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# Effect of Direct Seeding Mulch-Based Systems on Soil Carbon Storage and Macrofauna in Central Brazil

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# Summary

Soils represent a large carbon pool, approximately 1500 Gt, equivalent to almost three times the quantity stored in terrestrial biomass and twice the amount stored in the atmosphere. Any modification of land-use or land management can induce variations in soil carbon stocks, even in agricultural systems that are perceived to be in a steady state. These modifications also alter soil macrofauna that is known to affect soil carbon dynamics.

Direct seeding Mulch-based Cropping (DMC) systems with two crops per year without soil tillage have widely been adopted over the last 10 to 15 years in the Cerrado (central region) of Brazil. They are replacing the traditional soybean monocropping with fallow under conventional tillage (CT). The objective of this study was to examine how DMC practices affect soil organic carbon (SOC) dynamics and macrofauna (Rio Verde, Goias State). The approach was to determine soil C stocks and macrofauna in five fields under DMC aged 1, 5, 7, 11 and 13 years. In order to compare DMC systems with the native system of the region and previous land-use, a situation under native Cerrado (tree-savanna like vegetation) and a field conducted traditionally (CT) were also studied. Soil C stocks were calculated for the 0-10 and 0-40 cm soil depth and also for the first 400 kg m<sup>-2</sup> of soil to compare the same amount of soil and to suppress the potential artefact of soil compaction when sample is based on fix layer depth. Soil macrofauna was hand-sorted from soil monoliths (30 cm depth, TSBF method).

In our study, the annual rate of carbon storage was equal to *ca.* 1.6 MgC ha<sup>-1</sup>, which is in the range of values measured for DMC in different areas of Brazil, i.e., 0.4 to 1.7 MgC ha<sup>-1</sup> with the highest rates obtained in the Cerrado region.

Compared to natural vegetation, soil macrofauna in cropped systems was strongly modified. In CT, biomass and density were very low and much lower than in DMC systems. With increasing age of DMC, total macrofauna density increased and then decreased while total macrofauna biomass continuously increased due to a strong increase in Coleoptera larvae biomass. These modifications in macrofauna density and biomass are discussed with regard to soil SOC dynamics (decomposition, mineralization and physical protection).

# Key words

Brazil, Cerrado, carbon storage, earthworms, Coleoptera larvae

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# Introduction

From a global perspective soils represent an important terrestrial stock of organic carbon, holding approximately three times as much C as terrestrial vegetation and twice as much as the atmosphere (Amundson, 2001). The management and maintenance of soil carbon is therefore an integral part of the global carbon cycle. Land use change, inappropriate agricultural practices and climate change can all lead to a net release of C from soils to the atmosphere, exacerbating the problems of greenhouse gas release. The historic global loss of SOC is estimated to be 66-90 Pg (Lal, 2002). Most of this loss is the result of conversion of native ecosystems, such as forests, to agriculture. For a long time the depletion of SOC has been recognized as a major process of soil degradation in tropical environments (Nye and Greenland, 1960). Also SOC is a major determinant of soil fertility, water holding capacity and biological activity and is highly correlated to levels of above and below ground biodiversity (Carter, 2002).

The Cerrado or tropical savannah in central Brazil occupies about 23% of the national territory or about 2 million km<sup>2</sup> (Ab'Saber, 1971). While a large area of the Amazon forest was deforested to set up pastures, since the 1970s a rapid expansion of large-scale commercial agriculture took place in the Cerrado, driven by a set of government policies that were aimed at producing commodities for export (Klink et al, 1993). The major crops in the region are soybean, maize, rice and beans, traditionally grown under conventional soil tillage practices with the use of a disk harrow. However, during the last 10 to 15 years many farmers have adopted no-tillage (NT) practices with direct seeding into a mulch of plant residues (Direct seeding Mulch-based Cropping, DMC). Today, about 6 million ha of cropland are estimated to be under NT in the Cerrado. Erosion control is the major reason for switching from conventional tillage (CT) to DMC or NT. Another incentive for adoption of DMC is the reduction in labor time and costs. From the early 1990's onwards, the rate of expansion of DMC in the Cerrado region became faster than in the rest of Brazil (Bernoux et al., 2006).

In recent times much attention has been given to alternative tillage and cropping systems as a means to mitigate agricultural emissions of  $CO_2$  (Paustian et al., 1997; Follet, 2001). DMC systems represent a potential for soil C storage by increasing C inputs to the soil, reducing C losses due to soil erosion and by decreasing decomposition rates of SOC as a result of reduced mechanical soil disturbance. Furthermore, in Brazilian conditions, the possibility of an earlier seeding date with direct seeding often enables a second crop cycle with a commercial or cover crop. Consequently, more biomass is returned to the system each year. Bernoux et al (2006) reviewed the information available and reported that most studies give rates of carbon storage in the top 40 cm of the soil of 0.4 to 1.7 t C ha<sup>-1</sup> per year, with the highest rates in the Cerrado region.

As said above SOC is a major determinant of biological activity. No-tilled soils are generally characterized by higher densities of biota and especially of microorganisms. Only few studies considered the effect of DMC on soil macrofauna or the functional role of macrofauna in DMC (Marasas et al., 2001; Brown et al., 2002). Soil macrofauna is also known to control most of soil functions: organic matter dynamics, nutrient release, soil structure and associated physical properties (Lavelle & Spain, 2001; Blanchart et al., 2004). They can thus have a strong effect on SOC storage through (i) the incorporation of litter into the soil, (ii) the mixing of SOC with the mineral matrix and, (iii) the formation of stable macroaggregates that can protect SOC from mineralization.

Any modification of land-use or land management can induce variations in soil carbon stocks and soil macrofauna, even in agricultural systems that are perceived to be in a steady state. The objective of our study is to analyse the potentiality of DMC to store SOC and to increase the diversity, density and biomass of soil macrofauna in the Brazilian Cerrados. These parameters were analyzed along a chronosequence with different DMC plots aged from 1 to 13 years. These plots were compared with natural vegetation and a conventionally tilled plot.

# Material and methods

#### Study sites

The study area was located in the municipals of Rio Verde (17° 47' S, 51° 55' W) in the south-eastern part of the Goiás state on a plateau (750 m above sea level) in the centre of the Cerrado region. The climate in the region is humid tropical of savannah type. Mean annual rainfall is about 1600 mm, with a dry season from May till September. Potential evapo-transpiration is fairly constant throughout the year with a mean annual total of about 1500 mm. Mean minimum and maximum temperatures during the growing season are 17 and 27°C, respectively. The study area covered around 5000 ha of cropland that had been converted from natural savannah more than 25 years ago. Past cropping consisted of soybean mono-cropping with a fallow period and with the use of a harrow disk as tillage implement. Tillage depth was generally less than 15 cm with the major part of the surface crop residues buried into the soil. The first DMC systems were introduced in the region in 1990. DMC entails NT with the introduction of a cover crop (millet) or second commercial crop (maize or sorghum) following the main crop (soybean or maize).

Five fields were selected to represent a chronosequence of 1 to 13 years under continuous DMC aged 1, 5, 7, 11 and 13 years. In order to compare DMC systems with the native system of the region and previous land-use, a situation under native Cerrado (CER) and a field conducted conventionally (CT) were also studied. All situations had the same soil type (Geri-Gibbsic Ferralsol, FAO Soil Classification) with mean clay plus silt content ranging from 531 (CER) to 719 (DMC13) g kg<sup>-1</sup> soil and were situated on a similar topography. Soil pH determined in water was in the range from 5.5 to 5.9 for all cultivated situations, and the average was 4.8 for the situation under native vegetation.

# Measurements of soil carbon stocks

Soil carbon stocks were measured in October 2003. For each situation, soil was sampled in 18 profiles of 50 x 50 x 60 cm dimension and collected with cylindrical cores from five depths: 0-5 cm, 5-10 cm, 10-20 cm, 20-30 cm and 30-40 cm. Soil samples were air-dried and ground to pass through a <150  $\mu$ m sieve to determine the total soil organic carbon by dry combustion method using a carbon analyzer LECO<sup>®</sup> CR-2000. For each sample, bulk density was determined after weighing soil mass sampled and after the determination of the soil moisture.

Quantities of soil organic C stored in a fixed sample volume were calculated as the product of C concentration, soil thickness, and density of successive layer. But, this method do not fully account for variations in soil mass, and unambiguous comparisons among soil C storage require consideration of the masses of the soil involved. Thus, the method recommended by Ellert and Bettany (1995) and Ellert et al. (2002) was applied. The quantity of SOC for each profile was established in an "equivalent soil mass" determined for the first 400 kg m<sup>-2</sup> of soil (i.e., 4000 Mg ha<sup>-1</sup>).

#### Soil macrofauna samplings

Soil macrofauna was sampled in January 2004, in the middle of the rainy season. For each situation, nine soil monoliths ( $25 \times 25 \times 30$  cm) were excavated from each plot

after sampling litter layer and digging a trench around each monolith (modified TSBF method; Anderson and Ingram, 1993). Nine soil monoliths were located on a transect at regular intervals of ten meters between two monoliths. The first six monoliths were cut into three horizontal layers (0-10, 10-20 and 20-30 cm), and visible soil invertebrates were hand-sorted before being placed in a mixture of alcohol/formalin. In the last three monoliths, soil macrofauna were only sampled from the 0-10 cm layer. In the laboratory, invertebrates were identified at order/family level and then counted and weighed. Density (ind m<sup>-2</sup>) and biomass (g m<sup>-2</sup>) were calculated.

#### Statistical analysis

Results were analysed using the Statistica 6 Software (Statsoft France, 2001). Classical descriptive statistical analyses were carried out for the study of the different populations. Uni- and multilateral variance analyses were carried out to identify significant or non-significant effects on the variables. Mean comparisons were based on Tukey HSD (Honest Significant Difference) test.

# Results

### Soil carbon stocks

Soil bulk density varied from 0.87 to 1.28 Mg m<sup>-3</sup>, with an overall mean of 1.13 Mg m<sup>-3</sup> (Table 1). Soil bulk density showed significant differences both according to the depth and the studied land use, justifying thus the need to calculate C stocks on an equivalent soil mass. Bulk density of the 0-5 cm layer was significantly different of that of the 5-10 cm layer, and both were different from a third homogeneous group constituted by the layer below 10 cm. Homogeneous groups for the different land-use are given in Table 1.

Soil C stocks classically calculated per layer are summarized in Table 2. The same pattern among layers was observed for all situations. These values are in the range of observed C stock for this region and soil type (Bernoux et al., 2002).

Table 1. Mean soil bulk density (Mg m<sup>-3</sup>, n=18) for the different layers and situations studied. (CER = natural vegetation (Cerrado), CT = conventional tillage, DMC = Direct seeding mulch-based cropping system; the number following DMC indicates the age of the crop (in year))

Layer (cm)	CER	СТ	DMC1	DMC5	DMC8	DMC11	DMC13
0 - 5	0.87	1.03	1.15	1.13	1.11	0.97	0.96
5 - 10	1.01	1.11	1.20	1.17	1.20	1.04	1.04
10 - 20	1.19	1.14	1.22	1.18	1.28	1.11	1.11
20 - 30	1.20	1.13	1.26	1.19	1.22	1.07	1.10
30 - 40	1.23	1.11	1.24	1.17	1.26	1.05	1.09
Mean <sup>1</sup>	1.10a	1.10a	1.21b	1.17c	1.22b	1.05d	1.06d

<sup>1</sup> different letters indicate significant differences at p level of 0.05 using the Tukey HSD test.

**Table2.** Mean soil carbon stocks (Mg C ha<sup>-1</sup>, n=18) for the different layers and situations studied. (CER = natural vegetation (Cerrado), CT = conventional tillage, DMC = Direct seeding mulch-based cropping system; the number following DMC indicates the age of the crop (in year)).

Layer	CER	СТ	DMC1	DMC5	DMC8	DMC11	DMC13
0-5 cm	15.6	9.8	9.8	10.9	12.5	12.0	13.3
0-10 cm	28.1	19.9	18.9	20.7	23.1	22.7	24.9
0-20 cm	49.5	38.2	35.1	38.2	45.0	43.8	48.0
0-30 cm	67.4	53.7	50.1	53.8	61.3	61.9	66.8
0-40 cm	81.7	67.8	63.4	67.2	75.4	77.7	83.6

**Table 3.** Multiple linear regression between soil C stocks, soil clay contents and age of DMC situations. Legend : Beta = Standardized coefficient of regression or partial regression, ET = Error type

	Beta	ET of Beta	Coefficient	ET of Coefficient	t value (n =105)	p-level
Intercept			31.87	7.01	4.55	1.5 10-5
Age	0.66	0.07	1.30	0.143	9.52	< 10 <sup>-6</sup>
Clay content	0.24	0.07	0.42	0.12	3.44	8.4 10-4

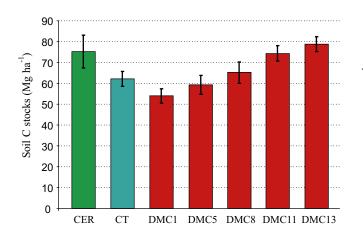


Figure 1. Soil C stocks based on soil mass (first 400 kg  $m^{-2}$  of soil) in the different situations studied (CER = natural vegetation (Cerrado), CT = conventional tillage, DMC = Direct seeding mulch-based cropping system; the number following DMC indicates the age of the crop (in year))

Soil C stocks established for the first 400 kg m<sup>-2</sup> of soil are showed in Figure 1. Carbon stocks ranged from 54.0 Mg C ha<sup>-1</sup> for DMC1 to 78.8 Mg C ha<sup>-1</sup> for DMC13 showing a regular increase. The associated standard deviation (bars in Figure 1) were in the range 3.5 - 5.1, except for the native situation (mean stock of 75.2 Mg C ha<sup>-1</sup> which showed the higher spatial variability with a standard deviation of 7.8 and minimum and maximum values of 58 and 90 Mg C ha<sup>-1</sup>.

The accumulation rate under DMC systems was calculated based on the age of the implementation expressed in months from the last seeding with conventional tillage (Figure 2). For this calculation, the field under conven-

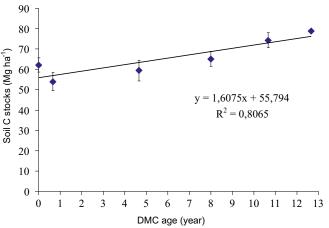
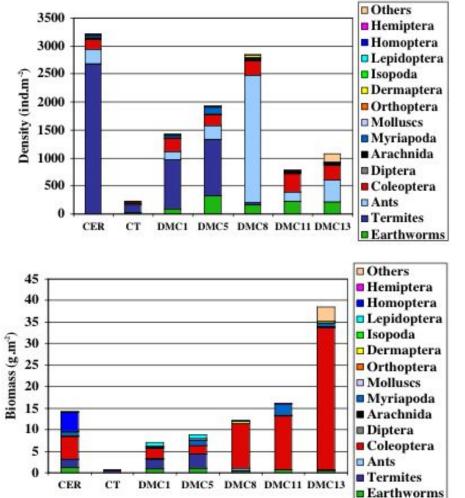


Figure 2. Carbon accumulation rate under direct seeding mulch-based cropping systems (DMC). Soil C stocks are based on soil mass (first 400 kg m<sup>-2</sup>)

tional tillage (CT) was considered as a DMC with an age of zero. The mean increase for this chronosequence was  $1.6 \text{ Mg C } ha^{-1} \text{ yr}^{-1}$ .

#### Soil macrofauna

A total number of 5449 individuals were hand-sorted from the monoliths of the seven situations: 1352, 124, 673, 902, 1409, 432, 557 in CER, CT, DMC1, DMC5, DMC8, DMC11, DMC13, respectively. The main taxa were termites (36% of collected fauna), ants (32%), Coleoptera (14%) and earthworms (11%). Mean densities were equal to 3258, 232, 1430, 1935, 2846, 793 and 1069 ind m<sup>-2</sup>, respectively in CER, CT, DMC1, DMC5, DMC8, DMC11, DMC13 (Figure 3). Termites were the most abundant invertebrates in soil



# Figure 3.

Density (ind m<sup>-2</sup>) of soil macrofauna taxa for different situations (CER = natural vegetation (Cerrado), CT = conventional tillage, DMC = Direct seeding mulch-based cropping system; the number following DMC indicates the age of the crop (in year))

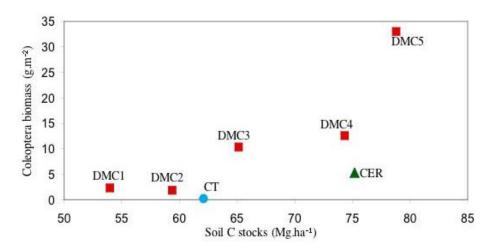
Figure 4. Biomass (g m<sup>-2</sup>) of soil macrofauna taxa for different situations (CER = natural vegetation (Cerrado), CT = conventional tillage, DMC = Direct seeding mulch-based cropping system; the number following DMC indicates the age of the crop (in year))

in CER (82% of the whole community), in CC (61%), in DMC1 (61%) and DMC5 (52%). Ants were dominant in DMC8 (80%) and in DMC13 (37%) while Coleoptera were dominant in DMC11 (41%). Earthworm densities ranged from 23 (CER) to 322 (DMC5) ind m<sup>-2</sup>. Each other taxonomic group represented less than 3% of mean density except Myriapoda (up to 6% of mean density in DMC5). Whether considering the 6 blocks sampled at 0-30 cm or the 9 blocks sampled at 0-10 cm, there were positive and significant correlations (P < 0.05) between earthworm and Coleoptera density, and between earthworm and Myriapoda density. Significant differences between situations were observed for earthworms whose density was higher in DMC5 than in CT (P < 0.05), for Coleoptera whose density was higher in DMC11 than in CT (P < 0.05), and for ants whose density was higher in DMC8 than in all other plots. Considering the mean of all DMC situations, it appeared that the total macrofauna density was significantly higher in DMC than in CT (P < 0.05).

Mean biomass was measured as 14.2, 0.8, 7.1, 8.9, 12.1, 16.2 and 38.6 g m<sup>-2</sup> in CER, CT, DMC1, DMC5, DMC8,

DMC11, DMC13, respectively (Figure 4). Homoptera larvae (4.5 g m<sup>-2</sup>) and Coleoptera (5.3 g m<sup>-2</sup>) biomasses were higher in CER. Coleoptera biomasses were higher in all DMC plots, except in DMC5 where termite biomasses were more important. In DMC11 and DMC13, Coleoptera biomass represented 73 and 85% of the whole community, respectively. In CER, 11 taxonomic groups were collected. The most diversified plot was DMC1 with 12 groups; the less diversity occurred in CT with only 8 taxonomic groups (Molluscs, Dermaptera, Isopoda, Lepidoptera, Homoptera and Hemiptera were absent from this plot).

Whether considering the 6 blocks sampled at 0-30 cm or the 9 blocks sampled at 0-10 cm, there were positive and significant correlations (P < 0.05) between Dermaptera and Coleoptera biomass, and between Dermaptera and ant biomass. Significant differences between situations were observed only for ants whose biomass was higher in DMC8 than in than in all other plots (P < 0.05). Considering the mean of all DMC situations, it appeared that the total macrofauna biomass was significantly higher in DMC than in CT (P < 0.05). We also found a positive correla-



#### Figure 5.

Relationship between Coleoptera biomass (g m<sup>-2</sup>) and soil C stock (Mg ha<sup>-1</sup>) for different situations (CER = natural vegetation (Cerrado), CT = conventional tillage, DMC = Direct seeding mulch-based cropping system; the number following DMC indicates the age of the crop (in year))

tion and significant correlation between Coleoptera biomass and soil C stock, when considering DMC situations only (Figure 5).

# Discussion

The accumulation rates of 1.6 Mg ha<sup>-1</sup>yr<sup>-1</sup> observed for this situation in Brazil is elevated while in the range of reported values. Bernoux et al. (2006) reviewed the C accumulation rates observed for Brazil. These authors reported for the Cerrado region carbon accumulation rates varying from 0.4 to 1.7 Mg C ha<sup>-1</sup> for the 0-40 cm layer. Soil C content and thus accumulation rates may depend on other factors that the age of DMC situations. A strong influence of the soil texture over C storage capacity of the soils is commonly reported. Therefore a multiple regression analysis was performed only for the DMC situations (CT field was considered as a DMC situation with an age of zero) considering the C stock for the first 400 kg m<sup>-2</sup> of soil as dependant variable and clay content and age as independent or predictor variables. Both clay content and age of the DMC situations had significantly partial correlation with the C stocks (Table 3). It is verified that the clay content influences the C stocks. Nevertheless the age effect is predominant with a partial regression coefficient of 0.66, i.e., nearly 44% of the original variability can be explained by the age.

In conventionally tilled situations soil macrofauna is characterized by low densities and biomasses if compared to natural situations (14 times less density and 18 times less biomass). This result is consistent with results generally obtained from literature for earthworms and arthropods (House & Parmelee, 1985; Fragoso et al., 1999; Marasas et al., 2001; Brown et al., 2003; Benito et al., 2004). Soil disturbance has generally a negative effect on invertebrate populations due to direct damage by the equipment, indirectly through loss of organic matter, and through changes in soil structure and water regime (Chan et al., 2001). In our study the installation of no-tillage systems induced a significant increase in total biomass and density of the whole soil fauna and of particular taxa. Earthworm density was significantly higher in DMC than in CT but it did not increase with time at the difference described by Brown et al. (2002) in South Brazil. These authors showed a significant increase in earthworm density with the age of no-tilled plots. In our experiments, although most of taxa presented higher density and biomass in DMC than in CT, there was no variations with time, except for the biomass of Coleoptera, which was significantly correlated with soil C stocks and the age of DMC plots (Figure 5). This increase in the biomass of Coleoptera is not due to an increase in their numbers but to an increase in their individual weight (it increases from 6.5 mg in CT to 132 mg in DMC13). The disappearance of termites after some years of no-tillage (from DMC8 afterwards) was in our knowledge never observed.

As reported from other studies, the biomass, density and diversity of soil macrofauna is greatly improved in DMC systems compared to conventionally tilled systems. This can strongly modify the soil functioning and especially soil carbon storage. The only significant differences between DMC (as a whole) and CT systems were measured for earthworms and Coleoptera density and for Coleoptera biomass. In our study sites, as well as in other sites from Brazil (Brown et al., 2001, unpub. data), Coleoptera are mainly scarab beetle larvae (white grubs). It appears that some white grub species can be rhizophagous (pests like Phyllophaga), some species can be beneficial saprophagous or coprophagous (Cyclocephala) and some species can be intermediate (Diloboderus). In our study sites, the high abundance of white grubs with no impact on root damage and plant production suggests that most of white grubs are saprophagous. These animals ingest soil organic matter (especially residues) and mix it with soil mineral particles, egest stable casts and create burrows. This can lead to the creation of hot-spots of soil enrichment in the upper 20-30 cm of soil, with significant increase in P and organic matter. White grubs should be considered among the soil engineers since their activities are similar to activities of earthworms (Lavelle et al., 1997). Earthworms are known to affect the dynamics of organic matter in the long term through the physical protection of organic matter in their casts (Martin, 1991).

## Conclusion

Compared to conventional systems, direct seeding mulch-based systems provide an ideal environment for the re-establishment of soil engineer (earthworms, white grubs), litter engineer (termites, ants, millipedes) and predator (spiders, centipedes) populations, thus leading to a higher biological activity and regulation in DMC systems. This high activity associated with the high abundance of soil and litter engineers, the presence of abundant crop residues and the absence of mechanical tillage can explain the increase in soil C stocks measured in DMC (or no-tilled) systems. More research will be necessary to relate soil C storage with the activity of soil macrofauna and especially of white grubs whose beneficial activity needs to be confirmed or refuted.

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