# University of São Paulo "Luiz de Queiroz" College of Agriculture

Soil quality response to land-use change for sugarcane expansion in Brazil
Maurício Roberto Cherubin

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Thesis presented to obtain the degree of Doctor in Science: Area: Soils and Plant Nutrition

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	Advisor: Prof. Dr. CARLOS CLEMENTE CERRI
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"Take care of the soil so it can take care of us"

Karlen and Rice (2015)

### **CONTENTS**

RESUMO	11
ABSTRACT	13
1 INTRODUCTION	15
References	20
2 SUGARCANE EXPANSION IN BRAZILIAN TROPICAL SOILS - EFFECTS OF LA	ND-
USE CHANGE ON SOIL CHEMICAL ATTRIBUTES	23
Abstract	23
2.1 Introduction	23
2.2 Material and Methods	25
2.2.1 Study sites	25
2.2.2 Land-use change sequence	30
2.2.3 Sampling and soil chemical attributes	32
2.2.4 Data analyses	32
2.3 Results	33
2.3.1 Soil organic carbon and total nitrogen	33
2.3.2 Soil acidity attributes and CEC <sub>pH7</sub>	35
2.3.3 Soil macro- and micro-nutrients	36
2.3.4 Correlation among soil chemical attributes	40
2.4 Discussion	42
2.4.1 Land-use change effects on soil organic carbon and total nitrogen	42
2.4.2 Land-use change effects on acidity attributes and CEC <sub>pH7</sub>	43
2.4.3 Land-use change effects on macro- and micro- nutrients	44
2.4.4 Soil chemical quality and its implications for sugarcane expansion	46
2.5 Conclusions	47
References	48
3 PHOSPHORUS POOLS RESPONSES TO LAND-USE CHANGE FOR SUGARC	ANE
EXPANSION IN WEATHERED BRAZILIAN SOILS	55
Abstract	55
3.1 Introduction	55
3.2 Material and Methods	57
3.2.1 Study sites and experimental design	57
3.2.2 Soil sampling and phosphorus fractionation	57
3.2.3 Soil attributes and macrofauna variables	58
3.2.4 Data analyses	59
3.3 Results	
3.3.1 Dynamic of P fraction levels under land-use change	60
3.3.2 Labile, moderately labile and non-labile P pool stocks	62
3.3.3 Biological, geochemical and total P pool stocks	
3.3.4 Relationship among P pools, soil chamical attributes and clay content	
3.3.5 Relationship between P pools and macrofauna	
3.4 Discussion	

3.4.1 Sugarcane expansion and its implications on P dynamic	69
3.4.2 Implications of macrofauna on P cycling	73
3.4.3 Phosphorus pools as soil quality indicators	74
3.5 Conclusions	75
References	76
4 SOIL PHYSICAL QUALITY RESPONSE TO SUGARCANE EXPANSION IN	BRAZIL
	83
Abstract	83
4.1 Introduction	83
4.2 Material and Methods	85
4.2.1 Study sites and experimental design	85
4.2.2 Sampling and soil physical measurements	85
4.2.3 Soil Physical Quality Index calculation	87
4.2.4 Data analyses	88
4.3 Results	89
4.3.1 Bulk density (BD) and soil degree of compactness (SDC)	89
4.3.2 Soil porosity	90
4.3.3 Soil water storage capacity (SWSC) and soil aeration capacity (SAC)	92
4.3.4 Field-satured hydraulic conductivity (K <sub>fs</sub> )	94
4.3.5 Soil resistance to penetration (SRP)	95
4.3.6 Soil Stability Structural Index (SSI)	96
4.3.7 Correlation between soil physical properties and SOC	96
4.3.8 Soil physical quality assessement	98
4.4 Discussion	100
4.4.1 Impacts of the LUC from native vegetation to pasture on soil physical attribute	s 100
4.4.2 Impacts of the LUC from pasture to sugarcane on soil physical attributes	102
4.4.3 Sugarcane expansion versus soil physical quality	103
4.5 Conclusions	106
References	106
5 ASSESSING SOIL STRUCTURAL QUALITY UNDER BRAZILIAN SUG	ARCANE
EXPANSION AREAS USING VISUAL EVALUATION OF SOIL STRUCTUR	E (VESS)
	113
Abstract	113
5.1 Introduction	113
5.2 Material and Methods	116
5.2.1 Study sites and experimental design	116
5.2.2 Sampling and VESS measurements	116
5.2.3 Relationship among VESS scores and quantitative soil physical properties	117
5.2.4 Data analyses	118
5.3 Results and Discussion	118
5.3.1 VESS sensitivity to detect LUC effects on soil structural quality	118
5.3.2 VESS score as an integrative soil structural quality indicator	127
5.4 Conclusions	130
References	130

6 A SOIL MANAGEMENT ASSESSMENT FRAMEWORK (SMAF) EVALUATIO	N OF
BRAZILIAN SUGARCANE EXPANSION ON SOIL QUALITY	135
Abstract	135
6.1 Introduction	135
6.2 Material and Methods	138
6.2.1 Study sites and experimental design	138
6.2.2 Soil sampling and laboratory analyses	138
6.2.3 Soil Management Assessment Framework	140
6.2.4 Data analyses	141
6.3 Results and Discussion	142
6.3.1 Soil chemical indicators	142
6.3.2 Soil physical indicators	145
6.3.3 Soil biological indicators	146
6.3.4 Overall Soil Quality Index and scores	148
6.3.4 Overall Soil Quality Index versus SOC stocks and VESS scores	151
6.4 Conclusions	154
References	154
7 SOIL QUALITY INDEXING STRATEGIES FOR EVALUATING SUGARO	CANE
EXPANSION IN BRAZIL	161
Abstract	161
7.1 Introduction	161
7.2 Material and Methods	164
7.2.1 Study sites and experimental design	
7.2.2 Soil sampling and analyses	164
7.2.3 Developing the soil quality indexes	166
7.2.4 Data analyses	173
7.3 Results and Discussion	173
7.3.1 Soil quality indicators	173
7.3.2 Soil quality indexing	176
7.3.3 What is the best indexing strategy for assessing sugarcane expansion impacts of	on soil
quality?	186
7.4 Conclusions	188
References	189
8 FINAL CONSIDER ATIONS	197

#### **RESUMO**

# Alterações na qualidade do solo devido a mudança de uso da terra para expansão da cana-de-açúcar no Brasil

Globalmente, o aumento da demanda de biocombustíveis têm intensificado a taxa de mudança de uso da terra (MUT) para expansão da produção de culturas para fins energéticos. No Brasil, a área de cana-de-acúcar aumentou 35% (3,2 Mha) na última década. A expansão do cultivo de cana-de-açúcar tem resultado em pastagens extensivas sendo submetidas a intensiva mecanização e ao uso de agroquímicos, implicando diretamente na qualidade do solo (QS). A hipótese testada nesse estudo foi que a MUT resulta na degradação da QS. Para tanto foi conduzido um estudo em três locais na região centro-sul, com objetivo de avaliar as modificações na QS devido a principal sequência de MUT (vegetação nativa - pastagem cana-de-açúcar) associada a expansão do cultivo de cana-de-açúcar no Brasil. Em cada uso da terra, amostras indeformadas e deformadas de solo foram coletadas nas profundidades de 0-10, 10-20 e 20-30 cm. Os atributos químicos e físicos do solo foram mensurados através de análises laboratoriais e à campo. Dados de atributos biológicos também foram incluídos no estudo. Inicialmente, os efeitos da MUT foram quantificados individualmente para cada um dos atributos do solo, e em seguida as alterações na QS global foram avaliadas através da Soil Management Assessment Framework (SMAF) e de seis índices de OS (IOS), desenvolvidos usando métodos com complexidade crescente. Os resultados demonstraram que a conversão da vegetação nativa em pastagem extensiva resultou na acidificação do solo, redução dos teores de carbono orgânico (COS) e macronutrientes (especialmente P), e severa compactação do solo, desequilibrando a relação entre ar e água e aumentando a resistência mecânica do solo ao crescimento radicular. Conversão da pastagem em cana-de-açúcar melhorou a qualidade química do solo através da correção da acidez e aumento dos macronutrientes. Apesar dessas melhorias, prolongado período de cultivo de cana-de-açúcar reduziu os teores de COS; e a maioria do P adicionado via fertilizantes acumulou em formas menos lábeis, confirmando o importante papel do P orgânico no fornecimento de P disponível às plantas em solos brasileiros. O cultivo de cana-de-açúcar teve impactos negativos nos atributos físicos do solo menos intensos do que aqueles gerados pelo uso com pastagem. Embora o preparo do solo para plantio e reforma da cana-de-açúcar reduziu a compactação do solo, os dados sugeriram que estes efeitos são de curta duração, ocorrendo a reconsolidação do solo e o aumento dos riscos de erosão ao longo do tempo. As alterações físicas do solo induzidas pela MUT foram detectadas tanto por meio de atributos quantitativos quanto por meio de avaliação visual da estrutura do solo (VESS), um método simples e diretamente aplicado no campo. A SMAF detectou eficientemente as alterações na QS devido a MUT. Além disso, todos os IQS desenvolvidos permitiram ranquear corretamente a QS entre os usos da terra. Assim, recomendamos que IQS mais simples e com melhor relação custo-benefício usando poucos indicadores chaves, tais como: pH, P, K, VESS e COS com ponderação proporcional entre os setores do solo (químico, físico e biológico) sejam usados como protocolo para avaliar a QS nas áreas de produção de cana-de-açúcar. Os resultados obtidos usando a SMAF e os IQS sugeriram que a conversão da vegetação nativa em pastagem extensiva reduziu a QS, degradando indicadores químicos, físicos e biológicos. Por outro lado, a conversão de pastagem em cana-de-açúcar não teve impactos na QS global, uma vez que a melhoria dos atributos químicos compensou os impactos negativos nos indicatores físicos e biológicos. Desta forma, esses resultados poderão ser utilizados como base científica pelos produtores, extencionistas e políticos para orientar estratégias de manejo que mantenham e/ou melhorem a QS e consequentemente a sustentabilidade da produção de cana-de-açúcar no Brasil.

Palavras-chave: Produção de etanol; Indicadores do solo; Índice de qualidade do solo; SMAF

#### **ABSTRACT**

#### Soil quality responses to land-use change for sugarcane expansion in Brazil

Globally, increasing demands for biofuels have intensified the rate of land-use change (LUC) for expansion of bioenergy crops. In Brazil, the world's largest sugarcane-ethanol producer, sugarcane area has expanded by 35% (3.2 Mha) in the last decade. Sugarcane expansion has resulted in extensive pastures being subjected to intensive mechanization and large inputs of agrochemicals, which have direct implications on soil quality (SQ). We hypothesized that LUC to support sugarcane expansion leads to overall SQ degradation. To test this hypothesis we conducted a field-study at three sites in the central-southern region, to assess the SQ response to the primary LUC sequence (i.e., native vegetation to pasture to sugarcane) associated to sugarcane expansion in Brazil. At each land use site undisturbed and disturbed soil samples were collected from the 0-10, 10-20 and 20-30 cm depths. Soil chemical and physical attributes were measured through on-farm and laboratory analyses. A dataset of soil biological attributes was also included in this study. Initially, the LUC effects on each individual soil indicator were quantified. Afterward, the LUC effects on overall SQ were assessed using the Soil Management Assessment Framework (SMAF). Furthermore, six SQ indexes (SQI) were developed using approaches with increasing complexity. Our results showed that long-term conversion from native vegetation to extensive pasture led to soil acidification, significant depletion of soil organic carbon (SOC) and macronutrients [especially phosphorus (P)] and severe soil compaction, which creates an unbalanced ratio between water- and air-filled pore space within the soil and increases mechanical resistance to root growth. Conversion from pasture to sugarcane improved soil chemical quality by correcting for acidity and increasing macronutrient levels. Despite those improvements, most of the P added by fertilizer accumulated in less plant-available P forms, confirming the key role of organic P has in providing available P to plants in Brazilian soils. Long-term sugarcane production subsequently led to further SOC depletions. Sugarcane production had slight negative impacts on soil physical attributes compared to pasture land. Although tillage performed for sugarcane planting and replanting alleviates soil compaction, our data suggested that the effects are short-term with persistent, reoccurring soil consolidation that increases erosion risk over time. These soil physical changes, induced by LUC, were detected by quantitative soil physical properties as well as by visual evaluation of soil structure (VESS), an on-farm and user-friendly method for evaluating SQ. The SMAF efficiently detected overall SQ response to LUC and it could be reliably used under Brazilian soil conditions. Furthermore, since all of the SQI values developed in this study were able to rank SQ among land uses. We recommend that simpler and more cost-effective SQI strategies using a small number of carefully chosen soil indicators, such as: pH, P, K, VESS and SOC, and proportional weighting within of each soil sectors (chemical, physical and biological) be used as a protocol for SQ assessments in Brazilian sugarcane areas. The SMAF and SQI scores suggested that long-term conversion from native vegetation to extensive pasture depleted overall SQ, driven by decreases in chemical, physical and biological indicators. In contrast, conversion from pasture to sugarcane had no negative impacts on overall SQ, mainly because chemical improvements offset negative impacts on biological and physical indicators. Therefore, our findings can be used as scientific base by farmers, extension agents and public policy makers to adopt and develop management strategies that sustain and/or improving SQ and the sustainability of sugarcane production in Brazil.

Keywords: Ethanol production; Soil indicators; Soil quality index; SMAF

#### 1 INTRODUCTION

Land-use change (LUC) processes have transformed a large portion of the planet's land surface, affecting directly the land capacity for provisioning ecosystem services (FOLEY et al., 2005). Increasing global demand to support bioenergy feedstock production has intensified LUC worldwide [(e.g., South America (LAPOLA et al., 2010; GODEMBERG et al., 2014), North America (WRIGHT; WIMBERLY, 2013), Europe (FISCHER et al., 2010), Asia (MUKHERJEE; SOVACOOL, 2014), Africa (GASPARATOS et al., 2015), Australia (GRUNDY et al., 2016)]. Globally, a gross land demand for bioenergy ranging from 50 to 200 Mha by 2050 was projected by Woods et al. (2015). Direct LUC refers to changes in land use that occur where bioenergy feedstock production becomes established, and generally includes both conversion from food or fiber production (including crop rotation patterns, conversion of pasture land, and changes in forest management) and conversion of natural ecosystems (KARP et al., 2015). Current LUC, especially forest conversion to agricultural land, has been and still is the primary driver of global deforestation and forest degradation in many countries, especially in the tropics (KARP et al., 2015). Therefore, to minimize environmental impact, expansion of biofuel crop production on non-cultivated land or marginal/degraded lands, such as extensive pasturelands, is currently being promoted.

Brazil is the world's largest sugarcane producer, and currently accounting for with about 40% of the global harvest, of which approximately a half is used to produce ethanol (COMPANHIA NACIONAL DE ABASTECIMENTO, 2016). Roughly one-third of the total global ethanol fuel production is provided for through Brazilian sugarcane, with small contributions from other Latin America countries (GOLDEMBERG et al., 2014). The evolution of sugarcane, sugar and ethanol production in Brazil is shown in Figure 1A. Historically sugarcane expansion has been concentrated in the central-southern region (Figure 1B), with 70% of this expansion occurring in extensive pasturelands (ADAMI et al., 2012). The vast area of degraded pasture in Brazil, coupled with opportunities for improvements in current ranching practices, could provide enough land for sugarcane production to meet the projected domestic demand for ethanol whilst meeting the demand for other ecosystem services (HORTA NOGUEIRA; CAPAZ, 2013; GOLDEMBERG et al., 2014; STRASSBURG et al., 2014).

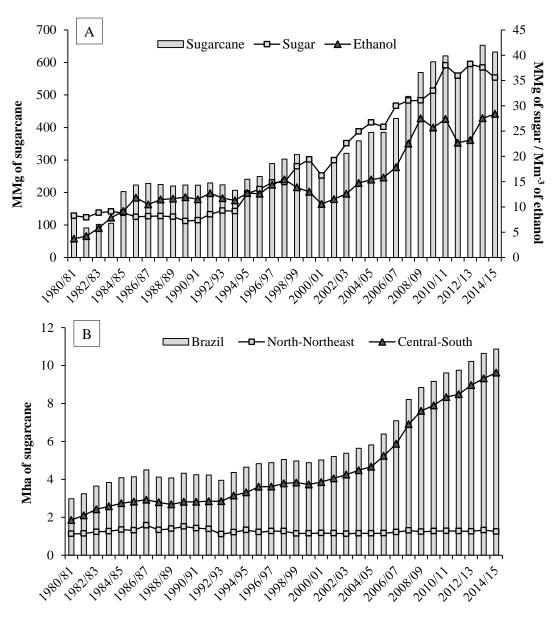


Figure 1 - Evolution of sugarcane, sugar and ethanol production (A) and sugarcane cropped area (B) in Brazil (UNIÃO DA INDÚSTRIA DE CANA-DE-AÇÚCAR, 2016)

Current preditions indicate that an additional of 6.4 Mha of sugarcane land will be required to meet the Brazilian demand for ethanol by 2021 (GOLDEMBERG et al., 2014). The accelerated pace of recent and projected sugarcane expansion, through which extensive pasture land has been subjected to intensive mechanization and agrochemical inputs has raised concerns regarding potential ecosystem impacts of LUC in Brazil. It is expected that maximizing the use of one ecosystem service (e.g., provision of biofuel and air quality) often leads to a sharp decline of other ecosystem services, particularly regulating services (e.g., C sequestration, soil retention, and water resource conservation) (FU et al., 2015). Thus,

identifying feasible and sensitive indicators for assessing trade-offs among various ecosystems services has become a challenge to the scientific community and stakeholders.

Soil quality (SQ) has been identified as a key component to assess the environmental sustainability of natural and anthropogenic ecosystems. Karlen et al. (1997) conceptualized SQ as the capacity of a specific kind of soil to function, within natural or managed ecosystem boundaries, to sustain plant and animal productivity, maintain or enhance water and air quality, and support human health and habitation. It is a complex functional concept and cannot be measured directly in the field or laboratory; but can be indirectly inferred by soil indicators. Indicators of SQ are those measurable soil properties and processes that have greatest sensitivity to changes in soil function and its ecosystem services (ANDREWS; KARLEN; CAMBARDELLA, 2004; ZORNOZA et al., 2015).

Assessment of SQ involves a three-step conceptual framework (Figure 2), including (i) indicator selection (chemical, physical and biological); (ii) indicator interpretation (linear or non-linear scoring curves); and (iii) integration into an overall SQ index (SQI) (KARLEN; DITZLER; ANDREWS, 2003). Assessment values are generally expressed as a fraction or percentage of full performance for soil functions such as crop productivity, nutrient cycling, or environmental protection (ANDREWS; KARLEN; CAMBARDELLA, 2004).

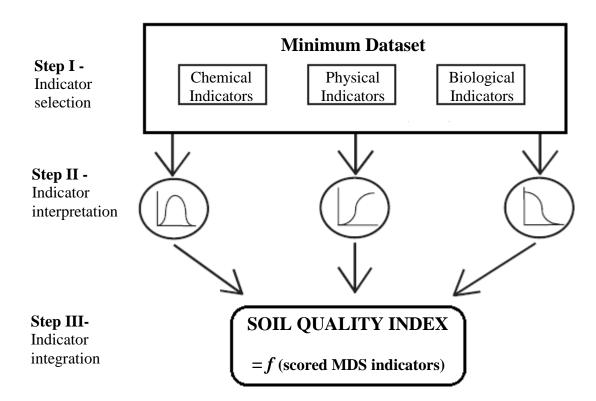


Figure 2 - Conceptual framework for the soil quality assessment (Adapted from Karlen, Ditzler and Andrews 2003)

Although this conceptual framework is broadly used for SQ assessments worldwide, different approaches have been tested to perform each one of the three steps (e.g., ANDREWS; KARLEN; MITCHELL, 2002; ANDREWS; KARLEN; CAMBARDELLA, 2004; MUKHERJEE; LAL, 2014). Soil quality indicators can be selected based on expert opinion, statistical procedures, decision rules, etc. Linear and non-linear curves can be used for scoring measured indicator values. Finally, scored values can be integrated into an overall index using simple additives or weighted additive methodologies. Each approach has advantages and disadvantages, and its performance dependent of the assessment's goals. Therefore, since there is no a universal method that can be used across multiple natural and anthropogenic ecosystems, many SQ assessment strategies have been developed and tested for specific purposes under particular environmental conditions worldwide. A distinguished example of the existing approaches is the Soil Management Assessment Framework (SMAF), which was initially developed and used by researchers in the USA on North American soils (ANDREWS; KARLEN; CAMBARDELLA, 2004); but it has been constantly enhanced by international collaborations, enabling to extend its use to other countries around world.

However, to our knowledge there is no any protocol or published studies evaluating SQ changes induced by the LUC for sugarcane expansion in Brazil. Therefore, we conducted a field study in central-southern Brazil, the largest sugarcane-producing regions of the world, for assessing SQ responses to the primary LUC sequence (i.e., native vegetation to pasture to sugarcane) associated with sugarcane expansion. For that, the specific objectives were to: i) evaluate the LUC impacts on soil chemical attributes; ii) investigate soil physical and structural changes induced by the LUC impacts; iii) integrate soil chemical, physical and biological responses to LUC into an overall SQ assessment using different approaches and frameworks; iv) establish a protocol for assessing SQ changes in Brazilian sugarcane areas. We tested the main hypothesis that the LUC sequence induces alteration on dynamic of soil chemical, physical and biological indicators, leading to overall SQ degradation and its impacts can detected by SQ indexing strategies.

To meet our objectives, this thesis is organized into eight chapters. Briefly, the **first** one presents a short introduction about the research topic studied. The **second** one addresses the LUC impacts associated to sugarcane expansion on soil chemical attributes and overall soil chemical quality. The **third** one evaluates soil phosphorus pool (labile, moderately labile and non-labile pools, as well as biological and geochemical pools) changes induced by the LUC for sugarcane expansion. The **fourth** one addresses the LUC impacts on soil physical properties and overall soil physical quality. The **fifth** one evaluates the sensitivity of the

Visual Evaluation of Soil Structure (VESS) method for detecting soil structure quality changes due to sugarcane expansion. The **sixth** provides the results of the first application of Soil Management Assessment Framework (SMAF) for evaluating overall SQ changes in Brazil. The **seventh** one investigates SQ changes induced by sugarcane expansion through six indexing strategies and provides a protocol for SQ assessment in Brazilian sugarcane areas. Finally, the **eigth** one provides the final considerations of this study.

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## 2 SUGARCANE EXPANSION IN BRAZILIAN TROPICAL SOILS - EFFECTS OF LAND-USE CHANGE ON SOIL CHEMICAL ATTRIBUTES

#### **Abstract**

Land-use change (LUC) for sugarcane ethanol production has raised concerns about its potential environmental impacts in Brazil. Soil quality is a key indicator to infer about the environmental sustainability of Brazilian ethanol production. Our objective was to quantify the effects of the most common LUC sequence associated with sugarcane expansion (i.e., native vegetation to pasture to sugarcane) on chemical attributes in tropical soils. Soil sampling was carried out in three study sites located in central-southern Brazil, primary sugarcane region of production and expansion of the world. Overall, long-term conversion from native vegetation to extensive pasture decreased soil organic carbon (SOC), total nitrogen (TN), available phosphorus, sulfur, calcium, magnesium and boron contents. In addition, the LUC led to soil acidification and decreased CEC<sub>pH7</sub>, indicating that pasturelands had poor soil chemical quality. The LUC from pasture to sugarcane increased soil nutrient levels and reduced the soil acidity due to inputs of lime and fertilizers. Despite that, increments of available P and base saturation are necessary to achieve ideal soil chemical conditions to sugarcane growth. Short-time (<5 years) conversion from pasture to sugarcane had no significant impacts on SOC and NT contents; however, after 20 years of sugarcane production significant losses were quantified. Overall, our findings suggest that sugarcane expansion in Brazil replacing pasturelands will promote improvements on soil chemical quality. Nevertheless, sugarcane expansion can be associated with management strategies to increase soil organic matter and improve the soil fertility, reducing the environmental and economic costs associated with ethanol production in Brazil.

Keywords: Soil chemical quality; Biofuel crops; Ethanol production; Environmental impacts; Soil fertility

#### 2.1 Introduction

Land use activities whether converting natural landscapes for human use or changing management practices on human-dominated lands have transformed a large proportion of the planet's land surface (FOLEY et al., 2005), with large short and long term environmental implications (LAMBIN; MEYFROIDT, 2011; TILMAN et al., 2011). The environmental impacts of agriculture include those caused by expansion (when croplands and pastures extend into new areas, replacing natural ecosystems) and those caused by intensification (when existing lands are managed to be more productive) (FOLEY et al., 2011).

In Brazil, the area currently cultivated with sugarcane is undergoing significant expansion due to the growing demand for bioethanol, driven by environmental, geopolitical and economic issues (LAPOLA et al., 2010; 2014; GOLDEMBERG et al., 2014; HERNANDES; BUFON; SEABRA, 2014). This is considered one of the main causes of LUC

in the central-southern region (LAPOLA et al., 2010; 2014; GOLDEMBERG et al., 2014; WALTER et al., 2014).

Brazil is currently the world's largest sugarcane producer, accounting for one-third of global harvest. An area of 9.0 Mha was cultivated during the 2015/2016 season (COMPANHIA NACIONAL DE ABASTECIMENTO, 2015), with 90% of the sugarcane cultivated in Brazil concentrated within the central-southern region under tropical soils, especially Oxisols and Ultisols. Although Brazilian sugarcane production is significant, an additional 6.4 Mha of sugarcane area would be required to meet the projected internal demand of ethanol by 2021 (61.6 billion L) (GOLDEMBERG et al., 2014). In central-southern Brazil, sugarcane expanded primarily onto pasturelands and annual croplands, with limited expansion into areas of native vegetation (ADAMI et al., 2012; EGESKOG et al., 2014). In the near future, sugarcane expansion is most likely to occur in areas previously used as extensive pasture (LAPOLA et al., 2010; ADAMI et al., 2012; EGESKOG et al., 2014; GOLDEMBERG et al., 2014).

This recent expansion in production to meet ethanol demand, in combination with the projected future expansion, has raised concerns about the potential environmental impacts of LUC in Brazil (HERNANDES; BUFON; SEABRA, 2014). Therefore, it is necessary investigate the LUC effects to assess of sustainability of expanding ethanol production. The greatest challenge is to define sensitive indicators that reflect local specificities of the environmental implications from LUC (GASPARATOS; STROMBERG; TAKEUCHI, 2011; EFROYMSON et al., 2013; FU et al., 2015).

The soil quality is identified as key component to assess the environmental sustainability of natural and anthropogenic ecosystems (KARLEN et al., 1997) and it has been proposed into current protocols such as the one elaborate by Better Sugar Cane Initiative "Bonsucro®" certification (BETTER SUGAR CANE INITIATIVE, 2011) and the "Global Bioenergy Partnership" by FAO (GLOBAL BIOENERGY PARTNERSHIP, 2011). To evaluate the effects of land use and soil managements on soil quality, soil chemical attributes can be used as potential indicators, such as: available macro- and micronutrients, acidity attributes, cation exchange capacity and soil organic carbon (SOC) (DORAN; PARKIN, 1994; VEZZANI; MIELNICZUK, 2011; CARDOSO et al., 2013; ZORNOZA et al., 2015)

Previous studies indicated that LUC affects soil chemical attributes in different ways. In a global meta-analysis Don, Schumacher and Freibauer (2011) showed that conversions from forest to pasture and/or cropland promoted SOC losses. Recently, a large study under tropical soils in central-southern Brazil showed that soil C stocks decreased following LUC

from native vegetation to pastures and then from pasture to sugarcane (MELLO et al., 2014). However, C stocks increases were found where cropland was converted to sugarcane (MELLO et al., 2014). On the other hand, Carvalho et al. (2009) showed the conversion from Brazilian's Cerrado into cropland under conservative management in an Oxisol increased SOC and nutrients (P, K, Ca, Mg) and reduced soil acidity. In this case, it was driven mainly by fertilizer application and liming to reduce soil acidity. Geissen et al. (2009) under Peruvian highland jungle (Amazon forest) region and Lindel, Åström and Öberg (2010) under different soils in tropical Southeast Mexico, concluded that the LUC did not lead to significant changes in soil chemical attributes, although, Geissen et al. (2009) observed that soils used as pastureland became acidified.

Therefore, our objective was to quantify effects of the most common LUC sequence associated with sugarcane expansion (i.e., native vegetation to pasture to sugarcane) on chemical attributes in tropical soils of central-southern Brazil. We hypothesized that i) LUC from native vegetation to extensive pasture leads to significant depletions of SOC and macronutrients and increase soil acidification; ii) sugarcane cultivation replacing extensive pasturelands can recover soil fertility; iii) pasturelands have lower soil chemical quality than sugarcane fields, constituting hotspots for sugarcane expansion in Brazil.

#### 2.2 Material and Methods

#### 2.2.1 Study sites

The study was carried out in central-southern Brazil, the largest sugarcane-producing region of the world (Figure 1). Three strategic and representative sites were chosen along a transect of approximately 1,000 km across this region: (i) Lat\_17S: located near Jataí city in the southwestern region of the Goiás state (Lat.: 17°56′16″S; Long.: 51°38′31″W) with a mean altitude of 800 m; (ii) Lat\_21S: located near Valparaíso city in the west region of the São Paulo state (Lat.: 21°14′48″S; Long.: 50°47′04″W) with a mean altitude of 425 m. (iii) Lat\_23S: located near Ipaussu city in the south-central region of the São Paulo state (Lat.: 23°05′08″ S; Long.: 49°37′52″ W), with a mean altitude of 630 m.

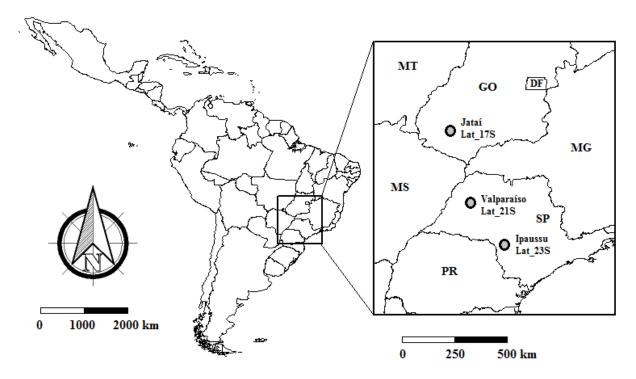


Figure 1 - Geographic location of study sites in central-southern Brazil

The climate classification in the studied sites are as follows: Lat\_17S: Awa type (Köppen classification) mesothermal tropical, with a mean annual temperature of 24.0 °C and annual precipitation of 1,600 mm (Figure 2A); Lat\_21S: Aw type (Köppen classification) humid tropical, with a mean annual temperature of 23.4 °C and annual precipitation of 1,240 mm (Figure 2B); Lat\_23S: Cwa type (Köppen classification) tropical, with annual mean temperature of 21.7 °C and annual precipitation of 1,470 mm (Figure 2C). Rainfall at all three sites is concentrated in the spring and summer (October to April), while the dry season is in the autumn and winter (May to September).

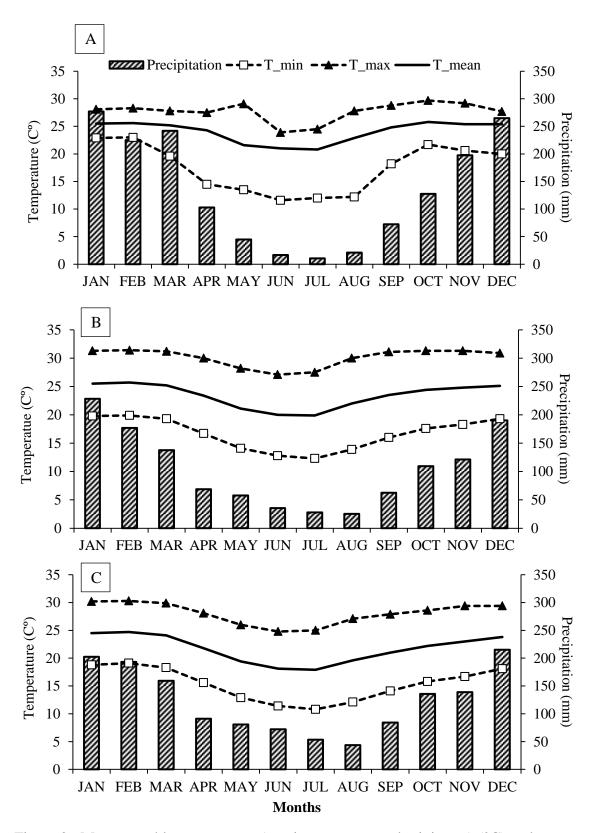


Figure 2 - Mean monthly temperature (maximum, mean and minimum) (°C) and mean annual precipitation (mm) in the region studied, where: A) Lat\_17S (Jataí - GO); B) Lat\_21S (Valparaíso - SP); C) Lat\_23S (Ipaussu - SP). Sources: CIIAGRO (http://www.ciiagro.sp.gov.br) and CEPAGRI (http://www.cpa.unicamp.br)

The soils at all three sites, classified as Oxisols, Ultisols and Alfisols, are characterized by highly weathered minerals (Ki and Kr weathering indexes have values < 2.0), typical of Brazilian tropical soils. A morphological description of 2-m deep soil profiles was carried out in January 2014, to classify the soils at each field site. From each soil horizon we collected samples for chemical, mineralogical and particle-size analyses. Overall, the soils have predominance of 1:1 minerals (kaolinite), Fe oxides (goethite, hematite) and Al oxide (gibbsite) (Figure 3). The profile description and the soils classification using criteria outlined by the USDA Soil Taxonomy (SOIL SURVEY STAFF, 2014) and Brazilian Classification System (SANTOS et al., 2013) as well as the parental material are presented in Table 1.

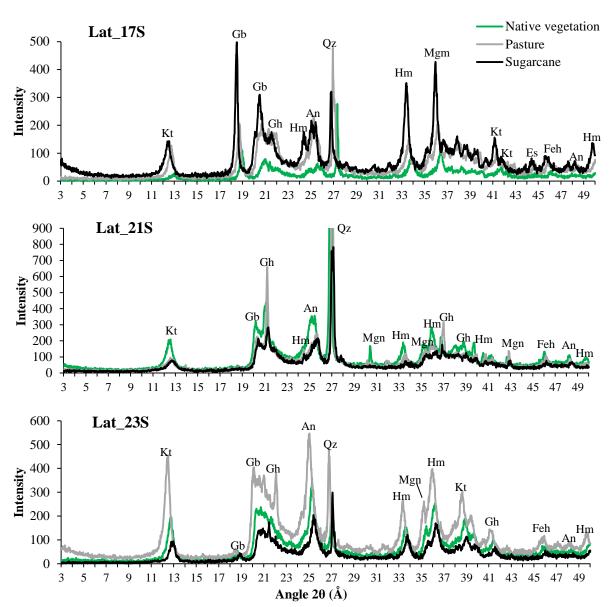


Figure 3 - Diffractograms of the silt+clay soil fraction (<0.05 mm Ø) for native vegetation, pasture and sugarcane at Lat\_17S, Lat\_21S and Lat\_23S, in central-southern Brazil

Table 1 - Profile description and classification of soils under native vegetation, pasture and sugarcane at the studied sites

Site§	Land Use		Depth	MO	pН	pН	ΔрН*	Index		BS†	Sand	Silte	Clay	Moist Color		sification§§	Geology and
Site	Land Use	попион	cm	g kg <sup>-1</sup>	$H_2O$	KCl	∆рп"	Ki	Kr	%		g kg <sup>-1</sup>		Worst Color	Soil Taxonomy	SiBCS	parental material‡
		$A_1$	0-21	20.91	4.5	3.7	-0.8	-	-	2.95	608	64	327	2.5YR 2.5/3			
	Native	$\mathbf{A}_2$	21-40	15.43	4.8	4.0	-0.8	-	-	3.24	570	55	375	2.5YR 2.5/4	clayey Anionic	Latossolo Vermelho distrófico típico	Paraná Basin (São Bento group) and
	Vegetation	$Bw_1$	40-108	7.07	5.0	4.6	-0.4	0.88	0.58	5.97	540	81	378	10R 3/3	Acrudox		
		$\mathbf{Bw}_2$	108-200+	4.15	5.2	5.4	+0.2	0.84	0.53	5.69	502	42	456	10R 3/3			Serra Geral geologic
	Pasture	$A_1$	0-19	11.35	5.0	4.0	-1.0	-	-	10.88	846	16	137	5YR 3/2		Latossolo Vermelho Amarelo distrófico	formation. The parental material is tholeitic basalt
		$A_2$	19-41	6.31	4.7	4.0	-0.7	-	-	4.32	835	14	150	5YR 3/3			
Lat_17S		AB	41-62	5.68	4.9	4.2	-0.7	-	-	7.05	812	25	164	5YR 3/3			
		Bw	62-200+	1.96	5.1	4.7	-0.4	1.05	0.74	7.26	806	18	176	5YR 3/4		(volcanic rocks)	
		Ap	0-13	14.57	6.5	5.5	-1.0	-	-	28.47	620	50	330	10R 4/4			with intertrappean
		À	13-35	12.48	4.8	4.4	-0.4	-	-	9.63	604	43	354	2.5YR 3/4	1 4	7 . 1 77 11	sandstone and
	Sugarcane	BA	35-61	10.55	5.2	4.4	-0.8	-	-	12.55	571	52	377	2.5YR 4/2	clayey Anionic	Latossolo Vermelho	diabase sills and dikes
		$Bw_1$	61-126	7.57	5.0	4.9	-0.1	0.76	0.47	9.81	541	33	426	2.5YR 3/3	Acrudox	distrófico típico	
		$Bw_2$	126-200+	4.85	4.8	5.5	+0.7	0.68	0.43	9.41	541	32	427	2.5YR 3/3			
		A	0-37	14.50	7.3	6.8	-0.5	-	-	92.76	744	55	202	2.5YR 3/2	loamy Typic Rhodudalf		Paraná Basin (Bauru group) and Adamantina geologic formation. The parental material is sandstone, with
	NT 4	AB	37-62	2.92	7.6	6.2	-1.4	-	-	85.77	724	49	226	2.5YR 3/3		Argissolo Vermelho	
	Native	Bt	62-126	3.52	7.4	6.6	-0.8	1.53	1.19	85.52	715	34	251	5YR 4/4		my Typic Amarelo eutrófico	
	Vegetation	Bc	126-152	4.78	7.7	6.5	-1.2	-	-	76.46	715	30	255	2.5YR 3/4			
		C	152-200+	2.95	7.4	6.6	-0.8	-	-	84.34	700	47	253	2.5YR 4/4			
Lat 210		A	0-35	10.79	5.5	4.2	-1.3	-	-	46.50	730	67	203	7.5YR 3/3	£ 1 T	Argissolo Vermelho Amarelo eutrófico	
Lat_21S	Pasture	BA	35-72	7.33	5.9	4.5	-1.4	-	-	52.92	644	53	303	5YR 3/4	fine-loamy Typic		
		Bt	72-200+	3.98	5.3	3.8	-1.5	1.88	1.4	21.75	668	53	279	5YR 4/4	Kandiudult	típico	
		A	0-26	14.44	6.4	5.4	-1.0	-	-	66.94	746	78	176	5YR 2.5/2			carystone, sitistone
	C	B1	26-84	4.58	5.6	4.9	-0.7	1.83	1.44	59.43	633	39	328	5YR 4/6	loamy Typic Hapludalf  Argissolo Vermelho Amarelo eutrófico		
	Sugarcane	B2	84-170	5.34	5.6	5.1	-0.5	1.68	1.37	69.33	628	41	331	7.5YR 5/8		and conglomerate	
		C	170-200+	4.85	6.3	5.4	-0.9	-	-	60.30	636	34	330	7.5YR 5/6		abrúptico	
		A	0-28	35.11	4.5	3.6	-0.9	-	-	17.17	168	137	694	2.5YR 3/3			
	Native	$Bt_1$	28-82	25.14	4.6	3.5	-1.1	1.69	1.33	7.05	122	168	710	2.5YR 4/4	clayey Rhodic	Nitossolo Vermelho	Paraná Basin (São
	Vegetation	$\mathbf{Bt}_2$	82-163	24.94	4.5	3.6	-0.9	1.72	1.35	3.89	153	181	665	2.5YR 3/3	Hapludox	alumínico típico	Bento group) and Serra Geral geologic
		C	163-200+	18.62	4.8	3.7	-1.1	-	-	6.24	156	179	665	2.5YR 3/4			
	Pasture	A	0-15	32.16	5.4	4.2	-1.2	-	-	44.06	236	178	586	2.5YR 2.5/4	clayey Rhodic Nitossolo Vermelho Kandiudox alumínico típico	formation. The	
Lat_23S		AB	15-51	20.64	5.1	4.1	-1.0	-	-	48.71	146	191	663	2.5YR 3/4		Nitossolo Vermelho	(volcanic rocks)
		Bt	51-148	16.20	5.2	3.8	-1.4	1.92	1.51	24.83	108	158	735	10R 4/3		alumínico típico	
		C	148-200+	10.99	5.3	3.9	-1.4	-	-	23.25	124	109	767	10R 3/4			
	Sugarcane	A	0-34	27.35	6.7	6.0	-0.7	-	-	78.77	229	99	672	2.5YR 2.5/3			— with intertrappean sandstone and
		$Bt_1$	34-73	14.90	6.9	5.0	-1.9	1.58	1.23	55.27	195	91	714	2.5YR 3/4	clayey Rhodic Nitossolo Vermelho Hapludox alumínico típico	diabase sills and dikes	
		$Bt_2$	73-158	11.58	6.4	3.7	-2.7	1.6	1.25	14.52	197	90	713	2.5YR 3/4			
		C	158-200+	13.33	5.4	4.2	-1.2	-	-	38.64	192	71	737	10R 3/6			

\$Lat\_17S, southwestern region of Goiás state (17°56′16″S, 51°38′31″W); Lat\_21S, west region of São Paulo state (21°14′48″S, 50°47′04″W); Lat\_23S, south-central region of São Paulo state (23°05′08″S, 49°37′52″W). \*ΔpH = pH<sub>KCl (1M)</sub> – pH<sub>H2O</sub>; \*\*Ki and Kr Indexes: degree of weathering, obtained by equations Ki = 1.7 x (% SiO<sub>2</sub>/%Al<sub>2</sub>O<sub>3</sub>) and Kr = 1.7 x % SiO<sub>2</sub>/[%Al<sub>2</sub>O<sub>3</sub>+(%Fe<sub>2</sub>O<sub>3</sub> x 0.64)]. Ki and Kr <2 indicates soil higher weathered; †BS = base saturation; \$\$USDA Soil Taxonomy (SOIL SURVEY STAFF, 2014) and Brazilian Classification System - SiBCS (SANTOS et al., 2013); \*According to geologic map of Brazil (MINISTÉRIO DE MINAS E ENERGIA, 1981).

#### 2.2.2 Land-use change sequence

To assess the effects of LUC on soil chemical attributes we adopted a chronosequence approach, where each one of three studied sites (Lat\_17S; Lat\_21S; Lat\_23S) included three land uses: native vegetation, pasture and sugarcane crop, representing the most common land use transition sequence in the south-central region of Brazil.

The three land uses are co-located adjacent to each other, to minimize the effects of climatic, topographic and soil variations on the soil quality indicators. Despite that concern, a textural difference between the soil under pasture (lower clay content) and the other land uses of the same chronosequence was observed at Lat\_17S (Table 1). Thus, we highlighted that all statements about LUC effects on P pools for this site must be interpreted carefully. When adopting a chronosequence approach the variability in soils within the chronosequence is not always possible to be controlled, but, the global lack of long-term studies evaluating the effects of LUC, and especially in Brazil, justifies to keep this synchronic approach in our study (SIQUEIRA NETO et al., 2010; COSTA JUNIOR et al., 2013).

Land use historical information and brief description of management operations conducted at the studied sites are shown in Figure 4 and Table 2.

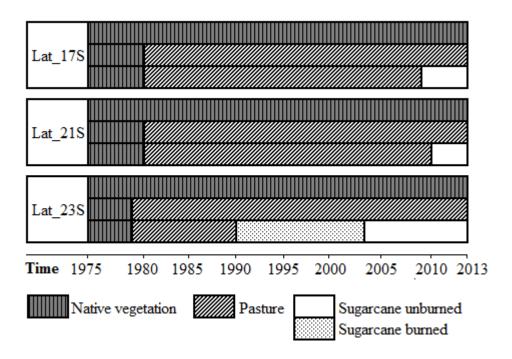


Figure 4 - Schematization of the land use history in the studied chronosequences

Table 2 - Land use historical information and brief description of management operations conducted at the studied sites

Site	Land use	Description
	Native vegetation	Cerradao forest formation, Cerrado biome, characterized by sclerophyllous and xeromorphic species. The vegetation is dense compared to the Cerrado <i>stricto sensu</i> (savanna).
Lat_17S	Pasture	Conversion from native vegetation to pasture occurred at 1980. Pasture is composed by tropical grasses of the genus <i>Brachiaria</i> , predominantly <i>B. decumbens</i> , <i>B. brizantha</i> and <i>B. ruziziensis</i> , and supports 1.5 AU ha <sup>-1</sup> full year. The conversion of native vegetation to pasture occurred of the beginning of the 1980s. The predominant species are of the <i>Brachiaria</i> genus, especially <i>B. decumbens</i> , <i>B. brizantha</i> e <i>B. ruziziensis</i> . The stocking rate is 1.5 UA ha <sup>-1</sup> along the all year.
Lat_175	Sugarcane	Sugarcane cultivar RB855453 was cultivated over part of the pasture at 2009. At that time soil was prepared by plowing and disking. The sugarcane mean yield since the implantation is $81.5$ ton $ha^{-1}$ . At the sampling time for chemical and biological analyses the sugarcane was in the third ratoon cropping of its cycle; while at soil sampling time for physical analyses the soil had been newly tilled for sugarcane replanting (chiseling and disking). Soil acidity was corrected with the application of $1.6$ ton $ha^{-1}$ of dolomitic lime. Also was applied 1 ton $ha^{-1}$ of gypsum before cropping to supply S and Ca and $150$ kg $ha^{-1}$ of $P_2O_5$ . Annually the crop fertilization is carried out by the application of $110$ kg $ha^{-1}$ N and $75$ kg $ha^{-1}$ K $_2$ O. Sugarcane has been mechanically harvested using a harvester ( $\approx 20$ Mg) and transported by a tractor + trailer ( $\approx 10 + 20$ Mg). Sugarcane production has not used controlled traffic system.
	Native vegetation	The local vegetation is seasonal semideciduous forest, Atlantic forest biome, in which a portion of the trees defoliates during the dry season. It is a transitional region, where the forest has more xeromorphic species than the wetter areas of the Atlantic forest, on the other hand presents less xeromorphic species than the Cerrado vegetation.
Lat_21S	Pasture	Conversion from native vegetation to pasture occurred at 1980. Pasture is composed by tropical grasses of the <i>Brachiaria</i> genus and supports 2 AU ha <sup>-1</sup> full year. Annually, the pasture receives 25 kg ha <sup>-1</sup> N, 6 kg ha <sup>-1</sup> P <sub>2</sub> O <sub>5</sub> , 23 kg ha <sup>-1</sup> K <sub>2</sub> O (mineral fertilizer).
	Sugarcane	Sugarcane cultivar SP791011 was cultivated over part of the pasture at 2010. At that time soil was prepared by plowing and disking. The sugarcane mean yield since the implantation is 80 ton ha <sup>-1</sup> . At the sampling time sugarcane was in the fourth ratoon cropping of its cycle. Soil acidity was corrected by liming. The sugarcane was annually fertilized with 11 kg ha <sup>-1</sup> N, 55 kg ha <sup>-1</sup> P <sub>2</sub> O <sub>5</sub> and 55 kg ha <sup>-1</sup> K <sub>2</sub> O (mineral fertilizer). Vinasse was applied to sugarcane in 2012 at an amount of 150 m <sup>3</sup> ha <sup>-1</sup> (corresponding to approximately 35 kg ha <sup>-1</sup> N, 30 kg ha <sup>-1</sup> P <sub>2</sub> O <sub>5</sub> and 300 kg ha <sup>-1</sup> K <sub>2</sub> O). Sugarcane has been mechanically harvested without controlled traffic system using machines similar to those described for Lat_17S.
	Native vegetation	The local vegetation is similar that described for Lat_21S site.
	Pasture	Conversion from native vegetation to pasture occurred at 1979. Pasture is composed by tropical grasses of the <i>Cynodon</i> genus, and supports 1 AU ha <sup>-1</sup> full year.
Lat_23S	Sugarcane	Sugarcane cultivar CTC6 was cultivated over part of the pasture at the beginning of the 1990s. At that time soil was prepared by plowing and disking. The sugarcane mean yield since the implantation is 85 ton ha <sup>-1</sup> . At the sampling time sugarcane was in the fifth ration cropping of its cycle. Soil acidity was corrected by liming. Annually the crop fertilization is carried out by the application of 45 kg ha <sup>-1</sup> of N (urea) plus 200 m <sup>3</sup> ha <sup>-1</sup> of vinasse (approximately 45 kg ha <sup>-1</sup> N, 40 kg ha <sup>-1</sup> P <sub>2</sub> O <sub>5</sub> and 400 kg ha <sup>-1</sup> K <sub>2</sub> O) and 25 ton ha <sup>-1</sup> of filter cake and boiler ash (approximately 75 kg ha <sup>-1</sup> N, 55 kg ha <sup>-1</sup> P <sub>2</sub> O <sub>5</sub> and 30 kg ha <sup>-1</sup> K <sub>2</sub> O). Sugarcane has been mechanically harvested since 2003 without controlled traffic system using machines similar to those described for Lat_17S. From 2013 around 50% of the sugarcane straw has been removed from the soil for electric energy production.

#### 2.2.3 Sampling and soil chemical attributes

Soil sampling was completed in January 2013 during the rainy season, when the sugarcane was in full growth close to harvest. Soil samples within each land use (i.e., native vegetation, pasture and sugarcane) were collected using a consistent grid pattern composed of nine points spaced 50 m apart, providing a total of 27 sampling points (3 land uses x 9 points) for each site or 81 sampling points for the three studied sites. Around each sampling point, composite samples consisting of 12 subsamples were collected using a Dutch auger, at three depths: 0-10, 10-20 and 20-30 cm. This provided a total of 243 disturbed soil samples for chemical analyses.

The soil chemical attributes studied were: soil organic carbon (SOC) and total nitrogen (TN) which were measured by dry combustion on a LECO<sup>®</sup> CN-2000 elemental analyzer (furnace at 1350 °C in pure oxygen); available phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), sulfur (S-sulphate), boron (B), cooper (Cu), manganese (Mn), iron (Fe), zinc (Zn), active acidity (pH<sub>CaCl2</sub> 0.01mol L<sup>-1</sup>), potential acidity (H+Al), base saturation (BS) and potential cation exchange capacity (CEC<sub>pH7</sub>) which were measured by analytical methods described in Raij et al. (2001).

#### 2.2.4 Data analyses

The normality of data was confirmed by Shapiro-Wilk's test (p>0.05), using the Statistical Analysis System - SAS v.9.3 (SAS Inc, Cary, USA), therefore no transformation of data was required. The data were analyzed using analysis of variance (ANOVA) using PROC GLM procedure. If the ANOVA F statistic was significant at (p<0.05), the means were compared using Tukey's test (p<0.05) by SAS v.9.3. To analyze the effects within each site, means were compared within each site, and to analyze the overall (regional scale) effects, means were compared considering each site as a block. The three soil depths were analyzed separately. A Pearson's correlation analysis (p<0.01 and p<0.05) was performed using PROC CORR procedure among all soil chemical attributes.

#### 2.3 Results

#### 2.3.1 Soil organic carbon and total nitrogen

The LUC to support sugarcane expansion induced significant SOC losses in the three study sites (Figure 5). At Lat\_17S and Lat\_21S the conversion from native vegetation to pasture resulted in SOC reductions in all soil layers, representing losses of approximately 40% in the 0-30 cm layer. In contrast, short-term (<5 years) conversion from pasture to sugarcane did not induced significant SOC changes (Figure 5A,C,E). At Lat\_23S, the SOC contents in natural ecosystem is higher than others study sites, averaged 37, 33 and 30 g kg<sup>-1</sup> for 0-10, 10-20 and 20-30 cm layers. The soils at this site have much higher clay content (Table 1). Conversion from native vegetation to pasture promoted significant SOC changes only in the subsurface layer (20-30 cm) (Figure 5C). After >20 years of conversion from pasture to sugarcane were observed severe SOC losses, decreasing from 36.4 to 18.9 g kg<sup>-1</sup> (0-10 cm), from 27.6 to 18.4 g kg<sup>-1</sup> (10-20 cm) and from 20.6 to 17.3 g kg<sup>-1</sup> (20-30 cm).

The TN response to LUC was similar to that quantified for SOC (Figure 5). At Lat\_17S and Lat\_21S conversion from native vegetation to pasture led to NT losses of approximately 42% and 55%, respectively (0-30 cm layer) (Figure 5 B,D,F). In general, short-term conversion from pasture to sugarcane had no negative impacts on TN, even some increases were found for the subsurface soil layers (10-20 and 20-30 cm) at Lat\_17S. Similar to verified for SOC, long-term conversion from native vegetation to pasture and then from pasture to sugarcane induced significant soil TN losses at Lat\_23S.

In the regional scale (three sites combined), the LUC induced significant SOC and TN depletions (Figure 6). Conversion from native vegetation to pasture decreased 28 and 18% of the SOC and NT contents, while conversion from pasture to sugarcane decreased 18 and 10%, respectively.

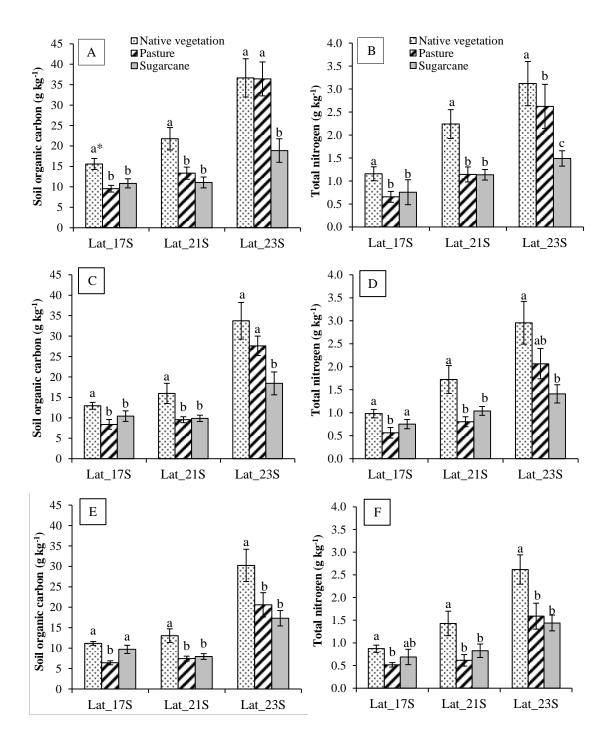


Figure 5 - Soil organic carbon (A;C;E) and total nitrogen (B;D;F) in the 0-10, 10-20 and 20-30 cm layers, respectively, under native vegetation (NV), pasture (PA) and sugarcane (SC) at Lat\_17S, Lat\_21S and Lat\_23S in central-southern Brazil. \*Mean values within each site in same depth followed by the same letter do not differ among themselves according to Tukey's test (*p*<0.05). Error bars denote standard deviation of the mean

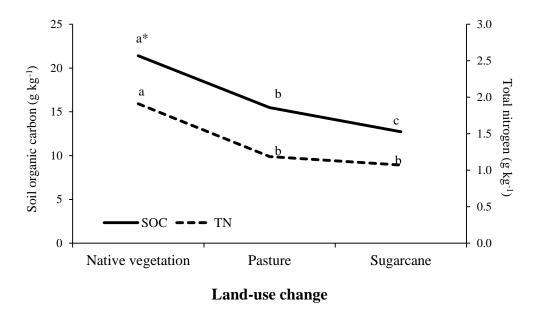


Figure 6 - Soil organic carbon (solid line) and total nitrogen (dashed line) for the 0-30 cm layer under native vegetation (NV), pasture (PA) and sugarcane (SC) in the regional scale in central-southern Brazil. \*Mean values of each element followed by the same letter do not differ among themselves according to Tukey's test (p<0.05). Sites were considered as blocks randomized

#### 2.3.2 Soil acidity attributes and CEC<sub>pH7</sub>

The soil pH measured at each site was lower of ideal range for plants growth (pH < 5.5-6.0) (Table 3). In general, conversion from native vegetation to pasture decreased pH and BS values. At Lat\_21S, the soil had higher pH and BS, and lower H+Al compared to the other sites and land uses (Table 3), most likely due to recent historical forest understory burning, because remains of ash were found on the soil surface. Sugarcane cultivation replacing pasture promoted significant decreases in both the active and potential acidity and consequently, BS was increased. However, in the sugarcane soils still remain acidity problems for crop, since the average pH (0-30 cm) was between 5.0 (Lat\_17S; Lat\_21S) and 5.5 (Lat\_23S), BS values were 45% (Lat\_17S), 60% (Lat\_21S) and 65% (Lat\_23S), and H+Al contents approximately of 30 mmol<sub>c</sub> dm<sup>-3</sup>. At the regional scale, this same response was confirmed, where sugarcane soils have lower acidity and higher BS, pasture soils have the greatest active acidity, and native vegetation sites have the highest potential acidity (soil electric charges are saturated with H<sup>+</sup> and Al<sup>3+</sup>). The LUC led to significant decrease of CEC<sub>pH7</sub> from native vegetation to pasture, but were not observed differences from pasture to sugarcane (Table 3) for all sites.

#### 2.3.3 Soil macro- and micro-nutrients

Overall, higher Ca and Mg contents were found under sugarcane (Table 4) as function of lime application to neutralize soil acidity. Native vegetation at Lat\_21S was an exception, because the effects of forest understory burning and ash (earlier commented) increased the Ca and Mg contents in the surface soil. There were significant improvements in K contents under pasture soils at the Lat\_21S and Lat\_23S compared to native vegetation and sugarcane (Table 4). Overall, K contents under pasture soils were 28 and 30% higher than native vegetation and sugarcane soils for the 0-30 cm layer. Native vegetation and sugarcane showed no significant K differences among themselves.

The lowest available P contents were found at Lat\_17S site (Table 4), where the soil is in more advanced weathering stage [i.e., lower Ki and Kr indexes (Table 1)]. Conversion from native vegetation to pasture significantly decreased available P by 40% at Lat\_17S, 65% at Lat\_21S and 27% at Lat\_23S, with an average of 42% at the regional scale for the 0-30 cm layer. Under sugarcane soils P contents were similar to those under pasture (Lat\_23S and regional scale) or under native vegetation (Lat\_17S and Lat\_21S). There were significant reductions of S-sulphate contents (Table 4) due to conversion from native vegetation to pasture at Lat\_23S and from pasture to sugarcane at the Lat\_21S and Lat\_23S. In contrast, there was no S differences between native vegetation and pasture at Lat\_17S; and sugarcane soil had the highest S values due to gypsum application before sugarcane planting (Table 2).

The soil B contents were negatively affected by the transition from native vegetation to pasture (Table 5). Part of those B losses was recovered by sugarcane cultivation under pasture; however, in all three sites the average B contents were <0.6 mg dm<sup>-3</sup>, level considered as adequate for Brazilian tropical soils (RAIJ et al., 1997). Other micronutrients, Cu, Mn, Fe and Zn (metal ions), in general, showed similar responses relative to impacts caused by LUC (Table 5). Thus, under pasture soils where the pH is lower, there were significant increases in Cu, Mn, Fe and Zn availability. Under sugarcane the concentration of Cu, Mn and Fe decreased, although the contents contained in soil are still considered high, and meet the crop nutritional demand. The Zn contents were <0.5 mg dm<sup>-3</sup> at Lat\_17S site (Oxisol more weathered), below the recommended range (0.6 - 1.0 mg dm<sup>-3</sup>) by Raij et al. (1997), requiring application of this nutrient.

Table 3 - Soil acidity properties and CEC<sub>pH7</sub> from 0-10, 10-20 and 20-30 cm soil layers under native vegetation (NV), pasture (PA) and sugarcane (SC) in central-southern Brazil

Soil	<u> </u>	0 - 10 cm			10 - 20 cm			20 - 30 cm				
properties§	NV	PA	SC	NV	PA	SC	NV	PA	SC			
Lat_17S												
pН	3.77 (±0.12) b*	3.72 (±0.07) b	5.06 (±0.18) a	3.79 (±0.12) b	3.76 (±0.09) b	5.06 (±0.25) a	3.87 (±0.10) b	3.80 (±0.10) b	4.84 (±0.32) a			
H+Al	92.84 (±17.05) a	55.08 (±7.46) b	30.01 (±6.61) c	78.05 (±8.90) a	46.79 (±6.96) b	30.07 (±4.92) c	64.75 (±7.86) a	39.12 (±4.54) b	33.07 (±5.60) b			
BS	8.61 (±2.11) b	8.54 (±1.90) b	50.52 (±10.98) a	6.89 (±1.30) b	7.54 (±2.65) b	49.32 (±9.22) a	6.77 (±1.57) b	9.28 (±4.70) b	40.97 (±9.28) a			
$CEC_{pH7}$	101.46 (±17.81) a	60.24 (±8.14) b	61.63 (±9.33) b	83.80 (±9.16) a	50.53 (±6.71) b	59.69 (±4.97) b	69.42 (±8.07) a	43.06 (±3.61) c	56.03 (±4.43) b			
				Lat	_21S							
pН	6.46 ( $\pm 0.56$ ) a	3.96 (±0.14) c	5.34 (±0.46) b	6.28 (±0.48) a	3.90 (±0.12) c	5.09 (±0.31) b	6.31 (±0.55) a	3.97 (±0.17) c	4.67 (±0.30) b			
H+Al	14.68 (±3.17) c	48.34 (±4.69) a	25.16 (±8.79) b	14.16 (±2.80) c	44.52 (±5.34) a	26.76 (±4.47) b	13.59 (±2.40) c	38.95 (±6.93) a	31.69 (±6.25) b			
BS	88.60 (±4.02) a	26.78 (±5.44) c	69.44 (±10.62) b	87.39 (±3.54) a	22.94 (±6.41) c	62.44 (±7.32) b	85.47 (±4.99) a	27.71 (±6.45) c	53.37 (±7.78) b			
$CEC_{pH7}$	137.61 (±34.22) a	66.03 (±3.90) b	82.85 (±8.93) b	116.21 (±17.65) a	57.67 (±3.43) c	71.82 (±5.62) b	99.42 (±21.23) a	53.58 (±5.23) c	68.11 (±7.80) b			
				Lat	_23S							
pН	3.72 (±0.25) c	4.71 (±0.06) b	5.49 (±0.60) a	3.72 (±0.16) c	4.53 (±0.10) b	5.33 (±0.59) a	3.69 (±0.16) c	4.48 (±0.13) b	5.43 (±0.59) a			
H+Al	152.83 (±41.55) a	52.50 (±6.78) b	29.65 (±10.21) b	156.60 (±36.76) a	55.76 (±8.12) b	35.88 (±14.03) b	152.68 (±34.94) a	53.71 (±11.43) b	33.59 (±14.29) b			
BS	17.04 (±9.09) c	51.84 (±4.55) b	68.62 (±15.08) a	14.73 (±7.27) c	47.39 (±5.22) b	64.68 (±17.37) a	13.52 (±6.50) c	43.35 (±8.47) b	65.45 (±17.53) a			
$CEC_{pH7}$	181.80 (±34.03) a	109.31 (±12.74) b	100.29 (±17.26) b	182.49 (±32.89) a			175.22 (±29.59) a	94.64 (±11.72) b	101.01 (±12.24) b			
	Regional scale											
pН	4.65 b**	4.14 c	5.29 a	4.60 b	4.06 c	5.16 a	4.62 a	4.08 b	4.96 a			
H+Al	86.78 a	52.12 b	28.22 c	82.94 a	49.02 b	30.90 b	77.01 a	43.93 b	32.75 b			
BS	38.08 b	29.14 b	62.64 a	36.34 b	25.96 b	58.81 a	35.25 b	26.78 b	52.79 a			
CEC <sub>pH7</sub>	140.29 a	79.01 b	80.87 b	127.50 a	71.38 b	79.26 b	114.69 a	63.76 b	74.05 b			

 $^{\$}$ pH CaCl<sub>2</sub>: potential of hydrogen in solution of CaCl<sub>2</sub> 0,01 mol L<sup>-1</sup> (1:2,5) – active acidity; H+Al: potential acidity (mmol<sub>c</sub> dm<sup>-3</sup>); BS: base saturation (%); CEC<sub>pH7</sub>: cations exchange capacity potential (mmol<sub>c</sub> dm<sup>-3</sup>); \*Mean values ("n" = 9) and standard deviation between brackets, mean values in line within each depth, followed by the same letter do not differ among themselves according to Tukey's test (p<0.05); \*\*Mean values ("n" = 27) considering the sites as blocks randomized.

Table 4 - Soil macronutrient contents from 0-10, 10-20 and 20-30 cm soil layers under native vegetation (NV), pasture (PA) and sugarcane (SC) in central-southern Brazil

Soil	0 - 10 cm				10 - 20 cm		20 - 30 cm					
macronutrients	NV	PA	SC	NV	PA	SC	NV	PA	SC			
Lat_17S												
Ca (mmol <sub>c</sub> dm <sup>-3</sup> )	4.24 (±1.28) b*	3.02 (±0.78) b	21.74 (±9.32) a	2.63 (±0.63) b	2.18 (±0.80) b	20.19 (±5.29) a	2.14 (±0.67) b	2.29 (±1.04) b	15.66 (±4.02) a			
Mg (mmol <sub>c</sub> dm <sup>-3</sup> )	$3.36 (\pm 0.89) b$	1.62 (±0.49) c	9.26 (±1.93) a	2.33 (±0.62) b	1.07 (±0.32) b	8.86 (±1.87) a	1.83 (±0.42) b	1.17 (±0.85) b	6.86 (±1.34) a			
K (mmol <sub>c</sub> dm <sup>-3</sup> )	1.02 (±0.12) a	0.61 (±0.13) b	0.61 (±0.18) b	$0.78 (\pm 0.09) a$	0.49 (±0.07) b	0.57 (±0.17) b	0.69 (±0.10) a	0.48 (±0.16) b	$0.46 (\pm 0.11) b$			
$P (mg dm^{-3})$	5.56 (±0.86) a	3.02 (±0.37) b	7.31 (±2.46) a	4.53 (±0.37) b	2.58 (±0.25) c	7.00 (±1.91) a	3.51 (±0.35) b	2.47 (±0.39) c	4.67 (±1.19) a			
$S (mg dm^{-3})$	$4.18 (\pm 2.47) b$	3.22 (±1.67) b	9.67 (±3.64) a	2.41 (±1.20) b	2.67 (±0.78) b	14.00 (±4.95) a	2.37 (±1.05) b	2.15 (±0.71) b	26.07 (±9.69) a			
				Lat_21S	5							
Ca (mmol <sub>c</sub> dm <sup>-3</sup> )	101.56 (±34.93) a	8.30 (±1.53) c	39.02 (±8.11) b	82.52 (±19.19) a	6.27 (±1.77) c	29.79 (±5.74) b	69.22 (±21.01) a	8.01 (±1.73) c	24.24 (±6.47) b			
Mg (mmol <sub>c</sub> dm <sup>-3</sup> )	18.62 (±3.58) a	5.21 (±0.99) b	15.59 (±3.87) a	16.66 (±3.01) a	$3.48 (\pm 0.85) c$	12.36 (±2.78) b	14.23 (±2.80) a	3.53 (±0.72) c	9.54 (±1.46) b			
K (mmol <sub>c</sub> dm <sup>-3</sup> )	$2.78 (\pm 0.50) b$	4.17 (±1.53) a	$3.09 (\pm 1.03)$ ab	$2.89 (\pm 0.47) a$	3.41 (±1.11) a	2.91 (±1.03) a	2.39 (±0.67) b	3.08 (±0.83) a	$2.63 (\pm 0.84)$ ab			
$P (mg dm^{-3})$	17.33 (±4.01) a	7.08 (±1.69) b	19.60 (±5.85) a	12.53 (±2.53) a	3.91 (±1.12) b	13.22 (±3.59) a	9.87 (±2.49) a	3.20 (±0.57) b	7.76 (±2.47) a			
$S (mg dm^{-3})$	8.52 (±1.82) a	8.46 (±1.98) a	5.19 (±1.60) b	8.11 (±1.46) ab	8.48 (±2.38) a	6.19 (±1.46) b	7.26 (±1.72) a	9.15 (±2.10) a	7.70 (±3.26) a			
				Lat_23S	5							
Ca (mmol <sub>c</sub> dm <sup>-3</sup> )	17.11 (±6.79) c	32.58 (±6.68) b	47.51 (±17.97) a	15.31 (±8.01) b	29.57 (±5.19) b	48.77 (±18.43) a	12.86 (±5.66) b	24.61 (±5.21) b	47.14 (±17.79) a			
Mg (mmol <sub>c</sub> dm <sup>-3</sup> )	9.06 (±3.41) b	19.88 (±2.67) a	20.06 (±6.98) a	8.14 (±3.05) b	16.16 (±2.10) a	19.32 (±7.02) a	7.38 (±2.51) c	12.27 (±4.34) b	18.30 (±6.32) a			
K (mmol <sub>c</sub> dm <sup>-3</sup> )	2.79 (±1.12) b	4.35 (±0.59) a	3.08 (±1.02) b	2.44 (±0.69) b	4.46 (±0.47) a	$2.32 (\pm 0.82) b$	2.31(±0.69) b	4.05 (±0.63) a	1.98 (±0.73) b			
$P (mg dm^{-3})$	14.29 (±3.26) a	11.47 (±3.72) ab	8.85 (±2.55) b	12.38 (±2.98) a	9.80 (±2.40) ab	8.58 (±2.76) b	10.98 (±2.70) a	7.56 (±2.88) b	7.18 (±2.62) b			
$S (mg dm^{-3})$	15.81 (±3.86) a	9.08 (±3.38) b	5.29 (±1.45) c	16.00 (±3.94) a	7.30 (±3.21) b	$5.19 (\pm 2.01) b$	15.85 (±3.85) a	6.74 (±3.90) b	5.50 (±3.16) b			
	Regional scale											
Ca (mmol <sub>c</sub> dm <sup>-3</sup> )	40.97 a**	14.88 b	35.65 a	33.49 a	12.67 b	32.91 a	28.07 a	11.64 b	28.32 a			
Mg (mmol <sub>c</sub> dm <sup>-3</sup> )	10.34 b	9.05 b	14.77 a	9.04 b	6.90 b	13.51 a	7.81 b	5.66 b	11.31 a			
K (mmol <sub>c</sub> dm <sup>-3</sup> )	2.19 b	3.00 a	2.23 b	2.04 b	2.79 a	1.93 b	1.80 b	2.54 a	1.68 b			
$P (mg dm^{-3})$	12.39 a	7.19 b	12.04 a	9.81 a	5.43 b	9.60 a	8.12 a	4.41 c	6.51 b			
S (mg dm <sup>-3</sup> )	9.51 a	6.86 b	6.77 b	8.84 a	6.15 a	8.46 a	8.49 ab	6.01 b	13.38 a			

<sup>\*</sup>Mean values ("n" = 9) and standard deviation between brackets, mean values in line within each depth, followed by the same letter do not differ among themselves according to Tukey's test (p<0.05); \*\* Mean values ("n" = 27) considering the sites as blocks randomized.

Table 5 - Soil micronutrient contents from 0-10, 10-20 and 20-30 cm soil layers under native vegetation (NV), pasture (PA) and sugarcane (SC) in central-southern Brazil

Soil		0 - 10 cm			10 - 20 cm			20 - 30 cm			
micronutrients	NV	PA	SC	NV	PA	SC	NV	PA	SC		
Lat_17S											
$B (mg dm^{-3})$	0.23 (±0.02) a*	0.15 (±0.01) b	0.15 (±0.02) b	$0.19 (\pm 0.02)$ a	$0.17 (\pm 0.02)$ ab	0.15 (±0.04) b	0.15 (±0.01) a	0.14 (±0.03) a	0.12 (±0.02) a		
Cu (mg dm <sup>-3</sup> )	3.23 (±0.37) a	0.67 (±0.07) b	3.20 (±0.45) a	3.17 (±0.16) a	0.66 (±0.09) b	3.09 (±0.25) a	3.05 (±0.24) a	0.64 (±0.02) b	3.27 (±0.64) a		
Mn (mg dm <sup>-3</sup> )	12.30 (±2.45) a	4.82 (±1.91) b	5.43 (±1.02) b	8.96 (±1.51) a	3.43 (±1.18) b	5.04 (±1.39) b	7.78 (±1.16) a	2.62 (±0.62) b	4.33 (±1.06) b		
Fe (mg dm <sup>-3</sup> )	55.88 (±5.12) b	133.71 (±51.64) a	21.16 (±3.78) b	42.25 (±5.44) b	72.18 (±10.45) a	20.75 (±2.66) c	32.77 (±1.37) b	50.78 (±6.31) a	20.29 (±2.51) c		
Zn (mg dm <sup>-3</sup> )	$0.32 (\pm 0.02)$ a	0.43 (±0.12) a	0.46 (±0.09) a	0.26 (±0.06) a	0.24 (±0.08) a	0.42 (±0.15) a	$0.23~(\pm 0.07)~ab$	0.15 (±0.03) b	0.33 (±0.14) a		
				Lat_2	1S						
$B (mg dm^{-3})$	$0.56 (\pm 0.07)$ a	0.16 (±0.02) c	0.34 (±0.06) b	$0.50 (\pm 0.04) a$	$0.15~(\pm 0.05)~c$	$0.38 (\pm 0.07) b$	$0.41 (\pm 0.02)$ a	0.16 (±0.01) b	$0.40 (\pm 0.07) a$		
Cu (mg dm <sup>-3</sup> )	$0.83 (\pm 0.12) b$	1.25 (±0.12) a	$1.00 (\pm 0.20)$ ab	$0.84 (\pm 0.09) b$	1.33 (±0.19) a	0.96 (±0.06) b	$0.85 (\pm 0.07) b$	1.15 (±0.20) a	$0.93 (\pm 0.04)$ ab		
Mn (mg dm <sup>-3</sup> )	34.08 (±4.99) a	15.78 (±5.90) b	16.43 (±2.39) b	34.16 (±4.21) a	13.29 (±2.45) b	15.67 (±2.65) b	29.44 (±2.30) a	13.71 (±3.49) b	17.44 (±2.96) b		
Fe (mg dm <sup>-3</sup> )	17.25 (±2.81) b	241.38 (±70.77) a	54.93 (±17.69) b	$15.16 (\pm 0.61) b$	164.98 (±31.59) a	52.42 (±4.89) b	12.56 (±0.92) b	88.14 (±31.77) a	46.84 (±5.15) b		
Zn (mg dm <sup>-3</sup> )	2.79 (±0.48) a	1.77 (±0.19) a	2.10 (±0.61) a	2.37 (±0.57) a	1.21 (±0.26) b	1.32 (±0.20) b	1.57 (±0.35) a	0.82 (±0.40) b	0.82 (±0.11) b		
				Lat_2	3S						
$B (mg dm^{-3})$	$0.55 (\pm 0.07)$ a	$0.26 (\pm 0.13) b$	$0.39 (\pm 0.04)$ ab	$0.59 (\pm 0.08)$ a	$0.34 (\pm 0.05) b$	0.33 (±0.06) b	$0.53 (\pm 0.03) a$	0.29 (±0.06) b	$0.20 (\pm 0.07) b$		
Cu (mg dm <sup>-3</sup> )	$1.64 (\pm 0.25) b$	$2.45 (\pm 0.35) a$	1.30 (±0.12) b	$1.59 (\pm 0.20) b$	$2.33 (\pm 0.10)$ a	1.22 (±0.06) c	$1.59 (\pm 0.22)$ ab	$2.19 (\pm 0.30)$ a	1.15 (±0.19) b		
Mn (mg dm <sup>-3</sup> )	49.81 (±11.31) ab	102.41 (±39.58) a	19.47 (±5.44) b	43.72 (±10.64) b	102.32 (±22.28) a	14.11 (±6.61) b	42.82 (±12.39) b	96.89 (±31.39) a	11.41 (±5.65) b		
Fe (mg dm <sup>-3</sup> )	91.06 (±32.06) a	123.93 (±16.27) a	24.79 (±10.02) b	92.53 (±18,61) a	81.62 (±19.53) a	20.75 (±4.79) b	78.91 (±14.50) a	65.42 (±13.01) a	17.49 (±5.10) b		
$Zn (mg dm^{-3})$	2.70 (±1.30) b	6.23 (±1.62) a	$0.85 (\pm 0.05) b$	2.41 (±1.19) a	3.36 (±0.79) a	0.69 (±0.10) b	2.07 (±1.39) a	2.72 (±1.28) a	$0.85 (\pm 0.47) a$		
Regional scale											
$B (mg dm^{-3})$	0.44 a**	0.19 c	0.28 b	0.42 a	0.22 b	0.29 b	0.36 a	0.20 b	0.25 b		
Cu (mg dm <sup>-3</sup> )	1.90 a	1.46 a	1.88 a	1.86 a	1.44 a	1.75 a	1.83 a	1.33 a	1.84 a		
Mn (mg dm <sup>-3</sup> )	32.06 ab	41.00 a	13.26 b	28.95 ab	39.68 a	11.60 b	26.68 ab	37.74 a	11.03 b		
Fe (mg dm <sup>-3</sup> )	54.73 b	166.34 a	34.43 b	49.98 b	106.26 a	31.41 b	41.41 b	68.11 a	29.18 b		
Zn (mg dm <sup>-3</sup> )	1.93 ab	2.81 a	1.16 b	1.68 a	1.60 ab	0.81 b	1.03 a	0.72 ab	0.65 b		

<sup>\*</sup> Mean values ("n" = 4) and standard deviation between brackets mean values in line within each depth, followed by the same letter do not differ among themselves according Tukey's test (p<0.05). \*\* Mean values ("n" = 12) considering the sites as blocks randomized.

## 2.3.4 Correlation among soil chemical attributes

Soil chemical properties were strongly correlated among themselves, as indicated by significant linear correlations for 79 (p<0.01) and 88 (p<0.05) of 120 pairs of soil properties studied (Table 6). The most significant positive correlation was observed for SOC and TN (r = 0.95), with the following regression [SOC = (11.17 x TN) + 1.10 (r<sup>2</sup> = 0.91)]. The SOC and TN were also correlated with the CEC<sub>pH7</sub> (r = 0.83; r = 0.87) and H+Al. In addition, SOC and TN were correlated with exchangeable anions P and S-sulphate, exchangeable cations K, Mg and Ca, and micronutrients Mn, Zn and B.

There were significant correlations between soil chemical properties that influence soil acidity [pH vs BS (r=0.95); pH vs H+Al (r=-0.64); BS vs H+Al (r=-0.63)] and with exchangeable cations, especially Ca and Mg [Ca vs pH (r=0.91); Mg vs pH (r=0.77)]. For the micronutrients, Fe and Cu were negatively correlated with pH and exchangeable cations; Contents of B, Zn and Mn were positively correlated with P and with the exchange cations K, Ca and Mg (Table 6).

Table 6 - Pearson's correlation coefficients (r) and probability of error (p) between soil chemical properties in areas of the land-use change (native vegetation – pasture – sugarcane) in central-southern Brazil

	TN	P	K Past	Ca	Mg	S	рН	BS	H+Al	CEC <sub>pH7</sub>	В	Cu	Fe	Mn	Zn
900	0.955*	0.498*	0.432*	0.210*	0.416*	0.303*	-0.033	0.034	0.596*	0.827*	0.564*	0.126	0.150	0.721*	0.714*
SOC	$< 0.0001^{\dagger}$	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	0.6112	0.6042	< 0.0001	< 0.0001	< 0.0001	0.1967	0.1261	< 0.0001	< 0.0001
TN	1.000	$0.604^{*}$	$0.437^{*}$	$0.338^{*}$	$0.437^{*}$	$0.332^{*}$	0.077	$0.132^{**}$	$0.551^{*}$	$0.873^{*}$	$0.681^{*}$	0.041	0.111	$0.676^{*}$	$0.707^*$
111		< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	0.2418	0.0445	< 0.0001	< 0.0001	< 0.0001	0.6784	0.2638	< 0.0001	< 0.0001
P		1.000	$0.470^{*}$	$0.620^{*}$	$0.643^{*}$	$0.132^{**}$	$0.496^{*}$	$0.560^{*}$	0.043	$0.578^{*}$	$0.746^{*}$	-0.097	-0.046	$0.482^{*}$	$0.631^{*}$
•			< 0.0001	<0.0001	<0.0001	0.0413	<0.0001	<0.0001	0.5052	<0.0001	<0.0001	0.3216	0.6420	<0.0001	<0.0001
K			1.000	0.314*	0.500*	0.022	0.193*	0.365*	-0.041	0.284*	0.455*	-0.220**	0.331*	0.675*	0.648*
				<0.0001	<0.0001	0.7306	0.0027	<0.0001	0.5230	<0.0001	<0.0001	0.0234	0.0005	<0.0001	<0.0001
Ca				1.000	0.783* <0.0001	-0.008 <i>0.9034</i>	0.907* <0.0001	0.874* <0.0001	-0.430* <0.0001	0.365* <0.0001	0.564* <0.0001	-0.302* 0.0016	-0.434* <0.0001	$0.257^* \ 0.0079$	0.358 <sup>*</sup> 0.0002
					1.000	0.9034	0.766*	$0.844^*$	-0.326*	0.371*	0.489*	-0.163	-0.305*	$0.0079$ $0.465^*$	0.0002 $0.503^*$
Mg					1.000	0.5258	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.0940	0.0015	<0.0001	<0.0001
~						1.000	-0.010	0.019	$0.280^{*}$	0.287*	0.161	0.286*	0.030	$0.222^{**}$	0.234**
S							0.8793	0.7720	< 0.0001	< 0.0001	0.1017	0.0030	0.7571	0.0223	0.0157
»II							1.000	$0.954^{*}$	-0.638*	0.085	$0.314^{*}$	-0.187	-0.521*	0.075	$0.195^{**}$
pН								< 0.0001	< 0.0001	0.1883	0.0011	0.0545	< 0.0001	0.4426	0.0455
BS								1.000	-0.631 <sup>*</sup>	0.090	$0.394^{*}$	-0.238**	-0.441*	$0.204^{**}$	$0.295^{*}$
Do									< 0.0001	0.1649	<0.0001	0.0142	<0.0001	0.0363	0.0021
H+Al									1.000	0.676*	0.295*	0.175	0.324*	0.208**	0.164
										<0.0001	0.0023	0.0729	0.0007	0.0327	0.0939
$CEC_{pH7}$										1.000	0.780* <0.0001	-0.051 0.6066	0.012 <i>0.9019</i>	0.484* <0.0001	0.509* <0.0001
•											1.000	-0.316*	-0.173	$0.362^*$	$0.419^*$
В											1.000	0.0010	0.0770	0.0001	<0.0001
_												1.000	-0.144	0.124	-0.013
Cu													0.1422	0.2068	0.8982
T.													1.000	0.165	$0.264^{*}$
Fe														0.0902	0.0063
Mn														1.000	$0.843^{*}$
14111															< 0.0001

<sup>\*</sup>Pearson's correlation coefficients significant (p<0.01) and \*\* (p<0.05). The correlations between micronutrients and other properties were calculated using "n"= 243. †Probability of error (p)

#### 2.4 Discussion

## 2.4.1 Land-use change effects on soil organic carbon and total nitrogen

Soil organic carbon and TN losses induced by long-term conversion from native vegetation to extensive pasture in tropical soils have been consistently reported in the literature (MURTY et al., 2002; DON; SCHUMACHER; FREIBAUER, 2011; ASSAD et al., 2013; MELLO et al., 2014). The key factors of these SOC and TN losses are the following: i) cutting and burning of native vegetation at the time of pasture establishment, where CO<sub>2</sub> and N volatile forms losses may occur through several mechanisms due to combustion (JUO; MANU, 1996); ii) exposure and SOM respiration due to soil disturbance by tillage (FELLER; BEARE, 1997; SIX; ELLIOTT; PAUSTIAN, 2000); iii) soil erosion and runoff (FELLER; BEARE, 1997; ZHANG; WANG; LI, 2015) and mineral N leaching (McGRATH et al., 2001); iv) low productivity of degraded pasture associated with continuous grazing and reduced organic litter inputs to the soil (MAIA et al., 2009; ASSAD et al., 2013; CARVALHO et al., 2014), typical characteristics of the Brazilian pasturelands as those here studied.

Impacts of LUC from pasture to sugarcane on SOC and NT are still little reported in the literature (ROSSI et al., 2013; MELLO et al., 2014). Our findings showed that a short-term (<5 years) transition from pasture to sugarcane, as observed at the Lat\_17 and Lat\_21S (Table 2; Figure 4) did not promote SOC losses for the 0-30 cm layer. On the other hand, long-term (>20 years) sugarcane production led to important SOC losses at Lat\_23S. This SOC depletions can be associated to several causes, such as soil tillage performed for planting, fertilization and sugarcane reformation (MELLO et al., 2014), long period of time (~10 years) with burning sugarcane harvesting system (CERRI et al., 2011; BORDONAL; FIGUEIREDO; LA SCALA JR, 2012; BRANDANI et al., 2015) and the recent crop residue removal for electricity production (CARVALHO et al., 2013). At Lat\_17S were observed higher SOC and TN content under sugarcane than pasture. These results are due to the effects of LUC, but also influenced by differences in soil texture (Table 1) between pasture area (lower clay content) and the others land uses of the same chronosequence. Clay content is one of the major controlling factors to protect, stabilize and storage organic C in the tropical soils (FELLER; BEARE, 1997).

Our findings showed SOC and NT losses in regional scale as response of LUC effects. These results agree with results reported in a global meta-analysis conducted by Don, Schumacher and Freibauer (2011), and measured effects reported by Assad et al. (2013) and Mello et al. (2014). Both SOC and TN are broadly recognized as key indicators of soil quality/health (DORAN; PARKIN, 1994; VEZZANI; MIELNICZUK, 2009; CARDOSO et al., 2013; ZORNOZA et al., 2015); therefore our data suggest that improved management practices are needed to prevent or minimize negative impact of sugarcane production on soil quality and other ecosystem services.

# 2.4.2 Land-use change effects on acidity attributes and CEC<sub>pH7</sub>

The high soil acidity found in all studied sites (Table 3) can be attributed to the natural acidity that is characteristic of highly weathered soils, such as Oxisols and Ultisols in Brazil. These low pH soils are associated with soil acidification processes driven by management practices (ANJOS et al., 2012). In croplands, soil acidity is increased by several factors, such as: uptake of basic cations by crops and removal of these during harvesting; inadequate soil management favoring erosion and exposure of subsurface soil horizons (more acidic); the use of N fertilizers, and through the oxidation of S and SOM (SOUZA; MIRANDA; OLIVEIRA, 2007).

Soil acidity may have been strongly neutralized through native vegetation burning and ash deposition on the soil surface (JUO; MANU, 1996; NGO-MBOGBA; YEMEFACK; NYECK, 2015). It seems to be the major reason of higher pH values in native vegetation at Lat\_21S. However, these effects were not observed after more than 30 years of burning in the conversion from native vegetation to pasture. Thereby, these results would indicate that over time pasture land use reduced pH, BS and the exchangeable cations (Ca and Mg). Soil acidity reductions and increases of Ca and Mg (Tables 3 and 4) observed in sugarcane fields are due to management with applications of lime during the sugarcane conversion, and each subsequent reformation cycle every 5-6 years (Table 2). Therefore, the sugarcane expansion under pasture still requires liming, because sugarcane-growing recommendations states a BS  $\geq$  60% requirement (RAIJ et al., 1997), and in pasture soils (land use prior to sugarcane) these BS values are lower (<10% at Lat\_17S; ~25% at Lat\_21S; ~50% at Lat\_23S).

Soil CEC<sub>pH7</sub> refers to the amount of negative charges at pH 7, which is not affected by addition of lime and fertilizer. Thus, CEC<sub>pH7</sub> is dependent upon the soil mineralogy and SOM content. Tropical soils, as those here studied, are dominated by variable-charge minerals with low CEC. Therefore, SOM is very important for the nutrient status of the soils, since its lower the point of zero charge and increase of CEC (FELLER; BEARE, 1997; ZECH et al., 1997).

It is confirmed in our dataset, where SOC and NT were strongly correlated with  $CEC_{pH7}$ , r = 0.83 (Table 6). Thus, depletions of SOM as response to LUC effects found in these areas (FRANCO et al., 2015) led to reductions of  $CEC_{pH7}$  from native vegetation to pasture and sugarcane.

## 2.4.3 Land-use change effects on macro- and micro- nutrients

The higher K contents were found under pasture soils (Table 4). It could be attributed to the heavy cycling, where up to 90% of the K taken up by plants is returned to the soil through of urine and feces (KAYSER; ISSELSTEIN, 2005) and low K losses from the system (extensive cattle with low stocking rate). The cycling of K in deeper soil layers and release of non-exchangeable K forms by aggressive root system of grasses. The external inputs of K fertilizers to degraded pasture (Lat\_21S) also increase K, in addition to the increases from the mineral animal feed supplements fed to cattle. In the native vegetation and sugarcane soils the K content were not significantly different, demonstrating that the application of mineral K fertilizers and organic residues (rich in K), such as vinasse (CHRISTOFOLETTI et al., 2013) to sugarcane maintains K levels equal to those found under native vegetation, despite significant K export in harvested sugarcane biomass.

Overall, low available P contents were found in all land uses studied (Table 4), characteristic of the tropical soils. According to Novais, Smyth and Nunes (2007) increases in the weathering degree leads to gradual changes in soil characteristics, resulting in increases of positives charge and ability of soil to adsorb and retain anions, such as phosphates. Under these conditions, P is strongly adsorbed (inner sphere complex) in Fe and Al oxides and kaolinites (FONTES; WEED, 1996; NOVAIS; SMYTH; NUNES, 2007), which is the predominant clay mineralogy in these study sites.

The LUC from native vegetation to pasture promoted available P contents depletions (Table 4). Recently a large regional survey in Brazil also concluded that LUC from native vegetation to extensive pasture led to decreases of available P stocks in the soil (GROPPO et al., 2015). This decrease may be associated with P exploitation over time by grazing cattle and absence of P fertilizer inputs. Several studies state that P plant-availability in pasture is strongly dependent on the mineralization of the P organic pools (AGUIAR et al., 2013; FONTES et al., 2014; NASH et al., 2014). Therefore, the pronounced SOM depletions in these sites (Figures 5 and 6), can be explains the severe degradation process of the Brazilian pasturelands. Available P increases in the transition from pasture to cropland (sugarcane in

this study) is frequently reported in the literature (NEGASSA; LEINWEBER, 2009), as response to application of mineral phosphate fertilizers. However, despite this increase (Table 4), the amount of P available to support sugarcane growth was not enhanced, since the levels were lower than the recommended 15 mg kg<sup>-1</sup> critical level (RAIJ et al., 1997).

Available P depletion observed in sugarcane soil at Lat\_23S may be due to application of only organic residues (filtercake, boiler ash and vinasse) from sugarcane industry (Table 2). These organic residues could be releasing available P to plants below that is removed in the harvested biomass, and probably, accumulating P in other organic and/or inorganic fractions that are less available in the soil. Therefore, future studies fractionating P forms will be crucial for understanding LUC impacts on plant-available P dynamics in the soil, and perhaps, identify indicator fractions for these effects (AGUIAR et al., 2013).

S-sulphate content depletions from native vegetation to pasture and from pasture to sugarcane (Table 4) are associated with i) native vegetation burning at the moment of conversion, where losses occur by volatilization of S (JUO; MANU, 1996), ii) reduction of organic matter inputs in the soil iii) SOM decreases with more intensive soil tillage in sugarcane fields. Practically the entire available S-sulphate in the soil comes from biological process of mineralization of SOM (ALVAREZ et al., 2007). Thus, SOM changes have a direct effect on the availability of S in the soil (KIRKBY et al., 2011). The SOC and NT losses induced by the LUC confirm the close linkage between S-sulphate and SOM.

Soil micronutrient availability is affected by several factors, such as: soil acidity; SOM; P availability; moisture, texture, soil mineralogy (Fe, Al and Mn oxides), land management and fertilization (WEI et al., 2006; ABREU; LOPES; SANTOS, 2007; SARKAR et al., 2014). Except the soil mineralogy and texture, these other factors are intensively affected by LUC process, modifying the availability of soil micronutrients to plants. The associated decrease in SOM in these areas (Figures 5 and 6) could be driving our observed decrease in B (Table 5), which has been shown to correlate with SOM, Fe and Mn oxides (SARKAR et al., 2014). The Cu, Mn, Fe and Zn responses to the LUC, with higher contents in pasture soils (Table 5), most likely is due to acidification process in these soils, which increase strongly their soil availability (ABREU; LOPES; SANTOS, 2007). The relationship between these micronutrients and acidity attributes was confirmed in this study (Table 6).

## 2.4.4 Soil chemical quality and its implications for sugarcane expansion

Soil chemical quality assessments are crucial to quantify the soil's capacity to provide nutrients for adequate plant growth. Our findings suggest that LUC promoted significant effects on soil chemical properties (Figure 7) and consequently on soil chemical quality. Summarily, conversion from native vegetation to pasture promoted the following effects: i) high depletions of soil attributes related to SOM content (i.e., SOC, TN, P, S, B and CEC<sub>pH7</sub>); ii) soil acidification process with decrease of pH, BS and exchange cations (Ca and Mg); iii) increase of metallic micronutrients availability (i.e., Cu, Mn, Fe and Zn). Therefore, conversion from native vegetation to pasture reduced the soil's capacity to support plant growth and productivity over time.

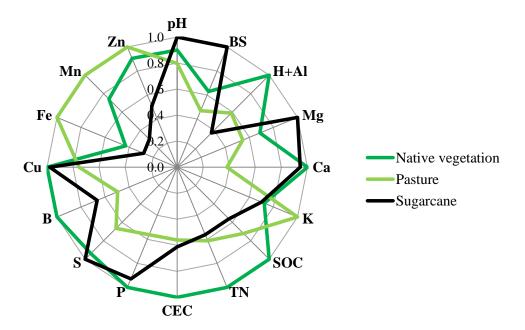


Figure 7 - Scores of the soil chemical attributes (0-30 cm layer) under land-use change (native vegetation - pasture - sugarcane) in central-southern Brazil. Mean values were relativized and transformed in scores ranging from 0 to 1, so for each soil attribute the land use that had the highest value received score 1 and other land uses received proportional values

Conversion from pasture to sugarcane resulted in a partial recovery of soil chemical quality on a relatively short timescale (<5 years) due to the successive applications of lime and fertilizers. Despite that, increments of available P and soil correction to BS  $\ge$ 60% are necessary to achieve ideal soil chemical conditions to sugarcane growth. Although, there was a decrease in soil micronutrient availability, the amount of available micronutrients to support plant growth was enhanced. Overall the LUC from pasture to sugarcane results in SOM

losses, as also reported by Mello et al. (2014). However, the payback time for these losses is short and overall it delivers substantial greenhouse gas emission savings when sugarcane ethanol is used to displace fossil based fuels (EGESKOG et al., 2014; MELLO et al., 2014). Based on our findings that indicated SOC losses in regional scale as response of LUC effects, associated to strong relationship among SOC with other soil chemical properties verified in this study, we conclude that SOC is an important indicator of soil degradation and could be useful for assessing soil quality in the sugarcane fields in Brazil.

Therefore, our study indicated that management strategies are necessary to maintain and/or improve the soil chemical quality and the sustainability of sugarcane production in Brazil. Mechanized harvesting associated with reduced tillage and maintenance of sugarcane straw on soil surface are key-factors to increase soil C sequestration, nutrient-cycling and improve soil quality in sugarcane fields (CERRI et al., 2011; BORDONAL; FIGUEIREDO; LA SCALA JR, 2012; BRANDANI et al., 2015). In addition, fertilization using organic residues from sugarcane industry (e.g., vinasse and filter cake) could be a feasible alternative to increase SOM (BRANDANI et al., 2015) and provide nutrients, especially K and P (CHRISTOFOLETTI et al., 2013; PRADO; CAIONE; CAMPOS, 2013) reducing the production costs with mineral fertilizers (ALMEIDA JUNIOR et al., 2011; SILVA; BONO; PEREIRA, 2014).

## 2.5 Conclusions

Our findings showed that long-term LUC from native vegetation to extensive pasture decreases SOC, TN, available P, S, Ca, Mg and B contents. In addition, the LUC induces to soil acidification and decrease of CEC<sub>pH7</sub>, indicating that pasture areas have poor soil chemical quality in the primary sugarcane producing region of Brazil.

Conversion from pasture to sugarcane leads to increase of macronutrients levels and reduction of soil acidity due to inputs of lime and fertilizers, improving soil fertility. Despite that, increments of available P and BS are necessary to achieve ideal soil chemical conditions to sugarcane growth. Decreases of available metallic micronutrients as response to soil acidity correction in sugarcane fields are not limiting to plants growth. Short-time (<5 years) conversion from pasture to sugarcane had no significant impacts on SOC and NT contents; however, after 20 years of sugarcane production significant losses were quantified. In regional scale, conversion from pasture to sugarcane induced 18 and 10% losses of the SOC and NT, respectively, for the 0-30 cm layer.

Overall, it is expected that sugarcane expansion in Brazil replacing areas currently occupied with pastures will promote improvement on soil chemical quality. Despite this, sugarcane expansion can be associated with management strategies to increase SOM and improve the soil fertility, reducing the environmental and economic costs associated with ethanol production in Brazil.

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# 3 PHOSPHORUS POOLS RESPONSES TO LAND-USE CHANGE FOR SUGARCANE EXPANSION IN WEATHERED BRAZILIAN SOILS

#### **Abstract**

Without proper management, land-use change (LUC) associated with producing sugarcane (Saccharum officinarum) for bioenergy in Brazil can lead to soil degradation and have negative implications on ecosystem functions. Phosphorus (P) depletion is one of the most frequent causes of land degradation in tropical environments, and as such, soil P pools have been identified as potential indicators of negative environmental impacts. We quantified soil P dynamics for the most common LUC sequence in sugarcane expansion areas (i.e., native vegetation to pasture to sugarcane), to determine if and how these changes could be used to evaluate environmental impacts of LUC in weathered Brazilian soils. Soil samples were collected from three areas in central-southern Brazil, representing the primary sugarcane-producing region of the world. Soil P fractionation was performed, and P stocks were calculated for the surface 30 cm. Soil chemical attributes and macrofauna data were correlated with P pools. Long-term conversion from natural ecosystems (Cerrado and Atlantic forest biome) to extensive pasture decreased total P stocks by 31.2% indicating progressive soil degradation in these areas. In contrast, the LUC from pasture to sugarcane increased total P stocks by 35.6%; nevertheless, fertilization management altered the soil P-cycle, causing P accumulation in less plant-available forms. Available P increases to support adequate sugarcane growth are still needed. Applying P using organic residues increased labile organic P and may be a complementary strategy for increasing nutrient supplies in sugarcane fields. Phosphorus availability showed significant positive correlations with other soil chemical properties and clay content, while the functional diversity of soil macrofauna was strongly correlated with labile and biological P. We conclude that P pools can be useful indicators for assessing LUC modifications on soil quality in the tropics, and recommend they be used to assess land degradation and environmental sustainability within sugarcane expansion areas in Brazil.

Keywords: Sugarcane production; Environmental sustainability; Phosphorus fractionation; Soil quality indicators; Soil macrofauna

## 3.1 Introduction

The global expansion of bioenergy crops has brought attention to the potential environmental impacts resulting from land-use changes (LUC) (ROWE; STREET; TAYLOR, 2009; WRIGHT; WIMBERLY, 2013; MUKHERJEE; SOVACOOL, 2014; WALTER et al., 2014; ANAYA; HUBER-SANNWALD, 2015). In Brazil, the largest sugarcane ethanol producer in the world, the area growing sugarcane expanded by 3.2 Mha between 2005 and 2015, totalizing approx. 9.1 Mha (COMPANHIA BRASILEIRA DE ABASTECIMENTO, 2015). However, Brazil will need an additional 6.4 Mha of sugarcane to meet domestic ethanol demand in 2021 (GOLDEMBERG et al., 2014). This additional area will primarily

come from areas that are currently in pasture (LAPOLA et al., 2010; GOLDEMBERG et al., 2014; WALTER et al., 2014).

Soil quality has been identified as a key environmental component for evaluating the sustainability of biofuel expansion (BETTER SUGAR CANE INITIATIVE, 2011; GLOBAL BIOENERGY PARTNERSHIP, 2011). Studies have been conducted to identify soil properties and processes that are suitable for evaluating soil quality, soil degradation and their impact on ecosystem services. Soil organic matter (SOM) depletion (DON; SCHUMACHER; FREIBAUER, 2011; MELLO et al., 2014; FRANCO et al., 2015) and loss of soil nutrients such as phosphorus (P) associated with the SOM (MacDONALD; BENNETT; TARANU, 2012; AGUIAR et al., 2013; HAMER et al., 2013; FONTE et al., 2014; FRANCO et al., 2015; NESPER et al., 2015) have been appointed as important indicators of soil quality degradation. Phosphorus is an important indicator, especially in tropical regions, because it is a major limiting factor for plant growth (ELSER et al., 2007; SHEN et al., 2011; ÅGREN; WETTERSTEDT; BILLBERGER, 2012; ELSER, 2012).

Soil macroinvertebrates, such as beetles, termites and earthworms can improve the availability of essential nutrients to plants in tropical soils (OUÉDRAOGO et al., 2005; CHAPUIS-LARDY et al., 2011; SEYMOUR et al., 2014). Studies have shown that macrofauna can significantly change the biogeochemical P cycle in the soil, usually leading to higher soil P availability (LE BAYON; BINET, 2006; CHAPUIS-LARDY et al., 2011). Therefore, LUC processes that induce deleterious effects on soil biodiversity probably also have adverse implications on P availability. Developing a better understanding of soil P dynamics in tropical soils undergoing LUC is necessary, not only to improve P-use efficiency, but also to reduce environmental risks (SHEN et al., 2011; MacDONALD et al., 2011; ELSER, 2012; STUTTER et al., 2015).

Soil P is present in organic and inorganic forms, ranging from ionic forms in solution to highly stable compounds with SOM and/or clayey minerals (NEGASSA; LEINWEBER, 2009; SHEN et al., 2011). Organic P (Po) is associated with soil microbial biomass and SOM, and generally includes compounds such as phosphomonoesters, phosphodiesters and organic polyphosphates (NASH et al., 2014; STUTTER et al., 2015). These compounds vary in P availability based on the degree of recalcitrance. Soil inorganic P (Pi) is primarily linked to amorphous and crystalline forms of iron (Fe), aluminum (Al) and calcium (Ca), and account for a large proportion of total soil P in tropical soils (FONTES; WEED, 1996; NOVAIS; SMYTH; NUNES, 2007; GAMA-RODRIGUES et al., 2014). Unfortunately, much of this P is unavailable for plant uptake.

The distribution of P between organic and inorganic compounds varies, reflecting soil use and composition of both natural ecosystems and agroecosystems (WRIGHT, 2009; AGUIAR et al., 2013; CREWS; BROOKES, 2014). Therefore, P fractionation and grouping into chemically defined pools, such as labile, moderately labile and non-labile (organic and inorganic) is useful for quantifying the fate of native and applied P in both systems (CROSS; SCHLESINGER, 1995; NEGASSA; LEINWEBER, 2009; WRIGHT, 2009; RISKIN et al., 2013; CREWS; BROOKES, 2014). Similarly, the distribution and dynamics of P pools can also be used to predict the effects of LUC on soil quality and to determine potential limitations when establishing new land uses (AGUIAR et al., 2013).

In a recent study, the microbe and plant-available P pool was not significantly affected by LUC in sugarcane expansion areas (FRANCO et al., 2015). The findings of Franco et al. (2015) also suggest that quantifying only the labile forms of P may not be suitable for assessing LUC effects on P dynamics in tropical soils, and that less labile forms of P can better elucidate LUC effects. Our objective was to quantify effects of the most common LUC sequence associated with sugarcane expansion (i.e., native vegetation to pasture to sugarcane), on soil P dynamics and thus determine environmental impacts of LUC on weathered Brazilian soils. We hypothesized that LUC from natural to anthropogenic ecosystems would result in soil P-pool modifications that could indicate soil degradation.

## 3.2 Material and Methods

## 3.2.1 Study sites and experimental design

The study sites and experimental design were described in the 2.2.1 and 2.2.2 items.

## 3.2.2 Soil sampling and phosphorus fractionation

Soil sampling was completed in January 2013. At each land-use four repetitions consisting of 12 subsamples each were collected from the 0-10, 10-20 and 20-30 cm depth around the center point using a Dutch auger. Samples were sieved through a 2-mm screen and dried at 50 °C prior to P determinations. Other soil chemical properties and particle sizes were analyzed and correlated with the P pools. Additionally, undisturbed soil samples were collected to measure soil bulk density using a metal cylinder (height 5 cm x internal diameter

5 cm, approx. 100 cm<sup>-3</sup>). Those values were subsequently used to calculate the P stocks for each layer.

Phosphorus pools were obtained by measuring inorganic P (Pi) and organic P (Po) as described by Hedley, Stewart and Chauhan (1982) and subsequently modified by Condron, Goh and Newman, (1985). Phosphorus was extracted sequentially from dry 0.5-g soil samples in the following order: anion exchange resin membrane (Pi<sub>resin</sub> fraction), 0.5 M of sodium bicarbonate (NaHCO<sub>3</sub>) (Pi<sub>bic</sub> and Po<sub>bic</sub> fractions), 0.1 M sodium hydroxide (NaOH) (Pi<sub>hyd01</sub> and Po<sub>hyd01</sub> fractions), 1.0 M of chloridric acid (HCl) (Pi<sub>HCl</sub> fraction) and 0.5 M of NaOH (Pi<sub>hyd05</sub> and Po<sub>hyd05</sub> fractions). The remaining soil was oven dried and digested with  $H_2SO_4 + H_2O_2$  (P<sub>residual</sub> fraction) to determine residual P. Phosphorus concentrations in the acid extracts were determined using the Murphy and Riley (1962) method. Inorganic P (Pi) fractions in the alkaline extracts (NaHCO<sub>3</sub> and NaOH) were determined using the Dick and Tabatabai (1977) method. After determining total P in the alkaline extracts by digesting with ammonium persulfate  $+ H_2SO_4$  in an autoclave, organic P (Po) was estimated by calculating the difference between total P and Pi in the various fractions.

Phosphorus fractions obtained by the Hedley fractionation were grouped into pools according to their plant availability as "Labile P" ( $Pi_{resin} + Pi_{bic} + Po_{bic}$ ), "Moderately labile P" ( $Pi_{hyd01} + Po_{hyd01} + Pi_{HCl}$ ) and "Non-labile P" ( $Pi_{hyd05} + Po_{hyd05} + P_{residual}$ ). In addition, P fractions were grouped as proposed by Cross and Schlesinger (1995) into biological and geochemical P pools, where the former includes all organic fractions ( $Po_{bic} + Po_{hyd01} + Po_{hyd05}$ ) and the latter includes all inorganic fractions and residual P ( $Pi_{resin} + Pi_{bic} + Pi_{HCl} + Pi_{hyd01} + Pi_{hyd05} + P_{residual}$ ).

## 3.2.3 Soil attributes and macrofauna variables

A dataset of soil chemical attributes and clay content provided by Cherubin et al.  $(2015)^1$  was used to verify their correlation with soil P pools. Those measurements were made on soil samples collected at the same sites and sampling times here reported for P fractionation.

The role of soil macroinvertebrates in mediating changes in P pools under the LUC scenarios was studied correlating data of soil P pools with data of macrofauna variables provided by Franco (2015) at the same site and time studied here. Briefly, at each soil

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<sup>&</sup>lt;sup>1</sup> Data presented in the chapter 2.

sampling point macrofauna numbers were determined using the standard Tropical Soil Biology and Fertility Institute soil monolith method (ANDERSON; INGRAM, 1993). Soil monoliths (25 x 25 x 10 cm) were extracted from 0-10, 10-20 and 20-30 cm soil layers and the macrofauna were sorted immediately after sampling. The faunal density was calculated as the number of individuals per surface unit (m²). Ecological indexes were calculated for assessing richness (Margalef's index), diversity (Shannon's index), evenness (Pielou's index) and dominance of species (Simpson's index) according to the methods described by Magurran (2004). The functional diversity of sampled macrofauna was determined through of taxa classification according to their ecological functions. This included species considered to be herbivorous (Dermaptera and Hemiptera), detritivorous (Blattodea, Diplopoda, Diptera, Gastropoda, Isopoda, Isoptera and Oligochaeta) or predators (Aranae, Chilopoda, Coleoptera, Formicidae, Hymenoptera and Scorpiones) (BROWN et al., 2001; JEFFERY et al., 2010).

## 3.2.4 Data analyses

The P stocks were calculated for each sampling point by multiplying P concentrations by soil bulk density and layer thickness (10 cm) before averaging the values for each site. Phosphorus stocks within pasture and sugarcane sites were adjusted to an equivalent soil mass corresponding native vegetation to measure effects of LUC on soil bulk density (LEE et al., 2009). An analysis of variance (ANOVA) was computed using PROC GLM procedure to test the influence of the LUC within each site on P pools. If the ANOVA was significant (p<0.05), the means were compared using Tukey's test (p<0.05). An additional ANOVA was computed to test the LUC effects in regional scale (global average at the results found at the three locations). In this case, each site was considered a block. Means were also compared using Tukey's test (p<0.05). A Pearson's correlation analysis (p<0.01 and p<0.05) was performed using PROC CORR procedure among P pools, soil chemical attributes and clay content. A Principal Component Analysis (PCA) was also performed using PROC FACTOR procedure to visualize the relationship between P pools and soil macrofauna variables. All statistical analyses were completed using the Statistical Analysis System – SAS v.9.3 (SAS Inc., Cary, USA).

## 3.3 Results

## 3.3.1 Dynamic of P fraction levels under land-use change

Soil P fractions in native vegetation, pasture and sugarcane land uses at the three sites are presented in Table 1. Although labile P fractions showed site-specific responses, there was an increasing trend within these fractions in sugarcane fields at Lat\_17S and Lat\_21S, presumably in response to mineral phosphate fertilization. However, P<sub>resin</sub> levels, the plant-available fraction used as a diagnostic for soil fertility, were very low, averaged 4.21, 8.63 and 3.64 mg kg<sup>-1</sup> for the 0-30 cm soil layer at Lat\_17S, Lat\_21S and Lat\_23S, respectively. Furthermore, this fraction represented between 0.4 to 2.0% of the total soil P. At Lat\_23S there was much less inorganic P in labile fractions (P<sub>resin</sub> and Pi<sub>bic</sub>) within sugarcane soil than within native vegetation or pasture soils (Table 1). However, use of organic wastes as fertilizers for sugarcane production increased the Po<sub>bic</sub> fraction (labile pool), which was higher than in native vegetation and pasture, but does not ensure adequate soil P<sub>resin</sub> levels for the sugarcane crop.

Long-term conversion from native vegetation to extensive pasture depleted soil P levels, especially for moderately labile ( $Pi_{hyd01}$ ,  $Po_{hyd01}$  and  $Pi_{HCl}$ ) and non-labile ( $Pi_{hyd05}$ ,  $Po_{hyd05}$  and  $P_{residual}$ ) P fractions; this depletion occurred at both Lat\_17S and Lat\_21S sites (Table 1). Sugarcane cultivation increased total P to levels similar or higher than those within native vagetation as a consequence of successive mineral and organic P fertilizer inputs. Fertilizer P in sugarcane areas was distributed among all P fractions, but the main effect was to mineral-associated fractions (inorganic P) which are strongly adsorbed and have low plant-availability (moderately and non-labile P fractions).

Table 1 - Phosphorus fraction levels (mg kg<sup>-1</sup>) under land-use change (native vegetation - NV, pasture - PA and sugarcane - SC) in central-southern Brazil

	Soil		Lat_17S		CV		Lat_21S		CV		Lat_23S		
P Fractions	Layer	NV	PA	SC	CV% -	NV	PA	SC	CV% —	NV	PA	SC	$\text{CV}_{\%}$
					La	abile P fracti	ons						
	0-10	3.85 b*	6.77 a	4.08 ab	29	9.19 b	6.01 b	13.34 a	17	11.64 a	10.73 a	3.88 b	29
P_resin	10-20	3.70 a	4.77 a	4.99 a	32	10.39 a	4.09 b	6.82 ab	29	8.01 a	7.25 a	3.78 b	37
	20-30	2.57 b	4.21 a	3.55 ab	19	7.16 a	2.61 b	5.73 a	24	7.03 a	5.59 a	3.25 b	21
	0-10	20.77 a	5.25 b	21.45 a	12	7.05 b	7.00 b	12.57 a	21	20.65 a	25.19 a	9.70 b	17
Pi_bic	10-20	18.04 a	3.94 b	21.44 a	17	4.51 b	3.72 b	6.89 a	21	22.24 a	21.56 a	9.02 b	20
	20-30	16.85 b	3.69 c	19.91 a	7	4.67 a	2.79 b	5.25 a	22	23.83 a	19.91 a	7.15 b	10
	0-10	1.85 c	27.94 a	9.41 b	36	32.54 a	33.54 a	38.87 a	19	10.74 b	10.72 b	23.82 a	21
Po_bic	10-20	5.00 b	25.74 a	9.42 b	21	33.57 a	26.28 a	38.16 a	19	17.66 b	3.85 c	25.56 a	19
	20-30	8.34 a	9.52 a	10.41 a	39	33.80 a	27.95 a	32.67 a	26	8.09 b	12.01 b	21.05 a	29
					Moderat	tely Labile P	fractions						
	0-10	58.06 a	0.90 b	62.33 a	10	32.46 a	37.07 a	28.14 a	14	59.85 b	68.04 ab	83.37 a	12
Pi_hyd0.1	10-20	53.30 b	0.70 c	72.06 a	18	24.27 b	34.74 a	21.89 b	16	58.81 b	58.06 b	86.55 a	18
	20-30	55.98 a	0.53 b	62.33 a	11	23.37 a	22.93 a	19.21 a	16	41.98 b	51.21 b	75.18 a	14
	0-10	30.60 b	2.55 c	72.64 a	22	41.05 b	58.85 b	89.13 a	18	183.03 a	99.29 b	42.66 c	28
Po_hyd0.1	10-20	21.02 b	1.56 c	69.56 a	22	79.69 a	30.25 b	99.22 a	26	86.84 a	68.68 a	24.69 b	29
	20-30	18.55 b	1.00 c	57.05 a	28	79.38 a	43.98 b	91.68 a	21	92.88 a	78.16 a	33.60 b	12
	0-10	2.38 b	0.42 c	4.50 a	31	10.13 a	7.93 a	14.17 a	33	7.94 b	14.02 a	14.81 a	25
Pi_HCl	10-20	2.31 b	0.23 c	4.76 a	43	9.99 a	4.23 b	8.17 a	42	7.08 a	10.25 a	11.60 a	26
	20-30	2.12 b	0.21 c	4.03 a	28	7.93 a	3.67 b	6.63 a	22	7.47 a	10.65 a	12.57 a	27
					Non-	Labile P frac	ctions						
Pi_hyd0.5	0-10	62.17 b	0.43 c	74.42 a	8	12.71 a	4.44 b	11.19 a	11	114.50 a	130.49 a	127.96 a	8
_ <b>,</b>	10-20	59.60 a	0.32 b	62.92 a	32	10.13 a	3.63 b	9.48 a	19	100.03 a	102.25 a	90.55 b	5
	20-30	58.08 a	0.20 b	54.91 a	35	8.62 b	6.25 c	12.10 a	13	101.85 b	96.60 b	135.98 a	14
Po_hyd0.5	0-10	29.82 b	63.33 a	73.25 a	17	125.49 a	61.84 b	79.40 b	20	29.08 a	11.42 a	33.17 a	67
	10-20	41.77 b	99.92 a	78.44 ab	30	129.11 a	63.73 b	120.21 a	33	19.97 b	36.64 b	72.40 a	37
	20-30	49.67 b	114.46 a	81.12 ab	31	162.58 a	83.30 b	98.56 b	26	45.32 a	31.93 a	20.96 a	56
	0-10	243.93 b	77.59 b	328.82 a	20	233.51 a	148.94 b	202.70 a	23	654.40 a	536.80 a	667.19 a	18
P_residual	10-20	232.35 b	67.43 b	356.24 a	27	217.93 ab	146.50 b	259.97 a	26	680.19 a	602.61 a	615.20 a	19
I_Iesiaaai	20-30	268.71 a	71.29 b	402.75 a	33	299.37 a	232.62 a	282.31 a	16	690.75 a	582.09 a	592.45 a	19
	20 30	200.71 u	71.270	102.75 a	33	Total P	232.02 u	202.31 u	10	070.73 <b>u</b>	302.07 u	372.13 u	17
	0-10	453.42 b	185.17 с	650.89 a	10	504.11 a	365.61 b	489.52 ab	14	1091.82 a	906.70 a	1006.46 a	11
P_total	10-20	437.09 b	204.60 c	679.83 a	15	519.59 a	317.16 b	570.79 a	11	1000.83 a	911.15 a	939.34 a	14
1_101111	20-30	480.86 b	205.11 c	696.07 a	19	626.91 a	426.09 b	554.14 ab	12	1019.21 a	888.17 a	902.19 a	11

<sup>\*</sup>Mean values in line within each site followed by the same letter do not differ among themselves according to Tukey's test (p<0.05).

## 3.3.2 Labile, moderately labile and non-labile P pool stocks

Labile P (0-30 cm) showed few significant differences among land uses. The native vegetation (Cerrado biome) at Lat\_17S (Figure 1A) had lower labile P stocks (82 kg ha<sup>-1</sup>) than the pasture (110 kg ha<sup>-1</sup>) and sugarcane (108 kg ha<sup>-1</sup>) areas. At Lat\_21S (Figure 1B), P stocks at pasture sites (122 kg ha<sup>-1</sup>) were significantly lower than native vegetation (150 kg ha<sup>-1</sup>) and sugarcane (177 kg ha<sup>-1</sup>). Labile P stocks were not affected by the three land uses studied within the Lat\_23S (Figure 1C), nor were they different on the regional scale (Figure 1D) averaging 94 and 115 kg ha<sup>-1</sup>, respectively. Therefore, in general, labile P stocks showed no change over time, although there was a small decreasing trend due to conversion from native vegetation to pasture and a slight increase with the conversion from pasture to sugarcane (Figure 2A). The labile P pool represents a small portion of total P reserves (Figure 1E), with relative contributions ranging from 5 (sugarcane) to 18% (pasture) at Lat\_17S; 9 (native vegetation) to 11% (pasture and sugarcane) at Lat\_21S; and a mean of 4% for all land uses at Lat\_23S. Considering the three sites, the average values ranged from 6 (native vegetation and sugarcane) to 8% (pasture) (Figure 1F).

The LUC effects were higher for P stocks in more recalcitrant pools, such as moderately labile and non-labile fractions. The LUC from native vegetation to pasture promoted a significant depletion (36%) in moderately labile P stocks (from 333 to 214 kg ha<sup>-1</sup>) and 32% depletion in non-labile P stocks (from 1422 to 961 kg ha<sup>-1</sup>) at the regional scale (Figure 1D). Overall, the change rates of moderately labile and non-labile P stocks due to conversion from native vegetation to pasture were -3.6 and -14.0 kg ha<sup>-1</sup>yr<sup>-1</sup>, respectively (Figure 2A). The LUC from pasture to sugarcane affected moderately labile and non-labile P pools differently at the three sites (Figure 1A-C), but in general, the results show a significant increase in the less labile forms in sugarcane soils (Lat\_17S and Lat\_21S). At the regional scale, these results were confirmed (Figure 1D), indicating that replacing pasture with sugarcane significantly increased moderately labile (39.5 kg ha<sup>-1</sup>yr<sup>-1</sup>) and non-labile (101.9 kg ha<sup>-1</sup>yr<sup>-1</sup>) P stocks (Figure 2B). We highlighted that part of this high P accumulation rates for regional scale are due to higher rates found at Lat\_17S (Figure 2B), where pasture soil presents less clay content than sugarcane soil.

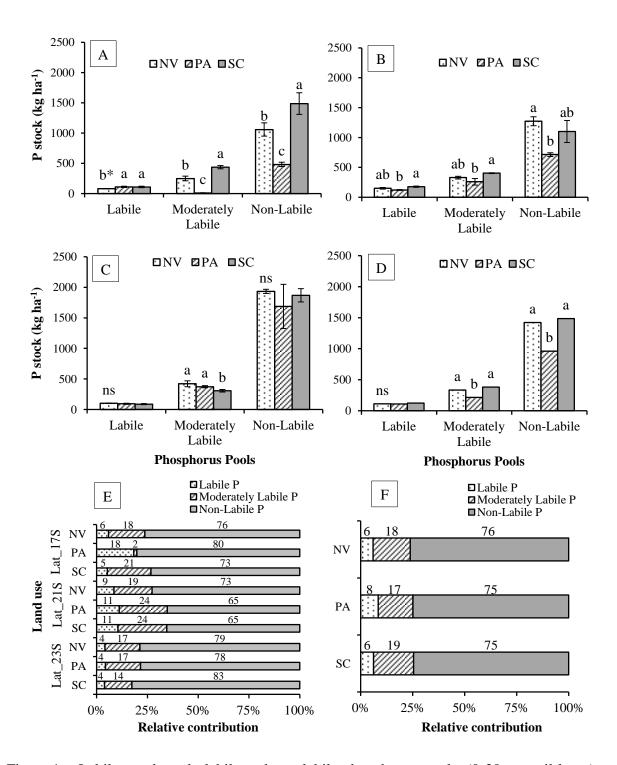


Figure 1 - Labile, moderately labile and non-labile phosphorus stocks (0-30 cm soil layer) at three sites: A) Lat\_17S, B) Lat\_21S, C) Lat\_23S and D) regional scale, and the relative contribution of each pool in the total P stock: E) at the three sites and F) on a regional scale, under land-use change (native vegetation - NV, pasture - PA, and sugarcane - SC) in central-southern Brazil. Error bars denote the standard deviation of the mean. \*Mean values within each phosphorus pool followed by the same letter do not differ among themselves according to Tukey's test (p<0.05); ns = non-significant

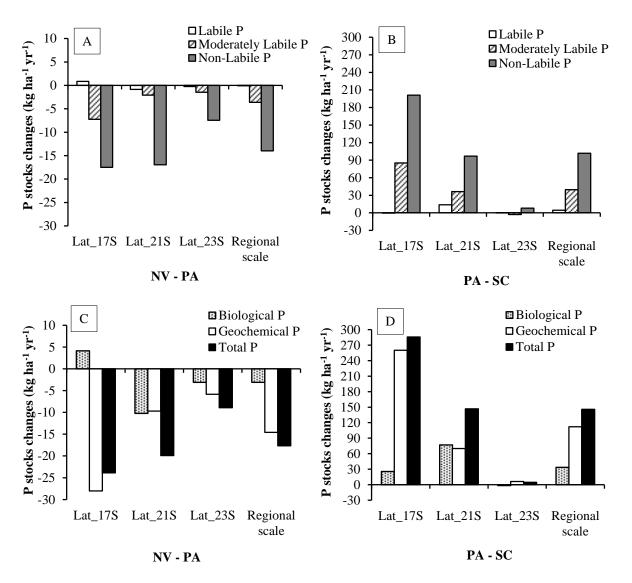


Figure 2 - Annual changes in labile, moderately labile and non-labile phosphorus stocks (A and B) and biological, geochemical and total phosphorus stock (C and D) for the 0-30 cm soil layer, as function of the land-use change from native vegetation to pasture (NV - PA) (left) and from pasture to sugarcane (PA - SC) (right) in central-southern Brazil

The moderately labile P pool contributed approximately 20% to the total P stock at the three sites, and averaged 18% in native vegetation, 17% in pasture and 19% in sugarcane (Figure 1E). In contrast, the non-labile P pool represented approximately 75% of the total P stock, with relative contributions ranging from 65 to 83% at the three sites studied (Figure 1E). At the regional scale, land use had no detectable effect on the soil P pools since relative values among all land uses were very similar, especially for moderately labile and non-labile pools (Figure 1F).

## 3.3.3 Biological, geochemical and total P pool stocks

Although there were few variations among land uses (Figure 3A), conversion from native vegetation to pasture decreased P stocks in both biological and geochemical pools (Figure 3C) at average rates of -3.1 kg ha<sup>-1</sup>yr<sup>-1</sup> and -14.6 kg ha<sup>-1</sup>yr<sup>-1</sup>, respectively (Figure 2C).

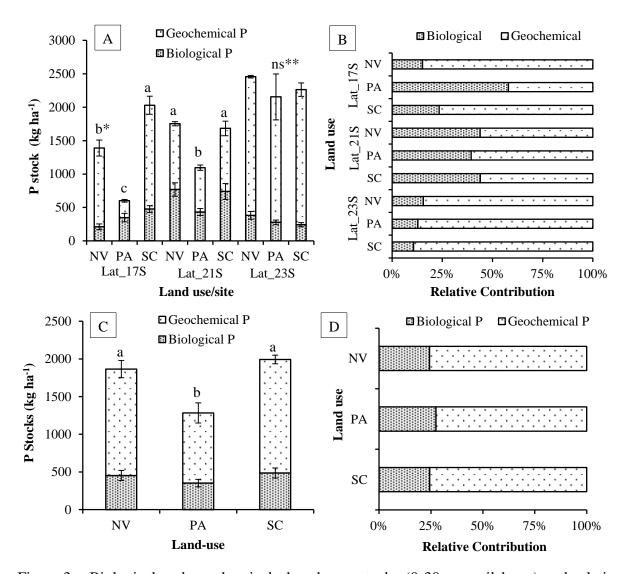


Figure 3 - Biological and geochemical phosphorus stocks (0-30 cm soil layer) and relative contribution of each pool at three sites (A and B) and on a regional scale (C and D) under land-use change (native vegetation - NV, pasture - PA, and sugarcane - SC) in central-southern Brazil. \*Mean values within each site followed by the same letter do not differ among themselves according to Tukey's test (*p*<0.05); ns = non-significant

In contrast, conversion from pasture to sugarcane increased both biological and geochemical P pools (Figure 3A, C) to levels similar to those observed under native vegetation. Overall, the biological P pool increased 33.7 kg ha<sup>-1</sup>yr<sup>-1</sup>, with the highest impact occurring within the geochemical P pool (112.1 kg ha<sup>-1</sup> yr<sup>-1</sup>) (Figure 2D).

Changes in total P stocks due to LUC (Figure 3) showed a consistent pattern but differed substantially in magnitude among the three sites. For example, 33 years after native vegetation was converted to pasture, P depletions ranged from 1,388 to 600 kg ha<sup>-1</sup> (-56.7%) at Lat\_17S, from 1,752 to 1,095 kg ha<sup>-1</sup> (-37.5%) at Lat\_21S, and from 2,457 to 2,154 kg ha<sup>-1</sup> (-12.3%) at Lat\_23S (Figure 3A). Considering the three sites together, in a regional scale, the average total P stock decreased from 1,866 in native vegetation soils to 1,283 kg ha<sup>-1</sup> in pasture soils. This total P decrease (-31.2%) occurred at an average rate of -18 kg ha<sup>-1</sup>yr<sup>-1</sup> (Figure 2C). Conversion of pasture to sugarcane increased total P stocks at all sites (Figure 3A). At the regional scale, total P stocks increased from 1,283 in pasture soils to 1,992 kg ha<sup>-1</sup> in sugarcane soils (Figure 3C) at an average rate of 145.6 kg ha<sup>-1</sup>yr<sup>-1</sup> (Figure 2D). Therefore, sugarcane soils currently have similar or higher P stocks than under native vegetation.

The relative proportion of biological and geochemical P pools within total P was affected by clay content (Figure 3B). Therefore, the P linked to organic composts had a higher contribution to total P stocks within sandy soil at Lat\_21S (ranging from 39 to 44%), than within clay soils at Lat\_23S (ranging from 11 to 16%). On average, biological P accounted for approximately 25% of the total P, but when analyzed separately, biological P represented more than 50% of labile and moderately labile P (Figure 4). Among the three land uses, biological P accounted for 58, 55, and 45% of the labile P pool within pasture, sugarcane and native vegetation sites, respectively. For moderately labile P, biological P accounted for 52, 51, and 57%, respectively.

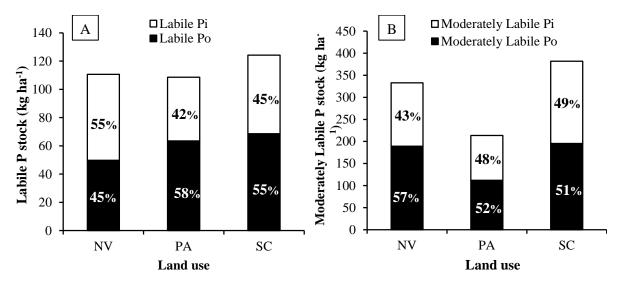


Figure 4 - Labile (A) and moderately labile (B) phosphorus stocks (0-30 cm) on biological (Po) and geochemical (Pi) pools under land-use change (native vegetation - NV, pasture - PA, and sugarcane - SC) in central-southern Brazil

## 3.3.4 Relationship among P pools, soil chamical attributes and clay content

Soil P pools are strongly correlated with other soil chemical properties and clay content, as indicated by significant linear correlations for 43 (p<0.01) and 50 (p<0.05) of the 60 pairs of soil properties studied (Table 2). Labile P was positively correlated with acidity attributes (BS, r = 0.51 and pH, r = 0.43), cations (Ca, r = 0.47; Mg, r = 0.44; K, r = 0.37), SOM (SOC, r = 0.23 and TN, r = 0.35) and CEC<sub>pH7</sub> (r = 0.33), but negatively (although not significantly) correlated with clay content. Moderately labile P was significantly correlated with all soil variables, showing even stronger positive relationships with clay content (r = 0.62), SOC (r = 0.65), TN (r = 0.65) and CEC<sub>pH7</sub> (r = 0.57). Non-labile P was similar to the moderately labile P, although it was not correlated with pH. Overall, soil acidity had a greater effect on labile P, while clay content, SOM (SOC and NT) and CEC<sub>pH7</sub> were more closely related to the moderately and non-labile P pools.

Table 2 - Pearson's correlation coefficients (r) and probability of error (p) among soil chemical attributes, clay content and soil P pools stocks in areas under land-use change (native vegetation - pasture - sugarcane) in central-southern Brazil

Soil	Phosphorus pools stocks <sup>b</sup>											
properties <sup>a</sup>	Labile	Mod. labile	Non-labile	Biological	Geochemical	Total						
Clare	-0.0978*	0.6207	0.8799	-0.3274	0.9249	0.8630						
Clay	$0.3139^{\dagger}$	< 0.0001	< 0.0001	0.0005	< 0.0001	< 0.0001						
SOC	0.2300	0.6552	0.6950	-0.0365	0.7243	0.7251						
SUC	0.0166	< 0.0001	< 0.0001	0.7079	< 0.0001	< 0.0001						
TN	0.3499	0.6542	0.6487	0.1572	0.6471	0.6904						
111	0.0002	< 0.0001	< 0.0001	0.1043	< 0.0001	< 0.0001						
S	0.1316	0.4737	0.3584	0.2124	0.3489	0.4009						
S	0.1746	< 0.0001	0.0001	0.0273	0.0002	< 0.0001						
K	0.3727	0.4132	0.3330	0.1609	0.3365	0.3768						
V	< 0.0001	< 0.0001	0.0004	0.0962	0.0004	< 0.0001						
Ca	0.4729	0.2439	0.2307	0.4617	0.1554	0.2609						
Ca	< 0.0001	0.0110	0.0163	< 0.0001	0.1083	0.0064						
Mg	0.4407	0.4871	0.4639	0.2678	0.4391	0.5046						
Mg	< 0.0001	< 0.0001	< 0.0001	0.0051	< 0.0001	< 0.0001						
рH	0.4293	0.2287	0.1465	0.4959	0.0728	0.1949						
рп	< 0.0001	0.0173	0.1304	< 0.0001	0.4542	0.0454						
BS	0.5145	0.3183	0.1955	0.5397	0.1245	0.2471						
DS	< 0.0001	0.0008	0.0426	< 0.0001	0.1992	0.0099						
CEC	0.3297	0.5690	0.6439	0.1216	0.6339	0.6690						
$\mathrm{CEC}_{\mathrm{pH7}}$	0.0005	< 0.0001	< 0.0001	0.2101	< 0.0001	< 0.0001						

<sup>&</sup>lt;sup>a</sup> Clay: clay content (g kg<sup>-1</sup>); SOC: soil organic carbon (g kg<sup>-1</sup>); TN: total nitrogen (g kg<sup>-1</sup>); S: sulfur (mg dm<sup>-3</sup>); K: potassium (mmol<sub>c</sub> dm<sup>-3</sup>); Ca: calcium (mmol<sub>c</sub> dm<sup>-3</sup>); Mg: magnesium (mmol<sub>c</sub> dm<sup>-3</sup>); pH: potential de hydrogen in solution of CaCl<sub>2</sub> 0,01M L<sup>-1</sup> (1:2.5); BS: base saturation (%); CEC<sub>pH7:</sub> potential cation exchange capacity (mmol<sub>c</sub> dm<sup>-3</sup>); <sup>b</sup> Phosphorus pools stocks (kg ha<sup>-1</sup>); \*Pearson's correlation were calculated using number of observations "n"=108; <sup>†</sup> Probability of error (p).

Biological P showed even higher positive correlations with acidity attributes (BS, r = 0.54 and pH, r = 0.50) and cations (Ca, r = 0.46 and Mg, r = 0.27), but was negatively correlated with clay content (r = -0.32). Geochemical P and clay contents had the highest positive correlation (r = 0.92). Geochemical P was correlated with all soil chemical properties, except the acidity attributes (pH and BS) and Ca. Biological P was also had lower correlations with soil chemical properties and clay content than geochemical P.

Total P stocks were positively correlated with all studied soil properties following the trend already observed for the geochemical P pool, emphasizing the highest correlations with clay content (r = 0.86) and SOM (SOC, r = 0.72 and NT, r = 0.69).

## 3.3.5 Relationship between P pools and macrofauna

The relationships between soil P pools and soil macrofauna are shown in Figure 5. First two axes explained 70% of the data variance. Although macrofauna variables were not associated with the soil total P, taxonomic richness, macrofauna diversity and evenness were closely and positively related to the labile P pool and biological P pool. Positive relationships were also found between the functional diversity of the macrofaunal community (i.e., the number of functional groups found in a sample) and the biological and labile P pools. The functional diversity was associated with the biological variables richness, diversity and evenness. In contrast, macrofauna density and dominance of species showed negative correlations with labile and biological P and with all the other biological variables. Total P content was strongly related with moderately-labile P, no-labile P and geochemical P pools.

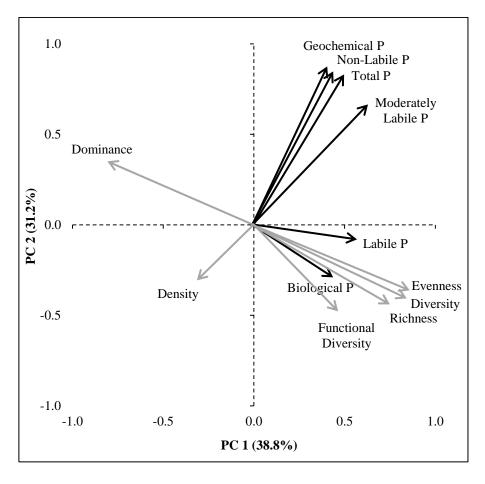


Figure 5 - Principal component analysis of the soil phosphorus pools (black lines) and macrofauna variables (gray lines)

#### 3.4 Discussion

## 3.4.1 Sugarcane expansion and its implications on P dynamic

Labile P fractions are the primary P source for plant growth, but those fractions represented less than 10% of the total soil P, which is similar to the proportion reported by Riskin et al. (2013) and Rodrigues et al. (2016). Overall, LUC from native vegetation to pasture had no significant effect in labile P pool (Figure 1D), although there was a decreasing trend within this pool for the P<sub>resin</sub> fraction, as reported by Cherubin et al. (2015). Another large, paired-comparison study recently carried out in Brazil (GROPPO et al., 2015) concluded that LUC from native vegetation to well-managed pasture led to increase of plant-available inorganic P. However, when assessed in a regional survey that better represented the status of the Brazilian extensive pastures (as those here studied) there were reductions of plant-available inorganic P stocks in pasture compared to native vegetation soils.

The conversion of natural ecosystems or pastures for agricultural purposes in tropical soils generally increases labile P pool due to low native P levels in natural ecosystem/pastures and inputs by phosphate fertilizers, as reported by Negassa and Leinweber (2009). The assumption that increasing pH increases plant-available P (HAYNES, 1984) was also confirmed by positive correlations between labile P and acidity attributes (Table 2). Our findings are in line with several studies carried out in Brazil. Pavinato, Merlin and Rosolem (2009) and Rodrigues et al. (2016) indicated that P applied through phosphate fertilization increased the inorganic labile and moderately labile P fractions in Oxisols from the Brazilian Cerrado. Aguiar et al. (2013) reported higher inorganic labile P levels under agricultural uses (alley cropping and no-tillage) compared to a newly cleared area, secondary Amazon forest and pasture. In the southeastern Amazon, Riskin at al. (2013) also observed that the continuum use of P fertilizer in soybean [Glycine max (L.) Merr.] fields led to increased inorganic labile P and decreased organic labile P compared to native vegetation. Groppo et al. (2015) found significant higher inorganic labile P stocks for the top soil layer (0-30 cm) in crop-livestock systems (49.50 kg ha<sup>-1</sup>) than in native vegetation (21.74 kg ha<sup>-1</sup>) in the Cerrado, Atlantic Forest, and Pampa biomes.

There was a higher contribution of organic P (58%) to the labile P pool for pasture areas (Table 1; Figure 4) representing significant P-recycling (COSTA et al., 2014; CREWS; BROOKES, 2014; FRANCO et al., 2015; STUTTER et al., 2015), where up to 85% of the P taken up by plants is returned to the soil through animal dung (NASH et al., 2014) and associated with greater soil biological activity (LAVELLE et al., 2014; FRANCO, 2015). These results confirm the hypothesis reported in the literature that plant-available P in pasture is strongly dependent on mineralization of the Po pool (AGUIAR et al., 2013; HAMER et al., 2013; CREWS; BROOKES, 2014; FONTE et al., 2014; NASH et al., 2014; FRANCO et al., 2015; NESPER et al., 2015; STUTTER et al., 2015). Long-term experiments in the USA (MOTAVALLI; MILES, 2002) and England (CREWS; BROOKES, 2014) have shown that native and perennial grass has capacity to maintain a greater proportion of native or fertilizer-P in relatively available organic forms compared to annual wheat (*Triticum aestivum*). These studies supported that increased P availability is due to a transformation of stable residual P into active Po. In addition, grazing appears to contribute to the regulation of labile P cycling in pasture conditions (COSTA et al., 2014). More recently, a large study carried out in the UK (STUTTER et al., 2015) found predominance of organic P forms (e.g., monoesters, diester, polyphosphates and other) in extensive grasslands soils, while inorganic P forms (orthophosphates) were more important in arable soils. Greater concentrations of microbial P forms in extensive grassland indicate presence of an active microbial population efficient in turnover of soil organic P in conditions of limited readily available inorganic P (STUTTER et al., 2015).

Overall, a depletion in P stocks was observed after 33 years of conversion from native vegetation to pasture, especially for the moderately and non-labile pools (Figures 1 and 2). These decreases are the result of P exploitation over time due to grazing without any P fertilizer inputs (GROPPO et al., 2015). This further emphasizes that inappropriate management of soil fertility by Brazilian farmers leads to soil degradation and becomes one of the major factors for low productivity (i.e., degraded pastures) in approximately 70% of Brazilian pasturelands (DIAS-FILHO, 2014; STRASSBURG et al., 2014). Recent studies stressed that soil structural degradation in pasture promotes a continuum of P depletions, especially organic forms associated with SOM, confirming that similar mechanisms act in the protection and stabilization of organic P and SOM in the soil (FONTE et al., 2014; NESPER et al., 2015). Although the linkage between SOM and biological organic P levels was not significant in this study, we found a strong correlation between total P stock and SOM (Table 2).

Expansion of sugarcane over extensive pasture led to P accumulation in less labile P pools, such as moderately labile and non-labile fractions (Figures 1 and 2), which are the primary fates of P from fertilizers in soil (NEUFELDT et al., 2000; NEGASSA; LEINWEBER, 2009; PAVINATO; MERLIN; ROSOLEM, 2009; WRIGHT, 2009; SHEN et al., 2011; RISKIN et al., 2013; RODRIGUES et al., 2016). These finding are consistent with other studies, that have found higher P accumulation in mineral-associated fractions (P sinks), particularly the Ca-bound fraction in temperate soils (CASTILHO; WRIGHT, 2008; NEGASSA; LEINWEBER, 2009; WRIGHT, 2009) as well as Fe- and Al-bound fractions in tropical soils (NEUFELDT et al., 2000; NEGASSA; LEINWEBER, 2009; PAVINATO; MERLIN; ROSOLEM, 2009; AGUIAR et al., 2013; RISKIN et al., 2013; RODRIGUES et al., 2016), such as the highly weathered soils evaluated in this study. When P from mineral fertilizers is added to soil it is quickly adsorbed or precipitated in mineral forms. First, an electrostatic attraction (reversible) occurs between the P (orthophosphate) and soil minerals (NOVAIS; SMYTH; NUNES, 2007), primarily on the edges of silicates, crystalline Fe and Al oxides, and amorphous Al oxides (FONTES; WEED, 1996). These attractions become stronger over time, and a specific adsorption or chemisorption (irreversible) occurs with a ligand-exchange (NOVAIS; SMYTH; NUNES, 2007).

An analysis of global agricultural P budgets, based in input and output rates, showed that the top quartile of P surpluses (accumulation in the soil) to be those with more than 13 kg ha<sup>-1</sup>yr<sup>-1</sup> (MacDONALD et al., 2011), indicating the high accumulation P rates found in our weathered Brazilian soils under sugarcane production (Figure 2) are among the highest P accumulation rates reported for world croplands. However, despite this high rate of total P accumulation, only a small amount was in labile forms (4.4 kg ha<sup>-1</sup>yr<sup>-1</sup>); consequently, the amount of available P to support sugarcane growth was not enhanced [i.e., P<sub>resin</sub> levels (Table 1) were lower than the recommended 15 mg kg<sup>-1</sup> critical level (ESPIRONELLO et al., 1997)]. In terms of environmental impacts, although a large amount of P has being annually accumulated in the soil, the strong sorption capacity of these weathered soils suggests that LUC associated with agricultural expansion in Brazil has a low risk for P losses to waterways (RISKIN et al., 2013) and subsequent eutrophication of water resources.

Phosphorus from organic residues (e.g., vinasse and filter cake) increased the organic labile P pool in sugarcane fields (see Lat\_23S, Table 1). In these conditions, similarly to pasture, the supply of P in forms available to plants depends on mineralization of organic P; nevertheless, lower soil biological activity in sugarcane fields (FRANCO, 2015) may result in lower turnover of soil organic P, and consequently, an insufficient P supply for the sugarcane crop. The release of P from crop residues is significantly reduced in systems where the P-status of crops and soils is low, requiring strong integration between organically cycled P with fertilizer management strategies (DAMON et al., 2014). Therefore, we suggest that fertilization strategies include a balance between organic and mineral P sources to improve both sugarcane yield and soil quality.

The geochemical and biological P pools represented around 75 and 25% of the total P stock, respectively. These proportions were similar to values reported in the literature (NEUFELDT et al., 2000; NZIGUHEBA; BÜNEMANN, 2005; PAVINATO; MERLIN; ROSOLEM, 2009; SHEN et al., 2011; RODRIGUES et al., 2016). However, when only labile and moderately labile pools are considered, biological P represents more than 50% of total P, indicating a great importance of SOM-cycling to P plant-availability in both natural ecosystems and agroecosystems. Soil organic matter is often the major storehouse of soil P, accounting for 30 to 65% of total P (SHEN et al., 2011). In highly weathered tropical soils the pools of organic P and decomposition of SOM are critical for plant productivity (CROSS; SCHLESINGER, 1995), and the maintenance of P in organic forms is the key for a long-term functioning and productivity of these soils (FONTE et al., 2014; NESPER et al., 2015; RODRIGUES et al., 2016), since the biological P pool is less susceptible to sorption reactions

(CROSS; SCHLESINGER, 1995). Future researches using techniques such as <sup>31</sup>P NMR are necessary to identify organic P forms (see NASH et al., 2014, STUTTER et al., 2015 and KRUSE et al., 2015) and their effects on P-cycling and P plant-availability in areas undergoing LUC for expansion of biofuel crops.

# 3.4.2 Implications of macrofauna on P cycling

Overall, biological P pool showed lower correlations with soil chemical properties and clay content than the geochemical P pool (Table 2), indicating that soil biological properties and processes (e.g., macrofauna, symbiotic actions between soil microorganisms and plants; actions of enzymes released by plant roots and microorganisms) and soil physical and structural conditions (FONTE et al., 2014; NESPER et al., 2015) should be considered to understand the dynamic of the biological P pool in the soil. This assumption was confirmed, since positive relationship was found between soil macrofauna diversity and biological P pool (Figure 5). The disconnection between total P contents and biological variables can be explained by the fact that the geochemical P pool (especially non-labile P forms) comprised around 75% of the total P at all the field sites (Figure 3).

In this study, higher densities of macrofauna animals were specially related to the wide dominance of termites in the communities of pasture soils at Lat\_17S and Lat\_23S (FRANCO, 2015). It was clearly the cause of the negative relationships between macrofauna density and the all the others biological variables. In accordance with our findings, studies have shown that functional diversity of the soil macrofaunal community affects decomposition processes and nutrient cycling (HEEMSBERGEN et al., 2004; ZIMMER; KAUTZ; TOPP, 2005; COLLISON; RIUTTA; SLADE, 2013). Heemsbergen et al. (2004) observed that functional dissimilarities between soil macrofauna species, irrespectively of the number of taxonomic groups, is a driver of ecosystem processes related to nutrient cycling due to facilitative interactions among species. Zimmer, Kautz and Topp (2005) reported a potential effect on ecosystem functioning through joint action of soil macrofauna even at low species diversity. It suggests that the taxa richness was associated with the enrichment of P labile forms as an indirect result of the positive correlation between functional diversity and taxa richness found in this study (Figure 5). We suggest that correlation between high soil P availability and the presence of a functional diverse soil macrofaunal community may be linked to a rapid turnover of an increased organic P content.

Some studies have described a significant enrichment of labile P forms as a result of the activity of macrofaunal groups such as coleoptera (LI et al., 2006), earthworms (OUEDRAOGO et al., 2005) and termites (RÜCKAMP et al., 2010). Li et al. (2006) showed an increased availability of soil P during the gut passage of scarabaeid beetle (Coleoptera) attributed to solubilization and enzymatic hydrolysis of organic P, and desorption of inorganic P. The contribution of earthworms to enhance soil P availability is attributed to higher pH of the gut content, changes in sorption complexes due to the competition for sorbing sites between phosphate and carboxyl groups of a mucus glycoprotein produced in its gut, and an increase in microbial activity during digestion (CHAPUIS-LARDY et al., 2011). Termites contribute to high levels of soil labile P by the transformation of organic P through enzymatic activity in the fresh biostructures (CHAPUIS-LARDY et al., 2011).

The LUC associated to sugarcane expansion in Brazil causes negative effects in both SOC (FRANCO et al., 2015) and soil biodiversity (FRANCO, 2015), which potentially aggravates the low plant availability of phosphorus in highly weathered soils, the main costly and limiting factor for agricultural productivity in tropical soils. Monitoring soil macrofaunal biodiversity to control functional simplification of the community may therefore be an efficient management strategy in sugarcane fields for increased soil P availability and likely for multiple ecosystem services.

# 3.4.3 Phosphorus pools as soil quality indicators

Phosphorus is an integral component to ecosystem management because it limits primary production in aquatic and terrestrial ecosystems (ELSER et al., 2007; ÅGREN; WETTERSTEDT; BILLBERGER, 2012; ELSER, 2012). Phosphorus is one of the primary limiting nutrients for agricultural production in Brazilian soils due to both inherent characteristics to strongly weathered soils and inadequate strategies of P application (NOVAIS; SMYTH; NUNES, 2007). Hence, P pools are highly influential on key soil functions related to nutrient fluxes and ecosystems services responsible for maintaining a suitable soil biotic habitat and sustaining plant growth (DORAN; PARKIN, 1994; LAVELLE et al., 2014). Soil quality indicators are defined as those soil properties and processes having the greatest sensitivity to changes in soil functions (ANDREWS; KARLEN; CAMBARDELLA, 2004). Therefore, for the soils we evaluated soil P pools appear to be useful soil quality indicators.

Our findings showed modifications of P pools in response to LUC associated with sugarcane expansion. Strong correlations among P pools and other soil properties broadly used to assess soil degradation, such as SOC, CEC and acidity attributes (DORAN; PARKIN, 1994; KARLEN et al., 2013), endorse the sensitivity of P pools to changes in soil functions. Furthermore, we observed significant correlations between taxonomic richness and diversity, and functional diversity of soil macrofauna communities with biological and labile P pools. Soil macrofauna have been indicated as an efficient biological indicators of modifications on soil quality as response to management or land-use changes (DORAN; ZEISS, 2000; ROUSSEAU et al., 2013; LAVELLE et al., 2014). Therefore, P pools warrant further investigations as potential soil quality/health indicators able to assess and monitor the soil degradation associated to sugarcane expansion for ethanol production in Brazil.

In addition, future studies should be carried out using an integrated approach to understand collectively the LUC effects on isolated indicators, for instance SOC stocks (MELLO et al., 2014; FRANCO et al., 2015), soil nutrients (CHERUBIN et al., 2015; FRANCO et al., 2015), soil biodiversity, soil structure and physical quality and others, to determine the environmental sustainability of Brazilian ethanol.

#### 3.5 Conclusions

This field study carried out across the primary sugarcane producing region in Brazil indicated that long-term conversions from native vegetation (Cerrado and Atlantic forest biome) to extensive pasture induced significant depletions of soil total P stocks. These results showed a critical process of soil chemical degradation, which helps to explain the low productivity in most of Brazilian pasturelands. Our findings suggest that under degraded pasture, the biological P pool plays crucial role for supplying available P to plants.

The LUC from pasture to sugarcane altered the P-cycle, increasing P stocks in moderately labile and non-labile pools. The fertilization management recovered total P stocks of sugarcane soils to similar or higher levels than those measured in natural ecosystems. However, although high accumulation rates of P were found, only a small and insufficient amount was kept in available forms to plants. Phosphorus inputs from organic residues (e.g., filter cake and vinasse) increased the organic labile P and may be a complementary strategy to supply P for sugarcane crop.

Phosphorus pools were significantly correlated with soil chemical attributes. Also, functional diversity of soil macrofauna community was positively related with increases of

soil P availability to plants, probably as a response to biological P pool increase and its greater turnover in the soil. Therefore, we conclude that P pools are useful indicators for detecting soil quality/health changes induced by LUC associated to sugarcane expansion in Brazilian tropical soils.

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# 4 SOIL PHYSICAL QUALITY RESPONSE TO SUGARCANE EXPANSION IN BRAZIL

#### **Abstract**

Globally, the rate of land-use change (LUC) is increasing rapidly to support biofuel feedstock production. In Brazil, sugarcane (Saccharum officinarum) expansion to produce ethanol is displacing degraded pastures. Intensive mechanization for sugarcane production, could impact soil physical quality in these areas. We evaluated a typical LUC sequence (i.e., native vegetation - pasture - sugarcane) on soil physical quality at three sites in centralsouthern Brazil. The attributes evaluated through on-farm and laboratory soil analyses were: bulk density, degree of compactness, macroporosity, microporosity and total porosity, waterfilled pore space, indexes of soil water storage and aeration capacity, soil resistance to penetration, field-saturated hydraulic conductivity and stability structural index. Calculations of mean weight diameter for the soil aggregates and visual evaluation of soil structure (VESS) scores were also included in this study. From those data we defined a minimum dataset for calculating an additive soil physical quality index (SPQI). Long-term conversion from native vegetation to pasture increased soil compaction (i.e., higher bulk density, degree of compactness and resistance to penetration values), decreased aeration porosity and water hydraulic conductivity, and consequently, created an unbalanced ratio between water- and airfilled pore space in the soil. Based on our SPQI, the soil's capacity to perform its physical functions decreased from 90% under native vegetation to 73% under pasture. Land-use change from pasture to sugarcane induced slight soil physical quality degradation, in which soil function was 68 and 56% of capacity. Overall, soil physical quality decreased under sugarcane fields, due to decreases in soil porosity, aeration and water hydraulic conductivity as well as increases in soil penetration resistance, structural degradation and erosion risk. Tillage operations performed during the sugarcane replanting had a short-term positive effect on soil physical quality, although over time it further decreased the resistance to erosion and structural degradation. Therefore, to convert degraded pasture to sugarcane in a sustainable manner, the soils should be managed in ways that increase the soil organic matter and minimize compaction. These actions are needed to prevent further soil physical quality degradation and to improve the sustainability of sugarcane ethanol production.

Keywords: Land-use change; Soil compaction; Soil physical functions; Ethanol production

#### 4.1 Introduction

Increasing global demand for biofuel has promoted the intensification of LUC around the world, imposing concerns about soil physical quality degradation and its negative implications on ecosystem function (GASPARATOS; STROMBERG; TAKEUCHI, 2011; FU et al., 2015). Brazil is the largest sugarcane ethanol producer in the world, having increased from 5.8 to 9.0 Mha between 2005 and 2015 (COMPANHIA NACIONAL DE

ABASTECIMENTO, 2015). To meet projected (2021) domestic supplies for ethanol in Brazil, an additional 6.4 Mha of sugarcane is required (GOLDEMBERG et al., 2014).

Historically sugarcane expansion has been concentrated in central-southern Brazil, with 70% of this expansion occurring in degraded pasturelands (ADAMI et al., 2012). The development of Brazilian agriculture has seen extensification of pasture into native vegetation with poor management practices. This has resulted in loss of soil organic carbon (SOC) and soil fertility (MELLO et al., 2014; CHERUBIN et al., 2015; FRANCO et al., 2015). It is estimated that 70% of Brazilian pasturelands are degraded or in the process of being degraded (DIAS-FILHO, 2014). The vast area of degraded pasture in Brazil, coupled with opportunities for improvements in current ranching practices, could provide enough land for sugarcane production to meet the projected demand for ethanol, while still meeting the domestic demand for other ecosystem services (LAPOLA et al., 2010; GOLDEMBERG et al., 2014).

In comparison to pasture, sugarcane production requires intensive mechanization resulting in changes to soil physical properties and related processes. Soil tillage is used to incorporate lime and fertilizer when sugarcane is first established, and again at approximately five-year intervals when sugarcane yields begin to decrease and it is replanted. With the recent shift to mechanized harvesting, intensive machinery traffic may be contributing to soil physical degradation in these areas. Studies have shown increased soil bulk density and soil strength (BAQUEIRO et al., 2012; BANGITA; RAO, 2012; SOUZA et al., 2014; 2015), with decreases in soil porosity, aeration, aggregation, water infiltration and available water in many sugarcane fields (BRAUNACK; McGARRY, 2006; CASTRO et al., 2013; FRANCO, 2015; HUNKE et al., 2015b).

Soil physical quality degradation has adverse impacts on root growth (OTTO et al., 2011; BAQUEIRO et al., 2012; SOUZA et al., 2014; 2015), often limiting uptake of water and nutrients, thus decreasing sugarcane yields (BANGITA; RAO, 2012; SOUZA et al., 2014). Decreased sugarcane productivity also decreases atmospheric CO<sub>2</sub> uptake by above and belowground biomass, resulting in lower organic C inputs and a gradual depletion of SOC (FRANCO et al., 2015). Decreasing soil physical quality may also reduce ecosystem functioning (FU et al., 2015), by accelerating soil C turnover (SIX; ELLIOTT; PAUSTIAN, 2000), decreasing SOC stocks (MELLO et al., 2014; FRANCO et al., 2015) and increasing CO<sub>2</sub> emissions (BALL, 2013; SILVA-OLAYA et al., 2013). In terms of biogeochemical processes, soil compaction reduces air-filled porosity favoring denitrification (N<sub>2</sub>O emissions) and methanogenesis (CH<sub>4</sub> emissions) (BALL, 2013). Runoff and soil erosion risks increase because physically degraded soils have lower water infiltration rates (HUNKE et al., 2015a,b)

and thus contribute sediment, nutrients, and pesticides to surface waters (GUCKER; BOECHAT; GIANI, 2009; HUNKE et al., 2015a,b). Furthermore, soil compaction can also modify or destroy native biological habitats, resulting in loss of biodiversity and ecosystem function (BENTON; VICKERY; WILSON, 2003).

Although many technical papers have been published, LUC effects on soil physical quality associated with sugarcane expansion in Brazil are still poorly documented. We conducted an on-farm study in the largest sugarcane-producing regions of Brazil to: i) quantify effects of the primary LUC sequence associated with sugarcane expansion (i.e., native vegetation to pasture to sugarcane) on soil physical properties, and ii) integrate the soil physical indicators of soil degradation into an additive index for assessing LUC impacts on soil physical functioning. We hypothesized that the LUC from native vegetation to pasture and then to sugarcane was resulting in continuous degradation of soil physical quality that could be detected by computing an overall Soil Physical Quality Index (SPQI).

#### 4.2 Material and Methods

# 4.2.1 Study sites and experimental design

The study sites and experimental design were described in the 2.2.1 and 2.2.2 items.

#### 4.2.2 Sampling and soil physical measurements

Soil sampling was carried out in January 2014. At each land use (i.e., native vegetation, pasture and sugarcane) soil samples were collected using a consistent grid pattern composed of nine sampling points spaced 50 m apart, providing a total of 27 sampling points for each site (i.e., total of 81 sampling points for the three sites). At each sampling point, a small trench (30 x 30 x 30 cm) was opened to collect disturbed and undisturbed soil samples from the 0-10, 10-20 and 20-30 cm layers, providing a total of 243 soil samples for soil physical analyses. Although root systems of tropical pastures and sugarcane can reach deeper soil layers, we limited our assessment to 30 cm because most of roots are concentrated in this layer (BALL-COELHO et al., 1992; KANNO et al., 1999) and this is the zone where more significant soil physical property changes are induced by land use and management practices. The sampling points for each land use were positioned in representative locations within the total area sampled. In native vegetation areas we avoided sampling close to ant or termite

nests, burrows of wild animals and big trees. In pasture areas, which were continuously and uniformly grazed, our major caution was to avoid sampling on the preferential cattle trampling paths, where the soil is much more compacted. Except at Lat\_17S where the soil had been recently tilled for sugarcane replanting, all sampling points in sugarcane fields were located within the inter-row position, which is homogeneously tracked during harvest operations.

Measurements of soil resistance to penetration (SRP) were taken around the soil sampling trenches to a depth of 30 cm using a digital penetrometer (PenetroLOG<sup>®</sup>). Five replicates were used to compute an average value for each sampling point. Field-saturated hydraulic conductivity ( $K_{fs}$ ) was measured using the 'simplified falling-head' method proposed by Bagarello, Iovino and Elrick (2004) and later used by Keller et al. (2012). Three replicate  $K_{fs}$  measurements for each sampling point were made using steel cylinders (height 15 cm x internal diameter 15 cm), inserted 8 cm into the soil and 330 ml of water applied according to Keller et al. (2012).

In the laboratory, disturbed soil samples were used to determine particle size using the hydrometer method (GEE; OR, 2002). The undisturbed soil samples were weighed (initial soil water content), saturated for 48 h by gradually raising the water level in a tray and weighed again. Soil water content at -6 kPa and -10 kPa water potentials were determined using tension tables similar to those described by Ball and Hunter (1988). The soil samples were then dried at 105°C for 48 hours and weighed again. Bulk density (BD, Mg m<sup>-3</sup>) was calculated by dividing the soil dry mass by volume of the cylinder. The maximum bulk density (BD<sub>max</sub>, Mg m<sup>-3</sup>) was estimated using a pedotransfer function described by Marcolin and Klein (2011), in which SOM and clay content are the input parameters. Based on the BD and  $BD_{max}$  the soil degree of compactness (SDC, %) was calculated [SDC = (BD/BD<sub>max</sub>) x 100]. Soil particle density (PD, Mg m<sup>-3</sup>) was determined from sub-samples (5g) using a gas pycnometer according to Flint and Flint (2002). The total porosity (TP, m<sup>3</sup> m<sup>-3</sup>) was calculated as TP = 1 - (BD/PD). Soil macroporosity (MaP, m<sup>3</sup> m<sup>-3</sup>) was computed as the difference between soil water content at saturation and at -6 kPa. Soil microporosity (MiP, m<sup>3</sup> m<sup>-3</sup>) was estimated as the soil water content at the -6 kPa. Water-filled pore space (WFPS) was calculated by dividing volumetric moisture at -6 kPa by TP as described by Wienhold et al. (2009). In addition, we calculated two indexes according to Reynolds et al. (2002): i) the soil water storage capacity (SWSC) defined as the ratio between water content at field capacity (FC, -10 kPa soil water potential) and TP (SWSC = FC/TP); ii) soil aeration capacity (SAC) calculated as the ratio between drained pores at soil water potential of -10 kPa (ACt) and TP (SAC = ACt/TP).

The risk of soil structural degradation was assessed using the "structural stability index" (SSI), according to Reynolds et al. (2009):  $SSI = [(SOC \times 1.724) / (silt + clay)]*100$ , where, SOC is the organic carbon content (g kg<sup>-1</sup>); 1.724 is a factor to convert SOC to SOM; silt and clay are particle size fractions (g kg<sup>-1</sup>).

Mean weight diameter of soil aggregates (MWD) and VESS scores were used to verify the correlation with other soil physical properties and calculate an index of SPQ. The MWD and VESS data were taken from Franco (2015) and Cherubin et al. (2016)<sup>2</sup>, respectively, which were measured from soil samples collected at the same sites and sampling times.

# 4.2.3 Soil Physical Quality Index calculation

An overall soil physical quality index (SPQI) was calculated to quantify LUC effects associated with sugarcane expansion in Brazil. Development of the SPQI followed three steps as outlined by Karlen and Stott (1994) and Karlen, Ditzler and Andrews (2003). The first step was to select appropriate SPQ indicators to represent and monitor four critical soil physical functions. They were to: f(i): support root growth; f(ii): supply water for plants and edaphic fauna; f(iii): allow gas exchange between soil and atmosphere (soil aeration); and f(iv): ability to resist erosion and soil degradation. These functions are essential to sustain plant productivity and maintain ecosystem services.

Based on published literature and the authors' experience, eight indicators (BD, VESS score, SWSC, K<sub>fs</sub>, MaP, SAC, MWD and SSI) were used as a minimum dataset to determine how well the four critical soil physical functions were being performed under the three land uses. The second step (indicator interpretation) involved transforming each indicator into a unitless value ranging from 0 to 1 for inclusion in the SPQI. The soil data from 0-10, 10-20 and 20-30 cm were averaged to 0-30 cm layer to calculate an overall soil physical quality index that better represents the whole soil profile assessed. The transformation was performed using a linear technique as described by Andrews, Karlen and Mitchell (2002). Indicators were ranked in ascending or descending order depending on whether a higher value was considered "good" or "bad" in terms of soil function. For 'more is better' indicators, such as

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<sup>&</sup>lt;sup>2</sup> Data presented in the chapter 5.

MaP,  $K_{fs}$  MWD and SSI, each observation was divided by the highest observed value such that the highest observed value received a score of 1. For 'less is better' indicators, such as BD and VESS, the lowest observed value (in the numerator) was divided by each observation (in the denominator) such that the lowest observed value receives a score of 1. For 'optimum' indicators, such as SWSC and SAC observations were scored as 'higher is better' up to a threshold value (SWSC = 0.66 and SAC = 0.33) then scored as 'lower is better' above the threshold.

For step 3, the transformed indicator values were multiplied by their weight. These results were summed within each soil physical function, resulting in a scored soil physical function. Subsequently, the individual scores of soil physical function were multiplied by their weight. Finally, these weighed scores were summed to calculate the SPQI. In Table 1 the step by step procedure used for developing the SPQI is shown. Soil physical function scores and SPQI scores for each sample were averaged and standard deviations were calculated for each land use. In addition, two scenarios were compared to verify soil physical quality effects of tillage operations performed during the sugarcane replanting.

Table 1 - Model of the soil physical functions framework and indicators used for developing the soil physical quality index (SPQI)

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Soil physical function <sup>‡</sup>	Weight I	Soil indicator§	Weight <b>II</b>	Transformed indicator value <sup>†</sup>	Soil indicator score (II x III)	Soil physical function score ∑(II x III) IV	Weighted soil physical function score (IV x I)	SPQI ∑(IV x I)
f(i)	0.25	BD	0.50	0.79	0.40	0.72	0.18	
	0.23	VESS	0.50	0.65	0.33	0.72	0.16	
f(ii)	0.05	SWSC	0.50	0.89	0.45	0.04	0.21	0.69
	0.25	$\mathbf{K}_{\mathrm{fs}}$	0.50	0.78	0.39	0.84		
f(iii)	0.25	SAC	0.50	0.39	0.20	0.50	0.15	<u>0.68</u>
		MaP	0.50	0.78	0.39	0.59	0.15	
f(iv)	0.25	MWD	0.50	0.54	0.27	0.50	0.45	
. /	0.25	SSI	0.50	0.64	0.32	0.59	0.15	

 $^{\$}$ f(i): support roots growth; f(ii): supply water for plants and edaphic fauna; f(iii): allow gases exchange between soil and atmosphere (soil aeration); and f(iv): ability resist erosion and physical degradation;  $^{\$}$ BD: bulk density, VESS: visual evaluation of soil structure, SWSC: soil water storage capacity index,  $K_6$ : field-saturated hydraulic conductivity, SAC: soil aeration capacity index, MaP: macroporosity, MWD: mean weight diameter of soil aggregates, SSI: structural stability index;  $^{\dagger}$ Indicator values obtained by linear transformation of the measured values (second step of index calculation).

#### 4.2.4 Data analyses

Data were tested for normality using Shapiro-Wilk's tests (p>0.05), indicating that data transformation was not required. An analysis of variance (ANOVA) was used to test LUC effects on soil physical properties. If the ANOVA results were significant (p<0.05), average soil physical property values were compared using Tukey's test (p<0.05). In addition, a Pearson's correlation analysis (p<0.01 and p<0.05) was performed using PROC CORR

procedure among soil physical attributes and SOC. The scores calculated for each soil physical function and the SPQI were compared using Tukey's test (p<0.05). All statistical analyses were carried out using the Statistical Analysis System – SAS v.9.3 (SAS Inc., Cary, USA).

#### 4.3 Results

#### 4.3.1 Bulk density (BD) and soil degree of compactness (SDC)

Conversion from native vegetation to pasture increased BD in all soil layers at all sites (Figure 1). Bulk density within the 0-30 cm layer was 23%, 19% and 28% lower in native vegetation compared to pasture at Lat\_17S, Lat\_21S and Lat\_23S, respectively. Conversion from pasture to sugarcane had no significant effect on BD except for the sugarcane field at Lat\_17S which was undergoing replanting (tillage operations) when soils were sampled. Tillage reduced BD in the surface layer (0-10 cm), but overall, BD tended to increase below 10 cm for all sites and land uses.

Land-use change had a significant effect on the SDC (Figure 1), showing an increasing transition from native vegetation to pasture of 14%, 19% and 23% at the Lat\_17S, Lat\_21S and Lat\_23S, respectively. Conversion from pasture to sugarcane had no significant effect on SDC, except in the upper layer (0-10 cm) of the sugarcane field undergoing replanting (Lat\_17S), where it was lower. Overall, SDC values ranged from 67 to 79% ( $\bar{x}$  73%) under native vegetation, 84 to 98% ( $\bar{x}$  90%) under pasture and 74 to 100% ( $\bar{x}$  90%) for sugarcane soils.

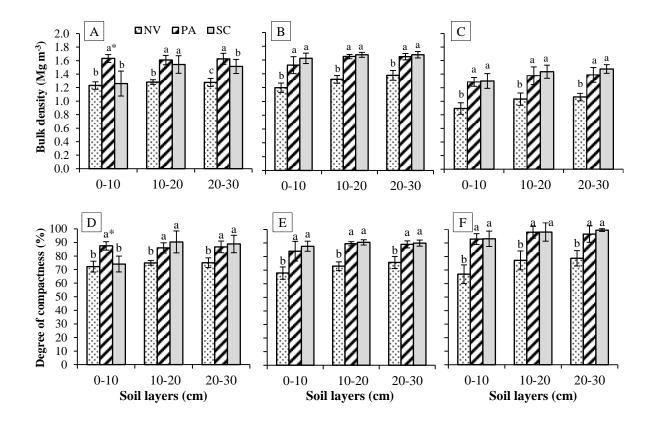


Figure 1 - Bulk density and soil degree of compactness from 0-10, 10-20 and 20-30 cm layers under native vegetation (NV), pasture (PA) and sugarcane (SC) at Lat\_17S, Lat\_21S and Lat\_23S in central-southern Brazil. \*Mean values within each site in same depth followed by the same letter do not differ among themselves according to Tukey's test (p<0.05). Error bars denote standard deviation of the mean

# 4.3.2 Soil porosity

Soil porosity components were very sensitive to LUC (Figure 2), especially macroporosity (MaP), Figure 2A-C. At Lat\_17S (Figure 2A), MaP was approximately 0.25 m³ m⁻³ in native vegetation and 0.17 m³ m⁻³ in pasture (0-30 cm). Tillage increased the MaP under sugarcane (0.27 m³ m⁻³) in the upper layer (0-10 cm), but within deeper layers, it was significantly lower than in pasture (10-20cm) or native vegetation (10-20 and 20-30 cm). At the Lat\_21S (Figure 2B) and Lat\_23S (Figure 2C) significant decreases in MaP were associated with LUC from native vegetation to pasture, with averages of 0.22 and 0.25 m³ m⁻³ in native vegetation and 0.07 (-68%) and 0.03 m³ m⁻³ (-88%) in pasture, respectively. Under sugarcane, the MaP values were similar to pasture, with average of 0.05 and 0.06 m³ m⁻³, respectively.

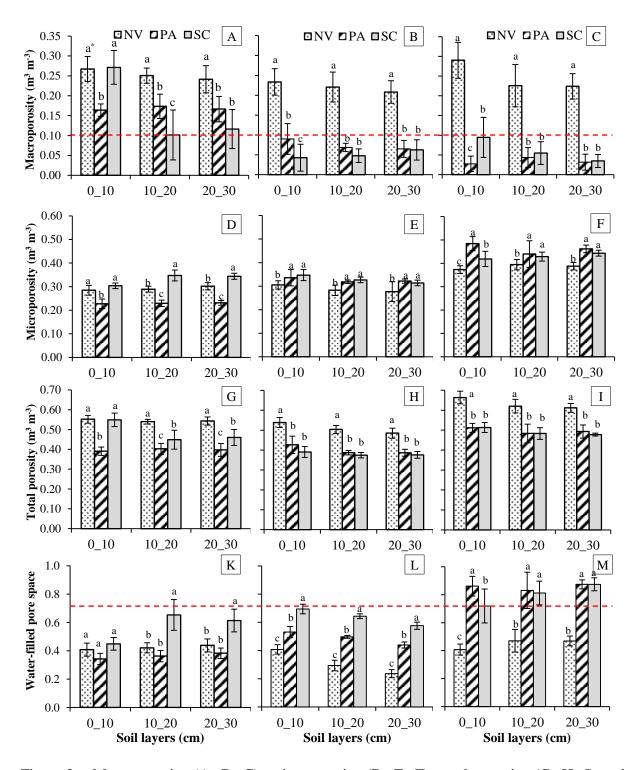


Figure 2 - Macroporosity (A; B; C), microporosity (D; E; F), total porosity (G; H; I) and water-filled pore space (K; L; M) from 0-10, 10-20 and 20-30 cm layers under native vegetation (NV), pasture (PA) and sugarcane (SC) at Lat\_17S (left), Lat\_21S (medium) and Lat\_23S (right) in central-southern Brazil. Red dashed lines indicate the critical limit to air diffusion (0.10 m³ m⁻³) and to start the anaerobic respiration (0.7) into the soil; \*Mean values within each site in same depth followed by the same letter do not differ among themselves according to Tukey's test (p<0.05); Error bars denote standard deviation of the mean

The microporosity (MiP) responded differently than the MaP to the effects of LUC, where MiP in sugarcane and pasture soils were greater than soils under native vegetation (Figure 2D-F). Total porosity (TP) decreased significantly due to LUC (Figure 2G-I), which was greater under native vegetation > sugarcane ≥ pasture. At the Lat\_17S site, MaP and MiP within the 0-10 cm layer were not significantly different under sugarcane compared to native vegetation. Overall, water-filled pore space (WFPS) increased significantly due to our LUC sequence (Figure 2K-M). At Lat\_17S and Lat\_23S the highest WFPS values (~0.6) were found in sugarcane soils. At Lat\_23S, WFPS values reached approximately 0.85 in pasture and sugarcane soils.

# 4.3.3 Soil water storage capacity (SWSC) and soil aeration capacity (SAC)

Overall, LUC induced significant changes in SAC and SWSC (Figure 3). At Lat\_17S, conversion from native vegetation to pasture had no significant impacts on SAC, with values of approximately 0.5. Significant reductions on SAC were verified in deeper layers (10-20 and 20-30 cm) in sugarcane, with values reaching 0.3 (Figure 3A). At Lat\_21S and Lat\_23S LUC effects on SAC (Figure 3B, C) were more significant, with higher values in soils under native vegetation averaging 0.57 and 0.43. Conversion from native vegetation to pasture decreased SAC to average values of 0.31 and 0.08, indicating reductions of 45% and 81%, respectively. Establishment of sugarcane under pasture, in general, had no significant influence on SAC.

Transition from native vegetation to pasture significantly increased SWSC at Lat\_21S and Lat\_23S reaching average values of 0.69 and 0.92 in pasture, respectively. Under sugarcane at Lat\_17S, significant increases in SWSC were observed in deeper layers when compared to native vegetation and pasture, with values of 0.72 for the 10-20 cm layer and 0.69 for the 20-30 cm layer (Figure 3A). At Lat\_21S and Lat\_23S there was no clear trend in upper soil layers (i.e., pasture had higher SWSC than sugarcane soils at one site, but a lower value at the other site). For deeper layers, there were no detectable differences between pasture and sugarcane soils (Figure 3B, C).

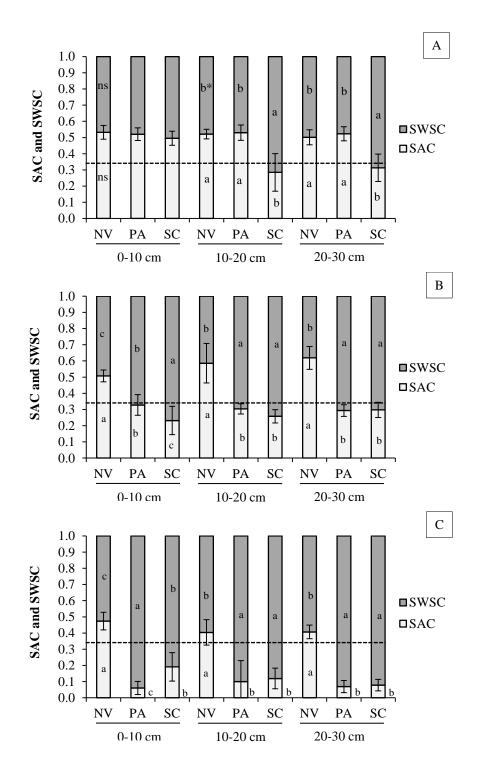


Figure 3 - Soil aeration capacity (SAC) and soil water storage capacity (SWSC) from 0-10, 10-20 and 20-30 cm layers under native vegetation (NV), pasture (PA) and sugarcane (SC) at Lat\_17S (A); Lat\_21S (B) and Lat\_23S (C) in central-southern Brazil. Dashed lines indicate the critical limit to root growth (ideal ratio = SAC 0.33 and SWSC: 0.66); \*Mean values within each site in same depth followed by the same letter do not differ among themselves according to Tukey's test (p<0.05); ns= non-significant; Error bars denote standard deviation of the mean

# 4.3.4 Field-satured hydraulic conductivity (K<sub>fs</sub>)

Higher  $K_{fs}$  values were obtained under native vegetation at Lat\_21S and Lat\_23S, with averages of 122 and 39 cm  $h^{-1}$ , respectively (Figure 4). At Lat\_17S,  $K_{fs}$  under native vegetation (147 cm  $h^{-1}$ ) was also high, but lower than within sugarcane which had just been tilled for replanting. The LUC from native vegetation to pasture resulted in drastic reductions (76, 98 and 97%) in  $K_{fs}$  at all sites. Under sugarcane,  $K_{fs}$  values were 97 and 98% lower than those measured within native vegetation soils at Lat\_21S and Lat\_23S (Figure 4B, C), but there was no difference when compared with values measured within pasture soils. On the other hand,  $K_{fs}$  increased from 36 cm  $h^{-1}$  in pasture to 250 cm  $h^{-1}$  in sugarcane at Lat\_17S (Figure 4A).

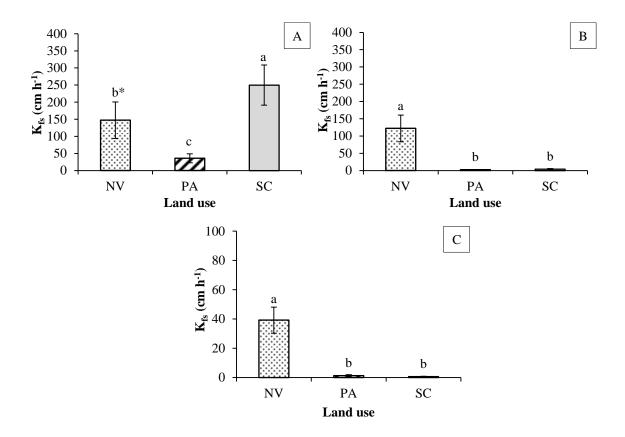


Figure 4 - Field-saturated hydraulic conductivity (K<sub>fs</sub>) under native vegetation (NV), pasture (PA) and sugarcane (SC) at Lat\_17S (A), Lat\_21S (B) and Lat\_23S (C) in central-southern Brazil. \*Mean values within each site followed by the same letter do not differ among themselves according to Tukey's test (*p*<0.05). Error bars denote standard deviation of the mean

#### 4.3.5 Soil resistance to penetration (SRP)

Soil resistance to penetration (SRP) was significantly higher in pasture and sugarcane compared to native vegetation, suggesting that LUC led to soil compaction and increased soil mechanical resistance to root growth (Figure 5). Conversion from native vegetation to pasture promoted more significant impacts on SRP, reaching maximum values of 2.51, 3.66 and 3.07 MPa at Lat\_17S, Lat\_21S and Lat\_23S, respectively.

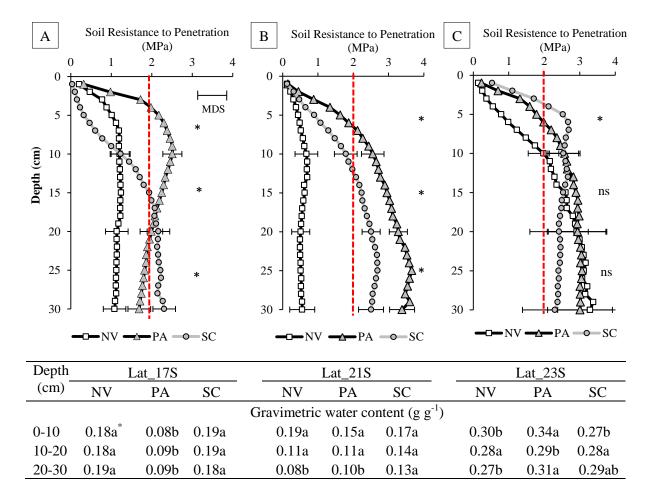


Figure 5 - Soil resistance to penetration and gravimetric water content from 0-30 cm layers under native vegetation (NV), pasture (PA) and sugarcane (SC) at Lat\_17S (A), Lat\_21S (B) and Lat\_23S (C) in central-southern Brazil. Red dashed lines indicate the critical limit to roots' growth (2 MPa);\*Mean values within each site in same depth followed by the same letter do not differ among themselves according to Tukey's test (*p*<0.05); ns: non-significant; Error bars denote standard deviation of the mean

There was an overall tendency to reduce SRP under sugarcane compared with pasture, with significant differences observed in the first 10 cm at Lat\_17S and in whole profile at Lat\_21S. However, soil moisture was lower in pasture than native vegetation and sugarcane at

Lat\_17S. Small differences were observed in soil moisture at Lat\_21S and Lat\_23S sites, although the differences were significant (p<0.05) in some layers (Figure 5).

# 4.3.6 Soil Stability Structural Index (SSI)

Overall native vegetation and pasture had higher SSI values (i.e., better soil structural stability) than sugarcane, mainly for the 0-10 cm layer (Figure 6). A decreasing trend on SSI values in depth was observed. Conversion from native vegetation to pasture induced site-specific changes on SSI. The SSI was significantly increased at Lat\_17S ( $\bar{x}$  SSI from 7.9 to 5.8%) and significantly decreased at Lat\_21S ( $\bar{x}$  SSI from 11.3 to 7.3%), and within deeper layers (10-20 and 20-30 cm) at Lat\_23S ( $\bar{x}$  SSI from 7.2 to 6.3%). Conversion from pasture to sugarcane consistently promoted significant decreases in SSI, with average SSI values for sugarcane soils of 4.3, 7.1 and 4.1% at Lat\_17S, Lat\_21S and Lat\_23S, respectively.

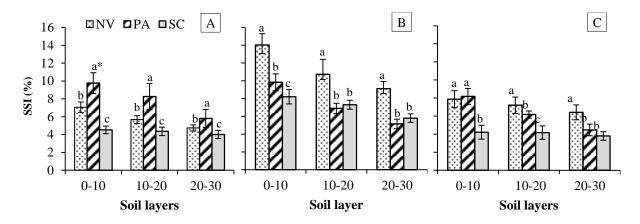


Figure 6 - Soil structural stability index - SSI (%) from 0-10, 10-20 and 20-30 cm layers under native vegetation (NV), pasture (PA) and sugarcane (SC) at Lat\_17S (A), Lat\_21S (B) and Lat\_23S (C) in central-southern Brazil. \*Mean values within each site in same depth followed by the same letter do not differ among themselves according to Tukey's test (*p*<0.05). Error bars denote standard deviation of the mean

#### 4.3.7 Correlation between soil physical properties and SOC

Significant correlation was observed for 72 (p<0.01) and 76 (p<0.05) of 91 pairs (combinations) of soil physical properties from LUC sites in central-southern Brazil (Table 2).

Table 2 - Pearson's correlation coefficients (r) and probability of error (p) among soil physical properties<sup>§</sup> and with soil organic carbon (SOC) in native vegetation, pasture and sugarcane land uses in central-southern Brazil

	Clay	BD	SDC	MaP	MiP	TP	WFPS	SWSC	SAC	SRP	K <sub>fs</sub>	MWD	VESS	SSI
	1.000*	-0.618	0.137	0.013	0.770	0.650	0.470	0.416	-0.416	0.289	-0.151	-0.080	0.138	-0.415
Clay		$< 0.0001^{\dagger}$	0.0402	0.8431	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	0.1828	0.2446	0.2489	< 0.0001
BD		1.000	0.679	-0.686	-0.282	-0.992	0.203	0.297	-0.297	0.291	-0.289	-0.059	0.347	0.009
БD			< 0.0001	< 0.0001	< 0.0001	< 0.0001	0.0020	< 0.0001	< 0.0001	< 0.0001	0.0099	0.4082	0.0040	0.1818
SDC			1.000	-0.893	0.394	-0.651	0.722	0.793	-0.793	-0.429	-0.497	-0.112	0.530	-0.367
SDC				< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	0.1199	< 0.0001	< 0.0001
MaP				1.000	-0.492	0.687	-0.748	-0.859	0.859	-0.507	0.676	0.026	-0.628	0.206
Mai					< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	0.7144	< 0.0001	0.0020
MiP					1.000	0.290	0.785	0.801	-0.801	0.333	-0.406	-0.010	0.369	-0.284
17414						< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	0.0002	0.9046	0.0020	< 0.0001
TP						1.000	-0.179	-0.284	0.284	-0.268	0.346	0.021	-0.325	-0.005
							0.007	< 0.0001	<0.0001	<0.0001	0.0019	0.7667	0.0070	0.9360
WFPS							1.000	0.922	-0.922	0.356	-0.437	-0.172	0.459	-0.368
								<0.0001	< 0.0001	<0.0001	<0.0001	0.0161	<0.0001	<0.0001
SWSC								1.000	<b>-1.000</b> <0.0001	<b>0.498</b> <0.0001	<b>-0.544</b> < 0.0001	-0.113 0.1161	<b>0.525</b> <0.0001	<b>-0.346</b> <0.0001
									1.000	<b>-0.498</b>	0.544	0.1101	-0.525	0.346
SAC									1.000	<0.0001	< 0.0001	0.113	< 0.0001	< 0.0001
~~~										1.000	-0.624	0.015	0.682	-0.429
SRP										1.000	< 0.0001	0.8260	< 0.0001	< 0.0001
T/											1.000	0.228	-0.524	-0.1599
$\mathbf{K}_{\mathbf{fs}}$												0.0464	0.0010	0.1617
MWD												1.000	-0.285	0.499
MIND													0.0221	< 0.0001
VESS													1.000	-0.396
<u> </u>														0.0006
SOC	0.742	-0.717	-0.125	0.134	0.646	0.685	0.245	0.227	-0.227	0.030	-0.255	0.331	-0.054	0.205
	< 0.0001	< 0.0001	0.0626	0.0460	< 0.0001	< 0.0001	0.0002	0.0006	0.0006	0.6393	0.0217	< 0.0001	0.6540	0.0015

 $^{\$}$ Clay: clay content; BD: bulk density, SDC: soil degree of compactness, MaP: macroporosity, MiP: microporosity, TP: total porosity, WFPS: water-filled pore space, SWSC: soil water storage capacity, SAC: soil aeration capacity, SRP: soil resistance to penetration,  $K_{fs}$ : field-saturated hydraulic conductivity; MWD: mean weight diameter of soil aggregates, VESS: visual evaluation of soil structure, SSI: structural stability index;  $^{*}$ Pearson's correlation coefficients significant (p<0.01) are in bold. The correlations were calculated using "n"= 243, except between  $K_{fs}$  and VESS and other properties, which were calculated using "n"= 81 and "n"= 72, respectively;  $^{†}$ Probability of error (p).

The highest correlation was observed between BD and TP (-0.99). The SWSC and SAC indexes were better correlated (>0.80) with WFPS, MaP and MiP. For  $K_{\rm fs}$  there was high positive correlation with MaP (0.68) and negative correlation with SRP (-0.62). VESS scores were correlated with most physical properties, highlighting greater positive and negative correlations with SRP (0.68) and MaP (-0.63). There was good correlation (0.50) between MWD and SSI.

Except for SRP and VESS scores, soil physical properties were strongly correlated with SOC, which was highest in soils with more clay. Overall, increased SOC resulted in a reduction in BD, SAC, SDC and  $K_{fs}$  and an increase in TP, MiP, MaP, WFPS, SWSC, MWD and SSI.

# 4.3.8 Soil physical quality assessement

Minimum dataset indicator scores for the two scenarios analyzed are shown in Figure 7. Conversion from native vegetation to pasture promoted severe reductions in  $K_{fs}$  and MaP scores and moderate reductions in VESS, BD and SSI scores. In contrast, pasture soils had the highest SAC, SWSC and MWD scores. For scenario I (Figure 7A), which did not include the sugarcane field that was being replanted, there was a major decrease in indicator scores, while for scenario II (Figure 7B) which included all sugarcane fields, tillage associated with replanting in one of three fields resulted in a marked increase in the  $K_{fs}$  score. Furthermore, small improvements in MaP, SAC and VESS scores were also verified. On the other hand, tillage reduced MWD and SSI scores.

The overall scores for each soil physical function and SPQI are presented in Table 3. For f(i) related to supporting root growth, a significant reduction in scores was observed in the sequence native vegetation > pasture > sugarcane. Under pasture this function decreased 24% (0.76) compared to maximum performance (1.00) under native vegetation. Tillage operations carried out during sugarcane replanting promoted significant improvement in f(i), increasing the score from 0.68 to 0.72 score, although, this was not enough to offset impacts of conversion from pasture to sugarcane. For f(ii) related to water supply for plants and edaphic fauna, conversion from native vegetation to pasture and then to sugarcane without tillage ( $SC_{wr}$ ) decreased scores markedly (from 0.84 to 0.55 and 0.43). However, the tillage operation in sugarcane increased water entry into the soil and resulted in scores (0.83) similar to those found in native vegetation. The f(iii) related to soil aeration was affected similarly to f(i), in which the scores decreased in the sequence native vegetation > pasture > sugarcane.

Tillage improved soil aeration compared to sugarcane without tillage ( $SC_{wr}$ ) as indicated by scores of 0.44 and 0.59. For f(iv) related to ability for resist against erosion and soil degradation, the results point out that under native vegetation and pasture there was no difference (0.93 and 0.94). On the other hand, within sugarcane fields, lower scores were found (0.59 in sugarcane and 0.68 in sugarcane without tillage). Beyond that, the f(iv) scores indicated that the sugarcane field being replanted, had a poor ability to resist erosion and subsequent degradation.

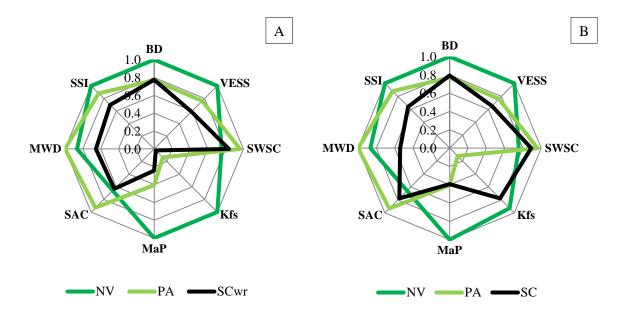


Figure 7 - Overall scores of the soil minimum dataset indicators for the 0-30 cm layer in the land-use change areas in central-southern Brazil, where A) scenario I: native vegetation (NV), pasture (PA) and sugarcane without replanting tillage ( $SC_{wr}$ ) when sampled; and B) scenario II: NV, PA and sugarcane including a field with replanting tillage (SC)

Overall, the additive SPQI was effective in classifying land uses according to the hypothesized potential of soil degradation in response to land-use intensification (Table 3). Therefore, conversion from native vegetation to pasture and then to sugarcane led to a reduction of SPQI. Finally, the SPQI was sensitive enough to identify that soil tillage operations had been carried out in the replanted sugarcane field, thus confirming its general ability to improve a soil's capacity to perform its physical functions, even though it had short-term effects.

Table 3 - Overall scores for each soil physical function and Soil Physical Quality Index (SPQI) values, within the 0-30 cm layer in the native vegetation (NV), pasture (PA) and sugarcane with replanting tillage (SC) and without replanting tillage ( $SC_{wr}$ ) in central-southern Brazil

Land use		SPQI				
Land use	f(i)	f(ii)	f(iii)	f(iv)	51 Q1	
NV	1.00 a*	0.84 a	0.83 a	0.93 a	0.90 a	
PA	0.76 b	0.55 b	0.67 b	0.94 a	0.73 b	
SC	0.72 c	0.83 a	0.59 c	0.59 c	0.68 c	
$SC_{wr}$	0.68 d	0.43 c	0.44 d	0.68 b	0.56 d	

 $<sup>\</sup>S$  f(i): support roots growth; f(ii): supply water for plants and edaphic fauna; f(iii): allow gases exchange between soil and atmosphere (soil aeration); and f(iv): ability to resist erosion and physical degradation; \*Mean values within each soil function followed by the same letter do not differ among themselves according to Tukey's test (p<0.05).

#### 4.4 Discussion

# 4.4.1 Impacts of the LUC from native vegetation to pasture on soil physical attributes

Long-term conversion (>30 years) from native vegetation to extensive pasture led to significant soil compaction (Figure 1). Sampled pastures had BD values that were >1.2 Mg m<sup>-1</sup> <sup>3</sup> in clay soils and >1.6 Mg m<sup>-3</sup> in sandy soils, suggesting a limiting condition for optimum plant growth on tropical soils (REICHERT; REINERT; BRAIDA, 2003; REICHERT et al. 2009). The SDC provides a more robust evaluation of soil compaction, because optimum BD and porosity values for crop growth vary considerably among soils (i.e., BD is strongly dependent on mineral composition and SOC). Optimal SDC had been associated with values between 80 and 90% (REICHERT et al., 2009). Our results suggest that soil compaction was close to the upper limit and even limiting grass growth, especially in high clay soils at Lat\_23S, where the SDC averaged 96% for the 0-30 cm layer. Soil compaction in pastures is primarily caused by SOC depletion and cattle trampling. Recently, Franco et al. (2015) verified that SOC losses, induced by conversion of native vegetation to pasture, resulted in an average C emission rate of 0.4 Mg ha<sup>-1</sup> yr<sup>-1</sup> in these sampled sites. Several studies have reported the crucial role that SOM plays with regard to resisting structural degradation in pastures (e.g., FONTE at al., 2014; NESPER et al., 2015). Low C inputs also have adverse implications on nutrient cycling and biological activity, further decreasing pasture productivity and increasing soil degradation. In addition, compressive forces due to continuous cattle trampling can induce soil compaction that has negative effects on soil physical quality and plant growth in pastures (GREENWOOD; McKENZIE, 2001; PIETOLA; HORN; YLI-HALLA, 2005; HERBIN et al., 2011; COSTA et al., 2012).

In this study, increased soil compaction reduced soil aeration and resulted in strong negative correlations between BD, SDC, and soil aeration properties (Table 2). Macroporosity and the SAC index were severely affected by conversion from native vegetation to pasture, with MaP values (Figure 2) falling below the critical limit of 10% (XU; NIEBER; GUPTA, 1992) and SAC values falling below 0.33 (Figure 3), which has been suggested as an ideal ratio for good soil aeration. These results are consistent with several other studies (e.g., HERBIN et al., 2011; RESENDE et al., 2012). In contrast, average values for MiP, WFPS and SWSC in pasture soils were higher than for native vegetation soils (Figures 2 and 3). Within the most compacted soils (Lat\_23S), WFPS values exceeded 0.7 indicating excess water and reduced soil aeration, which can induce denitrification (WIENHOLD et al., 2009). Furthermore, the SWSC reached 0.92 which is much higher than the 0.66 value that is considered ideal for good balance between air and water content in the soil (REYNOLDS et al., 2002; 2009). Although the pasture soils have greater SWSC than native vegetation soils, it does not mean that this water is easily accessible to roots. Soil compaction decreases the size and continuity of pores increasing water retention at lower soil water potentials and decreasing overall soil water availability.

Surface soil compaction also leads to reduced K<sub>fs</sub> (REICHERT et al., 2009; HUNKE et al., 2015b). Our results showed K<sub>fs</sub> reductions ranging from 76 to 98% in pasture soil compared to native vegetation (Figure 4). These cause and effect relationships were confirmed by inverse correlations between K<sub>fs</sub> versus BD and SDC and positive correlations between K<sub>fs</sub> versus TP and MaP (Table 2); a response similar to that found by Reichert et al. (2009). Several studies showing reduction in water infiltration and K<sub>fs</sub> from LUC effects in Brazilian Cerrado were compiled by Hunke et al. (2015a). In recent field-study Hunke et al. (2015b) found that conversion from native vegetation to pasture significantly reduced infiltration rates (-96%) and K<sub>fs</sub> (-92%), indicating high susceptibility to surface erosion and an increased potential for lateral nutrient transport to the surface waters, as reported by Gucker, Boechat and Giani (2009). Reductions of K<sub>fs</sub> were also consistently documented from trampling effects on grazing pasture (e.g., GREENWOOD; McKENZIE, 2001) even under low-intensity grazing (PIETOLA; HORN; YLI-HALLA, 2005). Higher soil mechanical resistance to root growth in pasture (Figure 5) is a key-factor limiting water access to the plants. We found SRP values >2 MPa (suggested as a critical limit to root growth) in all pasture sites. Geissen et al. (2009) concluded that permanent pasture in Mexico led to higher SRP and soil compaction. Costa et al. (2012) also found that an increased SRP associated with high-intensity grazing promoted lower root growth of *Panicum maximum*.

# 4.4.2 Impacts of the LUC from pasture to sugarcane on soil physical attributes

Conversion from long-term extensive pasture to sugarcane induced slight soil physical degradation. Sugarcane cultivation resulted in SOC losses (FRANCO et al., 2015) and relies on heavy mechanization (SOUZA et al., 2014; 2015), but significant differences in soil compaction (Figures 1 and 5), as evidenced by porosity (Figure 2), SAC and SWSC (Figure 3) and  $K_{fs}$  (Figure 4), were not observed when compared to pasture areas. Other studies also found no differences in BD and TP (RESENDE et al., 2012) or  $K_{fs}$  (HUNKE et al., 2015b) between long-term extensive pasture and sugarcane fields in Brazilian Cerrado, even though, both land uses had lower soil physical quality than areas with native vegetation.

Although soil physical degradation due to conversion from pasture to sugarcane was less intense than from native vegetation to pasture, soil physical conditions under sugarcane may be limiting plant growth. Soil compaction indicators (BD and SDC) were higher than established critical limits and the SRP values were >2 MPa, suggesting poor root growth (OTTO et al., 2011; SOUZA et al., 2014). Soil compaction in sugarcane fields due to intensive machine traffic is well documented in the literature (BRAUNACK; McGARRY, 2006; BAQUEIRO et al., 2012; CASTRO et al., 2013; SOUZA et al., 2014; 2015). Compressive forces are applied to soil by tractors, trailers and harvesters, causing a reduction in pore space and an increase in soil strength. Consequently, soil aeration properties were significantly affected, as shown by MaP <10% and SAC <0.33, which suggests a discontinuity in pathways of air-filled pores for gas diffusion (XU; NIEBER; GUPTA, 1992). The WFPS values were close 0.6 (Lat\_17S and Lat\_21S) or >0.7 (Lat\_23S), indicating limiting soil aeration which can induce greenhouse gas emissions to the atmosphere.

Soils within the sampled sugarcane fields showed a high risk for degradation since SSI values were generally  $\leq$ 5%. Lower soil structural resilience results in decreases in macroaggregation and MWD, lower  $K_{fs}$ , and poor VESS scores, and it suggests that areas under sugarcane expansion are at increased risk for runoff and soil erosion, both which may have adverse effects on soil and water quality.

Tillage performed during conversion of pasture to sugarcane and carried out during sugarcane replanting may be pointed out as the key-factor for alleviating soil compaction and maintaining soil physical conditions in a slightly better or similar condition in sugarcane

fields as under pasture. The effects of soil tillage during sugarcane replanting were verified at Lat\_17S, especially in the top layer (0-10 cm). Measurements showed a decrease in SDC (Figure 1) and SRP (Figure 5A) and an increase in MaP (Figure 2A), total porosity (Figure 2G), SAC (Figure 3A) and  $K_{fs}$  (Figure 4A). Tillage operations promote soil disturbance, create large pores and consequently alleviate soil compaction, improving soil physical conditions for sugarcane root growth. Unfortunately, soil physical remediation was restricted to the top soil layer, indicating that these mechanical operations were not successful in restoring the entire soil profile. Understanding the real impact of tillage is important for farmers, because the operations are expensive (CHAMEN et al., 2015) and when inadequately carried out could have adverse consequences on sugarcane yield.

Our findings also suggest that tillage performed for sugarcane replanting had a short-term effect on soil physical properties. This agreed with study by Centurion et al. (2007) who verified better soil physical conditions just after tillage followed by progressive soil physical degradation from the first to fourth ration. Short-term tillage improvements in soil physical quality may be associated with the disruption of macroaggregates (SIX; ELLIOTT; PAUSTIAN, 2000; FRANCO, 2015) which also favors more intense SOC losses (SILVA-OLAYA et al., 2013) and further structural degradation of the soil (Figure 6). This soil structural degradation is magnified by successive compaction events associated with machinery traffic, and leads to greater soil compaction throughout the sugarcane cycle (BRAUNACK; McGARRY, 2006; BAQUEIRO et al., 2012).

#### 4.4.3 Sugarcane expansion *versus* soil physical quality

The minimum dataset used to assess soil physical quality (0-30 cm layer) was able to rank LUC effects according to soil physical degradation (Figure 7; Table 3). Overall, the highest index for soil physical functioning was verified for soils under native vegetation, suggesting that they were functioning at 90% of their potential capacity. The soil physical function scores showed that under native vegetation the soil has full capacity to support root growth, since higher C inputs combined with the absence of soil perturbation ensures structural arrangement providing good soil physical conditions for growth and deep penetration of roots. The large air-filled porosity observed under native vegetation provides adequate soil aeration, but the reduced soil water retention induces an unbalanced ratio between air and water in the soil. Therefore, scores of f(ii) and f(iii) were not maximum (0.84 and 0.83) in these soils. Finally, higher SOM contents under native vegetation areas favor

formation of large aggregates and provide greater soil structural stability. Those inherent characteristics also suggest that soils beneath native vegetation have the ability to resist erosion and physical degradation.

Conversion from native vegetation to pasture promoted significant decreases in the soil's capacity to perform its physical functions (SPQI = 0.73). The extensive low-input management commonly used in pastures for more than 30 years led to soil compaction and reduced the soil's capacity to support of root growth - f(i), to supply water for plants and edaphic fauna - f(ii) and to allow gases exchange between soil and atmosphere - f (iii) compared to native vegetation soils. Despite that, based on our minimum dataset, we verified that pasture soils had high capacity to resist erosion and physical degradation. These results may be linked to greater macrofauna activity in pasture areas as suggested by Lavelle et al. (2014) and the aggressive root system of Poaceae (i.e., *Brachiaria* and *Cynodon*) which plays crucial role in soil aggregation and improvement of soil structural quality, especially in the top few centimeters (FONTE et al., 2014).

Sugarcane expansion into long-term extensive pasture land led to slightly negative changes in soil physical quality over time. Our findings showed that management practices adopted in sugarcane fields result in more dynamic changes in soil physical properties. This was illustrated by the one sugarcane field where recent tillage operations resulted in an overall soil capacity to perform physical functions of 68%, which was close to that observed in pasture (73%). Mechanical tillage alleviated soil compaction resulting in better aeration, water availability and favorable conditions for root growth. However, intensive sugarcane management using big and heavy machines promotes soil compaction over time. Our results indicated that soils in the fourth and fifth ratoon of sugarcane are functioning at only 56% of their potential. The measurements also suggest that sugarcane expansion into degraded pastures should follow management strategies that minimize soil physical degradation to ensure long-term economic and environmental sustainability for this production system.

Our findings confirmed an increasing risk for soil loss and degradation due to erosion in sugarcane fields that is aggravated by tillage operations during the sugarcane replanting. Soil perturbation increases structural degradation and SOC losses, decreasing the soil's capacity to resist erosion. In addition, after tillage operations, uncovered soil remains for weeks or months, creating increased water and wind erosion risks. Throughout the sugarcane production cycle, the main driver to soil erosion is compaction, which has deleterious effects on soil structure and water infiltration. Soil erosion is reported as one of major threats to sustainability of Brazilian sugarcane production system (MARTINELLI; FILOSO, 2007). In

the state of São Paulo, which is the core of the ethanol industry in Brazil, estimated rates of soil erosion in sugarcane fields are close to 60 Mg ha<sup>-1</sup> yr<sup>-1</sup> while in forests and pasture erosion rates do not exceed 2 Mg ha<sup>-1</sup> yr<sup>-1</sup> (WEILL; SPAROVEK, 2008). Thus, future research needs to evaluate the degree of soil erosion and associated soil and water resource degradation due to LUC in Brazilian sugarcane belt.

Based on our results, we emphasize the urgent need to adopt strategies to mitigate adverse effects of current sugarcane management practices on soil physical quality, minimizing the environmental impact of biofuel production. The season for sugarcane replanting, which extends from September to March, coincides with the highest rainfall period in central-southern Brazil. Therefore, a crop rotation plan including commercial crops [e.g., corn (Zea mays) or soybean (Glycine max)] or cover crops (e.g. Crotalaria juncea or Mucuna aterrima) could be implemented to prevent soil degradation by erosion, improve C allocation throughout the soil profile, N-fixation and nutrient cycling, break pest cycles and alleviate soil compaction. Machinery entry on the field, especially during harvest operations should be done under soil moisture conditions less favorable to compaction. Mathematical models that include soil and agricultural machinery characteristics should be developed as an auxiliary management guide to predict the impact of the entry of machines on soil physical quality (for example, see KELLER et al., 2015). In addition, more investments and research are necessary for developing machines adapted for implementing controlled traffic in sugarcane production system and for studying sugarcane planting strategies with different inter-row spacing, keeping non-trafficked zones between crop rows, as well as specific tillage zones, as discussed by Bangita and Rao (2012).

Finally, we suggest that soil physical quality changes should be evaluated and monitored throughout the sugarcane life cycle using indicators selected to calculate an overall SPQI, such as BD, VESS, MaP, SAC, K<sub>fs</sub>, SWSC, MWD and SSI. If sufficient resources (time and money) are not available for a detailed soil assessment including all of these indicators, we suggest using the VESS, which is an on-farm, inexpensive, simple to perform and easy to understand approach for evaluating soil physical quality. VESS scores are well-correlated with soil physical properties, and thus provide a first approximation of overall soil structure quality that can help farmers and land managers evaluate the impact of various management practices on sugarcane production.

#### 4.5 Conclusions

We investigated soil physical quality impacts of LUC associated with sugarcane expansion across the primary sugarcane-producing region in Brazil. Our hypotheses were confirmed. Conversion from native vegetation to long-term, extensive pasture increased soil compaction (i.e., higher values of BD, SDC and SRP), decreased aeration porosity and water hydraulic conductivity, and consequently, created an unbalanced ratio between water- and air-filled pore space in the soil. Based on our SPQI the native vegetation and pasture soils are functioning at 90 and 73% of their potential capacity, respectively.

The LUC from pasture to sugarcane induced slight soil physical quality degradation and resulted in soils functioning at between 56 and 68% of their capacity. Overall, sugarcane fields have critical soil compaction (BD and SDC) that is decreasing soil pore space (TP), soil aeration (MaP and SAC) and  $K_{\rm fs}$  and increasing soil strength (SRP) and the risk for soil erosion and structural degradation. Tillage operations performed approximately every five years when sugarcane is replanted have some short-term positive effects on soil physical quality; although overall it reduces the soil's ability to resist degradation due to erosive processes. Therefore, sugarcane expansion in Brazil should require the adoption and monitoring of management strategies that increase SOM and minimize soil compaction to reduce soil physical quality degradation and thus improve sustainability of sugarcane production in Brazil.

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# 5 ASSESSING SOIL STRUCTURAL QUALITY UNDER BRAZILIAN SUGARCANE EXPANSION AREAS USING VISUAL EVALUATION OF SOIL STRUCTURE (VESS)

### **Abstract**

Increasing global demand for biofuel has accelerated land-use change (LUC) in Brazil, primarily through the planting of sugarcane (Saccharum officinarum) to replace degraded pastures. The intensive mechanization associated with this LUC under tropical soils in Brazil has increased concerns regarding structural quality. Through decades of research focused on identifying sensitive indicators of soil degradation due to land use and management, the Visual Evaluation of Soil Structure (VESS) method has emerged as a simple, fast, reliable and accurate semi-quantitative approach for assessing soil structure changes. VESS integrates soil properties related to size, strength and porosity of aggregates, and root characteristics into a single score (Sq - structural quality) that ranges from 1 (good structural quality) to 5 (poor structural quality). Although the VESS method was developed for temperate soils, it has been used successfully as an indicator of soil and crop management practice effects on structural quality of tropical and subtropical soils. Our objectives were to evaluate soil structural quality changes associated with a LUC sequence (i.e., native vegetation to pasture to sugarcane) at three sites under Oxisols, Alfisols and Ultisols across central-southern Brazil using the VESS; and to correlate VESS scores with quantitative measurements of soil physical properties. Average VESS scores were 2.0, 2.7, and 3.1 for native vegetation, pasture, and sugarcane, respectively. Overall the VESS method was able to detect soil structural quality changes under LUC for sugarcane cultivation, indicating a decrease in soil quality from native vegetation through pasture to sugarcane. The VESS scores were significantly correlated with quantitative soil physical property measurements, suggesting VESS is a reliable indicator of soil structural quality in tropical soils. A VESS critical score (Sq= 3.0) seems to be suitable as a guide for management decisions. We conclude that VESS scores provide an efficient method to identify impacts of sugarcane expansion on soil structural quality, and recommend that VESS assessment be incorporated into monitoring protocols for evaluating not only sugarcane expansion areas, but also overall soil quality/health in Brazil.

Keywords: Land-use change; Visual method; Soil physical properties

### 5.1 Introduction

The large-scale cultivation of biofuel crops for bioenergy has caused widespread transformations in land use worldwide, either directly or through the replacement of other managed lands with food crops (FOLEY et al., 2005). Brazil, the world's largest producer of sugarcane ethanol [29.2 billion of liters per year (COMPANHIA NACIONAL DE ABASTECIMENTO, 2016)], is one of principal hotspots of these land-use changes (LAPOLA et al., 2014). In the last decade (2005-2015), the sugarcane area has increased from

5.8 to 9.0 Mha, with most of area being concentrated within the central-southern region (COMPANHIA NACIONAL DE ABASTECIMENTO, 2016). Nevertheless, in order to meet the projected domestic Brazilian supplies for ethanol by 2021, an additional 6.4 Mha of sugarcane will be required (GOLDEMBERG et al., 2014). Historically, in central-southern Brazil the expansion of agriculture has been done through the removal of native vegetation and the introduction of pasture, but often using poor management practices (LAPOLA et al., 2014), which has resulted in vast areas of low-productivity pasture (STRASSBURG et al., 2014) and poor soil quality (CHERUBIN et al., 2015; FRANCO et al., 2015). Therefore, conversion of extensive pasturelands to sugarcane is the most opportune and widespread landuse change to meet projected ethanol demands, thus avoiding direct competition for land with food crops and natural ecosystems (GOLDEMBERG et al., 2014; STRASSBURG et al., 2014).

Conversion of pastureland to sugarcane production requires intensive mechanization through large and heavy agricultural machines that impose unavoidable modifications to soil structure and to soil physical properties. Recent studies have shown that the LUC from pasture to sugarcane depletes soil organic carbon (SOC) stocks (MELLO et al., 2014; FRANCO et al., 2015) and increases soil susceptibility to compaction due to heavy and intense traffic during mechanical harvest and transport (BRAUNACK; McGARRY, 2006; LOZANO et al., 2013; SOUZA et al., 2014). Therefore, soil compaction has been identified as the main concern in modern sugarcane production systems in Brazil (LOZANO et al., 2013; SOUZA et al., 2014) and has been characterized by increases in bulk density, leading to a reduction in macroporosity and water infiltration (BRAUNACK; McGARRY, 2006; CASTRO et al., 2013; SOUZA et al., 2014). Furthermore, these physically and structurally degraded soils decrease root growth and sugarcane yield (OTTO et al., 2011; BAQUERO et al., 2012; SOUZA et al., 2014).

Impacts of sugarcane production on soil physical and structural quality have been traditionally assessed using quantitative soil physical properties such as bulk density, soil porosity, soil resistance to penetration, aggregate stability and macroaggregation that are indirectly related to soil structure (OTTO et al., 2011; LOZANO et al., 2013; SOUZA et al., 2014). However, measurements of these soil properties are relatively time consuming and expensive to analyze, and each sample provides an indication of the structural condition only at the point where it was taken within the soil profile (NEWELL-PRICE et al., 2013). Alternatively, visual methods of assessment of soil structure are characterized as simple, inexpensive, reliable and accurate, easy to perform, capable of producing results quickly and

being easily understood by researchers, advisers and farmers (BALL et al., 2007; GUIMARÃES et al., 2011; ASKARI; CUI; HOLDEN, 2013; GIAROLA et al., 2013; MONCADA et al., 2014). To make the assessment of soil physical quality simpler, spade methods based on the assessment of topsoil (first 25 cm) have been widely developed. This includes the "Visual Soil Assessment" (VSA) method developed by Shepherd (2009) and the "Visual Evaluation of Soil Structure" (VESS) method described by Ball, Batey and Munkholm (2007) and improved by Guimarães, Ball and Tormena (2011).

The VESS method was developed from the Peerlkamp method (BALL; BATEY; MUNKHOLM, 2007) and has been recognized as one of the simplest methods to employ while still including a variety of soil structure and rooting assessments (GUIMARÃES et al., 2013) in a way that Ball, Munkholm and Batey (2013) concluded can evaluate more than soil structural quality. The VESS method involves taking an undisturbed soil sample, breaking it up and visually assessing the size, shape, porosity and strength of aggregates, presence and state of roots and soil color (BALL; BATEY; MUNKHOLM, 2007; GUIMARÃES; BALL; TORMENA, 2011). These soil characteristics are integrated into a single numeric score (Sq structural quality) that ranges from 1 (good structural quality) to 5 (poor structural quality) that can subsequently be submitted to statistical analysis for decision making (MUNKHOLM; HECK; DEEN, 2013). Another distinctive feature of VESS is its ability to distinguish between topsoil layers width different structural characteristics. It is important, because evaluating soil layers individually rather than giving only a weighted average score enables to improve the choice of management practices adopted to preserve or improve overall structural soil quality (GIAROLA et al., 2010; GUIMARÃES; BALL; TORMENA, 2011; GUIMARÃES et al., 2013).

In recent years, VESS has been used to evaluate soil structure and soil quality under different land use and soil management (e.g., IMHOFF et al., 2009; GIAROLA et al., 2010; 2013; ASKARI et al., 2013; GUIMARÃES et al., 2013; MUELLER et al., 2013; MUNKHOLM; HECK; DEEN, 2013; CUI; ASKARI; HOLDEN, 2014; MONCADA et al., 2014; ABDOLLAHI et al., 2015). However, we are not aware of any studies using VESS for evaluating soil structure changes induced by sugarcane expansion in Brazilian tropical soils. Therefore, our objective was to apply the VESS method for assessing soil structural quality changes associated with a LUC sequence (i.e., native vegetation to pasture to sugarcane) at three field-sites across central-southern Brazil. We tested the hypotheses that i) the VESS method is able to efficiently detect soil structure changes induced by LUC and is a suitable indicator of soil quality in areas under sugarcane expansion in Brazil; ii) VESS Sq score is

negatively affected by LUC from native vegetation to pasture to sugarcane and, iii) VESS Sq score is correlated with quantitative soil physical properties.

### **5.2 Material and Methods**

### 5.2.1 Study sites and experimental design

The study sites and experimental design were described in the 2.2.1 and 2.2.2 items.

### 5.2.2 Sampling and VESS measurements

Soil sampling was carried out in January 2014. At each land-use site were collected four samples (i.e., totaling 36 soil samples). Sampling points were positioned in representative locations within each land use sampled. In native vegetation areas we avoided sampling close to ant or termite nests, burrows of wild animals and big trees. In pasture areas, which were continuously and uniformly grazed, our major caution was to avoid sampling on the preferential cattle trampling paths, where the soil is much more compacted. Except at Lat\_17S where the soil had been recently tilled for sugarcane replanting, all sampling points in sugarcane fields were located within the inter-row position, which is homogeneously tracked during harvest operations.

The VESS assessment and signature of scores were completed as described by Guimarães, Ball and Tormena (2011). At each sampling core, a mini-trench (30 x 30 x 30 cm in size) was dug out and then, using a spade, an undisturbed sample (20 x 10 x 25 cm deep – 5000 cm³ volume) was collected and transferred to a plastic tray. The soil evaluation included manual breakdown of soil aggregates along its fracture lines, identification of layers of contrasting structure, measurement of layer depth and assignment of a score by comparing the structure of the sample with the VESS chart, which contains descriptions and pictures of each proposed soil structure quality. The parameters used to describe soil structure included size, visible porosity, color, shape and strength of aggregates as well as the presence, number and distribution of roots (GUIMARÃES; BALL; TORMENA, 2011).

Since distinct layers were identified and a score was assigned for each layer identified according to the standard chart description, a final weighted score for each soil sample was calculated using equation 1.

$$VESS_{Sq} = \sum_{i=1}^{n} \frac{Sqi Di}{TD}$$
(1)

where, VESS<sub>Sq</sub> is the overall VESS score, Sqi and Di are respectively the score and depth of each identified soil layer, and TD is the total depth of soil sample.

Two additional strategies of scoring were also used: first, a weighted average of the Sq score was taken for the top (0-10 cm) and bottom (10-25 cm) soil layers at each site, and second, an overall Sq was taken using the depth and Sq scores of the naturally formed first and second soil layers.

The interpretation of VESS scores was conducted according to Ball, Batey and Munkholm (2007), which is based on requirements to change management practices to preserve soil structural quality: Sq=1 and Sq=2, good soil structural quality, requiring no changes in management practices; Sq=3, adequate soil structural quality; however there is need for improved soil management to avoid a further decline in soil quality. Therefore, Sq=3 is suggested as a critical limit to suitable crop production; Sq=4 and Sq=5, indicate poor soil structural quality, requiring urgent remedial management practices.

### 5.2.3 Relationship among VESS scores and quantitative soil physical properties

VESS is not a soil physical property but rather, it is a semi-quantitative measurement of soil structure. Thus, the VESS Sq score should be related to quantitative soil physical properties routinely used for studying soil structure in the laboratory and the field. At the same sampling time of the VESS assessments, soil resistance to penetration (SRP) measurements were taken and undisturbed soil samples (100 cm³) were collected. In the laboratory the following soil physical properties were determined: bulk density (BD) calculated by the ratio between soil dry mass and core volume; macroporosity (MaP) was computed as the difference between soil water content at saturation and soil water content at 6 kPa soil water potential; soil water storage capacity (SWSC) index, obtained as the ratio between water content at field capacity (-10 kPa soil water potential) and total porosity. These soil physical properties were used only for establishing functional relationships with VESS Sq

scores. A detailed description of the methods used in these measurements were provided by Cherubin et al. (2016)<sup>3</sup>.

### 5.2.4 Data analyses

The raw data were submitted to descriptive analysis and their normality was tested using a Shapiro-Wilk's test (p>0.05) through the Statistical Analysis System – SAS v.9.3 (SAS Inc., Cary, USA) software. Average values were compared using the confidence interval (p<0.05) according to Gabriel (1978). To test local effects of LUC, average VESS scores were compared within each site and to test overall LUC effects (regional scale) the average VESS scores of three sites were compared together. Linear regressions were performed using PROC REG procedure in SAS to verify the relationship among VESS scores and quantitative soil physical properties.

### **5.3 Results and Discussion**

### 5.3.1 VESS sensitivity to detect LUC effects on soil structural quality

The VESS method was capable of assessing soil structural quality changes under a wide textural range (15-66% clay content) in the Brazilian tropical soils studied. The VESS assessment took about 20 to 25 minutes from digging out the mini-trench to assigning the final score. It was more difficult to extract and breakdown samples from sugarcane soils indicating signs of soil compaction and damaged soil structure. It is also important to collect samples when soil moisture is close to field capacity, in order to minimize the physical effort associated with digging the mini-trench, extracting, and manipulating the samples as previously reported by Imhoff et al. (2009), Giarola et al. (2013) and Moncada et al. (2014). The step-by-step recommendations, pictures and criteria for Sq score differentiation available on the VESS chart properly enabled the identification of soil layers with different soil structural conditions. The shape of aggregate (approx. 1.5 cm diameter) was an important criterion for distinguishing between two scores when visual differentiation was not clear, especially for Sq scores between 3 and 4. We stress that user knowledge and experience are very important for assigning the Sq scores accurately. The visual evaluations were performed

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<sup>&</sup>lt;sup>3</sup>Data presented in the chapter 4.

by the same person in order to avoid potential Sq variability induced by different operators. Despite these limitations, several studies have shown that VESS scores have good reproducibility and that assessments can be accurately made by different, trained operators (BALL; BATEY; MUNKHOLM, 2007; CUI; ASKARI; HOLDEN, 2014).

The overall VESS scores ranged from 1.3 (native vegetation) and 4.0 (sugarcane), indicating a variation from good to poor soil structural quality among land uses (Table 1). Mean and median values had small differences and coefficient of variation values were below 25%. The normality of data was confirmed by Shapiro-Wilk's test (p>0.05).

Table 1 - Descriptive statistic of overall VESS scores for 0-25 cm layer in the native vegetation, pasture and sugarcane at three sites in central-southern Brazil

	Statistical parameters <sup>(1)</sup>										
Land use	Values				SD	Coefficients			W		
	Min	Mean	Median	Max	SD	CV	Cs	Ck	test <sup>(2)</sup>		
Lat_17S											
Native Vegetation	1.30	1.81	1.83	2.28	0.43	23.53	-0.19	-1.46	$0.99^{ns}$		
Pasture	1.61	2.00	2.01	2.36	0.31	15.35	-0.24	1.51	$0.95^{\text{ns}}$		
Sugarcane	1.80	2.49	2.65	2.85	0.47	18.84	-1.71	3.14	$0.82^{ns}$		
Lat_21S											
Native Vegetation	1.52	1.80	1.81	2.08	0.23	13.02	-0.06	0.05	$0.99^{ns}$		
Pasture	2.46	2.91	3.00	3.18	0.32	11.01	-1.33	1.50	$0.89^{ns}$		
Sugarcane	3.39	3.66	3.63	4.00	0.26	6.97	0.71	1.08	$0.97^{\text{ns}}$		
Lat_23S											
Native Vegetation	1.74	2.52	2.68	3.00	0.56	22.29	-1.26	1.13	$0.90^{\text{ns}}$		
Pasture	2.92	3.19	3.15	3.40	0.23	7.33	0.06	-4.54	$0.90^{\text{ns}}$		
Sugarcane	2.87	3.26	3.08	4.00	0.51	15.66	1.71	3.02	$0.82^{ns}$		

The standard deviation; CV (%): coefficient of variation; Cs: coefficient of skewness; Ck: coefficient of kurtosis; (2) W test: Shapiro-Wilk's test for normal distribution, where: (ns) non-significant by p < 0.05, indicating that the hypothesis of data are normally distributed was not rejected.

The VESS method was sensitive to local and regional soil structure changes induced by LUC for sugarcane production (Figures 1 and 2). Overall, conversion from native vegetation to pasture and then to sugarcane decreased soil structural quality for all soil layers (Figure 1). When the three sites were analyzed together (regional scale), overall VESS Sq scores significantly increased from Sq= 2.0 in native vegetation to Sq= 2.7 in pasture and then to Sq= 3.1 in sugarcane fields (Figure 2). We also identified a significant increase in scores from the top layer (0-10 cm depth) to the bottom layer (10-25 cm depth) for both local (Figure 1) and regional scale (Figure 2), indicating an increasing level of degradation in deeper layers. Guimarães et al. (2013) reasoned that assigning scores to individual layers provides

information that is more detailed and, therefore, allows more accurate management decision making than when only using a weighted average as described by Ball, Batey and Munkholm (2007). Using this approach, users can identify specific layers for sampling in case additional samples for quantitative analysis of soil quality indicators are desired (GUIMARÃES et al., 2013).

Considering the critical VESS score Sq= 3.0, as suggested by Ball, Batey and Munkholm (2007), it was verified that native vegetation soils, regardless of soil layer, have greater structural quality (Sq <3.0), supporting a suitable environment for root system growth and the exploitation of deeper soil layers. For pasture and sugarcane soils, poorer structural quality (Sq >3.0) was identified, mainly in the bottom layer (10-25 cm) at Lat\_21S and Lat\_23S, suggesting that management practice changes are needed to alleviate soil compaction. Our regional scale VESS assessment showed an overall score (Sq = 3.1) that was very close to the critical limit (Sq= 3.0) but it was even worse (i.e., Sq= 3.5) when the Lat\_17S site was not considered because it had been tilled for sugarcane replanting just before the time of sampling. These results confirm that current soil and crop management practices being used for sugarcane production are fostering a dangerous decline in soil structural quality and its consequent deleterious effects on sugarcane growth, development, and productivity. These results are supported by several other studies that show that current management practices can lead to soil compaction, which negatively affects sugarcane root system development and consequently, reduced yields (BRAUNACK; McGARRY, 2006; OTTO et al., 2011; BAQUERO et al., 2012; SOUZA et al., 2014).

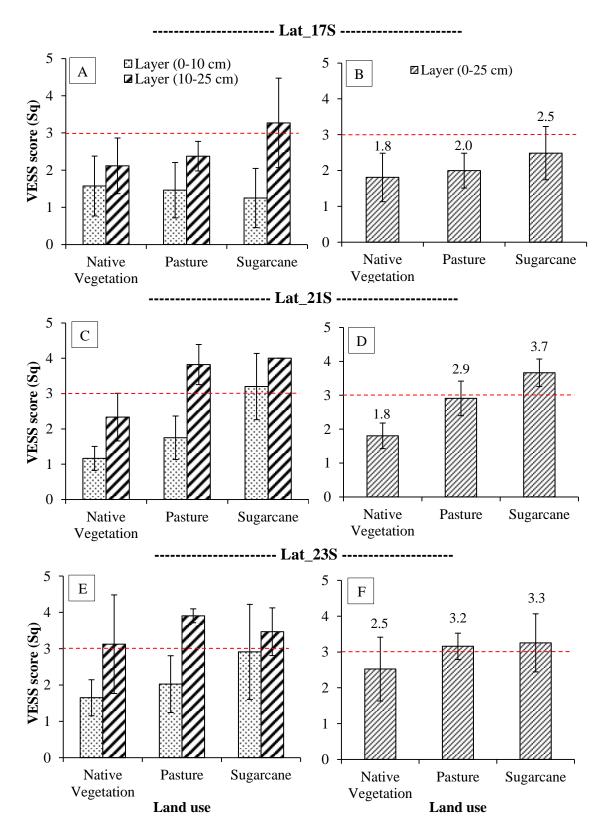
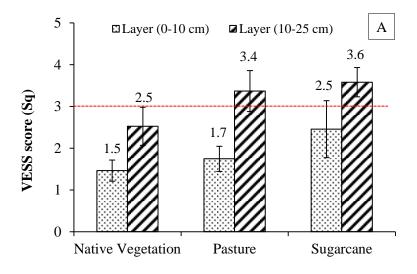


Figure 1 - VESS scores (Sq) for the top (0-10 cm) and bottom (10-25 cm) layers and overall Sq for total layer (0-25 cm) at Lat\_17S (A; B), Lat\_21S (C; D) and Lat\_23S (E; F) under land-use change sequence (native vegetation – pasture – sugarcane). Bars represent the confidence intervals (*p*<0.05). Red-dashed line indicated the critical VESS score (Sq= 3) to suitable root growth



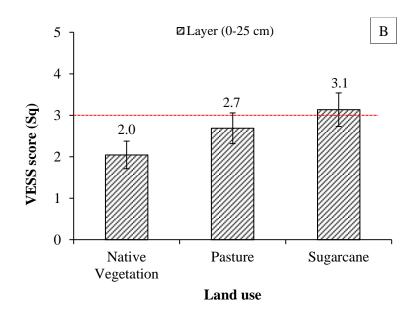


Figure 2 - VESS scores (Sq) for the top (0-10 cm) and bottom (10-25 cm) layers (A) and overall Sq for total layer (0-25 cm) (B) in regional scale under land-use change sequence (native vegetation – pasture – sugarcane). Bars represent the confidence intervals (*p*<0.05). Red-dashed line indicated the critical VESS score (Sq= 3) to suitable root growth

The depth of the top and bottom layers and their respective Sq scores are shown in Figure 3. The top layer depths were greater under native vegetation than in pasture or sugarcane and the VESS scores followed this order. Greater depth and higher Sq scores were observed in the bottom layer of sugarcane fields. A thicker bottom layer with lower soil structural quality suggests a strong limitation for sustainable yields using current sugarcane production practices. The differences in both depth and VESS scores induced by LUC can be clearly seen in Figure 4.

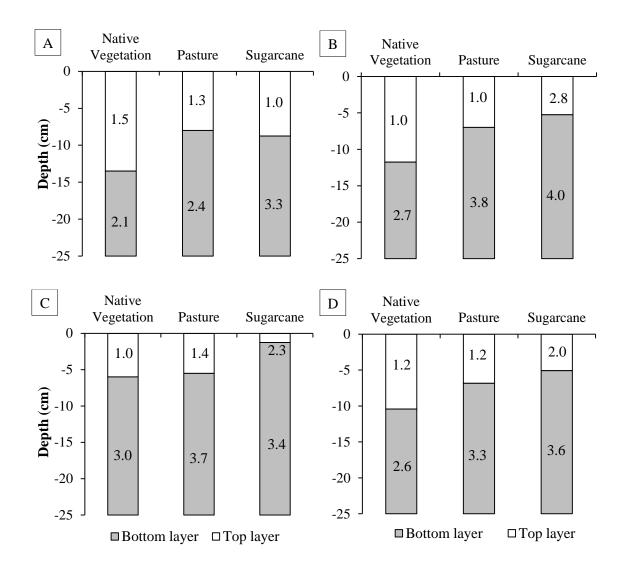


Figure 3 - Depth of distinct layers and VESS scores (inside of the bars) at Lat\_17S (A), Lat\_21S (B), Lat\_23S (C) and regional scale (D)

Overall, native vegetation soils had a top layer that was 35% or 50% deeper than in pasture or sugarcane soils, respectively (Figure 3D). The greater inputs of soil organic matter (SOM) and higher biological activity associated with absence of soil tillage and management are key factors for maintaining a deeper layer ( $\bar{x}$  10.4 cm) of better soil structural quality ( $\bar{x}$  Sq= 1.2) under native vegetation (Figure 4). Recently, Franco et al. (2015) verified that native vegetation had higher SOC stocks (0-30 cm layer) compared to pasture (average -26%) and sugarcane (average -36%) soils at these same sampling sites. In addition, field observations during VESS assessments indicated a high presence of earthworm burrows and evenness of root distribution in native vegetation soils. Organic carbon inputs associated with soil biota and root activity act as cementing agents throughout the soil aggregation process, gradually resulting in the formation of organic-mineral complexes (primary particles) that slowly form micro- and then macroaggregates (TISDALL; OADES, 1982). Soil aggregates physically

protect some SOM fractions, resulting in carbon pools with longer turnover times. This increase in C turnover time enables the organization of more complex and stable soil structure under native vegetation areas (SIX et al., 1998). Furthermore, the absence of tillage avoids disruption of aggregates and exposure of occlude SOM, that can ultimately lead to SOM decomposition.

# Land-Use Change Native Vegetation Pasture Sugarcane

Figure 4 - Illustration of the soil structural changes detected by VESS method due to effects of land-use change (e.g., native vegetation Sq=1.5; pasture Sq=2.5; sugarcane Sq=4.0) in central-southern Brazil

Long-term conversion from native vegetation to extensive pasture induced soil structure alterations in the sampled soil profile. We found a thinner layer ( $\bar{x}$  6.8 cm) of good soil structural quality ( $\bar{x}$  Sq= 1.2) only associated with the rhizosphere zone (Figure 4) overlying a compacted and deeper soil layer. The vigorous root systems of tropical grasses (e.g., the *Brachiaria* and *Cynodon* genera) can increase aggregate stability and improve soil structural quality (VEZZANI; MIELNICZUK, 2011; FONTE et al., 2012). Large root

systems promote high C inputs and act on the formation and stabilization of soil structure. Greater C stock in superficial soil layer was confirmed by Franco et al. (2015), who reported average SOC stock decreases of 6% from 0-10 to 10-20 cm layer and 18% from 0-10 to 20-30 cm layer in these pasture sites. Roots release a variety of exudates that have a cementing effect on soil particles and they can physically influence microaggregate formation via the compressing action of growing roots and in the entanglement of soil particles to form and stabilize macroaggregates (TISDALL; OADES, 1982; SIX et al., 2004; BRONICK; LAL, 2005). Roots also increase wet-dry cycling of adjacent soil, alter the ionic and osmotic balance in the rhizosphere through nutrient uptake and rhizodeposition and host a large population of micro- and macroorganisms that contribute to SOC and soil aggregation (TISDALL; OADES, 1982; SIX et al., 2004; BRONICK; LAL, 2005). In contrast, continuous cattle trampling and inadequate pasture management are the major drivers for soil compaction in pasturelands (NEWELL-PRICE et al., 2013; CUI; ASKARI; HOLDEN, 2014). Soil compaction limits growth and depth penetration of roots, decreasing deeper allocation of SOC and its potential improvement on soil structural quality. It results in a deeper bottom soil layer  $(\bar{x} 18.2 \text{ cm})$  of poor structural quality  $(\bar{x} \text{ Sq}=3.3)$ .

In sugarcane fields, a smaller soil surface layer depth ( $\bar{x}$  5.1 cm) with good structural quality ( $\bar{x}$  Sq= 2.0) and consequently larger bottom layer depth ( $\bar{x}$  19.9 cm) of poor soil structural quality ( $\bar{x}$  Sq= 3.6) can be associated with soil tillage operations performed during the establishment (land-use conversion) and replanting of sugarcane, SOC losses and intensive machinery traffic under favorable conditions for soil compaction. A thinner soil layer with good soil structural quality (lower Sq score) implies a reduced soil volume exploited by the sugarcane root system (OTTO et al., 2011; SOUZA et al., 2014). Under weather condition in central-southern Brazil, especially during dry periods, more fragile and superficial root systems induce chemical and physical stress to the plants that can lead to a decline in yield and the need for premature replanting operations. The soil tillage operations provide an intensive soil disturbance, inducing SOC losses to the atmosphere (SILVA-OLAYA et al., 2013; MELLO et al., 2014). In addition, sugarcane reformation is very expensive and requires appropriate weather to be carried out in an agronomically successful manner. Therefore, soil structural degradation decreases both environmental and economic sustainability of sugarcane production. The thicker surface layer of good soil structure observed in sugarcane fields (Figures 3 and 4) may be related to the positive effects of maintaining sugarcane straw on the soil surface. These results are supported by Franco et al. (2015), who reported reduction of SOC in deeper layers in these same sugarcane sites.

Dalchiavon et al. (2013) also verified that maintenance of sugarcane straw on the soil surface increases SOC stocks, decreases bulk density and soil resistance to penetration, and improves sugarcane yield.

Despite the differences between soils and length of sugarcane cultivation, a comparison of sugarcane fields at the different study sites (Figure 5) indicated that VESS Sq scores increased from sugarcane replanting (Lat\_17A) through initial growth (Lat\_23S) to full growth (Lat\_21S). Tillage operations conducted during planting or replanting (plowing and disking) promote soil disturbance and consequently alleviate soil compaction. Therefore, just after those operations VESS scores are lower, indicating better soil structural quality and adequate soil physical conditions for sugarcane growth. However, our data suggest that tillage performed for sugarcane replanting had a short-term effect on soil structure. Soil tillage operations promote disruption of macroaggregates favoring SOC losses (SIX et al., 1998; 2004) and inducing an increase in soil compaction over time (CASTRO et al., 2013). Its deleterious effects are further magnified by successive machinery operations used during the sugarcane production season (LOZANO et al., 2013; SOUZA et al., 2014).

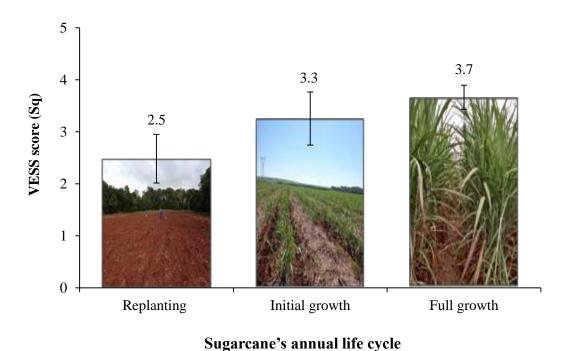


Figure 5 - Evolution of VESS scores during the sugarcane's annual life cycle, where, Lat\_17S: sugarcane replanting; Lat\_23S: initial growth; Lat\_21S: full growth. Bars represent the confidence intervals (p<0.05)

We emphasize that these results that show VESS score changes during the sugarcane cycle should be interpreted carefully, as our dataset was collected from different sites under distinct soil and weather conditions. Additional studies for evaluating impacts of sugarcane management on soil structure assessed by VESS should be carried out through the whole sugarcane cycle (about five years). Furthermore, subsoil compaction in sugarcane fields should also be evaluated using methodology such as SubVESS, proposed by Ball et al. (2015). We consider subsoil compaction an important concern since sugarcane roots have the potential to explore soil layers much deeper than 25 cm. Both VESS and SubVESS could be implemented at the same time for a complete evaluation of structural quality throughout the entire soil profile.

The VESS scores (Sq >3.0) found in this study suggest that important management changes are required to mitigate the negative effects of sugarcane cultivation on soil structure and improve its sustainability. Adoption of minimum tillage, harvesting without prior burning and maintaining straw on the soil surface could increase SOC and improve soil structure, making the soil more resistant to compressive machinery effects. Protocols that aim to have machinery enter fields under soil moisture that are less favorable to compaction in conjunction with controlled traffic strategies should also be encouraged to keep soil structure favorable for sustainable sugarcane production.

### 5.3.2 VESS score as an integrative soil structural quality indicator

Quantitative soil physical properties traditionally used to assess soil structure changes in sugarcane fields have been used for supporting management decisions. Since the VESS method integrates several soil physical properties into a single score, it is convenient that VESS scores are correlated with these quantitative parameters. Figure 6 shows that VESS Sq scores have a significant relationship with bulk density ( $r^2 = 0.33$  to 0.56), macroporosity ( $r^2 = 0.23$  to 0.44), index of soil water storage capacity ( $r^2 = 0.21$  to 0.50), and soil resistance to penetration ( $r^2 = 0.32$  to 0.52), regardless of the inherent properties of the soil (e.g., soil texture, moisture and SOC).

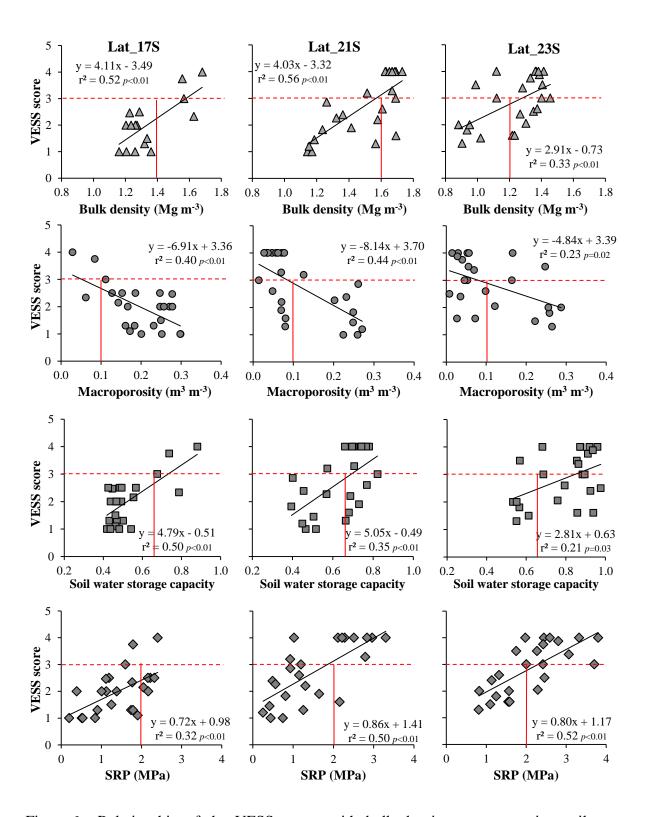


Figure 6 - Relationship of the VESS scores with bulk density, macroporosity, soil water storage capacity and soil resistance to penetration (SRP) at Lat\_17S (left), Lat\_21S (center) and Lat\_23S (right). Red-dashed lines indicate critical VESS score (Sq = 3) to suitable root growth; and red-solid lines indicate the critical limit to root growth [bulk density = 1.2 (clay soils), 1.4 (medium texture soils) and 1.6 Mg m $^{-3}$  (sandy soils); and SRP = 2 MPa], air diffusion (macroporosity = 0.10 m $^{3}$  m $^{-3}$ ), and balance between water and air (soil water storage capacity index = 0.66)

VESS scores were significantly associated with increases in bulk density, which lead to decreases in macroporosity and increases in water retention (especially in lower water potentials), unbalancing the relationship between air and water in the soil, and finally, increasing the soil impedance to root growth. Overall, the critical VESS score (Sq= 3.0) used to guide management changes was satisfactorily convergent with critical values to: bulk density, 1.2, 1.4 to 1.6 Mg m<sup>-3</sup> for clay, medium texture and sandy soils, respectively (REYNOLDS et al., 2002); macroporosity, 0.1 m<sup>3</sup> m<sup>-3</sup> (XU; NIEBER; GUPTA, 1992); soil water storage capacity, 0.66 (REYNOLDS et al., 2002) and soil resistance to penetration, 2 MPa (OTTO et al., 2011). VESS critical score (Sq= 3.0) proposed by Ball, Batey and Mulkholm (2007) for temperate soils seems appropriate as a guide for management decision in tropical soils.

These results suggest that VESS is an accurate and reliable semi-quantitative method that integrates physical functions (e.g., water availability, aeration and root growth) related to structural and physical quality of soils. Thus, VESS could be used as an alternative or complementary tool for assessing sugarcane expansion impacts on soil structural quality in Brazilian tropical soils. These results are consistent with Guimarães et al. (2013), who showed that VESS and the Least Limiting Water Range (a complex indicator) have converged to identify soil physical conditions highly restrictive to plant growth when Sq >3.5. In addition, quantitative soil physical properties have site-specific responses (Figure 6), which are highly influenced by inherent soil characteristics, and therefore, become one of the drawbacks of using one of these properties alone as an indicator of soil for structure or soil quality (NEWELL-PRICE et al., 2013).

In addition to the ability of VESS to detect soil structure/physical changes, as reported in this study, Mueller et al. (2013) reported that visual methods for soil assessment are useful diagnostic tools for monitoring and controlling overall soil quality over different scales, ranging from within-field to global. Recent studies confirmed that VESS can be used to validate quantitative soil quality indexes that encompassed soil chemical, physical and biological properties (ASKARI; HOLDEN, 2014; 2015). Therefore, the VESS method should be suggested to sugarcane producers as a practical, easily-performed and accurate tool for monitoring soil quality degradation. This also helps to reduce cost and time when evaluating soil quality over large sugarcane areas, typical in central-southern Brazil. A future challenge is the automated collection of samples to quantify VESS. This is needed to broaden its use, especially in extensive areas cultivated with sugarcane in Brazil. Finally, development of protocols and training of technicians and consultants on the proper application of VESS are

essential steps for its effective use in the assessment and monitoring of soil quality within sugarcane production systems.

### **5.4 Conclusions**

The VESS method was efficiently sensitive for detecting soil structural quality changes, demonstrating its potential for direct on-farm assessment. VESS scores were significantly correlated with quantitative soil physical properties, and the VESS critical score (Sq= 3.0) is a reliable guide for management decisions in Brazilian tropical soil under sugarcane cultivation. Land-use conversions from native vegetation to pasture and then to sugarcane led to degradation of soil structural quality. Therefore, sugarcane expansion fields must be monitored to prevent soil physical limitations that can negatively affect growth and yield of sugarcane. We conclude that VESS scores provide an efficient method for identifying soil structural quality degradation induced by LUC, and recommend that VESS evaluations be incorporated into monitoring protocols for evaluating soil quality/health in areas of sugarcane expansion in Brazil.

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## 6 A SOIL MANAGEMENT ASSESSMENT FRAMEWORK (SMAF) EVALUATION OF BRAZILIAN SUGARCANE EXPANSION ON SOIL QUALITY

### **Abstract**

The Soil Management Assessment Framework (SMAF) was developed to evaluate impacts of land use and management practices on soil quality (SQ), but its suitability for Brazilian tropical soils was unknown. We hypothesized that SMAF would be sensitive enough to detect SQ changes associated to sugarcane (Saccharum officinarum) expansion for ethanol production. Field studies were carried out at three sites across the central-southern region of Brazil, aiming to quantify impacts of a land-use change sequence (i.e., native vegetation – pasture – sugarcane) on SQ. Eight soil indicators were individually scored using SMAF curves developed primarily for North American soils and integrated into an overall Soil Quality Index (SQI) and in its chemical, physical and biological sectors. The SMAF scores were correlated with two other approaches used to assess SQ changes, soil organic carbon (SOC) stocks and Visual Evaluation of Soil Structure (VESS) scores. Our findings showed that the SMAF was an efficient tool for assessing land-use change effects on SQ of Brazilian tropical soils. The SMAF scoring curves developed using robust algorithms allowed proper assignment of scores for the soil chemical, physical and biological indicators assessed. The SQI scores were significantly correlated with SOC stocks and VESS scores. Long-term transition from native vegetation to extensive pasture promoted significant decreases in soil chemical, physical and biological indicators. Overall SQI suggested that native vegetation soils were functioning at 87% their potential capacity, while pasture soils were functioning at 70%. Conversions of pasture to sugarcane induced slight improvements on SQ, primarily because of improved soil fertility. Sugarcane soils are functioning at 74% of their potential capacity. Based on this study, management strategies are suggested to improve SQ and the sustainability of sugarcane production in Brazil.

Keywords: Land-use change; Ethanol production; Soil quality assessment; Soil quality indicators

### 6.1 Introduction

Soil quality/health (SQ) is a key factor required to achieve sustainable agricultural systems that will meet our increasing demands for food, feed, fiber and fuels. Therefore, in recent decades SQ has been discussed worldwide and become a major agenda item for the scientific community (KARLEN et al., 2008, KARLEN; PETERSON; WESTFALL, 2014). Soil quality was defined as the capacity of a specific kind of soil to function, within natural or managed ecosystem boundaries, to sustain plant and animal productivity, maintain or enhance water and air quality, and support human health and habitation (KARLEN et al., 1997). It is a product of inherent (e.g., parental material, climate, topography) and anthropogenic (e.g.,

tillage and cropping systems, land uses) interactions (KARLEN et al., 1997). Soil inherent attributes are governed by soil-forming processes, and are often relatively unresponsive to soil and crop management practices. On the other hand, dynamic soil properties (e.g., soil organic carbon, pH, soil aggregation, microbial biomass activity) are responsive to management practices and/or land use, but their change rates are dependent of the inherent soil attributes (KARLEN et al., 1997; 2008).

Land-use change processes have transformed a large proportion of the planet's land surface, affecting directly the capacity of soils to function (FOLEY et al., 2005). Increasing global demand for bioenergy feedstock production has intensified the land-use changes in many countries (FISCHER et al., 2010; WRIGHT; WIMBERLY, 2013; MUKHERJEE; SOVACOOL, 2014; GASPARATOS et al., 2015), and especially in Brazil (LAPOLA et al., 2010; GODEMBERG et al., 2014; BORDONAL et al., 2015). Brazil is the world's largest sugarcane producer (655 million tons) with about 40% of the global harvest (FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS, 2015). The sugarcane cropped area expanded from 5.8 to 9.0 Mha between 2005 and 2015 (COMPANHIA NACIONAL DE ABASTECIMENTO, 2015) and is projected to increase by 6.4 Mha to meet Brazilian domestic demand for ethanol by 2021 (GOLDEMBERG et al., 2014). Recent expansion has been concentrated in central-southern Brazil and 70% of the land-use change has occurred through conversion of extensive pasturelands (ADAMI et al., 2012). Sugarcane expansion initiatives have resulted in degraded pastures being subjected to intensive mechanization and inputs of agrochemicals (i.e., lime, fertilizer and pesticides) that have direct implications on SQ. Therefore, monitoring soil properties (indicators) altered by landuse change is crucial for identifying strategies that minimize SQ degradation and its negative implications on ecosystem functioning (FU et al., 2015; ZORNOZA et al., 2015).

To implement the concepts of SQ and its assessment, the Soil Management Assessment Framework (SMAF) was initially developed by researchers in the USA on North American soils (ANDREWS; KARLEN; CAMBARDELLA, 2004). The SMAF is a quantitative SQ evaluation method that emphasizes a dynamic view of SQ and involves detecting soil response to current or recent management decisions (ANDREWS; KARLEN; CAMBARDELLA, 2004; KARLEN et al., 2014). The SMAF uses a 3-step process to assess soil quality, including (i) indicator selection (chemical, physical and biological); (ii) indicator interpretation (non-linear scoring curves); and (iii) integration into an overall SQ index (SQI). Assessment values are generally expressed as a fraction or percentage of full performance for soil functions such as crop productivity, nutrient cycling, or environmental protection

(ANDREWS; KARLEN; CAMBARDELLA, 2004; KARLEN et al., 2013). Currently, the SMAF has scoring curves or interpretation algorithms for 13 indicators, which encompass physical properties: bulk density (BD), macroaggregate stability (AGS), plant-available water and water-filled pore space (WFPS); chemical properties: potential of hydrogen (pH), electrical conductivity, sodium adsorption ratio, extractable phosphorus (P) and potassium (K); and biological properties: soil organic carbon (SOC), microbial biomass carbon (MBC), potentially mineralizable N, and β-glucosidase (BG) activity (ANDREWS; KARLEN; CAMBARDELLA, 2004; WIENHOLD et al., 2009; STOTT et al., 2010). These scoring curves were developed and validated using datasets primarily from North America (USA, Canada, and Mexico), with the exception of WFPS (which included data from China), and BG (which included data from Brazil, Argentina and Italy), considering site-specific controlling factors (i.e., climate and/or inherent soil properties) that affect the score of each indicator (ANDREWS; KARLEN; CAMBARDELLA, 2004; WIENHOLD et al., 2009; STOTT et al., 2010).

The SMAF has been broadly used in the USA for assessing several situations and factors that affect both agricultural and natural systems at scales ranging from within-experimental field (plots) to regional (e.g., ANDREWS; KARLEN; CAMBARDELLA, 2004; WIENHOLD et al., 2006; ZOBECK et al., 2014; STOTT et al., 2013; KARLEN et al., 2014; VEUM et al., 2015) evaluations. In addition, SMAF has been tested in other countries around world [e.g., South Africa (SWANEPOEL et al. 2015), Ethiopia (ERKOSSA; ITANNA; STAHR, 2007; GELAW; SINGH; LAL, 2015) and Nepal (KALU et al., 2015)]. Data from Brazilian soils was limited in the development and validation of the SMAF, and to our knowledge, no other studies using SMAF as tool for assessing the impacts of current management practices and land uses on SQ in Brazil have been published. The SMAF could be an important, user-friendly tool for helping farmers, consultants, researchers and government officials make immediate and strategic decisions for improving SQ/health and agricultural sustainability.

Therefore, we conducted an on-farm study across the largest sugarcane-producing regions of Brazil to assess effects of the primary land-use change sequence associated with sugarcane expansion (i.e., native vegetation to pasture to sugarcane) on SQ for a wide range of soil textures using SMAF. We hypothesized that: (i) long-term conversion from native vegetation to extensive pasture led to significant SQ degradation; (ii) under current practices sugarcane production soils are recovering SQ attributes lost when used as pasturelands; and

(iii) SQ changes in Brazilian tropical soils under different land use and management systems could be detected by SMAF.

### **6.2 Material and Methods**

### 6.2.1 Study sites and experimental design

The study sites and experimental design were described in the 2.2.1 and 2.2.2 items.

### 6.2.2 Soil sampling and laboratory analyses

Soil samples within each land use (i.e., native vegetation, pasture and sugarcane) were collected using a consistent grid pattern composed of nine points spaced 50 m apart, providing a total of 27 sampling points (3 land uses x 9 points) for each site or 81 sampling points for the three studied sites. At each sampling point, a small trench (30 x 30 x 30 cm) was opened to collect undisturbed soil samples from the 0-10, 10-20 and 20-30 cm depths using metallic cylinders with volume of about 100 cm<sup>3</sup>. This provided a total of 243 undisturbed soil samples for soil physical indicators quantification. Around of each central trench, composite samples consisting of 12 subsamples were collected using a Dutch auger, at same three depths. This provided an additional of 243 disturbed soil samples for chemical and biological analyses.

Several soil indicators were analyzed. Chemical indicators included available P and K as well as active acidity (pH<sub>CaCl2</sub> 0.01M L<sup>-1</sup>) which were measured using analytical methods described in Raij et al. (2001). Physical indicators included bulk density (BD), calculated by dividing the soil dry mass by volume of the cylinder (100 cm<sup>3</sup>), and wet macroaggregate stability (AGS), determined using a vertical oscillator (Yoder, model MA-148) with three sieve-sizes (2000, 250, and 53  $\mu$ m) and a speed of 30 oscillations per minute for 10 minutes. The AGS (% macroaggregation) was calculated by summing aggregate mass for >2000 and >250  $\mu$ m classes, dividing by the total soil mass, and multiplying by 100. Particle-size was determined using a hydrometer method (GEE; OR, 2002). Biological indicators included: a) soil organic carbon (SOC), measured by dry combustion on a LECO<sup>®</sup> CN-2000 elemental analyzer (furnace at 1350 °C in pure oxygen); b) microbial biomass C (MBC), measured on three replicates of field-moist samples after fumigating for 24 h and extracting with 0.5M K<sub>2</sub>SO<sub>4</sub> (VANCE; BROOKES; JENKINSON, 1987). Organic C in the fumigated and non-

fumigated extracts was measured using a TOC-Vcs/cp analyzer attached in a Shimadzu<sup>®</sup> SSM-5000A (Shimadzu, Kyoto - Japan) before calculating biomass C with a correction factor of k = 0.33; and c) β-Glucosidase activity (BG) was measured using air-dried soil as described by Tabatabai (1994). The concentration of *p*-nitrophenol was determined in triplicate by measuring absorbance at 400 nm in a spectrophotometer, and results were expressed in mg of *p*-nitrophenol released kg<sup>-1</sup> soil h<sup>-1</sup>. Both MBC and BG activity were analyzed only for 0-10 cm soil layer.

Data of SOC stocks and Visual Evaluation of Soil Structure (VESS) scores were used to verify their relationship with SMAF scores. Those measurements were made on soil samples collected at the same sites and sampling times and previously reported by Franco et al. (2015) and Cherubin et al. (2015b), respectively. Briefly, SOC stocks were calculated for each soil layer by multiplying the SOC content of each one by the soil bulk density and the layer thickness (10 cm). To account for the effect of differing soil bulk densities (due to landuse change) on stock comparisons, the stocks within the pasture and sugarcane soils were adjusted to an equivalent soil mass based on measurements for native vegetation (LEE et al., 2009). Subsequently, individual SOC stocks for the 0-10, 10-20 and 20-30 cm layers were summed to provide a total SOC stock for 0-30 cm layer.

Visual Evaluation of Soil Structure (VESS) is a semi-quantitative approach developed by Ball, Batey and Munkholm (2007) and improved by Guimarães, Ball and Tormena (2011) for on-farm assessment of the soil physical and structural capacity to support plant growth. Briefly, a VESS assessment consists of digging out a small trench using a spade and collecting a block soil (20 x 10 x 25 cm - approx. 5000 cm<sup>-3</sup>). The VESS evaluation includes manual breakdown of soil aggregates along their weakness lines, identification of layers having contrasting structure, measurement of layer depth and assignment of a score by comparing the structure of the sample with the aggregated characteristics proposed by Guimarães, Ball and Tormena (2011). The latter, developed as a VESS key chart, contains descriptions, pictures, and a score for each soil structure quality rating. The criteria taken account to assign the score are related to shape, size, strength and visible porosity of aggregates, as well as biological activity and presence of root inter- or intra-aggregates. The soil structural quality scores range from 1 (good) to 5 (poor), with 3 being considered a critical limit for suitable plant growth (BALL; BATEY; MUNKHOLM, 2007).

### 6.2.3 Soil Management Assessment Framework

The SMAF was used as tool to evaluate the land-use change effects on SQ. The minimum dataset included eight soil indicators (pH, P, K, BD, AGS, SOC, MBC and BG) for 0-10 cm layer and six soil indicators (pH, P, K, BD, AGS and SOC) for 10-20 and 20-30 cm layers. The importance of each one of these indicators to soil functionality is consistently reported in the literature (e.g., ANDREWS; KARLEN; CAMBARDELLA, 2004; STOTT et al., 2010; CARDOSO et al., 2013; ZORNOZA et al., 2015). The pH, available P and K provide information about soil acidity and nutrient availability status. Macroaggregation stability and BD indicate soil structural and physical conditions, which affect soil aeration, water infiltration and storage and soil's ability to resist erosion process. Soil organic carbon, MBC and BG were chosen as biological indicators. The SOC plays crucial role in multiple soil processes including nutrient cycling and storage, food source for edaphic organisms and soil aggregation; while MBC and BG indicate the microbiological and biochemical activity of the soils.

This approach is consistent with the general SMAF guidelines, which recommend using a minimum of five indicators with at least one each representing soil chemical, physical and biological properties and processes (KARLEN et al., 2008). These indicators were scored by transforming mean measured values into 0 - 1 values using previously published algorithms (ANDREWS; KARLEN; CAMBARDELLA, 2004; WIENHOLD et al., 2009; STOTT et al., 2010), which were used to compute an overall SQI for each land use and studied site. Those algorithms account for organic matter, texture, climate, slope, region, mineralogy, weathering class, crop, sampling time, and analytical method effects on the various threshold values. For this study, the organic matter factor class (based on soil classification and used for scoring AGS, SOC, MBC and BG) was 4 (low OM) for all sites. The texture factor class (used for scoring BD, AGS, SOC, MBC and BG) was 2 (clay content ~17%) at Lat 21S and pasture in Lat 17S and 4 (clay contents >40%) at Lat 17S (except pasture) and Lat\_23S. The climate factor (used for scoring SOC, MBC and BG) was 1 (≥170° days and ≥550 mm of mean annual precipitation) for all sites. The seasonal factor, impacting MBC scores, was 2 (sampling on summer - January) for all sites. The Fe oxide, used for AGS scores, was 1 (Ultisol) for Lat\_21S and 2 (other soils) for other sites. The mineralogy factor class, used for scoring BD, were 3 (clay 1:1 and Fe and Al oxides) and the slope and weathering class factors, used for scoring P, were 2 (2-5% slope) and 2 (high weathering), respectively, for all sites. The method used to measure extractable P was resin (class 5). We

changed resin method factor from 3.1 to 1.25, to avoid overestimating the P scores under low P conditions in weathered soils. New crop factors, which affecting P and pH scores, needed to be added to SMAF spreadsheet for encompassing Brazilian natural vegetation (Cerrado and Atlantic forest), tropical grass (*Brachiaria* spp. and *Cynodon* spp.) and sugarcane. Phosphorus and pH thresholds for each "new crop" were set up using literature and expert's opinions. Optimum P and pH values were: 6 mg dm<sup>-3</sup> and 4.5 for Cerrado vegetation; 12 mg dm<sup>-3</sup> and 5.5 for Atlantic forest; 13 mg dm<sup>-3</sup> and 5.5 for pasture; and 16 mg dm<sup>-3</sup> and 6.0 for sugarcane (RAIJ et al., 1997). The SMAF algorithms are based on pH<sub>water</sub>, therefore, pH<sub>CaCl2</sub> was converted to pH<sub>water</sub> by the regression fitted in Ciprandi (1993), pH<sub>water</sub> = 0.890 + 0.922 pH<sub>CaCl2</sub> ( $r^2 = 0.97$ , p < 0.05). The SMAF scoring curve for K (WIENHOLD et al., 2009) is consistent with K recommendation classes adopted in Brazil (RAIJ et al., 1997).

In addition to individual indicator scores, an overall SQI was calculated by summing the scores and dividing by the number of indicators for each soil layer. The overall SQI was also subdivided into chemical (pH, P and K), physical (BD and AGS) and biological (SOC, MBC, and BG) sectors, as well as its relative contribution into overall SQI. This approach identifies the management areas of greatest concern (i.e., lowest index scores) so that land managers can be given better guidance on how to most efficiently restore or improve SQ at that specific location (STOTT et al., 2013, KARLEN et al., 2014).

### 6.2.4 Data analyses

An analysis of variance (ANOVA) was computed using PROC GLM procedure to test the influence of the land-use change within each site on individual soil indicators, SMAF scores and overall SQI values. If the ANOVA F statistic was significant (p<0.05), the means were compared using Tukey's test (p<0.05). The analyses were performed separately by depth. An additional ANOVA was computed to test the land-use change effects in regional scale (all sites simultaneously) on overall SQI and SQI-sectors scores for 0-30 cm layer. Means were also compared using Tukey's test (p<0.05). Finally, regression analyses were performed using PROC REG procedure between SMAF scores and SOC stocks within each site for 0-30 cm depth, and between SMAF scores and VESS scores for sites with contrasting texture (Lat\_21S: sandy soils; Lat\_23S: clay soils). All statistical procedures were completed using the software Statistical Analysis System – SAS v.9.3 (SAS Inc, Cary, USA).

### **6.3 Results and Discussion**

### 6.3.1 Soil chemical indicators

Soil chemical conditions were typical for tropical regions (Table 1). Soils of Cerrado biome in central-southern Brazil are highly weathering and characterized by high acidity and low nutrient availability, as shown by Lopes and Cox (1977). The transition from native vegetation to extensive pasture led to soil acidification and decreased nutrient levels, especially available P (Table 1). Soil acidification and nutrient depletions were the result of long-term (>30 years) soil use with continuous grazing and the absence of lime and fertilizer inputs as indicated by Cherubin et al. (2015a). Higher K levels under pasture at Lat\_21S and Lat\_23S could be attributed to several factors such as greater K cycling, lower K losses (KAYSER; ISSELSTEIN, 2005), and release of non-exchangeable K forms by aggressive root system of grasses (ROSOLEN; VICENTINI; STEINER, 2012).

The algorithms used in the SMAF were sensitive to detect the score changes for the chemical indicators under tropical conditions in Brazil (Table 2). As expected, we needed to add new "crop factors" into SMAF spreadsheet labeled as Brazilian Cerrado vegetation, Atlantic Forest vegetation, Brazilian tropical grasses (*Brachiaria* spp. and *Cynodon* spp.) and sugarcane. The SMAF scoring curves for pH and P have a parabolic shape denoting an optimum range, which takes into account crop-specific critical limits to sustain plant growth without causing environmental deleterious impacts (e.g., fresh water contamination). In general, the results showed that conversion of native vegetation to pasture decreased pH scores (average from 0.92 to 0.69) and P scores (average from 0.90 to 0.62), mainly at Lat\_17S and Lat\_21S sites (Table 2). The SMAF scoring curves for K also have a parabolic shape; however it was set up using a general response of crops to soil K levels according to Wienhold et al. (2009). Therefore, the K scores were lower than pH and P scores, especially at Lat\_17S (more weathered soil) with averages of 0.38 and 0.19 under native vegetation and pasture, respectively. For Lat\_21S and Lat\_23S, K scores increased from native vegetation (average from 0.67 and 0.76) to pasture (average from 0.76 and 0.96) (Table 2).

Table 1 - Mean values of the soil quality indicators from 0-10, 10-20 and 20-30 cm soil layers under native vegetation (NV), pasture (PA) and sugarcane (SC) in central-southern Brazil

	soutner	ı bıazıı		1	Moon india	otor volue	<u> </u>			
	Land		Mean indicator values  Chemical Physical Biological							
Site	use	рН	Chemical P K		BD AGS		SOC	Biological SOC MBC BG		
	usc	unit	mg dm <sup>-3</sup>	mg dm <sup>-3</sup>	Mg m <sup>-3</sup>	%	g kg <sup>-1</sup>	mg kg <sup>-1</sup>	μg g h <sup>-1</sup>	
-		unit	mg um	mg um	0-10 cm		g Kg	mg kg	<u> ид д п</u>	
Lat_17S	NV	$4.4 b^{\dagger}$	5.6 a	39.8 a	1.23 b	90.9 a	15.6 a	397.2 <sup>ns</sup>	49.7 <sup>ns</sup>	
	PA	4.3 b	3.0 b	23.8 b	1.63 a	92.8 a	9.5 b	301.5	40.0	
	SC	5.6 a	7.3 a	23.8 b	1.26 b	73.3 b	10.8 b	414.8	47.2	
	~ 0	2.0	10-20 cm							
	NV	4.4 b	4.5 b	30.5 a	1.28 b	88.7 b	12.9 a	_	_	
	PA	4.4 b	2.6 c	19.2 b	1.61 a	93.3 a	8.4 b	-	-	
	SC	5.6 a	7.0 a	22.3 b	1.54 a	78.5 c	10.4 b	-	-	
			20-30 cm							
	NV	4.5 b	3.5 b	27.0 a	1.28 b	88.7 b	11.2 a	-	-	
	PA	4.4 b	2.5 c	18.8 b	1.63 a	93.6 a	6.4 b	-	-	
	SC	5.4 a	4.7 a	18.0 b	1.51 a	84.4 b	9.7 a	-	-	
					0-10 cm					
	NV	6.8 a	17.3 a	108.7 b	1.20 b	92.1 a	21.8 a	870.4 a	122.6 c	
	PA	4.5 c	7.1 b	163.0 a	1.53 a	86.2 a	13.3 b	438.5 b	273.2 a	
	SC	5.8 b	19.6 a	120.8 ab	1.62 a	60.5 b	11.1 b	539.9 b	200.6 b	
					10-20 cm					
Lat_21S	NV	6.7 a	12.5 a	113.0 a	1.32 b	$72.8^{\text{ ns}}$	16.0 a	-	-	
	PA	4.5 c	3.9 b	133.3 a	1.65 a	86.2	9.5 b	-	-	
	SC	5.6 b	13.2 a	113.8 a	1.68 a	69.9	9.9 b	-	-	
			20-30 cm							
	NV	6.7 a	9.9 a	93.5 b	1.38 b	71.5 <sup>ns</sup>	13.1 a	-	-	
	PA	4.5 c	3.2 b	120.4 a	1.65 a	81.7	7.5 b	-	-	
	SC	5.2 b	7.8 a	102.8 ab	1.68 a	73.4	8.0 b	-	-	
					0-10 cm					
	NV	4.3 c	14.3 a	109.1 b	0.89 b	93.8 a	36.7 a	1978.7 a	337.8 a	
	PA	5.2 b	11.5 ab	170.1 a	1.30 a	95.8 a	36.4 a	2085.9 a	115.7 b	
	SC	5.9 a	8.8 b	120.4 b	1.33 a	84.3 b	18.9 b	928.6 b	53.8 c	
	NIT /	4.2	10-20 cm							
Lat_23S	NV	4.3 c		95.4 b	1.03 b	93.9 a	33.7 a	-	-	
	PA	5.1 b	9.8 ab	174.4 a	1.41 a	97.2 a	27.6 a	-	-	
	SC	5.8 a	8.6 b	90.7 b	1.44 a	83.3 b	18.4 b	-	-	
	NIV/	12 ~	11 0 6	20-30 cm						
	NV DA	4.3 c	11.0 a	90.3 b	1.06 b	92.0 ab	30.3 a	-	-	
	PA SC	5.0 b	7.6 b	158.4 a	1.39 a	96.8 a	20.6 b	-	-	
	SC	5.9 a	7.2 b	77.4 b	1.44 a	88.0 b	17.3 b	-	-	

§pH: potential of hydrogen, P: phosphorus, K: potassium, BD: bulk density, AGS: macroaggregate stability, SOC: soil organic carbon, MBC: microbial biomass carbon, BG: β-Glucosidase activity; †Mean values ("n" = 9) in column within each site and depth, followed by the same letter do not differ among themselves according to Tukey's test (p<0.05); <sup>ns</sup>: non-significant

Table 2 - Scores of the soil quality indicators from 0-10, 10-20 and 20-30 cm soil layers under native vegetation (NV), pasture (PA) and sugarcane (SC) in central-southern Brazil

	Brazii			т	ndicator CN	AAE soore	<b>N</b> C		
Site	Land		Chemica		ndicator SN	sical		Biologica	.1
Site	use	рН	P	K	BD	AGS	SOC	MBC	BG
		pii			0-10 cm	MOD	500	MDC	
	NV	0.99 a†	0.85 a	0.44 a	0.73 a	1.00 <sup>ns</sup>	0.96 a	1.00 <sup>ns</sup>	0.19 ns
	PA	0.50 c	0.39 b	0.22 c	0.38 b	1.00	0.78 b	0.95	0.15
	SC	0.89 b	0.78 a	0.29 b	0.71 a	1.00	0.74 b	1.00	0.19
	БС	0.07 0	0.70 <b>u</b>	0.27 0	10-20 cm	1.00	0.710	0.17	
I -4 170	NV	0.99 a	0.77 a	0.36 a	0.61 a	$1.00^{ns}$	0.88 a	-	-
Lat_17S	PA	0.52 c	0.29 b	0.18 c	0.41 b	1.00	0.65 b	-	-
	SC	0.89 b	0.78 a	0.28 b	0.32 b	1.00	0.69 b	-	-
					20-30 cm				
	NV	1.00 a	0.60 a	0.33 a	0.63 a	$1.00^{ns}$	0.77 a	-	-
	PA	0.54 c	0.26 b	0.18 c	0.40 b	1.00	0.40 c	-	-
	SC	0.79 b	0.54 a	0.23 b	0.32 b	1.00	0.63 b	-	-
					0-10 cm				
	NV	0.79 b	1.00 a	0.68 b	0.98 a	$1.00^{\rm ns}$	1.00 a	$1.00^{ns}$	0.91 b
	PA	0.63 c	0.88 b	0.81 a	0.56 b	1.00	0.96 a	1.00	1.00 a
	SC	0.91 a	0.99 a	0.71 ab	0.39 c	1.00	0.88 b	1.00	1.00 a
					10-20 cm				
Lat_21S	NV	0.83 a	0.98 a	0.70 a	0.94 a	$0.99^{\mathrm{ns}}$	0.98 a	-	-
Lat_213	PA	0.60 b	0.56 b	0.75 a	0.35 b	1.00	0.78 b	-	-
	SC	0.89 a	0.96 a	0.69 a	0.33 b	0.99	0.81 b	-	-
					20-30 cm				
	NV	0.82 a	0.96 a	0.62 b	0.85 a	$1.00^{ns}$	0.95 a	-	-
	PA	0.63 b	0.43 c	0.72 a	0.36 b	1.00	0.54 b	-	-
	SC	0.71 ab	0.80 b	0.65 ab	0.33 b	0.99	0.61 b	-	-
					0-10 cm				
	NV	$0.96^{\text{ns}}$	0.99 a	0.79 b	0.99 a	$1.00^{\rm ns}$	$1.00^{\rm ns}$	$1.00^{\rm ns}$	1.00 a
	PA	0.96	0.95 ab	0.96 a	0.61 b	1.00	1.00	1.00	0.84 b
	SC	0.89	0.86 b	0.84 b	0.58 b	1.00	0.98	1.00	0.23 c
					10-20 cm				
Lat_23S	NV	0.97 a	0.99 a	0.76 b	0.96 a	$1.00^{\rm ns}$	1.00 a	-	-
Lat_233	PA	0.91 ab	0.95 a	0.97 a	0.46 b	1.00	1.00 a	-	-
	SC	0.87 b	0.84 b	0.73 b	0.39 b	1.00	0.98 b	-	-
					20-30 cm				
	NV	$0.97^{\mathrm{ns}}$	0.98 a	0.74 b	0.97 a	$1.00^{ns}$	1.00 a	-	-
	PA	0.89	0.87 ab	0.94 a	0.45 b	1.00	0.99 a	-	-
	SC	0.89	0.76 b	0.67 b	0.33 b	1.00	0.98 b	-	-

§pH: potential of hydrogen, P: phosphorus, K: potassium, BD: bulk density, AGS: macroaggregate stability, SOC: soil organic carbon, MBC: microbial biomass carbon, BG: β-Glucosidase activity; †Mean values ("n" = 9) in column within each site and depth, followed by the same letter do not differ among themselves according to Tukey's test (p<0.05); <sup>ns</sup>: non-significant

The land-use change from pasture to sugarcane promoted overall improvements in soil chemical indicators. Sugarcane management including lime applications resulted in higher pH

values in all sites, with average increasing from 4.6 (pasture) to 5.6 (sugarcane) (Table 1) and average pH scores from 0.69 (pasture) to 0.86 (sugarcane) (Table 2). Applications of mineral fertilizer and complementary organic residues in sugarcane fields increased P levels and scores (from 0.47 to 0.81) and increased or maintained K levels (average scores 0.48 for pasture and 0.47 for sugarcane) at Lat\_17S and Lat\_21S. Although both P and K levels had improvements with sugarcane cultivation, they were still below of the critical limits, P >16 mg dm<sup>-3</sup> and K >120 mg dm<sup>-3</sup>, established by Raij et al. (1997). In contrast, lower P and K levels and scores were found in sugarcane field compared to pasture at Lat\_23S (Table 1), likely associated with the management of fertilization using insufficient amount of organic residues as verified by Cherubin et al. (2015a) and due to significant SOM depletions (FRANCO et al., 2015).

All measurements for pH, P and K were concentrated at the increasing part of SMAF parabolic curves, confirming that acidity and low plant available P and K levels are the limiting factors for sugarcane production under Brazilian weathered soils.

# 6.3.2 Soil physical indicators

The land-use change from native vegetation to pasture induced soil compaction by increasing BD values (Table 1). Many studies have shown that cattle trampling is the major driver for soil compaction under pasture (e.g., GREENWOOD; McKENZIE, 2001; PIETOLA; HORN; YLI-HALLA, 2005). In addition, low pasture productivity (shoot and roots) has been verified under compacted soils, reducing C inputs into the soil (MAIA et al., 2009; FRANCO et al., 2015), contributing to an increase in the soil structural degradation. The SMAF scoring curves for BD (less-is-better sigmoidal shapes), which take into account texture and mineralogical classes (ANDREWS; KARLEN; CAMBARDELLA, 2004), were able to identify alterations on BD due to land-use change effects (Table 2). The BD scores decreased from native vegetation (average 0.85) to pasture (average 0.44).

Regarding macroaggregation stability (AGS), higher values were found under native vegetation and pasture, ranging from >70% in sandy soils (Lat\_21S) and >90% in clay soils (Lat\_23S). High AGS values are typically reported in studies under weathered Brazilian soils (e.g., MADARI et al., 2005; BARTHÈS et al., 2008), being associated primarily with a clay mineral composition dominated by Fe and Al oxides and 1:1 mineral layering in these soils (SIX; ELLIOTT; PAUSTIAN, 2000). In addition, Franco (2015) verified that soil macrofauna abundance plays important role in the soil aggregation processes in tropical soils; therefore,

greater AGS under native vegetation and pasture are consistent with higher abundance of soil engineering invertebrates (i.e., earthworms and termites) in these areas (FRANCO, 2015). The SMAF scoring curves for AGS (more-is-better sigmoidal shapes) takes into account differences in SOM, texture and Fe oxides content (ANDREWS; KARLEN; CAMBARDELLA, 2004). However, for all possible variations of these factors, the maximum score (1.0) is assigned when the AGS values are higher than 50% (threshold value from which soil structural stability is optimum for environment protection and productivity goals). Therefore, using the current SMAF scoring curves, AGS scores was a non-sensitive indicator to detect land-use change impacts in tropical soils, reaching practically score 1.0 for all sites (Table 2). Macroaggregation stability has been globally used as SQ indicator (CARDOSO et al., 2013; KARLEN et al., 2013, 2014; STOTT et al., 2013; ZORNOZA et al., 2015) due to its crucial role on C stabilization and protection, mediating soil physical processes related to water and air dynamic and providing resistance against soil erosion. Therefore, additional SMAF scoring curves for AGS need to be developed for applying and detecting smaller changes caused by recent land use and management under well-aggregated tropical soils.

Conversion from pasture to sugarcane have been done through intensive mechanization raising the concern on soil compaction. Although BD had no significant differences between sugarcane and pasture, the values found in sugarcane [>1.2 Mg dm<sup>-3</sup> for clay soil and >1.6 Mg dm<sup>-3</sup> for sandy soils (Table 1)] are considered critical for sustaining adequate plant growth as shown by Reynolds et al. (2002). Using SMAF, the average BD score was 0.41 (Table 2), confirming that soil compaction is one of major driver to SQ degradation under sugarcane fields. Tillage operations carried out during sugarcane replanting (~ every 5 years) alleviated soil compaction (i.e., decreased BD), but this positive effect was limited to the surface layer (10 cm depth) in the sugarcane field at Lat\_17S and probably has short-term persistence as verified in other Brazilian soils by Silva et al. (2012). In addition, soil tillage promoted breakup of the macroaggregates and, SOC and macrofauna losses, decreasing AGS values under sugarcane production (Table 1). As discussed previously, even though AGS depletions were statistically significant, AGS scores were close to 1, generally equal those found under native vegetation and pasture (Table 2).

# 6.3.3 Soil biological indicators

Greater SOC contents were found under native vegetation, ranging from 11.2 to 36.7 g kg<sup>-1</sup> (Table 1), depending of soil taxonomic class, texture and climate. These factors are taken

into account in the SMAF scoring curves (more-is-better sigmoidal shapes) for SOC thus accounting for inherent soil characteristics that can affect the score (ANDREWS; KARLEN; CAMBARDELLA, 2004). The land-use change from native vegetation to pasture decreased SOC content (Table 1) and average scores from 0.95 to 0.79 (Table 2). These SOC losses in tropical regions are well documented in the literature (MAIA et al., 2009; MELLO et al., 2014; FRANCO et al., 2015) as result of conversion process and low C inputs due to low grass productivity and inadequate grazing management. The MBC values were high in all sites, especially in more clay soil (Lat\_23S). Conversions from native vegetation to pasture trend to decrease MBC at Lat\_17S and Lat\_21S (Table 1), similar to that observed for SOC, confirming the close relationship between MBC and SOC ( $r = 0.88 \ p < 0.01$ ). Regardless of site and effects of land-use change, the SMAF scores for MBC ranged from 0.95 to 1.0, without differences among land uses (Table 2). These results are consistent with study of Lopes et al. (2013), who defined that MBC values > 375 mg kg<sup>-1</sup> are classified as high under clayey Oxisols in Brazilian Cerrado.

The BG activity responses to land-use change were statistically different within each site. At Lat\_23S, BG values significantly decreased from native vegetation to pasture. In contrast, significantly higher BG was found under pasture compared to native vegetation at Lat\_21S, probably associated with higher pH under native vegetation soil (Table 1). The SMAF scores for BG decreased from native vegetation (1.0) to pasture (0.84) at Lat\_23S, there was a slight increase from native vegetation (0.91) to pasture (1.0) at Lat\_21S and there were not significant differences at Lat\_17S, in which the lowest scores were observed (Table 2). The SMAF scoring curves for BG were sensitive to alterations induced by land-use change. The inclusion of a dataset from Brazilian Cerrado soils for the development and validation of the SMAF-BG algorithms (STOTT et al., 2010) likely contributed to the good performance for the soils of this study. In addition, previously, Lopes et al. (2013) had verified that critical limits for BG activity defined as function of crop yield and SOC in clayey Brazilian Oxisols were consistent with SMAF-BG scores (i.e., values in the low and high interpretative classes were equivalent to SMAF-BG scores of 0.85 and 0.32, respectively).

Short-term transitions from pasture to sugarcane (<5 years) did not promote significant SOC changes at Lat\_17S and Lat\_21S (Table 1). However, after more than 20 years of sugarcane including approximately 10 years of burning pre-harvest, significant SOC depletion and reduced MBC and BG activity at Lat\_23S (Table 1) was observed. For that site, SOC scores showed a slight decrease from pasture (1.0) to sugarcane (0.98), MBC scores showed no differences and BG scores had marked depletion under sugarcane field (0.23).

These results are consistent with large studies recently carried out in central-southern Brazil by Mello et al. (2013) and Bordonal et al. (2015).

# 6.3.4 Overall Soil Quality Index and scores

Overall SQI and SQI sectors (i.e., chemical, physical and biological) for each depth and site are shown in Figure 1 and in regional scale (Figure 2) for the 0-30 cm. The SQI computed for each depths (0-10, 10-20, and 20-30 cm) indicated that SQ decreased in depth, regardless of the land use and site. Several factors contribute for improving SQ in the first centimeters, such as inputs of C from litter and crop residues on the soil surface, greater biological and biochemical activity, higher nutrient cycling and fertilizer inputs, better soil structure and physical resistance as well as better soil resilience to stress due to animal trampling and machinery traffic. We highlighted that SMAF scores calculated for deeper layers (<15 cm depth) must be carefully interpreted, since SMAF scoring algorithms were originally developed for near surface soils.

The highest SQI were verified in the native vegetation soils ranging from 0.72 to 0.77 at Lat 17S, from 0.87 to 0.92 at Lat 21S and from 0.94 to 0.97 at Lat 23S. In regional scale, average SQI suggests these soils are functioning at 87% of their potential capacity for the 0-30 cm layer (Figure 2). The SQI-sectors were also higher under native vegetation for all studied sites and depths (Figures 1 and 2). These results demonstrate that natural ecosystems are in dynamic balance, where chemical, physical and biological attributes act cooperatively in such way that soils perform their functions properly. Conversion from native vegetation to pasture promoted significant SQ degradation, with SQI values ranging from 0.51 to 0.77, 0.61 to 0.85 and 0.86 to 0.92 at Lat\_17S, Lat\_21S and Lat\_23S, respectively. In regional scale, average SQI suggests these soils are functioning at 70% of their potential capacity for the 0-30 cm layer (Figure 2). Long-term land use with extensive pasture led to chemical impoverishment of the soil, increasing soil compaction and its deleterious impacts on soil physical processes and negative impacts on biological indicators driven by SOC depletions, as evidenced by SQI-sectors scores in Figure 2B. Recent estimates suggest that 70% of Brazilian pasturelands are degraded or in the process of being degraded (DIAS-FILHO, 2014), and SQ degradation caused by inadequate management of pasture and animals is considered the major driver of this process. We believe that investigations using robust frameworks, such as SMAF, for assessing SQ under extensive pasture in Brazil could help farmers make the best decision about more sustainable uses for their lands and guide government's strategic planning for agriculture expansion and/or funding the adoption of strategies for recovery degraded pasturelands (e.g., Low-Carbon Agriculture program).

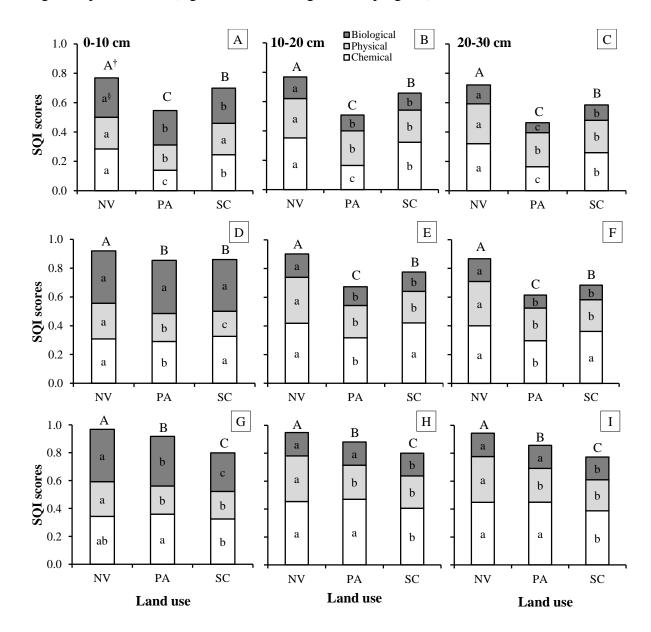


Figure 1 - Overall soil quality index (SQI) scores and the contribution of chemical, physical and biological attributes into the overall SQI under native vegetation (NV), pasture (PA) and sugarcane (SC) for the 0-10 (A;D;G - left), 10-20 (B;E;H - central) and 20-30 cm (C;F;I - right) layers at Lat\_17S (A;B;C), Lat\_21S (D;E;F) and Lat\_23S (G;H;I) in central-southern Brazil. †Mean SQI scores within each site in same depth followed by the same capital letter do not differ among themselves according to Tukey's test (*p*<0.05); §Mean sectors contribution within each site in same depth followed by the same lower letter do not differ among themselves according to Tukey's test (*p*<0.05)

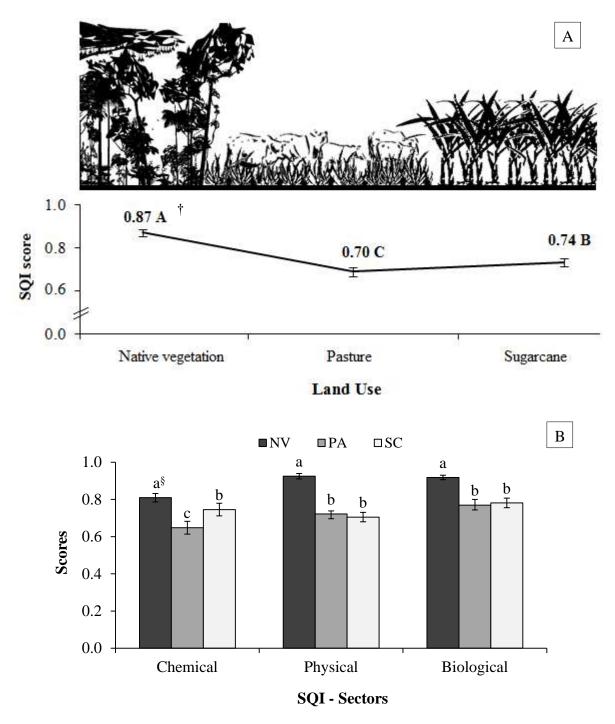


Figure 2 - Overall soil quality index (SQI) scores (A) and SQI sector (chemical, physical and biological) scores (B), 0-30 cm layer, for regional scale of land-use change [i.e., native vegetation (NV) – pasture (PA) – sugarcane (SC)] for sugarcane expansion in Brazil. Error bars denote standard deviation of the mean; †Mean SQI scores followed by the same capital letter do not differ among themselves according to Tukey's test (*p*<0.05); §Mean scores within each SQI sectors (chemical, physical and biological) followed by the same lower letter do not differ among themselves according to Tukey's test (*p*<0.05)

The sugarcane expansion under pasturelands improved SQ at Lat\_17S and Lat\_21S. For these sites, average SQI (0-30 cm) showed that sugarcane soils are functioning at 65 and

77% their potential capacity (Figure 1). At Lat\_23S, although SQI decreased under sugarcane, likely due to previous management involving burning pre-harvest and significant SOC losses (FRANCO et al., 2015) and current fertilization practices, the soil is functioning at 79% of its potential capacity (Figure 1). In regional scale (Figure 2) the SQI indicated that sugarcane expansion under extensive pasture led to slight, but significant improvement on SQ. Therefore, sugarcane soils are functioning at 74% their potential capacity within 0-30 cm layer. This SQ improvement was driven by inputs of lime and fertilizer, which significantly increased chemical SQI-sector (Figure 2B). These findings demonstrate how important the proper management of fertilization is in the agricultural systems for sustaining SQ in tropical regions. Physical and biological SQI-sectors had no differences between sugarcane and pasture soils, which had average decreases of 22 and 15% their physical and biological functioning capacity compared with native vegetation soils.

Our SQ assessment, based on SMAF scores, suggests that sugarcane cultivation has improved soil quality compared to extensive pasturelands. Therefore, sugarcane expansion reintegrates degraded pasturelands into a productive system, providing more economical and social benefits with positive environmental offsets [i.e., improving soil quality, saving greenhouse gas emissions (MELLO et al., 2014; BORDONAL et al., 2015) and alleviating deforestation of natural ecosystems (MELLO et al., 2014; GOLDEMBERG et al., 2014)]. However, to avoid future SQ decline in sugarcane fields we recommend the adoption of organic residues as complementary fertilization, minimum or no-tillage system associated to crop rotation, controlled machinery traffic), which ensure proper soil fertility to achieve nutritional demands of sugarcane crop, improve soil C sequestration and mitigate deleterious impacts from tillage and machine traffic on soil physical properties and processes.

# 6.3.4 Overall Soil Quality Index *versus* SOC stocks and VESS scores

Globally, SOC is the most common single indicator used for assessing impacts of the land-use changes and agricultural management practices on SQ and its multiple ecosystems services (CARDOSO et al., 2013; ZORNOZA et al., 2015). In Brazil, several studies have assessed the sustainability of biofuel crops expansion through SOC stock changes (FRAZÃO et al., 2014; MELLO et al., 2014; FRANCO et al., 2015). In the United States, Soil Conditioning Index was adopted by the USDA-NRCS to investigate effects of agricultural practices on SOC, and infer changes in SQ (USDA-NRCS, 2003). Zobeck et al. (2008, 2014)

compared agricultural management effects using Soil Conditioning Index and SMAF-SQI. The authors concluded that both methodologies were able to identify SQ changes; however, since SMAF includes several chemical, physical and biological indicators, it provides more detailed information about soil quality than Soil Conditioning Index.

Linear regressions between SOC stocks and SQI scores obtained using SMAF are shown in Figure 3. Soil organic carbon stocks explained between 53 to 78% of the variation in overall SQI. These finding support two important statements: (i) changes in SOC stocks results in modifications in physical, chemical and biological attributes of SQ, which are encompassed in the SMAF-SQI scores, supporting SOC as a universal indicator of SQ. However, when multiple indicators are used together the SQ assessment becomes more accurate and enables to identify which critical conditions need priority management (e.g., soil fertility, soil compaction, biological activity, etc); and (ii) these strong positive correlations validated SQI scores, since SOC stock is broadly recognized as a suitable endpoint for environmental protection and crop productivity management goals.

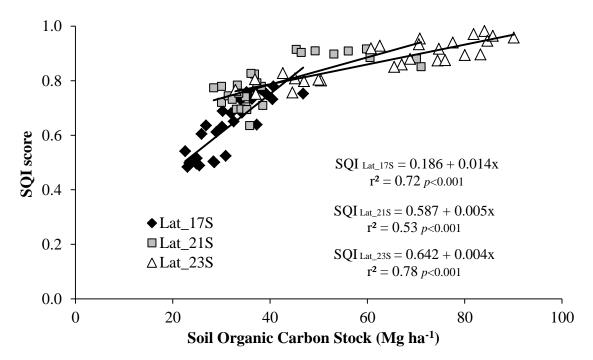


Figure 3 - Relationship between soil organic carbon stocks and overall soil quality index (SQI) scores, 0-30 cm layer, for the land-use change [i.e., native vegetation (NV) – pasture (PA) – sugarcane (SC)] at three sites (Lat\_17S, Lat\_21S and Lat\_23S) in central-southern Brazil

We also verified significant relationship between SQL<sub>physical</sub> sector and overall SQI scores with VESS scores (Figure 4). Our results showed that the variation in SQL<sub>physical</sub> sector

and overall SQI can be explained by VESS at 56 and 51% under sandy soils and at 32 and 25% under clay soils, respectively. Using the equations described in Figure 4 and the critical value of VESS= 3, it was verified that SQI<sub>physical</sub> and overall SQI reached values that corresponding to 76 and 82% of physical functioning and, 80 and 89% of overall functioning respectively for sandy and clay soils. We assume that a sharper decline in SQI<sub>physical</sub> and SQI scores must be observed when VESS scores are greater than 4, which were not found in the studied sites.

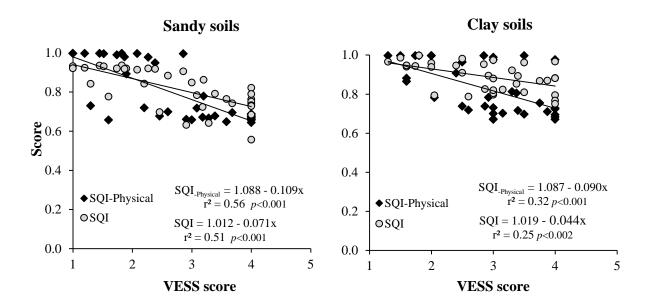


Figure 4 - Relationship among Visual Evaluation of Soil Structure (VESS) scores, overall soil quality index (SQI) and SQI-Physical sector scores under native vegetation, pasture and sugarcane areas in central-southern Brazil

These results suggest that VESS measures more than soil structural quality, with advantages of to be an on-farm method, simple to perform and easy to understand (GUIMARÃES; BALL; TORMENA, 2011; BALL; MUNKHOLM; BATEY, 2013; MULLER et al., 2013). Therefore, the VESS method could be used as a complementary tool for monitoring SQ in areas undergoing land-use change for sugarcane expansion in Brazil. In addition, we suggest that VESS could be further included into the SMAF or used to replace other soil physical properties. Thereby, studies in a wide range of soils and agricultural managements systems are necessary to developed reliable SMAF scoring curves for VESS.

#### **6.4 Conclusions**

This study was the first application of SMAF for assessing SQ changes in Brazil and confirmed our hypothesis that SMAF would be sensitive enough to detect SQ changes associated with sugarcane expansion. In general, the SMAF scoring curves developed primarily on North American soils properly assigned scores for soil chemical, physical and biological indicators included in this study. The SMAF indicator scores were useful for evaluating which sectors require priority management, while overall SQI score integrated all sectored information into a single value, enabling to detect global SQ changes induced by land-use change impacts. Overall SQI calculated by SMAF was positively correlated with SOC stocks ( $r^2 = 0.53$  to 0.78), which is recognized for its multiples ecosystems functions. In addition, SQI was negatively correlated with VESS scores ( $r^2 = 0.25$  to 0.51), a simpler semi-quantitative method that has showed potential for on-farm monitoring of SQ changes. Therefore, the SMAF was a reliable and efficient tool to detect the land-use change effects on SQ under Brazilian tropical conditions. However, futures studies are encouraged to adjust and validate SMAF algorithms using dataset from tropical soils and expanding its use around the world.

Our findings suggest that native vegetation land use had the greatest SQ, with soils functioning on average at 87% of their potential capacity. Replacing native vegetation by pasture decreased SQ to 70% of its potential capacity. Land-use change from pasture to sugarcane induced slight improvements on SQ, mainly driven by increasing on soil chemical quality. Overall, sugarcane soils are functioning at 74% of their potential capacity. Based in this study, management strategies that sustain proper soil fertility for sugarcane growth, increase soil C sequestration and alleviate soil compaction and erosion are recommended to improve SQ and the sustainability of sugarcane production in Brazil.

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# 7 SOIL QUALITY INDEXING STRATEGIES FOR EVALUATING SUGARCANE EXPANSION IN BRAZIL

#### **Abstract**

Increasing demand for biofuel has intensified land-use change (LUC) for sugarcane (Saccharum officinarum) expansion in Brazil. Assessments of soil quality (SQ) response to this LUC are essential for quantifying and monitoring sustainability of sugarcane production over time. Since there is not a universal methodology for assessing SQ, we conducted a fieldstudy at three sites within the largest sugarcane-producing region of Brazil to develop a SQ index (SQI). The most common LUC scenario (i.e., native vegetation to pasture to sugarcane) was evaluated using six SQI strategies with varying complexities. Thirty eight soil indicators were included in the total dataset. Two minimum datasets were selected: one using principal component analysis (7 indicators) and the other based on expert opinion (5 indicators). Nonlinear scoring curves were used to interpret the indicator values. Simple and weighted additive methods were used to combine individual indicator scores into an overall SQI. Long-term conversion from native vegetation to extensive pasture significantly decreased overall SQ. In contrast, conversion from pasture to sugarcane had no significant impact on overall SQ at the regional scale, but site-specific responses were found. In general, sugarcane production improved chemical attributes; however it has negative effects on physical and biological attributes. Overall, we found that simple, user-friendly strategies were as effective as more complex ones for identifying SQ changes. Therefore, as a protocol for SQ assessments in Brazilian sugarcane areas, we recommend using a small number of indicators [e.g., pH, P, K, Visual Evaluation of Soil Structure (VESS) scores and SOC concentration] and proportional weighting to reflect chemical, physical and biological processes within the soil. Our SQ evaluations also suggest that current approaches for expanding Brazilian sugarcane production by converting degraded pasture land to cropland can be a sustainable strategy for meeting increasing biofuel demand. However, management practices that alleviate negative impacts on soil physical and biological indicators must be prioritized within sugarcane producing areas to prevent unintentional SQ degradation over time.

Keywords: Land-use change; Ethanol production; Minimum dataset; Soil quality indexes

# 7.1 Introduction

Increasing global demand for biofuel has accelerated land-use change (LUC) to support bioenergy crops in many countries. In Brazil, the area devoted to sugarcane production increased from 5.8 to 9.0 Mha during the last decade (COMPANHIA NACIONAL DE ABASTECIMENTO, 2016). Even though Brazil is already the world's largest sugarcane producer, current predictions indicate that an additional 6.4 Mha of sugarcane will be needed to meet the domestic demand for ethanol by 2021 (GODEMBERG et al., 2014). Sugarcane expansion has primarily occurred on land previously occupied by extensive pastures (LAPOLA et al., 2010; GODEMBERG et al., 2014), most of which are degraded or in the process of being degraded (DIAS-FILHO, 2014; STRASSBURG et al., 2014). To obtain

long-term energy security, bioenergy systems will need to be agronomically and environmentally sustainable. Intensification of land use through mechanization and agrochemical inputs has direct implications on soil physical, chemical and biological properties and consequently on the quality/health of soils. To prevent unintended consequences, monitoring of soil property changes due to LUC is essential (FU et al., 2015; ZORNOZA et al., 2015). However, this research topic is still new in Brazil, and we are not aware of any protocol for evaluating soil quality (SQ) changes induced by sugarcane expansion in this region.

Soil quality was defined as the capacity of a specific kind of soil to function, within natural or managed ecosystem boundaries, to sustain plant and animal productivity, maintain or enhance water and air quality, and support human health and habitation (KARLEN et al., 1997). It is a complex functional concept and cannot be measured directly in the field or laboratory; but can be indirectly inferred by soil indicators (KARLEN et al., 1997; MUKHERJEE; LAL, 2014). Indicators of SQ are those measurable soil properties and processes that have greatest sensitivity to changes in soil function and its ecosystem services (ANDREWS; KARLEN; CAMBARDELLA, 2004; ZORNOZA et al., 2015). A wide range of soil chemical, physical and biological properties could be measured (CARDOSO et al., 2013; ZORNOZA et al., 2015), but due to cost it's not feasible to consider them all, and therefore it is necessary to select a minimum dataset (MDS). Several strategies have been used to define an appropriate MDS including principal component analysis (PCA) (LIMA; HOOGMOED; BRUSSAARD, 2008; ARMENISE et al., 2013; CHEN et al., 2013; ASKARI; HOLDEN, 2014; 2015; MUKHERJEE; LAL, 2014; SÁNCHEZ-NAVARRO et al., 2015), fuzzy sets (QI et al., 2009; XIA et al., 2015), expert opinion (ANDREWS; KARLEN; MITCHELL, 2002; ANDREWS; KARLEN; CAMBARDELLA, 2004) and farmer/local knowledge (LIMA; HOOGMOED; BRUSSAARD, 2011; TESFAHUNEGN; TAMENE; VLEK, 2011). According Doran and Parkin (1994) suitable SQ indicators should correlate well with ecosystem processes, integrate soil properties and processes, be accessible to many users, sensitive to management and climate, and, whenever possible, be components of existing databases. An example for reducing the number of potential SQ indicators was provided by Andrews, Karlen and Cambardella (2004) through their development of the Soil Management Assessment Framework (SMAF). Starting with an extensive list of 80 or more integrative measurements related to ecosystem processes and functions that reflect SQ, they developed scoring curves only for a small number (i.e., 10) of carefully selected indicators that could reliably detect SQ changes induced by agricultural management practices. In more recent studies, others have shown that small datasets can effectively characterize SQ within different ecosystems. Lima et al. (2013) compared SQ assessment using a total dataset (TDS) of 29 indicators, a MDS of eight indicators based on PCA, and an indigenous set of four indicators based on farmer knowledge to evaluate rice (*Oryza sativa* L.) production systems in southern Brazil. They concluded that the TDS provided the best assessment of SQ, but the smaller datasets showed the same SQ trends and thus provided meaningful information for land managers. Askari and Holden (2014; 2015) reduced the number of indicators using PCA from 21 to 3 and from 22 to 7, respectively, and verified that the MDS indicators were suitable to efficiently quantify SQ in grassland and arable fields in Ireland.

After defining a MDS, linear and non-linear techniques, each with their advantages and disadvantages, have been applied to interpret SQ indicators (ANDREWS et al., 2002; MASTO et al., 2008; ASKARI and HOLDEN, 2014, 2015). While linear methods are simple, user-friendly and require little knowledge of the indicator thresholds, non-linear methods can more often assign meaningful scores that better represent the soil functions being represented by the indicators (ANDREWS; KARLEN; MITCHELL, 2002)

Once individual indicators have been scored, it is often convenient, but not essential, to integrate them into an overall SQ index (SQI) that can be used to support decision making and selection of sustainable management practices (de PAUL OBADE; LAL, 2016). Currently, there is no comprehensive, universal SQI that can be used across multiple natural and anthropogenic ecosystems. Many indexing strategies have been developed and tested for specific purposes under particular environmental conditions around the world [e.g., in the 2002; U.S.A. (ANDREWS; KARLEN; MITCHELL, ANDREWS; KARLEN; CAMBARDELLA, 2004; MUKHERJEE; LAL, 2014; de PAUL OBADE; LAL, 2016), Brazil (LIMA et al., 2013), Argentina (ROMANIUK et al., 2011), Italy (ARMENISE et al., 2013), Spain (SÁNCHEZ-NAVARRO et al., 2015), Ireland (ASKARI; HOLDEN, 2014; 2015), South Africa (SWANEPOEL et al., 2015), India (MASTO et al., 2008) and China (QI et al., 2009; CHEN et al., 2013; LIU et al., 2015)]. The most user-friendly method to calculate a SQI is to simply add all indicator scores and then divide by the number of indicators (ANDREWS; KARLEN; MITCHELL, 2002; ANDREWS; KARLEN; CAMBARDELLA, 2004; KARLEN et al., 2013; MUKHERJEE; LAL, 2014). The major concern regarding this method is that when the number of indicators is unbalanced among chemical, physical and biological sectors, the overall SQI misrepresents the sector(s) having fewer indicators. On the other hand, several studies have used methods that assign weights for each indicator. Different criteria that have been used include soil function frameworks (KARLEN; STOTT, 1994; LIMA et al., 2013; MUKHERJEE; LAL, 2014), principal components loading (ANDREWS; KARLEN; MITCHELL, 2002; MASTO et al., 2008; MUKHERJEE; LAL, 2014; LIU et al., 2015; SÁNCHEZ-NAVARRO et al., 2015), partial least squares regression coefficients (de PAUL OBADE; LAL, 2016) and correlation with crop yield (NAKAJIMA; LAL; JIANG, 2015). Simple and weighted additive SQ indexing strategies provide site-specific responses (ANDREWS; KARLEN; MITCHELL, 2002; ASKARI; HOLDEN, 2014; 2015; MUKHERJEE; LAL, 2014), influenced by existing dataset, soil type, and effects of land use and management practices.

Developing more user-friendly and cost-effective strategies for assessing SQ changes induced by agricultural management practices, especially those associated to bioenergy feedstock production therefore remains a challenge for the scientific community (ANDREWS; KARLEN; MITCHELL, 2002; MUKHERJEE; LAL, 2014; de PAUL OBADE; LAL, 2016). Our goal was to develop a sensitive and reliable protocol for evaluating SQ impact associated with LUC occurring to increase Brazilian sugarcane production. To do so, we conducted a field-study at three sites where the primary LUC sequence (i.e., native vegetation to pasture to sugarcane) is occurring within of the largest sugarcane-producing region of Brazil. Six SQ indexing strategies with varying complexity were developed and tested. Our hypotheses were that: (i) the LUC sequence would result in SQ degradation; (ii) the SQI approach would be suitable to detect SQ changes due to LUC; and (iii) the simple, more user-friendly strategies would be able to detect SQ changes as effectively as more complex strategies.

#### 7.2 Material and Methods

#### 7.2.1 Study sites and experimental design

The study sites and experimental design were described in the 2.2.1 and 2.2.2 items.

# 7.2.2 Soil sampling and analyses

All soil samples were collected using a consistent experimental design that had four points spaced 50 m apart imposed within each land use. This provided 12 sampling points for each location or 36 sampling points for the three locations. A small trench (30 x 30 x 30 cm) was opened at each sampling point to collect both undisturbed and semi-undisturbed samples

from the 0 to 10-, 10 to 20- and 20 to 30-cm layers. This provided 108 samples for physical analyses, and 108 for soil aggregation and macrofauna analyses. An additional 108 disturbed samples were collected for chemical and biological analyses by compositing 12 subsamples taken from each soil layer with a Dutch auger.

Available phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), sulfur (S – sulphate), boron (B), cooper (Cu), manganese (Mn), iron (Fe), zinc (Zn), active acidity (pH<sub>CaCl2 0.01mol L</sub><sup>-1</sup>), potential acidity (H+Al), base saturation (BS) and cation exchange capacity (CEC<sub>pH7</sub>) were determined using analytical methods described by Raij et al. (2001). Soil resistance to penetration (SRP) and field-saturated hydraulic conductivity (Kfs) were measured at five and three locations, respectively, within ~5 m of each trench using a digital penetrometer (PenetroLOG®) and the 'simplified falling-head' method proposed by Bagarello, Iovino and Elrick (2004). Soil structural quality of the 20 x 10 x 25 cm monoliths from each trench was assessed using the Visual Evaluation of Soil Structure (VESS) method (BALL; BATEY; MUNKHOLM, 2007; GUIMARÃES; BALL; TORMENA, 2011). Particlesize was determined using the hydrometer method. Bulk density (BD) was determined based on the core method with 100 cm<sup>-3</sup>cylinder. Soil degree of compactness (SDC) was calculated as SDC = [(BD/BD<sub>max</sub>) x 100], where BD<sub>max</sub> is maximum bulk density, estimated using a pedotransfer function described by Marcolin and Klein et al. (2011). The total porosity (TP) was calculated as TP = 1 - (BD/PD), where, PD is particle density, determined using a gas pycnometer. Soil water content at -6 kPa and -10 kPa water potential was determined using tension tables as described by Ball and Hunter (1988). Soil macroporosity (MaP) was computed as the difference between soil water content at saturation and at -6 kPa. Soil microporosity (MiP) was estimated as the soil water content at the -6 kPa. Water-filled pore space (WFPS) was calculated by dividing volumetric moisture at -6 kPa by total porosity as indicated in Wienhold et al. (2009). We also calculated two indexes suggested by Reynolds et al. (2002): i) soil water storage capacity (SWSC) defined as the ratio between water content at field capacity (FC, -10 kPa soil water potential) and TP (SWSC = FC/TP); and ii) soil aeration capacity (SAC) calculated as the ratio between drained pores at soil water potential of -10 kPa (ACt) and TP (SAC = ACt/TP). A structural stability index (SSI) was calculated as suggested by Reynolds et al. (2009):  $SSI = [(SOC \times 1.724) / (silt + clay)]*100$ . Wet macroaggregate stability (AGS) was determined using a vertical oscillator (Yoder, model MA-148) with three sieves (2000, 250, and 53 µm) moving at a speed of 30 oscillations per min for 10 min. Percentage of macroaggregates was calculated by summing aggregate mass for >2000 and 250 μm classes, dividing by the total soil mass, and multiplying by 100. Mean

weight diameter (MWD) was calculated as the sum of the proportion of aggregates in each size fraction, with each proportion weighted by the mean diameter of aggregates in that size fraction. Soil organic carbon (SOC) and total nitrogen (TN) were determined by dry combustion on a LECO® CN-2000 elemental analyzer (furnace at 1350 °C in pure oxygen). Carbon and nitrogen within microbial biomass (MBC and MBN) were measured by fumigation/extraction as proposed by Vance, Brookes and Jenkinson (1987). Enzymatic activities of β-Glucosidase (BG) and acid phosphatase (AcP) were measured as described by Tabatabai (1994). Immediately after the sampling, soil macrofauna were carefully hand-sorted from each 25 x 25 x 30 cm soil block, according to the standard Tropical Soil Biology and Fertility Institute (TSBF) soil monolith method (ANDERSON; INGRAM, 1993). Invertebrates were classified into the taxonomic groups: Aranae, Blattodea, Chilopoda, Coleoptera, Dermaptera, Diplopoda, Diptera, Formicidae, others Hymenoptera, Gastropoda, Hemiptera, Isopoda, Isoptera, Oligochaeta, and Scorpiones. Macrofauna density was determined as the number of individuals per surface unit (m<sup>2</sup>). Ecological indexes were calculated for assessing richness (Margalef's index) and diversity (Shannon's index), according to the methods described by Magurran (2004).

# 7.2.3 Developing the soil quality indexes

Six SQI values were developed using different approaches (Figure 1), although each involved three common steps: selection of SQ indicators as an MDS, transformation of indicator values into unitless 0 to 1 scores using scoring curves, and integration into an overall index (KARLEN; DITZLER; ANDREWS, 2003). The SQIs were compared to identify the most appropriate strategy for assessing SQ changes induced by LUC associated with sugarcane expansion in Brazil. Soil data from the 0 to 10-, 10 to 20- and 20 to 30-cm layers were averaged to create a 0 to 30-cm layer that was then used to calculate an overall SQI that better represented the whole soil profile.

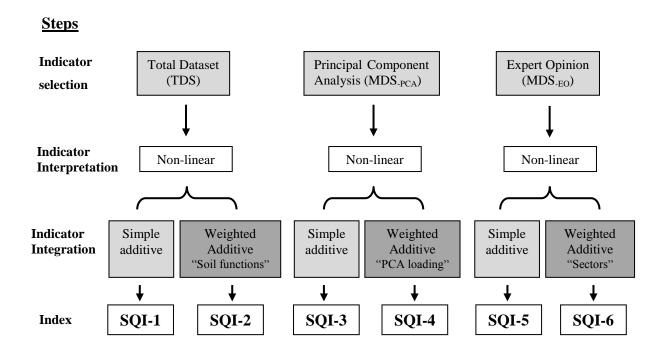


Figure 1 - Process diagram for the development of soil quality indexes tested in this study

# (a) Step 1- Indicator selection

Three indicator selection approaches were evaluated: (i) the Total Dataset (TDS) which included 38 indicators representing 14 chemical, 14 physical and 10 biological properties and processes; (ii) a MDS-PCA created using PCA on the TDS to reduce data redundancy and identify the most efficient indicators, without depending upon subjective, expert opinion or literature values; and (iii) a five indicator MDS-EO chosen based on expert opinion and literature review. For the MDS-PCA, only seven components with eigenvalues >1 (Kaiser's criteria) were retained and subjected to varimax rotation to enhance the interpretability of the components (Figure 2). Furthermore, for each component, only the indicators with loading values within 10% of the highest value were retained (ANDREWS; KARLEN; MITCHELL, 2002; CHEN et al., 2013; ASKARI; HOLDEN, 2014; 2015; LIU et al., 2015). When more than one indicator was retained, correlation values among them were analyzed. If the indicators were significantly correlated (p<0.01), only the one with the highest loading factor was retained in the MDS to avoid redundancy (ANDREWS; KARLEN; MITCHELL, 2002; CHEN et al., 2013; MUKHERJEE; LAL, 2014). The MDS-EO was selected taking into account the indicator's ability to detect soil function changes as well as the ease, practicality and cost-effectiveness for sampling, analysis and interpretation.

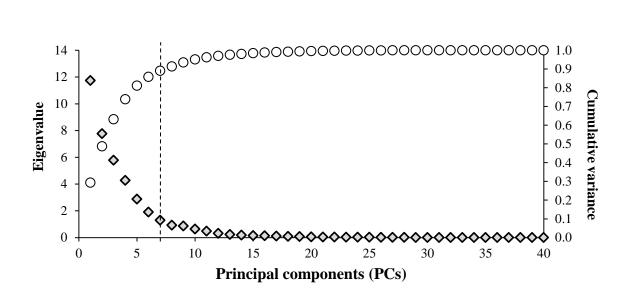


Figure 2 - Scree plot of principal component analysis

#### (b) Step 2- Indicator interpretation

All measured indicator values were transformed using non-linear scoring functions. Based on agronomic and environmental soil functions, each indicator was scored using one of the following curves: "more is better" (upper asymptote sigmoid curve), "less in better" (lower asymptote sigmoid curve), and "mid-point optimum" (Gaussian curve), as exemplified in Figure 3. The non-linear equations 1 and 2 were used for "more is better" and "less is better" scoring curve shapes, respectively. For "mid-point optimum" curve the equations 1 and 2 were jointly used in the increasing and decreasing parts of the curve, respectively.

$$Score = \frac{a}{\left[1 + \left(\frac{B - UB}{r - UB}\right)^{S}\right]} \tag{1}$$

Score = 
$$\frac{a}{\left[1 + \left(\frac{B - UB}{x - UB}\right)^{S}\right]}$$
Score = 
$$\frac{a}{\left[1 + \left(\frac{B - LB}{x - LB}\right)^{S}\right]}$$
(2)

where, Score is the unitless value of the soil indicator which ranging from 0 to 1, a is the maximum score which was equal to 1 in this study, B is the baseline value of the soil indicator where the score equals 0.5, LB is the lower threshold, UB is the upper threshold, x is the measured soil indicator value, and S is the slope of equation set to -2.5.

Threshold and baseline values for each soil indicator were based on literature references and expert's opinion, as presented in the Table 1. Indicator scoring calculations were performed using a Microsoft Excel® spreadsheet.

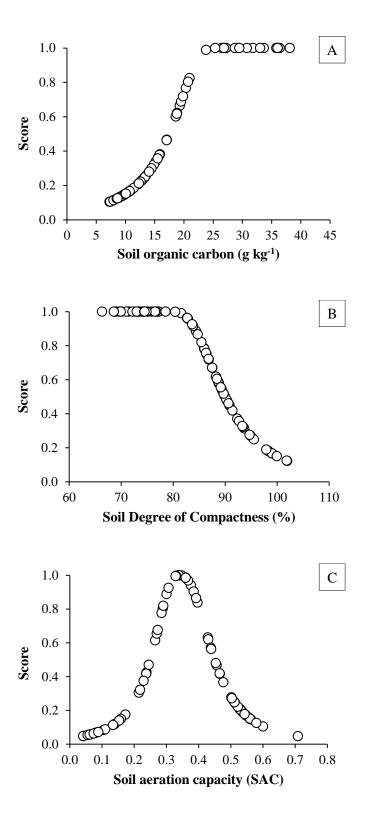


Figure 3 - Examples of the scoring curve shapes used for scoring each soil quality indicator.
A) more-is-better function; B) less-is-better function; and C) mid-point optimum function

Table 1 - Indicator thresholds and scoring curves

Indicator§	Unit	$\mathbf{L}\mathbf{T}^{\ddagger}$	LB	UT	UB	0	Scoring curve	Reference
				Chemica	ıl			
P	mg dm <sup>-3</sup>	2.0	8.0	16.0			More is better	Raij et al. (1997)
S	mg dm <sup>-3</sup>	2.5	5.0	10.0			More is better	Raij et al. (1997)
K	mmol <sub>c</sub> dm <sup>-3</sup>	0.4	0.8	1.6			More is better	Raij et al. (1997)
Ca	mmol <sub>c</sub> dm <sup>-3</sup>	2.0	4.0	8.0			More is better	Raij et al. (1997)
Mg	mmol <sub>c</sub> dm <sup>-3</sup>	2.0	4.0	7.0			More is better	Raij et al. (1997)
В	mg dm <sup>-3</sup>	0.1	0.3	0.6			More is better	Raij et al. (1997)
Cu	mg dm <sup>-3</sup>	0.1	0.4	0.8			More is better	Raij et al. (1997)
Fe	mg dm <sup>-3</sup>	2.0	5.0	12.0			More is better	Raij et al. (1997)
Mn	mg dm <sup>-3</sup>	0.6	2.5	5.0			More is better	Raij et al. (1997)
Zn	mg dm <sup>-3</sup>	0.3	0.6	1.2			More is better	Raij et al. (1997)
$CEC_{pH7}$	mmol <sub>c</sub> dm <sup>-3</sup>	50.0	75.0	150.0			More is better	CQFS-RS/SC (2004)
H+Al	mmol <sub>c</sub> dm <sup>-3</sup>	40.0	80.0	100.0			Less is better	Lima et al. (2013)
pH <sub>CaCl2</sub>	unitless	4.0	4.5	8.0	7.5	5.5	Optimum	Raij et al. (1997)
BS	%	20.0	40.0	80.0			More is better	Raij et al. (1997)
				Physica	l			
BD*	Mg m <sup>-3</sup>	1.1/1.3/1.5	1.25/1.45/1.65	1.4/1.6/1.8			Less is better	Reichert, Reinert and Braida (2003)
SDC	%	80.0	90.0	100.0			Less is better	Reichert et al. (2009)
SRP	MPa	2.0	3.0	5.0			Less is better	Arshad, Lowery and Grossman (1996)
MaP	$m^3 m^{-3}$	0.05	0.075	0.15			More is better	Reynolds et al. (2002)
MiP	$m^3 m^{-3}$	0.15	0.20	0.35			More is better	Expert opinion
TP	$m^3 m^{-3}$	0.20	0.35	0.50			More is better	Expert opinion
WFPS	unitless	0.15	0.30	0.90	0.80	0.60	Optimum	Wienhold et al. (2009)
SWSC	unitless	0.30	0.45	0.90	0.80	0.66	Optimum	Reynolds et al. (2002)
SAC	unitless	0.15	0.25	0.55	0.45	0.34	Optimum	Reynolds et al. (2002)
$K_{fs}$	cm h <sup>-1</sup>	2.0	7.5	15.0			More is better	USDA-NRCS (2001)
AGS	%	0.2	0.4	0.8			More is better	Expert opinion
MWD	mm	0.5	1.5	3.0			More is better	Spohn and Giani (2011)
VESS	score	1.5	3.5	5.0			Less is better	Ball, Batey and Munkholm (2007)
SSI	%	5.0	7.0	9.0			More is better	Reynolds et al. (2009)
				Biologica	al			
SOC	g kg <sup>-1</sup>	10.0	17.5	25.0			More is better	Lopes et al. (2013)
TN	g kg <sup>-1</sup>	1.0	1.75	2.5			More is better	Expert opinion
MBC	mg kg <sup>-1</sup>	200	275	350			More is better	Lopes et al. (2013)
MBN	mg kg <sup>-1</sup>	20	27.5	35			More is better	Expert opinion
BG	mg kg <sup>-1</sup> h <sup>-1</sup>	60	90	120			More is better	Lopes et al. (2013)
AcP	mg kg <sup>-1</sup> h <sup>-1</sup>	75	100	150			More is better	Expert opinion
Eworm	indiv m <sup>-2</sup>	25	100	200			More is better	Bartz, Pasini and Brown (2013)
MDens	indiv m <sup>-2</sup>	50	200	400			More is better	Expert opinion
MRich	unitless	0.0	0.5	1.0			More is better	Expert opinion
MDiver	unitless	0.4	0.8	1.6			More is better	Expert opinion

§P: phosphorus, S: sulfur, K: potassium, Ca: calcium, Mg: magnesium, B: boron, Cu: cooper, Fe: iron, Mn: manganese, Zn: zinc, CEC<sub>pH7</sub>: potential cation exchange capacity, H+Al: potential acidity, pH: potential of hydrogen in solution of CaCl<sub>2</sub> 0.01 mol L<sup>-1</sup> (1:2.5), BS: base saturation, BD: bulk density, SDC: soil degree of compactness, SRP: soil resistance to penetration, MaP: macroporosity, MiP: microporosity, TP: total porosity, WFPS: water-filled pore space, SWSC: soil water storage capacity, SAC: soil aeration capacity, K<sub>fs</sub>: field-saturated hydraulic conductivity; AGS: macroaggregation (>250μm) stability, MWD: mean weight diameter, VESS: visual evaluation of soil structure, SSI: structural stability index, SOC: soil organic carbon, TN: total nitrogen, MBC: microbial biomass carbon, MBN: microbial biomass nitrogen, BG:β Glucosidase activity, AcP: acid phosphatase activity, Eworm: number of earthworm, MDens: macrofauna density, MRich: macrofauna richness and MDiver: macrofauna diversity; ‡LT: lower threshold; LB: lower baseline; UT: upper threshold; UB: upper baseline; O: optimum; \*Threshold values are variable according to soil texture, in order clay, clay sandy and sandy soils, respectively.

# (c) Step 3- Indicator integration into an index

The indicator scores were integrated into indexes through two approaches, simple additive (Equation 3) used to calculate SQI-1, SQI-3 and SQI-5 (Figure 1); and weighted additive (Equation 4) used to calculate SQI-2, SQI-4 and SQI-6 (Figure 1).

$$SQI_{SA} \sum_{i=1}^{n} \frac{Si}{n}$$

$$SQI_{WA} \sum_{i=1}^{n} WiSi$$
(3)

where, *Si* is the indicator score, *n* the number of indicators integrated in the index and *Wi* the weighted value of the indicators. For the TDS, the indicators were weighted according to a framework developed based on five soil functions (Table 2), as suggested by Karlen and Stott (1994) and later used by Lima et al. (2013). Step by step procedure used for calculate the SQI-2 is shown in the Table 2. For the MDS<sub>-PCA</sub>, the indicators were weighted according with proportional variation explained by each principal component (i.e., % variance explained by each component divided by total cumulative variance of all components selected for the MDS). For the MDS<sub>-EO</sub> the indicators were weighted by chemical, physical and biological sectors, in which each one, regardless of number of indicators, had the same weight (33%) in the final index.

Sensitivity of SQ indexing strategies

The sensitivity of the SQ indexing strategies for detecting LUC impacts on SQ was calculated using equation 5, described by Masto et al. (2008).

Sensitivity (S) = 
$$SQI_{(max)} / SQI_{(min)}$$
 (5)

where,  $SQI_{(max)}$  and  $SQI_{(min)}$  are the maximum and minimum SQI observed within each SQ indexing strategies.

Table 2 - Model of soil functions framework and indicators used to develop the SQI-2

Soil Eurotion	Weight			Soil Indicator	·		_	Transformed	Indicator	Soil function score	Weighted soil	COL
Soil Function	Weight	Level 1	Weight	Level 2	Weight	Level 3	Weight	Indicator value <sup>†</sup>	score	$\sum (V*IV*III*II)$	function score	SQI
	I		II		III		IV	V	(V*IV*III*II)	VI	(VI*I)	∑(VI*I)
F(i) -	0.2	Nutrient	0.40	Macronutrients	0.80	TN	0.20	0.44	0.02816			
Storage,		availability				P	0.20	0.86	0.05504			
availability and						K	0.15	1.00	0.04800			
cycling of						Ca	0.15	1.00	0.04800			
nutrients						Mg	0.15	1.00	0.04800			
						S	0.15	0.93	0.04464			
				Micronutrients	0.20	В	0.20	0.92	0.01472			
						Cu	0.20	1.00	0.01600			
						Mn	0.20	1.00	0.01600			
						Fe	0.20	1.00	0.01600	0.000	0.100	
						Zn	0.20	1.00	0.01600	0.900	0.180	
		Acidity/Al	0.40	pH	0.25			0.96	0.09600			
		toxicity		H+Al	0.25			1.00	0.10000			
				BS	0.50			1.00	0.20000			
		Nutrient storage	0.15	CEC <sub>pH7</sub>	0.40			0.78	0.04680			
		and cycling		SOM	0.60	SOC	0.50	0.41	0.01845			
						MBC	0.25	1.00	0.02250			
						MBN	0.25	0.95	0.02138			
		Nutrient cycling	0.05	Enzyme activity	1.00	AcP	0.50	0.90	0.02250			
		, ,		, ,		BG	0.50	0.86	0.02150			
F(ii) –	0.2	Water infiltration	0.25	Kfs	0.70			1.00	0.17500			•••
Infiltration,		water infilitation		Correlated indicators	0.30	SOC	0.20	0.41	0.00615			
storage and						BD	0.50	1.00	0.03750			
availability of						Eworm	0.30	0.19	0.00428			
water, and soil		Water storage and	0.25	SWSC	0.50			0.42	0.05250			
aeration		availability		WFPS	0.30			0.65	0.04875	0.676	0.135	0.848
		avanaomity		MiP	0.10			0.91	0.02275			0.0-10
				Correlated indicator	0.10	TP	1.00	1.00	0.02500			
		Soil aeration	0.50	SAC	0.45			0.13	0.02925			
		Son acration	0.50	MaP	0.45			1.00	0.22500			
				Correlated indicator	0.10	TP	1.00	1.00	0.05000			
F(iii) –	0.2	SOC	0.10	Contracted management	0.10		1.00	0.41	0.04100			
Sustain biological	0.2	Microbial	0.30	MBC	0.50			1.00	0.15000			
activity		biomass	0.50	MBN	0.50			0.95	0.14250			
·		Edaphic	0.40	Eworm	0.10			0.19	0.00760			
		macrofauna	00	Mdens	0.20			1.00	0.08000	0.681	0.136	
				Mrich	0.30			0.79	0.09480	*****	*****	
				Mdiver	0.40			0.69	0.11040			
		Correlated	0.20	SWSC	0.50			0.42	0.04200			
		Indicators	0.20	SAC	0.50			0.13	0.01300			
F(iv) –	0.2	VESS	0.20		0.00			0.98	0.19600			
Sustain the plant	V.=	SRP	0.20					1.00	0.20000			
growth		Soil compaction	0.50	BD	0.50			1.00	0.25000			
· · · · ·		zon compuction	0.50	SDC	0.50			1.00	0.25000	0.984	0.197	
		Correlated	0.10	SOC	0.20			0.41	0.00820	0.701	U.171	
		Indicators	0.10	AGG	0.40			1.00	0.04000			
		-1101041013	0.10	TP	0.40			1.00	0.04000			
F(v) -	0.2	Structural stability	0.60	SSI	0.50		···	1.00	0.30000			•••
Ability to resist	0.2	Su uctural stability	0.00	AGG	0.25			1.00	0.15000			
degradation										1.000	0.200	
		W-4	0.40	MWD	0.25			1.00	0.15000			
		Water infiltration	0.40	Kfs	1.00			1.00	0.40000	1 1 1 2		

<sup>§</sup>Abbreviations are same as Table 1. †Indicator value obtained by non-linear transformation of measured values, as described in the second step of soil quality index calculation.

# 7.2.4 Data analyses

Data were tested for normality using Shapiro-Wilk's tests (p>0.05). The results indicated that no transformation was required. Principal component analysis was performed using PROC FACTOR procedure to select a MDS based on a statistical approach. An analysis of variance (ANOVA) was computed using PROC GLM procedure to test LUC effects on soil indicators and SQI scores. If the ANOVA F statistic was significant (p<0.05), the means were compared using Tukey's test (p<0.05). Linear correlations among SQI strategies were verified by Pearson's correlation analysis using PROC CORR procedure. All statistical procedures were completed using Statistical Analysis System – SAS v.9.3 (SAS Inc, Cary, USA).

#### 7.3 Results and Discussion

### 7.3.1 Soil quality indicators

Land-use change effects on the 38 soil quality indicators at each site are presented in Table 3. As typically reported for tropical soils, native vegetation sites were characterized by high acidity, low levels of soil organic matter (SOM) and plant-available macronutrients, suitable soil physical conditions, and high activity as well as diversity of edaphic fauna. Longterm conversion from native vegetation to extensive pasture significantly increased soil acidification (i.e., decreased pH and increased H+Al concentrations), depleted SOM (SOC and TN), available macronutrients, B and CTCpH7 and, increased micronutrient (Cu, Fe, Mn and Zn) availability. Poor long-term management, which typically includes continuous grazing without liming and/or applying fertilizer over time (DIAS-FILHO, 2014; STRASSBURG et al., 2014), is a major factor for SOC and nutrient depletion within Brazilian pastures. Conversion from native vegetation to pasture also degraded soil physical properties. Continuous cattle trampling coupled with SOC depletion, increased soil compaction (i.e., higher BD and SDC) and altered pore size and distribution (i.e., lower MaP and higher MiP). This subsequently reduced soil aeration (SAC), significantly decreased Kfs and available water, and may restrict root growth (i.e., higher SRP and VESS scores). Despite those changes, soil aggregate stability (AGS and MWD) was not affected by pasture establishment. Soil compaction and consequently physical degradation of pasturelands are well documented in the literature (e.g., GREENWOOD; McKENZIE, 2001; PIETOLA; HORN; YLI-HALLA, 2005; HUNKE et al., 2015).

Table 3 - Mean values of the 38 soil indicators (0-30 cm depth) in native vegetation (NV), pasture (PA) and sugarcane (SC) at three sites in central-southern Brazil

pasture (PA) and sugarcane (SC) at three sites in central-southern Brazil  Lucy S Lat_17S Lat_21S Lat_23S												
<b>Indicator</b> <sup>§</sup>	NV	PA	$\mathbf{SC}$	NV	PA	SC	NIX/	PA	SC			
_	1N V	ra		Chemical		SC	NV	PA	SC			
P (mg dm <sup>-3</sup> )	4.5 b*	2.6 c	6.7 a	12.9 a	5.1 b	9.8 a	14.5 a	10.9 ab	7.6 b			
S (mg dm <sup>-3</sup> )	4.3 b	3.6 b	17.3 a	8.6 a	9.1 a	9.8 a 7.7 a	14.5 a 16.4 a	10.9 ab	6.0 b			
K (mmol <sub>c</sub> dm <sup>-3</sup> )	0.8 a	0.5 b	0.4 b	2.7 a	3.1 a	2.5 a	3.0 b	4.4 a	2.0 b			
Ca (mmol <sub>c</sub> dm <sup>-3</sup> )	3.0 b	2.7 b	20.0 a	69.4 a	7.1 c	2.3 a 29.1 b	19.1 b	31.1 b	49.8 a			
Mg (mmol <sub>c</sub> dm <sup>-3</sup> )	2.4 b	1.3 b	20.0 a 8.7 a	17.6 a	4.1 c	13.0 b	9.9 b	17.8 ab	19.6 a			
B (mg dm <sup>-3</sup> )	0.2 a	0.1 b	0.1 b	0.5 a	0.2 c	0.4 b	0.6 a	0.3 b	0.3 b			
Cu (mg dm <sup>-3</sup> )	3.1 a	0.1 b	3.2 a	0.8 b	1.2 a	1.0 b	1.6 b	2.3 a	1.2 c			
Fe (mg dm <sup>-3</sup> )	43.6 b	85.6 a	20.8 b	15.0 c	1.2 a 164.8 a	51.4 b	87.5 a	90.3 a	21.6 b			
Mn (mg dm <sup>-3</sup> )	9.7 a	3.6 b	4.9 b	32.6 a	14.3 b	16.5 b	45.5 b	100.5 a	14.7 c			
Zn (mg dm <sup>-3</sup> )	0.5 a	0.3 a	0.4 a	2.2 a	1.3 b	1.4 b	2.4 b	4.1 a	0.8 b			
CEC <sub>pH7</sub> (mmol <sub>c</sub> dm <sup>-3</sup> )	78.6 a	54.3 b	60.3 b	2.2 a 104.6 a	60.9 c	71.0 b	2.4 b 169.5 a	103.0 b	105.2 b			
H+Al (mmol <sub>c</sub> dm <sup>-3</sup> )	78.0 a 72.4 a	49.7 b	31.2 c	104.0 a 14.9 c	46.6 a	26.5 b	109.5 a	49.8 b	33.8 b			
$pH_{CaCl2}$ (unitless)	3.7 b	3.7 b	5.0 a	6.1 a	3.9 c	5.0 b	3.8 c	4.6 b	5.4 a			
BS (%)	7.9 b	8.6 b	48.2 a	85.5 a	23.6 c	62.1 b	19.6 b	51.5 a	67.1 a			
DS (70)	1.70	0.00	40.2 a	Physical Physical	23.0 C	02.1 0	17.00	31.3 a	07.1 a			
BD (Mg m <sup>-3</sup> )	1.3 c	1.6 a	1.5 b	1.3 b	1.6 a	1.7 a	1.0 b	1.3 a	1.4 a			
SDC (%)	73.8 b	87.7 a	89.8 a	70.9 b	89.3 a	89.3 a	79.6 b	95.4 a	98.3 a			
SRP (MPa)	1.1 c	1.9 a	1.5 b	0.6 c	2.8 a	1.9 b	2.4 a	2.4 a	2.2 a			
$MaP (m^3 m^{-3})$	0.26 a	0.16 b	0.12 b	0.0 c	0.06 b	0.05 b	0.21 a	0.03 b	0.05 b			
$MiP (m^3 m^{-3})$	0.20 a 0.29 b	0.10 b	0.12 b	0.22 a 0.29 b	0.32 a	0.03 b	0.40 c	0.03 b	0.03 b			
$TP (m^3 m^{-3})$	0.55 a	0.23 c	0.46 b	0.27 b	0.32 a 0.39 b	0.32 a	0.40 c 0.61 a	0.48 a 0.51 b	0.49 b			
WFPS (unitless)	0.40 b	0.37 b	0.40 b	0.31 a	0.54 a	0.63 a	0.48 b	0.87 a	0.47 b			
SWSC (unitless)	0.40 b	0.49 b	0.62 a	0.37 b	0.71 a	0.72 a	0.40 b	0.93 a	0.88 a			
SAC (unitless)	0.53 a	0.47 b	0.31 b	0.59 a	0.71 a	0.72 a	0.39 a	0.07 b	0.12 b			
$K_{fs}$ (cm h <sup>-1</sup> )	130 b	48 b	358 a	129 a	3 b	4 b	46.9 a	1.7 b	0.12 b			
AGS (%)	90.0 a	92.7 a	79.2 b	80.5 a	84.5 a	66.7 b	93.7 b	96.7 a	87.0 c			
MWD (mm)	3.3 b	4.0 a	1.4 c	4.4 a	4.2 a	3.4 b	4.1 b	4.7 a	2.6 c			
VESS (score)	1.8 b	2.0 b	2.5 a	1.8 c	2.9 b	3.7 a	2.5 b	3.2 a	3.3 a			
SSI (%)	5.7 b	9.1 a	4.6 c	11.2 a	7.2 b	6.9 b	7.4 a	6.6 b	4.5 c			
221 (70)	3.7 0	).I u		Biologica		0.7 0	7.1 a	0.0 0	1.5 C			
SOC (g kg <sup>-1</sup> )	13.1 a	8.8 c	11.0 b	16.3 a	10.2 b	9.4 b	35.5 a	30.5 b	19.5 с			
TN (g kg <sup>-1</sup> )	1.0 a	0.5 b	0.9 a	1.7 a	0.9 b	1.0 b	3.1 a	2.3 b	1.5 c			
MBC (mg kg <sup>-1</sup> )	421.9 a	396.0 a	375.6 a	841.2 a	450.1 b	559.3 b	2049.5a	2238.2 a	1024.3 b			
MBN (mg kg <sup>-1</sup> )	41.0 a	22.6 b	17.0 b	75.7 a	30.1 b	21.7 b	98.4 b	161.9 a	43.3 c			
BG (mg kg <sup>-1</sup> h <sup>-1</sup> )	50.5 a	39.8 a	47.1 a	108.2 c	270.0 a	206.2 b	384.2 a	120.8 b	53.4 b			
$AcP (mg kg^{-1} h^{-1})$	204.5 a	154.2 b	138.2 b	151.6 b	256.2 a	229.4 a	324.3 a	326.2 a	167.8 b			
Eworm (indiv m <sup>-2</sup> )	8 a	4 a	4 a	20 b	248 a	36 b	12 b	60 a	4 b			
MDens (indiv m <sup>-2</sup> )	120 b	1428 a	40 b	664 a	772 a	148 b	516 a	888 a	72 b			
MRich (unitless)	0.4 a	0.2 a	0.3 a	0.9 a	0.5 a	0.4 a	0.6 a	0.4 ab	0.2 b			
MDiver (unitless)	0.4 a	0.2 ti	0.5 a	1.2 a	1.1 a	0.4 a	1.1 a	0.7 b	0.5 b			
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\*Mean values within each site followed by the same letter do not differ among themselves according to Tukey's test (p<0.05). Abbreviations are same as Table 1.

Soil biological changes were also observed due to conversion from native vegetation to pasture. Most biological indicators showed site-specific responses, although MBC and MBN tended to be lower within pasture soils, especially at the Lat\_17S and Lat\_21S sites. Enzyme activities (BG and AcP) showed a decreasing trend under pasture at Lat\_17S and Lat\_23S, but increased significantly at Lat\_21S. Variation in soil acidity, SOC, P availability, microbiological activity and other variables not assessed in this study, may be among the

controlling factors affecting enzyme responses at the various sites. Pastures soils generally had a higher density of macrofauna than native vegetation sites, but the increase was dominated by a few taxonomic groups such termites (mainly at Lat\_17S), ants, coleopterans and earthworms. Conversely, even though native vegetation samples had lower macrofauna populations, they had a higher richness and diversity of species. Our findings are consistent with others in the literature (BENITO et al., 2004; DECAENS et al., 2004), which generally state that macrofaunal community size in tropical soils tends to increase over time following conversion from native vegetation to pasture.

The LUC from pasture to sugarcane improved soil chemical quality. Liming and annual application of fertilizer (organic and/or mineral) reduced soil acidity and increased macronutrient availability. Short-term sugarcane establishment (<5 years) had no negative impacts on SOC and TN content at Lat\_17S and Lat\_21S, but as reported by Mello et al. (2014) and Franco et al. (2015), SOC and TN were depleted after more than 20 years (Lat\_23S) of sugarcane cultivation. Those decreases presumably are associated with the intensive tillage performed every five years (SILVA-OLAYA et al., 2013; MELLO et al., 2014) and more than 10 years of pre-harvest burning, which has been shown to deplete SOC over time (CERRI et al., 2011).

Conversion from pasture to sugarcane also negatively impacted on soil physical indicators, primarily those related to soil structure, such as AGS, MWD, VESS and SSI. Although tillage in preparation for sugarcane replanting (Lat\_17S) alleviated soil compaction (i.e., decreased BD and SRP; increased Kfs), our data suggest those positive effects have short-term persistence (i.e., primarily the first year, as reported by Centurion et al. (2007). Over the entire sugarcane cycle, intensive machinery traffic increases soil compaction again, leading to decreased of aeration, infiltration and water availability as observed at Lat\_21S and Lat\_23S. Short-term positive tillage effects on soil physical quality are most likely associated with SOC depletion, due to disruption of macroaggregates and exposure of physically and chemically protected C to microbial decomposition (SIX; ELLIOTT; PAUSTIAN, 2000), and the subsequent deleterious consequences on soil structure. In addition, several studies have shown that intensive machinery traffic in sugarcane fields has negative impacts on soil physical quality and often decreases sugarcane growth and yield (e.g., OTTO et al., 2011; SOUZA et al., 2014). Adverse impacts of current sugarcane management practices on soil physical and structural properties have also markedly increased soil loss and degradation by erosion when compared with native vegetation or pasture (WEILL; SPAROVEK, 2008), becoming one of major concern for a sustainable sugarcane production in Brazil (MARTINELLI; FILOSO, 2007).

Overall, LUC from pasture to sugarcane also has negative implications on soil biological indicators. Depletions of soil biota in sugarcane fields can be associated with quantitative and qualitative decreases in SOC. Franco et al. (2015) reported that sugarcane production depletes C input from C3 plants (forest) which is preferable by microorganisms, and that new C from C4 plants (i.e., pasture and sugarcane) was insufficient to offset those losses. Furthermore, intensification of land use and management, that includes considerable mineral fertilizer and pesticides inputs as well as the modification or destruction of native biological habitats by tillage, and soil compaction can led to a reduction or simplification in soil diversity and its ecosystem functions in sugarcane fields, as reported by Wagg et al. (2014).

# 7.3.2 Soil quality indexing

The three SQ indicator selection approaches (Figure 1) provided different datasets for index calculations. The TDS (38 indicators) provided a wide range of soil indicators and theoretically should have resulted in a more accurate (sensitive) assessment of SQ, due to the very comprehensive evaluation involving chemical, physical and biological soil properties and their interactions. The primary limitations of the TDS approach are the high cost, greater amount of time required for sampling and laboratory analyses, redundancy of indicators, and more complex data interpretation (QI et al., 2009; LIMA et al., 2013; ASKARI; HOLDEN, 2014). Using a PCA reduced the TDS to seven principal components (MDS-PCA) that explained approximately 90% of total variance (Figure 2 and Table 4). Only the indicator with highest loading factor within each PC was retained for the MDS-PCA, since the other highly weighted indicators were well-correlated (r>0.85; p<0.01) among themselves (Table 5). The seven selected indicators were: SOC, SAC, pH, K<sub>fs.</sub> Mdiver, BG and Mdens. Selecting SQ indicators using PCA has some advantages and disadvantages. According to Andrews, Karlen and Mitchell (2002) or Mukherjee and Lal (2014) PCA provides a less subjective method of indicator selection, which can help avoid bias and data redundancy. On the other hand, the PCA method requires a large dataset and is less "user friendly," thus imposing barriers to practical adoption for farm or regional scale SQ assessments. Furthermore, the selected indicators may not be meaningful for farmers and land managers (ANDREWS; KARLEN; MITCHELL, 2002).

Table 4 - Result of principal component analysis

	Principal Components													
	PC1	PC2	PC3	PC4	PC5	PC6	PC7	_						
Eigenvalues	10.33	7.92	5.76	3.17	2.53	2.31	2.19							
Variance (%)	27.19	20.84	15.15	8.35	6.67	6.08	5.75							
Cumulative (%)	27.19	48.03	63.18	71.53	78.20	84.27	90.03							
Soil Indicators				genvectors				Communalities						
P	0.685	-0.035	0.482	-0.018	0.122	0.421	0.076	0.901						
S	0.319	0.166	0.009	0.656	0.147	0.508	0.179	0.871						
K	0.553	0.395	0.194	-0.369	0.424	0.061	0.198	0.859						
Ca	0.243	-0.008	0.930	-0.132	0.035	0.044	-0.049	0.947						
Mg	0.407	0.343	0.797	-0.086	-0.037	0.027	-0.066	0.933						
В	0.610	-0.216	0.387	-0.248	0.130	0.517	-0.094	0.922						
Cu	0.170	0.047	-0.251	0.799	0.023	-0.328	-0.222	0.890						
Fe	0.030	0.257	-0.660	-0.329	0.269	0.106	0.416	0.867						
Mn	0.815	0.296	0.111	-0.060	0.204	-0.222	0.285	0.940						
Zn	0.736	0.198	0.199	-0.103	0.296	-0.080	0.328	0.833						
CEC	0.849	-0.114	0.076	-0.061	0.007	0.418	-0.183	0.951						
H+A1	0.580	-0.190	-0.656	0.052	-0.023	0.374	-0.144	0.967						
pН	-0.046	0.063	<u>0.981</u>	0.048	0.032	-0.028	-0.053	0.976						
BS	0.042	0.202	0.966	-0.028	0.099	0.031	-0.027	0.987						
BD	-0.832	0.375	-0.009	-0.167	-0.049	-0.065	0.286	0.950						
SDC	-0.131	0.900	-0.008	-0.040	-0.237	-0.080	0.054	0.895						
RP	0.123	0.727	-0.408	-0.244	-0.051	0.213	0.158	0.843						
MaP	0.187	-0.932	-0.147	0.144	-0.024	0.025	-0.179	0.980						
MiP	0.641	0.702	0.163	0.114	0.029	0.002	-0.143	0.964						
TP	0.817	-0.326	-0.016	0.249	0.019	0.035	-0.350	0.960						
WFPS	0.215	0.899	0.230	0.108	-0.022	-0.156	-0.097	0.954						
SWSC	0.184	0.964	0.073	0.051	-0.031	-0.037	-0.041	0.975						
SAC	-0.184	<u>-0.964</u>	-0.073	-0.051	0.031	0.037	0.041	0.975						
$K_{fs}$	-0.214	-0.302	0.141	0.858	-0.037	-0.067	-0.040	0.901						
AGS	0.607	-0.060	-0.477	-0.080	-0.235	-0.388	0.085	0.820						
MWD	0.448	-0.150	-0.097	-0.667	0.309	-0.119	0.383	0.934						
VESS	-0.011	0.832	0.111	-0.115	-0.039	0.322	0.010	0.823						
SSI	0.082	-0.586	0.227	-0.477	0.170	0.061	0.511	0.922						
SOC	0.963	0.150	-0.031	0.001	0.016	0.134	-0.052	0.972						
TN	0.929	0.063	0.055	-0.032	0.114	0.277	-0.056	0.963						
MBC	0.903	0.279	0.019	-0.087	-0.002	0.086	0.085	0.916						
MBN	0.866	0.160	0.082	-0.089	0.086	-0.209	0.202	0.883						
BG	0.367	0.099	-0.274	-0.210	0.342	0.729	0.125	0.928						
AcP	0.629	0.340	-0.402	-0.169	0.391	0.172	0.072	0.890						
Eworm	-0.163	0.215	-0.235	-0.003	0.322	0.128	0.520	0.518						
Mdens	0.065	-0.136	-0.134	-0.156	-0.237	-0.013	<u>0.746</u>	0.678						
Mrich	0.236	-0.396	0.186	-0.075	0.749	0.079	0.034	0.822						
Mdiver	0.131	-0.184	-0.020	0.015	0.909	0.129	-0.089	0.903						

Mdiver 0.131 -0.184 -0.020 0.015 0.909 0.129 -0.089 0.903

\*Bold values under each component were highly weighted (factor loading value within 10% of the highest values under the same principal component) and underlined-bold values were selected to minimum data set; \*Abbreviations are same as Table 1.

Table 5 - Pearson's correlation coefficients (r) among soil chemical, physical and biological indicators<sup>§</sup> in the land-use change areas in central-southern Brazil

	southern Brazil																		
	S	K	Ca	Mg	В	Cu	Fe	Mn	Zn	CEC	pН	BS	H+Al	BD	SDC	RP	MaP	MiP	TP
P	0.41	0.60	0.61	0.65	0.83	-0.12	-0.18	0.56	0.65	0.76	0.41	0.51	0.22	-0.58	-0.19	-0.05	0.08	0.47	0.53
S	1.00	0.13	0.02	0.11	0.24	0.32	0.02	0.25	0.26	0.40	0.02	0.06	0.35	-0.27	0.05	0.13	-0.05	0.36	0.30
K		1.00	0.35	0.53	0.46	-0.23	0.35	0.72	0.72	0.42	0.17	0.34	0.06	-0.23	0.16	0.39	-0.38	0.60	0.18
Ca			1.00	0.86	0.55	-0.32	-0.55	0.26	0.33	0.34	0.92	0.93	-0.46	-0.23	-0.05	-0.29	-0.09	0.32	0.20
Mg				1.00	0.50	-0.18	-0.43	0.48	0.47	0.42	0.79	0.86	-0.33	-0.25	0.24	0.03	-0.35	0.64	0.24
В					1.00	-0.34	-0.19	0.39	0.49	0.81	0.30	0.39	0.34	-0.60	-0.33	-0.14	0.24	0.29	0.54
Cu						1.00	-0.19	0.11	-0.01	-0.06	-0.20	-0.25	0.18	-0.31	-0.07	-0.17	0.19	0.21	0.40
Fe							1.00	0.18	0.20	-0.06	-0.64	-0.54	0.35	0.22	0.16	0.60	-0.28	0.02	-0.26
Mn								1.00	0.92	0.51	0.07	0.21	0.22	-0.45	0.13	0.23	-0.21	0.68	0.42
Zn									1.00	0.53	0.16	0.28	0.19	-0.42	0.01	0.15	-0.17	0.57	0.37
CEC										1.00	0.03	0.10	0.67	-0.84	-0.24	0.07	0.28	0.52	0.81
pН											1.00	0.97	-0.69	0.02	0.04	-0.36	-0.19	0.20	-0.03
BS												1.00	-0.64	0.02	0.13	-0.23	-0.32	0.33	-0.03
H+Al													1.00	-0.62	-0.26	0.22	0.42	0.17	0.62
BD														1.00	0.52	0.25	-0.60	-0.36	-0.99
SDC															1.00	0.67	-0.90	0.53	-0.47
RP																1.00	-0.64	0.46	-0.24
MaP																	1.00	-0.52	0.57
MiP																		1.00	0.40
TP																			1.00
WFPS																			
SWSC																			
SAC																			
$\mathbf{K}_{\mathbf{fs}}$																			
AGS																			
MWD																			
VESS																			
SSI																			
SOC																			
TN																			
MBC																			
MBN																			
BG																			
AcP																			
Eworm																			
Mdens																			
Mrich																			
Mdiver																			

Table 5 - Pearson's correlation coefficients (r) among soil chemical, physical and biological indicators<sup>§</sup> in the land-use change areas in central-southern Brazil. Continuation ...

		CIII DI azi				3.63375	TITOGO	COT	000	/DDA /	N. C.	3.5733	D.C.			1 (D	3.6D: 1	
	WFPS	SWSC	SAC	K <sub>fs</sub>	AGS	MWD	VESS	SSI	SOC	TN	MBC	MBN	BG	AcP	Eworm	MDens	MRich	MDiver
P	0.15	0.09	-0.09	-0.10	-0.03	0.30	0.16	0.27	0.69	0.78	0.66	0.57	0.51	0.37	-0.16	-0.01	0.37	0.21
S	0.19	0.22	-0.22	0.42	-0.05	-0.23	0.21	-0.18	0.40	0.44	0.35	0.19	0.42	0.27	0.06	-0.08	0.15	0.21
K	0.45	0.44	-0.44	-0.53	0.11	0.66	0.40	0.19	0.57	0.60	0.66	0.65	0.52	0.66	0.13	0.06	0.35	0.30
Ca	0.23	0.10	-0.10	-0.04	-0.25	0.08	0.10	0.28	0.22	0.31	0.24	0.27	-0.10	-0.22	-0.19	-0.14	0.28	0.08
Mg	0.59	0.46	-0.46	-0.16	-0.14	0.08	0.36	-0.03	0.42	0.44	0.46	0.42	-0.02	0.03	-0.16	-0.10	0.10	-0.03
В	-0.07	-0.09	0.09	-0.26	-0.05	0.36	0.05	0.39	0.62	0.74	0.56	0.45	0.54	0.37	-0.17	-0.03	0.45	0.30
Cu	0.17	0.10	-0.10	0.60	0.17	-0.45	-0.14	-0.61	0.12	0.03	0.03	0.11	-0.25	0.07	-0.10	-0.27	-0.17	-0.05
Fe	-0.02	0.16	-0.16	-0.48	0.28	0.49	0.18	0.12	0.07	0.04	0.12	0.11	0.54	0.57	0.43	0.34	0.01	0.15
Mn	0.46	0.42	-0.42	-0.32	0.43	0.55	0.21	0.12	0.77	0.73	0.84	0.91	0.26	0.68	0.05	0.13	0.20	0.15
Zn	0.35	0.30	-0.30	-0.33	0.25	0.58	0.16	0.26	0.70	0.70	0.71	0.79	0.34	0.63	0.10	0.11	0.28	0.23
CEC	0.06	0.05	-0.05	-0.24	0.35	0.30	0.01	0.10	0.88	0.91	0.73	0.56	0.54	0.50	-0.16	-0.02	0.34	0.24
pН	0.27	0.13	-0.13	0.17	-0.48	-0.17	0.12	0.13	-0.06	0.02	-0.02	0.05	-0.29	-0.42	-0.19	-0.22	0.14	0.00
BS	0.40	0.27	-0.27	0.04	-0.48	-0.07	0.28	0.12	0.04	0.13	0.12	0.16	-0.15	-0.24	-0.16	-0.21	0.17	0.04
H+Al	-0.21	-0.12	0.12	-0.16	0.52	0.20	-0.14	-0.07	0.62	0.59	0.45	0.27	0.57	0.59	-0.01	0.08	0.12	0.19
BD	0.11	0.18	-0.18	-0.05	-0.46	-0.21	0.31	-0.03	-0.76	-0.77	-0.58	-0.55	-0.26	-0.36	0.26	0.08	-0.37	-0.27
SDC	0.80	0.86	-0.86	-0.24	0.03	-0.23	0.67	-0.50	0.03	-0.09	0.15	0.03	-0.12	0.10	0.14	-0.03	-0.50	-0.36
RP	0.49	0.66	-0.66	-0.48	0.26	0.17	0.60	-0.28	0.28	0.22	0.39	0.22	0.46	0.50	0.37	0.07	-0.33	-0.10
MaP	-0.80	-0.87	0.87	0.34	0.20	0.07	-0.78	0.35	0.05	0.11	-0.11	-0.04	-0.01	-0.18	-0.24	0.01	0.33	0.19
MiP	0.84	0.83	-0.83	-0.24	0.27	0.03	0.57	-0.47	0.73	0.66	0.72	0.60	0.22	0.54	-0.01	-0.14	-0.05	0.01
TP	-0.06	-0.13	0.13	0.12	0.46	0.11	-0.29	-0.08	0.76	0.75	0.57	0.53	0.21	0.34	-0.29	-0.14	0.31	0.23
WFPS	1.00	0.94	-0.94	-0.19	0.01	-0.16	0.70	-0.59	0.32	0.21	0.40	0.31	-0.07	0.28	0.01	-0.15	-0.26	-0.15
SWSC		1.00	-1.00	-0.27	0.04	-0.14	0.77	-0.59	0.32	0.21	0.42	0.30	0.07	0.38	0.12	-0.11	-0.32	-0.16
SAC			1.00	0.27	-0.04	0.14	-0.77	0.59	-0.32	-0.21	-0.42	-0.30	-0.07	-0.38	-0.12	0.11	0.32	0.16
$\mathbf{K}_{\mathbf{fs}}$				1.00	-0.17	-0.63	-0.38	-0.20	-0.25	-0.25	-0.33	-0.27	-0.36	-0.47	-0.20	-0.21	0.00	0.01
AGS					1.00	0.38	-0.26	0.02	0.56	0.42	0.52	0.54	0.01	0.33	-0.02	0.24	-0.05	-0.09
MWD						1.00	-0.09	0.67	0.37	0.40	0.43	0.54	0.41	0.52	0.16	0.33	0.40	0.27
VESS							1.00	-0.39	0.12	0.11	0.29	0.11	0.29	0.32	0.18	-0.09	-0.32	-0.18
SSI								1.00	-0.01	0.10	0.04	0.19	0.17	-0.06	0.07	0.37	0.47	0.24
SOC									1.00	0.97	0.91	0.80	0.46	0.67	-0.11	0.00	0.21	0.16
TN										1.00	0.88	0.76	0.57	0.66	-0.08	-0.06	0.31	0.26
MBC											1.00	0.91	0.45	0.70	-0.07	0.05	0.13	0.09
MBN												1.00	0.26	0.62	-0.04	0.07	0.16	0.10
BG													1.00	0.70	0.28	0.03	0.27	0.38
AcP														1.00	0.21	0.03	0.22	0.34
Eworm															1.00	0.31	0.03	0.25
Mdens																1.00	0.07	-0.19
Mrich		00		0.4		0.05 1					11						1.00	0.86

Pearson's correlation coefficients significant at p<0.01 and p<0.05 are highlighted in dark gray and light gray cells, respectively; non-significant values are in white cells; <sup>§</sup>Abbreviations are same as Table 1.

The expert opinion approach reduced the TDS to five MDS-EO indicators (pH, P, K, VESS and SOC), with the first three being chemical indicators that are widely used to evaluate soil acidity and nutrient availability as well as to guide soil fertility management. As recommended by Doran and Parkin (1994), these indicators are desirable for SQ assessments because they are: easy to sample for, readily available in commercial laboratories at a low cost, and the results can be easily interpreted using pre-defined thresholds. The fourth indicator, VESS score, provides an integrative assessment of soil structural/physical quality through an easily-performed, low-cost, direct on-farm method (BALL; BATEY; MUNKHOLM, 2007; GUIMARÃES; BALL; TORMENA, 2011). VESS integrates soil properties related to size, strength and porosity of aggregates, roots and soil color into a single score, which ranges from 1 (good) to 5 (poor structural quality). The fifth indicator, SOC, is the most consistent indicator used for SQ assessments (ZORNOZA et al., 2015), because it influences multiple soil and ecosystem functions (LAL, 2004). Furthermore, SOC can be analyzed using the same sample collected for chemical indicators and it is routinely analyzed so most farmers have previous records for temporal comparisons. The MDS<sub>-EO</sub> approach was consistent with Andrews, Karlen and Cambardella (2004) and Karlen et al. (2008), who recommend that SQ assessments could be made using a minimum of five indicators provided there was at least one each representing soil chemical, physical and biological properties and processes. However, Andrews, Karlen and Mitchell (2002) did warn that expert opinion method does truly require expert knowledge of the entire system and may be subject to disciplinary biases.

Soil quality indicators were individually scored (Equation 1 and 2) and then, integrated using six strategies (Figure 1). The SQI scores for native vegetation, pasture and sugarcane (0-30 cm depth) at each site are shown in Figure 4. Overall, all six SQI approaches were able to detect SQ changes induced by LUC. Soils from native vegetation sites had significantly greater SQI values, except at Lat\_17S, where the soil was more weathered and consequently had very poor chemical quality (CHERUBIN et al., 2015). In general, LUC from native vegetation to pasture significantly decreased SQ, although the sensitivity among the SQ indexing strategies was slightly different. Conversion from pasture to sugarcane promoted site-specific SQ changes, leading to increases or decreases associated with inherent soil characteristics and historic of land use and management.

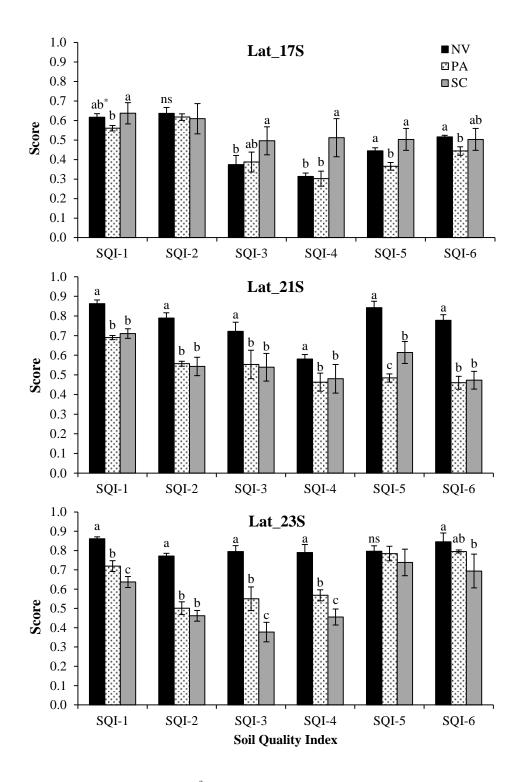


Figure 4 - Soil Quality Index (SQI)<sup>§</sup> scores under native vegetation (NV), pasture (PA) and sugarcane (SC), for the 0-30 cm depth, at three sites in central-southern Brazil. §SQI strategies: SQI-1: TDS/non-linear/simple additive, SQI-2: TDS/non-linear/weighted additive, SQI-3: MDS<sub>-PCA</sub>/non-linear/simple additive, SQI-4: MDS<sub>-PCA</sub>/non-linear/ weighted additive, SQI-5: MDS<sub>-EO</sub> /non-linear/simple additive, and SQI-6: MDS<sub>-EO</sub> /non-linear/weighted additive. \*Mean values within each index followed by the same letter do not differ among themselves according to Tukey's test (*p*<0.05)

At Lat\_17S, sugarcane cultivation increased SQ, primarily due to soil fertility improvement through lime and fertilizer applications. This was confirmed by SQIs calculated using strategies that gave greater weight to chemical indicators, such as SQI-4 and SQI-5. At Lat\_21S, conversion from pasture to sugarcane had essentially no influence on overall SQ, except when SQI-5 was used for the evaluation. In contrast, at Lat\_23S, SQI-1, SQI-3 and SQI-4 indicated that long-term sugarcane cultivation significantly decreased SQ likely due to significant SOM depletion (FRANCO et al., 2015). This in turn had negative implications on micro- and macro-faunal activity, cycling and availability of nutrients, and soil structure (Table 3).

At the regional scale, SQ changes induced by LUC for sugarcane expansion were consistently detected by all six indexing strategies (Figure 5). In general, higher absolute SQI values were observed when using the TDS (SQI-1 and SQI-2) followed by the SQIs from MDS-EO (SQI-5 and SQI-6) and SQIs from MDS-PCA (SQI-3 and SQI-4). An identical sequence was verified by Lima et al. (2013). Native vegetation soils had the highest SQI scores, suggesting they are functioning at 56 to 78% of their potential capacity for the 0-30 cm depth. These results support the hypothesis that natural ecosystems are more balanced, because chemical, physical and biological attributes act collectively, thus enabling soils to perform their functions properly. The SQIs indicated that long-term conversion from native vegetation to extensive pasture decreased SQ indexes by 15 to 23% (Figure 5), resulting in pasture soils that were functioning at between 44 to 66% of their potential capacity. Weighed indexes helped to clarify the reasons for overall SQ depletion within pasturelands. The SQI-2 scores (i.e., TDS weighted by soil function framework) indicated that pasture soils had reduced soil functions associated with storage and provision of water, as well as soil aeration (-32%), soil capacity to sustain plant growth (-18%), biological activity (-30%), ability to resist degradation (-22%), and although not statistically significant, the capacity for storage, provision and cycling of nutrients (-6%) when compared to soils under native vegetation (Figure 6). The SQI-4, weighted by PCA loading, showed that SQ depletions in pasturelands were mainly associated with significant decreases of SOC and Kfs (Figure 7A). Using only five selected, but weighted soil indicators (SQI-6) detected that SQ depletion due to conversion from native vegetation to pasture was associated with significant decreases in soil chemical (-23%), physical (-17%) and biological sectors (-22%) (Figure 7B). This was in agreement with results obtained using SQI-2, which was the most complex strategy.

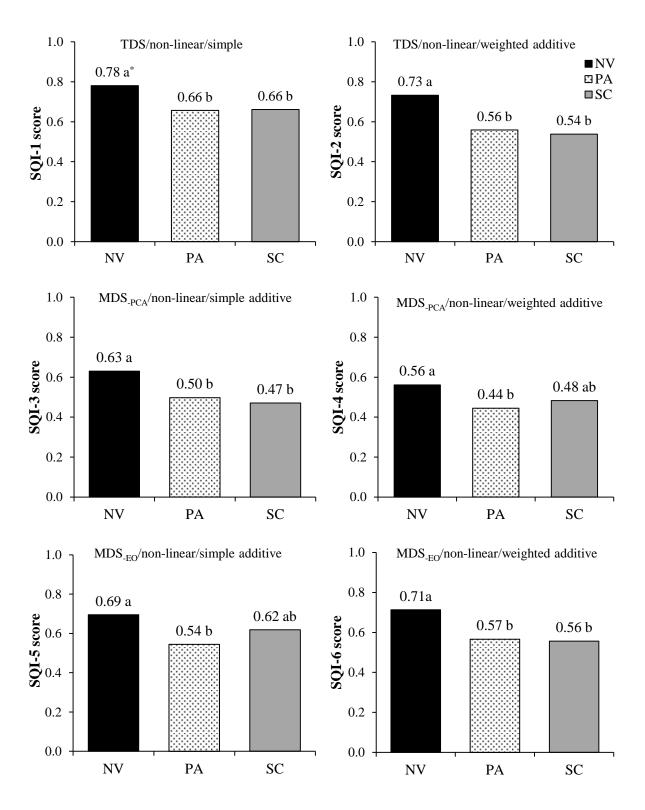


Figure 5 - Overall Soil Quality Index (SQI) scores under native vegetation (NV), pasture (PA) and sugarcane (SC), for the 0-30 cm depth, in central-southern Brazil. \*Mean values within each index followed by the same letter do not differ among themselves according to Tukey's test (p<0.05)

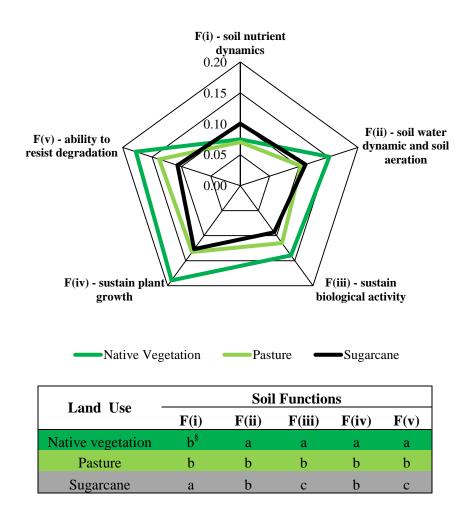


Figure 6 - Contribution of each soil functions in the SQI-2 under native vegetation, pasture and sugarcane in central-southern Brazil. Same letter within each soil function indicates that the mean values do not differ among land uses according to Tukey's test (*p*<0.05)

Our results effectively described the critical situation associated with most Brazilian pastureland. It is estimated that 70% of those areas are degraded or in the process of being degraded (DIAS-FILHO, 2014). Recently, a national-scale study verified that the current productivity (i.e., animal unit carrying capacity) of cultivated pasturelands is only 32-34% of their inherent potential (STRASSBURG et al., 2014). The low productivity of Brazilian pasturelands has multiple causes as reported by Strassburg et al. (2014). Among them are improper pasture management, including seedling failures and bare soil, continuous grazing, absence of liming, maintenance of soil fertility through fertilization, and uncontrolled erosion, which all lead to soil degradation over time (DIAS-FILHO, 2014; STRASSBURG et al., 2014).

At the regional scale, LUC from pasture to sugarcane showed no significant impact on overall SQ (Figure 5). The SQI scores suggest that sugarcane soils are functioning at 47 to

66% of their capacity. The SQI strategies showed sparse non-significant variations on SQ under sugarcane compared to pasture, ranging from -6% (SQI-3) to +13% (SQI-5). Respectively, SQI-3 and SQI-5 were the indexes that gave the lowest and the highest weight to chemical indicators. Therefore, improving soil fertility attenuated negative implications of sugarcane production on soil physical and biological indicators within overall SQ assessment. This was clearly demonstrated by weighted indexes (SQI-2, SQI-4 and SQI-6).

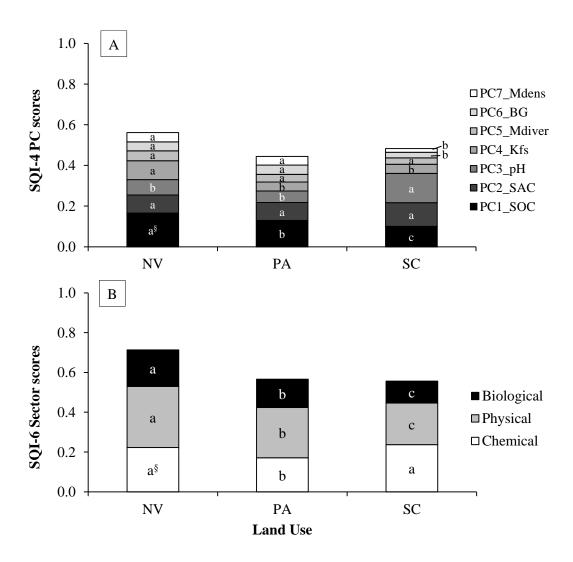


Figure 7 - Contribution of each principal component (PC) and soil sector in the SQI-4 (A) and SQI-6 (B), respectively under native vegetation (NV), pasture (PA) and sugarcane (SC) in central-southern Brazil.  $^{\$}$ Same letter within each PC or soil sector indicates that the mean values do not differ among land uses according to Tukey's test (p<0.05)

Conversion from pasture to sugarcane had one positive effect on soil functions – that related to nutrient dynamics. In contrast, significant adverse effects were observed in soil functions related to the capacities to sustain biological activity and resist to degradation (Figure 6). SQI-4 showed that under sugarcane only pH scores was improved, while SOC, BG

and Mdens scores decreased (Figure 7A). Finally, SQI-6 also was able to indicate that sugarcane production led to significant improvement on soil chemical indicators and decline on physical and biological indicators. Overall, these results indicate that sugarcane expansion over degraded pasturelands seems to be an opportune way to meet increasing domestic and global ethanol demands, avoiding direct competition for land with food crops and natural ecosystems, as reported by Goldemberg et al. (2014) and Strassburg et al. (2014). However, the results clearly indicated the necessity for improved management practices that can mitigate deleterious impacts of sugarcane production on soil physical/structural and biological indicators.

7.3.3 What is the best indexing strategy for assessing sugarcane expansion impacts on soil quality?

All six SQ indexing strategies were able to detect SQ changes induced by LUC, suggesting that any of them could be used for monitoring SQ in sugarcane expansion in Brazil (Figure 5). However, a sensitivity test showed there were slight differences among the strategies (Figure 8). The most complex strategy (SQI-2), which included the 38 indicator TDS and used weighting of the indicator scores provided by the soil function framework had greatest sensitivity to detect SQ changes due to LUC. In contrast, the least sensitive SQI was calculated using TDS without indicator weighting (SQI-1). These results suggest that using a meaningful method (e.g., soil functions) for weighting and integrating indicator scores into an index when a large dataset is available for SQ assessment is best, even though it is more complex than simple additive indexing and does not statistically modify the overall SQ assessment response (Figure 5).

There also is no consensus in the literature regarding the benefits of indicator weighting. Andrews, Karlen and Mitchell (2002) and Askari and Holden (2014) concluded that weighting the additive SQI did not change the relative SQI rankings for the treatments; therefore, this extra step was unnecessary for analyzing vegetable production or other systems. Mukherjee and Lal (2014) also reported similar effectiveness between simple and weighted indexes. They highlighted that appropriate weighting on scores can predict SQ with higher performance which was consistent with our findings. On the other hand, Askari and Holden (2015) showed that a simple additive linear SQI was the most efficient for detecting management practice impacts in arable soils.

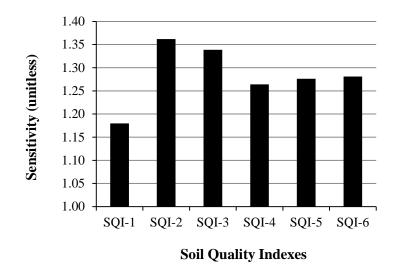


Figure 8 - Sensitivity values of SQ indexing strategies used to assess the land-use change (native vegetation - pasture - sugarcane) impacts on soil quality in central-southern Brazil

Both the MDS<sub>-PCA</sub> (SQI-3 and SQI-4) and MDS<sub>-EO</sub> (SQI-5 and SQI-6) strategies were effective for detecting SQ changes using a reduced number of indicators (Figure 8). Similar results were also reported by Andrews, Karlen and Mitchell (2002) and Lima et al. (2013), who both concluded that a reduced number of carefully chosen indicators could adequately provide the information needed for decision-making. From a practical perspective, this means that for any one indexing strategy to become the standard for research, large-scale SQ assessments, or to facilitate discussion and cooperation, it must be rapid, reliable, and economically feasible (QI et al., 2009). SQI-5 and SQI-6 strategies have an important advantage compared to SQI-3 and SQI-4, since the latter two require a large dataset in order to perform a PCA and select fewer indicators. Therefore, simple SQI strategies (SQI-5 and SQI-6), show excellent potential for monitoring SQ changes in sugarcane expansion areas in Brazil. More specifically, between SQI-5 and SQI-6, we suggest opting for SQI-6, which provides balanced weighting for chemical, physical and biological indicators.

The SQ indexing strategies were significantly correlated among themselves (Figure 9), confirming a close relationship between simple and more complex strategies. This was also reported by Mukherjee and Lal (2014). Decreasing correlations between SQI-1 *vs* SQI-3 (r=0.93), SQI-1 *vs* SQI-5 (r=0.78), SQI-2 *vs* SQI-4 (r=0.47), and SQI-2 *vs* SQI-6 (r=0.34) were observed. This indicates that as the indexes became simpler, correlations with more complex indexes that used the entire dataset were lower. However, despite lower correlations,

simple indexing strategies (e.g., SQI-5 and SQI-6) had the same statistical ability for ranking SQ responses due to LUC (Figure 5) as the more complex strategies.

Simple additive indexes had greater correlations among themselves than weighted additives (Figure 9), because the simple ones were calculated using only a different number of indicators, while weighted indexes also varied the weighting approaches.

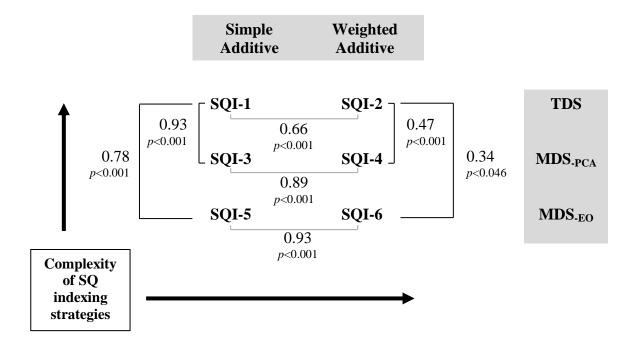


Figure 9 - Pearson's correlation coefficients and probability of error (p) among soil quality indexes (SQI) developed to assess the land-use change (native vegetation - pasture - sugarcane) impacts on soil quality in central-southern Brazil

Higher correlations were verified between simple additive and weighted additive indexes when fewer indicators were selected (i.e., SQI-1 *vs* SQI-2, r=0.66; SQI-3 *vs* SQI-4, r=0.89; SQI-5 *vs* SQI-6, r=0.92). Figure 8 confirms there was a decreasing trend for sensitivity differences between simple and weighted indexes derived from the same sequence, as the comparisons moved from more complex to simpler strategies.

## 7.4 Conclusions

All six indexing strategies efficiently detected SQ changes due to LUC for sugarcane expansion in Brazilian tropical soils. Both, PCA and EO approaches were useful to reduce the total dataset without any significant interference on SQ ranking among land uses. These results indicate that simple, easily-performed and more user-friendly SQI strategies (e.g., SQI-

5 and SQI-6) were as effective and suitable for detecting LUC effects on SQ as more complex SQI strategies (e.g., SQI-1, SQI-2, SQI-3 and SQI-4). Although simple additive and weighted additive SQIs were statistically similar, we recommend using weighted indexes, especially when the number of indicators is unbalanced among chemical, physical and biological components. Therefore, a SQI strategy using a small number of carefully chosen soil indicators, such as pH, P, K, VESS and SOC, and proportional weighting for indicator scores within of each soil sector (chemical, physical and biological) could be adopted as a protocol for SQ assessments in Brazilian sugarcane areas.

Our findings also suggest that long-term LUC from native vegetation to extensive pasture depleted overall SQ, driven by decreases in chemical, physical and biological indicators. In contrast, conversion from pasture to sugarcane had no significant impact on overall SQ, primarily, because chemical improvements offset negative impacts on biological and physical indicators. Therefore, sugarcane expansion into degraded pastureland seems to be a sustainable strategy to meet increasing demands for biofuels. Nevertheless, management practices that alleviate soil physical and biological degradation under sugarcane production must be prioritized to avoid or minimize SQ depletions over time.

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## **8 FINAL CONSIDERATIONS**

Global agreements to mitigate greenhouse gas emissions have raised the demand for low-carbon renewable fuels. Therefore, in recent decades the process of land-use change (LUC) has accelerated for expansion of bioenergy crops worldwide, and consequently has increased the pressure on soil and other natural resources. In Brazil, the world's largest sugarcane-ethanol producer, sugarcane area has expanded by 35% last decade. Land-use change for sugarcane expansion has resulted in extensive pastures being subjected to intensive mechanization and large inputs of agrochemicals (i.e., lime, fertilizer and pesticides). This land-use intensification has direct implications on soil quality (SQ), a key component for ecosystem functioning and consequently a key indicator for assessing the environmental sustainability of biofuel production. For this thesis, we hypothesized that LUC to support sugarcane expansion leads to overall SQ degradation. In order to test this hypothesis we conducted a field-study in three sites in the central-southern region, to assess the SQ response to the primary LUC sequence (i.e., native vegetation to pasture to sugarcane) associated to sugarcane expansion in Brazil.

The overall findings associated with this project show that: in *chapter 2* we concluded that LUC promoted significant impacts on soil chemical attributes. Long-term conversion from native vegetation to extensive pasture led to soil acidification as well as significant SOC and macronutrient depletions due to continuous cattle grazing coupled with the absence of lime and fertilizer inputs over time. In contrast, conversion from pasture to sugarcane decreased soil acidity and increased macronutrient levels. Improvements in soil chemical quality were in direct response to sugarcane management including liming and nutrient replenishment using mineral and/or organic fertilizers. However, long-term (~20 years) sugarcane production resulted in significant SOC depletions (0-30 cm layer). In *chapter 3*, we investigated soil P dynamics due to LUC, since P is the most limiting essential plant growth nutrient in weathered Brazilian soils. We verified that conversion from native vegetation to extensive pasture led to a sharp depletion of soil P pools. Under sugarcane production, soil total P stocks were increased; but most of P added by fertilizer accumulated in less plantavailable P forms. This confirmed the important role organic P has in providing available P to plants in these soils. Furthermore, we found a direct correlation between diversity of soil macrofauna and labile P, confirming the crucial role of soil biota on nutrient cycling and other related ecosystem services.

In chapter 4, our results showed that LUC had deleterious impacts on soil physical quality. Continuous cattle trampling associated to low productivity of pasture (i.e., lower C inputs in the soil) induced significant soil compaction, and created an unbalanced ratio between water- and air-filled pore spaces in the pasture soils. Intensive mechanization used for sugarcane production had a slight negative impact on soil physical properties compared to pasture land use. Tillage performed for sugarcane planting and replanting did alleviate some of the soil compaction associated with long-term pasture, but our data suggested the effects are short-term persistent, and that soil reconsolidation trends to occur over time. Furthermore, periodic tillage decreases soil resistance to erosion under sugarcane production. Overall, our findings suggest that native vegetation, pasture and sugarcane soils were physically functioning at 90%, 70% and from 56 to 68% their potential capacity, respectively. To identify a more user-friendly and cost-effective on-farm method for assessing soil physical changes due to sugarcane expansion, we tested the Visual Evaluation of Soil Structure (VESS) method that was discussed in chapter 5. Overall, the VESS was sensitive for detecting degradation in soil structural quality induced by LUC. In addition, VESS scores were well-correlated with quantitative soil physical attributes, confirming its utility as a potential tool that should be incorporated into protocols for on-farm assessments of SQ in Brazilian sugarcane production areas.

To provide an overall assessment of SQ changes induced by LUC for sugarcane production in Brazil, the second step of this project focused on developing a SQ index (SQI) that integrated soil chemical, physical and biological<sup>4</sup> indicators. We tested different approaches for evaluating SQ changes, since there is not a universal method for such assessments and this research topic is still relatively new in Brazil. In *chapter 6*, our initial use of the Soil Management Assessment Framework (SMAF), a tool developed and broadly used in the USA for assessing SQ changes under wide range of soils, land uses and cropping systems is discussed. The SMAF had never been used for Brazilian soils, so we worked to improve some of the scoring algorithms, thus allowing proper assignment of scores for the soil chemical, physical and biological indicators sampled under Brazilian tropical conditions. The results showed that the SMAF could be used as a reliable and efficient tool to detect SQ changes induced by LUC associated to sugarcane expansion in Brazil. Finnally, in *chapter 7*, we tested six approaches ranging from more complex to simple for developing a

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<sup>&</sup>lt;sup>4</sup> Biological data were measured from soil samples collected at the same sites and sampling time by Franco (2015).

comprehensible SQI. Our findings indicated that simple, easily-performed and more user-friendly SQ indexing strategies (i.e., about five indicators selected by expert opinion and integrated through simple additive or weighted method) were as suitable for detecting LUC effects on soil as more complex strategies [i.e., using the total dataset (38 indicators) and complex weighting procedures based on soil functions]. Therefore, our recommended SQI strategy using a small number of carefully chosen soil indicators, such as pH, P, K, VESS and SOC, and proportional weighting for indicator scores within of each soil sector (chemical, physical and biological) could be adopted as an economically and technically efficient protocol for SQ assessments in Brazilian sugarcane areas.

Overall, the SMAF and SQIs scores suggested that long-term conversion from native vegetation to extensive pasture depleted overall SQ, due to decreases in chemical, physical and biological indicators. On average the soil functioning decreased about 20% from native vegetation to pasture. In contrast, conversion from pasture to sugarcane had no additional negative impacts on overall SQ, mainly because chemical improvements offset negative impacts on biological and physical attributes. Therefore, the projected sugarcane expansion over degraded pastureland seems to be a sustainable strategy to meet increasing demands for biofuels. This suggests that our findings could be used as a scientific base by farmers, extension agents and public policy makers to adopt and develop management strategies that sustain proper soil fertility for sugarcane growth, increase C sequestration, alleviate soil physical and biological degradation for improving SQ and the sustainability of sugarcane production in Brazil.

We encourage future studies to test and validate our protocol for assessing and monitoring SQ changes in sugarcane production under different soils and management practices. Furthermore, we suggest correlating SQ scores with other key-ecosystem endpoints, such as primary productivity, biodiversity, and water and air quality for better understanding the overall environmental sustainability of sugarcane production system.