



Impact of tillage and N fertilization rate on soil N₂O emissions in irrigated maize in a Mediterranean agroecosystem



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ABSTRACT

In irrigated Mediterranean conditions there is a lack of knowledge about the best combination of tillage and N fertilization practices to reduce soil nitrous oxide (N₂O) emissions while maintaining maize productivity. The aim of this work was to investigate the effects of different soil management practices and synthetic N fertilization rates on soil N₂O emissions and their relationship with maize grain yield to determine the best management system to reduce yield-scaled N₂O emissions (YSNE) in a semiarid area recently converted to irrigation under Mediterranean conditions. A long-term tillage and N rate field experiment established in 1996 under barley (*Hordeum vulgare* L.) rainfed conditions, was converted to irrigated maize (*Zea mays* L.) in 2015. After the transformation to irrigation, the field experiment maintained the same tillage treatments and N fertilization rates. Three types of tillage (conventional tillage, CT; reduced tillage, RT; no-tillage, NT) and three mineral N fertilization rates (0, 200, 400 kg N ha⁻¹) were compared during three years (2015–2017) in a randomized block design with three replications. Soil N₂O emissions, water-filled pore space, soil temperature, mineral N content (as NH₄⁺ and NO₃⁻), denitrification potential and maize grain yield and above-ground N uptake were quantified. Moreover, the emission factor (EF) and YSNE were calculated. The results showed that the combination of NT and the highest rate of N fertilization led to greater N₂O emissions. Furthermore, the lowest N₂O fluxes were observed in CT when WFPS was below 40% and the highest N₂O fluxes were seen in NT when WFPS was above 60% coinciding with the greatest denitrification potential. Cumulative N₂O emissions in 2017 and 2015 followed the order 400 > 200 > 0 kg N ha⁻¹, while in 2016, rate of 400 and 200 kg N ha⁻¹ showed greater cumulative N₂O emission compared to the control. Only RT showed differences between growing seasons on cumulative N₂O emissions, with greater values in 2017 compared to 2015, and intermediate values in 2016. In all treatments, the N₂O EF was much lower than the default IPCC emission factor (1%). NT and RT increased the grain production compared to CT which was affected by severe soil crusting causing water deficit. Likewise, N fertilizer treatments significantly affected the YSNE, increasing with increasing fertilizer N application rate in the first year of study. Our data show that the use of NT or RT does not lead to more yield-scaled N₂O emissions than CT in Mediterranean agroecosystems recently converted to irrigation.

1. Introduction

Mediterranean climate is characterized by high evapotranspiration, relatively mild temperatures in winter and summer drought. Precipitation is highly variable, becoming deficient in some areas of the Mediterranean, leading to yield constraints. Consequently, rainfed areas are increasingly being converted to irrigation to stabilise or increase yields of traditional crops such as wheat or barley or to allow the establishment of more water demanding crops such as maize, alfalfa or fruit trees. Apart from an increase in crop yield, this conversion to

irrigated land also generates an increase in nitrogen fertilizer use which, if not adapted to the needs of the crop, can lead to adverse environmental impacts such as N₂O emitted to the atmosphere (Smith et al., 2008), soil nitrate leached (Quemada et al., 2013), or ammonia gas volatilized (Erisman et al., 2007). Irrigation increases soil water availability, which in combination with elevated temperatures, induces better conditions for biological activity, favouring denitrification. It is assumed that denitrification becomes the dominant mechanism when soil water-filled pore space is above 60%; due to low oxygen availability, rapidly increasing the rate of emission of N₂O (Skiba and Ball,

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2002).

In Mediterranean irrigated conditions, summer crops such as maize can have high productivity, which leads to significant requirements for N. The application of high rates of irrigation water combined with high rates of N offers an elevated potential for the formation of N₂O (Ellert and Janzen, 2008). Mineral N availability is a key process controlling soil N₂O emissions. An excess of mineral N accompanied by high N fertilizer rates increases soil mineral N losses as N₂O through higher nitrification and denitrification rates (Chantigny et al., 1998), increasing the EF. Authors such as Ma et al. (2010) and Hoben et al. (2011) have reported that an increase in N fertilization rates leads to higher N₂O emissions in maize. In the Mediterranean area, different studies have provided similar EF for maize production under sprinkler irrigation to the current IPCC default of 1% (Aguilera et al., 2013; Cayuela et al., 2017). However, in these previous works the impact of other management practices such as tillage on soil N₂O emissions was not elucidated.

Different to N fertilization, the impact of tillage on soil N₂O emissions is highly variable (Gregorich et al., 2008). The effects of conservation tillage on N₂O emissions depend on soil properties, climate conditions, and the number years since conservation tillage was implemented (van Kessel et al., 2013). Six et al. (2004) suggested that the emission of N₂O could be reduced when maintaining NT over time, as a result of an improvement in soil structure and porosity, thus reducing the formation of anaerobic microsites. For instance, use of NT is a means to conserve water and reduce soil organic matter losses compared with CT, and usually increases bulk density (Lampurlanés and Cantero-Martínez, 2003). This increase in bulk density reduces gas diffusivity, which combined with an increase in surface soil moisture, stimulates the probability of anaerobic conditions, favouring denitrification and N₂O fluxes (Mosier et al., 2002). On the other hand, long-term use of NT can improve soil structure (Pareja-Sánchez et al., 2017) and lower soil temperature, which in turn can reduce N₂O emissions relative to CT (Grandy et al., 2006). In past studies reporting tillage effects on N₂O emissions, several authors found greater fluxes under NT compared with CT (Baggs et al., 2003; Ball et al., 2008). However, others reported higher fluxes under CT (Elder and Lal, 2008). These differences between studies may be attributed to soil properties, climate conditions or the number of years under each treatment.

There is a need to identify practices that minimize net greenhouse gas emissions (Follett et al., 2005) while meeting agricultural production. Therefore, a good indicator of the performance of a cropping system in terms of productivity and environmental impact is the yield-scaled N₂O emissions (YSNE). This indicator is proposed as a metric of the important global challenge of ensuring food security whilst reducing N₂O emissions (Van Groenigen et al., 2010).

Over the last three decades, in the Mediterranean rainfed area of the Ebro Valley, NE Spain, RT and NT systems have been introduced with the purpose of mitigating soil erosion as well as for reducing production costs (Moreno et al., 2010). However, as in many arid and semiarid regions, rainfed areas are being converted to irrigation and changing to new more productive crops such as maize, which require more nitrogen input than the traditional winter cereal production. Nevertheless, in these newly irrigated areas, farmers are returning to adopt intensive tillage systems, which are common in irrigation production. The limited knowledge about the correct use of RT or NT systems in irrigated land, including their interactive effects with N fertilization, makes their adoption by farmers difficult and compromises the soil quality benefits attained with long-term NT use.

Different studies have focused on N fertilization strategies in irrigated maize under Mediterranean conditions (e.g. Martínez et al., 2017; Berenguer et al., 2009). However, to our knowledge none of them have tested the performance of conservation tillage and its interaction with N fertilization on irrigated maize productivity. Moreover, as far as we know, there are no studies that have investigated the interactions of fertilizer N rates and tillage practices on yield-scaled N₂O emissions in

maize production in irrigated Mediterranean conditions. Our main hypotheses were that i) reducing N fertilizer rate in combination with a decrease of tillage intensity, would reduce N₂O emissions, while ii) the possible greater N₂O emissions under NT would be compensated by greater grain yield. Therefore, according to that hypothesis, the objectives of the present study were to investigate the effects of different tillage systems and N application rates on maize grain yields and N₂O emissions and to determine the best combination to reduce YSNE.

2. Materials and methods

2.1. Site description and experimental design

The study was carried out in Agramunt, NE Spain (41°48' N, 1°07' E, 330 m asl). The climate is semiarid Mediterranean with a mean annual precipitation of 401 mm and potential evapotranspiration (PET) of 855 mm, (1984–2014). Mean annual air temperature is 14.1 °C.

A long-term field experiment was established in 1996 to compare three tillage systems (CT, RT and NT) and three increasing rates of mineral N fertilizer (0, 60 and 120 kg N ha⁻¹) under rainfed barley monoculture (Angás et al., 2006). In 2015 the experimental field was converted to irrigation with solid set sprinklers of 18 × 18 m spacing. Three successive maize growing seasons (2015–2017) were studied, corresponding to the typical irrigated cropping system in the area. After the conversion to irrigation, the field experiment maintained the same tillage treatments (CT, RT and NT) while N fertilization rates were adapted to maize (0, 200, 400 kg N ha⁻¹). Traditionally, farmer in the area apply N fertilizer rates ranging between 300 and 450 kg ha⁻¹ (Sisquella et al., 2004). Therefore, in our study the rate of 400 kg N ha⁻¹ reflects the typical scenarios used by some farmers and the medium rate (200 kg N ha⁻¹) aims to determine that N fertilizer application can be reduced by half to achieve optimal yields and reduce the environmental impact. The experiments were laid out in a randomized block design with three replications and plot size of 50 × 6 m. Site characteristics and soil properties are detailed in Table 1. The CT treatment consisted of one pass of rototiller (15 cm depth) followed by one pass of subsoiler (35 cm depth) and one pass of a disk plough (20 cm depth) before planting during March or April with almost 100% of the crop residues incorporated into the soil before planting. This tillage system represents the traditional practice for maize production in the area. The RT treatment consisted of one pass of a strip-till implement on the maize planting row to 25 cm depth reducing the surface tilled to 20%. Finally, NT consisted of a total herbicide application

Table 1

Soil characteristics of Ap horizon (0–28 cm depth) in 1996. Initial soil organic carbon content (SOC_i) (1996) and soil organic carbon content (SOC) (0–30 cm) in three tillage systems (conventional tillage, CT; reduced tillage, RT; no-tillage, NT) in 2015.

Soil characteristic	
Soil classification ^a	Typic Xerofluvent
pH (H ₂ O, 1:2.5)	8.5
EC _{1:5} (dS m ⁻¹)	0.15
P Olsen (ppm)	35
K Amm. Ac. (ppm)	194
Water retention (-33 kPa) (g g ⁻¹)	16
Water retention (-1500 kPa) (g g ⁻¹)	5
SOC _i (g kg ⁻¹)	7.6
Sand (%)	30.8
Silt (%)	57.3
Clay (%)	11.9
SOC (g kg ⁻¹)	
CT	7
RT	9
NT	9

^a According to the USDA classification (Soil Survey Staff, 2014).

(1.5 L ha⁻¹, 36% glyphosate) without soil disturbance. Planting was carried out with a pneumatic row direct drilling machine equipped with double disc furrow openers (model Prosem K, Solà, Calaf, Spain). The planting depth was adapted to each tillage system. Rotary residue row cleaners were installed to clear the path for the row unit openers. The N fertilizer rates were split in one pre-planting application with urea (46% N) in April, which was surface broadcasted and incorporated with tillage in CT and RT, with 50 kg N ha⁻¹ applied in the one splits in the 200 kg N ha⁻¹ rate being doubled in the 400 kg N ha⁻¹ rate. Afterwards, two top-dressing applications were carried out by broadcasting calcium ammonium nitrate (27% N), in May and July (V5 and V10 stages, respectively) with 75 and 75 kg N ha⁻¹ applied, respectively, in the two splits in the 200 kg N ha⁻¹ rate, being doubled in the 400 kg N ha⁻¹ rate. Mineral P and K fertilization was applied prior to maize planting based on soil analysis at rates of 154 kg ha⁻¹ P₂O₅ and 322 kg K₂O ha⁻¹, respectively, in the first two years. In the third year the levels of available P and K in the soil were appropriate for the crop, making unnecessary further P and K applications. In the three years maize (cv. Kopias) was planted late April at a rate of 90,000 seeds ha⁻¹ with a 73 cm width between rows. Irrigation began in April and ended in September being supplied to meet the estimated evapotranspiration of the crop (ET_c) minus the effective precipitation, which was estimated as 75% of precipitation (for any precipitation > 5 mm) (Dastane, 1978). Weekly ET_c was calculated from the corresponding values of PET and the crop coefficient (K_c). Potential evapotranspiration was computed with the FAO Penman–Monteith method from meteorological data obtained from an automated weather station located near the experimental site. Crop coefficients (K_c) were estimated based on crop development, ranging between 0.3 and 1.2. Irrigation was carried out every 3 to 4 days when crop evapotranspiration was lower (April, May, June and September) and with a daily frequency in July and August, when the crop water needs were higher. Harvesting was done at the beginning of November with a commercial combine. Afterwards, crop residues were chopped and spread over the soil. During the periods between crops in winter the soil was maintained free of weeds with an application of glyphosate at 1.5 L ha⁻¹.

2.2. Soil N₂O emissions and denitrification potential

During the three years studied, the emission of N₂O from soil was measured with the non-steady-state chamber method (Hutchinson and Mosier, 1981), using the same chambers described by Plaza-Bonilla et al. (2014). Two polyvinylchloride rings (31.5 cm internal diameter) were inserted into the soil to a depth of 5 cm. Chambers of 20-cm height were constructed with same material. A metal fitting was attached in the center of the top of the chamber and was lined with two silicon-Teflon septa as sampling port. To reduce internal temperature fluctuations the chambers were covered with a reflective insulation layer (model Aislatermic, Arelux, Zaragoza, Spain). Soil N₂O fluxes were measured in two observations per plot, with weekly measurements during the growing season (April to November), greater measurement intensity during fertilizer applications (i.e. 24 h. prior and 3 h., 24 h. and 48 h. after) and measured every 21 days in the periods between crops in winter (November to March). Gas samples were taken at 0, 20 and 40 min after the closure of the chamber and stored into 15 mL Exetainer® borosilicate vials (model 038 W, Labco, High Wycombe, UK). Samples were subsequently analyzed by a gas chromatography system (7890A, Agilent, Santa Clara, CA, United States) equipped with an electrical conductivity detector (ECD) and an HP-Plot Q column (30 m long, 0.32 mm of section and 20 μm) with a pre-column 15 m long of the same characteristics. The injector and oven temperatures were set to 50 °C. The temperature of the detector was set to 300 °C, using a 5% methane in Argon gas mixture as a make-up gas at a flow of 30 mL min⁻¹. The system was calibrated using analytical grade standards (Carbueros Metálicos, Barcelona, Spain). Gas fluxes were calculated taking into account the linear increase in the N₂O concentration

inside the chamber with time (40 min) and correcting the values for air temperature.

Soil denitrification potential was determined 5 days after the three fertilizer applications of the second maize season (2016) by quantifying the activity of denitrifying enzyme as described by Groffman et al. (1999). 25 g of fresh soil and 25 mL of a solution containing 1 M glucose, 1 nM KNO₃ and 1 g L⁻¹ chloramphenicol were added into 125 mL hermetic glass jars. The jars were sealed and repeatedly flushed with N₂ for 2 min in order to create anaerobic conditions. Afterwards, acetylene 5% was added to the jars to determine denitrification potential (Estavillo et al., 2002). The jars were incubated in an orbital shaker at room temperature. After incubation at 30 and 90 min, 15 ml gas samples were removed from the jar headspace using a syringe and then stored in vials. Sample N₂O concentration was analyzed by gas chromatography as described above.

2.3. Soil sampling and plant analysis

At the same sampling dates as soil N₂O emissions measurements, soil samples (0–5 cm depth) were obtained for mineral N (as ammonium, NH₄⁺, and nitrate, NO₃⁻) and gravimetric moisture determination in two observations per plot. Soil temperature (10 cm depth) was measured using a handheld probe (TM65, Crison). Soil gravimetric moisture was transformed into water-filled pore space (WFPS) using soil bulk density (BD), which was measured monthly at two positions per plot, and assuming a theoretical particle density of 2.65 g cm⁻³. Soil NH₄⁺ and NO₃⁻ contents were quantified by extracting 50 g of fresh soil with 100 mL of 1 M KCl. The extracts were analyzed with a continuous flow autoanalyzer (Seal Autoanalyzer 3, Seal Analytical, Norderstedt, Germany).

At harvest, maize above-ground biomass and grain yield were measured by collecting plant samples of two central rows 2–5 m long, depending on plant density, in three sampling areas per plot. The number of plants and ears was counted and registered. Afterwards, a sub-sample of two entire plants and five ears were taken to determine the yield components and moisture. The sub-sample was oven-dried at 60 °C for 48 h and weighed. Next, the grain was threshed and weighed. Grain moisture was adjusted to 14% moisture content. These determinations allowed calculating the total above-ground biomass as well as maize yield components: number of plants per square meter, number of ears per plant and thousand kernels weight (TKW). Grain and above-ground biomass N concentration were determined by dry combustion (Dumas method) (Truspec CN, LECO, St Joseph, MI, USA). Afterwards, N content of the grain and the rest of above-ground biomass were calculated by multiplying the biomass of each fraction by its N concentration. Above-ground N uptake was calculated by the sum of N content in both fractions.

2.4. Cumulative N₂O emissions, emission factor and yield-scaled N₂O emissions

Cumulative N₂O emissions were quantified with the trapezoid rule, differentiating three maize growing seasons from April to November in 2015, 2016, and 2017, and two periods between maize crops from November 2015 to March 2016 and from November 2016 to March 2017. Yield-scaled N₂O emissions were calculated dividing the cumulative N₂O emission in CO₂ equivalents (assuming a global warming potential of 298 as suggested by IPCC, 2013) by maize grain yield (dry matter), for each maize growing season.

The EF was calculated for each year using the following equation:

$$EF (\%) = (E_i - E_0) / (N \text{ Rate}_i) \times 100 \quad (1)$$

here E_i are the cumulative N₂O emissions from the i treatment (kg N₂O-N ha⁻¹), E_0 are the cumulative N₂O emissions (kg N₂O-N ha⁻¹) from the control treatment without N fertilizer, and $N \text{ Rate}_i$ is the N

fertilization rate in the *i* treatment (kg N ha^{-1}). Note that to complete the cumulative N_2O emissions for 2017; we assumed that the emissions of the period between crops in winter are equal to those measured in the season 2016–2017.

2.5. Statistical analysis

Statistical analyses were performed with the statistical package JMP 13 (SAS Institute Inc, 2018). Data were checked for normality by plotting a normal quartile plot. All data complied with normality. A repeated measures analysis of variance (ANOVA) was performed with tillage, N fertilization, sampling date or year or period and their interactions as effects. Sampling date was used as an effect to analyse WFPS, soil ammonium and nitrate contents, N_2O emissions, and denitrification potential. Period (i.e. growing seasons and winter periods between crops) was used as an effect to analyse cumulative N_2O emissions. Finally, year was used as an effect to analyse above-ground biomass, grain yield, N-uptake, and YSNE. When significant, differences among treatments were identified at 0.05 probability level of significance with a Tukey HSD test.

3. Results

3.1. Weather conditions during the experimental period

Mean air temperatures were 19.3, 18.8 and 18.8 °C for the maize season in 2015, 2016 and 2017 respectively. Meanwhile in periods between crops in winter 2015–2016 and 2016–2017 mean air temperatures were 7.7 and 7.1 °C, respectively (Fig. 1a). Cumulative rainfall was 226, 151 and 78 mm for 2015–2017, respectively, during the maize growing season. In the same growing seasons the amount of water applied by irrigation was 631, 672 and 696 mm, respectively (Fig. 1a). During the periods between crops, rainfall was 108 mm and 106 mm in 2015–2016 and 2016–2017, respectively.

3.2. Soil temperature, WFPS, soil bulk density, soil ammonium and soil nitrate content

Mean soil temperatures at the 10-cm soil depth were 18.6, 17.1 and 19.8 °C for in the 2015, 2016 and 2017 maize seasons, respectively.

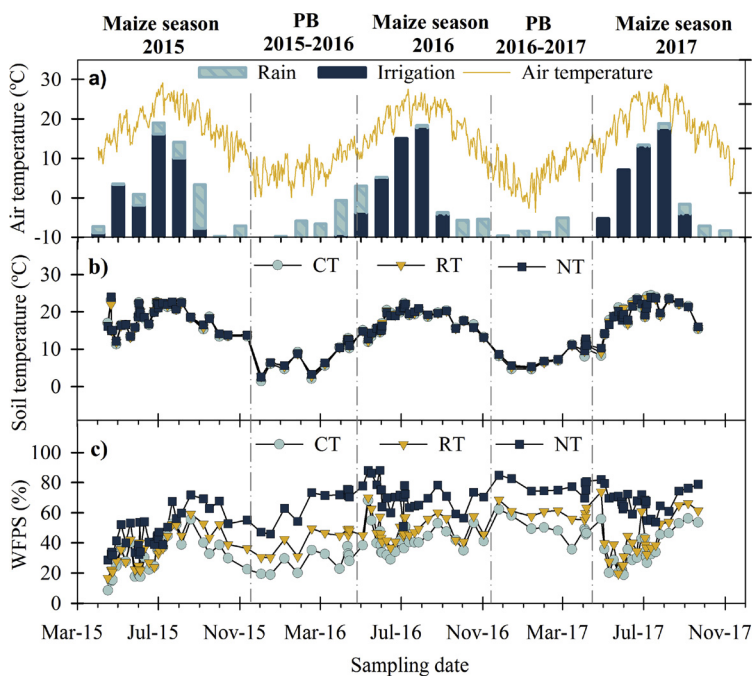


Fig. 1. Monthly precipitation and irrigation (light blue and dark blue columns, respectively) and daily air temperature (continuous line) (a), soil temperature (10 cm depth) (b), and soil water-filled pore space (WFPS) (0–5 cm depth) (c) in plots managed under conventional tillage (CT), reduced tillage (RT) and no-tillage (NT) during the 2015, 2016 and 2017 maize growing seasons and periods between crops in winter (PB 2015–2016 and PB 2016–2017). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article).

Meanwhile in periods between crops in 2015–16 and 2016–17, mean soil temperatures were 6.9 and 8.7 °C, respectively (Fig. 1b). Mean WFPS (0–5-cm soil depth) for CT, RT and NT were 36, 44 and 63%, respectively, as average of the three years of sampling (Fig. 1c).

Soil bulk density (BD) (0–5-cm soil depth) was significantly affected by the interaction between tillage and N fertilization and tillage and sampling date (Table 2). In this regard, soil BD followed the order $\text{NT} > \text{RT} > \text{CT}$, when applying 0, 200 and 400 kg N ha^{-1} (1.46, 1.42 and 1.40 g cm^{-3} for 0 kg N ha^{-1} , 1.43, 1.41 and 1.36 g cm^{-3} for 200 kg N ha^{-1} and 1.46, 1.40 and 1.36 g cm^{-3} for 400 kg ha^{-1} , respectively).

Soil NH_4^+ and NO_3^- contents (0–5-cm soil depth) were significantly affected by the interaction between tillage, N fertilization and sampling date (Table 2). Mean soil NH_4^+ values remained low ($< 5 \text{ kg NH}_4^+ \text{ N ha}^{-1}$) during most of the period studied and increased rapidly after N fertilizer applications (Fig. 2). Soil NO_3^- content peaked after fertilization events (Fig. 3). The application of increasing N rates were accompanied by increasing amounts of NO_3^- in the soil surface (0–5 cm) during the subsequent month, and this trend was of a greater magnitude under CT (Fig. 3).

3.3. Soil N_2O emissions and denitrification potential

Soil N_2O fluxes ranged from $-0.24 \text{ mg N}_2\text{O-N m}^{-2} \text{ d}^{-1}$ (CT-200 on 1st July 2015) to $3.29 \text{ mg N}_2\text{O-N m}^{-2} \text{ d}^{-1}$ (NT-400 on 7th July 2016) (Fig. 4). The interaction between tillage, N fertilization and sampling date had a significant effect on soil N_2O emissions (Table 2). Several N_2O emission peaks occurred during the maize growing period, which were observed within a few days after N fertilizer application (Fig. 4). In the three maize seasons, NT presented the highest N_2O emission values in most sampling dates compared with RT and CT, showing the rate of 400 kg N ha^{-1} had greater soil N_2O emission under NT and 200 kg N ha^{-1} rates (Fig. 4). For instance, for the NT tillage system, the average soil N_2O emission for the 0, 200 and 400 kg N ha^{-1} rates (considering the three maize seasons) were 0.08, 0.29 and 0.52 $\text{mg N}_2\text{O-N m}^{-2} \text{ d}^{-1}$, respectively. In the case of the CT system, the average emission values dropped to 0.04, 0.18 and 0.27 $\text{mg N}_2\text{O-N m}^{-2} \text{ d}^{-1}$ for the 0, 200 and 400 kg N ha^{-1} rates, respectively (Fig. 4). Increases in soil N_2O fluxes also occurred after pre-planting fertilizer application in maize season 2015 only under NT (Fig. 4). Conversely, in the two periods between crops in winter, all N_2O fluxes observed were lower

Table 2
Analysis of variance (P-values) of soil bulk density (BD), soil water-filled pore space (WFPS), soil ammonium and nitrate contents (0–5 cm depth), soil N₂O emissions, denitrification potential, cumulative N₂O emissions for each maize growing season (2015–2017) and period between crops in winter (2015–2016 and 2016–2017), grain yield, above-ground N uptake and yield-scaled N₂O emissions (YSNE), as affected by tillage, N fertilization rate, date/year/period and their interactions.

Source of variation	BD	WFPS	Soil ammonium (0–5 cm)	Soil Nitrate (0–5 cm)	N ₂ O emissions	Denitrification potential	Cumulative N ₂ O emissions	Grain yield	Above-ground N uptake	YSNE
Tillage (Till)	< 0.001	< 0.001	ns	< 0.001	< 0.001	< 0.001	ns	< 0.001	< 0.001	ns
N fertilization (Fert)	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	0.01	< 0.001	< 0.001	< 0.001	< 0.001
Date/Year/Period	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
Till*Fert	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	ns	ns	< 0.001	< 0.001	ns
Till*Date/Year/Period	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	0.008	0.01	0.01	0.006	ns
Fert*Date/Year/Period	ns	0.02	< 0.001	< 0.001	< 0.001	ns	< 0.001	< 0.001	< 0.001	0.04
Till*Date/Year/Period*Fert	ns	ns	< 0.001	0.003	0.001	ns	ns	< 0.001	ns	ns
ns, non-significant										

than 0.3 mg N₂O-N m⁻² d⁻¹ without significant differences between treatments.

Soil denitrification potential was significantly affected by the interaction between tillage and N application date and N fertilization single effect (Table 2). Soil denitrification potential just after pre-planting fertilizer application was higher under NT compared to CT with intermediate values under RT, while no differences between tillage systems were found after top-dressing N applications (Fig. 5). In turn, the application of 200 and 400 kg N ha⁻¹ led to greater soil denitrification potentials compared to the control, with mean values of 0.46, 0.48 and 0.22 g N₂O-N g soil⁻¹ min⁻¹, respectively.

3.4. Cumulative soil N₂O emissions and emission factor

The interaction between N fertilization rates and maize growing season and between tillage system and maize growing season had a significant effect on cumulative N₂O emissions (Table 2). In the 2015 and 2017 growing seasons, cumulative N₂O emissions followed the order 0 < 200 < 400 kg N ha⁻¹. In 2016, the N rates of 200 and 400 kg N ha⁻¹ showed greater values compared to the control (Fig. 6a). No-tillage and CT did not show differences on cumulative N₂O emissions between growing seasons. Differently, under RT differences between maize seasons were found, with greater cumulative N₂O emission in 2017 compared to 2015 and intermediate values in 2016 (0.57, 0.30 and 0.35 kg N₂O-N ha⁻¹, respectively). However, no differences between N rates or between tillage systems were found in the periods between crops in winter 2015–2016 and 2016–2017 (Fig. 6a).

The EF showed the greatest value when applying 200 kg N ha⁻¹ (0.20%) compared to the application of 400 kg N ha⁻¹ (0.18%) as an average of the three years studied (Table 3). Meanwhile, the EF ranged between 0.16 and 0.23% and between 0.10 and 0.22%, under NT and CT respectively, when applying 400 kg N ha⁻¹.

3.5. Maize grain yield, above-ground N uptake and yield-scaled N₂O emissions

The interaction between tillage and N fertilization and their interaction with year had a significant effect on maize grain yields (Table 2). In 2016 and 2017, the application of 200 (12,760 and 10,425 kg ha⁻¹, respectively) and 400 kg N ha⁻¹ (13,067 and 10,879 kg ha⁻¹, respectively) led to greater yields than the control treatment (6870 and 4297 kg ha⁻¹, respectively). In 2015 and 2017, grain yields were higher under NT (11,406 and 9844 kg ha⁻¹, respectively) and RT (9548 and 9278 kg ha⁻¹, respectively) than under CT (5594 and 6478 kg ha⁻¹, respectively), without differences between tillage treatments in 2016. No differences between tillage systems on grain yield were observed in the control treatment, as an average of the three years studied (Fig. 6b). In contrast, greater grain yield was observed under NT compared to CT with intermediate values in RT when applying 200 kg N ha⁻¹. Moreover, as an average of years, greater grain yield was observed under NT and RT when 400 kg N ha⁻¹ were applied, in comparison with CT at the same rate (Fig. 6b).

Maize above-ground N uptake was significantly affected by the interaction between tillage and N fertilization and by the interaction between N fertilization and year (Table 2). In this regard, greater above-ground N uptake was observed under NT than CT, with intermediate values in RT when applying 200 kg N ha⁻¹, (243, 186 and 223 kg ha⁻¹, respectively). Moreover, greater above-ground N uptake was found under NT when applying 400 kg N ha⁻¹ followed by RT and finally by CT at the same rate (295, 240 and 172 kg ha⁻¹, respectively) as an average of the different years covered by the experiment. In 2015, 2016 and 2017 greater above-ground N uptake was observed under the application of 200 (197, 241 and 214 kg N ha⁻¹, respectively) and 400 kg N ha⁻¹ (178, 277 and 252 kg N ha⁻¹, respectively) compared to the control (123, 111 and 79 kg N ha⁻¹, respectively).

Yield-scaled N₂O emissions were significantly affected by the

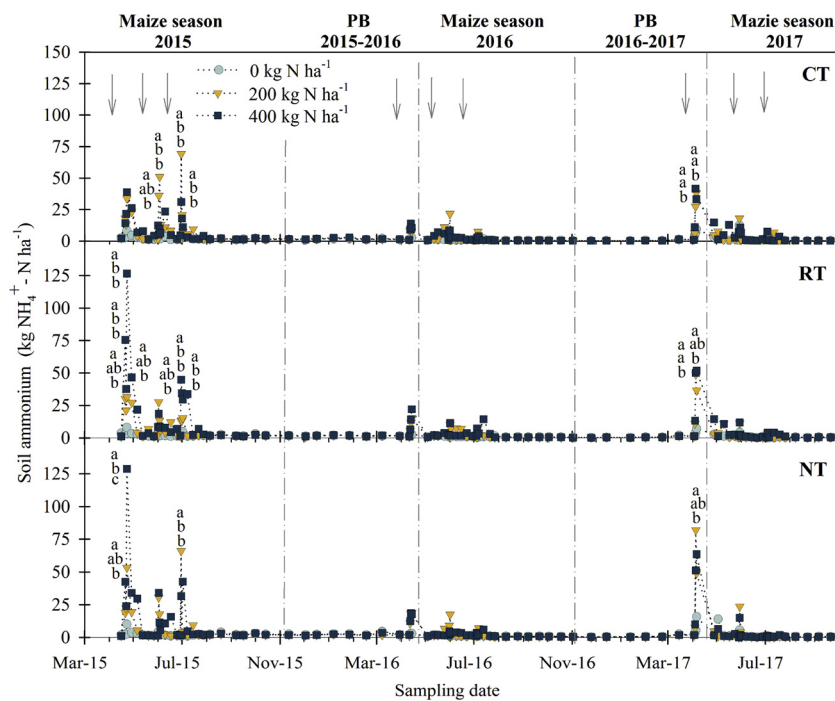


Fig. 2. Tillage system (CT, conventional tillage; RT, reduced tillage; NT no-tillage) and N fertilizer rate (0, 200, 400 kg N ha⁻¹) effects on soil ammonium (NH₄⁺-N) (0–5 cm depth) during the 2015, 2016 and 2017 maize growing seasons and periods between crops in winter (PB 2015–2016 and PB 2016–2017). Arrows indicate dates of N fertilizer application. For a given date and tillage treatment, different lower case letters indicate significant differences between N fertilization rates at $P < 0.05$.

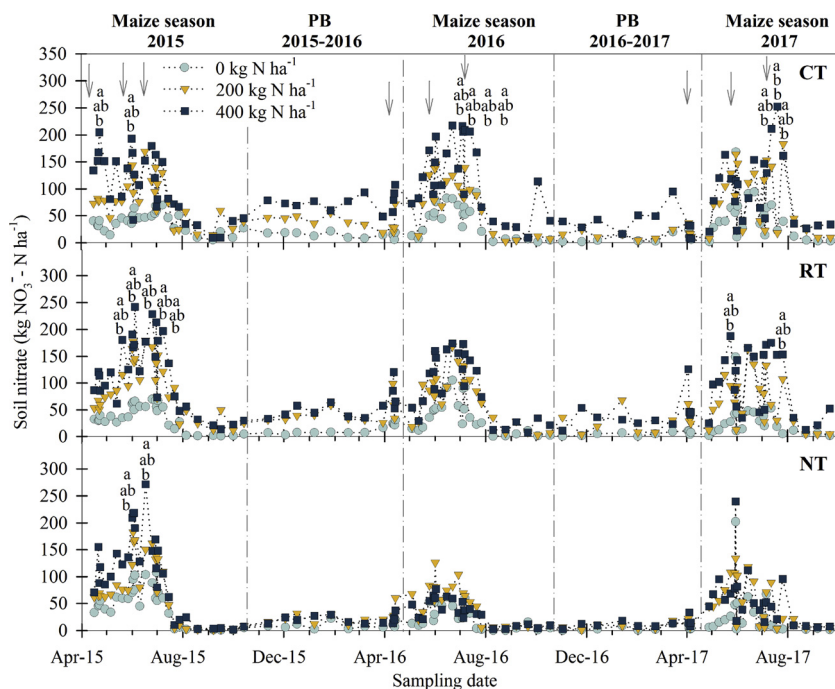


Fig. 3. Tillage system (CT, conventional tillage; RT, reduced tillage; NT no-tillage) and N fertilizer rate (0, 200, 400 kg N ha⁻¹) effects on soil nitrate (NO₃⁻-N) (0–5 cm depth) during the 2015, 2016 and 2017 maize growing seasons and periods between crops in winter (PB 2015–2016 and PB 2016–2017). Arrows indicate dates of N fertilizer application. For a given date and tillage treatment, different lower case letters indicate significant differences between N fertilization rates at $P < 0.05$.

interaction between N fertilization and year (Table 2). In 2015, YSNE showed greater values when applying 400 kg N ha⁻¹, compared to the control and the rate of 200 kg N ha⁻¹. Differently, no significant differences between treatments were found in 2016 and 2017, although a trend of greater YSNE at higher N rates was observed (Fig. 6c).

4. Discussion

4.1. Impacts of tillage and N fertilization rates on soil N₂O emission

When converting rainfed Mediterranean agroecosystems to irrigation, conservation tillage systems like no-tillage and strip-tillage should be maintained since increase the content of organic matter and

therefore the fertility of the soil, leading to sustainable crop production (Pareja-Sánchez et al., 2019) although there may be an increase in N₂O emissions. This study, carried out during three maize seasons, has demonstrated that soil tillage combined with mineral N fertilization rate exerts a significant impact on soil N₂O emissions in Mediterranean irrigated conditions, increasing N₂O emissions when N application was higher under no-tillage. In this regard, different studies in irrigated Mediterranean conditions have shown that high rates of N fertilizer, lead to greater soil N₂O fluxes (Meijide et al., 2007; López-Fernández et al., 2007; Álvaro-Fuentes et al., 2016; Guardia et al., 2017). However, the present study demonstrates that the effect of N fertilizer on N₂O emissions in Mediterranean irrigated areas is determined by soil tillage. The different tillage systems studied influenced N₂O emissions

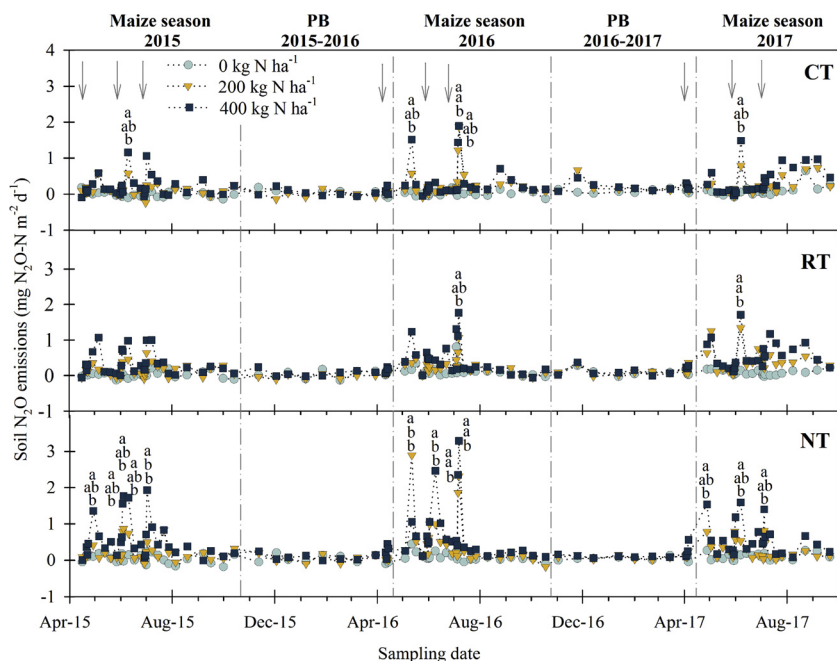


Fig. 4. Tillage system (CT, conventional tillage; RT, reduced tillage; NT no-tillage) and N fertilizer rate (0, 200, 400 kg N ha⁻¹) effects on soil N₂O emissions during the 2015, 2016 and 2017 maize growing seasons and periods between crops in winter (PB 2015–2016 and PB 2016–2017). Arrows indicate dates of N fertilizer application. For a given date and tillage treatment, different lower case letters indicate significant differences between N fertilization rates at *P* < 0.05.

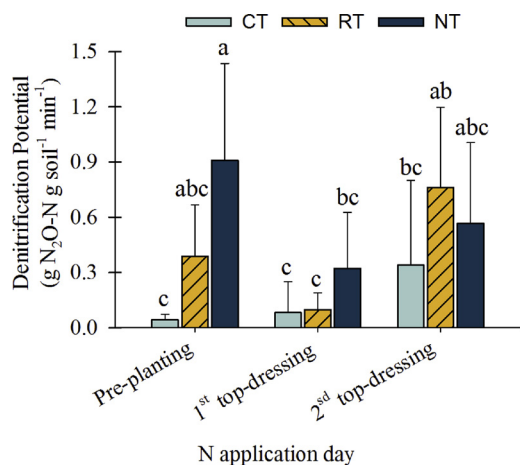


Fig. 5. Tillage system (CT, conventional tillage; RT, reduced tillage; NT no-tillage) effects on soil potential denitrification 5 days after pre-planting N fertilizer application, 1st top-dressing application and 2nd top-dressing application during the 2016 maize growing season. Different lower case letters indicate significant differences between tillage systems at *P* < 0.05. Vertical bars indicate standard deviation.

through variations in the WFPS and mineral nitrogen content, which play a substantial role in N₂O emissions, by influencing microbial activity and water distribution in the soil matrix (Rees et al., 2013). In our study, the soil properties that were mostly affected by the tillage treatments were soil physical properties, especially BD and soil structural degradation. An increase in BD under NT treatment could lead to greater WFPS and, therefore, higher N₂O emissions under NT than CT, as numerous authors have shown (Hansen et al., 1993; Ruser et al., 1998, 2006). The authors observed a strong increase in N₂O emissions under soils with higher bulk density, which were primarily a result of an increase of the WFPS. However, in our study, also soil structural degradation could be an important factor, in the CT treatment caused by the formation soil surface crusting, which avoided the entry of water into the soil profile. The main process behind soil crusting in this trial was the breakdown of dry-sieved (Pareja-Sánchez et al., 2017). This physical degradation led to changes in WFPS, NO₃⁻ or NH₄⁺ that could influence N₂O emissions as explained throughout the discussion. In our

study, the results clearly show that the highest rates of N fertilization had major impacts on N₂O emission under NT (Fig. 4). In all three tillage systems, the highest N₂O fluxes occurred within a few days after N fertilization, contributing about 60% of the total emissions in the three years studied. Exceptionally, in the first year of study, NT was the only tillage system that showed a N₂O peak associated with the pre-planting fertilizer application. This could be due to the incorporation of the fertilizer by tillage (CT and RT) in very dry soil conditions, since irrigation began a week after the N application.

During the three years of study, the highest N₂O fluxes were generally observed when WFPS was above 60% under NT with 400 kg N ha⁻¹. However, on some specific dates, the N₂O emissions were higher with a low WFPS in the CT treatment. As above, CT and RT show some WFPS values > 40% resulting in lower emissions. N fertilizer and soil moisture are the two main factors influencing soil N₂O emissions (Gao et al., 2014). In this study, soil water content and soil bulk density were higher under NT than under CT, which resulted in generally higher levels of WFPS. Under NT, greater denitrification rates could also be stimulated by the greater levels of SOC in NT compared to the CT systems (Plaza-Bonilla et al., 2013; Álvaro-Fuentes et al., 2014). It is well known that denitrifying bacteria require available C as an energy source before the reduction of added nitrogen can occur (Saggar et al., 2013). In our conditions, it was likely that a fast nitrification of the ammonium to nitrate could have been the main N₂O production process which is justified by the low levels of soil NH₄⁺ (4.8 kg NH₄⁺ - N ha⁻¹ as an average of three years of study) and the low WFPS, especially in CT and RT treatments (< 40 and 50%, respectively, as an average of three years of study). Differently, under NT, in some periods, denitrification could have also produced N₂O emissions due to the higher WFPS (> 60% as an average of three years of study) as observed by other authors (Venterea et al., 2005). This last assumption is supported by the greater denitrification potential of NT treatment compared CT and RT observed in the study.

During the periods between crops (winter months) N₂O emissions were low and did not show significant differences between treatments. The low N₂O emissions during these periods could be explained by the soil temperatures, which were lower than 8 °C leading to low activity levels of nitrifying bacteria in the soil (Smith et al., 2010).

As N₂O emissions are mainly driven by soil moisture and soil mineral N levels, careful management of agricultural practices involving

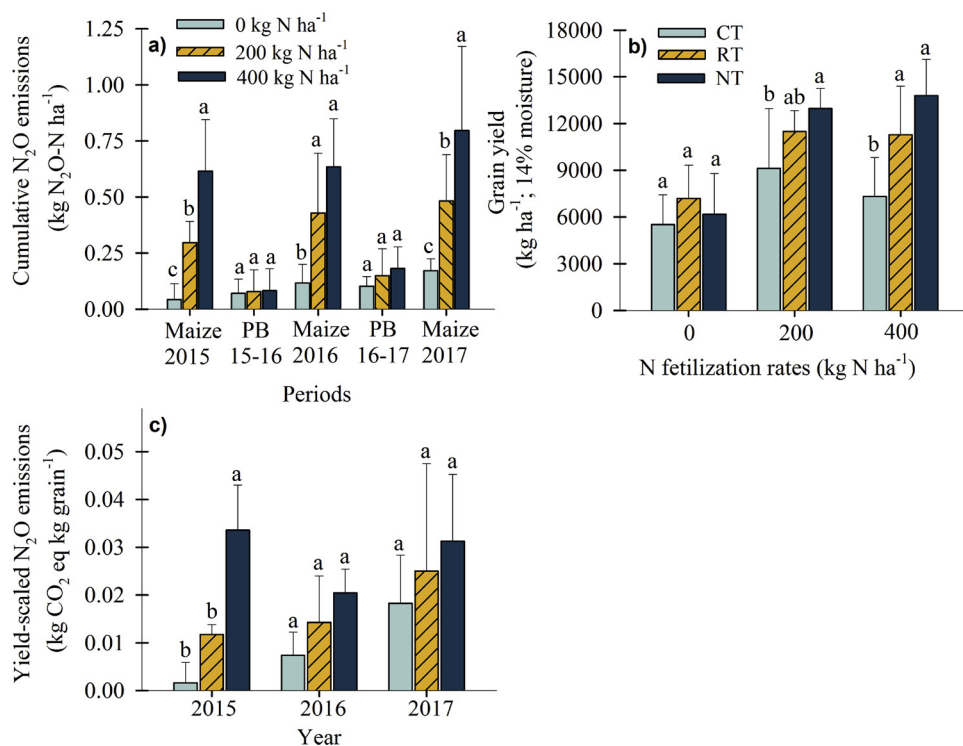


Fig. 6. Nitrogen fertilizer rate (0, 200, 400 kg N ha⁻¹) effects on cumulative N₂O emissions (a) and yield-scaled N₂O emissions (c), and tillage system (CT, conventional tillage; RT, reduced tillage; NT, no-tillage) effects on grain yield (b). Values correspond to three consecutive maize growing seasons (2015–2017) and two periods between crops in winter (PB 2015–2016 and PB 2016–2017). Different lowercase letters indicate significant differences between N fertilization rates for a given period (a and b) and significant differences between tillage systems for a given N fertilization rate (c) at $P < 0.05$. Vertical bars indicate standard deviation.

Table 3

Soil N₂O emission factor (EF) (%) in 2015, 2016 and 2017 as affected by N fertilization rate (200 and 400 kg N ha⁻¹) and tillage system (CT, conventional tillage; RT, reduced tillage; NT, no-tillage). Average of the three years studied in rate of 200 and 400 kg N ha⁻¹.

Year	Tillage system	EF (%)	
		200 kg N ha ⁻¹	400 kg N ha ⁻¹
2015	CT	0.09	0.10
	RT	0.07	0.15
	NT	0.17	0.22
2016	CT	0.33	0.20
	RT	0.12	0.12
	NT	0.27	0.23
2017	CT	0.31	0.22
	RT	0.29	0.22
	NT	0.13	0.16
Average		0.20	0.18

fertilization, tillage and irrigation are very important when it comes to minimizing gaseous losses (Cayuela et al., 2017). Management can be key through proper irrigation use which can reduce N₂O emission (Franco-Luesma et al., 2019). For example, performing the irrigation according to the needs of the crop as measured in this experiment. Another example would be not applying irrigation water immediately after fertilization could decrease N₂O emissions. Also, N fertilizer rate adapted to the needs of the crop could lead to a decrease of N₂O. Moreover, delaying the timing of application of N fertilizer may have helped to reduce N₂O emissions (Venterea et al., 2012).

4.2. Cumulative N₂O emissions and emission factor

Previously, in the same experimental field, under rainfed CT barley cumulative N₂O emissions were lower compared to the values found in our study in irrigated conditions (0.43 vs. 0.52 kg N₂O-N ha⁻¹, respectively), and in NT this difference was even greater compared to irrigated conditions (0.33 vs. 0.63 kg N₂O-N ha⁻¹, respectively) (Plaza-Bonilla et al., 2014) due to the increased soil moisture and N fertilization rate in the irrigation experiment. The lower increase of N₂O

emissions in CT (only 17%) could be due to the low WFPS which was caused by surface crusting that reduced the infiltration of water into the soil (Pareja-Sánchez et al., 2017). In CT the lower production of maize biomass as well as the reduced availability of water in the soil negatively influenced crop N uptake and led to an accumulation of soil nitrate. Although a higher soil NO₃⁻ content was observed under CT, N₂O emissions remained low since WFPS values were generally below 40% under this tillage system. Therefore, the physical properties through soil structural degradation had a greater influence on N₂O emissions.

In all three maize growing seasons, the greatest cumulative soil N₂O emissions were obtained with the highest N rates (400 kg ha⁻¹) and declined as the rate of N decreased. The high cumulative N₂O emissions found in the treatments with the greatest N fertilization rate could be related to the high NO₃⁻ concentration in the soil when applying high rates of N, favoring denitrification. The addition of N fertilizer increases soil mineral N losses as N₂O through higher nitrification and denitrification rates (Bouwman et al., 2002).

In our study the EF (the percentage of fertilizer N applied that is emitted on-site as N₂O) of irrigated maize was lower than the default 1% factor currently proposed by the IPCC (IPCC, 2006). The highest EF estimated in our experiment was 0.24% for CT when applying 200 kg N ha⁻¹ and 0.20% in NT when applying 400 kg N ha⁻¹, as an average of the three years of studied. In a meta-analysis of N₂O emissions in Mediterranean cropping systems, Cayuela et al. (2017) showed that irrigated maize production presents an average EF of 0.83%, a value higher than the ones obtained in our study. This disagreement could be explained by different causes. One hypothesis could be soil texture, which was fine-textured in our study. Soil texture affects soil N₂O production through its influence on soil aeration which, in turn, modulates nitrification and denitrification processes. Cayuela et al., 2017 suggested that larger EFs could be expected from coarse (EF: 0.58%) and medium-textured soils (EF: 0.48%) compared to fine textured soils (EF: 0.27%). This last value agrees with the one found in our study and would confirm that fine-textured Mediterranean soils usually present low N₂O EF. This could be due to the fact that in fine-textured soils, aeration is lower and therefore less oxygen is available for the microorganisms in microsites, even in rather low WFPS levels. Under these

conditions microorganism would further reduce N_2O decreasing the amount of this gas emitted to the atmosphere (Šimek and Cooper, 2002). Another hypothesis that could explain the low N_2O EF found in our study would be related to the management of irrigation. In order to reduce the emission of N_2O as much as possible, we did not apply irrigation water immediately after N fertilization, maintaining soil WFPS at low levels, avoiding the rapid burst of N_2O emission usually found in other experiments (e.g. Álvaro-Fuentes et al., 2016). Irrigation water was applied 3 days after fertilizer application at low and frequent rates equivalent to crop needs. When the concentration of nitrate in the soil is high and the WFPS is low the emission of N_2O could be reduced. Venterea et al. (2011) in rainfed maize in Minnesota, with a mean annual precipitation of 879 mm, obtained EF in the range of 0.14 to 0.42% of the applied N (146 kg N ha^{-1}) as averaged across all treatments. They concluded that the timing of fertilizer application could reduce N_2O emissions leading to lower EF values. Through increasing the number of N applications during the growing season would result in reduced N_2O emissions (Li et al., 2012) since, split applications, performed to more closely match N uptake demands by maize.

4.3. Impacts of tillage and N fertilization rates on maize productivity and yield-scaled N_2O emissions

In this study, averaged over 3 years, grain yields in NT and RT were similar when applying 200 and 400 kg N ha^{-1} , while CT showed the lowest yields at both N rates (Fig. 6b). The lack of yield difference between 200 and 400 kg N ha^{-1} could be attributed to the high initial N availability for crop growth in the plots fertilized with 200 kg N ha^{-1} . Therefore, these data suggest that the use of less aggressive tillage practices, such as no-tillage and strip-tillage, as well as the reduction of N fertilization, could be viable options to stabilize or, even, increase crop yields. Moreover, it could lead to a decrease of N_2O emissions to the atmosphere simultaneously, saving production costs in comparison with the traditional management based on conventional tillage with high rates of mineral N. Hence, it is interesting to analyze N_2O emissions in relation to the yield obtained, since it provides a good indicator of the environmental impacts of intensive agricultural production systems. In our study, an increase in the N rate led to an increase in the yield-scaled N_2O emissions only in the first out three years, although a similar trend was observed in the subsequent two years (Fig. 6c). But, as explained before, similar yields were obtained with both 400 kg N ha^{-1} and 200 kg N ha^{-1} . These results suggest that optimal N rates can produce maximum yields while reducing annual yield-scaled emissions by 40%. Moreover, although the yield-scaled N_2O emissions did not differ significantly between tillage treatments, a marked trend existed in the rates found between tillage systems, in the order $CT > RT > NT$. Conventional tillage was greatly affected by soil degradation and led to lower plant density inducing lower grain yield, compared to NT that showed greater grain yield. These results suggest that although cumulative N_2O emissions under CT are lower, the reduction in crop yield in CT led to an increase in yield-scaled emission compared to NT. Conversely Venterea et al. (2011), in a maize-soybean rotation in SE Minnesota (USA), observed that the yield-scaled N_2O emission for CT was 40.7% lower than in NT with a urea fertilizer N input of 146 kg N ha^{-1} . In their case, averaged over 3 years study, the grain yield were 14.2% lower in NT than in CT. Lower yield NT was attributed to cooler soil temperatures in the spring, which may inhibit early-season plant development (Venterea et al., 2011). Our results demonstrated that in order to keep yield-scaled N_2O emissions low, it is necessary to obtain adequate crop productivity. Reducing yield-scaled emissions is consistent with the aim of ensuring the sustainability of production and minimizing environmental impacts (Powelson et al., 2011).

5. Conclusions

In Mediterranean irrigated maize systems, a reduced tillage strategy

is key to maintaining high yields. In this study, we found that the increase of WFPS under NT had a major effect on N_2O emissions especially when combined with the high rate of N fertilization that increased soil mineral N. The yield-scaled N_2O emissions did not significantly differ between tillage treatments since greater grain yield under NT offset the higher N_2O emissions. However, the use of a high N rate led to an increase in the yield-scaled N_2O emissions in the first year of study. In this cropping system and climate regime, the mean N_2O EF measured was 0.19%, much lower than the 1% factor currently default by the IPCC. Therefore, the results of this work confirm that the IPCC default EF often overestimates the emissions of N_2O in Mediterranean areas.

When converting rainfed Mediterranean systems to irrigation, conservation tillage should be maintained for sustainable maize production. No-tillage is an adequate technological opportunity for the transition from rainfed to irrigated land. If some problems arise under NT during this period, such as those related to crop residue management and/or soil compaction in the planting row, the implementation strip-tillage would be a key alternative. Moreover, the use of an appropriate N fertilizer rate according to crop needs may achieve a yield advantage while decreasing soil N_2O emissions, independently of the tillage treatment. This combination of management strategies is important to reduce N_2O emissions as well as enhance crop productivity.

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