



Does grassland introduction into cropping cycles affect carbon dynamics through changes of allocation of soil organic matter within aggregate fractions?



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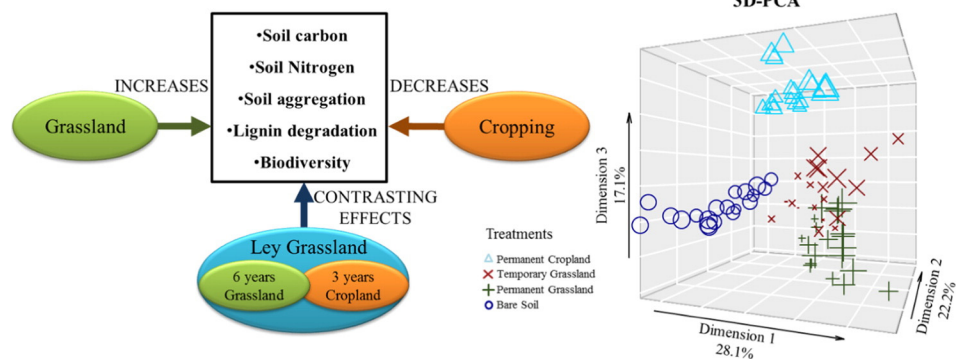
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HIGHLIGHTS

- Changes on soil organic matter dynamics under different land uses are still poor understood.
- Lignin biomarkers and isotopic signature were measured in water stable aggregates.
- Lignin was preserved and carbon was stored in larger aggregates under grassland.
- Litter quality and land use reduced carbon turnover in fine fractions of ley grassland.
- Grassland footprint is still relevant after three years of continuous cropping.

GRAPHICAL ABSTRACT



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ABSTRACT

Implementation of ley grassland into crop rotation could have positive influence in soil ecosystem services such as C storage. The periodical changes of land-use plus the *in situ* labelling given by the introduction of maize crops under ley grassland induce differences in soil organic matter (SOM) that could be traced either by stable isotopes or by the characterization of plant biomarkers such as lignin derived phenols. Evaluation of SOM dynamics is often limited by the complexity of soil matrix. To override these limitations, a hierarchical approach to decompose the soil mosaic into aggregates has been proposed in this study. Soil and plant samples were collected from a long-term experimental area in Lusignan (western France). Soils from four different treatments (bare fallow, permanent maize, permanent grassland, and ley grassland based on 6 years of grassland followed by 3 years of maize) were sampled, fractionated into water stable aggregates, and finally analysed for carbon, nitrogen, and lignin contents, as well as for ¹³C isotopic signature.

Soils under ley and permanent grassland stored higher amount of SOM in larger aggregates and preserved more efficiently the lignin stocks than the corresponding samples under permanent maize. Contemporary, finer fraction of ley grassland showed higher mean residence time of organic carbon, probably due to a legacy effect of the previous years under grassland. Even if maize derived SOM was identified, the grassland footprint was still dominating the ley grassland soils, as described by the principal component analysis. Strong correlation between these results and the quality and stoichiometry of the vegetal litter returned to soil were found, evidencing the

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needs for a comprehensive evaluation at a molecular level of all the parameters modified by land-use changes, including tillage, to understand the potential for carbon storage of different agroecosystems.

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1. Introduction

Achieving a sustainable equilibrium among agricultural practices and environment is the main goal for agronomic research nowadays (Smith et al., 2015a, b; Keesstra et al., 2016). Land management should tend to a sustainable agriculture to guarantee or improve crop yields and soil quality, while limiting greenhouse gases emissions (Lal, 2000; IPCC, 2013). Perennial systems, such as grassland, or alternation of grassland and cropland have been indicated as more efficient than permanent cropping systems in terms of C_{org} sequestration and increasing biodiversity and nutrient availability of the agroecosystem (Lemaire et al., 2011; Gelfand et al., 2013; Lemaire et al., 2015; Parras-Alcántara et al., 2015; Rumpel et al., 2015). Therefore, temporary or ley grassland should be considered not only in terms of fodder production for livestock, but also as a landscape area for realizing some essential ecosystem services, such as absorbing negative environmental impacts resulting from intensive agriculture (Lemaire et al., 2015). For example, nitrate leaching was greatly reduced after the introduction of ley grassland into a cropping cycle (Kunrath et al., 2015).

Soil organic matter (SOM) plays a key role for agroecosystems, given that increasing carbon stocks concomitantly improves soil structure, fertility, and crop yields (Six et al., 1999; Lal, 2002). The storage of organic compounds in soil is mainly achieved by promoting the retention of the litter proceeding from previous crops and/or the incorporation of amendments. However, the mechanisms that underlie the incorporation of C_{org} from residual plant litter into the mineral soil remain widely unknown, since they are widely variable depending on edaphoclimatic characteristics and land-use (Kögel-Knabner and Ziegler, 1993; Gleixner et al., 2002; Franzluebbers, 2004; Panettieri et al., 2015). Furthermore, the complexity of soil samples has always represented a major barrier for molecular-scale analyses of SOM (Derenne and Nguyen Tu, 2014).

In recent studies, the combination of a hierarchic approach provided by soil fractionation and successive chemical characterization of SOM pools has been used as a way to identify different behaviours of SOM at a molecular level (Ludwig et al., 2015; Panettieri et al., 2015). In addition, stable isotope analyses were extensively used to measure the turnover of SOM under different conditions, especially for land-use changes and for conversion from C3 to C4 vegetation (Balesdent et al., 1987; Balesdent and Mariotti, 1996; Dignac et al., 2005). Larger aggregate fraction was identified as the most reactive to changes in land-use, and aggregate disruption induced by tillage has been correlated to carbon losses, worsening of soil physical properties and soil quality (Six et al., 2000a; Bronick and Lal, 2005; Panettieri et al., 2013; Six and Paustian, 2014). However, other authors found more pronounced changes in chemical composition of SOM in smaller size fractions when grassland was changed to arable land (Leifeld and Kögel-Knabner, 2005).

While disaggregating forces are often exogenous or related to changes in land-use, aggregation is mainly led by intrinsic factors of the soil-plant-water system, as for example root activity (Rasse et al., 2005) and the composition of organic inputs added to soil (Hu et al., 2016). Green litter or mucilage demonstrated to greatly increase aggregation at short term, whereas more stabilized OM sources as compost or humified substances have an impact of lower magnitude but more prolonged in time (Abiven et al., 2009).

A deeper follow up of OM accrual after introduction of ley grassland into cropping systems has been carried out using specific plant biomarkers and *in situ* labelling (Armas-Herrera et al., 2016). Among biomarkers, lignin derived compounds are specifically synthesized by

plants and largely stored in soil via selective interactions within some of the compartments of soil matrix (Clemente and Simpson, 2013).

Lignins represent approximately 20% of litter input into soils (Thevenot et al., 2010) and is characterized by slow decomposition rates, thereby being a major controlling compound of litter degradation once added to soil (Sanaullah et al., 2010). However, recent studies found faster turnover for lignin phenols than for total SOM, and consequently different mechanisms for stabilization/degradation of lignins were proposed (Dignac et al., 2005; Thevenot et al., 2013). The hypothesis about the presence of two different pools of lignin characterized by fast and slow degradation rate, respectively, has been described by Lobe et al. (2002) and further confirmed by the model proposed by Rasse et al. (2006) in which 92% of lignin derived compounds are not stabilized within the soil matrix and, thus, quickly degraded. Therefore, a molecular study of lignin fate in soil constitutes a valid approach to the understanding of SOM behaviour (Heim and Schmidt, 2007a).

We hypothesized that ley grassland rotation could improve the net carbon storage at mid- to long-term, changing the dynamics of SOM accrual. Furthermore, we hypothesized that changes in land-use affect the SOM dynamics in specific compartments of the soil matrix so that the overall effect could be hidden when complete soil is analysed, as summarized in Fig. 1. Long term studies investigating soil organic matters dynamics in ley grassland rotations are scarce and showed contrasting effects related to the type of rotation (Studdert et al., 1997), but no available studies have been targeted to the molecular level.

This experiment consisted of maize cropping as in continuous monoculture or as part of temporary grassland. The treatments were established on fields previously cultivated with C3 plants to label by *in situ* natural ^{13}C enrichment the litter input during the cropland phase. We sampled soils, which had been for 9 years under permanent grassland (I), or under permanent cropland (II), left bare for 9 years (III) and under temperate grassland for 6 years (IV). The latter treatment was sampled after the 3 years of crop in order to detect the legacy effect of the ley grassland. The presence of bare fallow plots permitted to compare the persistence of SOM and its allocation within the soil aggregates in absence of fresh inputs and soil perturbation, against the three different treatment analysed. Soil aggregates were isolated to overcome soil matrix complexity using a hierarchical approach. We analysed bulk soil and aggregate samples for stable carbon isotopes as well as lignin biomarkers.

The aims of this study were to (1) explore soil fractions of water stable aggregates to investigate how land-use with changing quantity and quality of litter input (bare fallow, continuous cropping, ley and

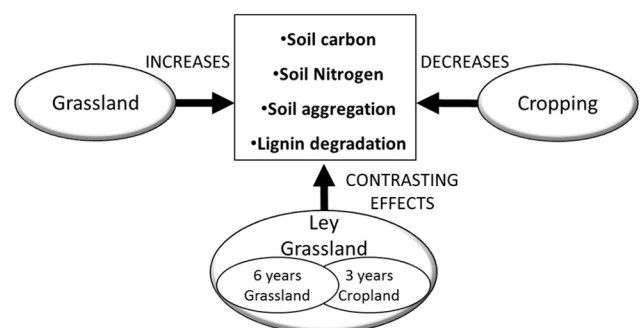


Fig. 1. Simplified schema of interactions between different land-uses and soil parameters analysed in this study.

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