Europ. J. Agronomy 64 (2015) 1–7

Contents lists available at ScienceDirect

European Journal of Agronomy

journal homepage: www.elsevier.com/locate/eja

# The use of $^{13}C$ and $^{15}N$ based isotopic techniques for assessing soil C and N changes under conservation agriculture

Kenza Ismaili, Mohammed Ismaili*, Jamal Ibijbijen

*Université Moulay Ismail, Faculté des Sciences BP 11201 Zitoune, Meknès 50000, Morocco*

A R T I C L E I N F O

*Article history:*
Received 18 July 2013
Received in revised form 13 October 2013
Accepted 2 December 2014
Available online 12 December 2014

*Keywords:*
Conservation agriculture
$^{13}C$
$^{15}N$
Soil organic matter
Tillage

A B S T R A C T

A four years experiment was conducted to investigate the effect of tillage and addition of crop residues in wheat–faba bean rotation. The soil was fertilized with a total of 150 kg nitrogen (N) ha$^{-1}$ enriched with 9.96 percent nitrogen-15 ($^{15}N$) atom excess, in four applications. The first crop was corn, a $C_4$ plant cropped under till (T) and no-till (NT) conditions. Percent N derived from fertilizer (Ndff) by corn was 37.12–48.62. The leaves had the lowest delta carbon-13 ($\delta^{13}C$) values of −12.7 and the seeds the highest (−11.8). Soil $\delta^{13}C$ was affected by addition of $C_4$ plant residues. Soil under residue and till treatment (RT) had the highest percent $^{15}N$ values. Residues and no-till (RNT) had the lower percent $^{15}N$ values. At the end of the corn crop soil percent $^{15}N$ was 0.211, 0.26, and 0.253 in the three soil depths. Residues and tillage increased significantly the Ndff of wheat: from 6.43 to 6.46 kg N ha$^{-1}$ for no residues no-till (NRNT) and no-residues and till (NRT) and from 11.1 to 13 kg N ha$^{-1}$ in RNT and RT treatments. In wheat nitrogen derived from residues (Ndfr) was 4.68 and 1.53 kg N ha$^{-1}$ in grain and residues, respectively. Residues and tillage affected significantly soil C, N, $^{15}N$, and $\delta^{13}C$ from seeding to two months after and have no effect at harvest. The interaction was always significant. After four years the $^{15}N$ fertilizer contributed only with 1.5–2.85 kg N ha$^{-1}$ in NRNT and NRT, respectively, and 3.3–5.63 kg N ha$^{-1}$ in RNT and RT, respectively. Cumulated N recovered during the three growing seasons following corn was 8.59–11.07 kg N ha$^{-1}$ for NRNT and NRT; 20.24–15.6 kg N ha$^{-1}$ for RT and RNT, respectively. Residues increased N mineralization by 50 percent and the quantity of $^{15}N$ available to plants increased with tillage.



## 1. Introduction

Conservation agriculture (CA) or sustainable agriculture maintains or enhances soil C and N fertility by suppression of soil tilling and no incorporation of added organic matter. In consequence yield and soil structure are improved. The evolution of the organic matter and the interaction of the mineral fertilizers is an important research aspect. However the use of $^{13}C$ and $^{15}N$ in studies of utilization and evolution of fertilizers and relative contribution of $C_4$ and $C_3$ plants to soil organic matter is not new (Drinkwater et al., 1998). Similarly there are many studies concerning fate of N mineral or organic fertilization using $^{15}N$ labelled fertilizers. Drinkwater et al. (1998) in a 15-year study compared the conventional system of maize/soybean rotation using mineral fertilizers with two organic crop rotations and found that organic methods are as productive as conventional ones, in addition to significant qualitative and quantitative improvement in soil fertility and structure. In a thorough literature review Giller et al. (2009) showed positive effect of CA on water availability and soil N (reduced soil evaporation, reduced water runoff, increased water infiltration and reduced soil temperature). Long term sustainability of crop production and carbon sequestration in soil are strategies to mitigate climate change (Galloway et al., 2008), in addition to soil fertility giving particular attention to soil nitrogen (N mineralization and immobilization) (Nannipieri & Eldor, 2009). Recently-formed soil organic matter (SOM) may follow different pathways of mineralization and/or stabilization depending on soil properties and climate conditions (von Lutzow et al., 2006). Crop residues reduce the effect of drought and improve water and N storage in soil and protect soil from high temperatures (Cassman et al., 2003). Blesh and Drinkwater (2013) constructed N mass balances for a gradient of farm types and concluded that diversified farmers were more likely to adjust their management practices in response to environmental variability and that agroecosystems will require reducing N inputs and increasing C availability.

* Corresponding author. Personal address: N°9, Rue All Attorounj, Secteur 24, lot 9 Bloc H, Riad-Agdal, Riad, 10100, Rabat. Tel.: +212 6 64 55 12 01; fax: +212 5 35 58 38 08.

*E-mail addresses:* kenza0505@hotmail.com (K. Ismaili), ismailih2000@yahoo.fr (M. Ismaili), jamal_ibijbijen@yahoo.fr (J. Ibijbijen).

http://dx.doi.org/10.1016/j.eja.2014.12.005
1161-0301/

Soil temperature and humidity affect the activity of soil micro organisms and induces a rapid SOM degradation (Grall et al., 2006; Van der Stelt et al., 2007). Nitrogen that remains immobilized will lead to reduced N losses via leaching and/or denitrification (Scow, 1997). The use of labelled organic matter by $^{13}C$ and $^{15}N$ isotopes is a best method to study the evolution of added organic and mineral fertilizers and the interaction between C and N in soil. The use of natural abundance of stable isotopes in plant ecology have focused largely on variations in bulk delta carbon-13 ($\delta^{13}C$) (Bowling et al., 2008), nitrogen isotopic composition of soil solution (Billy et al., 2010), Strontium/Calcium ratio and oxygen stable isotopes (Fink et al., 2010). Smith et al. (2000) showed that $\delta^{13}C$ values of soil can be used to evaluate C sequestration through plant residues addition to soil. The wide diversity of $\delta^{13}C$ values of grassland plants was found to be due to environmental conditions (Dungait et al., 2010). Previous studies have reported differences in leaf bulk $\delta^{13}C$ values between life forms (Zheng and Shangguan, 2007). Cerri et al. (1985) used differences in $\delta^{13}C$ values of $C_3$ plants (−24 to −34) and $C_4$ plants (−6 to −19) to evaluate water use efficiency in arid environments.

The $^{13}C$ natural abundance technique provides useful data to test SOM models in complex cropping systems in which both $C_3$ and $C_4$ plants are intercropped or rotated (Diels et al., 2004). The $^{13}C$ natural labeling in combination with modelling was used by Diels et al., 2000 to study long-term SOM changes in agroforestry systems through investigation of yearly changes of $\delta^{13}C$ in plant materials. Schwartz et al., 2007 used $\delta^{15}N$ and $\delta^{13}C$ of DNA extracted from soil to investigate soil N and C. Carbon isotope discrimination ($\delta^{13}C$) was correlated with grain yield and N availability under water stress conditions Monneveux et al. (2005); Dercon et al. (2006); Pansak et al. (2007); Stewart et al. (1995). The relationship and slope between available $NO_3$-N and $\delta^{13}C$ values suggests that differences in $\delta^{13}C$ were more related to availability of N than to differences in water availability.

Crop residues labelled with nitrogen-15 ($^{15}N$) were used to investigate, SOM degradation, available soil N and SOM-N pools (Ismaili et al., 2003). The form of N application; i.e., organic versus inorganic or in combination, may affect the amount of N and carbon that becomes sequestered in soil (Moran et al., 2005). Using data from 572 peer-reviewed studies Ladha et al. (2005), calculated that the average recovery of $^{15}N$-fertilizer by field grown maize, rice, and wheat in the grain was 30 percent which increased to 44 percent when straw was included. At all harvests, plant $^{15}N$ in the no residue treatments was lower than in the residue treatments showing a positive effect of the residues on soil nitrogen. Vanlauwe et al. (2001) showed that the combination of applying inorganic N and Senna (*Senna siamea*) residues improved N-fertilizer use efficiency by maize in low yielding production systems in West Africa. Compared to the control treatment without residue management, the N fertilizer use efficiency increased from 28 to 47 percent.

The objectives of this experiment were to study the fate of C and N in conservative agriculture: (i) follow the changes in SOC after adding $C_4$ plant residues (corn) enriched with $^{15}N$ under tilled and not-till treatments, (ii) evaluate the effects of CA system on soil N and C sequestration, and (iii) estimate the effect of corn residues and tillage on the $\delta^{13}C$ values of soil.

## 2. Material and methods

A four year experiment was conducted as part of a FAO/IAEA coordinated research project (CRP) to investigate CA system based on wheat (*Triticum durum* Oum Rbia G3 2004)–faba bean (*Vicia faba* L.) rotation. The study was completed on farm in the region of Meknes, at the center of Morocco, with an average rainfall of 500 mm year. The previous crop was fallow. The soil was sandy loam well drained, of poor fertility and low water holding capacity. Four replicates were used. The first crop was corn (*Zea mays* var. rugosa), a $C_4$ plant seeded in Spring 2005 and harvested in July 2005, on 16 plots (16 $m^2$) fertilized with 150 kg N $ha^{-1}$ in four applications: 25 kg N $ha^{-1}$ at seeding, 25 kg N $ha^{-1}$ two weeks after seeding, 50 kg N $ha^{-1}$ one month after seeding and 50 kg N $ha^{-1}$ two months after seeding. Nitrogen was added as ammonium sulphate enriched with 9.96 percent atom $^{15}N$ excess at all applications. The second, third, and fourth applications were sprayed on the plots, soil and plant. Soil was previously amended with phosphate (100 kg $P_2O_5$ $ha^{-1}$) and potassium (40 kg $K_2O$ $ha^{-1}$). The corn crop was irrigated as needed, but the following wheat and faba bean crops were only rain fed (El Alami and Ismaili, 2007b). The second crop was wheat, a $C_3$ plant grown under no-till and till and with or without corn residues left on the soil. The wheat was seeded in fall 2005 and harvested in June 2006. The roots and crowns of the corn crop were left in the soil in all treatments; however two control treatments were added where corn was not grown in the plots and where residues removed from no-residues treatment plots were added to soil to estimate the direct effect of corn residues on soil N and C. For the first corn crop, the treatments were: NT = no-till and T = with tillage. After the first corn crop and in plots where corn was grown, the treatments were NRNT = no residues, no-till, NRT = no residues, with tillage, RT = with residues, with tillage, RNT = with residues and no-till. In the plots where corn was not grown the treatments were no-corn RT and no-corn RNT.

The wheat was sampled three times during the growing season: at one month after seeding, two months after seeding and at the end of the season. The third crop was faba bean seeded on fall 2006 and harvested in may 2007, and the forth crop was wheat seeded in fall 2007 and harvested in June 2008. The corn crop was used to enrich the soil with $C_4$ plant residues. At the end of the growing season, when the corn was at maturity and the leaves were still green we harvested the plants. One square meter was sampled and the leaves, stems, seeds, come, come petals and (crowns + roots) were separated and weighted then a sub sample was taken from each part and weighted then dried in the oven at temperature 64 °C for 72 h to determine dry weight of each plant part in the sub sample then in the sample and in one hectare. The sub sample of each plant part was grounded to determine %N, %C, percent atom $^{15}N$ excess at excess and $\delta^{13}C$. The total mass of residues was calculated as the total dry matter of all parts of the corn plant (not seeds) of both treatments T and NT. Total C of residues was calculated as the dry matter of the different parts of corn (not seeds) multiplied by percent C of related part in T and NT. The percent N, percent C, percent atom $^{15}N$ excess and $\delta^{13}C$ of residues were calculated as a weighted value of the same parameters of plant parts returned as residues to the soil. In the control plots roots and crowns were not included in the residues added to soil.

The main objective of the experiment was to investigate the effect of corn as a tracer of C in the soil and estimate the effect of added residues on soil $\delta^{13}C$. The $^{15}N$ fertilizer was added to label soil and residues as one system without separating the two sources. So we were referring at the addition of N fertilizer (150 Kg N $ha^{-1}$) in all crops and not the N in the residues. Only in the control plots where no corn was grown as previous crop, we could refer to the added residues and calculate the N derived by wheat from residues and separate the effects of the residues and roots, crowns, and soil N derived from fertilizer. So we were interested on how much N was used by the crops, after corn and how much was left in the soil, and how this system affects soil parameters.

Soil was sampled at harvest time of corn crop before seeding the wheat, one month after seeding the wheat, two months after seeding wheat and at wheat harvest to determine the evolution of percent C, percent N, percent $^{15}N$ and $\delta^{13}C$ (Van Kessel et al., 1994) at three different soil depths: (0–25 cm), (25–50 cm),