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Mechanical control of clover improves nitrogen supply and growth of wheat in winter wheat/white clover intercropping

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Abstract

The major objective for clover in a winter wheat/white clover intercropping system is to supply nitrogen (N) for the wheat. A field experiment was repeated in 2 years on a loamy sand in Denmark to investigate the possibilities for increasing N supply to the winter wheat by cutting and mulching the clover between the wheat rows. The clover was cut with a weed brusher on three different dates in each year.

Intercropped wheat with unbrushed clover had a lower grain yield than wheat as a sole crop. Brushing increased wheat N uptake and wheat grain yields. Intercropping with two or three brushing dates gave higher wheat yields than wheat as the sole crop. The largest increases in grain N uptake, $21-25 \text{ kg N ha}^{-1}$, were obtained for the brushings around wheat flag leaf emergence. The highest yield increases with a single brushing, $0.98-1.11 \text{ Mg DM ha}^{-1}$, were obtained when brushing was performed during the stem elongation phase. The largest grain yields for treatments with two brushings were obtained with a first brushing at start of stem elongation and a second around flag leaf emergence. The first brushing probably provided N to increase the wheat leaf area index and thus the light interception, while the second brushing provided N to sustain the leaf area during grain filling and reduced clover biomass and therefore competition for water. Intercropping wheat and clover increased grain N concentrations by 0.11-0.39%-point compared with wheat as a sole crop. Intercropping may thus offer possibilities for improving the bread-making quality of organically grown wheat.

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1. Introduction

Low-input farming systems, such as arable organic farming, often have limited access to nitrogen (N), and this often limits the productivity of these systems (Berry et al., 2002; Olesen et al., 2002). Many of these farming systems rely on biological N fixation (BNF) to supply N for intercropped cereals and/or for subsequent crops (Elgersma et al., 2000; Mueller and Thorup-Kristensen, 2001). In intercropping systems, legumes can provide N for the intercropped cereals through N transfer (Xiao et al., 2004), while long-term transfer through effects on soil organic N is most important for yield improvements at the crop rotation level (Olesen et al., 2002).

White clover (*Trifolium repens* L.) is known to have a high capacity for BNF, and the fixed N is partly made available

to associated crops through mineralised root and nodule N (Ledgard and Steele, 1992). Annual N inputs from BNF to a grass-clover mixture can be up to 300–500 kg N ha⁻¹ (Vinther and Jensen, 2000), although values of 150–300 kg N ha⁻¹ year⁻¹ may be more typical (Elgersma et al., 2000). The amount of N supplied from BNF in grass-clover mixtures is extremely variable, depending on the proportion of clover, the defoliation regime and soil moisture availability. The amount of N transferred from the legume to the grass component can also be quite variable, but the proportion rarely exceeds 10% of BNF (Ledgard et al., 1985; Høgh-Jensen and Schjørring, 2000).

Intercropping of cereals and clover offers a possibility to provide N to the cropping system while growing a cereal crop (Anil et al., 1998). In a winter wheat/white clover intercrop, winter wheat (*Triticum aestivum* L.) is seeded into an already established stand of white clover (Jones and Clements, 1993). The wheat can be seeded into rototilled strips to reduce early competition between the clover and winter wheat (Thorsted et al., in press).

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Previous experiments on winter wheat/white clover systems have shown interspecific competition for both above- and belowground resources (M.D. Thorsted, unpublished data). Belowground competition for water and N is particularly important for wheat grain yields, whereas above-ground competition primarily affects the growth of the clover (Thorsted et al., in press). When the cereal crop is less competitive (e.g. due to low plant densities), grain yields may also be reduced by above-ground competition (Jones and Clements, 1993). Grain N concentrations, however, generally increased by intercropping with clover.

The direct transfer of N from a N2-fixing legume to an intercropped cereal is usually insufficient for cereal production (Stern, 1993). There is evidence that the transfer of N from legumes to grass in mixed pastures is promoted by defoliation of the legume, which results in root and nodule death (Trannin et al., 2000). Thus, methods that reduces the above-ground clover biomass may contribute N to the intercropped cereal, both through above-ground residues and through increased release of below-ground N. Thorsted et al. (2002a) used a brush weeder to cut the clover between oat rows to increase N recycling and reduce interspecific competition in oat/white clover intercropping. This increased oat grain N yields and grain N concentration. Mechanical control of the clover may therefore be an option to reduce interspecific competition and to increase N transfer from clover to wheat in winter wheat/white clover intercropping systems.

The objective of the experiment presented here was to investigate the possibilities for reducing interspecific competition and increasing the transfer of N from clover to winter wheat by brushing off the clover at various times during the vegetative growth of winter wheat.

2. Materials and methods

A field experiment was performed twice starting in 1999 and 2000 at Research Centre Foulum in Denmark (56°30′N, 9°35′E). The soil is loamy sand (9% clay, 3% organic matter) and is classified as a typic hapludult according to the Soil Taxonomy System (Nielsen and Møberg, 1985).

2.1. Experiment design

Clover was undersown in spring oats (*Avena sativa* L.) in spring 1999 and in another field in 2000 by evenly distributing 8 kg seeds ha⁻¹ in 36 and 32 plots, respectively. The oat crop was harvested and the grain and straw removed on 3 September 1999 and 25 August 2000. In the plots with clover, the clover was cut and the cuttings removed just before seeding the wheat. There were four plots without clover. In 1999, the plots without clover were created by scraping off the upper 5 cm soil in four plots that had clover. In 2000, the clover-free plots were created by not seeding clover at all. A Maschio rotary cultivator was used to rototill 9 cm wide bands 25 cm apart in the clover sward, and winter wheat was sown in the bands with a Nordsten seeding machine mounted on the rotary cultivator on 8 October 1999 and on 21 September 2000. The target plant density was 400 plants m⁻², and the direction of the rows was East–West.

The white clover cultivar was Milo (DLF-Trifolium, Denmark), and the winter wheat cultivar was Stakado (Abed, Denmark). Inorganic fertiliser was applied at the rate of $70\,\mathrm{kg}\,\mathrm{N}\,\mathrm{ha}^{-1}$, $17\,\mathrm{kg}\,\mathrm{P}\,\mathrm{ha}^{-1}$ and $52\,\mathrm{kg}\,\mathrm{K}\,\mathrm{ha}^{-1}$ by even distribution on 10 April 2000 and on 26 April 2001.

The experiment was a randomised complete block design with four replicates. There were nine experimental treatments giving a total of 36 plots. The plot size was $3 \text{ m} \times 18 \text{ m}$. The treatments included intercrops of winter wheat and white clover, winter wheat as a sole crop, and different combinations of three different timings of brushing the white clover (Table 1). The clover and weeds were brushed/cut between cereal rows with a brush weeder (Bärtschi-Fobro, Hüswil, Switzerland). The brushing was done from zero to three times at three different dates prior to wheat growth stage GS39 on the BBCH scale (Lancashire et al., 1991). Brushing was done on 10 April (GS23), 4 May (GS31) and 18 May (GS37) in 2000. In 2001 brushing was done on 4 May (GS30), 18 May (GS31) and 31 May (GS39). The weed brusher cut off the plant material at the soil surface in 11 cm wide bands, and this plant material was left on the ground. The brush weeder was also used in the plots without white clover.

2.2. Measurements

Grain yields were determined by harvesting a $1.5 \text{ m} \times 9.15 \text{ m}$ subplot. The area outside the harvest plot was used for plant sampling. The plots were harvested at maturity (15 August 2000 and 23 August 2001) using a combine harvester. Grain weight was determined by weighing 600 dried grains per plot. Aboveground biomass of wheat, clover and weeds was harvested in two samples of $50 \text{ cm} \times 50 \text{ cm}$ per plot. To assess the above-ground clover biomass removed at each brushing, two $11 \,\mathrm{cm} \times 50 \,\mathrm{cm}$ samples in the clover bands were cut prior to each brushing in 2001. Clover N content was not measured, but an estimate of 3% N of clover dry matter was used in the calculations (Thorsted et al., 2002b). Sampling of above-ground plant material was done on three dates from 2 May to 13 August in 2000, and on four dates from 30 May to 2 August in 2001. The above-ground plant material was separated into wheat, clover and weed fractions in 2001, but clover and weeds were pooled in 2000. Clover and weeds were not sampled on the first sample date in 2001.

The dry weight of grain and all plant samples were determined after oven drying at 80 °C for 24 h. Total N in the wheat biomass and grain was determined by the Dumas method on finely milled samples from each plot (Hansen, 1989).

2.3. Statistical analyses

The yield and biomass data were analysed by one-way ANOVA using the MIXED procedure of SAS statistical analysis system (SAS Institute, 1999). Since the exact timing of the brushings varied between the 2 years, data for each cropping year was analysed separately with block as random variable. To obtain variance homogeneity, all data from the plant samplings of above-ground biomass was transformed using the natural logarithm. The grain yield was not transformed.

Effects of intercropping (with/without clover) and dates of brushing of clover on winter wheat dry matter grain yield, dry matter grain weight, harvest index (HI) and grain N concentration and content, and wheat above-ground N uptake at the last sample date in 2000 and 2001

Treatment	ent			2000						2001					
Index	Brushing dates	ig dates	Clover	Grain yield (Mg ha ⁻¹)	H	Grain weight (mg)	N conc. (%)	N conc. (%) Grain N content Plant N uptake (kg N ha ⁻¹)	Plant N uptake (kg N ha ⁻¹)	Grain yield (Mg ha ⁻¹)	Ħ	Grain weight (mg)	N conc. (%)	Grain weight N conc. (%) Grain N content Plant N uptake (mg) (kg N ha ⁻¹) (kg N ha ⁻¹)	Plant N uptake (kg N ha ⁻¹)
	1 2	3	I												
a	1	ı	+	3.60	0.41	47.2	1.66	60.0	76	4.13	0.38	47.8	1.57	65.0	108
q	+	1	+	3.94	0.38	46.1	1.58	62.3	112	5.18	4.0	47.6	1.56	6.08	119
c	+	ı	+	4.59	0.44	45.2	1.60	73.4	1117	5.15	0.43	47.8	1.58	81.5	121
q	1	+	+	4.71	0.43	49.1	1.82	85.4	134	4.87	0.42	49.7	1.76	85.9	123
в	+	ı	+	5.00	0.45	4.5	1.58	79.0	121	5.30	4.0	46.9	1.56	82.8	119
f	+	+	+	4.73	0.45	48.6	1.68	79.7	120	5.59	0.46	49.0	1.61	90.3	126
00	+	+	+	5.20	0.45	47.3	1.67	8.98	126	5.31	0.47	48.6	1.64	87.3	117
h	+	+	+	5.33	0.43	46.6	1.65	88.1	139	5.80	0.48	47.5	1.55	0.06	120
i	+	+	I	4.86	0.47	45.9	1.47	71.3	86	5.36	4.0	48.4	1.37	73.7	112
S.E.D.				0.16	0.02	0.7	0.03	3.1	7	0.14	0.02	0.4	0.03	2.7	∞
S.E.D.	is the star	S.E.D. is the standard error of difference.	of differenc	ě.											

The wheat harvest index (HI) was calculated by dividing the dry matter grain yield by dry above-ground wheat biomass at the last sampling date.

3. Results

3.1. Wheat grain yield

Brushing on any single date increased grain yield (Table 1). On average, brushing one or more times increased grain yield by 31%. In 2000, the highest yields were obtained from the two brushing dates in May, whereas the first brushing in April only gave a small yield increase. In 2001, the first two brushings (early May) resulted in higher yield than brushing in late Mav.

The average yield increase from a single brushing was 0.81 and $0.94\,\mathrm{Mg}\,\mathrm{ha}^{-1}$ in 2000 and 2001, respectively (Table 1). The additional average yield increase from a second brushing was $0.56\,\mathrm{Mg}\,\mathrm{ha}^{-1}$ in 2000 and $0.33\,\mathrm{Mg}\,\mathrm{ha}^{-1}$ in 2001, and a third brushing resulted in an additional average yield increase of 0.35 and $0.40\,\mathrm{Mg}\,\mathrm{ha}^{-1}$ in 2000 and 2001, respectively. The yield increase of an additional brushing decreased with the number of brushings.

Intercrops on average had 4% lower grain yield than wheat as a sole crop (Table 1). However, all intercrops that had been brushed twice or more had grain yields very close to or higher than the wheat sole crop.

3.2. Harvest index and grain weight

The harvest index (HI) was increased by brushing in 2001, whereas HI increased in 2000 for the two last brushing dates only (Table 1). There was a clear tendency towards higher HI when clover was brushed two or three times compared to one brushing only.

The grain weight was reduced by brushing on the first two dates in both years, but was increased by the last brushing (Table 1).

3.3. Grain N uptake and concentration

Wheat N content increased on average by 31% when clover was brushed one or more times (Table 1). The greatest increase in grain N content (25 kg N ha $^{-1}$ in 2000 and 21 kg N ha $^{-1}$ in 2001) was obtained with the final brushing alone in both years, and there was only a small additional increase in grain N content from additional brushings.

The wheat sole crop had on average $10 \,\mathrm{kg} \,\mathrm{N} \,\mathrm{ha}^{-1}$ higher grain N content than the unbrushed wheat intercrop. However, the mean grain N content of the brushed intercrops averaged $10 \,\mathrm{kg} \,\mathrm{N} \,\mathrm{ha}^{-1}$ higher than that of the wheat sole crop.

Grain N concentration was slightly reduced by brushing on the first two dates in both years, but it was increased by brushing on the last date (Table 1). Intercropped wheat had considerably higher N concentration than wheat as a sole crop in both years.

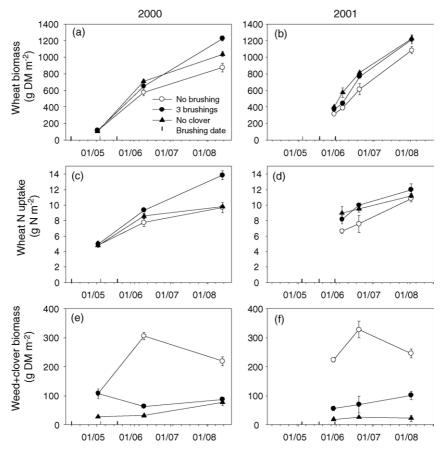


Fig. 1. Development of above-ground dry matter in wheat in 2000 (a) and 2001 (b), N uptake in wheat above-ground biomass in 2000 (c) and 2001 (d) and above-ground dry matter in clover and weeds in 2000 (e) and 2001 (f). Symbols indicate no brushing of clover in intercrop (), brushing of clover at all three dates in intercrop () and wheat sole crop brushed three times between wheat rows (). Vertical marks indicate dates of brushing. Bars show the standard error.

3.4. Wheat biomass and N uptake

At the second sampling in 2000, the second brushing was the only treatment giving a significant increase in wheat biomass (P=0.002). At the third sampling there was significantly higher biomass for all of the brushing dates (P<0.03), data not shown). In 2001, brushing on the first date increased wheat biomass on the first and second sampling date (P<0.01). The largest wheat biomass was obtained in both years by brushing on all three dates (Fig. 1a and b).

The N content in above-ground wheat biomass at the final sampling was similar for the wheat as a sole crop and the unbrushed wheat intercrop in both years (Fig. 1c and d). Brushing increased total above-ground N uptake of wheat by an average of $27 \, \mathrm{kg} \, \mathrm{N} \, \mathrm{ha}^{-1}$ in 2000 and $13 \, \mathrm{kg} \, \mathrm{N} \, \mathrm{ha}^{-1}$ in 2001 (Table 1). Later brushings increased N uptake more than earlier brushings in 2000, whereas there was no significant difference among brushing dates in 2001.

3.5. White clover and weed biomass

The clover + weed biomass was reduced by similar amounts by brushing in both years (Fig. 1e and f). The clover + weed biomass remained fairly stable over time during June and July after the last brushing in all treatments. The above-ground clover biomass measured several days after brushing in 2001 was

reduced to $40-50\,\mathrm{g}\,\mathrm{DM}\,\mathrm{m}^{-2}$ (Table 2). Thus, not all above-ground N in the clover was recycled by the brushings. However, these estimates of clover biomass covered both wheat rows and clover strips, and some clover was probably also growing in the wheat rows. The estimates of above-ground N in clover (Table 3) prior to brushing were from the clover strips and therefore probably represent a more accurate estimate of the amount of above-ground N recycled.

The amount of N recycled by brushings increased when brushing was delayed from the tillering stage to the stem elongation stage in wheat (Table 3). Multiple brushings did not recycle substantially more N than one single late brushing, probably because the clover needs to re-establish its leaf area after brushing, and this reduces photosynthesis and BNF.

Weed biomass was low in all treatments in 2001 (Table 2). Brushing tended to reduce weed biomass at the first sampling date. However, by the last sampling date there were no differences.

4. Discussion

4.1. Recycling of N

The increase in N content in above-ground wheat biomass from brushing was on average $20\,\mathrm{kg}\,\mathrm{N}\,\mathrm{ha}^{-1}$. The average

Table 2
Mean clover and weed biomass at three sampling dates in 2001

Treatment			Clover bioma	ass (g DM m ⁻²)		Weed biomass $(g DM m^{-2})$		
Index	Brushing	Clover	30 May	20 June	2 August	30 May	20 June	2 August
a		+	192 bc	293 f	233 f	32 cd	26 abc	4 a
b	+	+	131 bc	157 cde	169 def	18 bcd	29 abc	10 a
c	-+-	+	48 a	100 bcde	120 cde	16 abcd	12 ab	19 a
d	+	+	230 с	71 abc	67 abcde	18 bcd	29 c	10 a
e	++-	+	48 a	87 bcd	105 bcde	7 abd	28 abc	17 a
f	+ - +	+	123 bc	53 ab	49 abc	14 abcd	18 abc	15 a
g	-++	+	89 b	48 ab	73 abcd	7 ab	17 abc	22 a
ĥ	+++	+	41 a	48 ab	79 abcd	12 abd	18 abc	15 a
i	+++	_				18 bcd	24 abc	7 a

Analysed on log-transformed data and retransformed to the original unit. Within each column, means followed by different letters are significantly different at the 0.05 significance level based on *t*-tests.

increase in grain N content from brushing was also $20 \, kg \, N \, ha^{-1}$. This suggests that the entire additional N uptake was found in harvested grain N. This large additional N uptake in the grains may be explained by the late availability of the N from the brushings, which will tend to favour the growing grains because no additional N is needed to sustain the leaf area. Based on the estimates of rate of N uptake in above-ground wheat biomass, the extra increase of N in the unbrushed wheat intercrop versus the wheat sole crop was ca. $10 \, kg \, N \, ha^{-1}$, which is in line with values obtained in other wheat/clover intercrop studies (Thorsted et al., in press). Thus, brushing approximately tripled the N transfer from the clover to the wheat.

The N use efficiency (Eff) from the recycled N from the brushing in 2001 can be estimated by dividing the increase in grain N content by the N in above-ground clover brushed (Tables 1 and 3):

$$Eff = 100 \times \frac{N_{\rm ub} - N_{\rm uc}}{N_{\rm brush}}$$

where $N_{\rm ub}$ is the grain N in the brushed intercrop treatment (kg N ha⁻¹), $N_{\rm uc}$ the grain N in the unbrushed intercrop treatment (kg N ha⁻¹) and $N_{\rm brush}$ is the amount of N brushed off by the weed brusher in the brushed intercrop treatment (kg N ha⁻¹).

The estimated average N use efficiency of all brushing treatments was 17%. This is considerably lower than the typical N use efficiency of 40–50% obtained for mineral N fertiliser application to winter wheat in Denmark (Olesen et al., 2003; Petersen and Djurhuus, 2004). The largest N use efficiency (28%) was obtained for the first single brushing, and the smallest for the last

Table 3 Estimated amounts of N in above-ground clover biomass (kg N ha $^{-1}$) at brushing dates in 2001

Treatment	4 May	18 May	31 May
b	56	=	_
c	_	86	_
d	_	_	156
e	65	67	_
f	62	_	137
g	_	96	40
h	45	62	29

single brushing (13%). Smaller reductions in N use efficiency have been found from delaying mineral N fertilisation (Olesen et al., 2003). The estimated N use efficiencies are probably overestimated, since the brushings will also result in release of N from clover roots (Trannin et al., 2000). About 40% of total N accumulated in white clover is situated below the cutting height (Jørgensen and Ledgard, 1997).

There are several possible explanations for the relatively low N use efficiency. First, the white clover will compete with the wheat for soil N. Secondly, the mulched clover must be mineralised before it becomes available to the crops. The high N concentration of the clover will usually ensure a high mineralisation rate (Breland, 1994). Since the clover was not incorporated in the soil, it is possible that the generally dry conditions during May and June may have reduced the mineralisation rate or restricted the leaching of mineral N or dissolved organic N (Hadas et al., 2004). There may have been gaseous losses of NH₃, N₂O and N₂ from the clover mulch. Substantial ammonia volatilisation and nitrous oxide emissions from N-rich mulches have been documented (Larsson et al., 1998). Finally, efficiency of any resource decreases with the resource's abundance. While N levels here were low compared to very high productivity cereal production systems, these levels are still high with respect to natural or pasture systems, where N is very limiting and therefore used very efficiently.

4.2. Interspecific competition

Brushing not only affected the transfer of N from the clover to the wheat, it reduced competition from clover and weeds. It is likely that the wheat was primarily affected by competition from the clover intercrop, because weed biomass was low in both years.

Above-ground competition for light plays an important role for the early growth of winter wheat in this intercropping system (Thorsted et al., in press). This may partly explain the increase in wheat biomass observed in the treatments with early brushing of the clover. In some cases, there was a reduction in wheat biomass immediately following the brushings, probably caused by mechanical damage to the wheat from brushing.

Above-ground competition for light also plays a role for reducing the growth of the clover (Thorsted et al., in press). After stem elongation in wheat, the light available for growth of clover is severely reduced, and this probably limits BNF in the clover until the wheat is harvested.

The harvest index (HI) of conventionally grown winter wheat is usually 0.44-0.48 (Hay, 1995; Shearman et al., 2005), which is in line with the HI measured for the sole crop (Table 1). The HI was considerably lower in the unbrushed wheat intercrop. Except for the first brushing in 2000, brushing raised the HI of the wheat intercrop to the level of the sole crop. A low HI is generally caused by factors reducing assimilate availability during grain filling. Due to the differences in crop height there is little influence from competition for light on the wheat crop during grain filling (Thorsted et al., in press). It is more likely that the lower HI of the unbrushed intercrop was caused by competition for water during grain filling, which has been shown to lead to reduction in HI (Pan et al., 2003). The larger leaf area index of the intercrop, as seen from the higher clover biomass (Fig. 1e and f), would lead to higher evapotranspiration, using up more of the available soil moisture. This may have been a particular problem in 2001, where there was a potential soil water deficit of 162 mm during the period April to July (Thorsted et al., in press). Alternatively, the increase in HI with brushing could be an effect of wheat plant size (Weiner, 2004).

4.3. Timing of brushings

The largest yield increases were obtained for single brushings during the stem elongation phase (stages GS31 to GS37) in the first half of May. For these brushings, consistent average grain dry matter yield increases of 0.98–1.11 Mg ha⁻¹ were obtained, and the grain N content was increased by 13–25 kg N ha⁻¹. Early brushings resulted in less N being recycled from the clover. On the other hand, the N use efficiency from early brushings was higher than from late brushings. The net effect was an increase in grain N with increasing delay in brushing.

Despite the higher grain N content with later brushings, this did not lead to higher grain yields. The small yield effect of late available N has also been seen in conventional fertiliser trials (e.g. Olesen et al., 2003). This occurs because the yield under conditions of ample water supply is determined by the cumulated light interception of the crop canopy, which in turn is largely determined by early N uptake (Grindlay, 1997). An increase in the N uptake during the vegetative stage affects light interception during a large part of the growing season, whereas N uptake after anthesis will have little or no effect.

The effect of brushings on grain N concentration depended on the balancing effects of brushings on the grain yield and on the grain N content. Early brushings increased grain yield, resulting in lower grain N concentrations, whereas late brushings increased N concentration.

The highest grain yields in both years were obtained with three brushings. However, identical grain N content and comparable yields were obtained with only two brushings (treatment g in 2000 and treatment f in 2001). This suggests that the first

brushing should be performed at GS30/31 to increase the wheat leaf area index, and the second brushing at GS37 to sustain the leaf area during grain filling and reduce competition for water. Late brushing will also increase grain N concentration and grain weight (Table 1). However, the optimal timing will probably depend on the actual climatic and soil conditions and on the architecture of the intercropping system.

4.4. Perspectives

Grain N concentration in the intercropped wheat was considerably higher that of wheat grown alone. Similar effects have been observed in other white clover/winter wheat intercropping experiments (Bergkvist, 2003; Thorsted et al., in press). Grain N concentration was maintained and sometimes increased by clover brushings (Table 1). Protein content is an important factor determining the bread-making quality of wheat, which is an important issue in organic farming (Berry et al., 2002). Intercropping of white clover and winter wheat may help ensure good bread-making quality in organic farming, and this could make mechanical control of clover profitable. Brushing is energyintensive, however, and is currently used only in high-value vegetable crops. There is a need to develop other less costly methods of cutting the clover between the wheat rows. In conventional farming, spraying clover with herbicides may also offer possibilities to trigger leaf senescence and N release (Jones, 1992; Bergkvist, 2003).

In sole clover crops, 75–94% of the total clover N may be derived from BNF (Jørgensen et al., 1999). In the present study, the above-ground N in clover brushed of in the period from 4 May to 29 May 2001 was estimated at 56–199 kg N ha⁻¹. Assuming that 85% of clover N was derived from BNF, this would correspond to a N input of 48–169 kg N ha⁻¹. Only a small proportion of this N fixation was removed in the grains of the intercropped wheat. Continued growth of the white clover after harvest of the wheat will contribute additionally to BNF. This may leave a considerable amount of N in the soil and the clover, which can contribute to the N supply of subsequent crops (Olesen et al., 2002) or to increased nitrate leaching losses (Eriksen, 2001).

5. Conclusions

Brushing of the clover in the winter wheat/white clover intercrop was found to increase wheat N uptake and wheat grain yields. The grain dry matter yield was increased by brushings during the stem elongation phase (GS31 to GS37), whereas grain N concentration was increased by brushings at flag leaf emergence (GS37 to GS39). The intercropped wheat increased grain N concentrations by 0.11–0.39% and may offer possibilities to improve the bread-making quality of organically grown wheat. Mechanical control of the clover in the intercrop can increase wheat N supply and reduce interspecific competition, improving both crop yield and grain quality. However, brushing is a costly operation, and other less costly measures to control the growth of the clover will need to be developed before this system is adopted in practice.

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References

- Anil, L., Park, R.H., Phipps, R.H., Miller, F.A., 1998. Temperate intercropping of cereals of cereals for forage: a review of the potential for growth and utilization with particular reference to the UK. Grass Forage Sci. 53, 301–317
- Bergkvist, G., 2003. Effect of white clover and nitrogen availability on the grain yield of winter wheat in a three-season intercropping system. Acta Agric. Scand., Sect. B, Soil Plant Sci. 53, 97–109.
- Berry, P.M., Sylvester-Bradley, R., Philipps, L., Hatch, D.J., Cuttle, S.P., Rayns, F.W., Goslin, P., 2002. Is the productivity of organic farms restricted by the supply available nitrogen? Soil Use Manage. 18, 248–255.
- Breland, T.A., 1994. Measured and predicted mineralization of clover green manure at low temperatures at different depths in two soils. Plant Soil 166, 13, 20
- Elgersma, A., Schlepers, H., Nassiri, M., 2000. Interaction between perennial ryegrass and white clover under contrasting nitrogen availability: productivity, seasonal patterns of species composition, N₂ fixation, N transfer and N recovery. Plant Soil 221, 281–299.
- Eriksen, J., 2001. Nitrate leaching and growth of cereal crops following cultivation of contrasting temporary grasslands. J. Agric. Sci., Camb. 136, 271–281.
- Grindlay, D.J.C., 1997. Towards an explanation of crop nitrogen demand based on the optimization of leaf nitrogen per unit leaf area. J. Agric. Sci., Camb. 128, 377–396.
- Hadas, A., Agassi, M., Zhevelev, H., Kautsky, L., Levy, G.J., Fizik, E., Gotessman, M., 2004. Mulching with composted municipal solid wastes in the Central Nevev, Israel. II. Effect on available nitrogen and phosphorus and on organic matter in soil. Soil Till. Res. 78, 115–128.
- Hansen, B., 1989. Determination of nitrogen as elementary N, an alternative to Kjeldahl. Acta Agric. Scand. 39, 113–118.
- Hay, R.K.M., 1995. Harvest index: a review of its use in plant breeding and crop physiology. Ann. Appl. Biol. 126, 197–216.
- Høgh-Jensen, H., Schjørring, J.K., 2000. Below-ground nitrogen transfer between different grassland species: direct quantification by ¹⁵N leaf feeding compared with indirect dilution of soil ¹⁵N. Plant Soil 227, 171–183.
- Jones, L., 1992. Preliminary trials using a white clover (*Trifolium repens* L.) understorey to supply the nitrogen requirements of a cereal crop. Grass Forage Sci. 47, 366–374.
- Jones, L., Clements, R.O., 1993. Development of a low input system for growing wheat in a permanent understorey of white clover. Ann. Appl. Biol. 123, 109–119.
- Jørgensen, F.V., Jensen, E.S., Schjorring, J.K., 1999. Dinitrogen fixation in white clover grown in pure stand and mixture with ryegrass estimated by the immobilized ¹⁵N isotope dilution method. Plant Soil 208, 293–305.
- Jørgensen, F.V., Ledgard, S.F., 1997. Contribution from stolons and roots to estimates of the total amount of N₂ fixed by white clover (*Trifolium repens* L.). Ann. Bot. 80, 641–648.

- Lancashire, P.D., Bleiholder, H., van den Boom, T., Langelüddeke, P., Strauss, R., Weber, E., Witzenberger, A., 1991. A uniform decimal code for the growth stages of crops and weeds. Ann. Appl. Biol. 119, 561–601.
- Larsson, L., Ferm, M., Kasimir-Klemedtsson, Å., Klemedtsson, L., 1998. Ammonia and nitrous oxide emissions from grass and alfalfa mulches. Nutr. Cycl. Agroecosyst. 51, 41–46.
- Ledgard, S.F., Freney, J.R., Simpson, J.R., 1985. Assessing nitrogen transfer from legumes to associated grasses. Soil Biol. Biochem. 17, 575–577.
- Ledgard, S.F., Steele, K.W., 1992. Biological nitrogen fixation in mixed legume/grass pastures. Plant Soil 141, 137–153.
- Mueller, T., Thorup-Kristensen, K., 2001. N-fixation of selected green manure plants in an organic crop rotation. Biol. Agric. Hort. 18, 345–363.
- Nielsen, J.D., Møberg, J.P., 1985. Klassificering af jordprofiler fra forsøgsstationer i Danmark. Tidsskr. Planteavl 89, 157–168.
- Olesen, J.E., Rasmussen, I.A., Askegaard, M., Kristensen, K., 2002. Whole-rotation dry matter and nitrogen grain yields from the first course of an organic farming crop rotation experiment. J. Agric. Sci., Camb. 139, 361–370.
- Olesen, J.E., Jørgensen, L.N., Petersen, J., Mortensen, J.V., 2003. Effects of rates and timing of nitrogen fertiliser on disease control by fungicides in winter wheat. 1. Crop yield and nitrogen uptake. J. Agric. Sci., Camb. 140, 1–13.
- Pan, X.Y., Wang, G.X., Yang, H.M., Wie, X.P., 2003. Effect of water deficits on within-plot variability in growth and grain yield of spring wheat in northwest China. Fields Crops Res. 80, 195–205.
- Petersen, J., Djurhuus, J., 2004. Sammenhæng mellem tilførsel, udvaskning og optagelse af kvælstof i handelsgødede, kornrige sædskifter. DJF rapport Markbrug 102, Danish Institute of Agricultural Sciences, Tjele, Denmark.
- SAS Institute, 1999. SAS/STAT User's Guide, The SAS system for Windows, Version 8. SAS Institute, Cary, NC.
- Shearman, V.J., Sylvester-Bradley, R., Scott, R.K., Foulkes, M.J., 2005. Physiological processes associated with wheat yield progress in the UK. Crop Sci. 45, 175–185.
- Stern, W.R., 1993. Nitrogen fixation and transfer in intercrop systems. Field Crops Res. 34, 335–356.
- Thorsted, M.D., Olesen, J.E., Koefoed, N., 2002a. Intercropping of oats (Avena sativa L.) with different white clover (Trifolium repens L.) cultivars. Effects on biomass development and oat yield. J. Agric. Sci., Camb. 138, 261–267.
- Thorsted, M.D., Soegaard, K., Koefoed, N., 2002b. Yield and quality of oat/white clover intercrops. Grassland Sci. Europe 7, 94–95.
- Thorsted, M.D., Olesen, J.E., Weiner, J. Width of wheat rows and clover strips influence grain yield in winter wheat/white clover intercropping. Field Crops Res., in press.
- Trannin, W.S., Urquiaga, S., Guerra, G., Ibijbijen, J., Cadisch, G., 2000. Interspecies competition and N transfer in a tropical grass-legume mixture. Biol. Fertil. Soils 32, 441–448.
- Vinther, F.P., Jensen, E.S., 2000. Estimating legume N_2 fixation in grass-clover mixtures of a grazed organic cropping system using two ^{15}N methods. Agric. Ecosyst. Environ. 78, 139–147.
- Weiner, J., 2004. Allocation, plasticity and allometry in plants. Perspect. Plant Ecol. Evol. Syst. 6, 207–215.
- Xiao, Y., Li, L., Zhang, F., 2004. Effect of root contact on interspecific competition and N transfer between wheat and fababean using direct and indirect ¹⁵N techniques. Plant Soil 262, 45–54.