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REGULAR ARTICLE

Rain regime and soil type affect the C and N dynamics in soil columns that are covered with mixed-species mulches

Akhtar Iqbal · Sohaib Aslam · Gonzague Alavoine · Pierre Benoit · Patricia Garnier · Sylvie Recous

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Abstract
Aims The role of precipitation on the decomposition of residue mulches is of primary importance for the adequate management of nutrients in no-tilled agrosystems. The objective of this work was to understand the interactions between water dynamics and crop residue quality and their effect on carbon (C) and nitrogen (N) mineralization.
Methods The decomposition of two residue mixtures (wheat + alfalfa and maize + lablab) left at the surface of repacked soil columns, was studied under controlled conditions, at 20 °C over 84 days. Simulated rain pulses were either light and frequent or heavy and infrequent. A loamy soil (Luvisol) and a sandy soil (Ferralsol) were used.
Results The maize/lablab mulch remained wetter between rain pulses which induced greater decomposition than the wheat/alfalfa mulch. Frequent/light rain pulses maintained the mulches wetter between pulses than infrequent/heavy rain pulses, and therefore these mulches decomposed faster. The loamy soil favored the moistening of the mulch layer in contact with the soil which enhanced its decomposition, compared to the sandy soil.
Conclusions The water dynamics (water content of the mulches and soil, evaporation, and drainage) was highly modified by residue quality, rain regime and soil type, which in turn significantly affected the mineralization of C and N.

Keywords Conservation agriculture · Crop residue · Decomposition · Drought · Mulching · Rain pulses

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A. Iqbal · G. Alavoine · S. Recous
INRA, UMR614 Fractionnement des AgroRessources et Environnement, F-51100 Reims, France

G. Alavoine · S. Recous (✉)
UMR614 Fractionnement des AgroRessources et Environnement, Université Reims-Champagne Ardenne, 51100 Reims, France
e-mail: sylvie.recous@reims.inra.fr

S. Aslam · P. Benoit · P. Garnier
INRA, UMR1091 Environnement et Grandes Cultures, F-78500 Thiverval-Grignon, France

P. Benoit · P. Garnier
UMR1091 Environnement et Grandes Cultures, AgroParisTech, F-78500 Thiverval-Grignon, France

Present Address:
A. Iqbal
COMSATS Institute of Information Technology, Abbottabad 22060, Pakistan

Present Address:
S. Aslam
Forman Christian College, Lahore 54600, Pakistan

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Introduction

The widespread implementation of conservation agriculture has occurred in many parts of the world and is considered a promising alternative to intensive agriculture because it prevents soil degradation, controls erosion and improves soil fertility and crop yield (FAO 2008; Derpsch et al. 2010; Scopel et al. 2013). Direct seeding with permanent vegetation cover is emerging as a means to sustain agricultural yields, and this type of agriculture, which is based on the presence of living and dead plant biomass and permanent soil cover, may increase the soil carbon content and provide nitrogen for subsequent crop growth, decreasing the dependence on inorganic N fertilization (Erenstein 2003; Maltas et al. 2009; Neto et al. 2010; Mupangwa et al. 2013). Farmers from developed countries also must cope with several production constraints to mitigate the negative impacts of agriculture, and they are confronted with varying degrees of economic and climate risks. These farmers are now focusing on developing alternative cropping systems.

Even if conservation agriculture has been shown to be efficient in terms of soil fertility management, uncertainty regarding the efficacy of conservation agriculture with respect to crop yields has been attributed to the complexity of interacting biophysical factors and process pathways and drivers (Scopel et al. 2013; Brouder and Gomez-Macpherson 2014). Thus, the role of individual management options and their interactions with other management options and climatic conditions must be better understood (Brouder and Gomez-Macpherson 2014; Lee et al. 2014). In this general context, the process and drivers that result in the decomposition of residue mulches left at the soil surface by no-till practices have received increasing attention in recent years, particularly the role of precipitation in arid or semi-arid ecosystems, under tropical conditions with alternating wet and dry seasons, and with a possible redistribution of rainfall throughout the growing season under global climatic changes. Such studies are primarily intended to determine the effect of drought stress on residue decomposition (e.g., Vanlauwe et al. 1995; Seneviratne et al. 1998; Schimel et al. 1999). The results indicate a high control of mulch decomposition by the precipitation pattern (in addition to temperature), which affects the soil water content and microbial decomposition activities (Kumar and Goh 2000). The total amount of water received and frequency and intensity of rainfall control mulch degradation and nutrient release, and the rain regimes modify the soil water content and interact with crop residue placement (Vanlauwe et al. 1995; Abera et al. 2014; Lee et al. 2014) and residue quality (Tian et al. 2007; Sanaullah et al. 2012; Abera et al. 2014; Marinari et al. 2014); the precipitation regime has a stronger impact when the residues are left at the soil surface (Coppens et al. 2007; Lee et al. 2014).

Integration of legumes and cereal crops in the cropping systems or their succession in rotations are commonly practiced by smallholder farmers throughout the tropics (Giller et al. 2011) and in conservation agriculture, with legume residues providing significant amounts of N to subsequent crops. Therefore, the mixing of crop residues with different characteristics in different amounts is a common situation in such systems. The individual components of these mixtures might provide extra nutrients that may enhance decomposition of the mixture, e.g., N for decomposers (Berklund et al. 2013), and they might also improve the microenvironmental conditions for decomposition (Makkonen et al. 2013). Thus so far, limited research has been performed on the residue quality, particularly on the physical characteristics of the residue mulches, including the water retention capacity of the plant tissues (Garnier and Laurent 1994; Iqbal et al. 2013) and their behavior under rain (Coppens et al. 2007). This litter trait is expected to influence the moisture of the mulch after rain and its evolution during evaporation periods, which was shown by Coppens et al. (2006; 2007) with rye and rape residues under simulated rain pulses.

Our aim was to understand the interactions between mulch quality, rain regime and soil type on the water, carbon (C) and nitrogen (N) dynamics in soils during mixed-species mulch decomposition. The main hypothesis was that the characteristics of residue mulches, which differ between crops and change during decomposition, affect their water retention properties and alter their contact with the soil. In turn, these factors affect soil processes, such as water, C and N fluxes. Therefore, we used an experimental approach with repacked soil columns and two mixed-species mulches with plants that are representative of conservation agriculture in temperate and tropical conditions, and we imposed two contrasting regimes of rain that varied in intensity and frequency. Although we are aware that this design is a simplified and standardized representation of mulch decomposition at the soil surface and of rain regimes in the context of conservation agriculture, this approach allowed us to assess the relative importance of these individual drivers and their interactions while minimizing the confounding effects than would occur under field conditions.

Materials and methods

Soils and crop residues

The first soil was a silty loam Luvisol (FAO classification) that was sampled from La Cage, the experimental site of the Institut National de la Recherche Agronomique (INRA) near Versailles, France (48° 50′ 23.41″ N, 1° 56′ 50.36″ E). The field had not been tilled for more than 10 years, and the main crops in the rotation (maize-wheat-pea-wheat) were direct-seeded with a permanent plant cover. The second soil was a sandy Ferralsol (FAO classification) that was sampled near Lake Alaotra, Madagascar (17°41.33′ S, 48° 27.58′ E) on plots that had not been tilled for 4 years. The abbreviations LAC and ALA will be used for the "La Cage" and "Alaotra" soils, respectively. The main characteristics of the two soils are presented in Table 1. During sampling, two layers were distinguished that correspond to the 0 to 5 cm soil depth and 5 to 25 cm soil depth. The sampled soils were sieved (<4 mm) and stored separately in bags at 4 °C until further use. The water retention curve of each soil layer was determined using pressure plates (Klute 1986).

We selected two crop associations to prepare experimental residue mulches: the first situation was representative of an agricultural situation in Madagascar where maize (*Zea mays*) is grown in association with lablab (*Dolichos Lablab*); the second situation was representative of organic farming in France where wheat (*Triticum aestivum*) is grown in association with alfalfa (*Medicago sativa*). The aboveground portions of the plants were collected from farmers' fields in France or Madagascar, and the proportion of the two plant species in the association was determined. Only the leaves and stems were retained, and their proportion for each crop was determined. The residues were chopped into 3-to-4-cm-length pieces (Fig. 2), and the two mulch residues were composed according to the actual proportion of each plant organ, which was actually determined on plants harvested in the field for this experiment. The mulch of maize plus lablab (ML) was composed of 63 % maize residue (60 % stems + 40 % leaves) and 37 % lablab residue (87 % stems + 13 % leaves). The mulch of wheat plus alfalfa (WA) was composed of 83 % wheat residue (64 % stems + 36 % leaves) and 17 % alfalfa residue (62 % stems + 38 % leaves). The elemental and biochemical composition of the initial crop residues was determined and is given in Table 3 and Fig. 3, respectively. Briefly, ML mulch consisted of 255 g soluble fraction kg^{-1} DM, 262 g hemicellulose kg^{-1} DM, 392 g cellulose kg^{-1} DM and 92 g lignin-like fraction kg^{-1} DM; WA mulch was composed of 220 g soluble kg^{-1} DM, 295 g hemicellulose kg^{-1} DM, 395 g cellulose kg^{-1} DM and 90 g lignin-like fraction kg^{-1} DM. The initial C:N ratio of the individual plants (stem + leaves) was quite different, with 32.7 for lablab, 57.4 for maize, 13.8 for alfalfa and 278.0 for wheat. But the C:N ratio of the mixtures

Table 1 Selected properties of the soils that were used

		LAC		ALA	
		0–5 cm	5–25 cm	0–5 cm	5–25 cm
Clay	g kg^{-1}	148	163	153	157
Silt	g kg^{-1}	655	648	270	251
Sand	g kg^{-1}	197	189	577	592
Organic Carbon	g kg^{-1}	20.7	9.9	32.5	27.4
Total Nitrogen	g kg^{-1}	1.8	0.9	2.9	2.3
C:N		11.4	10.7	11.2	11.7
Total Carbonate	g kg^{-1}	<1	<1	<1	<1
pH (H_2O)		6.4	7.1	5.7	5.8
CEC	cmolc kg^{-1}	10.6	9.6	18.0	16.9

was quite similar at 60.4 and 62.0 for the ML and WA mixtures, respectively.

Experimental design and protocol

The experimental conditions are summarized in Table 2. The experimental setup was similar to that described by Aslam et al. (2014). PVC cylinders (15.4-cm inner diameter, 35 cm height) with a small hole at their base for drainage were used. Each cylinder was filled with 25 cm of moist sieved soil, and it was filled from −35 to −15 cm with soil collected from the 5 to 25 cm depth and then from −15 to −10 cm with the soil from the 0 to 5 cm depth. Before building the columns, each soil layer was adjusted to the water content at pF 2.5 that was derived from the water retention curves (data not shown). To fill the cylinder from −35 to −15 cm, 5.6 kg of LAC and 3.7 kg of ALA (equivalent dry weight) moist soils were divided into four subsamples, which were successively compacted homogeneously at 1.5 g cm^{-3} (LAC) and 1 g cm^{-3} (ALA) to a predefined volume of the cylinder (Table 2). The surface of each compacted soil sub-layer was loosened before the addition of the next soil subsample to maintain continuity in the arrangement of the soil particles. In the top layer (0 to 5 cm) of LAC, 1.2 kg (equivalent dry weight) of soil was compacted at 1.3 g cm^{-3}. For ALA, 0.93 kg (equivalent dry weight) of soil was compacted at 1 g cm^{-3} (Table 2). A mixture of maize + lablab (ML) or wheat + alfalfa (WA) was placed as a mulch on the soil surface, with 14.1 g of residue (equivalent dry mass) per soil column, corresponding to a crop residue biomass addition of 7.6 t per ha.

Two rainfall treatments were assigned using simulated rainfall pulses to create fluctuating soil water content conditions. These treatments were inspired from weather patterns in temperate and tropical climates. "Light and Frequent" rain (LF_R) was applied twice a week at 6 mm hr^{-1} for 20 min (2 mm of rainfall equivalent applied at each rain pulse); thus, these columns received a total of 16 mm of rain equivalent per month. "Heavy and Infrequent" rain (HI_R) was applied every 2 weeks at 20 mm hr^{-1} for 24 min (8 mm of rain equivalent applied at each rain pulse); thus, these columns also received a total of 16 mm of water per month (Table 2). This choice was a compromise to represent contrasting rainfall intensities and frequencies and to fulfill the experimental constraint of applying the same amount of water per month and at the end of the experiment. The rain simulator consisted of capillary tubes (inner diameter of 0.5 mm) that were equally distributed over the surface (186 cm^2) of the column. The rain intensity was controlled by adjusting the water pressure of the pumps that were attached to the simulators. Deionized water was used, and the soil columns were weighed before and after each rain pulse to calculate the exact amounts of water that were (i) lost by evaporation during incubation between two rains and (ii) applied with each rain. There were a total of 24 rain pulses for the LF_R treatment and 6 rain pulses for the HI_R

Table 2 Experimental conditions for the treatments that were used in the soil column experimental approach. "a" represents the 0 to 5 cm soil layer, and "b" represents the 5 to 25 cm soil layer

Description of experimental conditions	ML-HIR-ALA	ML-HIR-LAC	ML-LFR-LAC	WA-LFR-LAC
Humidity of soil (*g H_2O g^{-1}*)	0.4^a /0.38^b	0.25^a /0.22^b	0.25^a /0.22^b	0.25^a /0.22^b
Bulk density of soil (*g cm^{-3}*)	1.0^a /1.0^b	1.3^a /1.5^b	1.3^a /1.5^b	1.3^a /1.5^b
Equivalent soil dry weight (*kg per column*)	0.9^a /3.7^b	1.2^a /5.6^b	1.2^a /5.6^b	1.2^a /5.6^b
Temperature *(°C)*	20	20	20	20
Duration of incubation (*day)s*	84	84	84	84
Total number of rain pulses	6	6	24	24
Intensity of rain applied (*mm hr^{-1}*)	20	20	6	6
Water applied at each rain (*mm)*	8	8	2	2
Total amount of water applied (*mm)*	48	48	48	48
Interval between rain pulses (*days)*	14	14	3 and 4	3 and 4
Initial mulch dry matter (*g per column)*	14.1	14.1	14.1	14.1
Mulch thickness *(cm)*	3 to 4	3 to 4	3 to 4	3 to 4

treatment, with corresponding numbers of wetting and drying periods (Table 2). No leachate was recovered at the bottom of the columns because the amount of rain of each pulse almost replaced the amount of water that was lost by evaporation, without the addition of excess water.

The three factors, i.e., the type of mulch, type of soil and rainfall pattern, were combined into four different treatments (Fig. 1). The effect of the crop residue quality was investigated by comparing the maize + lablab (ML) and wheat + alfalfa (WA) mulches covering LAC soil with the LF_R pattern. The effect of the soil type was investigated with the maize + lablab mulch (ML) under the HI_R pattern with the two soils (ALA and LAC). The effect of the rainfall pattern was investigated with the maize + lablab (ML) mulch covering the LAC soil. The four treatments were termed ML-HIR-ALA, ML-HIR-LAC, ML-LFR-LAC and WA-LFR-LAC. The mulch residues (ML and WA) were pre-moistened with a sprayer and placed homogenously on the soil surface of each column before beginning the experiment. In total, 53 soil columns with mulches on the soil surface were prepared and divided into 3 sets that served different purposes. A total of 36 columns (4 treatments × 3 destructive sampling dates × 3 replicates) were used to measure the dynamics of mulch mass, soil C, N and microbial pools during decomposition, and 8 columns (4 treatments × 2 replicates) were used for the continuous measurements of CO_2 and N_2O fluxes and non-destructive sampling of the soil solution. A total of 9 columns were destroyed at day 0 prior to any rain application to analyze the initial conditions for the soils and mulches. The 44 other columns were incubated in the dark at 20 °C for 84 days and received rain pulses according to the rainfall treatments. After mulch placement, a pesticide solution containing s-metolachlor and glyphosate molecules was applied homogeneously onto each column at day 0; this was performed according to a companion study on pesticide transport in mulched soils (Aslam et al. 2014). This work showed that pesticide application had no effect on the total microbial respiration or carbon mineralization.

Destructive samplings were performed at days 0, 14, 41 and 84 after the start of the experiment. The mulch residues were gently collected from the soil surface and weighed, dried at 40 °C until a constant weight and maintained for analysis. The soil of each column was sliced into three layers: 0 to 5 cm, 5 to 15 cm and 15 to 25 cm. Soil sub-samples from each layer were analyzed immediately (see below section) or frozen (−20 °C) until analysis.

Semi-continuous and continuous measurements

The emissions of CO_2 and N_2O were calculated from the accumulation rate of CO_2 and N_2O in the headspace of the columns (Fig. 2). During a 22-min period, the columns were sealed with a cover that was connected to an infrared gas analyzer (INNOVA-1412-Photoacoustic field gas monitor, Denmark). A fan on the inside of the closed chamber mixed the air to a homogeneous state. The slope of a linear regression of the increase in the CO_2 or N_2O concentration over time was used to calculate the fluxes. The measurements were collected 5 times per week except for the days during which the columns received rain, for which two measurements

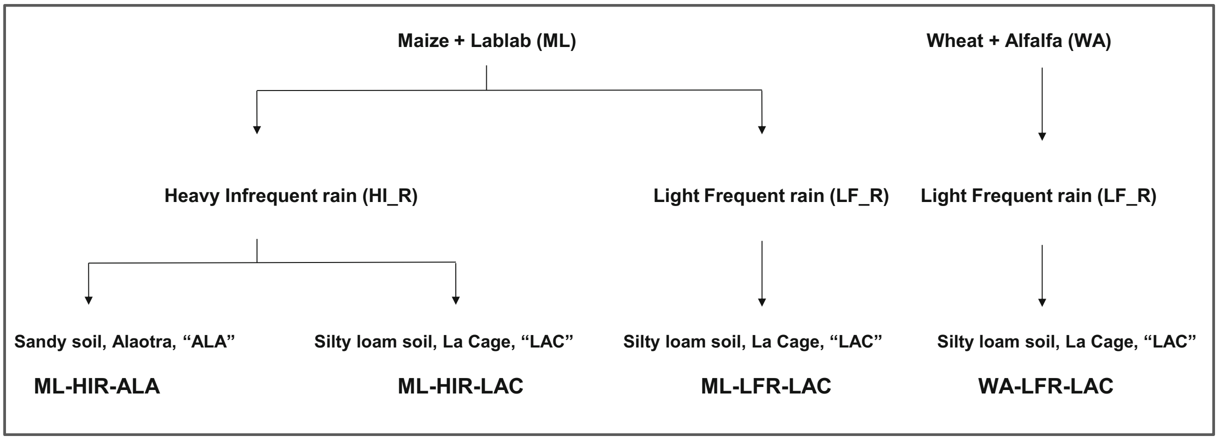

Fig. 1 Schematic diagram representing the experimental design with the different treatments

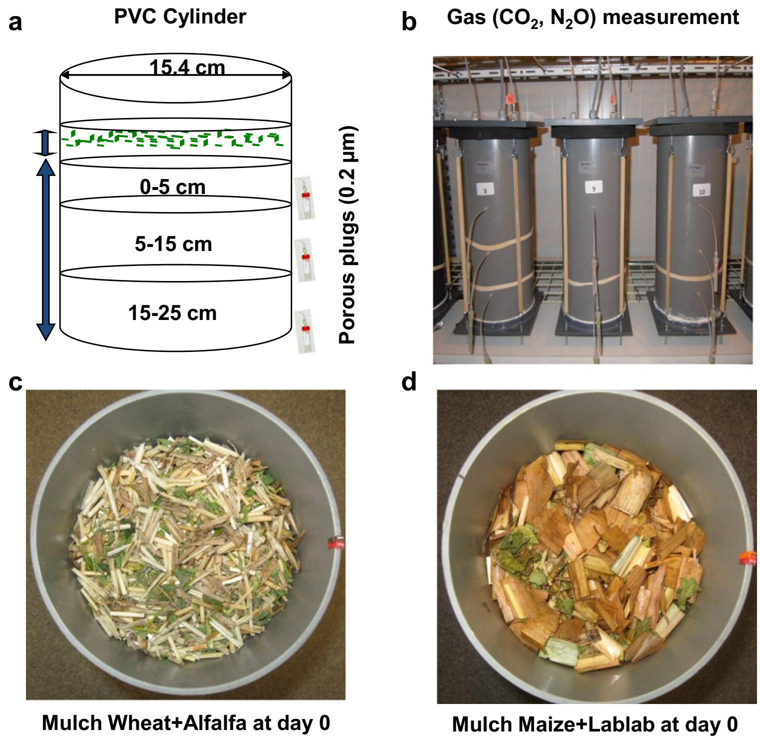

Fig. 2 Pictures of the columns with gas measurement equipment, design of the column constitution and initial mulches at the soil surface in the columns of wheat + alfalfa (WA) and maize + lablab (ML)

were collected before and after the rain pulse. In the latter case, the measurements were collected within 2–3 h after the rain pulse.

The water potential was measured at the −3 and −15 cm depths with tensiometers, and the volumetric water content was measured through soil water content sensors (EC H2O Dielectric Aquameter sensors, Decagon Devices, Inc.). The soil solution (10 ml) was sampled 12 h after rain every 2 weeks at depths of −2, −10 and −18 cm (Rhizon MOM 10 cm, Rhizosphere Research Products) (Fig. 2). The samples were stored at −20 °C until analysis for ammonium, nitrate N and soluble C. The soluble C that was determined in the soil solutions sampled after each rainfall was considered 'mobile' dissolved organic C (DOCm).

Chemical analysis

At each sampling date (0, 14, 41 and 84 days), the gravimetric water content of the soils was determined after drying at 105 °C for 24 h. The microbial biomass C was determined on moist sub-samples of soils (50 g dry-weight basis) via the fumigation extraction method that was proposed by Vance et al. (1987). The soluble carbon from the soil samples that were fumigated with chloroform or unfumigated was extracted by 0.025 M K_2SO_4 (soil:extractant ratio = 1:4), agitated at 40 rpm for 30 min, and then centrifuged at 5000 rpm for 10 min at 20 °C. The supernatant was filtered, and the filtrate was maintained at −20 °C until further analysis. The extracted soluble C was analyzed with an analyzer (1010, O.I. Analytical), and the amount of microbial C was calculated as the difference between the microbial C of the fumigated and unfumigated samples of the same soil, with a coefficient K_{EC} = 0.38 (Vance et al. 1987) to convert the amount of extracted C into microbial C. The soil mineral N was extracted with 1 M KCL, and the ammonium and nitrate concentrations of the extracts were measured by colorimetry.

The nitrate in the soil solutions was measured by colorimetry (TRAACS 2000, Bran and Luebbe, Norderstedt, Germany), and the soluble C was analyzed using an auto analyzer (1010, OI Analytical, College station, Texas, USA). The method used oxidation at 100 °C in persulfate medium, measuring the CO_2 that was produced by infrared (Barcelona 1984).

The mulches that were recovered from the surface of the soil columns were ground to 4 mm and used for biochemical fractionation using a proximate analysis (Goering and Van Soest 1970). The total C and N concentration of the mulch residues were also determined using an elemental analyzer (NA 1500, Carlo Erba, Milan, Italy).

Statistical analysis

A three-way analysis of variance (ANOVA) was used for soil mineral N and microbial C in different column layers (variables presented in Fig. 5), with the treatment, time and column layer as the three factors. A two-way analysis of variance (ANOVA) was used for mulch mass loss, mulch total C, mulch total N, C:N, mulch N, mulch C, mulch water content, soil mineral N content, CO_2 and N_2O measurements (variables presented in Tables 3, 4 and Fig. 4), with the treatment and time as the two factors. A two-way repeated-measure ANOVA was used to determine the effect of the treatment and sampling date on the soil pore water concentrations for soluble C and NO_3^-. The analyses were conducted with SigmaPlot.

Results

Mulch dynamics

The dry mass of mulches decreased for all of the treatments during decomposition, with significant differences between treatments. The largest loss in mass was at day 84 for the ML-LFR-LAC (−30 % of initial mass) treatment, whereas the lowest loss was observed for ML-HIR-ALA (−11 % of the initial mass) (Table 3). The values for ML-HIR-LAC (−16 % initial mass) and WA-LFR-LAC (−13 % initial mass) were intermediate.

The N concentration of the ML mulches increased during decomposition and resulted in a decrease in the C:N ratio. For the WA mulch (WA-LFR-LAC), the N concentration decreased until day 41, after which it increased slightly (Table 3). The decrease in the C:N ratio was more pronounced for ML-LFR-LAC, which was also the most decomposed compared with the other treatments. As a result, the amount of mulch N did not follow the pattern of mass and C losses: we observed a slight increase in the total mulch N (g/m^2) for ML mulch compared with the initial mulch N, whereas there was a net loss of N for the WA mulch.

The C loss from the mulch followed the dynamics of mulch mass and ranked the treatments accordingly (Table 3). Overall, the ML mulch lost more C when it was decomposing on the surface of the loamy soil (LAC) with frequent rainfall (ML-LFR-LAC) compared with the other treatments. The WA mulch decomposed slowly compared with ML using the same soil and same pattern of rain. The differences between the two rain patterns (ML-HIR-LAC vs. ML-LFR-LAC), two mulches (ML-LFR-LAC vs. WA-LFR-LAC) and two soils (ML-HIR-ALA vs. ML-HIR-LAC) were statistically significant ($p \leq 0.05$) at day 84. The ML mulch on ALA soil remained drier during decomposition compared with ML on LAC soil. The ML mulch with frequent rain (LFR) was always wetter than the ML mulch with infrequent rain (HIR). In addition, the ML mulch remained wetter than the WA mulch under the same rainfall pattern (Table 3).

The mean initial chemical composition of the ML and WA mulches did not show significant differences despite the different assemblages of mulches (Fig. 3). ML had a slightly higher initial soluble concentration and a lower hemicellulose concentration compared with WA, but their cellulose and lignin concentrations were similar. During the initial decomposition (0 to 14 days), the mass loss was mainly caused by the decrease in the soluble pool for both of the mulches. The hemicelluloses and cellulose fractions decreased with decomposition. The composition of the remaining mulches changed slightly over time, with a decrease in the relative size of the soluble fraction (approximately −5 %) and increase in the relative size of the lignin fraction (+ 5 %); the proportion of hemicelluloses and cellulose fractions was stable.

CO_2 and N_2O emissions

The ML and WA mulches differed in their initial rate of mineralization (Fig. 4a and b), with a much higher rate of CO_2 with WA compared with ML during the first 6 days of incubation. From day 14, the opposite trend was observed, with higher rates of mineralization from the ML treatment compared with WA. Overall, the rates of mineralization decreased from the start of incubation onwards. The effect of rain application translated into peaks in CO_2 emission after each rain, i.e., every 2 weeks in the HIR scenario, and lasted approximately 3 days. In the LFR scenario, the peaks were more frequent, which was expected. The cumulative CO_2 that evolved during the 84 days of incubation, which was calculated from the daily rates of mineralization, amounted to 54±2 g of CO_2-C m^{-2} for ML-HIR-ALA, 126±9 g for ML-HIR-

Table 3 Dry mass, Carbon (C) and Nitrogen (N) concentrations, C, N and water contents of mulches during decomposition at 20 °C

Sampling time (days)	Treatment	Mass g/m^2	Total C *g/kg residue*	Total N *g/kg residue*	C:N ratio	Mulch-N g/m^2	Mulch-C g/m^2	Mulch water content $g\ H_2O/g$ *residue*
0	ML	$758^{a/A}$	$442±17.6^{a/A}$	$7.4±0.5^{a/A}$	$60.4±6.1^{a/A}$	$5.6±0.4^{a/A}$	$335±0.1^{a/A}$	$0.12±0.02^{a/A}$
	WA	$758^{a/A}$	$442±16.0^{a/A}$	$7.4±1.3^{a/A}$	$62.0±13^{a/A}$	$5.6±1.0^{a/A}$	$335±0.0^{a/A}$	$0.13±0.01^{a/A}$
14	ML-HIR-ALA	$743±4^{a/A}$	$444±13.7^{a/A}$	$8.9±1.4^{b/A}$	$51.2±9.7^{b/A}$	$6.6±0.4^{b/A}$	$330±2.0^{ab/A}$	$0.38±0.08^{b/A}$
	ML-HIR-LAC	$718±34^{b/A}$	$445±15.6^{a/A}$	$9.2±0.8^{b/A}$	$48.6±4.2^{b/B}$	$6.6±0.3^{b/A}$	$319±15.3^{b/A}$	$0.38±0.06^{b/A}$
	ML-LFR-LAC	$619±14^{b/B}$	$448±15.6^{a/A}$	$9.6±1.6^{b/A}$	$47.6±8.0^{b/B}$	$6.0±0.1^{b/B}$	$278±6.4^{b/B}$	$1.15±0.32^{b/B}$
	WA-LFR-LAC	$730±2^{a/A}$	$449±15.7^{a/A}$	$5.1±0.7^{b/B}$	$89.5±13^{b/C}$	$3.7±0.0^{b/C}$	$328±0.8^{a/AC}$	$0.88±0.57^{b/B}$
41	ML-HIR-ALA	$703±22^{b/A}$	$449±10.9^{a/A}$	$9.8±1.5^{b/A}$	$46.5±7.3^{c/A}$	$6.9±0.2^{bc/A}$	$315±10.0^{bc/A}$	$0.36±0.06^{b/A}$
	ML-HIR-LAC	$622±44^{c/B}$	$444±15.1^{a/A}$	$10.2±0.9^{c/A}$	$43.7±4.4^{b/A}$	$6.4±0.5^{b/A}$	$276±19.6^{c/B}$	$0.55±0.22^{b/A}$
	ML-LFR-LAC	$563±26^{c/C}$	$447±17.8^{a/A}$	$10.6±0.3^{bc/A}$	$42.3±2.1^{b/A}$	$6.0±0.3^{b/AB}$	$252±11.6^{c/C}$	$1.23±0.25^{b/B}$
	WA-LFR-LAC	$674±29^{b/A}$	$443±26.5^{a/A}$	$5.4±0.5^{bc/B}$	$82.2±12^{bc/B}$	$3.7±0.2^{b/C}$	$298±12.8^{b/D}$	$0.69±0.18^{b/A}$
84	ML-HIR-ALA	$672±12^{c/A}$	$443±15.5^{a/A}$	$9.7±0.9^{b/A}$	$46.1±5.3^{c/A}$	$6.5±0.1^{bd/A}$	$298±5.1^{d/A}$	$0.28±0.05^{b/A}$
	ML-HIR-LAC	$631±36^{c/A}$	$444±19.2^{a/A}$	$9.1±0.5^{bd/A}$	$48.9±3.4^{b/A}$	$5.7±0.2^{ac/B}$	$280±16.1^{c/B}$	$0.52±0.03^{b/B}$
	ML-LFR-LAC	$540±26^{c/C}$	$433±19.9^{a/A}$	$9.1±0.9^{bd/A}$	$47.7±5.0^{b/A}$	$4.9±0.3^{c/C}$	$234±11.4^{d/C}$	$0.82±0.14^{b/C}$
	WA-LFR-LAC	$661±16^{b/A}$	$430±20.7^{a/A}$	$6.1±0.4^{cd/B}$	$71.0±6.7^{ac/B}$	$4.0±0.1^{c/D}$	$285±6.9^{b/AB}$	$0.32±0.09^{ac/A}$

The mulches were sampled at days 0, 14, 41 and 84. ML is the maize + lablab mixture, WA is the wheat + alfalfa mixture. HIR and LFR represent the Heavy Infrequent and Light Frequent rains treatments, respectively. LAC and ALA represent the loamy soil and sandy soil treatments, respectively. Lowercase letters (a, b, c and d) represent the differences between the incubation times for a given treatment, while the uppercase letters (A, B, C and D) represent the differences between the treatments at a given incubation time with >95 % confidence. The values are the mean of three replicates (n=3)±SD

Table 4 Mineral N content (g m^{-2}) in the 0 to 25 cm soil depth column at 0, 14, 41 and 84 days of incubation and net mineralization between day 0 and day 84. Lowercase letters (a, b, c and d) represent the differences between the incubation times for a given treatment, while the uppercase letters (A, B, C and D) represent the differences between the treatments at a given incubation time with >95 % confidence

Treatment	d0	d14	d41	d84	Net change at d84
g m^{-2}					
ML-HIR-ALA	10.3±0.1 $^{a/A}$	10.8±0.1 $^{b/A}$	12.2±0.1 $^{c/A}$	14.4±0.2 $^{d/A}$	4.1±0.2 ^{A}
ML-HIR-LAC	4.1±0.1 $^{a/B}$	3.9±0.1 $^{a/B}$	4.5±0.7 $^{a/B}$	9.6±1.9 $^{b/B}$	5.5±1.7 ^{B}
ML-LFR-LAC	4.1±0.1 $^{a/B}$	2.7±1.0 $^{ab/B}$	5.6±0.6 $^{ac/C}$	8.9±1.2 $^{d/BC}$	4.8±1.0 ^{AB}
WA-LFR-LAC	4.1±0.1 $^{a/B}$	3.6±2.1 $^{a/B}$	2.8±0.4 $^{a/D}$	7.3±2.1 $^{b/CD}$	3.2±1.9 ^{AC}

The data are mean values of 3 replicates (n=3)±SD

LAC, 131±5 g for ML-LFR-LAC and 124±7 g for WA-LFR-LAC, demonstrating a strong effect for the type of soil and little effect for the type of mulch and the rain pattern (no statistical difference between the three treatments with same soil LAC) despite differences in the kinetics of the evolved CO_2.

The highest N_2O fluxes were observed immediately after the first rain with the LAC soil, and no flux was measured afterwards (Fig. 4c and d). For this soil, the peaks lasted 1 to 2 days and reached 95 and 112 mg N-N_2O m^{-2} day^{-1} with ML mulch and 53 mg N-N_2O m^{-2} day^{-1} for WA mulch. With the ALA soil, the

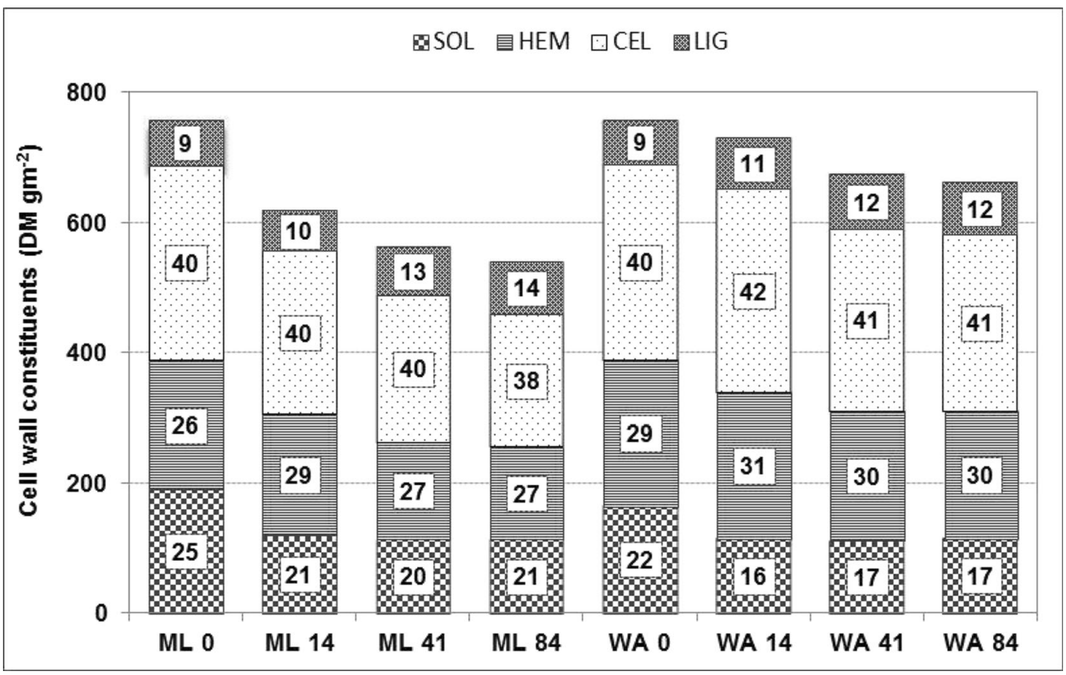

Fig. 3 Changes in mass of the different biochemical fractions of mulches of maize + lablab (ML) and wheat + alfalfa (WA) residues during decomposition at day 0, day 14, day 41 and day 84 of decomposition in the loamy soil from La Cage (treatments ML-LFR-LAC and WA-LFR-LAC). The fractions are ranked as Soluble, Hemicellulose, Cellulose and Lignin-like fractions from base to top of each bar. Labels within the bars indicate the relative proportion (% of total DM) of each fraction at each date. Data are mean values of 3 replicates (n=3)

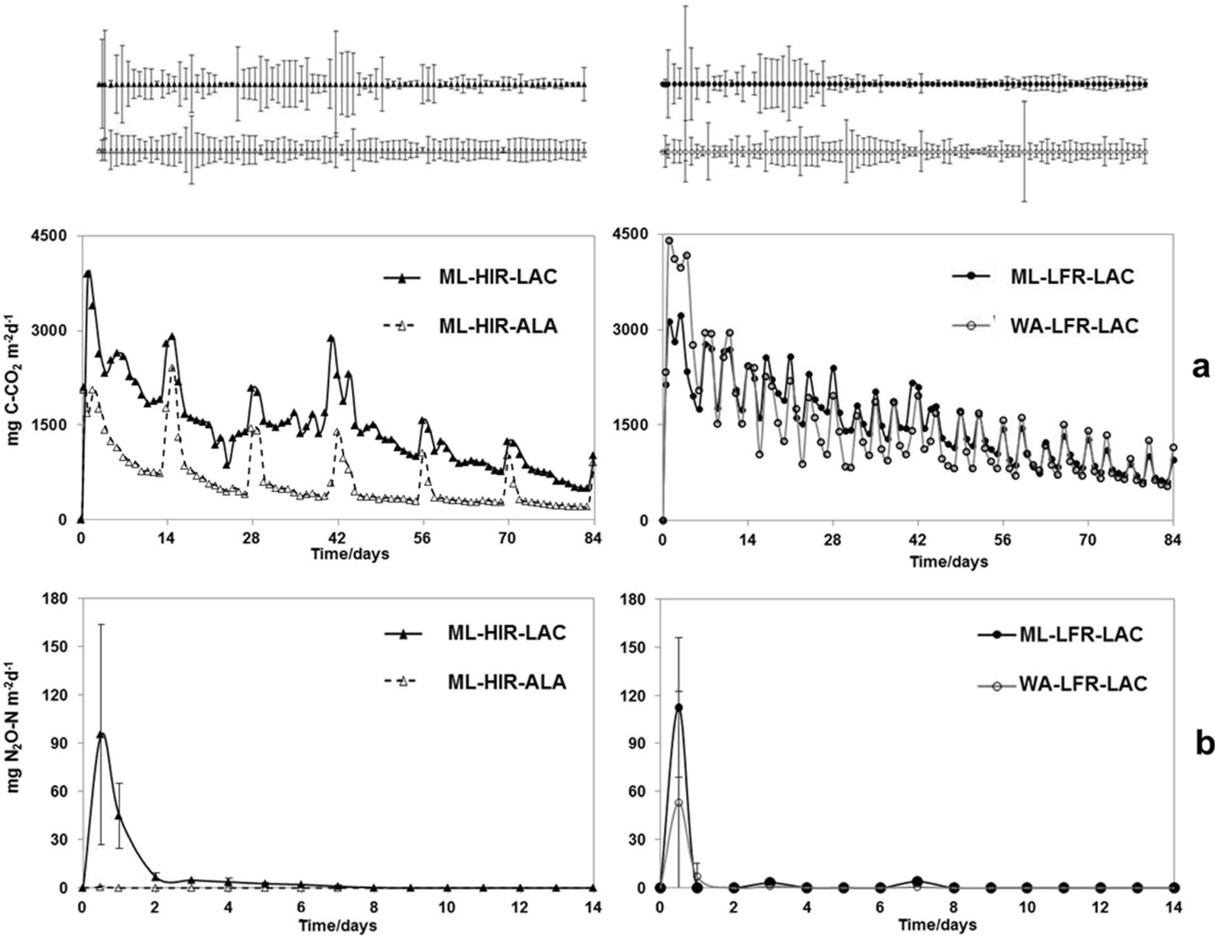

Fig. 4 Fluxes of CO_2 (**a**) and N_2O-N (**b**) that evolved from soil columns with the ML and WA mulch residues throughout 84 days of decomposition under different treatments. The data are mean values of 2 replicates. Standard error of the means are given above the figures, as an indication of the variability over time

emission was almost nil, with a maximum of 0.4 mg N-N_2O m^{-2} day^{-1} after the first rain.

Soil water dynamics

The rainfall pattern and soil type affected the water dynamics in the soils. For light the LF_R scenario, the evaporation cycle lasted only 3 days, whereas for HI_R scenario, it lasted 14 days. These differences in the rainfall regimes also created differences in the water flow. For the LAC soil with the LF_R regime, the matric potential ranged from 0 to −25 hPa and 0 to −10 hPa at depths of 3 cm and 15 cm, respectively (Aslam et al. 2014). With the HI_R regime, the soil matric potentials were always lower as a result of longer periods of evaporation and ranged from 0 to −700 hPa and 0 to −450 at depths 3 cm and 15 cm, respectively. For both of the treatments, the matric potentials were lower in the surface layer than in bottom layers because of evaporation. For the ALA soil with the HI_R regime, the matric potential ranged from −21 to −50 hPa and from −12 to −35 hPa at depths of 3 cm and 15 cm, respectively (Supplementary material Fig. 2).

Dynamics of inorganic N and microbial biomass C

The two soils varied strongly in their initial inorganic N contents (the amount of NH_4^+-N was negligible in every case), with 10.3 g inorganic N m^{-2} for the ALA soil and 4.1 g inorganic N m^{-2} for the LAC soil throughout the 0 to 25 cm depth (Table 4). In the ALA soil, the total amount of inorganic N increased from day 0 to day 84. For the three treatments with the LAC soil, a net decrease was first observed (until day 14 or day 41, depending on the treatment), and then a net increase was observed between day 41 and day 84. The net mean N mineralization, calculated as the difference in inorganic N in the columns between day 0 and day 84, was + 4.1 g N m^{-2} for ML-HIR-ALA and +5.5, +4.8 and + 3.2 g N m^{-2} for ML-HIR-LAC, ML-LFR-LAC and WA-LFR-LAC, respectively (Table 4). The soil type (ML-HIR-ALA vs. ML-HIR-LAC) and mulch quality (ML-LFR-LAC vs. WA-LFR-LAC) induced significant differences in the net N mineralization at day 84 ($p \leq 0.05$). The total N recovered in the soil inorganic pool plus mulch over time (supplementary material Table 2), showed that the largest difference resulted from the initial quality of mulch (WA vs. ML), while the type of soil or the rain regime had little effect.

The distribution of inorganic N in the soil columns differed over time and between treatments, particularly between the two soils. In the ALA soil (ML-HIR-ALA), a strong depletion in nitrate occurred in the 0 to 5 cm layer between day 0 and day 14, which translated into a large increase in the 5 to 15 and 15 to 25 cm layers because of the leaching of nitrate (Fig. 5a). Then, a net increase occurred on day 41 and day 84. In the corresponding treatment with the LAC soil (ML-HIR-LAC), there was little change in the nitrate content and distribution up to day 41, after which a strong increase was observed at day 84 in every layer, but mostly in the 0 to 5 cm layer (Fig. 5b).

Initially the soil microbial C was 500 (0 to 5 cm) and 304 mg C kg^{-1} dry soil (5 to 25 cm) for the ALA soil (Fig. 5c) and 455 (0 to 5 cm) and 168 mg C kg^{-1} dry soil (5 to 25 cm) for the LAC soil (Fig. 5d), indicating a strong gradient of microbial C with the soil depth. As the mulch decomposition proceeded, we observed a decrease in the microbial biomass C for every treatment in the 0 to 5 cm layer. In the 5 to 15 and 15 to 25 cm layers, there was no significant change in microbial C during incubation (Fig. 5c and d).

Soluble C and nitrate in the soil solution

The soluble C (DOC) was measured in the soil solution that was extracted from the soil columns every 2 weeks after re-wetting events (Supplementary material Table 3)

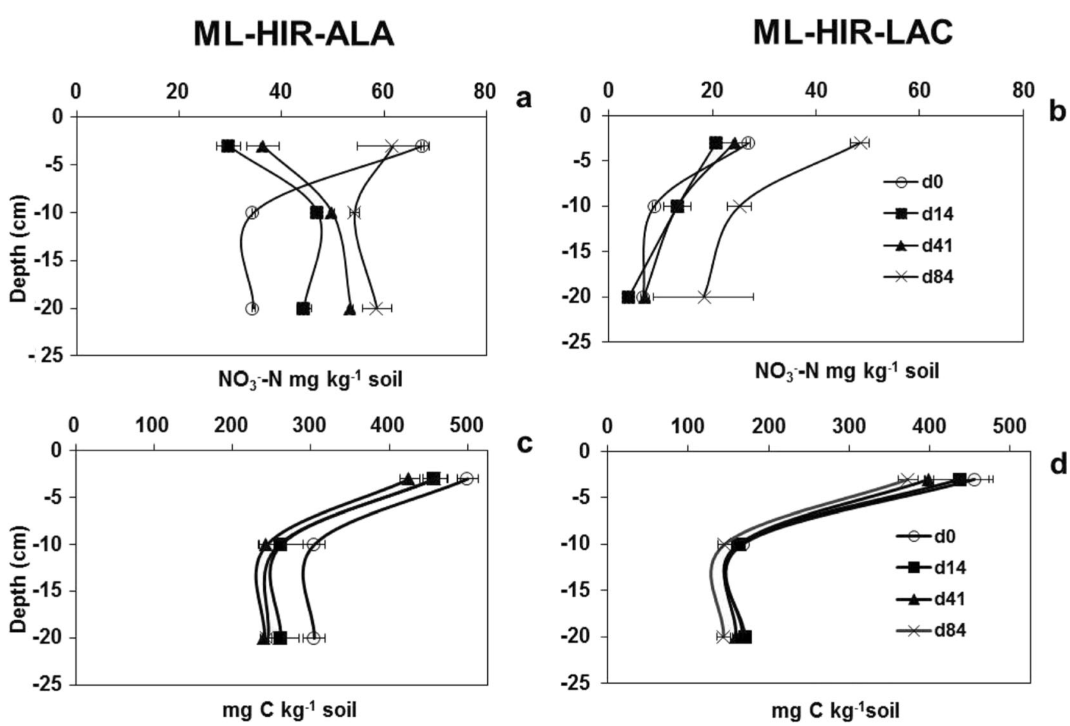

Fig. 5 Comparison of the distribution of mineral N (**a**, **b**) and microbial biomass C (**c**, **d**) in the soil for the three soil layers of the soil columns (0 to 25 cm depth) at 0, 14, 41 and 84 days of incubation in the LAC) and AL under the HIR regime with ML mulch. The data are the mean values of 3 replicates (n=3)

at −2, −10 and −18 cm depths. Initially, the ALA soil contained 7 mg C L^{-1} solution in the 0 to 5 cm layer and 4 mg C L^{-1} in the 5 to 25 cm layer. The LAC soil contained 24 and 7 mg C L^{-1} in the 0 to 5 cm and 5 to 25 cm soil layers, respectively. The differences between the two soils were maintained during the incubation time for every layer. The application of the first rain on the mulches resulted in an increase in the soluble C concentration of the soil solution (measured twelve hr after the rain) in the 0 to 5 cm layer, with 26 mg (ML-HIR-ALA), 41 mg (ML-HIR-LAC), 31 mg (ML-LFR-LAC) and 23 mg (WA-LFR-LAC) of C L^{-1}.

The nitrate concentration in the soil solution increased rapidly for all the treatments, especially in the 0 to 5 cm layer. The increase was larger for the LAC soil compared with the ALA soil in the 0 to 5 cm layer, equivalent in the 5 to 15 cm layer, and lower in the 15 to 25 cm layer. Regarding the two mulches, there was no significant differences in the amount, distribution and evolution of nitrate in the soil solution throughout the incubation (Supplementary material Table 4).

Discussion

Effects of the chemical and physical characteristics of the mulch

The two mulches that were selected as experimental models to mimic residues from crop associations consisted of two crops that were mixed in different proportions, each having different proportions of plant parts. As expected, the individual residues varied in their characteristics, but the final mixture of maize + lablab (ML) and wheat + alfalfa (WA) did not show large differences in their mean chemical composition and mean C:N ratio (approximately 60). However we observed a faster and higher decomposition of ML compared with that of WA throughout the incubation period. These differences in the decomposition rates translated into significant differences in the chemical composition of the remaining mulches that were collected during decomposition. In our study, we did not determine the individual mass loss for each plant in the mulches because it was impossible to separately recover the small particles of alfalfa and distinguish them from wheat. However, our results can only be explained by the fact that the dynamics of the decomposition of individual components of each mixture varied and suggest that the interactions between the physical and chemical properties of each species occurred during decomposition between the mixture components. These interactions, also called non-additive effects, indicate that the decomposition or C or N mineralization of a mixture of crop residues or plant litters is different from what could be predicted from the weighted mean of individual components of the mixture (Gartner and Cardon 2004). These non-additive effects are attributed to the heterogeneity or dissimilarity in chemical traits of the components of the mixture (Gartner and Cardon 2004; Redin et al. 2014), particularly the N content (Berklund et al. 2013). In a previous study (Iqbal et al. 2013), residue maximal water retention (in g of H_2O per g of residue) was higher for maize stem (3.10±1.12 g g^{-1}) than for wheat (2.33±0.46 g g^{-1}), alfalfa (1.28±0.04 g g^{-1}) and lablab (1.32±0.11 g g^{-1}) stem residues. In the present experiment, the moisture of ML mulch was higher than that of WA at each sampling date under the same rain-evaporation pattern. Makkonen et al. (2013) showed with leaf litters that the litter water-holding capacity (WHC) was closely related to the non-additive effects in mixtures of both poor- and high-quality litter types and concluded that the physical characteristics of some plant litters may improve the microenvironmental (climatic) conditions for decomposers and promote the decomposition of their co-occurring litter species in litter mixtures. Wardle et al. (2003) also suggested a possible role for slowly decomposing litters in facilitating the breakdown of litters from other species through greater moisture retention in the litter layer. Therefore, we believe that in the ML mulch, the maize residue maintained a high moisture, which aided in the decomposition of lablab; we cannot exclude that lablab residue which had a higher N concentration than maize residue (C:N=32.7 vs. 57.4), might by turn favor maize decomposition ("N hypothesis"). Conversely, in the WA mixture, alfalfa (C:N=13.8) represented only 17 % of the total dry matter of the mulch and probably did not provide enough N to significantly modify the decomposition of wheat residue, which had a very high C:N of 278. Therefore, we assume that the evolution of mulch decomposition had been driven by an interaction between the physical and chemical properties of the mixtures, with one litter type providing a "physical advantage" (water retention) and the other litter type providing a "chemical advantage" (nutrient availability). In this study, the effect was more prominent in the ML mixture because of the larger difference in the maximum water retention between the individual components of the mulch (maize and lablab).

Remarkably, these differences in the decomposition of mulches induced also large differences in the dynamics of mulch-N between the two treatments despite a similar initial C:N ratio of the WA and ML mixtures (approximately 60). Changes in the mulch N concentration (or C:N ratio) over time result from the balance between the N loss because of residue mineralization and the N increase (or retention) because of N assimilation by the decomposing microorganisms growing on the mulch particles (Schomberg et al. 1994). At day 84, 57 % (WA) and 86 % (ML) of the initial mulch-N had been mineralized, respectively, which is equivalent to 3.2 (WA) and 4.8 (ML) g N m^{-2}.

Regarding fluxes to the atmosphere, the quality of the mulches influenced C-CO_2 emissions but did not significantly change the N_2O emission. The soil that was covered with WA mulch emitted more CO_2 than did the soil that was covered with ML mulch during the first days of decomposition. The high-soluble pool (equivalent to 45 % of the total DM) of alfalfa compared with other residues can explain this rapid mineralization. This difference was transient because of the fast depletion of the soluble pool. The peaks of CO_2 after each rain resulted from the wetting of mulch particles and enhancement of decomposition, which was observed by Coppens et al. (2006, 2007) under similar experimental conditions with rapeseed and rye residues decomposing on the soil surface. Concerning N_2O emissions, a single but high peak of N_2O (equivalent to 600 to 1200 g N ha^{-1} day^{-1}) was recorded after the first rain. The observed rate was in the high range of values that were obtained for single emission during a plant growing cycle (Hénault et al. 2012). The occurrence of a single peak despite high N availability and a high moisture content of the soil in the column, suggests the importance of the mulch-derived soluble C in the top of the soil, in combination with available N-NO_3 and partial anaerobic conditions under the mulch just after rain. This phenomenon has also been reported by Baggs et al. (2003), who attributed the N_2O emission to the availability of readily degradable C from the rye in the presence of anaerobic conditions under mulch. De Troyer et al. (2011) showed with ^{13}C-labeled maize residues incorporated into the soil that the residue application released a pulse of dissolved organic C that was quickly consumed (<3 days), which is consistent with the rapid depletion that we observed in the soil columns.

Therefore, the type of mulch had a high impact on all of the fluxes, such as the dynamics and rates of decomposition, CO_2 emission and N net mineralization. Despite a similar mean composition, the heterogeneity of the mulch mixtures had a strong effect on these processes. Under our experimental conditions, the water retention properties of the plant residues seemed to have a major effect on the decomposition kinetics and associated fluxes. Several authors have demonstrated that mulch decomposition and/or soil microbial activity are strongly affected by the climatic conditions, particularly precipitation under mulching conditions (Tian et al. 2007; Sanaullah et al. 2012; Lee et al. 2014; Marinari et al. 2014), but none of these studies related mulch decomposition or CO_2 emissions to mulch water retention properties, which were never characterized.

Effects of the soil characteristics

The effect of the soil characteristics on the dynamics of mulch decomposition was analyzed by comparing the ML mulch under the HI_R rain pattern overlying the two different soils that were used in this study (LAC and ALA). The two soils differed greatly by their texture, organic content, water retention properties and ability to mineralize C and N. The two soils behaved differently, particularly with large differences in the water potential in the soil columns. The soil type also modified the dynamics of the decomposition of the mulches, and the mulch moistures were always higher in the LAC soil compared with those in the ALA soil (except at 14 days), even if the differences were not significant. We therefore hypothesize that the possible re-rewetting of the mulch by the underneath soil layer had an impact on its decomposition. This is understandable if we consider that the decomposition of mulch was primarily limited by moisture and not by quality, which was shown previously by Coppens et al. (2007) under similar experimental conditions.

The drainage in the sandy ALA soil was much higher than that in the loamy LAC soil, and this translated into a rapid transport of nitrate down the soil profile after rain. In contrast, no emission of N_2O was observed in the ALA soil, which might have been caused by the higher drainage capacity of the soil and low nitrate content in the upper layer combined with a lower soil density under our experimental conditions. This result indicates that the ALA and LAC soils would be susceptible to different environmental risks under similar climatic conditions.

In this experiment, we chose not to use "intact" soil columns that were collected from the field and preserved as is, which was difficult to achieve, especially with the soil of Madagascar; such conditions would have required a large number of columns per treatment to offset the high spatial variability of soil properties, which was not possible. Therefore, we chose to reconstruct the columns with sieved soil, respecting the gradient of properties and activities that are observed in situ between the surface (0 to 5 cm layer) and underlying layer (5 to 25 cm layer) and with a soil density that was equivalent to that of the field. Several authors (e.g., Shipitalo et al. 2000) have demonstrated that under conservation tillage, the macropore network that is formed by earthworm burrows is not disrupted by tillage and thus permits the downward flow of water and solutes at a higher rate than if the movements occurred only through the soil matrix. In our system of homogeneous soil columns, we can assume that the transport of water and solutes under the mulch was different from that of the undisturbed soils that are found in no-till fields, but the processes in the mulch itself may only be slightly affected by the repacking.

Effect of the rainfall pattern

As hypothesized, the dynamics of mulch decomposition and the total loss of ML mulch C after 84 days were affected by the rain pattern. The ML mulch with light and frequent rainfall remained 2–3 times wetter throughout the incubation period compared with the same mulch under heavy infrequent rainfall. We conclude that frequent rain allowed the mulch water content to remain above the threshold that is favorable for microbial activity. Schimel et al. (1999) reported that the biomass and its activity were strongly related to the moisture content of the litter, and proposed a moisture value of 0.5 g H_2O g^{-1} litter (residue) as a threshold for the measureable microbial activity in litter. Vanlauwe et al. (1995) proposed that the percentage loss of residue dry matter was better correlated with the number of rain events than with the total amount of rain, which is confirmed by our results. Coppens et al. (2007) used a model and showed that a homogeneous distribution of rain (compared with heavy infrequent pulses) had little effect on the decomposition of rape residues that were incorporated into a soil, whereas it had a strong effect on the decomposition of the same but surface-applied residues. Lee et al. (2014) investigated the effects of crop residue placement in the soil with simulated rainfall pulses in the laboratory and an experimental design that was similar to our work (large infrequent vs. small frequent rain pulses). These authors also reported a strong interaction between the frequency of rain pulses and the placement of crop residues (in mulch at the surface vs. buried into the soil).

The kinetics of the CO_2 fluxes (from the soil and from mulch mineralization) responded clearly to the rain regime with the enhancement of mineralization after each rain and rapid decrease between rains; however, the calculated cumulative C mineralization did not show significant differences between the two rain treatments throughout the 84-day incubation. This result was also found by Lee et al. (2014) in the study that is described above. In the case of HIR treatment, more C-CO_2 evolved from the mineralization of soil OM at the end of the incubation (84 days) than with the LFR treatment, as shown with control columns without mulch added (Supplementary material, Fig. 1). Regarding N dynamics, the greater concentration of nitrate in the soil solution in the LFR treatment than that in the HIR treatment, suggests that regular rain application (LFR treatment) favored the leaching of solutes. We also observed a faster immobilization and remineralization of inorganic N in the case of frequent rain compared with infrequent rain resulting from the more rapid decomposition in the LFR treatment, indicating a greater microbial activity and turnover with frequent rains.

Conclusion

This work demonstrates that the characteristics of residue mulches strongly affect their decomposition, which was expected, and that the mean initial characteristics of a mixture of crop residues that mimics field situations with crop associations cannot fully predict their subsequent C and N mineralization. This result emphasizes that better understanding of short-term interactions between different crop residues decomposing together (particularly N and water interactions) is required. This work also demonstrates the role of water retention characteristics of crop residues in the control of mulch decomposition during dry periods. Therefore, we confirmed our main hypothesis that in situations of decomposition at the soil surface (as explored in this study), the decomposition of mulch is primarily under the control of the crop residue moisture, which is under the control of the intrinsic capacity of the crop residue to store water. In contrast, N_2O fluxes and nitrate transport in the soils, which are important to consider for their potential environmental impacts, were greatly affected by the soil type and not particularly affected by the composition of mulches and rainfall pattern under the experimental conditions that were investigated here. Overall, the water dynamics, i.e., evaporation, drainage, and moisture of the mulch and soil, were the main processes that strongly interacted with the three studied factors. Consequently, any climate that has a more regular distribution of rainfall will be more favorable to high residue decomposition under no-tilled conditions. In contrast, a climate with strong fluctuations in the amount and distribution of rainfall, such as those encountered under sub-tropical and tropical conditions, will cause more hazards in the rate of decomposition of residue mulches and thus in the protection of the soil surface and the retention of nutrients. In future studies, scenario modeling using different plant species and climates would help in the generalization and discussion of our laboratory results and contribute to the adaptation of agricultural practices to environmental conditions.

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