

UNIVERSIDADE FEDERAL DE SANTA MARIA
CENTRO DE CIÊNCIAS RURAIS
PROGRAMA DE PÓS-GRADUAÇÃO EM CIÊNCIA DO SOLO

Lineu Trindade Leal

**DINÂMICA DO CARBONO INFLUENCIADA PELA QUALIDADE DE
RESÍDUOS CULTURAIS, TEOR INICIAL DE CARBONO E TEXTURA
EM DOIS SOLOS SUBTROPICAIS**

Santa Maria, RS
2020

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CULTURAIS, TEOR INICIAL DE CARBONO E TEXTURA EM DOIS SOLOS
SUBTROPICAIS**

Tese de Doutorado apresentada ao Programa de Pós-Graduação em Ciência do Solo da Universidade Federal de Santa Maria (UFSM), como requisito parcial para a obtenção do título de **Doutor em Ciência do Solo**.

Orientador: Dr. Sandro José Giacomini

Santa Maria, RS
2020

Leal, Lineu Trindade

Dinâmica do carbono influenciada pela qualidade de resíduos culturais, teor inicial de carbono e textura em dois solos subtropicais / Lineu Trindade Leal.- 2020.
90 p.; 30 cm

Orientador: Sandro José Giacomini

Coorientador: Denis Angers

Tese (doutorado) - Universidade Federal de Santa Maria, Centro de Ciências Rurais, Programa de Pós Graduação em Ciência do Solo, RS, 2020

1. Sequestro de carbono 2. Efeito Priming 3. Resíduos culturais 4. Estabilização de Carbono I. Giacomini, Sandro José II. Angers, Denis III. Título.

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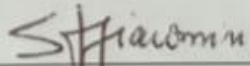
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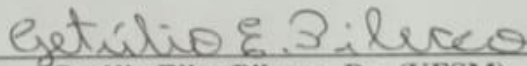
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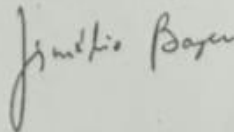
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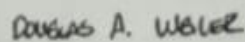
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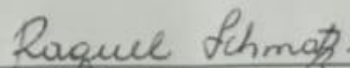
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Santa Maria, RS
2020

Dedico à minha esposa Andriéli.

AGRADECIMENTOS

A Deus, que sempre iluminou meus caminhos.

Ao meu pai, minha mãe e meus irmãos pelo amor, carinho e apoio incondicional.

À Universidade Federal de Santa Maria e ao Programa de Pós-graduação em Ciência do Solo pela oportunidade de realização do curso de doutorado.

Ao professor Sandro J. Giacomini pela amizade, apoio, confiança e orientação na execução deste trabalho.

Aos pesquisadores do “Agriculture and Agri-Food Canada”, Martin Chantigny, Denis Anger e Johanne Tremblay e Marie-Line Leclerc pela acolhida, ensinamentos e experiências oportunizadas nesta instituição renomada. Ao pesquisador David Pelster e sua família, Lilia e Mathias, pela hospitalidade e carinho, fazendo eu me sentir em casa em Quebec.

Ao pesquisador da Embrapa, Anderson Santi, e ao professor da UFRGS, Cimélio Bayer, pela doação dos solos utilizados no experimento desta tese.

À minha amada esposa Andriéli pelo amor, carinho e companheirismo. Pelo apoio para realização deste doutorado. Pela alegria maior de estarmos à espera do Vicente para aumentar e alegrar ainda mais a nossa família.

Aos bolsistas de iniciação científica Daniela, Emanuel, Giovani, Gustavo, João, Mathias, Rafael e Roberta por todo apoio na realização dos experimentos.

Aos colegas de pós-graduação Adriane, Ana, Bruno, Douglas, Getúlio, Guilherme, Heitor, Janquieli, Pâmela, Patrick, Rosemar, e Stefen pela amizade e convivência.

À Elci e ao Fabiano pelo apoio e dedicação nas análises realizadas no LABCEN.

À banca avaliadora pela contribuição e sugestões de melhoria no trabalho.

Ao Heverton pelo cordialidade e eficiência nos atendimentos na secretaria do PPGCS.

Ao CNPq pela concessão da bolsa de doutorado sanduíche.

Ao meu chefe Ricardo Furtado e colegas do MAPA, pelo apoio e compreensão pela minha ausência durante o período de licença para realização deste doutorado.

As demais pessoas, contribuíram de alguma forma para a realização deste trabalho.

Muitíssimo obrigado!

RESUMO

DINÂMICA DO CARBONO INFLUENCIADA PELA QUALIDADE DE RESÍDUOS CULTURAIS, TEOR INICIAL DE CARBONO E TEXTURA EM DOIS SOLOS SUBTROPICAIS

AUTOR: Lineu Trindade Leal
ORIENTADOR: Sandro José Giacomini

A qualidade dos resíduos culturais e o teor de carbono (C) no solo influenciam a dinâmica do C dos resíduos em solos agrícolas. No entanto, existe um entendimento limitado do destino do C dos resíduos e do balanço líquido de C no solo após a adição dos resíduos, particularmente em solos sob plantio direto a longo prazo. Para avaliar isso, resíduos marcados com ^{13}C (parte aérea e raízes de soja e sorgo) foram incorporados em solos com distintas textura e conteúdo de C (arenoso alto-C, arenoso baixo-C, argiloso alto-C e argiloso baixo-C) e incubados por 360 dias em condições de laboratório. A liberação de C-CO₂ e ^{13}C -CO₂ foi avaliada continuamente e a quantificação do ^{13}C remanescente nos resíduos e no solo aos 28, 180 e 360 dias. No solo coletado aos 360 dias foi realizado o fracionamento físico da matéria orgânica do solo (MOS) (<53 μm , 53-250 μm e >250 μm). A mineralização do C dos resíduos foi influenciada pela qualidade e quantidade adicionada e também pelo teor inicial de C do solo, havendo interação significativa entre os fatores. Os solos com alto C propiciaram uma maior decomposição dos resíduos. A quantidade de C mineralizado da MOS após a adição dos resíduos foi 1,6 vezes maior nos solos com alto C do que com baixo C. Isso, indica que a de ciclagem do C nos solos com alto C é rápida comparada a solos com baixo C. Por outro lado, nos solos com alto C não foi observada a ocorrência de maior efeito priming (EP) acumulado após 360 dias, conforme era esperado. As raízes, em geral, apresentam menor EP do que os resíduos de parte aérea, já que apresentam menor índice de decomposição ao final do período. O maior EP acumulado foi observado no solo arenoso baixo-C, onde não foi verificada uma fase tardia de EP negativo, conforme observado nos demais solos. A recuperação de C dos resíduos no solo aos 360 dias sofreu efeito da interação entre o teor de C e o tipo de resíduo, sendo que a qualidade dos resíduos apresentou maior efeito nos solos pobres em C, onde resíduos de soja incrementaram mais C do que os resíduos de sorgo. As raízes também se destacaram por contribuir com maior quantidade de C na fração fina dos solos pobres em C, enquanto que nos solos ricos em C ocorreu um maior do acúmulo nas frações grosseiras. O balanço líquido de C no solo indicou que as quantidades de C derivado dos resíduos superaram as perdas de C mineralizado da MOS apenas no solo com baixo C. Os resultados mostram uma interação complexa entre características dos resíduos culturais e do solo que atuam sobre a dinâmica de retenção de C novo no solo, assim como sobre a intensidade das perdas de C do solo via mineralização da MOS. As intensas perdas de C nos solos com alto-C (chegando a 11% do teor inicial de C) e a maior eficiência de estabilização de C nos solos com baixo-C ressaltam a importância de estratégias para estabilização de C em camadas subsuperficiais no sistema plantio direto.

Palavras-chave: Sequestro de Carbono. Efeito Priming. Estabilização de Carbono. Resíduos Culturais.

ABSTRACT

CARBON DYNAMICS INFLUENCED BY CROP RESIDUE QUALITY, INITIAL CARBON CONTENT AND TEXTURE IN TWO SUBTROPICAL SOILS

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ADVISOR: Sandro José Giacomini

Crop residues quality and initial soil carbon (C) content influence the residue-derived C dynamic in agricultural soils. However, there is a limited understanding about the fate of residue C and the net C balance in the soil after residues application, particularly in soils subjected to long-term no-till systems. To address this, ^{13}C labeled residues (shoots and roots of soybean and sorghum) were incorporated in soils with different textures and initial soil C content (sand high-C, sand low-C, clay high-C and clay low-C) and incubated in laboratory conditions for 360 days. The C-CO₂ e ^{13}C -CO₂ were continuously measured and the ^{13}C in remaining residues and in soil were measured after 28, 180 and 360 days. At 360 days, it was performed the size fractionation of soil organic matter (SOM) (<53 μm , 53-250 μm e >250 μm). The residue-derived C mineralization was influenced by quality and quantity of residues, initial soil C content, and their interactions. High-C soils provided a greater residue mineralization. After 360 days, the average of mineralized soil organic C (SOC) in soils amended with crop residues was 1.6 greater in high-C than in low-C soils, indicating that SOC cycling is faster in high-C than in low-C soils. On the other hand, high-C content was not determinant to increase the cumulate priming effect (PE) at long-term. Overall, roots induce less PE over time compared to shoot residues, since it also had less residue-C mineralization. The highest PE cumulative was observed in sand low-C soil, which did not show a late negative PE phase as other soils. The residue-derived C recovery was influenced by initial soil C content and residues type interaction, with greater effect of residues quality in low-C than high-C soils, with soybean residues contributed more to soil C than sorghum residues. The roots also included more residue-derived C in the fine fraction in low-C than in high-C soils, while in high-C soils tended show greater residue-derived C recovery in coarse fractions (coarse sand and fine sand). The net C balance showed that amounts of residue-derived C recovery in the soil exceeded the losses of mineralized SOC only in low-C soils. The results show a complex interaction between residues and soil characteristics that affect the new C retention in the soil and the intensity of soil C losses. The high losses of C in high-C soils (reaching to 11% of initial soil C content) and the greater efficiency of residue-C stabilization in low-C soils, highlight the importance of strategies to achieve the residue-C stabilization in subsurface layers of no-till systems.

Keywords: Carbon Sequestration. Priming Effect. Carbon Stabilization. Crop Residues.

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1 INTRODUÇÃO

A matéria orgânica do solo (MOS) é sugerida como indicador-chave da qualidade do solo sendo utilizado em diversos estudos de avaliação da sustentabilidade de sistemas de produção agrícola (Amado et al., 2006; Conceição et al., 2005; Mielniczuk, 1999). A MOS possui alta correlação com atributos do solo relacionados às funções básicas do solo, como estabilidade de agregados, susceptibilidade à erosão, infiltração e retenção de água, capacidade de troca de cátions (CTC), disponibilidade de nutrientes para as plantas, lixiviação de nutrientes, atividade biológica, entre outros, que justificam a sua importância como indicador de qualidade (Mielniczuk, 1999). Além disso, a MOS está diretamente ligada a mitigação das mudanças climáticas através do aumento do sequestro de C no solo.

Após o avanço do sistema de plantio direto (SPD) no Brasil, estudos tem se dedicado a avaliar os sistemas de cultivo, mostrando que o SPD é uma importante ferramenta que pode contribuir para o sequestro de carbono (C), enquanto o plantio convencional costuma atuar como fonte de carbono (C) para a atmosfera (Bayer et al., 2004; Campos et al., 2011; Conceição et al., 2005). O incremento de C no solo depende de um balanço positivo entre os fatores de entrada e saída deste no solo. Assim, culturas que produzem maiores quantidades de resíduos tem maior potencial de adição de C e conseqüentemente maior acúmulo de MOS (Campos, 2006).

Diversas pesquisas foram realizadas na busca de identificar sistemas de cultivo com capacidade de melhorar a qualidade do solo com ênfase para os estoques de C e N do solo (Amado et al., 2006; Campos et al., 2011; Conceição et al., 2005; Heenan et al., 2004; Poirier et al., 2016; Vergutz et al., 2010; Zanatta et al., 2007). Além do tipo de manejo do solo adotado e da quantidade de resíduos orgânicos adicionados ao solo, a eficiência de retenção de C novo no solo também pode sofrer influência da composição química dos resíduos culturais, genericamente denominado de qualidade dos resíduos (Cyle et al., 2016; Schmatz et al., 2017). A qualidade tem influência direta sobre a biomassa microbiana e sua atividade (Babujia et al., 2010; Sauvadet et al., 2016) e por conseqüência sobre a quantidade de C estabilizado no solo (Cotrufo et al., 2013; Haddix et al., 2016).

Entretanto, a adição de resíduos culturais estimula a atividade microbiana do solo gerando alteração na taxa de decomposição da MOS, o que é conhecido como efeito priming (EP) (Fontaine et al., 2003). O aumento na velocidade de mineralização da MOS (EP positivo) pode reduzir taxa líquida de sequestro de C no solo a valores muito baixos ou até negativos

(Shahbaz et al., 2017a; Wu et al., 2019; Zhang et al., 2017). Em geral, solos mais ricos em C e N são propensos a apresentarem EP em maior intensidade, sendo que o mesmo aumenta com a quantidade de substâncias orgânicas adicionadas ao solo, especialmente aquelas com maiores concentrações de fração solúvel (Kuzakov et al., 2000). Portanto, o EP passa a ser ainda mais importante a medida que o solo acumula mais C, aumentando a intensidade de saída de C do solo e consequentemente dificultando a manutenção dos níveis de sequestro de C no solo.

A MOS tem sido amplamente estudada quanto a sua química, dinâmica, interações e transformações microbianas, buscando entender melhor o tempo de permanência de cada componente no solo, o papel dos produtos microbianos e a modelagem da matéria orgânica (Paul, 2016). O aprofundamento deste conhecimento permitirá uma melhor gestão dos sistemas agrícolas na busca da sustentabilidade do solo como recurso imprescindível para a manutenção da vida. A estabilização da matéria orgânica pode ocorrer por proteção física no interior de microagregados, estabilização química por associação com minerais silte e argila, ou bioquimicamente pela formação de compostos recalcitrantes (Six et al., 2002). A fração mais estável da MOS está ligada aos minerais silte e argila. Considerando que existe uma correlação entre a textura do solo e a quantidade de matéria orgânica estabilizada por associação à esses minerais, é assumido que cada solo possui uma capacidade de proteção da matéria orgânica nesta fração que está diretamente ligada a sua textura (Hassink, 1997; Plaza et al., 2013; Stewart et al., 2008). Assim, tem se evoluído no entendimento que o solo possui um nível de saturação de C a partir do qual o teor de matéria orgânica tende à estabilidade (Castellano et al., 2012; Six et al., 2002; Stewart et al., 2007; West and Six, 2007).

Neste contexto, foi conduzida uma incubação em condições de laboratório pelo período de 360 dias. Os resíduos de parte aérea e raízes de soja e de sorgo, marcados com ^{13}C , foram incorporados em quatro solos, com texturas e teores iniciais de C distintos (arenoso alto-C, arenoso baixo-C, argiloso alto-C e argiloso baixo-C) coletados em experimentos de longa duração (35 anos) no Rio Grande do Sul. Os resultados são discutidos em dois artigos científicos e integrados na discussão geral. O primeiro artigo apresenta a dinâmica de decomposição dos resíduos culturais e sua influência na mineralização do carbono orgânico do solo (COS) e consequentemente o efeito priming (EP) gerado no período. O segundo artigo engloba os dados de C novo retidos nas frações físicas da MOS.

1.1 Hipóteses

Resíduos culturais de maior qualidade química induzem maior efeito priming do que resíduos de menor qualidade.

O efeito priming é maior em solos com maior teor de C orgânico.

Quanto menor o teor inicial de C orgânico do solo maior será a quantidade de C proveniente do resíduo estabilizado na fração silte e argila.

O efeito da qualidade dos resíduos culturais sobre a estabilização de C novo na fração silte e argila no solo é maior em solos com menor teor de C do que em solos mais ricos em C.

1.2 Objetivo Geral

Compreender a relação entre o teor inicial de C do solo e a qualidade e quantidade dos resíduos culturais adicionados sobre a dinâmica de C no solo.

1.3 Objetivos Específicos

Quantificar o efeito da adição de resíduos ao solo sobre a mineralização da MOS (efeito priming) durante um ano de incubação.

Quantificar a retenção de C novo no solo, proveniente dos resíduos culturais, estabilizados nas frações físicas da MOS.

Quantificar o balanço líquido de C no solo em função da adição de resíduos culturais.

2 ARTIGO 1 - INTERACTION BETWEEN CROP RESIDUES AND SOIL CARBON CONTENT ON PRIMING EFFECT INTENSITY IN TWO SOIL TYPES¹

2.1 Abstract

Crop residues is the main source of carbon (C) to soil, but the residue addition may accelerate soil organic carbon (SOC) mineralization and impact the amount of C stored in the soil. However, it is largely unknown to what extent the soil C content and residue type affect the priming effect (PE). The aim of this study was to investigate the effect of initial SOC content, and quality and quantity of crop residues on mineralization of C derived from residues and its priming on native SOC decomposition in two soil textures. We mixed ¹³C-labelled crop residues (shoots and roots of soybean and sorghum) in four soils (high-C sand, low-C sand, high-C clay and low-C clay) and incubated for 360 days. The C mineralization (% of C added) at 360 days ranged from 11 to 37% in the roots and from 36 to 57% in the shoots. The SOC mineralization was influenced by the soil C content, residues types and their interaction. At 360 days, the cumulative SOC mineralization was 1.6 times greater in high-C soils than in low-C soils, indicating that SOC cycling time is short in high-C than in low-C soils, which can be due a greater proportion of unprotected SOC in higher-C soils. Contradictory to expectations, high-C content was not determinant to increase the cumulative PE at long-term. We observed negative PE a late stage in the high-C soils and low-C clay soil, which could be related to the theory of microbial succession that increase the SOC mineralization in unamended soils. This behavior was not observed in the low-C sand, which presents the highest PE cumulative at 360 days. Overall, roots induce less PE over time compared to shoot residues, since it also had less residue-C mineralization. Therefore, the PE may change intensity and direction at long-term, being strongly influenced by interaction between crop residues (shoots and roots) and soil characteristics.

Keywords: Priming effect, crop residue quality, soil organic carbon.

2.2 Introduction

¹ Artigo elaborado conforme normas da Revista Soil Biology and Biochemistry

Soil organic carbon (SOC) is a key compound of soil quality, playing multiple ecosystem functions in agricultural soils (Campos et al., 2011). Crop residues are fundamental for microbial activity and for maintenance of SOM levels. Depending on plant species and harvest time, crop residues have different chemical composition (soluble fraction, cellulose, hemicellulose, lignin, N content, etc.), which influences the decomposition dynamic and microbial processes (Abiven et al., 2005; Bertrand et al., 2006; Lian et al., 2016; Redin et al., 2014b). Labile residue components are used more efficiently by microbes, thus leading to greater production of microbial products that are precursors of stable SOC in mineral-associated organic matter (Cotrufo et al., 2013; Haddix et al., 2016).

Addition of crop residues may increase the short-term rate of SOC decomposition by microorganisms, a phenomenon called priming effect (PE) (Kuzyakov et al., 2000). However, the mechanisms that induce the priming effect, its strength, and its impact on SOC dynamic are not well understood. The intensity of priming effect may be dependent on the amount of residues added, but the response is usually not proportional to increase in the addition rate and the priming per unit C-added declines with increasing residues inputs (Xiao et al., 2015). Blagodatskaya and Kuzyakov (2008) reported that PE intensity increased linearly with labile C addition equivalent to up to 15% of soil microbial biomass C induce linear increase of PE; however, while addition rates higher than 50% of microbial biomass C induced an exponential decrease or even negative PE values. The PE intensity may also depend on soil type (Frøseth and Bleken, 2015; Schmatz et al., 2017; Zhang and Marschner, 2017) and nutrients (C, N, P) availability (Chen et al., 2014; Hicks et al., 2019). In general, PE tend to show greater response to inputs in C- and N-rich than in poor soils (Kuzyakov et al., 2000).

In this context, the objectives of our study were to understand the effects of initial SOC content, and the quality and quantity of crop residues added on (i) the mineralization of crop residue C and nitrogen (N), and (ii) the PE of SOC mineralization in the short and longer term, in two different soil textures, using ^{13}C labelled crop residues. We hypothesized that: (i) crop residues with greater quality would induce greater PE than lower quality residues; and (ii) PE would be greater in C-rich than C-poor soils, whatever the soil texture.

2.3 Materials and methods

2.3.1 Soil sampling

Two soils were collected in two long-term experiments started in 1985 in the state of Rio Grande do Sul, Southern Brazil. A sandy clay loam (sandy soil), classified as a Typic Paleudalf (Soil Survey Staff, 2014), was collected at the Federal University of Rio Grande do Sul (30° 50' S, 51° 38' W). A clay soil, classified as an Oxisol (Soil Survey Staff, 2014), was collected at the Embrapa Wheat Research Centre (28°15'S, 52°24'W). For both soils, the 0-5 cm and 10-15 cm layers were collected separately, with the objective of obtaining soils with different SOC contents but similar texture. The soils were gently crumbled, sieved at <4 mm in the field, and visible organic residues were removed by hand. Field-moist sieved soils were homogenized by thorough mixing, sub-sampled, and stored in dark plastic bags at room temperature for 20 days until incubation. The soil mineral particle distribution (Soil Survey Staff, 1972), total organic carbon (dry combustion), total mineral N (1M KCl exchangeable), Mehlich-I extractable phosphorous, and pH in water (1:1) were measured (Table 1).

2.3.2 Planta residues

Sorghum (*Sorghum bicolor* L.) and soybean (*Glycine max* L.) were grown in greenhouse. When the plants reached the V2 vegetative stage, weekly pulse labelling with $^{13}\text{CO}_2$ started and lasted until the plants had flowered. Pulse labelling was done using a system adapted from Sangster et al. (2010). Once a week, the plants were enclosed in a chamber made of $0.8 \times 0.8 \times 0.3$ m (length, width, height) polymethyl methacrylate segments that were stacked according to plant height, using a final segment closed on top. Pulses of $^{13}\text{CO}_2$ were generated from the reaction of 2 M HCl with a solution of $\text{NaH}^{13}\text{CO}_3$ (33 atom % ^{13}C).

The plants were harvested at physiological maturity. Plants shoots were cut close to the ground and separated into leaves and stems. The roots were thoroughly washed on a 2-mm sieve under a stream of tap water. All plant parts were oven-dried at 45°C to a constant weight, and cut to 1-cm pieces. Leaves and stems were combined in a 1:1 mass ratio (hereafter called shoot residues). A subsample of roots and shoots was ground to 1 mm with a knife mill (willey type) to determine the soluble (SOL), cellulose (CEL), hemicellulose (HEM) and lignin (LIG) fractions according to Van Soest, (1963). Briefly, a neutral digestion (0.3 g of plant residue in 30 mL of neutral detergent solution) and an acid digestion (0.6 g of residue plants in 60 mL of acid detergent solution) were performed in a digester block at 150°C for 1 h. Both digested samples were vacuum-filtered in a fritted crucible and then washed with distilled water followed by 30-40 mL of acetone. The fibers remaining in crucible were dried at 105°C for 12

h. The SOL was taken as the difference between initial sample weight and weight of fibers remaining after neutral digestion. The HEM content was taken as the difference between weight of fibers remaining after neutral and acid digestion. The fibers remaining after acid digestion were further digested in 12 M H₂SO₄ for 3 h, and the CEL content was taken as the weight loss. The crucible with remaining material was then put in a muffle furnace at 500 °C for 3 h, and the LIG content was taken as the weight loss.

Another plant subsample was oven-dried at 65°C for 48 h to determine dry matter (DM) concentration. This sample was then finely ground to measure total C and N concentrations using an elemental analyser (FlashEA 1112, Thermo Finnigan, Milan, Italy), and ¹³C abundance with an isotope ratio mass spectrometer (IRMS) (DELTA V Advantage, Thermo Fisher Scientific, Bremen, Germany) after combustion in an elemental analyser (Flash EA 2000, Thermo Fisher Scientific, Bremen, Germany). The characteristics of crop residues are given in Table 2.

2.3.3 Incubation conditions and experimental design

The four soil types described in section 2.1 were used in this incubation and were defined as high-C sand, low-C sand, high-C clay and low-C clay. One day before initiating the incubation, the mineral N concentration of the soils was standardized at 60 mg kg⁻¹ by the addition of KNO₃, the soil water content was adjusted to 70% water field capacity, and soils were homogenized to set the experimental units. The aboveground plant residues were mixed with the soils at rate equivalent to 4, 8 or 12 Mg ha⁻¹. Root residues were added only at a rate of 4 Mg ha⁻¹. A non-amended control treatment was included with each soil type, for a total of 36 treatments. Each treatment was replicated three time for a total of 108 experimental units arranged as a completely randomized design. Three sets of 108 experimental units were prepared to allow for destructive sampling in the course of incubation (see section 2.5).

The crop residues were moistened with 3 mL g⁻¹ of deionized water, thoroughly mixed with 107.9 g of soil (dry mass equivalent) in a tray, and transferred in a 110-mL cylindrical acrylic container (5.0 cm in diameter and 5.0 cm in height). To ensure a uniform density, the soil-residue mixture was placed into the container in two steps. Half of the soil mixture was placed in the container and compressed to a height of 2.5 cm. The remaining soil mixture was then placed on the compressed soil and compressed to a final height of 5.0 cm. This allowed to

reach a final bulk density of 1.1 g cm^{-3} . The incubation was conducted for 360 days in the dark at $25 \pm 1 \text{ }^\circ\text{C}$.

2.3.4 CO_2 release

The acrylic containers with compressed soil were placed in 1-L glass jars equipped with sealing cap and septum to monitor C mineralization. A small beaker with 4 mL of water was placed in each jar to minimize soil water loss, and a 37-mL beaker with 10 mL of 1M NaOH was placed above the acrylic container (on a support to avoid contact with the soil surface) to trap the CO_2 released (Stotzky, 1965).

The total CO_2 released was measured by replacing the NaOH trap after 3, 6, 11, 17, 28, 42, 56, 77, 98, 126, 153, 180, 225, 270, 314 and 360 days of incubation. On each sampling date, the carbonates trapped in NaOH were precipitated to BaCO_3 with 2 ml of 2 M BaCl_2 solution (Schmatz et al., 2017). The remaining NaOH in the trap was then titrated with 1 M HCl using phenolphthalein as indicator. Each time that NaOH traps were renewed, the glass jars were kept opened for 10-15 minutes to equilibrate the soil atmosphere with ambient air. The jars were then weighed to adjust the soil moisture and closed again until the next trap replacement date.

The proportion of C mineralized that was derived from the crop residues was assessed by quantification of $^{13}\text{CO}_2$ in the NaOH traps, as described by Schmatz et al. (2017). Briefly, after the titration step, the BaCO_3 precipitate in each trap was recovered by vacuum filtration on a fiberglass filter (Whatman 42; porosity $1.2 \text{ } \mu\text{m}$). The filter with precipitate was dried in an oven at 65°C for a day. The dried precipitate was removed from the filter with a spatula, ground, and analyzed for ^{13}C isotope abundance with the IRMS described in section 1.2.

2.3.5 Soil Mineral N

The jars were destructively sampled at 28, 180, and 360 days of incubation to determine soil mineral N concentration by extracting moist soil in 1 M KCl (30 min shaking; 1:4, soil:solution ratio). The soil suspension was left on the bench for 30 min and the supernatant was filtered (paper filter Unifil C42, particle size retention 1-2 μm). The filtered extracts were analysed for $\text{NH}_4^+\text{-N}$ and $\text{NO}_3^-\text{-N}$ concentrations by continuous flow colorimetry (SAM Plus, Skalar Analytical, Breda, Netherlands).

2.3.6 Calculations and statistical analyses

The apparent mineralization of crop residue C (C_{\min}) was calculated as follows:

$$C_{\min} = (CO_{2\text{ total}} - CO_{2\text{ s}})$$

where, $CO_{2\text{ total}}$ is the amount of C released from soil amended with residues, and $CO_{2\text{ s}}$ is the amount of C released from the same soil without residues. The actual mineralization of residue C was calculated as follows:

$$^{13}C_{\min} = CO_{2\text{ total}} \times (At\%^{13}C_r - At\%^{13}C_s)$$

$$^{13}C_{\min} (\%) = (^{13}C_{\min} / ^{13}C_{\text{ad}}) \times 100$$

where, $^{13}C_{\min}$ is the amount of residues- ^{13}C mineralized in mg kg^{-1} , and $^{13}C_{\min} (\%)$ is the proportion (%) of C added that was mineralized; $At\%^{13}C_r$ and $At\%^{13}C_s$ are the ^{13}C abundance (atom %) in soil with and without crop residues, respectively; and $^{13}C_{\text{ad}}$ is the quantity of ^{13}C initially added to the soil.

The amount of CO_2 -C derived from SOC (SOC_CO_2) could be calculated as:

$$SOC_CO_2 = CO_{2\text{ total}} - ^{13}C_{\min}$$

where, $CO_{2\text{ total}}$ is the amount of C released from soil amended with residues.

The PE of SOC mineralization caused by residue addition was calculated as the difference between the apparent and actual C mineralization:

$$PE = C_{\min} - ^{13}C_{\min}$$

The PE value calculated at a given sampling date represent the cumulative effect from the beginning of incubation to the given sampling date.

An analysis of variance (ANOVA) was performed for each soil texture separately to test the effect of initial soil C content, residue type and their interactions on residue-C and SOC mineralization, and cumulative PE at days 6, 28, 98, 180 and 360. The Mixed procedure of SAS with repeated measures (SAS Institute, 2002) was used. Homogeneity of variances was previously checked. Contrast analysis was performed when significant effects were found ($P < 0.05$). We used the following contrasts to test crop residue effects: rate 4 shoot sorghum vs rate 4 shoot soybean; rate 8 shoot sorghum vs rate 8 shoot soybean; rate 12 shoot sorghum vs rate 12 shoot soybean; sorghum root vs soybean root; rate 4 root vs rate 4 shoot; rate 4 sorghum root vs rate 4 sorghum shoot; rate 4 soybean root vs rate 4 soybean shoot. When the C content showed significant interaction with residue type, we performed contrasts for each C content separately. The effects were considered significant at $P < 0.05$. A second ANOVA was carried out to test the effects of addition rate of shoot residues and its interaction with initial soil C

content and shoot residue type (soybean or sorghum). Posteriorly, the followed contrasts were performed: soybean *vs* sorghum; rate addition linear effect; rate addition quadratic effect; C content per plant; C content per rate linear; C content par rate quadratic; Plant per rate linear; Plant per rate quadratic; C content x Plant per rate linear; C content x Plant per rate quadratic).

2.4 Results

2.4.1 Residue C mineralization

In all amended soils, residue-derived C mineralization showed a classic two-stage kinetic of decomposition, with an initial phase of rapid mineralization, that lasted for the first 50 to 100 d when the main portion of C mineralization occurred, followed by a second phase when C mineralization levelled off (Fig. 1). The proportion of residue-C mineralization varied depending on initial soil C content and residue type during all experimental time (Table 3), with significant interaction between these factors in both soils textures. This interaction show a greater effect of residues in high-C than in low-C soils. In high-C sand, sorghum root mineralized more C than soybean root, while this difference was not significant in low-C sand. In the same way, the residue-C mineralization from soybean rate 4 was faster than sorghum rate 4, in high-C clay, but was not significant in low-C clay.

At a comparable addition rate (4 Mg ha^{-1}), C mineralization from roots was less than shoot residues (Fig. 1; Table 3), in both soil textures. The cumulative mineralization of roots (% of C added) at 360 d ranged from 11% for soybean roots in the low-C clay to 37% for sorghum roots in the high-C sand. Shoot residues mineralized more rapidly, reaching 36 to 57% of added C, depending on soil type. Carbon mineralization increased linearly with shoot addition rate in both soils. This effects was generally similar with both soil C contents and with both residue types (soybean or sorghum).

2.4.2 SOC mineralization

The SOC mineralization was dependent on initial soil C content, residue type and addition rate. In sandy soils, the contrast between control (non-amended soil) and other treatments (amended soil) demonstrate significant difference, all experimental time (Table S3), confirming the PE effect that change SOC decomposition rates after the crop residues addition.

This contrast was not significant at 180 and 360 d in clay soil, which indicates that PE lead toward neutrality at long-term in this soil (Tables S3 and S4).

The indigenous SOC mineralization rate was faster at the beginning of the incubation and the rate gradually declined during the first 30-40 d of incubation (Fig. S1) and remained constant thereafter for most amended soils (Figs. S1 and S2). The mineralization of SOC continued in all treatments until the end of incubation. The SOC mineralization rate declined more rapidly in the low-C than high-C soils (Fig. S1). As a result, the total amounts of SOC released as CO₂ after 360 d were 1.5 to 4 times greater in high-C than low-C soils, depending on treatment (Fig. S2).

After 77 d, the cumulative SOC mineralized in the unamended control soils was 511 mg C kg⁻¹ soil in the high-C sand, 121 mg C kg⁻¹ in the low-C sand, 661 mg C kg⁻¹ in the high-C clay, and 179 mg C kg⁻¹ in the low-C clay (Fig. S1). After 360 d, SOC mineralization in the control soils totaled 1715, 403, 2886 and 1016 mg C kg⁻¹, respectively (Fig. S2). These amounts corresponded to 8, 5, 11 and 7% of the initial soil C content, respectively (Fig. 4a). Interestingly, in both soil textures, the proportion of SOC mineralized after 360 d was 1.6 times greater in high-C than low-C soils, indicating greater lability of C present in high-C soils, or greater recalcitrance of C present in low-C soils.

All treatments with residues increased indigenous SOC mineralization compared to the non-amended soil, especially during the first 77 d when residues decomposition was more intensive (Fig. S1). From Day 77 to 126 days, SOM mineralization rate in the control soils became greater than in the amended soils, except in the low-C sand where mineralization rates remained relatively similar between the control and the amended treatments (Fig. S2). The contrast between control and other treatments was significant every experimental time in sand soil, while it was no significant in last two evaluation dates (180 and 360 d) in clay soil (Table S3). As a result, after 360 d the amounts of SOC mineralized was less with soybean and sorghum shoots applied at 4 Mg ha⁻¹ than in the control high-C sand, less with all soybean residues than in the control high-C clay, and less with all soybean residues and sorghum shoots (4 Mg ha⁻¹) than in the control low-C clay (Fig. 4a), resulting in negatives values to cumulated PE. On the other hand, all residue treatments lead to greater SOC mineralized compared to the control in the low-C sand.

2.4.3 Mineral N

The soil mineral N was growing over experimental time, being always greater in high-C soils than low-C soils. The addition rate was significant to mineral N just at 28 days, while the other factors (C content and residue type) were significant in all dates (28, 180 and 360). The contrast also showed difference between soybean and sorghum shoot residues in both soils (sandy and clayey) during all experimental time, which was found just at first date for contrast between roots residues or when compared shoot and root (Tables S5 and S6).

2.4.4 Priming Effect

The C content was significant to PE on most dates, while residue type and addition rate were significant during all experimental time (Tables 5 and 6). At 360 d just sand soil had significant interaction C content x Residues (Tables 6). This interaction showed behavior difference between shoot and root residues when compared in initial soil C content different. The addition rate (quantity) showed no interaction with the other factors in most evaluations. At 360 d just quantity x C content was significant in clay soil for PE. This interaction indicate that addition rate of residue influences more in low-C than in high-C clay soil.

In the sandy soils, the cumulative PE increased until 18 (high-C) to 28 (low-C) days (Fig. 2). With most residue types, the values then remained stable until the end of incubation in the low-C sand, whereas they slowly increased until Day 153 in the high-C sand and declined thereafter (Fig. 3). In the clayey soils, cumulative PE reached a maximum between 77 to 98 days of incubation (Figs 2 and 3). The PE values tended to be negative until Day 226 to 270 in the low- and high-C clay, respectively, and cumulative PE reached negatives values in several treatments. After this period, cumulative PE remained stable until the end of incubation.

The C content and residue type showed significant effect on PE, in almost every experimental period. At the end (360d) just C content was no significant in clay soil. The highest cumulative PE value was observed in the low-C sand, reaching to 447 mg C kg⁻¹ soil (8 Mg ha⁻¹ soybean shoots), while in the other soils it reached a maximum of 395 mg C kg⁻¹ soil (high-C sand with 12 Mg ha⁻¹ soybean shoots), 66 mg C kg⁻¹ soil (high-C clay with 4 Mg ha⁻¹ sorghum shoots), and 130 mg C kg⁻¹ soil (low-C clay with 12 Mg ha⁻¹ of sorghum shoots), as shown in Fig. 4b. These PE values are equivalent to 5.4, 1.9, 0.2 and 0.9% of initial SOC, respectively.

In both soil types, the PE values varied as a function of initial soil C content and residue type (Table 5). An interaction between initial soil C content and residues types was found at day 6 in both soils and was mainly reflected in greater PE values with sorghum than soybean

residues in the low-C than high-C soils. An interaction was also found at Day 360 in the sandy soils mainly because cumulative PE values were lower with shoots (4 Mg ha^{-1}) than roots (both species) in the high-C sand, whereas values were similar (soybean) or greater (sorghum) with shoots than roots in the low-C sand (Fig. 3).

In the early phase of incubation (Fig. 2), sorghum shoot residues tended to induce greater cumulative PE than soybean shoot residues, especially at the greater addition rates (8 and 12 Mg ha^{-1}), in all soil except the high-C sand. This difference between plant species was maintained in the later part of incubation in the clayey soils, whereas PE values in the presence of soybean shoots became gradually similar to (high-C sand) or greater than (low-C sand) with sorghum residues at the end of incubation (Fig. 3). Soybean root residues induced greater PE in the early phase of incubation, especially in the sandy soils (Fig. 2), and this difference was maintained in the sandy soils until the end of incubation (Fig. 3), whereas PE values gradually became greater with sorghum roots than soybean roots in the clayey soils. At 126 days of incubation and afterwards, the cumulative PE induced by roots became greater than shoots residues for both species (at same addition rate; 4 Mg ha^{-1}) in the high-C sand, whereas the reverse was observed in the low-C sand (Fig. 3). In the late part of incubation PE values also tended to be greater with sorghum roots than sorghum shoots in the low-C clay.

2.5 Discussion

2.5.1 Residue C mineralization

2.5.1.1 Soil type

In general, residue C mineralized more slowly in clayey than sandy soils, especially at lower addition levels (Fig. 1). At the highest addition rate, residue C mineralization was similar between soil textures. Schmatz et al. (2017) adding residues in soil surface (3 Mg ha^{-1}), also showed faster residue mineralization in a sandy soil than clayey soil until 180 days. Physical characteristics of soil, such as soil texture, porosity, and enzymes and O_2 diffusion are important drivers of the capacity of a soil to physically protect organic C by occlusion within aggregates (Plaza et al., 2013; Pulleman and Marinissen, 2004; Six et al., 2002). Frøseth and Bleken (2015) explained the lower rates of residue-C decomposition in clayey soils by the lower diffusion rates of exo-enzymes towards C substrates, thereby reducing access of organic substrates to soil

microbes. Investigating the residence times of ^{14}C -labelled ryegrass residues over five years, Saggiar et al., (1997) showed that clay surface area regulates C turnover rate and residence times in the soil, and could be a good predictor of organic matter decomposition rates and stabilization in different soils. Therefore, the greater decomposition of crop residues in our sandy than in clayey soils could be explained by a better protection of crop residues and organic compounds residue derived by clay particles.

2.5.1.2 Residue quality and application rate

Slower decomposition rate for roots than shoot residues was already reported (Shahbaz et al., 2017; Trinsoutrot et al., 2000), and was attributed to a greater abundance of recalcitrant constituents like lignin, tannins, cutin and suberin in root tissues. The anatomy of root tissues is also important. For instance, the peripheral location of the suberin-lignin complex can restrict the accessibility of inner structures to enzymes and thus reduce access to easily decomposable compartments (Abiven et al., 2005). Studying roots from 20 different species and four botanical families, Redin et al. (2014a) did not find a good correlation between the lignin content and C mineralization, whereas the mineralization constant (k value) was negatively correlated to hemicelluloses and positively correlated to N content. Trinsoutrot et al. (2000) also demonstrated that the kinetics of C mineralization was different among oilseed rape parts (pod wall > stem > roots) and was greater with N-rich than N-poor residues. Other characteristics such as the lignin-to-N ratio (Rasse et al., 2005), and the lignin to lignin plus cellulose ratio (the lignocellulose index [LCI]) were suggested as good indices to compare chemical quality of plant residues (Whittinghill et al., 2012). However, in our study the lignin content as well as the LCI and lignin-to-N ratio (Table 2) did not clearly reflect the slower root decomposition as soybean shoots had greater lignin content and lignin-to-N ratio than sorghum roots and the highest LCI.

Several studies showed that residues with similar lignin content may decompose at different rates. The quality of lignin (monomers and condensation level) could explain this difference. For instance, plant tissues containing condensed lignin, as occur in xylem vessels and perivascular fibers, are generally the most resistant structures (Bertrand et al., 2006). Bertrand et al. (2006) also suggested that the proportions of vascular tissue and sclerenchyma cells in plant material are key determinants of plant tissue decomposability. On the other hand, Rasse et al. (2005) estimated that root-derived C has a mean residence time in soil 2.4 times

greater than shoot-derived C, but only about one fourth of this difference was explained by chemical recalcitrance. They suggested that SOM protection mechanisms (e.g., physical protection through encrustment with soil mineral particles, mycorrhiza and root-hair activities; chemical interactions with metal ions) may be more important for root than shoot tissues. The relatively low C content of root materials (34.4 - 35.4%) in our study suggests that, even though thorough washing was done, a pellicle of fine mineral particles was likely present at time of incorporation to soil, which physically protected the surface of roots from being attacked by microorganisms. The preferential incorporation of aluminium (Al) and iron (Fe) into roots tissues could be another factor contributing to the lower decomposition rate of roots noticed in the present study (Rasse et al., 2005). The plants were grown in an acid soil that had these metals available in the soil solution (Al - 1.2 cmolc L⁻¹; Fe - 1545.9 mg L⁻¹). This likely favoured their accumulation into the roots and interfered with microbial decomposition. It is possible that soil-derived C attached to the root surface was more recalcitrant than plant roots, but we believe that the first two factors (physical protection and greater root Al and Fe concentrations) were the dominant drivers. Therefore, we believe that roots showed slower decomposition than shoot materials due to a combination of several chemical and physical factors.

In the early phase of incubation (first 28 d), soybean shoots (lower C/N ratio) decomposed faster than sorghum shoots (Fig. 1; Table 3). While this difference appeared in contradiction with the greater lignin content of soybean residues (Table 2), it agreed with the higher SOL and N concentrations in soybean shoots, corroborating previous studies that attributed faster decomposition rates in the short term to higher SOL and N concentrations (Cobo et al., 2002; Redin et al., 2014a). This early difference between shoot types was even greater in the low-C soils (Fig. 1; Table 3), which may be related to lower nutrient availability in these soils.

In the high-C soils, the addition rate had greater influence on residues decomposition than residues quality. The rates of mineralization were similar among addition rates in low-C soils, whereas, in high-C soils, greater mineralization rate was found at the lowest residue addition rate. For both soil textures, all treatments at 4 Mg ha⁻¹ (except to soybean shoots added to sandy soil) induced a greater mineralization in high-C than low-C soils (Fig. 1; Table 3), in the earlier phase. This could be attributed to greater microbial biomass and nutrient availability in high-C soils (Table 1), which likely supported faster residue decomposition. At greater residue addition rate, however, this difference between high-C and low-C soils was not

observed, indicating a saturation of the capacity of high-C soils to degrade fresh organic residues.

After adding plant residues at two different rates (5.4 and 10.8 g dry mass kg⁻¹ soil) and incubating for 120 d, Shahbaz et al. (2017) found less CO₂ released from roots (29% of added C) than from leaves and stems (46 and 38%, respectively) at the low addition rate. At the high addition rate, however, roots had mineralization rate similar to the low rate (29% of added C), whereas leaves and stems decomposed 1.17 (leaves) to 1.30 (stems) times more rapidly than at the low addition rate. These rates of mineralization are similar to those found in our study after 126 d, which averaged 20-12% of root-C and 43-36% of shoot-C, in the sandy and clayey soils, respectively.

2.5.2 SOC mineralization

The effect of quality and quantity of residues added on indigenous SOC mineralization were more obvious in the low- than in the high-C soils. The residue quality effect was greater in the early phase of incubation (Fig. S1), when decomposition of the most available substrates likely occurred. In this phase, the sorghum shoot residues (with high C:N ratio) generally induced a greater mineralization of indigenous SOC than soybean residues, resulting in greater PE (Fig. 2). Greater SOC decomposition is often reported after the addition of residues with high C:N ratio and is generally attributed to the microbial attack of indigenous SOM to supply the lack of N in fresh residues (Kuzyakov et al., 2000).

The temporal dynamics of cumulative PE observed in the low-C sand was similar to results showed by Schmatz et al. (2017) using a similar soil type, where high PE values were measured during a brief initial phase of intensive decomposition, followed by a long period with near zero PE values resulting in stable cumulative PE values with time. In contrast, Shahbaz et al. (2017) reported a different kinetics, where PE was lower during the early phase of intensive residue mineralization (up to 3 weeks) and increased when residue mineralization rate declined over time. They attributed this finding to the preferential residue decomposition, generating low or even negative PE on SOC mineralization during the early phase of residue decomposition.

The three other soil types (high-C soils and low-C clay soil) were generally associated with negative PE during the second phase of incubation (Fig. 3). These negative values essentially reflected the increase in SOC mineralization rate in the unamended control soils.

The late increase in SOC mineralization in the unamended soils could be explained by the hypothesis of microbial community succession. The hypothesis states that, in the early stage of fresh organic matter decomposition, soil microbial community is mainly dominated by r-strategists (mainly bacteria), characterized by fast growth and high turnover rates, while K-strategists (mainly fungi) with slow growth and turnover rates predominate at later decomposition stages (Fontaine et al., 2003; Zhou et al., 2017). Usually, r-strategists rapidly take up the most available substrates, whereas the slow-growing K-strategists rely more on more recalcitrant polymerised compounds, such as lignocellulose (Fontaine et al., 2003). Fontaine et al. (2011) suggested that the decomposition of recalcitrant soil C pool is dependent on the presence of fresh C. They also suggested that fungi are the main cellulose decomposer and PE drivers, and are able to adjust their degradation activity to nutrient availability. Besides, Chen et al. (2014) suggested that the soil N availability is determinant to the priming of SOM mining, with an increasing contribution of K species under N limitation. In our unamended soils, it is thus likely that a microbial community succession occurred, where r-strategist species decomposed the most easily available fraction of SOM and used soil available N during the first 77 days, and then declined and were gradually replaced with slow-growing K-strategist species able to use the most stable fractions of SOM. This would explain the increase in SOC decomposition rates in the control soils during the second phase of the incubation (Fig. S2), and some negative PE values in amended soils (except the low-C sand) past that point. Our hypothesis is supported by the fact that soils with residue addition showed low and stable rates of residue-C (Fig. 1) and SOC mineralization (Fig. S2) during the second phase of incubation.

Previous studies have proposed that PE is greater in C- and N-rich soils than in poor soils (Kuzyakov et al., 2000). Our results indicate that this may not always be the case, especially under prolonged incubation. After 360 d, greater PE was found in the high-C than in the low-C clay at the lowest shoots residue addition rate (4 Mg ha^{-1} ; Fig. 4b), with roots following the same trend, in agreement with Kuzyakov et al. (2000). While, in higher addition rates, the PE showed reverse trend, with greater values in low-C than in high-C clay. In sandy soil, all treatments tended to induce greater cumulative PE in low- than in the high-C sandy. Our results highlight the need for long-term studies on PE, as proposed in others studies (Cui et al., 2017; Fontaine et al., 2011; Schmatz et al., 2017; Zhang et al., 2017).

After 360 d, the cumulative PE was lower in clayey than in sandy soils, for all treatments and at both soil initial C contents (Fig. 4b). Frøseth and Bleken (2015) also observed that PE-induced SOC mineralization was about 3 times higher in sandy than in clayey soil, when

expressed as mol of CO₂ mineralized per mol of initial SOC. However, they found the opposite trend when PE-induced SOC mineralization was expressed per kg of soil, and values in sandy soils were then about 80% of values observed in the clayey soils. In contrast, in an experiment with crop residues added on the soil surface, Schmatz et al. (2017) reported greater PE in a clayey than in a sandy soil. After 815 d of incubation with maize residues, Zhang et al., (2017) reported net positive PE in a clayey soil (Mollisol), and net negative PE in a sandy soil (Alfisol). The greater PE often observed in clayey than in sandy soils were generally attributed to greater C content, nutrient availability, and microbial biomass in the clayey soils (Schmatz et al., 2017; Zhang et al., 2017). Our results indicate that other factors such as residues quality and application rate, as discussed above, can interact with soil texture and influence the PE dynamics. This would explain the variable results reported in the literature. Therefore, it is worth highlighting the relevance of our study where two soils textures with similar SOC content (clay with 22.9 g C kg⁻¹ for high-C clay vs. 26.4 for high-C sand; 11.4 g C kg⁻¹ for low-C clay vs. 12.6 for low-C sand) were compared in a long-term incubation.

In the high-C sand and low-C clay, PE increased when the amount of residue added increased from 4 to 8 Mg ha⁻¹, but did not change significantly when addition rate was raised from 8 to 12 Mg ha⁻¹ (Fig. 4b). This is in agreement with Guenet et al. (2010) who observed that the PE intensity did not increase linearly with increasing wheat straw addition rate. They suggested that PE may level off as C input increases because the SOM fraction subject to PE is limited or, alternatively, because of the co-occurrence of two antagonist mechanisms, a positive PE induced by the increase in active microbial biomass accelerating SOM mineralization, and a negative PE induced by the preferential decomposition of added residues decreasing SOM mineralization. Such a non-linear response of PE was also reported earlier (Xiao et al., 2015). They argued that the change in substrate preference by decomposer organisms is associated to a shift in microbial community composition. They also suggested that fungal communities were responsible for the non-linear response of PE, as fungal phospholipid fatty acids (PLFAs) increased with greater litter inputs, while bacterial PLFAs were not altered. Our results reinforce the idea that PE has a non-linear response to increasing C inputs. We believe that the smaller effect of addition rates observed in the low-C sand could be due the high rate of C addition in proportion to initial SOC (C added represented 18-54% of initial SOC depending on rate). These amounts may have saturated the enzymatic system of the low-C sand, which contained less nutrients and less microbial biomass as compared to the high-C soils (Table 1). On the other hand, the high-C clay did not show a significant effect of addition rate on

cumulative PE. In that soil, the non-linear response of PE residue rate may be attributable to the preferential decomposition of added residues over the SOC, given the large amount of residue-C available in the soil.

In our study, the influence of residue type on cumulate PE varied with soil type. In the high-C sand, soybean shoots residues induced higher PE than sorghum shoots (Fig. 4). This soil had a high proportion of SOC present as easily available C (41% of initial SOC present in the coarse + fine sand fraction; Table S1 – supplementary data). In addition, compared to the unamended control, the treatments with soybean shoots generally caused net soil N mineralization (up to 41% more mineral N at the highest addition rate than in the control), whereas sorghum shoots caused net N immobilization, with on average 18% less mineral N than the control (Fig. S3). We suggest that the narrower C/N ratio of soybean shoots combined with greater SOC and N availability in the high-C sand were responsible for the higher PE intensity with soybean than sorghum shoots in this soil.

In the clayey soils, we observed a reverse trend of greater PE intensity with sorghum than soybean residues, especially in the high-C where the effect of residue quality was obvious 360 days after residue addition (Fig. 4b). In contrast with the sandy soils, the clayey soils had most of their initial SOC present as stable C (only 3 to 17% of initial SOC associated with the coarse + fine sand fraction; Table S1 – supplementary data). The relative SOC stability may also influence PE. Zhang et al., (2017) observed that PE was less responsive in soils with a higher proportion of stable SOC until 160-350 days of incubation, whereas PE was as responsive or even more responsive to C inputs in the soils with more stable SOC than in the soil with more labile SOC after 815 d of incubation. It is likely that our clay soils, with more stable SOC, supported greater population of K-strategists when compared with sandy soils, and that low-C soils contained more K-strategists than high-C soils. Thus, the wider C/N ratio of sorghum residues likely stimulated the growth of these microbes, which were able to mineralize more stable SOM (Fontaine et al., 2003), thereby raising PE intensity.

Priming effect has been generally found to be greater after addition of residues with wide C/N ratio, which is generally attributed to the need for stimulated microorganisms to mine SOM-N, thereby accelerating SOC mineralization (Kuzyakov, 2010). However, residue C/N ratio does not always show interaction with PE intensity. In an 80-d incubation in a soil with 1% C content, Guenet et al. (2010) observed that narrowing C/N ratio of inputs did not affect PE intensity. They suggested that shifts in microbial community characteristics, such as its C/N ratio, or its C assimilation efficiency, besides of N requirement could be met from re-

mineralization of microbial N. Chen et al. (2014) showed that mineral N addition accelerated mineralization of native SOM if added with sucrose, whereas the mineral N addition with plants residues accelerated the residue-C mineralization, supporting the microbial SOM mining theory that in low N easily availability stimulates the decomposition of recalcitrant SOM to acquire N. They suggested that the SOC mineralization is primed by C inputs and controlled by N availability. Schmatz et al. (2017) also showed that PE intensity can be modulated by the amounts of soluble C present in fresh plant residues, and found higher PE where residues had greater soluble fraction.

Contrary to our expectations, even though root residues had slower rates of decomposition, it induced PE intensity similar to shoots at the same input rate (4 Mg ha⁻¹). This finding is in agreement with Shahbaz et al. (2017) who demonstrated that roots may induce as intense PE as shoots, despite slower decomposition rate.

2.6 Conclusions

In summary, we found that the magnitude and direction of PE depends on several soil and crop residue characteristics, and their interactions. The PE increased with the addition rate, but its evolution is not proportional to the increasing in the addition rate. This behavior indicates that higher addition rates lead to greater absolute values of PE, but less PE produced by unit of residue-C added in the soil. In the same time, the texture showed a clear trend to influence the PE, being greater in sandy soil than in clayey soil.

Our study could contribute with knowledge about the priming effect, showing its existence in different intensity, varying over time. We could demonstrate that there are numerous factors influencing the way residue-C and soil-C are mineralized after amending the soil with fresh residues addition. However, it is difficult to predict the PE, and SOM models may have to include these numerous factors to better simulate the evolution of SOM over time. It is also important to highlight that PE may change intensity and direction at long-term, showing that long experiments maybe required to better understand the actual net effect of soil amendments on SOM priming.

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Tables

Table 1 Selected characteristics^a of the four soil types used for incubation.

Soil	Coarse Sand	Fine Sand	Silt	Clay	C	<53 μm	>53 μm	Mineral N	P	pH
	g kg ⁻¹				% of SOC		mg kg ⁻¹	mg dm ⁻³		
High-C sand	296	277	226	201	22.9	67	33	56.11	68.9	5.0
Low-C sand	286	261	220	233	11.4	94	6	15.05	15.1	4.6
High-C clay	85	188	240	487	26.4	89	11	52.28	80.2	5.0
Low-C clay	67	186	211	536	12.6	98	2	4.61	46.2	4.7

^a Coarse Sand = 2 to 0.2 mm; Fine Sand = 0.2 to 0.05 mm; Silt = 0.05 to 0.002 mm; Clay <0.002 mm; C = Carbon; P = Mehlich-I extractible phosphorus.

Table 2 Selected characteristics^a of soybean and sorghum residues used in the incubation.

Residue type		C	N	SOL	CEL	HEM	LIG	LCI	LIG/N	C/N	¹³ C
		g kg ⁻¹									Atom %
Soybean	Shoots	414.6	14.1	281	431	155	135	0.187	9.57	29.3	1.537
Sorghum		412.6	6.2	265	351	336	47	0.064	7.58	66.3	1.496
Soybean	Roots	344.3	23.2	277	357	205	157	0.218	6.77	14.8	1.605
Sorghum		354.4	9.2	261	351	291	74	0.103	8.04	38.7	1.601

^a C = total C, N = total N, SOL = soluble fraction, CEL = cellulose, HEM = hemicellulose, LIG = lignin, LCI = lignocellulose index = LIG/(CEL + HEM + LIG).

Table 3 Results of analysis of variance (ANOVA) and contrast analyses of the effects of initial soil C content, residue type, and their interaction on mineralized residue-C (% of C added) at 6, 28, 98, 180 and 360 days, as a function of soil texture.

	Mineralized Residue-C (% added)				
	6	28	98	180	360
Sandy Soil					
CContent	<0.001	<0.001	0.003	<0.001	<0.001
Residues	<0.001	<0.001	<0.001	<0.001	<0.001
CContent x Residues	<0.001	0.032	0.041	0.022	0.023
Contrasts					
High-C					
sorghum vs soybean-shoots rate 4	<0.001	0.707	0.316	0.183	0.088
sorghum vs soybean-shoots rate 8	<0.001	0.021	0.220	0.440	0.631
sorghum vs soybean-shoots rate 12	<0.001	0.425	0.657	0.430	0.144
sorghum vs soybean-roots	<0.001	0.322	0.003	0.001	0.002
shoots vs roots (rate 4)	<0.001	<0.001	<0.001	<0.001	<0.001
shoots vs roots-sorghum (rate4)	<0.001	<0.001	<0.001	<0.001	0.001
shoots vs roots-soybean (rate4)	<0.001	<0.001	<0.001	<0.001	<0.001
Low-C					
sorghum vs soybean-shoots rate 4	<0.001	0.018	0.235	0.271	0.234
sorghum vs soybean-shoots rate 8	<0.001	0.004	0.228	0.251	0.318
sorghum vs soybean-shoots rate 12	<0.001	0.017	0.445	0.962	0.643
sorghum vs soybean-roots	0.275	0.876	0.291	0.303	0.364
shoots vs roots (rate 4)	<0.001	<0.001	<0.001	<0.001	<0.001
shoots vs roots-sorghum (rate4)	0.006	<0.001	0.001	0.001	0.002
shoots vs roots-soybean (rate4)	<0.001	<0.001	<0.001	<0.001	<0.001
Clay Soil					
CContent	<0.001	<0.001	<0.001	<0.001	0.007
Residues	<0.001	<0.001	<0.001	<0.001	<0.001
CContent x Residues	0.003	0.026	0.021	0.001	0.014
Contrasts					
High-C					
sorghum vs soybean-shoots rate 4	<0.001	0.004	0.026	0.035	0.017
sorghum vs soybean-shoots rate 8	0.085	0.510	0.533	0.544	0.526
sorghum vs soybean-shoots rate 12	0.010	0.432	0.101	0.189	0.753
sorghum vs soybean-roots	0.165	0.719	0.371	0.268	0.027
shoots vs roots (rate 4)	<0.001	<0.001	<0.001	<0.001	<0.001
shoots vs roots-sorghum (rate4)	<0.001	<0.001	0.001	0.002	<0.001
shoots vs roots-soybean (rate4)	<0.001	<0.001	<0.001	<0.001	<0.001
Low-C					
sorghum vs soybean-shoots rate 4	<0.001	0.009	0.236	0.472	0.834
sorghum vs soybean-shoots rate 8	<0.001	0.001	0.572	0.746	0.273
sorghum vs soybean-shoots rate 12	<0.001	0.001	0.028	0.742	0.515
sorghum vs soybean-roots	0.009	0.412	0.159	0.054	0.010
shoots vs roots (rate 4)	<0.001	<0.001	<0.001	<0.001	<0.001
shoots vs roots-sorghum (rate4)	<0.001	<0.001	<0.001	<0.001	0.001
shoots vs roots-soybean (rate4)	<0.001	<0.001	<0.001	<0.001	<0.001

Table 4 Results of analysis of variance (ANOVA) and contrast analyses of the effect of the quantity of shoot residue added and its interactions with initial soil C content, shoot residue type (soybean and sorghum) on mineralized residue-C (% of C added) after 6, 28, 98, 180 and 360 days of incubation, as a function of soil texture.

	Mineralized Residue-C (% C added)				
	6	28	98	180	360
	Sandy				
Quantity	<0.001	<0.001	0.002	0.004	0.007
CContent xQuantity	0.069	0.333	0.401	0.223	0.282
Shoot *Quantity	0.027	0.336	0.646	0.501	0.320
CContent* Shoot*Quantity	0.870	0.793	0.744	0.678	0.520
Contrasts					
dose lin	<0.001	<0.001	<0.001	0.001	0.002
dose quad	0.275	0.720	0.840	0.922	0.896
CContent x dose lin	0.038	0.150	0.192	0.089	0.120
CContent x dose quad	0.285	0.778	0.787	0.924	0.843
Shoot per dose lin	0.011	0.657	0.959	0.700	0.429
Shoot per dose quad	0.368	0.163	0.359	0.274	0.200
CContent x Shoot per dose lin	0.825	0.695	0.660	0.433	0.316
CContent x Shoot per dose quad	0.638	0.584	0.536	0.708	0.599
	Clayey				
Quantity	<0.001	<0.001	<0.001	0.003	0.008
CContent x Quantity	0.016	0.025	0.019	0.003	0.003
Shoot x Quantity	0.033	0.146	0.098	0.072	0.067
CContent x Shoot*Quantity	0.044	0.086	0.228	0.185	0.200
Contrasts					
dose lin	<0.001	<0.001	0.001	<0.001	0.002
dose quad	0.149	0.538	0.613	0.620	0.662
CContent x dose lin	0.012	0.010	0.008	<0.001	0.001
CContent x dose quad	0.078	0.068	0.094	0.156	0.206
Shoot x dose lin	0.016	0.258	0.257	0.116	0.108
Shoot x dose quad	0.249	0.046	0.028	0.071	0.076
CContent x Shoot x dose lin	0.339	0.136	0.117	0.173	0.189
CContent x Shoot x dose quad	0.020	0.023	0.118	0.195	0.212

Table 5 Results of analysis of variance (ANOVA) and contrast analyses of the effects of initial soil C content, residues type, and their interaction on cumulative priming effect (mg C kg⁻¹ soil) after 6, 28, 98, 180 and 360 days of incubation, as a function of soil texture.

	Cumulate priming effect (mg C kg ⁻¹ soil)						
	6	28	98	180	360		
Sandy Soil							
CContent	<0.001	<0.001	0.036	0.087	<0.001		
Residues	<0.001	<0.001	<0.001	<0.001	<0.001		
CContent x Residues	<0.001	0.097	0.838	0.263	0.022		
Contrasts	High-C	Low-C			High-C	Low-C	
sorghum vs soybean-shoots rate 4	0.075	0.044	0.899	0.447	0.166	0.044	0.271
sorghum vs soybean-shoots rate 8	<0.001	<0.001	0.004	0.528	0.799	0.027	0.792
sorghum vs soybean-shoots rate 12	<0.001	<0.001	0.058	0.631	0.180	0.003	0.965
sorghum vs soybean-roots	0.003	<0.001	<0.001	0.001	0.001	0.096	0.038
shoots vs roots (rate 4)	0.393	0.149	0.381	0.344	0.870	<0.001	0.071
shoots vs roots-sorghum (rate4)	0.005	0.001	0.053	0.176	0.622	0.002	0.202
shoots vs roots-soybean (rate4)	0.056	0.060	0.455	0.979	0.471	0.005	0.176
Clayey Soil							
CContent	0.085	<0.001	<0.001	<0.001	<0.001	0.309	
Residues	<0.001	<0.001	<0.001	<0.001	<0.001	0.002	
CContent x Residues	0.032	0.857	0.682	0.180	0.180	0.055	
Contrasts	High-C	Low-C			High-C	Low-C	
sorghum vs soybean-shoots rate 4	0.067	0.091	0.036	0.163	0.122	0.478	0.063
sorghum vs soybean-shoots rate 8	0.023	<0.001	<0.001	0.013	0.119	0.207	0.177
sorghum vs soybean-shoots rate 12	<0.001	<0.001	<0.001	<0.001	0.002	0.006	0.124
sorghum vs soybean-roots	0.002	<0.001	0.417	0.337	0.316	0.222	0.041
shoots vs roots (rate 4)	0.007	0.012	0.148	0.046	0.699	0.424	0.829
shoots vs roots-sorghum (rate4)	<0.001	<0.001	0.042	0.011	0.572	0.401	0.692
shoots vs roots-soybean (rate4)	0.653	0.848	0.573	0.812	0.985	0.767	0.486

Table 6 Results of analysis of variance (ANOVA) and contrast analyses of the effect of the quantity of shoot residue added and its interactions with initial soil C content and shoot residue type (soybean and sorghum) on cumulate priming effect (mg C kg⁻¹ soil) after 6, 28, 98, 180 and 360 days of incubation, as a function of soil texture.

	Cumulate Priming Effect (mg C kg ⁻¹ soil)				
	6	28	98	180	360
	Sandy				
Quantity	<0.001	<0.001	0.003	<0.001	<0.001
Ccontent x Quantity	0.116	0.852	0.761	0.705	0.015
Shoot x Quantity	0.002	0.119	0.581	0.651	0.887
Ccontent x Shoot xQuantity	0.133	0.642	0.893	0.613	0.525
Contrasts					
dose lin	<0.001	<0.001	0.001	<0.001	<0.001
dose quad	0.088	0.039	0.193	0.035	0.025
CContent x dose lin	0.346	0.745	0.553	0.419	0.012
CContent x dose quad	0.062	0.650	0.669	0.871	0.104
Shoot x dose lin	0.001	0.206	0.841	0.973	0.729
Shoot x dose quad	0.054	0.096	0.313	0.361	0.735
CContent x Shoot x dose lin	0.048	0.476	0.752	0.684	0.263
CContent x Shoot x dose quad	0.980	0.547	0.728	0.373	0.946
	Clayey				
Quantity	<0.001	<0.001	<0.001	<0.001	0.026
Ccontent x Quantity	0.217	0.421	0.462	0.113	0.035
Shoot x Quantity	0.002	0.199	0.286	0.419	0.948
Ccontent x Shoot xQuantity	0.125	0.437	0.828	0.316	0.600
Contrasts					
dose lin	<0.001	<0.001	<0.001	<0.001	0.011
dose quad	0.253	0.740	0.755	0.737	0.366
CContent x dose lin	0.328	0.616	0.639	0.052	0.013
CContent x dose quad	0.146	0.228	0.254	0.309	0.490
Shoot x dose lin	<0.001	0.121	0.118	0.209	0.997
Shoot x dose quad	0.385	0.356	0.951	0.600	0.747
CContent x Shoot x dose lin	0.373	0.403	0.791	0.152	0.644
CContent x Shoot x dose quad	0.066	0.329	0.585	0.512	0.374

Figures

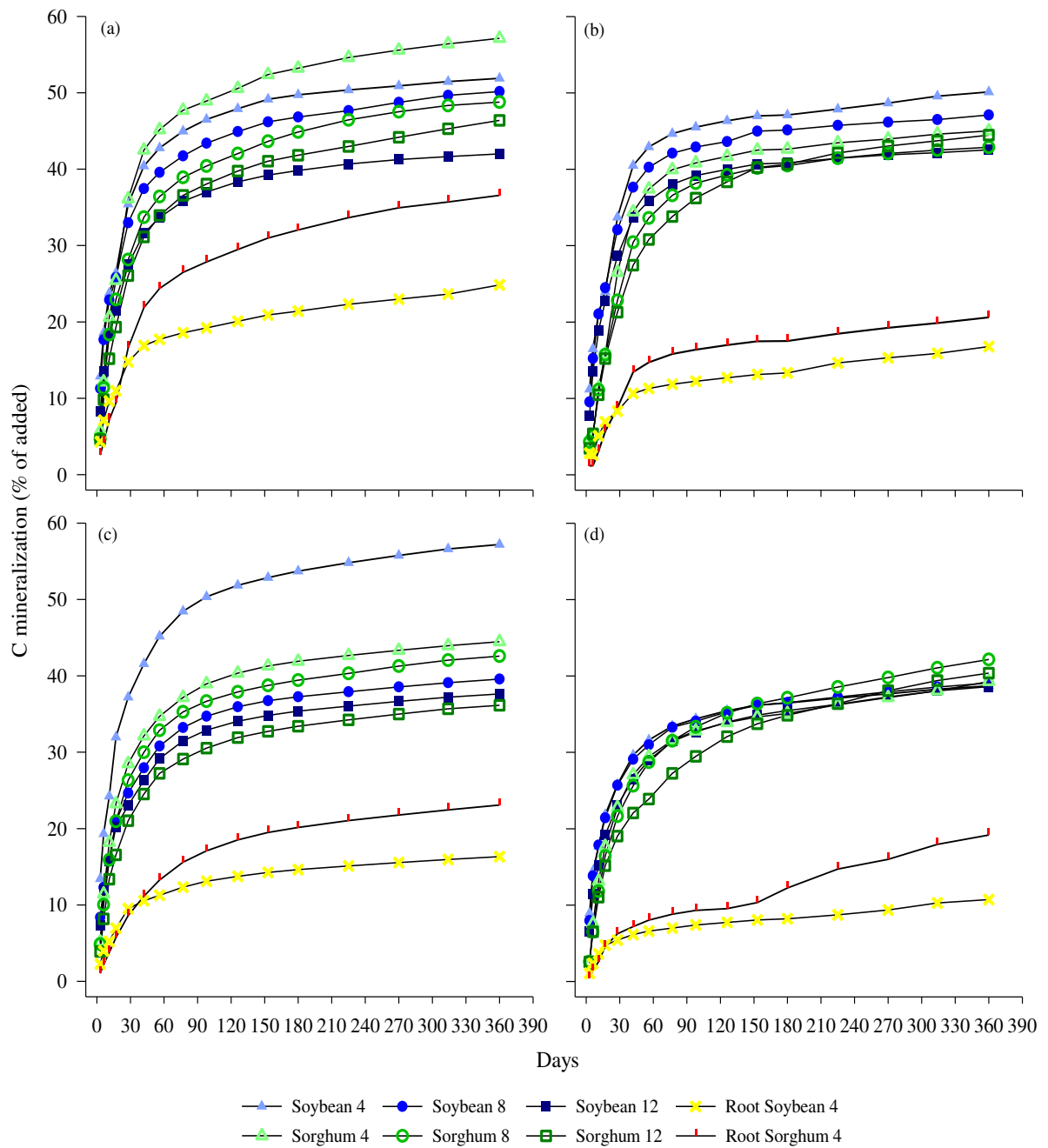


Fig. 1. Residue-derived C mineralization during 360 days in different soils (a – high-C sand; b – low-C sand; c – high-C clay; d – low-C clay) amended with different types and amounts of crop residues.

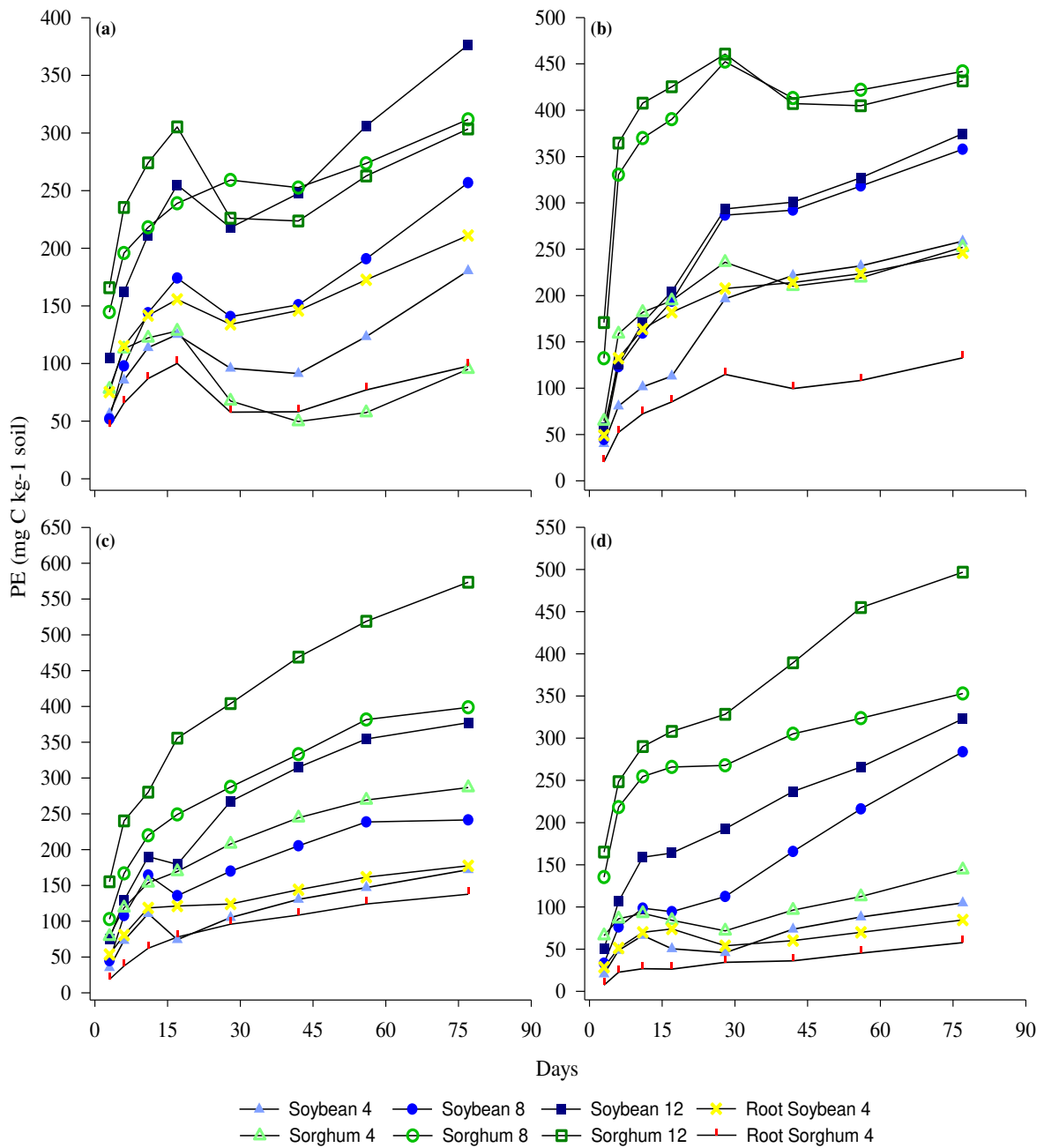


Fig. 2. Cumulative Priming Effect (mg kg^{-1}) for the first 77 days of incubation in different soils (a – high-C sand; b – low-C sand; c – high-C clay; d – low-C clay) amended with different types and amounts of crop residues. Note the different Y-axis scales for high- vs. low-C soils.

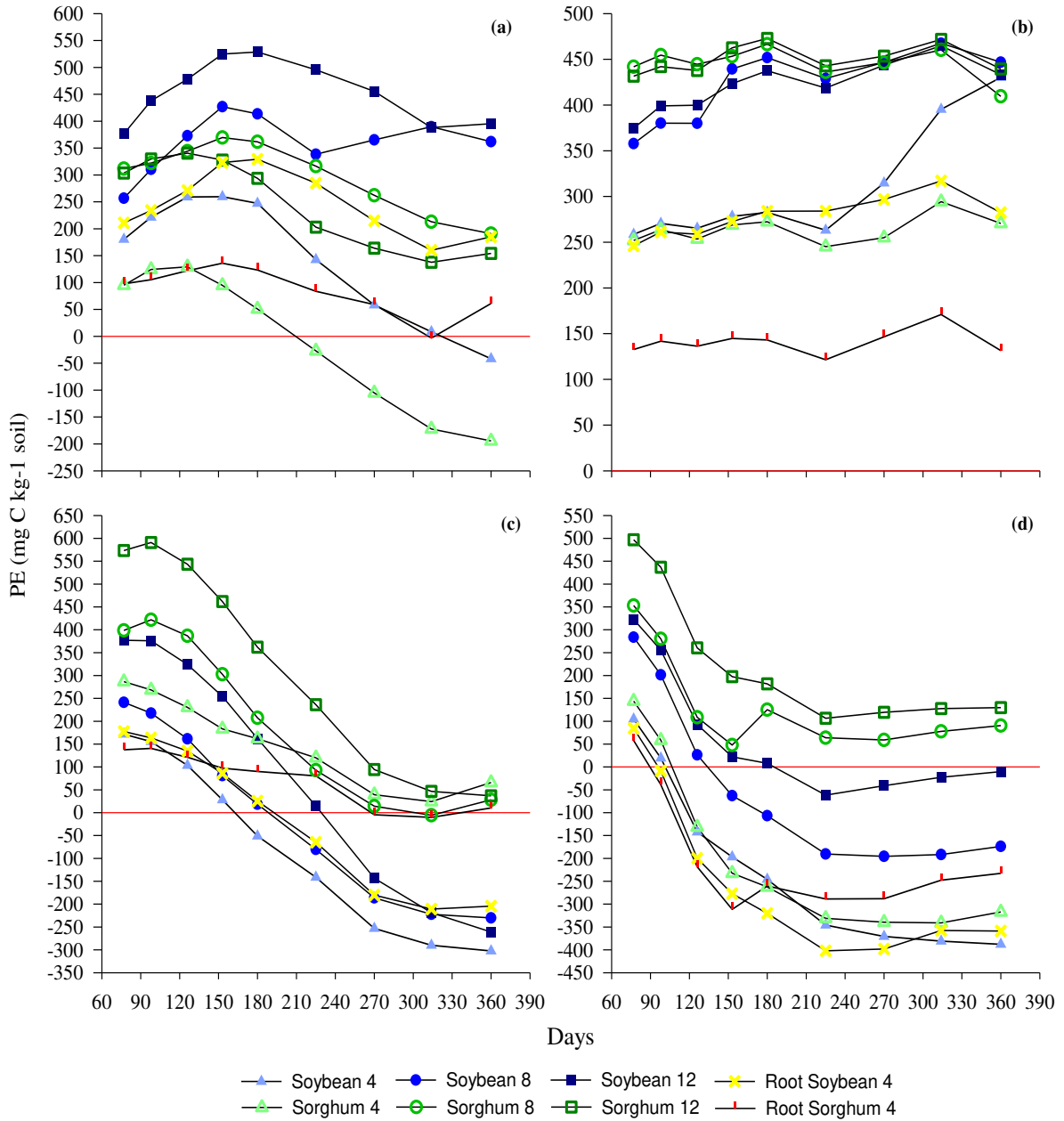


Fig. 3. Cumulative Priming Effect (mg kg⁻¹) from day 77 to 360 of incubation in different soils (a – high-C sand; b – low-C sand; c – high-C clay; d – low-C clay) amended with different types and amounts of crop residues. Note the different Y-axis scales for high- vs. low-C soils.

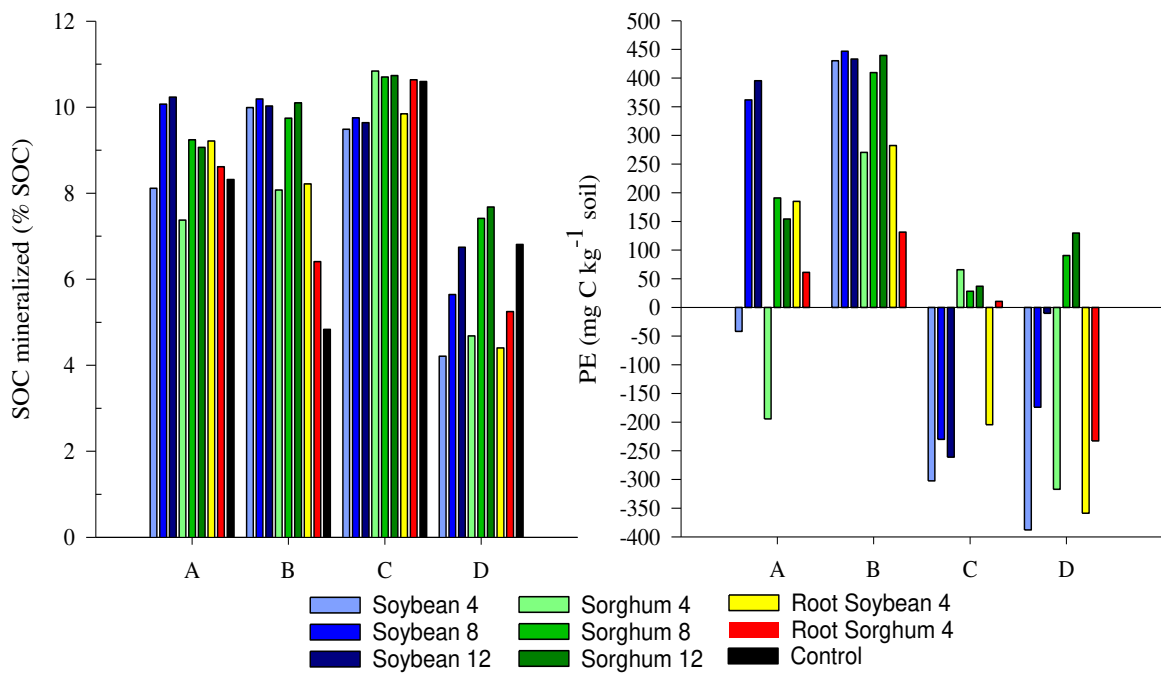


Fig. 4. Cumulative soil organic carbon mineralization and priming effect after 360 days of incubation in different soils (A – high-C sand; B – low-C sand; C – high-C clay; D – low-C clay) amended with different types and amounts of crop residues.

SUPPLEMENTARY DATA

Table S1 Total N and soil organic carbon (SOC) fractionated by mineral size in each soil of incubation.

Soil	C content	Coarse Sandy	Fine Sandy	Silt and Clay
% of total N				
Sandy	C-High	14%	19%	67%
Sandy	C-Low	2%	4%	94%
Clayey	C-High	4%	8%	89%
Clayey	C-Low	0%	2%	98%
% of SOC				
Sandy	C-High	18%	23%	58%
Sandy	C-Low	3%	4%	93%
Clayey	C-High	6%	11%	83%
Clayey	C-Low	1%	2%	97%

Table S2 Amount of C and N added in each soil, in relation to the initial soil organic C (SOC) and total N concentrations.

Residue	Quantity (Mg ha ⁻¹)	High-C sand	Low-C sand	High-C clay	Low-C clay
		C added (% of initial SOC)			
Soybean	4	7%	18%	6%	10%
Soybean	8	15%	36%	11%	20%
Soybean	12	22%	54%	17%	30%
Sorghum	4	7%	18%	6%	10%
Sorghum	8	15%	36%	11%	20%
Sorghum	12	22%	54%	17%	30%
Root Soybean	4	6%	15%	5%	8%
Root Sorghum	4	6%	15%	5%	9%
		N added (% of initial total N)			
Soybean	4	2%	5%	2%	3%
Soybean	8	4%	10%	4%	7%
Soybean	12	7%	14%	5%	10%
Sorghum	4	1%	2%	1%	2%
Sorghum	8	2%	4%	2%	3%
Sorghum	12	3%	6%	2%	5%
Root Soybean	4	4%	8%	3%	6%
Root Sorghum	4	1%	3%	1%	2%

Table S3 Results of analysis of variance (ANOVA) and contrast analyses of the effects of C content and residue type, and their interaction on cumulative SOC mineralized (mg C kg⁻¹ soil) after 6, 28, 98, 180 and 360 days of incubation, as a function of soil texture.

	Cumulate SOC mineralized (mg C kg ⁻¹ soil)									
	6		28		98		180		360	
Sandy Soil										
CContent	0.187		<0.001		<0.001		<0.001		<0.001	
Residues	<0.001		<0.001		<0.001		<0.001		<0.001	
Ccontent xResidues	<0.001		0.002		0.533		0.401		0.032	
Contrasts	High-C	Low-C	High-C	Low-C	Both soils	Both soils	High-C	Low-C		
control vs others	<0.001	<0.001	<0.001	<0.001	<0.001	0.001	0.026	<0.001		
sorghum vs soybean-shoots rate 4	0.061	0.044	0.569	0.580	0.447	0.127	0.061	0.271		
sorghum vs soybean-shoots rate 8	<0.001	<0.001	0.034	0.035	0.528	0.777	0.038	0.792		
sorghum vs soybean-shoots rate 12	<0.001	<0.001	0.867	0.033	0.631	0.139	0.005	0.965		
sorghum vs soybean-roots	0.002	0.001	0.144	0.003	0.001	0.014	0.123	0.038		
shoots vs roots (rate 4)	0.370	0.149	0.686	0.145	0.344	0.887	<0.001	0.071		
shoots vs roots-sorghum (rate4)	0.003	0.001	0.882	0.031	0.176	0.723	0.004	0.202		
shoots vs roots-soybean (rate4)	0.044	0.060	0.114	0.828	0.979	0.402	0.008	0.176		
Clayey Soil										
CContent	<0.001		<0.001		<0.001		<0.001		<0.001	
Residues	<0.001		<0.001		<0.001		<0.001		<0.001	
Ccontent xResidues	0.036		0.759		0.058		0.103		0.093	
Contrasts	High-C	Low-C	Both soils		Both soils	Both soils	Both soils			
control vs others	<0.001	0.005	0.008		<0.001	0.913	0.065			
sorghum vs soybean-shoots rate 4	0.067	0.091	0.036		0.163	0.122	0.033			
sorghum vs soybean-shoots rate 8	0.023	<0.001	0.000		0.013	0.119	0.013			
sorghum vs soybean-shoots rate 12	<0.001	<0.001	0.000		0.001	0.002	0.033			
sorghum vs soybean-roots	0.002	<0.001	0.417		0.337	0.316	0.092			
shoots vs roots (rate 4)	0.007	0.012	0.148		0.046	0.699	0.576			
shoots vs roots-sorghum (rate4)	<0.001	0.001	0.042		0.011	0.572	0.904			
shoots vs roots-soybean (rate4)	0.653	0.848	0.573		0.812	0.985	0.387			

Table S4 Results of analysis of variance (ANOVA) and contrast analyses of the effect of the quantity of shoot residue added and its interactions with initial soil C content and shoot residue type (soybean and sorghum) on cumulative SOC mineralized (mg C kg⁻¹ soil) after 6, 28, 98, 180 and 360 days of incubation, as a function of soil texture.

	Cumulative SOC mineralized (mg C kg ⁻¹ soil)				
	6	28	98	180	360
	Sandy				
Quantity	<0.001	<0.001	0.003	<0.001	<0.001
Ccontent x Quantity	0.116	0.852	0.761	0.705	0.015
Shoot x Quantity	0.002	0.119	0.581	0.651	0.887
Ccontent x Shoot xQuantity	0.133	0.642	0.893	0.613	0.525
Contrasts					
dose lin	<0.001	<0.001	0.001	<0.001	<0.001
dose quad	0.088	0.039	0.193	0.035	0.025
CContent x dose lin	0.346	0.745	0.553	0.419	0.012
CContent x dose quad	0.062	0.650	0.669	0.871	0.104
Shoot x dose lin	0.001	0.206	0.841	0.973	0.729
Shoot x dose quad	0.054	0.096	0.313	0.361	0.735
CContent x Shoot x dose lin	0.048	0.476	0.752	0.684	0.263
CContent x Shoot x dose quad	0.980	0.547	0.728	0.373	0.946
	Clayey				
Quantity	<0.001	<0.001	<0.001	<0.001	0.026
Ccontent x Quantity	0.217	0.421	0.462	0.113	0.035
Shoot x Quantity	0.002	0.199	0.286	0.419	0.948
Ccontent x Shoot xQuantity	0.125	0.437	0.828	0.316	0.600
Contrasts					
dose lin	<0.001	<0.001	<0.001	<0.001	0.011
dose quad	0.253	0.740	0.755	0.737	0.366
CContent x dose lin	0.328	0.616	0.639	0.052	0.013
CContent x dose quad	0.146	0.228	0.254	0.309	0.490
Shoot x dose lin	<0.001	0.121	0.118	0.209	0.997
Shoot x dose quad	0.385	0.356	0.951	0.600	0.747
CContent x Shoot x dose lin	0.373	0.403	0.791	0.152	0.644
CContent x Shoot x dose quad	0.066	0.329	0.585	0.512	0.374

Table S5 Results of analysis of variance (ANOVA) and contrasts showing the effect of C content (high-C and low-C), residues type (9 treatments) and their interaction on cumulate mineral N (mg N kg⁻¹ soil) assessed at 6, 28, 98, 180 and 360 days, performed for each soil texture.

	Mineral N (mg N kg ⁻¹ soil)		
	28	180	360
	Sandy		
CContent	<0.001	<0.001	<0.001
Residues	<0.001	<0.001	<0.001
Ccontent x Residues	<0.001	<0.001	0.266
Contrasts			
soybean vs sorghum	<0.001	<0.001	<0.001
root soybean vs sorghum	0.002	<0.001	0.108
shoot soybean vs sorghum	<0.001	<0.001	<0.001
Rate4 sorghum vs Rate4 soybean	<0.001	<0.001	0.004
Rate4 root vs Rate4 shoot	0.002	0.185	0.285
Rate4-sorgho root vs shoot	<0.001	0.853	0.165
Rate4-soybean root vs shoot	0.113	0.169	0.913
CContent x soybean vs sorghum	0.030	0.541	0.192
CContent x root soybean vs sorghum	0.792	0.102	0.753
CContent x shoot soybean vs sorghum	0.007	0.780	0.184
CContent x Rate4 sorghum vs Rate4 soybean	0.144	0.187	0.241
CContent x Rate4 roots vs Rate4 shoot	0.010	0.006	0.307
CContent x Rate4-sorghum roots vs shoot	0.001	0.001	0.838
CContent x Rate4-soybean roots vs shoot	0.253	0.257	0.216
	Clayey		
CContent	<0.001	<0.001	<0.001
Residues	<0.001	0.004	<0.001
Ccontent x Residues	<0.001	0.386	<0.001
Contrasts			
soybean vs sorghum	<0.001	<0.001	<0.001
root soybean vs sorghum	0.014	0.690	0.316
shoot soybean vs sorghum	<0.001	<0.001	<0.001
Rate4 sorghum vs Rate4 soybean	<0.001	0.214	0.008
Rate4 root vs Rate4 shoot	<0.001	0.759	0.012
Rate4-sorgho root vs shoot	<0.001	0.787	0.007
Rate4-soybean root vs shoot	<0.001	0.471	0.376
CContent x soybean vs sorghum	0.059	0.216	0.096
CContent x root soybean vs sorghum	0.659	0.481	0.001
CContent x shoot soybean vs sorghum	0.021	0.301	0.924
CContent x Rate4 sorghum vs Rate4 soybean	0.244	0.608	0.009
CContent x Rate4 roots vs Rate4 shoot	0.032	0.321	0.786
CContent x Rate4-sorghum roots vs shoot	0.045	0.727	0.081
CContent x Rate4-soybean roots vs shoot	0.507	0.296	0.166

Table S6 Results of analysis of variance (ANOVA) and contrasts showing the effect of C content (high-C and low-C), shoot residue type (soybean and sorghum), quantity added (4, 8 and 12 Mg ha⁻¹) and their interaction on mineral N (mg N kg⁻¹ soil) assessed at 6, 28, 98, 180 and 360 days, performed for each soil texture.

	Mineral N (mg N kg ⁻¹ soil)		
	28	180	360
	Sandy		
CContent	<0.001	<0.001	<0.001
Shoot Type	<0.001	<0.001	<0.001
CContent* Shoot	0.007	0.788	0.086
Quantity	<0.001	0.037	0.202
CContent x Quantity	<0.001	0.144	0.422
Shoot x Quantity	<0.001	0.004	<0.001
CContent x Shoot x Quantity	<0.001	0.003	0.010
Contrasts			
CContent high vs low	<0.001	<0.001	<0.001
soya vs sorgho	<0.001	<0.001	<0.001
dose lin	<0.001	0.017	0.082
dose quad	0.200	0.319	0.215
CContent x shoot	0.007	0.788	0.086
CContent x dose lin	<0.001	0.175	0.315
CContent x dose quad	0.833	0.138	0.922
Shoot x dose lin	<0.001	0.002	<0.001
Shoot x dose quad	0.284	0.256	0.570
CContent x Shoot per dose lin	<0.001	0.081	0.744
CContent x Shoot per dose quad	0.367	0.001	0.012
	Clayey		
CContent	<0.001	<0.001	<0.001
Shoot Type	<0.001	<0.001	<0.001
CContent* Shoot	0.042	0.338	0.928
Quantity	<0.001	0.320	0.141
CContent x Quantity	0.029	0.446	<0.001
Shoot x Quantity	0.002	0.023	<0.001
CContent x Shoot x Quantity	0.352	0.159	0.342
Contrasts			
CContent high vs low	<0.001	<0.001	<0.001
soya vs sorgho	<0.001	<0.001	<0.001
dose lin	<0.001	0.164	0.052
dose quad	0.186	0.581	0.830
CContent x shoot	0.042	0.338	0.928
CContent x dose lin	0.100	0.242	<0.001
CContent x dose quad	0.023	0.651	0.160
Shoot x dose lin	0.001	0.012	<0.001
Shoot x dose quad	0.585	0.217	0.629
CContent x Shoot per dose lin	0.950	0.708	0.263
CContent x Shoot per dose quad	0.161	0.064	0.344

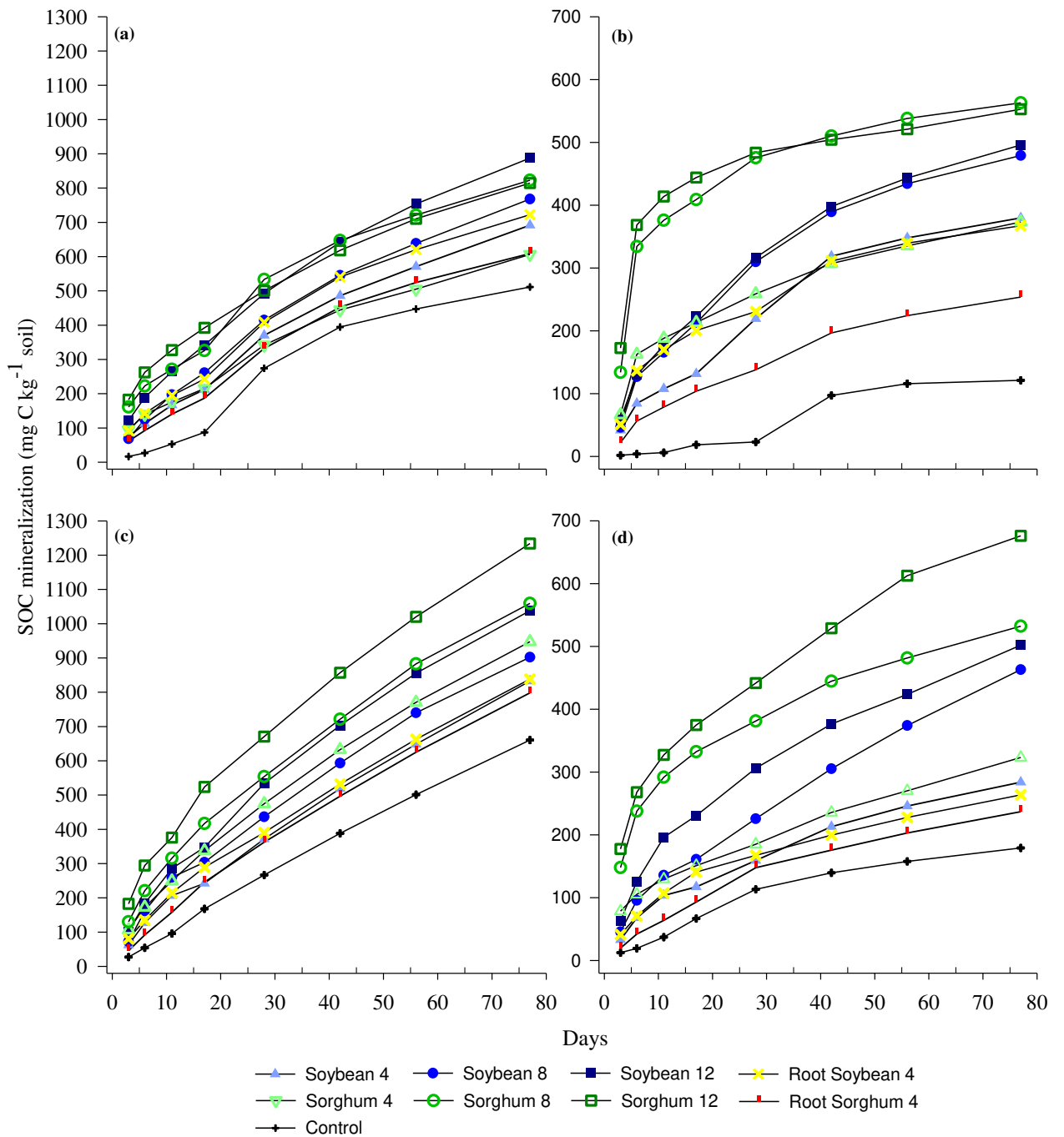


Fig. S1 Soil organic carbon (SOC) mineralization for the first 77 days of incubation in different soils (a – high-C sand; b – low-C sand; c – high-C clay; d – low-C clay) amended or not (control) with different types and amounts of crop residues. Note the different Y-axis scales for high- vs. low-C soils.

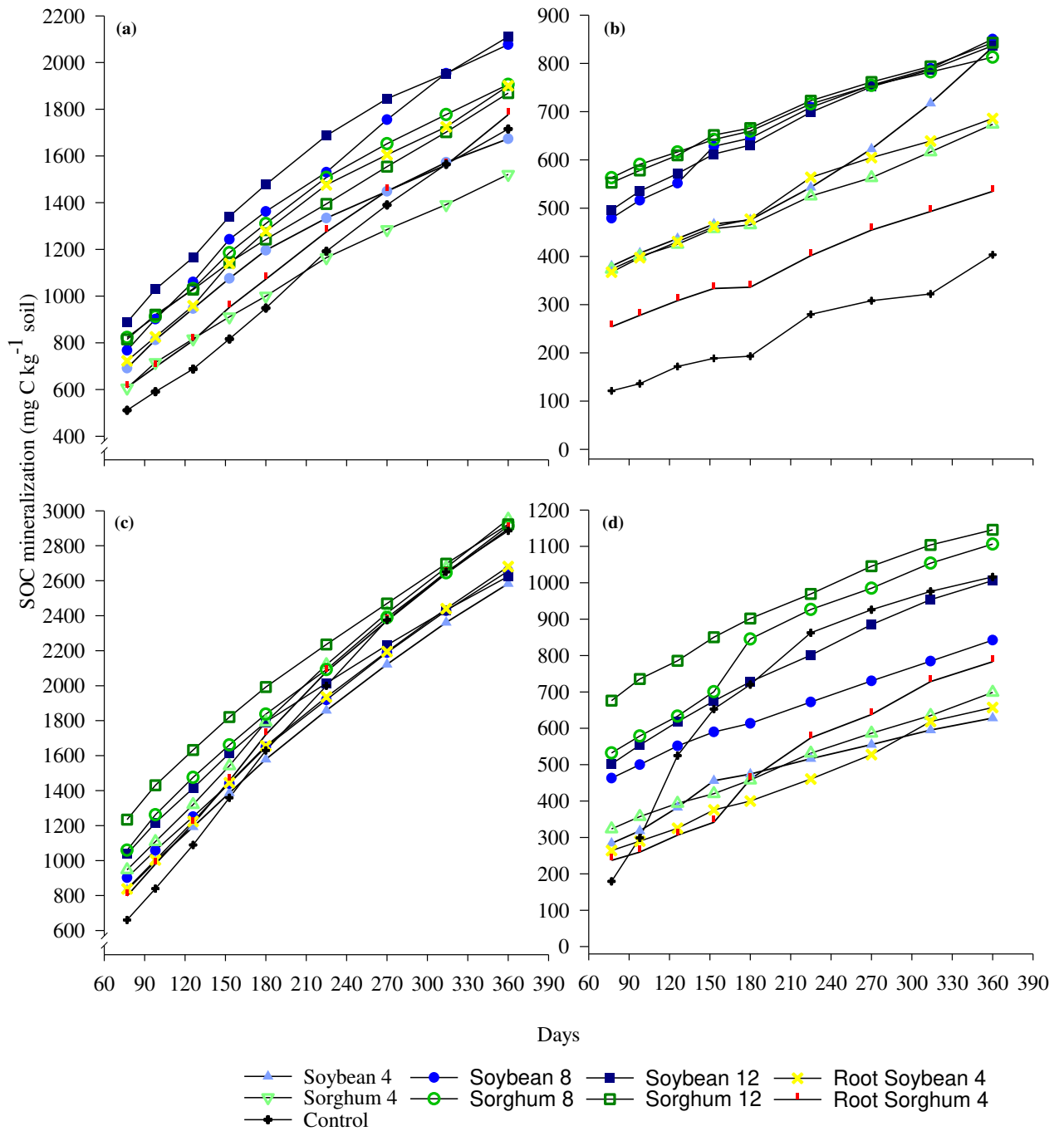


Fig. S2 Soil organic carbon (SOC) mineralization from day 77 to 360 of incubation in different soils (a – high-C sand; b – low-C sand; c – high-C clay; d – low-C clay) amended or not (control) with different types and amounts of crop residues. Note the different Y-axis scales for high- vs. low-C soils.

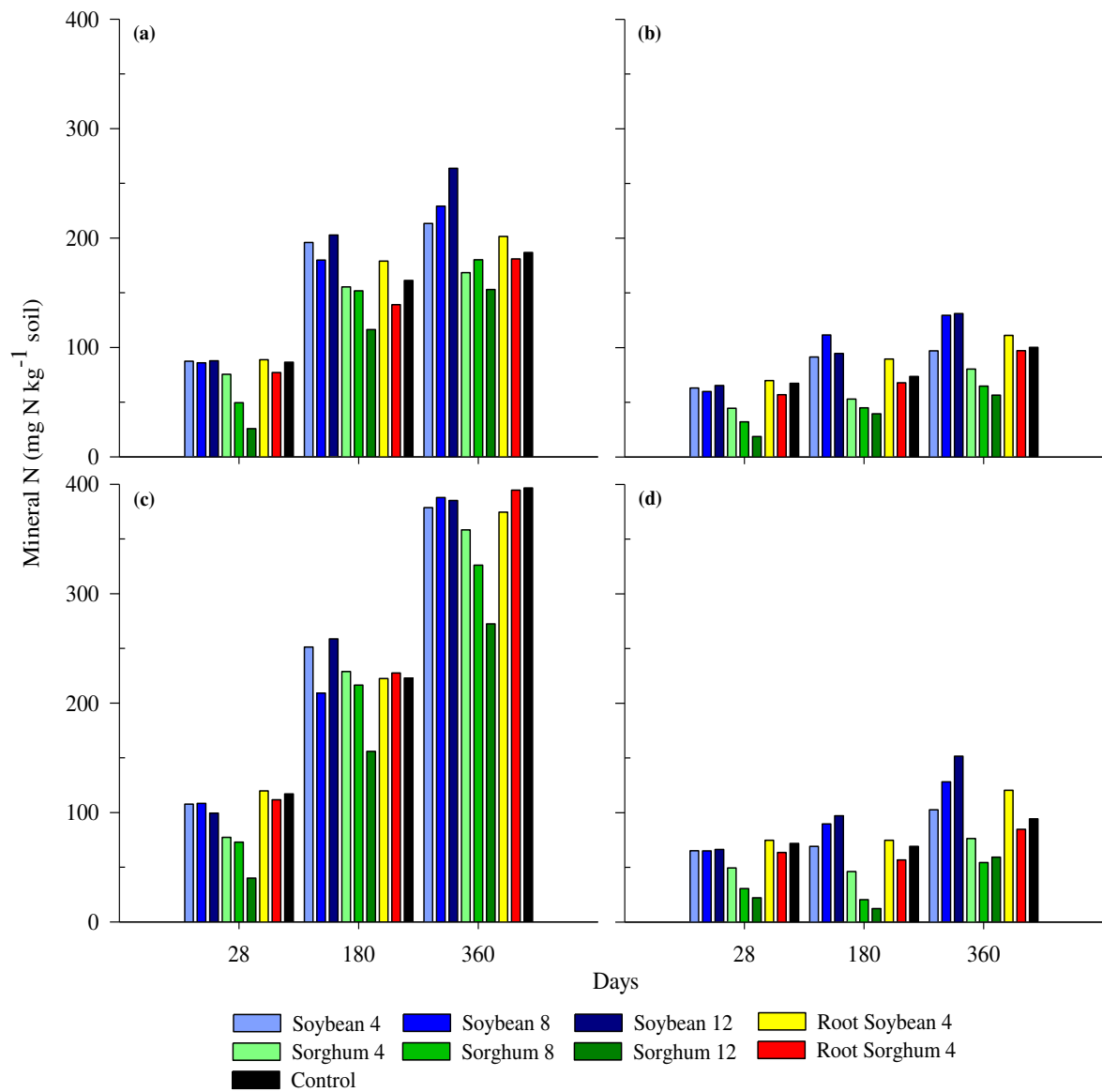


Fig. S3 Mineral N (mg kg⁻¹) in different soils (a – high-C sand; b – low-C sand; c – high-C clay; d – low-C clay) amended with different types and amounts of crop residues after 28, 180 and 360 days of incubation.

3 ARTIGO 2 - SOIL CARBON CONTENT AND RESIDUE QUALITY EFFECTS ON CARBON DISTRIBUTION IN PHYSICAL SOIL FRACTIONS²

3.1 Abstract

Carbon (C) sequestration is largely studied in crop fields because its importance to soil quality and climate change mitigation. The quantity of C stabilized in soil is governed by several factors, highlighted the quality and quantity of residues, soil texture and initial soil C content. However, is not well understood how these factors interact to determine the retention of new C in the soil after residues addition. We performed a laboratory incubation for 360 days, using ¹³C-labeled residues (shoot and root of soybean and sorghum) mixed in two soils with different textures, each one with two initial C content (sand high-C, sand low-C, clay high-C, clay low-C). Shoots residues were added in three addition rates (4, 8 and 12 Mg ha⁻¹), while roots residues were added just in the 4 Mg ha⁻¹ rate. We observed that addition rate and residue quality had effect on amount of residue-C retained in the soil during all experimental time. At 360 days, the initial C content influenced the residue-C retention (% of C added) in the silt and clay fraction and in the fine sand fraction. The percent averages of residue-C retained in the fraction associated to silt and clay minerals were greater in low-C than in high-C soils (25% x 22%, in sand soil; and 34% x 23%, in clay soil). On the other hand, high-C soil showed a compensatory effect contributing with greater amount of C retained in the fine sand fraction. Residue quality showed greater effect in low-C soils than in rich soils, since soybean contributed with a greater proportion of residue-C retained in the fine fraction than sorghum residues (38% for soybean x 22% for sorghum, in low-C; and 22% for soybean x 23% for sorghum, in high-C soils). We also highlight root residues that showed high efficiency to add new C in low-C soils. Lastly, we could demonstrate that the rate of residues addition is the main factor to determine the amount of residue-C retained in the soil. The residue quality affects the efficiency of new C retention in soil, especially in the fine fraction, but its effect interacts with the soil initial C content, with higher new C in low-C soils than in high-C soils.

Keywords: crop residue, residue quality, carbon sequestration, carbon stabilization

² Artigo elaborado conforme normas da Revista Soil Biology and Biochemistry

3.2 Introduction

The soil organic carbon (SOC) is fundamental to the sustainability of agricultural soils, maintaining the soil fertility and the productive capacity of agroecosystems. Crop residues are the main source of SOC (Schmatz et al., 2017) and management options have been proposed to sequester carbon in agricultural soils to mitigate global climate change (Dungait et al., 2012; Lal, 2002; Paustian et al., 2000). The SOC is the main pool of organic C, since soils worldwide store more carbon than terrestrial vegetation and the atmosphere (Schmidt et al., 2011).

Nowadays, have been shown that the amount of new C stabilized in the soil after adding residues is governed by several factors, such as quantity and chemical quality of residues added (Mitchell et al., 2018), soil texture (Hassink, 1997), soil C content (Poirier et al., 2013), soil fertilization (Wu et al., 2019), among others. However, it is still not completely understood how these factors interact to determine the amount and the turnover of new C stabilized in the soil.

The physical, chemical and biochemical mechanisms of C stabilization in the soil are largely studied currently regarding their importance for the C cycle (Plaza et al., 2013; Six et al., 2002; Verchot et al., 2011). The C associated to silt and clay fractions have been highlighted because tends to show turnover slower and have older average age (Balesdent, 1996; Lützow et al., 2006). Thus, there is a positive correlation between soil texture and the amount of soil organic matter (SOM) associated to silt and clay, suggesting that each soil has a C protective capacity that is directly linked to the soil texture (Hassink, 1997; Stewart et al., 2008). This strong correlation between soil texture and the C stabilization associated with mineral surfaces led to evolution of conceptual models of soil C saturation (Six et al., 2002; Stewart et al., 2007; West and Six, 2007).

Microbes and microbial products are crucial to sequestration C in the fraction of SOM associated with mineral surfaces (Plaza et al., 2013). The Microbial Efficiency-Matrix Stabilization (MEMS) model suggest that labile plant constituents are the main source of microbial products, relative to input rates, which these products of decomposition become important precursors of stable SOM by promoting aggregation and through strong chemical bonding to the mineral soil matrix (Cotrufo et al., 2013). Castellano et al. (2015) linked crop residues quality effect with the soil C saturation concept, suggesting that high quality residues tend to show greater efficiency than low quality residues to stabilize SOM in soil, but this quality effect decrease when soil approaches the C saturation.

In this context, the objective of this study was understand how the residue quality and quantity interact with the initial soil C content to determine the amount of new C in SOM size fractions, in two soil textures. Crop residues ^{13}C -labelled (shoot and root of soybean and sorghum) were mixed in four soils (two different textures, each one with two different initial soil C content) and incubated for 360 days under laboratory conditions. We hypothesized that: (i) at lower initial SOC concentration, greater it will be the residue-C stabilization in the silt and clay fraction; (ii) crop residue quality should affect the residue-C stabilization in the silt and clay fraction, with greater effect in low-C than in high-C soils.

3.3 Materials and methods

3.3.1 Soils

The soils were collected in two long-term experiments started in 1985 in Rio Grande do Sul State, Southern Brazil. The sandy clay loam (sandy soil), classified as a Typic Paluedalf (Soil Survey Staff, 2014), was collected at the Federal University of Rio Grande do Sul (30° 50' S, 51° 38' W). The clay soil, classified as Oxisol (Soil Survey Staff, 2014), was collected at the Embrapa Wheat Research Centre (28°15'S, 52°24'W). For both soils, the 0-5 cm and 10-15 cm layers were collected separately, with the objective of obtaining soils with different concentrations of soil organic carbon (SOC) but similar texture. The four soils were gently crumbled, sieved at <4 mm, and visible organic residues were removed by hand. Field-moist sieved soils were homogenized by thorough mixing, sub-sampled and stored in dark plastic bags at room temperature for 20 days until incubation. The main soil characteristics are presented in Table 1.

3.3.2 Plant residues

The plant residues used in this study were from sorghum (*Sorghum bicolor* L.) and soybean (*Glycine max* L.) grown in greenhouse, which were labelled with ^{13}C . When the plants reached the V2 vegetative stage, weekly pulse labelling with $^{13}\text{CO}_2$ started and lasted until the plants had flowered. Pulse labelling was done using a system adapted from Sangster et al. (2010). Briefly, once a week the plants were enclosed in an acrylic chamber (polymethyl methacrylate) made of 0.8 x 0.8 x 0.3 m (length, width, height) segments that were stacked

according to plant height, using a final segment closed on top. Pulses of ^{13}C were generated from the reaction of 2 M HCl with a solution of $\text{NaH}^{13}\text{CO}_3$ (33 atom % ^{13}C). Plants shoots were cut close to the ground and separated into leaves and stems. Roots were washed in running water over a sieve (2 mm). All plant parts were dried in oven at 45°C to a constant weight, and cut to 1 cm pieces. Leaves and stems were then combined in a 1:1 mass ratio. A subsample was ground to 1 mm (in a knife mill) to analyse the soluble (SOL), cellulose (CEL), hemicellulose (HEM) and lignin (LIG) according to Van Soest (1963). A neutral digestion (0.3 g of plant residue in 30 ml of neutral detergent solution) and an acid digestion (0.6 g of residue plants in 60 ml of acid detergent solution) were performed in the digester block at 150°C for 1 h. After the samples were filtered by vacuum using a filter crucible and then washed with distilled water and 30-40 ml of acetone. The fibres were dried at 105°C for 12 h. The SOL was determined from the difference in the initial weight and after neutral digestion. The difference between the total neutral fiber and acid detergent fiber determined the HEM content. Next, the acid detergent fiber was digested in H_2SO_4 12 M for 3 h to determine the CEL content by difference, and lastly, the remaining was burned in a muffle furnace at 500°C for 3 h to determine the LIG content also by difference. Another subsample was oven-dried at 65°C for 48 h to determine dry matter (DM) concentration. This sample was then finely ground to measure total C and N concentrations using an elemental analyser (FlashEA 1112, Thermo Finnigan, Milan, Italy), and ^{13}C with an isotope ratio mass spectrometer (IRMS) (DELTA V Advantage, Thermo Fisher Scientific, Bremen, Germany) after combustion in an elemental analyser (Flash EA 2000, Thermo Fisher Scientific, Bremen, Germany). The characteristics of crop residues are given in Table 2.

3.3.3 Experimental design and incubation conditions

The four soil types were mixed with either aboveground or root residues from soybean and sorghum. The aboveground plant residues (leaves and stems mixed in equal quantities) were added at rates equivalents to 4, 8 and 12 Mg ha^{-1} , whereas root residues were added only at 4 Mg ha^{-1} ; a no-residue control was included for total of nine residue treatments per soil type, resulting in 36 treatments replicated three times. One day before incubation set up, the soils received KNO_3 to standardize the content of mineral N to 60 mg kg^{-1} and water to adjust soil moisture to 70% of field capacity. In each experimental unit, 107.9 g soil (dry weight basis) were thoroughly mixed with crop residues that had been moistened with 3 mL g^{-1} of deionized

water. The soil-residue mixture was then placed in a 110 mL cylindrical acrylic pot (5.0 cm in diameter and 5.0 cm in height) in two steps: one half of the soil-residue mixture was placed in the pot and compressed to a height of 2.5 cm. The remaining soil-residue mixture was placed above the compressed soil, and compressed to a total height of 5.0 cm, to reach a final bulk density of 1.1 g cm^{-3} . Three sets of acrylic pots were prepared (total of 324 experimental units) for destructive soil sampling at pre-determined dates as described in the next sections. Two sets of acrylic pots were incubated in 2-L glass jars and one set of acrylic pots was incubated 1-L glass jars, which was used to assess C mineralization by measuring CO_2 release as described in Chapter I. A small beaker with 4 mL of water was placed in each jar to minimize soil water losses. All jars were incubated in the dark at $25 \pm 1 \text{ }^\circ\text{C}$ and were opened periodically for a few minutes, to renovate the atmosphere, and the pots were weighed and water added as required to adjust soil moisture.

3.3.4 Sampling and analytical measurements

Destructive samplings were done at 28, 180 and 360 days of incubation. The acrylic pots were removed from glass jars and each unit was gently crumbled to recover all visible remaining crop residues by hand, and sieved to $<4 \text{ mm}$. A subsample of moist soil (20 g) was taken to measure mineral N concentration as described below. The other portion of soil was air-dried. The recovered residues were dried in oven at 65°C for 48 h, and weighed. The residues and a subsample of the air-dried soil were finely ground (in mortar) to determine total C and N by dry combustion, and ^{13}C isotope abundance by mass spectrometry as described in section 3.3.2.

Soil mineral N concentration (NH_4^+ and NO_3^-) was determined by extracting moist soil in 1 M KCl (30 min shaking; soil:solution ratio, 1:4). The soil suspension was left on the bench for 30 min and the supernatant was filtered (paper filter Unifil C42, particle size retention 1-2 μm). The filtered extracts were analysed by continuous flow colorimetry (Skalar Analytical, Netherlands).

The size fractionation of the soil samples was carried out by mixing 10 g of air-dried soil with 50 mL of deionized water and 10 glass beads (3 mm diameter) in a 150-mL glass snap-cap. The slurries were shaken (horizontal shaker; 120 stroke per min) for 16 h to disrupt aggregates (Diochon et al., 2016). After shaking, the slurries were passed through two sieves of decreasing mesh size (250 and 53 μm) using distilled water and a washing bottle until the

wash water was clear. The fractions retained on sieves (>250 μm and 53-250 μm) and the fraction that has passed (<53 μm) were dried at 60 $^{\circ}\text{C}$ to a constant weight. The fractions were weighed and finely ground (in mortar) to determine total C and N and ^{13}C and ^{15}N isotope abundance as described in section 3.3.2.

3.3.5 Calculations and statistical analysis

The recovery of residue-derived C and N in whole soil and fractions was calculated as proposed previously (Poirier et al., 2013). Firstly, the $\delta^{13}\text{C}$ (in per mill) values were calculated according to the equation:

$$\delta^{13}\text{C} = \left[\frac{(^{13}\text{R}_{\text{sample}} - ^{13}\text{R}_{\text{standard}})}{^{13}\text{R}_{\text{standard}}} \right] \times 100$$

where $^{13}\text{R}_{\text{sample}}$ and $^{13}\text{R}_{\text{standard}}$ are $^{13}\text{C}/^{12}\text{C}$ ratio in soil samples and standard (international Vienna-Pee Dee Belemnite), respectively. The proportions of C and N in whole soil and fractions that was derived from crop residue was calculated as follow:

$$f_{\text{residue}} = (\delta_{\text{tr}} - \delta_{\text{c}}) / (\delta_{\text{r}} - \delta_{\text{c}})$$

where f_{residue} is the portion of C present in whole soil or in a given fraction that is derived from crop residues (in grams of residue-C per gram of SOC), δ_{tr} is the $\delta^{13}\text{C}$ of whole soil or fractions that was amended with crop residues; δ_{c} is the $\delta^{13}\text{C}$ of unamended (control) whole soil or fractions; and δ_{r} is the $\delta^{13}\text{C}$ of the initial crop residues added to the soil. For whole soil and fractions, the δ_{c} was calculated as the average of measurements made on all control soils or fractions. The recovery of C from decomposing residues was calculated with the following equation:

$$\text{Residue}_C = f_{\text{residue}} \times T$$

where Residue_C = amount of residue-derived C or N recovered in the soil or each fraction; and T = total amount of C in whole soil or fractions (g kg^{-1} of soil).

The total recovery of C and N in the different soil fractions was on average 89% and 98% of total C and N found in whole soil samples, respectively. Therefore, we adjusted the fractionation values proportionally to the whole soil content, respecting that whole soil C and N content are 100% of recover.

The net soil C balance was calculated at Day 360 of incubation, between inputs (residue-C stabilized in the soil and remaining residue-C) and output (SOC mineralization), using the follow equation:

$$\text{Net C balance} = \text{Residue-C}_{\text{ws}} + \text{Remaining-C} - \text{SOC-CO}_2$$

where Net C balance is the final result (in mg C kg⁻¹ soil) at Day 360 comparing the new C inputs and the SOC sink; Residue-C_{ws} is the sum of C recovered in every organic matter size fraction (whole soil); Remaining-C is the amount of C recovered in the visible remaining residues recovered by hand (undecomposed); and SOC-CO₂ is the amount of C mineralized from SOC.

An analysis of variance (ANOVA) was performed for each soil texture separately to test the effect of initial soil C content, residue type and their interactions on residue-C recovered in each size fraction (in % of C added) measured at 360 days, and residue-C recovered in the whole soil (in mg C kg⁻¹ soil) and mineral N measured at Days 28, 180 and 360. The mixed procedure of SAS with repeated measures (SAS Institute, 2002) was used. Homogeneity of variances was previously checked. Contrast analysis was performed when significant effects were found. We used the following contrasts to test crop residue effects: soybean vs sorghum; soybean root vs sorghum root; soybean shoot vs sorghum shoot; rate 4 soybean vs rate 4 sorghum; rate 4 root vs rate 4 shoot; rate 4 sorghum root vs rate 4 sorghum shoot; and rate 4 soybean root vs rate 4 soybean shoot. When the initial soil C content showed significant interaction with residue type, the following contrasts were used: C content x soybean vs sorghum; C content x root soybean vs sorghum; C content x shoot soybean vs sorghum; C content x Rate4 sorghum vs Rate4 soybean; C content x Rate4 roots vs Rate4 shoot; C content x Rate4-sorghum roots vs shoot; C content x Rate4-soybean roots vs shoot). The effects were considered significant at $P \leq 0.05$. A second ANOVA was carried out to test the effects of addition rate of shoot residues and its interaction with initial soil C content and shoot residue type (soybean or sorghum). Posteriorly, the followed contrasts were performed: soybean vs sorghum; rate addition linear effect; rate addition quadratic effect; C content per plant; C content per rate linear; C content per rate quadratic; Shoot per rate linear; Shoot per rate quadratic; C content x Shoot per rate linear; C content x Shoot per rate quadratic.

3.3.6 Residue-C and SOC mineralization

The total CO₂ released was measured by replacing the NaOH trap after 3, 6, 11, 17, 28, 42, 56, 77, 98, 126, 153, 180, 225, 270, 314 and 360 days of incubation. The proportion of C mineralized that was derived from the crop residues was assessed by quantification of ¹³CO₂ in the NaOH traps (Schmatz et al., 2017). Posteriorly, the SOC mineralization was calculated by difference between the total C mineralized and the residue-C mineralized.

These data were not shown in this paper, because they are compounding another paper about priming effect that is being written. Then, just the values of cumulative SOC mineralized were used to calculate the net soil C balance as final result in this paper.

3.4 Results

3.4.1 Soil Mineral N

The mineral N showed significant interaction between initial soil C content and residues types (Table 3), in short-term (28 d) and medium-term (180 d). At 360 days, both factors were significant, but without interaction. Overall, in all dates of evaluation, soybean showed greater amounts of soil mineral N than sorghum treatments, such as the high-C had mineral N content than low-C soils, in both soil textures. Sorghum residues lead to increasing N immobilization with the increment of additions rate (Figure 1). Roots residues showed stable over the time, keeping similar level to the soil non-amended (control), regardless of soil type.

3.4.2 Residue-derived C recovery in bulk soil

The residue-derived C recovery in whole soil (absolute amounts, mg kg⁻¹) was strongly affected by residue type (Table 3 and 4) and quantity in both soils (Table 5 and 6). The soil C content and its interaction with residue type and quantity had effect on amount of residue-derived C only in the short term (up to 180 days). At 360 days the interaction between soil C content and residue type was observed only when considering the proportion of C added (% of C added) that was recovered in soil. The residue-C recovery in whole soil was relatively constants over experimental time (Fig. 2). The residue effect on residue-derived recovery (mg C kg⁻¹ soil) decreased between 28 and 360 days in sandy soil, while in clay soil, it persist throughout the experiment period.

At 360 days, in soil textures, a difference in the amount of residue-C in the soil was observed only between shoot and roots regardless of species, with the roots promoting a higher amount of residue-C in soil than shoot (Fig. 3). Considering the proportion of residue-derived C (% of C added) that was recovered in whole clayey soil, the C content had significant interaction with contrasts soybean vs sorghum; root soybean vs root sorghum; ; soybean 4 vs sorghum 4; root 4 vs shoot 4; and soybean root vs soybean shoot. In sandy soil it was observed significant contrasts in interaction with the initial C content: root 4 vs shoot 4 (Table 3 and 4). These interaction contrasts are significant because the effect of residues type is more relevant in low-C than in high-C soils (Fig. 3). In sand soil, the residue-derived C recovery in low-C reached to 29%, for soybean residues, and 18% for sorghum residues (mean of all addition rates), while in high-C it was recovery 21 and 25%, respectively. In the meantime, the recovery in clay soil was 33% for soybean and 23% for sorghum, in low-C, while in high-C it was 25 and 24%, respectively.

The amounts of residue derived C in whole soil increases linearly with increasing amounts of shoots in both soils, following the order: 12 Mg ha⁻¹ > 8 Mg ha⁻¹ > 4 Mg ha⁻¹. However, the portion of residue derived C (% of initial C input) recovered in both soils do not differ between quantities regardless of crop residues (22% for soybean and 21% for sorghum, averages of rates in sand soil; and 25 and 22%, respectively, averages in clay soil).

3.4.3 Residue-derived C recovery in size fractions of SOM

In both textures soils the residue-derived C recovery (% of initial C input) in silt and clay fraction was influenced by the interaction between C content and residues types, while coarse and fine sand fractions was influenced only by the residues types and C content, respectively (Table 3 and 4; Fig. 4). Soils with low-C content showed greater response to residues quality, especially on recovery of residue-C in the fine fraction of soil. In low-C soils in C content, soybean shoots trend to be more efficient than sorghum to include new C into the whole soil and in the fine fraction (silt and clay). In these soils, shoot soybean contributed with the SOC in the fine fraction with 13-23% of C added and sorghum shoots included 9-15% of C added. Whereas, in high-C soils, the amount of new C included in the most stable fraction showed similar efficiency between soybean and sorghum shoots (9-18 and 10-18 % of C added, respectively).. Roots had greater amounts of recovered residue-C in the soil than shoots,

especially in the coarse sand fraction, since it had slower decomposition and more non-decomposed residues particles could be occluded in the soil.

The efficiency of accumulation of residue-C in the fine fraction was greater in low-C soils (16.1 and 14% of C added, sandy and clayey, respectively) than in high-C soils (12.1 and 11.1% of C added, sandy and clayey, respectively).

The addition rate of residues did not show effect on the efficiency of residue-derived C recovery in clay soil. In sand soil, the significant effect was found just on coarse sand fraction, indicating that the percent of residue-C recovery in this fraction increase with the addition rate.

3.4.4 Net soil C balance

The net soil C balance of C inputs (via crop residues addition) and C losses (from indigenous SOC mineralization) at long-term (day 360) showed interaction between the initial soil C content and residues type (Table 7). High-C soils lost high amounts of indigenous SOC, resulting in negative net C balance regardless of residue type (Fig. 5 and 6), with average loss of 903 and 1713 mg C, in sand and clay soil, respectively. Low-C soils had proportionally less expressive losses of SOC, trending to positive C balances (average 430 and 337 mg C, in sand and clay soil, respectively), especially with medium and high addition rates (8 and 12 Mg ha⁻¹).

The efficiency of C sequestration increase with the addition rates. Soybean residues tend to sequester more C than sorghum in low-C soils (643 vs 397 mg C kg⁻¹, respectively). This behavior also was observed in high-C clay soil, although with negative, where soybean reached -1360 and sorghum -1780 mg C kg⁻¹ soil. Just in high-C sand soil, it was observed reverse behavior with soybean residues showing C balance more negative than sorghum residues (-892 and -711 mg c kg⁻¹ soil, respectively).

3.5 Discussion

3.5.1 Effect of initial soil C content

In both textures, low-C soils had greater percentage of residue-C associated with silt and clay minerals fraction. The greater efficiency of residue-C accumulation in soil poorer in C can

be linked to the higher amount of available mineral surfaces to C adsorption by organo-mineral association. The amount of SOC stabilized in association with silt and clay minerals has high correlation with the soil texture (Hassink, 1997; Six et al., 2002; Stewart et al., 2007), being that soils with low-C in this fraction have greater capacity to stabilize more C from residues added (West and Six, 2007). In agree with our results, in an incubation for 2.5 years, Stewart et al. (2008) showed that in six out of the seven sites, the soils with lower C content (from C-horizon) had greater proportion of residue-C stabilized than in soils higher C content (from A-horizon), confirming the hypothesis that the C saturation deficit has effect on the amount of new C stabilized. Similar result also was observed by Poirier et al. (2013), when they added high amounts of residues ($>10 \text{ g C kg}^{-1}$ soil), where the surface soil (higher C content) stabilized less residue-C in the fine fraction ($<50 \mu\text{m}$) compared to subsoil with low-C.

Overall, the greater percent of residue-C in the fine sand fraction of high-C soils compensated the lower proportion recovered in the silt and clay fraction. This compensatory effect can be a response to soil C saturation in the fine fraction, which is stabilized by mineral association (Stewart et al., 2007). Thus, our first hypothesis tested is partially confirmed since the residue-C stabilization in the fine fraction has a significant interaction with the initial C content of the soil.

3.5.2 Interaction between residues quality and initial soil C content

Our results showed strong interaction between the initial C content of soil and the residues type, that influence the proportion of residue-derived C in the whole soil and in silt and clay size fraction on both soils at 360 days. We observed a general trend of greater efficiency to retain more C from soybean residues compared to sorghum residues, especially in low-C soils, decreasing the residue effect in high-C soils. The greater sorghum residue-C retention in high-C compared to low-C soils suggest that the greater N availability in soils richer in SOM may compensate the residue nutrient limitation, favoring a residue decomposition with more efficiency. Currently, have been proposed that residues with high quality (characterized main by high fraction soluble, low C/N ratio, low phenol content and high decomposition rate) are more efficient than low quality residues to accumulate C associated with the mineral soil fraction. The labile constituents are utilized by microbes with higher efficiency becoming the main source of microbial products. Thus, the microbial products of decomposition are the main precursor of stable SOM in association with the mineral soil matrix (promoting aggregation and

strong chemical bonding), such as proposed in the Microbial Efficiency-Matrix Stabilization (MEMS) framework by Cotrufo et al. (2013). Haddix et al. (2016), using differentially labeled plant material, showed that structural component was found only in the light fraction of SOM, while the metabolic component was found in all size SOM fractions (sand, silt, and clay fraction), reinforcing the importance of labile plants input as precursor of the mineral-bonded SOM. In contrast, Castellano et al. (2015) in a review, noticed that 55% of papers checked did not verified effect of residues quality on efficiency stabilization of new C in the soil, suggesting that the residue quality effect is linked to the level of C saturation of the soil, being this effect decrease with increasing of soil SOC content stabilized in the mineral association. Therefore, based on MEMS framework, Castellano et al. (2015) proposed that effect of litter quality on SOM stabilization is modulated by the extent of soil C saturation, where high-quality residues are stabilized in SOM with greater efficiency in higher C saturation deficit. In other words, the quality residues effect decrease with the increase of SOM content stabilized in mineral association. Our results are in agreement with this theory, since the quality effect was observed with higher relevance in low-C than high-C soils.

The roots was the most efficient residues to include new C in the clayey soils, analysing the whole soil. It is linked to greater proportion of residue-C from roots recovered in the coarse sand fraction, which also was observed in the sandy soil. The clay soil should have more ability to protect physically roots particles trough encrustment with minerals. Morphological characteristics such as anatomy of tissues (peripheral location of the suberin-lignin complex), the greater proportion of vessel tissues and its lignin type (Bertrand et al., 2006) and specific protection mechanisms (Rasse et al., 2005) may contribute to explain the more difficult mining of roots and consequently accumulate of residue-C in the coarse sand fraction. Shahbaz et al. (2017) adding high amount of residues (5.04 g kg⁻¹ soil; equivalent to 18 Mg ha⁻¹ assuming 25 cm depth and a bulk density of 1.5 g cm⁻³) showed that a greater portion of roots was incorporated into aggregates with decreased mineralization when compared to stalk and leaves.

The higher percentage of roots residue-C retained in the coarse sand fraction was compensated by lower proportions recovered in the silt and clay fraction, compared to shoots residues. This fact reinforce the evidences that roots has an important physical protection that retarded its decomposition and transformation to thinner fractions. In the sandy soil, roots residues had different behavior between the levels of initial soil C content, where residue-C retained in the fine fraction was greater in low-C soil, highlighting the soybean roots that reached to 27% of C added, being the most efficient treatment to include C in this fraction. This

fact may be due lower capacity of physical protection in the sandy soil that allowed greater degradation of these residues.

This effect from residues type was less observed in high-C soil, which agree with our hypothesis that residues quality should affect the residue-C retention in the soil with greater effect in low-C than in high-C soils. Researches about the efficiency of retention of residue-C in the soil remain controversial about the residues quality influence (Córdova et al., 2018; Dungait et al., 2012; Gentile et al., 2011; Redin et al., 2014b; Schmatz et al., 2017). The residues quality show influence on C mineralization rates (from residues and from SOM) changing the dynamic of priming effect (extra SOC mineralization) that may modify the balance of C in the soil (between C input and C loss). Schmatz et al. (2017) noticed that the residue quality modified the priming effect (PE) and the C balance, but it did not influenced the amount of recovered new C in the soil after a year. In a short-term experiment (46 days), Córdova et al. (2018) observed that high quality residues had faster mineralization and higher C accumulation in the mineral-associated organic matter (MAOM) in relation to lower quality residues, but the amount of MAOM-C accumulated per unit of residue-C mineralized was lower in soils amended with high quality residues. It is common in the literature, to be found results about residues quality where its effect on rates of decomposition and C accumulation is greater in earlier phases with a gradual decrease of differences over time (Martens, 2000). The rates of decomposition decrease fast with the high soluble fraction and are not sustained over the medium-term due to exhaustion of this most labile fraction (Cotrufo et al., 2013). This trend also was observed here in the dynamic of residue decomposition (data not showed).

The trend to loss more SOC in high-C than in low-C soils, associated to greater efficiency of new C retention in low-C soils cooperate to high-C soils, resulted in net soil C balances extremely different depending on the initial soil C content. Xu et al. (2019), using maize residue, observed negative soil C balance with 10 g C kg⁻¹ addition rate and positive values in addition rates 50 to 100 g C kg⁻¹ soil. They also relate that high addition rates of crop residues combined with long-term fertilization increase the C sequestration rate in soils with low SOC content.

High-C soils have proportionally less stabilized C in the fine fraction of soil and greater percent of labile C (in the coarse and fine sand fractions), leading to a faster C turnover, since the percent of SOC mineralized in these soils were greater than low-C soils (average 9,6% and 7,5%, respectively; data not shown). Then, the amounts of new C recovered in the soil were not enough to compensate the C output in high-C soils, independently of treatment. Only low-C

soils had positive C balances at long-term, showing general trend to sequester C in the soil. These results confirm that initial soil C content and its lability are fundamental to define size of C losses, influencing directly the net soil C balance. Our results, show that although residue type and addition rates influence the efficiency of C stabilization in the soil, the size of soil sink (influenced by initial SOC) is dominant to determine the successful, or not, to the C sequestration. The interaction between residues type and C content show greater effect of residue quality in low-C than in high-C soil, which soybean residues tend to provide more C sequester than sorghum, following the similar behaviour already observed in the amount of new C recovered in the soil. Therefore, results of net soil C balance reinforce the relevance of residue quality for low-C soils. Schmatz et al. (2007) reported negative C balance was found with vetch in both soil textures (sand and clay) and with pea in the clay soil, while wheat residue provide positive C balances in both soils. They relate negative C balance with the greater priming effect (additional native SOC mineralization) observed when added residues with greater initial soluble C fraction (vetch and pea).

3.6 Conclusions

The initial SOC content interact with residue quality to govern the amount of residue-C stabilized in the fraction associated to minerals silt and clay. The addition of residues in low-C soils reach greater percent amounts of residue-C recovered in the fine fraction of soil, which indicate that C stabilization by association with silt and clay minerals is more efficient in low-C than in high-C, independently of the texture, confirming our first hypothesis. It is reinforced by the compensatory retention of residue-C in the fine sand fraction of high-C soils.

In low-C soils, sorghum shoot residues were less efficient than soybean to add new C in the fraction associated to silt and clay minerals. Therefore, our second hypothesis was confirmed, since the residue quality show greater effect in low-C soils than in high-C soils. We also should highlight root residues that show highest capacity to add new C in low-C soils, both in the whole soil and in the fine fraction.

Our study could reinforce that residue quality is an important factor that influences the efficiency of residue-C retention in the soil. However, its effect depends on the interaction with the soil C content. High quality residues tend to be more efficient to include new C and N in the soil, but this quality effect decrease in soils rich in C, which corroborates with our second hypothesis.

3.7 References

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Tables

Table 1 Selected characteristics^a of the four soil types used for incubation.

Soil	Coarse sand	Fine sand	Silt	Clay	C	% of SOC		Mineral N	P	pH
	g kg ⁻¹					<53 µm	>53 µm	mg kg ⁻¹	mg dm ⁻³	
High-C sand	296	277	226	201	22.9	67	33	56.11	68.9	5.0
Low-C sand	286	261	220	233	11.4	94	6	15.05	15.1	4.6
High-C clay	85	188	240	487	26.4	89	11	52.28	80.2	5.0
Low-C clay	67	186	211	536	12.6	98	2	4.61	46.2	4.7

^a Coarse Sand = 2 to 0.2 mm; Fine Sand = 0.2 to 0.05 mm; Silt = 0.05 to 0.002 mm; Clay <0.002 mm; C = Carbon; P = Mehlich-I extractible phosphorus.

Table 2 Selected characteristics^a of soybean and sorghum residues used in the incubation.

Residue type		C	N	SOL	CEL	HEM	LIG	LCI	LIG/N	C/N	¹³ C	
		g kg ⁻¹										Atom %
Soybean	Shoots	414.6	14.1	281	431	155	135	0.187	9.57	29.3	1.537	
Sorghum		412.6	6.2	265	351	336	47	0.064	7.58	66.3	1.496	
Soybean	Roots	344.3	23.2	277	357	205	157	0.218	6.77	14.8	1.605	
Sorghum		354.4	9.2	261	351	291	74	0.103	8.04	38.7	1.601	

^a C = total C, N = total N, SOL = soluble fraction, CEL = cellulose, HEM = hemicellulose, LIG = lignin, LCI = lignocellulose index = LIG/(CEL + HEM + LIG).

Table 3 Analysis of variance (ANOVA) performed for sandy soils to test the effect of initial C content and residues type and their interactions on measured variables and results of analysis of contrasts.

	Residue-C recovery in whole soil						Residue-C recovery in size fractions of SOM			Mineral N		
	28 d	180 d	360 d	28 d	180 d	360 d	CS	FS	SC	28 d	180 d	360 d
	mg kg ⁻¹			% of C added						mg kg ⁻¹		
CCont	0.586	0.056	0.872	0.362	0.049	0.636	0.915	<0.001	0.003	<0.001	<0.001	<0.001
Residus	<0.001	<0.001	<0.001	0.008	0.090	0.194	0.001	0.183	0.004	<0.001	<0.001	<0.001
Ccontent x Residues	0.001	0.110	0.140	0.044	0.157	0.010	0.060	0.057	<0.001	<0.001	0.001	0.266
Contrasts												
soybean vs sorghum	<0.001	0.080	0.057	0.110	0.393	0.102	0.180	0.036	0.007	<0.001	<0.001	<0.001
root soybean vs sorghum	0.634	0.562	0.141	0.795	0.519	0.105	0.208	0.025	<0.001	0.002	<0.001	0.108
shoot soybean vs sorghum	<0.001	0.050	0.184	0.002	0.509	0.587	0.663	0.671	0.478	<0.001	<0.001	<0.001
4-sorghum vs 4-soybean	0.750	0.404	0.053	0.868	0.467	0.069	0.221	0.014	<0.001	<0.001	<0.001	0.004
4-root vs 4-shoot	<0.001	0.012	0.037	0.005	0.015	0.038	0.001	0.872	0.497	0.002	0.178	0.285
4-sorghum root vs 4-sorghum shoot	<0.001	0.004	0.075	0.013	0.085	0.459	0.002	0.161	0.021	<0.001	0.894	0.165
4-soybean root vs 4-soybean shoot	0.129	0.092	0.120	0.067	0.032	0.027	0.047	0.233	0.153	0.113	0.080	0.913
CContent x soybean vs sorghum	0.294			0.239	0.595	0.001			<0.001	0.030	0.538	<0.001
CContent x root soybean vs root sorghum	0.566			0.654	0.473	0.034			0.108	0.792	0.095	<0.001
CContent x shoot soybean vs shoot sorghum	0.376			0.146	0.717	0.008			0.003	0.007	0.778	<0.001
CContent x 4-sorghum vs 4-soybean	0.329			0.321	0.276	0.005			0.005	0.144	0.180	<0.001
CContent x 4-roots vs 4-shoot	0.004			0.010	0.046	0.019			0.001	0.010	0.004	<0.001
CContent x 4-sorghum roots vs shoot	0.006			0.041	0.159	0.286			0.006	<0.001	0.006	<0.001
CContent x 4-soybean roots vs shoot	0.103			0.063	0.093	0.019			0.057	0.253	0.152	<0.001

CS = Coarse sand; FS = Fine sand and SC = Silt and Clay

Table 4 - Analysis of variance (ANOVA) performed for clay soils to test the effect of initial C content and residues type and their interactions on measured variables and results of analysis of contrasts.

	Residue-C recovery in whole soil						Residue-C recovery in size fractions of SOM			Mineral N		
	28 d	180 d	360 d	28 d	180 d	360 d	CS	FS	SC	28 d	180 d	360 d
	mg kg ⁻¹			% of C added						mg kg ⁻¹		
CContent	0.956	0.035	0.618	0.805	0.000	0.117	0.342	0.029	0.033	<0.001	<0.001	<0.001
Residues	<0.001	<0.001	<0.001	0.000	0.000	0.000	<0.001	0.267	<0.001	<0.001	<0.001	<0.001
Ccontent x Residues	0.027	0.001	0.150	0.081	0.001	0.010	0.169	0.155	0.023	0.006	0.340	<0.001
Contrasts												
soybean vs sorghum	<0.001	0.005	0.017	0.001	0.013	0.013	0.094	0.205	0.018	<0.001	<0.001	<0.001
root soybean vs sorghum	0.100	0.424	0.043	0.142	0.373	0.003	0.068	0.255	0.483	0.001	0.690	0.316
shoot soybean vs sorghum	<0.001	0.005	0.078	0.000	0.019	0.247	0.536	0.415	0.019	<0.001	<0.001	<0.001
4-sorghum vs 4-soybean	0.026	0.315	0.015	0.030	0.469	0.007	0.092	0.114	0.045	<0.001	0.213	0.008
4-root vs 4-shoot	<0.001	<0.001	<0.001	0.000	0.000	0.000	<0.001	0.102	0.002	<0.001	0.759	0.012
4-sorghum root vs 4-sorghum shoot	<0.001	<0.001	0.011	0.003	0.000	0.018	0.006	0.243	0.117	0.002	0.778	0.007
4-soybean root vs 4-soybean shoot	<0.001	<0.001	0.009	0.001	0.000	0.000	<0.001	0.239	0.004	<0.001	0.476	0.376
CContent x soybean vs sorghum	0.047	0.778			0.330	0.062			0.015	0.056		0.096
CContent x root soybean vs root sorghum	<0.001	0.969			0.934	0.209			0.809	0.445		0.001
CContent x shoot soybean vs shoot sorghum	0.796	0.780			0.292	0.145			0.004	0.033		0.924
CContent x 4-sorghum vs 4-soybean	0.007	0.065			0.037	0.053			0.076	0.216		0.009
CContent x 4-roots vs 4-shoot	0.250	0.015			0.004	0.025			0.949	0.029		0.786
CContent x 4-sorghum roots vs shoot	0.407	<0.001			0.001	0.081			0.122	0.045		0.081
CContent x 4-soybean roots vs shoot	0.018	0.589			0.400	0.137			0.144	0.535		0.166

CS = Coarse sand; FS = Fine sand and SC = Silt and Clay;

Table 5 Analysis of variance (ANOVA) performed for sandy soils to test the effect of initial C content, crop residue type (only shoots residues treatments) and residues quantity and their interactions on measured variables and results of analysis of contrasts.

	Residue-C recovery in whole soil						Residue-C recovery in size fractions of SOM			Mineral N		
	28 d	180 d	360 d	28 d	180 d	360 d	CS	FS	SC	28 d	180 d	360 d
	mg kg ⁻¹			% of C added						mg kg ⁻¹		
CContent	0.003	0.836	0.236	0.010	0.905	0.143	0.676	<0.001	0.435	<0.001	<0.001	<0.001
Plant	<0.001	0.050	0.184	0.003	0.511	0.587	0.665	0.674	0.468	<0.001	<0.001	<0.001
Ccontent x Plant	0.309	0.237	0.038	0.149	0.718	0.008	0.717	0.216	0.003	0.007	0.788	0.086
Quantity	<0.001	<0.001	<0.001	0.975	0.009	0.295	0.017	0.422	0.664	<0.001	0.037	0.202
CContent x Quantity	0.158	0.909	0.317	0.382	0.791	0.212	0.056	0.176	0.044	<0.001	0.144	0.422
Plant x Quantity	0.004	0.368	0.721	0.459	0.998	0.476	0.295	0.459	0.278	<0.001	0.004	<0.001
CContent x Plant x Quantity	0.829	0.030	0.851	0.557	0.015	0.580	0.593	0.564	0.596	<0.001	0.003	0.010
Contrasts												
CContent high vs low	0.003	0.836	0.236	0.010	0.905	0.143	0.676	<0.001	0.435	<0.001	<0.001	<0.001
soybean vs sorghum	<0.001	0.050	0.184	0.003	0.511	0.587	0.665	0.674	0.468	<0.001	<0.001	<0.001
dose lin	<0.001	<0.001	<0.001	0.887	0.002	0.123	0.013	0.201	0.911	<0.001	0.017	0.082
dose quad	0.893	0.890	0.666	0.863	0.515	0.922	0.011	0.843	0.374	0.200	0.319	0.215
CContent x plante	0.309	0.237	0.038	0.149	0.718	0.008	0.717	0.216	0.003	0.007	0.788	0.086
CContent x dose lin	0.175	0.781	0.357	0.683	0.552	0.888	0.078	0.702	0.369	<0.001	0.175	0.315
CContent x dose quad	0.165	0.786	0.132	0.189	0.702	0.082	0.020	0.072	0.019	0.833	0.138	0.922
Plant x dose lin	0.001	0.165	0.719	0.222	0.979	0.309	0.752	0.265	0.288	<0.001	0.002	<0.001
Plant x dose quad	0.547	0.957	0.414	0.869	0.950	0.508	0.112	0.594	0.229	0.284	0.256	0.570
CContent x Plant x dose lin	0.559	0.013	0.966	0.288	0.004	0.308	0.304	0.393	0.318	<0.001	0.081	0.744
CContent x Plant x dose quad	0.872	0.675	0.619	0.912	0.365	0.861	0.690	0.530	0.904	0.367	0.001	0.012

CS = Coarse sand; FS = Fine sand and SC = Silt and Clay

Table 6 Analysis of variance (ANOVA) performed for clay soils to test the effect of initial C content, crop residue type (only shoots residues treatments) and residues quantity and their interactions on measured variables and results of analysis of contrasts.

	Residue-C recovery in whole soil						Residue-C recovery in size fractions of SOM			Mineral N		
	28 d	180 d	360 d	28 d	180 d	360 d	CS	FS	SC	28 d	180 d	360 d
	mg kg ⁻¹			% of C added						mg kg ⁻¹		
CContent	0.500	0.335	0.507	0.480	0.121	0.583	0.059	0.004	0.246	<0.001	<0.001	<0.001
Plant	<0.001	0.005	0.079	0.000	0.033	0.214	0.537	0.442	0.022	<0.001	<0.001	<0.001
Ccontent x Plant	0.787	0.780	0.200	0.708	0.332	0.120	0.124	0.874	0.006	0.042	0.338	0.928
Quantity	<0.001	<0.001	<0.001	0.825	0.311	0.293	0.112	0.829	0.217	<0.001	0.320	0.141
CContent x Quantity	0.215	0.936	0.240	0.299	0.650	0.203	0.134	0.704	0.111	0.029	0.446	<0.001
Plant x Quantity	0.080	0.011	0.505	0.700	0.221	0.455	0.563	0.422	0.273	0.002	0.023	<0.001
CContent x Plant x Quantity	0.744	0.021	0.486	0.722	0.032	0.386	0.532	0.766	0.284	0.352	0.159	0.342
Contrasts												
CContent high vs low	0.500	0.335	0.507	0.480	0.121	0.583	0.059	0.004	0.246	<0.001	<0.001	<0.001
soybean vs sorghum	<0.001	0.005	0.079	0.000	0.033	0.214	0.537	0.442	0.022	<0.001	<0.001	<0.001
dose lin	<0.001	<0.001	<0.001	0.803	0.121	0.137	0.051	0.639	0.174	0.000	0.164	0.052
dose quad	0.544	0.583	0.964	0.533	0.960	0.655	0.445	0.701	0.264	0.186	0.581	0.830
CContent x plante	0.787	0.780	0.200	0.708	0.332	0.120	0.124	0.874	0.006	0.042	0.338	0.928
CContent x dose lin	0.554	0.778	0.776	0.832	0.372	0.527	0.485	0.598	0.158	0.100	0.242	<0.001
CContent x dose quad	0.100	0.945	0.161	0.125	0.845	0.097	0.061	0.522	0.110	0.023	0.651	0.160
Plant x dose lin	0.038	0.135	0.966	0.930	0.387	0.411	0.605	0.895	0.138	0.001	0.012	<0.001
Plant x dose quad	0.390	0.339	0.286	0.440	0.126	0.346	0.353	0.198	0.545	0.585	0.217	0.629
CContent x Plant x dose lin	0.889	0.610	0.496	0.806	0.161	0.789	0.504	0.906	0.530	0.950	0.708	0.263
CContent x Plant x dose quad	0.454	0.096	0.226	0.417	0.013	0.182	0.370	0.478	0.148	0.161	0.064	0.344

CS = Coarse sand; FS = Fine sand and SC = Silt and Clay

Figures

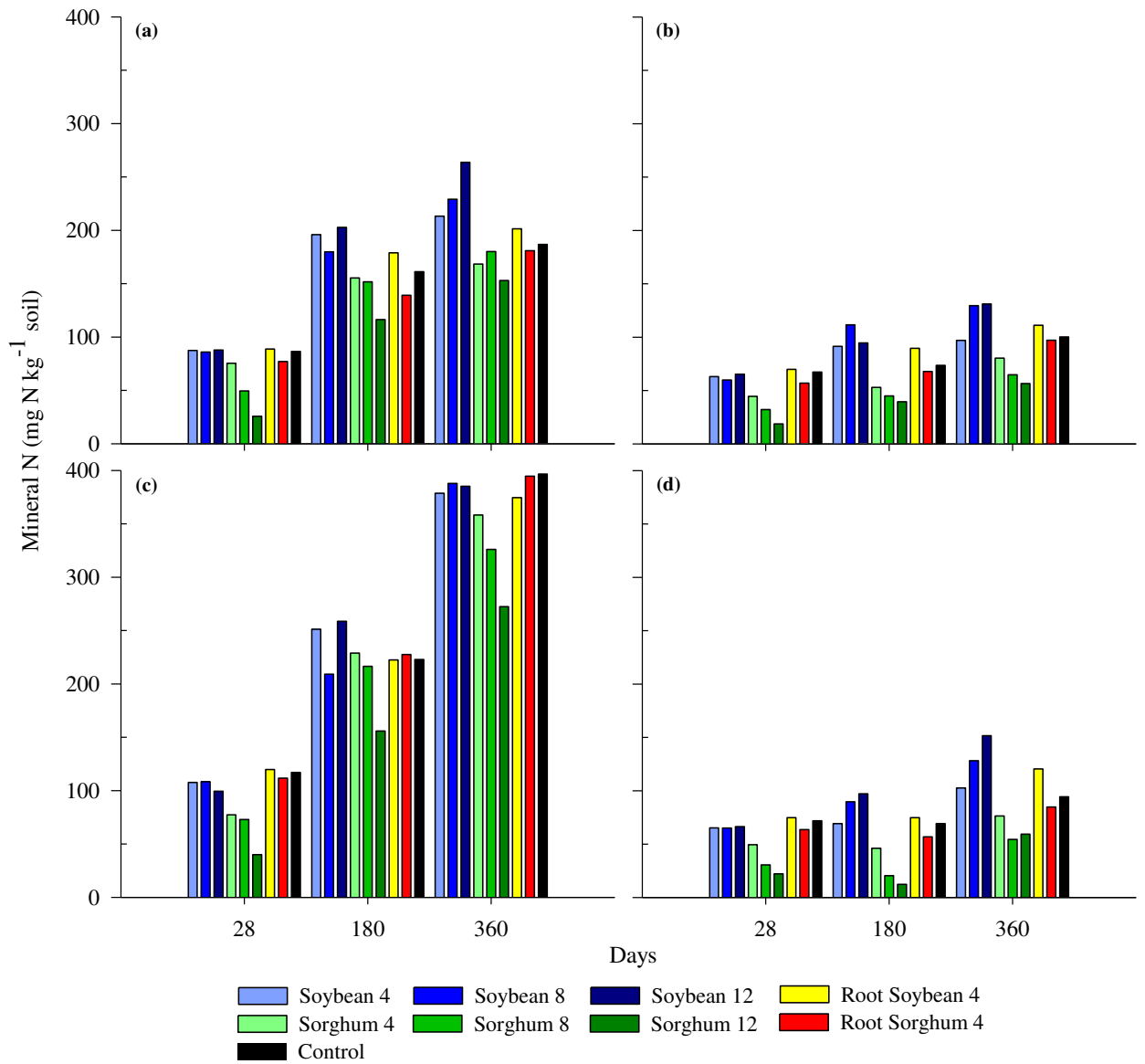


Fig. 1. Mineral N (mg kg⁻¹) in each incubated soil (a – high-C sand; b – low-C sand; c – high-C clay; d – low-C clay) at 28, 180 and 360 days of incubation.

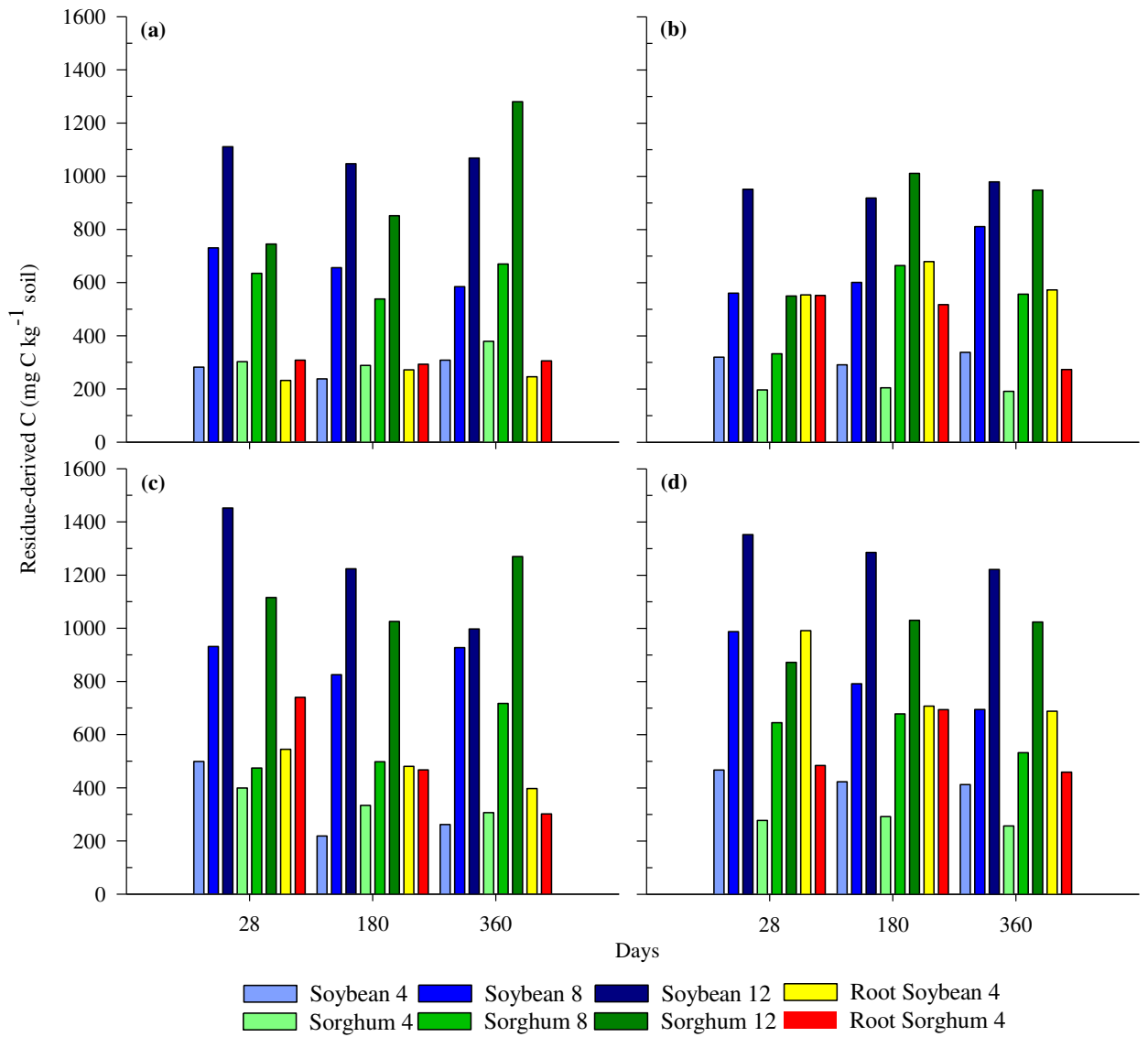


Fig. 2. Residue-derived C recovery in the whole soil (mg kg⁻¹) measured at 28, 180 and 360 days of incubation.

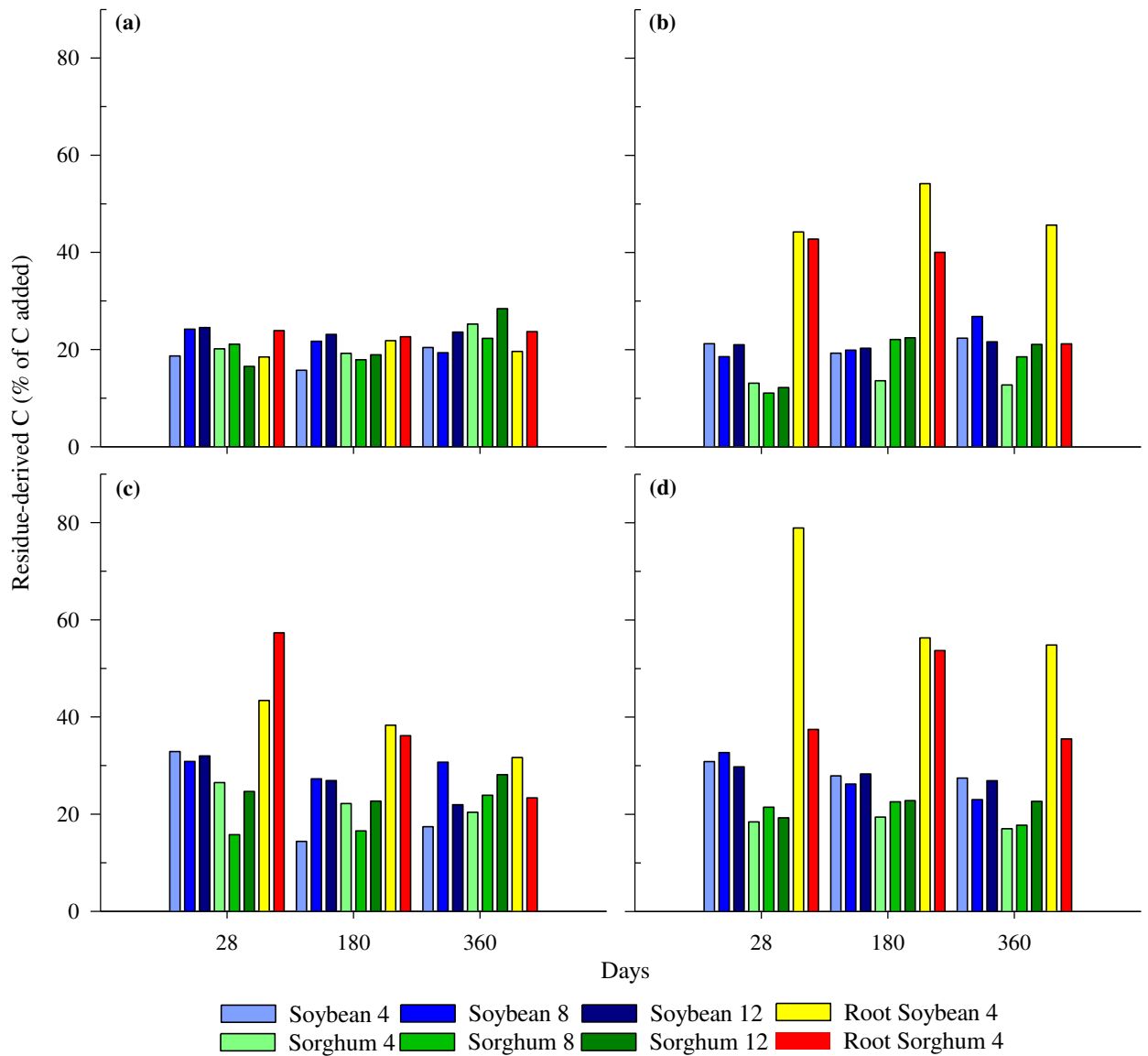


Fig. 3. Residue-derived C recovery in the whole soil (% of C added) measured at 28, 180 and 360 days of incubation. (a – high-C sand; b – low-C sand; c – high-C clay; d – low-C clay).

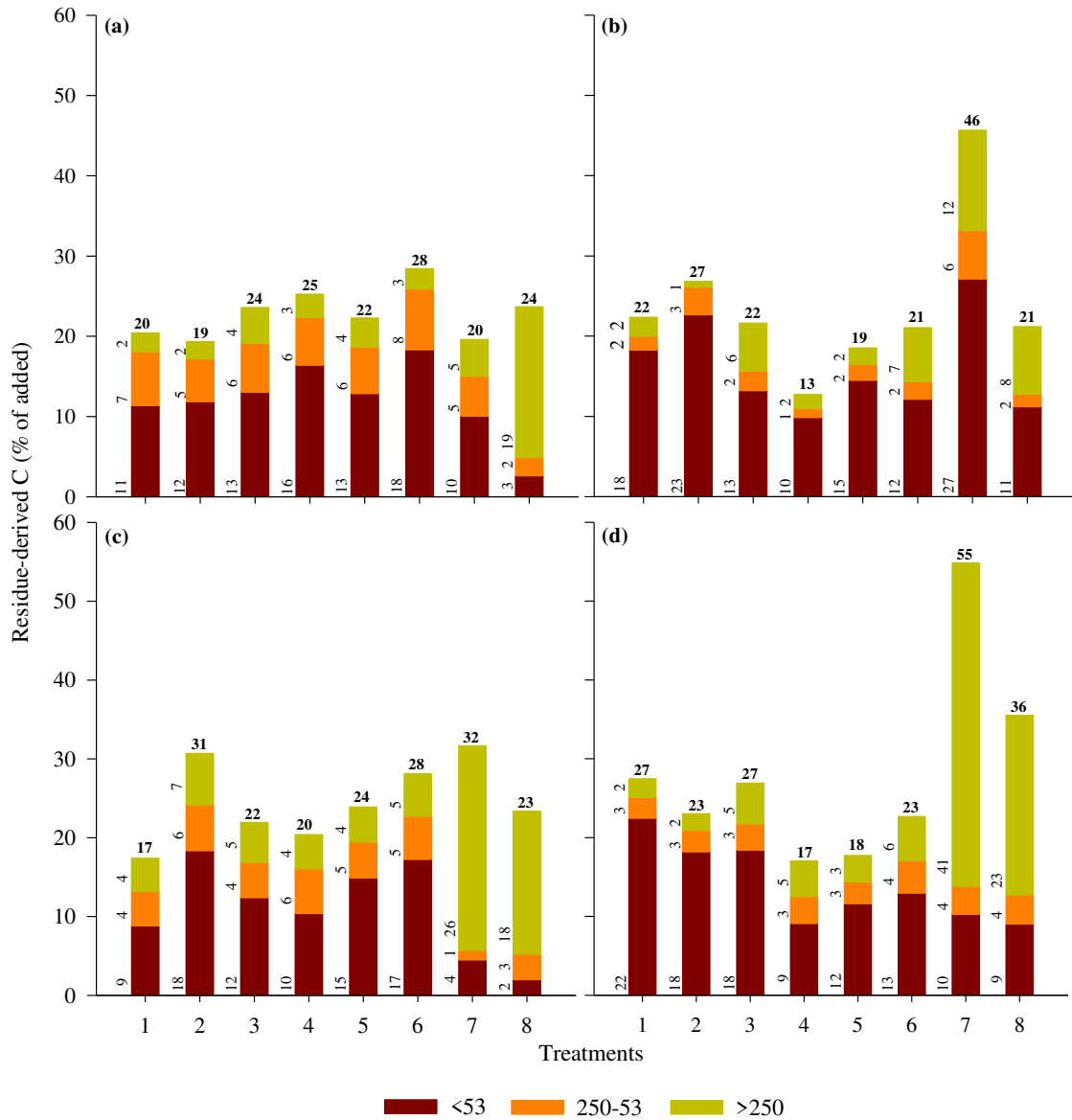


Fig. 4. Residue-derived C recovery in three size fractions of SOM (coarse sand, >250 μm; fine sand, 53 – 250 μm and silt and clay, < 53 μm) in different soil types (a – high-C sand; b – low-C sand; c – high-C clay; d – low-C clay) amended with different types and amounts of crop residues (1 – Soybean, 4 Mg ha⁻¹; 2 – Soybean, 8 Mg ha⁻¹; 3 – Soybean, 12 Mg ha⁻¹; 4 – Sorghum, 4 Mg ha⁻¹; 5 – Sorghum, 8 Mg ha⁻¹; 6 – Sorghum, 12 Mg ha⁻¹; 7 – Soybean Root, 4 Mg ha⁻¹; 8 – Sorghum Root, 4 Mg ha⁻¹; 9 – Control, without residue) at day 360.

4 DISCUSSÃO GERAL

O teor inicial de C no solo teve forte influência sobre a quantidade de MOS mineralizada após a adição dos resíduos, apresentando maiores quantidades de C mineralizado nos solos mais ricos em C, como era esperado. Entretanto, esta tendência não se reflete em maior efeito priming (EP) acumulado a longo prazo (360 dias). Assim, o maior teor de C no solo não foi determinante para um maior EP ao final do período avaliado, com os maiores valores de EP acumulado observados no solo arenoso com baixo teor inicial de C. Em geral, solos mais ricos em C e N são propensos a apresentarem maior EP, sendo que o mesmo aumenta com a quantidade de substâncias orgânicas adicionadas ao solo, especialmente com aquelas ricas em C solúvel (Kuzuyakov et al., 2000). Essa tendência foi observada neste trabalho até 98 e 180 dias para os solos arenoso e argiloso, respectivamente. Entretanto, ao final do período de incubação (360 dias), o EP acumulado foi maior no solo arenoso baixo-C comparado ao solo rico, enquanto que no solo argiloso o EP apresentou valores semelhantes nos dois níveis de C. A exceção do solo arenoso baixo-C, foi observado nos demais solos uma fase tardia de EP negativo, que reduziu os valores acumulados ao final do período e minimizou as diferenças entre os solos. A fase de EP negativo ocorre porque a mineralização do C do solo passa a ser mais intensa no tratamento controle do que nos tratamentos que receberam a adição de resíduos. Esta intensificação é justificada pela hipótese de sucessão da comunidade microbiana, onde na fase inicial predominam microrganismos de crescimento rápido (principalmente bactérias), decompondo rapidamente compostos mais lábeis, que gradativamente são substituídos por microrganismos de crescimento mais lento (predominantemente fungos) que aumentam a sua participação a medida que restam compostos mais recalcitrantes e diminui a disponibilidade de N (Fontaine et al., 2003; Zhou et al., 2017).

Em três dos quatro solos incubados, os resíduos de parte aérea de sorgo apresentaram uma tendência a acumular um maior EP ao final do período (360 dias) do que os resíduos de soja. O EP causado pela adição de resíduos orgânicos com elevada relação C/N tem sido justificado pelo efeito de mineração de N (Chen et al., 2014), onde ocorre o aumento da decomposição da MOS para suprir a baixa disponibilidade do nutriente no resíduo. Em contrapartida, o solo arenoso, rico em C, foi o único em que os resíduos de parte aérea de soja apresentaram maior EP que os resíduos de sorgo. Este comportamento pode estar ligado às diferenças nas características da MOS presente neste solo, o qual apresenta os menores percentuais de C e N presentes na fração associada aos minerais silte e argila (58% do C e 67%

do N presentes no solo), em detrimento de uma maior concentração nas frações areia grossa e areia fina que totalizam 42% do C e 33% do N do solo. A MOS presente nas frações areia costumam apresentar uma maior C/N e a menor estabilidade em relação à fração silte e argila. Portanto, a adição de resíduos de soja pode ter favorecido a decomposição das frações menos estáveis da MOS a partir do aumento da disponibilidade N no solo proveniente da mineralização deste tipo de resíduo.

Os solos mais pobres em C apresentaram tendência de estabilizar mais C na fração associada aos minerais silte e argila do solo, especialmente quando receberam a adição de resíduos de soja ou raízes. Já os resíduos de parte aérea de sorgo apresentaram comportamento similar entre os solos ou com tendência a maior eficiência nos solos mais ricos em C. Portanto, a interação entre o tipo de resíduo e o teor de C do solo influencia a eficiência de retenção de C novo no solo. Assim, os resíduos de soja (com melhor qualidade química) confirmaram o maior potencial para maior eficiência retenção de C novo na fração silte e argila nos solos com baixo teor inicial de C, sendo que essa diferença de eficiência entre os tipos de resíduos é reduzida em solos mais ricos em C, corroborando com o proposto por Castellano et al. (2015). Compostos mais lábeis quando decompostos resultam em mais produtos microbianos que são precursores de MOS associada aos minerais (Cotrufo et al., 2013; Haddix et al., 2016). Já em solos ricos em MOS, apresentam uma elevada disponibilidade de N mineral (Tabela 1 – Artigo I) podendo compensar a baixa disponibilidade nos resíduos de menor relação C/N. Isso pode justificar porque a eficiência de retenção de C novo na fração fina do solo sofreu menor influência da qualidade dos resíduos adicionados.

Como resultado final, observa-se que o conteúdo de C inicial do solo e o tipo de resíduo apresentaram interação sobre o balanço líquido de C no solo ao final do período experimental (Tabela 1). Os solos com alto teor de C apresentaram altas quantidades de C mineralizado da MOS, resultando em balanços líquidos negativos, independentemente do tipo de resíduo adicionado (Figuras 1 e 2). Os solos pobres em C inicial, por sua vez, apresentam maior facilidade para apresentar balanço líquido positivo, sendo os resultados mais favoráveis quando adicionados resíduos de melhor qualidade química e/ou quando aumentada a taxa de adição de resíduo ao solo. Xu et al. (2019) também verificaram balanço líquido de C negativo no solo após a adição de resíduos de milho na taxa de 10 g C kg⁻¹ de solo, atingindo valores positivos nas taxas de adição de 50 e 100 g C kg⁻¹ de solo. Os autores também ressaltam que as altas taxas de adição de resíduos, combinadas com a aplicação de fertilizantes de longo prazo, aumentam a taxa de sequestro de C no solo com menor teor inicial de SOC. Por vezes, a recuperação de C

novo na fração fina do solo se mostra maior em solos ricos em MOS do que em solos pobres, especialmente em taxas de adição de resíduos menos elevadas, conforme observado por Poirier et al. (2013) em que até a dose de 10 g C kg^{-1} de solo a retenção de C foi superior no solo mais rico, enquanto que em doses superiores a curva não linear de acúmulo de C na fração fina do solo sugere a aproximação do nível de saturação de C no solo rico.

No presente estudo, possivelmente a maior proporção de C presente nas frações menos estáveis da MOS (frações areia grossa e areia fina), nos solos ricos em C, foi determinante para estimular uma decomposição do C do solo em maior intensidade do que os níveis verificados nos solos mais pobres em C. O percentual de C mineralizado nos solos ricos em C foi 1,6 vezes maior do que os percentuais mineralizados nos solos pobres, confirmando um menor tempo de ciclagem do C e maior labilidade da MOS presente nos solos ricos. Assim, as quantidades de C adicionado não foram suficientes para compensar as perdas de C nos solos mais ricos, independentemente do tipo e das quantidades de resíduos adicionados. Em contrapartida, nos solos com baixo teor inicial de C, observa-se balanço líquido de C positivo, especialmente a partir da dose de 8 Mg ha^{-1} de resíduo adicionado. Além disso, os resíduos de parte aérea de soja apresentaram uma tendência a gerar balanços de C mais positivos do que os resíduos de sorgo, quando adicionados nos solos com menor teor inicial de C, reforçando a relevância da qualidade dos resíduos neste tipo de solo. Essa diferença se torna ainda mais relevante se analisarmos a quantidade de C proveniente dos resíduos recuperado nas frações físicas da MOS (desconsiderando a quantidade de C remanescente nos resíduos), onde os resíduos de melhor qualidade química se mostram ainda mais eficientes em adicionar C novo na MOS.

5 CONCLUSÃO GERAL

A maior concentração inicial de C no solo implica em uma mineralização do COS nativo mais intensa. Entretanto, a longo prazo, isso não resulta em um maior efeito priming acumulado. As taxas de adição de resíduos mais elevadas resultam em maiores valores absolutos de efeito priming, mas proporcionalmente menores em relação à quantidade de C adicionado.

Os solos pobres em C, tendem a apresentarem maior eficiência de retenção de C na fração fina do solo (associada aos minerais silte e argila) quando recebem resíduos de melhor qualidade química (maior teor de N, menor relação C/N e maior fração solúvel). No mesmo sentido, o efeito da qualidade dos resíduos sobre a eficiência de retenção de C na fração fina do solo é mais significativo nos solos pobres em C, onde resíduos de soja foram mais eficientes do

que os resíduos de sorgo, enquanto que nos solos ricos em C as diferenças entre resíduos foram reduzidas.

Por fim, a qualidade dos resíduos contribui para um balanço de C mais positivo nos solos pobres em C, especialmente sob taxas de adição mais elevadas. Enquanto isso, nos solos ricos em C, os tratamentos utilizados não sofrem suficientes para compensar as perdas de C do solo.

Do ponto de vista do plantio direto, os resultados apresentados mostram que a capacidade de retenção de C novo possui um gradiente no perfil do solo, chamando a atenção para a importância de estratégias de manejos que visem a incorporação de C nas camadas mais profundas do solo, onde a eficiência de retenção nas frações mais estáveis da MOS é maior.

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<<https://doi.org/10.1016/j.soilbio.2017.08.014>>.

Tabelas

Tabela 1 - Resultados da análise de variância (ANOVA) mostrando o efeito do conteúdo inicial de C (C inicial), dos resíduos e sua interação sobre o balanço líquido de C no solo e análise de contrastes.

	Balanço líquido de C (mg C kg ⁻¹ soil)	
	Solo arenoso	
C inicial	<0,001	
Resíduos	<0,001	
C inicial x Resíduos	0,001	
Contrastes	Baixo-C	Alto-C
controle vs outros	<0,001	<0,001
sorgo vs soja	0,131	0,016
sorgo vs soja-parte aérea	0,137	0,076
sorgo vs soja-parte aérea 4 Mg ha ⁻¹	0,276	0,228
sorgo vs soja-parte aérea 8 Mg ha ⁻¹	0,016	0,139
sorgo vs soja-parte aérea 12 Mg ha ⁻¹	0,194	0,651
sorgo vs soja-raiz	0,763	0,056
parte aérea vs raiz (4 Mg ha ⁻¹)	0,002	0,057
parte aérea vs raiz-sorgo (4 Mg ha ⁻¹)	0,032	0,313
parte aérea vs raiz-soja (4 Mg ha ⁻¹)	0,004	0,083
	Solo argiloso	
C inicial	<0,001	
Resíduos	<0,001	
C inicial x Resíduos	0,052	
Contrastes	Baixo-C	Alto-C
controle vs outros	<0,001	<0,001
sorgo vs soja	0,004	0,011
sorgo vs soja-parte aérea	0,003	0,017
sorgo vs soja-parte aérea 4 Mg ha ⁻¹	0,323	0,081
sorgo vs soja-parte aérea 8 Mg ha ⁻¹	0,172	0,077
sorgo vs soja-parte aérea 12 Mg ha ⁻¹	0,002	0,183
sorgo vs soja-raiz	0,573	0,300
parte aérea vs raiz (4 Mg ha ⁻¹)	0,253	0,113
parte aérea vs raiz-sorgo (4 Mg ha ⁻¹)	0,305	0,127
parte aérea vs raiz-soja (4 Mg ha ⁻¹)	0,547	0,438

Figuras

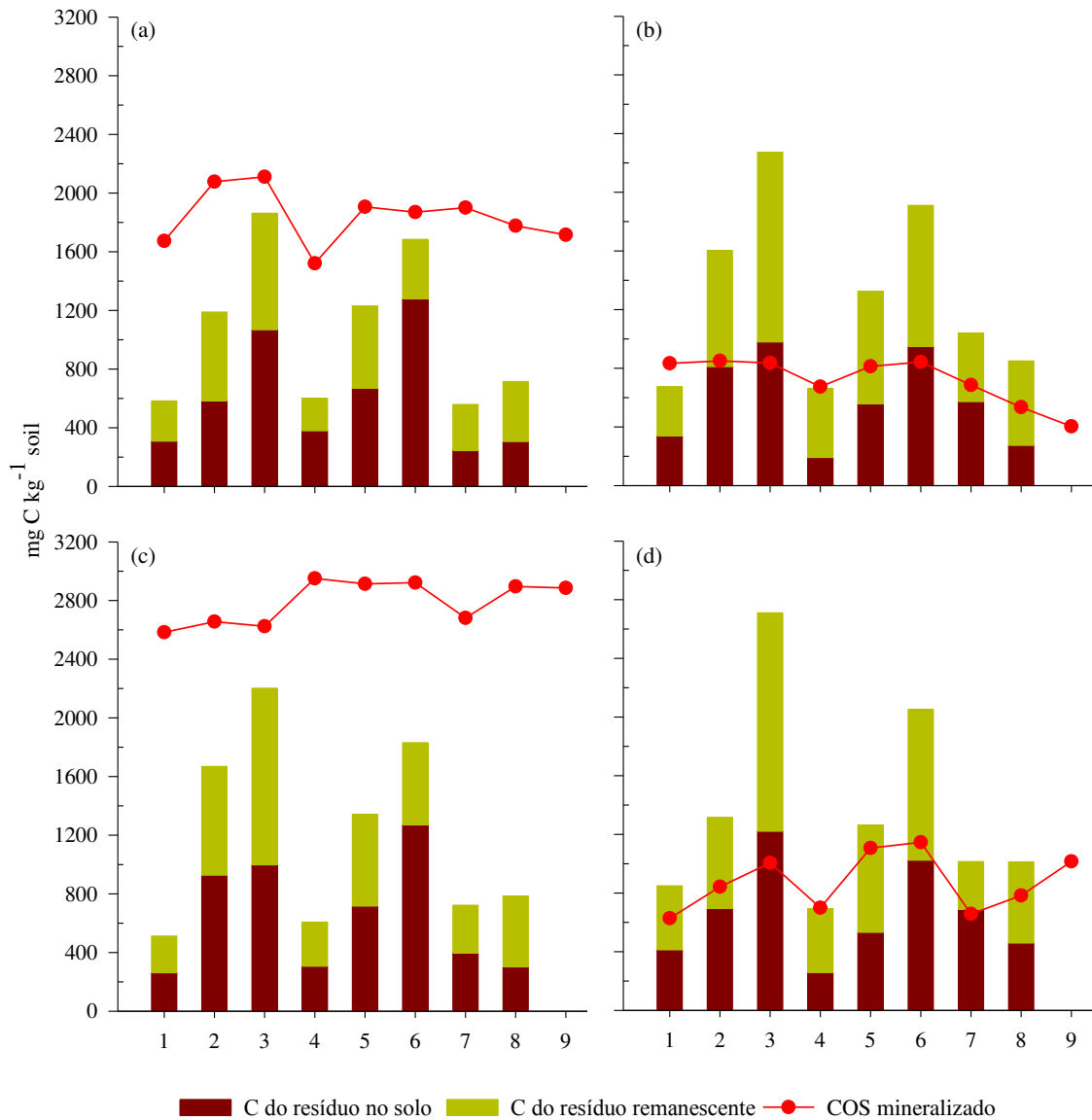


Figura 1. Quantidades de C dos resíduos no solo (C recuperado), remanescente nos resíduos (C remanescente) e C mineralizado da MOS (COS mineralizado) em nove tratamentos (1 - soja 4 Mg ha⁻¹; 2 - soja 8 Mg ha⁻¹; 3 - soja 12 Mg ha⁻¹; 4- sorgo 4 Mg ha⁻¹; 5 - sorgo 8 Mg ha⁻¹; 6 - sorgo 12 Mg ha⁻¹; 7 – raiz de soja 4 Mg ha⁻¹; 8 – raiz de sorgo 4 Mg ha⁻¹; 9 – controle/sem resíduo), mensurados após 360 dias de incubação em quatro tipos de solo (a - arenoso alto-C; b - arenoso baixo-C; c – argiloso alto-C; d – argiloso baixo-C).

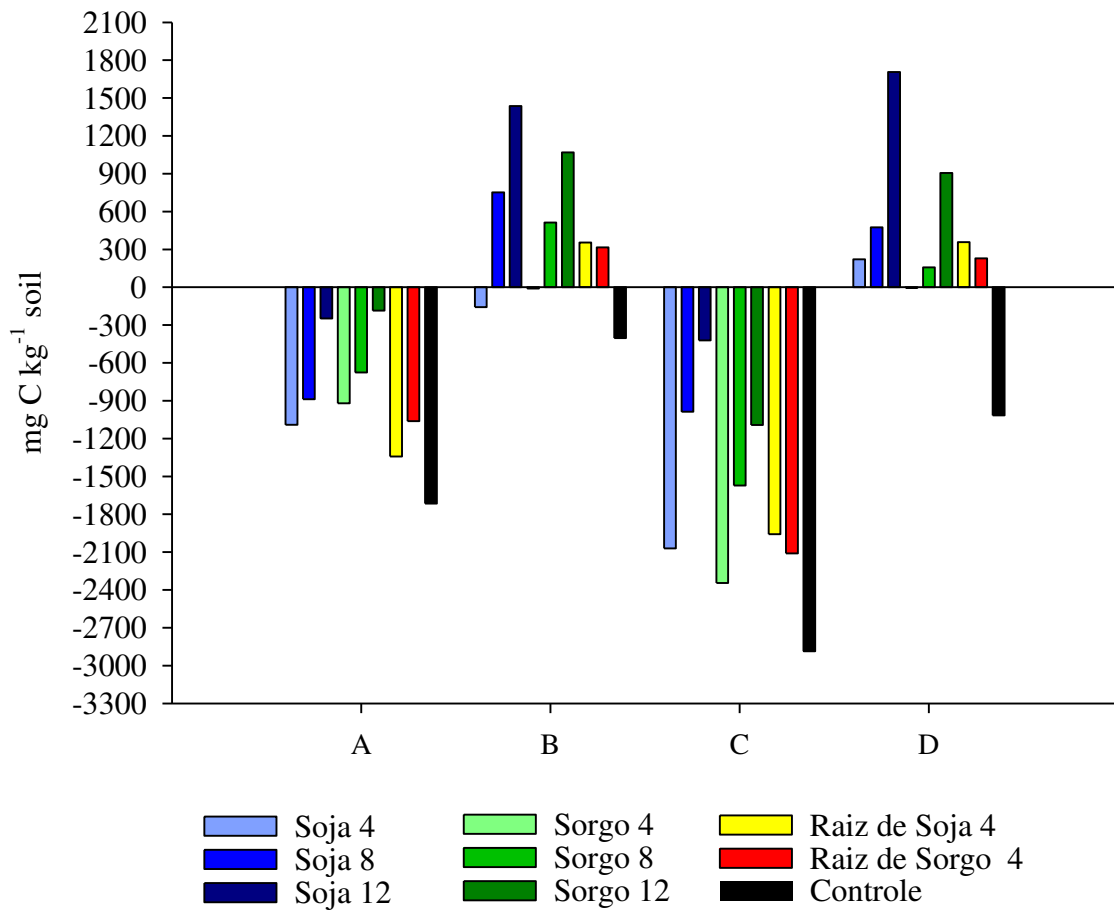


Figura 2. Balanço líquido de C no solo após 360 dias de incubação em quatro solos (A - arenoso alto-C; B - arenoso baixo-C; C - argiloso alto-C; D - argiloso baixo-C). Balanço de C (mg C kg⁻¹ de solo) = C recuperado no solo + C nos resíduos remanescentes - C-CO₂ mineralizado do solo.