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The fate of cumulative applications of ^{15}N-labelled fertiliser in perennial and annual bioenergy crops

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ABSTRACT

The fate of nitrogen (N) fertiliser applied to bioenergy crops is a key issue to allow high biomass production while minimising environmental impacts due to N losses. The aim of this study was to follow the fate in the soil-plant system of N fertiliser applied to perennial (*Miscanthus* × *giganteus* and switchgrass), "semi-perennial" (fescue and alfalfa) and annual (sorghum and triticale) bioenergy crops. Crops received ^{15}N-labelled fertiliser (urea ammonium nitrate solution) during 4 or 5 successive years on the same subplots, at a rate varying from 24 to 120 kg N ha^{-1} yr^{-1}. Biomass production, N and ^{15}N removal at harvest were measured each year. The ^{15}N recovery in crop residues, non-harvested crop parts and soil was measured at the end of the ^{15}N-labelling period. Perennial crops had higher biomass production but generally lower ^{15}N recovery in harvested biomass than other crops, particularly when harvested late (end of winter). At the end of the 4 or 5-year period, the proportion of ^{15}N recovered in harvested biomass was 13–34% for perennials, 23–38% for semi-perennials and 34–39% for annual crops. Perennial crops stored large amounts of N in their belowground organs; the mean ^{15}N recovery in these organs was 12%, corresponding to a N storage flux of 14 kg N ha^{-1} yr^{-1}. The ^{15}N recovery in soil (including crop residues) was higher for perennials (average 36%) than semi-perennials (28%) and annual crops (19%), corresponding to a N immobilisation rate of 43, 15 and 12 kg N ha^{-1} yr^{-1} respectively. The mean overall ^{15}N recovery in the soil-plant system was 69% in perennials, 61% in semi-perennials to 56% in annual crops, suggesting that important fertiliser losses occurred through volatilisation and denitrification. Perennial bioenergy crops had the better efficiency by storing fertiliser-N in soil organic matter and living belowground biomass used as N reserves for succeeding years.

1. Introduction

Bioenergy production from crops has been supported to contribute to the production of renewable energy in response to the challenges of climate change and depletion of fossil resources (Don et al., 2011). However, the use of conventional food crops to produce biofuels has raised a lot of concerns about its environmental consequences (*e.g.* Galloway et al., 2008; Smith and Searchinger, 2012). The large nitrogen (N) requirements of these first generation bioenergy crops may be harmful to the greenhouse gas balance of biofuels (Crutzen et al., 2008). The development of new conversion technologies and biorefineries allows considering a wide range of new bioenergy crops (Ragauskas et al., 2006; Sanderson and Adler, 2008; Somerville et al., 2010). Among them, perennial C4 crops such as miscanthus and switchgrass are considered as promising because of their high biomass production with low nutrient requirements and expected low greenhouse gas emissions (Don et al., 2011; Jørgensen, 2011; Monti et al., 2012; Cadoux et al., 2014). However, even with these crops, N fertilisation may still be necessary to maintain high yields and soil fertility on the long term (Cadoux et al., 2012; Monti et al., 2012; Cadoux et al., 2014). Fertiliser-N use efficiency of bioenergy crops is therefore a key issue to allow high biomass production while minimising environmental impacts due to N losses.

There are various ways to define and measure fertiliser-N efficiency. Two different approaches are widely used in the literature: (1) the apparent recovery which is based on the difference in N uptake between a crop receiving N fertiliser and a reference plot without N applied (*e.g.* Cassman et al., 2002) and (2) the actual recovery or ^{15}N recovery which is the fraction of labelled

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N that is taken up by a crop following application of ^{15}N-labelled fertiliser (Hauck and Bremner, 1976). Both methods can give similar or dissimilar results whether or not the uptake of inorganic soil N is different between fertilised and unfertilised treatments. "Pool substitution" between fertiliser-N and soil mineral N can lead to a higher apparent than actual recovery (Jenkinson et al., 1985). Nevertheless, only the ^{15}N method allows to determine the fate of the fertiliser-N in the different compartments of the agro-ecosystem (*e.g.* crop and soil) and therefore the overall losses of fertiliser-N (Gardner and Drinkwater, 2009). Few studies have analysed the fate of ^{15}N-labelled fertiliser applied to bioenergy crops. Christian et al. (2006) and Pedroso et al. (2014) analysed the effect of a single ^{15}N fertilisation pulse during 3 successive years on miscanthus and switchgrass respectively. They found a rather low ^{15}N recovery in the harvested biomass (14–39%) and that belowground organs and soil represented important N sinks. Pedroso et al. (2014) also pointed out the effect of crop management, *i.e.* harvest date, on the ^{15}N recovery and partitioning. However, no study has compared the ^{15}N recovery of different bioenergy crops at the same site.

Only a few experiments have followed the recovery of ^{15}N in the soil–plant system on the long term. In arable cropping systems, a small proportion of the residual ^{15}N, *i.e.* the labelled fertiliser-N remaining in soil (mainly in organic form) and crop residues after harvest, is re-mineralised each year from the soil organic matter pool and can be recovered by the following crops or lost through N leaching or gaseous losses (Glendining et al., 2001; Macdonald et al., 2002; Sebilo et al., 2013). The amount of ^{15}N remaining in the soil-plant system after the year of application is likely to be greater with perennial bioenergy crops than with annual crops because of the presence of perennial organs. The ^{15}N stored in perennial organs can be used by the crop in the subsequent years and partly recovered at harvest, as shown by Christian et al. (2006) for miscanthus. Using cumulative applications of ^{15}N in the same plots over several growing seasons could allow to integrate part of the long-term fate of the residual ^{15}N and to reduce the variability in plant N uptake and fertiliser-N losses due to climate conditions and agronomical context (*i.e.* age of the crop, other stresses, etc.). In this study, we used cumulative applications of ^{15}N-labelled fertiliser for four or five years to determine (1) the fate of fertiliser applied to perennial, semi-perennial and annual bioenergy crops in the soil-plant system, and (2) the interaction with crop management, *i.e.* harvest date of perennial crops and N fertiliser rate for all crops. We hypothesised that perennial crops would export smaller amounts of ^{15}N through harvests than the other crops but would store larger amounts of ^{15}N in perennial organs and soil organic matter, leading to an equal or improved overall ^{15}N recovery in the soil-plant system.

2. Materials and methods

2.1. Site and experimental design

The study is based on an ongoing long-term experiment established in 2006 at the INRA experimental station in Estrées-Mons, northern France (49.872 N, 3.013 E) called "Biomass & Environment" (B&E). The soil is a Haplic Luvisol (IUSS Working Group WRB, 2006). Soil characteristics are given in Table S1 (Appendix A). Over the period 2006–2011, the mean annual temperature was 10.6 °C, the mean rainfall and potential evapotranspiration were 673 and 737 mm yr^{-1} respectively. Before 2006, the field had been cultivated for many years with annual crops, winter wheat being the most common crop.

The experiment was initiated to study the production and the environmental impacts of a wide range of bioenergy crops. It compares eight "rotations": four with C4 perennial crops (monocultures), two with C3 "semi-perennial" crops (destroyed every two or three years) and two with C3/C4 annual crops (Table 1). The perennial crops are miscanthus (*Miscanthus* × *giganteus* Greef & Deuter ex Hodkinson & Renvoize) and switchgrass (*Panicum virgatum* cv. Kanlow). They are harvested either early in October (E) or late in February (L). The semi-perennial crops are tall fescue (*Festuca arundinacea*) and alfalfa (*Medicago sativa*). Annual crops are fibre sorghum (*Sorghum bicolor* (L.) Moench cv. H133) and triticale (×*Triticosecale* Wittmack). The experiment also includes two nitrogen treatments (N- and N+) with fertiliser-N rates depending on the crops (Table 2). The rationale for defining the N rates was explained by Cadoux et al. (2014).

Table 1
Rotations of the B&E long term experiment.

Rotation	2006	2007	2008	2009	2010	2011
Mis E	Mis n.h.	Mis E	Mis E	Mis E	Mis E	Mis E
Mis L	Mis n.h.	Mis L	Mis L	Mis L	Mis L	Mis L
Swi E	Swi n.h.	Swi E	Swi E	Swi E	Swi E	Swi E
Swi L	Swi n.h.	Swi L	Swi L	Swi L	Swi L	Swi L
Fes-Alf	CC/Fes	Fes	Fes	Alf	Alf	Fes
Alf-Fes	Alf	Alf	Alf	Fes	Fes	Alf
Sor-Tri[a]	CC	Sor	Tri/CC	Sor	Tri/CC	Sor
Tri-Sor[a]	Sor	Tri/CC	Sor	Tri/CC	Sor	Tri/CC

Mis = miscanthus, Swi = switchgrass, Fes = fescue, Alf = alfalfa, Sor = fiber sorghum, Tri = triticale, CC = catch crop; E = early harvest, L = late harvest, n.h. = not harvested.
[a] Rotations with catch crops (oats in 2006, rye in 2007, mustard in 2008, oat–vetch mixture in 2009 and mustard–clover mixture from 2010 to 2011) which were sown every year in late August or early September between triticale and sorghum.

The 2.7 ha field was divided into two parts in order to facilitate cropping operations and limit competition between plants due to differences in canopy height: (1) a split-block design in the west part for perennial crops with "rotations" in the main plots (miscanthus early, miscanthus late, switchgrass early, switchgrass late) and N fertilisation rates in the subplots (N− and N+), and (2) a split-plot design in the east part for the other crops with rotations in the main plots (fescue-alfalfa, alfalfa-fescue, sorghum-triticale and triticale-sorghum) and N fertilisation rates in the subplots (N− and N+). Each of the two parts comprised three replicate blocks and 24 subplots of 360 m^2 (Fig. S1, Appendix A). Soil analyses performed in 2006 revealed a slightly higher clay content in the west than in the east part (180 ± 27 *vs.* 148 ± 19 g kg^{-1} in the 0–30 cm layer, Table S1 in Appendix A).

At the start of the experiment, the field was mouldboard ploughed at a depth of *ca.* 25 cm. After seedbed preparation, miscanthus was planted in April 2006 (1.5 rhizome m^{-2}) and switchgrass sown in June 2006 (seed rate = 15 kg ha^{-1}). In 2006, perennial crops were not harvested because of the low biomass production during the first year of growth. Their aboveground biomass was cut and left on the soil surface. Semi-perennial crops were sown in 2006, 2009 and 2011, usually in April. Before sowing, the previous crop (alfalfa or fescue) was destroyed in late autumn with a cultivator and a disc harrow (15 cm deep) in 2008 and mouldboard ploughed (*ca.* 22 cm deep) in 2010. These crops were harvested in two or three cuttings depending on years, with the last cut in October. Annual crops were tilled superficially (12–15 cm deep) without inversion ploughing. Sorghum was sown in late May and harvested in late September. Triticale was sown in mid-October and harvested in late July or early August. The N fertiliser was surface-applied from 2007 onwards as UAN solution (urea ammonium nitrate) containing 390 g N l^{-1} (50% urea, 50% NH_4NO_3). Perennial crops received a single annual application in late April. Fescue received N fertiliser at the beginning of each cycle of regrowth and seedling crops were not fertilised before the first