Chicory increases the level of N fixation by companion legumes

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Abstract

For chicory (*Cichorium intybus*) to be successfully incorporated into short term pasture phases in mixed farming systems, it needs to be grown with a companion legume to meet its high nitrogen (N) requirements. It was hypothesised that chicory's superior ability to scavenge for N will deplete the system of mineral N more than a perennial grass, thus increasing the level of biological N fixation by companion legumes. A glasshouse study was conducted where chicory and perennial ryegrass (*Lolium perenne*) were grown as monocultures or in 50:50 mixtures with either lucerne (*Medicago sativa*) or subterranean clover (*Trifolium subterraneum*). Monocultures of lucerne or subterranean clover were also included. All pasture treatments containing chicory were shown to deplete the soil mineral N by approximately 110 mg N pot^{-1} more than the perennial ryegrass treatments. Pure subterranean clover treatments had significantly greater (P<0.05) estimated levels of biological N fixation (1818 mg N pot^{-1}) compared with lucerne treatments (801 mg N pot^{-1}). Combining subterranean clover with chicory in a 50:50 mixture resulted in 43.1 mg N fixed per g of legume dry matter (DM), which was significantly greater (P<0.05) than all other treatments including the pure subterranean clover treatment (29.3 mg N fixed per g of legume DM). Growing chicory and lucerne mixtures of 50:50 increased (P<0.05) estimated N fixed by 7.4 mg N fixed per g of legume DM compared to the pure lucerne treatments (24.2 mg N fixed per g of legume DM). It was concluded that when chicory is grown in 50:50 mixtures with subterranean clover or lucerne, the superior ability of chicory to deplete soil mineral N increases the level of N fixation by the companion legumes compared to when the legumes were grown in 50:50 mixtures with perennial ryegrass.

Key Words

Lucerne, subterranean clover, perennial ryegrass, soil mineral N

Introduction

If chicory is to be successfully introduced into phased pasture-crop farming systems it is important that the contribution of companion pasture legume species to the N economy of the following cropping phase is not disrupted. The impact that chicory may have on the N fixation efficiency of companion legume species, such as subterranean clover and lucerne, when grown in mixtures has not been thoroughly investigated.

The quantity of N fixed by legumes is a function of the DM accumulated, the N concentration of that DM, the proportion of this N derived from the atmosphere and the concentration of NO_3^- in the soil (Peoples and Baldock 2001). Typically for an annual legume species, such as subterranean clover growing in combination with other annual species, the symbiotic dependence on atmospheric N ranges from 65% - 95% (Bolger et al. 1995). For lucerne the dependence is much lower, ranging between 40 – 80% (Peoples and Baldock 2001). With increasing dependence on atmospheric N, legumes increase the quantity of N fixed and ultimately, the total N within the system. This dependence on atmospheric N can rise throughout the growing season as the soil mineral N is utilised by more competitive pasture species (Ledgard and Steele 1992).

The use of perennial grasses in mixtures with legumes has been shown to slightly improve the efficiency of N fixation by legumes due to the continual consumption of readily available soil N from the root zone by the perennial species (Dear et al*.* 1999). As chicory has been shown to be a highly efficient scavenger for soil N (Hogh-Jensen et al*.* 2006) it is hypothesised that chicory would be a more effective scavenger for soil N than a perennial grass such as perennial ryegrass; thus increasing the level biological N fixation by the companion legumes.

Methods

Soil was collected from the 0 – 15 cm depth interval of a Red Chromosol soil (Isbell 1996) derived from granite, located on the Charles Sturt University farm, Wagga Wagga NSW. The area was under a long term pasture comprising of mixed annual broadleaf and grass species with little to no legume content. The soil was passed through a 5 mm screen and mixed thoroughly with a cement mixer to obtain a homogenous sample.

Chemical analysis of the soil indicated the soil pH was 5.96 (1:5 1M KCl) and mineral N (NH_4^+, NO_3^- and NO_2^-) was 43.9 mg N kg^{-1}. The exchangable cations were extracted and concentrations determined by atomic absorption spectroscopy. The effective cation exchange capacity of the soil was 6.73 cmol+/kg, which was given by the sum of exchangeable Ca^{2+}, Al^{3+}, Mg^{2+}, Mn^{2+}, Na^+ and K^+ that were 4.51, 0, 0.64, 0.01, 0.02, 1.56 cmol+/kg, respectively. The field capacity of the soil occurred at a gravimetric moisture content of 18 g g^{-1}. To avoid drying the collected soil, subsamples were taken to determine its gravimetric moisture content enabling the equivalent mass of oven dried soil used to be calculated.

Each pot was lined with a plastic bag to create a closed system within the pot. The equivalent of 11.7 kg of oven dry soil was added to each pot and packed to a bulk density of 1.3 Mg m^{-3}. After the pots were filled with soil, pots were watered to 90 % field capacity by weight and 120 mL of basal fertiliser treatments were applied evenly to the soil surface. A basal fertiliser treatment ensured all nutrients, except N, were not limiting. Pots were watered daily to weight and maintained at 90 % field capacity for the remainder of the experiment.

A factorial pot experiment was conducted using a randomised block design with four replicates. Chicory and perennial ryegrass were grown as monocultures or in 50:50 mixtures with either lucerne or subterranean clover. Mixture ratios for these treatments were based on plant numbers. Monocultures of lucerne and subterranean clover were also included. Approximately 16 imbibed seeds of the respective plant treatments were sown to a depth of 0.5 cm into each pot. A commercial peat inoculant (rhizobial strains WU95 and WSM826, for lucerne and subterranean clover, respectively) was applied to the soil surface on the day of sowing in all pots and then watered in. Following germination the seedlings were thinned to 8 plants per pot. Therefore for the 50:50 chicory/legume mixtures there were 4 chicory plants and 4 legume plants. In addition, a no plant treatment was included to monitor the change in soil mineral N during the experiment when no plant uptake occurred. The experiment was conducted in a naturally lit, temperature controlled (25^{o}C day) glasshouse at the Wagga Wagga Agricultural Institute. After 16 weeks the pots were sampled and plants were separated into shoots and roots to determine dry matter yield and total N of plant material. Soil was analysed for mineral concentration.

Nitrogen fixation was estimated using the N-difference method, where the pot N (soil mineral N + plant N) of the non-legume treatment was subtracted from the pot N of the respective legume mixture treatment. N fixed = (soil mineral N+ plant N in legume pot) **-** (soil mineral N + plant N in non-legume pot) **Equation.1**

Therefore, the average pot N of the chicory monoculture was subtracted from the pot N of the chicory-legume mixtures. An average pot N for the non legume monocultures was subtracted to determine N fixed for the legume monocultures.

Results

All the chicory treatments depleted soil mineral N concentrations to a greater extent than the perennial ryegrass ($P<0.05$) (Figure 1). When chicory was grown with subterranean clover in a 50:50 mixture soil mineral N was lower (47 mg N pot^{-1}) than the chicory monoculture (90 mg N pot^{-1}) ($P<0.05$) (Figure 1). There was no difference in soil mineral N between perennial ryegrass treatments ($P>0.05$), while both the lucerne (335mg N pot^{-1}) and subterranean clover (338mg N pot^{-1}) monocultures resulted in the highest concentration of soil mineral N ($P<0.05$). All treatments had significantly depleted the soil mineral N compared to the 589 mg N pot^{-1} in the no plant treatment.

In the treatments containing the companion legumes the change in pot N above the zero value can be attributed to N fixation (Figure 2). The 50:50 mixtures of chicory and subterranean clover (1107 mg N pot^{-1}) resulted in greater ($P<0.05$) levels of N fixation compared to the 50:50 mixtures of perennial ryegrass and subterranean clover (576 mg N pot^{-1}) (Figure 2). Similar trends ($P<0.05$) were observed when chicory (633 mg N pot^{-1}) and perennial ryegrass (502 mg N pot^{-1}) were grown in 50:50 mixtures with lucerne (Figure 2). The subterranean clover monoculture (1818 mg N pot^{-1}) had higher levels of N fixation ($P<0.05$) than all other treatments and was more than 2 times that of the lucerne monoculture (801 mg N pot^{-1}) (Figure 2).

As chicory depleted more soil mineral N compared to perennial ryegrass when grown in 50:50 mixtures with companion legumes, there was a greater proportion of the pot N present as plant N compared to when perennial ryegrass was grown in the same mixtures. In the 50:50 mixtures of chicory and subterranean clover 95% of the pot N was present as plant N, whereas, only 64% of the pot N was plant N for the 50:50 mixtures of perennial ryegrass and subterranean clover. In comparison, the proportion of total pot N represented by plant N for the lucerne treatments was lower than those of subterranean clover with 86% and 38% of the pot N present as plant N for the 50:50 mixture of lucerne with chicory and perennial ryegrass, respectively. The greater proportion of pot N being present as plant N in the chicory treatments suggests that there is more total N in the system for these treatments (Figure 2), which was being utilised more effectively.

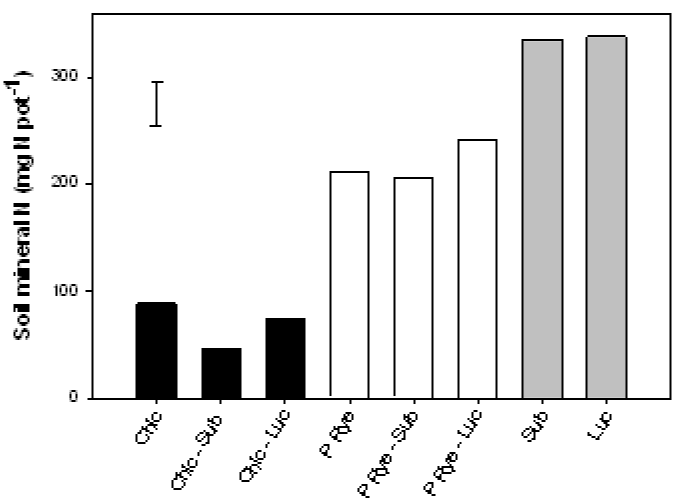

Figure 1: Soil mineral N concentration for the chicory, perennial ryegrass, lucerne and subterranean clover treatments when grown as monocultures or in 50:50 mixtures. Bar indicates L.S.D at *P*=0.05.

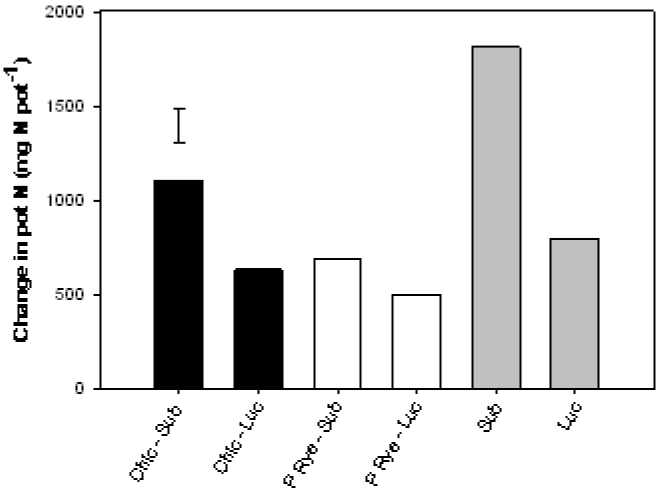

Figure 2: The change in pot N (soil mineral N and total plant N) over 16 weeks for the chicory, perennial ryegrass, lucerne and subterranean clover treatments when grown as monocultures or in 50:50 mixtures (See Equation 1). Bar indicates L.S.D at *P*=0.05.

The change in estimated N fixation efficiency by the companion legumes either grown in monocultures or in 50:50 mixtures is presented in Figure 3. Growing subterranean clover (43 mg N g legume DM^{-1}) or lucerne (32 mg N g legume DM^{-1}) in a 50:50 mixture with chicory was shown to increase the efficiency of N fixation ($P<0.05$) in comparison to the subterranean clover (29 mg N g legume DM^{-1}) or lucerne (24mg N g legume DM^{-1}) monocultures, respectively (Figure 3). In comparison, growing subterranean clover or lucerne in 50:50 mixtures with perennial ryegrass resulted in a reduction in N fixation efficiency ($P<0.05$) compared to the monocultures of subterranean clover and lucerne treatments, respectively (Figure 3). The reduction in estimated N fixation efficiency by subterranean clover and lucerne when grown in 50:50 mixtures with perennial ryegrass compared to legume monocultures was an unexpected result. It was hypothesised that perennial ryegrass would deplete the soil mineral, thus resulting in either a similar or greater estimated N fixation efficiency than the legume monocultures. The estimates are a derived value and the finding may be confounded by the fact that soil mineral N was used in the equation to estimate N fixation or it could suggest that N fixation by the companion legume was in some way being inhibited when grown in a 50:50 mixture with perennial ryegrass. This requires further investigation.

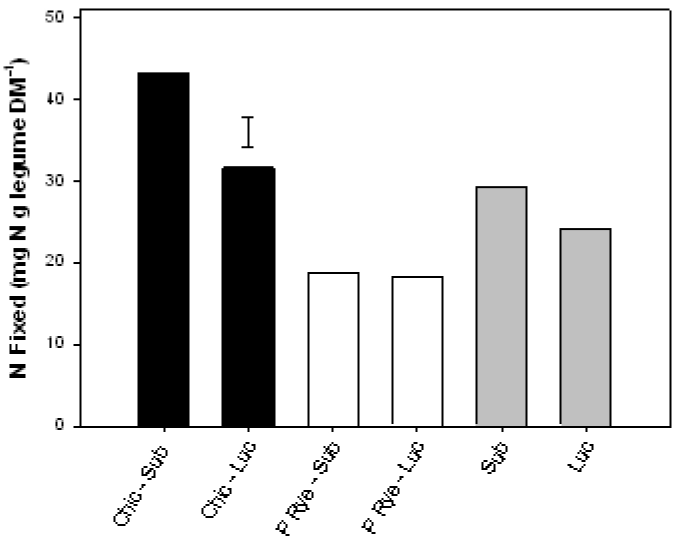

Figure 3. Estimated N fixed per g of legume DM for the chicory, perennial ryegrass, lucerne and subterranean clover treatments when grown as monocultures or in 50:50 mixtures. Bar indicates L.S.D at *P*=0.05.

Conclusion

Over the 16 weeks of the experiment chicory significantly depleted soil mineral N to a greater extent than perennial ryegrass. This resulted in increases in both the quantity and efficiency of N fixation for subterranean clover and lucerne when grown in a 50:50 mixture with chicory. Regardless of the treatment combination, the subterranean clover treatments had greater levels of estimated N fixation in comparison to the lucerne treatments. If chicory is grown in 50:50 mixtures with either subterranean clover or lucerne it could be concluded that there would be a greater quantity of N in the system compared to 50:50 perennial ryegrass-legume mixtures. The additional N accrued under the 50:50 chicory-legume mixtures was utilised very effectively with a high proportion of the N being present in the plant. The greater quantity of N present under a chicory-legume pasture is a direct result of chicory depleting the concentrations of soil mineral N more effectively than perennial ryegrass thereby increasing the legume dependence on atmospheric N. Given the assumptions involved with estimating N fixation by the N difference technique used in this experiment further studies have been conducted using the natural abundance technique to confirm the effect of chicory on N fixation by companion legumes.

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