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SOIL SCIENCE

Isotopic abundance of ^{13}C and contribution of eucalyptus biomass to soil organic matter conversion

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ABSTRACT: *It has become possible to evaluate the conversion of soil organic matter (SOM) in pastures and arboreal crops due to the difference between the photosynthetic cycles of Eucalyptus (C3) and most grasses (C4). The auto analyzer method coupled to the IRMS (Isotope Ratio Mass Spectrometer) in the present study evaluated the ^{13}C content in soil profiles of Eucalyptus plantations of different ages (2, 10 and 21 years), in natural regeneration areas and natural grazing fields, and estimated the SOM conversion of each crop type of. The initial management of all sampled areas was natural pasture. The following profile layers were evaluated: 0-5, 5-10, 10-20, 20-30, 30-40, 40-50, 50-70 and 70-90cm, and the contribution of Eucalyptus biomass over the years of farming was estimated in the SOM conversion process. After 2 years of planting Eucalyptus, the beginning of pasture carbon conversion process occurred in the surface layer (0-5cm). Ten years after planting, the process of converting organic matter by arboreal crops reached the layers up to 20cm. After 21 years of planting and in natural regeneration areas, the entire profile has already been changed by planting Eucalyptus and native tree species.*
Key words: *cycle C3, cycle C4,* ***Eucalyptus grandis****.*

Abundância isotópica do ^{13}C e contribuição da biomassa de eucalipto na conversão da matéria orgânica do solo

RESUMO: *A avaliação da conversão da matéria orgânica do solo (MOS) entre pastagens e cultivos arbóreos, tornou-se possível graças a diferença entre os ciclos fotossintéticos do eucalipto (C3) e da maioria das gramíneas (C4). Com o autoanalisador acoplado ao IRMS (isotope ratio mass spectrometer) e a avaliação da abundância isotópica de ^{13}C, o presente estudo estimou a conversão da MOS em perfis do solo sob plantio de eucaliptos de diferentes idades (2, 10 e 21 anos), em área de regeneração natural e em campo de pastagem natural. O manejo inicial de todas as áreas amostradas era de pastagem natural. Foram avaliadas as seguintes camadas do perfil: 0-5, 5-10, 10-20, 20-30, 30-40, 40-50, 50-70 e 70-90cm. Assim como estimada a contribuição proveniente da biomassa de eucaliptos ao longo dos anos de cultivo no processo de conversão da MOS. Após 10 anos de plantio, o processo de conversão da matéria orgânica nos cultivos arbóreos atingiu as camadas até 20cm. Após 21 anos de plantio e na área de regeneração natural, todo o perfil já sofreu alteração pelo plantio de eucaliptos e espécies arbóreas nativas.*
Palavras-chave: *ciclo C3, ciclo C4,* ***Eucalyptus grandis****.*

INTRODUCTION

In recent decades, Eucalyptus (***Eucalyptus grandis***) began to be planted in many native grassland areas in the south of Rio Grande do Sul State. Eucalyptus plantation in these areas has been criticized for several different reasons, but dryness and nutritional soil impoverishment are the most often mentioned. Nutritional soil impoverishment is the decline in soil organic matter (SOM) content in soil over years of planting and the excessive removal of nutrients with little contribution to them from Eucalyptus cultivation (VIANA, 2004). Many studies have been conducted regarding carbon behavior in areas where Eucalyptus trees are planted and which once were native forests and native grasslands. Reports indicated an organic carbon decline in the initial years after the introduction of Eucalyptus forests. Carbon content decline in forest soils occurred mainly in the first years of cultivation and it is related to land use change. This behavior was observed in the first 0.40m depth (CASTRO FILHO et al., 1991; NILSSON & SCHOPFHAUSER, 1995). After 20-40 years of cultivation, the SOM content may return almost to the starting point, depending on site management (SPECHT & WEST, 2003). In some places, after several Eucalyptus cycles, SOM levels recovered, but to intermediate levels between degraded pastures and native forest soils (LEITE et al., 2010). It is critical to know SOM conversion in those areas for proper

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planning of management practices and environmental impacts of crops in different environments where the Eucalyptus is grown.

Understanding the SOM dynamics in areas converted from pasture to arboreal crops, such as Eucalyptus for example, is easier if the management of the types of vegetation in the area is known, and with the later determination of the natural isotopic abundance of carbon in the area. The variation in the natural abundance of ^{13}C has been used to assess the impact of different land uses and managements in SOM fractions in various parts of Brazil (SA et al., 2001; SISTI et al., 2004). In nature, approximately 98.89% of all carbon are ^{12}C and 1.11% is ^{13}C and the value of these two isotopes may vary as a result of fractionation during physical, chemical and biological natural processes (BOUTTON, 1991a). The natural abundance of C allows characterizing SOM and is related to the photosynthetic pathway of C isotope fixation of the predominant vegetation. Type C3 plants (e.g. eucalyptus) discriminate part of ^{13}C compared to ^{12}C, resulting in lower soil levels of ^{13}C; and plants of the C4 type (most grasses) fix ^{13}C in higher proportions than type C3, which creates differences in the natural abundance of ^{13}C (GOLCHIN et al, 1995; MARTINELLI et al., 2009). To estimate the relative SOM contribution of each vegetation type, it is possible to apply the isotope mixture model (MARTINELLI et al., 2009), according to equation (1):

$$(C4\%) = \left(\frac{\delta^{13}\text{soil} - \delta^{13}\text{eucalyptus}}{\delta^{13}\text{pasture} - \delta^{13}\text{eucalyptus}}\right) \times 100$$

Where: C4%: percentage of SOM stemming from the C4 vegetation in the soil; $\delta^{13}C$: delta value provided by the IRMS equipment for the evaluated soil, C3 plant tissue (eucalyptus) and C4 plant tissue (pasture grass).

This model is used when there are two distinct sources of isotopes and their relative contributions can be estimated by this equation, which is simply a mass balance combined with an isotopic balance expression (MARTINELLI et al., 2009). It can be used in studies where there is an exchange of C3 species for C4 or vice versa, as it allows verifying the impact of these different area managements (MARTINELLI et al, 2009; COSTA et al, 2009; PEGORARO et al 2011). This study aimed to quantify the natural abundance of ^{13}C in Eucalyptus plantations of different ages, in a natural regeneration area, and in an adjacent pasture under the same conditions for planting and, from that, to estimate the SOM conversion between native pasture and areas planted with Eucalyptus and native species.

MATERIALS AND METHODS

The study area is located in the municipality of São João do Polesine, in the central region of Rio Grande do Sul, at the transition between the Plateau and the Central Depression regions. The climate presents Cfa, according to the Köppen classification. The annual average minimum temperature is from about 14°C and average annual maximum of 25°C. The annual rainfall average is 1700mm (BURIOL et al., 1979).

The study was conducted in Eucalyptus forests aged in 2(E1), 10(E2) and 21 (E3) year-old forests, native pasture (NP) and natural regeneration area (NR). The approximate area location for sampling in E1 is 29°39’23 (latitude) and 53°25’28 (longitude) in E2 is 29°39’16 (latitude) and 53°26’07 (longitude), E3 is 29°39’20 (latitude) and 53°26’10 (longitude), NP is 29°39’25 (latitude) and 53°26’06 (longitude) and NR is 29°39’28 (latitude) and 53°26’10 (longitude). Sites had no prior preparation or subsequent application of chemical lime and/or fertilizer, only controlling for ants. The E3 area was in its second planting cycle and E2 and E1 in the first cycle. Species planted in the area was ***Eucalyptus grandis***. The tree which was cut was about 10 years old usually used to make fence posts. The natural regeneration area has contained the expected species for this region, such as ***Luehea grandiflora*** (açoita cavalo) and ***Cinnamomum zeylanicum*** (caneleira), since for 21 years no kind of management was performed in the area. Native pasture (CN) was dominated by typical grasses of the Pampa biome region, ***Paspalum notatum*** (capim forquilha) and ***Axonopus jesuiticus*** (missionary grass), and the area did not undergo any mechanical interferences and/or fertilizations of any kind, and was used for extensive cattle pastures.

The native pasture (NP) was dominated by typical grasses of the Pampa biome region such as ***Paspalum notatum*** and ***Axonopus jesuiticus*** and the area did not suffer mechanical treatment and/or fertilization, and has been used for extensive grazing of cattle. This area was not managed in the last 21 years.

At each location (E1, E2, E3, NP and NR) three trenches were opened approximately 20m apart, measuring 0.6m long x 0.4m wide and 0.5m deep. Samplings at depths greater than 50cm were done with a Dutch design auger. The soil samples were taken at depths: 0-0.05, 0.05-0.1, 0.1-0.2, 0.2-0.3, 0.3-0.4, 0.4-0.5, 0.5-0.7 and 0.7-0.9m. Sampling in Eucalyptus forests (E1, E2 and E3) was performed between the planting lines and on a slope with the same condition. At each depth of each trench three sub-samples were collected to compose a sample and then were placed in plastic bags. According to the FAO soil classification (WRB, 2006), the soil from NP belongs to the Haplic Cambisols class, the soils from NR, E2 and E1 are classified as Regosols and the soil from E3 area as Arenosols. In total, 15 trenches with eight depths of each trench were excavated for 120 soil samples.

In NR, leaves of various species present in the area were collected. In NP, three sampling points measuring 50cm x 50cm were delimited for the subsequent cutting of the aerial part of grasses. . Once they had been sampled, roots and macroscopic residues of soil samples were removed manually after being dried at 60°C to constant weight, ground and sieved to 1mm.

Aliquots used for analysis were previously macerated in an agate stone mortar and pestle ensuring that they were uniformly ground. Plant tissue samples were stored in paper bags, dried at constant weight at a temperature below 55°C and ground in a Wiley mill. In an auto analyzer (Flash 1112 Advantage model) coupled to the IRMS (isotope ratio mass spectrometer) (Delta V Advantage IRMS model), both from Thermo Scientific, between 1 and 2mg of sample (soil and plant tissue) were used for $\delta^{13}C$ and C content determination. The values of isotopic abundance of carbon ($\delta^{13}C$) in soil and plant tissue samples were determined and the SOM conversion based on vegetation estimated according to equation 1.

Results were obtained from the analysis and characterization via auto analyzer. Sets whose values were greater or equal to 95% ($P<0.05$) for each sample done in triplicate were considered accurate. The comparison between the soil layers, within the same profile and same depth for each site, was performed by a T-test (Student) with $P<0.05$, or 95% confidence. The input factors were $\delta^{13}C$ value and carbon content representing true triplicates (n=3). The Excel software was generally used.

RESULTS AND DISCUSSION

Eucalyptus leaves collected at the site presented a value of -30.3‰ abundance of ^{13}C, while the plant tissue of the grasses in this area presented a value of -14.98‰ and the mean of the values of 13C of the leaves in the NR area were -32.2‰. Values experimentally found were consistent with what is reported in the literature wherein photosynthetic cycle C3 plant tissues exhibit lower isotopic values ($\delta^{13}C$), ranging from -20.0 to -32.0‰, with an average -27.0‰, while the $\delta^{13}C$ values of C4 species are larger, ranging from -9.0‰ to -17.0‰, averaging -13.0‰. (BOUTTON, 1991b; SILVA et al., 2013). In soil, the greater $\delta^{13}C$ values reported in this study were found in the surface layer (0-5cm) NP (-19.19‰) (Table 1), which indicated the largest contribution coming from C-C4 plants (in this case predominantly grasses) in SOM (SMITH & EPSTEIN, 1971) and the remaining layers showed values close to each other, or similar isotopic values along the profile.

From the isotopic dilution calculation, it was possible to follow the SOM carbon conversion process among the types of management studies (Table 1). Values obtained with the isotopic dilution calculation were limited to 100% considering a margin of error that may overstate conversion values and undermine the interpretations. During the plant material decomposition process enrichment of 1 to 2‰ ^{13}C occurred in SOM due to loss of ^{12}C atoms of the vegetation and soil (MARTINELLI et al., 2009). It is also necessary to evaluate the effects of the ^{13}C and ^{14}C concentration decline which occurred mainly in the atmosphere in the last century. This took place at the start of the industrial era, since the emission rate of CO_2 increased at 4% per year, which caused a dilution effect of these isotopes, the so-called Suess's effect (SUESS, 1955; EKDAL & KEELING, 1973). Isotopic decrease in the ^{13}C over the soil profile can reach up to 3‰ due to this phenomenon (MARTINELLI et al., 2009).

In the 2 - year old Eucalyptus (E1) only the surface layer (0-5cm) showed a significant decrease in ^{13}C values regarding the subsurface layer (Table 2). Thus it showed an amount of carbon already derived from planting Eucalyptus (29.9%), while the other layers continued to have 100% of the carbon from pasture. Ten years after planting (E2) there was significant decrease in the amount of 13C in the profile between layers 5-10cm, 10-20cm and 20-30cm, and this was reflected in the carbon values derived from eucalyptus. In this profile up to 20cm deep there was interference from the Eucalyptus management in carbon content and at greater depths it was observed that the isotopes were maintained similarly to the natural pasture site (NP). Comparing the results in E1 and E2, it is possible to see the evolution of the carbon cycling process covering the profile, as in E2 there is differentiation in ^{13}C values between the layers 0-5 and 5-10cm (Table 2). A 10-20cm layer already has 8.2% carbon from Eucalyptus possibly through their roots, which denoted the beginning of pasture carbon conversion process in this layer.

Carbon values that have already originated from planting eucalyptus in the organic matter of this area contradict studies that say that the rate of carbon substitution at places where eucalyptus was implemented is low, with a contribution of only 5%

Table 1 - Isotopic Abundance ^{13}C and % carbon from different management areas.

Layers (cm)	Isotopic Abundance $\delta^{13}C$(‰)	Carbon from NP (%)	Carbon from Euc (%)	Carbonfrom NR (%)
---Native Pasture (NP)---				
0-5	-19.19	100	0	
5-10	-21.59	100	0	
10-20	-21.74	100	0	
20-30	-21.67	100	0	
30-40	-21.57	100	0	
40-50	-21.56	100	0	
50-70	-20.68	100	0	
70-90	-21.50	100	0	
---Eucalyptus aged 02 (E1)---				
0-5	-22.59	70.1	29.9	
5-10	-21.33	100*	0	
10-20	-20.69	100*	0	
20-30	-19.68	100*	0	
30-40	-19.68	100*	0	
40-50	-19.81	100*	0	
50-70	-20.26	100*	0	
70-90	-18.80	100*	0	
---Eucalyptus aged 10 (E2)---				
0-5	-26.58	34.2	65.8	
5-10	-24.15	71.2	28.8	
10-20	-22.44	91.8	8.2	
20-30	-20.47	100*	0	
30-40	-20.49	100*	0	
40-50	-20.10	100*	0	
50-70	-20.21	100*	0	
70-90	-20.96	100*	0	
---Eucalyptus aged 21 (E3)---				
0-5	-29.30	9.6	90.4	
5-10	-25.03	62.0	38.0	
10-20	-24.20	71.7	28.3	
20-30	-24.80	64.6	35.4	
30-40	-24.47	67.0	33.0	
40-50	-24.75	63.8	36.2	
50-70	-24.87	56.8	43.2	
70-90	-24.32	69.8	30.2	
---Natural Regeneration (NR)---				
0-5	-28.01	21.0		79.0
5-10	-26.65	42.1		57.9
10-20	-26.33	46.5		53.5
20-30	-25.61	54.7		45.3
30-40	-25.32	57.2		42.8
40-50	-25.18	59.2		40.8
50-70	-24.92	56.3		43.7
70-90	-24.05	71.3		28.7

*Values whose isotopic dilution calculation resulted in values greater than 100%.

of carbon derived from eucalyptus in the 0-20cm layer after 20 years of cultivation (PULRLNIK et at 2009) and an annual mean rate of substitution in the d0-5cm layer of 0.43% in a 32-year planting (LIMA et al.; 2006), both in clayey feral sols. This low carbon exchange rate is attributed to lack of soil preparation in the study area, mild weather conditions (dry winter and altitude), very clayey soil texture and plenty of iron

Table 2 - Probability values (p) t test (Student) obtained in the comparisons of $\delta^{13}C$ values between pairs of adjacent layers of the soil profile at a same location and between pairs from different locations in the same layer of the soil profile.

---Layers (cm)---								
Places	0-5/5-10	5-10/10-20	10-20/20-30	20-30/30-40	30-40/40-50	40-50/50-70	50-70/70-90	
NP	0.0450*	0.8318	0.8113	0.8356	0.9743	0.0747	0.5462	
E1	0.0191*	0.0516	0.0400*	0.7719	0.8793	0.3419	0.4817	
E2	0.1061	0.0501*	0.0216*	0.9440	0.4592	0.6908	0.4781	
E3	0.0086*	0.0210*	0.3711	0.4909	0.4296	0.4948	0.3286	
NR	0.0726	0.0734	0.0403*	0.1248	0.7400	0.5566	0.0726	
---Depths (cm)---								
Places	0-5	5-10	10-20	20-30	30-40	40-50	50-70	70-90
NP – E1	0.0729	0.8291	0.2895	0.1245	0.0321	0.0681	0.2536	0.0678
E1 – E2	0.0009**	0.0438**	0.1322	0.1844	0.1263	0.5570	0.6885	0.1246
E2 – E3	0.0245**	0.3088	0.0884	0.0118**	0.0047	0.0054**	0.0033**	0.0698
E3 – NR	0.0360**	0.2432	0.1136	0.2223	0.1633	0.5773	0.7905	0.7680
NP – E3	0.0047**	0.0969	0.0718	0.0674	0.0032**	0.0066**	0.0087**	0.1174
NP – NR	0.0026**	0.0066**	0.0010**	0.0104**	0.0078**	0.0573**	0.0184**	0.017**
E1 – E3	0.0028**	0.0094**	0.0072**	0.0245**	0.0119**	0.0148**	0.0296**	0.0034**

*P values less than 0.05 indicate significant differences in the respective pair of layers. **P values less than 0.05 indicate significant differences in the respective pair of locations. NP - native pasture; E1 - eucalyptus aged 02; E2 - eucalyptus aged 10; E3 - eucalyptus aged 21; NR - natural regeneration area.

and aluminum oxides (PULROLNIK et al., 2009), and compounds which can form coordination complexes with the SOM in order to stabilize it (ZINN et al., 2005). In this study, it can be inferred that the carbon conversion rate between grass land and Eucalyptus may be higher due to different soil and edaphoclimatic conditions as described earlier, as higher average rainfall, low aluminum content, sandy soil types and less protection from SOM. The difference between the thicknesses of the evaluated layers (0-5 and 0-10cm) can also facilitate the overestimation of grassland carbon that was converted.

However, it is necessary to take into account the variability of ^{13}C values, a fact that should lead to a cautious interpretation of these data. The differences between the absolute values ^{13}C in the surface layers E1 and NP are small and not significant. Thus, the calculation of isotopic dilution may give us overestimated carbon conversion values in the area. In this way the value of carbon reported in the surface layer from planting eucalyptus as E1 (29.9%) may not be considered an absolute quantitative evaluation, although it can be used to indicate the advance of the cycling process of this element in the surface layer.

After 21 years of cultivation (E3), area E3 presented a differentiation of $\delta^{13}C$ values between the layers up to 20cm, which qualifies a sharp reduction of natural isotoposes of carbon, and at deeper layers these values were stabilized (-24‰). According to the isotope dilution model it can be inferred that all the organic matter in the profile has already undergone interference from Eucalyptus planting, probably through carbon cycling coming from the plant root system. The 21-year plantation areas (E3) and pasture area (NP) showed constant values of ^{13}C from 10cm and 5cm depth, about -24‰ and -21‰ respectively. The ^{13}C content in the NP area possibly had been stable for 21 years and then eucalyptus was planted and the isotopic composition of soil again attained a situation of equilibrium. As the evaluated areas are of different soil classes, it is important to note that this may end up influencing the dynamics of the distribution and occurrence of isotopes between the layers of the assessed profile.

In NR, ^{13}C values reported in the evaluated profile layers ranged from -28‰ to -24‰. There was no significant variation in these values between the layers and it shows that there is possibly a tendency for the stabilization of the abundance of isotopes in the area. All the profile layers in the E3 and NR already have been influenced by management and carbon content derived from these plants can cause a significant increase in SOM in these areas in a few decades. This behavior was confirmed in a Brazilian Atlantic Forest area that, after 35 years of natural regeneration, presented almost 100% of C from tree species and a typical C3 cycle. This change occurred in the entire profile and can be attributed to the C input derived from the roots of woody plants, even at depths greater than 40cm, where it is expected that the dynamics of C from the surface was slower not contributing significantly to the increase of SOM (COUTINHO et al., 2010).

It is also important to note that the total carbon exchange cannot occur completely and take years due to the presence of organic carbon material belonging to stabilized and protected pasture. In general, most of the humified carbon in SOM is in most recalcitrant forms and in greater interaction with the soil mineral colloidal matrix (STEVENSON, 1994), a mechanism that hinders the access of microorganisms and their enzymes to the organic substrate (BALESDENT et al., 2000).

The difference in isotopic abundance in the surface layer between the E1, E2 and E3 was significant (Table 2). After 2 years of Eucalyptus (E1) it has not been possible to see significant differences in the natural values of 13C abundance between the NP sampled in any of the layers. This behavior is confirmed by BALIEIRO et al. (2008), wherein replacement of the ***Panicum maximum*** pasture (capim mombaça) by ***Eucalyptus grandis*** did not significantly alter the isotopic composition of SOM relative to pasture in the surface layers and also COUTINHO et al. (2010) who reported no significant differences in carbon isotope composition after 4 years of planting Eucalyptus. This fact is evidenced in this study by $\delta^{13}C$ values that, in the surface layer of E1, tended to decrease (-22.59‰) and the consequent start of carbon conversion in the area and the rest of the profile remained at values that do not differ from those f reported in pasture area.

The value of natural isotopic abundance between E3 and NR showed no difference, except for the surface layer, which confirms that the profile of both has already suffered interference from the planting of tree crops with an entire C3 cycle. These areas compared showed that management differences are likely to interfere with the conversion of C between pasture and arboreal crops, contributing to carbon maintenance. The areas evaluated in this study had some characteristics that may limit the implementation of other agricultural crops that required better physical and chemical soil properties, such as low fertility, wide variation in slope and high sand content in some profiles. However, assessing the SOM conversion data we can infer that this type of tree farming can bring benefits to the soil such as the maintenance of carbon content and soil protection of steeper slopes, reducing erosions occurrence of erosions.

CONCLUSION

The ^{13}C isotopic abundance indicates that eucalyptus stands contributed to higher SOM-up values in the first 20cm layer, after 10 years of cultivation, and it also has an effect throughout the total soil profile after 21 years of cultivation, similarly to what happened in natural regeneration areas with tree species.

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REFERENCES

BALESDENT, J et al. Relationship of soil organic matter dynamics to physical protection and tillage. **Soil and Tillage Research**, v.53, p.230, 2000. Available from: <http://www.sciencedirect.com/science/article/pii/S0167198799001075>. Accessed: jun. 10, 2015. doi: 10.1016/S0167-1987(99)00107-5.

BALIEIRO, F. C. et al. Soil carbon and nitrogen in pasture soil reforested with eucalyptus and guachape. **Revista Brasileira de Ciência do Solo**, v.32, 1260p, 2008. Available from: <http://www.scielo.br/pdf/rbcs/v32n3/a33v32n3.pdf>. Accessed: jun. 05, 2015. doi: 10.1590/S0100-06832008000300033.

BOUTTON, T. W. 13C/12C fractionation and its utility in terrestrial plant studies. In: COLEMAN, D. C.; FRY, B. **Carbon isotope techniques**. San Diego: Academic, p.200, 1991.

BOUTTON, T. W. Stable carbon isotopes ratios of natural materials. In: COLEMAN, D.C.; FRY, B. (Eds.). **Carbon isotopes techniques**. New York: Academic, p.200, 1991.

BURIOL, G. A. et al. Monthly and yearly charts of average temperatures, the maximum average temperatures and average minimum temperature of the Rio Grande do Sul state. **Revista do Centro de Ciências Rurais**, v.9, supl, p.1-53, 1979.

CASTRO FILHO, C. et al. Tillage methods and soil and water conservation in southern Brazil. **Soil Tillage**, v.20, p.283, 1991. Available from: <http://www.sciencedirect.com/science/article/pii/016719879190043W>. Accessed: jun. 03, 2015. doi: 10.1016/0167-1987(91)90043-W.

COSTA, O. V. et al. Estoque de carbono do solo sob pastagem em área de tabuleiro costeiro no sul da Bahia. **Revista Brasileira de Ciência do Solo**, v.33, p.1145, 2009. Available from: <http://www.scielo.br/scielo.php?pid=S010006832009000500007&script=sci_

abstract&tlng=pt>.Accessed: jun. 03, 2015. doi: 10.1590/S0100-06832009000500007.

COUTINHO, R. P. et al. Estoque de carbono e nitrogênio e emissão de N2O em diferentes usos do solo na Mata Atlântica. **Pesquisa Agropecuária Brasileira**, v.45, n.2, p.203, 2010. Available from: <http://www.scielo.br/scielo.php?script=sci_arttext&pid=S0100-204X2010000200011>. Accessed: jun. 03, 2015. doi: 10.1590/S0100-204X2010000200011.

EKDAL, C. A.; KEELING, C. D. Atmospheric carbon dioxide and radiocarbon in the natural carbon cycle I: Quantitative deductions from records at Mauna Loa Observatory and at the South Pole. In: WOODWELL, G.M; PECAN, E.V. (Eds.). **CO^2 production from fossil fuel combustion and kilning of limestone**. New York: Brookhaven National Lab. Carbon and the biosphere. Carbon and the Biosphere conference Upton, p.85, 1973.

GOLCHIN, A. et al. Structural and dynamic properties of soil organic matter as reflected by^{13}C natural abundance, pyrolysis mass spectrometry and solid-state ^{13}C NMR spectroscopy in density fractions of Oxisol under forest and pasture. **Journal Soil Research**, v.33, 76p, 1995. Available from: <http://www.publish.csiro.au/sr/SR9950059>. Accessed: jun. 03, 2015. doi: 10.1071/SR9950059.

LEITE, F. P. et al. Alterations of soil chemical properties by eucalyptus cultivation in five regions in the Rio Doce Valley. **Revista Brasileira de Ciência do Solo**, v.34, p.831, 2010. Available from: <www.scielo.br/scielo.php?script=sci_arttext&pid=S0100-06832010000300024>. Accessed: jun. 03, 2015. doi: 10.1590/S0100-06832010000300024.

LIMA, A. M. N. et al. Soil organic carbon dynamics following afforestation of degraded pastures with eucalyptus in Southeastern Brazil. **Forest Ecology and Management**, v.235, p.231, 2006. Available from: <http://www.sciencedirect.com/science/article/pii/S0378112706008449>. Accessed: jun. 03, 2015. doi: 10.1016/j.foreco.2006.08.331.

MARTINELLI, L. A. et al. **Desvendando questões ambientais com isótopos estáveis**. São Paulo: Oficina de Textos, 2009. p.144.

NILSSON, S.; SCHOPFHAUSER, W. The carbon-sequestration potential of a global afforestation program. **Climate Change**, v.30, n.3, p.293, 1995. Available from: <http://link.springer.com/article/10.1007/BF01091928>. Accessed: mai. 06, 2015. doi: 10.1007/BF01091928.

PEGORARO, R. F. et al. Estoques de carbono e nitrogênio nas frações da matéria orgânica em Argissolo sob eucalipto e pastagem. **Ciência Florestal**, v.21, n.2, p.354, 2011. Available from: <https://periodicos.ufsm.br/cienciaflorestal/article/view/3230>. Accessed: mai. 06, 2015. doi: 10.5902/198050983230.

PULROLNIK, K. et al. Estoques de carbono e N em frações lábeis e estáveis da matéria orgânica de solos sob eucalipto, pastagem e cerrado no Vale do Jequitinhonha - MG. **Revista Brasileira de Ciência do Solo**, v.33, p.1136, 2009. Available from: <http://www.scielo.br/scielo.php?script=sci_arttext&pid=S0100-06832009000500006>. Accessed: mai. 06, 2015. doi: 10.1590/S0100-06832009000500006.

SÁ, J. C. M. et al. Organic matter dynamics and carbon sequestration rates for a tillage chronosequence in a Brazilian Oxisol. **Soil Science Society of America Journal**, v.65, p.1499, 2001. Available from: <https://dl.sciencesocieties.org/publications/sssaj/abstracts/65/5/1486?access=0&view=pdf>. Accessed: mai. 06, 2015. doi: 10.2136/sssaj2001.6551486x.

SILVA, V. E. et al. Composição lignocelulósica e isótopica da vegetação e da matéria orgânica do solo de uma turfeira tropical: I - composição florística, fitomassa e acúmulo de carbono. **Revista Brasileira Ciência Solo**, v.37, n.1, 133p, 2013. Available from: <http://www.scielo.br/scielo.php?script=sci_arttext&pid=S0100-06832013000100013>. Accessed: mai. 06, 2015. doi: 10.1590/S0100-06832013000100013.

SISTI, C. P. J. et al. Change in carbon and nitrogen stocks in soil under 13 years of conventional or zero tillage in southern Brazil. **Soil Tillage Research**, v.76, p.58, 2004. Available from: <http://www.sciencedirect.com/science/article/pii/S016719870300196X>. Accessed: mai. 06, 2015. doi: 10.1016/j.still.2003.08.007.

SMITH, B. N.; EPSTEIN, S. Two categories of $^{13}C/^{12}C$ ratios for higher plants. **Plant Physiology**, v.47, 384p, 1971. Available from: <http://www.plantphysiol.org/content/47/3/380>. Accessed: mai. 06, 2015. doi: 10.1104/pp.47.3.380.

SPECHT, A.; WEST, P. W. Estimation of biomass and sequestered carbon on farm forest plantations in northern New South Wales, Australia. **Biomass & Bioenergy**, v.25, n.4, 379p, 2003. Available from: <http://www.sciencedirect.com/science/article/pii/S0961953403000503>. Accessed: mai. 07, 2015. doi: 10.1016/S0961-9534(03)00050-3.

STEVENSON, F. J.; COLE, M. **Cycles of soil**: carbon, nitrogen, phosphorus, sulfur, micronutrients. New York: John Wiley, 1999. p.427.

SUESS, H. E. Radiation concentration in modern wood. **Science**, v.122, n.3166, p.417, 1955.

VIANA, M. B. **O eucalipto e os efeitos ambientais do seu plantio em escala**. Brasília: Biblioteca Digital da Câmara dos Deputados, p.29, 2004.

WORLD REFERENCE BASE FOR SOIL RESOURCES. (WRB). **A framework for international classification, correlation and communication**. Food and Agriculture Organization of the United Nations. Rome: IUSS/ISRIC/FAO, 2006. p.145. (World Soil Resources Reports, 103).

ZINN, Y. L. et al. Changes in soil organic carbon stocks under agriculture in Brazil. **Soil Tillage Research**, v.84, p.40, 2005. Available from: <http://www.sciencedirect.com/science/article/pii/S0167198704001862>. Accessed: mai. 07, 2015. doi: 10.1016/j.still.2004.08.007.