

Nitrate accumulation in the soil profile: Long-term effects of tillage, rotation and N rate in a Mediterranean Vertisol

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ABSTRACT

Excessive application of fertiliser in agriculture can have detrimental effects, one of which is diffuse contamination with nitrates. An 18-year field study was conducted on a typical rainfed Mediterranean Vertisol to determine the effects of the tillage system, crop rotation and N fertiliser rate on the long-term NO_3^- -N content in the soil profile (0–90 cm). The experiment was designed as a randomised complete block with a split-split plot arrangement and 3 replications. The main plots tested the effects of the tillage system (no-tillage and conventional tillage); the subplots tested crop rotation, with two 2-yr rotations (wheat-wheat, wheat-fallow, wheat-chickpea, wheat-faba bean and wheat-sunflower); and the sub-subplots tested the N fertiliser rate (0, 50, 100 and 150 kg N ha⁻¹). The nitrate content increased with time. The tillage system showed an inconsistent effect on nitrates, although, overall, nitrate levels were higher under conventional tillage than with no-tillage. The wheat-faba bean rotation induced a larger accumulation of soil nitrates. Nitrates usually accumulated to a greater extent in the 30 to 60-cm depth of soil. As a rule, farmers should know the amount of residual N existing in the soil prior to crop fertilisation in order to avoid over-fertilisation.

Keywords: no-tillage, wheat, faba-bean, chickpea, sunflower, rainfed

1. Introduction

Agriculture is recognised as a source of nonpoint pollution, and one major concern is the contamination of surface and deep waters by nitrates (Borin, 1997). Inadequate agricultural practices, particularly intensive agriculture, can lead to excessive fertilisation

of the system. Over-fertilisation is commonly employed as a form of insurance, especially by farmers in developed countries (Fageria and Baligar, 2005). The amount of N fertiliser required under semiarid climates is largely dictated by the seasonal rainfall. Experiments on dry Vertisols in southern Spain (Garrido-Lestache et al., 2004; López-Bellido et al., 1996 and 2000) showed that wheat yield increased with an N fertiliser application rate of up to 100 kg ha⁻¹ during wet years but had little or no response to N fertiliser during dry years. The goal is to supply the correct amount of N when the crop needs it and where the crop can access it, while minimising losses to the environment (Schlegel et al., 2005). The damaging effects, both environmental and economic, can be reduced by studying the efficiency of N fertiliser in soil. The efficiency of the N applied to satisfy the N demand of the crop depends on factors such as the type of fertiliser, the timing of application and seasonal trends (Blankenau et al., 2002; Borghi, 2000; Huggins and Pan, 1993). López-Bellido et al. (2005, 2012) studied strategies of splitting and N timing in wheat crop to ensure an adequate amount of N in a rainfed Mediterranean system. According to Dinnes et al. (2002), these strategies play an important role in the reduction of losses of NO₃⁻-N from crop root zones.

Tillage and crop rotation with legume crops are two management practices that can influence the N dynamics of soil-plant systems. Some studies have revealed changes in the efficiency of available N under reduced tillage and no-tillage conditions (Campbell et al., 1993; Peoples and Herridge, 1990; Rao and Dao, 1992). Growing several species of crops together or sequentially may utilise nutrients more efficiently than monoculture if the different species exploit a larger soil volume or different parts of the profile (Francis, 1989). Legume crop residue, particularly green manure, is an effective source of N (Bremer and van Kessel, 1992; Haynes et al., 1993). When released in synchrony with crop N demand, crop residue N is a particularly desirable source of N because losses to the environment are minimised (Stute and Posner, 1995). However, Bremer and van Kessel (1992) showed that the influence of crop residues on plant-available N depends on how they affect the net mineralisation of other soil N sources.

The efficient capture of nitrate from the topsoil requires a high rooting density. Given favourable soil conditions, winter wheat has one of the most rapidly growing and prolific root systems of all arable crops (Barraclough et al., 1991). Nitrate in groundwater can originate from many sources, but N fertiliser and the mineralisation of soil organic N are regarded as primary components in the NO₃ contamination of groundwater supplies (Keeney, 1989). Long-term N fertility studies have shown that residual soil NO₃⁻-N increases when N fertilisation rates exceed that needed for maximum yield (Halvorson

and Reule, 1994; Porter et al., 1996; Raun and Johnson, 1995; Westerman et al., 1994). In addition, there is a substantial carryover effect of fertiliser N from one growing season to the next in cropped soils, especially under semiarid Mediterranean conditions (Corbeels et al., 1998), which contributes to the progressive accumulation of nitrates in the soil.

There is scarce information on the long-term effect of agricultural practices on soil NO_3^- -N in rainfed Mediterranean Vertisol. The objective of this study was to determine the impact of tillage system, crop rotation, and N fertiliser on soil NO_3^- -N accumulation and depth distribution in a Vertisol under dryland Mediterranean conditions in 18-yr.

2. Materials and methods

2.1. Site and experimental design

Field experiments were conducted in Córdoba, southern Spain ($37^\circ 46' \text{ N}$ and $4^\circ 31' \text{ W}$, 280 m a.s.l.), on a Vertisol (Typic Haploxererts) typical of the Mediterranean region, where rainfed cropping is the standard practice (Table 1). The study took place over an 18-year period (1992–1993 to 2009–2010) within the framework of a long-term experiment called “Malagón” that began in 1986 and was designed as a randomised complete block with a split-split plot arrangement and 3 replications. The main plots tested the effects of the tillage system (no-tillage and conventional tillage); subplots tested 2-year crop rotations (wheat-wheat, wheat-fallow, wheat-chickpea, wheat-faba bean and wheat-sunflower); and sub-subplots tested N fertiliser rate (0, 50, 100 and 150 kg N ha^{-1}). Each rotation was duplicated in a reverse crop sequence to obtain data for all crops on a yearly basis. The area of each sub-subplot was 50 m^2 (10 by 5 m).

2.2. Crop management

The no-tillage plots were seeded with a no-till seed drill. Weeds were controlled with glyphosate [N-(phosphonomethyl) glycine] + MCPA [(4-chloro-2-methylphenoxy) acetic acid] at a rate of 0.5 + 0.5 L a.i. ha^{-1} prior to planting. The conventional tillage treatment included mouldboard ploughing and disk harrowing and/or vibrating tine cultivation to prepare a proper seedbed. During the chickpea and faba bean growing season, weeds were controlled by means of cyanazine [2-(4-chloro-6-ethylamino-1,3,5-triazin-2-yl-amino)-2-methyl propionitrile] at 2 L a.i. ha^{-1} . Ascochyta blight (*Didymella rabiei*) in

chickpea was controlled with chlorothalonil [2, 4, 5, 6 – tetra–chloroisophthalonitrile] at 0.75 a.i ha⁻¹. Glyphosate was applied on faba bean plots at rate of 0.065 L a.i. ha⁻¹ as a post-emergence spray when broomrape (*Orobanche crenata* Forsk) was approximately 0.5–1 cm high (García-Torres et al., 1987).

As is common practice in the area, kabuli chickpea (cvs Pedrosillano and Zoco) was planted in 0.38-m-wide rows in February at a seeding rate of 80 kg ha⁻¹; faba bean (cv Alameda) was planted in 50-cm-wide rows in early December at 170 kg ha⁻¹; sunflower (various hybrid cultivars) was planted in 50-cm-wide rows in February at 5 kg ha⁻¹; and hard red spring wheat (cvs Cajeme and Gazul) was planted in 18-cm-wide rows in late November at 150 kg ha⁻¹. Each year, the wheat plots were also supplied with P fertiliser at a rate of 65 kg P ha⁻¹; the fertiliser was incorporated following the standard practice in the conventional tillage soil and was banded with drilling in the no-tillage plots.

2.3. Soil analysis

Three soil core samples were collected from each wheat plot prior to sowing at a depth of 0–90 cm using a manual Eijkelkamp auger. Every three years, samples were divided into three portions: 0 to 30, 30 to 60 and 60 to 90 cm for analysis of each soil depth. Due to the high clay content of the soil, the samples had dried before grinding and sieving. Samples were crumbled, as finely as possible, and spread out on trays in order to facilitate air dry for 48 h. Subsequently, they were ground, sieved (2 mm), and kept frozen in plastic bags at –30°C until analysis. Soil nitrate was determined by a colorimetric method: Griess Illosvay modified by Barnes and Folkard (1951) and Bremner (1965). This method involves the quantitative extraction of nitrate from soils using 2.0 M KCl. Nitrate is determined by reduction to nitrite via a cadmium reactor. A continuous flow colorimeter was used for this purpose (QUATRO, Bran Luebbe, Norderstedt, Germany). Soil electrical conductivity was measured in a 1:5 soil-water extract, using a Crison GLP 31 conductivity meter (Crison Instruments, Barcelona, Spain) calibrated against a reference NaCl solution in the wheat-sunflower rotation with 150 kg N ha⁻¹ in 2010.

2.4. Statistical analysis

We analysed the data using a mixed-model analysis of variance (ANOVA) with three fixed factors: tillage system, crop rotation and N rate. The year was considered a random factor in this work due to unpredictable weather conditions under rainfed Mediterranean conditions (Gómez and Gómez, 1984). Nitrate levels were subjected to ANOVA using a

split-split plot design combined over the years, tillage system, preceding crop and N rate following the error term according to McIntosh (1983). When their effects were calculated as being significant, the treatment methods were compared using Fisher's protected least significant difference (LSD) test with a significance threshold at $P = 0.05$. The ANOVAs were performed using Statistix 8.1 (Analytical Software, 2005) to determine the treatment effects.

3. Results

3.1. Year effects

All main effects and interactions were significant in the soil nitrate content except tillage system x rotation x N rate (Table 2). Over the entire experiment, an increase in the amount of nitrate accumulated in the total soil profile (0-90 cm) was observed across the years (Fig. 1). From the first year of analysis (1992) until 2009, the nitrate levels increased by 77 kg ha^{-1} (from 37 to $114 \text{ kg NO}_3^- \text{-N ha}^{-1}$, respectively) in the total soil profile, which represents an increase of 200% of $\text{NO}_3^- \text{-N}$ in the soil in 18 years (Fig. 1).

3.2. Tillage effects

The tillage system also affected the levels of nitrates. Summing up the results of the whole time span of 18 years, the nitrate concentrations were higher on average under conventional tillage compared to no-tillage (112 and $104 \text{ kg NO}_3^- \text{-N ha}^{-1}$, respectively). Significant differences were found in only five years: 1995, 2006, 2007 and 2008, where conventional tillage was superior to no-tillage; in 2004, the opposite occurred. Figure 1 shows how these differences existed essentially only in the final period and not in the early years. The trend lines tend to separate as tillage system implementation takes more time, favouring conventional tillage nitrate accumulation. Over the years, there were not significant differences in soil nitrate by tillage system at rates of 0 and 150 kg N ha^{-1} applied to wheat, but there were differences in rates of 50 kg N ha^{-1} (94 and 83 kg N ha^{-1} in CT and NT, respectively) and 100 kg N ha^{-1} (121 and 110 kg N ha^{-1} in CT and NT, respectively).

3.3. Rotation effects

Different crop rotations with wheat gradually induced differences in the soil nitrate concentration. Averaged across years, there was a higher amount of nitrate in the wheat-faba bean rotation (139 kg ha⁻¹), followed by wheat-wheat, wheat-fallow, wheat-chickpea and wheat-sunflower (124, 117, 104 and 55 kg NO₃⁻-N ha⁻¹, respectively), which are all significantly different. The soil nitrate content also varied according to the rotations over time (Fig. 2). In the first year of the study, the nitrate content was largely similar in all rotations, and no significant differences were found. However, as time progressed, significant differences were evident between these rotations (Fig. 2). Overall, the nitrate content increased as the years progressed, except in the wheat-sunflower rotation, in which nitrate levels remained virtually constant (Fig. 2).

3.4. N rate effects

Averaged across years, the nitrate content was highest with application of 150 kg N ha⁻¹, followed by 100, 50 and 0 kg N ha⁻¹, and nitrate content varied significantly between all application rates (145, 116, 88 and 82 kg NO₃⁻-N ha⁻¹, respectively). As the years passed, there was a clear trend of increasing nitrate in soil for all rates of N fertiliser applied (Fig. 3). In addition, the slope was greater as the rate of N fertiliser applied increased. In the first year of the analysis (1992), the nitrate content was very similar for all rates of N fertiliser application. However, as time passed, the differences became greater among the different rates, increasing the nitrate content of the soil throughout the period 1.8, 2.4, 3.7 and 5 times for rates of 0, 50, 100 and 150 kg N ha⁻¹, respectively (Fig. 3).

3.5. Nitrate distribution in the soil profile

The nitrate content, as time passed, was higher at all soil depths (Fig. 4). However, soil nitrate increased more rapidly at depths of 30–60 and 60–90 cm, even though there was a decrease in the same depths between 2006 and 2009 (Fig. 4). The average nitrate content was 26 kg ha⁻¹ in the first 30 cm of soil, 46 kg ha⁻¹ at 30–60 cm and 39 kg ha⁻¹ at 60–90 cm. At first (1995), the nitrate content was lower in the deepest layer of the soil (60–90 cm) and higher at 0–30 and 30–60 cm. In subsequent years, nitrate accumulated more in the deeper soil profiles (Fig. 4).

The tillage system x crop rotation x N rate x soil depth interaction for 18-yr period at 3-yr intervals was also significant (Fig. 5). In general, the nitrate levels increased, as did the rate of N application, in virtually all rotations (Fig. 5). The highest values were found at a

rate of 150 kg N ha⁻¹ for rotations of wheat-wheat and wheat-faba bean. Nitrates usually accumulated to a greater extent at a depth of 30-60 cm for each rotation and N rate applied. The tillage system was also significant in some cases (Fig. 5); in these cases, conventional tillage favoured the accumulation of nitrates in the soil, while the opposite occurred almost exclusively in the rotation with faba bean. There were more significant differences in the 30–60 cm layer, for which nitrate content was always higher in conventional tillage compared to no-tillage.

The nitrate content at 0–30 cm did not differ among different N application rates in any rotation except in wheat-wheat, where there were higher nitrate levels at rates of 100 and 150 kg N ha⁻¹ than at 0 to 50 kg N ha⁻¹ (Fig. 5). The content of nitrates at 30–60 and 60–90 cm was greater with 100 kg N ha⁻¹ than with 0 and 50 kg N ha⁻¹ in all rotations, and there were virtually no significant differences in nitrate content between these rates in any rotation (Fig. 5). At 150 kg N ha⁻¹, the soil accumulated more nitrate in wheat-fallow, wheat-faba bean and wheat continuous rotations (Fig. 5). In wheat-chickpea and wheat-sunflower, there were no differences between the rates of 100 and 150 kg N ha⁻¹ (Fig. 5).

The soil electrical conductivity measured in the wheat-sunflower rotation with 150 kg N ha⁻¹ in 2010 showed an increase with depth. The values were 0.44, 1.8 and 2.4 dS m⁻¹ at depths of 0–30, 30–60 and 60–90 cm, respectively.

4. Discussion

The development of nitrate levels over time varied depending on the year, tillage system and previous crop. In our experiment, the values of nitrate levels in the 0–90 cm soil profile ranged between 37 and 191 kg N ha⁻¹, depending on the year. Herridge et al. (1998) observed that nitrate in the top 0.9 m of the soil at sowing varied between 19 and 158 kg N ha⁻¹, with amounts reflecting site characteristics, fertiliser N rate and previous fertiliser N utilisation. Under the rainfed Mediterranean Vertisol studied, periods of intensive accumulation of mineral N can be followed by N depletion, depending on rainfall and crop growth, resulting in fluctuations in the content of nitrates between years. Moreover, in heavy rainfall years - which also occur occasionally - denitrification can be significant due to poor soil aeration of Vertisols, although there are disagreements between studies (Probert et al. 1987). However, a particularly marked increase of nitrates in the soil was observed from the early to final years. Corbeels et al. (1998) pointed out the carryover effect of N fertiliser from one growing season to the next in soils under

rainfed Mediterranean conditions can be substantial, but in this study, long term effects of continuous treatment must be also taken into account.

Overall, the nitrate content was higher under conventional tillage than with no-tillage. Several studies have shown similar results to ours (Borin, 1997; Eck and Jones, 1992; Grant and Lafond, 1992; McConkey et al. 2002; Soon et al., 2001). To explain this result, these studies suggest a lower net N mineralisation under no-tillage due to slower decomposition and more N immobilisation and nitrification differences. Also, López-Bellido et al. (1997) noted a more marked translocation in NT soil because it was favoured by cracks formed in Vertisols during the drought period. The tillage system did not affect nitrate accumulation where there was an excess or deficiency of N fertiliser (rates of 150 and 0 kg N ha⁻¹, respectively). Differences were observed only at critical rates of response to N fertiliser (50 and 100 kg N ha⁻¹), where in both cases the nitrates were higher in conventional tillage than in no-tillage conditions.

The development of nitrate in the soil gradually differentiated with crop rotations, and there was a greater accumulation of nitrate in the wheat-faba bean rotation. According to O'Connor et al. (2010), two mechanisms could lead to these differences: a higher rhizodeposit mineralisation of legumes and lower utilisation of nitrates by the faba beans compared with wheat. Burns (1980) noted that the effective maximum rooting depth for nitrate in faba beans may be less than half that of wheat, which means faba beans use less soil nitrate. The wheat monoculture also accumulated large amounts of nitrates in the soil, but this was due to it receiving an annual rate of N as opposed to rotations where only cereals were fertilised. The soil nitrate content in wheat monoculture was followed by the wheat-fallow rotation. Schlegel et al. (2005) showed that fallow systems can accumulate significant amounts of NO₃⁻-N in the soil profile from mineralisation during an extended fallow period. As reported by López-Bellido et al. (2000), the rotation that had the lowest accumulation of soil nitrates was wheat-sunflower, showing that sunflower is an excellent complement to wheat in terms of N utilisation (López-Bellido and López-Bellido, 2001). Connor and Jones (1985) showed that the sunflower root density is high even at a depth of 120 cm under rainfed conditions, which means the sunflower crop can explore and absorb the NO₃⁻-N in the deeper layers of soil. Thanks to this exhaustive exploitation of sunflower, the amount of nitrates in rotation with wheat remains virtually unchanged over time.

The results obtained with the variation in fertiliser N rates were similar to those of Kücke and Kleeberg (1997), who reported that reduction in N fertilisation decreased the nitrate

contents in soil solutions. The nitrate content at 30–60 and 60–90 cm soil developed similarly, with a clear increase over time. However, it was the 30–60 cm depth that showed the highest accumulation of nitrate in all years of the study. Probert et al. (1987) reported that because leaching in Vertisols is usually not excessive, there is often a spike in of NO_3^- -N at some depth in the soil profile, which is also caused by the cracks in these soils. The decline in nitrate content for 30–60 and 60–90 soil depth from 2006 to 2009 could be due to higher grain yield in 2006–2009 period than in 2003–2006 (3 times higher). Although, as mentioned previously, the nitrate accumulation in the soil was at its highest at 30–60, equal or even increased nitrate levels were often observed at 60–90 cm (Fig. 4). Also, Probert et al. (1987) indicated that the content of NO_3^- -N in Vertisols is not high in the surface horizons, a result which was also found in this experiment. The amount of soil nitrates accumulated at 0–30 cm, besides being the smallest of the three depths, showed practically no increase over the years. López-Bellido et al. (1997), during the first years of the same experiment, reported higher levels of nitrates in the upper layer, which is consistent with the data obtained in the first year of our study (1995).

As we increased the rate of N fertiliser, the pattern of the vertical distribution of nitrates became less linear in wheat-fallow and wheat-wheat, and major differences were observed between depths (Fig. 5). In the rotations with legumes, such differences between depths were observed for all rates of N. This behaviour was also noted by O'Connor et al. (2010). Nitrate accumulation was consistently higher in the 30–60 cm soil layer in virtually all rotations and rates of N. The tillage system, in contrast, did not follow a predetermined pattern (Fig. 5).

The soil electrical conductivity showed an increase with depth. According Ju et al. (2007), the electrical conductivity shows a highly significant positive correlation with the NO_3^- -N concentration and can also be used as an index of salinisation. He also reported that the substantial accumulation of NO_3^- -N may have been an important factor contributing to soil salinity. According to all the above results, the soil in our experiment would produce a deep saline accumulation due to over-fertilisation of N. López-Bellido et al. (2005) found, for the same experiment, that plants extract a maximum of 50–60% of the N fertiliser applied due to the importance of residual N, with the remaining N being native to the soil. Therefore, N fertiliser rates of over 150 kg N ha^{-1} may have led to continuous over-fertilisation of the crop and a considerable carryover effect of N fertiliser.

5. Conclusions

Nitrate content shows inconsistent behaviour according to the tillage systems used, while the crop rotations clearly show great differences amongst them, with faba bean inducing the highest accumulation of nitrate in the soil.

In the rainfed Mediterranean Vertisol studied, a progressive increase in N content was observed in the soil profile as a function of N fertiliser. Any applied N unused by the crop progressively increases the N reserves in the soil, which can produce an over-fertilisation of the system and a reduced response of the cereal to fertiliser.

The continuous application of high rates of N fertiliser, especially in wet years, can have detrimental effects leading to soil contamination due to nitrate accumulation. In Vertisols, the residual mineral N is stored stably in the soil profile, playing an important role in the practice of fertilisation. Consequently, the best strategy to establish the optimal rate of N fertiliser for wheat should be decided upon according to precipitation and should be based specifically on the amount of residual N in the soil at sowing and in the preceding crop.

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References

Analytical Software, 2005. Statistix 8.1. Tallahassee, FL, USA.

Barnes, H., Folkard, A.R., 1951. The determination of nitrates. *Analyst* 76, 599–603.

Barraclough, P.B., Weir, A.H., Kuhlmann, H., 1991. Factors affecting the growth and distribution of winter wheat roots under UK field conditions, in: McMichael, B.L., Persson, H. (Eds.), *Plant roots and their environment*. Elsevier Science, pp. 410–417.

- Blankenau, K., Olf, H.W., Kuhlmann, H., 2002. Strategies to improve the use efficiency of mineral fertilizer nitrogen applied to winter wheat. *J. Agron. Crop. Sci.* 188, 146–154.
- Borghetti, B., 2000. Nitrogen as determinant of wheat growth and yield, in: Satorre, E.H., Slafer, G.A. (Eds.), *Wheat ecology and physiology of yield determination*. Food products press, New York, pp. 67–84.
- Borin, M., 1997. Effects of agricultural practices on nitrate concentration in groundwater in north east Italy. *Ital. J. Agron.* 1, 47–54.
- Bremer, E., van Kessel, C., 1992. Plant-available nitrogen from lentil and wheat residues during a subsequent growing season. *Soil Sci. Soc. Am. J.* 56, 1155–1160.
- Bremner, J.M., 1965. Inorganic forms of nitrogen, in: Black, C.A., Evans, D.D., Ensminger, L.E., White, J.L., Clark, F.E. (Eds.), *Methods of soil analysis, Part 2*. Agron. Monogr. 9. ASA and SSSA, Madison, WI, pp. 1179–1237.
- Burns, I.G., 1980. Influence of the spatial distribution of nitrate on the uptake of N by plants: a review and a model for rooting depth. *J. Soil Sci.* 31, 155–173.
- Campbell, C.A., Zentner, R.P., Selles, F., McConkey, B.G., Dyck, F.B., 1993. Nitrogen management for spring wheat grown annually on zero-tillage: Yield and nitrogen use efficiency. *Agron. J.* 85, 107–114.
- Connor, D.J., Jones, T.R., 1985. Response of sunflower to strategies of irrigation. II. Morphological and physiological responses to water shortage. *Field Crops Res.* 12, 91–103.
- Corbeels, M., Hofman, G., van Cleemput, O., 1998. Residual effect of nitrogen fertilization in a wheat-sunflower cropping sequence on a Vertisol under semi-arid Mediterranean conditions. *Eur. J. Agron.* 9, 109–116.
- Dinnes, D.L., Karlen, K.L., Jaynes, D.B., Kaspar, T.C., Hatfield, J.L., Colvin, T.S., Cambardella, C.A., 2002. Nitrogen management strategies to reduce nitrate leaching in tile-drained midwestern soils. *Agron. J.* 94, 153–171.

- Eck, H.V., Jones, O.R., 1992. Soil nitrogen status as affected by tillage, crops, and crop sequences. *Agron. J.* 84, 660–668.
- Fageria, N., Baligar, V., 2005. Enhancing nitrogen use efficiency in crop plants. *Adv. Agron.* 88, 97–185.
- Francis, C.A., 1989. Biological efficiencies in multiple-cropping systems. *Adv. Agron.* 42, 1–37.
- García-Torres, L., Romero, F., Mesa, J., 1987. Agronomic problems and chemical control of broomrape (*Orobanche* spp.) in Spain, studies review, in: Weber, H.C., Forstreuter, W. (Eds.), *Proc. of the 4th Symp. on Parasitic Flowering*.
- Garrido-Lestache, E., López-Bellido, R.J., López-Bellido, L., 2004. Effect of N rate, timing and splitting and N type on bread-making quality in hard red spring wheat under rainfed Mediterranean conditions. *Field Crops Res.* 85, 213–236.
- Gómez, K.A., Gómez, A.A., 1984. *Statistical procedures for agricultural research*. Wiley, New York.
- Grant, C.A., Lafond, G.P., 1992. The effects of tillage systems and crop rotations on soil chemical properties of a Black Chernozemic soil. *Can. J. Soil Sci.* 74, 301–306.
- Halvorson, A.D., Reule, C.A., 1994. Nitrogen fertilizer requirements in a annual dryland cropping system. *Agron. J.* 86, 315–318.
- Haynes, R.J., Martin, R.J., Goh, K.M., 1993. Nitrogen fixation, accumulation of soil nitrogen, and nitrogen balance for some field-grown legume crops. *Field Crops Res.* 35, 85–92.
- Herridge, D.F., Marcellos, H., Felton, W.L., Turner, G.L., Peoples, M.B., 1998. Chickpea in wheat-based cropping systems of northern New South Wales III. Prediction of N₂ fixation and N balance using soil nitrate at sowing and chickpea yield. *Aust. J. Agric. Res.* 49, 409–418.
- Huggins, D.R., Pan, W.L., 1993. Nitrogen efficiency component analysis: an evaluation of cropping system differences in productivity. *Agron. J.* 85, 898–905.

- Ju, X.T., Kou, C.L., Christie, P., Dou, Z.X., Zhang, F.S., 2007. Changes in the soil environment from excessive application of fertilizers and manures to two contrasting intensive cropping systems on the North China Plain. *Environ. Pollut.* 2, 497–506.
- Keeney, D.R., 1989. Sources of nitrate to groundwater, in: Follet, R.F. (Ed.), *Nitrogen management and groundwater protection. Developments in agricultural and managed-forest ecology* 21. Elsevier, New York, pp. 23–24.
- Kücke, M., Kleeberg, P., 1997. Nitrogen balance and soil nitrogen dynamics in two areas with different soil, climatic and cropping conditions. *Eur. J. Agron.* 6, 89–100.
- López-Bellido, L., Fuentes, M., Castillo, J.E., López-Garrido, F.J., Fernández, E.J., 1996. Long-term tillage, crop rotation, and nitrogen fertilizer effects on wheat yield under Mediterranean conditions. *Agron. J.* 88, 783–791.
- López-Bellido, L., López-Bellido, F.J., Fuentes, M., Castillo, J.E., Fernández, E.J., 1997. Influence of tillage, crop rotation and nitrogen fertilization on soil organic matter and nitrogen under rain-fed Mediterranean conditions. *Soil Tillage Res.* 43, 277–293.
- López-Bellido, L., López-Bellido, R.J., Castillo, J.E., López-Bellido, F.J., 2000. Effects of tillage, crop rotation, and nitrogen fertilization on wheat under rainfed Mediterranean conditions. *Agron. J.* 92, 1054–1063.
- López-Bellido, R.J., López-Bellido, L., 2001. Efficiency of nitrogen in wheat under Mediterranean conditions: effect of tillage, crop rotation and N fertilization. *Field Crops Res.* 71, 31–46.
- López-Bellido, L., López-Bellido, R.J., Redondo, R., 2005. Nitrogen efficiency in wheat under rainfed Mediterranean conditions as affected by split nitrogen application. *Field Crops Res.* 94, 86–97.
- López-Bellido, L., Muñoz-Romero, V., Benítez-Vega, J., Fernández-García, P., Redondo, R., López-Bellido, R.J., 2012. Wheat response to nitrogen splitting applied to a Vertisols in different tillage systems and cropping rotations under typical Mediterranean climatic conditions. *Eur. J. Agron.* 43, 24–32.

- McConkey, B.G., Curtin, D., Campbell, C.A., Brandt, S.A., Selles, F., 2002. Crop and soil nitrogen status of tilled and no-tillage systems in semiarid regions of Saskatchewan. *Can. J. Soil. Sci.* 489–498.
- McIntosh, M.S., 1983. Analysis of combined experiments. *Agron. J.* 75, 153–155.
- O'Connor, G.E., Evans, J., Black, S., Fettell, N., Orchard, B., Theo, R., 2010. Influence of agronomic management of legume crops on soil accumulation with nitrate. *Nutr. Cycl. Agroecosyst.* 86, 269–286.
- Peoples, M.B., Herridge, D.F., 1990. Nitrogen fixation by legumes in tropical and subtropical agriculture. *Adv. Agron.* 44, 155–223.
- Porter, L.K., Follett, R.F., Halvorson, A.D., 1996. Fertilizer nitrogen recovery in a no-till wheat-sorghum-fallow-wheat sequence. *Agron. J.* 88, 750–757.
- Probert, M.E., Fergus, I.J., Bridge, B.J., McGarry, D., Thompson, C.H., Russell, J.S., 1987. The properties and management of vertisols. C.A.B. International. IBSRAM. U.K., p.36.
- Rao, S.C., Dao, T.H., 1992. Fertilizer placement and tillage effects of nitrogen assimilation by wheat. *Agron. J.* 84, 1028–1032.
- Raun, W.R., Johnson, G.V., 1995. Soil-plant buffering of organic nitrogen in continuous winter wheat. *Agron. J.* 87, 827–834.
- Schlegel, A.J., Grant, C.A., Havlin, J.L., 2005. Challenging approaches to nitrogen fertilizer recommendations in continuous cropping systems in the great plains. *Agron. J.* 97, 391–398.
- Soon, Y.K., Clayton, G.W., Rice, W.A., 2001. Tillage and previous crop effects on dynamics of nitrogen in a wheat-soil system. *Agron. J.* 93, 842–849.
- Stute, J.K., Posner, J.L., 1995. Synchrony between legume nitrogen release and corn demand in the upper midwest. *Agron. J.* 87, 1063–1069.
- Westerman, R.L., Boman, R.K., Raun, W.R., Johnson, G.V., 1994. Ammonium and nitrate nitrogen in soil profiles of long-term winter wheat fertilization experiments. *Agron. J.* 86, 94–99.

Table 1. The properties of the Vertisol used in field experiments. Córdoba (Spain).

	Soil depth (cm)		
	0-30	30-60	60-90
Fine sand (g kg ⁻¹)	127	143	187
Silt (g kg ⁻¹)	179	152	26
Clay (g kg ⁻¹)	694	705	787
pH (water)	7.7	7.6	7.6
Organic matter (g kg ⁻¹)	10.2	7.4	5.3
Calcium carbonate equivalent (g kg ⁻¹)	75	93	71
CEC (cmol kg ⁻¹)	46.5	36.6	30

Table 2. Analysis of variance (mean squares) of the soil nitrate content at sowing as affected by year, tillage system, crop rotation and N rate over 18-year period.

Source	df	Mean squares
Year (Y)	14	218131***
Tillage (T)	1	25905***
Y × T	14	18184***
Rotation (R)	4	414593***
Y × R	56	30946***
T × R	4	16346***
Y × T × R	56	5970***
N rate (N)	3	389615***
Y × N	42	21658***
T × N	3	2772*
R × N	12	51616***
Y × T × N	42	3058***
Y × R × N	168	4670***
T × R × N	12	1408
Y × T × R × N	168	2220***

*, *** Significant at the 0.05 and 0.001 probability level

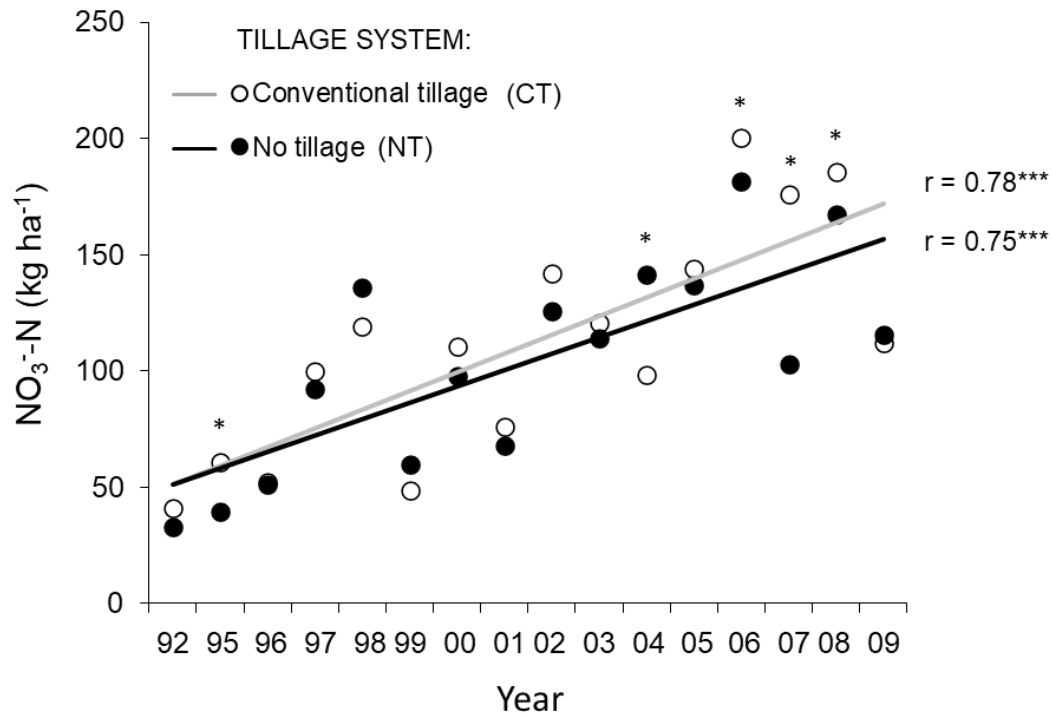


Fig. 1. Development of nitrate content in the soil profile (0-90 cm) as affected by tillage system for a 18-yr experiment at Córdoba in Spain.

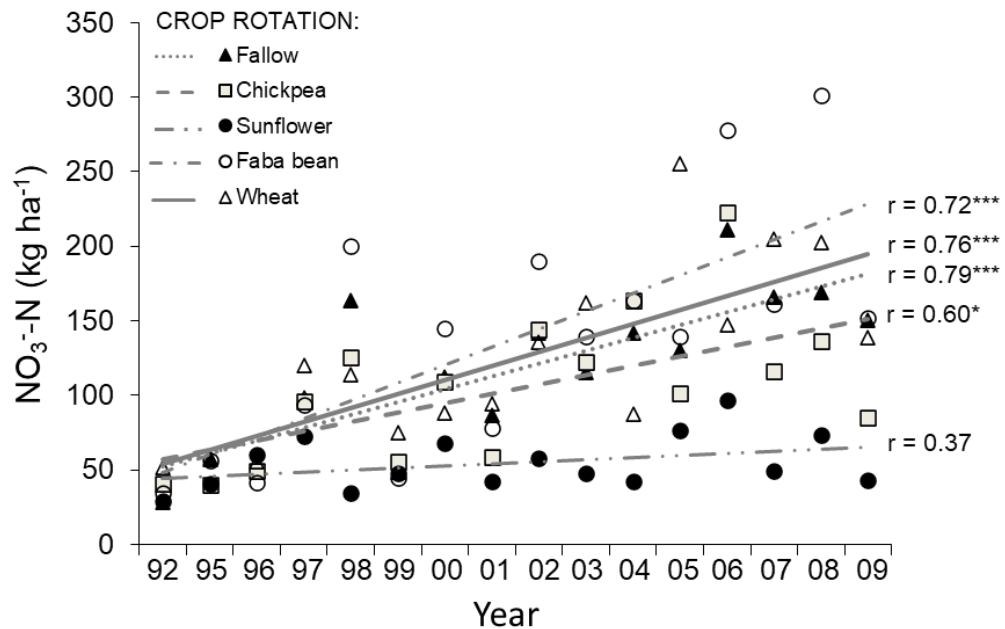


Fig. 2. Development of nitrate content in the soil profile (0-90 cm) as affected by crop rotation (wheat-wheat, wheat-fallow, wheat-chickpea, wheat-faba bean and wheat-sunflower) for a 18-yr experiment at Córdoba in Spain.

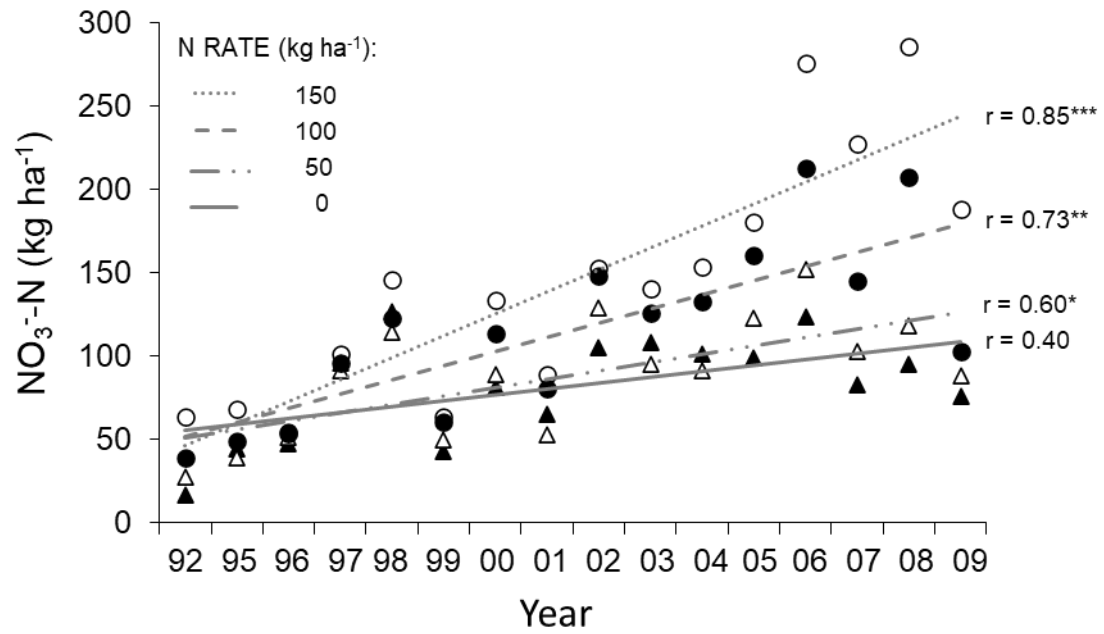


Fig. 3. Development of nitrate content in the soil profile (0-90 cm) as affected by N rate for a 18-yr experiment at Córdoba in Spain.

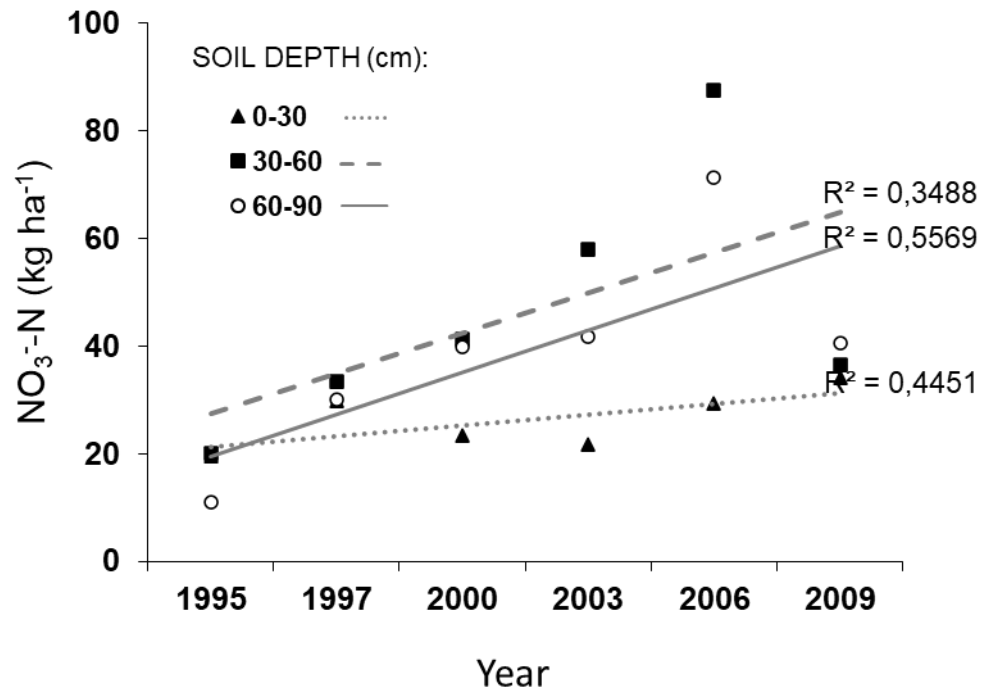


Fig. 4. Development of nitrate content in different soil depths for a 18-yr experiment at Córdoba in Spain.

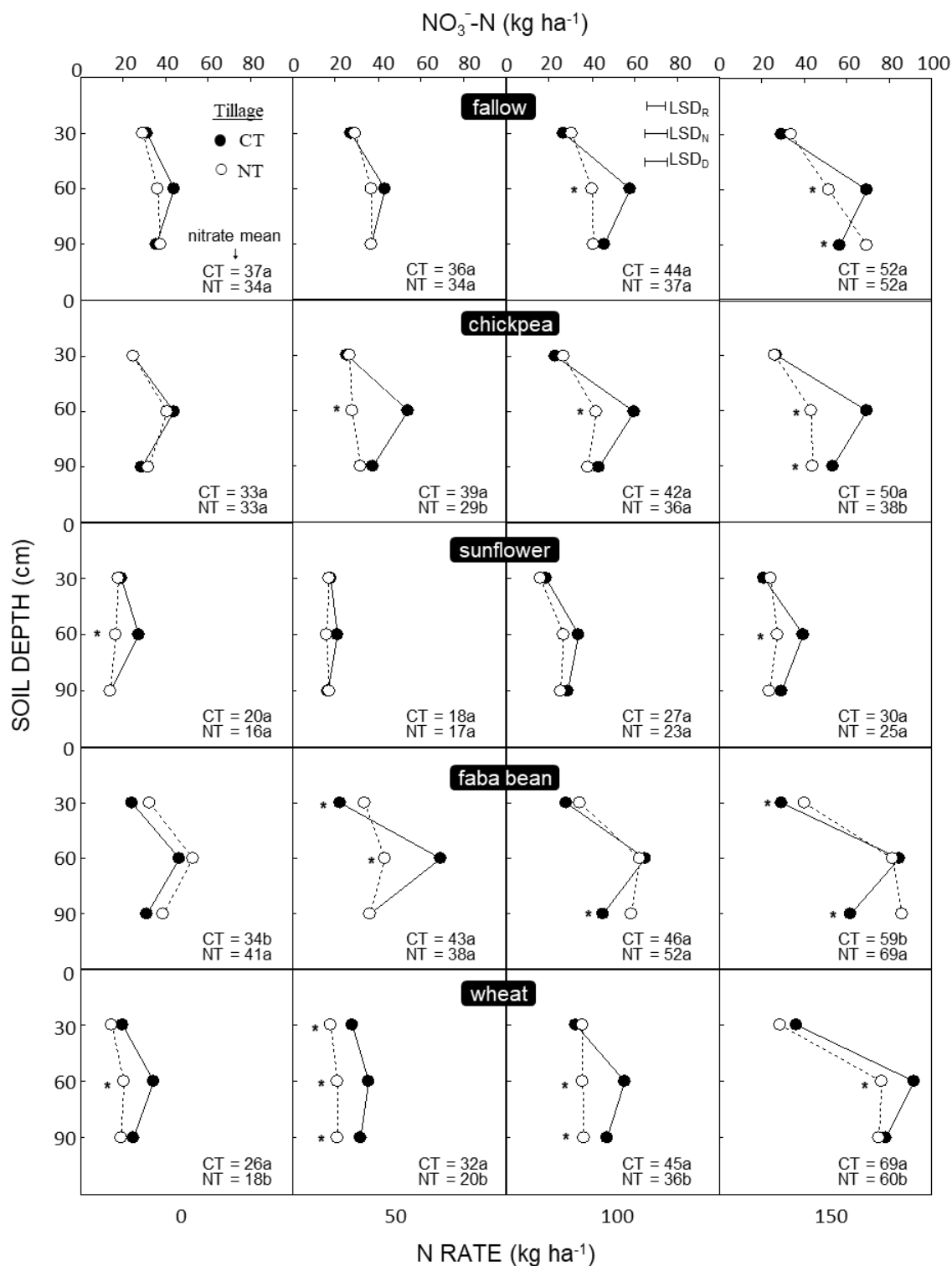


Fig. 5. Soil nitrate content as influenced by tillage system (CT: conventional tillage, NT: no-tillage), crop rotation (wheat-wheat, wheat-fallow, wheat-chickpea, wheat-faba bean and wheat-sunflower), N fertiliser rate and soil depth for a 18-yr period at 3-yr intervals. The asterisk represents significant differences between tillage systems. Horizontal bars represent LSD for comparison: LSD_D, same tillage system, crop rotation and N rate; LSD_N, same tillage system and crop rotation; LSD_R, same tillage system.