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ABSTRACT

Under semiarid Mediterranean conditions irrigated maize has been associated to diffuse nitrate pollution of surface and groundwater. Cover crops grown during winter combined with reduced N fertilization to maize could reduce N leaching risks while maintaining maize productivity. A field experiment was conducted testing two different cover crop planting methods (direct seeding versus seeding after conventional tillage operations) and five different cover crops species (barley, oilseed rape, winter rape, common vetch, and a control (bare soil)). The experiment started in November 2006 after a maize crop fertilized with 300 kg N ha⁻¹ and included two complete cover crop-maize rotations. Maize was fertilized with 300 kg N ha⁻¹ at the control treatment, and this amount was reduced to 250 kg N ha⁻¹ in maize after a cover crop. Direct seeding of the cover crops allowed earlier planting dates than seeding after conventional tillage, producing greater cover crop biomass and N uptake of all species in the first year. In the following year, direct seeding did not increase cover crop biomass due to a poorer plant establishment. Barley produced more biomass than the other species but its N concentration was much lower than in the other cover crops, resulting in higher C:N ratio (>26). Cover crops reduced the N leaching risks as soil N content in spring and at maize harvest was reduced compared to the control treatment. Maize yield was reduced by 4 Mg ha⁻¹ after barley in 2007 and by 1 Mg ha⁻¹ after barley and oilseed rape in 2008. The maize yield reduction was due to an N deficiency caused by insufficient N mineralization from the cover crops due to a high C:N ratio (barley) or low biomass N content (oilseed rape) and/or lack of synchronization with maize N uptake. Indirect chlorophyll measurements in maize leaves were useful to detect N deficiency in maize after cover crops. The use of vetch, winter rape and oilseed rape cover crops combined with a reduced N fertilization to maize was efficient for reducing N leaching risks while maintaining maize productivity. However, the reduction of maize yield after barley makes difficult its use as cover crop.

Keywords:

cover crops, direct seeding, maize, nitrogen, SPAD, ,tillage

Abbreviations:

DM, dry matter; KM, kernel mass; ET, evapotranspiration.

1. INTRODUCTION

Monoculture maize in semiarid conditions can be a high yielding crop (15 Mg ha⁻¹ of grain), but has a high water and N input demand, with total plant N uptake of 300 kg N ha⁻¹ and over (Moreno et al., 1996; Berenguer et al., 2009). Management of irrigation water and N fertilizer have been recognized as the main factors controlling N leaching risks and diffuse nitrate pollution of surface water and groundwater in irrigated semiarid areas of Spain (Diez et al., 1997; Cavero et al., 2003; Causapé et al., 2004; Isidoro et al., 2006) and of USA (Pratt et al., 1984; Klocke et al., 1999).

Reducing N fertilizer rates applied to maize can decrease N leaching risks and several works have studied the effect of N rates on the return flows from irrigated or rainfed fields (Martin et al., 1994; Diez et al., 2000; Sogbedji et al., 2000). However, due to the uncertainty for adjusting maize N fertilizer requirements under field conditions, often farmers apply N fertilizer rates that exceed maize N requirements in order to avoid risks of yield losses. Data from surveys in the Ebro River Basin (a semiarid irrigated area of Spain) indicate that rates of 318 - 453 kg N ha⁻¹ yr⁻¹ are applied every year by farmers (Cavero et al., 2003; Isidoro et al., 2006). When excess of N fertilizer is applied, residual N at harvest can be leached during the intercrop period of maize (October to April) (Moreno et al., 1996), depending on the unpredictable rainfall distribution under semiarid conditions. Moreover, N can be lost during the beginning of maize growing season when irrigation water applied exceeds crop evapotranspiration (Moreno et al., 1996; Salmerón et al., 2010).

N leaching risks also depend on irrigation management (Martin et al., 1994; Schepers et al., 1995; Pang, et al., 1997; Diez et al., 2000; Cavero et al., 2003; Causapé et al., 2006). Sprinkler irrigation allows high irrigation efficiencies, which can reach values close to 95% in sprinkler irrigated watersheds (Cavero et al., 2003), but some leaching fraction is generally needed in semiarid areas to prevent soil salinization problems in the long term (Oster, 1994). Surface irrigation systems usually result in lower irrigation efficiencies and higher N leaching losses (Isidoro et al., 2006).

Improving irrigation and N fertilizer management can reduce significantly N leaching losses (Diez et al., 2000). Adequately managed sprinkler irrigation combined with split N fertilizer applications should minimize N losses in maize, but results from monitored sprinkler irrigated watersheds indicate significant annual losses ranging from 25 to 50 kg N ha⁻¹ (Cavero et al., 2003). This suggests that adequate management of irrigation and N fertilizer should be complemented with other strategies to minimize N leaching.

Cover crops in humid climates are known to reduce N leaching during winter when precipitation is high (McCracken et al., 1994; Martinez and Guiraud, 1990; Ball-Coelho et al., 2004; Tonitto et al., 2006). Growing winter cover crops before irrigated maize under semiarid conditions is not a common practice, as winter precipitation is usually low. However, cover crops have proved to be useful to reduce N leaching risks by depleting residual soil N (Gabriel and Quemada, 2011) and reducing N leaching at the start of irrigation and during maize growing season (Salmerón et al., 2010). Cover crops reduced nitrate concentration in drainage water, whereas drainage volume was unaffected during the maize growing season (Salmerón et al., 2010). This enabled a reduction in nitrate leaching while maintaining an adequate leaching fraction, which is a key factor to avoid salt accumulation in irrigated areas (Oster, 1994).

Residual N is not only an important potential source for water pollution but also the producer can save money if this N is used by the next crop instead of being lost with drainage water. When winter cover crops are incorporated into the soil, part of the N contained in the cover crop residue can be mineralized (Stivers-Young, 1998) becoming available to the next crop. Therefore, the optimum N fertilizer rate to the subsequent maize crop should be reduced, as otherwise, N inputs in the system would be higher than without a cover crop, and it is likely that N losses would be greater in the long term (Thomsen and Kristensen, 1999; Hansen et al., 2000). In addition, a reduction of N fertilizer applied to maize will reduce total costs associated with cover crops, promoting their use by farmers. However, N fertilizer rates applied to maize after a cover crop should be well adjusted in order to avoid maize yield losses. Salmerón et al. (2010) found that maize grain yield can be reduced after non-legume cover crops in irrigated Mediterranean conditions because cover crop depleted the residual soil N after maize harvest but not all the N on the cover crops biomass was available for the following maize crop.

Inadequate release of N from biomass of non-legume cover crops has been found under similar (Wyland et al., 1995) and different climate (Clark et al., 2007a,b).

One constraint to the use of winter cover crops after maize is the short period of time available to plant the cover crop before frost. Direct seeding allows an earlier seeding of cover crops after maize harvest compared with seeding after conventional tillage because direct seeding does not require soil tillage operations prior to seeding. Moreover, soil tillage operations require that soil is not too wet. Besides, direct seeding reduces planting costs. However, emergence of small-seed cover crops such as *Brassica* species could be hampered due to soil crusting and coarse maize crop residues in the field when direct seeding is used. Emergence of white mustard (*Sinapis alba* L.) cover crop has been reported to be affected by humidity and temperature, but not by reduced tillage and previous crop residues (Dorsainvil et al., 2005). It is important to evaluate cover crop growth under different planting techniques and conditions in order to have a proper establishment of the cover crop.

The objectives of this study were: (1) to quantify the biomass and N uptake of different winter cover crops with two planting methods in a monoculture maize system under irrigation and, (2) to evaluate the effect of these cover crops on soil N dynamics, soil water content, and maize yield.

2. MATERIALS AND METHODS

2.1 Site and experimental design

The experiment was carried out from 2006 to 2008 in an experimental field at the Estación Experimental de Aula Dei (CSIC) located in the Ebro Valley (41°43'N; 0°49'W, 225 m altitude) in Zaragoza, Spain. The climate is Mediterranean semiarid with mean annual maximum and minimum daily air temperatures of 20.9 and 8.5°C, respectively, yearly average precipitation of 322 mm, and yearly average reference evapotranspiration of 1,100 mm. The soil is a clay loam (27% sand, 51% silt and 26% clay) classified as Typic Xerofluvent (Table 1). The soil was analyzed for pH (extract 1:2.5 in water), C (Walkley-Black) and N concentration (Kjeldahl),

CaCO₃ (potenciometry) and water content at field capacity and wilting point (Richard pressure plates) (Table1). The field was cropped with maize during three years previous to the start of the experiment.

The experimental design was a split plot with two factors and 3 replicates. The main factor studied was the planting method of the cover crops: direct seeding after maize harvest with notillage (DS) or seeding after soil preparation with conventional tillage operations after maize harvest (CT). Direct seeding allowed earlier planting dates, the soil was untilled, and the previous maize residue was on the soil surface. On the other hand, seeding after conventional tillage had later planting dates and the soil was tilled with the maize residue incorporated into the soil. These are the differences between the two planting methods under real field conditions and this is what we tested. The second factor studied was the different winter cover crop species: winter barley (*Hordeum vulgare* L., *cv*.Hispanic), oilseed rape (*Brassica napus* L., *cv*. Madrigal), winter rape (*Brassica rapa* L., *cv*. Perko), common vetch (*Vicia sativa* L., *cv*. Armantes), and a control treatment with bare soil during winter. The size of each experimental plot was 6 m by 18 m.

The experiment was started in November 2006 after a maize crop fertilized with 300 kg N ha⁻¹. Maize grain was harvested each year with a commercial combine that chopped the maize stubble and left it on the soil. In the DS treatment maize residue was left on the soil surface, whereas in the CT treatment it was incorporated immediately after maize harvest with a disc harrow. Cover crops were seeded with a commercial seed drill (SD-1203, Solá, Calaf, Spain) as soon as possible after maize harvest (Table 2), at seeding rates of 180, 7, 12 and 110 kg ha⁻¹ for barley, oilseed rape, winter rape and common vetch, respectively. In the DS treatments cover crops were seeded directly, whereas in the CT treatments, seedbed was prepared with a stubble cultivator before seeding. In the control treatment (bare soil) of the cover crop species factor (hereafter control treatment), the same soil tillage practices than for the cover crop species factor (barley, oilseed rape, winter rape and common vetch) were implemented within each planting method studied. Some irrigation was provided (40 mm in 2006 and 51 in 2007) after the winter cover crop species is emergence. In the following spring after cover crop growth in 2007 and 2008, the cover crops were mechanically incorporated into the soil with a power tiller (Table 2).

Maize cultivar 'Pioneer PR34N43' was planted on the reported dates in Table 2 at a plant density of 87,000 plants ha⁻¹ with a row distance of 0.75 m. Maize was fertilized with 300 kg N ha⁻¹ in the control treatment and this rate was split in three equal applications (100 kg N ha⁻¹): at preplant as urea (46% N), and two side-dress applications as urea ammonium nitrate solution (UAN, 32% N) at V6 and V12 growing stages. In the cover crop treatments pre-plant N application was reduced to 50 kg N ha⁻¹. P and K were applied before maize planting at a rate of 100 and 150 kg ha⁻¹ of P₂O₅ and K₂O, respectively. The pre-plant application of N was done by hand because it was different in the control treatment. The side-dress applications were made injecting the liquid UAN solution into the irrigation water system (fertigation).

Maize was irrigated using a solid set sprinkler irrigation system with spacing of 18 m x 18 m obtaining an application rate of 5 mm h⁻¹. Previous studies in the same field reported a high irrigation uniformity, close to 90% (Cavero et al., 2008). The weekly irrigation requirements were calculated from the daily ETo (estimated with the Penman-Monteith equation) and the crop coefficients, according to the FAO procedures (Allen et al., 1998) and considering an irrigation efficiency of 85%. The volume of irrigation applied was measured with an electromagnetic flow meter (Promag 50, Endress+Hauser, Reinach, Switzerland) which has a measurement error of \pm 0.5%. Total water applied as irrigation plus precipitation during maize growing season was 890 and 750 mm for 2007 and 2008, respectively. Weed and pest control was made according to the standard practices of the area to ensure an adequate growth of the maize crop.

2.2 Cover crops and maize growth analysis.

Cover crops were sampled before being incorporated into the soil by harvesting in each plot the aboveground biomass contained in two samples of 0.5 m² and making a composited sample of them. The samples were taken to represent the variability within each plot. The sample was then oven dried at 65°C, weighed and finely ground before total N and C analysis by combustion (TruSpec CN, LECO, St. Joseph, MI, USA).

Leaf greenness of maize was measured during the growing season with a chlorophyll meter (SPAD-502, Minolta Camera Co., Ltd., Japan). Measurements were done on the

youngest fully developed leaf until the silks emerged and later on the ear leaf. The average from 30 readings in different plants within each plot was calculated.

Maize was harvested on the dates reported in Table 2. All the ears in 2 rows of 10 m length per plot (15 m²) were hand harvested to determine yield, number of grains per square meter, and unit kernel mass (KM). The plants contained in a 2 rows x 2 m section (3 m²) were harvested and the grain was separated from the rest of the plant. Grain and plants were dried at 65°C, weighed and ground prior to analyses of total N and C similarly to the cover crops biomass. Grain yields are reported on the basis of 140 g kg⁻¹ moisture content.

Maize stalks to evaluate the end-of-season nitrate test were collected at harvest time from 15 plants following the procedure described by Binford et al. (1992). In all cases the sheaths were removed from the stalks, then oven dried at 65 °C and ground. A subsample of 2 g was extracted with 50 mL of KCI 2N, shaken for 30 min, filtered through a cellulose filter (Whatman no. 1) and analyzed with a continuous flow analyzer by spectrophotometry UV-Vis (THERMO-OPTEK, Iris Advantage Ers Duo, Thermo Fisher Scientific, Massachusetts, USA).

2.3 Soil analysis

Soil was sampled each year before cover crop incorporation and after maize harvest. Two soil cores from each experimental plot were taken with a 5 cm diameter hand auger (Eijkelkamp Agrisearch Equipment BV, The Netherlands) and the two samples were combined per depth in 0.3 m increments to 1.2 m depth. In the second year, soil was sampled to 2.1 m depth with an auger coupled to a tractor and soil samples were combined in 0.3 m increments as well. The soil was fresh-sieved to pass a 2 mm sieve, and 10 g were extracted with 30 ml of KCl 2N solution for determination of $NO_3^{-}N$ and $NH_4^{+}-N$ concentrations colorimetrically with a continuous flow analyzer (AA3, Bran+Luebbe, Norderstedt, Germany). Another subsample was dried at 105°C to constant weight for gravimetric water content determination. Gravimetric water content was converted to volumetric water content using a bulk density of 1.46 g cm⁻³, obtained from previous studies in the same experimental field.

An N budget was calculated for the maize crop period considering the 0 to 1.2 m soil layer. Inputs considered were soil mineral N before cover crop incorporation ($N_{inorg I}$) and N applied as fertilizer (N_F). Outputs included were soil mineral N at maize harvest ($N_{inorg H}$) and maize N uptake (N_{uptake}). Soil mineral N is the sum of NO_3^- -N and NH_4^+ -N. The unbalance term (ΔN) of equation (1) would include N mineralization – N losses by drainage leaching and by volatilization and denitrification. N mineralization includes soil, maize stover and cover crop biomass net mineralization.

$$\Delta N = N_{inorgH} + N_{uptake} - N_{inorg I} - N_F$$
^[1]

An N budget was calculated similarly for the intercrop period considering the 0 to 1.2 m soil layer. The soil mineral N after maize harvest of previous year ($N_{inorg H}$) was considered as input. Outputs included were soil mineral N before cover crop incorporation ($N_{inorg I}$) and cover crop N uptake ($N_{cc-uptake}$). The unbalance term (ΔN) of equation (2) would include N mineralization – N losses by drainage leaching and by volatilization and denitrification. N mineralization includes soil and maize stover net mineralization.

$$\Delta N = N_{inorgH} + N_{CC-uptake} - N_{inorgI}$$
^[2]

2.4 Statistical analysis

Data were analyzed using analysis of variance through the General Linear Model (GLM) procedure of the SAS 9.1 software (SAS Institute, 2004). Multiple comparisons among treatments were performed using Fisher's Protected LSD test at P = 0.05. Values of soil N_{inorg} and maize stalk NO₃⁻-N were transformed prior to analyses by the function y=log(x) to obtain homogeneity of variance. Multiple comparisons among treatments for total soil N_{inorg} in the soil profile (0 to 1.2 m) were performed using Fisher's Protected LSD test at P = 0.10.

3. RESULTS

3.1 Cover crop aboveground biomass and N uptake

Cover crop aboveground biomass in 2007 ranged from 714 to 6,993 kg ha⁻¹, and was affected by the planting method and by the cover crop treatment, but it was not affected by the interaction of the two factors studied (Table 3). The earlier planting date in DS treatment produced higher cover crop biomass compared to CS (1.7 Mg ha⁻¹ higher averaging all cover crops). Barley produced the highest cover crop biomass, which was more than double than the other cover crops. In 2008, cover crop biomass was lower in general, ranging from 427 to 3,148 kg ha⁻¹, and the interaction of the planting method and the cover crop species independently of the planting method. However, the biomass of barley was not significantly affected by the planting method. Winter rape and vetch with conventional planting had similar biomass productions than barley CS, whereas the rest of the cover crops and planting methods had significantly lower biomass productions (< 1 Mg ha⁻¹).

In 2007, N uptake of cover crops ranged from 18 to 116 kg N ha⁻¹ and was affected by the planting method and almost by the cover crop species (P=0.053). The earlier planting time with direct seeding (12 days early) allowed a greater accumulation of N in the aboveground biomass of the cover crops this year (50 kg N ha⁻¹ more). Barley had a tendency to produce the highest N uptake in 2007, followed by winter rape, vetch and oilseed rape. N uptake of cover crops in 2008 was lower than in 2007 due to the lower biomass production, and was not affected by the planting method or the cover crop species.

In 2007, N concentration in the cover crop plants was only affected by the cover crop species, whereas in 2008 was affected by the cover crop*planting method interaction. In both seasons, barley had a lower N concentration (average of 1.5%) compared to the other cover crops (average of 3.1%). Oilseed rape, winter rape and vetch had similar concentrations in 2007, whereas in 2008 winter rape CS had a lower N concentration than the other cover crops species. The C:N ratio had a similar response than N concentrations. Barley presented C:N ratios higher than 26 (average of 31) whereas the other species had values lower than 20 (average of 14).

3.2 Maize grain yield, yield components and N uptake

Maize grain yield and yield components were affected by the cover crop treatment but not by the cover crop planting method or by the interaction of the two factors. For this reason, only the cover crop treatments results are presented in Table 4. In 2007, the barley cover crop decreased the maize grain yield by 3.9 Mg ha⁻¹ compared to the control (Table 4). The other cover crop treatments produced similar maize grain yield than the control. In 2008, the barley and oilseed rape cover crops slightly decreased (\approx 1 Mg ha⁻¹) the maize grain yield compared to the control, but the winter rape and common vetch treatments produced a similar maize grain yield than the control.

Total aboveground biomass of maize was significantly reduced in the barley and winter rape treatments compared to the control (6 and 2 Mg ha⁻¹ less, respectively) during the 2007 season (Table 4). In the subsequent year the aboveground biomass was not significantly affected by the cover crop treatment. In 2007, the kernel mass and the number of grains per m² were only significantly reduced after the barley cover crop. In 2008, the kernel mass was not affected by the cover crop treatment.

In 2007, maize grain and plant N concentration were significantly lower in maize after barley compared to the control. The maize grain N concentration was higher in the vetch treatment compared to the other cover crop species. In 2008, grain N concentration was not affected by the cover crop treatment, but plant N concentration was reduced in maize after barley, oilseed rape and winter rape compared to the control. Similarly to grain N concentration, grain N uptake was reduced in maize after barley in 2007 compared to the control, and was higher in the vetch treatment compared to the other cover crop species. In 2008, grain N uptake was reduced in maize after barley in 2007 compared to the control, and was higher in the vetch treatment compared to the other cover crop species. In 2008, grain N uptake was not affected by the cover crops. Compared to the control, total N uptake (grain + plant) was greatly reduced in maize after barley in 2007 (94 kg N ha⁻¹ less than the control), and in a lesser extent in all the cover crop treatments in 2008 (20 – 40 kg N ha⁻¹ less than the control). The vetch cover crop treatment had a higher total N uptake than the other cover crop species with the exception of winter rape in 2008, which had similar total N uptake.

Corn stalk nitrate concentrations at harvest were significantly lower than the control in the barley and oilseed cover crop treatments in 2007, and in the barley, oilseed rape and winter rape cover crops treatments in 2008 (Table 4).

The SPAD readings of maize leaves were not affected by the cover crop planting method. There was a significant effect of cover crop species factor on SPAD although the results were different depending on the year and date of measurement (Table 5). SPAD values in 2007 were lower in maize after barley and winter rape at the V8 maize stage compared to the control treatment (bare soil). At the V14 stage there were no differences between treatments, whereas at flowering (R1) and maturity (R5) SPAD values in maize after barley were lower than the control treatment. In 2008, SPAD values were in general lower than the control in maize after barley, oilseed rape and winter rape at the V6 and V10 stages, but no significant differences were found at the V12 stage or later. The regression of SPAD values measured at the R1 stage with relative grain yield showed a significant relationship in 2007 (R^2 =0.71; P<0.001), but not in 2008 (R^2 =0.12; P=0.066) (Figure 1).

3.3 Soil water content

Rainfall during the intercrop period was very different in the two years (241 mm in 2006-2007 and 92 mm in 2007-2008). Soil water content in spring before incorporating the cover crops was affected by the cover crop treatment but not by the planting method (Figure 2). Cover crops reduced soil water content compared to the control in both years, as revealed by contrast of significance of control vs. cover crop treatments (P<0.05 in 2007; P<0.001 in 2008). The differences of available water with the control treatment after the cover crops in the whole soil profile (0-1.2 m depth) ranged only from 5 mm (oilseed rape) to 12 mm (barley) in 2007 due to the heavy rains prior to cover crop incorporation. In 2008, decreases in soil available water compared to the control (Figure 2a). In 2008, barley was the cover crop that removed more soil water, as deep as 0.9 m (Figure 2c) which is consistent with the higher biomass production of this cover crop (Table 3). Vetch reduced soil water content to 0.6 m soil depth in 2008, whereas winter rape reduced soil water content in the 0 to 0.3 m soil layer and oilseed rape did not affect soil water content.

At the beginning of maize growing season (around V6 stage), soil water content was similar for all treatments in the 0 to 0.3 m soil layer (data not shown). Similarly, no differences in soil water content were found at maize harvest in all the soil profile (Figure 2b and d).

3.4 Soil mineral N and N balance

Soil inorganic N in spring, before incorporating cover crops into the soil, was affected by the cover crop planting method depending on the cover crop species treatment. This interaction is explained because the oilseed rape treatment in spring 2007, and the oilseed rape and vetch in 2008, had a greater soil N depletion when the cover crops were seeded with conventional tillage than when direct seeded, probably due to a poor establishment of these cover crops when direct seeded. Because this effect of cover crops planting method in soil inorganic N content was relatively small and associated to the indirect effect on cover crop establishment, the average values of the different cover crop treatments are presented in Figure 3 and Table 6.

In 2007, barley and oilseed rape reduced significantly the soil N_{inorg} to 0.6 m depth in spring before its incorporation to the soil (Figure 3a). Common vetch reduced soil N_{inorg} in the 0.3 to 0.6 m layer. In 2008, barley reduced soil N_{inorg} significantly to the 0.6 m depth (Fig. 3c) and winter rape to 0.3 m soil depth. Soil N_{inorg} content below this depth was also on average lower when a cover crop was grown compared to the control in both years, although not significantly different. Considering the soil profile up to 1.2 m, barley reduced soil mineral N compared to bare soil in 112 and 71 kg N ha⁻¹ in 2007 and 2008, respectively (Table 6). The other cover crops reduced soil N_{inorg} to a lesser extent and not statistically significant, on average 60 kg N ha⁻¹ in 2007 and 33 kg N ha⁻¹ in 2008 (Table 6). Soil inorganic N in the 1.2 to 2.1 m layer in the second year of the experiment was not significantly different between treatments in spring. However, the soil inorganic N in this layer was on average 21 kg N ha⁻¹ lower after the barley and vetch cover crops compared to the control.

At maize harvest, the soil N_{inorg} content was similar for all the treatments (Figure 3 b and d), except for a reduced soil N content in the 0.3 to 0.6 m soil layer where cover crops were grown compared to the control in 2007 (Figure 3 b). Considering all the soil profile (0 to 1.2 m depth), soil inorganic N at maize harvest was lower in maize after barley in both years

compared to the control (Table 6), on average 104 and 28 kg N ha⁻¹ lower in 2007 and 2008, respectively. In maize after the other cover crops, soil N_{inorg} had a tendency to be lower compared to the control, but this reduction was not statistically significant. Soil N_{inorg} content at maize harvest in the 1.2 to 2.1 m soil depth was not affected by the cover crop treatments (Table 6).

During the first intercrop period there was a positive N balance (net N mineralization – N losses) ranging from 26 to 63 kg/ha (Table 7) and without significant differences between the cover crop treatments. However, during the second intercrop period there was a negative N balance (N immobilization or N lost), but similarly no statistical differences were found between the cover crops treatments. During the first maize crop period only the vetch treatment showed a significant positive balance, while the other treatments showed a negative one or close to zero (oilseed rape). During the second maize crop period there was a slight negative balance in the control treatment while there was a small positive balance when a cover crop was grown (except in oilseed rape).

4. DISCUSSION

4.1 Cover crop growth as affected by planting method and species

Growing a cover crop after maize harvest is not common in the irrigated areas of the Ebro River Basin and other semiarid irrigated areas in Spain due to the limited information available about how to manage the cover crops. In one of the two years all cover crops studied produced biomass and aboveground N content in the high range of those reported under similar (Gabriel and Quemada, 2011) and other conditions (Stenberg et al., 1999; Thomsen, 2005; Kramberger et al., 2009; Maltas et al., 2009). The relatively long maize intercrop period from October - November to March enabled the cover crops to grow significantly and without high supplemental irrigation (40 to 51 mm), making this practice a possible option in the study area.

Direct seeding of the cover crops enabled to plant them 8 to12 days earlier compared to seeding after conventional tillage operations. Earlier planting dates have been reported to

increase cover crop growth and N uptake (Stenberg, 1998). This was the case in the first year of the experiment when direct seeding allowed earlier planting time and resulted in higher cover crop biomass productions and N uptake. However, direct seeding had a detrimental effect on common vetch, oilseed rape and winter rape in 2008, which resulted in a poorer establishment compared to conventional seeding. This could be explained by a poorer emergence due to the maize stover residue which mechanically hampered plant emergence of these species. Optimum planting date for *Brassica* species and vetch in the area when grown as cash crops is one month earlier than the planting date used in the experiment, and this could also explain the poor growth of these crops in 2008. Barley was the cover crop that produced more biomass, probably due to the fact that planting date was optimal for this species in the area.

Although barley ensured a good plant establishment and high biomass it always resulted in lower biomass N concentration and higher C:N ratio compared to the other cover crop species. This could be partially due to the dilution effect of the higher biomass production of barley. The high C:N ratio in barley (ranging from 26 to 39) increases the risk of N immobilization processes when it is incorporated to the soil, as C:N ratios above 25 have been related to N immobilization (Ranells and Wagger, 1996; Kaye and Hart, 1997; Kuo and Jellum, 2000). Oilseed rape, winter rape and vetch had a lower C:N ratio (ranging from 11 to 20), but could only produce significant biomass and N accumulation provided there is a good crop establishment.

The cover crop planting method had no significant effect on soil N dynamics and maize grain yield response and, therefore, only the effect of the cover crop factor is discussed in the subsequent discussion sections.

4.2 Cover crop effect on soil water content and N dynamics.

The lower soil water content in spring after the cover crops compared to bare soil was the result of cover crop transpiration. In the first year of the experiment, the high rainfall during cover crop growing season (241 mm), with high precipitation events in spring close to the time of cover crop incorporation, was the reason for the similar soil water content of the cover crop treatments compared to the control (bare soil). However, in the second year the reduction of soil water content due to the cover crop transpiration was higher due to the lower rainfall during all cover crop growing season (92 mm). Even though the irrigation applied to the maize crop was the same in all cover crop treatments, the similar soil water content at the V6 maize stage and at harvest in all treatments indicates that the cover crop growth did not reduce significantly the available water for the maize crop, as soil water differences were small and disappeared with the start of irrigation.

Cover crops reduced significantly soil N_{inorg} content in spring compared to bare soil up to 0.6 m soil depth, and soil N below this layer also had a tendency to be lower after a cover crop. Deeper soil N depletions were expected for the *Brassica* species, as previously reported (Thorup-Kristensen, 2001). However, the poor establishment of the *Brassica* crops and the late planting date could explain the relatively lower soil N depletion compared to barley. The highest soil N depletions observed after barley can be explained by the higher biomass production and high N uptake in this cover crop. These soil N reductions can avoid N leaching during the intercrop period with occasional heavy precipitations that can occur in semiarid conditions during this time, such as in March-April 2007 (184 mm). Furthermore, cover crop transpiration resulted in lower soil water content in early spring, which will reduce drainage and the associated N leaching risks during the first irrigation events in the next maize crop (Salmerón et al., 2010).

At maize harvest, the reduction of soil inorganic N content observed after the cover crop treatments reduces N leaching risks during the next intercrop period. Deep placed soil N_{inorg} is more likely to be moved below the next crop root depth and therefore lost by leaching. Subsoil N content (below 1 m) has proved useful to indicate differences among treatments in N leaching losses under more humid conditions (Thorup-Kristensen et al., 2009). However, no differences were found below 1.2 m soil depth in our experiment. Recent studies (Salmerón et al., 2010) under irrigated conditions similar to this experiment found that most N leaching occurs during maize growing season with the first irrigations, which agrees with the increase of nitrate lost in drainage water observed after side-dress N applications in watershed studies in the same area (Causapé et al., 2004; Isidoro et al., 2006). Consequently, the reduction in soil N_{inorg} content in spring before the start of irrigation could be the most limiting factor determining N leaching

losses under irrigated conditions. A cover crop that depletes residual soil N after maize harvest and a proper N fertilization management to the subsequent maize crop could be an efficient way to reduce N leaching while not compromising water and N requirements to maize.

4.3 Cover crop effect on maize yield

Maize grain yield reductions were observed in the barley cover crop treatment in 2007 (decrease \approx 4 Mg ha⁻¹) and in a lesser extent in the barley and oilseed rape cover crop treatments in 2008 (decrease \approx 1 Mg ha⁻¹). In all the other cases, the cover crop biomass incorporated into the soil, the soil inorganic N content after the cover crop growth and the reduced N fertilizer applied (250 kg N ha⁻¹) were sufficient to fulfill maize N requirements similarly to the control supplied with an extra 50 kg N ha⁻¹. Therefore, winter rape, vetch and in the first year oilseed rape, proved to be efficient in reducing soil N content compared to the control and the associated N leaching risks, while maintaining maize crop yield with lower fertilizer N inputs.

The decrease of maize yield found after barley in both years and oilseed rape in 2008 can be explained by an N deficiency of maize plants, as indicated by the lower SPAD values, the lower maize N uptake, and the lower end of season maize stalk nitrate concentrations in these treatments compared to the control. The higher decrease of maize yield after the barley cover crop treatment in 2007 was related with a stronger N deficiency.

N deficiency can occur if N release from organic sources is not synchronized with the N demand by the crop (Magdoff, 1991; Wyland et al., 1995; Cavero et al., 1997; Clark et al., 2007a). A timed N release is especially relevant in crops with determinate growth habit and that have a high N demand in a short period of time such as maize (Magdoff, 1991; Salmerón et al., 2010). The maize N deficiency after the barley cover crop was probably due to the high C:N ratio of the barley biomass, that caused N immobilization (clearly shown in the 2007 N balance) and therefore a low availability of this N during the maize growing season. Incorporation of cover crops or other plant residues with high C:N ratio has been reported to decrease soil N inorganic and reduce N availability to maize (Baggs et al., 2000; Sainju et al., 2005; Clark et al., 2007a; Starovoytov et al., 2010). Some works have found that the incorporation of cover crop

residues with C:N ratios above 25 result in N immobilization (Ranells and Wagger, 1996; Kaye and Hart, 1997). An earlier incorporation of the barley cover crop residue could have reduced the C:N ratio in the cover crop biomass (Clark et al., 2007a). Moreover, the results suggest that a cereal-legume biculture could be an efficient combination to obtain a cover crop residue with a higher N concentration (Ranells and Wagger, 1997; Sainju et al., 2005; Clark et al., 2007a), which would be interesting to study in our area. The maize yield reduction after the oilseed rape cover crop in 2008 could be explained by the low biomass produced and therefore the small amount of N mineralized from the cover crop as shown in the N balance. N immobilization is not likely to be the cause for the N deficiency in these treatment, as C:N ratio in this cover crop was below the proposed threshold for immobilization of 25 (Ranells and Wagger, 1996; Kaye and Hart, 1997; Kuo and Jellum, 2000). High N availability after vetch was clearly shown as N net mineralization during the maize crop occurred both years, which resulted in maize yield similar to the control.

Our results agree with previous works where maize yield after cover crops was greatly dependent on the quality of the cover crop residue (C:N ratio) incorporated into the soil (Starovoytov et al., 2010). High N availability after legume cover crops, and reduced N availability after cereal cover crops have been reported (Ranells and Wagger, 1996; Baggs et al., 2000; Clark et al., 2007a,b; Starovoytov et al., 2010). High precipitations during winter and spring as well as irrigation management will likely affect the cover crop effect on soil N availability as compared to a control with bare soil during winter. Therefore, N recommendations to maize based merely on cover crop N content incorporated into the soil and/or based on soil N content after a cover crop will more likely fail to give optimum maize yields. Cover crop quality, soil N_{inorg} and climate and irrigation conditions should be taken into account, what can be difficult under field conditions.

N fertilizer recommendation tools that allow in season N fertilizer applications could be useful to adjust N fertilizer rates to maize after cover crops. SPAD measurements in maize leaves can indicate N deficiencies when compared with a well fertilized area (Varvel et al., 2007). SPAD values were able to detect maize N deficiencies early in the season both years which resulted in yield reduction both years. When the N deficiency was more important, the SPAD values were lower at later maize stages (R1 and R5) and consequently the yield decrease was more important (\approx 4 Mg ha⁻¹). This tool has been found useful to show N status in maize after cover crops (Miguez and Bollero, 2006). Therefore, the use of SPAD can be a valuable tool when using cover crops because of the uncertainty of N availability due the mineralization-immobilization processes of cover crops residue.

Maize stalk nitrate concentration at maize harvest is an end-of-season diagnostic test of N fertilizer management (Binford et al., 1992; Blackmer et al., 1994; Hooker et al., 1999). In our work the maize stalk nitrate was efficient detecting differences between treatments and was well related to maize yield. Thus, this tool was able to detect N deficiency in the barley treatment in 2007, with NO₃-N contents (35 mg kg⁻¹) much lower than the threshold of 250 mg kg⁻¹ for N deficiency proposed by Binford et al. (1992). In the other treatments and years the stalk nitrate concentrations were within the range for sufficiency (300 to 1,800 mg kg⁻¹ of NO₃⁻-N) to achieve maximum or nearmaximum yield. In all cases, maize stalk nitrate concentrations were below 1,800 mg kg⁻¹ of NO₃⁻-N which indicates that maize was not over fertilized.

5. CONCLUSIONS

Direct seeding of cover crops allowed an earlier planting both years and resulted in greater biomass production and N uptake in the first year, but not in the second year due to a poor establishment of the non-cereal cover crops. Thus, if cover crops establishment problems are anticipated, the use of direct seeding is not recommended for oilseed rape, winter rape and vetch, but it is recommended for barley.

Barley was the cover crop that produced higher biomass although the N uptake was not statistically different from the other cover crops species. Besides, the C:N ratio was higher in barley.

Cover crop treatments reduced soil inorganic N in spring and at maize harvest, reducing the N leaching risk.

Growing cover crops during the intercrop period and reducing the N fertilizer applied to maize by 50 kg N ha⁻¹ did not affect the maize yield in the case of winter rape and vetch, and slightly reduced the maize yield (-7%) in the case of oilseed rape in one of the years, However, when barley was grown as cover crop the maize yield was reduced by 1 to 4 Mg ha⁻¹ (-6% and -25%, respectively) because of maize N deficiency caused by low N availability due to insufficient N mineralization and/or lack of synchronization with maize N uptake.

SPAD measurements in maize leaves were useful to detect N deficiency in maize after cover crops.

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Depth	рН	С	Ν	CaCO ₃	Sand	Silt	Clay	FC [†]	WP
m				%	, 0			m ³	m ⁻³
0.0 - 0.3	8.4	0.86	0.110	30.9	26.5	45.4	28.1	0.351	0.197
0.3 - 0.6	8.4	0.72	0.102	31.6	24.0	46.9	29.1	0.351	0.217
0.6 - 0.9	8.4	0.44	0.088	30.7	17.4	50.0	32.6	0.344	0.196
0.9 - 1.2	8.6	0.38	0.075	30.3	19.1	50.3	30.6	0.329	0.171

Table 1. Soil characteristics of the experimental field.

FC: Field capacity (-0.033 MPa); WP: Wilting point (-1.5 MPa)

Operation	Da	ate
	2007	2008
Cover crop		
Planting (DS)	3 Nov. 2006	30 Oct. 2007
Planting (CT)	15 Nov 2006	7 Nov. 2007
Incorporation	12 Apr. 2007	7 Apr. 2008
Maize	·	·
Planting	8 May 2007	25 Apr. 2008
Harvest	23 Oct. 2007	24 Oct. 2008

Table 2. Dates of cover crop and maize planting, cover crop incorporation and maize harvest.

DS: cover crop direct seeded CS: cover crop seeded after conventional tillage operations

Table 3. Cover crop aboveground biomass, N uptake and concentration, and C/N ratio before its incorporation into the soil in spring depending on the planting method and the cover crop species during the two years of the experiment.

					Ν		_	
Factor	Bio	mass	Upt	ake	Conce	ntration	C:I	N ratio
	2007	2008	2007	2008	2007	2008	2007	2008
		kg h	a ⁻¹		0	%		
Planting method (P								
value)	0.018	NS	0.043	NS	NS	NS	NS	NS
Direct seeding (DS) Conventional	3854 a	1208	101 a	34	2.90	2.36	16.7	22.8
seeding (CS)	2104 b	1557	49 b	35	2.58	2.40	17.4	22.1
Cover crop species								
(P value)	0.001	0.001	0.053	NS	0.001	0.001	0.001	0.001
Barley	5555 a	2668	93	35	1.68 b	1.26	27.1 a	35.8
Oilseed rape	1962 b	482	61	21	3.08 a	3.00	12.8 b	13.5
Winter rape	1765 b	1339	80	32	3.44 a	2.47	13.5 b	17.6
Vetch	2228 b	1041	72	52	3.14 a	3.44	13.0 b	12.6
Planting method *								
Cover crop species								
(P value)	NS	0.031	NS	NS	NS	0.015	NS	0.017
Barley DS	6993	3148 a	116	45	1.65	1.42 d	28.0	32.8 b
Barley CS	4116	2187 ab	70	24	1.71	1.09 d	26.1	38.8 a
Oilseed rape DS	2315	427 d	77	-	3.28	-	11.2	-
Oilseed rape CS	1609	536 cd	46	17	2.88	3.00 b	14.4	13.5 d
Winter rape DS	2416	715 cd	87	22	2.94	3.42 a	16.3	11.7 e
Winter rape CS	714	1963 b	18	45	3.57	1.99 c	11.6	20.6 c
Vetch DS	2956	540 cd	102	42	3.44	3.23 a	12.2	13.4 de
Vetch CS	1501	1542 bc	43	56	2.83	3.59 a	13.7	12.1 de

DS: cover crop direct seeded

CS: cover crop seeded after conventional tillage operations

Table 4. Maize grain yield (at 140 g kg⁻¹ moisture content), total aboveground biomass, kernel mass (KM), grain number per m², N concentration in grain and plant, N uptake in grain and plant and stalk NO₃-N, for each cover crop species and year. Values followed by the same letter are not significantly different (P>0.05). The planting method and the interaction of planting method and cover crop species factor were not significant (P>0.05).

Cover crop species	Grain	Total biomass	KM	Grain number	N conce	entration	N up	otake	Stalk NO₃⁻-N
•					Grain	Plant	Grain	Total	
	Mg	ha ⁻¹	g unit⁻¹	nº m⁻²	9	%	kg	ha ⁻¹	mg kg⁻¹
					2007				
Control	15.6 a	26.7 a	0.373 a	3709 a	1.32 ab	0.70 a	181 ab	272 ab	1081 a
Barley	11.7 b	20.7 c	0.337 b	3048 b	1.16 c	0.54 b	121 c	178 c	35 c
Oilseed rape	15.1 a	25.5 ab	0.389 a	3444 a	1.29 b	0.76 a	172 b	265 ab	500 b
Winter rape	14.6 a	24.7 b	0.374 a	3459 a	1.25 bc	0.65 ab	162 b	240 b	761 ab
Vetch	15.7 a	26.7 a	0.391 a	3560 a	1.42 a	0.68 a	197 a	285 a	1071 a
P value	0.001	0.001	0.003	0.006	0.003	0.020	0.001	<0.0001	0.001
					2008				
Control	14.6 a	24.1	0.371	4469 a	1.43	0.83 a	183	277 a	1328 a
Barley	13.7 bc	22.2	0.381	4081 b	1.33	0.70 bc	161	232 c	469 c
Oilseed rape	13.6 c	21.8	0.371	4148 b	1.36	0.70 bc	163	232 c	808 bc
Winter rape	14.0 abc	22.4	0.373	4252 ab	1.42	0.68 c	174	244 bc	794 bc
Vetch	14.4 ab	22.9	0.393	4167 b	1.39	0.77 ab	176	255 b	984 ab
P value	0.035	NS	NS	0.018	NS	0.014	NS	0.002	0.001

Table 5. Average SPAD values of maize leaves in the different cover crop species treatments during the maize growing season in 2007 and 2008. Values followed by the same letter are not

2 3 4 significantly different (P>0.05). The planting method and the interaction of planting method and cover crop species factor did not affect the SPAD variable.

Cover crop sp	ecies				SPAD		
					2007		
		V8			V14	R1	R5
Control		50.7 ab			49.7	57.4 a	56.4 a
Barley		42.4 d			47.7	54.8 b	46.9 b
Oilseed rape		49.0 bc			48.9	57.5 a	55.6 a
Winter rape		48.1 c			49.6	56.8 a	54.2 a
Vetch		52.5 a			49.0	57.7 a	55.5 a
	P value	0.001			NS	0.001	0.001
					2008		
		V6	V10	V12	V15	R1	R5
Control		41.4 a	52.5 a	51.4	44.1	57.7	57.9
Barley		37.3 c	50.8 c	52.1	44.7	57.8	55.9
Oilseed rape		40.1 ab	51.3 bc	50.6	44.0	56.5	56.0
Winter rape		39.7 b	51.9 ab	51.6	44.6	58.2	56.1
Vetch		41.0 ab	52.0 a	52.2	44.7	57.3	57.0
	P value	0.001	0.001	NS	NS	NS	NS

V6: 6 leaves stage; V8: 8 leaves; V10: 10 leaves; V12: 12 leaves; V14: 14 leaves; R1: silking; 7 8 R5: dent.

 $\overline{28}$

2 3

Table 6. Soil inorganic N content (N_{inorg}) measured before incorporating the cover crops in spring and after maize harvest in the different soil layers depending on the cover crop species

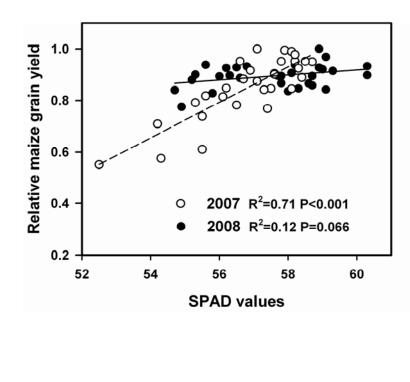
treatment.

			Soil N	V inorg		
Cover crop species	Before inc	orporation o	of cover crops	Afte	er maize har	vest
	2007	2	2008	2007	2	008
-	0-1.2 m	0-1.2 m	1.2-2.1 m	0-1.2 m	0-1.2 m	1.2-2.1m
			kg l	าa⁻¹		
Control	202 a	105 a	79	202 a	66 a	52
Barley	90 b	34 b	57	98 b	38 b	29
Oilseed rape	134 ab	73 a	62	128 ab	46 ab	41
Winter rape	157 a	73 a	73	119 ab	50 ab	49
Vetch	131 ab	70 a	59	160 ab	57 a	45
P value	0.057	0.006	NS	0.093	0.067	NS

- **Table 7**. N balance during the maize intercrop period and cropping season depending on crop
- 2 cover species. AN represents the unbalance term of the balance (net N mineralization N
- 3 losses).

Cover crop species Int Control Barley Oilseed rape Winter rape Vetch <i>P value</i>	63 26 31 33 45	Cropping season kg ha ⁻¹ 2007 -28 a -63 a 9 ab -40 a
Barley Oilseed rape Winter rape Vetch	63 26 31 33	2007 -28 a -63 a 9 ab -40 a
Barley Oilseed rape Winter rape Vetch	63 26 31 33	2007 -28 a -63 a 9 ab -40 a
Barley Oilseed rape Winter rape Vetch	26 31 33	-63 a 9 ab -40 a
Oilseed rape Winter rape Vetch	31 33	9 ab -40 a
Winter rape Vetch	33	-40 a
Vetch		
	45	
P value		63 b
	NS	0.036
		2008
Control	-98	-10
Barley	-29	15
Oilseed rape	-41	-3
Winter rape	-14	20
Vetch	-56	36
P value	NS	NS

- **Figure 1.** Relationship between relative maize grain yield and SPAD values of maize
- leaves at silking (R1) in 2007 and 2008. For each year, data from all treatments were
 pooled (n=30).



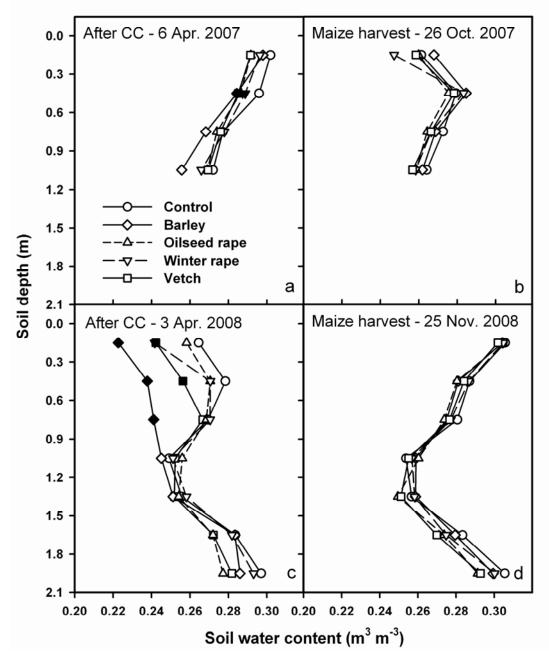




Figure 2. Soil water content in spring before the cover crops (CC) were incorporated (a and c) and in fall after maize harvest (b and d) in the two years of the experiment. The filled black symbols indicate significant differences compared to the control treatment within each soil depth at P<0.05 after ANOVA.

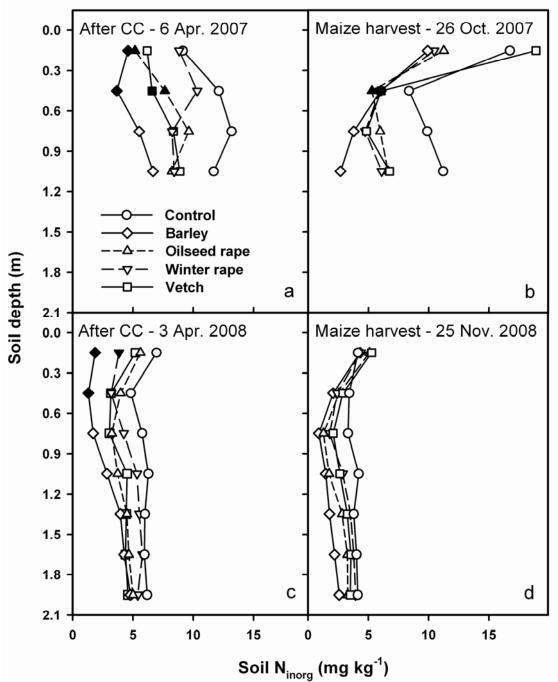


Figure 3. Soil mineral N (Ninorg) content in spring before the cover crops (CC) were 6 incorporated (a and c) and in fall after maize harvest (b and d) in the two years of the 7 experiment. The filled black symbols indicate significant differences compared to the . 8 9 control treatment within each soil depth at P<0.05 after ANOVA.

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