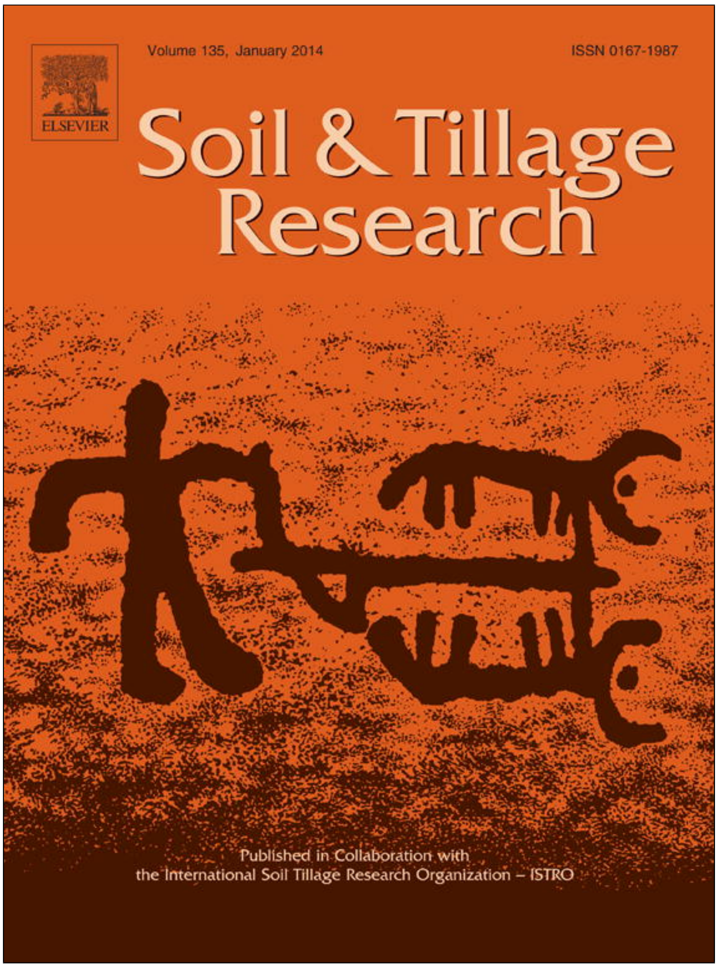
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Soil organic carbon dynamics under different tillage systems in rotations with perennial pastures

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A B S T R A C T

Physical fractionation and ^{13}C determinations are useful techniques for soil organic carbon (SOC) dynamics studies. Changes in SOC content, distribution and origin were assessed after 9.5-year crop-perennial (C3 species) rotation on a Uruguayan Mollisol under conventional tillage (CT) and no-tillage (NT). Soil samples were collected at depths of 0–6, 6–12 and 12–18 cm in 1994 and 2003. Determinations were made of total SOC, particulate organic matter C (POM-C) and mineral-associated organic matter C (MAOM-C). In addition, ^{13}C determinations were made on the total sample and the different particle size fractions. None of the studied variables were affected significantly by the tillage system. SOC levels in 2003 did not differ significantly from those of 1994 at any of the studied depths. However, changes were found in fraction distribution. Within 0–18 cm of the soil surface, POM-C decreased by 63%, whereas MAOM-C did not vary significantly. After 9.5 years, only 14.5% of SOC within 0–18 cm of the soil surface was young SOC. The largest proportion was incorporated within 0–6 cm of the soil surface and in the coarsest physical fractions of organic matter. Only 17% of the estimated C input from crops for the study period was retained by the topsoil. The estimated half-life of SOC within the upper 18 cm of soil was 28 years. Within this layer, the C half-life varied from less than 5 years for POM-C to more than 400 years for MAOM-C. These results suggest that agricultural rotation systems including perennial pastures are capable of maintaining SOC levels even under CT. However, C cycling and other ecosystem processes may be altered due to the significant loss of labile organic matter. The use of ^{13}C analysis enabled the estimation of parameters relevant to the modeling of SOC dynamics.

1. Introduction

The soil organic carbon (SOC) stock results from the balance between inputs resulting from the decomposition of plant and animal residues and outputs due to erosion and microbial oxidation (Alvarez and Steinbach, 2006a; Urquiaga et al., 2007). Residue decomposition rates depend largely on the physical and biochemical characteristics of each type of organic material, as well as on the interaction of the latter with the mineral fraction and microorganisms occurring in a particular soil type, in addition to the environmental conditions, mainly temperature and moisture (Urquiaga et al., 2007).

Traditionally, research into SOC dynamics has focused on determining the productive functions of soil. Over the past decades, however, the environmental potential of soil dynamics has also gained in importance, since soil can act as either a source or a sink of CO_2, a greenhouse gas (Balesdent and Mariotti, 1996; Urquiaga et al., 2007).

Not only may a certain soil management procedure result in net SOC gains or losses but it can also lead to changes in the composition of soil organic matter, even if no significant changes in SOC content are to be detected. Determinations of ^{13}C have been used in several SOC studies (Gregorich et al., 1995; Balesdent and Mariotti, 1996; Andriulo et al., 1999a; Collins et al., 1999; Urquiaga et al., 2007). This technique is based on the difference between the ^{13}C proportion of C_3 species and that of C_4 species (average $\delta^{13}C$: −27‰ and −12‰, respectively) and on the fact that SOC retains the $\delta^{13}C$ signal of vegetation. Thus, a change in the photosynthetic cycle of vegetation growing on a soil will be reflected in a change in the isotopic composition of SOC. Based on the above, the origin of organic matter may be traced, and the path and dynamics of its transformations studied (Balesdent and Mariotti, 1996).

The physical fractionation of soil organic matter (SOM) according to particle size or density has also been used in several SOC studies (Christensen, 2001; Urquiaga et al., 2007). Experimental physical fractionation efforts have been aimed at isolating SOM pools according to turnover rates. Those SOM fractions

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corresponding to the size of the coarsest soil particles (sand size), referred to as particulate organic matter (POM), consist of only partially decomposed plant material with a C/N ratio nearly as high as that of the starting plant material and a higher decomposition rate than that associated with silt and clay carbon (Christensen, 2001). The coarse fractions have been demonstrated to be more sensitive to changes associated with soil use and management (Cambardella and Elliot, 1992; Bayer et al., 2001; Morón and Sawchik, 2003; Galantini et al., 2004). The quantification of the progressive incorporation of new C into the different SOC fractions enables the elucidation of the transformation and stabilization paths of organic carbon (Balesdent and Mariotti, 1996).

Generally, ^{13}C determinations and physical fractionation methodologies have been used to assess the effect of vegetation changes (C_3 to C_4 species, or vice versa), based on monoculture studies. In contrast, crop-pasture rotation systems are commonly used for grain and livestock production in Uruguay (García Préchac et al., 2004). Traditionally, soil tillage has been used before each crop and at pasture planting. Over the past decades, the expansion of no-tillage systems has enabled a reduction in the amount of tillage, minimizing SOM losses due to mineralization and erosion. Although several studies have focused on how these systems contribute to changes in the SOC stock (Morón, 2003; Terra et al., 2006; Salvo et al., 2010), little has been reported on the contributions of new crop and pasture vegetation to the total SOC stock or its individual fractions.

Therefore, the objective of this study was to quantify the change in SOC content, distribution and origin after a 9.5-year crop-pasture (C_3) rotation under conventional tillage (CT) and no-tillage (NT) on a soil with a history of several cops under CT.

2. Materials and methods

2.1. Experimental conditions and management procedure

This study was conducted within the framework of an ongoing long term field trial iniciated 1993, at the Mario A. Cassinoni experimental station of Uruguay's Universidad de la República (Faculty of Agronomy). Located at a 10-km distance from the city of Paysandú (32°21′S 58°02′W), the Station is comprised within a sub-humid region with annual, winter and summer average temperatures of 17, 12 and 24 °C, respectively. The soil at this site is classified (USDA) as a fine, mixed, active, thermic Typic Argiudoll on a slope of less than 1% with an A horizon 18 cm deep, $pH_{(H2O)}$ 5.7 (soil water relation v:v = 1:2.5), and clay, silt and sand contents of 289, 437, 273 g kg^{-1}, respectively.

The framework experiment combines different crop-pasture rotations with CT and NT (Salvo et al., 2010). Among these rotations, that with the highest proportion of C_3 species during the period from 1994 to 2003 (treated under both CT and NT) was selected for this study.

In a distant past, the natural vegetation of the experimental site was a mixture of C_3 and C_4 species. The highest occurrence and abundance were observed, among C_4 species, for *Botriochloa laguroides* DC., *Paspalum dilatatum* Poir., *Paspalum notatum* Flűgge and *Setaria vaginata* Spreng, and, among C_3 species, for *Briza subaristata* Lam., *Bromus auleticus* Trin., *Bromus catharticus* Vahl., *Piptochaetium stipoides* Hack., *Stipa hyalina* Nees, *S. megapotamica* Spreng, *Sterculia setigera* Presl. (García et al., 2005). The soil cover prior to the commencement of this study may be characterized according to the following two periods. Over the 1970–1986 period, crop-pasture rotations under CT included C_3 and C_4 species. Then, the plot was planted with perennial pastures of C_3 species (*Festuca arundinacea* Schreb., *Lotus corniculatus* L. and *Trifolium repens* L.), which had progressively been invaded by *Cynodon dactylon* (a C_4 species) by the start of the experiment (1993).

Table 1
Crop sequences of crop-pasture rotations under CT and NT from 1993 to 2003.

Year	Winter crop	Summer crop
93/94	Barley	Sorghum
94/95	Wheat	Sunflower
95/96	Wheat with PP[a]	PP
96/97	PP	PP
97/98	PP	PP
98/99	Fallow	Corn
99/00	Wheat	Fallow
00/01	Wheat	Soybean
01/02	Fallow	Sunflower
02/03	Wheat with PP	PP
2003	PP	–

PP: perennial pasture (*Festuca arundinacea* Schreb., *Lotus corniculatus* L. and *Trifolium repens* L.).
[a] Pasture was sod together with wheat to reduce sawing costs. Wheat is harvested for grain, after which pasture is grazed.

The crop-pasture rotation used in the experiment consisted of three years of crops (two crops per year) and three years of livestock-grazed pastures (Table 1). A chisel plow and an excentric disk harrow were used for CT at a depth of 15–20 cm. On average, four plowing operations were used for soil preparation in winter, while two operations were used for summer crops. Agrochemical management (fertilizers, herbicides, insecticides and fungicides) was conducted according to the requirements of each tillage system.

Wheat (*Triticum aestivum* L.) (C_3) and barley (*Hordeum vulgare* L.) (C_3) were cultivated in winter, and sunflower (*Helianthus annus L.*) (C_3) and soybean (*Glycine max* L.) (C_3) in summer (Table 1). Pasture mixes included *F. arundinacea* Schreb., *L. corniculatus* L. and *T. repens* L., which were cultivated between 1995 and 1998 (first cycle) and from 2002 to 2003 (first year of the second cycle). Only two C_4 crops were seeded during the study period: sorghum in the 93/94 summer (South hemisphere) and corn in the 98/99 summer (Table 1). A randomized complete block design with three replications was used for the analysis. Plot size was 10 × 50 m.

2.2. Sampling and measurements

Soil samples were collected in January 1994 (summer in the southern hemisphere) before Sorgum harvest, for the two tillage types at depths of 0–6, 6–12 and 12–18 cm. At the end of the first year of pastures, in June 2003, the soil was sampled again at depths of 0–3, 3–6, 6–12, 12–18 cm. Prior to collecting each sample, aboveground crop residues were removed from the soil surface. Samples were composed of 20 cores per plot. At each soil depth, undisturbed samples were collected to determine bulk density. To such end, ring samplers 3 cm high and 5.4 cm wide were used, which were introduced vertically in the soil. Three replicates per plot were collected.

Soil samples for carbon analysis were sieved to less than 2 mm and physically fractionated according to Cambardella and Elliot (1992), separating coarse and fine particulate organic matter (POM-C > 200 μm, POM-C > 50 μm, respectively) from mineral-associated organic matter (MAOM-C; smaller than 50 μm). Carbon content and ^{13}C isotopic abundance were determined for each depth and size fraction by dry combustion (IRMS Thermo Finnigan Delta Plus coupled to a Flash EA 112 elemental analyzer). The C recovery from the three fractions averaged 90% of total SOC. MAOM-C was calculated as the difference between total SOC and POM-C.

Natural ^{13}C abundance, expressed in delta (δ) units, indicating the isotopic ratio of the sample relative to that of the Pee Dee

Belemnite (PDB) standard, may be calculated as follows:

$$\delta^{13}C\,(‰) = \left[\frac{(^{13}C/^{12}C) - (^{13}C/^{12}C)_{PDB}}{(^{13}C/^{12}C)_{PDB}}\right] \times 1000 \quad (1)$$

where $^{13}C/^{12}C$ is the isotopic ratio of the study material and $^{13}C/^{12}C_{PDB}$ that of the reference standard (PDB).

2.3. Estimates and calculations

2.3.1. Percentage of young carbon

The carbon input from new vegetation was calculated as follows:

$$\alpha(\%) = 100 \times \frac{(\delta - \delta 0)}{(\delta 1 - \delta 0)} \quad (2)$$

where α is the ratio of young C to total C, expressed as a percentage, δ is the $\delta^{13}C$ of SOC at $t = 1$, δ_0 is the $\delta^{13}C$ of SOC at $t = 0$, and δ_1 is $\delta^{13}C$ of new vegetation residues.where, δ was the $\delta^{13}C$ of SOC under each treatment in 2003; δ_0 was considered equal for both treatments and thus obtained as the average $\delta^{13}C$ of SOC under CT and NT in 1994; δ_1 was determined as the average $\delta^{13}C$ of C_3 species grown during the study period (average $\delta^{13}C$: −27.89) (Table 2).

2.3.2. Organic carbon retention in soil

The percentage of organic C retained in the soil ($C_{retained}$) was calculated as follows:

$$C_{Retained}(\%) = \frac{SOC_{young}}{C_{input}} \times 100 \quad (3)$$

where SOC_{young} was the amount of young C present in 2003, calculated from $\delta^{13}C$ values according to Eq. (2), and C_{input} was the quantity of C input as plant residues from 1994 to 2003.

The amount of C incorporated within the topsoil (0 to 18 cm) due to root death and new vegetation harvest residues was calculated as follows:

- Based on reported cereal crop yields obtained using rotation, a harvest index of 35% (based on the total aboveground dry matter) was assumed for wheat, soybean and sunflower (Alvarez and Steinbach, 2006b), with an aboveground/root ratio (AG/R) of 5 (Bolinder et al., 2007).
- As pasture yield data were not available, a conservative national estimate for pastures of the same species was used (Crempien, 1978). It was assumed that only 20% of the total biomass produced by the pastures returned to the soil and that the remaining 80% was removed by livestock. The AG/R ratio was assumed to be 0.7 (Bolinder et al., 2007).
- For all crops and pastures, it was assumed that the root exudate production amounted to 65% of root dry matter (Bolinder et al., 2007) and that 90% of roots were located within the upper 18 cm of soil, as well as that 40% of plant residue dry matter was C
- (Alvarez and Steinbach, 2006b).

Table 2
$\delta^{13}C$ values for C_3 crops.

Culture	$\delta^{13}C$
Soybean	−27.32
Sunflower	−27.50
Wheat	−27.88
Perennial pasture	−28.84
Average	−27.89

Calculation example for C input made by crops:

$$CCInput = (PHR + R + RE) \times C\% \quad (4)$$

where CCinput is the input of C made by crop (Mg ha^{-1}), PHR is the amount of above-ground post harvest residue (excluding harvest product), R is the amount of root residue in the evaluated depth, RE is the root exudates and C% is the C percentage of the vegetation.

PHR, R and RE were calculated as indicated by the following equations:

$$PHR = \frac{CY}{HI} - CY \quad (5)$$

where CY is the crop yield (Mg ha^{-1}) and HI is the harvest index, all in dry matter basis.

$$R = \frac{PHR + CY}{S:R} \times 0.9 \quad (6)$$

where S:R is de shoot:root ratio and 0.9 corresponds to the percentage of root mass found in the evaluated depth.

$$RE = R \times 0.65 \quad (7)$$

where 0.65 is the amount of the root exudates per unit of root.

Decomposition constant and half-life of soil organic carbon and its fractions

To calculate the C decomposition constant (k), an exponential SOC content was assumed, such that:

$$C_{(t)} = C_0 \exp^{(-kt)} \quad (8)$$

where $C_{(t)}$ is the amount of original C (input from existing vegetation prior to the experiment) still present in 2003, C_0 is the SOC content at the start of the experiment (*i.e.*, in 1994), k is the decomposition constant, and t is the time elapsed since the vegetation change (*i.e.*, 9.5 year).

Following the estimation of k, the C half-life (HL), *i.e.*, the time in which C_0 is reduced by half, was calculated based on Eq. (5):

$$\frac{C_0}{2} = C_0 \exp^{(-kt)} \quad (9)$$

solving for t in this equation, and considering t = HL:

$$HL = \frac{\ln(2)}{k} \quad (10)$$

3. Statistical analysis

An analysis of variance was performed by SAS GLM procedure (SAS Institute, 1990). Tukey's least significant difference (LSD) ($P < 0.05$) was used to separate the means of the two treatments where effects were found to be significant.

4. Results and discussion

4.1. Changes in SOC content and composition

No significant SOC differences (either in g kg^{-1} or in Mg ha^{-1}) were detected at any of the studied depths between CT and NT in 2003 or between 2003 and 1994 values (Fig. 1(A)). The calculated SOC concentration within the upper 18 cm was 21.4 g kg^{-1} in 1994 and 19.8 and 20.0 g kg^{-1} in 2003 for CT and NT treatments, respectively.

It has been suggested that equivalent soil masses, instead of fixed depths, should be used for comparisons of SOC stock between soils subjected to either treatment (Ellert and Bettany, 1995; Alvarez and Steinbach, 2006c). Here, a fixed depth of 18 cm was used because the soil mass found at this depth did not differ between treatments or vary over the years (unpublished data). The

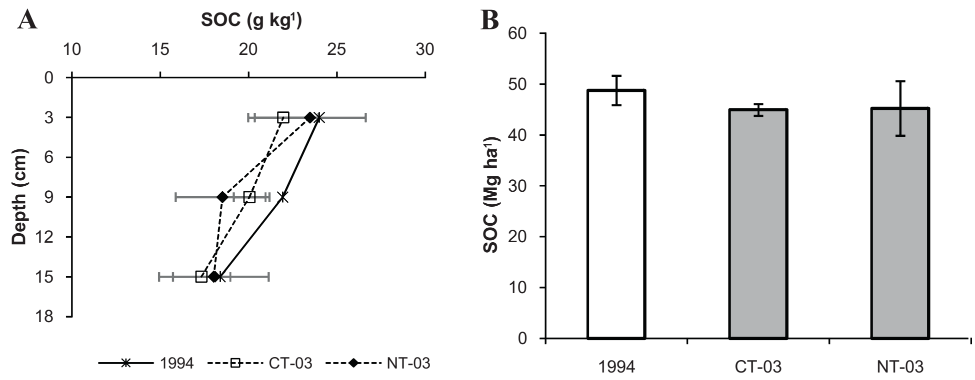

Fig. 1. SOC in 1994 and 2003 for CT and NT. (A) Carbon concentration (g kg^{-1}) according to depth. (B) SOC stock (Mg ha^{-1}) within the upper 18 cm of soil. Note: CT-03: crop-pasture under conventional tillage in 2003; NT-03: crop-pasture under no-tillage in 2003. Points in (B) indicate the middle point at the sampling depth (0–6, 6–12, 12–18 cm). Horizontal error bars (A) and vertical error bars (B) indicate ±1 standard deviation for each mean value.

1994 SOC content was 48.75 Mg ha^{-1}, decreasing in 2003 to 44.93 and 45.22 Mg ha^{-1} for CT and NT, respectively, with no significant differences between treatments (Fig. 1(B)). Thus, after 9.5 years of crop-pasture rotations, SOC levels did not differ significantly from the baseline values (1994). In turn, in these crop-pasture systems, CT did not lead to increased SOC losses with respect to NT.

The rotation systems compared in the present study do not represent the highest contrast from the standpoint of their impact on the soil. Both have short periods of bare soil due to the inclusion of two crops per year and pastures in the rotations. This generates a continuous residue input and reduces the time in which soil is susceptible to erosion (especially under tillage), the main cause of SOC losses under Uruguayan conditions (Clérici et al., 2004).

Similar results were obtained in long-term experiments conducted in Uruguay on soils with a higher slope (*ca.* 3%) but considering a similar depth to that of this study (15 to 20 cm). A crop-pasture rotation system under CT for 40 years (50% of which time pastures with perennial grasses and legumes were grown) did not lead to significant changes in SOC content with respect to the baseline (Morón, 2003). Likewise, in another experiment using NT, soils under rotation systems including a certain proportion of pastures had a similar SOC content to that of soil with natural pastures after an assessment period of 8 years (Terra et al., 2006).

Díaz-Zorita et al. (2002) found an interaction between tillage and rotation systems, where SOC level maintenance depended on the inclusion of perennial pastures when annual crops were seeded with CT, while NT systems may not need the inclusion of pastures to maintain SOC levels. However, some studies reported greater total organic C contents under perennial grass systems than under conservation-tilled annual crops (Ismail et al., 1994; Janzen et al., 1998). Crop-pasture rotations, even under CT, are associated with the concept of environmental and productive sustainability (Díaz Rossello, 2003; Díaz-Zorita et al., 2002; García Préchac et al., 2004). The extent of SOC increase in these rotations depends on management factors such as pasture phase duration, pasture type, grazing, and nutrient management during the cropping phase (Guo and Gifford, 2002).

Whereas no significant differences in SOC content were found between treatments or over the study period, variations in SOC composition were observed between years. At all depths, a marked decrease in POM-C throughout the study period (Table 3) reflected a lower proportion of POM-C in SOC in 2003 than in 1994 ($P < 0.05$). Within the upper 18 cm of soil, the POM-C-to-SOC ratio (expressed as a percentage) was 18.2% in 1994 and 6.8% in 2003, considering the average of the two treatments ($P < 0.05$). At the same depth, POM-C decreased to 63%, amounting to a reduction from 9.05 to 3.31 Mg ha^{-1} ($P < 0.05$), while MAOM-C did not show significant changes. Consistently with reported data (Cambardella and Elliot, 1992; Bayer et al., 2001; Morón and Sawchik, 2003), these results suggest that POM-C is a more sensitive indicator of changes in soil use and management than SOC. The loss of POM-C has been reported to have an impact on nutrient dynamics (Chan, 1997; Galantini et al., 2004), microbial activity (Alvarez et al., 1998) and soil macroaggregate formation and stability (Chan, 1997; Six et al., 2002).

The observed reduction in POM-C with time amounted to a greater percentage in the coarser POM fraction (>200 μm), the degree of reduction increasing with depth (Table 3). From 1994 to 2003, POM-C (CT and NT average) decreased by 53%, 71% and 79% at depths from 0 to 6, 6 to 12 and 12 to 18 cm, respectively. According to Gale and Cambardella (2000), in relatively undisturbed soil, POM is derived primarily from roots and these do their major biomass contribution in the near surface soil layers. Hence, as the greatest and most continuous input of fresh residues occurs at the soil surface (Franzluebbers, 2002) a greater degree of replacement of lost POM-C should be expected to have occurred within this stratum.

A slight increase in MAOM-C was observed over the study period at nearly all depths. However, these changes were not significant. An average decrease in POM-C of 5.74 Mg ha^{-1} within the upper 18 cm of soil was greater than the associated reduction

Table 3
Carbon content (Mg ha^{-1}) of each physical fraction at different depths in 1994 and following CT and NT in 2003.

Depth (cm)	Treatment	SOC physical fraction (Mg C ha^{-1})		
		POM-C > 200	POM-C > 50	MAOM-C
0–6	1994	2.31 a[a]	2.31 a	14.25 a
	CT-03	0.77 b	1.15 a	14.50 a
	NT-03	1.01 b	1.41 a	14.97 a
6–12	1994	0.96 a	1.45 a	13.91 a
	CT-03	0.18 b	0.64 b	14.52 a
	NT-03	0.16 b	0.44 b	13.78 a
12–18	1994	0.82 a	1.21 a	11.55 a
	CT-03	0.11 b	0.27 b	12.79 a
	NT-03	0.14 b	0.34 b	12.96 a
0–18	1994	4.08 a	4.97 a	39.70 a
	CT-03	1.07 b	2.05 b	41.81 a
	NT-03	1.31 b	2.19 b	41.72 a

[a] Within a given depth range in each column, different letters indicate significant differences between treatments ($P < 0.05$) according to the Tukey test.
Note: CT-03: crop-pasture under conventional tillage in 2003; NT-03: crop-pasture under no-tillage in 2003; POM-C > 200: fraction of particulate organic matter larger than 200 μm in size; POM-C > 50: fraction of particulate organic matter of size between 50 and 200 μm; MAOM-C: C in mineral-associated organic matter.

in total SOC, the latter amounting to 3.67 Mg ha^{-1}. The above suggests that a large portion of POM-C has been mineralized and lost to the atmosphere as CO_2, but a certain portion must have been incorporated as MAOM-C, which trended higher by approximately 2.0 Mg ha^{-1} over the study period.

The redistribution of SOC from labile fractions to humidified fractions has also been reported (Cambardella and Elliot, 1992) in a study using a 20-year wheat culture under NT, which was only tilled once, to break out the native short-grass prairie grass sod, before the establishment of long-term NT. Similar results were obtained by Galantini et al. (2004) in a study conducted in Argentina. They studied the variation in the organic fraction dynamics during a nine-year system rotating three years of wheat (*T. aestivum* L.) and three years of clover (*Trifolium pratense* L.). The reported POM-C content was lower than that of the reference soil (non-cultivated pasture), while the MAOM-C content was similar to (slightly higher than) that of the reference. MAOM-C varied only slightly between rotation phases, showing higher values in the pasture phase. In contrast, POM-C increased considerably during the pasture phase and decreased during the phase of wheat cultivation.

Results presented in this paper are not in agreement with those of a previous 8-year study conducted in Uruguay using different forage crop rotation systems managed under NT, according to which, the POM-C content within the upper 15 cm of soil beneath permanent pastures (natural pastures including legumes) did not differ with treatments that included pastures in the rotation (Terra et al., 2006). These differences in results may be related to: (i) a higher proportion of C_3 crops in the rotation systems used in this study, as POM-C has been observed to be higher in rotation systems with a higher proportion of C_4 crops (Salvo et al., 2010); (ii) the fact that crops were used over longer time periods than pastures (6 and 4 years, respectively) in this study, so that the effects of pastures on the fractions of soil C may not have become apparent over the time of this study.

4.2. Changes in the $^{13}C/^{12}C$ ratio of soil organic carbon

The change in vegetation (mixture of C_3 and C_4 replaced by C_3 species) led to differences in SOC $\delta^{13}C$ between 1994 and 2003 (Fig. 2).

In the reference year (1994), the average $\delta^{13}C$ of SOC within the upper 18 cm was −17‰—*i.e.*, a value closer to that characteristic of

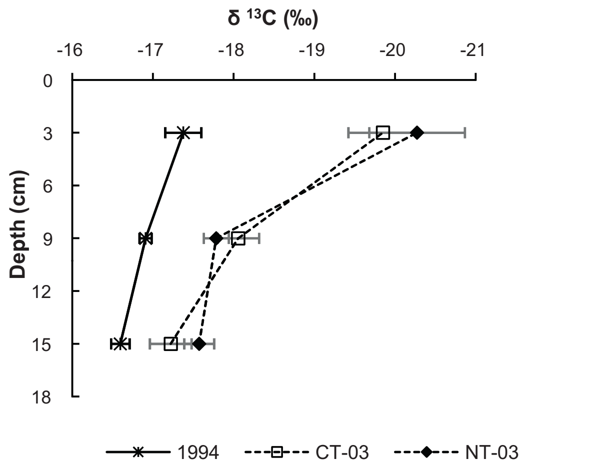

Fig. 2. $\delta^{13}C$ of SOC at different depths in samples collected in 1994 and 2003. Note: CT-03: crop-pasture under conventional tillage in 2003; NT: crop-pasture under no-tillage in 2003. Points in the figure indicate the middle point at the sampling depth (0–6, 6–12, 12–18 cm).

C_4 species ($\delta^{13}C$: −12‰) than to that of C_3 species ($\delta^{13}C$: −27‰) (Gregorich et al., 1995). This was due to the presence of native C_3/C_4 mixed vegetation as pasture, followed by a management that may have altered the relative prevalence of species with different photosynthesis cycle. Although the soil was under a C_3 pasture for 7 years prior to the commencement of the experimental stage, it had been invaded by *C. dactylon* (C_4 species, $\delta^{13}C$: −14.15 ‰).

Fig. 2 shows an increasing ^{13}C proportion in SOC with increasing soil depth in all the treatments. This is consistent with previous work, even using pure systems of C_3 or C_4 species. Such behavior has been attributed to the isotopic fractionation produced by decomposer organisms, which use preferentially ^{12}C and, thus, concentrate ^{13}C in the organic matter remnants (Boutton, 2002). The observed pattern is in line with the expected behavior, as a higher proportion of humidified SOC was found with increasing soil depth, where organic matter is exposed to microbial activity for a longer time period (greater $\delta^{13}C$) than young C deposited in the proximity of the soil surface (Boutton, 2002).

Whereas SOC was significantly impoverished in ^{13}C within the upper 18 cm of soil after 9.5 years of crop and C_3 pasture rotation (Fig. 2), no significant differences were found between CT and NT treatments regardless of depth. $\delta^{13}C$ decreased by 2.7 units (CT and NT average) within the upper 6 cm of soil, and by 1 and 0.8 units at depths of 6–12 cm and 12–18 cm, respectively ($P < 0.05$). This reflects the net soil carbon input associated with the incorporation of plant residues within 0–18 cm of the soil surface, although inputs were greater within the upper 6 cm of topsoil.

No significant differences in $\delta^{13}C$ according to depth were found between CT and NT within a given fraction (Table 4). At each soil depth, differences in $\delta^{13}C$ between 1994 and 2003 increased with increasing size of the SOC physical fraction. As coarse POM corresponds to the youngest residues (Christensen, 2001; Galantini et al., 2004), the associated $\delta^{13}C$ is similar to that of the most recently incorporated residues (Table 4). Regardless of depth, both POM size factions were significantly impoverished in ^{13}C ($P < 0.05$). Considering the upper 18 cm of soil, $\delta^{13}C$ differences over the study period within fractions amounted to 5.5 and 3.3‰ for fraction sizes larger than 200 μm and between 50 and 200 μm, respectively (average CT–NT).

An impoverishment in ^{13}C was also observed for MAOM-C, mainly in the upper 6 cm of soil, where ^{13}C differed by 1.1‰ ($P < 0.05$) (Table 4). However, as no significant variation with time was observed over the study period in the other depths, the humification of plant remains appears to have occurred mainly within the upper 6 cm of soil.

The percentage of young C – *i.e.*, the amount of SOC in 2003 incorporated as residues of C_3 species seeded after 1994 (Table 5) – was calculated according to Eq. (2). This variable did not differ between CT and NT irrespective of depth.

Young carbon within the upper 6 cm of soil represented *ca.* 25.5% of total SOC, decreasing to 9.2% at a depth of 6–12 cm and to 7.1% at 12–18 cm (Table 5). This reflects the incorporation of 4.26, 1.36 and 0.94 Mg C ha^{-1} within each depth range (CT–NT average), the amount incorporated within the upper 6 cm of soil being 1.85-fold greater than that incorporated between 6 and 18 cm. This shows a greater C input in the proximity of the surface even under CT systems, consisting of aboveground and root remains of the cultivated crops and perennial grasses, consistently with the stratification found in systems that include perennial pastures and/or systems with no tillage (Franzluebbers, 2002).

The greatest rate of C input was found in the POM fractions located within 0 to 6 cm of the soil surface (Table 4), with young C amounting to 80.5% of POM-C > 200 and 52.2% of POM-C > 50, respectively (average CT–NT). At depths of 6–12 and 12–18 cm, the proportion of young C represented *ca.* 52% of POM-C > 200 and 31% of POM-C > 50. Young C represented only 12.4% of MAOM-C at

Table 4
$\delta^{13}C$ of each physical fraction at different depths in 1994 and following CT and NT in 2003; and young C of each physical fraction measured at the end of period.

Depth (cm)	Treatment	SOC physical fraction					
		POM-C > 200	POM-C > 50	MAOM-C	POM-C > 200	POM-C > 50	MAOM-C
		$\delta^{13}C$ (‰)			Young C (%)		
0–6	1994	−19.25 a[a]	−19.38 a	−17.44 a	–	–	–
	CT-03	−26.25 b	−23.49 b	−18.62 b	81.10 a	48.40 a	11.19 a
	NT-03	−26.15 b	−24.16 b	−18.88 b	79.93 a	56.01 a	13.79 a
6–12	1994	−18.66 a	−18.85 a	−17.02 a	–	–	–
	CT-03	−23.48 b	−21.60 b	−17.62 a	52.21 a	30.46 a	5.18 a
	NT-03	−23.41 b	−21.78 b	−17.39 a	51.47 a	32.36 a	3.42 a
12–18	1994	−18.73 a	−18.45 a	−16.79 a	–	–	–
	CT-03	−23.31 b	−20.95 b	−16.83 a	50.12 a	26.43 a	0.36 a
	NT-03	−23.74 b	−21.25 b	−17.05 a	54.53 a	29.67 a	2.33 a

[a] Within a given depth range in each column, different letters indicate significant differences between treatments ($P < 0.05$) according to the Tukey test.
Note: CT-03: crop-pasture under conventional tillage in 2003; NT-03: crop-pasture under no-tillage in 2003; POM-C > 200: fraction of particulate organic matter larger than 200 μm in size; POM-C > 50: fraction of particulate organic matter of size between 50 and 200 μm; MAOM-C: C in mineral-associated organic matter.

Table 5
$\delta^{13}C$ and SOC values in 1994 and 2003, and young C incorporated over the period, according to depth.

Depth (cm)	Treatment	SOC		Young C	
		$\delta^{13}C$ (‰)	Mg ha^{-1}	%	Mg ha^{-1}
0–6	1994	−17.38 a[a]	18.86 a	–	–
	CT-03	−19.85 b	16.42 a	23.47 a	3.85 a
	NT-03	−20.27 b	17.40 a	27.47 a	4.67 a
6–12	1994	−16.91 a	16.32 a	–	–
	CT-03	−18.06 b	15.34 a	10.46 a	1.60 a
	NT-03	−17.78 b	14.38 a	7.97 a	1.13 a
12–18	1994	−16.60 a	13.57 a	–	–
	CT-03	−17.22 b	13.17 a	5.53 a	0.73 a
	NT-03	−17.57 b	13.44 a	8.66 a	1.16 a

[a] Within a given depth range, different letters in the column indicate significant differences ($P < 0.05$) between treatments according to the Tukey test.
Note: CT-03: crop-pasture under conventional tillage in 2003.
NT-03: crop-pasture under no-tillage in 2003.

0–6 cm, and 4.1% and 1.5% of MAOM-C at 6–12 and 12–18 cm, respectively. These results suggest that humification may be a slower process at greater depths, presumably because there are fewer roots, which results in smaller C inputs and reduced microbial activity.

The use of tillage did not significantly affect the relative amount of young C in SOC; nor did it affect the composition of the different size fractions (Tables 4 and 5). The amount of young C incorporated into the topsoil (0–18 cm; CT–NT average) was 6.56 Mg C ha^{-1}, amounting to 14.5% of total organic C. The percentage of C incorporated as a result of recent vegetation growth was 61.6%, 37.2% and 6.1% in the POM-C > 200 μ, POM-C > 50 μ and MAOM-C fractions, respectively. It should be noted that the $\delta^{13}C$ of the unfractionated samples differed from the weighted mean based on the $\delta^{13}C$ values obtained for the three physical fractions (Tables 3 and 4). Thus, the total amount of young C, obtained by addition of the contributions from the three fractions, was 33% lower than the value calculated for the complete sample. Data from the unfractionated samples are associated with a smaller error – and are therefore more reliable – than data obtained adding the $\delta^{13}C$ values of the fractionated samples. Similar behavior also was reported by Balesdent et al. (1988).

The carbon input to soil from the biomass of C_3 species over the time period was estimated at 38.63 Mg ha^{-1} (Table 6), of which only 6.56 Mg C ha^{-1} was retained in the soil until 2003 (young carbon), representing 17% of the incorporated amount (average for the two tillage treatments). In the case of CT, approximately 83% of C appears to have been released to the atmosphere. In order to calculate carbon losses under NT, it should be necessary to deduct the amount of C retained in the residues on the soil surface (no data available). Gale and Cambardella (2000), using ^{14}C in a simulated no-till oat experiment, concluded that only 11% of the original ^{14}C remained in residue on the soil surface after a year incubation, whereas 16% was incorporated to the soil, and 66% was breathed as $^{14}CO_2$. These authors concluded as well that 75% of the new C retained under NT came from root inputs.

In our study, the inclusion of two C_4 crops means that increases in $\delta^{13}C$ may have been higher than those reported, and so may the

Table 6
Accumulated C_3 crop yield, accumulated pasture production, estimated C input to soil, and percentage of C retention after 9.5 year (CT and NT).

Treatment	Accumulated yield (W/Sun/Soy[a]) (Mg DM ha^{-1})	Accumulated production[b] (perennial pasture)	C input from C_3 species in the upper 18 cm of soil		Carbon retained by soil (%)
			Mg ha^{-1}	Mg ha^{-1}	
CT-03	17.09 a	20.99	37.99	4.00	16.3
NT-03	18.29 a	20.99	39.28	4.13	17.7

[a] W: wheat, Sun: sunflower, Soy: soybean.
[b] Pasture production data used in forage budgets, according to Crempien (1978).
Note: CT-03: crop-pasture under conventional tillage in 2003 NT-03: crop-pasture under no-tillage in 2003; DM: dry matter.

percentage of young C. Thus, the actual amount of C incorporated into the soil could have been slightly greater than reported, as the percentage released to the atmosphere is expected to have been lower.

Similar results were found after a 13-year soybean monoculture initially planted on a natural pasture (Typic Argiudoll) (Pergamino, Argentina), finding that only 13% of the C incorporated as residues was retained by the soil (Andriulo et al., 1999a). Gregorich et al. (1995) (Ontario, Canada) reported that over 75% of the amount of incorporated C returned to the atmosphere as CO_2 in a 25-yr continuous corn crop under tillage. Andriulo et al. (1999a) have also reported retention values of 10 to 22% of C in studies conducted in France, Brazil and Canada. The above C retention rates are markedly lower than the 33% reported in the classic literature (Brady and Weil, 2002). The retention rate is expected to vary with, *i.a.*, annual mean temperature, amount and quality of biomass input, and soil physical characteristics and management (Andriulo et al., 1999a; Collins et al., 1999; Bayer et al., 2006).

The average decomposition constant (CT–NT average) of original C was 0.025 yr^{-1} ($C_{(t)}$: 38.51 Mg ha^{-1}, C_0: 48.75 Mg ha^{-1}, 1, with *t*: 9.5) with a half-life (HL) of 28 years. This decomposition constant is similar to values obtained in cold and temperate regions, with yearly decomposition rates below 2% (Balesdent et al., 1990). Andriulo et al. (1999b) reported decomposition constants between 0.022 and 0.036 yr^{-1} for five crop systems in Argentina. According to work cited by Bayer et al. (2006), these values are greater in tropical and subtropical regions, reaching up to 10% (0.10 yr^{-1}). The decomposition or humification coefficient is affected by climate (temperature and rainfall), soil characteristics (texture and mineralogy) and soil management (Bayer et al., 2006).

The carbon HL was drastically smaller for the POM-C fractions. The fractions POM-$C > 200\ \mu$ and POM-$C > 50\ \mu$ showed a HL of 2.6 ($k = 0.27\ yr^{-1}$) and 4.6 ($k = 0.15\ yr^{-1}$) years, respectively (average of CT–NT). In contrast, the HL of MAOM-C was 425 years, indicating a high degree of inaccessibility to decomposer organisms and the presence of strong organic-mineral interactions, although all organic matter stabilization processes may be acting in a combined manner in this fraction (Lutzow et al., 2007).

Consistently with the results reported herein, Andriulo et al. (1999b) reported that the HL of the readily mineralizable fraction (active fraction of a modified version of the Hénin–Dupuis model, (Hénin and Dupuis, 1945) varied from 2.5 to 4 years. This modified model version is a three compartments model, separating soil organic matter into an active fraction (supplied from crop residues) and a stable fraction (Janssen, 1984; Andrén and Kätterer, 1997). On the other hand, Cambardella and Elliot (1992) reported higher HL values (13 years) for POM derived from a grass-like pasture following a 20-yr wheat crop. While these authors suggested that POM might represent a large part of the "slow pool" of the multicompartment model proposed by Parton et al. (1987), the turnover times (1/*k*) reported here for the different POM fractions (3.7 for the fraction greater than 200 μm, and 6.7 years for the fraction between 50 and 200 μm) appear to be consistent with those of the "active pool" proposed by this model (1–5 years).

Differences found in the HLs of different physical fractions confirm the heterogeneity of constituent substances of the organic pool of soil and the different retention mechanisms acting on these fractions (Christensen, 2001; Lutzow et al., 2007).

5. Conclusions

After 9.5 years, crop-pasture rotation systems maintained initial SOC levels within the upper 18 cm of soil, even under CT, and can thus be considered sustainable production systems. However, SOC became less labile, with possible implications for C cycling and other ecosystem functions.

The natural abundance of ^{13}C enabled the detection of biomass remains recently incorporated (C_3 species) within 18 cm of the soil surface (maximum studied depth), finding decreasing levels with increasing depth. Young C was incorporated mainly in the coarser fractions of SOC and only 17% of the amount of incorporated C was retained by the soil.

The half-life of original C found in topsoil was 28 years, decreasing to less than 5 years in the case of POM-C and increasing to more than 400 years in the case of C associated with the mineral fraction. This confirms the heterogeneity of the substances that compose SOC and the different degrees of association with mineral particles. The estimation of parameters related to SOC dynamics – *i.e.*, necessary for its modeling – contributes to an improved understanding of the underlying processes.

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