Impact of land use changes on soil carbon pools, gross nitrogen fluxes and nitrifying and denitrifying communities

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Abstract
The COSMOS-Flux project aimed at studying two situations that have important environmental impacts at a larger scale : the conversion tillage ⇆ no tillage where different tillage systems have been applied for 14 years at the start of experiment; the conversion grassland ⇆ annual crop where the introduction of temporary grassland into rotations is studied. The characterization of upper layers of soil for C and N pools, mineralization, immobilization and nitrification of N, along with characteristics of the nitrifying and denitrifying bacterial communities (activity, size and structure) were followed during 18 to 36 months after conversion. We observed that the tillage of soils untilled for 14 years, or the ploughing of the 5-year old grassland were major disturbances for the soils, which led to a very fast evolution of soil organic matter pools, N fluxes and microbial activities towards the characteristics observed for tilled and arable situations. Conversely, the shifts from till to no-till, and the establishment of grassland on soil previously cropped with annual species did not change significantly their soil characteristics at the time scale of the study. Among soil environmental variables, soil organic carbon appeared as a key driver of the observed responses.

Key Words
Environmental impact, land use, organic carbon, microbial function, tillage.

Introduction
Use and land management were long regarded as a problem to benefit primarily local, but now they appear as a factor with implications broader (Foley *et al.* 2005). Indeed, changes in uses and land management related to agriculture occurred and still occur over very large areas of the globe. These changes are very different: it may be conversion of forests into farmland or pasture, or crop intensification, for example. For over a decade and especially since the Rio Summit (2001), awareness of environmental issues related to climate change resulting from human activity, the role of vegetation cover and soil have significantly stepped up the questioning the impact of patterns of land use and agricultural practices. Management methods and their changes can have significant impacts on ecological processes and services provided by ecosystems such as the regulation of climate and biogeochemical cycles, the erosion control, soil formation, the role of 'habitat for living organisms, food production, biodiversity, etc. These practices and developments affect the ecosystem services either directly through their chemical or physical effects, or indirectly through their effects on biodiversity (composition change, number of species and / or alteration of the capacity of the species) (Diaz *et al.* 2007). In this context, microorganisms play a fundamental role in the functioning of the ecosystem including their role in biogeochemical cycles, they are key players in soils subjected to human influence.

The first objective of this project was to study how the characteristics of soil organic matters (quantity and quality), those of soil microbial communities (diversity, size, activity), and fluxes of C and N are coupled in the soil. The second objective of this project was to study how this coupling may be affected by changes in practice and effectively regulates the response of soils. Two situations that have important environmental impacts at a larger scale were investigated: the conversion tillage ⇆ no tillage and the conversion grassland ⇆ annual crop.

Materials and Methods
Experimental site
To study the conversion grassland ⇆ annual crop, the experimental site ORE ACBB (Agro-ecosystems, biogeochemical cycles and biodiversity) in Lusignan, France was chosen, which is studying the role of the introduction of leys in crop rotations (corn-wheat-barley). To study the conversion tillage ⇆ no tillage, the experimental site of Arvalis, Boigneville, France was chosen. On this site, a rotation pea-wheat-barley

received similar agricultural practices and the only differentiating factor for 14 years was the direct sowing (no tillage) and annual plowing. In both situations, the existing plots were split into two in order to follow the usual practices (controls), i.e. pasture aged 5 years and rotation of annual crops at Lusignan, direct sowing annual ploughing at Boigneville on half of the surface, while the other half underwent conversion: pasture ploughed or installed new pasture, ploughing of no tilled plots or abandonment of tillage. The choice to conduct parallel situations either in conversion or as reference treatment (control) (i.e. 4 treatments in parallel per experimental site), is essential to assess the evolution over time of several monitored parameters. Each site was sampled five times for 1.5 to 3 years, with more frequent sampling just after conversions.

Measurements on sampled soils
The characterization of upper layers of soil (0-20 cm and 0-30 cm for Boigneville and Lusignan, respectively) for C (total organic C, soluble C, biomass C and respiration) and N pools (total and mineral N), mineralization, immobilization and nitrification of N, along with characteristics of the nitrifying and denitrifying bacterial communities (activity, size and structure) were followed during 18 to 36 months after conversion. Soils from the 4 treatments were sampled at time 0, +3 weeks, +3 months, + 1 year, +2 years and + 3 years after the conversion, sieved. To assess the activity of nitrifying and denitrifying bacteria, we chose to measure the potential activity of nitrification and denitrification: these measures are often called Nitrifying Oxidizing Enzyme Activity (NOEA) (Smorczewski and Schmidt, 1991) and denitrifying Enzyme Activity (DEA) (Tiedje *et al*. 1989) in literature. This type of measurement is achieved by incubation of fresh soil samples to the laboratory in optimal conditions for achieving these bacterial activities in terms of temperature, oxygenation, and substrate availability. To determine gross N fluxes, we used the method of enrichment-dilution isotope ^{15}N. The gross mineralization is measured by enriching the pool of NH_4^+ with $^{15}NH_4^{\ +}$ and incubating during 24 hours at 15°C (Recous *et al*. 1999). Variations of isotopic abundance and quantities of ammonium nitrate and organic N were measured at the beginning and end of incubation. The rates of mineralization, immobilization and nitrification were calculated using FLUAZ software (Mary *et al*. 1998).

Results
The results first confirm other results that grassland, like direct sowing, install a large gradient of organic carbon in soils, one because residues crop are left on the surface of the soil where they decompose, another because the aerial litter and root of perennial grassland species accumulate in the first horizons below the surface soil. Apart from a possible increase in the total stock of organic C in soil (especially in grassland), a problem which was not the purpose of this project, the distribution of C, including the increased concentration in the “unperturbed” situations, causes a cascading series of "properties" for these layers of soil: increased amount of soluble carbon, increased heterotrophic microbial biomass and soil respiration, increased associated microbial transformations of nitrogen mineralization and immobilization, nitrification, denitrification . As many results on the two experimental sites were produced by this large project, results are presented in Figures 1 and 2 as examples.

The second important result concerns the effects of management changes. In the cases studied, the tillage of plots untilled during 14 years, and the ploughing of grassland aged 5 years, the common point is both the mechanical action exerted on the soil surface layer and the mixing of soil layers previously undisturbed, leading to the destruction of the gradient of organic matter and microbial communities and activities. In reality it is not possible to distinguish the two, however, (i) the first (mechanical) as a consequence of making "accessible" to the decomposition and mineralization of organic forms which were not: organic carbon " protected "by the structure of soil tillage on the one hand, root and aerial parts including roots at various stages of decomposition of the meadow destroyed, (ii) the second factor, the mixing of the layers, leads in terms of organic matter to immediate dilution of organic compartments, and microbial enzymes that cause medium-term adaptation of microbial communities to resources and new environmental conditions. The interpretations of the observed effects of these management changes, multi-factors in terms of the processes studied are complex and cannot actually access only a resultant of two effects.

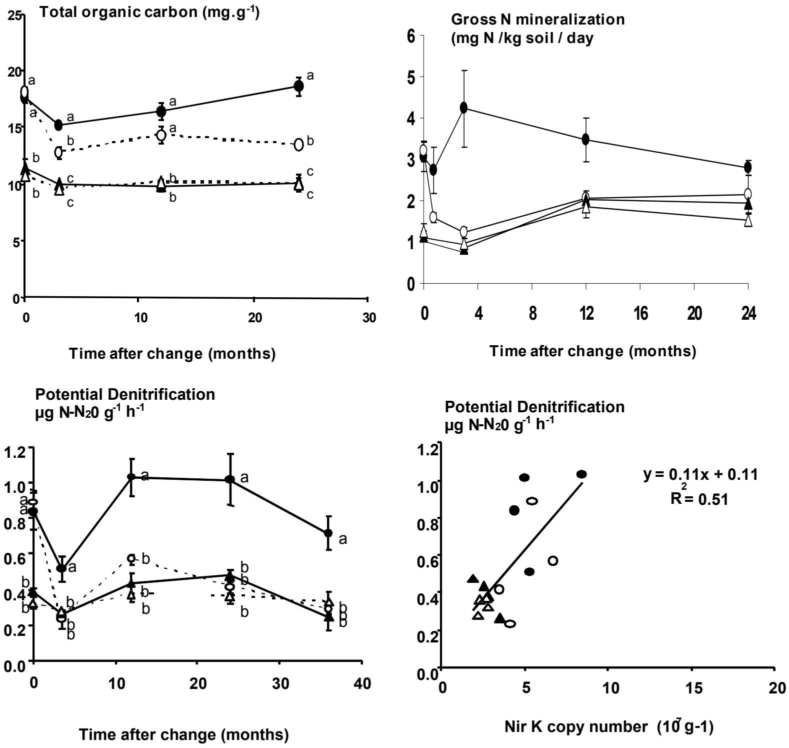

Figure 1. Changes in total organic carbon, the gross N mineralization, denitrification potential and relationship with denitrifier abundance (assessed by nir K copies) in grassland/arable crop conversion, at Lusignan site. ● continuous grassland, ○ploughed grassland, ▲ continuous annual crop rotation, △ establishment of grassland.

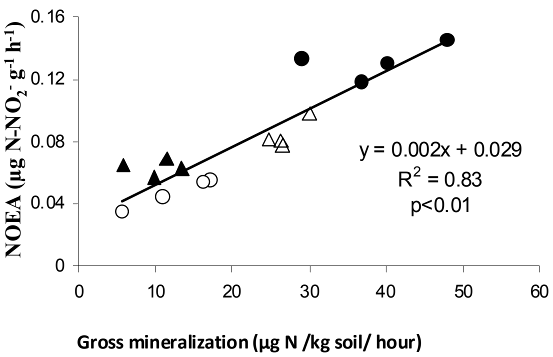

Figure 2. Correlation between nitrite oxidizer enzyme activity (NOEA) and gross N mineralization for soil samples taken from the layer 0-5cm, 2 weeks after the change of soil management on the Boigneville site. ● Continuous no till; ○ abandonment of no tillage; ▲ continuous annual ploughing; △ abandonment of annual ploughing.

Conclusions

The results of this project can draw several important conclusions about the effects of changes in use and management of soils but also transitions between these modes of management. This issue is important for example in the practice of direct sowing, which often includes periodical ploughing, or the establishment of agricultural scheme in conservation farming, or insertion of leys in rotations which aim to promote nutrient management based more on recycling organic materials. The results showed that in "reference" treatments, e.g. till vs. no-till soil, grassland vs. arable soil, soils had contrasted characteristics due to the significant gradient in the accumulation of organic C in the upper layer of no-till and grassland soils. We observed that the tillage of soils untilled for 14 years, or the ploughing of the 5-year old grassland were major disturbances for the soils, which led to a very fast evolution of soil organic matter pools, N fluxes and microbial activities towards the characteristics observed for tilled and arable situations. These effects result both from the cultivation of soil and the mixing of soil layers. Conversely, the shifts from till to no-till, and the establishment of grassland on soil previously cropped with annual species did not change significantly their soil characteristics at the time scale of the study, i.e. 18 months at Boigneville and 36 months at Lusignan sites, as these treatments appear as a cessation of disturbance rather than a disturbance. The results also provided the hierarchy of factors influencing the microbial communities involved in the nitrification and denitrification processes. The genetic structure of microbial communities hardly explained changes in their activity. Community size strongly explained changes in activity level for nitrite oxidizers, not denitrifiers. Among soil environmental variables, soil organic carbon appeared as a key driver of the observed responses.

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