No-tillage crop rotations, C sequestration and aspects of C saturation in a subtropical Ferralsol

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Abstract
We assessed the 17-years contribution of no-tillage crop rotations to C accumulation in bulk soil and in physical fractions up to 20-cm depth of a subtropical Ferralsol of Brazil. The wheat-soybean succession was the reference. The alfalfa system with maize at each three years showed the highest C accumulation (0.44 t/ha/yr). The bi-annual rotation of ryegrass (hay)-maize-ryegrass-soybean sequestered 0.32 t C/ha/yr. Among the two bi-annual crop-based systems of black oat-maize-wheat-soybean and hairy vetch-maize-wheat-soybean, the legume vetch-based system, although having lower C addition, sequestered more C (0.28 t/ha/yr) than the grass black oat-based system (0.16 t/ha/yr). Most of the C accumulation took place in the mineral associated fraction (71 to 95 %, in the 0-5 cm layer) compared to the particulate organic matter fraction. There was a linear relationship between annual C input and total and fraction C stocks, and no evidence for C saturation could be confirmed yet. Crop-forage systems and crop-based systems with legume represent viable strategies to increase soil organic C stocks in no-tillage soils of subtropical Brazil, in a C addition range whose contribution is not limited by an eventual C saturation.

Key Words
Crop-forage rotation, physical fractionation, alfalfa, legumes

Introduction
The conversion of conventional to no-tillage farming is far from being the ultimate possible achievement in terms of SOC accumulation in subtropical Brazilian soils. The challenge now is to develop and improve crop rotation schemes with high phytomass-C inputs that maximize the benefits of no-tillage as a strategy to promote CO_2-C sequestration and soil quality. The cultivation of forage species has shown benefits in terms of SOC accumulation (Franzluebbers *et al.,* 2001), possibly because of the high photosynthate accumulation in roots stimulated by grazing or hay cut. The cultivation of legume cover crops has also been reported to increase SOC stocks in no-tillage soils (Sisti *et al.,* 2004).

In discussions about soil as a C sink generally rises the question whether soils have or not a finite capacity of C storage, the so called C saturation, in which the steady-state C pool no longer increase regardless of increases in annual C input (Stewart *et al.,* 2007). This issue remains to be better clarified and more studies are required in order to come to consistent conclusions about it, especially for tropical and subtropical soils.

The objective of this study was to assess the long-term contribution of no-tillage crop rotations of wide range of phytomass-C addition, some including forages (hay) or winter cover crops, to the accumulation of organic C in bulk soil and in associated physical fractions of a subtropical Ferralsol; and thus infer about the capability of those rotations to promote C sequestration and to reach an eventual C saturation level.

Methods
Field experiment and soil sampling
The study was based on a long-term field experiment (17 years) conducted in the experimental station of ABC Foundation for Agricultural Assistance (Ponta Grossa-PR, Brazil), on a sandy clay Ferralsol (FAO). Climate is subtropical, classified as Cfb (Köppen). The experiment was established during the winter of 1989. Crop rotation treatments are distributed in 7.0 × 10.5-m plots, according to the randomized complete block design, with four replicates. Six crop rotation systems were selected: (i) Wheat (*Triticum aestivum* L.) in winter – soybean (*Glycine max* (L.) Merr) in summer [W-S], representing the reference or base-line system; (ii) Black oat (*Avena strigosa* Schreb.) in winter – maize (*Zea mays* L.) in summer – wheat – soybean [O-M-W-S], with black oat as winter cover crop; (iii) Hairy vetch (*Vicia villosa* Roth) in winter –

maize – wheat – soybean [V-M-W-S], with hairy vetch as legume winter cover crop; (iv) Hairy vetch – maize – black oat – soybean – wheat – soybean [V-M-O-S-W-S], with hairy vetch and black oat as cover crops; (v) Ryegrass (*Lolium multiflorum* Lam.) in winter – maize – ryegrass – soybean [R-M-R-S], where ryegrass is hayed; (vi) Alfalfa (*Medicago sativa* L.), with maize cropping each three years [A-M]. Samples from the 0-5, 5-10 and 10-20 cm layers were collected with spatula in December 2006 at two sampling points per plot. Undisturbed soil cores were collected in each layer for determination of soil bulk density.

Estimate of annual C addition, soil C analysis and physical fractionation
The mean annual C addition was estimated from historic information about grain yield or aboveground dry matter yield (including hay) (Molin, 2008). The total C addition (shoot and root) was calculated according to the procedure suggested by Bolinder *et al.* (2007), with some modifications.

Samples were analysed by dry combustion to determine total organic C (TOC). The granulometric physical fractionation was based on dispersion with hexametaphosphate solution and size-separation of particulate organic matter (POM) (>53 µm) (Cambardella and Elliott, 1992). POM was analysed by dry combustion. The C in mineral associated fraction (min-C) was determined from the difference between TOC and POM-C. The TOC stocks were corrected against the equivalent mass of soil in W-S reference system (Sisti *et al.,* 2004). The stocks of POM-C and min-C were also corrected. To calculate the annual C sequestration rate in the 0-20 cm layer of each crop rotation system, the W-S was considered as the base-line system.

Results

Total organic carbon
According to the TOC content in the whole 0-20 cm layer (Figure 1), cropping systems were separated into four main groups: A-M (52.2 t C/ha) > R-M-R-S = V-M-W-S (average of 49.7 t C/ha) > O-M-W-S = V-M-O-S-W-S (average 47.1 t C/ha) > W-S (44.7 t C/ha), demonstrating the C accumulation capacity of the two forage-based systems of alfalfa and ryegrass and of the legume-based bi-annual crop rotation with vetch. These C stocks are partly related to the annual C addition of each crop rotation, as can be viewed from the linear regression between annual C addition (shoot plus root) and the TOC stock in the 0-20 cm layer (Figure 2). The A-M system had the highest annual C addition (8.25 t C/ha) and thus showed the highest TOC stock, while the W-S system, with the lowest C addition (5.08 t C/ha), showed the lowest TOC stock.

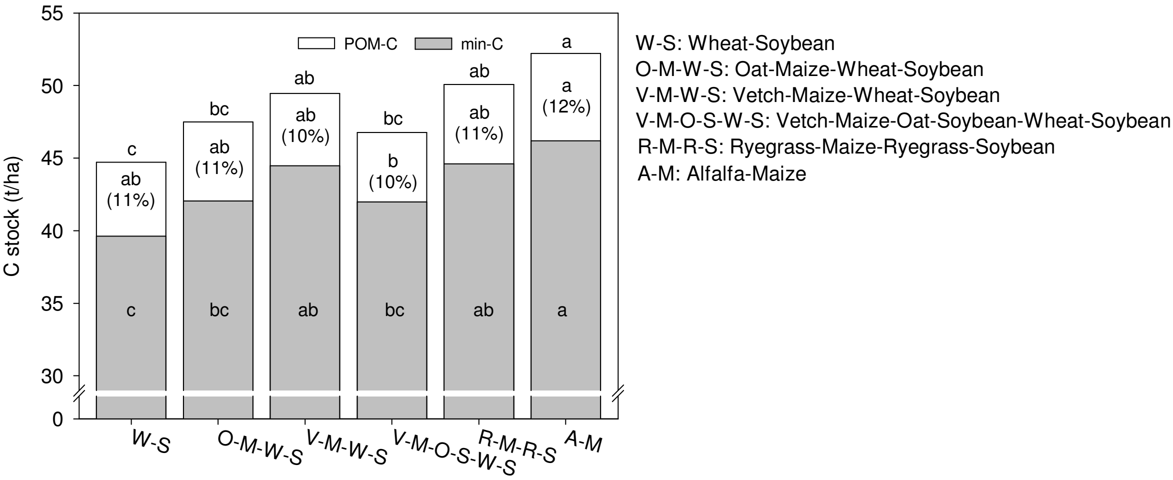

Figure 1. Organic C stock in whole soil, in particulate organic matter fraction (POM-C) and in mineral associated fraction (min-C), in the 0-20 cm layer. Numbers in parenthesis are the percentage of total C stock in the POM-C fraction. Letters compare crop rotation systems, according to Tukey's test (*P*<0.10).

Among the systems with oat or vetch winter cover crops, the V-M-W-S system contained almost 2 t/ha more C than in O-M-W-S system (Figure 1). This is intriguing by considering that V-M-W-S had 8 % less C addition (Figure 2) but 4% more C in soil, throwing some light to the hypothesis that the legume based system was more efficient in converting added C into soil organic matter C. Drinkwater *et al.* (1998) observed that quantitative differences in C addition did not account for the observed changes in SOC stocks across three crop rotations, but suggested that qualitative differences in plant and litter composition had a significant impact, and this would explain why legume-based systems retained more SOC although showing lower C addition. In line to this idea, it is important to considered the contribution that proteinaceous legume-N might have on organo-mineral interaction and thus C stabilization. Kleber *et al.* (2007) have

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recently proposed a conceptual model of organo-mineral interaction in which nitrogenous (proteinaceous) organic compounds are believed to be preferentially adsorbed on mineral surfaces; a proposal reinforced by the fact that mineral associated organic matter generally have low C:N ratio compared to the bulk soil organic matter (Diekow *et al.,* 2005). But until now all these are speculations that remains to be clarified in future studies seeking to deeply understand the role of legumes into C sequestration processes in no-tillage soils.

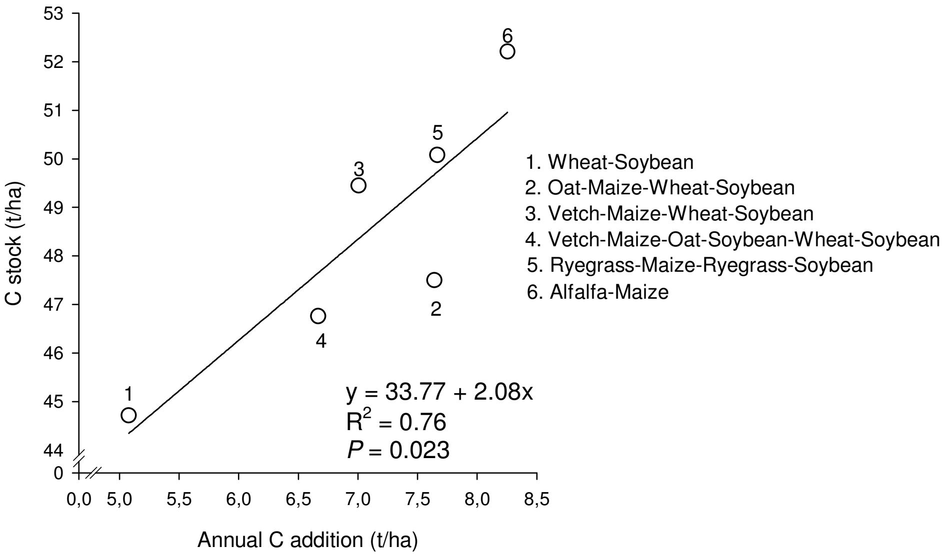

Figure 2. Relationship between annual C addition (shoot plus root) and total organic C stock (TOC) in whole soil of the 0-20 cm layer.

The tri-annual V-M-O-S-W-S rotation, although being a legume-based system, showed lower C stocks compared to the bi-annual V-M-W-S or O-M-W-S. That relies on the fact that in V-M-O-S-W-S system maize is cropped in one and soybean in two out of three summers, while in the bi-annual systems maize and soybean are cropped in the same proportion. Since soybean has a lower phytomass production compared to maize, this rendered a smaller average annual C addition and C stocks in V-M-O-S-W-S system (not shown).

Taking the W-S as the base-line system, the changes in SOC stocks varied from 4.5 % increment in V-M-O-S-W-S to 16.7 % increment in A-M system. Considering the 17 years of experimental duration, the average annual C sequestration rate ranged in the following order: V-M-O-S-W-S (0.12 t C/ha) < O-M-W-S (0.16 t C/ha) < V-M-W-S and R-M-R-S (average of 0.30 t C/ha) < A-M (0.44 t C/ha).

Organic carbon accumulation in physical fractions

The min-C represented most (89 %) of the TOC stock in the 0-20 cm layer (Figure 1). It was also the fraction that stored most of the C sequestered by each cropping system, in proportions that reached 71 to 95 % of the total C sequestration in the 0-5 cm layer and in proportions that were even higher for the two deeper layers and the whole 0-20 cm layer (not shown). This highlights the fundamental role of the mineral associated organic matter fraction in contributing to C sequestration by no-tillage crop rotations. Two major stabilization mechanisms of organic matter are involved: (i) the direct organo-mineral interaction through ligand-exchange process and (ii) the occlusion of organic matter, even of that directly associated to mineral surfaces, inside stable microaggregates. Denef *et al.* (2007) found in a previous study in two subtropical Brazilian Oxisols the mineral associated C (in microaggregates inside macroaggregates) stored most of the C accumulation obtained by adopting NT against CT and concluded that this was a very important long-term stabilized fraction for C sequestration. Although most of the accumulation ultimately occurs in the min-C fraction, one has to consider the role of POM in serving as a "bridge" fraction between the free POM fragments recently derived from plant residues and the min-C pool. Plant residue C does not immediately follows the path to become stabilized on mineral surfaces or microaggregates, but instead it first rely on the physical protection offered by macroaggregates when it is still in the POM form, so that later it can be slowly incorporated into microaggregates and mineral associated fractions instead of being promptly mineralized (Six *et al.,* 2000).

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Soil C saturation
With a range in annual C input (5.08 to 8.25 t C/ha/yr), there was a clear trend of a linear relationship between annual C input and C stocks in soil, and this was observed for the TOC (Figure 2), POM-C and min-C pools (not shown) in 0-20 cm layer. No evidence for C saturation in bulk soil could be confirmed, but it was possible to conclude that if C saturation exists for this soil, it will occur at C inputs far beyond than those obtained even in the best rotation systems practicable in agricultural systems in Southern Brazil.

Conclusion
The crop-forage rotation systems of semi-perennial alfalfa and annual ryegrass contribute more to soil organic C sequestration than rotation systems bases only in crops (wheat, soybean and maize) and winter cover crops (black oat or vetch). A significant contribution of C input comes from root and aboveground material like leaf-falls from those species. In crop-based systems, the rotation with legume winter cover crop (vetch) contributes more to soil C accumulation than the rotation with grass winter cover crop (black oat), although the later has higher C input. No clear evidence can so far be presented to explain why legumes contribute more than grasses cover crops to soil C accumulation. Most of the C accumulation due to crop rotation takes place in the mineral associated fraction. The turnover of the particulate C pool inside macroaggregates is crucial to further C accumulation in the mineral fraction. No evidence for C saturation in total as well as in physical fraction C pools is found for this subtropical Ferralsol after being subjected to 17 years of no-tillage crop rotations of wide range of phytomass-C addition. If C saturation exists for this soil, it will occur at C inputs far beyond than those evaluated in this study and practicable in agricultural systems in Southern Brazil.

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