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Quantification and modelling of carbon and nitrogen fate in alternative cropping systems experiments on the long term

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par

Bénédicte AUTRET

**Quantification and modelling of carbon and nitrogen fate in
alternative cropping systems experiments on the long term**

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Abstract

Agricultural activities can lead to imbalanced carbon (C) and nitrogen (N) dynamics compared to natural terrestrial eco-systems, causing potential damages for soil, water and air quality. Among these prejudices, decreased soil C and N stocks, increased nitrate leaching in waters and gaseous N emissions towards the atmosphere are of a major concern. To reduce these environmental impacts, innovative and sustainable farming systems are promoted, such as low inputs cropping systems, “conservation” agriculture or organic farming. At a larger scale, the diffusion of such systems could play an important role in mitigating and adapting agriculture to climate change, assuming their assessment in the long term. The objectives of this work were i) to quantify the long term impact of different alternative cropping systems on the fate of C and N in the soil-plant-atmosphere system and ii) to simulate C and N dynamics with the agro-environmental model STICS. For this purpose, we studied three long-term field trials, namely the experiment of La Cage (France) established in 1998, the DOK (Switzerland) started in 1978 and the Foulum Organic (Denmark) established in 1998. The methodological approach combined experimentation and modelling. While La Cage trial enabled an *in situ* quantification of soil organic C and N storage, N leaching, nitrous oxide (N₂O) emissions and greenhouse gas (GHG) balance in alternative cropping systems compared to conventional, the Swiss and Danish experiments were used for *in silico* estimation of the C and N fates in organic cropping systems, after adaptation of the STICS model to simulate new cropping practices, followed by calibration and evaluation of the model.

After 16 years, significant annual SOC and SON accumulation was found under conservation agriculture (630 kg C and 54 kg N ha⁻¹ yr⁻¹) and organic farming (280 kg C and 31 kg N ha⁻¹ yr⁻¹) on 0-30 cm at La Cage, whereas no significant change was observed in the low input and the conventional systems. We measured the specific SOC and SON mineralization rates (per unit of SOC and SON) of the four systems through a four-month soil incubation and did not detect any difference between systems. Using these results and the AMG model, we conclude that the higher C and N storage in soil observed in the conservation and organic systems was mainly driven by increased C and N inputs deriving from cover crop and catch crop residues, rather than by the effect of no tillage practiced in conservation agriculture. The N surplus, *i.e.* the difference between N inputs and N exports at the field scale, varied widely between treatments, from +47 to +181 kg N ha⁻¹ yr⁻¹ in low input and conservation systems, respectively.

The fate of this N surplus also varied between systems with wide variations in SON storage and gaseous losses but no differences in N leaching. The cumulative N₂O emissions measured continuously during 3.3 years were highly correlated with the calculated gaseous N losses (volatilization and denitrification), with higher losses in the conservation system. Our results allowed to establish a full GHG balance, which was high and positive in conventional and low input systems, smaller in conservation system, and lowest and negative in the organic system. These trends were maintained when the GHG balance was expressed per unit of N input or N exported. Therefore the four agricultural systems dissimilarly impacted the N fate. N fate could not be predicted by the N surplus. The GHG balance is a much better indicator of the environmental impact of cropping systems relative to C and N fluxes.

In the Danish and Swiss experiments, the soil-crop model STICS was used to mimic crop production, N uptake and N surplus. The model was first adapted and evaluated to simulate organic farming systems involving a wider diversity of crops and practices than in conventional systems. The model could satisfactorily simulate crop production, N uptake, N surplus and SON storage in the organic and conventional systems of these two long-term experiments. Model outputs suggested that the N fate could be contrasted according to fertilization and crop management, and that N losses were not systematically reduced in organic compared to conventional cropping systems.

This study challenges the frequent belief that alternative cropping systems necessarily improve the global C and N environmental impacts of agriculture. It points out the complex interactions occurring between alternative practices, and the compensations which can occur between C and N processes. Some of them leading to increased soil C and N stocks, decreased N leaching and gaseous N emissions. Further work should focus on assessing the on-farm performances of these practices on C and N fates and evaluating STICS model in other alternative cropping conditions.

Keyword: carbon storage, nitrogen storage, organic matter turnover, nitrogeN surplus, nitrate leaching, N₂O emissions, GHG balance, organic farming, conservation agriculture, low-input, no-till, modelling

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CHAPTER

1.

Overall framework: environmental impacts
of alternative cropping systems

1.1 General context

Since the end of the Second World War, intensive agriculture has gradually imposed itself on the various forms of pre-existing farming systems, until becoming the paradigm of agricultural production (Pingali, 2012), also called “conventional farming”. The use of synthetic nitrogen fertilizers, which production process was developed by Haber and Bosch at the beginning of the 20th century, has revolutionized agriculture and markedly increased the crop production. Thus, European countries have been able to achieve quantitatively the objective of food self-sufficiency, but not without social and environmental damages (Billen *et al.*, 2011; Sutton *et al.*, 2011). Indeed, although N is most often the limiting element of crop productivity, the massive use of synthetic N fertilizers led to excess N in ecosystems with increased N leaching and gaseous N emissions, causing numerous damages to water bodies and atmosphere (Robertson & Vitousek, 2009; Sutton *et al.*, 2011; Erisman *et al.*, 2013). Lassaletta *et al.* (2014) estimated that 47% of the reactive N applied in agricultural systems is actually converted into harvested biomass, meaning that about 50% of the N fertilizer contributes to the losses listed above (Figure 1.1). Notably, high nitrate (NO₃) concentration in surface waters is responsible for eutrophication, characterized by an algal bloom that prevents light from penetrating beneath water surface layers, and therefore killing plants living below.

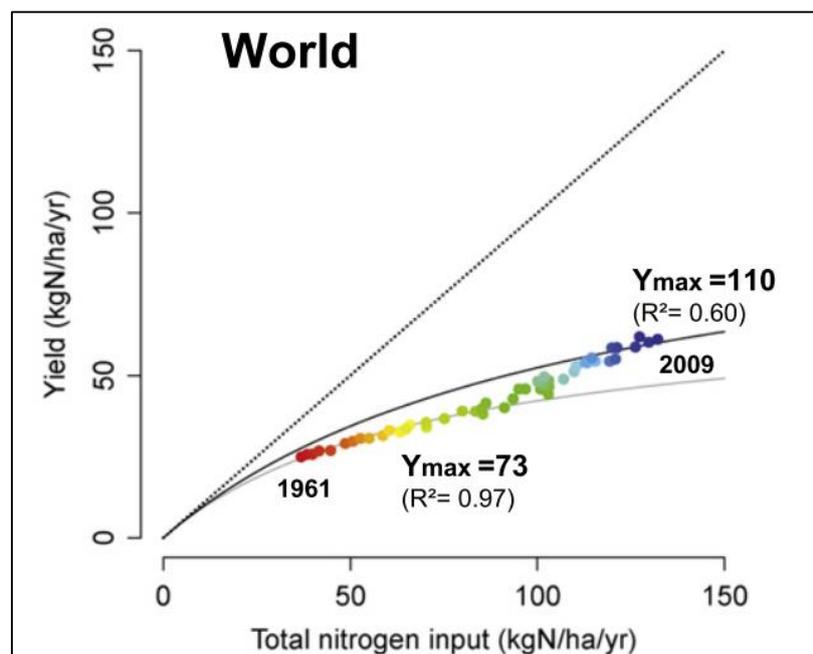


Figure 1.1. Trajectory followed by global world cropping systems in the crop yield in protein harvested versus the total N inputs to the cropland soil (Lassaletta *et al.*, 2014).

The agricultural sector is also responsible of 10 to 12% of total anthropogenic greenhouse gases (GHG) emissions (Smith *et al.*, 2014), among which the nitrous oxide (N₂O), the most potent natural GHG, represents 70% of the total GHG emissions from agriculture (Braker & Conrad, 2011). These N related pollutions are recognized as threats to human health, being responsible among other disorders of numerous cancers, respiratory illnesses, stomach and kidney failures (Erisman *et al.*, 2013). Concomitantly, the geographical separation of livestock and cereal productions led to an unbalanced repartition of carbon and nitrogen between territories (Mignolet *et al.*, 2012), with nutrients depletion in stockless regions and excess in regions of intensive animal husbandry (Goulding *et al.*, 2008).

The awareness of these impacts led to the implementation of environmental legislation by the European Union (EU), in order to alleviate pressures from agricultural activities while ensuring quantitative food security. For instance, the Nitrate Directive (91/676/CEE), implemented in 1991, aimed at reducing the nitrate concentration in surface and ground waters below the threshold of 50 mg NO₃ l⁻¹ by promoting the use of good farming practices. Later in 2000, this directive became part of the Water Framework Directive (2000/60/CE) implemented to achieve the objectives of "good ecological status" of water bodies. More recently, the Directive 2016/2284/EU defined national ceilings for nitrogen oxides (NO_x) and ammonia (NH₃) emissions in order to reach the 2030 objectives of the "Clean Air Programme", but options to reduce emissions have not been defined yet. Soil protection is for its part not totally ensured, as a Soil Framework Directive defined in 2006 was withdrawn in 2014 by the European Commission. However, the 7th EU Environment Action Program recognized that soil needed to be adequately protected by 2020, with efforts to increase soil organic matter. More recently, the "4 per 1000" initiative (<http://4p1000.org/>), launched at the COP21 conference of Paris in 2015, has proposed to increase soil carbon stocks of agricultural areas by 4‰ per year, which could offset 20 to 35% of global anthropogenic CO₂ emissions (Minasny *et al.*, 2017). Therefore, compliance with the EU policies, coupled with the awareness of the mitigation power of agriculture on global warming, raised the need for defining sustainable and resilient agricultural practices.

1.2 Alternative cropping systems – definition

1.2.1 From conventional to alternative practices

Conventional agriculture aims at maximizing crop production per unit of cropped area. It requires both large investments and intensive use of agricultural inputs (energy, fertilizer, equipment), leading to numerous adverse effects relative to soil, air and water pollution. Costs of these detrimental effects are not fully known, but Sutton *et al.* (2011) estimated that N related pollutions could represent between 70 and 320 billion of euros per year to the EU. The changeover of agriculture towards more sustainable practices is essential for the purposes of mitigating the environmental impacts while keeping satisfactory yields, in order to meet food need of the world population expected to be 50% larger than at present by 2050 (Tilman *et al.*, 2002). A sustainable agriculture is supposed to be “environmentally sound, resource-conserving, economically viable and socially supportive” while maintaining productivity (Sadok *et al.*, 2008). The development of sustainable agriculture requires the implementation of “alternative cropping systems” (Doré *et al.*, 2011), now referred to by the umbrella term of “agro-ecology”, replacing the dominant form of agriculture by systems favouring ecological processes (Estevez & Domon, 1999). In particular, alternative cropping systems are currently defined at the plot scale while agro-ecological practices combine several scales (Figure 1.2).

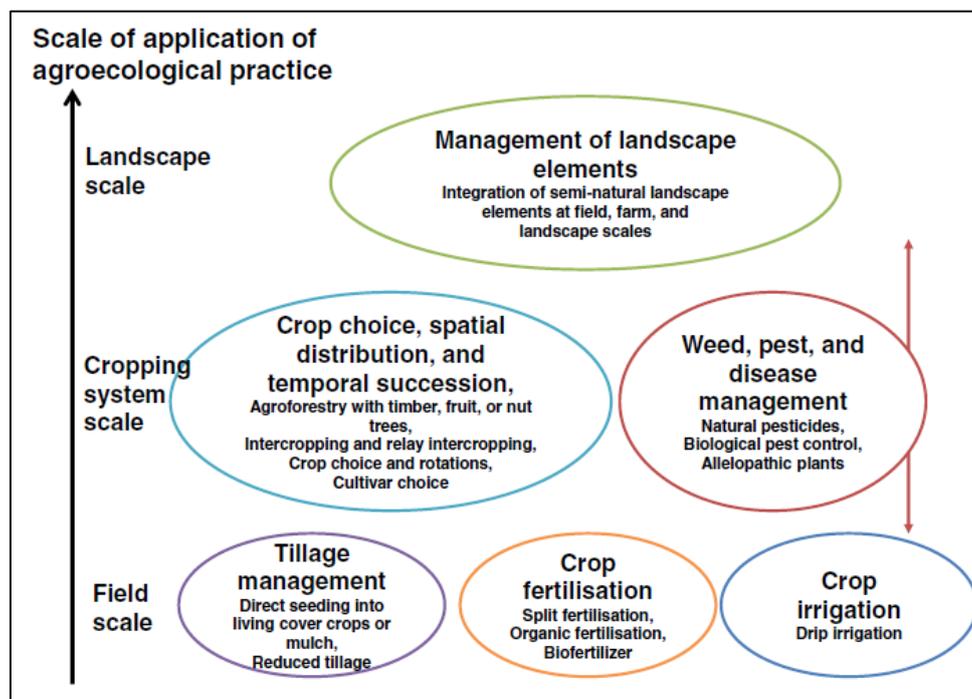


Figure 1.2. Different categories of agro-ecological practices, applied at different scale, from field scale to landscape scale. The arrow of “weed, pest, and disease management” indicates an application at all level (Wezel *et al.*, 2014).

These systems do not meet a single set of specifications because they attempt to provide local answers to questions asked in a more global perspective, particularly in the case of the impact of agriculture on the environment. However, they can be characterized by common interrelated practices like the use of on-farm resources such as animal manures and legumes to provide plant nutrients; crop residues, cover crops, and conservation tillage to control soil erosion and runoff; crop rotations to control weeds, insects and diseases; and conservation tillage to save energy and reduce operation costs. More globally, alternative cropping systems “advocates smaller farm units, reduced energy use, greater farm self-sufficiency, [...] conservation of finite resources” (Beus & Dunlap, 1990). In principle, alternative cropping systems do not seek maximum yield, but optimal production in relation to the constraints of the environment. Therefore, they have a strong dependence on natural resources and low dependence on chemical inputs. The most widespread alternative cropping systems in Europe are listed here, along with their main features:

- Reasoned agriculture: appeared in France in the 70’s and defined by (Paillotin, 2000) as a “set of practices that aim at reinforcing the positive impacts of agricultural practices on the environment and to reduce the negative effects, without questioning the economic profitability of the farms”. No common rules related to reasoned agriculture have been defined within the EU (Angelucci & Mundler, 2007). Overall, it seeks to substitute the “blind” struggle against weeds and pests by a “reasonable fight” based on thresholds of crop tolerance (Ferron, 1999). Thus, the use of pesticides is not systematic but controlled according to the needs of the crops along with an adjusted fertilization (Blouet *et al.*, 2003). However, the existing marketing around reasoned agriculture was criticised as it was promoted as a prototype breaking with the conventional agriculture when in fact maintaining the use of pesticides and sustained by the main partners of intensive agriculture.
- Integrated agriculture: defined by Boller *et al.* (2004) in the guidelines for the IOBC as a “*farming system that produces high quality food [...] by using natural resources and regulating mechanisms to replace polluting inputs and to secure sustainable farming*”. It is often presented as an extension of the concept of “integrated protection” to “integrated production”, the first being restricted to a limited use of pesticides while the second enlarges this limitation to fertilizers use. The main principles of integrated farming are the diversification of crop rotation to reduce the risk of pest and weed pressure, soil protection during winter to reduce the risk of nitrate leaching and targeted application of fertilizers and pesticides (Morris & Winter, 1999; Boller *et al.*, 2004; Nemecek *et al.*, 2011).

- Conservation agriculture: officially promoted and defined by the FAO as “an agriculture based on three main principles: the minimal soil disturbance or absence of deep ploughing, the diversification of crops in the rotation and a permanent plant soil coverage” (FAO, www.fao.org/ag/ca/). Conservation agriculture principles emanate from conservation tillage, which includes no tillage. In theory, these three principles should be applied simultaneously, but this is rarely true. For example, many published studies refer to “conservation” whereas only no-till is applied. The three principles have their consistency: weeds pressure can be reduced by a maximum soil coverage by auxiliary crops. The maintenance of a permanent or semi-permanent plant cover is promoted as a natural soil protection against rain and erosion, but also as a resource for macro- and micro- soil organisms. Among these organisms, earthworm communities are supposed to improve soil structure, providing a biological tillage through soil bioturbation (FAO, 2001). The permanent cover crops can control weeds and reduces the need for herbicides (Kassam et al., 2009). However, the use of pesticides is not systematically reduced in conservation agriculture, particularly herbicides which are the only way to control weeds and reduce the growth of auxiliary crops (Sans et al., 2011).
- Organic agriculture: defined by the FAO (www.fao.org/organicag/oa-home/en/) as “*an overall production management system that excludes the use of synthetic fertilizers and pesticides and genetically modified organisms, minimizes air, water and soil pollution and optimizes the health and productivity of interdependent communities of plants, animals and humans*”. Organic farming is based on ecological processes, biodiversity and cycles tailored to local conditions, rather than on the use of inputs with adverse effects. Hence, the notion of self-sufficiency is central in organic farming with minimized external inputs (Watson et al., 2002a). The non-use of herbicides often results in significant tillage in order to limit competition with weeds, even though no tillage or reduced tillage can also be implemented in organic farming (Peigné et al., 2007).

Despite a growing interest for the alternative cropping systems presented above (National Research Council, 2010), their implementation and understanding still need to be improved in order to achieve environmental objectives laid down by the European Directives.

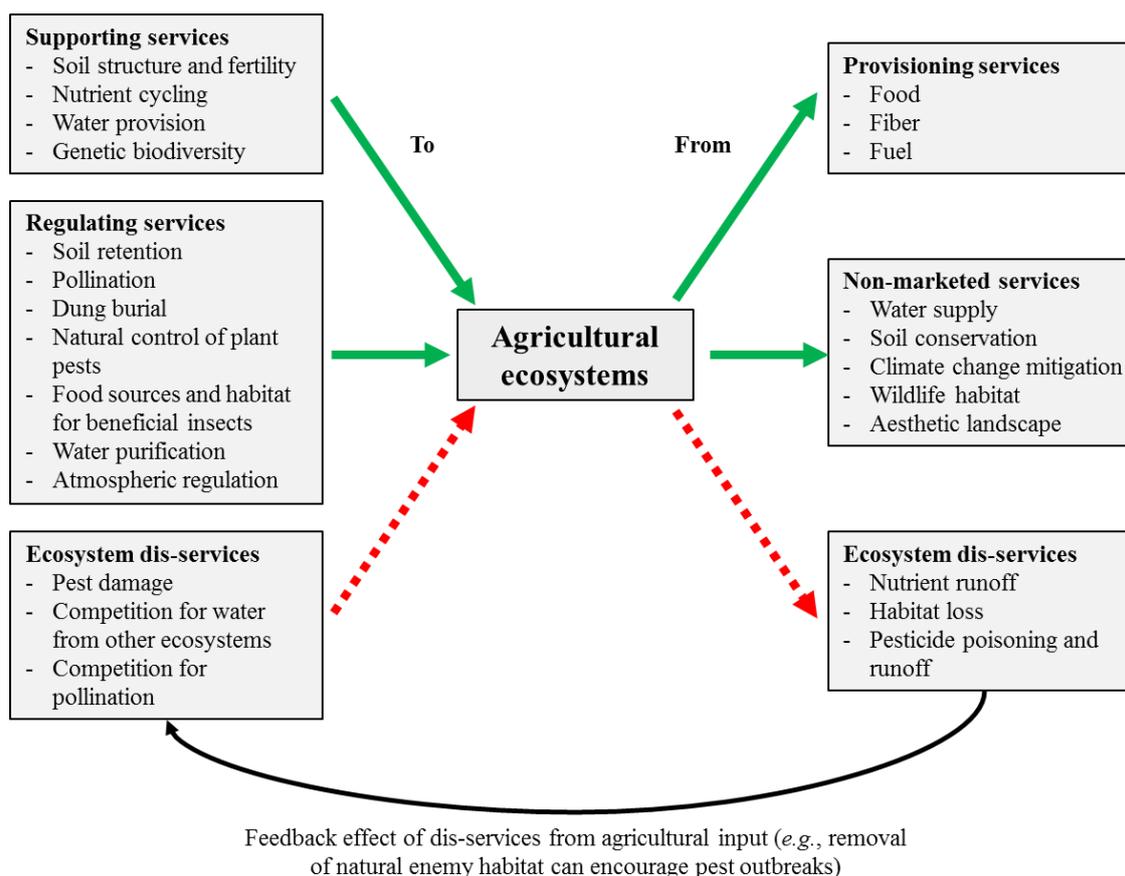


Figure 1.3. Ecosystem services and dis-services to and from agriculture. Green arrows indicate services, whereas red dashed arrows indicate dis-services (adapted from Zhang *et al.*, 2007).

Table 1.1. Properties of natural ecosystems compared with conventional and alternative agroecosystems (adapted from Pretty, 2008).

Property	Natural ecosystem	Conventional agroecosystem	Alternative agroecosystem
Productivity	medium	high	medium
Species diversity	high	low	medium
Functional diversity	high	low	medium-high
Biomass accumulation	high	low	medium-high
Nutrient recycling	closed	open	semi-closed
Trophic relationships	complex	simple	intermediate
Natural population regulation	high	low	medium-high
Resilience	high	low	medium
Dependence on external inputs	low	high	medium
Human displacement of ecological processes	low	high	low-medium

1.2.2 Alternative practices and ecosystem services

Agriculture provides and relies upon important ecosystem services, that were defined by the Millennium Ecosystem Assessment as “the benefits people obtain from ecosystems” (MEA, 2005). Agriculture modifies three categories of ecosystems services: the supporting services (*e.g.* soil formation, nutrient recycling), the provisioning services (*e.g.* food, water, oxygen) and the regulating services (*e.g.* climate regulation, carbon sequestration). The main goal of agriculture is to optimize one provisioning service which is food production (Figure 1.2). The production relies on regulating services, such as soil fertility and pollination, and can be decreased by “dis-services” (Zhang *et al.*, 2007) such as water shortage and pest damages. Simultaneously to the production process, agriculture can produce “negative externalities”, like soil, water and air pollution by oversupplied nutrients and pesticides (Roy *et al.*, 2009).

Compared to conventional agro-ecosystems, alternative agro-ecosystems are supposed to increase the non-marketed services, to reduce the ecosystem “dis-services” while maintaining provisioning services, their properties being closer to natural ecosystems (Table 1.1). Numerous studies reported the environmental performances of alternative cropping systems (Mondelaers *et al.*, 2009; Scopel *et al.*, 2012; Palm *et al.*, 2014; Ponisio *et al.*, 2015; Reganold & Wachter, 2016). For instance, an increased biodiversity at the plot and landscape scale has been reported in conservation agriculture (Palm *et al.*, 2014) and organic agriculture (Bengtsson *et al.*, 2005; Reganold & Wachter, 2016). In particular, greater floral and faunal diversity have been reported in organic agriculture, with more cultivated crops, diverse birds, insects and soil microflora (Kennedy *et al.*, 2013; Tuck *et al.*, 2014). A greater abundance of arthropods and microbial communities has also been observed in conservation agriculture compared to conventional agriculture (Rodríguez *et al.*, 2006; González-Chávez *et al.*, 2010), in the topsoil layer. A larger abundance of earthworms was found in conservation agriculture but with a lower functional diversity (Pelosi *et al.*, 2016). More diverse functionalities of microbial communities can also be observed (Lynch, 2012; Tuck *et al.*, 2014), with a 30% increase in species richness under organic agriculture compared to conventional (Bengtsson *et al.*, 2005). Thus, alternative agro-ecosystem tend to offer better conditions to wildlife (Mondelaers *et al.*, 2009; Scopel *et al.*, 2012), willing to regulate pests populations, increase yield with an enhanced pollination, and participate to organic matter decomposition and nutrient cycling (Loranger-Merciris *et al.*, 2006). Soil erosion is generally reduced in alternative agro-ecosystems such as organic (Arnhold *et al.*, 2014; Soriano *et al.*, 2014) or conservation agriculture (Palm *et al.*, 2014), often linked with the higher soil coverage during winter. In addition, the decrease of soil and water

pollution is particularly important in organic agriculture (Stolze *et al.*, 2000; Alföldi *et al.*, 2002), in which chemical fertilizer and pesticide use is forbidden. However, although reduced, the use of pesticides in integrated and conservation agriculture can lead to water and soil pollution (Baker *et al.*, 2002; Kay *et al.*, 2009).

Finally, alternative cropping systems are often mentioned for their role in adapting and mitigating climate change, through optimal recycling of nutrients and organic matter turnover (Gattinger *et al.*, 2012). However, controversies emerge from the literature concerning soil C and N storage (Govaerts *et al.*, 2009; Leifeld & Fuhrer, 2010; Tuomisto *et al.*, 2012), reduction of nitrate leaching and greenhouse gas emissions (Mondelaers *et al.*, 2009; Snyder *et al.*, 2009; Palm *et al.*, 2014).

1.3 Carbon and nitrogen cycles in agro-ecosystems

1.3.1 Global trends in carbon and nitrogen cycles

The soil organic matter (SOM) can be defined as all organic soil compounds including living biomass and dead organic matter, *i.e.* plant and animal residues at various stages of decomposition (Calvet *et al.*, 2011). The latter is involved in soil fertility as it influences soil structure through the formation of aggregates that promote water retention, aeration and structural stability of the soil, along with nutrients availability for soil organisms and plants cultivated in agriculture (Citeau, 2008; Chenu *et al.*, 2014). The carbon (C) and nitrogen (N) represent a significant proportion of the SOM, about 58% for the C, the proportion of N being variable depending on soils (C/N ratio varying between 8 and 12 in arable soils). In natural ecosystems, the soil organic C and N are coupled and cycle together, perturbations in the N cycle having repercussions on the carbon cycle, and *vice-versa* (Zaehle, 2013). Since the start of the industrial era, the terrestrial ecosystems C and N cycles have been strongly disturbed and uncoupled (Figure 1.3).

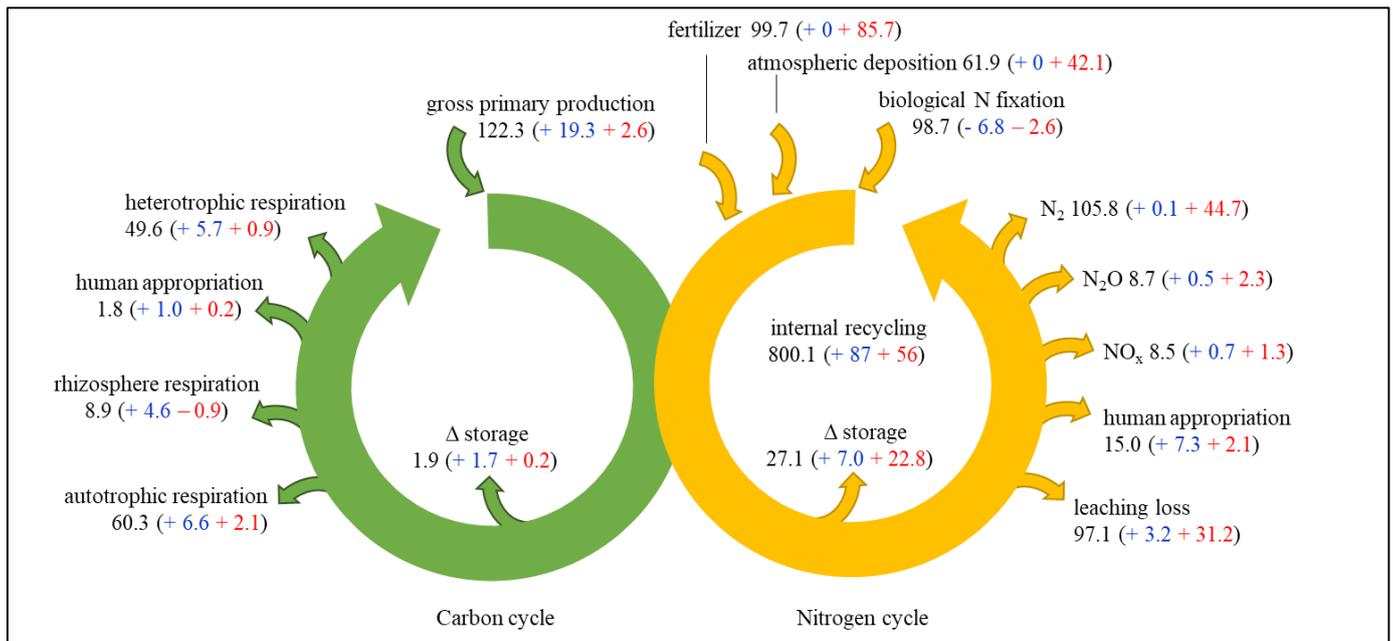


Figure 1.4. The global carbon and nitrogen cycles in terrestrial ecosystems (estimation for the 2001–2010 period). The values in parentheses are the changes from the pre-industrial equilibrium fluxes (1860) owing to land-use, climate and atmospheric CO₂ change (blue) and anthropogenic nitrogen additions (red). Carbon fluxes: Pg C yr⁻¹; nitrogen fluxes: Tg N yr⁻¹. NO_x, N₂O and N₂ emissions are from soils only (adapted from Zaehle, 2013).

Notably, agricultural activities impact on C and N cycles has been shown, including:

- The shortening of crop rotations, *i.e.* the decrease of crop diversity in the rotation, and the withdrawal of perennial grasslands in favour of annual crops, lowering the SOM stocks (Matson *et al.*, 1997; Drinkwater & Snapp, 2007);
- The massive application of N fertilizers, increased by a factor of 4.4 between 1961 and 2009, versus a smaller increase in the N exported from cultivated land, resulting in N losses 7.3 times higher in 2009 than in 1961 (Lassaletta *et al.*, 2016).
- The land use change caused a total loss of 40 to 90 Gt of C through cultivation and disturbance since 1850, the current rates of C loss from cultivated soil being of about 1.6 ± 0.8 Gt C yr⁻¹ (Lal, 2009).

1.3.2 Soil organic matter turnover in agro-ecosystems

1.3.2.1 Soil organic matter compartments

In agro-ecosystems, the SOM content is the result of two opposed fluxes: on one side the inputs of organic matter through returned crop residues (*e.g.* straw, senescent leaves, dead roots) and application of organic fertilizers (*e.g.* manure, slurry, sludge, green manure), and on the other

side the losses of SOM by mineralization, leaching and erosion (Liu *et al.*, 2006; Chenu *et al.*, 2014). In order to predict the dynamics of SOM, compartmental models identify dynamical “pools” (Figure 1.4), characterized by a specific potential decomposition rate and factors controlling the effective decomposition rate (Six *et al.*, 2002; von Lützow *et al.*, 2008). The mean residence time (MRT) of each soil organic pool is the ratio of the amount of SOC contained in this pool to its incoming C flux, also equal to its output C flux since this calculation refers to steady state. Thus, SOM models include various pools with different MRT, at least three pools (Figure 1.4): a labile pool, which is easily degraded (MRT from 1 day to 1 year), an active pool (MRT from 1 year to a few years) and a stable pool, which turns over in decades or centuries. Both labile and active pools originate predominantly from plant, animal, bacterial and fungal residues, the second being also supplied by degradation products from the first. The stable pool originates from labile and active pools and includes the largest part of soil organic C (Torn *et al.*, 2009). In this pool, the physical protection of C by soil aggregates and the physico-chemical protection by adsorption, humification or complexation with mineral particles all contribute to the slow degradation rate (Six *et al.*, 2002). For cultivated soils, the mean MRT of SOC (all pools mixed) has been estimated at 61 ± 9 years in a synthesis of 10 studies (Six & Jastrow, 2002). It is higher in the deepest soil layers (Fontaine *et al.*, 2007).

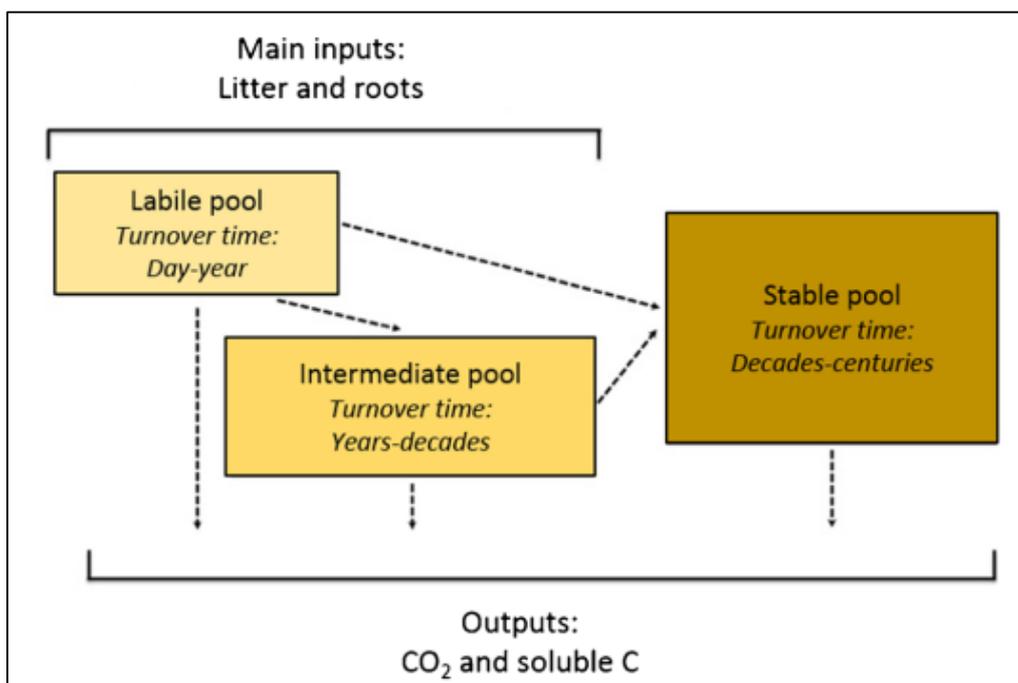


Figure 1.5. Conceptual pools of soil C depending on its turnover time: labile, intermediate and stable pools (Dignac *et al.*, 2017).

1.3.2.2 Soil organic matter decomposition

The decomposition of SOM is a complex process that depends on many factors including soil minerals (clay, carbonates, iron oxides, ...), climate (temperature, rainfall) and agricultural practices (residue management, soil tillage...). SOM decomposition can be defined as the combination of three processes (Baldock & Skjemstad, 2000): i) the conversion of organic matter from a chemical structure to another in response to enzymatic attacks and chemical reactions; ii) the incorporation of organic C and N into heterotrophic decomposers, and iii) the mineralization. C mineralization is the conversion of organic C into CO₂ by heterotrophic organisms under aerobic conditions, and into methane (CH₄) under anoxic conditions. N mineralization leads to release of ammonium (NH₄⁺) through the ammonification process (gross mineralization). Nitrification then converts ammonium into nitrite (NO₂⁻) first and then nitrate (NO₃⁻), both steps being realized by autotrophic bacteria (Benoit *et al.*, 2015). The denitrification process converts NO₂⁻ and NO₃⁻ into gaseous N compounds that are nitric oxide (NO), N₂O and N₂. It involves heterotrophic microorganisms, whose activity is maximum in anoxic environment, in presence of NO₃ and C sources. The nitrifier denitrification can also lead to the production of NO, N₂O and N₂, from the nitrite produced during the nitrification.

The rate and fate of organic residues decomposition depend on the amount and nature of the residues, environmental factors and the nature of the microbial populations. These populations have a low C:N ratio, lower than that of crop residues. Since their N requirement during the decomposition process is rarely satisfied by the N mineralized from the residue, they take up soil mineral N, leading to N immobilization. The higher is the C:N ratio of organic residue, the greater is immobilization intensity. On the long term, decomposers die and part of their C and N is transformed into stable SOM (humification).

1.3.3 Carbon and nitrogen fluxes from cultivated field towards the environment

The C and N mineralized during decomposition of SOM may represent a source of pollution for the environment. Most of the C and N outputs from cultivated land occurs through the crop biomass exportation: grain for cereals, aboveground biomass for silage maize or leys cuts. Besides these exportations, different C and N losses may occur from agricultural lands: C and N leaching, gaseous emissions of CO₂, CH₄, N₂ and N₂O and volatilization of NH₃ (Figure 1.6), the relative proportions of these losses being site dependent.

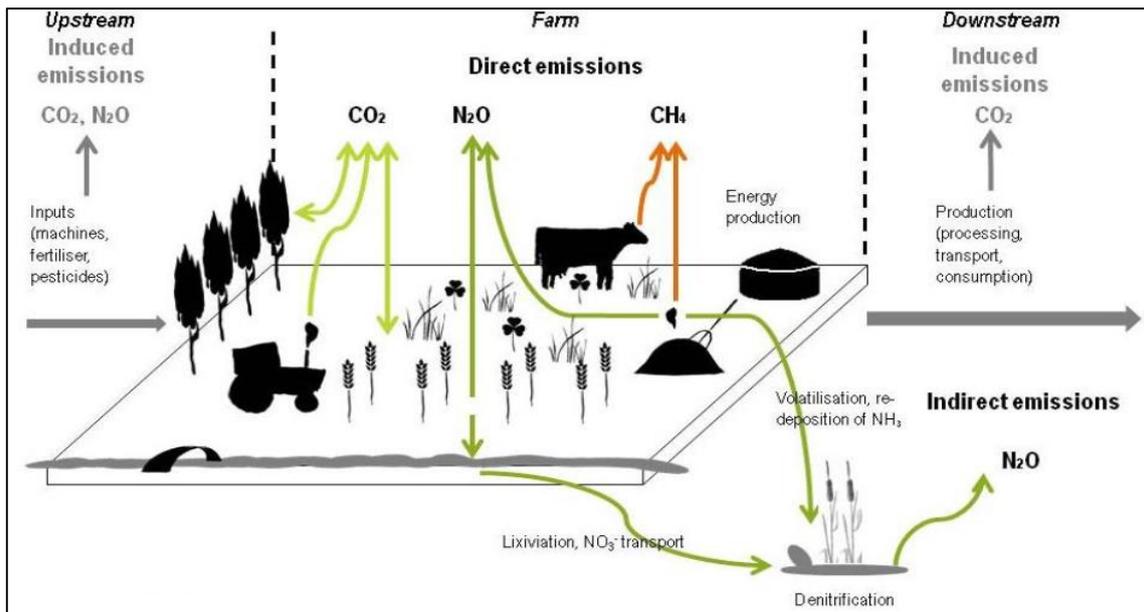


Figure 1.6. Direct, indirect and induced CO_2 , N_2O and CH_4 emissions occurring upstream and downstream agricultural systems (Pellerin *et al.*, 2013).

The mineral N leaching is the physical transfer of soluble mineral N, mainly in the form of NO_3^- , in soil solution to the surface or ground water. Leaching is conditioned by the intensity of water drainage and the concentration of nitrate in the soil, varying according to the net soil organic N mineralization of nitrogen of the soil and the mineral N fertilization. In Europe, it occurs mainly in winter time, during which evapotranspiration is minimal and thus drainage is highest. Leaching of dissolved organic C (DOC) and N (DON) can also occur but in a smaller range. In their review, Van Kessel *et al.* (2009) estimated DON losses at $12.7 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ in agricultural systems through runoff and erosion, while Nachimuthu & Hulugalle (2016) reported DOC losses between 0 and $1072 \text{ kg C ha}^{-1} \text{ yr}^{-1}$. The leached N can also be denitrified all along the N cascade from surface and ground waters (Galloway *et al.*, 2003; Butterbach-Bahl *et al.*, 2011). Gaseous N emissions into the atmosphere are caused by denitrification, leading to N_2 and N_2O emissions, mainly linked with N fertilization along with biotic and abiotic factors (Bessou *et al.*, 2011). Although N_2O losses represent a small percentage of total N losses, they have a huge impact on the total GHG balance, N_2O global warming potential (GWP) being 296 times greater than that of CO_2 for a 100-year time span. Finally, the volatilization of NH_3 is a physicochemical process that occurs in the presence of ammonium at the soil surface. It is favoured by low soil humidity, high pH and temperatures.

1.4 Carbon and nitrogen impacts of cropping systems on the long term

Evaluating the environmental sustainability of alternative agricultural systems is a key issue for their implementation with actors of agricultural activities (Tilman *et al.*, 2002). It requires a long term monitoring to consider slow processes affecting C and N dynamics (West & Post, 2002; Möller, 2009), themselves influenced by climate variations over years. Different agro-environmental indicators have been developed to assess environmental impacts of agricultural practices and predict the effects of agro-environmental policies (Langeveld *et al.*, 2007; Makowski *et al.*, 2009). For instance, calculating the rate of C storage in soil organic matter allows to approximate the potential mitigation of CO₂ emissions by a given cropping system. The N surplus, *i.e.* the difference between total N inputs and outputs, has been promoted as a proxy of potential losses from agricultural systems (van Beek *et al.*, 2003; Galloway *et al.*, 2003). At a more global scale, the GHG balance is an indicator of the contribution of agricultural practices to GHG emissions, by expressing the total emissions in a common unit (tons of CO₂ equivalent). This indicator encompasses both C and N impacts, including storage in soil organic matter and losses through leaching and gaseous emissions (N₂O, CH₄, CO₂...). Finally, beyond the use of indicators requiring measurements of several parameters, agro-environmental models also constitute a valuable tool to predict the long term performances of agricultural practices on C and N balances, in order to achieve more applied objectives related to cropping systems understanding and management (Brun *et al.*, 2006).

1.5 How do alternative cropping systems impact C and N cycles?

1.5.1 Diversification of crop rotations

Compared to conventional agriculture, the whole crop rotation is generally redesigned in alternative agricultural systems and characterized by longer successions, increased crop diversity and insertion of perennial crops, associated crops and/or catch crops. Longer and more diversified crop rotations may exert a positive impact on SOC stocks. McDaniel *et al.* (2014) found that adding one or more crops in rotation to a monoculture increased total soil C by 3.6% and total N by 5.3%. They attributed this to a higher production of microbial products often reported in these complex rotations. Crop rotations involving perennial forages tend to stabilise soil organic matter at a higher level than crop rotations involving fallow periods (Hansen *et al.*, 2000). Including perennials in cropping systems allows to decrease the risk of

N leaching (Crews & Peoples, 2005; Lemaire *et al.*, 2015). However, when returned to soil, legume perennials may result in important N mineralization enhancing NH_3 volatilization and N_2O emissions (Ball *et al.*, 2007; Askegaard *et al.*, 2011; Nadeem *et al.*, 2012) and leaching (Hansen *et al.*, 2000).

The association of crops within the same field, particularly the combination of cereal and legume, called “intercropping”, gives greater yields per unit area by making more efficient use of available nutrients (Lithourgidis *et al.*, 2011). Intercropping is recognized to increase SOM content (Duchene *et al.*, 2017). Cong *et al.* (2015) found a 4% and 11% increase in C and N content respectively after 7 years in an intercrop compared to a conventional system. The authors explain that the increased root biomass observed with associated crops could reduce N leaching across the soil profile by the complementarity in location and timing of N uptake between crop species. Other types of cropping systems not considered here are also possible tools to increase agriculture sustainability, such as agroforestry, *i.e.* the introduction of trees within cultivated fields, which rate of soil C storage can average $350 \text{ kg C ha}^{-1} \text{ yr}^{-1}$ over 0-100 cm (Cardinael *et al.*, 2015).

Introducing catch crops between two main crops has been shown to reduce N leaching (*e.g.* Constantin *et al.*, 2012; Tribouillois *et al.*, 2016). The trapping efficiency of the catch crop depends on the species: non legumes can reduce nitrate leaching up to 70% (Tonitto *et al.*, 2006), while legume catch crops are less efficient (Justes *et al.*, 2012; Valkama *et al.*, 2015). Furthermore, catch crops have been shown to sequester significant amounts of carbon in soils in spite of their relative small production. The review of Mary *et al.* (2012) gives a mean C storage rate of $290 \pm 150 \text{ kg C ha}^{-1} \text{ yr}^{-1}$ under temperate climate (average of 16 studies and 15 years). In their meta-analysis, Poeplau & Don (2015) report a similar rate: $320 \pm 80 \text{ kg C ha}^{-1} \text{ yr}^{-1}$. The global effect of catch crops on N_2O emissions is not yet established since lower emissions are recorded during catch crop growth (Basche *et al.*, 2014) but higher fluxes are observed after destruction (Kaye & Quemada, 2017).

Cover crops, *i.e.* auxiliary crops which grow beneath the main crop, allow a permanent soil coverage, limiting soil erosion and weed spreading, and often improve soil fertility and SOC content. In addition, belowground C inputs contributes more effectively to the relatively stable C pool than an equivalent amount of above ground C-input (Rasse *et al.*, 2005; Kätterer *et al.*, 2011). Particularly, the cultivation of deep rooting crops would promote the amount of carbon stored in the deep soil layers, although root C fluxes to soil are poorly understood because of

high uncertainties on total root biomass and associated exudation (Rumpel & Kögel-Knabner, 2011).

1.5.2 Crop fertilization

Crop fertilization is often modified in alternative cropping systems, mineral fertilizers being reduced or replaced by organic fertilizers and/or reactive N produced by biological N fixation. Splitting mineral N fertilization in several applications has been fostered in order to better match mineral N availability and crop N demand. It can increase fertilizer efficiency without decreasing crop yields for a variety of crops in diverse areas (Robertson & Vitousek, 2009). While allowing a better uptake of nutrient by crops, its impact on N leaching seems limited (Zebarth *et al.*, 2009). It may contribute to reduce NH₃ and N₂O emissions (Sitthaphanit *et al.*, 2009; Liu *et al.*, 2016). Another alternative practice is the reduction of the total amount of fertilizer applied compared to conventional rate, but a moderate reduction in N rate has little effect on N leaching on short term while it may have a stronger impact on long term (Beaudoin *et al.*, 2005; Constantin *et al.*, 2010). Its effect on SOC storage is debated: Khan *et al.* (2007) reported a negative effect, while others reported an increase in SOC from increased biomass returned to the soil after harvest (Jarecki & Lal, 2003), particularly with a higher belowground biomass in agricultural systems (Lu *et al.*, 2011).

The use of external organic fertilizers (*e.g.* manure, slurry, sewage sludge) is common in alternative cropping systems. Unlike mineral fertilizers, nutrients in organic fertilizers are not immediately available for the plant, as their decomposition spreads over time, with possible phases of N immobilization, varying according to the environmental conditions (Seufert *et al.*, 2012), resulting in temporal mismatches between nutrient availability and crop demand (Pang & Letey, 2000). Thereby, nutrient surplus, not assimilated by plants, may trigger N losses. Simultaneously, regular manure application has been demonstrated to maintain (Schulz *et al.*, 2014) or increase SOC stocks (Gattinger *et al.*, 2012; Powlson *et al.*, 2012; Maltas *et al.*, 2013). In their meta-analysis, Maillard & Angers (2014) estimated a relative SOC stock change factor of 1.26 ± 0.14 related to long term cumulative manure application (> 20 yr), compared to systems receiving mineral fertilization.

The inclusion of legumes as a green manure in alternative cropping systems has also been advocated for replacing chemical N fertilizers, supplying easily absorbable N for the subsequent crop (Cherr *et al.*, 2006; Fustec *et al.*, 2010). One main feature of the returned legumes residues

is its rather low C:N ratio, leading to a short immobilization phase and a subsequent release of mineral N (Bolger *et al.*, 2003; Fornara *et al.*, 2009). Fertilizer use efficiency was increased when legumes were used as green manure, with reduced N losses compared to synthetic fertilizers (Robertson & Vitousek, 2009; Ponisio *et al.*, 2015). However, soil incorporation of legume residues has been shown to increase the risk of N₂O emissions; harvesting the legume residues and recycling them into biogas may be a way to reduce gaseous emissions (Stinner *et al.*, 2008).

1.5.3 Reduced tillage

Reduced tillage means either reduction of tillage operations or decrease in tillage intensity (lower tilled depth, suppression of full inversion tillage) and may reach the total suppression of tillage except at sowing (“no-tillage”). No-tillage has been adopted in some alternative cropping systems in order to minimize soil disturbance, enhance soil aggregation and water infiltration (Guo *et al.*, 2016). A modification of the behaviour of the crop residues, left as a mulch on the top soil layer, is proposed by some authors, with modified physical and chemical conditions of their decomposition or humus mineralization (Oorts *et al.*, 2007). Furthermore, no-tillage has been promoted as a mitigation practice able to increase SOC storage, particularly during the previous decade (*e.g.* West & Post, 2002; Lal, 2004). However, this conclusion has been questioned during the last years because of methodological issues. Hence, it has been shown that shallow soil sampling could over-estimate the SOC stocks since reduced till redistributes C in soil profile, with higher C content in the top soil (0-10 cm) and a lower content below 15 cm compared to full inversion tillage (Baker *et al.*, 2007; Angers & Eriksen-Hamel, 2008; Luo *et al.*, 2010b). Moreover, the lack of diachronic comparisons and/or initial soil characterization has been pointed out (Neto *et al.*, 2010; Costa Junior *et al.*, 2013). In their meta-analysis, Luo *et al.* (2010b) found on average no effect of no-tillage on total SOC stocks over 30 cm. This was confirmed by Dimassi *et al.* (2013, 2014) in two long-term experiments at Boigneville in France. The impact of no-tillage on GHG gases has also been investigated, but with inconsistency in results, related with the lack of long-term observations and the high temporal and spatial variability in N₂O and CH₄ emissions (Palm *et al.*, 2014).

1.6 Research questions and purposes of the thesis

The state of art indicated that numerous studies evaluated the short term C and N impact of alternative cropping practices. However, few of them compared the effect of all these practices together in more complex systems and on the long term (Arrouays, 2002; Leifeld *et al.*, 2009). Such studies are essential to strengthen the understanding of the impact of alternative agro-ecosystems on slow processes affecting C and N dynamics in the long term (West & Post, 2002; Möller, 2009), themselves influenced by climate variations over years.

In this framework, the ENBIO research project (experimental agri-ENvironmental evaluation of agri-BIOlogical cropping systems) was set up in 2013 and led by Nicolas Beaudoin (INRA, AgroImpact) and coordinated by “Agro Transfert Ressources et Territoires”. The project was shared between i) the study of the spatial variability of production, water drainage and leaching in alternative systems; ii) the study of the temporal variability of water, carbon and nitrogen budgets in alternative systems and iii) the calibration of the STICS model in organic cropping systems. The two latter topics are addressed in this thesis, with the objective of answering the following questions:

- Do alternative cropping systems modify N surplus and the fate of this surplus in soil, water and atmosphere?
- What can explain extra C storage in alternative cropping systems: changes in soil tillage or auxiliary legume crops?
- Are the soil C and N mineralization rates modified by alternative cropping practices?
- Do alternative cropping systems have better GHG balance than conventional systems, when expressed per unit of area, production or N exported?
- Does the STICS model allow to simulate the C and N dynamics in alternative cropping systems?

Research hypotheses are specific to each type of alternative system studied and were therefore formulated in each chapter. The two main purposes of this work were as follows:

- evaluating experimentally the main C and N fluxes of alternative cropping systems, in comparison with conventional systems;
- modelling these cropping systems with the agro-environmental model STICS, in order to predict their long-term effect on C and N fluxes.

To a broader scale, this work aims at specifying the mitigation potential of alternative agriculture in the context of climate change.

1.7 Organisation of the document

The first chapter presents the general framework of the thesis. Chapter 2 focuses on the effect of alternative cropping systems on SOC storage in one long-term experiment, while their impact on C and N mineralization is presented in the third chapter. The N fate and GHG balance of these systems are described in the fourth chapter. The fifth chapter presents the modelling of C and N fluxes in two other long term experiments including organic farming. The last chapter concludes on results and perspectives for the future. Chapters 2-5 are written in the form of scientific papers (published or in preparation). The structure of the manuscript is summarised in Figure 1.7.

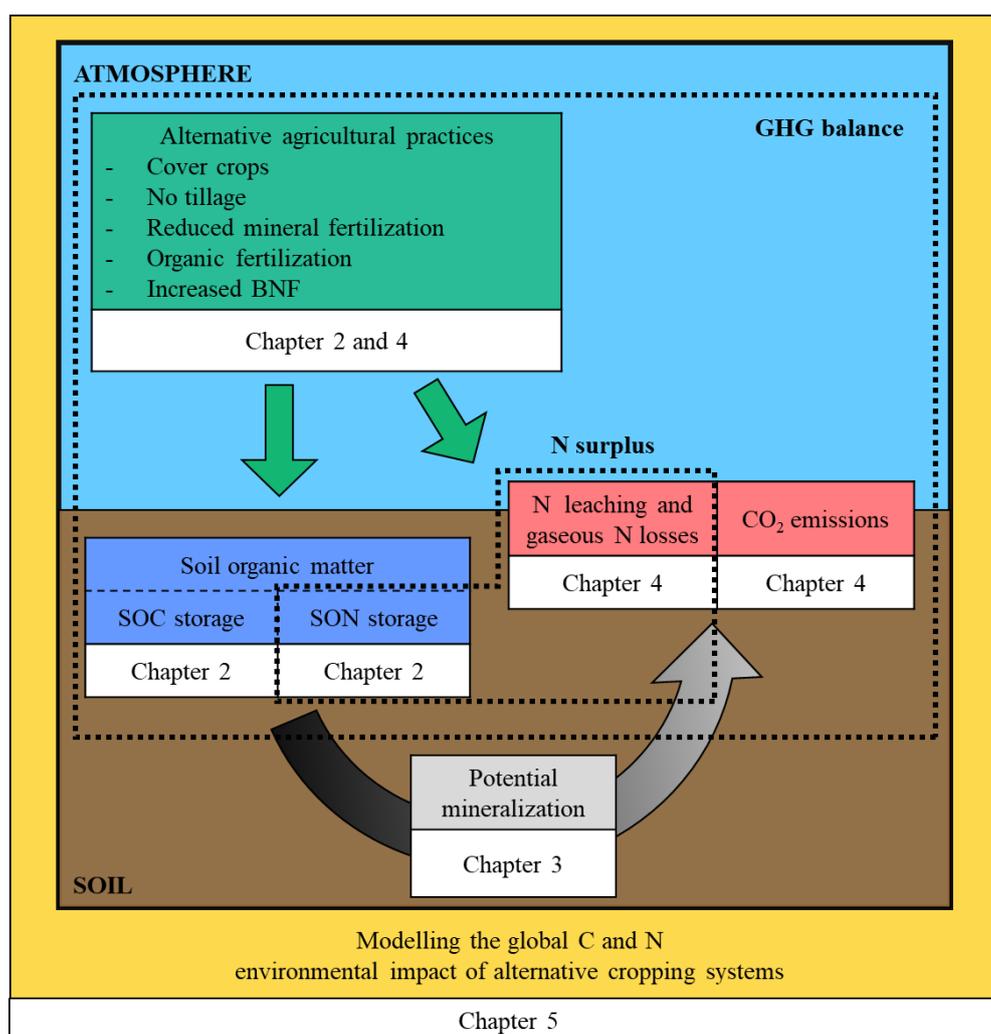


Figure 1.7. Graphical summary of the thesis chapters.

1.8 Experiments

Four criteria were retained to select the field experiments: i) arable cropping systems (with no or moderate organic fertilizer); ii) frequent measurements of C and N related fluxes (SOC and SON stocks, SWC and SMN contents, ...); iii) treatments including low input and organic systems and iv) long term experiments allowing to account for climate variability, cumulative effects and slow processes (C and N storage). Three European field experiments were selected in the Northern part of Europe (Figure 1.8):

- at Therwil (Switzerland) for the “DOK” experiment, started in 1978 and managed by the FiBL and Agroscope (Zürich);
- at Foulum (Denmark) for the “Foulum organic” experiment, started in 1997 and managed by the Foulum Aarhus University;
- at Versailles (France) for “La Cage” experiment, started in 1998 and managed by the “UMR Agronomie” of INRA-AgroParisTech (Thiverval-Grignon).

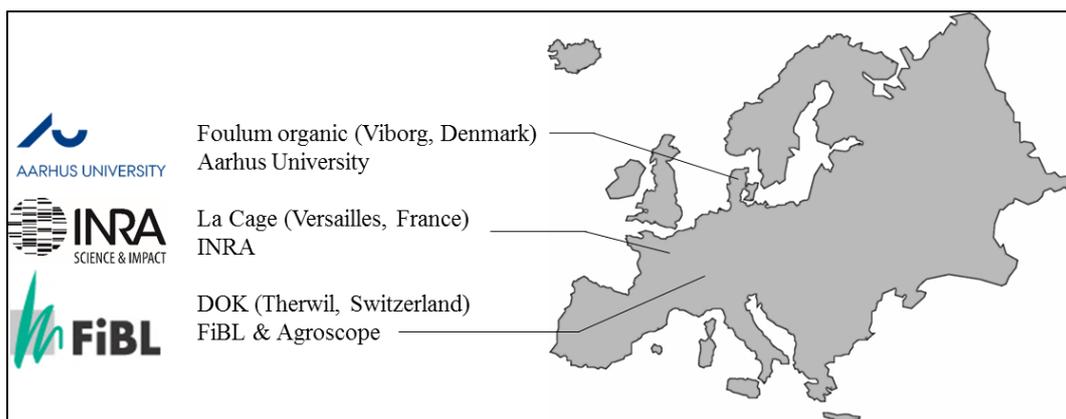


Figure 1.8. Localisation of the three European experimental sites.

These experiments compare the effects of alternative agricultural systems, such as organic farming, conservation agriculture or low inputs cropping systems, on crop production, nitrate pollution and carbon storage, in varied pedo-climatic situations (Table 1.2). Each has its own particular characteristics for the crop rotation, the type of fertilization, the doses of organic fertilizer and N applied.

Table 1.2. Main features of the three experiments studied.

Experiment	Age (starting year)	Soil type	Annual rainfall mm	Mean temperature °C	Cropping systems
DOK (Switzerland)	39 (1978)	Haplic Luvisol	872	9.5	Conventional, unfertilized, organic and biodynamic
Foulum (Denmark)	20 (1997)	Typic Hapludult	626	7.3	Conventional and organic
La Cage (France)	19 (1998)	Luvisol	675	11.0	Conventional, integrated, organic and conservation agriculture

1.9 Methodological approach

The strategy consisted in coupling *in situ*, *in vitro* and *in silico* experiments.

For the *in situ* experiment, we added a specific follow-up in the long term experiment of La Cage from 2014 to 2017, in order to measure SOC and SON changes, NO₃ and N₂O losses. We measured SOC and SON contents in 2014, soil mineral nitrogen and water contents three times a year from 2013 to 2017 and made a continuous monitoring of greenhouse gas emissions (CO₂, N₂O) from April 2014 to July 2017. We selected agri-environmental indicators to characterize the performances of the alternative practices compared to conventional agriculture. We used both a diachronic approach to compare the evolution of SOC stocks over time, and a synchronic approach to compare cropping systems at a given date. We calculated the N surplus, N leaching and gaseous losses and the total GHG balance in each of the four systems.

The *in vitro* experiment consisted in soil incubation performed on soils taken in La Cage, in order to test whether the potential C and N mineralization rate differed or not between systems. Results from incubations were compared to mineralization rate estimated by modelling with AMG model (Saffih-Hdadi & Mary, 2008).

The *in silico* experiment was performed with a dynamic model, a useful tool for evaluating the long term effects of alternative cropping systems on water, C and N balances. Various models

have already been evaluated and validated for predicting the long-term turnover of soil organic matter in various pedo-climatic conditions, mainly for conventional cropping systems. Modelling C and N dynamics in alternative cropping systems on the long-term has received much less attention. We used the STICS model (Brisson *et al.*, 1998, 2002) to simulate crop production, C and N storage and N surplus in the DOK and the Foulum experiments.

CHAPTER

2.

Alternative arable cropping systems: a key to increase soil organic carbon storage? Results from a 16 year field experiment

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Highlights

- ▶ SOC stocks were investigated down to 30 cm in 4 cropping systems of a 16-yr trial.
- ▶ No temporal change of SOC stocks was observed in conventional and low input systems.
- ▶ SOC stocks increased by 12% in the organic and 24% in the conservation systems.
- ▶ Modelling showed that C inputs from cover crops could explain SOC storage.
- ▶ The simulated SOC mineralization did not differ between systems including no-till.

Abstract

Alternative cropping systems such as conservation agriculture and organic farming are expected to decrease negative impacts of conventional systems through sequestration of organic carbon in soil and mitigation of greenhouse gas emissions. We studied soil organic carbon (SOC) dynamics in the long-term (16 years) field experiment “La Cage” (France) which compares four arable cropping systems, free from manure application, under conventional (CON), low input (LI), conservation agriculture (CA) and organic (ORG) management. Bulk densities and SOC concentrations were measured at different dates between 1998 and 2014. SOC stocks were calculated at equivalent soil mass taking into account bulk density variations and SOC redistribution across the different soil layers. We analyzed the evolution of SOC stocks and compared it with outputs of the simulation model AMG. The rate of change in SOC stocks in the old ploughed layer (ca. 0-30 cm) during the 16 years was 0.08, 0.02, 0.63 and 0.28 t ha⁻¹ yr⁻¹ in the CON, LI, CA and ORG systems respectively and significantly differed from 0 in the CA and ORG treatments. The AMG model satisfactorily reproduced the observed evolution of SOC stocks in the old ploughed layer in all treatments. A Bayesian optimization procedure was used to assess the mean and the distribution of the most uncertain parameters: the SOC mineralization rate and the C inputs derived from belowground biomass of cover crops which were fescue (*Festuca rubra*) and alfalfa (*Medicago sativa*). The model thus parameterized was able to predict SOC evolution in each block and soil layer (0-10, 10-20 and 20-30 cm). There was no significant difference in SOC mineralization rates between all cropping systems including CA under no-till. In particular, the increased SOC storage in CA was explained by higher carbon inputs compared to the other cropping systems (+1.72 t C ha⁻¹ yr⁻¹ on average). The CA and ORG systems were less productive than the CON and LI systems but the smaller C inputs derived from cash crop residues were compensated by the extra inputs from additional crops (fescue and alfalfa) specifically grown in CA and ORG, resulting in a positive carbon storage in soil. We conclude that alternative arable systems have potential to sequester organic carbon in temperate climate conditions, through higher carbon input rather than by the effect of reduced soil tillage.

2.1 Introduction

Soil is one of the major components of the biosphere, delivering various essential ecosystems services. It constitutes the main terrestrial carbon sink, containing 1500 Gt of carbon across one m depth (Batjes, 1996). Farming practices impact this compartment through modification of carbon inputs coming from crop residues or organic fertilizers and indirectly by affecting soil organic carbon (SOC) turnover through soil disturbance. Optimized farming practices with high organic inputs, permanent plant cover and reduced soil tillage can play an essential role in soil carbon sequestration, defined by difference with a reference cropping system (*e.g.* Luo *et al.*, 2010a), and thus in mitigating climate changes (West & Post, 2002; Freibauer *et al.*, 2004; Powlson *et al.*, 2011). Combining these practices can generate alternative cropping systems differing from the dominant paradigm of conventional systems as they share similar inspirations such as sustainable development of agriculture with the improvement of environmental performance (Beus & Dunlap, 1990).

During the last twenty years, alternative cropping systems have been tested including some which may be less profitable for farmers (Eltun *et al.*, 2002). Conservation, organic and integrated agriculture are examples of alternative systems with expected environmental benefits, including a greater soil organic carbon sequestration, depending on the implemented practices. Conservation agriculture is characterized by the suppression of soil tillage, more diversified crop successions and permanent plant cover. No-tillage systems are often included in this category (Corsi *et al.*, 2012), although they often do not fulfill the last two criteria. Another alternative cropping system is organic agriculture which aims at minimizing its impact on soil, water and air quality. Systemic prevention of weeds, pests and diseases, combined with nutrient self-sufficiency is the core of sustainable organic production (Lammerts van Bueren *et al.*, 2002) since external inputs should be limited (Watson *et al.*, 2002a). In such a farming system, crop production is mainly based on organic fertilizers (*i.e.* manure, compost), green manures and frequent tillage most often essential to control weeds. Low input system, also known as integrated system, combines some practices applied in organic or conservation systems, as it promotes natural regulation in the farming system in order to limit the use of external inputs and sustain farm income (Eltiti, 1992). Overall, reduced intensity in soil tillage, reduced and better adjusted fertilization, increased frequency of cover crops and weaker use of pesticides are the main features that distinguish alternative from conventional system.

Existing reviews on SOC storage in alternative *vs.* conventional systems report contradictory results. They can arise from the difficulty of fulfilling all methodological requirements such as

measurements of the initial state, measurements of C concentration and bulk density at a sufficient depth (at least 0-30 cm in order to include variation of the ploughing depth in the time) in order to calculate SOC stocks at equivalent soil mass between different dates. Higher SOC stocks were recorded in some studies dealing with cropping systems similar to conservation agriculture in which ploughing was stopped and the number of crops increased in the rotation for a same duration (West & Post, 2002; Calegari *et al.*, 2008). However, recent meta-analyses selecting studies conducted with an adequate methodology revealed that SOC sequestration potential in no-till systems had been over-estimated (LUA A VIRTO 2011). Concerning organic cropping systems, several studies agreed on their ability to store more SOC than conventional ones (Mondelaers *et al.*, 2009; Leifeld & Fuhrer, 2010; Gomiero *et al.*, 2011; Tuomisto *et al.*, 2012). These authors mainly attributed the extra C storage to a greater application of livestock manure in the organic systems. However, Leifeld *et al.* (2013) indicated that the proportion of conventional and organic systems in the meta-analysis of Gattinger *et al.* (2012) was unbalanced in terms of systems with external carbon inputs (27% and 92% respectively), leading to a misinterpretation. Since organic fertilizer (including manure) addition rate is a major driver of SOC sequestration, its uneven distribution makes the comparison between organic and conventional systems difficult and hampers the identification of possible other drivers, such as crop rotation and nature of carbon inputs (Leifeld *et al.*, 2009). Finally, the number of experiments comparing conventional and alternative arable systems without livestock manure is scarce.

Here, we studied a long term experiment (16-yr) including four purely arable cropping systems without manure fertilization. Our objectives were to: i) compare SOC stocks in these systems; ii) predict the dynamics of SOC stocks with a simulation model and iii) understand the drivers of C storage with the help of modelling. The evolution of SOC stocks between 1998 and 2014 was simulated using the simple AMG model (Saffih-Hdadi & Mary, 2008). We tested two hypotheses: i) SOC stocks can evolve differently due to variations in carbon inputs between cropping systems and ii) the mineralization rate of SOC is unaffected by the type of cropping system.

Table 2.1. Physical and chemical properties of the soil at “La Cage” (layer 0-25 cm) measured at the start of experiment in 1998.

Cropping system	Block	Clay	Fine silt	Coarse silt	Fine sand	Coarse sand	Org. C	Total N	CaCO ₃	pH _{H2O}	CEC	
		<2 µm	2-20	20-50	50-200	200-2000						
							(g kg ⁻¹)					(cmol+ kg ⁻¹)
CON	1	184	175	413	205	25	9.90	1.01	2.50	7.55	12.35	
	2	171	202	408	195	25	9.30	0.92	0.83	7.40	11.55	
LI	1	153	178	329	291	49	11.55	1.18	0.67	7.45	12.60	
	2	165	197	432	184	23	9.15	0.93	0.50	7.05	10.10	
CA	1	150	173	303	312	64	11.05	1.12	0.83	7.35	11.30	
	2	174	186	404	213	25	9.55	0.97	0.83	7.35	11.15	
ORG	1	177	181	411	208	24	9.45	0.94	0.33	7.35	11.50	
	2	161	165	342	282	51	8.90	0.90	0.67	7.50	11.60	

CON = conventional, LI = low input, CA = conservation agriculture, ORG = organic farming.

2.2 Materials and methods

2.2.1 Site and soil characteristics

The study was conducted at the long-term experimental site of “La Cage”, Versailles, France (48°48' N, 2°08' E) established in 1998 by INRA. Before 1998, the whole site was conducted under a conventional management. The purpose of the experiment is to evaluate the agronomic, economic and environmental performances of three alternative systems compared to a conventional cropping system which is representative of arable farming in Northern France. During the studied period (1998-2014), the mean annual temperature, precipitation and potential evapotranspiration were 11.3°C, 627 and 673 mm respectively. The soil is a well-drained deep Luvisol (IUSS Working Group WRB, 2006) (Table 2.1). Minimum and maximum clay content varies between 150 and 184 g kg⁻¹, with a mean value of 167 g kg⁻¹ over the whole field. In 1998, at the start of the experiment, the ploughed layer (0-25 cm) had a mean organic C content of 9.49 g kg⁻¹, a C:N ratio of 9.6 and a pH of 7.38.

2.2.2 Cropping systems

Four cropping systems are compared: a conventional (CON), a low input (LI), a conservation agriculture (CA) (direct seeding with permanent plant cover called cover crop) and an organic farming (ORG) system. The experimental site is divided in two blocks. Each block consists of four plots, each plot corresponding to one cropping system. The plots are divided into two subplots of 0.56 ha, each of them supporting a different crop of the rotation. Wheat is grown every year in one of the two subplots. A detailed presentation of crop rotations, soil management and fertilization in each treatment is given in Table 2.2. Each cropping system had its own management specificity:

1) Tillage. Ploughing occurred every year in CON and ORG, except after pea crops, and one out of two years in LI, before pea and rapeseed. No tillage was done in CA.

2) Fertilization. N fertilization varied every year according to crop and system. The mean amount of mineral N fertilizer applied over the 16 year period was 143, 114, 104 and 0 kg N ha⁻¹ yr⁻¹ for CON, LI, CA and ORG, respectively. No manure or external organic fertilizer was

Table 2.2. Crop rotations, soil tillage and nitrogen fertilization management at “La Cage” over the period 1998-2014.

Management		CON	LI	CA	ORG
Crop rotation	1998	pea	pea	pea (2)	wheat
	1999	wheat	wheat	wheat : fescue	rapeseed
	2000	rapeseed	rapeseed	corn : fescue	wheat
	2001	wheat	wheat	wheat : fescue (2)	pea
	2002	pea	pea	pea : fescue (2)	wheat
	2003	wheat	wheat	wheat (3)	alfalfa
	2004	rapeseed	rapeseed	corn (4)	alfalfa
	2005	wheat	wheat	wheat	wheat
	2006	pea	pea	pea	rapeseed
	2007	wheat	wheat	wheat	wheat
	2008	rapeseed	rapeseed	wheat : alfalfa	wheat
	2009	wheat	wheat	alfalfa	alfalfa
	2010	pea	pea	wheat : alfalfa	alfalfa
	2011	wheat	wheat	alfalfa : fescue	wheat
	2012	rapeseed (1)	rapeseed (1)	oat : alfalfa	wheat
2013	wheat	wheat	alfalfa	alfalfa	
2014	pea	pea	wheat : alfalfa	alfalfa	
Ploughing		each year	every 2 years	no till	each year
Nitrogen fertilization (kg N ha ⁻¹ yr ⁻¹)		143	114	104	10
Yield (t ha ⁻¹ yr ⁻¹)*	wheat	9.7	8.9	6.7	5.4
	pea	4.2	4.5	3.7	2.6
	rapeseed	4.5	3.8	-	0.8

* Grain yields is given for comparison, at 15% moisture content. When grown with a cover crop, main crop is followed by ":" and the name of the cover crop. Catch crop following a main crop is in brackets (1 = white mustard, 2 = oat and vetch, 3 = clover, 4 = oat). CON = conventional, LI = low input, CA = conservation agriculture, ORG = organic farming.

applied in the experiment, except in ORG in the first cropping period before 2009, during which guano and feather meal were used which represented $10 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ on average. P and K were provided to all cropping systems except ORG through the application of mineral fertilizers.

3) Crop protection. A systematic use of pesticides was done in CON in order to avoid any yield limiting factor, whereas their application was made in LI and CA only when the damage threshold was exceeded. No pesticides were used in ORG, according to the European specifications for organic farming.

4) Rotation. A four year rotation was applied in the CON and LI systems during the whole period, 1998-2014: rapeseed (*Brassica napus L.*), winter wheat (*Triticum aestivum L.*), spring pea (*Pisum sativum L.*) and winter wheat. The rotation was modified in the CA and ORG systems because of a progress strategy and the integration of technical innovation. Additional main crops differed according to the treatment: maize (*Zea mays L.*) grown two years in CA instead of rapeseed, and alfalfa (*Medicago sativa*) in CA and ORG.

5) Crop residue management. Main crops were harvested for grain at maturity and crop residues were left at soil surface for all of the treatments. Alfalfa was cut three to four times per year in ORG and CA and left on soil surface as a green manure except for the first cut which was removed from the plots in ORG. Catch crops, grown during fall and winter between two main crops in order to avoid nitrate losses, were oat (*Avena sativa L.*), vetch (*Vicia sativa L.*), white mustard (*Sinapis alba L.*) and fodder raddish (*Raphanus sativus L.*). Cover crops, grown only in CA under the main crop in order to protect the soil, were alfalfa and fescue (*Festuca rubra*). Cover and catch crops grown in CA were chemically destroyed or rolled before seeding the cash crop.

The frequency of occurrence of the various crops (main crops, catch crops and cover crops) from 1998 to 2014 varied among the four cropping systems (Fig. 2.1). Winter wheat frequency was about 50% in each cropping system in order to be representative of regional practices. Winter rapeseed and spring pea were less represented in CA and ORG than in CON and LI systems, but CA and ORG included a significant proportion of alfalfa (18% and 34% respectively) which did not appear in the other systems. The specificity of the CA system was its higher frequency of catch crops and above all the presence of a cover crop.

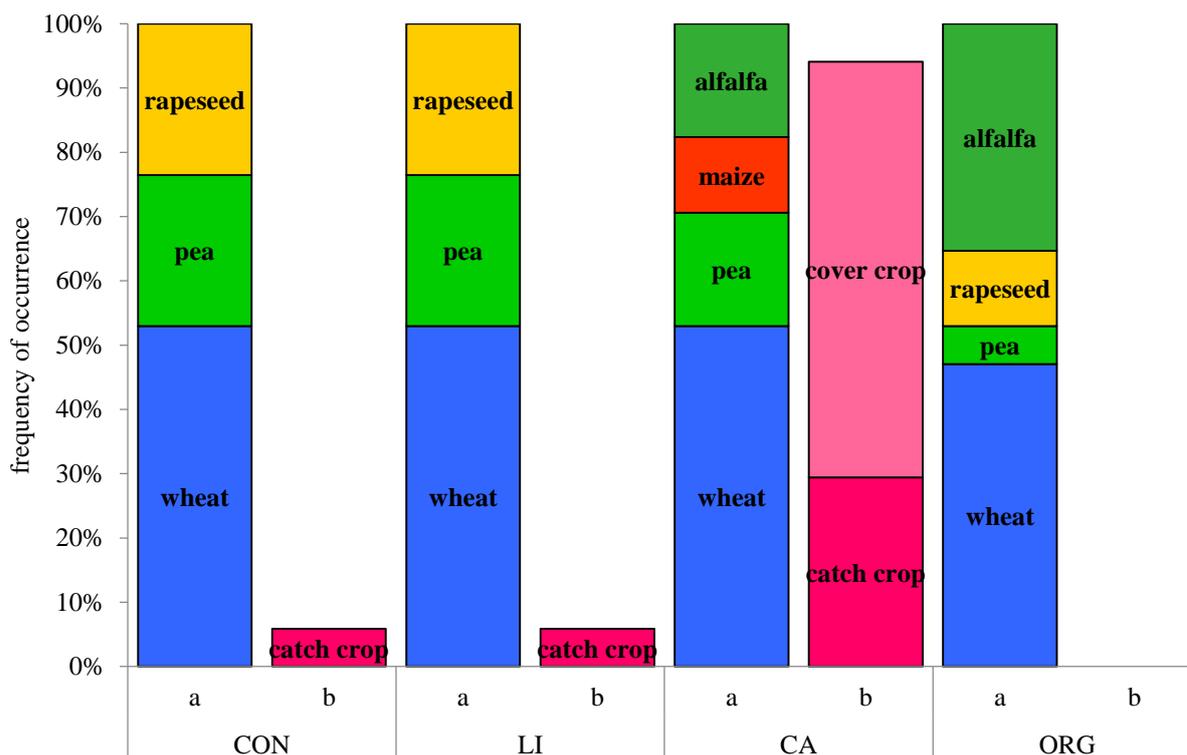


Figure 2.1. Cumulative frequencies of occurrence of a) main crops and b) auxiliary crops in the 4 cropping systems (mean values over the period 1998-2014). CON = conventional, LI = low input, CA = conservation agriculture, ORG = organic farming.

2.2.3 Crop yields and residue biomass

Crop yields were determined every year from 1998 to 2014 based on grain collected by the combine harvester. The biomass of aboveground (AG) residues of the main crops returned to soil was estimated using the harvest index of each crop which was 0.54, 0.60, 0.31 and 0.46 for wheat, pea, rapeseed and maize respectively (Dubrulle *et al.*, 2004). The AG biomass of catch crops and cover crops totally returned to soil was not measured but estimated using crop growth allometric equations (see Appendix 1.A). It was assessed using a relationship based on the thermal time, *i.e.* the cumulative temperature above the crop base temperature. In the case of alfalfa as main crop, the exported cut was measured at mowing time and the cuttings left on the soil were estimated using regional references. When alfalfa was grown as cover crop, we used relationships depending on the date of cutting and regrowth: one for the period of establishment after seeding at the end of summer, one for autumn regrowth and the last for spring and summer regrowth.

Belowground (BG) biomass was not quantified in the experiment. BG biomass of main crops was assumed to be independent of AG biomass and calculated using references from Dubrulle

et al. (2004): it was 2.33, 2.33, 0.98 and 2.24 t DM ha⁻¹ for wheat, rapeseed, pea and maize respectively. In the case of catch crops and cover crops, we assumed that their BG biomass was proportional to their AG biomass because they derived from younger, unripe plants. The BG/AG ratio was set at 1.6 for fescue (Vertès *et al.*, 2002), 0.6 for alfalfa (Thiébeau *et al.*, 2011) and 0.7 for catch crops (Constantin *et al.*, 2010). The conversion of dry mass to C content was made by assuming a 42% and 38% carbon content in the AG and BG residues, respectively (Justes *et al.*, 2009).

2.2.4 Soil sampling and analysis

The soil sampling strategy was designed to calculate SOC stocks on an equivalent soil mass (ESM) basis (Ellert & Bettany, 1995) over a depth at least equal to the deepest tillage event. The ploughing depth was ca. 30 cm before 1998 and shallower afterwards, about 25 cm. The SOC measurements were carried out at the experimental site on both blocks in 1998 and 2014, and only in block 2 in 2000, 2003 and 2011. In February 1998, twenty soil samples were taken in each plot over 30 cm depth. In May 1998, March 2000, March 2003 and March 2011, six soil samples per plot were taken in block 2 down to 30 cm in plots where wheat was grown. In April 2014, six soil cores per plot were taken in both blocks down to 60 cm using a hydraulic gauge of 6 cm diameter. A single soil layer (0-30 cm) was analysed for samples taken in February 1998, March 2000, March 2003 and March 2011. The soil cores were divided in 3 layers in May 1998 (0-10, 10-20 and 20-30 cm), and 5 layers in April 2014 (0-10, 10-25, 25-30, 30-35 and 35-60 cm). Soil was homogenized, coarse residues (> 2 mm) and visible roots were removed by hand picking. Soil samples were oven dried for 48h at 35°C and sieved (2 mm). A soil subsample of 20 g was finely ground in a ball mill (PM 400, Retsch, Germany) and an aliquot taken for carbon analysis. The Dumas method was used for carbon analysis using an elemental analyzer (EURO EA, Eurovector, Italy). The CaCO₃ content was measured by acid decarbonation (NF ISO 10693). Inorganic C represented on average 0.08 g C kg⁻¹ (Table 2.1) and was subtracted from total C to obtain the organic C.

Bulk density was measured for three layers (0-10, 10-20 and 20-30 cm) simultaneously with soil sampling in 1998, 2000, 2003 and 2011 using a steel cylinder of 98 cm³ inserted vertically in the soil. Soil was weighed after drying during 48 h at 105 °C. The same method was used to determine the bulk density on the 0-5 cm layer in 2014. A second method was used in 2014 to measure the bulk density every 5 cm in the layers between 5 and 40 cm with a gamma-densitometer (LPC-INRA, Angers, France).

Coarse particulate organic matter (cPOM) was determined in soil samples taken in 2003 and 2014 by particle size separation. A sample of 50 g of 2 mm sieved and air dried soil was dispersed under water on a 200 μm sieve. Coarser particles (200-2000 μm) were washed out in a bucket, floating particles (cPOM) collected and oven dried at 60°C before being crushed and analysed for C concentration.

2.2.5 Calculations of soil mass and SOC stock

SOC stocks were calculated on ESM basis at different depths, particularly over the old ploughing depth, using measurement of bulk densities and organic C concentrations. To facilitate calculations, the soil was discretized into elementary layers of 1 mm thickness. The soil mass at a fixed depth z (in mm), $M(z)$ (in t ha^{-1}), can be calculated as the sum of soil masses of z elementary layers, as follows:

$$M(z) = 10 \sum_{k=1}^z \rho(k) \quad (2.1)$$

where $\rho(k)$ is the bulk density of the elementary layer k (g cm^{-3}), k varying from 1 to 600 mm. A reference soil mass M_R (in t ha^{-1}) was considered, corresponding to the old ploughing depth of the CON system (30 cm) which was estimated in 1998 at 4300 t ha^{-1} . For the subsequent years, the z value corresponding to M_R was determined by fulfilling the equation: $M(z) = M_R$. We also considered three other soil mass references in order to analyse the SOC evolution in the soil profile: L1 (*ca.* 0-10 cm) and L2 (*ca.* 10-20 cm) with a fixed mass of 1300 t ha^{-1} for each layer, L3 (*ca.* 20-30 cm) with a fixed mass of 1700 t ha^{-1} of soil, L4 (*ca.* 30-40 cm) with a fixed mass of 1400 t ha^{-1} and L5 (*ca.* 40-60 cm) with a fixed mass of 2800 t ha^{-1} .

The cumulative SOC stock $QC(z)$ (in t ha^{-1}) in the layer 0- z is:

$$QC(z) = 0.01 \sum_{k=1}^z \rho(k) \cdot C(k) \quad (2.2)$$

where $C(k)$ is the SOC concentration in the elementary layer k (g kg^{-1} dry soil). Since the measured values of bulk densities and SOC concentrations refer to macro-layers (L1 to L5), $\rho(k)$ and $C(k)$ were supposed to be equal to their respective values in these macro-layers.

2.2.6 Statistical analysis

Statistical analyses were performed using the R software (R Core Team, 2010). Since the number of true replicates in the experiment was low (two randomized blocks), each of the two subplots (not randomized) was considered as replicate thus producing four pseudo replicates. This choice resulted from the weakness of the experimental design, which forces us to be

conservative with our results as explained by (Hurlbert, 1984). (Henneron *et al.*, 2014) and (Pelosi *et al.*, 2015), analysing the soil organisms on the same site, have described the rationale supporting this choice: i) the entire experiment had the same crop management before 1998, ii) soil sampling was done in large plots (0.56 ha) and samples were taken far enough from each other to be considered as independent, and iii) the pre-existing topographic and pedological gradients were controlled by blocking. Indeed, our measurements relative to SOC concentrations and stocks made in 1998 show that the intra-plot variability (between subplots) was as important as the inter-plot variability (within blocks), as indicated by the comparison of variances ($F=1.83$, $p<0.05$). Furthermore, our objective was to compare not only SOC stocks at given dates but also the temporal variations of SOC stocks between cropping systems. These variations, calculated as the difference between final and initial SOC stocks measured in each subplot, can be considered as true replicates, if we assume that possible interactions between the effect of cropping systems and the initial SOC stocks were of second order of magnitude.

Analyses of variance (ANOVA) were performed on measurements made in 1998, 2000 and 2014 to test the effect of cropping system on SOC stocks for all layers L1, L2, L3 and L1-3, and only on L1-3 for 1999, 2001, 2003 and 2011. A separate ANOVA was done to compare the SOC concentrations and stocks of 1998 and 2014 for each treatment and the change in SOC stocks between 1998 and 2014 for each treatment. The assumptions of ANOVA were checked by visually examining the residuals against predicted values and using the Shapiro-Wilk and Levene's tests. The existence of significant effects ($p<0.05$) was followed by a post-hoc comparison test of means with the SNK.test from the agricolae package (De Mendiburu, 2014). When normality and homoscedasticity were not respected, a Kruskal-Wallis test was applied followed by means comparison using the kruskal.test from the agricolae package (De Mendiburu, 2014).

2.2.7 Simulation of SOC stocks evolution

2.2.7.1 AMG model

The simulation of SOC stocks evolution was made over the 1998-2014 period using the AMG model (Andriulo *et al.*, 1999; Saffih-Hdadi & Mary, 2008). AMG is a simple soil simulation model with an annual time step, which considers three compartments of organic matter: crop residues, active and stable humified organic matter. AMG was successfully evaluated to

simulate SOC evolution in Argentina (Andriulo *et al.*, 1999; Milesi Delaye *et al.*, 2013) and in 9 long term experiments (Saffih-Hdadi & Mary, 2008). The model uses the following equations:

$$QC = C_S + C_A \quad (2.3)$$

$$\frac{dC_A}{dt} = \sum_i m_i h_i - k C_A \quad (2.4)$$

where QC is the SOC stock ($t \text{ ha}^{-1}$), C_S is the stable carbon stock ($t \text{ C ha}^{-1}$), C_A is the active carbon stock ($t \text{ C ha}^{-1}$), m_i is the annual carbon input of organic residue i ($t \text{ ha}^{-1} \text{ yr}^{-1}$), h_i is the humification coefficient of the residue i and k is the mineralization rate of the soil active fraction (yr^{-1}). In the case where carbon input rate is constant every year, equations (3-4) can be integrated as:

$$QC = C_S + (C_0 - C_S)e^{-kt} + \sum_i \frac{h_i m_i}{k} (1 - e^{-kt}) \quad (2.5)$$

where C_0 is the initial SOC stock ($t \text{ ha}^{-1}$). The second term represents the residual amount of old carbon initially present and the third term is the humified carbon formed since the initial time. The mineralization rate k is dependent on pedoclimatic conditions and calculated as follows:

$$k = k_0 \cdot f(A) \cdot f(T) \quad (2.6)$$

where k_0 is the potential mineralization rate (yr^{-1}), A the clay content (g kg^{-1}) and T the temperature ($^{\circ}\text{C}$). The functions and parameters are described in Saffih-Hdadi and Mary (2008).

2.2.7.2 Modelling steps

The evolution of SOC stocks of each cropping system was first simulated on block 2 in which SOC was measured more frequently than block 1. Required data for modelling were SOC stocks at different dates, carbon inputs from crop residues returned to the soil, clay content and mean annual temperature. Saffih-Hdadi and Mary (2008) proposed values for some of the parameters based on observations in long term experiments. They recommended a value of 65% for the C_s/C_0 ratio, which is the proportion of stable carbon in the soil, considered as equivalent in all the systems at the start of the experiment in 1998. They also proposed a humification rate for straw residues (h_a) with a value of 0.21 that we used here for all types of AG residues. The humification rate for BG residues (h_r) was set at 0.42, *i.e.* two times greater than for AG residues, in accordance with several studies showing that roots and rhizodeposits contribute more to humification than aboveground residues (Balesdent & Balabane, 1996; Clapp *et al.*, 2000; Kristiansen *et al.*, 2005). Modelling was made in four steps:

- Step 1: SOC stocks in layer L1-3 were simulated for each cropping system with default values of the model and compared with observed data (simulation S0). Using default parameters, we calculated the mineralization coefficient ($k = 0.044 \text{ yr}^{-1}$), BG inputs from fescue residues in the CA system ($mf = 0.46 \text{ t C ha}^{-1} \text{ yr}^{-1}$), BG inputs from alfalfa residues ($ma = 0.58 \text{ t C ha}^{-1} \text{ yr}^{-1}$ in CA and $0.44 \text{ t C ha}^{-1} \text{ yr}^{-1}$ in ORG system).
- Step 2: the mineralization rate k of each of the four cropping systems was optimized, as well as the belowground inputs derived from fescue (mf) and alfalfa (ma), yielding six fitted parameters (simulation S6). The optimization procedure embedded in AMG software is based on a Bayesian method with a MCMC algorithm.
- Step 3: the mineralization rate k was assumed to be the same in each cropping system and was optimized, as well as the mf and ma parameters, so that 3 parameters were optimized (simulation S3).

Step 4: the evolution of SOC stocks was simulated in layer L1-3 for block 1 and then for each layer (L1, L2 and L3) of block 2 using the values of k , mf and ma parameters obtained in simulation S3. In the case of the multi-layer simulation, the C inputs had to be allocated into each layer. We assumed that the C inputs derived from aboveground material were proportional to the amounts of cPOM measured in each layer. We tested two hypotheses concerning BG carbon inputs allocation among soil layers: i) an equal C input into each layer, ii) an allocation proportional to cPOM content.

Table 2.3. Mean annual carbon inputs coming from crop residues (cash crops, cover crops and catch crops) over the 1998-2014 period.

		CON			LI			CA			ORG		
		AG	BG	AG+BG	AG	BG	AG+BG	AG	BG	AG+BG	AG	BG	AG+BG
		(t C ha ⁻¹ yr ⁻¹)											
Main crop	Cash crops	3.23 ^a	0.80 ^b	4.03	2.95 ^a	0.80 ^b	3.74	1.84 ^a	0.65 ^b	2.50	1.19 ^a	0.61 ^b	1.80
	Alfalfa ^c							0.41	0.16	0.57	0.58	0.47	1.04
Cover crop ^c	Fescue ^d							0.39	0.49	0.88			
	Alfalfa ^e							0.70	0.42	1.12			
Catch crop ^{a,f}		0.04	0.03	0.07	0.04	0.03	0.07	0.21	0.14	0.35			
Organic residues ^{a,g}											0.02		0.02
Total inputs		3.27	0.82	4.09	2.99	0.82	3.81	3.55	1.86	5.41	1.79	1.07	2.87

AG = aboveground inputs, BG = belowground inputs. CON = conventional, LI = low input, CA = conservation agriculture, ORG = organic farming.

^a measured

^b estimated with AMG model

^c estimated by modeling (see appendix 1.A)

^d Fescue, clover or fodder radish, period 1998-2009

^e Alfalfa, period 2010-2014

^f oat and vetch, white mustard or fodder radish

^g guano, feather meal

The statistical criteria used to evaluate the model were the mean difference (MD) and the root mean square error (RMSE), both expressed in t C ha⁻¹:

$$MD_{jk} = \frac{1}{n_{jk}} \sum_{i=1}^{n_{jk}} (QC_{ijk} - \widehat{QC}_{ijk}) \quad (2.7)$$

$$RMSE_{jk} = \sqrt{\frac{1}{n_{jk}} \sum_{i=1}^{n_{jk}} (QC_{ijk} - \widehat{QC}_{ijk})^2} \quad (2.8)$$

where n is the number of observation for the treatments j and the layer k, QC_i and \widehat{QC}_i are the observed and estimated SOC stocks for the observation i, treatment j and layer k.

2.3 Results

2.3.1 Crop yields and residues

Mean wheat yields decreased in the following order: CON > LI > CA > ORG (Table 2.2). They varied between 9.7 t ha⁻¹ yr⁻¹ and 5.4 t ha⁻¹ yr⁻¹ and were strongly related to the mineral N fertilizer rate ($R^2 = 0.80$, $p < 0.05$). Pea yields were similar for CON and LI (4.3 t ha⁻¹ yr⁻¹ on average) and smaller in CA and ORG (3.1 t ha⁻¹ yr⁻¹ on average). Rapeseed yields were 4.5 and 3.8 t ha⁻¹ yr⁻¹ for CON and LI respectively and much smaller in ORG with 0.8 t ha⁻¹ yr⁻¹ (due to pest attacks) hence this crop was stopped after two years in ORG. The mean amount of AG residues from main crops (wheat, pea and rapeseed) was estimated at 7.1, 6.4, 4.9 and 3.6 t DM ha⁻¹ yr⁻¹ for CON, LI, CA and ORG respectively (results not shown). The estimated amounts of BG residues were less different between cropping systems, with a mean value of 1.9 t DM ha⁻¹ yr⁻¹.

Details on carbon inputs from AG and BG residues are given in Table 2.3. The total inputs from the main crops varied between 1.80 and 4.03 t C ha⁻¹ yr⁻¹ and decreased in the following order: CON > LI > CA > ORG. Belowground inputs represented on average 0.71 t C ha⁻¹ yr⁻¹, corresponding to 20% to 34% of the total inputs. Alfalfa as main crop was only cultivated in CA and ORG. The total inputs coming from this crop were estimated at 0.57 and 1.04 t C ha⁻¹ yr⁻¹ in CA and ORG respectively, the belowground material representing 29% and 45% of this input. Cover crops only concerned the CA system and consisted of fescue until 2009 and alfalfa since 2010. The estimated inputs from fescue were 0.88 t C ha⁻¹ yr⁻¹ and those coming from alfalfa as a cover crop were 1.12 t C ha⁻¹ yr⁻¹, about half of these amounts deriving from root material. Catch crops grown in CA represented an additional input of 0.35 t C ha⁻¹ yr⁻¹.

Finally, total carbon inputs were higher in CA (5.41 t C ha⁻¹ yr⁻¹) than in the three other systems which received 4.09, 3.81 and 2.87 t C ha⁻¹ yr⁻¹ for CON, LI and ORG respectively. Cover crops, catch crops and alfalfa biomass compensated for the lower main crop production in CA and ORG, since they contributed to 54% and 37% of the total C inputs

2.3.2 Bulk densities

The measured bulk densities at the initiation of the experiment did not differ between plots, both in individual layers and in the old ploughed layer L1-3 (Table 2.4). There was also no significant difference between layers, unsurprisingly since the field was regularly ploughed (and therefore homogenized) down to about 30 cm before the onset of the experiment. In 2014, bulk density also did not differ between cropping systems both in individual layers and in layer L1. However, a significant change occurred between 1998 and 2014. Bulk density increased in all layers and all treatments, even though the change was not significant in LI and ORG systems. The mean bulk density increased from 1.40 to 1.50 g cm⁻³. The most detectable effect was found in the CA system where bulk density in the layers L2 and L3 was significantly greater than in the other systems, reaching 1.58 g cm⁻³. This high value was compensated by a lower value in the upper layer, so that the mean value over the old ploughed layer was about the same in all systems. Bulk density values must be analyzed with care as they also vary within a year. However, bulk density evolution in CA system is consistent with an observation often made in no-till systems, *i.e.* a decrease in the upper layer (0-10 cm) and an increase below (10-30 cm) compared to conventional tilled systems (*e.g.* Dimassi *et al.*, 2014).

Table 2.4. Bulk densities (g cm⁻³) measured in three soil layers in 1998 and 2014.

	Layer	Depth (cm)	ESM	CON		LI		CA		ORG	
			(t ha ⁻¹)	(g cm ⁻³)							
1998	L1	~0-10	1300	1.33 (0.03)		1.35 (0.07)		1.35 (0.04)		1.36 (0.08)	
	L2	~10-20	1300	1.41 (0.04)		1.43 (0.09)		1.39 (0.08)		1.46 (0.07)	
	L3	~20-30	1700	1.41 (0.04)		1.46 (0.06)		1.39 (0.08)		1.48 (0.08)	
	L1-3	~0-30	4300	1.38 (0.03)		1.41 (0.05)		1.38 (0.06)		1.43 (0.07)	
2014	L1	~0-10	1300	1.42 (0.09)	NS	1.43 (0.08)	NS	1.38 (0.02)	NS	1.50 (0.03)	*
	L2	~10-20	1300	1.52 (0.07)	*	1.53 (0.11)	NS	1.57 (0.02)	**	1.48 (0.02)	NS
	L3	~20-30	1700	1.54 (0.07)	*	1.55 (0.07)	NS	1.59 (0.03)	**	1.51 (0.01)	NS
	L1-3	~0-30	4300	1.49 (0.07)	*	1.50 (0.08)	NS	1.51 (0.02)	**	1.50 (0.01)	NS

Values in brackets are standard deviations. Asterisks indicate significant difference between years (* $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; NS = not significant).

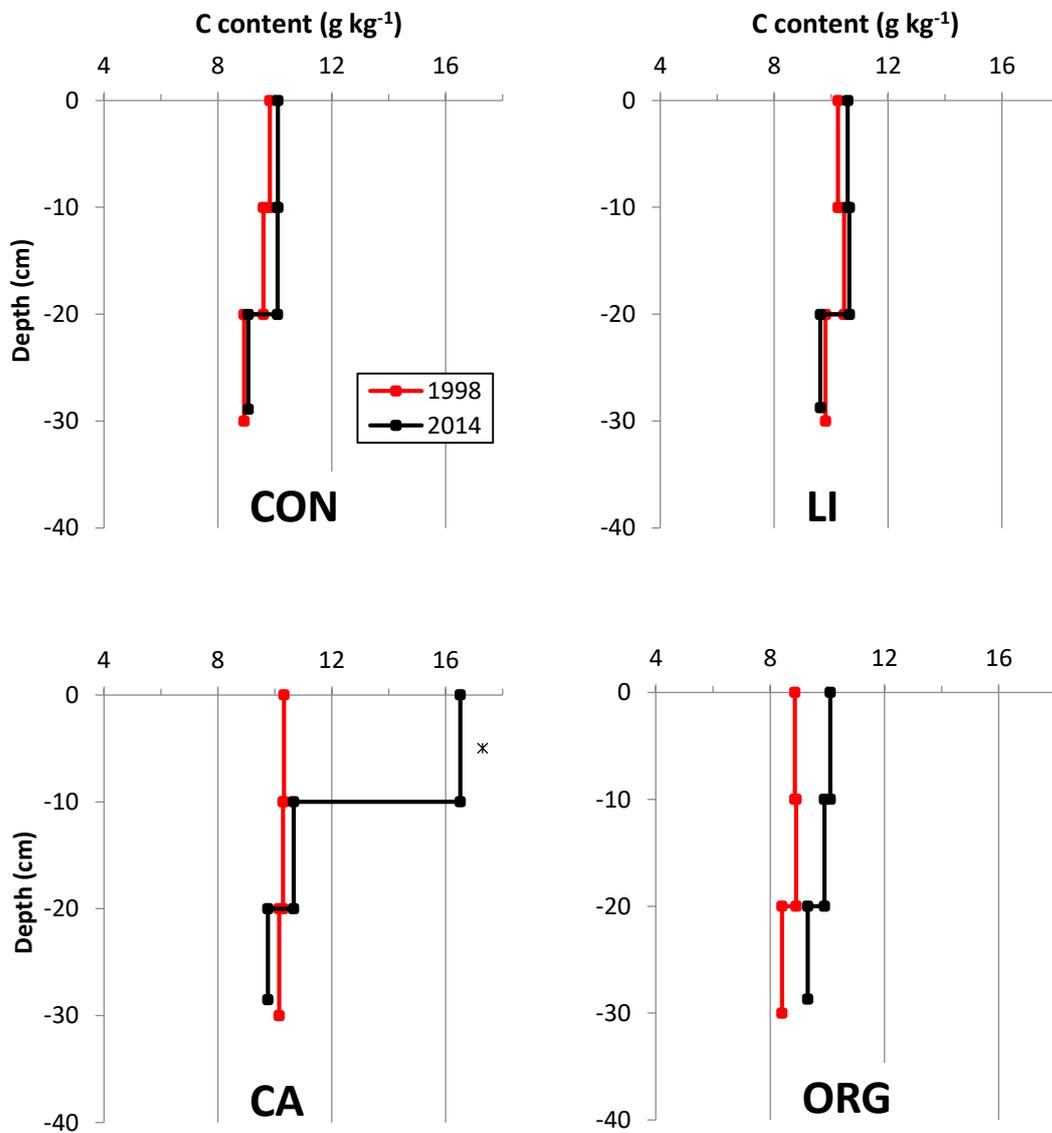


Figure 2.2. Profile of carbon concentration in the soil in 1998 and 2014. Depths correspond to fixed equivalent soil masses (see Table 2.4). Asterisks indicate significant evolution between 1998 and 2014 (* $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$). CON = conventional, LI = low input, CA = conservation agriculture, ORG = organic farming.

2.3.3 SOC concentrations in 1998 and 2014

The profiles of SOC concentration among soil layers measured in 1998 and 2014 are presented in Figure 2.2 for each cropping system. In 1998, no significant difference was found between systems except for ORG. The initial SOC concentration in ORG was smaller than in the other cropping systems (8.70 g kg^{-1} vs 9.76 g kg^{-1} on average, $p < 0.10$). In 2014, the distribution varied between the four cropping systems. SOC concentration in the first layer did not differ significantly between CON, LI and ORG (mean value = 10.2 g kg^{-1}), but was 61% higher in the CA system (16.5 g kg^{-1} , $p < 0.001$). No significant difference was found between cropping systems in any of the deeper layers (L2, L3, L4, L5). The mean SOC concentration in these layers was 10.5 , 9.5 , 5.3 and 4.1 g kg^{-1} , respectively. Over the whole ploughed layer (L1-3), CA had a significantly higher carbon concentration compared to the three other systems (+22%, $p < 0.05$). This was also true for layer L1-5, equivalent to 0-60 cm, since SOC concentration was 19% higher in CA compared to the other systems ($p < 0.01$): 8.54 vs 7.14 g kg^{-1} respectively. A significant temporal change in SOC concentration was detected between 1998 and 2014 in the CA system on the L1 layer ($+0.39 \text{ g kg}^{-1}$, $p < 0.05$) but neither in the other cropping systems nor in the other layers.

2.3.4 cPOM concentration in 2014

The distribution of coarse particulate organic matter measured in 2014 in soil layers L1, L2 and L3 is presented in Table 2.5. cPOM concentration did not differ between CON, LI and ORG systems in any layer. The mean value in the old ploughed layer (L1-3) was $0.44 \pm 0.01 \text{ g C kg}^{-1}$. cPOM concentration was much more stratified in the CA system: it was significantly higher in CA than in CON, LI and ORG in the upper layer L1 (0.85 vs 0.51 g C kg^{-1} on average) and lower in layers L2 and L3. A full compensation occurred between layers since cPOM concentration in the old ploughed layer of CA system was equal to that of other systems: $0.42 \pm 0.01 \text{ g C kg}^{-1}$. cPOM carbon concentration in layer L1-3 represented between 3.5% and 4.6% of SOC; this was comparable to previous measurements made in 2003 (Balabane *et al.*, 2005) which varied between 2.8% and 5.0%. The rather low content of cPOM indicates a fast decomposition of crop residues in all soil layers and all systems including CA

Table 2.5. Coarse particulate organic matter (cPOM) measured in 2014: carbon concentration (g C kg⁻¹) and proportion of total soil organic carbon.

Layer	Depth (cm)	ESM (t ha ⁻¹)	CON		LI		CA		ORG	
			(g kg ⁻¹)	%SOC						
L1	~0-10	1300	0.55 (0.12)	b 5.4%	0.50 (0.09)	b 4.7%	0.85 (0.21)	a 5.1%	0.48 (0.11)	b 4.7%
L2	~10-20	1300	0.53 (0.17)	a 5.2%	0.51 (0.12)	a 4.8%	0.33 (0.09)	b 3.1%	0.54 (0.10)	a 5.5%
L3	~20-30	1700	0.31 (0.12)	a 3.4%	0.30 (0.07)	a 3.2%	0.18 (0.06)	b 1.8%	0.34 (0.07)	a 3.6%
L1-3	~0-30	4300	0.45 (0.13)	a 4.6%	0.42 (0.07)	a 4.2%	0.42 (0.10)	a 3.5%	0.44 (0.06)	a 4.5%

Table 2.6. SOC stocks (t C ha⁻¹) measured in 1998 and 2014 in three soil layers (mean of the two blocks).

	Layer	Depth (cm)	ESM (t ha ⁻¹)	CON		LI		CA		ORG	
				(t C ha ⁻¹)		(t C ha ⁻¹)		(t C ha ⁻¹)		(t C ha ⁻¹)	
1998	L1	~0-10	1300	12.8 (1.0)	a	13.3 (2.0)	a	13.4 (2.5)	a	11.5 (1.4)	a
	L2	~10-20	1300	12.5 (1.0)	a	13.6 (2.7)	a	13.4 (1.7)	a	11.6 (1.4)	a
	L3	~20-30	1700	15.2 (1.9)	a	16.7 (3.3)	a	15.1 (5.0)	a	14.3 (1.5)	a
	L1-3	~0-30	4300	40.4 (3.5)	a	43.6 (8.0)	a	41.9 (8.7)	a	37.4 (4.3)	a
2014	L1	~0-10	1300	13.1 (1.1)	a NS	13.7 (1.9)	a NS	21.5 (2.9)	b *	13.1 (1.3)	a NS
	L2	~10-20	1300	13.1 (1.4)	a NS	13.8 (1.8)	a NS	13.9 (2.0)	a NS	12.9 (0.7)	a NS
	L3	~20-30	1700	15.4 (2.0)	a NS	16.3 (2.1)	a NS	16.6 (2.2)	a NS	15.8 (1.2)	a NS
	L4	~30-40	1400	6.7 (1.0)	a	7.3 (1.4)	a	8.4 (1.1)	a	7.2 (1.2)	a
	L5	~40-60	2800	10.7 (5.8)	a	11.8 (5.5)	a	12.2 (1.2)	a	11.4 (4.5)	a
	L1-3	~0-30	4300	41.7 (4.2)	a NS	43.9 (5.3)	a NS	51.9 (6.6)	b NS	41.8 (2.6)	a NS
	L1-5	~0-60	8500	58.8 (5.8)	a	63.0 (5.5)	a	72.6 (7.0)	b	60.4 (4.5)	a

Values in brackets are standard deviations. Asterisks indicate significant difference between years (* $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; NS = not significant). Different letters indicate significant differences between rotations (Newman-Keuls, CI = 95%). ESM = equivalent soil mass, CON = conventional, LI = low input, CA = conservation agriculture, ORG = organic farming.

2.3.5 SOC stocks in 1998 and 2014

At the start of the experiment, SOC stocks did not differ significantly between plots in any layer ($p < 0.05$), but tended to be lower in the ORG plots. The mean SOC content in the ploughed layer (L1-3) was 40.8 t ha^{-1} (Table 2.6). In 2014, the average SOC stocks at ESM were significantly higher in CA than in CON, LI and ORG which did not differ from each other ($p < 0.05$). The mean SOC amount in layer L1-3 was 51.9 t ha^{-1} in CA and $42.5 \pm 1.2 \text{ t ha}^{-1}$ in the three other systems. The SOC stocks over a greater depth (ca. 0-60 cm, layer L1-5) also differed significantly with 72.6 t ha^{-1} for CA against $60.7 \pm 2.1 \text{ t ha}^{-1}$ on average for CON, LI and ORG ($p < 0.01$). The main difference occurred in the upper layer L1 which contained 21.5 t ha^{-1} in CA, *i.e.* 8.1 t ha^{-1} higher than in the other systems. No difference between systems was detectable in layers L2 and L3.

The temporal variation of SOC stocks between 1998 and 2014 in each block is given in Table 2.7. SOC stocks in block 1 were always higher than the corresponding ones in block 2 (average difference = 5.9 t C ha^{-1}). The SOC change after 16 years was very similar between blocks in spite of the initial heterogeneity in SOC stocks. The mean change was 1.3, 0.3, 10.0 and 4.4 t ha^{-1} in the CON, LI, CA and ORG systems, respectively. The SOC increase was significant only in CA and ORG systems. If we consider the conventional system as a reference, the mean rate of C sequestration was $0.55 \text{ t ha}^{-1} \text{ yr}^{-1}$ in the conservation agriculture system and $0.20 \text{ t ha}^{-1} \text{ yr}^{-1}$ in the organic system.

Table 2.7. Changes in SOC stocks during the 1998-2014 period in the old ploughed layer.

	CON		LI		CA		ORG	
	(t C ha ⁻¹)							
Block 1	2.5	(2.4)	-0.9	(3.0)	11.3	(3.5)	4.9	(1.9)
Block 2	0.0	(1.5)	1.6	(1.7)	8.7	(2.7)	3.9	(1.8)
All blocks	1.3	(1.2) a	0.3	(2.0) a	10.0	(2.2) c	4.4	(1.0) b

Values in brackets are standard deviations. Different letters indicate significant differences between cropping systems (Newman-Keuls, CI = 95%). CON = conventional, LI = low input, CA = conservation agriculture, ORG = organic farming.

2.3.6 Simulating the evolution of SOC stocks in the old ploughed layer

Table 2.8 presents the results of the three steps of simulation (S0, S6, S3) in terms of parameter values and statistical criteria giving the quality of fit for SOC stocks in layer L1-3 of block 2. In simulation S0, the simulations were conducted with the default parameters of the model and with C inputs indicated in Table 2.3. The total C inputs ranged from 2.87 t C ha⁻¹ yr⁻¹ in the ORG system to 5.41 t C ha⁻¹ yr⁻¹ in the CA system. The simulation gave a good agreement with the observed measurements, since the mean difference did not exceed 1.64 t C ha⁻¹ (in CA system), and the root mean square error (on average 1.69 t C ha⁻¹) was close to the mean standard deviation of measurements (2.10 t C ha⁻¹). The positive MD reveals a slight underestimate of observed values by the model, particularly for the CA system. In simulation S6, which included the optimization of six parameters, the quality of fit was slightly improved compared to simulation S0 but the improvement was small since the average MD was 0.27 t C ha⁻¹ (instead of 0.85 in S0) and the average RMSE equalled 1.47 t C ha⁻¹ (instead of 1.69). The Bayesian optimization procedure resulted in a modification of calculated inputs; it suggested that the belowground inputs derived from fescue (mf) had been underestimated in CA whereas those derived from alfalfa (ma) had been overestimated. However, the confidence intervals of mf and ma were large and included the default values considered in simulation S0. The mineralization rates (k) optimized for each of the four cropping systems were smaller than the default value used in S0 (0.041 vs 0.044 yr⁻¹) and all contained within the confidence intervals. No significant difference was found between the k values of the four cropping systems.

These observations led us to simulation S3, in which a single mineralization coefficient was adopted for all treatments. This simulation S3 gave a similar quality of fit compared to S6, with a slightly higher MD (0.34 vs 0.27 t C ha⁻¹ on average) and RMSE (1.56 vs 1.47 t C ha⁻¹). The optimized values of mf were higher than the default values (0.73 vs 0.49 t C ha⁻¹ yr⁻¹), suggesting that the fescue grown as cover crop had an important root turnover and produced greater amounts of rhizodeposits than initially assessed. In contrast, the optimized values of ma inputs were very close to the default value (4% higher). The optimized mineralization rate k was slightly smaller than the default value (0.041 vs 0.044 yr⁻¹).

Table 2.8. Mean value of the default or optimized parameters (*ma*, *mf* and *k*) for the simulations (S0, S3 and S6) of SOC evolution between 1998 and 2014.

	Total C inputs	BG C inputs	Fixed or optimized parameters						Statistical criteria		
			<i>mf</i>		<i>ma</i>		<i>k</i>		MD	RMSE	SD
			(t C ha ⁻¹ yr ⁻¹)				(yr ⁻¹)		(t C ha ⁻¹)		
S0. Simulation with default parameters											
CON	4.09	0.82					0.044	0.43	2.24	2.02	
LI	3.81	0.82					0.044	0.41	1.21	1.08	
CA	5.41	1.86	0.49		0.58		0.044	1.64	2.06	3.93	
ORG	2.87	1.07			0.47		0.044	0.91	1.25	1.38	
S6. Bayesian optimization (6 parameters)											
CON	4.09	0.82					0.051 (0.037 - 0.070)	1.04	2.24	2.02	
LI	3.81	0.82					0.041 (0.031 - 0.051)	0.00	1.39	1.08	
CA	5.62	2.05	0.80 (0.18 - 1.67)		0.47 (0.07 - 1.57)		0.037 (0.015 - 0.076)	-0.04	1.49	3.93	
ORG	2.70	0.98			0.38 (0.06 - 1.26)		0.034 (0.024 - 0.054)	0.08	0.76	1.38	
S3. Bayesian optimization (3 parameters)											
CON	4.09	0.82					0.041 (0.035 - 0.051)	0.14	2.34	2.02	
LI	3.81	0.82					0.041 (0.035 - 0.051)	0.12	1.31	1.08	
CA	5.67	2.12	0.73 (0.28 - 1.19)		0.60 (0.19 - 1.14)		0.041 (0.035 - 0.051)	0.49	1.58	3.93	
ORG	2.89	1.09			0.49 (0.14 - 0.85)		0.041 (0.035 - 0.051)	0.59	1.00	1.38	

mf = mean annual belowground inputs derived from fescue, *ma* = mean annual belowground inputs derived from alfalfa. Fitted parameters are shown in bold types. Confidence intervals are in brackets. Statistical criteria: MD = mean difference, RMSE = root mean square error, SD = standard deviation. CON = conventional, LI = low input, CA = conservation agriculture, ORG = organic farming.

The temporal evolution of observed and simulated SOC stocks in CON, LI, CA and ORG systems in block 2 is presented in Figure 2.3. The three simulations (S0, S6 and S3) could correctly reproduce the dynamics of SOC evolution between 1998 and 2014 in the four cropping systems. The variability of SOC measurements in the CON system was greater than in other systems and hampers the comparison between observed and simulated values. The three simulated kinetics were very close each other in the LI and ORG systems, and close to observed data. The poorer simulation was obtained in CA system with the direct simulation S0 which underestimated observations made in 2000, 2003 and 2011. The better agreement obtained with simulations S3 and S6 during the 16-year period is mainly due to the increased values of C inputs derived from the fescue cover crop.

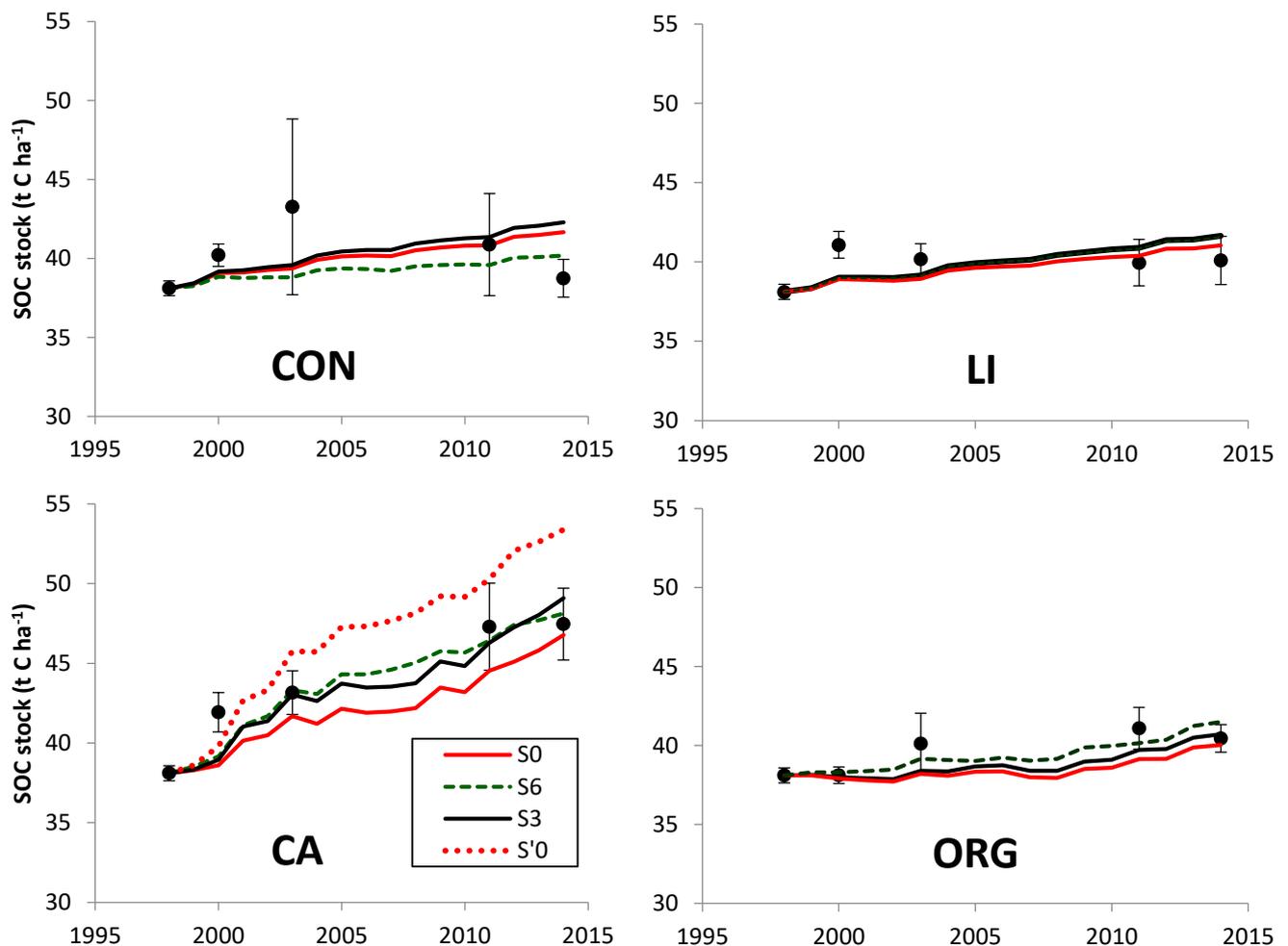


Figure 2.3. Evolution of SOC stocks in the old ploughed layer (L1-3) from 1998 to 2014 in block 2: observed (symbols) and simulated values (lines) in the four cropping systems. S0 = simulation with original parameters; S6 = simulation with 6 parameters optimized; S3 = simulation with 3 parameters optimized (see Table 2.8); S'0 = simulation with a reduced mineralization rate in CA, according to Paustian et al (2000).

2.3.7 Simulation of SOC evolution in block 1 and elementary layers of block 2

In the last step, we evaluated the ability of the model to simulate the SOC stock in the L1-3 layer of block 1 with the parameter values obtained in simulation S3 for block 2. Figure 2.4 shows that the model succeeded rather well in predicting the SOC stocks in 2014 in spite of the spatial variability between blocks, except for the CON treatment which had a greater intra-variability. This result points out the interest of a detailed characterization of the spatial variability of SOC contents at time 0 in long term experiments. During the 16-year period, the observed rate of change in SOC stock (mean of two blocks) was 0.08, 0.02, 0.63 and 0.28 t C ha⁻¹ yr⁻¹ for CON, LI, CA and ORG respectively. It is rather close to the simulated rate of change which was 0.30, 0.06, 0.63 and 0.39 t C ha⁻¹ yr⁻¹ respectively.

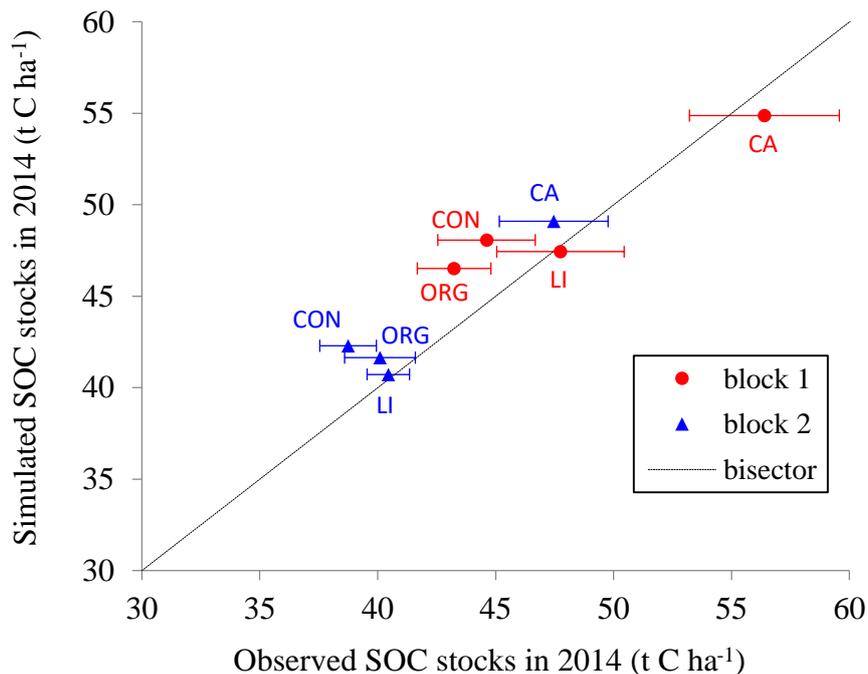


Figure 2.4. Observed and simulated SOC stocks over the old ploughed layer (L1-3) in 2014 for each block and each cropping system. CON = conventional, LI = low input, CA = conservation agriculture, ORG = organic farming.

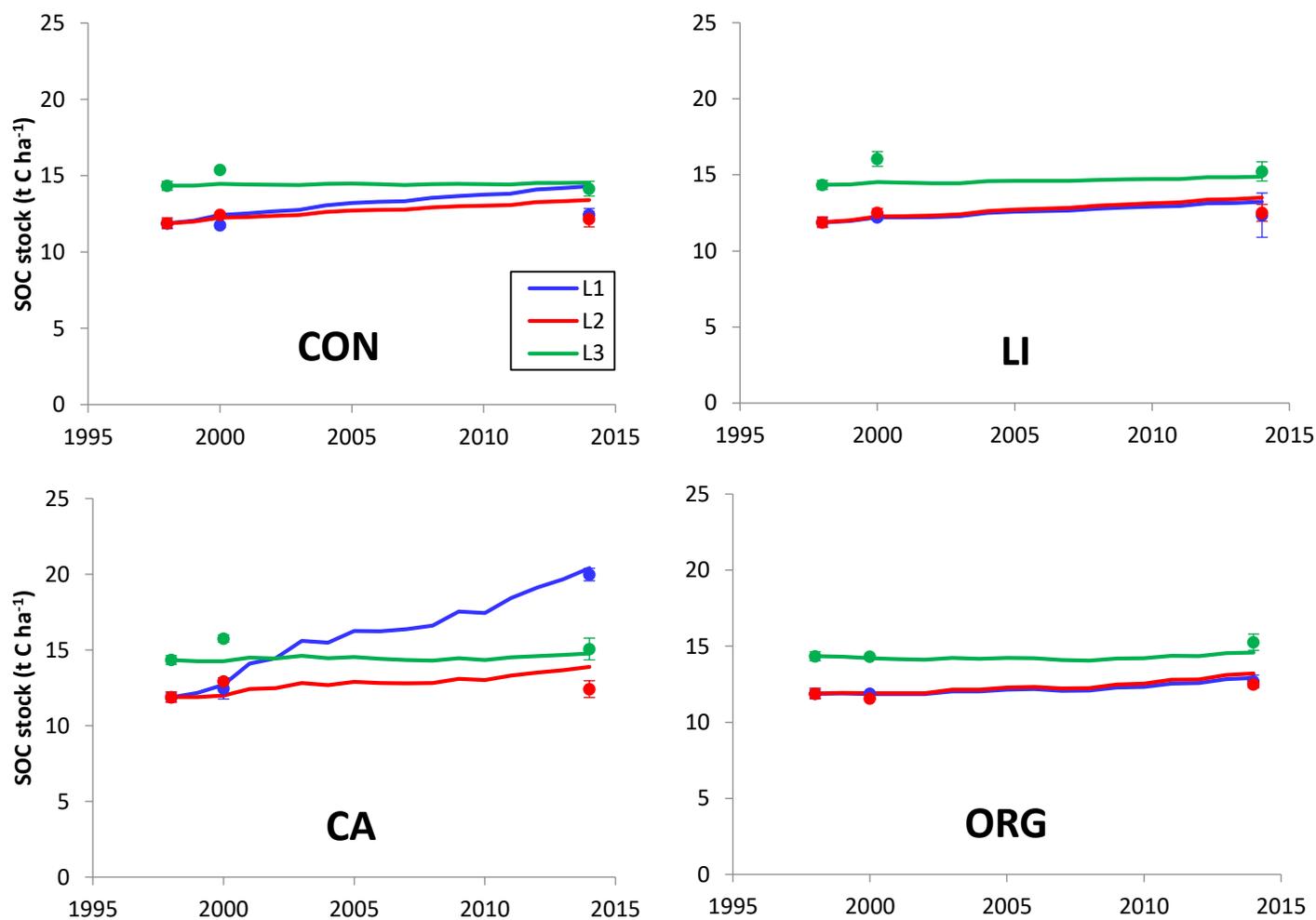


Figure 2.5. Evolution of SOC stocks observed (symbols) and simulated (lines) from 1998 to 2014 in block 2 of each cropping system. Simulations correspond to S3 with optimized values of m_f and m_a and carbon input distribution in layers L1 (~0-10 cm), L2 (~10-20 cm) and L3 (~20-30 cm) proportional to cPOM concentration. CON = conventional, LI = low input, CA = conservation agriculture, ORG = organic farming.

We also evaluated the ability of the model to predict the evolution of SOC stock in each layer (L1, L2 and L3) of block 2. The mineralization rate was assumed to be similar in each soil layer and equal to the value determined in simulation S3 ($k = 0.041 \text{ yr}^{-1}$). The C inputs derived from fescue and alfalfa (optimized values of m_f and m_a resulting from simulation S3) were distributed between the three layers according to each allocation hypothesis made earlier, *i.e.* C allocation either proportional to cPOM content or identical in each layer. Observed and simulated SOC stocks are presented in Figure 2.5. The quality of fit was about the same for each hypothesis made for belowground inputs (results not shown). Measurements indicated that SOC stocks in layers L2 and L3 were almost stable in all systems. Simulations gave good results for all treatments and all layers, except for the layer L1 in the CON system and the layer L2 in the CA system, whose SOC concentrations were all slightly overestimated.

2.4 Discussion

The “La Cage” experiment is characterized by alternative cropping systems with specific crop rotations (*i.e.* with cover or catch crops), no use of livestock manure or other exogenous organic fertilizer and a mineral fertilization in CON, LI and CA. We compare our results here with other studies on similar farming practices and for soil measurements made at least over 0-30 cm depth.

2.4.1 Yields and inputs of crop residues

The main crop yields observed in CON were similar to the regional production levels for wheat, pea and rapeseed over the same period. In the LI system, characterized by reduced N fertilization (-21%), ploughing frequency and use of pesticides, main crop yields were slightly smaller, with the average wheat yield reaching 91% of CON. (Loyce *et al.*, 2012) reported similar results for wheat grown under an integrated cropping system with management similar to LI in 28 field experiments in France between 1999 and 2002, with an average yield of 8.4 t ha^{-1} representing 94% of the conventional yield. In comparison, the ORG system which received neither mineral fertilizer nor farmyard manure, wheat yields were much lower, averaging only 55% of CON. Entz *et al.* (2005) studied a similar organic system in a pure grain rotation, a green manure-grain rotation and an alfalfa-grain rotation without manure application for 12 years. Similar to our results, they found smaller yields in the organic compared to conventional system with a decrease of 63, 46 and 14% for the three rotations respectively.

Seufert *et al.* (2012) reported in their meta-analysis that organic wheat yields reached 60% of conventional yields (75% when considering all cereals crops). Lastly, in our CA system, with a fertilization rate reduced by 28%, wheat yield represented 69% of the CON system. We compared these yields with two meta-analyses of observations made in no-tilled cropping systems, both showing a smaller gap with conventional yields. Firstly, the meta-regression carried out by Van den Putte *et al.* (2010) pointed out that yields of no-till systems in northern Europe were on average 8.5% lower than those obtained in conventionally tilled systems. Secondly, Pittelkow *et al.* (2015) found in their meta-analysis of 678 studies that cereal yields grown under no-till systems reached 95% of conventional yields.

Lower yields result in lower amounts of crop residues (aboveground + belowground) returned to soil which in turn affects SOC stocks. Thus, in our experiment, the estimates of total C inputs derived from cash crops ranked as follows: CON (4.03 t C ha⁻¹ yr⁻¹) > INT(3.74) > CA (2.50) > ORG (1.80). However, the CA system is characterized by the frequent or permanent cultivation of catch crops and cover crops associated with the main crop. When also accounting for these additional crops, the C input ranking becomes: CA (5.41 t C ha⁻¹ yr⁻¹) > CON (4.09) > LI (3.81) > ORG (2.87), revealing that the lower inputs associated with lower yields of the main crops were compensated by the inputs from cover crops, in particular from root materials. Bell *et al.* (2012) estimated these inputs at 2.25 and 1.50 t C ha⁻¹ yr⁻¹ in their CON and ORG systems respectively, *i.e.* close to our values.

2.4.2 SOC storage in relation with cropping systems

The main change in SOC stock over the 16 studied years was found in the CA system (+10.0 t C ha⁻¹, *i.e.* +24%), followed by ORG (+4.4 t C ha⁻¹, *i.e.* +12%) whereas the other systems did not change significantly (+3% and +1% for CON and LI). The highest SOC increase observed in the CA system can be attributed to one or several specific practices: no-tillage, permanent soil cover and crop diversification. These effects are often reported separately by authors. However, the effect of tillage *sensu stricto* (all other factors being equal) on SOC sequestration is still controversial. In fact, the more recent meta-analyses comparing no till or reduced tillage versus full inversion tillage indicate that the mean C sequestration over a depth greater or equal to 30 cm over a period of about 15 years is negligible on average (Luo *et al.*, 2010a) or small (mean value = 3.4 t C ha⁻¹; Virto *et al.*, 2011). Virto *et al.* (2011) have shown that the tillage effect is partly attributable to changes in C inputs. Moreover, climatic factors such as annual precipitations can also explain variations in SOC sequestration in no-till soils, as demonstrated

by Blanco-Moure *et al.* (2013) and Dimassi *et al.* (2014). The latter authors established linear relationships between the rate of change in SOC content and the water balance during the studied period. If we apply these relationships to our conditions with a relative dry climate (mean annual precipitation = 627 mm and water balance = -108 mm), we conclude that the effect of no-tillage per se in the CA system may have led to a carbon sequestration rate of 0.17 t C ha⁻¹ yr⁻¹ between 1998 and 2014 in the layer 0-5 cm, and 0.11 t C ha⁻¹ yr⁻¹ in the layer 0-30 cm. Therefore, the no-till effect by itself would have contributed 20% to the total sequestration rate (0.55 t C ha⁻¹ yr⁻¹). Hence, the change in crop rotation is more likely to explain SOC increase than the no-till practice. Furthermore, the amount of crop residues has been shown to be an essential factor in SOC storage (*e.g.* Luo *et al.*, 2010a,b; Powlson *et al.*, 2011). At “La Cage”, the additional crops introduced in the CA system contributed to increase the total C inputs. The impact of C inputs was also studied by Mary and Justes (2012) and Poeplau and Don (2015) who analysed long-term studies focusing on catch crops or cover crops. The authors reported mean SOC sequestration rates of 0.29 and 0.32 t C ha⁻¹ yr⁻¹ respectively during a period comparable to ours (15 and 12 years) when these additional crops were introduced. However, a weak correlation was found at La Cage between changes in SOC stocks and total C inputs ($R^2 = 0.38$, $p < 0.10$), revealing that SOC stock changes were also due to other factors, such as the type of crop residue depending on species.

Indeed, the effect of crop diversity can also influence SOC storage Luo *et al.* (2010b) studied the effect of this practice in a meta-analysis focusing on the impact of agricultural practices on SOC changes in Australia over a mean duration of 11-yr. They found that increasing crop diversity (rotation vs monoculture) had a small effect on SOC stocks (+5%) but a larger effect (+18%) when perennial legumes were introduced in the rotation. More recent studies underline the relevance of this finding: Tiemann *et al.* (2015) observed an influence of crop diversity on SOC stocks that was mainly explained by the introduction of a legume cover crop (pure red clover or mixed with rye). In a fescue-alfalfa rotation, Ferchaud *et al.* (2016) also found an important SOC storage rate of 0.87 ± 0.28 t C ha⁻¹ yr⁻¹ compared to a rotation of annual crops (sorghum-triticale) during a 6 yr period. Similarly, Albuquerque *et al.* (2015) found a higher SOC sequestration rate in a alfalfa-maize sequence compared to a winter wheat-soybean rotation (+0.50 t C ha⁻¹ yr⁻¹ over 0-100 cm), both systems being in no-tillage for 21 yr. Although it is difficult to disentangle the importance of these practices on SOC storage, some authors managed to study them jointly. Taking the oat-maize rotation as the baseline of their study, Conceição *et al.* (2013) found higher SOC sequestration in diversified cropping system during

18 years. More precisely, the addition of a legume cover crops (vetch or cowpea) to the cropping system increased the SOC sequestration for tilled and no-tilled systems (+0.28 and +0.34 t C ha⁻¹ yr⁻¹ respectively). This increase was higher than the effect of an exclusive change to no-tillage in the simple or diversified rotation (+0.18 and +0.25 t C ha⁻¹ yr⁻¹ respectively).

In our ORG system, we found a significant increase in SOC stock between 1998 and 2014 (+12%), whereas SOC stocks did not vary significantly in the CON and LI systems. This confirms the relevance of long-term studies with a good characterization of all plots at time 0 to detect true differences between treatments (Neto *et al.*, 2010). There are four possible explanations for this increase: i) the lower (although not significantly) initial SOC stock in ORG compared to the other treatments which was therefore more distant from the equilibrium level; ii) the possible C inputs derived from additional weeds, even though weeds were rather well controlled by mechanical operations; iii) the possibly higher C input derived from roots in ORG compared to CON due to enhanced root growth in nutrient limited conditions as shown by Chirinda *et al.* (2012); iv) the presence of perennial legumes in the crop rotation, as explained above.

One example of the impact of C input on SOC sequestration in organic cropping systems in particular is presented by Pimentel *et al.* (2005). They measured higher SOC sequestration in an organic crop rotation including a hairy vetch catch crop compared to a conventional rotation (+7% on the 0-30 cm depth), under similar C inputs. Similarly, Reid *et al.* (2015) found that the root turnover rate of alfalfa was as important as that of perennial ryegrass (3.7 yr⁻¹) which is known to have a high sequestration rate (*e.g.* Soussana *et al.* 2007). Conversely, after 18 years of experiment comparing organic and conventional systems including alfalfa in the rotation, Bell *et al.* (2012) found no significant difference in SOC stocks over 0-30 cm. Our results, which suggest that the perennial crop alfalfa had an essential contribution to SOC storage in the ORG system through important root turnover and rhizodeposit inputs, are consistent with Pimentel *et al.* (2005) and Reid *et al.* (2015) but not with Bell *et al.* (2012). The difference with Bell *et al.* (2012) is most likely explained by the fact that alfalfa was less frequently cut in their study (twice a year) so that the root turnover was lower.

2.4.3 SOC distribution over the old ploughed layer in CA

The carbon sequestration observed in the CA system was remarkably high in the upper layer (0-10 cm). Several studies under CA also found similar results. For example, Diekow *et al.*

(2005) analyzed the impact of converting conventional systems to conservation agriculture in Brazil. The conversion into a CA system with a legume cover crop (lablab or pigeon pea) resulted in an increase of 12-16 t C ha⁻¹ in the 0-17 cm layer after 17 years, compared to a no-till system without permanent cover.

The two meta-analyses comparing SOC distribution in soil layers in no-till versus tilled systems show that tillage induced both positive and negative SOC variations in the soil profile: Luo *et al.* (2010a) found a mean increase of 3.1 t C ha⁻¹ in 0-10 cm and a decrease of 3.1 t C ha⁻¹ in 10-30 cm; Angers and Eriksen-Hamel (2008) found a relative increase of 30% in 0-10 cm and a decrease of 26% in 10-30 cm. This distribution is attributable to the effect of tillage per se. Likewise, we observed an increase in the CA system (compared to CON) in the layer 0-10 cm, but no decrease in the deeper layers, indicating that the additional C inputs in the CA system due to cover crops were responsible for the major part of SOC sequestration. Besides the amount of C inputs, the specific nature of the cover crop is likely to have contributed to this sequestration, particularly with a greater proportion of root material compared to annual crops. In support of this, Constantin *et al.* (2010) and Poeplau and Don (2015) have shown that the conversion of catch crop or cover crop residues into stable organic matter was highly efficient. Particularly, the greater contribution of roots to C humification compared to aerial residues has been shown in several studies (Balesdent & Balabane, 1996; Johnson *et al.*, 2006). Rasse *et al.* (2005) calculated that the mean residence time of root-derived C in soils was 2.4 times that of shoot-derived C.

2.4.4 Simulation of SOC storage

Our objectives for using the simple AMG model were: i) to estimate the carbon inputs derived from belowground parts of cover crops and ii) to test whether the C mineralization rates could differ between systems, particularly the CA system which was no tilled. Modelling allowed us to simulate the effects of no-tillage and of crop residue inputs in the CA system separately. The model was able to correctly simulate SOC evolution in all cropping systems. The quality of fit was slightly improved by optimizing 3 or 6 parameters compared to the default values. The optimization procedure applied to each system individually (simulation S6) gave relative large confidence intervals for the mineralization rates which did not differ significantly from each other. The optimization made with a common mineralization rate (S3) gave an equivalent quality of fit, indicating that the hypothesis of similar mineralization rates between systems, including CA, could explain the observed data. In the literature, the effect of no-till on the long-

term on mineralization rates is controversial. For instance, Paustian *et al.* (2000) compiled the results of three studies calculating the mean residence time of SOC (MRT, inverse of the mineralization rate) in long-term experiments comparing no-till and conventional tillage, using the natural ^{13}C tracing technique. The MRT (average of the three sites) was found to be 1.9 times higher in the no-till systems. However, using the same technique, Haile-Mariam *et al.* (2008) did not find any difference in MRT between no-till and tilled systems in three other long term experiments in USA. To address this controversy, we tested the results of Paustian *et al.* (2000) at “La Cage” by assuming that the MRT in CA system was 1.9 times greater than in the other systems. The model thus parameterized overestimated SOC stocks in the CA system starting from 2003 and diverging continuously after (simulation S'0 in Fig. 3). Therefore, the hypothesis of a much greater MRT in the CA system was rejected. This conclusion is consistent with results obtained by Oorts *et al.* (2006, 2007) for carbon and nitrogen both in laboratory and in situ and with the review made by Mary *et al.* (2014) showing small or insignificant differences in nitrogen mineralization rates in long-term experiments only differing in tillage techniques.

2.5 Conclusion

We quantified SOC stocks and their temporal dynamics over 16 years in the long-term experiment of “La Cage” (northern France) comparing conventional and alternative cropping systems. The SOC stocks did not change throughout time in conventional (CON) and low-input (LI) systems, slightly increased in the organic (ORG) system and increased markedly in the top soil layer (0-10 cm) of the conservation agriculture (CA) system. The amount and nature of C inputs, particularly the additional belowground inputs due to cover crops in the CA system, were able to explain this temporal evolution. Another significant result in the CA system was the constancy of SOC stocks in the layer 10-30 cm compared to the other treatments and also throughout time, contrasting with the SOC decline often found in this layer in no-tilled situations. Modelling SOC stocks evolution was successfully achieved for all of the cropping systems with an optimized common mineralization rate and higher belowground inputs from alfalfa and fescue. Our results stress the importance of long-term studies with initial and dynamic characterization of SOC stocks throughout time which allows a better understanding of the relevant components of farming practices in carbon storage, highlighting the effect of permanent plant cover.

2.6 Acknowledgements

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CHAPTER

3.

Similar mineralization rates of soil organic carbon and nitrogen in different alternative arable cropping systems

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Highlights:

- ▶ C and N mineralization were compared in four cropping systems after 17 years
- ▶ Incubations were realized on disturbed or undisturbed soil cores
- ▶ C and N mineralized were greater in undisturbed than in disturbed soil samples
- ▶ Specific C and N mineralized (per unit of SOC or SON) did not differ between the four systems

Abstract

No tillage is believed to mitigate greenhouse gas emissions from agricultural activities by increasing soil carbon stocks, assuming a limited mineralization of soil organic carbon (SOC) and nitrogen (SON). Regrettably, the impact of cropping practices on SOC and SON mineralization rates is often confined to superficial soil layer and disrupted soil. We compared SOC and SON mineralization rates at the long term experiment of La Cage (France) in four different cropping systems, under conventional (CON), low input (LI), conservation agriculture (CA) and organic (ORG) management. Disturbed soil samples from the 0-27 cm soil layer were incubated in laboratory for four months to record C and N mineralization in all treatments, while undisturbed soil samples from CON and CA were incubated to account for soil tillage effect. Physical disturbance of soil samples decreased the soil C and N mineralization rates. Model fitting showed that the size of the labile pool contributing to C mineralization was much larger in undisturbed soil cores (*e.g.* 565 in CON and 377 mg C kg⁻¹ in CA) than in disturbed samples (100 and 90 mg C kg⁻¹ respectively). A higher abundance of labile SOC in undisturbed soil cylinders (coarse plant residues and fine roots) compared to disturbed soils samples would explain this result. No differences were found between the four cropping systems in their specific rate of mineralization, expressed *vs* SOC, SON or microbial biomass C. For undisturbed soil cores, similar mineralization in CA and CON may result from the balance between higher amount of coarse labile OM and less favourable soil structure for decomposition in CA. For disturbed soil samples, similar mineralization rates suggested that the OM decomposability and the environmental conditions for decomposers were similar between cropping systems. Overall, these results confirmed the equivalent SOC mineralization rate regardless the cropping treatments as previously estimated *in silico* (Autret *et al.*, 2016), validating the hypothesis of unmodified mineralization rates according to alternative cropping practices, including no-tillage. Hence, our results, together with the increased SOC stocks observed in CA and ORG treatments, suggest that increased biomass returns to soil or changes in microbial physiology may be the main drivers of SOC storage.

3.1 Introduction

Agricultural practices can play a major role in mitigating GHG emissions of agriculture through decreased GHG emissions from soils and increased soil organic carbon (SOC) stocks (Smith *et al.*, 2008). A range of agricultural practices are promoted to increase SOC stocks, such as the crop diversity in the rotation, residue return, cover crops, agroforestry or no tillage (Smith *et al.*, 2008; Stockmann *et al.*, 2013; Paustian *et al.*, 2016). While extensively reviewed at the global scale (see previous references) or at the national and regional scale (*e.g.* VandenBygaart *et al.*, 2008; Pellerin *et al.*, 2013), there is still a large uncertainty on the potential SOC storage rate of the different practices. The effect of some of these practices has been re-evaluated with smaller SOC storage rates than previously thought, *e.g.* no-tillage (Luo *et al.*, 2010a; Virto *et al.*, 2012). Alternative cropping systems that have been identified to provide environmental benefits combine different practices. Organic agriculture, in which synthetic fertilizers and pesticides are absent, combines the use of organic fertilizers and green manures to provide nutrients to crops with frequent tillage to control weeds. Conservation agriculture combines the absence of tillage with permanent plant cover and more diversified rotations. Low input systems, also called integrated systems, combine practices applied in both organic and conservation systems with better adjusted fertilization, diversified rotations compared to conventional systems and reduced tillage frequency. Most of the literature dealing with SOC storage in soils concerns agricultural practices rather than cropping systems.

Cropping systems and agricultural practices may increase SOC stocks by either increasing OC inputs to soil or decreasing OC outputs by mineralization or by erosion. In fact, very few studies allow to compare the contribution of these two levers, increasing OC inputs and decreasing OC outputs, to changes in SOC stocks with management. Two recent studies suggest that increased SOC storage is due to increased OC inputs and not to modified mineralization rates of SOC. In an alley cropping agroforestry system, Cardinael *et al.* (2016) found that biomass-C inputs to soil were increased compared to a reference plot thanks to the tree rows and showed in a modelling exercise that these inputs could explain the observed increases in SOC stocks in the agroforestry plot. In a previous study (Autret *et al.*, 2016), we monitored SOC stocks after the implementation of alternative cropping systems, *i.e.* low input (LI), organic (ORG), conservation agriculture (CA), compared to a conventional cropping system (CON). We found that in 17 years, SOC stocks were increased in the CA and to a lesser extent in the ORG system. OC inputs to soil, measured frequently from the start of the experiment, were increased in the

CA system compared to the conventional system. The model AMG (Saffih-Hdadi & Mary, 2008) was able to reproduce the evolution of SOC stocks in the 4 systems with a single decomposition constant. This suggests that mineralisation rates of SOC are not affected by the cropping system, and in particular are the same in systems with or without tillage.

No tillage is however considered to decrease SOC mineralization rates compared to conventional tillage because of less favourable local climatic conditions and better physical protection of organic matter in soil aggregates (Balesdent *et al.*, 2000). Indeed, using ^{13}C natural abundance, the mean residence time of SOC under no tillage was found to be 2.1 times higher (Balesdent *et al.*, 1990), 1.9 time higher (Ryan *et al.*, 1995), and 1.7 times higher (Six *et al.*, 1998) under no tillage than under full inversion tillage for the equivalent of the ploughed layer (Paustian *et al.*, 2000). Modelling the dynamics of SOC in long term experiments, the decay rate constants (k) of SOC under no tillage were found to be 0.73 (Huggins *et al.*, 2007), 0.48 ± 0.12 (Chatskikh *et al.*, 2009) and 0.88 (Dimassi, 2013) of that under full inversion tillage, also for the equivalent of the ploughed layer. Another way to estimate the decomposition rate of SOC is to measure mineralization rates in incubation experiments. However, published results are very variable and are often based on very superficial layers of soil (*e.g.* 0-5 or 0-10 cm) biasing the analysis since these layers are relatively enriched in SOC and in easily decomposable organic matter under no till. Incubation experiments often use disturbed soil samples, *i.e.* the initial soil structure is disrupted and soil samples are sieved. This disruption was shown to increase the mineralization rates of SOC and SON (Balesdent *et al.*, 2000) also providing an explanation for the effects of tillage on mineralization of soil organic matter (Beare *et al.*, 1994), *i.e.* to protect soil organic matter from decomposition in aggregates not frequently disturbed by tillage operations nor exposure to rain at the soil surface (Balesdent *et al.*, 2000).

Here, we aimed at testing the conclusion of Autret *et al.* (2016) that mineralization rates of SOC were unaffected under alternative cropping systems in a temperate Luvisol. We hypothesized that mineralization rates of C and N would be the same in soil samples from the different cropping systems. To test this, we sampled soil in the surface layer (0-27 cm) in plots under the different cropping systems, incubated the soil in the laboratory, measured the net C and N mineralization over time, *i.e.* the gross mineralization minus N immobilization by soil microorganisms. As soil was disturbed and sieved prior to incubation, we also measured mineralization rates on incubated intact cores to evaluate the effect of this disruption, hypothesising that mineralization rates would be higher in disrupted soils.

3.2 Materials and methods

3.2.1 Site and soil characteristics

We investigated soil samples from the long term field experiment of “La Cage” located in Versailles, France (48°48' N, 2°08' E). The experiment was started in 1998 to evaluate the agronomic, economic and environmental performances of low input (LI), conservation agriculture (CA) (direct seeding with permanent plant cover called cover crop) and organic farming (ORG) systems compared to conventional farming (CON) (Balabane *et al.*, 2005; Debaeke *et al.*, 2009; Pelosi *et al.*, 2009; Henneron *et al.*, 2014). Long term annual mean temperature is 11.3 °C and annual rainfalls average 627 mm. The soil is a well-drained deep Luvisol (IUSS Working Group WRB, 2006), which mean particle size distribution is 17 % clay, 56 % silt and 27 % sand. At the start of the experiment, the ploughed layer (0-25 cm) had a C/N ratio of 9.6, a pH of 7.38 and a mean organic C content of 9.49 g kg⁻¹, against 10.10, 10.57, 10.11 and 16.52 g kg⁻¹ in CON, LI, ORG and CA respectively in 2014 (Autret *et al.*, 2016).

3.2.2 Cropping systems

The field experiment is arranged in a randomized complete block design, divided into two blocks, themselves divided into four plots for each cropping system. Each plot is divided into two subplots of 0.56 ha, so that two different crops of the crop rotation are present each year, wheat being grown every year in one of the two subplots. A detailed presentation of crop rotations, soil management and fertilization was described by (Autret *et al.*, 2016). Briefly, crop rotation was mainly based on rapeseed (*Brassica napus L.*), winter wheat (*Triticum aestivum L.*) and spring pea (*Pisum sativum L.*) but differed in CA and ORG for some years, with the replacement of rapeseed by maize (*Zea mays L.*) in CA or the introduction of alfalfa (*Medicago sativa*) in CA and ORG. The CA system was characterized by the presence of a permanent soil cover, composed of fescue (*Festuca rubra*) until 2010 and alfalfa thereafter, grown under the main crop and during the intercrop. The CON treatment is characterized by a soil and crop management representative of the Paris Basin cereal production, with annual soil ploughing, the absence of organic amendment, a mineral N fertilization (~143 kg N ha⁻¹ yr⁻¹) and a systematic use of pesticides. Compared to the CON system, the LI cropping system is managed with a less intensive soil tillage (one out of two years), a reduced mineral N fertilization (~114 kg N ha⁻¹ yr⁻¹) and a limited use of pesticides. The CA system had a reduced mineral N fertilization (~104 kg N ha⁻¹ yr⁻¹) as well, but the absence of soil tillage resulted in a systematic

use of herbicides to destroy preceding main crops, cover crops and weeds. The ORG system was managed according to the European specifications for organic farming, *i.e.* without any application of synthetic N fertilizer or pesticides. Thus, soil ploughing frequently occurred in ORG to control the weed development.

3.2.3 Soil sampling and analysis

Soil sampling was performed in February 2016 in all plots of the experiment, under rapeseed and winter wheat. Two types of soil samples were collected (Figure 3.1):

- disturbed soil samples: 6 soil cores were taken randomly per plot in both blocks, down to 27 cm depth using a cylindrical probe of 6 cm diameter. Soil cores were homogenized (or “disturbed”) and pooled for each plot, leading to a total of 16 soil samples of approximately 6 kg each. Coarse residues and visible roots were removed manually;
- undisturbed soil samples: 6 soil cores were taken randomly in 8 plots corresponding to the CON and CA systems, down to 27 cm depth using polyethylene (PE) tubes of 6 cm diameter. The 48 soil cores were kept intact (or “undisturbed”) in their PE tubes and visible crop residues located on the soil surface were removed manually. These undisturbed soil cores were sampled in order to evaluate the effect of no tillage (CA) on C and N mineralization with regard to a conventionally ploughed system (CON).

The total 64 soil samples (16 disturbed and 48 undisturbed) were kept in a cold room at 4°C during 2 weeks until the start of the incubations, in order to limit the process of mineralization.

3.2.4 Incubations

The details of the incubation conditions and the preparation of the soil samples are given in Figure 3.1. A one-week pre-incubation was realized in order to reactivate the microbial flora and avoid any flush of C and N mineralization. Soil samples were incubated at a temperature of 20°C and a soil moisture adjusted to 18.3% (g/g, equivalent to $pF = 3$), for a total incubation period of 4 months.

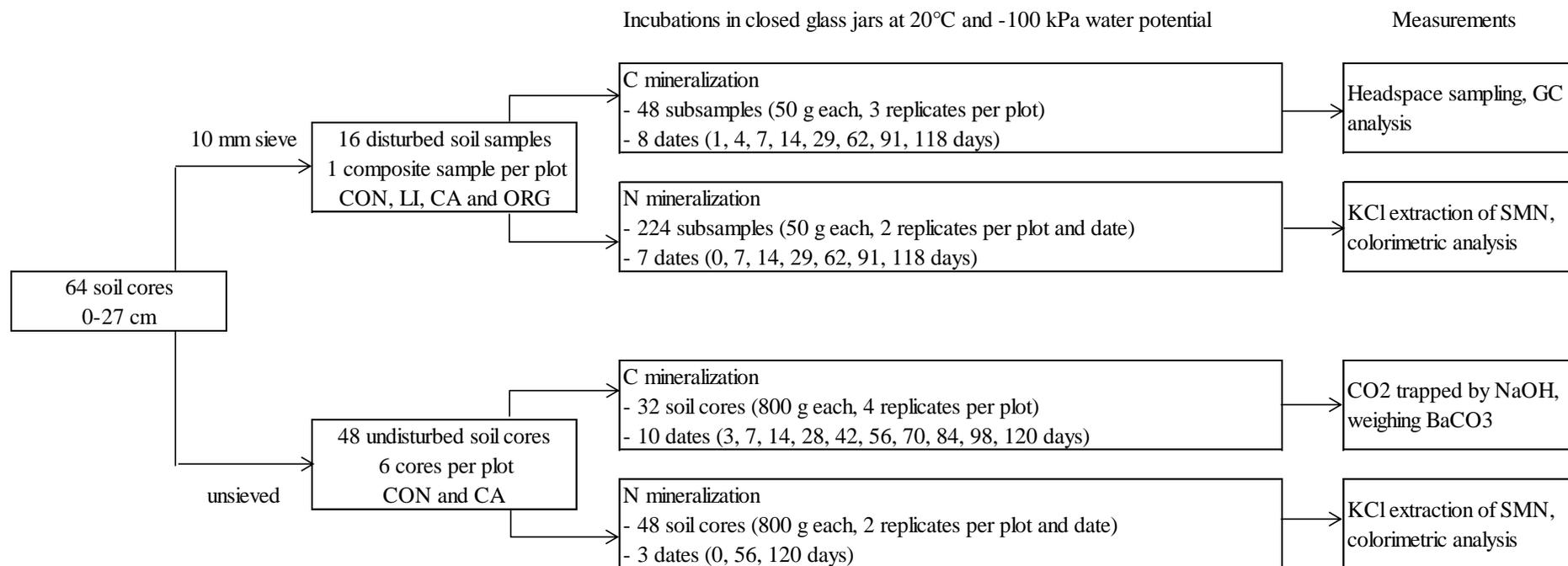


Figure 3.1. Scheme of the soil sampling, sieving, incubation and measurements protocol for each type of soil sample (undisturbed and disturbed).

The 16 disturbed soil samples were forced through a 10 mm sieve and slightly air-dried for 72h. Soil moisture was measured on a 5 g soil sample and adjusted for required incubation conditions. For C mineralization, three soil samples of 50 g each were placed in 0.5 L jars hermetically closed, with a small beaker of water to avoid soil drying. The follow-up of CO₂ emissions was carried out over 8 dates (day 1, 4, 7, 14, 29, 62, 91 and 118). Net N mineralization was determined on three replicated soil samples at three dates (day 0, 29 and 118), and on a single soil sample at four other dates (day 7, 14, 62 and 91), given the destructive nature of the measurement.

Undisturbed cores were prepared for incubation as follows. The soil cylinders were capped by the bottom with a cheesecloth cover, taped to the outer wall of the cylinder. Each sample was placed in a 2 L incubation jar on a 10 mm thick crosspiece in order to allow free gas diffusion through the base of the core. A 10 mL cup of water was placed in the incubation jar to avoid soil drying. Two beakers of 20 mL of NaOH (1M) were also inserted in the jar in order to trap the CO₂ emitted during each incubation interval, allowing to trap 225 mg CO₂-C kg⁻¹ soil. This amount appeared to be satisfactory *a posteriori* since the maximum emission recorded was 93 mg CO₂-C kg⁻¹ soil. Similarly, the O₂ concentration calculated in the jars' atmosphere never dropped below 11%, ensuring permanent oxic conditions in all incubated soil cores. CO₂ measurements were made at days 3, 7, 14, 28, 42, 56, 70, 84, 98 and 120. The destructive mineral N measurements were carried on 8 replicated soil cores at three dates: day 1, day 56 and day 120. The moisture content during the incubation, measured *a posteriori*, appeared to be slightly higher than in the disturbed soils: it was 20.8% in the CON system and 21.8% in the CA system on average during the 120 days.

3.2.5 C, N and microbial biomass

The 16 disturbed soil samples were analysed for their characteristics, each in triplicate aliquots. Organic carbon and total nitrogen concentration were measured with a CN elemental analyser (Carlo Erba NA 2000, Milan, Italy) after drying the samples and ball milling them until they passed a 200 µm mesh sieve. The microbial biomass-C was determined on field moist samples by fumigation-extraction (Vance *et al.*, 1987).

3.2.6 C and N mineralization measurements

The measurement of C mineralization differed between the undisturbed and the disturbed soil samples because of laboratory constraints. The CO₂ emitted in the undisturbed soil samples was trapped into NaOH solution. A solution of barium dichloride (BaCl₂) 1M was then added, leading to the formation of a precipitate (BaCO₃) which was filtered onto a glass fiber filter under vacuum, dried out and weighed. The amount of C mineralized (*mC*) was calculated as follows:

$$mC = mBaCO_3 \cdot \frac{M_C}{M_{BaCO_3}} \quad (3.1)$$

where *mBaCO₃* is the mass of barium carbonate, *M_C* and *M_{BaCO₃}* are the molar mass of C (12 g mol⁻¹) and BaCO₃ (197 g mol⁻¹), respectively.

For the disturbed soil samples, an air sample extracted from each incubation jar was injected into a gas chromatograph MICRO-GC (Agilent 3000A, Santa Clara, CA, USA), separating gases according to their molecular weight. The headspace CO₂ concentration was measured by a thermal conductivity detector (GC-TCD), expressed in parts per million and converted into mg C. The residual CO₂ in the jars' atmosphere was removed at the beginning of incubation and after each measurement by flushing jars with reconstituted CO₂-free air. The agreement between the two methods of measurement of mineralized C was checked successfully in the CON treatment using replicated samples.

The net N mineralization was determined similarly in disturbed and undisturbed soil samples. At each measurement date, the soil samples were taken from the incubations jars, placed in the freezer and thawed subsequently at 4°C during 24h before soil analysis. Mineral N was extracted on a 50 g soil sample after 30 minutes shaking with 100 mL of a KCl solution (1M). The mineral N concentration - ammonium (NH₄) and nitrate (NO₃) - was measured by continuous flow-colorimetry (Skalar Analytical, The Netherlands). Net N mineralization from incubated soil samples was calculated by subtracting the average mineral N content at the start of the incubation from the mineral N content measured at each measurement date.

3.2.7 Simulation of C and N mineralization kinetics

The C and N mineralization kinetics determined over the 120-days incubation was fitted to a model for each cropping system and each incubation technique (disturbed or undisturbed)

separately. On the basis of the results, a linear-exponential model with two pools was used to simulate the absolute and specific C mineralization, along with the C mineralization per unit of microbial C. The cumulative amount of mineralized C was fitted to the following model:

$$C = A \cdot (1 - e^{-k \cdot t}) + B \cdot t \quad (3.2)$$

where C is the absolute or relative amount of C mineralized (mg C kg⁻¹ soil or mg C g⁻¹ SOC or mg C mg⁻¹ Cmic), A is the rapidly mineralizable C (same unit), k is its mineralization rate constant (day⁻¹), t is the incubation time (days) and B the mineralization rate of the slow mineralizing pool (mg C kg⁻¹ soil, mg C g⁻¹ SOC or mg C mg⁻¹ Cmic). The optimization of parameters A , B and k was made with the Excel solver tool by minimizing the root mean square error (RMSE) between observed and simulated data, expressed in mg C kg⁻¹ soil, mg C g⁻¹ SOC or mg C mg⁻¹ Cmic:

$$RMSE_j = \sqrt{\frac{1}{n_j} \sum_{i=1}^{n_j} (QC_{ij} - \widehat{QC}_{ij})^2} \quad (3.3)$$

where n_j is the number of observations in treatment j , QC_{ij} and \widehat{QC}_{ij} are the observed and estimated mineralized C for the observation i and treatment j . The fitting procedure included four steps:

- Step 1: A , B and k were optimized for each plot and for disturbed (O1) and undisturbed (O'1) soil samples;
- Step 2: A and B were optimized for each plot, for disturbed (O2) and undisturbed (O'2) soil samples. k was set to the average value found in step 1 for each cropping system;
- Step 3: A and B were optimized for each plot for disturbed (O3) and undisturbed (O'3) soil samples. k was set to the average value found in step 2 for all cropping systems;
- Step 4: it concerned only C mineralization kinetics in undisturbed soil cores (O'4). A and B were optimized for each plot. k was set to the average value found in O3 for all systems.

N mineralization kinetics was determined on a reduced number of points and appeared to follow a more linear evolution than C kinetics. Therefore, absolute or specific N mineralization was described by a simple linear model:

$$N = D \cdot t \quad (3.4)$$

where D is the mineralization rate ($\text{mg N kg}^{-1} \text{ d}^{-1}$ or $\text{mg N g}^{-1} \text{ SON d}^{-1}$).

3.2.8 Statistical analysis

All the data were analysed under version 3.1.2 of R (R Core Team, 2014). Although the number of true replicates in the experiment was low (two randomized blocks), each of the sub-plots was considered as a replicate, thus producing four pseudo-replicates per cropping system. We tested the effect of the four different cropping systems on several variables: C and N mineralized on day 120, parameters of the mineralization curves, soil C and N contents, C/N ratio and microbial biomass-C at the start of the incubation. We also tested the effect of the previous and current crops on these variables, but did not detect any significant effect on C and N mineralization (data not shown). The mean of the three replicates of each subplot and of each variable was used for statistical analysis, *i.e.* four values per cropping system and per date of measurement for disturbed samples. For undisturbed samples, the mean of the four soil cores from each subplot was used, *i.e.* four values per cropping system and per date of measurement, except for mineral N at day 0 and day 56, where only one soil core was available for each subplot. All data were analysed by ANOVA, and the LSD test of the package *agricolae* (De Mendiburu, 2014) was applied for comparison of treatment means when significant effects were observed, with block as random variable. The normal distribution of model residues was checked using the Shapiro-Wilk and Levene tests. In case of discordance, the non-parametric test of Kruskal-Wallis was used, followed by means comparison using the `kruskal.test` from the *agricolae* package.

3.3 Results

3.3.1 Soil characteristics

The surface layer of soil samples in the different systems exhibited the same characteristics (Table 3.1), except in the conservation agriculture system with significantly higher organic carbon and total nitrogen contents ($p < 0.05$). Microbial biomass in absolute values as well as in proportion of total organic carbon tended to be higher in the CA system but not significantly.

Table 3.1. Soil organic C (SOC), total N (SON) and microbial biomass C (MBC) concentrations measured in disturbed soils (0-27 cm). Values in brackets are standard deviations. Letters indicate significant differences between cropping systems ($p < 0.05$).

Cropping system	SOC		SON		MBC		MBC/SOC	
	g kg ⁻¹				mg C kg ⁻¹		%	
CON	10.9 (1.2)	b	1.10 (0.12)	b	127 (11)	a	1.2% (0.1%)	a
LI	10.8 (1.5)	b	1.09 (0.14)	b	155 (50)	a	1.4% (0.3%)	a
CA	13.0 (1.6)	a	1.33 (0.15)	a	202 (20)	a	1.6% (0.1%)	a
ORG	10.7 (0.5)	b	1.10 (0.05)	b	151 (14)	a	1.4% (0.2%)	a

3.3.2 Mineralization of C and N in disturbed soils

At the end of incubation of disturbed soils (day 120), the cumulative amounts of C and N mineralized expressed per unit of soil mass did not differ significantly between the four systems, whatever the crop (Figure 3.2.a). The specific rates of mineralization, expressed per unit of soil C (Figure 3.2.c) or microbial biomass carbon (Figure 3.2.e) did not differ either between systems.

Carbon mineralization kinetics were close to each other. They were satisfactorily modelled with the two pools model, with small RMSEs. The decomposition rate optimized for each plot in simulation O1 (differing in cropping system and in the 2016 crop) was significantly smaller in LI compared to CA, but they did not significantly differ from CON and ORG (Table 3.2). Conversely, pool sizes did not differ between system in O1, whatever their unit of expression. Optimizing mineralization rates for each plot with a single mineralization rate per system (simulation O2) led to the same result as in O1, while differences were detected for pool sizes. Hence, a significantly smaller B parameter was found in LI compared to CA and ORG when expressed in mg kg⁻¹ d⁻¹ and in mg g⁻¹ SOC d⁻¹, while the A parameter was significantly higher in LI than in CA when expressed in mg g⁻¹ SOC and in g g⁻¹ MBC. Parameters A and B optimized with a common k value of 0.028 d⁻¹ for all systems (simulation O3) did not differ significantly between systems.

The N mineralization kinetics were well simulated by a linear model, the slope of which was similar between systems (Table 3.3). It is however noticeable that the treatments CA and ORG seemed to have a greater mineralization rate than the other two cropping systems, consistently with the trend found for C mineralization (Figures 3.2.a and b). The absence of statistical significance might be due to an insufficient number of replicates. The specific N mineralization rates (per unit of SON) did not differ significantly between systems.

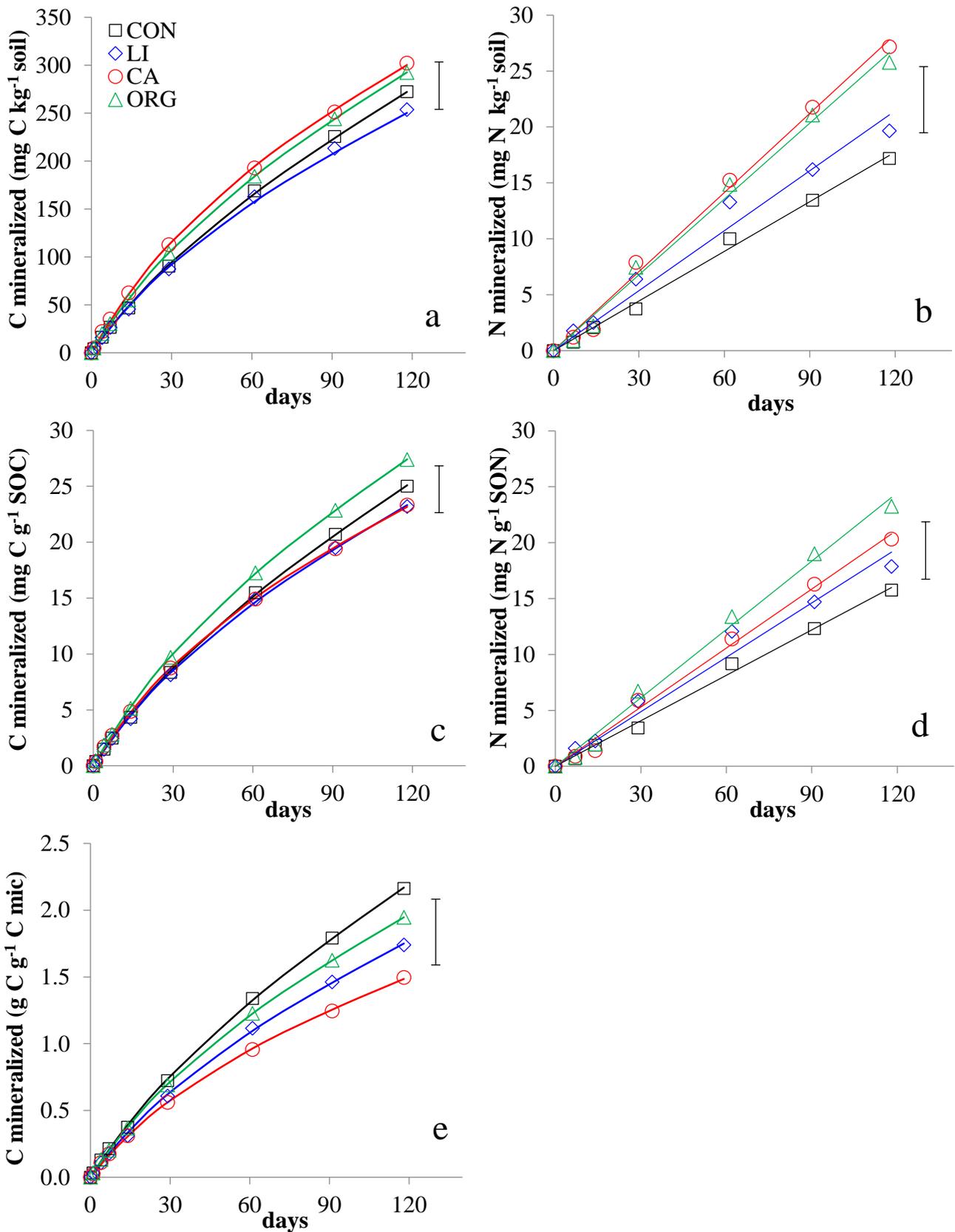


Figure 3.2. C and N mineralization kinetics in disturbed soil samples: a, b) per kg of soil; c, d) per g of SOC or TN; e) per mg of microbial biomass C. Dots are observed values and full lines represent the fitted curves of C and N mineralization (simulation O3, see Table 3.2). Vertical bars represent the mean confidence intervals ($p < 0.05$) at the end of incubation.

Table 3.2. Optimized parameters (A and B) obtained in curve fitting to the C mineralization model (eq. 3.2) of disturbed soil samples. Values in brackets are standard deviations. Letters indicate significant differences between cropping systems ($p < 0.05$).

optimization procedure	cropping system	mineralization rate		absolute C mineralization				specific C mineralization (per unit of SOC)				specific C mineralization (per unit of MBC)								
		k		A	B	RMSE	A	B	RMSE	A	B	RMSE								
		d^{-1}		mg kg ⁻¹	mg kg ⁻¹ d ⁻¹	mg kg ⁻¹	mg g ⁻¹ SOC	mg g ⁻¹ SOC d ⁻¹	mg g ⁻¹ SOC	g g ⁻¹ MBC	mg g ⁻¹ MBC d ⁻¹	g g ⁻¹ MBC								
O1	CON	0.021 (0.010)	<i>ab</i>	150 (103)	<i>a</i>	1.3 (0.5)	<i>a</i>	5.3	12.3 (6.2)	<i>a</i>	0.13 (0.04)	<i>a</i>	0.5	1.16	0.72	<i>a</i>	10.6	4.48	<i>a</i>	0.04
	LI	0.019 (0.005)	<i>b</i>	153 (59)	<i>a</i>	1.0 (0.4)	<i>a</i>	6.4	14.8 (7.1)	<i>a</i>	0.09 (0.03)	<i>a</i>	0.6	1.14	0.76	<i>a</i>	6.7	2.63	<i>a</i>	0.04
	CA	0.039 (0.006)	<i>a</i>	92 (10)	<i>a</i>	1.8 (0.3)	<i>a</i>	4.0	7.1 (0.9)	<i>a</i>	0.14 (0.02)	<i>a</i>	0.3	0.46	0.06	<i>a</i>	8.8	1.06	<i>a</i>	0.02
	ORG	0.028 (0.012)	<i>ab</i>	135 (86)	<i>a</i>	1.5 (0.3)	<i>a</i>	6.5	12.5 (7.9)	<i>a</i>	0.14 (0.03)	<i>a</i>	0.6	0.88	0.54	<i>a</i>	10.1	2.52	<i>a</i>	0.04
O2	CON	0.022 (0.010)	<i>ab</i>	100 (48)	<i>a</i>	1.5 (0.3)	<i>ab</i>	6.1	9.2 (4.6)	<i>ab</i>	0.14 (0.03)	<i>a</i>	0.6	0.81	0.45	<i>ab</i>	12.0	4.48	<i>a</i>	0.05
	LI	0.018 (0.005)	<i>b</i>	146 (27)	<i>a</i>	1.0 (0.3)	<i>b</i>	6.8	14.0 (1.0)	<i>a</i>	0.09 (0.03)	<i>b</i>	0.6	0.83	0.54	<i>a</i>	8.8	2.63	<i>a</i>	0.10
	CA	0.039 (0.006)	<i>a</i>	90 (17)	<i>a</i>	1.8 (0.4)	<i>a</i>	5.0	6.9 (1.8)	<i>b</i>	0.14 (0.01)	<i>a</i>	0.4	0.45	0.11	<i>b</i>	8.9	1.06	<i>a</i>	0.03
	ORG	0.032 (0.012)	<i>ab</i>	90 (24)	<i>a</i>	1.7 (0.2)	<i>a</i>	7.9	8.4 (1.9)	<i>ab</i>	0.16 (0.02)	<i>a</i>	0.7	0.60	0.18	<i>ab</i>	11.6	2.52	<i>a</i>	0.05
O3	CON	0.028 (0.011)		79 (38)	<i>a</i>	1.7 (0.4)	<i>a</i>	6.5	7.3 (3.7)	<i>a</i>	0.15 (0.03)	<i>a</i>	0.6	0.63	0.35	<i>a</i>	13.2	3.50	<i>a</i>	0.05
	LI	0.028 (0.011)		88 (17)	<i>a</i>	1.4 (0.3)	<i>a</i>	8.0	8.1 (0.5)	<i>a</i>	0.13 (0.03)	<i>a</i>	0.8	0.60	0.13	<i>a</i>	10.0	4.10	<i>a</i>	0.06
	CA	0.028 (0.011)		123 (22)	<i>a</i>	1.5 (0.4)	<i>a</i>	7.7	9.5 (2.4)	<i>a</i>	0.12 (0.02)	<i>a</i>	0.6	0.61	0.15	<i>a</i>	7.6	1.00	<i>a</i>	0.04
	ORG	0.028 (0.011)		102 (27)	<i>a</i>	1.6 (0.2)	<i>a</i>	8.1	9.5 (2.1)	<i>a</i>	0.15 (0.03)	<i>a</i>	0.8	0.68	0.20	<i>a</i>	10.9	1.30	<i>a</i>	0.05

O1: A, B and k optimized for each plot

O2: A, B and k optimized for each plot, with a common k per system

O3: A, B and k optimized for each plot, with a common k for all systems

Table 3.3. Absolute and specific N mineralization rates (parameter D in eq. 3.4) in disturbed soil samples. Values in brackets are standard deviations. Letters indicate significant differences between cropping systems ($p < 0.05$).

cropping system	absolute N mineralization rate		specific N mineralization rate (per unit of SON)	
	mg N kg ⁻¹ d ⁻¹		mg N g ⁻¹ SON d ⁻¹	
CON	0.15	(0.02) a	0.13	(0.03) a
LI	0.17	(0.02) a	0.15	(0.02) a
CA	0.23	(0.07) a	0.18	(0.07) a
ORG	0.22	(0.03) a	0.20	(0.03) a

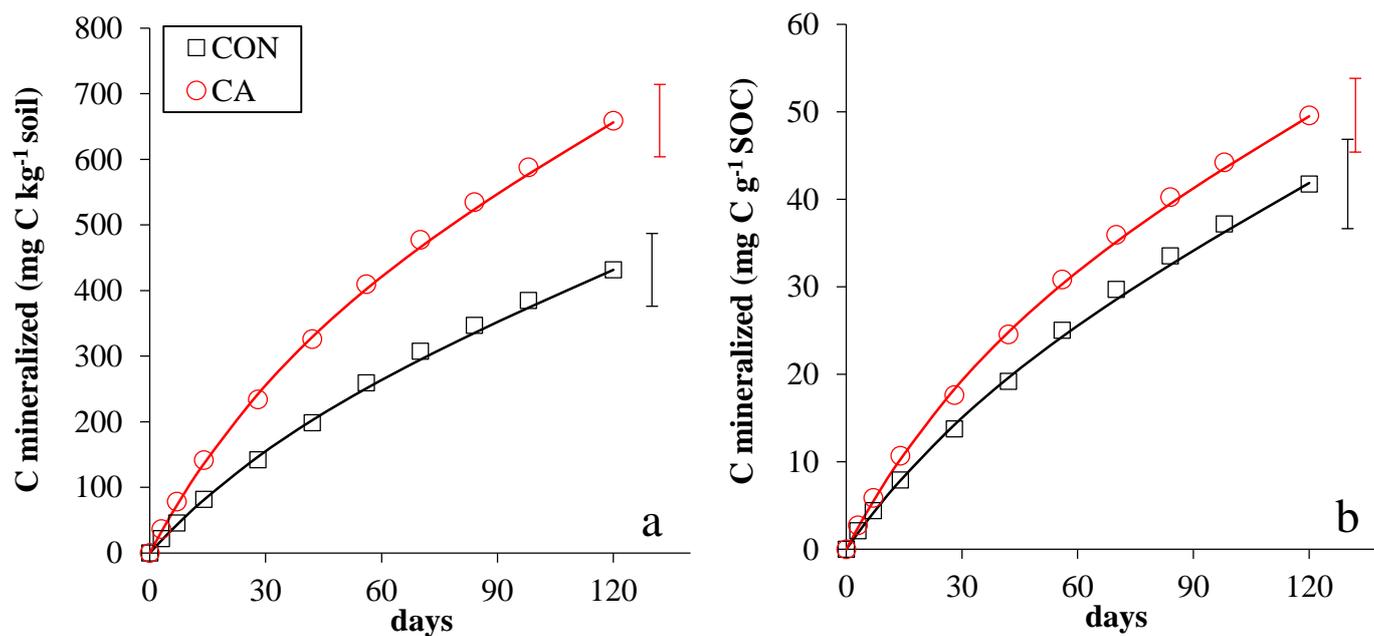


Figure 3.3. C mineralization kinetics in undisturbed soil samples: a) per kg of soil; b) per g of SOC. Dots are observed values and full lines represent the fitted curves of C mineralization (optimization O'4, see Table 3.5). Vertical bars represent the mean confidence intervals ($p < 0.05$).

Table 3.4. Optimized parameters (A, B and k) obtained in curve fitting to the C mineralization model (eq. 3.2) of undisturbed soil samples. Values in brackets are standard deviations. Letters indicate significant difference between cropping systems ($p < 0.05$).

optimization procedure	mineralization rate	absolute C mineralization				specific C mineralization (per unit of SOC)		
		k d^{-1}	A $mg\ kg^{-1}$	B $mg\ kg^{-1}\ d^{-1}$	RMSE $mg\ kg^{-1}$	A $mg\ g^{-1}\ SOC$	B $mg\ g^{-1}\ SOC\ d^{-1}$	RMSE $mg\ g^{-1}\ SOC$
O'1	CON	0.014 (0.009) <i>a</i>	484 (299) <i>a</i>	0.8 (1.1) <i>a</i>	3.7	47 (30) <i>a</i>	0.07 (0.10) <i>a</i>	0.36
	CA	0.021 (0.009) <i>a</i>	462 (194) <i>a</i>	2.2 (1.3) <i>a</i>	5.2	34 (13) <i>a</i>	0.17 (0.10) <i>a</i>	0.39
O'2	CON	0.010 (0.000)	565 (189) <i>a</i>	0.3 (0.4) <i>b</i>	4.1	52 (17) <i>a</i>	0.04 (0.04) <i>b</i>	0.39
	CA	0.021 (0.000)	377 (44) <i>a</i>	2.5 (0.3) <i>a</i>	5.9	28 (4) <i>a</i>	0.19 (0.02) <i>a</i>	0.44
O'3	CON	0.017 (0.000)	246 (83) <i>b</i>	1.8 (0.2) <i>a</i>	4.4	24 (8) <i>a</i>	0.17 (0.03) <i>a</i>	0.43
	CA	0.017 (0.000)	473 (55) <i>a</i>	2.0 (0.3) <i>a</i>	6.0	36 (5) <i>a</i>	0.15 (0.02) <i>a</i>	0.45
O'4	CON	0.028 (0.000)	145 (48) <i>b</i>	2.4 (0.4) <i>b</i>	5.3	14 (4) <i>a</i>	0.24 (0.05) <i>a</i>	0.52
	CA	0.028 (0.000)	280 (34) <i>a</i>	3.2 (0.3) <i>a</i>	6.3	21 (3) <i>a</i>	0.24 (0.02) <i>a</i>	0.47

O'1: A, B and k optimized for each plot

O'2: A, B and k optimized for each plot, with a common k per system

O'3: A, B and k optimized for each plot, with a common k for CON and CA

O'4: A and B optimized for each plot, with k determined previously in O3

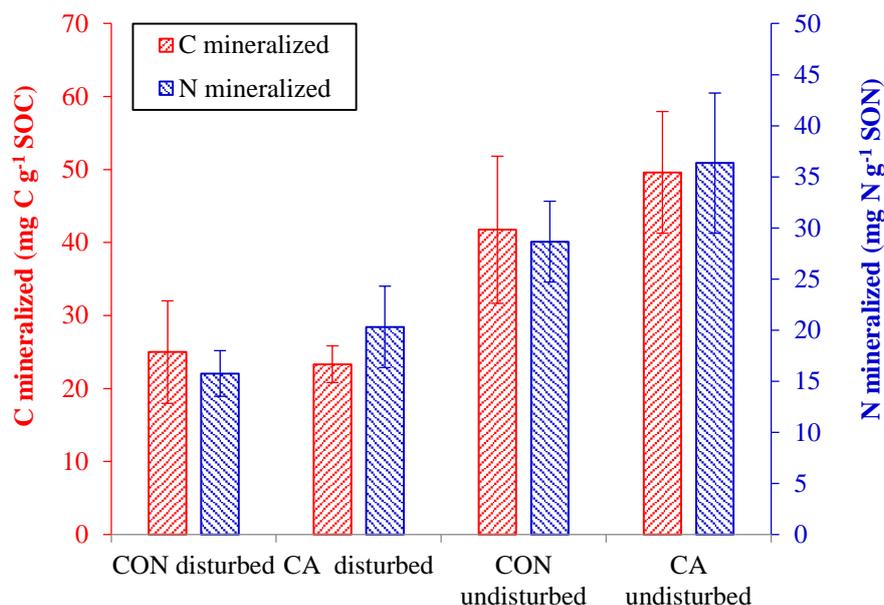


Figure 3.4. Specific amounts of C and N mineralized in disturbed and undisturbed soil samples of CON and CA systems at the last day of incubation for the 0-27 cm soil layer (day 120). Bars represent the standard deviations.

Table 3.5. Absolute and specific N mineralization rates in each layer of undisturbed soil cores (mean rate measured in 15 replicated cores during 120 days). Values in brackets are standard deviations. Letters indicate significant differences in each layer between the two cropping systems ($p < 0.05$).

cropping system	soil layer cm	absolute N mineralization rate			specific N mineralization rate (per unit of SON)		
		mg N kg ⁻¹ d ⁻¹			mg N g ⁻¹ SON d ⁻¹		
CON	0-5	0.27	(0.06)	<i>a</i>	0.25	(0.06)	<i>a</i>
	5-10	0.21	(0.05)	<i>a</i>	0.19	(0.04)	<i>a</i>
	10-27	0.22	(0.06)	<i>a</i>	0.20	(0.06)	<i>a</i>
	0-27	0.23	(0.04)	<i>a</i>	0.24	(0.03)	<i>a</i>
CA	0-5	0.75	(0.21)	<i>b</i>	0.32	(0.09)	<i>a</i>
	5-10	0.49	(0.14)	<i>b</i>	0.31	(0.08)	<i>a</i>
	10-27	0.31	(0.06)	<i>a</i>	0.26	(0.06)	<i>a</i>
	0-27	0.41	(0.08)	<i>b</i>	0.30	(0.06)	<i>a</i>

3.3.3 Mineralization of C and N in undisturbed soils

Undisturbed soil cores from conservation agriculture plots mineralized significantly more carbon (+53% at day 120) than those from conventional plots (Figure 3.3). The difference between the two systems was smaller (+19%) for specific mineralization and not significant. Optimizing decomposition model parameters independently between cropping systems (simulations O'1 and O'2) provided values which did not differ significantly (Table 3.4). This surprising result is attributed to the uncertainty in parameter estimation which is itself due to the correlation between the three parameters. This uncertainty disappeared when a common k value was applied to both systems (simulation O'3): in this case, the A parameter differed markedly between the two systems (with a much greater value for CA system). The difference was even greater when k was set at the value determined in the disturbed cores ($k = 0.028 \text{ d}^{-1}$, simulation O'4) since both A and B were greater in the CA than in the CON system. In contrast, the model parameters of the specific mineralization kinetics (per unit of SOC) did not differ between systems, whatever the optimization procedure. The N mineralization results showed exactly the same trends than the C mineralization (Table 3.5). The absolute N mineralization rate was much greater in CA than CON whereas the specific N mineralization rate was similar in both systems. The separation between soil layers indicated that the difference occurred mainly in the two upper layers (0-5 and 5-10 cm). The upper layer mineralized almost three times faster in the conservation than in the conventional system. No significant difference could be detected in specific N mineralization whatever the layer, indicating that the differences in net N mineralization were strongly related to the variations in SOC and SON contents.

It is noticeable that the amounts of C and N mineralized were much larger in undisturbed than in disturbed soil samples, and this was true in both systems (Figure 4). The effect of soil conditioning (disturbed vs undisturbed) was greater than that of the cropping system: the undisturbed:disturbed ratio varied from 1.67 to 2.13 whereas the CA:CON ratio varied from 0.93 to 1.27.

3.4 Discussion

3.4.1 Physical disturbance effect

In incubation studies, the soil is most often incubated after some degree of disruption of the soil structure and sieving in order to decrease the variability between soil samples and for practical constraints. We hypothesised that disturbing the soil prior to incubation would increase the mineralization of C and N because of a de-protection of the organic matter located within the soil structure, and possibly a better oxygen availability in the sieved soil. Indeed, several experiments comparing the net N mineralization in intact soil cores *vs* sieved soil showed that mineralization rates were increased in sieved soils (Sierra, 1992; Stenger *et al.*, 2002; Ringuelet & Bachmeier, 2002; Li *et al.*, 2013). It is noticeable that all these studies used the Stanford and Smith's leaching technique which does not provide similar conditions of moisture in disturbed and undisturbed soils. Other studies found no effect of physical disturbance on N mineralization rate after the possible initial flush effect (Cabrera & Kissel, 1988; Franzluebbers, 1999; Zhao *et al.*, 2010; Moberg *et al.*, 2013; Curtin *et al.*, 2014). In our study, we observed a negative effect of soil disturbance on C and N mineralization rates which were higher in undisturbed soil cores. Model fitting showed that the size of the labile pool contributing to C mineralization was much larger in undisturbed soil cores (*e.g.* 565 in CON and 377 mg C kg⁻¹ in CA) when a single mineralization rate was set per system (optimization O'2, Table 3.4) than in disturbed samples (100 and 90 mg C kg⁻¹ respectively) (optimization O2, Table 3.2). One possible explanation is a higher abundance of labile SOC in undisturbed soil cylinders than in disturbed soils samples that had been sieved to 10 mm and from which visible coarse plant residues and fine roots had been manually eliminated. At the end of incubation, the undisturbed soil cylinders were manually sorted out and were found to contain biological attributes: germinated seed (average 1.5 per soil core), insect (1.8), living root (0.6), earthworm (0.8). This living or labile organic matter could explain why more C and N was mineralized from undisturbed than disturbed soil samples. Another possible explanation is that our disturbed soil had been pre-incubated for 2 weeks and the flush of mineralization due to disturbance might have been over. Indeed, in several studies where disturbed soil was incubated right after disruption a flux of mineralization was observed for a few days and afterwards the disturbed and undisturbed soil mineralization curves were parallel (Cabrera & Kissel, 1988; Zhao *et al.*, 2010). Another explanation would be the higher soil moisture in the undisturbed soils, that would be more favourable to mineralization.

3.4.2 Effect of cropping system on mineralization rates

We observed no significant effect of the cropping system on the absolute rate of C and N mineralization, except for undisturbed soil cores in which the conservation agriculture had a higher rate than the conventional system. In fact, there was a consistent trend in having higher rates of C and N mineralization in disturbed soil samples of CA and ORG systems compared to CON and LI but which was not significant. The four systems did not show any difference in their specific rate of mineralization, expressed *vs* SOC, SON or microbial biomass C. This result is fully consistent with the hypothesis made by Autret *et al.* (2016) who simulated the long-term variation of SOC in the same experiment. The authors found no significant SOC change in the LI and CON systems, a moderate increase in the ORG system and a high increase in the CA system after 17 years. They could simulate satisfactorily the evolution of SOC stocks by accounting for the crop residues inputs using a two pools model and assuming a single decomposition constant value for all systems. This assumption is confirmed by the present study, both for C and N mineralization and for disturbed and undisturbed soil samples. It suggests that the main driver of the SOC variation among these four cropping systems is the amount of organic inputs (which varied widely between systems) rather than the variation in soil tillage (varying from permanent no till to annual ploughing) which may not affect the specific mineralization rate constants (Autret *et al.*, 2016).

Several previous studies showed higher mineralization rates of C and N in organic cropping systems than in conventional ones, but these could be explained by the applied organic fertilizers inputs (Gunapala & Scow, 1998; Carpenter-Boggs *et al.*, 2000). Regarding the effects of tillage, most studies focused on the superficial soil layers, *e.g.* 0-5 cm, and found higher mineralization rates of C and N in no till soils, easily ascribed to the relative enrichment in organic matter and particulate organic matter content in soil surface layer (*e.g.* Beare *et al.*, 1994). However, no difference in C and N mineralization was observed between tilled and untilled soils when deeper layers were included in the comparison (Oorts *et al.*, 2006; Jacobs *et al.*, 2010) and results are contrasted when considering the whole ploughed layer (Beare *et al.*, 1994; Franzluebbers *et al.*, 1995).

The similarity in mineralization rates observed in the four cropping systems suggests that either the decomposability of the organic matter is similar in the different cropping systems or that the environmental conditions for decomposers are the same. Regarding the decomposability of the organic matter, the organic inputs to soil are quite similar in the different cropping systems

as wheat is cropped on year over two, and legumes (pea, alfalfa) are cropped in all systems, rapeseed is cultivated in CON, LI and ORG (Autret *et al.*, 2016). There are slight differences however, as the frequency of rapeseed, alfalfa and pea is not the same in the different systems, and there is no rapeseed in CA but maize was cultivated instead in *ca* 10% of the cropping seasons. There is no organic fertilization in either plot of the experiment. No major differences in the biochemical quality of the plant inputs are then expected between the different cropping systems. Regarding the environmental conditions, the soils have close pH, similar C/N ratio. However, the frequency of tillage is very contrasted between the systems and is expected to affect soil structure and relatively increase the physical protection of organic matter in the CA soils compared to others (Six *et al.*, 2000). While a more stable aggregated structure was observed early in the CA soil compared to others (Balabane *et al.*, 2005), there was no difference in bulk density in 2014 for the \approx 0-30 cm layer (Autret *et al.*, 2016). In the incubated undisturbed cores, two opposed processes might explain the similar mineralization rates of C and N in the CON and CA cylinders: there was more coarse labile organic matter in the CA cylinders (see above) but the soil structure may have been less favourable to decomposition. In the case of disturbed soil, sieved to 10 mm, the similar mineralization rates of C and N over 120 days could mean either that there is no difference in the importance of physical protection among systems, or that the physical protection essentially affects organic matter with residence times much longer than captured in a 120 days incubation (with an average k of 0.028 d^{-1} , the turnover time of the labile pool was of 36 days, Table 3.2).

Our results suggest that the observed increased C and N stocks in the conservation agriculture system at La Cage are essentially due to the observed increased biomass inputs (Autret *et al.*, 2016). However, in the case of the organic agriculture system, the observed small increase of SOC stocks in 17 years, along with smaller OC inputs than in the CON system (Autret *et al.*, 2016) and mineralization rates of C and N similar to that in the CON system raise a problem. Similar observations of maintained or increased SOC stocks with reduced OC inputs in legume based organic agriculture were previously made (Gregorich *et al.*, 2001; Syswerda *et al.*, 2011). One possible explanation lies in the physiology of microorganisms. Kallenbach *et al.* (2015) recently showed that the carbon use efficiency of soil microorganisms was higher in an organic agriculture trial than in its conventional agriculture reference plot. This was explained by changes in the microbial community structure as well as changes in the physiology of microorganisms in nutrient poor environments. For a given amount of OC inputs to soil, a higher carbon use efficiency would result in more OC being left, *i.e.* stored in soil for an

unchanged rate of mineralization. This process requires more investigations in the alternative cropping systems studied here (ORG, LI, CA), all characterized by a reduction in fertilizer inputs to soil and for the ORG and CA by a higher proportion of legumes being grown either as a cash crop or as a cover crop.

3.5 Conclusion

The effects of alternative arable cropping systems on soil organic matter can result from modified organic inputs to soil or modified rates of mineralization of C and N from soil. Here we found that soils from alternative arable cropping systems in temperate area, *i.e.* a legume based organic agriculture system and a conservation agriculture system, that resulted in increased carbon storage in soil over 17 years compared to a conventional one, exhibited similar mineralization rates of C and N when measured in incubation. This suggests that the observed increased SOC stocks are due to increased biomass returns to soil or to changes in microbial physiology. Overall it suggests that increasing biomass returns to soil, as above ground or below ground plant material is an efficient strategy to increase soil organic carbon stocks, both to maintain and increase soil quality and to mitigate climate change. For the later objective a full greenhouse gas balance is indeed needed for alternative arable cropping systems.

3.6 Acknowledgements

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CHAPTER

4.

Can alternative cropping systems mitigate nitrogen losses and improve GHG balance? Results from a 19-yr experiment in Northern France

Article in preparation for *Agronomy, ecosystem and environment*

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Highlights

- ▶ N fate and GHG balance were investigated in four arable cropping systems over 19 years
- ▶ Conventional (CON), low input (LI), conservation (CA) and organic (ORG) systems
- ▶ N recovery ranked as ORG > LI > CON > CA and gaseous N losses in the opposite order
- ▶ CA system resulted both in high C and N storage in soil and high N₂O emissions
- ▶ GHG balance ranked as CON > LI > CA > ORG, ORG being the most favourable system.

Abstract

Alternative cropping systems are promoted to reduce nitrogen (N) losses in the environment and mitigate greenhouse gas (GHG) emissions. However, these supposed benefits are not fully known, rarely studied together and on the long term. Here, we studied the N inputs, N exports, soil organic N (SON) storage, N leaching, gaseous N emissions and GHG balance in a 19-yr field experiment comparing four arable cropping systems without manure fertilization, under conventional (CON), low-input (LI), conservation agriculture (CA) and organic (ORG) managements. The annual N surplus, i.e. the difference between total N inputs and exports, was lowest in LI with $47 \text{ kg ha}^{-1} \text{ yr}^{-1}$, intermediary for CON and ORG with 67 and $68 \text{ kg ha}^{-1} \text{ yr}^{-1}$ and highest in CA with $181 \text{ kg ha}^{-1} \text{ yr}^{-1}$. CA and ORG received high amounts of N derived from biological fixation from alfalfa. The annual SON storage rates markedly differed between cropping systems: $\text{CA} (54 \text{ kg ha}^{-1} \text{ yr}^{-1}) > \text{ORG} (31) > \text{CON} (13) = \text{LI} (3)$. N leaching, calculated using soil mineral N measurements, reached an average of $20 \text{ kg ha}^{-1} \text{ yr}^{-1}$ and did not significantly differ between treatments. The gaseous N emissions (volatilization + denitrification), calculated as the difference between N surplus, SON storage and N leaching, ranged from $19 \text{ kg ha}^{-1} \text{ yr}^{-1}$ in ORG to $106 \text{ kg ha}^{-1} \text{ yr}^{-1}$ in CA. N_2O emissions were continuously monitored with automatic chambers during 3.3 years. They varied from $1.33 \text{ kg ha}^{-1} \text{ yr}^{-1}$ in ORG to $4.24 \text{ kg ha}^{-1} \text{ yr}^{-1}$ in CA system and were highly correlated with gaseous N emissions. The GHG balance varied widely between systems: it was highest in CON and LI, with 1989 and $1443 \text{ kg CO}_{2\text{eq}} \text{ ha}^{-1} \text{ yr}^{-1}$ respectively. In CA, the GHG balance was much more favourable ($124 \text{ kg CO}_{2\text{eq}} \text{ ha}^{-1} \text{ yr}^{-1}$), despite important N_2O losses which partly offset the benefit of SOC storage. ORG was the system with the smallest GHG balance ($-149 \text{ kg CO}_{2\text{eq}} \text{ ha}^{-1} \text{ yr}^{-1}$), acting as a CO_2 sink in the long term. Similar trends were observed when GHG was expressed per unit of N input or N exported. The four agricultural systems dissimilarly impacted the N fate which could not be predicted by the N surplus. Complementary predictors of N losses and GHG balance are required to obtain a true overview of the C and N environmental impacts of cropping systems.

4.1 Introduction

The objective of increasing crop yields to meet the worldwide increasing food demand has put pressure onto the use of nitrogen (N) fertilizers since sixty years, the amount of synthetic N fertilizer applied being multiplied by 7 between 1960 and 1995 (Tilman *et al.*, 2002). However, the N use efficiency (NUE), *i.e.* the ratio between crop N offtake and N inputs, is often less than 50% (Tilman *et al.*, 2002). The unrecovered N in the crop can be stored in the soil or released in the environment throughout the “nitrogen cascade” (Galloway *et al.*, 2003) as dinitrogen (N₂), nitrogen oxides (NO_x), nitrous oxide (N₂O), ammonia (NH₃) and nitrate (NO₃). These N losses cause different types of damages for the environment, with eutrophication of rivers, algal blooms in estuaries or slimes under forest (Sutton *et al.*, 2011) and for human health (WHO, 2013; Habermeyer *et al.*, 2015). In addition, the emission of greenhouse gases (GHG) such as N₂O are of major concern, because of their high global warming potential of 296 over a 100-year time span.

The awareness of this situation led to the implementation of regulations or initiatives such as the Nitrate Directive in Europe (91/676/CEE), which aims at reducing N losses from agriculture to the groundwater, or the “4 per 1000” initiative launched by the COP21 in 2015 (<http://4p1000.org>) in order to yearly increase SOC stocks by 4‰. To meet these objectives, alternative farming practices have been promoted, such as the reduction of mineral N fertilization, the establishment of catch crops, the cultivation of legume crops as organic N fertilizer, the introduction of perennial crops in arable systems, the suppression of tillage... An ideal cropping system combining these practices would result in high N exportations, high NUE and low N surplus (Eurostat, 2016).

Most studies focused on evaluating separately various cropping practices with regard to N losses in the environment. Hence, straw incorporation, catch crops and reduced N fertilization practices are known to reduce N leaching (Beaudoin *et al.*, 2005; Constantin *et al.*, 2010; Hansen *et al.*, 2015), while gaseous N emissions can be decreased by reduced fertilization and increased by no-till (Constantin *et al.*, 2010). These improved practices are often clustered in alternative farming systems, such as low-input farming, organic farming or conservation agriculture. The combination of these practices may lead to a harder understanding of alternative cropping systems impacts on the N flows. Although they are generally assumed to be environmental-friendlier than conventional farming systems, because of their low nutrient inputs, improved biodiversity and/or low soil disturbance, the biochemical processes involved

in N fate still need to be clarified. While the positive effect of the reduced fertilization on N leaching is commonly accepted, the effect of organic farming and conservation agriculture is still a matter of controversy. Conservation agriculture is characterized by no-tillage and permanent living mulch growing under the main crop (Soane *et al.*, 2012) and its impact on N flows is unclear. For example, it may either result in a reduced N leaching by an immobilization of N of the topsoil layer during crop residues decomposition or increase it because of preferential water flows occurring under no-till (Kay *et al.*, 2009). Palm *et al.* (2014) report inconsistent results concerning N₂O emissions which could be increased with a higher moisture at soil surface in CA or decreased in the long term. In organic farming systems, N fertilization is mainly ensured by organic fertilizers and crop residues derived from N fixing legumes that need to be mineralized before becoming available (Thorup-Kristensen & Dresbøll, 2010). The N inputs in stockless organic systems mainly derive from the second source and are usually lower than for conventional systems. A greater N sequestration seems to occur with these inputs compared to mineral N inputs (Kramer *et al.*, 2002). Gaseous N emissions from organic cropping systems are expected to be smaller since soil mineral N is scarcer than in conventional systems highly fertilized (Skinner *et al.*, 2014b).

At the field scale, the N surplus of a cropping system is calculated as the difference between N inputs and N exported by crop harvests. N surplus is often considered as an indicator of the negative impact of agricultural practices on the environment (OECD, 2001). N surplus must be calculated over long enough period, in order to catch the long term effect of a cropping system on soil structure and texture, soil microbial biomass quantity and diversity.

Few studies have simultaneously estimated the components of the N fate (N surplus, N leaching, SON storage, gaseous N emissions) and the GHG balance of alternative cropping systems on the long term. This is the objective of this paper which analyses the long-term experiment of “La Cage”, comparing conventional (CON), low input (LI), conservation agriculture (CA) and organic (ORG) cropping systems. Autret *et al.* (2016) previously assessed the changes in soil organic carbon (SOC) stocks in these four cropping systems. In this paper, we i) compared changes in SON stocks between systems from 1998 to 2014; ii) quantified the N losses by comparing the N surplus to the SON stocks changes, N leaching and N₂O emissions and iii) established a GHG balance of each cropping system on the short term (2014-2017) and the long term (1998-2014). The N surplus was calculated using records of N inputs and exportations between 1998 and 2016. The SON stocks measured made in 1998 and 2014 were used to estimate the SON sequestration rates. N leaching was calculated using measurements of soil

mineral N and LIXIM model (Mary *et al.*, 1999). The direct N₂O emissions were continuously measured during 3 years between 2014 and 2017. We hypothesized that the four cropping systems could vary widely in the magnitude of C and N fluxes and in their environmental impacts.

4.2 Material and methods

4.2.1 Experimental site

The study was conducted at the experimental site of “La Cage” at Versailles, France (48°48' N, 2°08' E) as described by Autret *et al.* (2016). The experiment was established in 1998 in order to assess the agronomic, economic and environmental performances of three alternative systems compared to a prevalent conventional cropping system of Northern France. Before the experimental establishment, the site was evenly conducted under a conventional management. The climate is oceanic temperate with mean annual temperatures of 11.3°C and mean annual precipitations of 673 mm. The soil is an artificially drained deep Luvisol (IUSS Working Group WRB, 2006). The mean clay content measured in 1998 is 167 g kg⁻¹ over the whole field and the pH in water is 7.38 (Appendix 2.A).

4.2.2 Cropping systems and management

Four cropping systems are compared: a conventional (CON), low input (LI), conservation agriculture (CA) and organic farming (ORG) system. They differ in soil tillage, crop succession and protection, fertilization and crop residues management. Soil was ploughed every year in CON and ORG, and only one out of two years in LI. The CA system, consisting in direct seeding with a permanent plant cover, is conducted in no-till since 1998. The crop rotation of CON and LI was the following: rapeseed (*Brassica napus L.*), winter wheat (*Triticum aestivum L.*), spring pea (*Pisum sativum L.*) and winter wheat. It slightly differed in CA where maize (*Zea mays L.*) was grown two years instead of rapeseed. The main difference occurred in the ORG system in which alfalfa (*Medicago sativa*) was introduced thereafter to replace pea and rapeseed. All crop residues were left on the soil surface at harvest. Alfalfa cuts were returned to the soil in CA and ORG systems in the respective proportion of 50% to 75%. Catch crops were grown only in the CA system, being composed of oat (*Avena sativa L.*), vetch (*Vicia sativa L.*), white mustard (*Sinapis alba L.*) or fodder radish (*Raphanus sativus L.*). A permanent cover crop, composed of

festuca (*Festuca rubra*) in the first years and alfalfa thereafter, was maintained under the main crop in CA. It was chemically destroyed at least every 4 years, before seeding pea crops. Pesticides were used in the CON, LI and CA systems for weed, pest and disease control; their application rate was lower in LI and CA compared to CON system. The ORG system did not receive any pesticide, according the European specifications for organic farming. Phosphate fertilizer was applied at a mean rate of 14, 14, 9 and 3 kg P ha⁻¹ yr⁻¹ and potassium at a rate of 26, 26, 18 and 6 kg K ha⁻¹ yr⁻¹ in CON, LI, CA and ORG systems respectively. Mineral N fertilizers were applied to non-legume crops of CON, LI and CA systems. The N fertilizer rate of each crop was calculated in mid-winter according to a balance-sheet method and split in 2 or 3 applications. The N fertilization rate was highest in CON system and was reduced on average by 22% in LI and 29% in CA systems. In the ORG system, organic N fertilizers (feather meal and guano) were applied solely on wheat during the first years and in 1999 and 2005 for rapeseed, at a low rate (6% of the CON system). No other manure or organic fertilizer was applied.

4.2.3 Measurements

The timeline of N related measurements made at La Cage between 1998 and 2017 is summarized in Figure 4.1.

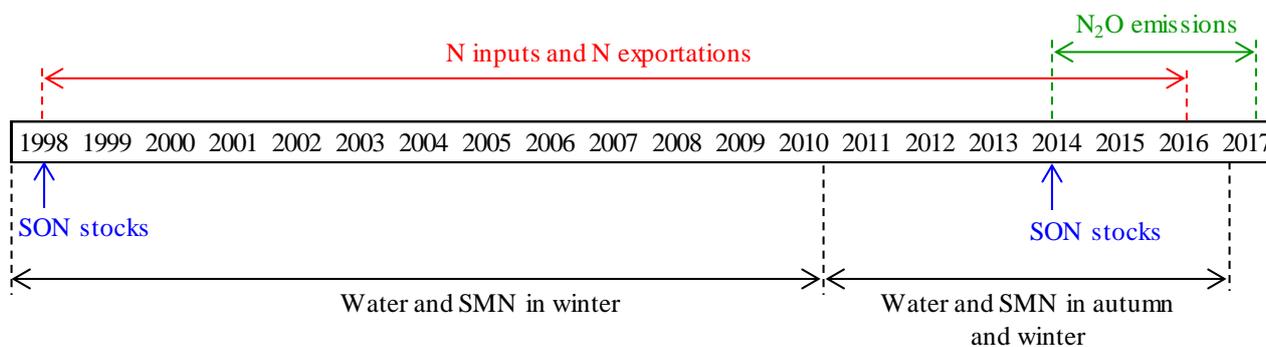


Figure 4.1. Sequences of N measurements made between 1998 and 2017 and used in this paper.

4.2.3.1 Crop yields and N uptake

Grain yield was determined based on the quantity of grain collected by the combine harvester and average values of dry matter content of grain. Alfalfa yield was calculated as the sum of the biomass content of exported cuts which was measured directly, and the aboveground biomass returned to soil when alfalfa was not exported which was estimated using regional references (Autret *et al.*, 2016). When not available, N content in harvested grains and

aboveground biomass was assumed to be equal to national reference values (Appendix 2.B.1). They were used to estimate N offtake by main crops (Table 4.1). Crop residues, cover crops and catch crops were returned to the soil and therefore not accounted for in the N outputs.

4.2.3.2 *Soil water and mineral N contents*

Soil mineral N (SMN) and water contents were measured three times per year: after harvest (average date July 30), in autumn (average November 2) and winter (average February 11) between November 2012 and February 2017. Three soil samples were collected in each subplot down to 90 cm depth, corresponding to the maximum rooting depth. Soil cores were divided into three layers of 30 cm and frozen until mineral N extraction. The gravimetric water content was determined after 48h drying at 105°C. Mineral N was extracted from 100 g of soil shaken in 200 mL of a potassium chloride solution (1M). Nitrate (NO₃) and ammonium (NH₄) concentrations of the solution were measured by continuous-flow colorimetry (Khan *et al.*, 2007). Bulk densities used to calculate SMN stocks were taken from previous measurements made in 2014 (Autret *et al.*, 2016).

4.2.3.3 *SOC and SON stocks*

The SOC and SON concentrations and bulk densities were measured at different dates between 1998 and 2014, in order to calculate SON stocks at equivalent soil mass (ESM) basis (Ellert & Bettany, 1995) over a depth at least equal to the deepest tillage event. Twenty samples were taken in each subplot down to 30 cm depth in 1998, whereas six samples were picked up in each subplot at 60 cm depth in 2014 with a hydraulic gauge of 6 cm diameter (Autret *et al.*, 2016). Soil samples were coarsely crushed, oven dried for 48h at 35°C and sieved (2 mm). Soil subsamples were finely ground in a ball mill (PM 400, Retsch, Germany) before measurement of the total nitrogen content by Dumas (dry combustion) with an elemental analyser (EURO EA, Eurovector, Italy). Bulk density was measured in 1998 for the layers 0-10, 10-20 and 20-30 cm and in 2014 on the top soil layer 0-5 cm, using a steel cylinder of 98 cm³. Soil was weighed after drying during 48 h at 105 °C. A gamma-densitometer (LPC-INRA, Angers, France) was used in 2014 to measure the bulk density every 5 cm in the layers from 5 to 40 cm depth.

4.2.3.4 *N₂O emissions*

Direct N₂O emissions were monitored quasi continuously between April 2014 and July 2017, using automatic chambers as described by Bessou *et al.* (2011). On April 8, 2014, twelve automatic, large size chambers (0.70 m x 0.70 m) were installed in four plots of the experimental site, representing the four cropping systems. Three chambers were installed at a mean distance of 10 m each other in each plot of 0.56 ha. They were maintained in the plot throughout the growing season and removed only for the main operations (sowing, mechanical weeding, harvest and disk ploughing), receiving the same management than the rest of the plot. Chambers were connected to two infra-red gas analysers, one for CO₂ (LiCor 820, LiCor Biosciences, USA) and the other for N₂O (Thermo 46c, Thermo Fisher Scientific, USA) installed in an enclosure left in situ. The automatic chambers were closed sequentially four times a day during 20 minutes (starting at 0:00, 6:00, 12:00 and 18:00 hours GMT). The CO₂ and N₂O air concentrations were measured every 10 seconds during the closure. Additional corrections (besides those provided by the manufacturer) were made to account for the (small) interferences between N₂O and CO₂ concentrations. The N₂O concentration were converted into fluxes by fitting the concentration kinetics to a linear or an exponential model. The smaller measurable flux using this method is lower than 1 g N₂O-N ha⁻¹ day⁻¹.

4.2.4 Calculations

4.2.4.1 *Biological N fixation*

The biological nitrogen fixation (BNF) of legume crops was estimated, either for legumes as main crop, as a cover crop undersown in the main crop, or as a catch crop preceding the main crop. The total quantity of fixed N was calculated as the sum of the contributions of each category of grown legumes. BNF was calculated using the empirical relation established by Anglade *et al.* (2015a) between the amount of N derived from atmosphere (Ndfa) and the N yield of the legume crop:

$$Ndfa = \alpha \cdot Ny + \beta \quad (4.1)$$

where α and β are the specific slope and intercept coefficients of a given crop (Appendix 2.B.2), and Ny is the nitrogen yield, defined as the total nitrogen accumulated in the aboveground biomass (kg N ha⁻¹), calculated as follows:

$$N_y = Y \cdot N_c / NHI \quad (4.2)$$

where Y is the harvested crop yield (t DM ha⁻¹), N_c is the nitrogen content of the dry matter (g kg⁻¹), and NHI is the nitrogen harvest index (ratio of N in the harvested material to N in total aboveground biomass). N_y was determined using the following variables: i) the measured grain yield for pulses (faba bean, lupin, pea and soybean), ii) the estimates of aboveground biomass for the other legumes (alfalfa, vetch and clover), iii) an average value of measured N content for pea, and iv) standard values of N content for the other species (CORPEN, 1988; Parr *et al.*, 2011; Anglade *et al.*, 2015a). The N_{dfa} was finally corrected by a multiplicative factor accounting for belowground contributions (BGN-F) which varied between species, in order to estimate the total BNF (Anglade *et al.*, 2015a).

4.2.4.2 N surplus

The N surplus (N_{sur}) relative to the soil-plant system was calculated for each of the four cropping systems according to OECD (2001) by subtracting the total N exportation from the total N inputs. A positive N surplus indicates that N losses occur in the environment and/or that SON stock increases, whereas a negative surplus means a soil impoverishment. The annual surplus (kg ha⁻¹ yr⁻¹) was calculated as:

$$N_{sur} = N_{fert} + N_{fix} + N_{atm} - N_{exp} \quad (4.3)$$

where N_{fert} is the annual N fertilization, N_{fix} the N derived from symbiotic fixation, N_{atm} the atmospheric N deposition and N_{exp} the N exported from the field by harvests, all values expressed in kg ha⁻¹ yr⁻¹. The values of N_{fert} and N_{exp} were recorded each year whereas N_{fix} was calculated as indicated previously. N_{atm} was estimated based on the European Monitoring and Evaluation Program (<http://www.emep.int/>), providing an annual deposition of 12.9 kg ha⁻¹ yr⁻¹ at the regional scale in France over the 2000-2015 period.

4.2.4.3 N leaching

Water drainage, net N mineralization and N leaching were calculated using both measurements of water and SMN contents and the LIXIM model (Mary *et al.*, 1999). LIXIM simulates simultaneous N mineralisation and water and nitrate transfer between soil layers at a daily time-step, fitting simulated water and mineral N stocks to observed values. We used the adaptation made by Beaudoin *et al.* (2005) to account for the concomitant crop N uptake which can occur

when soil is not completely bare fallow during winter. Results showed that leaching predictions were only very slightly impacted by crop N uptake.

Input data for LIXIM model were the mean daily temperature and precipitation, the potential evapotranspiration, soil bulk density, soil water contents at permanent wilting point and field capacity of each layer, soil water and SMN contents in autumn and winter. When soil was covered by a crop (catch crop, cover crop, main crop or rapeseed regrowth), the date of seeding, depth of rooting, root growth rate, base temperature and N absorption rate were also provided. The depth of rooting, root growth rate and base temperature of each crop were set to values given in Appendix 2.C. The AET / PET ratio was set at 0.5 for a bare soil and 0.6 or fitted by the model when a crop was present in winter. The net N mineralization rate was optimized, as well as the nitrogen absorption rate, by fitting the simulated values of water and SMN contents to the observed values.

Drainage and leaching were estimated in two steps for two different periods. First, they were calculated for the five drainage seasons during which water and SMN stocks were measured both in autumn and winter, *i.e.* 2012 (only November), 2013, 2014, 2015, 2016 and 2017 (only February). The annual nitrate concentration in the drained water ($[NO_3]_w$, in $mg\ NO_3\ L^{-1}$) was estimated by LIXIM during these four years. A linear relationship was then established between this concentration and the SMN stock measured over 90 cm in February ($[NO_3]_s$) as follows:

$$[NO_3]_w = 0.96 * [NO_3]_s \quad R^2 = 0.91, n=16 \quad (4.4)$$

This relationship was used to estimate nitrate leaching from 1998 to 2012, during which water and mineral N stocks were only measured once a year in February. The N leached L ($kg\ N\ ha^{-1}\ yr^{-1}$) was calculated as follows:

$$L = D. [NO_3]_w / 443 \quad (4.5)$$

where D is the amount of drained water below the soil profile ($mm\ yr^{-1}$) and $[NO_3]_w$ the nitrate concentration in the drained water ($mg\ NO_3\ L^{-1}$). The annual drainage D was estimated with a drainage simulator based on evapotranspiration, precipitation and soil available water capacity measurements.

4.2.4.4 Gaseous N losses

Based on N surplus, N leaching and SON storage estimated between 1998 and 2016, we calculated the total gaseous N losses (G , in $\text{kg ha}^{-1} \text{ yr}^{-1}$) as proposed by Mary *et al.* (2002) with the following equation:

$$G = N_{sur} - (N_{stored} + L) \quad (4.6)$$

where N_{stored} is the SON sequestration rate which was measured between 1998 and 2014. This equation assumes that the variation in soil mineral N between the beginning and end of the period is negligible, which is true. G includes all N emissions through denitrification ($\text{N}_2 + \text{N}_2\text{O}$), nitrification ($\text{N}_2\text{O} + \text{NO}_x$) and volatilization (NH_3), without detail of their respective proportions.

4.2.4.5 GHG balance

The total emissions of greenhouse gases at La Cage experiment results from total equivalent CO_2 losses, deriving from the soil and crop management of each cropping. We calculated the annual GHG balance (GHG_b , in $\text{kg CO}_2\text{eq ha}^{-1} \text{ yr}^{-1}$) as follows:

$$\text{GHG}_b = F + M + 296 \cdot \frac{44}{28} (\text{direct } \text{N}_2\text{Oe} + \text{indirect } \text{N}_2\text{Oe}) - \frac{44}{12} \text{SOC}_{storage} \quad (4.7)$$

where F is the amount of CO_2 emitted during the fertilizer synthesis (in $\text{kg CO}_2 \text{ ha}^{-1} \text{ yr}^{-1}$), M the amount of CO_2 emitted during the plant and soil management (in $\text{kg CO}_2 \text{ ha}^{-1} \text{ yr}^{-1}$), *direct* N_2Oe the amount of N_2O emitted by the soil ($\text{kg N}_2\text{O-N ha}^{-1} \text{ yr}^{-1}$), *indirect* N_2Oe the amount of N_2O emitted throughout the N cascade ($\text{kg N}_2\text{O-N ha}^{-1} \text{ yr}^{-1}$) and $\text{SOC}_{storage}$ the amount of carbon yearly stored in the soil ($\text{kg C ha}^{-1} \text{ yr}^{-1}$). F was calculated as the product of the amount of fertilizer applied per hectare and the corresponding emission factors which were $6.17 \text{ kg CO}_2\text{eq kg}^{-1}$ of N for ammonitrate, $1.30 \text{ kg CO}_2\text{eq kg}^{-1}$ of phosphate (P) and $0.54 \text{ kg CO}_2\text{eq kg}^{-1}$ of potassium (K) for binary P-K fertilizers (Gac *et al.*, 2011). M was obtained by multiplying the amount of fuel consumed per hectare for soil and crop management (Appendix 2.D) by the emission factor of $0.81 \text{ kg CO}_2\text{eq per liter of fuel consumed}$ (ADEME, 2010). The direct N_2Oe emissions were measured for the 2014-2017 period. They were used to calculate the impact factors relative to mineral fertilizers and organic N residues for each system during three years. These factors were then used to estimate the *direct* N_2O emissions from 1998 to 2013. The *indirect* N_2Oe emissions were estimated with the emission factor defined by IPCC (2006),

namely 0.75% of the leached N being transformed into N₂O all along the N cascade in groundwater, rivers and estuaries. Finally, the $SOC_{storage}$ was taken from by Autret *et al.* (2016) who measured it during the 1998-2014 period, with 78, 22, 625 and 277 kg C ha⁻¹ yr⁻¹ for CON, LI, CA and ORG respectively.

4.2.5 Statistical analysis

Statistical analyses were performed using the R software (R Core Team, 2017). The weakness of the experiment was its design with only two randomized blocks, previously underlined by Henneron *et al.* (2014), Pelosi *et al.* (2015) and Autret *et al.* (2016). However, these authors considered each of the two sided subplots of a given cropping system as replicate, thus producing four pseudo replicates. This choice was supported by different arguments and particularly the fact that the intra-plot variability (between subplots) was as important as the inter-plot variability (within blocks), as indicated by the comparison of variances ($F=1.83$, $p<0.05$) made by Autret *et al.* (2016).

Analyses of variance (ANOVA) were performed to test the effect of cropping system on 1) SON stocks in 1998 and 2014; 2) N inputs, N exportations and N surplus from 1998 to 2016; 3) N leaching between 1998 and 2017; 4) N₂O emissions between April 2014 and July 2017 and 5) GHG balance. A separate ANOVA was done to compare the change in SON stocks between 1998 and 2014 for each treatment. The assumptions of ANOVA were checked by using the Shapiro-Wilk and Levene's tests. The existence of significant effects ($p<0.05$) was followed by a post-hoc comparison test of means with the SNK.test from the agricolae package (de Mendiburu, 2014). When normality and homoscedasticity were not respected, a Kruskal-Wallis test was applied and followed by means comparison using the kruskal.test of the agricolae package.

4.3 Results

4.3.1 N inputs and offtake by main and auxiliary crops

The mean input of fertilizer-N over the 19 years experiment was 199, 144 and 167 kg N ha⁻¹ yr⁻¹ for wheat crops and 189, 169 and 98 kg N ha⁻¹ yr⁻¹ for rapeseed in the CON, LI and CA cropping systems, respectively (Table 4.1). It was much lower in the ORG system, with an average of 12 kg N ha⁻¹ yr⁻¹ (wheat) and 44 kg N ha⁻¹ yr⁻¹ (rapeseed), both applied as organic fertilizer. The N input derived from symbiotic fixation, related to the legume crop N yield, varied between species. It was estimated at 132, 134, 161 and 84 kg N ha⁻¹ yr⁻¹ for pea crop in the CON, LI, CA and ORG systems, respectively. Alfalfa, whether grown as a main crop in CA and ORG or as a cover crop mixed with a cereal in CA, provided the highest N inputs, ranging from 185 to 460 kg N ha⁻¹ yr⁻¹. BNF represented 16%, 20%, 51% and 80% of total N inputs for CON, LI, CA and ORG systems, respectively. After 2008, organic N fertilizers were no longer applied to the ORG system which then relied exclusively on legume crops to inject reactive N. The part of BNF in total N inputs also increased with time in the CA system, due to the more frequent use of legumes as cover crop and green manure, from 30% until 2008 to an average of 77% since 2009.

N outputs (offtake) are directly linked to the harvest of crop grains and aerial biomass of alfalfa. Average wheat yields varied between cropping systems, respectively 8.3, 7.6, 6.1 and 4.7 t DM ha⁻¹ in CON, LI, CA and ORG. They were correlated to the N fertilizer rate ($r = 0.51$, $n=152$, $p<0.001$). The yields of spring pea and winter rapeseed were also lower in the ORG than in the other cropping systems. The mean N offtake through wheat grains was 149, 134, 115 and 76 kg N ha⁻¹ yr⁻¹ respectively. It represented 53 to 60% of the total N offtake.

4.3.2 Annual N surplus

The average N surplus calculated between 1998 and 2016 was clearly positive for all systems (Table 4.2). It varied from 47 to 152 kg N ha⁻¹ yr⁻¹ and ranked as follows: LI < CON = ORG < CA. The smaller N surplus observed in LI compared to CON results from lower N inputs (162 kg N ha⁻¹ yr⁻¹) and a similar symbiotic N fixation. The N surplus of the ORG system did not differ from the CON system, despite its very low fertilizer input (9 kg N ha⁻¹ yr⁻¹) but the presence of alfalfa generated high N input (93 kg N ha⁻¹ yr⁻¹), three times greater than in the CON system. The very high N surplus observed in CA ($p<0.001$) results from the important

Table 4.1. Crop frequency, mean N inputs from fertilizers and BNF (biological N fixation), mean N offtake by crops and mean grain yield per crop and cropping system during the whole period (1998-2016). Values in brackets are standard deviations between years.

Cropping system	Main crop	Cover crop	Crop frequency	N fertilizer	BNF	N offtake	Grain yield
				kg N ha ⁻¹ yr ⁻¹			t DM ha ⁻¹
CON	wheat		50%	199 (25)		149 (27)	9.5 (1.8)
	rapeseed		26%	189 (31)		118 (23)	4.3 (0.8)
	pea		24%		132 (46)	93 (33)	4.3 (1.6)
LI	wheat		50%	144 (27)		134 (20)	8.7 (1.4)
	rapeseed		26%	169 (15)		95 (25)	3.4 (0.9)
	pea		24%		134 (49)	94 (36)	4.4 (1.7)
CA	wheat	fescue	26%	154 (19)		125 (13)	7.7 (1.1)
	wheat	alfalfa	21%	181 (36)	199 (42)	102 (21)	6.3 (1.5)
	wheat	clover	3%	166	112	120	7.3
	rapeseed	alfalfa	5%	98 (4)	152 (10)	87 (55)	3.1 (2.0)
	pea		16%	8	161 (73)	82 (35)	3.8 (1.6)
	oat	alfalfa	3%		460	44	2.6
	maize		11%	154 (36)	82 (99)	85 (34)	6.1 (2.4)
	alfalfa		16%		341 (40)	64 (5)	9.8 (0.7) *
ORG	wheat		50%	12 (23)		76 (28)	5.5 (1.9)
	rapeseed		8%	44 (40)		22 (20)	0.8 (0.7)
	pea		5%		84 (22)	57 (16)	2.7 (0.7)
	barley-pea		3%		91	67	2.9
	lupin		3%		219	142	3.2
	soyabean		3%		137	84	1.5
	alfalfa		29%		346 (42)	74 (42)	10.7 (0.9) *

* total aerial biomass produced for alfalfa as a main crop

amounts of symbiotic N fixation, especially from alfalfa accounting for 61% of the total BNF inputs. The N surplus was positive in all cropping systems, indicating that N had been stored in the soil and/or lost through leaching and gaseous emissions in all systems.

Table 4.2. Mean annual values of N input, N exported and N surplus (1998 - 2016). Total N input is the sum of N fertilization, BNF and atmospheric N deposition. Values in brackets are standard deviations between replicates. Different letters indicate significant differences between cropping systems ($p < 0.05$).

Cropping system	N fertilization	BNF	atmospheric N deposition	total N input	total N exported	N surplus
	kg N ha ⁻¹ yr ⁻¹					
CON	149 (9) a	31 (10) c	13	194 (3) b	128 (6) a	66 (5) b
LI	117 (8) b	32 (14) c	13	161 (6) c	114 (7) b	47 (3) c
CA	106 (4) c	153 (27) a	13	271 (23) a	95 (7) c	176 (27) a
ORG	9 (1) d	116 (7) b	13	139 (7) d	72 (6) d	67 (2) b

4.3.3 SON storage

The SON stocks calculated at ESM in the 0-30 cm layer (old ploughed layer) in 1998 and 2014 are presented in Table 4.3. Small differences, but not significant, were found between treatments in SON stocks in 1998 ($p < 0.05$), the average SON value being 4.27 t N ha⁻¹. In 2014, SON stocks were much higher in CA than in the three other systems which did not differ significantly each other. SON stocks in the layer 30-60 cm did not differ between systems (results not shown), indicating that most SON variations occurred in the upper layer. The change in SON stocks during the 16 years varied between 0.04 and 1.06 t N ha⁻¹. It was significantly different from 0 in the CA and ORG systems but not in the other systems. The average rates of N sequestration were 13, 3, 54 and 31 kg N ha⁻¹ yr⁻¹ in CON, LI, CA and ORG respectively.

Table 4.3. SON stocks at equivalent soil mass in 1998 and 2014 and SON storage rate between 1998 and 2014. Values in brackets are standard deviations. Letters indicate significant differences between cropping systems ($p < 0.05$).

Cropping system	SON stock 1998	SON stock 2014	SON storage rate (1998-2014)
	t N ha ⁻¹	t N ha ⁻¹	kg N ha ⁻¹ yr ⁻¹
CON	4.13 (0.37) a	4.34 (0.58) b	13 (12) c
LI	4.53 (0.69) a	4.57 (0.95) b	3 (23) c
CA	4.47 (0.43) a	5.33 (0.76) a	54 (15) a
ORG	3.94 (0.25) a	4.44 (0.45) b	31 (9) b

Table 4.4. Mean annual drainage, SMN stocks in autumn and winter, N leached and NO₃ concentration in drained water for each cropping system. NO₃ concentrations are weighted by water drainage. N leached and NO₃ concentrations were estimated with LIXIM model between 2013 and 2017 and calculated during the remaining years using eq. 4.4 and 4.5.

Drainage season	Drained water	CON				LI				CA				ORG			
		SMN autumn	SMN winter	N leached	[NO ₃ ⁻]	SMN autumn	SMN winter	N leached	[NO ₃ ⁻]	SMN autumn	SMN winter	N leached	[NO ₃ ⁻]	SMN autumn	SMN winter	N leached	[NO ₃ ⁻]
		mm yr ⁻¹	kg ha ⁻¹	kg ha ⁻¹	kg ha ⁻¹ y ⁻¹	mg L ⁻¹	kg ha ⁻¹	kg ha ⁻¹	kg ha ⁻¹ y ⁻¹	mg L ⁻¹	kg ha ⁻¹	kg ha ⁻¹	kg ha ⁻¹ y ⁻¹	mg L ⁻¹	kg ha ⁻¹	kg ha ⁻¹	kg ha ⁻¹ y ⁻¹
1998-1999	290		15	9	14		22	17	25		27	23	35		23	18	28
1999-2000	377		22	20	23		24	24	29		20	20	23		27	23	27
2000-2001	401		18	10	11		23	17	19		63	56	61		31	28	30
2001-2002	271		39	27	44		53	39	64		32	17	28		49	29	48
2004-2005	123		59	19	67		55	18	64		41	10	35		96	26	94
2005-2006	99		57	16	71		89	27	120		48	11	49		38	8	37
2006-2007	206		16	8	17		47	27	59		50	30	64		69	31	67
2007-2008	104		19	6	25		12	4	16		19	6	25		24	6	24
2008-2009	35		61	5	67		48	4	49		45	1	17		73	6	71
2009-2010	69		28	5	34		32	6	39		28	4	24		38	6	37
2011-2012	172		15	4	10		26	0	10		55	24	61		30	11	29
1998-2012	195		32	12	27		39	17	38		39	18	41		45	17	40
2012-2013	183	113	30	66	160	92	38	51	122	40	28	20	47	46	29	40	98
2013-2014	226	35	19	19	38	45	27	21	40	42	38	22	43	55	43	23	45
2014-2015	158	59	21	9	26	65	28	10	29	38	17	9	24	38	38	7	19
2015-2016	116	29	3	6	22	27	8	6	22	25	9	5	19	58	27	11	42
2016-2017	26	74	70	0	3	61	61	1	12	84	80	0	5	73	71	0	6
2012-2017	142	62	29	20	63	58	32	18	55	46	35	11	35	54	42	16	51
1998-2017	178		31	14	36		37	17	42		38	16	40		44	17	42

4.3.4 N leaching

The measured SMN stocks and the calculated drainage, N leaching and NO₃ concentrations in drained water are presented in Table 4.4. The SMN stocks in autumn were only measured from 2012 to 2016. The mineral N content in autumn averaged over these five years was close between systems: 62, 58, 46 and 54 kg N ha⁻¹ in the CON, LI, CA and ORG systems respectively. The highest value (113 kg N ha⁻¹) was found in CON in 2012 and the lowest (25 kg N ha⁻¹) in CA in 2015. The SMN stocks found in winter during the same years were lower, respectively 29, 32, 35 and 42 kg N ha⁻¹. The winter SMN during the rest of experiment (1998-2012) were slightly higher but with the same ranking between systems. Although no significant difference was found, the ORG system which received no mineral N fertilizer tended to have the highest SMN stocks in February, varying between 23 and 96 kg N ha⁻¹.

The calculated drainage did not vary significantly between cropping systems but varied markedly between years, from 26 to 401 mm yr⁻¹. The average drainage over the whole period (1998-2017) was 178 mm yr⁻¹. Similarly, the amounts of N leached did not differ significantly between systems but widely among years. The mean amounts of leached N over the whole period were 17, 21, 21 and 22 kg N ha⁻¹ yr⁻¹ for CON, LI, CA and ORG respectively. The nitrate concentration in drained water did not differ significantly between systems with an average value of 51 (period 2012-2017) and 41 mg NO₃ L⁻¹ (whole period), *i.e.* slightly smaller than the maximum content of 50 mg NO₃ L⁻¹ set by the Nitrate Directive (91/676/CEE). The concentrations calculated during the last five years differed more between systems (35 mg NO₃ L⁻¹ in CA vs 63 mg NO₃ L⁻¹ in CON), suggesting that the improvement in the management of the non-conventional systems might reduce nitrate leaching losses in the near future.

4.3.5 N surplus and gaseous N losses

The greatest source of uncertainty in the calculation of the N surplus lie in the BNF input from alfalfa in the CA and ORG systems, with two components: 1) the estimate of aboveground biomass and 2) the belowground N derived from BNF (Table 4.5). We conducted a sensitivity analysis to determine the change in the N surplus in response to a variation in aboveground production of alfalfa and in the ratio of total BNF (above + belowground) to BNF in aboveground biomass (BGN factor in Anglade *et al.* 2015a). Biomass production varied between 90 and 110% of the nominal value (range determined using the measurements made

in the last years) and three values of the BGN factor were tested: 1.4, 1.7 and 2.1 corresponding to its minimum, average and maximum value as presented by Anglade *et al.* (2015a). The N surplus thus calculated varied widely, from 150 to 220 kg N ha⁻¹ yr⁻¹ in CA and 47 to 103 kg N ha⁻¹ yr⁻¹ in ORG system (Table 4.5). The average values were close to those previously calculated, 180 ± 18 kg N ha⁻¹ yr⁻¹ for CA and 71 ± 18 kg N ha⁻¹ yr⁻¹ for ORG. It is noticeable that the difference between the two cropping systems was much more stable, 109 ± 4 kg N ha⁻¹ yr⁻¹. This indicates that despite the rather large uncertainty in the estimates of N surplus, the CA system is characterized by a much higher surplus than the ORG system.

Table 4.5. Sensitivity analysis of N surplus (kg N ha⁻¹ yr⁻¹) to the aboveground biomass and BGN factor of alfalfa for the conservation agriculture (CA) and organic (ORG) cropping systems. Values in brackets are standard deviations. The N surplus used in the study is shown in bold.

Cropping system	BGN factor	Relative variation of AG biomass				
		90%	100%	110%	mean	sd
CA	2.1	191	205	220	180	(23)
	1.7	164	176	188		
	1.4	150	160	170		
ORG	2.1	81	92	103	71	(18)
	1.7	58	67	75		
	1.4	47	54	62		
Difference	2.1	110	113	117	109	(4)
CA-ORG	1.7	106	109	112		
	1.4	103	105	108		

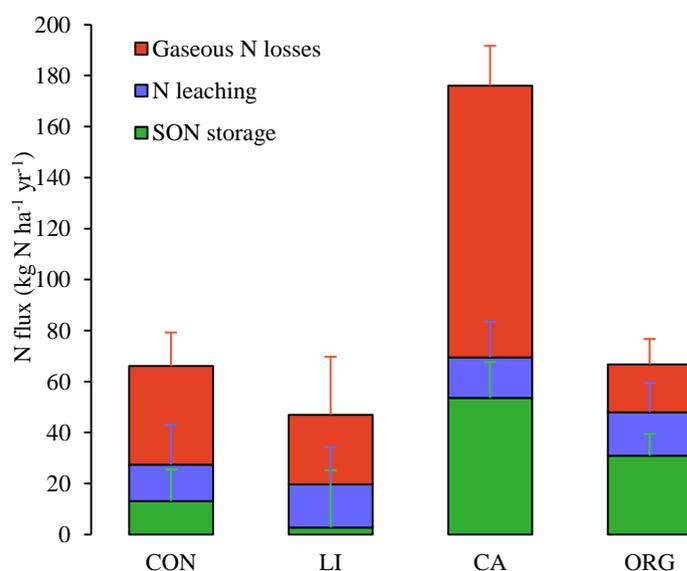


Figure 4.2. Partitioning of the N surplus between SON storage, N leaching and gaseous N emissions (see eq. 4.6). Gaseous N losses were calculated as the difference between the N surplus and the sum of SON storage and N leaching. Error bars represent the standard deviations.

The unrecovered N, *i.e.* the difference between the N surplus and the sum of N stored in soil and N leached, corresponds to the gaseous N losses (denitrification + volatilization). Figure 4.2 displays the N surplus and its partitioning into its three components over the 1998-2017 period. N surplus was mainly correlated with gaseous losses ($r=0.97$), moderately with SON storage ($r=0.91$) and not correlated with N leaching ($r=-0.12$). The gaseous losses differed widely among systems: they were about three times greater in the CA system ($106 \text{ kg N ha}^{-1} \text{ yr}^{-1}$) than in the three other systems. They were smallest in the ORG ($19 \text{ kg N ha}^{-1} \text{ yr}^{-1}$) and intermediate in the CON and LI systems (39 and $27 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ respectively). Similar differences were obtained when calculations were made only during the period 2014-2017.

4.3.6 N₂O emissions

Emissions varied widely between systems and throughout time (Table 4.6). Over the whole monitoring period (April 2014 – July 2017) the highest emissions were recorded in the CA system ($11.96 \text{ kg N}_2\text{O-N ha}^{-1}$), the lowest in the LI and ORG system (3.51 and $4.39 \text{ kg N}_2\text{O-N ha}^{-1}$) and intermediate in the CON system ($6.85 \text{ kg N}_2\text{O-N ha}^{-1}$). Emissions occurred mainly but not exclusively after fertilizer application, in response to rainfall events, indicating that denitrification was the main source of N₂O production. The ORG system produced very small emissions except during the last period (November 2016 – July 2017) under wheat established after the destruction of alfalfa, contrasting with the other systems which had small emission rates during this period. This is attributed to the very dry season.

Table 4.6. Cumulative N₂O-N fluxes (kg N ha^{-1}) measured continuously during 6 periods (4 crops and 3.3 years). Values in brackets are standard deviations. Letters indicate significant differences between cropping systems ($p < 0.05$) at each period.

Measurement period dd/mm/yy	Duration days	Crop	Cropping system				
			CON	LI	CA	ORG	
08/04/14	20/07/14	104	wheat	1.50 (0.55) b	0.93 (0.19) b	2.98 (0.90) a	0.22 (0.07) c
02/09/14	08/07/15	310	rapeseed*	1.44 (0.14) b	1.30 (0.15) b	2.51 (0.39) a	0.51 (0.04) c
10/07/15	19/10/15	102	fallow*	0.17 (0.05) a	0.17 (0.10) a	0.09 (0.05) a	0.23 (0.02) a
10/11/15	26/07/16	260	wheat	2.31 (0.54) b	0.86 (0.16) c	5.21 (1.39) a	0.54 (0.13) d
31/08/16	19/10/16	50	fallow*	0.38 (0.08) b	0.15 (0.03) c	0.55 (0.09) a	0.57 (0.18) a
16/11/16	16/07/17	243	wheat	1.04 (0.10) b	0.11 (0.02) d	0.62 (0.14) c	2.32 (0.56) a
08/04/14	16/07/17	1069	all	6.85 (1.48) b	3.51 (0.88) c	11.96 (3.29) a	4.39 (0.95) c

* alfalfa for the ORG system

Table 4.7. GHG balance and components at La Cage estimated during the 2014-2017 period (with N₂O monitoring) and the whole experimental period (1998-2017).

Period	Cropping system	<i>F</i>	<i>M</i>	<i>direct</i> N ₂ O _e	<i>indirect</i> N ₂ O _e	SOC storage rate	GHG balance		
							kg CO _{2eq} ha ⁻¹ yr ⁻¹	kg CO _{2eq} kg ⁻¹ N input	kg CO _{2eq} kg ⁻¹ N exported
2014-2017	CON	861 a	281 a	1088 b	30	286 **	1975 a	10.2 a	15.5 a
	LI	644 b	251 b	558 c	33	81 **	1405 a	8.7 a	12.3 a
	CA	606 b	131 c	1900 a	31	2292 **	377 b	1.4 b	4.0 b
	ORG	0 c	243 b	698 c	36	1016 **	-39 c	-0.3 c	-0.5 c
1998-2017	CON	927 a	272 a	1153 *	50	286 a	2116 a	10.9 a	16.6 a
	LI	728 b	249 b	736 *	59	81 a	1690 a	10.5 a	14.8 a
	CA	659 b	161 c	1645 *	56	2292 c	229 b	0.8 b	2.4 b
	ORG	44 c	255 b	576 *	60	1016 b	-81 c	-0.6 c	-1.1 c

F is the equivalent amount of CO₂ emitted during fertilizer synthesis and application.

M is the equivalent amount of CO₂ emitted by tractors during crops and soil management.

Direct and indirect N₂O_e are the N₂O fluxes directly emitted by the soil-crop system and throughout the N cascade, respectively.

SOC storage rates were measured during the 1998-2014 period (Autret et al., 2016).

* calculated using N₂O emission factors measured during the period 2014-2017

** assumed to be similar to the period 1998-2014

The N₂O emission factor relative to the total N inputs (mineral + organic N) during the whole monitoring period was 1.19%, 0.92%, 1.49% and 1.42% for the CON, LI, CA and ORG systems, respectively. These values are close to IPCC references. In addition, N₂O emissions were highly correlated with total gaseous losses ($r=0.97$, $p<0.001$). This result suggests that a large part of the gaseous losses originated from denitrification.

4.3.7 Global GHG balance

The GHG balance calculated for each cropping system is presented in Table 4.7. The upper part of the table shows the GHG balance during the 2014-2017 period, during which N₂O fluxes were continuously measured, whereas the lower part concerns the 19-yr period (1998-2017).

Emissions deriving from the synthesis of fertilizers F were highest in CON (927 kg CO₂eq ha⁻¹ yr⁻¹) and smallest in ORG (44 kg CO₂eq ha⁻¹ yr⁻¹) which received small amounts of organic fertilizers in the early years of the trial. The CO₂ emissions related to agricultural operations varied less between systems, from 272 kg CO₂eq ha⁻¹ yr⁻¹ in CON to 161 kg CO₂eq ha⁻¹ yr⁻¹ in CA. The differences were mainly due to the absence of soil tillage in CA, ploughing being particularly fuel consuming. The indirect N₂O emissions occurring during the N cascade contributed very little (average 71 kg CO₂eq ha⁻¹ yr⁻¹) to the GHG balance. The direct N₂O emissions and the direct CO₂ emissions (estimated by the SOC variation) were the main sources of variability. Taking into account the annual SOC sequestration, the net GHG balance was estimated for each cropping system on the 1998-2017 period and ranked as follows: CON (2116 kg CO₂eq ha⁻¹ yr⁻¹) = LI (1690) > CA (229) > ORG (-81). A similar ranking and amplitude of variation was obtained for the period 2014-2017, showing the robustness of the calculations. The ranking was conserved when the GHG balance was expressed per unit of N input or unit of exported N. Intensive mineral N fertilization and mechanization, associated with a poor SOC sequestration rate lead to high GHG emissions in the CON, as well as in LI even with reduced farming intensity. The CA system had a much better GHG balance due to its very high SOC storage rate. However, its high N₂O emissions offset this beneficial effect, resulting in a positive GHG balance. Finally, the ORG system was the only cropping system leading to a negative GHG balance.

4.4 Discussion

We have studied the long term impact of alternative cropping systems on N fate, particularly N uptake by crops, SON storage, N leaching and gaseous N emissions, and also on the GHG balance, including direct and indirect emissions. Figure 4.3 summarizes the results and compares C and N impacts (graded between 0 and 1) of each cropping system to a reference value (scoring 0.75).

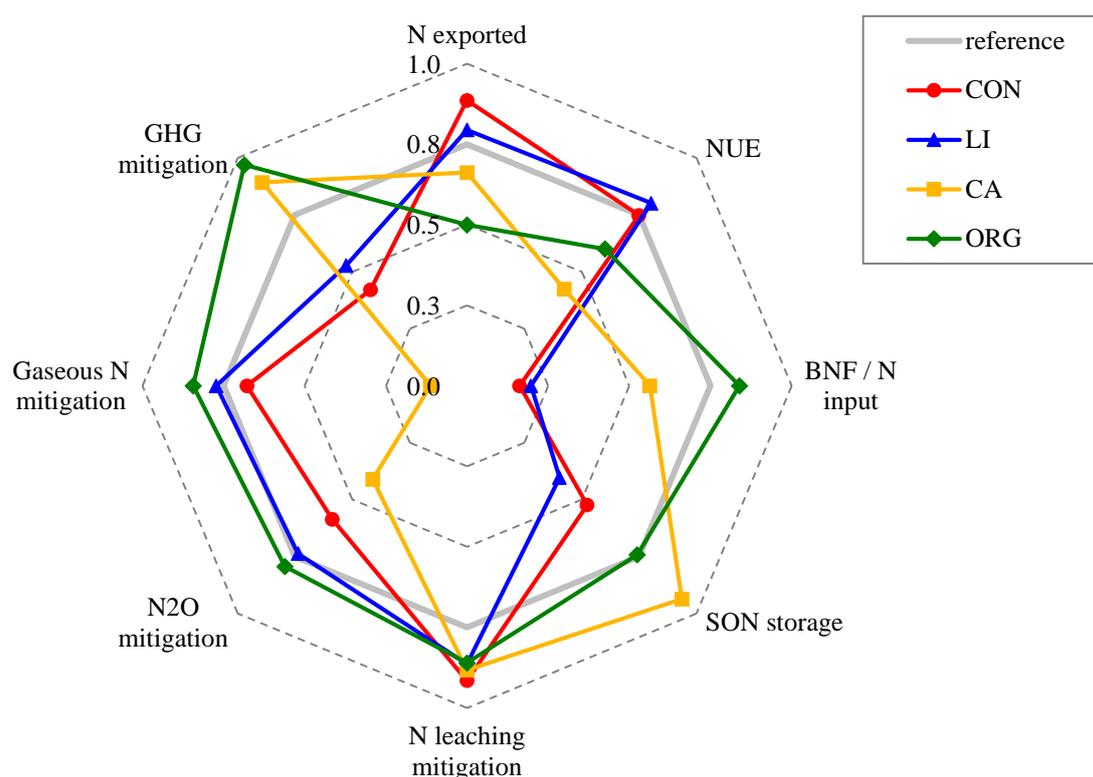


Figure 4.3. Global evaluation of the environmental C and N impact of the four cropping system. Each dimension was scored between 0 (adverse impact) and 1 (beneficial impact). The scores were attributed based on mean reference values: 108 kg N ha⁻¹ (average N exported in France, Eurostat), 66% (NUE, OECD), 75% (BNF/ N input), 50 mg NO₃ L⁻¹ (N leaching), 1.5 kg N₂O-N ha⁻¹ yr⁻¹ (N₂O emissions), 8 p mille (SON storage), 1000 kg CO₂eq ha⁻¹ yr⁻¹ (GHG balance), 30 kg N ha⁻¹ yr⁻¹ (gaseous N losses).

4.4.1 N use efficiency

The NUE of each cropping system can be compared to the reference value of 0.66 obtained on average for France during the 2000-2014 period (Eurostat, 2016). During the 19 years of the experiment, LI system yielded the highest NUE, 0.71, CON and ORG obtained respectively 0.66 and 0.52, and the lowest was found in CA system, 0.35. This latter value was explained by the high organic N inputs derived from cover crop residues which were preferentially stored

in the soil rather than used for plant uptake. Given that straw was systematically returned to soil during the experiment in all cropping systems, the differences in NUE refer solely to the efficiency of conversion of the supplied N, either by inorganic fertilizers or through BNF, into crop uptake and grain N. Aronsson *et al.* (2007) reported similar NUE values in Sweden: 0.54 for an organic system relying on green manure, 0.68 for a conventional system, but higher efficiency in an organic system with animal manure (0.70). In Italy, higher NUE were found for an organic system (average 0.74 over 16 years) than for a conventional (average 0.51) (Migliorini *et al.*, 2014). Lin *et al.* (2016) also reported higher NUE for organic systems in a 20-yr experiment in Germany.

4.4.2 N surplus, an ambiguous indicator

The N surplus is often presented as an indicator of the N losses in arable fields (OECD, 2001), useful to compare management practices at annual time step. However, a positive N surplus may also reflect a SON storage. In our experiment, the N surplus was almost three times higher in CA than in the other systems (181 vs 64 kg N ha⁻¹ yr⁻¹), in spite of a smaller addition of mineral N fertilizer compared to the CON system. The important amounts of symbiotic fixed N combined with smaller N exportations are responsible of the very high N surplus in CA. The introduction of legume cover crops in conservation agriculture is used to provide N for the subsequent crop after crop residues mineralization and allow to reduce mineral N fertilization (Scopel *et al.*, 2012). Blesh and Drinkwater (2013) made contrasted observations in fields from the Mississippi River Basin, where legumes and complex crop rotations including annual and perennial species were grown. They found smaller surpluses in these cropping systems compared to mineral N based systems (<10 and 35 kg N ha⁻¹ yr⁻¹ respectively). The difference between their results and ours can be explained by the large proportion of alfalfa cuts returned to soil in La Cage experiment: 25% to 33% in the CA system and 70% in ORG. The N surplus in the LI and CON systems is equal or slightly greater than the mean value (47 kg N ha⁻¹ yr⁻¹) reported by Poisvert *et al.* (2017) in the same region over the period 2000-2010. The ORG system is characterized by an unusual high N surplus (67 kg N ha⁻¹ yr⁻¹) compared to other published references. This results from the high frequency of legumes in the rotation (42%), particularly alfalfa (29%) whose cuts were mostly returned to soil. The asynchrony between the crop N uptake and the release of mineral N from decomposing residues, described by Crews and Peoples (2005), may have limited N uptake and increased the N surplus.

4.4.3 SON storage

SOC and SON measurements realized in 1998 and 2014 indicated that N storage occurred in all treatments but was significant only in the CA and ORG systems. The mean rate of sequestration was 54 and 31 kg N ha⁻¹ yr⁻¹ respectively. These results are consistent with Autret *et al.* (2016) who found significant SOC sequestration in both systems. The authors attributed the SOC increase of these systems to important crop residues and root inputs, deriving from supplementary catch crops and cover crops. SOC sequestration rates can be compared to the yearly increase of 4 ‰ targeted by the “4 per 1000” initiative to mitigate CO₂ emissions. They were 1.8, 0.5, 14.1 and 7.0 ‰ for CON, LI, CA and ORG systems respectively (Autret *et al.*, 2016). SON sequestration rates were close: 3.2, 0.6, 12.0 and 7.8 ‰ respectively; the conservation and organic systems thus demonstrated a high potential of C and N storage in soil. If the important storage could be expected in the CA system (González-Sánchez *et al.*, 2012), the storage in the ORG system is more surprising. The positive impact of organic systems on SOC/SON storage claimed by some studies (Gattinger *et al.*, 2012; Lin *et al.*, 2016) is debated (Leifeld *et al.*, 2013) and/or attributed to higher application of organic fertilizer in organic farming experiments (Leifeld & Fuhrer, 2010). In our experiment, very low amounts of organic fertilizer were applied in the ORG system. We hypothesize that the storage in ORG (and also CA) mainly result from the important amount of legume residues, particularly alfalfa, which provided both C and N substrate needed for C and N sequestration in soil: large C inputs were demonstrated by Autret *et al.* (2016) and large N surpluses were shown in this study. This is fully consistent with Van Groenigen *et al.* (2017) who pointed out the importance of N required for SOC sequestration.

4.4.4 N leaching

The N leaching mitigation (Figure 4.3) was assessed by comparing the NO₃ concentrations in drained water to a reference threshold of 50 mg L⁻¹ defined by the Nitrate Directive. The average leaching losses over the 1998-2017 period was 20 kg N ha⁻¹ yr⁻¹, without any difference between cropping systems. This value was lower than the average N leaching reported by Benoit *et al.* (2014) in the same region for conventional cropping systems (32-77 kg N ha⁻¹ yr⁻¹) and closer to those reported for organic farms (13-37 kg N ha⁻¹ yr⁻¹). N leaching was obviously not related at all with N surplus. The correlation between N leaching and N surplus, reported by Billen *et al.* (2013) for conventional systems, does no longer apply when comparing these cropping systems involving diverse cropping practices and/or rotation. Other studies also confirmed this

poor or absence of correlation in arable cropping systems (Sieling & Kage, 2006; Pugesgaard *et al.*, 2017).

A moderate reduction in mineral fertilizer N has been shown to have small effects on N leaching if the reference system is not over-fertilized (*e.g.* Constantin *et al.*, 2010). This was the case of LI compared to CON. In the case of CA system, opposite effects occurred: the presence of permanent cover crop favoured catching of mineral N whereas alfalfa destruction and decomposition during autumn and winter increased SMN. The absence of tillage in this system did not seem to reduce N leaching, in good agreement with previous results (Oorts *et al.*, 2007; Mary *et al.*, 2014; Hansen *et al.*, 2015). The similar N leaching observed in the ORG system is more surprising with regard to the literature. Organic cropping systems generally lead to a decrease of N leaching because of a lower level N fertilizer applied, as highlighted by Tuomisto *et al.* (2012) in their meta-analysis. Syswerda *et al.* (2012) also found smaller nitrate losses in an organic system using legume catch crops compared to a mineral N fertilizer-based conventional system; the average nitrate concentration in drained water over 11 years was decreased to 40 instead of 84 mg NO₃ L⁻¹, respectively. In our study, the amount of drained water was smaller which likely reduced the differences between treatments. Furthermore, the favourable effect of low fertilizer rate was probably offset by the risky phase of alfalfa destruction. Several authors have mentioned that the poor synchrony between mineral N availability derived from alfalfa residues and the subsequent crop uptake could increase N leaching (Crews and Peoples, 2005; Aronsson *et al.*, 2007).

4.4.5 Gaseous N emissions

The N₂O emissions monitored continuously during 3.3 years are assumed to be representative of the four systems, at least during the last years. The emission factors calculated for this period are close to the average values recommended by IPCC, *i.e.* 1% of the N inputs. The CA system was characterized by high rates of N₂O emissions after fertilization events, much higher than those observed in the LI system, although both systems received almost the same amount of fertilizer-N. Such difference may result from the absence of tillage or the presence of the living mulch or both effects. The influence of no tillage on N₂O emissions is not clear yet, since opposite results have been reported. In their meta-analysis, van Kessel *et al.* (2013) did not point out differences between tilled or no-tilled systems during the first ten years, and found lower emissions in no-till systems after 10 years under dry climates. Therefore, tillage does not seem to be the reason for the differences observed at La Cage. The mulch formed by dead crop

residues on the top soil layer together with the more humid micro-climate created by the living cover crop may have played an important role in the emissions recorded in the CA system. Shan and Yan (2013) have shown in their meta-analysis that the presence of mulch of plant residues stimulates N₂O emissions compared to their incorporation in soil. Crop residues, left as a mulch on the top soil layer, have been shown to increase soil moisture content of soil surface and exacerbate the N₂O emissions under annual or perennial crops (Peyrard *et al.*, 2016, 2017). Moreover, residues deriving from alfalfa probably increased N₂O emissions in the CA system, since legumes residues are known to induce higher N losses than non-legume (Basche *et al.*, 2014).

The total gaseous N emissions, assessed by the N mass balance, correspond to the sum of NH₃ volatilization, production of N₂ and N₂O by denitrification and NO_x and N₂O by nitrification, all fluxes being stimulated by mineral N fertilization (De Klein *et al.*, 2006). In our study these losses were not correlated with the fertilizer rate, but with the N surplus ($r = 0.97$, $p < 0.001$). They were also well correlated with N₂O emissions ($r = 0.97$, $p < 0.001$), which suggests that denitrification was the major source of leak to the atmosphere. The CA system exhibited the highest N losses (average 106 kg N ha⁻¹ yr⁻¹) and ORG the smallest ones (19 kg N ha⁻¹ yr⁻¹). If we assume that NH₃ volatilization represented 14% of the synthetic N fertilizer applied (Bouwman *et al.*, 2002) and that losses through nitrification 1% of the nitrified N (Rolland *et al.*, 2008), then the N₂O/(N₂+N₂O) ratio would vary from 5% (CA) to 15% (CON).

4.4.6 The GHG balance, and ultimate environmental indicator

Our GHG balance accounts for most sources of CO₂ emissions, including mineral fertilizer synthesis, fuel combustion due to crop and soil management, direct and indirect N₂O emissions from soil, groundwater, rivers and estuaries and net CO₂ emissions from soil (assessed using SOC change rate). The average GHG balance was 2116 kg CO_{2eq} ha⁻¹ yr⁻¹ in the conventional system over the 19 years, and 20% smaller in the LI system. Due to its very C sequestration rate, the CA system had a much more favourable balance, emitting only 229 kg CO_{2eq} ha⁻¹ yr⁻¹ to the atmosphere. The best situation was found in the ORG system which was a sink for the atmosphere (-81 kg CO_{2eq} ha⁻¹ yr⁻¹). The relative differences are maintained when the GHG is expressed per unit of N input or N exported instead of area unit.

Very few studies have quantified the GHG balance of such arable alternative cropping systems without manure application. Most studies focused on gross GHG emissions, without

considering SOC storage and/or N₂O emissions. Six *et al.* (2004) estimated a negative GHG balance in no-till systems compared to conventional tillage, but they mentioned the large uncertainty of their estimation, related to the variability of N₂O emissions.

Mary *et al.* (2014) also compared tilled versus no-tilled systems in a long-term experiment at Boigneville (France) in which SOC stocks had been monitored for 41 years and N₂O emissions for three years. They found high values of the GHG balance both for no-till (2250 kg CO_{2eq} ha⁻¹ yr⁻¹) and ploughed systems (2930 kg CO_{2eq} ha⁻¹ yr⁻¹). Aguilera *et al.* (2015) estimated the carbon footprint of rainfed crops under conventional and organic management in 8 Spanish farms. The estimated higher net GHG emissions from conventional than organic systems when expressed per unit of area (1024 vs 361 kg CO_{2eq} ha⁻¹ yr⁻¹) and also per unit of production (315 vs 182 g CO_{2eq} kg⁻¹). The authors attributed the low carbon footprint of organic management to reduced CO₂ emissions deriving from synthetic fertilizers use and lower direct N₂O emissions since SOC sequestration rates were small and similar in conventional and organic systems.

Compared to the N surplus, the GHG balance can be presented as an ultimate indicator giving a wider evaluation of cropping systems, in the context of the global climate change mitigation with alternative cropping managements. In our study, the GHG balance completely reshuffled the ranking obtained with the N surplus among cropping systems. The N₂O losses accounted for a small share (around 5%) of the total N losses in the four cropping systems, whereas they represented on average 49% of total GHG emissions. In the CA system, N₂O emissions offset 66% of the SOC sequestration rate which nevertheless reached the top range achievable in arable cropping systems. The importance of assessing N₂O fluxes on the global warming potential was previously pointed out by Six *et al.* (2004) who compared tilled and no-till systems. Using a simulation model, Li *et al.* (2005) predicted that C sequestration often goes along with increased N₂O emissions in alternative cropping systems. Our results support the idea that no-till management, and even conservation agriculture system, should not be seen as an ultimate solution to mitigate global warming (VandenBygaart, 2016). Hence, the need for a complete evaluation of the GHG balance in designing alternative cropping systems is critical, yet being a difficult task (Skinner *et al.*, 2014b).

4.5 Conclusion

The N surplus, N fate and the GHG balance were compared in the conventional and three alternative arable cropping systems over 19 years in La Cage experiment (Northern France). The four systems had contrasted impacts on C and N cycles: they had similar effects on nitrate leaching, but very different ability to sequester C and N in soil, and very different gaseous losses, including denitrification and N₂O losses. Their mitigation potential was not reflected by their N surplus. The alternative systems all improved the GHG balance, slightly for the low input system, markedly for the conservation agriculture system systems and even better in the organic cropping system which led to a negative GHG balance. In conservation agriculture, the high N₂O emissions partially offset the very high carbon sequestration rate observed in the soil.

Agricultural policies targeting a single environmental objective, such as the “4 per 1000” initiative (Minasny *et al.*, 2017), must be considered with great vigilance since they may potentially overestimate the CO₂ sequestration potential (*e.g.* White *et al.*, 2017). Our results clearly demonstrate that the full GHG balance has to be considered when comparing the potential of new management practices. Hence, an appropriate assessment of the environmental impact of a cropping system should be based on a global evaluation, considering both C and N fluxes modified by the farming practices and not be limited to a patchy indicator. Our study confirms the interest of long term monitoring to accurately evaluate the impact of alternative systems. Moreover, there is a need to investigate other CA or ORG systems varying in the nature and importance of legume residue returns which constitute the main alternative to mineral N fertilizer for injecting reactive nitrogen into soils. This investigation can be done using experiments, modelling or both.

4.6 Acknowledgements

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CHAPTER

5.

Long term modelling of crop production and nitrogen fate in organic cropping systems

Article in preparation for *Science of the Total Environment*

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Highlights

- ▶ The fate of C and N was quantified and modelled in two long term experiments
- ▶ STICS model was improved in order to simulate organic farming (OF) systems
- ▶ STICS was able to reproduce crop production, N surplus and SON stocks
- ▶ OF did not systematically differ from conventional in their N surplus and N losses

Abstract

Although organic cropping systems are promoted for their environmental benefits, little is known about their long term impact on nitrogen (N) fate in the soil-plant-atmosphere system. STICS model was used to simulate crop production, N surplus, nitrate leaching, gaseous N losses and changes in soil organic N in two long-term experiments: DOK in Switzerland (39-yr) and Foulum organic in Denmark (19-yr). Four treatments were considered in each experiment: two conventional treatments with (CONFYM) or without manure (CONMIN), organic with manure (BIOORG) and unfertilized treatment (NOFERT) at DOK; conventional (C4-CC+IF) and three organic treatments, one with catch crops only (O4+CC-M) and two including catch crops and clover-grass with (O2+CC+M) or without manure (O2+CC-M), at Foulum. STICS model was calibrated in the conventional treatments and could satisfactorily simulate crop production and N exported in all treatments, with a slight bias for water and mineral N contents in soil. The temporal evolution of soil organic N stocks was well captured along with the annual N surplus. N surplus greatly differed between treatments at DOK, from -58 (NOFERT) to +21 kg N ha⁻¹ yr⁻¹ (CONFYM), and only from -9 (O4+CC-M) to +21 kg N ha⁻¹ yr⁻¹ (O2+CC+M) in Foulum. The simulated N leaching did not differ between treatments at DOK, while it was reduced by 41% in O4+CC-M compared to C4-CC+IF at Foulum, due to the combined effects of catch crops and absence of mineral N fertilizer. N volatilization was greatest in CONFYM and O2+CC+M. N denitrification was highest in BIOORG at DOK and 53% higher in fertilized treatments at Foulum. Predicted changes in soil N pools were always negative (from -18 to -78 kg N ha⁻¹ yr⁻¹), consistently with the measured N surpluses, depending on fertilization and crop rotation. The model simulated important N fluxes towards deep root residues (1-14 kg N ha⁻¹ yr⁻¹), *i.e.* below the ploughed layer, particularly in systems including a clover-grass ley. The fate of this deep N (supposed to accumulate in soil) was not simulated. This study showed that STICS model was able to mimic the N fate in organic and conventional cropping systems with common formalisms. Our simulations stressed out the fact that arable organic systems did not systematically induce lower losses than conventional ones, providing a progress margin for increasing N use efficiency of these systems.

5.1 Introduction

Given the negative side-effects of conventional agriculture on the environment (e.g. Alcamo *et al.*, 2003; Sutton *et al.*, 2011), the development of organic cropping systems has met a resounding success, with an increase of 74% of the organic agricultural lands in Europe over the last decade (Willer & Lernoud, 2017). Organic farming is often depicted as an opportunity for mitigating climate change by limiting C and N losses towards the environment and reducing pesticides pollution (Mondelaers *et al.*, 2009; Tuomisto *et al.*, 2012). The impact of organic farming on soil carbon stocks, nitrate leaching or nitrous oxide (N₂O) emissions has been examined in recent papers (Gattinger *et al.*, 2012; Aguilera *et al.*, 2013; Benoit *et al.*, 2014; Skinner *et al.*, 2014b). Long term assessment of these impacts implies costly and time consuming field experiments. Coupling data acquisition and soil crop modelling on the long term gives access to hardly measurable variables and allows to catch the potential impacts of cropping systems on C and N cycles (Möller, 2009; Constantin *et al.*, 2012). Process-based dynamic models have been developed and assessed for simulating yields and environmental impact of conventional cropping systems (Hansen *et al.*, 1990; Brisson *et al.*, 1998; Jones *et al.*, 2003; Keating *et al.*, 2003; Stöckle *et al.*, 2003), but few of them have considered stockless organic systems. Furthermore, crop succession effects are rarely considered (Lorenz *et al.*, 2013) and C and N impacts often considered separately, either for carbon (Leifeld *et al.*, 2009) or nitrogen (Berntsen *et al.*, 2006). Simulating accurately the long term effect of more diversified crop rotations and management practices - as found in organic cropping systems - on C and N fluxes simultaneously remains a scientific challenge, requiring efforts in model parameterization and validation.

Arable organic farming systems often include diversified crops, catch crops, relay cover crops (Amossé *et al.*, 2014) or intercrops mixing legumes and cereals (Thiessen Martens *et al.*, 2001), or pluri-annual crops like mixed leys including legumes (Teasdale *et al.*, 2004; Stinner *et al.*, 2008). Some of these techniques involve undersowing an auxiliary leguminous crop in an established main crop, resulting in a well-developed cover crop after harvest, able to produce a high biomass and add extra N through symbiotic fixation. The destruction of mixed leys and cover crops releases nutrients in soil, particularly N, available for the subsequent crops (Fustec *et al.*, 2010; Amossé *et al.*, 2014). Another expected benefit of these supplementary crops is the increase in SOC stocks in the tilled layer receiving crop residues (Autret *et al.*, 2016; Blanco-Canqui *et al.*, 2017) and the important root deposition due to herbaceous species (Poorter *et al.*, 2015) increased with the diversity of species in the crop mixture (Lange *et al.*, 2015). However,

the mismatch between the release of N through residues N mineralization and the N demand of the next crop may lead to significant nitrate leaching (Olesen *et al.*, 2009; Valkama *et al.*, 2015) and cover crops residues, particularly in mulch, tend to promote N₂O emissions (Rochette & Janzen, 2005; Basche *et al.*, 2014).

STICS is a soil-crop model that simulates crop growth and the cycles of nitrogen, carbon and water with their associated environmental impacts (Brisson *et al.*, 1998, 2008). It has been positively evaluated for simulating the impact of agricultural practices on soil carbon balance (Wattenbach *et al.*, 2010), N mineralization (Gabrielle *et al.*, 2002), nitrate leaching (Poch-Massegú *et al.*, 2014; Constantin *et al.*, 2015; Plaza-Bonilla *et al.*, 2015) and N₂O emissions (Peyrard *et al.*, 2017; Plaza-Bonilla *et al.*, 2017), over a wide range of cropping and pedo-climatic conditions (Coucheney *et al.*, 2015). STICS can simulate varied agricultural management practices related to organic matter inputs, catch crops (Beaudoin *et al.*, 2008; Constantin *et al.*, 2012) and intercrops (Corre-Hellou *et al.*, 2009). A recent improvement of the model allows simulating perennial crops including their root turnover (Strullu *et al.*, 2015). Thus, given its genericity, robustness and diversity of variable outputs, STICS model has the potential to simulate C-N dynamics in organic cropping systems on the long term.

In this work, the scientific strategy consisted in coupling experiments and modelling to compare conventional and organic arable cropping systems varying in rate and form of N inputs. Our objective was to evaluate the ability of STICS for predicting yield, N surplus, changes in soil organic N, N leaching and gaseous losses in organic cropping systems on the long term. We compiled data from two long term experiments comparing conventional and organic systems, namely the DOK experiment (39-yr) located in Therwil (Switzerland), and the Foulum organic experiment (19-yr) (Denmark). We hypothesized that crop production could be predicted by STICS in organic systems managed with a good control of weeds and that the organic matter turnover formalisms evaluated previously in conventional farming are valid in organic systems.

Table 5.1. Crop rotation and fertilization management for each treatment in DOK and Foulum long term experiments.

Treatments	DOK (Switzerland)				Foulum (Denmark)					
		CONMIN	CONFYM	NOFERT	BIOORG	C4-CC+IF	O4+CC-M	O2+CC-M	O2+CC+M	
Duration	yr	39				19				
Crop rotation ^a	1 st cycle	1978-1984	PO/WW/cc/CB/WW/WB/CG ^b /CG			1997-2000	SP-SB/O/WW/TR	SP-SB/cc/O/WW/WC	WW/cc/SP-SB/cc/SB/CG ^c	
	2 nd cycle	1985-1991	PO/cc/WW/cc/B/cc/WW/WB/CG/CG			2001-2004	LU/WW/O/SB	LU/WW/cc/O/cc/SB/cc	WW/cc/LU-SB/cc/SB/CG	
	3 rd cycle	1992-1998	PO/WW/cc/B/WW/CG/CG/CG			2005-2008	SP-SB/PO/WW/SB	FB/cc/PO/WW/cc/SB/cc	PO/WW/cc/SB/CG	
	4 th cycle	1999-2005	PO/WW/cc/SO/cc/SM/WW/CG/CG			2009-2012	SP-SB/SW/PO/SB	SP-SB/cc/SW/cc/PO/SB/cc	PO/cc/SB/AL/AL	
	5 th cycle	2006-2012	SM/WW/cc/SO/PO/WW/CG/CG			2013-2017	H/SP-SB/SW/O	H/cc/SP-SB/cc/SW/cc/O/cc	SW/cc/PO/cc/SB/CG	
	6 th cycle	2013-2017	SM/SO/WW/cc/PO/cc/SM/WW/CG/CG							
Catch crops		different mixtures of rye, vetch, oat, rapeseed, sunflower, legumes, grass.				mixtures of ryegrass, chicory, fodder radish, clover, black medic, seradella, birdsfoot-trefoil, subterranean clover, vetch.				
Residues management	main crop	exported	exported	exported	exported	returned	returned	returned	returned	
	catch crop	returned	returned	returned	returned	returned	returned	returned	returned	
	clover-grass cuts	exported	exported	exported	exported			returned	exported ^d	
Mineral N fertilization (kg N ha ⁻¹ yr ⁻¹)		97	101	-	-	55	-	-	-	
Organic N fertilizer (kg N ha ⁻¹ yr ⁻¹)		-	54	-	94	-	-	-	50	

AL: alfalfa (*Medicago sativa* L.); B: beetroot (*Bet vulgaris* L.); CB: white cabbage (*Brassica oleracea* L.); CG: clover-grass ley; H: hemp (*Cannabis sativa* L.); LU: lupin (*Lupinus albus* L.); O: oat (*Avena sativa* L.); PO: potato (*Solanum tuberosum* L.); SB: spring barley (*Hordeum vulgare* L.); SM: silage maize (*Zea mays* L.); SO: soybean (*Glycine max* L.); SP: spring pea (*Pisum sativum* L.); SW: spring wheat (*Triticum aestivum* L.); TR: triticale (\times *Triticosecale* Wittm. ex A. Camus); WB: winter barley (*Hordeum vulgare* L.); WC: winter cereal; WW: winter wheat (*Triticum aestivum* L.). The "-" stands for associated crops and "cc" for catch crops.

^a three different crops of the succession are cultivated each year at the DOK, four at Foulum.

^b clover-grass ley composed of a mixture of red clover (*Trifolium pratense* L.); white clover (*Trifolium repens* L.); cock's-foot (*Dactylis glomerata* L.); fescue (*Festuca rubra* L.); timothy-grass (*Phleum pratense* L.); perennial ryegrass (*Lolium perenne* L.); kentucky bluegrass (*Poa pratensis* L.)

^c clover-grass ley composed of a mixture of perennial ryegrass, white clover and red clover.

^d returned to soil before 2007

5.2 Material and methods

5.2.1 Experimental sites and cropping systems

Two long term experiments including arable organic farming were used in this study: 1) the DOK trial, set up in 1978 in Therwil, Switzerland (47°30'N, 7°33'E); 2) the Foulum experiment, initiated in 1997 at the Research Centre of Foulum in Aarhus University, Denmark (56°30'N, 9°34'E). Both experiments were set up to evaluate the agronomic and ecological effects of organic cropping systems. Only the main features of these experiments will be presented here, more precisions can be found in Mäder *et al.* (2002) for DOK experiment, Olesen *et al.* (2000) and Askegaard *et al.* (2011) for Foulum experiment. Both sites compare organic and conventional cropping systems, but they differ by their duration, crop species, length of the rotation and fertilization management (Table 5.1).

Four treatments were selected among the eight treatments available in the DOK trial. This selection represented a total of 48 parcels (4 treatments x 3 crops of the rotation present each year x 4 replicates) arranged in a split-plot block design. The CONMIN treatment was managed as integrated farming (according to the Swiss national guidelines of integrated plant production), exclusively receiving mineral fertilization (nil between 1978 and 1985). The CONFYM treatment was managed as the CONMIN treatment, but with additional organic fertilizers through manure and slurry applications. The NOFERT treatment received neither organic nor inorganic fertilizers since the start of the experiment. The organic treatment, BIOORG, received solely organic fertilizers, without addition of mineral fertilizers nor pesticides. The rate of application of organic fertilizer was set at 1.4 livestock unit ha⁻¹, which corresponds to average application rates of 2.2 and 2.0 t DM ha⁻¹ yr⁻¹ since 1978 in CONFYM and BIOORG, respectively. The total amount of N applied averaged 0, 95, 154 and 92 kg N ha⁻¹ yr⁻¹ in NOFERT, CONMIN, CONFYM and BIOORG treatments, respectively. Soil ploughing was done at around 20 cm depth in all treatments. Weed pressure was mechanically controlled in the BIOORG and NOFERT treatments, while herbicides and pesticides were used in CONMIN and CONFYM treatments when the infection threshold was exceeded.

The Foulum experiment had a factorial design comprising three factors that were i) the presence (2) or absence (4) of a clover-grass ley in the crop rotation; ii) the inclusion (+CC) or exclusion (-CC) of catch crops undersown in the main crop in spring and iii) the addition (+M) or the

absence (-M) of manure. All crops of rotations 2 and 4 were represented every year in each of two completely randomized blocks. Among all treatments, we selected three organic (O) treatments: one treatment without manure application and excluding the clover-grass ley (O4+CC-M), including the clover-grass ley (O2+CC-M) and one treatment including application of manure (O2+CC+M). The average amount of external organic fertilizer applied in O2+CC+M was of 0.59 t DM ha⁻¹ yr⁻¹, as manure and pig slurry, the composition of which varied between years. All organic treatments were managed without pesticides use, according to the European regulation for organic farming. One conventional treatment was also studied (C4-CC+IF), receiving inorganic fertilizers but without clover-grass ley and without catch crop. This treatment had been managed without N fertilization (and pesticides) until 2004, before being converted into a conventional treatment (Askegaard *et al.*, 2011). The amount of total N applied to soil averaged 23 and 51 kg N ha⁻¹ yr⁻¹ in C4-CC+IF and O2+CC+M respectively. All crop residues were returned to soil at harvest for main crops and during the mechanical destruction for catch crop and clover-grass leys. Prior to 2005, the clover-grass ley cuts were left to decompose on the soil in O2+CC-M and O2+CC+M, whereas they were exported from the field in O2+CC+M thereafter.

5.2.2 Climate and soil characteristics

Prior to the initiation of the experiments, soils were characterized in 1977 for DOK and in 1996 at Foulum (Table 5.2). DOK soil is classified as Haplic Luvisol and Foulum soil as a Mollic Luvisol (IUSS Working Group WRB, 2006). The two soils have a contrasted texture, with high silt content (71%) and low sand content (12%) at DOK, and low silt (14%) and high sand content (77%) at Foulum. Clay content is higher at DOK (16% vs 9%). The initial soil organic C content (SOC) was lower at DOK (16.6 vs 22.8 g kg⁻¹) whereas the initial soil organic N content (SON) was similar between sites. In the DOK trial, the initial organic N content was estimated based on the initial SOC content and the final soil C:N ratio measured in 2016. The difference in C/N ratio (8.9 vs 13.0) could result from the difference in the previous land use: arable crops at Foulum vs grassland at DOK. The experimental sites also differed by their climatic conditions, with mean annual precipitation and air temperature of 860 mm and 10.7°C at the DOK (1977-2016), against 716 mm and 8.2°C at the Foulum experiment (1996-2016).

Table 5.2. Topsoil characteristics used for initializing the STICS simulations in DOK and Foulum experiments in 1977 (0-20 cm) and 1996 (0-25 cm), respectively.

Treatment	DOK				Foulum				
	CONMIN	CONFYM	NOFERT	BIOORG	C4-CC+IF	O4+CC-M	O2+CC-M	O2+CC+M	
Texture class	Haplic Luvisol				Mollic Luvisol				
Clay	g kg ⁻¹	167	145	162	151	85	101	90	88
Silt	g kg ⁻¹	700	709	707	714	129	150	138	149
Sand	g kg ⁻¹	113	126	114	114	785	749	772	763
Organic C	g kg ⁻¹	16.2	15.2	18.1	16.7	21.4	24.2	21.5	23.9
Total N ^d	g kg ⁻¹	1.81	1.70	2.03	1.86	1.66	1.81	1.71	1.79
C:N ratio ^d		8.9	8.9	8.9	9.0	12.9	13.3	12.5	13.4
CaCO ₃	g kg ⁻¹	2.2	2.2	2.5	2.3	0	0	0	0
pH _{H2O}		6.18	6.29	6.21	6.30	6.45	6.43	6.59	6.50
Bulk density	g cm ⁻³	1.32	1.32	1.32	1.31	1.42	1.42	1.42	1.42
WFC ^a	g kg ⁻¹	296	301	306	305	192	192	192	192
WPWP ^b	g kg ⁻¹	133	135	138	137	82	82	82	82
PAW ^c	mm	322	327	332	330	234	234	234	234

^a water content at field capacity

^b water content at permanent wilting point

^c plant available water on 150 cm

^d total N calculated at the DOK with the organic C content in 1977 and C/N ratio of 2016

5.2.3 STICS model improvement

STICS is a deterministic soil-crop model simulating agricultural variables (crop development, biomass production, N uptake, N fixation, ...) and environmental variables (soil water, C and N fluxes). Initial soil characteristics (N content, C:N ratio, clay content, ...), daily climatic data, crop characteristics and agricultural practices must be given as input data. Potential crop development and growth are simulated using specific plant parameters, and abiotic stress factors (related to temperature, water or nitrogen) are applied to calculate effective growth rates. The soil is divided into layers, characterized by their water content at field capacity, permanent wilting point and bulk density. Organic matter decomposition in soil is simulated with three compartments: fresh organic matter, microbial biomass and humified organic matter, the latter being composed of an active and a stable fraction (Figure 5.1). C and N fluxes between these pools depend on their C:N ratio, soil temperature, water content and mineral N content, and potential mineralization parameters: decomposition rate of residues, C assimilation yield by the microbial biomass, decay rate of biomass and humification rate (Nicolardot *et al.*, 2001). The parameters of crop residues and organic fertilizers decomposition have been calibrated on large datasets of laboratory incubations.

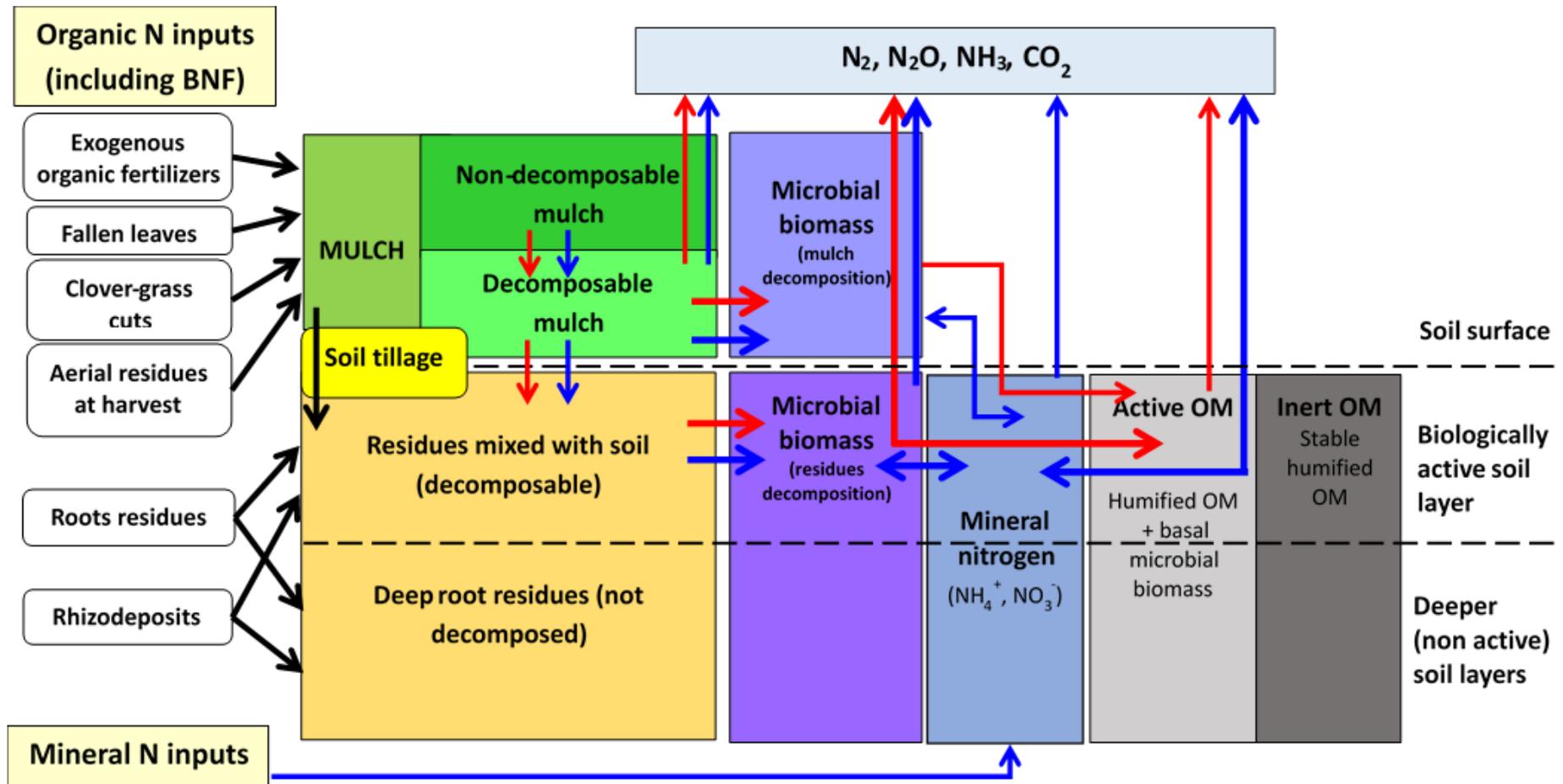


Figure 5.1. Soil C and N compartments and incoming and outgoing C-N fluxes in STICS model. Blue arrows stand for C fluxes and red arrows for N fluxes.

The N mineralized from humified organic matter depends on a potential mineralization rate, related to clay, CaCO₃ and SON contents, and the temperature and moisture conditions of the biologically active soil layer. The transfer of nitrate in soil is described with the mixing cell concept, simulating solute dispersion. Gaseous N losses (NH₃, N₂ and N₂O) are simulated either empirically (fraction of fertilizer lost) or more mechanistically (Peyrard *et al.*, 2017).

A research version of STICS was used in this study in order to widen the range of possibilities offered by the latest standard version (v8.4). We improved the version (v8.42) evaluated by Strullu *et al.* (2015), in order to i) run successive simulations including intercrops; ii) run simulations of clover-grass over successive years; iii) undersow a cover crop in an already established crop and simulate its further growth after harvest of the main crop; iv) return to soil part of grassland cut as a green manure and v) simulate a C-N mineralization peak during the year following grassland destruction. The latter process was mimicked by an artificial input of organic matter, from 2.5 to 5 t DM ha⁻¹ yr⁻¹ according to the grassland age, with a low C/N ratio (12). This add-on is justified by observations made in grassland soils, such as fast release of N after grassland destruction, accumulation of particulate organic matter under grassland (Vertès *et al.*, 2002), increase in microbial biomass (Attard *et al.*, 2016) and deposition of legume nodules rich in nitrogen as proposed by Christensen *et al.* (2009), not simulated by STICS model.

A new parameterization was applied to winter wheat, spring and winter barley, triticale and winter faba-bean using independent datasets obtained in other organic farming experiments. The mixed grassland, consisting in a mixture of grass and legume, was simulated using the existing fescue plant file, in which the biological N fixation (BNF) was activated and calibrated.

5.2.4 Experimental data used for modelling

Data collected throughout the 39 or 19-yr experiment were used for model evaluation. These data concerned the aboveground biomass and N content measured during the crop growth and/or at harvest, along with soil organic carbon (SOC) and nitrogen (SON) contents, soil water content (SWC) and soil mineral nitrogen (SMN) contents at different dates. Details about measurement methodologies are given in previous studies for DOK (Mäder *et al.*, 2007; Leifeld *et al.*, 2009; Mayer *et al.*, 2015) and Foulum (Askegaard *et al.*, 2011; Doltra *et al.*, 2011; Petersen *et al.*, 2013). Complementary measurements of SWC and SMN were realized three

times per year between 2015 and 2017 in the DOK experiment, in order to evaluate the predictions of soil water content and SMN evolution over three successive drainage seasons.

The measured data relative to N inputs and outputs allowed to calculate the N surplus, as follows:

$$N_{sur} = N_{fert} + N_{fix} + N_{atm} - N_{exp} \quad (5.1)$$

where N_{fert} is the N fertilization (mineral + organic), N_{fix} the N input deriving from symbiotic fixation, N_{atm} the atmospheric N deposition and N_{exp} the N exported from the field at harvest, all values in kg N ha⁻¹ yr⁻¹. N_{atm} was estimated based on the European Monitoring and Evaluation Program (<http://www.emep.int/>), providing an annual deposition of 17 and 14 kg ha⁻¹ yr⁻¹ in Switzerland and Denmark for the 1980 -2015 period. The values of N_{fert} and N_{exp} were annual data provided by experimenters, and N_{fix} was calculated for the leguminous crops with the equation proposed by Anglade *et al.* (2015a):

$$N_{dfa} = \alpha \cdot N_y + \beta \quad (5.2)$$

where α and β are the slope and intercept coefficients, specific of each crop (see Appendix 2.B.2), N_y is the nitrogen yield, defined as the total nitrogen accumulated in the aboveground biomass, and calculated as follows:

$$N_y = Y \cdot N_c \cdot NHI^{-1} \quad (5.3)$$

where Y is the harvested crop yield (t DM ha⁻¹), N_c is the nitrogen content in the dry matter (g kg⁻¹), and NHI is the nitrogen harvest index defined as the ratio of N contained in the harvested material to the total N in the aboveground biomass. Nitrogen yield was determined using the measured grain yield for pulses (fababean, lupin, pea and soybean), the estimates of aboveground biomass for the other legumes (alfalfa, vetch and clover), an average value of measured N content for pea, and standard values of N content for the other leguminous species (CORPEN, 1988; Parr *et al.*, 2011; Anglade *et al.*, 2015a). The total amount of N derived from atmosphere in legumes was calculated as the product of N_y and a factor accounting for belowground contributions ($BGN-F$), which varied between legume species (Anglade *et al.*, 2015a).

5.2.5 Simulation strategy

The initialization of the SOM pools involved the initialization of the amount of both carbon and nitrogen (through C:N ratios). In the DOK trial, SWC at field capacity was set as the median of the highest values of SWC measured in mid-winter, and SWC at permanent wilting point was set at 45% of SWC at field capacity. The soil water retention curves established at Foulum (Djurhuus & Olesen, 2000) enabled to define SWC at field capacity ($pF = 2.5$) and permanent wilting point ($pF = 4.2$). The bulk density, which is fixed in the current version of the model, was set at 1.32 g cm^{-3} in the 0-20 cm soil layer at DOK (Leifeld *et al.*, 2009) and 1.42 g cm^{-3} in the 0-30 cm soil layer at Foulum (Djurhuus & Olesen, 2000). The depth of the biologically active layer (“mineralization depth”) was assumed to be 25 cm in both experiments, corresponding to the ploughing depth plus 10% (Brisson *et al.*, 2008). The conventional treatments (CONMIN and C4-CC+IF) closest to cropping systems usually simulated with STICS were selected for model calibration, while all other treatments were used for the model evaluation.

The model was first calibrated against the conventional treatments with a trial-error method. The objective was to find the best compromise in the quality of fit for crop production, N uptake, SWC and SMN contents. During the calibration process, several plant parameters were changed to reach a good simulation of crop growth and N uptake, particularly radiation use efficiency and root traits of beetroot, hemp and silage maize. Continuous simulations were used to calibrate the active fraction of soil organic matter, in order to correctly simulate the evolution of soil organic matter observed in the field.

5.2.6 Model assessment

The model was evaluated both in the conventional treatments used for calibration and the organic treatments used for independent testing, against SWC, SMN, crop biomass, N content and yield, N surplus and soil organic N. A good prediction of these variables is required to trust the C and N fluxes simulated by the model, particularly C and N deposition by crop residues, C and N mineralization, N leaching and gaseous N emissions.

We characterized the model performance by calculating complementary statistical criteria based on the comparison of observed and simulated data. They allowed us to estimate the magnitude of model errors and model ability to reproduce observed data variability for each

output variable. They include the mean difference (*MD*) and the root mean square error (*RMSE*) which are calculated as follows:

$$MD = \frac{1}{n} \sum_{i=1}^n (S_i - O_i) \quad (5.4)$$

$$RMSE = \sqrt{\frac{1}{n} \cdot \sum_{i=1}^n (S_i - O_i)^2} \quad (5.5)$$

where *O* and *S* are the observed and the simulated values respectively and *n* is the number of observed-simulated pairs. *MD* gives the bias of the model, whereas *RMSE* gives an estimation of the magnitude of the model error. It can be decomposed into two components describing the systematic error (*RMSE_s*) and the unsystematic error (*RMSE_u*), calculated as follows (Willmott, 1981):

$$RMSE_s = \sqrt{\frac{1}{n} \cdot \sum_{i=1}^n (\bar{S}_i - O_i)^2} \quad (5.6)$$

$$RMSE_u = \sqrt{\frac{1}{n} \cdot \sum_{i=1}^n (S_i - \bar{S}_i)^2} \quad (5.7)$$

with \bar{S}_i deriving from the following linear regression of predicted vs. observed values: $\hat{S}_i = a + bO_i$, *a* and *b* being the slope and intercept of the regression, respectively. *RMSE_s* gives the systematic bias of the model, while *RMSE_u* reveals the dispersion of the simulated values. A prevalence of systematic error means that there was an error during model calibration and parameterization or that the model misses important process(es) needed to simulate with accuracy the behaviour of the soil–crop system. Unsystematic error is linked to i) inputs or measurements uncertainty or ii) effect of exceptional environmental conditions or biotic stresses not taken into account by the model.

We considered that model predictions are satisfactory (acceptance criterion) if two conditions are fulfilled: i) the bias (estimated by *MD* and *RMSE_s*) is small; ii) the model error (*RMSE_u*) is equal or lower than the data variability in measurements (standard deviation).

5.2.7 Statistical analysis

The mean annual variables related to C and N balances were analysed statistically for each experiment, using a repeated measures mixed model with cropping system as fixed effect. Analysis of variance (ANOVA) was performed to test the effect of cropping system on the previous C and N fluxes affected. Residues of the model were verified by the Shapiro-Wilk and Levene test respectively. When needed, a BoxCox transformation was used in order to normalize the data. When significant differences among treatments were identified, a LSD test was applied at the 5% probability level of significance. In case of discordance, the non-parametric test of Kruskal-Wallis was used, followed by means comparison with the *kruskal.test* from the *agricolae* package (De Mendiburu, 2014).

5.3 Results

5.3.1 Evaluation of STICS for biomass production and N uptake

The global evaluation of STICS model for the aerial biomass and the crop N content is displayed in Table 5.3. In the conventional treatments, the simulations with calibration gave a slight overestimation of the harvested biomass (1.1 and 0.8 t DM ha⁻¹), and an average RMSE of 2.3 and 2.2 t DM ha⁻¹ at DOK and Foulum, respectively. The bias was slightly higher in the validation treatments (1.6 and 1.1 t DM ha⁻¹ on average at DOK and Foulum). The RMSEs was always lower than the RMSE_u, indicating that model hardly simulated the data variability. Conversely, the model could satisfactory reproduce the dispersion of harvest yields, reaching highest values for potato (16.2 t DM ha⁻¹) and lowest for white cabbage (0.3 t DM ha⁻¹). The exported biomass was overestimated in the unfertilized treatments (NOFERT and O2+CC-M).

In spite of the calibration, the exported N in harvested biomass was slightly underestimated in the conventional treatments of both experiments ($MD = 12$ and 22 kg N ha⁻¹). In the DOK trial, the model error mainly came from dispersion ($RMSE_u = 40$ kg N ha⁻¹) rather than a systematic error ($RMSE_s = 17$ kg N ha⁻¹). In the Foulum experiment, the difference between $RMSE_u$ and $RMSE_s$ was lower, with 35 and 27 kg N ha⁻¹ respectively. The evaluation gave better results. In DOK, N content in exported biomass was well simulated in the BIOORG treatment, with a lower $RMSE$ (38 kg N ha⁻¹), underestimated in CONFYM and overestimated in NOFERT. At Foulum, the exported N in biomass was well simulated in the organic treatments of rotation 2 ($MD = 3$ kg N ha⁻¹), and slightly overestimated in the O4+CC-M treatment ($MD = -12$ kg N ha⁻¹ on average).

The exported biomass in clover-grass cuts was well predicted in the conventional treatments, with a mean difference of 0.3 t DM ha⁻¹ in CONMIN. The corresponding N exported was slightly overestimated (+11 kg N ha⁻¹) with an important $RMSE_s$ (30 kg N ha⁻¹). The evaluation phase showed an overestimation of the exported biomass of clover-grass (including O2+CC+M, the only treatment in which cuts were exported at Foulum). The corresponding N exportations were overestimated by 14 kg N ha⁻¹ on average, the mean $RMSE_s$ (29 kg N ha⁻¹) being equivalent to that of calibration.

Table 5.3. Performance of STICS model for the dataset used for the calibration (bold) and the evaluation of aboveground biomass and N content. Values in brackets are standard deviations.

		DOK				Foulum			
		CONMIN	CONFYM	NOFERT	BIOORG	C4-CC+IF	O4+CC-M	O2+CC-M	O2+CC+M
exported biomass ^a (t DM ha ⁻¹)	n	64	64	64	63	86	86	64	64
	X obs	5.4 (2.8)	5.8 (3.0)	2.5 (1.0)	4.5 (2.2)	4.1 (2.3)	3.1 (1.3)	3.6 (1.3)	4.1 (1.5)
	X sim	6.5 (3.5)	7.0 (3.9)	4.7 (2.5)	5.9 (3.1)	4.9 (3.2)	3.7 (2.4)	4.9 (3.5)	5.3 (3.6)
	RMSE	2.4	2.4	3.3	2.8	2.3	2.2	3.2	3.3
	RMSEs	1.1	1.2	2.3	1.3	0.8	0.7	1.5	1.3
	RMSEu	2.2	2.1	2.4	2.5	2.1	2.1	2.9	3.0
exported N at harvest ^a (kg N ha ⁻¹)	n	69	69	69	69	83	83	63	63
	X obs	126 (41)	139 (43)	66 (30)	104 (41)	88 (49)	66 (39)	68 (27)	80 (28)
	X sim	114 (50)	123 (51)	80 (51)	99 (45)	66 (35)	54 (35)	72 (52)	82 (53)
	RMSE	44	57	41	42	44	39	46	46
	RMSEs	17	30	14	18	35	25	5	2
	RMSEu	40	48	38	38	27	30	46	46
exported clover- grass cuts (t DM ha ⁻¹)	n	182	182	182	182				62
	X obs	2.6 (1.3)	2.9 (1.2)	1.6 (0.9)	2.5 (1.2)				3.4 (1.9)
	X sim	2.9 (1.0)	3.1 (1.0)	2.4 (0.8)	2.9 (1.0)				4.0 (2.8)
	RMSE	1.1	1.0	1.2	1.0				2.4
	RMSEs	0.7	0.6	1.0	0.7				0.7
	RMSEu	0.8	0.8	0.8	0.8				2.3
exported N in clover-grass cuts (kg N ha ⁻¹)	n	177	177	177	177				56
	X obs	69 (35)	78 (35)	46 (27)	69 (35)				73 (40)
	X sim	79 (24)	90 (25)	56 (16)	77 (21)				99 (58)
	RMSE	38	40	29	34				62
	RMSEs	30	31	24	28				31
	RMSEu	23	25	15	19				53

total aerial biomass ^b (t DM ha ⁻¹)	n	241	241	241	241	62	89	123	119
	X obs	5.1 (5.0)	5.6 (5.3)	3.1 (3.1)	4.8 (4.6)	10.6 (3.9)	6.9 (3.8)	5.3 (3.4)	5.8 (4.1)
	X sim	5.6 (5.2)	6.0 (5.6)	4.4 (3.8)	5.3 (4.7)	10.2 (3.0)	6.4 (3.8)	5.8 (4.5)	6.5 (5.2)
	RMSE	1.8	2.1	2.2	1.6	3.3	2.9	3.4	3.7
	RMSEs	0.5	0.4	1.3	0.5	2.2	1.2	0.5	0.7
	RMSEu	1.7	2.1	1.8	1.6	2.4	2.7	3.3	3.7
total aerial N uptake ^b (kg N ha ⁻¹)	n	240	240	240	240	58	85	111	107
	X obs	96 (61)	107 (65)	61 (39)	89 (54)	138 (66)	93 (60)	88 (41)	91 (45)
	X sim	101 (50)	114 (54)	71 (42)	96 (45)	100 (41)	66 (32)	98 (57)	109 (57)
	RMSE	43	48	37	40	65	58	58	72
	RMSEs	25	28	16	24	56	51	23	33
	RMSEu	35	39	33	32	33	27	53	64

n = number of observed/simulated data pairs, *X obs* = mean of measured values, *X sim* = mean of simulated values, *RMSE* = root mean square error, *RMSEs* = systematic RMSE, *RMSEu* = unsystematic RMSE

^a *except clover-grass cuts*

^b *grain, stubble and straw*

The model simulated correctly the aerial crop biomass after calibration of the conventional treatments, with a small mean difference of 0.4 and -0.4 t DM ha⁻¹ at DOK and Foulum, respectively. Their respective *RMSE_u* were 1.7 and 2.4 t DM ha⁻¹. The N accumulated in aerial biomass was well simulated in the DOK calibration treatment (*MD* = 5 kg N ha⁻¹), but underestimated at Foulum (*MD* = -38 kg N ha⁻¹). The evaluation showed small differences between observed and simulated aboveground biomass with a *MD* of 0.7 and 1.1 t DM ha⁻¹ at DOK and Foulum, respectively. The corresponding N content simulated in aerial biomass varied according to treatments, with a general overestimation for all treatments at DOK and in organic treatments of rotation 2 at Foulum, and an underestimation in O4.

On the basis of the acceptance criterion, we conclude that model was satisfactory for predicting crop biomass, for both harvested organs and total aboveground biomass. The N accumulated in aboveground biomass was not always well captured (*RMSE_u* equal or greater than the standard deviations of measurements) despite a parametrization of plant files. The bias which appears in some treatments is likely to be related with a poor simulation of soil mineral N content.

5.3.2 Evaluation of STICS for soil water and mineral N

The results of simulation of soil water and nitrate contents are presented in Table 5.4, on the 0-90 cm soil layer for DOK and 0-25 cm soil layer for Foulum. Soil water content was well simulated in conventional treatments, model residuals being low in both experiments with a *RMSE* of 24 and 16 mm for DOK and Foulum respectively. The *RMSEs* was lower than *RMSE_u* at DOK (13 vs 21 mm, respectively) whereas the opposite result was found at Foulum (15 vs 6 mm, respectively), indicating a bias in simulating soil water content in the sandy soil. Similar results were found in the organic treatments, with an average *RMSE* of 24 and 18 mm for DOK and Foulum, respectively. The *RMSEs* was also lower than the *RMSE_u* at DOK (15 vs 19 mm) and *vice versa* at Foulum (16 vs 6 mm on average). Soil water was therefore satisfactorily simulated by the model.

Table 5.4. Performance of STICS for predicting soil water and nitrate contents (0-90 cm at DOK and 0-25 cm at Foulum). Treatments used for the calibration are in bold, other treatments were used for the evaluation.

		DOK				Foulum			
		CONMIN	CONFYM	NOFERT	BIOORG	C4-CC+IF	O4+CC-M	O2+CC-M	O2+CC+M
soil water content (mm)	n	73	73	28	76	34	31	27	34
	X obs	322 (39)	322 (37)	334 (34)	322 (40)	87 (19)	98 (16)	88 (16)	95 (21)
	X sim	322 (34)	318 (34)	339 (26)	323 (33)	86 (7)	85 (8)	84 (9)	86 (8)
	RMSE	25	15	31	26	16	18	14	21
	RMSEs	13	7	21	15	15	17	12	20
	RMSEu	21	13	22	21	6	7	8	2
soil nitrate content (kg N ha ⁻¹)	n	116	119	42	118	74	17	53	57
	X obs	47 (36)	56 (40)	56 (45)	50 (37)	42 (51)	9 (9)	16 (27)	23 (6)
	X sim	25 (26)	31 (31)	26 (26)	28 (37)	21 (24)	6 (3)	9 (11)	12 (2)
	RMSE	36	40	48	40	50	9	32	43
	RMSEs	30	32	43	32	45	9	30	41
	RMSEu	21	24	22	25	22	3	11	13

n = number of observed/simulated data pairs, *X obs* = mean of measured values, *X sim* = mean of simulated values, *RMSE* = root mean square error, *RMSEs* = systematic *RMSE*, *RMSEu* = unsystematic *RMSE*

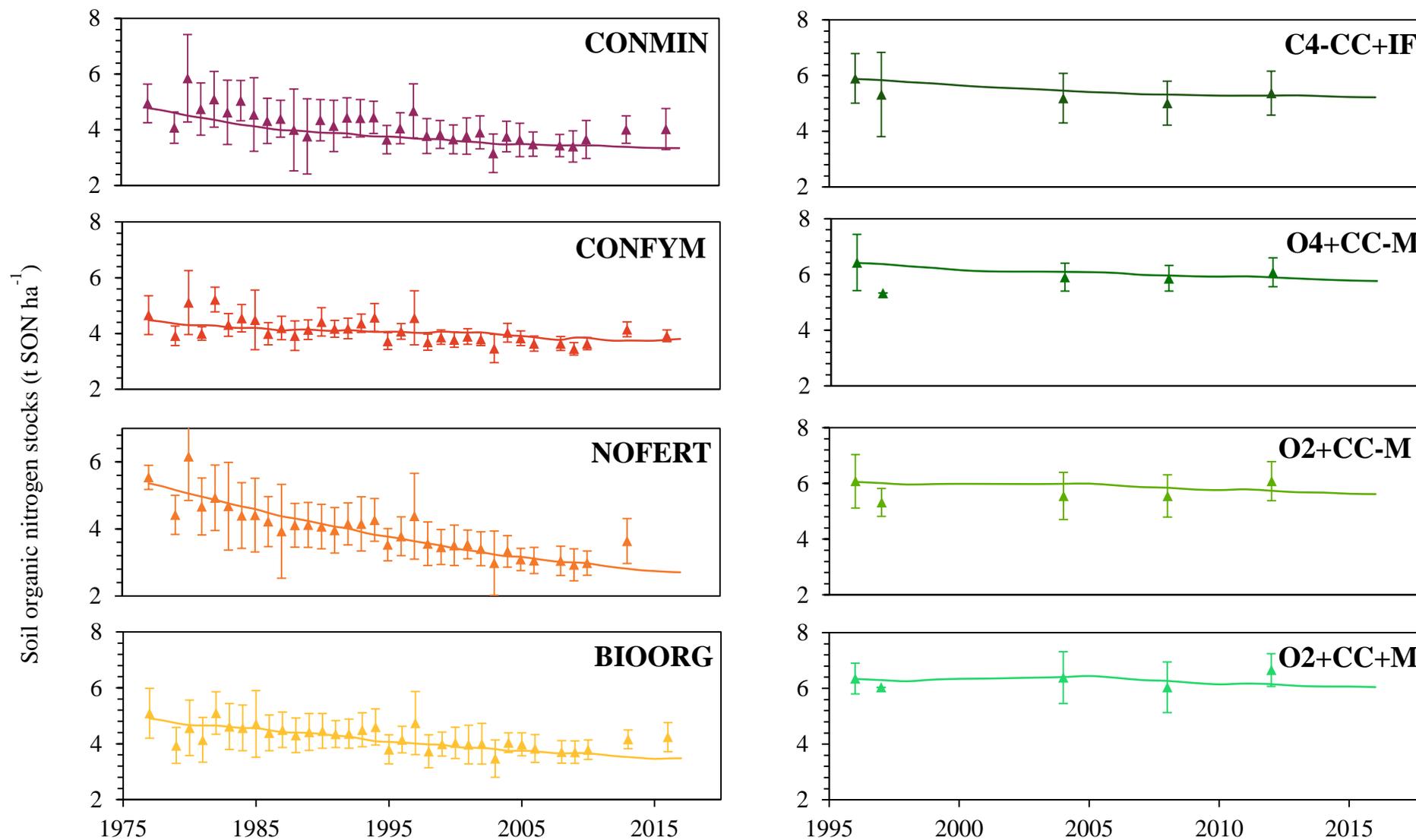


Figure 5.2. Temporal evolution of SON stocks in DOK trial (left side) over 0-20 cm and in Foulum experiment (right side) over 0-25 cm. Symbols (▲) display the observed SON stocks, \pm SD. Lines are simulated values, based on simulations of single parcels ($n = 3$ at DOK, $n = 4$ at Foulum).

Soil mineral N was markedly under-estimated by the model in all treatments of the DOK trial. The bias was -22 kg N ha^{-1} in the CONMIN and -21 kg N ha^{-1} in the C4-CC+IF treatment. This poor agreement was mainly explained by the model error than its dispersion, the $RMSEs$ being higher than the $RMSEu$ for both experiments. The model predicted more satisfactorily SMN at Foulum, with a mean difference of -7 kg N ha^{-1} . However, the $RMSEs$ were always higher than the $RMSEu$ for the evaluation. The model could simulate data variability, but with a systematic error.

5.3.3 Organic C and N balances

The temporal evolution of observed and simulated SON stocks is shown at Figure 5.2. The SON stocks evolution was very well simulated in both experiments. SON stocks decreased markedly in all treatments of the DOK experiment. In this trial, the decrease in SON stocks was well captured by the model, possibly with a slight underestimation in the last years. The model error ($RMSE = 0.38 \text{ t ha}^{-1}$) was much lower than the measurement error (mean standard deviation = 0.63 t ha^{-1}). In the Danish experiment, the observed SON stocks slightly decreased throughout time. The model could capture this slow decrease for all treatments. The model error ($RMSE = 0.38 \text{ t ha}^{-1}$) was also much lower than the measurement error (mean standard deviation = 0.71 t ha^{-1}), confirming the satisfactory quality of prediction.

The simulated annual rate of changes of soil organic C and N stocks are summarized in Table 5.5, along with the components of the C and N balance over the whole soil profile. These estimations are done for the entire period considered, *i.e.* 39 years for DOK and 19 years at Foulum. In the DOK experiment, the rate of SON change ranged as follows: CONFYM < BIORG = CONMIN < NOFERT. Similar trends were observed for changes in SOC stocks. N contained in deep root residues (dead roots below the ploughing layer) increased with time, since their decomposition was not simulated by the model, at the rate of $8\text{-}14 \text{ kg N ha}^{-1} \text{ yr}^{-1}$.

Table 5.5. Simulated balance of soil organic C and N. Values are the mean of 39 years and three replicates for DOK, 19 years and four replicates for Foulum.

		DOK				Foulum			
		CONMIN	CONFYM	NOFERT	BIOORG	C4-CC+IF	O4+CC-M	O2+CC-M	O2+CC+M
Organic C (kg C ha ⁻¹ yr ⁻¹)	changes in SOC stocks ^a	-476 c	-172 a	-713 d	-359 b	-368 ab	-326 a	-387 b	-319 ab
	changes in deep root C ^a	341 a	372 a	278 a	334 a	82 c	145 b	232 a	197 ab
input fluxes	organic fertilizer ^b	0 c	1107 a	0 c	1010 b	0 b	1 ab	-226 c	31 a
	crop residues ^a	1501 ab	1584 a	1223 c	1416 b	1928 d	2338 c	3391 a	2951 b
	dead roots ^a	1305 ab	1413 a	1032 c	1275 b	719 b	647 b	1088 a	1172 a
	mineralization ^a	2940 c	3903 a	2688 d	3728 b	2935 b	3143 b	4500 a	4343 a
	output fluxes								
Organic N (kg N ha ⁻¹ yr ⁻¹)	changes in SON stocks ^a	-39 b	-15 a	-78 c	-39 b	-31 b	-28 b	-26 ab	-18 a
	changes in deep root N ^a	12 ab	14 a	8 b	12 ab	1 c	4 b	10 a	9 a
input fluxes	organic fertilizer ^b	0 c	54 b	0 c	64 a	0 c	0 b	1 b	20 a
	crop residues ^a	71 b	67 a	39 c	48 b	41 d	74 c	138 a	109 b
	dead roots ^a	54 ab	63 a	36 c	53 b	12 b	16 b	42 a	47 a
	mineralization ^a	152 b	185 a	144 c	193 a	83 c	114 b	197 a	185 a
output fluxes									

^a simulated data and ^b observed data

Letters indicate significant differences between cropping systems for each field experiment ($p < 0.05$).

The C and N input fluxes derived from organic fertilizer, crop residues and total dead roots (over the whole soil profile) were highest in CONFYM and smallest in NOFERT. The N mineralization rate varied from 144 (NOFERT) to 193 (BIOORG) kg N ha⁻¹ yr⁻¹.

In the Foulum experiment, the rate of SON change varied between -31 (C4-CC+IF) to -18 (O2+CC-M) kg N ha⁻¹ yr⁻¹. Similar trends were observed for changes in SOC stocks. The main factor determining N fate was the rotation and not the treatment. N contained in deep root residues increased at small rate in rotation 4 (1-4 kg N ha⁻¹ yr⁻¹) and faster in rotation 2 with grass ley (9-10 kg N ha⁻¹ yr⁻¹). The N inputs derived from crop residues and total dead roots were much lower in rotation 4 than in rotation 2, due to the inclusion of clover-grass in the latter rotation. The annual N mineralization rate also varied widely between rotations: 83-114 kg N ha⁻¹ yr⁻¹ in rotation 4 and 185-197 kg N ha⁻¹ yr⁻¹ in rotation 2.

5.3.4 N surplus

The simulated N surplus varied among experiments and treatments (Table 5.6). In DOK trial, N surpluses varied between treatments and ranked as follows CONFYM < BIOORG < CONMIN < NOFERT, the N surplus being positive only in the CONFYM treatment. Less contrasted N surpluses were found at Foulum, and only the O4+CC-M treatment had a negative N surplus, significantly lower than the other treatments. The differences in N surpluses result from the diversity in quality and quantity of inputs and outputs.

In DOK trial, total N inputs were highest in the CONFYM treatment, 62% deriving from fertilization and 31% from the BNF. N inputs were similar in the CONMIN and BIOORG treatments (185-191 kg N ha⁻¹ yr⁻¹), 50% coming from fertilization and 41% from BNF. BNF contributed to 80 % of total N inputs in NOFERT. Total N exportations were highest in CONFYM and lowest in NOFERT.

At Foulum, the O2+CC+M treatment had the highest N inputs, 38% deriving from fertilization and 51% from the BNF. Total N inputs did not differ significantly between C4-CC+IF and O2+CC-M, but had different origins, particularly in O2+CC-M treatment which N inputs relied by 81% on BNF. The total N outputs followed the same ranking as N inputs between treatments, the highest N exportations occurring in O2+CC+M (120 kg N ha⁻¹ yr⁻¹), in which clover-grass cuts were exported.

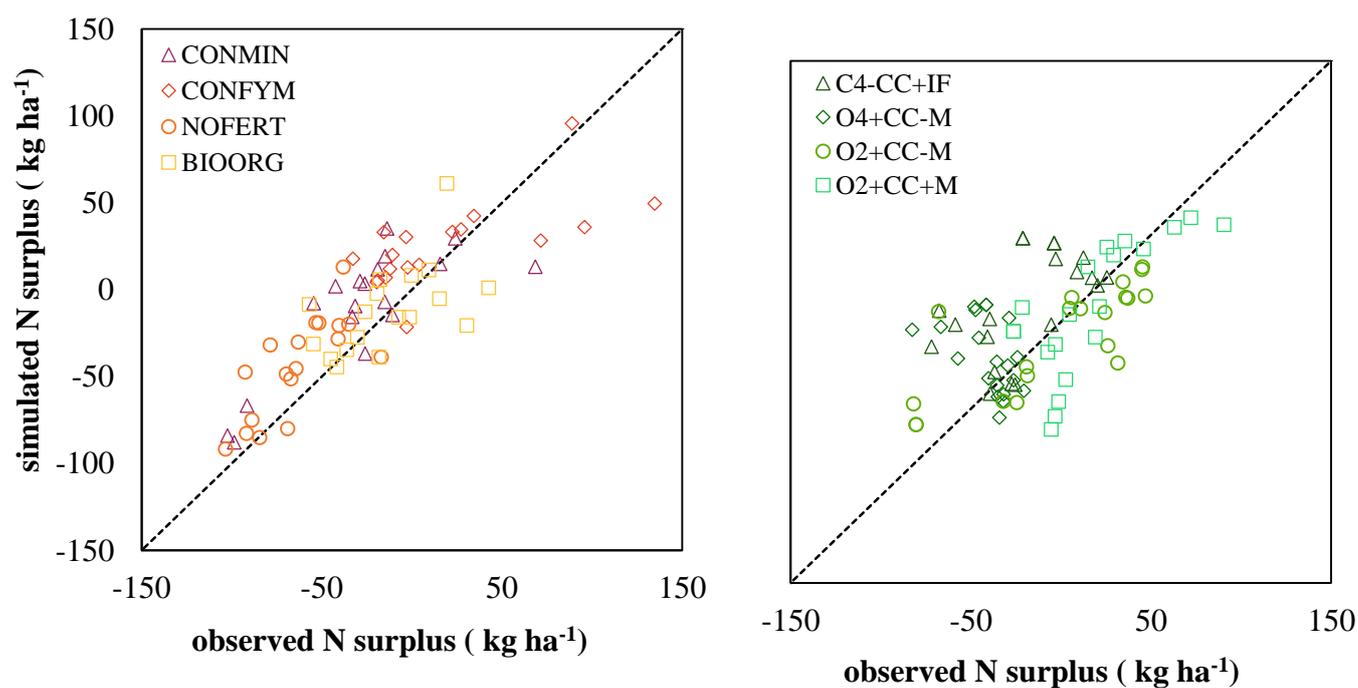
Table 5.6. Mean values of N inputs, N exported and N surplus ($\text{kg N ha}^{-1} \text{ yr}^{-1}$). Total N input is the sum of mineral and organic N fertilization, BNF and atmospheric N deposition.

	DOK				Foulum			
	CONMIN	CONFYM	NOFERT	BIOORG	C4-CC+IF	O4+CC-M	O2+CC-M	O2+CC+M
N fertilization ^b	97 b	155 a	0 c	94 b	56 a	1 b	1 b	54 a
BNF ^a	78 a	79 a	66 a	74 a	15 c	35 b	72 a	72 a
atmospheric N deposition ^a	17	17	17	17	15	16	16	16
total N input ^a	191 b	251 a	82 c	185 b	86 b	52 c	89 b	141 a
total N exported ^a	205 b	230 a	141 d	192 c	77 b	60 c	78 b	120 a
N surplus ^a	-13 c	21 a	-58 d	-7 b	9 a	-8 b	12 a	21 a

^a simulated data and ^b observed data

Letters indicate significant differences between treatments in each field experiment ($p < 0.05$).

Figure 5.3. Comparison of the simulated and observed N surplus. Each dot refers to the average N surplus for a given crop cycle for each treatment in the DOK (A) and Foulum (B) experiments.



The simulated surplus was compared to the ‘observed’ surplus, calculated for each treatment and each crop cycle (Figure 5.3). For the DOK, the observed surplus varied between -103 and $135 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ and the simulated surplus between -92 and $96 \text{ kg N ha}^{-1} \text{ yr}^{-1}$. Both variables were well correlated ($R = 0.81$), but the model slightly overestimated the N surplus, by $11 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ on average. The correlation was smaller in Foulum experiment ($R = 0.58$), the conventional treatment being overestimated by $9 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ on average while the O2+CC-M and O2+CC+M treatments were underestimated by $8 \text{ kg N ha}^{-1} \text{ yr}^{-1}$.

5.3.5 Nitrogen fate

The components of the N surplus in each treatment are presented in Figure 5.4. A positive surplus implies N losses in the environment and/or positive soil N storage whereas a negative surplus implies a decline in soil organic N. In the DOK experiment, the N surplus varied widely between treatments, and most of its variation resulted in changes in soil N pools. Changes in deep root residues (below the ploughed layer) did not differ between treatments. N losses were small and did not differ significantly between treatments, whether through leaching ($6 \text{ kg N ha}^{-1} \text{ yr}^{-1}$), denitrification ($9 \text{ kg N ha}^{-1} \text{ yr}^{-1}$) and volatilization ($3 \text{ kg N ha}^{-1} \text{ yr}^{-1}$).

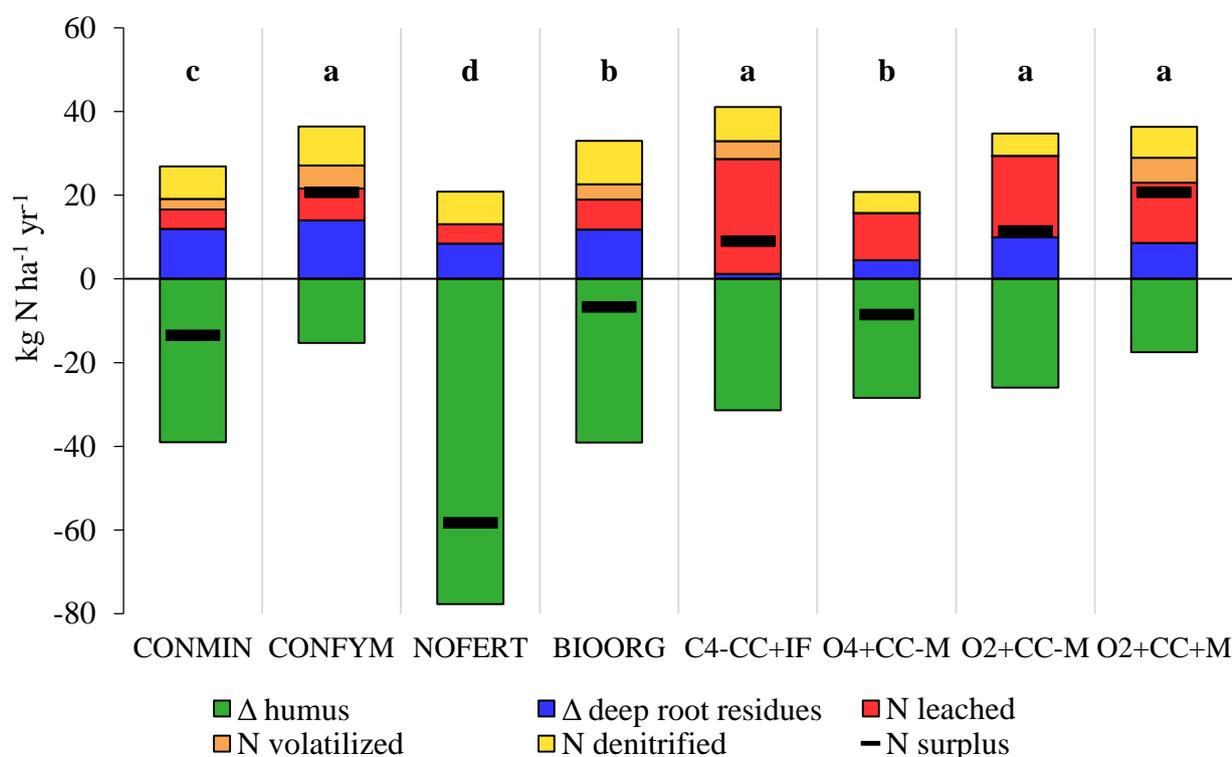


Figure 5.4. Decomposition of the simulated N surplus between changes in SON stocks, N leaching and gaseous N emissions.

In Foulum experiment, N surplus varied little between treatments whereas its fate differed significantly. If changes in SON stocks and deep root residues were mainly affected by the rotation, N losses varied with treatments. N leaching was smallest in treatment O4+CC-M (11 kg N ha⁻¹ yr⁻¹) and highest in the conventional treatment (27 kg N ha⁻¹ yr⁻¹). The gaseous losses were similar in C4-CC+IF and O2+CC+M, on average 13 kg N ha⁻¹ yr⁻¹ for both denitrification and volatilization. They were smaller (5 kg N ha⁻¹ yr⁻¹) in the O4+CC-M and O2+CC-M treatments, and derived from denitrification only.

5.3.6 N mineralization

The temporal evolution of the simulated annual mineralization of N derived from humified organic matter (SON) and organic residues and the total N mineralization is given in Figure 5.5. In DOK trial, the annual mineralization from SON pool was 149 kg N ha⁻¹ yr⁻¹ varied between treatments. It reached 172 kg N ha⁻¹ yr⁻¹ for CONFYM and BIOORG, and was significantly lower in CONMIN and NOFERT (average 127 kg N ha⁻¹ yr⁻¹). Conversely, the mineralization deriving from crop residues was similar in all treatments, averaging 21 kg N ha⁻¹ yr⁻¹. Annual variations were linked to manure application and clover-grass destruction. The total N mineralization was also higher in CONFYM and BIOORG (189 kg N ha⁻¹ yr⁻¹) compared to NOFERT and CONMIN (148 kg N ha⁻¹ yr⁻¹), and the differences between treatments increased after 1990.

At Foulum, the SON mineralization was very stable across years, while the net mineralization rate of crop residues varied widely between treatments and years. The latter was high in rotation 2 (95 kg N ha⁻¹ yr⁻¹), low in O4+CC-M (28 kg N ha⁻¹ yr⁻¹) and very low in C4-CC+IF (-5 kg N ha⁻¹ yr⁻¹). In addition, a temporal shift was observed in rotation 2 after 2006, with a net decline in residue-derived mineralization. The total N mineralization exacerbated the differences in humus and residues N mineralization between treatments, with differences appearing early after the start of the experiment. Over the 19 yr period, the mean amount of N mineralized was 99 kg N ha⁻¹ yr⁻¹ in rotation 4 and 188 kg N ha⁻¹ yr⁻¹ in rotation 2.

Hence, significant differences between treatments originated from humus mineralization at the DOK, while it originated from residues mineralization at Foulum. They highlight the variability of N availability between organic systems, according to their management.

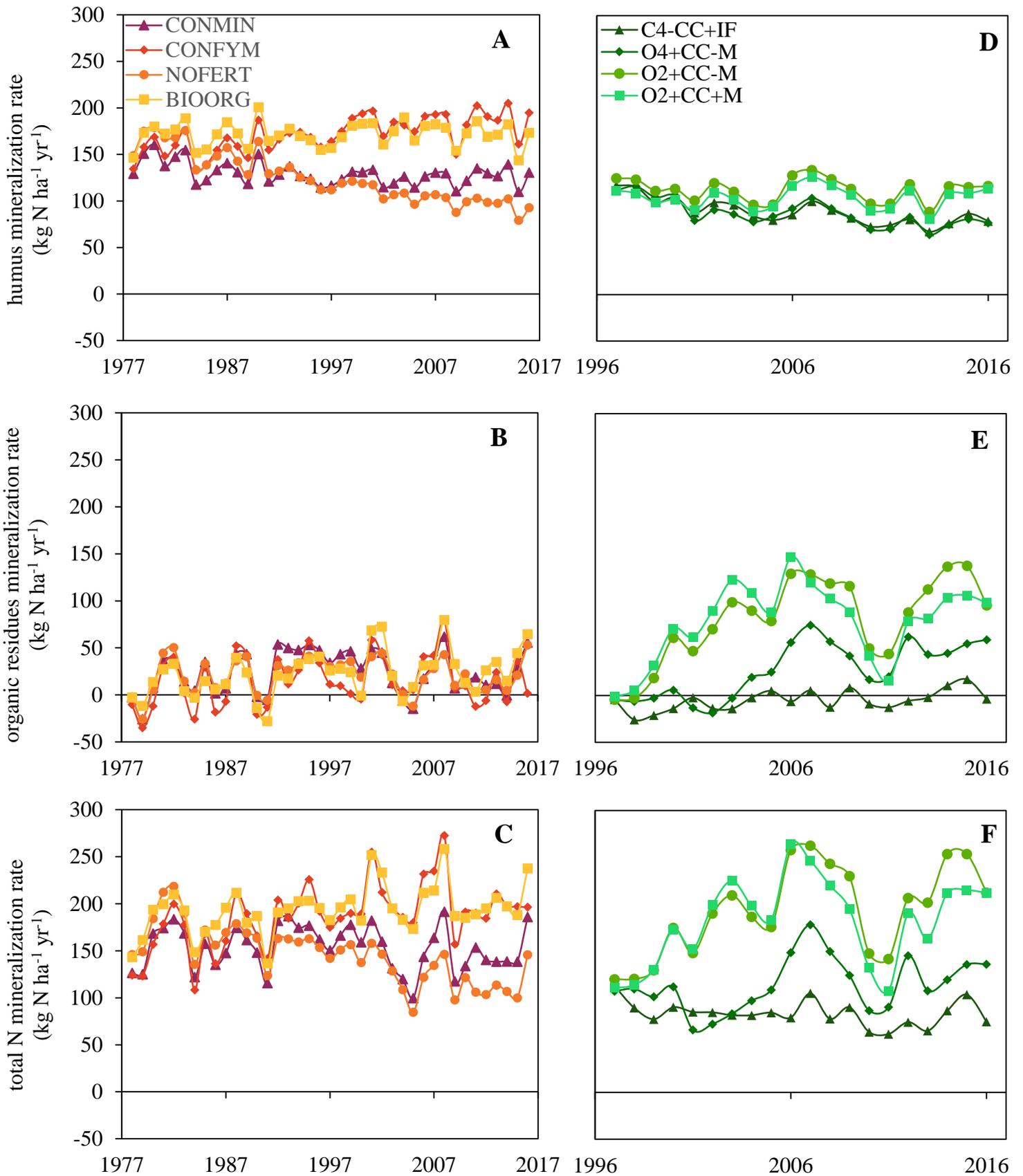


Figure 5.5. Simulated annual mineralization rate of humified N, organic residue and total N at the DOK (A, B and C) and Foulum (E, F and G) experiments, respectively.

5.4 Discussion

5.4.1 Performance of STICS model

The model satisfactorily simulated crop biomass, both exported biomass (grains, tuber, clover-grass cuts) and total aboveground biomass. The quality of prediction was similar for organic and conventional systems, validating our hypothesis. Harvested N and aboveground N contents were more poorly simulated, confirming results obtained by Coucheney *et al.* (2015). Differences between observed and simulated crop N content are related to the ability of the model to simulate soil water and mineral N available for crops as N demand from the crop.

SWC was accurately simulated in DOK trial as well as Foulum. However, the measurements at Foulum were only available in the upper layer, so that it is difficult to ensure that water content was well simulated in the whole profile. SMN was systematically underestimated (by 45- 48%) all treatments of DOK and in the conventional treatment at Foulum, but a better prediction was obtained in the organic treatments of Foulum (relative MD = -19% to +13%). However, SMN measurements were too scarce to ensure a systematic underestimation all along the experiment. For example, the small amounts of SMN measured under the clover-grass ley were well reproduced by the model. The evolution of SMN contents after clover-grass destruction was partly but not fully mimicked. This underestimation may be due to the root turnover of the clover-grass ley, with clover having a higher turnover than grasses, therefore leading to higher N inputs from clover-grass leys (Rasmussen *et al.*, 2008), in contrast with the single root turnover applied in the plant file used for simulation.

Finally, the underestimation of crop yield for some years can be explained by the fact that the model did not take into account crop stresses linked with potassium and phosphorus shortage. Oehl *et al.* (2002) reported a significant decrease in soil P content in the NOFERT treatment from 1978 to 2002 at the DOK, potentially leading to low yields compared to the simulations. In addition, biotic stresses such as diseases, pests and competition with weeds for nutrients are not considered by the STICS model, *e.g.* fungal disease in organic wheat (Gunst *et al.*, 2006) and potato late blight (Zihlmann *et al.*, 2004) at the DOK or weed pressure at Foulum (Olesen *et al.*, 2007). Unsatisfactory plant file parametrization could also decrease the quality of simulations, showing a need for improving orphan crops, such as beetroot, hemp, faba-bean, especially for organic cultivars.

5.4.2 Simulation of soil N surplus

The N surplus has been promoted as an environmental indicator revealing the potential N losses from cultivated lands (OECD, 2001). According to Oenema (2005), a reduction of the N surplus should decrease the risk of N losses. However, it should be noted that a positive N surplus may indicate low losses and a potential N storage in soil organic nitrogen pool (Poudel *et al.*, 2001; Watson *et al.*, 2002b; Anglade *et al.*, 2015b). Despite a small overestimation of the N surplus, the model could correctly reproduce its large variability among treatments and years, varying from -276 to +331 kg N ha⁻¹ yr⁻¹ (observed) and -235 to +348 kg N ha⁻¹ yr⁻¹ (simulated). Anglade *et al.* (2015b) found a N surplus 26% lower in organic than in conventional cropping systems, partly due to the smaller inputs (-12 %) in organic systems. We observed a similar ranking in the DOK trial, where the N surplus was lower in BIOORG than in CONFYM, the former receiving 39% less N inputs. Compared to CONMIN, the BIOORG received similar N inputs but exhibited a higher surplus. In Foulum, the N surplus did not differ significantly between the C4-CC+IF and O2+CC+M treatments, receiving similar amount of N inputs. Therefore, organic systems receiving as much N input as conventional can have equal or higher N surplus (Reganold & Wachter, 2016). Thus, the difference between conventional and organic treatments cannot be detected by the N surplus, while they may result in contrasted N losses.

5.4.3 Drivers of N leaching in organic systems

Part of the N surplus is converted into N losses. Among these losses, nitrate leaching which depends on soil type can be well simulated by the model as shown by Coucheney *et al.* (2015). We simulated a low leaching in the DOK trial (6 kg N ha⁻¹ yr⁻¹ on average), similar in the conventional, the low input and the organic treatments. In our study, the underestimation of SMN might have led to underestimate N leaching. Only one paper reported measurements of SMN under potato crop from 1999 to 2002 in DOK, after destruction of the previous clover-grass ley (Zihlmann *et al.*, 2004). The authors found that SMN contents were slightly higher in manured treatments than in CONMIN, increasing the risk of leaching in the following autumn-winter period. The higher, although not significant, leaching simulated in CONFYM and BIOORG (7 kg N ha⁻¹ yr⁻¹) compared to CONMIN and NOFERT (4 kg N ha⁻¹ yr⁻¹) is consistent with this conclusion. Hence, it appears that N leaching was mainly regulated by the type of soil cover rather than the amount of N inputs (CONMIN *vs* CONFYM) or type of fertilizer used (mineral fertilizer *vs* manure and slurry).

In the Foulum experiment, the model indicated that organic treatments significantly impacted nitrate leaching. Compared to the conventional treatment, which had the highest leaching (28 kg ha⁻¹ yr⁻¹), the N leaching was significantly reduced by 60% in O4+CC-M, where no manure was applied and a catch crop was grown during autumn and winter. Askegaard *et al.* (2011) found in the same experiment that the use of catch crops could reduce nitrate leaching by 7 to 63%. Therefore, including catch crops in the organic treatments should have contributed to reduce nitrate leaching. Indeed, STICS simulated a 24% reduction in leaching in the two organic treatments (rotation 2) compared to the conventional (rotation 4). Conversely, the inclusion of a clover-grass ley in the rotation had no significant effect on leaching compared to the organic rotation 4 (Askegaard *et al.*, 2011) or to the conventional treatment (Pugesgaard *et al.*, 2017). Mondelaers *et al.* (2009) reported no correlation between the proportion of grass in the rotation and N leaching in simulation studies.

Our simulations corroborate the finding that N leaching is relatively insensitive to the source and amount of fertilizer N applied (Stopes *et al.*, 2002; Stark *et al.*, 2006; Brozyna *et al.*, 2013), up to levels of fertilization close to the economic optimum. (Benoit *et al.*, 2014) tempered this result, explaining that the leached N concentration would vary according to type of organic fertilizers applied, with increased N concentrations when poultry manure or vinasse is applied, and decreased for compost. In fact, the date of application of organic fertilizer seems to be crucial, unless a cover crop can take over to retain the excess of mineral N available.

5.4.4 Gaseous N losses affected by the fertilization

Besides N leaching, the model simulated gaseous N losses by volatilisation and denitrification. The predicted values can hardly be compared to observations since measurements are difficult, scarce and made on the short term (Chirinda *et al.*, 2010; Skinner *et al.*, 2014a; Li *et al.*, 2015; Pugesgaard *et al.*, 2017). In DOK trial, STICS predicted the highest denitrification rate (N₂+N₂O) in CONFYM. The highest N₂O emissions measured in 2013 by Skinner *et al.* (2014a) were also found in this treatment. However, these authors found no differences in N₂O emissions between CONMIN and BIOORG, while simulated denitrification was higher in BIOORG. We simulated a lower denitrification in NOFERT compared to other treatments, while authors did not find differences with BIOORG and CONMIN. Volatilization ranged from 0 to 6 kg N ha⁻¹ yr⁻¹, the smallest values being simulated in NOFERT and the highest in CONFYM. No assessment of N volatilization is available in the DOK experiment.

At Foulum, we simulated small losses through denitrification, which were 33% higher in fertilized treatments than in unfertilized. Volatilization losses occurred in the conventional and O2+CC+M treatments only. Pugesgaard *et al.* (2017) estimated annual denitrification and volatilization at Foulum for the 2006-2009 period. Their estimates of denitrification were higher than our simulations: 12 vs 8 kg N ha⁻¹ yr⁻¹ in the conventional treatment, and 16 vs 8 kg N ha⁻¹ yr⁻¹ in O2+CC+M. Their estimate of volatilization in the conventional treatment was close to ours (3 vs 4 kg N ha⁻¹ yr⁻¹), but 2 to 4 times higher than ours in the organic treatments.

Soil N₂O production is a complex phenomenon, as it may be influenced by numerous factors such as the soil water content, temperature, pH, mineral N and readily available C. All these factors may be modified by agricultural practices such as the supply of N fertilizers and the incorporation of crop residues. In our study, an average higher denitrification was simulated at the DOK compared to the Foulum, that may be linked with the differences of soil pH, slightly smaller in the former than in the latter (6.25 vs 6.49, respectively). High N₂ and N₂O emissions were simulated at the Danish experiment in the conventional and the O2+CC+M treatments, while they were lower in O2+CC-M and O4+CC-M: they were mainly driven by the amount of N fertilizer applied. At DOK trial, increased denitrification was found in systems receiving external organic N inputs (manure and slurry), *i.e.* in BIOORG and CONFYM.

5.4.5 Long term evolution of soil organic N pools

Besides the N losses, variations in soil organic N pools can represent a source or a sink of N in the N surplus. Our second hypothesis was validated, since SON dynamics was equally well simulated in conventional and organic systems with the same formalism. Contrasted changes in SON content were simulated in the DOK experiment. The evolution of SON stocks was correctly modelled after decreasing the initial proportion of active soil organic matter from 35% to 55%. This change is consistent with the previous history of the experiment, since grassland is known to increase carbon storage and therefore the proportion of active fraction, compared to arable cropping. Leifeld *et al.* (2009) modelled satisfactorily the SOC stocks evolution at the DOK from 1978 to 2006 with RothC model. They justified their slight underestimation of SOC stocks in CONFYM by the N input rate, three times higher than in other treatments, which could have accelerated the decomposition of SOM, thus decreasing the SOC stock. Such an hypothesis was not necessary in our modelling which predicted satisfactorily both SON and SOC in the CONFYM treatment.

In the Foulum experiment which had a previous history of arable cropping, the proportion of active fraction was maintained to its default value, 35%. Changes in SON stocks were also well simulated, with an average decrease of -34 and -4 kg N ha⁻¹ yr⁻¹ in rotation 4 and 2, respectively over 19 years. Pugesgaard *et al.* (2017) estimated the variation of SON stocks in the same experiment by subtracting N losses to the N surplus calculated for the 2006-2009 period. Their results were very close to ours, -30 and +2 kg N ha⁻¹ yr⁻¹, in rotation 4 and 2.

Contrasted annual changes in soil humified N content were simulated for the DOK experiment. The evolution of SON stocks was correctly modelled after decreasing the share of stable soil organic matter from 65% to 45%, to account for the past grassland use of the experiment. While a high annual SON decrease was observed in NOFERT (-78 kg N ha⁻¹ yr⁻¹), lower decrease was simulated in CONMIN and BIOORG (-39 kg N ha⁻¹ yr⁻¹) and in CONFYM (-15 kg N ha⁻¹ yr⁻¹). For the Foulum experiment, changes in soil humified N stocks were correctly simulated, with an average decrease of -34 and -4 kg N ha⁻¹ yr⁻¹ in rotation 4 and 2, respectively over 19 years. In their study, Pugesgaard *et al.* (2017) estimated the variation of SON stocks by subtracting N losses to the N surplus calculated for the 2006-2009 period. Their results were very close to our findings, as they found a decrease in humified N stocks in the conventional treatment C4-CC+IF (-30 kg N ha⁻¹ yr⁻¹) and a very low increase in treatments of rotation 2 (+2 kg N ha⁻¹ yr⁻¹).

Changes in organic N pool were affected by crop management in both experiments, affecting the amount and type of organic residues (0-25 cm) and in deep root deposits (25-100 cm). Organic residues were crop residues (straw, stubble and root localized in the 0-25 cm soil layer), clover-grass cuts, catch crop residues and organic fertilizers (manure and slurry), mineralized according to specific model parameters. Particularly, the amount of dead roots inputs was increased with N fertilization for both experiments, as in STICS model the biomass portioning to roots depends on aboveground biomass production. Thus, the shoot-to-root ratio is not actually impacted by fertilization level in the model. Among all crops, clover-grass contributed to a large part of the root inputs, like at Foulum where we simulated four times more root N inputs in organic treatments of rotation 2 (including clover-grass) compared to the conventional treatment. These root inputs represented from 4 to 175 kg N ha⁻¹ according to the length of the simulation unit, with an average C/N ratio of 20. Similar root N inputs were reported previously, with up to 156 kg N ha⁻¹ in the top 20 cm of soil at the destruction of a three year old clover-grass ley whose C/N ratio as close to 20 (Eriksen & Jensen, 2001). However, studies often reported the root C and N inputs recovered at harvest, not reflecting the previous turnover

simulated by the model. Conversely, a low amount of aboveground crop residues was returned to soil in rotation 4 at Foulum, with straw and stubbles whose C/N ratio averaged 33 and 60 respectively. Therefore, these elevated ratios would explain the lower N mineralization from residue simulated in O4+CC-M and immobilization of N in microbial biomass in C4-CC+IF, leading to the mining of the SON stocks.

Lastly, the evolution of the deep root residues pool was linked with root deposition in the depth, below the mineralization layer (25-150 cm). This pool could only be increased in the STICS model as no decomposition is considered in this soil layer, which is a common simplification in agro-environmental models. This assumption is linked with the chemical recalcitrance of root tissues (Lorenz & Lal, 2005), the decomposition of which may occur over decades (Rasse *et al.*, 2005; Rumpel & Kögel-Knabner, 2011), particularly because of the poor soil aeration, low temperature and reduced microbial activity. Root deposition is linked with the definition of the plant rooting, whose corresponding parameters are hardly accessible and poorly studied (Sanaullah *et al.*, 2011). However, the simulation tended to show important root biomass inputs below the soil mineralization layer, especially at the DOK where it increased from 8 to 14 kg N ha⁻¹ yr⁻¹. The mineralization of these deep root residues would lead to higher mineral N availability, either absorbed by crops, leached or accumulated in soil organic matter. Jenkinson & Coleman (2008) intended the modelling of subsoils root residues with the dynamical model RothC-26.3, by adding parameters to consider downward C flows and the slower decomposition of deep root residues. However, the simplified pedo-climatic conditions with homogeneous soil temperature and humidity on 1 m of soil may lead to discrepancy between simulations and reality. Hence, further data would be required in order to parametrize the mineralization of deep root residues in the long term (Rumpel & Kögel-Knabner, 2011), with accurate measurements of deep soil characteristics, deep C and N inputs and C and N losses.

5.5 Conclusion

We simulated the crop production and the related C and N fluxes modified in conventional vs organic cropping systems in the long term experiments of DOK (Switzerland) and Foulum (Denmark) with the agro-environmental model STICS. The STICS version used was found to reproduce well the aboveground crop biomass in conventional and organic treatments, while the crop N uptake simulation was simulated with less precision. This gap can be related to the general underestimation of SMN content. Therefore, the quality of simulations of the different

N impacts such as soil N storage, nitrate leaching, NH_3 volatilization and N_2 and N_2O denitrification may be flawed. We simulated the N surplus, as an indicator of the potential losses from cropping systems. We found that contrasted N surplus can reflect similar type of N losses at the DOK, whereas close N surplus can reveal contrasted N fate and losses at Foulum. The model gave an insight of potential C and N processes modified in conventional crop management *vs* organic, *i.e.* crop rotation, fertilization and soil tillage. Simulations revealed that the N related environmental impacts depended on different managements considered by the model: thus, leaching was more affected by crop rotation, gaseous losses mostly by fertilization while soil N pools were sensitives to both crop rotation and fertilization. These cross effects between cropping management and N impacts revealed that not one cropping system in its entirety can be a solution to decrease all types of N losses.

This result sustains the idea that agro-environmental models can strongly participate to the understanding of the long term behaviour of cropping systems, and to the quantification of agricultural impacts in contrasted pedo-climatic situations and crop management. However, while most impacts can be simulated by STICS model, the latter does not consider the evolution of root accumulation in soil depth, below the mineralization layer. The root biomass is impacted by cropping management though, and further studies are required to properly parametrize roots biomass production, C and N inputs and their fate in deeper soil layer, especially under and after clover-grass ley destruction.

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CHAPTER

6.

General conclusions and perspectives

6.1 Main results and achievements

The aim of this thesis project was to evaluate, to better understand and to predict the long term impact of alternative cropping systems on soil C and N fates. These alternative cropping systems refer to several agricultural systems, from conservation agriculture to organic farming. The latter were developed to resolve pollution issues related to the intensive and unbalance managements of nutrients and pesticides in conventional systems. In order to achieve this work, we studied three long term experiments comparing different alternative and conventional agricultural systems and located in Northern Europe: “La Cage” experiment started in 1998 (Versailles, France), the “DOK” experiment started in 1978 (Therwil, Switzerland) and the “Foulum organic” started in 1997 (Foulum, Denmark). In addition of the measurements carried out between 2014 and 2017, the long term monitoring of the three experiments allowed the acquisition of data regarding C and N dynamics and needed for a long term assessment of the alternative practices. . Here, we present the synthesis of our main results, answering the research questions formulated in the first chapter. The soil C and N fluxes and compartment affected by the different alternative cropping systems of La Cage experiment are summarized in Figure 6.1.

6.1.1 Storing carbon and nitrogen under alternative cropping systems

The quantification and modelling of the temporal evolution of soil organic C and N stocks allowed us to disentangle the cross-effects of alternatives cropping practices.

After 16 years of differentiations, we found that **soil C and N storage** was mainly relying on organic matter input, rather than no-tillage at La Cage experiment (Chapter 2 and 4). The rates of change in SOC stocks in the old ploughed layer (ca. 0-30 cm) during the 16 years were 0.63 and 0.28 t ha⁻¹ yr⁻¹, while the rates of change of SON stocks were 54 and 31 kg N ha⁻¹ yr⁻¹ in the CA and ORG systems, respectively. Particularly, this increase was localized on the first 10 cm of soil in CA, whereas it was it was evenly distributed on the ploughed layer in ORG. The CA and ORG systems were less productive than the CON and LI systems but the smaller C inputs derived from cash crop residues were compensated by the extra inputs from additional crops (fescue and alfalfa) specifically grown in CA and ORG, resulting in a positive carbon storage in soil. Our hypothesis concerning the contrasted evolution of C and N related to variations in organic matter inputs between cropping systems was thus validated.

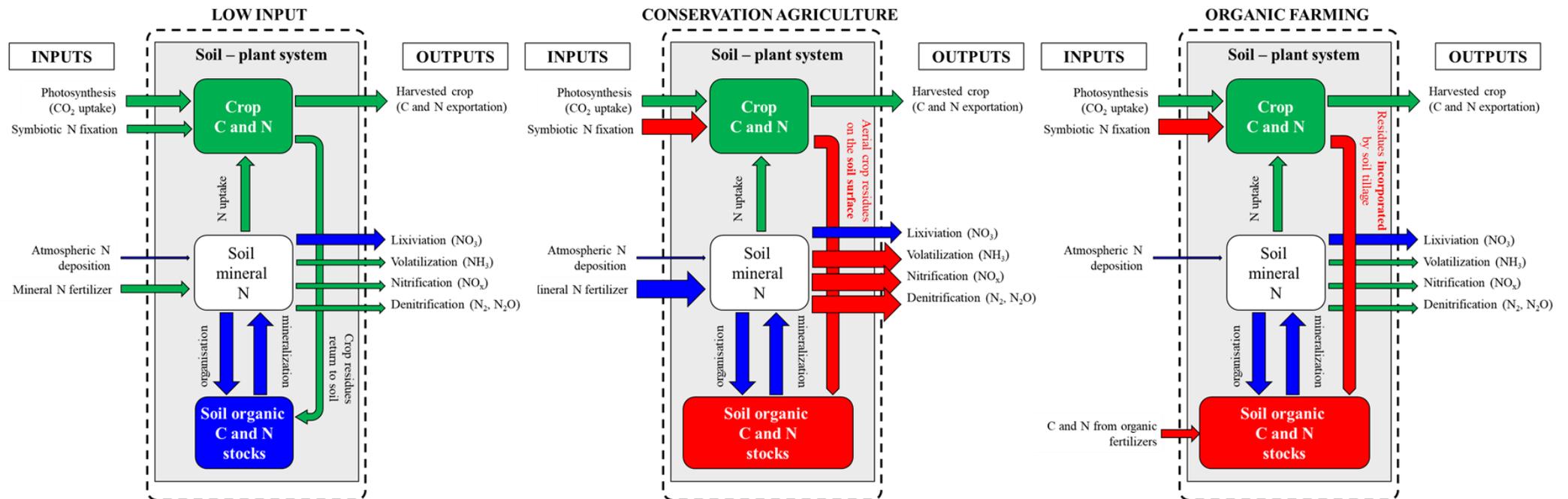


Figure 6.1. Modified C and N fluxes and compartments based on observations and calculations made at La Cage experiment (Versailles, France). Red, blue and green filling stand for increased, unchanged or decreased flow/compartments compared to the conventional system, respectively.

In particular, the increased SOC storage in CA system was explained by higher carbon inputs compared to the other cropping systems ($+1.72 \text{ t C ha}^{-1} \text{ yr}^{-1}$ on average).

In addition, the four months' soil incubations showed no differences of soil C and N specific mineralization rate between treatments of La Cage experiment, including under no tillage and for both disrupted and undisrupted soil samples (Chapter 3). This result confirmed the observation made beforehand with the AMG model, satisfactorily predicting SOC stocks evolution from 1998 to 2014 with no difference in SOC mineralization rates between all cropping systems (Chapter 2). Hence, our results together with the increased SOC stocks observed in CA and ORG treatments suggest that increased biomass returns to soil or changes in microbial physiology may be the main drivers of SOC storage, rather than by a reduced soil tillage or a lower C and N mineralization rate.

Simulating the long term evolution of SON stocks at the DOK and Foulum experiments was achieved with STICS model (Chapter 5), after complementing the conceptual scheme and implementing new options of simulation. These options enable to consider successive simulations including associated crops, to sow a crop under the established main crop while making possible its development in a successive simulation, to simulate clover-grass with transmission of root system over successive simulations and to return clover-grass cuts to soil. Thus, after the destruction of the previous grassland at the DOK, the model simulated a lower decrease of SON stocks in fertilized treatments receiving manure and/or mineral N fertilizer and important crop residues, rather than in the unfertilized treatment leading to lower biomass residues. Likewise, the SON stocks were more maintained at Foulum in treatments including a clover-grass ley in the crop rotation, leading to important belowground inputs.

Finally, our work shows the interest of alternative cropping systems including higher organic matter inputs in order to increase the SOC and SON stocks. This result confirms findings of recent meta-analysis made on the topic, concluding on the fact that increased C inputs to soil is a more efficient solution to store soil C (Pellerin *et al.*, 2013; Poeplau & Don, 2015) rather than limiting the output by no-tillage to reduce organic matter mineralization (Luo *et al.*, 2010b; Virto *et al.*, 2012). In addition, it showed the need to better estimate the amount of biomass input related to root system, appearing as one of the major source of soil C and N, especially in deep soil layers.

6.1.2 Reducing nitrogen losses in alternative cropping systems

The annual **N surplus**, resulting from the difference between total N inputs and N exportations, was calculated as a proxy to estimate the N losses from the different cropping systems studies for La Cage experiment (Chapter 4) and for the DOK and Foulum experiments (Chapter 5). When negative, the N surplus may be associated with soil N pool mining, whereas a positive N surplus signify a N surplus, whether stored in soil, lost by leaching or emitted through N gas. We found contrasted impacts of the alternative cropping systems on the annual N surplus, depending on fertilization management and on the amount of N exported (Chapter 4). This may be linked with the N mineralization of supplementary alfalfa cuts left on the soil in the ORG system. In addition, we found that for an equivalent N surplus, the fate of the N surplus could be contrasted according to agricultural practices, whether it was stored in soil organic matter, lost by leaching or by gaseous emissions (Chapter 5). Similarly, no correlation was found between the mean N surplus and N leaching calculated over eleven organic cropping systems in Northern France (Rakotovololona *et al.*, submitted). Hence, we conclude on the insufficiency of this indicator to give a complete picture of the fate of N in these systems, whether stored in soil organic matter, lost by leaching or by gaseous emission (Chapter 4 and 5). Theoretically, the relationship between the N surplus and N losses should occur when the SON has reached an equilibrium; however, this horizon is virtual since the systems are always evolving.

The soil N losses were quantified directly by measurements of soil water content, mineral N stocks and soil N₂O emissions for La Cage experiment (Chapter 4) or indirectly by modelling of N leaching, volatilization and denitrification for the DOK and Foulum experiments (Chapter 5). **Nitrate leaching** calculated with LIXIM model did not significantly differ between treatments of La Cage experiment, neither for treatments of the DOK experiments when simulated with STICS. However, leaching was reduced by 60% at Foulum experiment in one organic treatments including catch crop and excluding manure application compared to the conventional treatment. Hence, soil N mineralization rate and crop rotation appeared to be the main driver of N leaching, regardless of the type and amount of N fertilizer applied. In addition, the increased mineralization following the clover-grass destruction needed to be accounted for by an artificial input of easily decomposable organic matter, releasing mineral N to the subsequent crop.

At La Cage experiment, the **gaseous N emissions** by volatilization and denitrification estimations varied from $15 \pm 9 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ in the organic treatment up to $106 \pm 17 \text{ kg N ha}^{-1}$

yr⁻¹ in conservation agriculture (Chapter 4). These losses were highly correlated with the measured N₂O emissions monitored continuously during more than three years and reaching up to 4.15 kg N₂O-N ha⁻¹ yr⁻¹ under conservation agriculture, while they were the lowest in low input and organic farming. The focus on dynamical emissions showed that the application of mineral N fertilizers and the restitution of fresh biomass (from alfalfa cuts or chemical destruction of the cover crop) were mainly responsible for the peak of N₂O emissions. A reduction in N₂O emissions was also observed when total N inputs were reduced. Simulation of cumulative gaseous N losses with STICS (Chapter 5) were in accordance with these observations; however, further research and improvements are needed for the model to be able to account for the high temporal variations of N₂O emissions.

6.1.3 The greenhouse gas balance: an absolute indicator

We calculated the global **GHG balance** for each cropping system of La Cage experiment, accounting for equivalent CO₂ emissions, related to fertilizer synthesis, fuel consumption and soil direct and indirect N₂O emissions and CO₂ storage, related to the soil C accumulation (Chapter 4). The annual GHG balance varied widely between systems with +2.1 t CO₂eq ha⁻¹ yr⁻¹ in the conventional treatment, +1.7 in low input, +0.3 in conservation agriculture and -0.1 in organic farming. This ranking was similar when the GHG balance was expressed per unit of N input or exported N. Hence, the soil carbon storage calculated in conservation agriculture over the 0-30 soil layer was counterbalanced by important N₂O losses as well as important N losses whereas N losses were lowest in the ORG system. Hence, we showed that SOC storage or N losses alone are incomplete indicators of the environmental impact of agriculture, and that using the GHG balance was a more precise indicator to give a complete estimation of the C and N environmental impact of a cropping system.

Finally, this thesis confirmed the interest of long term study to accurately evaluate the impact of agricultural systems on C and N fates. In fact, we considered sufficiently long periods to account for slow processes affecting SOC and SON dynamic in relation with agricultural practices, along with more rapid and punctual processes such as N leaching and N₂O emissions.

6.2 Interest and drawbacks of the methodological approach

Our methodological approach consisted in coupling experimentation and modelling. The experimental part of the work was based on three fields experiment, comparing conventional and alternative cropping systems for more than 15 years in different pedo-climatic situations. Using both experimentation and modelling allowed to i) improve the understanding of short term and long term phenomenon affecting carbon and nitrogen cycling and ii) investigate in the future the impacts of spatial variability of each system.

The measurements related to soil organic C and N stock were made with a relatively precise methodology. In fact, soil depth, soil bulk densities and soil C and N contents were all measured at the start and thereafter allowing an estimation of soil C and N stocks at equivalent soil mass. However, the experiments were not designed at their start for the purpose of our study, explaining why some types or frequency of measurements were missing here. In addition, long term experiments are the place of numerous simultaneous studies, meaning that not all types of measurements can be done, especially for trials like the DOK with small field plots, while larger field plots of La Cage experiment enable the achievement of frequent measurements.

One of the major drawback of the calculation of the N surplus is the need for component of its calculation that are not always measured, especially the biological nitrogen fixation and the atmospheric deposition. Estimations of these components can lead to uncertainties in the N surplus calculation, and thereafter in the total gaseous N losses estimated by subtracting N storage and leaching to the N surplus. However, the continuous monitoring of soil N₂O emissions at La Cage experiment from April 2014 to May 2017 enable to have a very precise image of the gas. Contrastingly, most studies reporting soil N₂O emissions are based on punctual measurements that may lead to an incomplete estimation of total losses, due to the temporal variability of N₂O emissions. The major weakness of the experimental part of work is the lack of more frequent measurements of soil water and mineral N, that would allow a better estimation of N leaching. Finally, the simultaneous evaluation of both C and N fluxes allowed us to consider several environmental impacts and thus to give a global evaluation of the cropping systems, including the GHG balance. Very few studies considered all these impacts together.

Another limit of the experimental design can be the type of crop rotations compared in the long term experiment of La Cage. There, crop rotation differed in conservation agriculture and

organic farming compared to the conventional treatment, with the introduction of alfalfa in a short rotation composed of wheat and pea. In fact, the cultivation of alfalfa was conditioned by the lack of availability of harvesting equipment, leading to the restitution of alfalfa cuts to the soil in the conservation agriculture and organic farming treatments. Although this management may not reflect the actual cuts management in other French arable cropping systems, this restitution can also be taken as an organic N fertilizer in the organic cropping system. Furthermore, crop rotations and managements compared at the DOK and Foulum experiments were more realistic, as the experiments were designed in order to compare systems close to local farmers' issues in their respective countries.

The modelling was used to simulate the long term impact of these systems, in order to access not measured variables. Applying STICS in organic farming systems was a scientific challenge as regard to the SOM turn-over and the pests pressure. Our modelling approach assumed that N was the main limiting factor of production, but other limiting factors could actually determine crop production, such as the soil potassium and phosphorus availability, along with the biotic stresses caused by pest damages and the competition for nutrients with weeds. This item was insured thanks to the postulate that the studied experiments were scarcely impacted by weeds and pests. In addition, we could not conclude on the relation between the reduced amount N-fertilizer and rhizodeposition in alternative cropping systems. In fact, belowground crop residues and rhizodeposition were not measured in this study, these variable representing a full time research topic. The model could simulate root deposition but the latter was proportional to the aboveground biomass leading systematically to lower root biomass in stressed systems.

6.3 Perspectives and advices

6.3.1 Supplementary data

Some measurements and analysis made in the frame of this thesis are not presented in detail here. Particularly, the continuous measurement of soil N₂O emissions made at La Cage experiment, from April 2014 to May 2017, will have to be analyse in detail. The exploration of these data may give a precise insight of the local on N₂O dynamics according to the type of crop, the soil humidity and temperature on 0-20 cm soil layer, the fertilization and soil managements. In addition, the measurement of soil water and mineral N contents made at the DOK experiment between February 2015 and February 2017, used for STICS calibration, could

also be used by experimenters in order to evaluate the relation between soil water and N dynamics and crop yields for the corresponding years.

6.3.2 Modelling environmental impacts: research perspectives

The major interest of modelling is the possibility to predict variables not measured and to generalise results of modelling of alternative cropping systems to bigger space and time scales. Here, we could estimate N leaching and gaseous losses with STICS model. However, modelling challenges still need to be tackled. Hence, further modelling research is needed to precise root biomass growth and decomposition: in the frame of the RHIZOCARB research project, Xiaogang Yin (INRA, P3F) will focus on the parameterization of STICS root module for typical cereal/grass crops and their impacts on SOC storage in long term French experiments, while in the frame of “AESN STICS Prairies” project taken on by Hughes Clivot (INRA, UR AgroImpact), the simulation of long term grassland production, water, C and N dynamics will be improved in relation with root modelling improvements. Moreover, there is still a need to evaluate the STICS model against others alternative cropping systems and to carry on virtual experiment about variability of their response to different pedo-climatic conditions, type and amount of crops residues or fertilization management. The calibration of this new version will still require gathering reliable datasets and global evaluation as currently done for any standard version (Coucheney *et al.*, 2015). Hence, modelling of La Cage experiment should be tested in the frame if the “Climate-CAFÉ” research project, which aims at evaluating adaptation strategies of agricultural systems in the context of climate change. Finally, the implementation of clover-grass plant file in will need to be done, by considering the legume and the grass separately in order to account for specific crop growth, nutrients need and root turn over.

6.3.3 Which alternative cropping systems for the least environmental impacts?

Contrasted types of alternative cropping systems were compared in our work, all coming with their benefits and drawbacks regarding impacts on C and N fluxes. Therefore, not one cropping systems can be seen as a complete solution to reduce agricultural pollution, and one should rather consider practices that can together be combined in the best way to decrease pollutions. For instance, we showed that when efforts were focussed on soil C storage by increased organic matter inputs in no tilled system, the side effect was the emissions of N₂O, finally leading to a positive GHG balance (Chapter 2 and 4).

However, regarding the main results of our work, we propose the inclusion of the following levers when designing alternative and sustainable cropping systems:

- The generalization of catch crops in order to reduce N leaching in autumn and winter should be generalized, as shown before in several studies. Besides, they can participate to the increase of soil organic matter stocks by their root residues, and thus an increase of soil fertility. When sown sufficiently early into the standing cash crops (before its harvest), they can also shade out weeds and reduce competition for nutrient with catch crop.
- The inclusion of perennials in crop rotation can help increasing soil organic matter stocks, particularly from the root deposition in different soil layers. However, we have seen that its returned on the top soil layer may favour denitrification and thus N₂O losses. To avoid these losses, the exportation of perennials cuts for livestock feeding or for biogas production can be envisaged.
- The use of associated crop (*e.g.* cereal and grain legume) in order to increase the N use efficiency thanks to different plant nutrients need in time (according to specific crop growth) and space (according to specific rooting depth).
- Limiting the use of synthetic fertilizer, in order to reduced CO₂ emissions due to the fertilizer synthesis, and NH₃ volatilization and N₂O emissions happening at application at the field scale. Instead, the use of organic fertilizer can be promoted, provided that the crop need and the organic fertilizer mineralization are synchronized, that can be solved by the practices listed above, *i.e.* the maximizing of soil coverage and the association of crops.
- Reducing N losses from may be accomplished with additional policies aiming at reducing or eliminating late season ploughing of catch crops (Finney *et al.*, 2015), but should not be promoted as an efficient practice for increasing soil carbons stocks. Conversely, soil tillage should be preserved as an interesting tool to limit the use of herbicide when weed pressure threshold is exceeded.

All these levers need to be coherently integrated to the current cropping systems which are expected to meet the specifications of the global change attenuation and adaptation.

6.3.4 Which advices for policies maker?

The use and the diffusion of environmental indicators should be carefully considered. As shown, the GHG balance was the most complete indicator when evaluation both C and N fluxes affected by cropping systems. Agricultural policies targeting a single environmental objective, such as the “4 per 1000” initiative, must be considered with great vigilance since they may

potentially lead to an adverse effect such as N₂O emissions from decomposed mulch in conservation agriculture, while the mulch can simultaneously be a tool to limit water evaporation and to increase the water table recharge. Hence, an appropriate assessment of the environmental impact of a cropping system should be based on a global evaluation, considering both C and N fluxes modified by the farming practices and not be limited to one indicator. Such assessment will require further monitoring of long term studies with contrasted crops successions and pedo-climatic situations. To a larger extent, the N footprint indicator has been developed to show the potential loss of N to the environment from the production and consumption of food and fossil fuels (Shibata *et al.*, 2016). Their approach attempt to involve not only production sectors such as agriculture but also consumers' sector, that creates the demand for the food produced.

In addition to the implementation of alternative cropping systems, research is needed to other develop levers participating in N losses reduction, particularly with increased knowledge of crop traits and genetic improvement. In fact, closing the gap between expected yields and actual ones by increased N use efficiency by crops should help minimizing losses of nutrients to the environment (Foley *et al.*, 2011), the yield gaps being nowadays up to 20–30% for organic production (Seufert *et al.*, 2012; Ponisio *et al.*, 2015). In addition, reducing N losses from agricultural activity is strongly linked with a reduction of food waste, which account for 30 to 40% of the total production (Gustavsson *et al.*, 2011). Some authors proposed a shift in the human diet to reduce these losses, the transition toward more plant-based diets potentially reducing food-related greenhouse gas emissions by 29 to 70% (Springmann *et al.*, 2016).

Finally, this study was focussed on environmental impact related to C and N fates in agro-ecosystems. Yet, attention should be paid to the fact that pesticides used in agriculture may also generate persistent pollution of soil, air and water resources, leading to health hazardous effects for living creatures (Gilden *et al.*, 2010), that we did not consider in our evaluation.

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Appendices

Appendix 1.A. Formulae used to calculate aboveground biomass production of cover crops and catch crops.

	Crop and management	Formula	Definition and Parameters	Reference
Eq.1	Catch crop and cover crop (apart from alfalfa)	Weibull $DM = a \cdot (1 - e^{-b \cdot \Sigma(T-Tb)^c})$	DM = crop biomass (t ha ⁻¹) for oat and fescue: $a = 5.84, b = 0.71, c = 2.67$ for clover: $a = 4.37, b = 0.38, c = 4.42$ for fodder radish: $a = 6.09, b = 0.59, c = 1.76$	(Laurent <i>et al.</i> , 1995)
Eq.2	Alfalfa cover crop establishment in early september, and spring and summer regrowth.	$LAI = e^{(a \cdot \Sigma(T-Tb)) - b}$	LAI = leaf area index in autumn $a = 0.0025$ and $b = 5$ in spring $a = 0.0024$ and $b = 0$ in summer $a = 0.0035$ and $b = 0$	(Beaudoin & Thiébeau, unpublished) (Justes <i>et al.</i> , 2002)
Eq.3		$PAR = Rg \cdot f$	PAR = photosynthetically active radiation (MJ m ⁻²) Rg = global radiation (MJ m ⁻²) f = photosynthetically active fraction $f = 0.48$	(Varlet-Grancher <i>et al.</i> , 1982)
Eq.4		$PARi = PAR \cdot g \cdot (1 - e^{-k \cdot LAI})$	$PARi$ = intercepted PAR (MJ m ⁻²) $g = 0.97$ and $k = 0.88$	(Gosse <i>et al.</i> , 1982, 1984)
Eq.5		$DM = PARi \cdot RUE$	RUE = radiation use efficiency	
Eq.6	Alfalfa cover crop autumn growth	$LAI_{pot} = c \cdot \Sigma(T - Tb)$	LAI_{pot} = potential LAI $c = 0.0092$	(Coulmier, 1990)
Eq.7		$\epsilon_{ipot} = g \cdot (1 - e^{-k \cdot LAI_{pot}})$	ϵ_{ipot} = potential interception efficiency	id
Eq.8		$PAR_{pot} = \epsilon_{ipot} \cdot PAR$	PAR_{pot} = potentially intercepted PAR (MJ m ⁻²)	
Eq.9		$LAI = -\frac{1}{k} \cdot \log(1 - \frac{PAR_{pot} - PAR_{sen}}{g \cdot PAR})$	PAR_{sen} = PAR not intercepted due to senescent leaves $PAR_{sen} = 0.02$	id

Eq.10	$\varepsilon i = g. (1 - e^{-k.LAI})$	$\varepsilon i =$ interception efficiency	id
Eq.11	$PAR_i = \varepsilon i. PAR$		
Eq.12	$SEN = j. (LAI_{pot} - LAI)$	$SEN = LAI$ of senescent leaves $j = 0.25$	id
Eq.13	$DM = i. PAR_i - SEN$	$i = 0.015$	id
Eq.14	$DM_m = DM. LER_m$	$DM_m =$ biomass of the cover crop associated with a cash crop (t ha ⁻¹) $LER_m =$ land equivalent ratio of the cover crop for alfalfa, fescue and fodder radish: $LER_m = 0.5$ for clover: $LER_m = 0.65$	(Szumigalski & Van Acker, 2008; Shili-Touzi, 2009)
Eq.15	Alfalfa as main crop $DM = DM_{ref}(i)$	$i =$ cutting number $DM_{ref}(1) = 5.0$ $DM_{ref}(2) = 9.0$ $DM_{ref}(3) = 10.5$ $DM_{ref}(4) = 12.5$ $DM_{ref}(5) = 13.0$	

Appendix 2.A. Physical and chemical properties of La Cage experiment's soil (layer 0-25 cm) measured in 1998.

Soil texture	(g kg ⁻¹)	Clay	(<2 µm)	167
		Fine silt	(2-20)	182
		Coarse silt	(20-50)	380
		Fine sand	(50-200)	236
		Coarse sand	(200-2000)	35
		Organic mater	(g kg ⁻¹)	Org. C
		Total N		0.99
CaCO ₃	(g kg ⁻¹)			0.9
pH _{H₂O}				7.4
Total CEC	(cmol+ kg ⁻¹)			11.5

Appendix 2.B.1. Parameters of the regression model and belowground nitrogen factor for the estimation of biological nitrogen fixation (Anglade *et al.*, 2015a).

Crop species	α	β	BGN
Alfalfa	0.81	-13.9	1.70
Clover	0.78	3.06	1.60
Faba bean	0.73	5.45	1.53
Lentil	0.64	3.32	1.40
Pea	0.66	4.32	1.33
Soybean	0.66	4.32	1.50
Lupin	0.64	5.45	1.50

Appendix 2.B.2. Standard values of nitrogen content of crops harvested organ (CORPEN, 1988).

Crop species	N content
	kg N t ⁻¹ DM
Barley	17.1
Lupin	51.6
Maize	16.4
Oat	19.6
Pea	37.0
Rapeseed	30.5
Soybean	68.0
Alfalfa	26.0

Appendix 2.C. Standard values of the depth of roots at the start of the experiment, root growth rate and base temperature used in LIXIM when a crop cover existed in winter time (Brisson *et al.*, 2008).

Crop species	depth of roots	root growth rate	base temperature
	cm	GDD	°C
Wheat	-2	0.12	0
Rapeseed	-2	0.12	0
Pea	-2	0.12	0
Alfalfa 1 st year	-2	0.06	3
Alfalfa 2 nd year	-90	0.06	3
Mustard	-2	0.09	3.3

Appendix 2.D. Fuel consumption per agricultural device (Lorin, 2010) (references from the "Bureau de Coordination du Machinisme Agricole").

Agricultural machinery	Fuel consumption
	L ha ⁻¹
Sprayer	1.3
Centrifugal spreader	0.9
Plow	27.6
Rotary harrow	14.7
Seeder	4.4
Direct seeder machine	8.5
Harvest combine	20.5
Crusher	11.5
Weeder harrow	5.6
Rotary mower	5.2
Hay and forage harvester	4.1
Round baler press	6.2
Flat roller	2.9
Hoe	2.9
Stubble cultivator	13.2

Appendix 3. Résumé substantiel en français.

L'activité agricole est à l'origine de déséquilibres dans les cycles du carbone (C) et de l'azote (N) au sein des écosystèmes terrestres naturels, entraînant une diminution des stocks de C et N dans le sol, une augmentation de la lixiviation du nitrate et des pertes d'azote par voie gazeuse. Pour réduire ces impacts environnementaux, la recherche de systèmes de cultures plus autonomes en intrants est une nécessité impérieuse pour améliorer le bilan social, énergétique et environnemental de l'agriculture. Ainsi, une solution est de développer des systèmes « agro-écologiques », économes en intrants et valorisant les services éco-systémiques, tels que les systèmes à bas niveau intrants, l'agriculture de conservation ou l'agriculture biologique. Ces systèmes ont un bilan environnemental *a priori* positif en termes de pression de pesticides et de maintien de la biodiversité mais incertain quant aux impacts liés aux composés azotés (NO₃, N₂O) ou carbonés (C organique du sol, CO₂), lorsqu'ils sont exprimés par unité de production. Les systèmes alternatifs incluant des apports de fumier ou de compost favorisent le stockage de carbone et d'azote organique. En leur absence, un déstockage est possible sachant que le stockage du carbone dans le sol dépend d'abord des quantités de résidus restitués au sol. Toutefois, la séquestration de carbone pourrait être plus forte en système alternatif par la fréquence élevée des cultures auxiliaires (couverts permanents, culture intermédiaire, cultures pluriannuelles). Ce facteur pourrait s'ajouter à ceux de la quantité de résidus enfouis et de leur ratio C/N, déjà identifiés comme déterminants de l'évolution des stocks de carbone et d'azote organiques en systèmes conventionnels. De plus, la modélisation du stock d'humus à long terme est sensible à la prédiction des restitutions, à la caractérisation des effluents apportés et à la prise en compte de la biomasse racinaire. De nombreuses études ont évalué l'impact à court terme de C et N des pratiques culturales alternatives. Cependant, peu d'entre elles ont comparé l'effet de ces pratiques dans des systèmes plus complexes et sur le long terme (Arrouays, 2002, Leifeld et al., 2009). Ces études sont essentielles pour renforcer la compréhension de l'impact des agroécosystèmes alternatifs sur les processus lents affectant les dynamiques C et N à long terme (West & Post, 2002, Möller, 2009), eux-mêmes influencés par les variations climatiques au cours des années. Dans ce cadre, le projet de recherche ENBIO (évaluation agro-ENVironnementale expérimentale des systèmes de culture agro-BIOlogiques) a été mis en place en 2013, dirigé par Nicolas Beaudoin (INRA, AgroImpact) et coordonné par «Agro Transfert Ressources et Territoires». Ce projet s'est divisé entre i) l'étude de la variabilité spatiale de la production, du drainage de l'eau et de la lixiviation dans des systèmes alternatifs ; ii) l'étude de la variabilité temporelle des bilans d'eau, de carbone et d'azote dans des systèmes alternatifs et

iii) l'étalonnage du modèle STICS dans les systèmes de culture biologique. Ces deux derniers thèmes sont traités dans cette thèse, avec l'objectif de répondre aux questions suivantes : les systèmes de culture alternatifs modifient-ils le surplus d'azote et le devenir de cet excédent dans le sol, l'eau et l'atmosphère ? Qu'est-ce qui peut expliquer le stockage supplémentaire de carbone dans les systèmes de culture alternatifs : les changements dans le travail du sol ou dans les cultures auxiliaires ? Les taux de minéralisation de C et N du sol sont-ils modifiés par des pratiques culturales alternatives ? Les systèmes alternatifs de culture ont-ils un meilleur équilibre des GES que les systèmes conventionnels, lorsqu'ils sont exprimés par unité de surface, production ou azote exporté ? Le modèle STICS permet-il de simuler la dynamique C et N dans les systèmes de culture alternatifs ? Les hypothèses de recherche sont spécifiques à chaque type de système alternatif étudié et ont donc été formulées dans chaque chapitre. Les deux principaux objectifs de ce travail étaient les suivants i) évaluer expérimentalement les principaux flux de C et N des systèmes de culture alternatifs, en comparaison avec les systèmes conventionnels et ii) modéliser ces systèmes de culture avec le modèle agro-environnemental STICS, afin de prévoir leur effet à long terme sur les flux C et N.

Cette thèse avait pour objectif de quantifier l'impact à long terme des différents systèmes de culture sur le devenir du carbone et de l'azote dans le système sol-plante-atmosphère et de simuler la dynamique de ces éléments avec le modèle agro-environnemental STICS. À cette fin, nous avons étudié trois essais de longue durée :

- à Therwil (Suisse) pour l'essai "DOK", initié en 1978 et géré par le FiBL et Agroscope (Zürich);
- à Foulum (Danemark) pour l'essai "Foulum organic", démarré en 1997 et géré par l'Université Foulum Aarhus;
- à Versailles (France) pour l'essai "La Cage", initié en 1998 et géré par l'UMR Agronomie de l'INRA-AgroParisTech (Thiverval-Grignon).

Quatre critères ont été retenus pour sélectionner les dispositifs expérimentaux : il devait s'agir de dispositifs présentant i) des systèmes de culture arable, ii) des mesures fréquentes des flux liés au C et N (stocks de SOC et SON, teneur en eau du sol, ...), iii) des traitements incluant des systèmes de culture alternatifs et iv) des essais de long terme permettant de tenir compte de la variabilité du climat, des effets cumulatifs et des processus lents affectants les cycles de C et N.

La stratégie de travail a consisté en un couplage des expériences *in situ*, *in vitro* et *in silico*. Pour l'expérience *in situ*, nous avons réalisé un suivi spécifique de l'essai de long terme de La

Cage de 2014 à 2017, afin de mesurer les changements de SOC et de SON, les pertes de NO_3 et de N_2O . Nous avons mesuré les teneurs en SOC et SON en 2014, teneur en azote et en eau du sol trois fois par an de 2013 à 2017 et suivi en continu les émissions de gaz à effet de serre (N_2O) d'avril 2014 à juillet 2017. Nous avons sélectionné des indicateurs agroenvironnementaux pour caractériser les performances des pratiques alternatives par rapport à l'agriculture conventionnelle. Nous avons utilisé une approche diachronique pour comparer l'évolution des stocks de SOC dans le temps et une approche synchronique pour comparer les systèmes de culture à une date donnée. Nous avons calculé le surplus azoté, les pertes par lixiviation et les pertes gazeuses et le bilan total de GES dans chacun des quatre systèmes. L'expérience *in vitro* a consisté en une incubation du sol réalisée sur des sols prélevés à La Cage, afin de tester si les vitesses spécifiques de minéralisation de C et N différaient ou non entre les systèmes. Les résultats des incubations ont été comparés au taux de minéralisation estimé par modélisation avec le modèle AMG (Saffih-Hdadi & Mary, 2008). L'expérience *in silico* a été réalisée avec un modèle dynamique, un outil utile pour évaluer les effets à long terme d'autres systèmes de culture sur les bilans hydriques, C et N. Divers modèles ont déjà été évalués et validés pour prédire le renouvellement à long terme de la matière organique du sol dans diverses conditions pédoclimatiques, principalement pour les systèmes de culture conventionnels. La modélisation des dynamiques C et N dans les systèmes alternatifs de culture à long terme a été quant à elle beaucoup moins étudiée. Nous avons utilisé le modèle STICS (Brisson et al., 1998, 2002) pour simuler la production végétale, le stockage de C et N et le surplus de N dans les essais DOK et Foulum.

Dans un premier temps, nous avons étudié la dynamique du carbone organique du sol (COS) dans l'expérience de terrain de "La Cage" qui compare quatre systèmes de culture arable, sans application de fumier, en agriculture conventionnel (CON), bas intrant (LI), en agriculture de conservation (CA) et en agriculture biologique (ORG). Les densités apparentes et les concentrations de COS ont été mesurées à différentes dates entre 1998 et 2014. Les stocks de COS ont été calculés à masse équivalente en tenant compte des variations de densité apparente et de la redistribution du COS entre les différentes couches de sol. Nous avons analysé l'évolution des stocks de SOC et l'avons comparée aux sorties du modèle de simulation AMG. Les vitesses de variation des stocks de COS dans l'ancienne couche labourée (environ 0-30 cm) au cours des 16 années étaient respectivement de 0,08, 0,02, 0,63 et 0,28 $\text{t ha}^{-1} \text{an}^{-1}$ dans les systèmes CON, LI, CA et ORG. et significativement différent de 0 dans les traitements CA et ORG. Le modèle AMG a reproduit de manière satisfaisante l'évolution observée des stocks de

COS dans l'ancienne couche labourée dans tous les traitements. Une méthode d'optimisation bayésienne a été utilisée pour évaluer la moyenne et la distribution des paramètres les plus incertains : le taux de minéralisation du COS et les apports en C dérivés de la biomasse souterraine des cultures de couverture comme la fétuque (*Festuca rubra*) et la luzerne (*Medicago sativa*). Le modèle ainsi paramétré était capable de prédire l'évolution du COS dans chaque bloc et couche de sol (0-10, 10-20 et 20-30 cm). Il n'y avait pas de différence significative dans les taux de minéralisation du COS entre tous les systèmes de culture, y compris le système CA en semis direct. En particulier, l'augmentation du stockage de COS dans CA a été expliquée par des apports de carbone plus élevés par rapport aux autres systèmes de culture (+1,72 t C ha⁻¹ an⁻¹ en moyenne). Les systèmes CA et ORG étaient moins productifs que les systèmes CON et LI, mais les plus petits intrants de carbone dérivés des résidus de cultures ont été compensés par les apports supplémentaires de cultures supplémentaires (fétuque et luzerne) spécifiquement cultivées en CA et ORG, résultant en un bilan carbone positif. Nous concluons que les systèmes arables alternatifs ont le potentiel de séquestrer le carbone organique dans des conditions climatiques tempérées, à travers un apport de carbone plus élevé plutôt que par l'effet du labour réduit du sol.

Dans un second temps, nous avons comparé les taux de minéralisation du COS et de l'azote organique du sol (NOS) des quatre systèmes de l'essai de La Cage. Des échantillons de sol remaniés provenant de la couche de sol de 0-27 cm ont été incubés en laboratoire pendant quatre mois afin de suivre la minéralisation de C et N dans tous les traitements, tandis que des échantillons non perturbés de CON et CA ont été incubés. La perturbation physique des échantillons de sol a diminué les taux de minéralisation du C et du N dans le sol. L'ajustement du modèle a montré que la taille du pool de carbone labile contribuant à la minéralisation du carbone était beaucoup plus élevée dans les carottes non perturbées (par exemple 565 dans CON et 377 mg C kg⁻¹ dans CA) que dans les échantillons de sol remaniés (100 et 90 mg C kg⁻¹ respectivement). Une plus grande abondance de COS labile dans les cylindres de sol non perturbés (résidus végétaux grossiers et racines fines) par rapport aux échantillons de sols remaniés expliquerait ce résultat. Aucune différence n'a été trouvée entre les quatre systèmes de culture dans leur taux de minéralisation spécifique, exprimé par rapport au COS, NOS ou au C de la biomasse microbienne. Pour les carottes non perturbées, une minéralisation similaire en CA et CON peut résulter de l'équilibre entre une plus grande quantité de matière organique labile et une structure du sol moins favorable à la décomposition en CA. Pour les échantillons de sol perturbés, des taux de minéralisation similaires ont suggéré que la capacité de

décomposition de la matière organique et les conditions environnementales pour les décomposeurs étaient similaires entre les systèmes de culture. Dans l'ensemble, ces résultats ont confirmé des vitesses spécifiques de minéralisation du carbone équivalents, indépendamment des traitements cultureux, validant l'hypothèse de vitesses spécifiques de minéralisation non modifiés par les pratiques culturales, y compris le non-labour. Par conséquent, nos résultats, combinés aux stocks accrus de carbone organique du sol observés dans les traitements CA et ORG suggèrent que des rendements accrus de la biomasse dans le sol ou des changements dans la physiologie microbienne pourraient être les principaux facteurs de stockage du carbone organique du sol.

Nous avons ensuite étudié la balance azotée, le stockage organique du N, la lixiviation de N, les émissions de gazeuses de N et l'équilibre des GES dans le dispositif de La Cage. Le surplus d'azote, soit la différence entre les intrants N totaux et les exportations, était le plus bas en LI avec $47 \text{ kg ha}^{-1} \text{ an}^{-1}$, intermédiaire pour CON et ORG avec 67 et $68 \text{ kg ha}^{-1} \text{ an}^{-1}$ et le plus élevé en CA avec $181 \text{ kg ha}^{-1} \text{ an}^{-1}$. CA et ORG ont reçu de grandes quantités de N provenant de la fixation biologique de la luzerne. Les taux annuels de stockage du SON différaient nettement entre les systèmes de culture : $\text{CA} (54 \text{ kg ha}^{-1} \text{ an}^{-1}) > \text{ORG} (31) > \text{CON} (13) = \text{LI} (3)$. La lixiviation de l'azote, calculée en utilisant des mesures de N minéral du sol, a atteint une moyenne de $20 \text{ kg ha}^{-1} \text{ an}^{-1}$ et ne différait pas significativement entre les traitements. Les émissions gazeuses d'azote (volatilisation + dénitrification), calculées comme la différence entre l'excédent de N, le stockage de NOS et la lixiviation de N, allaient de $19 \text{ kg ha}^{-1} \text{ an}^{-1}$ dans l'ORG à $106 \text{ kg ha}^{-1} \text{ an}^{-1}$ dans l'AC. Les émissions de N_2O ont été surveillées en continu avec des chambres automatiques pendant 3,3 ans. Elles variaient de $1,33 \text{ kg ha}^{-1} \text{ an}^{-1}$ dans l'ORG à $4,24 \text{ kg ha}^{-1} \text{ an}^{-1}$ dans le système CA et étaient fortement corrélées aux émissions d'azote gazeux. Le bilan des émissions de GES varie considérablement d'un système à l'autre : il est le plus élevé dans CON et LI, avec 1989 et $1443 \text{ kg éq CO}_2 \text{ ha}^{-1} \text{ an}^{-1}$ respectivement. En CA, le bilan des GES était beaucoup plus favorable ($124 \text{ kg éq CO}_2 \text{ ha}^{-1} \text{ an}^{-1}$), malgré d'importantes pertes en N_2O qui ont partiellement compensé les avantages du stockage du COS. L'ORG était le système ayant le plus petit bilan de GES ($-149 \text{ kg éq CO}_2 \text{ ha}^{-1} \text{ an}^{-1}$), agissant comme un puits de CO_2 à long terme. Des tendances similaires ont été observées lorsque le bilan GES était exprimé par unité d'azote apporté ou d'azote exporté. Les quatre systèmes agricoles ont eu des effets dissemblables sur le sort de l'azote, qui ne pouvaient être prédits par le surplus azoté. Des prédicteurs complémentaires des pertes d'azote et de l'équilibre des GES sont nécessaires pour

obtenir une véritable vue d'ensemble des impacts environnementaux C et N des systèmes de culture.

Enfin, dans un dernier temps, le modèle STICS a été utilisé pour simuler la production végétale, le surplus d'azote, la lixiviation des nitrates, les pertes gazeuses azotées et les variations du N organique du sol dans deux essais de long terme DOK en Suisse (39 ans) et Foulum au Danemark (19 ans). Quatre traitements ont été considérés dans chaque dispositif : deux traitements conventionnels avec (CONFYM) ou sans fumier (CONMIN), organiques avec du fumier (BIOORG) et un traitement non fertilisé (NOFERT) à DOK; conventionnel (C4-CC + IF) et trois traitements biologiques, l'un avec des cultures intermédiaires seulement (O4 + CC-M) et deux autres avec des cultures intermédiaires et prairie avec (O2 + CC + M) ou sans fumier (O2 + CC- M), à Foulum. Le modèle STICS a été calibré dans les traitements conventionnels et a permis de simuler de façon satisfaisante la production de cultures et le N exporté dans tous les traitements, avec un léger biais pour les teneurs en N et en eau dans le sol. L'évolution temporelle des stocks de N organique du sol a été bien capturée, ainsi que le surplus azoté. Ce surplus de N varie fortement entre les traitements au DOK, de -58 (NOFERT) à +21 kg N ha⁻¹ an⁻¹ (CONFYM), et seulement de -9 (O4 + CC-M) à +21 kg N ha⁻¹ yr⁻¹ (O2 + CC + M) dans Foulum. La lixiviation simulée de N ne différait pas entre les traitements à DOK, alors qu'elle était réduite de 41% en O4 + CC-M par rapport à C4-CC + IF à Foulum, en raison des effets combinés des cultures intermédiaires et de l'absence d'engrais azoté minéral. La volatilisation de N était la plus élevée dans CONFYM et O2 + CC + M. La dénitrification par N était la plus élevée dans BIOORG au DOK et 53% plus élevée dans les traitements fertilisés au Foulum. Les variations de stocks d'azote du sol ont toujours été négatives (de -18 à -78 kg N ha⁻¹ an⁻¹), conformément aux excédents de N mesurés, en fonction de la fertilisation et de la rotation des cultures. Le modèle simule d'importants flux de N liés aux résidus de racines profondes (1-14 kg N ha⁻¹ an⁻¹), c'est-à-dire en dessous de la couche labourée, en particulier dans les systèmes comprenant une prairie. Le devenir de cet azote profond (supposé s'accumuler dans le sol) n'a pas été simulé. Cette étude a montré que le modèle STICS était capable d'imiter le devenir de l'azote dans les systèmes de culture biologiques et conventionnels avec des formalismes communs. Nos simulations ont souligné le fait que les systèmes biologiques arables n'induisaient pas systématiquement des pertes plus faibles d'azote que les systèmes conventionnels, fournissant une marge de progression pour l'augmentation de l'efficacité d'utilisation de ces systèmes.

Cette thèse a permis de mettre en évidence la nécessité de précautions quant à l'utilisation et la diffusion des indicateurs environnementaux. Comme montré, la balance GES est l'indicateur le plus complet pour l'évaluation des flux C et N affectés par les systèmes de culture. Les politiques agricoles ciblant un seul objectif environnemental, comme l'initiative «4 pour 1000» pour le carbone ou la Directive Nitrate, doivent être considérées avec une grande vigilance car elles peuvent potentiellement être contreproductives, comme l'illustre l'exemple du stockage de carbone en surface du sol en agriculture de conservation tandis que des émissions de N_2O y sont aussi accrues. Par conséquent, une évaluation appropriée de l'impact environnemental d'un système de culture devrait être basée sur une évaluation globale, en considérant les flux C et N modifiés par les pratiques agricoles et ne pas se limiter à un seul indicateur. Une telle évaluation nécessite un suivi plus poussé des études à long terme avec des successions de cultures et des situations pédoclimatiques contrastées. Dans une plus large mesure, l'indicateur d'empreinte azoté (N footprint) a été développé pour mettre en évidence la perte potentielle de N dans l'environnement par la production et la consommation d'aliments et de combustibles fossiles (Shibata et al., 2016). Cette approche tente d'impliquer non seulement des secteurs de production tels que l'agriculture mais aussi le secteur de la consommation, créant la demande pour la nourriture produite. En plus de la mise en place de systèmes de cultures durables, une recherche approfondie est nécessaire pour développer d'autres leviers participant à la réduction des pertes d'azote, en particulier avec une meilleure connaissance des cultures et l'amélioration génétique. En fait, combler l'écart entre les rendements attendus et les rendements réels en augmentant l'efficacité d'absorption de l'azote devrait permettre de minimiser les pertes dans l'environnement (Foley et al., 2011). De plus, la réduction des pertes d'azote liées à l'activité agricole est fortement liée à une réduction des déchets alimentaires, qui représentent 30 à 40% de la production totale (Gustavsson et al., 2011). Certains auteurs ont proposé un changement dans l'alimentation humaine pour réduire ces pertes, la transition vers des régimes alimentaires à base de plantes réduisant potentiellement les émissions de gaz à effet de serre liées aux aliments de 29 à 70% (Springmann et al., 2016). Enfin, cette étude s'est concentrée sur l'impact environnemental lié aux destins C et N dans les agro-écosystèmes. Il conviendrait toutefois de prendre en considération le fait que les pesticides utilisés en agriculture peuvent également générer une pollution persistante des sols, de l'air et des ressources en eau, et entraîner des effets nocifs pour les êtres vivants (Gilden et al., 2010).

Title : Quantification and modelling of carbon and nitrogen fate in alternative cropping systems experiments on the long term

Keywords : organic farming, conservation agriculture, carbon, nitrogen, leaching, nitrogen surplus, greenhouse gas, modelling, STICS

Abstract : Agricultural activities can lead to imbalanced carbon (C) and nitrogen (N) dynamics compared to natural terrestrial eco-systems, causing potential damages for soil, water and air quality. Among these prejudices, decreased soil C and N stocks, increased nitrate leaching in waters and gaseous N emissions towards the atmosphere are of a major concern. To reduce these environmental impacts, innovative and sustainable farming systems are promoted, such as low inputs cropping systems, “conservation” agriculture or organic farming. The objectives of this work were i) to quantify the long term impact of different alternative cropping systems on the fate of C and N in the soil-plant-atmosphere system and ii) to simulate C and N dynamics with the agro-environmental model STICS. For this purpose, we studied three long-term field trials: the experiment of La Cage (France) established in 1998, the DOK (Switzerland) started in 1978 and the Foulum Organic (Denmark) established in 1998. The methodological approach combined experimentation and modelling. While La Cage trial enabled an in situ quantification of soil organic C and N storage, N leaching, nitrous oxide (N₂O) emissions and greenhouse gas (GHG) balance in alternative cropping systems compared to conventional, the Swiss and Danish experiments were used for in silico estimation of the C and N fates in organic cropping systems, after adaptation of the STICS model, followed by calibration and evaluation of the model. Significant annual SOC and SON accumulation was found under conservation agriculture and organic farming at La Cage, whereas no significant change was observed in the low input and the conventional systems. No difference of specific SOC and SON mineralization rates was found between systems in vitro or in silico : we conclude that the higher C and N storage in soil observed in the conservation and organic systems was mainly driven by increased crop residues, rather than by the effect of no tillage practiced in conservation agriculture. The N surplus, *i.e.* the difference between N inputs and N exports at the field scale, varied widely between treatments. The fate of this N surplus also varied between systems with wide variations in SON storage and gaseous losses but no differences in N leaching. The cumulative N₂O emissions measured continuously for three years were highly correlated with the calculated gaseous N losses (volatilization and denitrification), with higher losses in the conservation system. These calculations allowed establishing a full GHG balance. Therefore the four agricultural systems dissimilarly impacted the N fate, which could not be predicted by the N surplus alone. The GHG balance is a much better indicator of the environmental impact of cropping systems relative to C and N fluxes. In the Danish and Swiss experiments, the soil-crop model STICS was used to mimic crop production, N uptake and N surplus. The model was first adapted and evaluated to simulate organic farming systems. The model could satisfactorily simulate crop production, N uptake, N surplus and SON storage in the organic and conventional systems of these two longterm experiments. Model outputs suggested that the N fate could be contrasted according to fertilization and crop management, and that N losses were not systematically reduced in organic compared to conventional cropping systems. This study challenges the frequent belief that alternative cropping systems systematically improve the global C and N environmental impacts of agriculture.

Titre : Quantification et modélisation du devenir du carbone et de l'azote de systèmes de culture alternatifs en situation expérimentale de longue durée.

Mots-clés : agriculture biologique, agriculture de conservation, carbone, azote, lessivage, surplus azoté, gaz à effet de serre, modélisation, STICS

Résumé : L'activité agricole peut entraîner un déséquilibre des cycles du carbone (C) et de l'azote (N) dans les écosystèmes terrestres naturels et entraîner une diminution des stocks de C et N dans le sol, une augmentation de la lixiviation du nitrate et des pertes d'azote par voie gazeuse. Pour réduire ces impacts environnementaux, la mise en place de systèmes agricoles innovants et durables est encouragée, tels que les systèmes à bas niveau intrants, l'agriculture de conservation ou l'agriculture biologique. Les objectifs de cette thèse sont i) de quantifier l'impact à long terme des différents systèmes de culture sur le devenir du carbone et de l'azote dans le système sol-plante-atmosphère et ii) de simuler la dynamique de ces éléments avec le modèle agro-environnemental STICS. À cette fin, nous avons étudié trois essais de longue durée : l'essai de La Cage (France) établi en 1998, l'essai DOK (Suisse) débuté en 1978 et l'essai Foulum (Danemark) créé en 1998. Alors que l'essai de La Cage a permis une quantification in situ du stockage du carbone et de l'azote organiques du sol, de la lixiviation de l'azote, des émissions de protoxyde d'azote (N_2O) et de la balance des gaz à effet de serre pour des systèmes de culture alternatifs, les essais danois et suisses ont permis l'estimation in silico du devenir du C et N en agriculture biologique, après adaptation du modèle STICS pour simuler de nouvelles pratiques culturales. Après 16 années d'expérimentation, une accumulation annuelle significative de SOC et de SON a été observée en agriculture et en agriculture biologique à La Cage, alors qu'aucun changement significatif n'a été observé dans les systèmes conventionnels et bas intrants. La minéralisation spécifique de SOC et SON des quatre systèmes, simulée sur AMG et mesurée lors d'incubation des sols pendant quatre mois, s'est montrée équivalente entre systèmes. Le stockage de C et N observé dans les systèmes de conservation et biologiques s'explique principalement par l'augmentation des résidus de cultures plutôt que par l'effet du non-labour en agriculture de conservation. De plus, le surplus azoté (différence entre apports et exportations d'azote) a été calculé pour chaque système de culture. Le devenir de l'excédent d'azote a été estimé entre stockage de N dans le sol, pertes gazeuses et lixiviation de l'azote. Les émissions cumulatives de N_2O mesurées en continu pendant plus de trois ans sont fortement corrélées avec les pertes totales calculées de N par voie gazeuse (volatilisation et dénitrification), ces pertes étant les plus importantes dans le système de conservation. Enfin, la réalisation d'un bilan complet des émissions de GES a montré de fortes différences entre système et des phénomènes de compensation entre stockage et perte de C et N. Le modèle sol-culture STICS a ensuite été utilisé pour simuler le devenir de l'azote dans les essais DOK et Foulum. Après une adaptation du modèle, sa calibration et son évaluation ont été réalisées permettant de simuler de façon satisfaisante les rendements, l'absorption de N, le surplus de N et l'évolution des stocks de SON dans les systèmes conventionnels et biologiques. Les simulations suggèrent que le devenir de l'azote dans ces systèmes peut être contrasté en fonction de la fertilisation et de la gestion des cultures et que les pertes d'azote ne sont pas systématiquement réduites en agriculture biologique par rapport au conventionnel. Cette thèse remet en question les appréciations simplistes qui associent systématiquement systèmes de culture alternatifs et diminution des impacts environnementaux liés aux cycles de C et N.