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Nitrogen transfer between clover and wheat in an intercropping experiment

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Summary

A novel approach to the problem of improving nitrogen supply in organic farming is to use intercropping of cereals with a legume to provide nitrogen transfer within a season and/or to following crops. The affects of intercropping were studied in a column experiment using mixtures of winter wheat (*Triticum aestivum* cv. *Claire*), with white clover (w.c.) (*Trifolium repens* cv. *Barblanca*) and with red clover (r.c.) (*Trifolium pratense* cv. *Britta*). The effects of cutting and removal above ground clover material with and without additional soil disturbance were compared to leaving clover plants in situ and intercropped with wheat in a split root design. Wheat and clover plants, as monocultures, were used for the controls. ^{15}N ammonium nitrate solution was applied. The wheat seeds were sown into the column without nitrogen. We found that the cutting treatment produced the highest yield of wheat. Available ammonium-N in the soil was greatest in the clover control treatment for the column with only red clover roots and in the cutting+soil disturbance treatment for the column with only white clover roots. Available nitrate-N was greatest in the soil disturbance treatment in the column with clover and wheat roots for both red and white clover. The cutting treatment produced the highest yield of wheat.

Key words: Intercropping; winter wheat; red clover; white clover; N fixation; ^{15}N-labelling

Introduction

There is a world-wide interest in developing more sustainable approaches to agriculture at a time when the use of mineral fertilisers and chemicals is being critically assessed (Bergkvist, 2003). In organic farming, synthetic fertilisers as well as pesticides are not allowed and the main aim is to keep a constant-closed nutrient cycle on farms, to protect environmental quality and to enhance beneficial biological interactions and processes (Altieri, 1995). In the UK, organic farming systems are traditionally based on ley arable crop rotations (Sanders, 2003). The aim of organic crop rotations is to achieve a balance between crops which reduce soil fertility and crops which re-establish fertility.

Crop yields from organically farmed systems have been shown to be lower than those from conventional farms (Nieberg & Schultze, 1996). The N concentrations in organically grown wheat have also been found to be significantly lower than those in conventional systems (high N input) (Gooding *et al*., 1999). This suggests that the yield reduction may be a consequence of restricted N supply (Berry *et al*., 2002), with the availability of nitrogen be different at different developmental stages.

However, a number of studies have shown that organic farming systems regularly result in a positive N balance (i.e. that the input is greater than the output) (Watson *et al*., 2002), which leads to the suggestion that the problem is the timing of N supply rather than the amount. Moreover, the mechanism by which the nitrogen transfer between legumes and non- fixing plants taking place is still poorly understood. The overall aims of this study were to quantify transfer of nitrogen from clover to wheat and to examine the relative contributions of shoots and roots to N transfer.

Materials and Methods

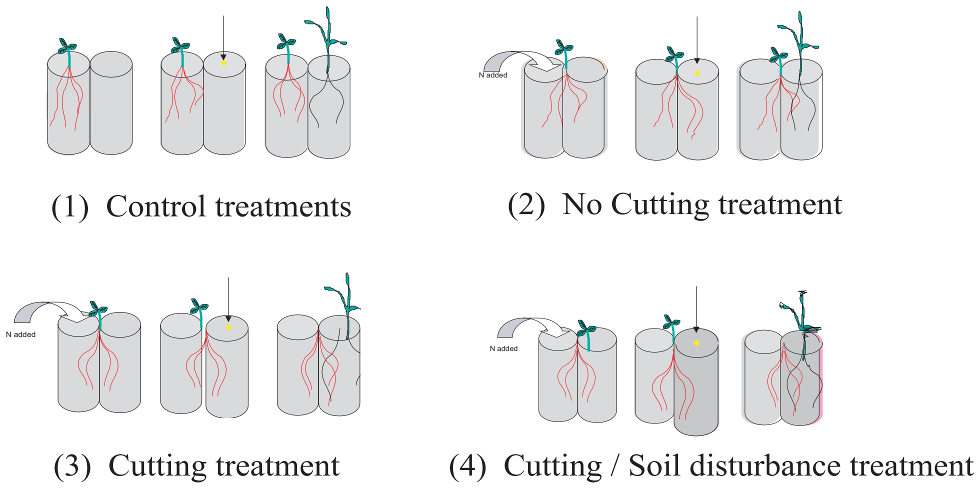
Plant culture

The experiment was set up in a greenhouse at the Bush Estate in Edinburgh (lat. 55^o 51'N, long. 3^o 12' W). Mixtures of winter wheat (*Triticum aestivum* cv. Claire), with white clover (w.c.) (*Trifolium repens* cv. Barblanca) and with red clover (r.c.) (*Trifolium pratense* cv. Britta) were used. The seeds of the clover cultivars were surface sterilised, germinated at room temperature for 24 h and sown in terragreen. After 15 days, the roots had been cut to 3 cm and only the top-two branches remained on each plant, so as to enhance the growth of the two branches and then split them one in each column. The plants were transplanted to glass tubes filled with terragreen and left for another 25 days. During this period, the temperature was 20^{o}C and the photoperiod was 24 h per day.

Experimental design

The plants were transplanted to PVC columns (400 mL) and filled with approximately 3 kg of a sandy loam soil. The columns were about 30 cm high, 5 cm in diameter and each was placed in a plastic tray. All the columns were manually irrigated with the minus-N Long Ashton nutrient solution (Hewitt & Smith, 1975) and were maintained with a constant level of liquid at the base. Each experimental unit was made up of a pair of adjacent columns. The soil used was sieved (9.525 mm) to remove any roots, plants, and stones. The total number of units was 32 (16 per variety).

Four different treatments of growing units were prepared and named according to clover treatment (1) Controls (2) No cutting (3) Cutting and (4) Cutting+soil disturbance, and each pair was glued together (Fig. 1). Each unit had one column containing only clover (red or white) roots growing and one column containing clover (red or white) roots and wheat roots, except the controls where in each column was growing only clover or wheat. The clover plants had been transplanted such that the roots were permitted to grow either in one column only (control treatment) or with the root system split between two columns. In sets two, three and four, which simulate intercropping, one month after planting with clover roots in two columns, an application of ^{15}N ammonium nitrate in solution was made to one of the two columns at 15 g N m^{-2}. The wheat seeds were sown after one month into the column without nitrogen. When the wheat was sown, the clover shoots kept growing *in situ* in the first (control) and second (no cutting) treatment. In the third treatment (cutting) and fourth treatment (cutting+disturbed treatment), the clover shoots were cut and removed from the top of the columns Additionally, the soil was disturbed to simulate ploughing in the fourth treatment. One month after the wheat was sown, the wheat plants were harvested. The clover shoot yields were measured for each treatment at the end of the second month for the third and fourth treatment and at the end of the third month for the third and fourth.



(1) Control treatments

(2) No Cutting treatment

(3) Cutting treatment

(4) Cutting / Soil disturbance treatment

Fig. 1. The four treatments at the end of the first month (LH column pair), second month (centre column pair) and third month (RH column pair) of the experiment.

Results

The wheat plants grown in the cutting treatment with the red clover donor and the cutting+soil disturbance treatment had the tallest and heaviest shoots, and heaviest roots. The non-cutting treatment was significantly different from the other treatments and had the weakest and lightest wheat plants. In the columns containing clover roots, highest ammonium-N concentrations were observed in the soil of the control with red clover and the soil disturbance treatment for white clover. The non-cutting treatment had the highest amount of ammonium-N in the soil in the

Table 1. *Ammonium-N levels in the soil DM comparing the columns growing only red (*Trifolium pratense *cv.Britta) or white (*Trifolium repens *cv. Barblanca) clover roots with the columns growing wheat and clover roots (n=8)*

Growing columns / Treatments	Red Clover	Red clover and wheat	White Clover	White clover and wheat
Controls	1.76 ± 0.08	0.63 ± 0.07	1.06 ± 0.24	0.75 ± 0.03
Non Cutting	1.15 ± 0.12	1.45 ± 0.05	1.23 ± 0.16	1.40 ± 0.04
Cutting	0.79 ± 0.14	0.76 ± 0.11	1.35 ± 0.18	0.71 ± 0.09
Cutting and Soil Disturbance	0.75 ± 0.09	0.62 ± 0.06	1.50 ± 0.75	0.79 ± 0.16

Table 2. *Nitrate-N levels in the soil DM comparing the columns growing only red (*Trifolium pratense *cv.Britta) or white (*Trifolium repens *cv. Barblanca) clover roots with the columns growing wheat and clover roots (n=8).*

Growing columns / Treatments	Red Clover	Red clover and wheat	White Clover	White clover and wheat
Controls	0.60 ± 0.11	0.95 ± 0.31	17.40 ± 2.71	1.14 ± 0.34
Non Cutting	0.91 ± 0.27	0.50 ± 0.02	0.65 ± 0.01	0.21 ± 0.02
Cutting	16.05 ± 2.35	2.65 ± 1.08	2.99 ± 3.48	0.91 ± 0.25
Cutting and Soil Disturbance	6.76 ± 4.90	13.0 ± 1.72	13.02 ± 1.73	2.07 ± 1.96

column containing clover and wheat roots (Table 1). There were significant differences in nitrate-N between the treatments and the columns containing wheat or clover (Table 2).

In the wheat shoots and roots, the highest amounts of total N were measured in the cutting treatment intercropping with red clover and the cutting+soil disturbance treatment intercropping with white clover (FigS 2 & 3). In the wheat plants, the excess of ^{15}N and the transfer of ^{15}N were measured by mass spectrometry and were highest in the cutting treatment intercropping with red clover and cutting+soil disturbance treatment intercropping with white clover, but the values were low (Fig. 4). The clover plants in the non-cutting treatment were significantly different between

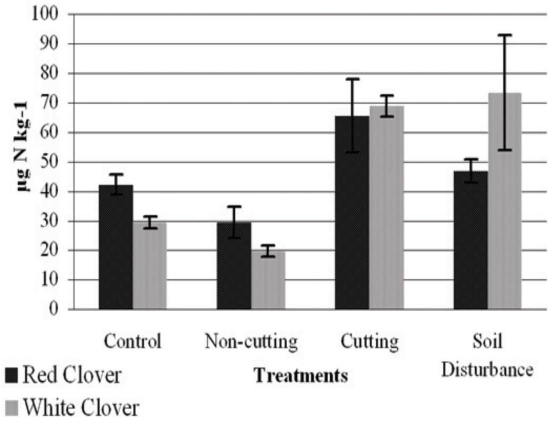


Fig. 2. The amount of total N in the wheat shoots grown alone (control) or with red or white clover plants.

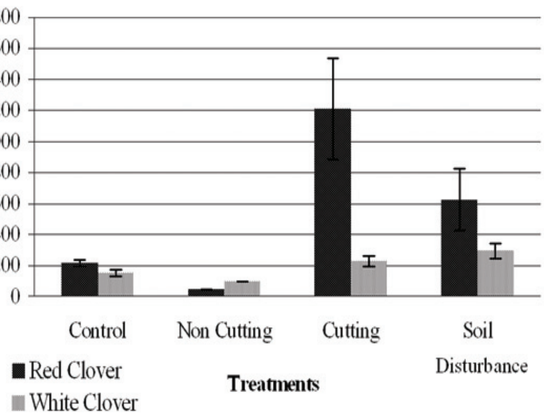


Fig. 3. The amount of total N in the wheat roots grown alone (control) or with red or white clover plants.

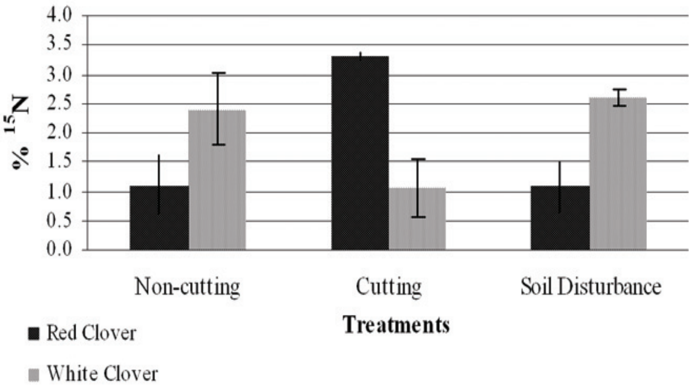


Fig. 4. The transfer of 15N in the wheat plants in the different treatments with red or white clover plants.

clover types and had the highest amount of ^{15}N excess and showed the highest levels of ^{15}N transfer.

Discussion

Direct transfer of N between plant species has long been of interest to ecologists and agronomists as a mechanism of nutrient flow within systems (Paynel *et al.*, 2001; Paynel & Cliquet, 2003). Thorsted *et al.* (2005) found that intercropping winter wheat with clover resulted in wheat grain yield decreases of 10–25% compared with wheat only. This was likely to have been caused by interspecific competition for light and N during vegetative growth and for soil water during grain filling.

In this experiment, the cutting only treatment produced the highest yield of wheat. It seems that this treatment provided the ideal environment for N mineralisation. The lower yield in the cutting+soil disturbance treatment is possibly due to greater immobilisation (since the disturbance of the soil could encourage soil organic matter decomposition and then immobilisation). Moreover, the clover plants were able to efficiently recover the applied ^{15}N. Rates of transfer were low, and the soil disturbance and cutting treatments did not lead to a consistent increase in transfer of N. Further work is needed to determine the mechanism by which N is transferred and to establish if the quantities of N transferred change over longer periods of growth. These results have potential practical applications in low input systems. It is possible that drilling cereals into a cut clover crop or cutting strips of clover grown between strips of cereals may enhance the crop's nutrition and cereal growth and increase yield. Finally, the main advantage is the development of a more fertile soil with improved structure, aeration and more organic matter.

Acknowledgements

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