The effects of the tillage system on chickpea root growth

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ABSTRACT

A well-developed root system is crucial for plant growth, especially under dryland farming conditions. A two-year field study (2003–2004 and 2005–2006) was conducted to determine the effects of the tillage system on root growth in chickpea (*Cicer arietinum* L.) grown in continuous rotation with wheat (*Triticum aestivum* L.) on a typical Vertisol in southern Spain as part of the long-term "Malagon" experiment begun in 1986. The tillage treatments were either no tillage (NT) or conventional tillage (CT), and the experiment was designed as a randomized complete block with three replications. Both soil cores and a minirhizotron were used to evaluate the root system. Measurements of the root parameters were performed at different depths and included the following: root length, root biomass, root nitrogen and root length density. Root length measurements were performed during five chickpea growth stages. The CT was more favourable than NT for chickpea root development (0.34 mm cm^{-3} versus 0.18 mm cm^{-3}), which is one of the factors that induced higher yields during the drier year. The nitrogen content of the roots represented 15% of the total N extracted by the plant. The measured root lengths were larger when using the soil core method than with the minirhizotron (2.5 mm cm^{-3} versus 1.3 mm cm^{-3}), which can be attributed to the cracks that occur in Mediterranean Vertisols that can separate the tube from the soil, resulting in the underestimation of the root length.

Keywords: no tillage; conventional tillage; Vertisol; root length; root biomass; minirhizotron.

1. INTRODUCTION

Plant roots are a fundamental component of terrestrial ecosystems and are important for balancing water and nutrients in the soil (Spedding et al., 2004). The root system, with its extensive but structured development, is considered to be an evolutionary response to the spatio-temporal variability in resource supply and the associated constraints on growth (Harper et al., 1991). Crop species differ in the rate of root growth (Liu et al., 2011) and how the roots are distributed within the soil profile.

In Mediterranean climates, chickpea is traditionally planted in early spring. Dryland chickpea production is dependent on the irregular and generally scarce rainfall and on the residual soil moisture (López-Bellido, 2008). Crop performance under water-related stress conditions is closely related to the root system development (Abdelhamid, 2010). Under water-limiting conditions, the morphology of crop root systems is a crucial determinant for the capacity for nutrient uptake and water extraction by crop plants (Fageria, 2004), influencing aboveground growth and biomass yield. A root that has developed during the early growth stages of the plant can effectively exploit the water in the soil, especially in semiarid areas in which plant establishment is often limited by low water availability (Lee et al., 1996; Lilley and Kirkegaard, 2007). Roots with a longer length or more tips increase the nutrient supply to the plant to a greater extent than those with shorter roots or fewer root hairs (Dong et al., 1995).

Root traits such as root depth and root biomass (RB) have been identified as the most promising plant traits in chickpea for terminal drought tolerance, as these help extract available soil

moisture (Abdelhamid, 2010). However, Zaman-Allah et al. (2011) indicated that the temporal pattern of water uptake by roots, more than root growth, is critical for understanding water management and the adaption to terminal drought. Several key attributes of chickpea roots, such as their high water absorption efficiency per unit root length density, their ability to change the rooting pattern across soil depths to efficiently access the available soil moisture and their ability to produce a larger root surface area per unit root biomass, seem to make chickpea the best choice for dryland cropping systems compared with other legumes or cereals (Tilahun and Schubert, 2003; Benjamin and Nielsen, 2006).

The minirhizotron system is a non-destructive technique for studying the dynamics of crop root systems. Root system dynamics are instrumental in maintaining the biological and chemical equilibrium within the soil and modulating the changes to soil quality (Zobel and Wright, 2005). Vertisols are fine-textured soils that contain swelling clay minerals, and they can develop wide and deep cracks during prolonged dry seasons. Vertisols have particular management requirements as well as specific problems with tillage. The mechanical impedance and lack of aeration in soil can both be alleviated by conventional tillage, although this practice may accelerate the loss of soil moisture (Agrawal et al., 1989; Gupta and Woodhead, 1989). In contrast, minimal or zero tillage systems and the retention of stubble can improve the soil structure, increase the organic matter content (Blair and Crocker, 2000) and soil water storage capacity (O´Leary and Connor, 1997), improve chemical fertility (Chan et al., 1999) and conserve water (Carroll et al., 1997). However, crops may also suffer from the formation of soil cracks, which can accelerate the loss of soil moisture under zero or minimum tillage (Rathore et al., 1998).

The aim of the present study was to determine the response of chickpea root growth to two different tillage systems within the framework of a long-term field experiment on a rain-fed Vertisol using soil cores and a minirhizotron to estimate the root length, root biomass and root nitrogen during the period of legume growth.

2. MATERIALS AND METHODS

2.1. Site and experimental design

Field experiments were conducted in Córdoba, southern Spain (37º46' N, 4º31' 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 a 3-year period (2003–2004 and 2005–2006); data for 2004–2005 were discarded because a severe drought prevented the installation of minirhizotrons in the soil. The study was conducted within the framework of a long-term experiment named ''Malagón'', started in 1986, and designed as a randomized complete block with a split–split plot arrangement and four blocks. Main plots were tillage system [no-tillage (NT) and conventional tillage (CT)]; subplots were crop rotation, with four 2-year rotations (wheat–sunflower (*Helianthus annuus* L), wheat–chickpea, wheat–faba bean (*Vicia faba* L) and wheat–fallow) and continuous wheat; sub-subplots were N fertilizer rate (0, 50, 100, and 150 kg N ha^{-1}) applied to wheat. Each rotation was duplicated in reverse crop sequence in order to obtain data for all crops on a yearly basis. The area of each sub-subplot was 50 m^2 (10 by 5 m).

Since this study was conducted to independently evaluate the influence of tillage system on chickpea root growth in continuous rotation with wheat, using only the 100 kg N ha^{-1} rate applied to wheat, the design was a randomized complete block with three replications.

2.2. Crop management

No-till plots were seeded with a no-till seed drill. Weeds were controlled with glyphosate + 2-methyl-4-chlorophenoxyacetic acid (MCPA) at a rate of 0.5 + 0.5 L active ingredient ha^{-1} prior to planting. The conventional till treatment included moldboard ploughing (25–30 cm depth) and disk harrowing and/or vibrating tine cultivation (10–15 cm depth) several times to grind clods. The crop residues were not removed by either tillage treatment; residues remained as mulch on NT treatments and were incorporated in CT treatments.

Chickpea (cv. Zoco) was planted in 48 cm-wide rows in February at a seeding rate of 384,600 seed ha^{-1} with an average thousand seeds weight of 260 g. Nitrogen fertiliser (100 kg N ha^{-1}) was applied to the preceding wheat (*Triticum aestivum* L) plots as ammonium nitrate. Half of the N was applied before sowing (incorporated by disk harrowing in conventional till plots and surface broadcast in no-till plots). The remaining N was applied as a top dressing at the beginning of wheat tillering. Each year, the preceding wheat plots were also supplied with P fertiliser as calcium superphosphate at a rate of 65 kg ha^{-1}; the fertiliser was incorporated in conventional till soil and banded with a drill in the no-till plots. Soil-available K was adequate (530 mg kg^{-1}).

At harvest, a 1-m^2 area at the centre of each chickpea plot was sampled. From this sample, aboveground biomass was measured by drying plants at 80 ºC to a constant weight. The

chickpea was harvested in early June each year by using a 1.5-m wide Nursemaster elite plot combine (30 m² per plot).

2.3. Measurements

2.3.1. Soil coring

Cylindrical soil cores were randomly sampled and in triplicate at the centre of each plot and on planting rows, using an 8 cm-diameter bi-partite root auger (Eijkelkamp, NL). The first sample was taken on a line from the centre of the plot and the other two were taken on lines separated by 2-3 meters in the opposite direction. Manschadi et al. (1998) found differences between soil core taken on the row and between rows only in the first 15 cm. We adopted the criterion of taking soil core samples from the sowing line, since this is where the minirhizotron tubes were installed and one of our objectives was to perform a comparative study of the root system using both methods. Each location was sampled at seven depths (0–10, 10–20, 20–30, 30–40, 40–55, 55–70 and 70–85 cm). Sampling was carried out during full flowering of the chickpea (growth stage 65) (Hack et al., 1992). Prior to processing, soil samples were immediately frozen at -30 ºC to avoid root decomposition.

Roots were washed using Calgon (a 10% sodium hexametaphosphate and sodium bicarbonate solution) as a dispersant. After 12 hours in this solution, the roots were rinsed in water and collected on a sieve with a 0.2-mm mesh screen. Debris and dead roots were manually removed from live roots. The criteria of distinguishing live from dead roots are typically based on colour (separating white or pale brown roots from darker materials) and physical appearance (e.g. branched, able to bend, some elasticity) according to Gregory (1994). The roots were scanned and the images were processed to determine length, using the specific image-processing

software package CIAS version 2.0 (CID 2002). They were then dried at 40 ºC for 24 hours and weighed. N content was measured by the Dumas Combustion Method.

The following indices were calculated: root length density (RLD), i.e. length of roots per unit volume of soil (mm cm^{-3}); root biomass (RB), i.e. root weight per unit area (kg ha^{-1}); and root N content, i.e. the amount of N in the roots per unit area (kg ha^{-1}).

2.3.2. Minirhizotron

Measurements of the root length (RL) were performed using the CI-600 root growth monitoring system (CID, Inc. Camas, WA 98607 USA) fitted with a scanner head for collecting images, a laptop computer with the CI-400 Computer Image Analysis Software (CIAS) and standard clear 1.8-m soil tubes (50.8 mm internal diameter) with end caps. After the emergence of the chickpea plants, tubes were installed permanently at the centre of each plot on the sowing line, 45º from the vertical as recommended by Johnson et al. (2001). An auger of the same external diameter as the tube was used to facilitate close tube/soil contact. The scanner was then inserted into each tube to a depth of 100 cm. Images were captured with the aid of an automatic indexing handle at six depths, which were equivalent to 0–15, 15–30, 30–50, 50–65, 65–80 and 80–100 cm (given the angle of the tube at 45º off the vertical). Images were captured at the following stages of development (Hack et al., 1992): 5–6-leaves (16), 8–10-leaves (19), full-flowering (65), pod-setting/filling (75) and pod-ripening stages (89). Measurements were performed between March and May in each of the two study years.

The images were processed with the WinRhizotron® software (Regent Instruments Inc.), which provided values for the RL (mm cm^{-2}) for each plot and each chickpea growth stage under the two tillage systems tested.

To compare the two methods, the root length, as measured using the minirhizotron, was converted to units of volume using the empirical conversion method described by Itoh (1985). This method assumes that the RL of all of the roots around the frame of the scanned tube is observed at a depth of field of 3 mm. The RL per unit soil volume is obtained using the equation RLD = L/(AxDOF), where RLD is the volumetric root length density (m m^{-3}), L is the root length observed in the minirhizotron frame (m), A is the minirhizotron frame area observed (m^2) and DOF is the depth of field (m).

2.4. Statistical analysis

The annual data for each variable over the total 2-yr period were subjected to analysis of variance (ANOVA), using a randomized block design combined over years and an error term according to McIntosh (1983). Year was considered a random effect, whilst tillage system, soil depth and growth stage considered fixed effect. Means were compared using Fisher's protected least significant difference (LSD) test at $P < 0.05$. The LSDs for comparisons of the different main effects and interaction terms were calculated using the appropriate standard error terms. The Statistix v. 8.1 (Analytical Software, 2005) package was used for this purpose.

2.5. Weather conditions

The rainfall and its distribution varied considerably between the two years (Fig. 1). The 2003–2004 year had 704 mm of rainfall, while the 2005–2006 year had only 402 mm of rain. The average annual rainfall in the area over 30 years was 584 mm. The rainfall in the month prior to sowing during each year was 13 and 66 mm in 2005–2006 and 2003–2004, respectively. The

2004–2005 year was discarded because the low rainfall (263 mm) resulted in the soil not having sufficient moisture to install the minirhizotron.

3. RESULTS

3.1. Biomass yield and N uptake

Grain yields and N uptake during the 2003–2004 year were approximately twice those from the 2005–2006 year (Table 1). In the 2003–2004 year, there were no differences between crop biomass and N uptake related to the tillage system (Table 1). In 2005–2006, there was a higher grain yield, straw yield and grain N uptake with conventional tillage (CT) compared with no tillage (NT) (Table 1).

3.2. Root length

Each of the treatments resulted in significant differences for the root length (RL) (Table 2). In 2003–2004, the values were higher than those in 2005–2006 (0.33 mm cm^{-2} versus 0.19 mm cm^{-2}, respectively). In addition, the RL under CT was significantly higher than under NT (0.34 and 0.18 mm cm^{-2}, respectively).

The RL increased progressively from sowing to flowering in both years of the study. The longest RL was observed at the flowering stage followed by the pod-setting/filling, pod-ripening, 8–10-leaves and finally 5–6-leaves stage (Fig. 2). The differences between the different phenological stages were significant except for those between the pod-ripening and the 8–10-leaves stages. In 2003–2004, there was a marked significant decrease in the RL after

flowering until maturity. In contrast, in 2005–2006, there was no such decrease in the RL, and the RL at the 5–6-leaves growth stage of 2005–2006 was almost zero (Fig. 2).

In general, during the two years of the study and during all growth stages, the RL decreased as the soil depth increased (Fig. 2). The percentage of the RL in the first layer of soil (0–15 cm) was higher in 2003–2004 than in 2005–2006 (38% and 27%, respectively), whereas in the deeper layers (65–100 cm), the opposite trend was observed (3% and 8% for the years 2003–2004 and 2005–2006, respectively). This behaviour was most evident at the pod-setting/filling stage (Fig. 2).

At the chickpea of 5–6- and 8–10-leaves growth stages, there were few significant differences as a result of the tillage system during the two years of the study (Fig. 2). However, after 8–10 leaves, there were significant differences in favour of CT in the most superficial layers (Fig. 2).

3.3. Root biomass

Overall, there was no significant difference in the root biomass (RB) with respect to year or tillage system. However, the RB in 2003–2004 was higher with NT than with CT in the surface soil layer (0–10 cm) and was the lowest with NT in the next layer (10–20 cm) (Fig. 3). In 2005–2006, there were no significant differences between tillage systems. As with RL, the RB decreased with increasing soil depth in both years (Fig. 3).

In 2003–2004, the RB was 368 kg ha^{-1}, whereas in 2005–2006, it was 279 kg ha^{-1}. There were no significant differences between the years. In 2003–2004, 84% of the RB was concentrated

in the top 30 cm of soil, while in 2005–2006, only 69% was. In the deepest 45 cm of the soil profile, the RB was three times higher in 2005–2006 than in 2003–2004.

The root:shoot biomass ratio was 0.14 and 0.18 in 2003–2004 and 2005–2006, respectively. This ratio was 0.14 and 0.20 for CT and NT, respectively.

3.4. Root nitrogen

The concentration of nitrogen in the roots did not differ between treatments. The differences that were found are likely because the total amount of root nitrogen (RN) depends directly on the RB. Consequently, the RN behaviour pattern was similar to the RB pattern. The RN was higher under conditions of NT than under CT in the first layer of soil (0–10 cm), but the opposite trend occurred in the second layer (10–20 cm). In 2003–2004, RN was 9.3 kg N ha^{-1} in the entire soil profile (0–85 cm), while in 2005-2006 it was 7.2 kg N ha^{-1}, although this difference was not significant (Table 1). The RN did not differ in the 3 most superficial soil layers (0–10, 10–20 and 20–30 cm), but it decreased progressively from 30 cm to the deepest layer (70–85 cm). Root nitrogen accounted for 13% and 16% of total N extracted by the plant in the years 2003–2004 and 2005–2006, respectively.

3.5. Comparison between the soil core and minirhizotron methods

During the flowering stage in both years of the study, the root length density (RLD) obtained using the soil core method was higher than that obtained using the minirhizotron (2.3 versus 1.7 mm cm^{-3} in 2003–2004 and 2.7 versus 0.9 mm cm^{-3} in 2005–2006). In the wetter year (2003–2004), the RLD did not differ between the two methods except in the first soil layer (0–

10 cm) (Fig. 4). In the drier year (2005–2006), there were differences in the first 55 cm of the soil (Fig. 4). In both years, the values were typically higher using the soil core than with the minirhizotron method.

4. DISCUSSION

Greater rainfall during the chickpea-growing season in 2003–2004 led to the largest values for RL throughout the entire experiment. Liu et al. (2011) indicated that chickpea had longer roots and a larger number of tips under high water conditions than under low water conditions, although statistical differences were observed only at the 0–40 cm depth. In our case, there were no significant inter-annual differences in the deeper profiles. Similarly, Benjamin and Nielsen (2006) found that irrigation increased the root surface area density for chickpea only in the topsoil layer.

Chickpea has the ability to change its root distribution across soil depths depending on the soil moisture availability (Abdelhamid, 2010). In our study, the concentration of roots in the most superficial soil layer was higher in the wettest year, in contrast with the deeper layers, where the proportion of roots was higher in the driest year. This trend was most evident at the pod-setting/filling stage. Ali et al. (2002) and Benjamin and Nielsen (2006) also indicated that the proportion of RLD distributed at deeper soil layers was shown to be higher under lower soil moisture conditions. Given the progressively decreasing moisture content and (increasing temperature) of typical chickpea growing environments, the ability to maximise the extraction of water from the soil, particularly during the pod-filling stage, should provide an important advantage, making the root system an essential part of drought tolerance along with early or appropriate maturity (Serraj et al., 2004).

Optimal crop establishment is critical for the crop's further development, and a key factor in this process is the soil moisture. In the drier year (2005–2006), almost no root system was observed until the stage of 8–10 leaves. This phenomenon could be attributed to a higher amount of rainfall recorded in the wetter year during the month prior to sowing (2003-2004, 5 times higher than in the drier year) because during the sowing period and the 5–6 leaves stage, the rainfall amount was similar for both years.

Overall, CT enhanced the root development of chickpea. Rathore et al. (1998) also found a higher root density under CT when compared with NT. The differences between tillage systems become remarkable beginning at the flowering stage. Rathore et al. (1998) attributed these differences to a higher soil temperature and water availability during the flowering and grain-filling stages under CT compared with NT. During the wetter year, water was not limited during the critical crop development stages, so the yield of chickpea was similar in both tillage systems although the RL was shorter with NT than with CT. In contrast, during the drier year, the large amount root development with the CT system induced higher availability of water which led to yields that were much higher than with NT.

The RB during the wetter year was closely related to the RL during flowering, except in the first layer of soil (0–10 cm). In this layer, the RB, unlike the RL, was higher under the NT system than under the CT system. During the same year and at the same location, Muñoz et al. (2011) also reported a longer RL in faba-bean with CT than with NT at the beginning of flowering and an increased RB in the first layer of soil under NT during flowering. In this case, CT produced more root growth per unit of RB in the most superficial layer of soil, which benefits the most from tillage (aeration, number of pores, etc.). This result could mean that

plants have smaller diameter roots under CT and larger diameter roots under NT, which may be attributable to an increased resistance to root penetration in the first layer of soil for NT compared with CT. Rathore et al. (1998) found increased resistance to penetration in NT than in CT for chickpea cultivation. According to Shein and Pachepsky (1995), under these conditions, roots have a tendency to thicken.

Jackson et al. (1996) found that mature crops had a root:shoot biomass ratio of approximately 0.1. Gan et al. (2009) reported a ratio of 0.2 at maturity for chickpea. The values obtained in our experiment were between 0.12 and 0.22. Gregory (2006) indicated that the root: shoot biomass ratio increased as conditions became harsher. In our case, this ratio was higher when the year was drier and under the NT system, which indicates unfavourable conditions for the roots under these conditions. Klepper (1992) also reported that under limited soil moisture conditions, root growth is reduced less than shoot growth.

The quantity of root nitrogen (RN) is important because it is part of the contribution of the roots to soil fertility, especially in rain-fed systems in which organic matter is typically scarce. Under our experimental conditions, the root nitrogen concentration was very consistent. Differences in RN were due to the amount of RB obtained with the different treatments. The N in the recovered roots represented 15% of the total N extracted from the plant. In an earlier study, Khan et al. (2002) found 11% of the total N in the roots.

The RLD was higher when measured with the soil core method in the first layer of soil during the two years of study. Most studies have found that roots measurements made with a minirhizotron in the upper soil layer are underestimated because the installation of the minirhizotron tube prevents a good soil-tube contact surface. The differences that existed

throughout most of the profile in the drier year could be due to the cracks that clay soils develop as they dry, which allow light to penetrate deep into the soil and possibly into the minirhizotron tube (Dubach and Russelle, 1995).

5. CONCLUSIONS

Conventional tillage favoured an increased root length in chickpea. During the wetter year, in the absence of water restriction, the grain yields tended to be similar under CT and NT. In contrast, during the drier year, the larger root development under CT induced higher yields than under NT. The root:shoot biomass ratio increased both when the year was drier and under NT. This result would indicate unfavourable conditions under which the plants respond by increasing their RB rather than their aboveground biomass.

The root length obtained using the soil core method was longer than the value obtained with the minirhizotron. This variation was more pronounced in the drier year, when it was observed through the 55-cm soil depth. However, in the wetter year, the disparity was seen only in the first layer of soil. These differences are attributed to cracks that Mediterranean Vertisols develop under dry conditions and that lead to the separation of the soil from the minirhizotron tube, resulting in an underestimation of the RL.

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Table 1. Chickpea biomass and N uptake as affected by year and tillage in a continuous rotation with wheat at Córdoba (Spain).

Year	Tillage system	Biomass (kg ha^{-1})			N uptake (kg ha^{-1})		
		Grain	Straw	Roots	Grain	Straw	Roots
2003-2004	CT	1343a†	1431a	337a	52a	15a	7.8a
	NT	1104a	1258a	398a	39a	17a	10.8a
2005-2006	CT	903a	1129a	306a	32a	16a	8.0a
	NT	393b	729b	251a	12b	15a	6.4a

†Within treatment (year and tillage) means followed by the same letter are not significantly different at P< 0.05 according to LSD.

Table 2. Significant effects of year, tillage system, soil depth and growth stage on root length; and year, tillage system and soil depth on root biomass and root nitrogen in chickpea crop over 2 year period.

Source	Root length (mm cm^{-2})	Root biomass (kg ha^{-1})	Root nitrogen (kg ha^{-1})
Year (Y)	*	ns	ns
Tillage (T)	**	ns	ns
Y × T	ns	ns	ns
Soil Depht (D)	***	***	***
Y × D	**	**	ns
T × D	*	*	**
Y × T × D	ns	*	*
Growth stage (S)	***		
Y × S	***		
T × S	***		
D × S	***		
Y × T × S	**		
Y × D × S	***		
T × D × S	***		
Y × T × D × S	***		

*, **, *** Significant at the 0.05, 0.01 and 0.001 probability level
ns not significant

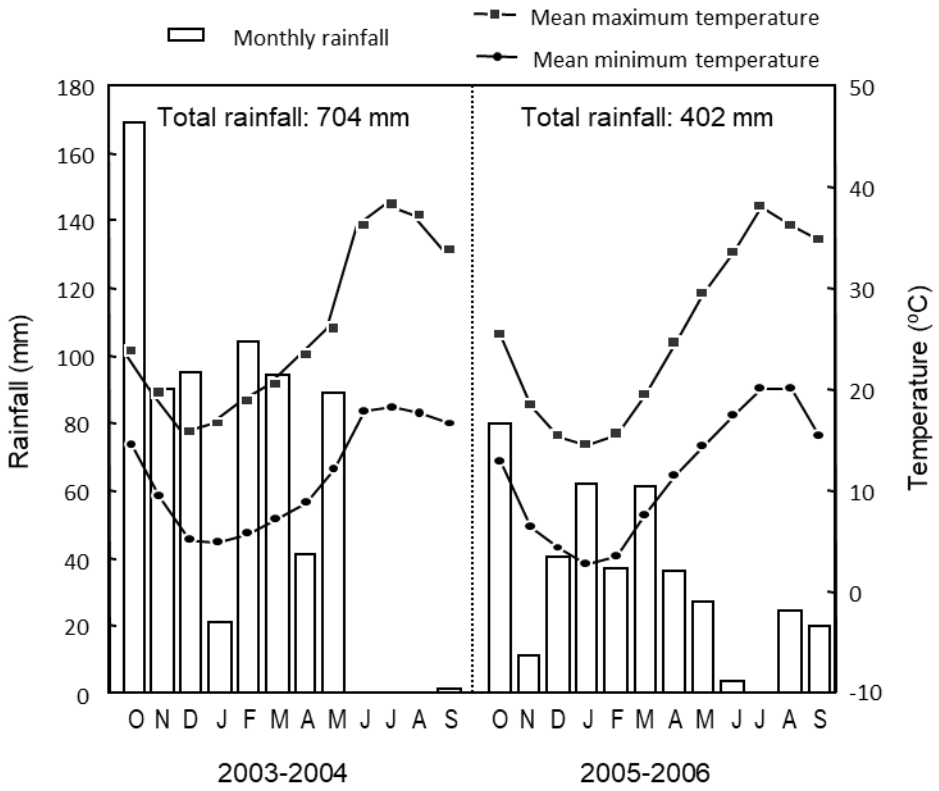

Figure 1. Monthly and annual rainfall, mean maximum and minimum temperatures over the 2-year study period at Córdoba (Spain).

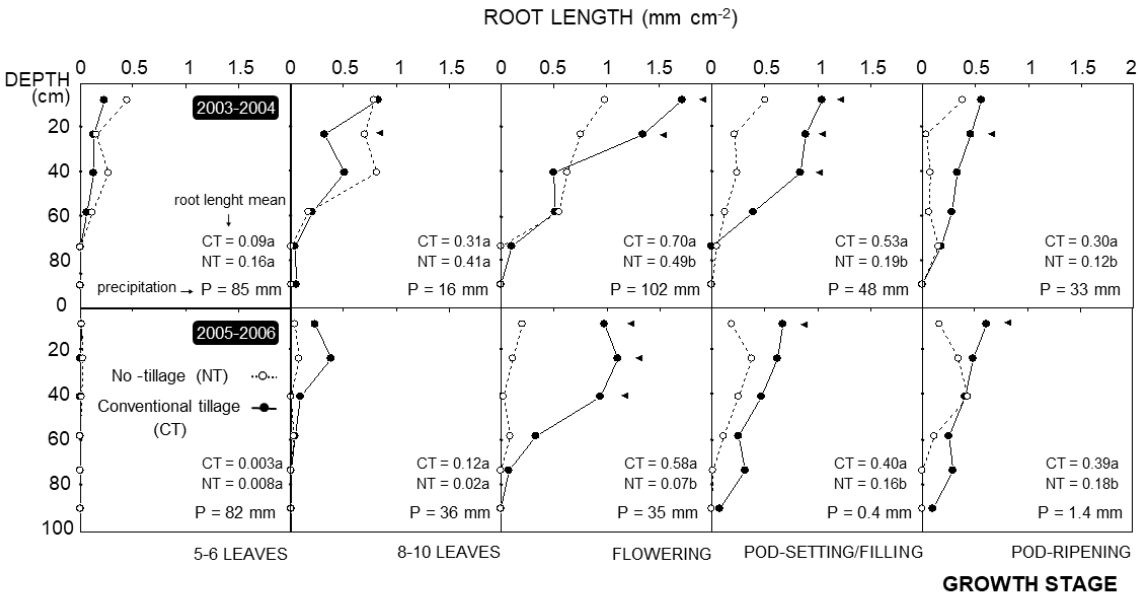

Figure 2. Chickpea root length as influenced by year and tillage system (CT: conventional tillage, NT: no tillage) for different soil depths and growth stages (Hack et al., 1992). Precipitation represents the amount between the growth stage indicated and the preceding one. The triangle (◂) represents significant difference between tillage systems.

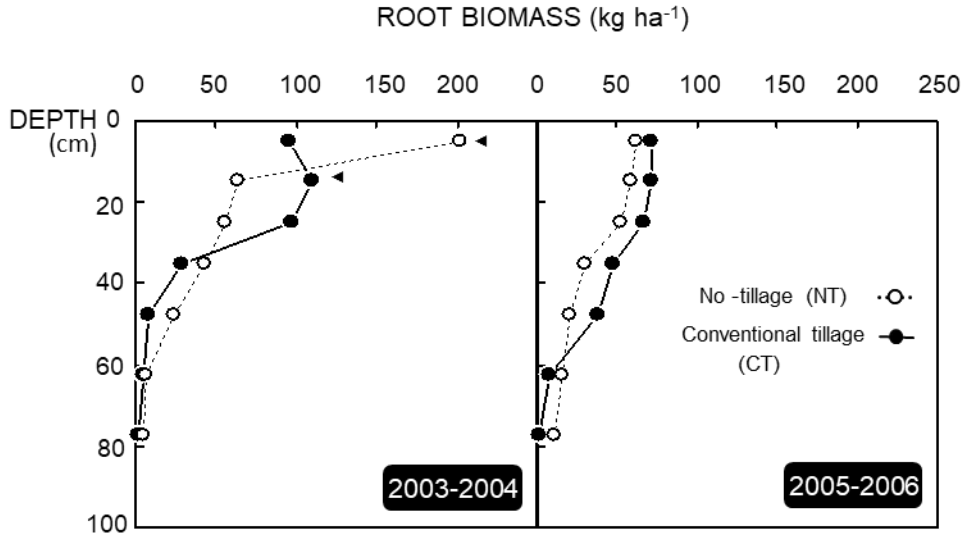

Figure 3. Chickpea root biomass as influenced by year and tillage system for different soil depths. The triangle (◂) represents significant differences between tillage systems.

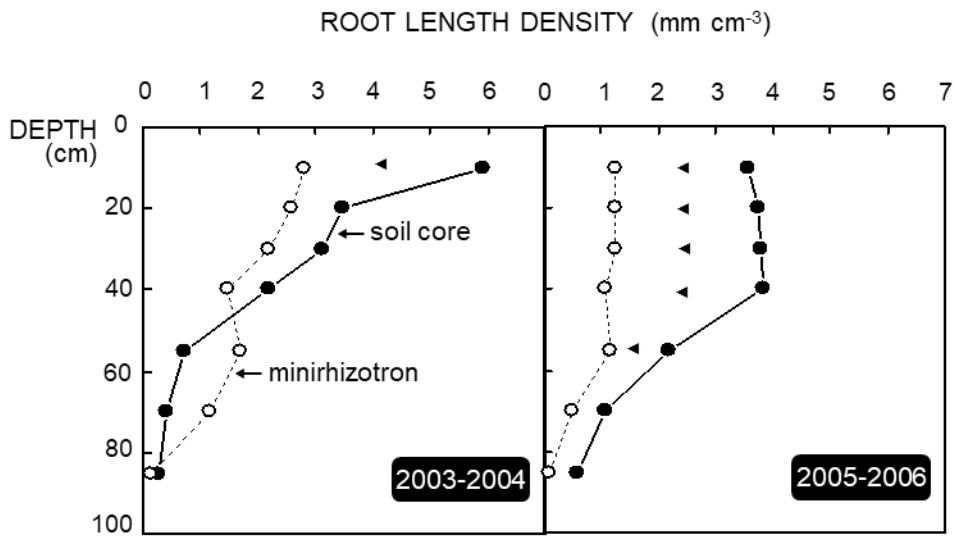

Figure 4. Chickpea root length density as influenced by method (soil core and minirhizotron) for different soil depths. The triangle (◂) represents significant differences between methods.