5.01; P = 0.014) and *Gigasporaceae* (F = 9.31; P < 0.001), and none of the three showed an effect of irrigation or an interaction (Fig. 2b). The recovery in the second year was not proportional among them. *Acaulosporaceae* was the only family whose density exceeded the undisturbed level, rising from 24.8 to 65.0 spores per 50 cm3, and its relative abundance increased from 42.5 to 47.9%. *Gigasporaceae* also doubled in absolute terms, from 5.7 to 11.7, so its participation in the community was maintained rather than lost. Permutational analysis confirmed that community composition differed among removal times (pseudo-F = 5.27; P = 0.001) but not among irrigation systems (pseudo-F = 1.70; P = 0.253), and the separation is visible in the ordination (Fig. 3a).

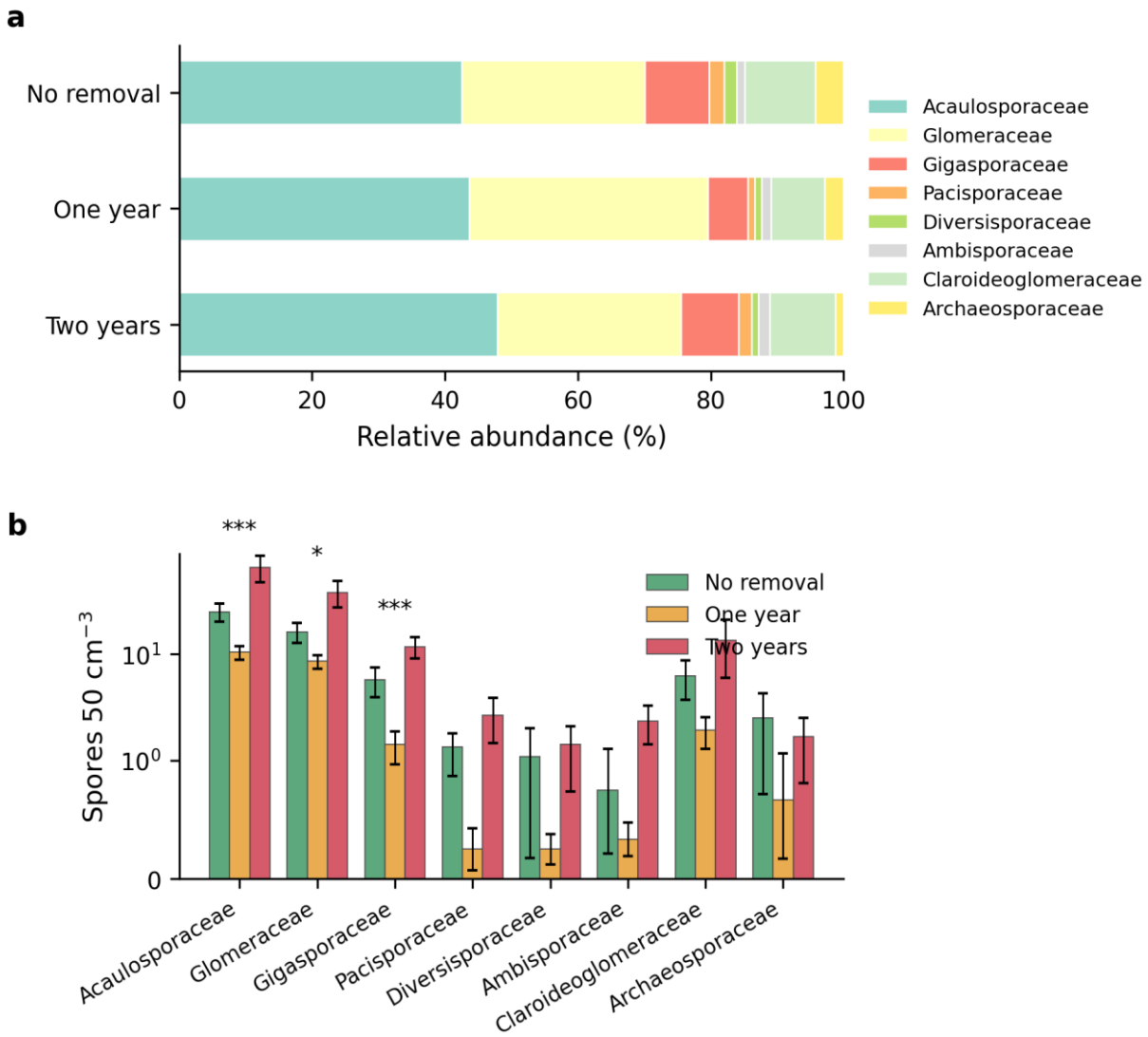

Fig. 2. Composition of the arbuscular mycorrhizal fungal community under three straw removal times. a relative abundance of families, b mean spore density of each family on a symmetric logarithmic scale, with bars showing the standard error of the mean and asterisks indicating the

significance of the straw removal effect, where one asterisk denotes P < 0.05 and three denote P < 0.001

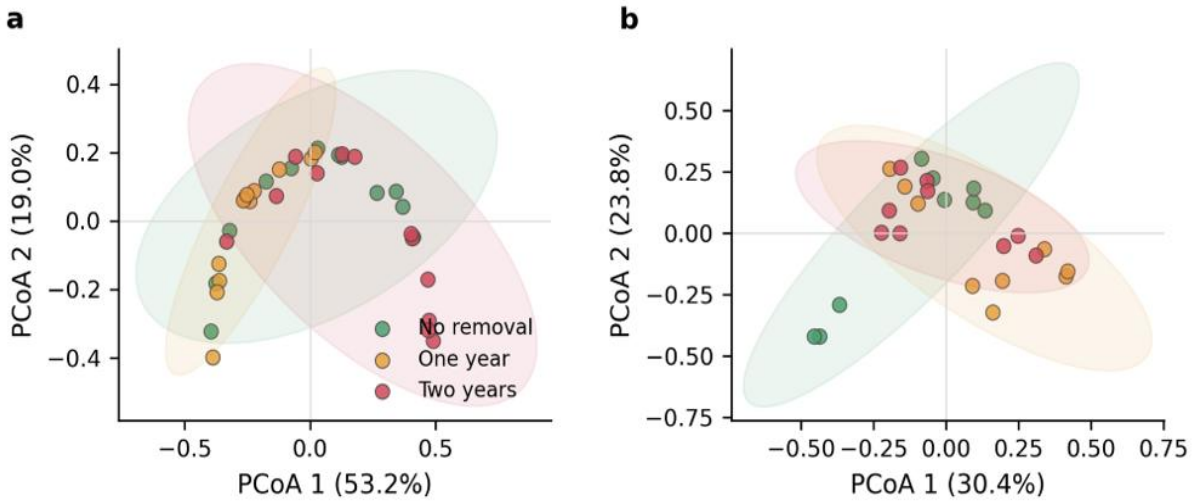

Fig. 3. Principal coordinates analysis on the Bray-Curtis distance for the community of arbuscular mycorrhizal fungal families and b the community of molecular targets, with samples coloured by straw removal time. Ellipses enclose the 95% confidence region of each group and the percentage of variation explained is given on each axis

5.2 Molecular targets

The molecular targets behaved differently from the mycorrhizal community. Interactions between straw removal and irrigation were significant for *Trichoderma spp.* (P = 0.005), *Bacillus subtilis* (P < 0.001), *Bacillus amyloliquefaciens* (P = 0.022) and *Azospirillum brasilense* (P < 0.001), so a single removal effect cannot be stated for these taxa (Table 3, Fig. 4). Decomposition of the simple effects showed that the direction of the response changed with the water regime. *Trichoderma spp*. peaked after one year under the reel system, rising from 5213 to 113 711 copies per gram (F = 20.87; P = 0.002), but declined steadily under the centre pivot (F = 21.77; P = 0.002). *B. subtilis* fell sharply after one year under the reel system (F = 278.20; P < 0.001) and rose under rainfed conditions (F = 38.15; P < 0.001).

Table 3. Molecular targets quantified by real-time PCR under three straw removal times and three irrigation systems.

Target	Detection (%)	Empty cells	F removal	P removal	P irrigation	P interaction
Trichoderma spp.	100.0	0	18.34	0.052 ns	0.076 ns	0.005
Bacillus subtilis	100.0	0	4.79	0.727 ns	0.038	<0.001

Bacillus amyloliquefaciens	100.0	0	8.20	0.023	0.829 ns	0.022
Pseudomonas fluorescens	22.2	7	not applicable	<0.001	0.213 ns	0.103 ns
Azospirillum brasilense	66.7	3	not applicable	0.947 ns	0.004	<0.001
Bacillus megaterium	77.8	2	not applicable	0.083 ns	<0.001	0.313 ns
Total beneficial organisms	100.0	0	22.74	0.014	0.002	0.492 ns
Fusarium oxysporum	100.0	0	3.39	0.243 ns	0.989 ns	<0.001
Pythium sp.	100.0	0	2.73	0.301 ns	<0.001	0.709 ns
Rhizoctonia solani	77.8	2	not applicable	0.123 ns	0.272 ns	<0.001
Beneficial to pathogen ratio	100.0	0	15.75	0.004	0.030	0.449 ns

Probabilities were obtained by unrestricted permutation with 4999 permutations. The F ratio is reported only for targets in which no cell of the design was empty, since a zero within-cell variance makes the F distribution an unreliable reference. Detection is the percentage of samples in which the target was detected. ns not significant

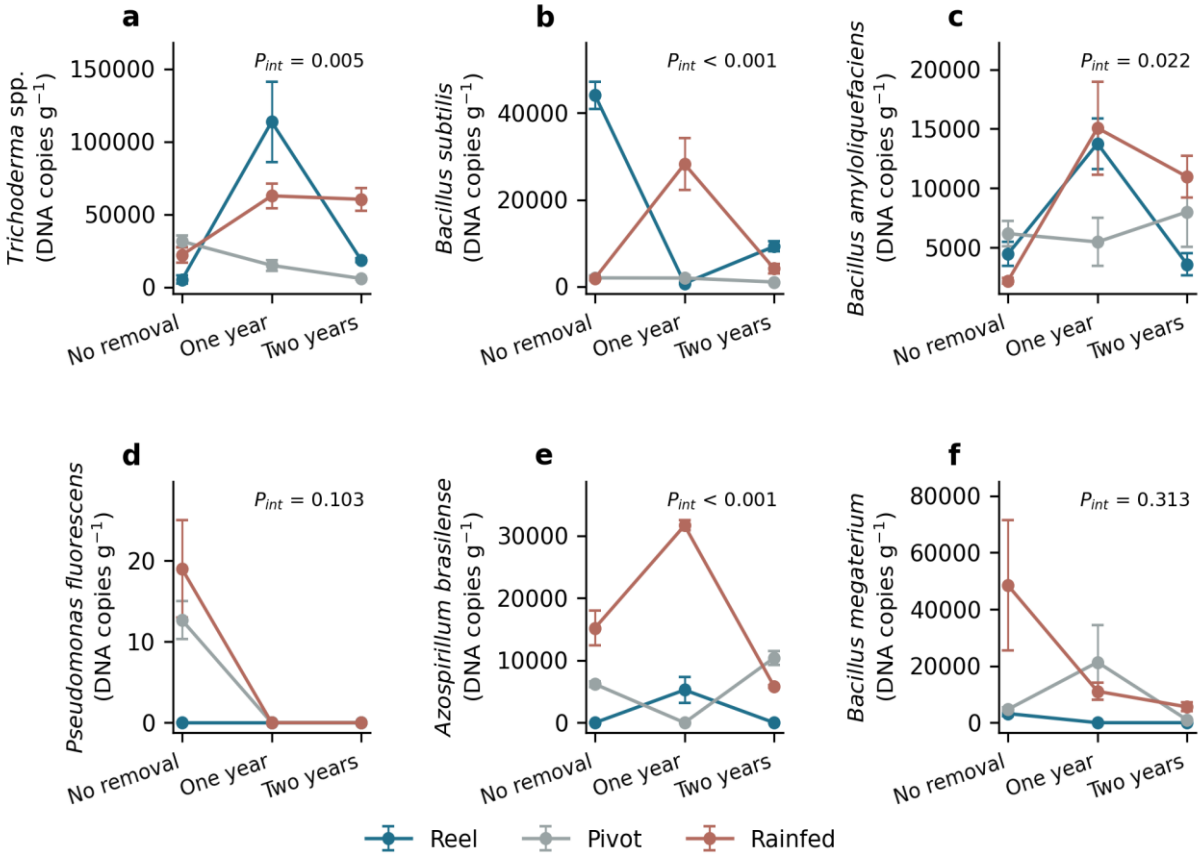

Fig. 4. Abundance of six beneficial taxa quantified by real-time PCR as a function of straw removal time within each irrigation system. a Trichoderma spp., b Bacillus subtilis, c Bacillus amyloliquefaciens, d Pseudomonas fluorescens, e Azospirillum brasilense, f Bacillus megaterium. Symbols are means and vertical lines the standard error of the mean. The probability of the interaction between straw removal and irrigation system is given in each panel

Pseudomonas fluorescens was the exception. It was detected in only 22.2% of the samples, always in undisturbed soil, and disappeared wherever straw had been removed, under every irrigation system ($P < 0.001$ for removal, with no interaction). *Bacillus megaterium* and the total abundance of beneficial organisms were governed mainly by irrigation ($P < 0.001$ and $P = 0.002$, respectively), with the total also responding to removal ($P = 0.014$).

5.3 Pathogens and the balance of the community

The three pathogens were detected in most samples. *Pythium sp.* responded to irrigation alone ($P < 0.001$), with the highest densities under rainfed conditions. *Fusarium oxysporum* and *Rhizoctonia solani* showed strong interactions ($P < 0.001$ in both cases), and in the case of *R. solani* the direction reversed between systems: under the centre pivot it rose from 47 539 copies per gram in undisturbed soil to 589 321 after two removals, while under rainfed conditions it fell to zero over the same gradient (Fig. 5a to 5c).

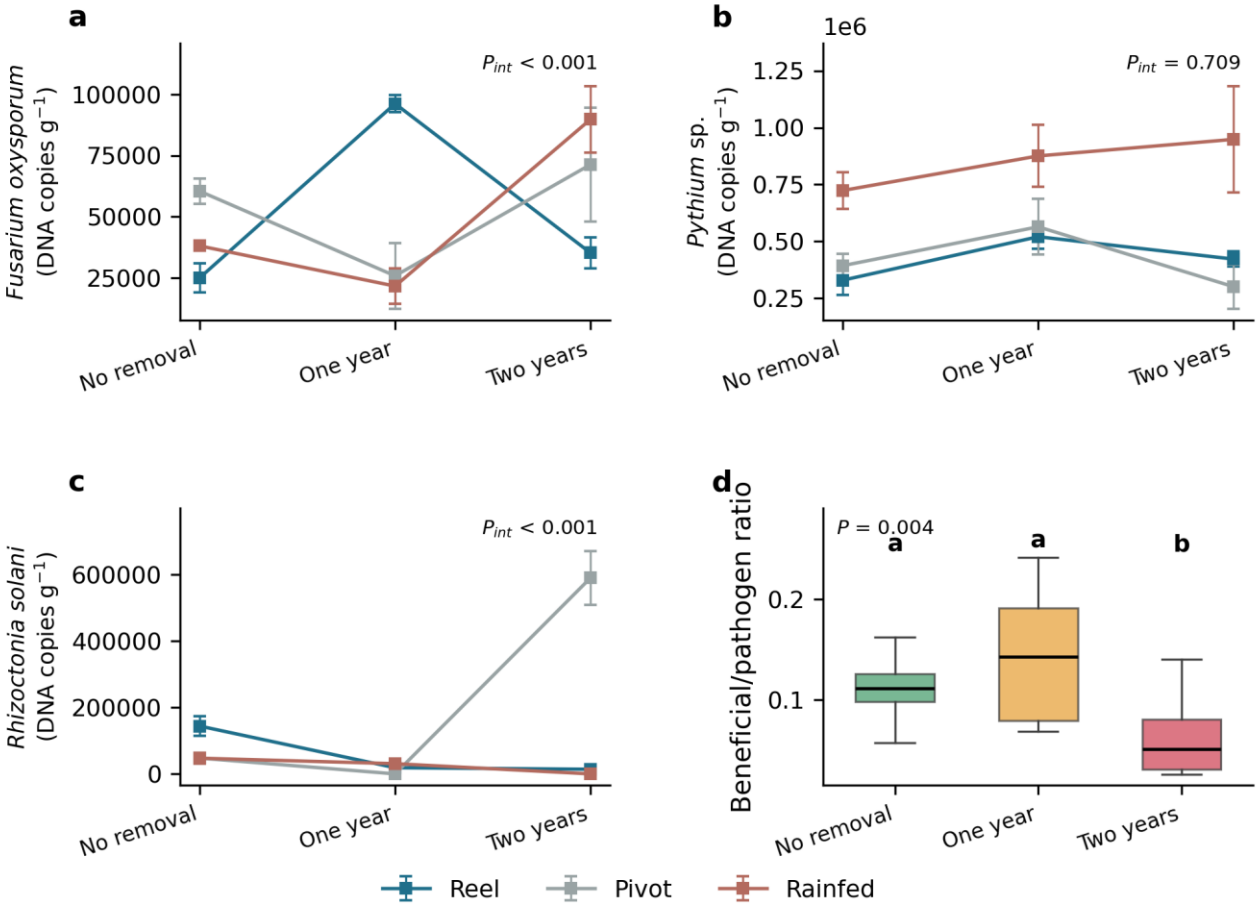

Fig. 5. Soil-borne pathogens and the balance of the rhizosphere community. a Fusarium oxysporum, b Pythium sp., c Rhizoctonia solani as a function of straw removal within each

irrigation system, with the probability of the interaction shown in each panel, and d ratio between the total abundance of beneficial organisms and the total abundance of pathogens, for which neither the irrigation system nor the interaction was significant. Different letters indicate differences by the Tukey test at 5% probability

The ratio between beneficial organisms and pathogens summarized the effect of straw management without the confounding of the individual taxa. It responded to removal ($P = 0.004$) and to irrigation ($P = 0.030$) without an interaction, and declined after two removals from 0.11 in undisturbed soil and 0.14 after one year to 0.06 (Fig. 5d). Spore density was positively correlated with organic matter and with calcium, cation exchange capacity and base saturation, whereas no correlation was found between the soil chemical attributes and either root colonization or the total abundance of beneficial organisms.

6. DISCUSSION

Two factors were examined and they did not act on the two microbial groups in the same way. The mycorrhizal community responded to straw removal alone, with no effect of the water regime and no interaction, while most molecular targets were governed by the combination of the two. This separation supports the hypothesis with which the study began and has a practical consequence: conclusions about free-living rhizosphere organisms obtained under one irrigation system should not be transferred to another, whereas the mycorrhizal response appears transferable across the three systems studied here.

The trajectory of sporulation was not the monotonic increase that a simple stress model would predict. Spore density and family richness fell to a minimum after one year and returned by the second year to values indistinguishable from undisturbed soil. Neither end of this trajectory measures symbiotic performance, since root colonization did not change at any point. Arbuscular mycorrhizal fungi are obligate biotrophs whose carbon comes from the host and not from the residue, so the pulse of labile carbon released by the first disturbance brought them no benefit, while the host was still buffered against the water and thermal stress that develops once the cover is permanently absent. The recovery in the second year is consistent with the allocation to resistant propagules described for AMF under adverse conditions (Souza et al. 2015; Moura et al. 2025), and comparable sporulation under stress has been reported for degraded areas of the Cerrado by Moura and Cabral (2019). The dissociation between a reproductive descriptor and a functional descriptor indicates that the two should not be treated as interchangeable indicators of soil quality, a point of practical value for monitoring programmes that rely on spore counts alone.

What did change unambiguously is the balance among families. The recovery in the second year was disproportionate, and Acaulosporaceae was the only family to exceed its undisturbed density, which raised its relative abundance and drove evenness down. Gigasporaceae, often described as sensitive to desiccation, doubled in absolute density over the same gradient and maintained its share of the community, so the compositional shift observed here is a matter of disproportion rather than of local loss. Family richness assessed from spore morphology is in any case conditional on sporulation, since a taxon is recorded only when it produces enough spores to be recovered from the extracted volume, and the richness curve therefore tracks the spore density curve. Evenness, which is not inflated by the appearance of

rare taxa in the same way, is the descriptor that isolates the compositional signal, and it was the only one of the four diversity measures to show the effect. Reporting a single aggregate index would have concealed it.

The molecular targets require a different reading. Where the interaction was significant the direction of the removal effect changed between irrigation systems, which is what would be expected of organisms that depend directly on carbon availability and on soil moisture rather than on a host plant. *Trichoderma* spp. illustrates this clearly: the transient peak in the first year, which under the reel system reached more than twenty times the undisturbed density, did not occur under the centre pivot, where the population declined steadily. Supplemental water accelerates residue decomposition and shortens the pulse of labile carbon, so the same removal event delivers a different substrate history depending on the water regime. This dependence also indicates that inoculation programmes applied to fields under intensive straw removal may encounter a habitat unable to sustain the introduced populations, a limitation reported for sugarcane by de Alcantara Neto et al. (2025), and that the outcome of such programmes should be expected to vary with irrigation.

Pseudomonas fluorescens behaved as the single unambiguous indicator among the molecular targets. Its detection was restricted to undisturbed soil under all three systems, and it disappeared wherever straw had been removed, without interaction. Because it was detected in only a fifth of the samples, the appropriate reading is one of presence and absence rather than of abundance, and on that reading the loss is complete. Taxa of this genus are known for high carbon demand and low tolerance to desiccation (Kong and Liu 2022), which is consistent with a response governed by the disappearance of the surface cover itself rather than by the amount of water applied.

Quantifying pathogens alongside beneficial organisms proved necessary rather than ancillary. Taken individually, *Rhizoctonia solani* would support opposite conclusions depending on the system in which it was measured, since it increased more than tenfold under the centre pivot and disappeared under rainfed conditions across the same removal gradient. The ratio between beneficial organisms and pathogens, by contrast, responded to straw removal without an interaction and fell by nearly half after two removals. An aggregate balance of this kind is less sensitive to the idiosyncrasy of individual taxa and is the descriptor we would recommend for monitoring, since it addresses the agronomic question directly, namely whether the rhizosphere is becoming more or less favourable to the crop.

The correlations between spore density and organic matter, calcium, cation exchange

capacity and base saturation indicate that biological and chemical deterioration are coupled through the interruption of residue input. In highly weathered soils of the Cerrado, where the organic fraction accounts for a large share of the exchange capacity, this coupling is tighter than in soils with a more active clay fraction (Primo et al. 2024; Silva Gomes et al. 2026). The absence of correlation with root colonization reinforces the interpretation that colonization is governed mainly by the demand of the host plant and not by the fertility gradient.

Some limitations should be considered. The study was conducted in commercial fields, which brings agronomic realism but prevents strict control of spatial heterogeneity, and part of the residual variance may have a spatial origin that the design does not isolate. Identification at family level from spore morphology underrepresents taxa that sporulate little during the sampling period and makes richness dependent on sporulation intensity, as discussed above; a molecular survey would separate these components and is the natural continuation of this work. The molecular quantification covers nine selected taxa and measures gene copies, which include DNA from non-viable cells, so it expresses potential rather than active abundance. Several targets were absent from entire cells of the design, which is why permutation was used in place of the F distribution, and for those targets presence and absence carries more information than the mean. Finally, a two-year series captures the initial trajectory of the process without establishing whether the system stabilizes at a new equilibrium.

7. CONCLUSIONS

Straw removal and irrigation act on the sugarcane rhizosphere through different routes. The mycorrhizal community responded to straw management alone, in a trajectory that dips after one year and recovers by the second, while root colonization remained unchanged throughout, so neither spore counts nor spore-derived richness report soil biological quality on their own. What the second-year leaves behind is a less even community dominated by Acaulosporaceae. Most molecular targets, by contrast, responded to the two factors jointly, and their direction reversed between irrigation systems, which means that results obtained under one water regime should not be extrapolated to another. *Pseudomonas fluorescens* and the ratio between beneficial organisms and pathogens were the two descriptors that responded to straw removal consistently across systems, and both deteriorated once the residue was withdrawn. Maintaining straw cover therefore remains necessary to preserve the functional balance of the soil microbiome, and partial or alternating removal deserves investigation as a way of reconciling the energy use of the biomass with the biological sustainability of the system.

8. STATEMENTS AND DECLARATIONS

8.1 Conflict of interest

On behalf of all authors, the corresponding author states that there is no conflict of interest.

REFERENCES

Alvares, C. A., Stape, J. L., Sentelhas, P. C., de Moraes, J. L. G., & Sparovek, G. (2014). Köppen's climate classification map for Brazil. Meteorologische Zeitschrift, 22(6), 711–728.

Azevedo, L. C. B., Stürmer, S. L., & Lambais, M. R. (2014). Early changes in arbuscular mycorrhiza development in sugarcane under two harvest management systems. Brazilian Journal of Microbiology, 45(3), 995–1005. https://doi.org/10.1590/S1517-83822014000300032

Bach, H. J., Tomanova, J., Schloter, M., & Munch, J. C. (2002). Enumeration of total bacteria and bacteria with genes for proteolytic activity in pure cultures and in environmental samples by quantitative PCR mediated amplification. Journal of Microbiological Methods, 49(3), 235–245. https://doi.org/10.1016/S0167-7012(01)00370-0

Castioni, G. A. F., Cherubin, M. R., Bordonal, R. O., Carvalho, J. L. N., Menandro, L. M. S., & Franco, H. C. J. (2019). Straw removal affects soil physical quality and sugarcane yield in Brazil. BioEnergy Research, 12, 1000–1014.

Chen, S., Xiang, X., Ma, H., Penttinen, P., Zheng, T., Huang, X., & Fan, G. (2022). Response of soil bacterial communities in wheat rhizosphere to straw mulching and N fertilization. Frontiers in Microbiology, 13, 982109. https://doi.org/10.3389/fmicb.2022.982109

Cherubin, M. R., Bordonal, R. O., Castioni, G. A., Guimarães, E. M., Lisboa, I. P., Moraes, L. A. A., Menandro, L. M. S., Tenelli, S., Cerri, C. E. P., Karlen, D. L., & Carvalho, J. L. N. (2021). Soil health response to sugarcane straw removal in Brazil. Industrial Crops and Products, 163, 113315. https://doi.org/10.1016/j.indcrop.2021.113315

de Alcântara Neto, F., Pinheiro, D. A., Rocha, S. M. B., Leite, M. R. L., Costa, R. M., da Silva, J. M., Ventura, S. H., Costa, M. K. L., Sousa, T. K. dos S. A., et al. (2025). Bacterial community in sugarcane rhizosphere under Bacillus subtilis inoculation and straw return. Soil Systems, 9(2), 44. https://doi.org/10.3390/soilsystems9020044

dos Santos, H. G., Jacomine, P. K. T., Anjos, L. H. C., Oliveira, V. A., Lumbreras, J. F., Coelho, M. R., Almeida, J. A., Araujo Filho, J. C., Oliveira, J. B., & Cunha, T. J. F. (2025). Sistema brasileiro de classificacao de solos (6th ed.). Embrapa.

Fanai, A., Bohia, B., Lalremruati, F., et al. (2024). Plant growth promoting bacteria (PGPB)-induced plant adaptations to stresses: An updated review. PeerJ, 12, e17882. https://doi.org/10.7717/peerj.17882

Formann, S., Hahn, A., Janke, L., Stinner, W., & Sträuber, H. (2020). Beyond sugar and ethanol production: Value generation opportunities through sugarcane residues. Frontiers in Energy Research, 8, 579577.

Fraiture, M.-A., Gobbo, A., Papazova, N., & Roosens, N. H. C. (2022). Development of a taxon-specific real-time PCR method targeting the Bacillus subtilis group to strengthen the control of genetically modified bacteria in fermentation products. Fermentation, 8(2), 78. https://doi.org/10.3390/fermentation8020078

Galdos, M. V., Cantarella, H., Hastings, A., Hillier, J., & Smith, P. (2017). Environmental sustainability aspects of second generation ethanol production from sugarcane. In M. S. Buckeridge & A. P. De Souza (Eds.), Advances of Basic Science for Second Generation Bioethanol from Sugarcane (pp. 177–195). Springer. https://doi.org/10.1007/978-3-319-49826-3_10

Gerdemann, J. W., & Nicolson, T. H. (1963). Spores of mycorrhizal Endogone species extracted from soil by wet sieving and decanting. Transactions of the British Mycological Society, 46(2), 235–244. https://doi.org/10.1016/S0007-1536(63)80079-0

Gerin, D., Pollastro, S., Raguseo, C., De Miccolis Angelini, R. M., & Faretra, F. (2018). A ready-to-use single- and duplex-TaqMan-qPCR assay to detect and quantify the biocontrol agents Trichoderma asperellum and Trichoderma gamsii. Frontiers in Microbiology, 9, 2073. https://doi.org/10.3389/fmicb.2018.02073

Giovannetti, M., & Mosse, B. (1980). An evaluation of techniques for measuring vesicular arbuscular mycorrhizal infection in roots. New Phytologist, 84(3), 489–500. https://doi.org/10.1111/j.1469-8137.1980.tb04556.x

Gmach, M. R., Kaiser, K., Cherubin, M. R., & Cerri, C. E. P. (2021). Soil dissolved organic carbon responses to sugarcane straw removal. Soil Use and Management, 37(2), 270–281.

Gobbi, P. C., & Mattos, M. L. T. (2024). Compatibility and antimicrobial activity of Trichoderma spp. combined with diazotrophs and growth-promoting bacteria. Australian Journal of Crop Science.

Guo, Y., Bei, Q., Dzomeku, B. M., Martin, K., & Rasche, F. (2022). Genetic diversity and community composition of arbuscular mycorrhizal fungi associated with root and rhizosphere soil of the pioneer plant Pueraria phaseoloides. iMeta, 1(4), e51. https://doi.org/10.1002/imt2.51

Juntahum, S., Jongrungklang, N., Kaewpradit, W., Lumyong, S., & Boonlue, S. (2020). Impact of arbuscular mycorrhizal fungi on growth and productivity of sugarcane under field conditions. Sugar Tech, 22(3), 451–459. https://doi.org/10.1007/s12355-019-00784-z

Kaminsky, L. M., & Bell, T. H. (2022). Novel primers for quantification of Priestia megaterium populations in soil using qPCR. Applied Soil Ecology, 180, 104628. https://doi.org/10.1016/j.apsoil.2022.104628

Kong, Z., & Liu, H. (2022). Modification of rhizosphere microbial communities: A possible mechanism of plant growth promoting rhizobacteria enhancing plant growth and fitness. Frontiers in Plant Science, 13, 920813. https://doi.org/10.3389/fpls.2022.920813

Morais, M. C., Siqueira-Neto, M., Guerra, H. P., Satiro, L. S., & Cerri, C. E. P. (2020). Trade-offs between sugarcane straw removal and soil organic matter in Brazil. Sustainability, 12(22), 9363.

Moura, J. B. de, & Cabral, J. S. R. (2019). Mycorrhiza in Central Savannahs: Cerrado and Caatinga. In Mycorrhizal Fungi in South America. Springer International Publishing.

Moura, J. B. de, Valentim, N. M., Ventura, M. V. A., & Vieira Junior, W. G. (2017). Taxa de colonização micorrízica sob diferentes sistemas de cultivo no Cerrado em cana-de-açúcar. Diálogos & Ciência, 2(40), 60–66.

Moura, J. B., Vieira Junior, W. G., Filho, L. C. L., de Souza, R. F., dos Santos Lucas, L., & Ramos, M. L. G. (2025). Mycorrhizal fungi in Brazilian tropical biomes. In Q.-S. Wu (Ed.), Management of Mycorrhizal Symbiosis for Mycoremediation and Phytostabilization (pp. 219–243). Elsevier. https://doi.org/10.1016/B978-0-443-24863-4.00013-3

Ninkuu, V., Liu, Z., Qin, A., Xie, Y., Song, X., & Sun, X. (2025). Impact of straw returning on soil ecology and crop yield: A review. Heliyon, 11(2), e41651. https://doi.org/10.1016/j.heliyon.2025.e41651

Oehl, F., Sieverding, E., Palenzuela, J., Ineichen, K., & da Silva, G. A. (2011). Advances in Glomeromycota taxonomy and classification. IMA Fungus, 2(2), 191–199. https://doi.org/10.5598/imafungus.2011.02.02.10

Phillips, J. M., & Hayman, D. S. (1970). Improved procedures for clearing roots and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. Transactions of the British Mycological Society, 55(1), 158–161. https://doi.org/10.1016/S0007-1536(70)80110-3

Pimentel, M. L., Cerri, C. E. P., & Cherubin, M. R. (2024). Quantity, quality and physical protection of soil carbon in response to sugarcane straw removal. Soil and Tillage Research, 236, 105934.

Primo, A. A., Lustosa Filho, J. F., Marota, H. B., Tonucci, R. G., da Silva, I. R., & de Oliveira, T. S. (2024). Different composition of plant residues as a driver of microbial community structure and soil organic matter composition: A microcosm study. Pedobiologia - Journal of Soil Ecology, 106, 150985. https://doi.org/10.1016/j.pedobi.2024.150985

Romero, F., Hilfiker, S., Edlinger, A., Held, A., Hartman, K., Labouyrie, M., & van der Heijden, M. G. A. (2023). Soil microbial biodiversity promotes crop productivity and agro-ecosystem functioning in experimental microcosms. Science of the Total Environment, 885, 163683. https://doi.org/10.1016/j.scitotenv.2023.163683

Santos, H. G., Jacomine, P. K. T., Anjos, L. H. C., Oliveira, V. Á., Lumbreras, J. F., Coelho, M. R., & Cunha, T. J. (2018). Sistema brasileiro de classificação de solos (5th ed.). Embrapa.

Schenck, N. C., & Pérez, Y. (1990). Manual for the Identification of VA Mycorrhizal Fungi (3rd ed.). Synergistic Publications.

Senkovs, M., Nikolajeva, V., & Makarenkova, G. (2021). Influence of Trichoderma asperellum and Bacillus subtilis as biocontrol and plant growth promoting agents on soil microbiota. Annals of Microbiology, 71(1), 1–12.

Silva Gomes, M., Mauad, M., Oliveira, A. C., & Mercante, F. M. (2026). Tillage systems and straw levels: Soil biological quality and sugarcane yield. Revista Caatinga.

Sousa Junior, J. G. A., Cherubin, M. R., Oliveira, B. G., Cerri, C. E. P., & Cerri, C. C. (2018). Three-year soil carbon and nitrogen responses to sugarcane straw management. BioEnergy Research, 11, 249–261.

Souza, R., Ambrosini, A., & Passaglia, L. M. P. (2015). Plant growth-promoting bacteria as inoculants in agricultural soils. Genetics and Molecular Biology, 38, 401–419. https://doi.org/10.1590/S1415-475738420150053

Stets, M. I., Alqueres, S. M. C., Souza, E. M., Pedrosa, F. O., Schmid, M., Hartmann, A., & Cruz, L. M. (2015). Quantification of Azospirillum brasilense FP2 bacteria in wheat roots by strain-specific quantitative PCR. Applied and Environmental Microbiology, 81(19), 6700–6709. https://doi.org/10.1128/AEM.01351-15

Wang, M., Qi, X., Shi, Y., Zhao, J., & Ahmad, S. (2023). Sugarcane straw returning is an approaching technique for the improvement of rhizosphere soil functionality, microbial community, and yield. Frontiers in Microbiology, 14, 1133973. https://doi.org/10.3389/fmicb.2023.1133973

Xu, X., Zhang, W., Liu, Y., Chen, L., Li, Y., Wang, J., Zhang, X., & Shen, G. (2025). Long-term straw return enhances soil microbial co-occurrence patterns and network stability in a rice-wheat cropping system. Frontiers in Microbiology, 16, 1533839. https://doi.org/10.3389/fmicb.2025.1533839

Yao, X., Guo, H., Zhang, K., Zhao, M., Ruan, J., & Chen, J. (2023). Trichoderma and its role in biological control of plant fungal and nematode disease. Frontiers in Microbiology, 14, 1160551. https://doi.org/10.3389/fmicb.2023.1160551

Zhang, M., Dang, P., Haegeman, B., Han, X., Wang, X., Pu, X., Qin, X., & Siddique, K. H. M. (2024). The effects of straw return on soil bacterial diversity and functional profiles: A meta-analysis. Soil Biology & Biochemistry, 195, 109484. https://doi.org/10.1016/j.soilbio.2024.109484