Sowing time and tillage practice affect chickpea yield and nitrogen fixation 2. Nitrogen accumulation, nitrogen fixation and soil nitrogen balance

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Summary. Following long-term studies at Warra, on the western Darling Downs, chickpea (*Cicer arietinum*) was selected as a useful grain legume cash crop with potential for improvement of its nitrogen (N) fixing ability through management. This 2-year study examined the effect of sowing time and tillage practice on dry matter yield, grain yield (Horn *et al.* 1996), N accumulation, N₂ fixation, and the subsequent soil N balance. Generally, greater N accumulation resulted from sowing in late autumn–early winter (89–117 kg N/ha) than sowing in late winter (76–90 kg N/ha). The amount of N₂ fixed was low in both years (15–32 kg N/ha), and was not significantly affected by sowing time or tillage. The

Introduction

In grain legumes, nitrogen (N) accumulation is closely linked to biomass accumulation and seed growth (Muchow et al. 1993), but there is need for quantitive information specific to chickpea (Cicer arietinum) in Australian conditions. In Australian trials across a range of tropical and subtropical environments, using 3 grain legumes (not chickpea), Muchow et al. (1993) found that net N accumulation was linearly related to net aboveground biomass. Early sowing of chickpea resulted in greater chickpea biomass (Siddique and Sedgley 1986; Horn *et al.* 1996) and could be expected to result in greater N accumulation if chickpea behaves as other grain legumes (Muchow et al. 1993). Evidence suggests that the soil nitrate (NO_3) level is critical to the initiation of chickpea nodulation and affects dry matter (DM) accumulation and distribution, and N₂ fixation, with N₂ fixation decreasing as soil NO₃-N increases (Doughton et al. 1993; Herridge et al. 1995). The effect of tillage practice on N accumulation and N₂ fixation by chickpea does not appear to have been studied, but N₂ fixation by

potential for N_2 fixation was reduced in both years due to high initial soil nitrate levels and low total biomass of chickpea because of low rainfall. Nitrogen accumulation by grain was higher under zero tillage (ZT) than conventional tillage (CT) for all sowing times, and this affected the level of grain N export.

The consequence of low N_2 fixation and high N export in chickpea grain was a net loss of total soil N, (2-48 kg N/ha under CT and 22-59 kg N/ha under ZT). Management practices to ensure larger biomass production and lower soil nitrate-N levels may result in increased N_2 fixation by chickpea and thus a positive soil N balance.

soybean was found to be higher under zero tilage (ZT) (236 kg N/ha) than under conventional tillage (CT) (132 kg N/ha) (Herridge and Bergersen 1988).

The objective of this part of our study was to examine the influence of sowing time and tillage practice on N accumulation and N_2 fixation by chickpea and its subsequent contribution to the overall soil N balance.

Materials and methods

Details of the trial site, experimental design, cultural practice and statistical analysis are reported in Horn *et al.* (1996). Briefly, this 2-year study was conducted on a Vertisol, which had been cultivated for 57 years mainly for cereal cropping and contained 0.072% total N in the 0-10 cm depth. The field experiment was established in May 1992. The initial treatments were CT and 3 sowing dates of chickpea with 8 replicates, arranged in a randomised complete block. The ZT treatments commenced after the wheat crop harvest in November 1992. In 1993, the treatments were 3 sowing dates of chickpea by 2 tillage treatments

(CT as in 1992 and ZT) by 4 replicates. Desi type chickpea (cv. Barwon) was sown at 65 kg/ha on 1 May, 27 May and 7 August in 1992, and 28 April, 8 June and 21 July in 1993. Chickpea seed was inoculated with appropriate rhizobium, treated with Apron (a.i. metalaxyl, at 1.5 g/kg seed) to control phytophthora root rot, and then sown with 222 kg/ha of granulated single superphosphate with copper (Cu) and zinc (Zn), to supply 18 kg phosphorus/ha, 2 kg Cu/ha and 2 kg Zn/ha.

Soil sampling and analysis

Soils were sampled for moisture and NO₃-N before sowing and after harvest in 1992 and 1993, with an additional sampling during the fallow in February 1993. Two cores were taken per plot to 150 cm depth, segmented at 10-cm intervals to 30 cm and at 30-cm intervals to 150 cm. Cores were then bulked to 1 sample per plot per depth.

Soil was air-dried at 35°C and ground to <2 mm. Soil NO₃-N was extracted by shaking 10 g of soil for 1 h in 100 mL of 2 mol KCl/L, and after filtration through Whatman No. 40 paper, the extract was analysed by an automated method (Best 1976).

Plant sampling and analysis

Samples of chickpea were taken at intervals during the growing season, from flowering to harvest, to establish the time of maximum DM yield, in order to estimate the time of maximum N₂ fixation. Aboveground plant samples were collected from 2 m of row in each plot. Flowering was defined as 50% of plants having commenced flowering. Samples for ¹⁵N natural abundance were also taken at the time of DM sampling. A chickpea plant and a reference milk thistle (*Sonchus oleraceus* L.) plant (at least 30 cm apart) were sampled in duplicate in both halves of each plot. The same procedure was followed in both years. Milk thistle was largely absent from the 27 May 1992 sowing as a result of tillage during the sowing operation.

Plant samples were dried at 65° C and ground to <0.5 mm before analysis for total N using Kjeldahl digestion and automated analysis (Buresh *et al.* 1982). Analysis of ¹⁵N was undertaken as described by Doughton *et al.* (1993).

Nitrogen fixation

 $\delta^{15}N$ value. One $\delta^{15}N$ unit, expressed in parts per thousand, is the difference between sample ¹⁵N and atmospheric ¹⁵N (0.3663 atom % ¹⁵N) divided by atmospheric ¹⁵N. $\delta^{15}N$ values for chickpea and milk thistle were calculated from:

 δ^{15} N = 1000 [(atom % ¹⁵N sample - 0.3663)/0.3663].

 Table 1. Effect of sowing time on total N and grain N accumulation

 (kg N/ha) by chickpea at maximum dry matter yield in 1992 and 1993

Sowing date	Total N	Grain N	
	1992		
1 May	115	79	
27 May	92	78	
7 August	90	47	
l.s.d. $(P = 0.05)$	n.s.	21	
	1993		
28 April	89	64	
8 June	117	73	
21 July	76	30	
1.s.d. $(P = 0.05)$	27	21	

%*Ndfa.* Nitrogen derived from the atmosphere by chickpea was calculated from Ledgard and Peoples (1988):

%Ndfa = 100
$$\left(\frac{\delta^{15}N \text{ reference plant} - \delta^{15}N \text{ chickpea}}{\delta^{15}N \text{ reference plant} - 2.10}\right)$$

where $\delta^{15}N$ reference plant and $\delta^{15}N$ chickpea are the ¹⁵N atom % of N in milk thistle and chickpea tops, and the $\delta^{15}N$ of fixed N for chickpea is 2.10 (Doughton *et al.* 1993).

 N_2 fixed. The amount of N₂ fixed (kg/ha) was calculated using chickpea crop N yield (kg/ha) and %Ndfa by chickpea, both measured at maximum DM yield:

 N_2 fixed = N yield x (%Ndfa/100).

Table 2. Effect of sowing time and tillage practice on total N, grain N and vegetative N accumulation (kg N/ha) by chickpea in 1993 CT, conventional tillage; ZT, zero tillage

Sowing	Flow	ering	Max. D	M stage	Har	vest	
date	CT	ZT	CT	ZT	CT	ZT	
Total N							
28 April	40.0	62.8	97.3	80,9	_		
8 June	34.9	66.4	121.3	113.3		_	
21 July	67.0	56.7	78.8	73.5		_	
l.s.d. $(P = 0.05)$							
Sowing date x tilla	nge 3	0.7	n.	s.	-		
		Grain	N				
28 April	—	_		_	38.8	78.8	
8 June	—			_	67.0	74.4	
21 July	—		_		29.6	44.9	
l.s.d. $(P = 0.05)$							
Sowing date x tilla	ige -		-	_	3	1.0	
Vegetative N							
28 April	40.0	62.8	27.6	21.8	_	—	
8 June	34.9	66.4	46.6	41.8		—	
21 July	67.0	56.7	49.7	43.0	_		
l.s.d. $(P = 0.05)$							
Sowing time x tilla	nge 3	30.7	n	1.8.		_	



Figure 1. The relationship between DM and N accumulation by chickpea in 1992 and 1993. \bullet Early plantings, \blacksquare mid plantings, \blacktriangle late plantings. $y = 22.766 + 0.016 \times DMA (r^2 = 0.84, P < 0.05).$

Results

Nitrogen accumulation

The greatest total N accumulation was achieved from the 1 May 1992 and 8 June 1993 sowings. The smallest N accumulation resulted from the late sowings in both years.

There was no significant difference in total N accumulation between sowing times in 1992, but in 1993, total N accumulation from the 8 June sowing was significantly greater than from the other 2 sowings (Table 1). Total N accumulation through the growing season was generally greater under ZT, though this trend was reversed at the time of maximum dry matter yield (Table 2). The sowing time x tillage interaction was significant in the 8 June sowing at flowering, with greater total N accumulation under ZT than CT (Table 2).

Table 3. Effect of sowing date and tillage practice on $\delta^{15}N$ (parts per thousand) of chickpea and milk thistle, and %Ndfa by chickpea in 1993

CT, conventional tillage; ZT, zero tillage

Sowing date	δ ¹⁵ N				%Ndfa	
	Milk thistle		Chickpea			
	CT	ZT	CT	ZT	СТ	ZT
28 April	13.6	11.3	9.4	9.4	34.2	27.9
8 June	11.3	11.1	10.2	10.5	14.9	13.6
21 July	10.5	11.2	8.4	7.3	17.6	30.3
1.s.d. $(P = 0.05)$	3.0			n	.S.	

Table 4. Effect of sowing date (not significant) on %Ndfa and N_2 fixation in 1992 and 1993

%Ndfa	N ₂ fixation (kg N/ha)
1992	
11.8	13.6
13.7	12.6
16.0	14.4
1993	
31.0	29.5
14.3	17.0
24.0	19.0
	%Ndfa 1992 11.8 13.7 16.0 1993 31.0 14.3 24.0

Grain N accumulation by chickpea was significantly higher in the earlier sowings (65% in 1992 and 113% in 1993) than the late sowings (Table 1). Greater grain N accumulation occurred under ZT (mean 66 kg N/ha) than under CT (mean 45 kg N/ha) in 1993 (Table 2). The sowing time x tillage interaction was significant only for the 28 April sowing (Table 2).

Accumulation of N by vegetative matter (total DM minus grain) reached a peak during pod fill in both years and then declined. The sowing time x tillage interaction was significant only at flowering in the 8 June 1993 sowing, where chickpea accumulated 90% more N under ZT (66.4 kg N/ha) than under CT (34.9 kg N/ha) (Table 2). Nitrogen accumulation under ZT was 31% higher (mean 62.0 kg N/ha) than under CT (mean 47.3 kg/ha) at flowering.

Chickpea N accumulation and DM were linearly related, with slightly higher N concentration in 1992 than in 1993 (0.018 kg N/kg DM in 1992 v. 0.013 kg N/kg DM in 1993) (Fig. 1).

Nitrogen fixation

In both years, %Ndfa was similar for all sowing times, except for a trend to lower %Ndfa in the 8 June 1993 sowing (Table 3). There was no tillage effect or sowing time x tillage interaction for %Ndfa in 1993 (Table 3).

Table 5. Effect of sowing date and tillage practice on N_2 fixation and N balance (kg/ha) in 1993

CT, conventional tillage; ZT, zero tillage

Sowing date	Tillage practice	N ₂ fixation	Grain N export	Soil N gain
28 April	CT	32	34	-2
-	ZT	27	79	-52
8 June	CT	19	67	-48
	ZT	15	74	-59
21 July	CT	15	30	-15
	ZT	23	45	-22
1.s.d. $(P = 0.05)$		n.s.	31	55

The amount of N₂ fixed (mean values across sowing times) was in the range 12.6–14.4 kg N/ha in 1992 and 17.0–29.5 kg N/ha in 1993 (Table 4). There was no sowing time x tillage interaction for N₂ fixed by chickpea in 1993 (Table 5). All treatments showed a loss from the soil N pool, with significantly greater grain N export occurring from ZT (79 kg N/ha) than from CT (34 kg N/ha) in the 28 April sowing (Table 5).

Discussion

Nitrogen accumulation

In grain legumes, N accumulation is closely linked to biomass accumulation and seed growth (Muchow *et al.* 1993). This experiment shows that chickpea exhibits a similar relationship (Fig. 1). The relationship exists irrespective of sowing time or the fact that lower total N accumulation occurred in later sowings than in earlier sowings (Table 1). This suggests that agronomic influences such as available soil water may have a major influence on biomass accumulation and subsequently on N accumulation. This is confirmed by Beech and Leach (1988), who reported that in below- and above-average rainfall conditions N accumulation (calculated) was 38 and 177 kg N/ha, respectively.

Seed N accumulation followed a similar pattern to total N accumulation, with greater accumulation in the earlier sowings than the late sowings. The importance of adequate moisture is highlighted in studies by Beech and Leach (1988) on a Vertisol at Dalby. Below-average rainfall resulted in seed N accumulation of 96 kg N/ha (seed yield 1990 kg/ha) and above-average rainfall in seed N accumulation of 232 kg N/ha (seed yield 5060 kg/ha).

Similar trends occurred in vegetative N accumulation, with greater N accumulation occurring in the autumn sowings than in the late winter sowings, as a result of greater DM accumulation in the autumn sowings. This concurs with Beck *et al.* (1991) and Beech and Leach (1988).

As with total DM yield, the effect of tillage practice on N distribution was evident only at harvest; 47% greater mean N accumulation by chickpea grain was achieved from ZT plots (66 kg N/ha) than from CT plots (45 kg N/ha). However, the sowing time x tillage interaction was significant for ZT at harvest in the 28 April sowing time (Table 2) and is probably due to the availability of more soil water below 60 cm depth, which chickpea would utilise from flowering stage onwards.

Nitrogen fixation

The ¹⁵N natural abundance method used to determine %Ndfa and the choice of milk thistle as a reference plant have been found to be appropriate for chickpea on Vertisols in the Darling Downs, southern Queensland.

Beck *et al.* (1991) used a non-nodulating chickpea as a reference crop for chickpea. The matching of legume to reference plant is important (Chalk 1985) because the concentration of 15 N in available soil N may vary during the growing season and a plant dissimilar in growth pattern to chickpea may take up 15 N of different concentration. The milk thistle used as a reference plant in this study is a dicotyledon like chickpea and has a similar growth pattern.

The advantages of late autumn and early winter sowing over late winter or spring sowing, which were evident in DM (Horn *et al.* 1996) and N accumulation in this study, are not carried through into fixed N₂. The %Ndfa values in 1992 (12–16%) and 1993 (14–31%) are low in comparison with other reported values (62%, Hossain *et al.* 1995; 55–80%, Beck *et al.* 1991; and 61-63%, Beck 1992).

The amount of N_2 fixed in 1993 ranged (in the different tillage practices) from 15 to 32 kg N/ha for the 28 April and 8 June sowings and from 15 to 23 kg N/ha for the 21 July sowing (Table 5). This compares with the 89–115 kg N/ha for late autumn sowing and 3–6 kg N/ha for spring sowing reported by Beck *et al.* (1991).

The low %Ndfa and subsequently low N_2 fixed in this trial are unlikely to be a result of poor reference plant selection. It is more likely that low %Ndfa occurred because high soil NO₃-N levels allowed the chickpea to utilise this source of N rather than fixing atmospheric N₂. Doughton *et al.* (1993) reported that N₂ fixation decreases as soil NO₃-N increases, to the extent that chickpea fixed 85% of crop uptake at 8 kg NO₃-N/ha, reducing to 20% fixed at 324 kg NO₃-N/ha (Fig. 2). This was confirmed in studies reported by Herridge *et al.* (1995).

Pre-sowing soil NO3-N in this trial was high, with a mean value in the root zone (0-120 cm) of 162 kg N/ha in 1992 and 110 kg N/ha in 1993 (data not shown). Nitrogen fixation, according to Doughton et al. (1993), could be expected to be 55-60 kg N/ha in 1992 and 65-70 kg N/ha in 1993. Amounts fixed in our study were considerably lower with 12-14 kg N/ha in 1992 and 17-30 kg N/ha in 1993 (Table 4), which can be explained by lower total biomass produced in dry seasons as a result of differences in available soil water. The trial reported by Doughton et al. (1993), was conducted in average to good rainfall seasons in the mid 1980s at a site with average annual rainfall of 960 mm and potential evapotranspiration of 1227 mm. Our study was conducted in below-average rainfall seasons at a site with annual average rainfall of 680 mm and potential evapotranspiration of 1700 mm. Thus, while N₂ fixation probably decreased with increasing soil NO₃, biomass and N_2 fixation for a particular value of soil NO₃ may decrease as rainfall decreases and potential evapotranspiration increases.



Figure 2. The effect of soil NO_3 -N on N_2 fixation by chickpea in 1993.

That there was no tillage effect on N_2 fixation or interaction between sowing time and tillage was probably also due to the overiding influence of high soil NO₃.

Nitrogen balance

The negative soil N balance shown in Table 5 is due to the removal of N in chickpea grain not being balanced by N₂ fixation. After soil NO₃-N, the next major determinant of N₂ fixation is the quantity of biomass produced (Hossain *et al.* 1995), and this is primarily influenced by soil moisture and nutrient availability. In this experiment, low N₂ fixation was due to a combination of dry seasonal conditions, low biomass production in chickpea and high soil NO₃.

It is not possible to deduce the benefit of a legume crop to soil N status solely from the calculation of N_2 fixation without making allowance for the removal of grain and possibly straw from the system. The simple N balance presented in Table 5 assumes that the major N input is from atmospheric N_2 fixation and the major output is in grain. It takes no account of the contribution of chickpea roots to organic soil N or processes such as NO_3 leaching and denitrification, ammonia volatilisation, or non-symbiotic N_2 fixation. The negative N balance, however, is of concern in the longterm maintenance of soil N. Legume crops may not consistently obtain most of their N requirements from N_2 fixation, and can use variable amounts of soil N, sometimes well in excess of 100 kg N/ha (Evans *et al.*

1989). Hence their contribution to soil N can be highly variable. Evans et al. (1989) reported that gains in soil N are likely when the legume obtains at least 50% of crop N from N₂ fixation. They found threshold values for lupins and field peas were about 30 and 60 kg N/ha fixed, respectively. Chickpea contributed 65 kg N/ha to soil N when the proportion fixed was 83%, but when the proportion fixed was 59% there was a net loss from the soil of 7 kg N/ha (Evans et al. 1989), suggesting that chickpea needs to obtain at least 60% of crop N from N₂ fixation before any contribution is made to soil N. However, other interrelated factors such as grain removal, soil NO3 at sowing, and legume biomass all affect the overall contribution of chickpea to soil N. In this study, chickpea did not contribute significantly to soil N because of high soil NO₃-N, low soil moisture reducing biomass production, the low %Ndfa for 1993 (14–34%, Table 3) and considerable grain N removed from the system.

Conclusions

Growing chickpea in this experiment resulted in a net loss from the soil-plant system of 2-59 kg N/ha. Major factors in this loss were removal of N in grain (30-79 kg N/ha) and low amounts of N₂ fixation (15-32 kg N/ha) as a result of high soil NO₃-N and low chickpea biomass production during seasons of below-average rainfall. Management of the cropping system to reduce soil NO₃ to a level where chickpea will fix N₂ instead of using soil NO₃-N is essential if chickpea is to have a useful role in sustainable cropping systems, even though it is an attractive cash crop.

The use of ZT, while not directly contributing to N_2 fixation, did result in generally higher DM yields and has potential to be a useful management option. The benefit of ZT may be more evident in normal rainfall seasons on the Darling Downs or in higher rainfall climates, where there is greater potential for water to be stored at depth in the profile during the fallow. Early sowing is also beneficial. However, the success of a particular sowing time depends on an adequate supply of available soil water for rapid chickpea establishment, but low soil NO₃-N levels for maximum N₂ fixation.

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