**SLU EkoForsk Research Program**

# Final report

## Multifunctional cover crops for stockless organic farming systems


**Erik Steen Jensen**
**Cropping Systems Ecology**

**Department of Biosystems and Technology**
**Faculty of Landscape Architecture, Horticulture and Crop Production Science**
**Swedish University of Agricultural Sciences**


SLU

**Front cover page photo:** Mixture of oil radish and common vetch cover crops in October 2012 (E.S. Jensen)

## Foreword

This document is the final reporting of the project: "**Multifunctional cover crop for stockless organic farming systems"** funded by the SLU EkoForsk research program 2011-2013. Due difficult weather conditions it was not possible to establish cover crops in the autumn of 2011. Consequently, the whole project was postponed one year.

Preliminary research results were presented at the FoU-day within organic agriculture on the 10 April 2014 in Linköping. The preliminary results have been presented in several other seminars and in lectures. This report forms the basis for an international publication and a popular scientific article.

Alnarp 2015-03-27

Erik Steen Jensen

**Summary**

Cover crops (CC) are essential components of arable systems, since they deliver multiple ecosystems services. They may retain nitrogen (N) within systems by their "N catch crop function", which is of particular importance in stockless organic systems, and they may supply symbiotically fixed N, which contribute to their "green manuring function" as well as supply organic carbon to build soil fertility. Cover crops can contribute these services, but more knowledge is required on the multifunctionality of CC species or species mixtures and how these crops influence the soil fertility and nutrient use in rotational sequences.

The aim of the project is to determine the efficiency of CC for stockless organic systems for soil mineral N retention, $N_2$ fixation and organic matter supply and the effect of these crops on subsequent crops in the rotation. Mixtures and sole CCs of common vetch, hairy vetch, oil radish and winter rye will be studied in an organic rotation after cultivation of spring barley and spring pea in a 3-year project. Multifunctional annual CCs will be compared with an under-sown grass-clover CC and the effect of CC measured in a subsequent spring wheat crop.

The biomass production of cover crops varied between 1.0 and 4.1 Mg ha$^{-1}$. The results showed that the mixtures had higher biomass yields than the sole crop vetches, but the mixture dry matter yields were similar to the oil radish or winter rye sole crop yields. A positive relation between soil mineral N content after harvest of main crops (greater after pea than after barley) and the proportion of non-legume was hypothesized. It was not possible to find support for the hypothesis in the frost-killed CC mixture of oil radish and common vetch, but the hypothesis was accepted for the grass-clover and the hairy vetch-winter rye mixtures.

Hairy vetch was the most efficient $N_2$ fixing crop, fixing almost 40 kg N ha$^{-1}$ in mixture with winter rye. Common vetch fixed much less. The catch crop function of CCs was estimated by determining the soil N accumulation in legumes and non-legumes of the CCs. In the frost-killed CCs the accumulation of soil N was similar in oil radish and the mixture of oil radish and common vetch, but greater in the mixture than in common vetch. For over-wintering CCs the hairy vetch and winter rye CC mixture did not recover more soil N than the grass-clover mixture or the sole crop winter rye CC. The symbiotic $N_2$ fixation by the common vetch component in the CC mixture did not result in a greater total N content of the mixture than of oil radish. However, the over-wintering hairy vetch-winter rye mixtures provided the same efficiency in catch crop function and in addition to this 10 to 26 kg N ha$^{-1}$ extra N in its green manure function, due to symbiotic $N_2$ fixation.

The biomass C-to-N ratios were generally lower than 20. With wider ratios it is likely that, at least for some time, there will net immobilization of N. The grass-clover CC biomass had C/N of 23 to 27. The sole crop vetches had C-to-N ratios of 11-14 and the oil radish and winter rye 14-19. The mixtures had intermediate C-to-N ratios of 11-16, which would be more optimal for net N mineralization, but restricting net release and leaching more than the sole crop vetches. The highest subsequent spring wheat grain yields and total N accumulation in the short term were obtained for the vetch cover crops and the mixtures of vetches and non-fixing species oil radish and winter rye. The undersown grass-clover mixture contributed with the greatest amount of carbon, which is important in the long-term for soil organic matter building, but the net immobilization of N in the spring and regrowth of perennial ryegrass in the spring wheat, probably led to the low spring wheat grain yields.

As average of years and precrops the vetch containing CCs had the strongest positive effect on the spring wheat yield. Common vetch and hairy vetch sole crop CCs on average increased the spring wheat yield 15% over the no CC treatment; after barley hairy vetch increased the spring wheat yield with 21-25%. The common vetch-oil radish and the hairy vetch-winter rye increased the grain yield by 14 and 5%, respectively. The grass-clover and winter rye CCs reduced the spring wheat yield with 12% relative to the non-CC control. Oil radish enhanced the yield by 9%.

It is concluded that establishing a cover crop mixture of hairy vetch and winter rye is an efficient was of maximizing the catch crop and green manure functions of a cover crop in a stockless organic crop rotation. The CC mixture management, e.g. placement in the rotation, the proportion of hairy vetch and winter rye in the mixture and incorporation time and technique needs further research.

**Contents** Page

## 1. Introduction

Organic farming systems are based on principles of efficient retention and recycling of nitrogen to reduce the requirement for inputs of new reactive N and prevent losses to the environment (Jensen, 1991; Beck-Friis et al.,1994; Schröder et al., 1998; ). Eco-functional intensification (Niggli et al., 2008) based on soil biodiversity, soil organic matter and ecosystem services are key components for maintaining soil fertility and quality (Carter et al., 2004). A high degree of soil cover throughout the year is a fundamental prerequisite for sustainable soil management and this is achieved quiet easily in systems with pastures.

Cover crops (CC) may contribute many ecosystem services in agroecosystems (Schipanski et al., 2014). In arable organic systems cover crops are used for nutrient retention, reducing nutrient leaching and $N_2O$ emission and enhancing biomass production ($CO_2$ capture/carbon sequestration) during periods with no main crops (Knudsen et al., 2006; Papendick and Elliot, 1984). In addition to their role in nutrient retention, CCs: supply organic matter to soils (Sainju et al., 2002), improve the soil structure by enhanced aggregate stability and greater porosity and reduce the requirement for soil tillage (Stokholm, 1979). Cover crops (legumes) may symbiotically fix dinitrogen, protect soils against erosion (Laloy and Bielders, 2010) contribute to agrobiodiversity and they are tools to manage weeds by competition and allelopathic substances, diseases and pests (Thorup-Kristensen et al., 2003; Campiglia et al., 2010). In addition CCs may be harvested and used as feed or biomass feedstock in bioenergy production. Via their potential multiple functions, there is an increasing interest in using CCs for enhancing agronomic main crop performance. Thus, CCs are essential components of arable systems that thrive towards greater sustainability.

The cover crop should be designed for specific goals and functions within a specific crop rotation and system context. The many challenges of agriculture for sustainable development and production of food require a multifunctional approach for the redesign of future cropping systems (IAASTD, 2009). Multiple functions and efficient use of natural resources may not be obtained by a single CC species, but rather with mixtures (Thorup-Kristensen et al, 2003, Wortman et al., 2012).

Reducing nitrate leaching is a key process in which CCs may be very efficient (Jensen, 1991a; Knudsen et al., 2006), and in stockless organic agricultural systems it is of particular importance to retain and recycle nutrient sources and to enhance the inputs of nitrogen from symbiotic $N_2$ fixation and organic matter. Cover crop multifunctionality for N retention, nitrogen fixation and organic matter acquisition would benefit these systems. Grass-clover may be undersown main crops as a CC, but it is of interest to develop more multifunctional CC mixtures for arable systems based on annual CC species, due to problems with too much clover in rotations. Secondly, the effect of grass-clover CCs on nitrogen mineralization-immobilization turnover in spring may lead to net immobilization of N, due to the high C/N ratio of the incorporated materials (Jensen, 1991a; Hauggaard-Nielsen et al., 2012). It has been shown that 30-40% of the N added in a ryegrass CC was recovered in the subsequent spring barley (Thomsen, 1993), but it is essential to enhance the use efficiency of N from incorporated CCs by a better synchronization of the nitrogen availability with subsequent crop requirement for N (Jensen, 1991b). The synchrony may be optimized by the composition of the CC and the management methods.

Nitrogen dynamics following a given crops depend on the specific crop N use efficiency, the residual soil mineral N and the C/N ratio of the crop residues as well and the climatic conditions during autumn and winter. Thus the N availability will vary in space within the field and time (Hauggaard-Nielsen et al., 2009). After a cereal the soil inorganic in the autumn is typically lower than after a grain legume (Jensen, 1991a; Hauggaard-Nielsen et al., 2012), but variable across the field. Thus in some part of the fields a CC efficient in acquiring soil N mineral N may be required, whereas in other parts low levels of N may create good conditions for symbiotic $N_2$-fixation. This has led to term *Ecological Precision Farming* (EPF) (Jensen et al., 2015) in which a mixture of a legume and non-legume self-regulate across the field according to the spatial availability of soil mineral N.

## 2. Objectives and Hypotheses

The aim of this project is to determine the potential of annual multi-species cover crop mixtures after a cereal (barley, *Hordeum vulgare* L.) and a grain legume (pea, *Pisum sativum* L.) for stockless systems with the main functionalities:

- soil mineral N retention
- symbiotic nitrogen fixation during autumn
- organic matter supply, and
- N supply to the following main crop.

Two types of cover crops were used frost-killed and over-wintering types established shortly after main crop harvest in August. The frost-killed CCs were common vetch (CV, *Vicia sativa*) and oil radish (OR, *Raphanus sativus oleiformis)*. The overwinterings CCs were hairy vetch (HV, *Vicia villosa*) and winter rye (WR, *Secale cereale*). These CCs were grown as sole crops (SC) or CC mixtures. A perennial ryegrass (*Lolium perenne)*-white clover (*Trifolium repens)* CC, undersown to main crops were used for comparison.

The following main hypotheses were tested:

i. The biomass production of the CC mixtures are greater than sole crop CCs
ii. Cover crop mixtures of CVxOR and HVxWR have greater recovery of soil inorganic N during autumn/winter than a perennial ryegrass (*Lolium perenne)*-white clover (*Trifolium repens)* CC and the individual species as sole CCs.
iii. CC mixtures with common and hairy vetch have a greater symbiotic nitrogen fixation than white clover in autumn and early spring.
iv. The pre-crop will influence the dynamics of subsequent CC mixture component competition and composition via its effect on available soil N in the autumn. It is hypothesized that there is a positive correlation between soil mineral N after the pre-crop and the proportion of non-legume CC biomass. A higher amount of soil mineral N is expected after the main crop grain legume than after the cereal.
v. The release of nitrogen from incorporated mixtures of CVxOR and HVxWR is in better synchrony with the subsequent crop N requirement than after sole crop CCs and a grass-clover resulting in enhanced CC N recovery in the subsequent crop, due to a more optimal C/N ratio of the CC biomass for net N release and synchronization with the subsequent crop N demand.

## 3. Materials and Methods

### *3.1 Site and soil*

The hypotheses will be tested in field experiments at the SLU Alnarp organic crop rotation (55º39´N, 13º05´E; KRAV-certified since 1993) during 2012-2013 and 2013-2014. The experiment was initially established 2011, but due to an extraordinary wet autumn 2011, it was impossible to establish CCs after main crops. The experiment was cancel and the whole project was postponed one year. The sites has sandy loams with c. 14% clay in 2012-2013 and 22% clay in 2013-14, pH 7.8, soil organic matter content 7.1% and 3.3% in the two experiments, respectively. The P content on the soil in the second experiment was somewhat lower than in the first experiment.

### *3.2 Cropping system sequence and CC treatments*

The cropping systems sequence was main crops of pea (80 plants $m^{-2}$) and barley (300 plants $m^{-2}$, fertilized 50 kg N $ha^{-1}$ in Biofer), followed by CC treatments in the autumns and spring wheat in the subsequent year (Figure 1, Table 1)

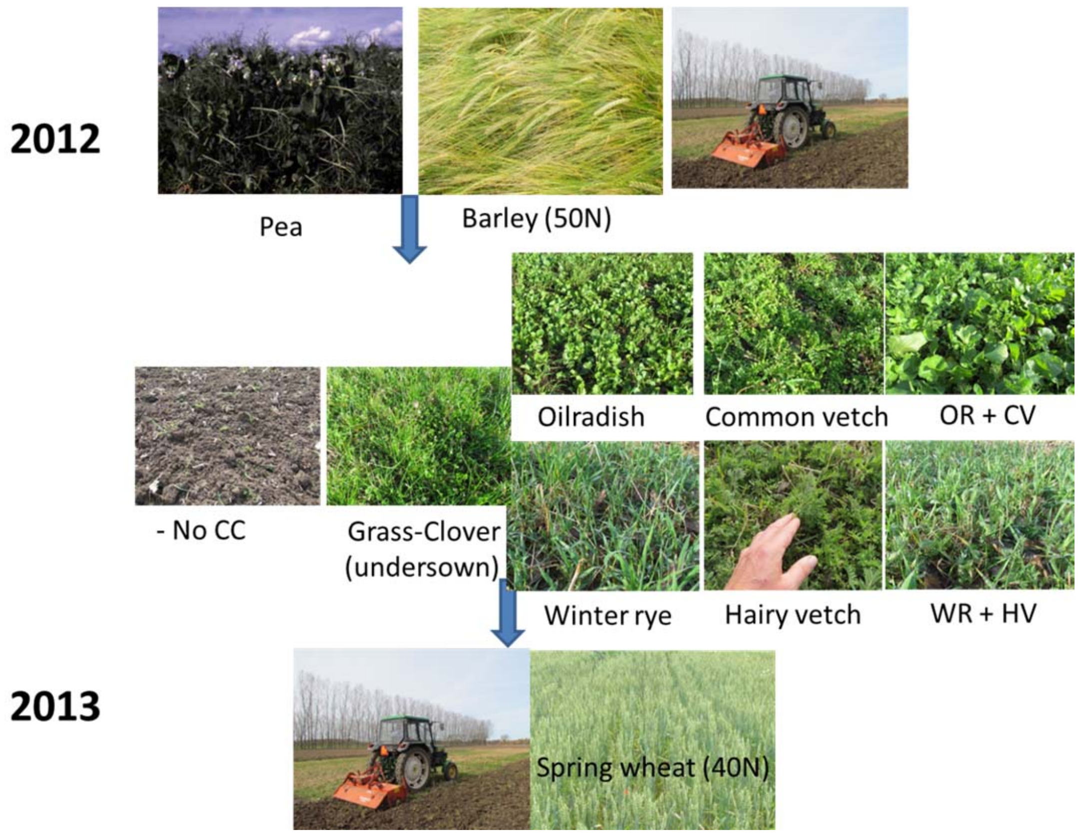



**Figure 1**. Cropping rotation sequence (shown for 2012-2013). Field plots were rotor-tilled after main crop harvest in August and the annual CCs were sown as soon as possible. In the spring CCs were rotor-tilled in April and sown with spring wheat. 40N and 50N indicate that the crop received 40 and 50 kg N $ha^{-1}$ in Biofer.

The CCs are established after main crops spring barley and pea to potentially obtain two levels of soil inorganic N in autumn after harvest of the precrops (Figure 1). The CC crops were: I) Perennial ryegrass-white clover mixture under-sown the main crops in the spring (90 % of normal sowing rate in perennial ryegrass and 10% of normal sowing rate in white clover) and incorporated in the subsequent spring along with overwintering annual CCs, II) sole crops and mixtures of oil radish (1.6 g $m^{-2}$ in SC) and common vetch (8 g $m^{-2}$ in SC) sown as soon as possible after main crop harvest in August and residual frost-killed biomass incorporated in spring, and III) sole crops and mixtures of winter rye (15 g $m^{-2}$ in SC) and hairy vetch (8 g $m^{-2}$ in SC) established as soon as possible after harvest of the main crop and incorporated in following spring (Table 1). The sowing rates of annual CC mixtures are 50%:50% of the respective sole crop sowing rates. Spring wheat was sown in 2013 and 2014 (300 pl. $m^{-2}$) after incorporation by rotor-tilling of the CCs and fertilized 40 kg N $ha^{-1}$ in Biofer. Cultivars of the crops species used are shown in Table 2.

**Table 1.** Crop management and analyses in field experiments 2012-2013 and 2013-2014.

| Field activity | Field experiment 2012-13 | Field experiment 2013-14 |
|---|---|---|
| Sowing of pea and barley | 13 April 2012 | 3 April 2013 |
| Under-sowing of grass-clover CC | 14 April 2012 | 3 April 2013 |
| Harvest pea and barley | 5 August 2012 | 3 August 2013 |
| Sowing of cover crops | 5 August 2012 | 9 August 2013 |
| Soil sampling for mineral N | 16 August 2012 | 13 September 2013 |
| Autumn growth analysis of CC | 26 October 2012 | 1 November 2013 |
| Spring growth analysis of CC | 9 April 2013 | 3 April 2014 |
| Sowing of spring wheat | 10 April 2013 | 29 April 2014 |
| Harvest of spring wheat | 27 August 2013 | 16 August 2014 |

### *3.3 Experimental design*

Crops will be established CC in plots 2 x 9 m in four replicate blocks using a split-plot design with main crops a main plots and CC treatments as split plots.

**Table 2.** Species and cultivars used in experiments

| Species | Experiment 2012-2013 | Experiment 2013-2014 |
|---|---|---|
| Barley (*Hordeum vulgare* L.) | Marcelo | Tam Tam |
| Pea (*Pisum sativum* L.) | Clara | Clara |
| Perennial ryegras (*Lolium perenne* L.) | Aberdart | Aberdart |
| White clover (*Trifolium repens* L.) | AberCrest | AberCrest |
| Common vetch (CV) (*Vicia sativa* L.) | Candy | Candy |
| Oil radish (OR) (*Raphanus sativus* L.) | Colonel | Colonel |
| Winter rye (WR) (*Secale cereale* L. ) | Amilo | Amilo |
| Hairy vetch (HV) (*Vicia villosa* L.) | Hunga villosa | Hunga villosa |
| Spring wheat (*Triticum aestivum* L.) | Diskett | Dacke |

### *3.4 Measurements and harvests*

Yields of main crops of pea and barley were determined by harvesting whole plants within 2 x 1 $m^2$ within each of the four blocks of pea and barley. In 2013 heavy lodging resulted in serious seed losses. Crops were dried at 55ºC for 24-48 hours and separated in straw and grain. Milled grain and straw samples weighed into tin capsules for determination of total N by elemental analysis using a Thermo Scientific Flash 2000 elemental analyser.

Soil inorganic N in the 0-20 and 20-50 cm soil depth was determined by soil sampling using a Eijelkamp soil drill and 2 x 10 samples per block of respective main crops. The 10 samples from each main crop block and soil layer were pooled into a total of 2 x 2 x 2 x 4 samples. Soil samples were extracted with 2 M KCl with a soil:KCl ratio of 1:10, shaking for 1 hour and centrifugation. Ammonium and nitrate were determined in the liquid using an Autoanalyzer 3.0 and simultaneous ammonium and nitrate determination. Soil dry matter determination was determined by drying in 20 h at 105ºC.

Cover crops were sampled for biomass production and botanical composition from 0.5 $m^2$ frame in one end of the plot. The cover crop were separated in non-fixing and $N_2$ fixing species and in 2013 emerged pea plants form seeds lost during harvest were sampled separately. The SC vetch crops contained non-fixing weeds, which were sampled separately.

Samples were dried at 55º C for 24-48 hours, weighed and milled for total N and $^{15}N$ natural abundance determination by isotope ratio mass spectrometry at Risø National Laboratory, DTU, Denmark using stable isotope ratio mass spectrometry (CE Instruments EA 1110) coupled in continuous flow mode to an isotope ratio mass spectrometer (Finnigan MAT DeltaPlus). The proportion of symbiotic $N_2$ and soil N acquisition and in CCs were determined by stable nitrogen isotope methodology based on $^{15}N$ natural abundance (Peoples et al., 1997, Unkovich et al. 2008). The proportion of $N_2$ fixation in cover crop legumes were determined from the equation:

$$\% \text{ Pfix} = 100 \times (^{15}Na_{reference} - {}^{15}Na_{legume})/(^{15}Na_{reference} - \beta).$$

$^{15}Na_{reference}$ is the natural abundance of an non-fixing reference plant growing in the same plot as the legume (mixture species or non-fixing weed), $^{15}Na_{legume}$ is the natural abundance of the legume, and β is the $^{15}N$ abundance of the test legume dependent completely on $N_2$ fixation for growth. β for the vetches were set to -0.70 (Unkovich et al., 2008), for pea to -0.72 (Knudsen et al., 2004) and white clover -1.4 (Eriksen and Høgh-Jensen, 1998) . The proportion of N derived from the soil in legumes was estimated as; %Psoil=100 - %Pfix. All N in non-legumes were assumed to be derived from soil inorganic N sources.

Spring wheat following CCs were harvested medio to late August by harvesting 0.5 $m^2$ within the plot. Subsequently all weed within the 0.5 $m^2$ plot were collected. Subsequently whole plots were harvested by experimental combiner (data not shown). Plant materials were dried (55ºC 24 h) and the wheat separated in grain and straw, weighed and total N determined milled samples using elemental analysis (see above). Weeds were dried (50ºC 24 h) and total dry matter determined. The protein concentration in spring wheat was calculated as 6.25 × %N in grain.

### *3.5 Statistical analysis*

Data was analysed statistically using Minitab 16 and GLM for analysis of variance. $LSD_{0.95}$was used for comparison of means if main effects: main crop or CC treatment or the interaction was significant.

## 4. Results

### *4.1 Main crops*

Pea and spring barley developed well in 2012 and 2013. Barley yielded similarly in the two years, on average 5.3 t $ha^{-1}$, whereas the pea yield was much lower in 2013 due to seed losses (Table 3, Appendix 1). Assuming a ratio between pea grain and straw of 2.0 as in 2012, the 2013 grain yield would have been close to 7 Mg $ha^{-1}$. Many pea seeds emerged during the autumn, which made it necessary to separately harvest pea plants in the plots.

The pea straw N concentration was rather low, 1.1-1.3% N or a C/N ratio of c. 36, which would not result in significant net mineralization, during the first months. The amount of soil mineral N was always greater in the 30 cm layer below the plough-layer than in the plough-layer, and only slightly greater after the pea compared to barley in 2013 (ca. 12 kg N/ha) (Table 3).

**Table 3.** Grain and straw dry matter and nitrogen accumulation and soil mineral N (0-20, 20-50 cm, kg $NO_3$-N+$NH_4$-N $ha^{-1}$) after harvest 2012 and 2013.Soil sampling 2012: 11days after crop harvest. 2013: 41 days after crop harvest. Mean of eight replicates ± standard deviation

| DM or N | Barley 2012 | Pea 2012 | Barley 2013 | Pea 2013 |
|---|---|---|---|---|
| Grain Mg $ha^{-1}$, DM | 5.33 ± 0.59 | 4.52 ± 0.45 | 5.20 ± 0.20 | 2.41 ± 0.27 |
| Straw Mg $ha^{-1}$, DM | 3.04 ± 0.31 | 2.27 ± 0.26 | 4.11 ± 0.21 | 3.47 ± 0.50 |
| %N grain | 1.45 ± 0.06 | 3.28 ± 0.18 | 1.42 ± 0.07 | 3.27 ± 0.39 |
| %N straw | 0.73 ± 0.03 | 1.29 ± 0.08 | 0.67 ± 0.05 | 1.06 ± 0.26 |
| Grain kg N $ha^{-1}$ | 75 ± 10.0 | 148 ± 15.2 | 74 ± 4.4 | 70 ± 8.8 |
| Straw kg N $ha^{-1}$ | 22 ± 2.5 | 45 ± 2.4 | 28 ± 3.0 | 37 ±13.0 |
| | | | | |
| Soil min N 0-20 cm | 10 ± 4.0 | 13 ± 8.0 | 8.7 ± 3.7 | 15.6 ± 7.9 |
| Soil min N 20-50 cm | 20 ± 11.0 | 21 ± 11.1 | 19.1 ± 4.2 | 24.1 ± 3.3 |
| Soil min N 0-50 cm | 30 | 34 | 27.8 | 39.7 |

### *4.2 Dry matter production of cover crops*

4.2.1 Frost-killed CC

Cover crops of common vetch (CV) and oil radish (OR) and the mixture of the two CCs (CV+OR) were sown within 9 days after harvest of main crops (Table 1). Table 4 shows dry matter production, nitrogen concentration and C/N ratio of the aboveground CC biomass in the two experiments. Figure 2 shows the total dry matter production of the CCs.

The sole crop of common vetch produced only about 0.3 to 0.8 Mg $ha^{-1}$ and germinated barley and non-fixing weeds had similar biomass production (Table 4). Oil radish had much greater biomass production in sole and mixed cropping than the CV, from 1.0 to 1.5 Mg $ha^{-1}$. In 2013 the germinated pea seeds constituted a high proportion of the biomass and they were harvested separately (Table 4). Total DM production of the CV, OR and CV+OR cover crops incl. other weeds after pea, but excluding pea, were 1.0, 1.5 and 1.5 Mg $ha^{-1}$, respectively. The total dry matter was higher in the CCs with germinated pea seed in 2013 (Figure 2). The total biomass production of the frost-killed CC mixture was similar to the OR sole crop, but higher than the CV sole crop. The differences were not significant, but in 2013 there was a significant effect of pre-crop ($P<0.05$). The hypothesis I for frost-killed CC is rejected.

**Table 4.** Dry matter production, % N and C/N ratio of aboveground CC biomass harvested in the late autumn of species which are frost-killed. fix: $N_2$ fixing species (CV and pea), nfix: non $N_2$-fixing species (OR, barley and non-fixing weeds), pea: germinated pea seeds. Means ± standard error (n=4)

| Year and pre-crop | Cover crop | DM fix (Mg/ha) | DM nfix (Mg/ha) | DM pea (Mg/ha) | %N fix | %Npea | %N nfix | C/N CC weighed |
|---|---|---|---|---|---|---|---|---|
| **2012 Frost-killed cover crops** | | | | | | | | |
| Pea | CV | 0.34 ± 0.18 | 0.36 ± 0.06# | NA | 4.77 ± 0.58 | NA | 2.70 ± 0.11 | 11.2 ± 0.81 |
| Barley | CV | 0.32 ± 0.05 | 0.66 ± 0.11§ | NA | 4.37 ± 0.14 | NA | 3.01 ± 0.33 | 13.7 ± 1.53 |
| Pea | OR | 0.22 ± 0.05* | 1.16 ± 0.10 | NA | 4.50 ± 0.12 | NA | 3.54 ± 0.15 | 12.1 ± 0.49 |
| Barley | OR | NA | 1.16 ± 0.13 | NA | NA | NA | 2.92 ± 0.09 | 15.1 ± 0.46 |
| Pea | CV+OR | 0.28 ± 0.10 | 1.14 ± 0.15 | NA | 4.54 ± 0.13 | NA | 3.81 ± 0.66 | 11.3 ± 0.48 |
| Barley | CV+OR | 0.20 ± 0.03 | 1.11 ± 0.04 | NA | 4.40 ± 0.15 | NA | 2.58 ± 0.14 | 16.1 ± 0.79 |
| **2013 Frost-killed cover crops** | | | | | | | | |
| Pea | CV | 0.34 ± 0.03 | 0.62 ± 0.08 | 0.79±0.66 | 3.55 ± 0.21 | 3.7±0.2 | 3.44 ± 0.20 | 12.0 ± 0.52 |
| Barley | CV | 0.59 ± 0.09 | 0.73 ± 0.14 | NA | 4.13 ± 0.15 | NA | 2.70 ± 0.12 | 13.8 ± 0.62 |
| Pea | OR | NA | 1.50 ± 0.17 | 0.92±0.14 | NA | 3.5±0.2 | 2.98 ± 0.30 | 14.3 ± 1.30 |
| Barley | OR | NA | 1.52 ± 0.06 | NA | NA | NA | 2.31 ± 0.07 | 19.1 ± 0.55 |
| Pea | CV+OR | 0.34 ± 0.10 | 1.16 ± 0.29 | 1.22±0.37 | 3.55 ± 0.12 | 3.2±0.2 | 2.75 ± 0.22 | 15.3 ± 0.50 |
| Barley | CV+OR | 0.24 ± 0.02 | 1.00 ± 0.06 | NA | 3.99 ± 0.09 | NA | 2.78 ± 0.31 | 15.0 ± 0.84 |

* incl. germinated pea seeds, # weeds, § weeds and germinated barley seeds, NA: Not available

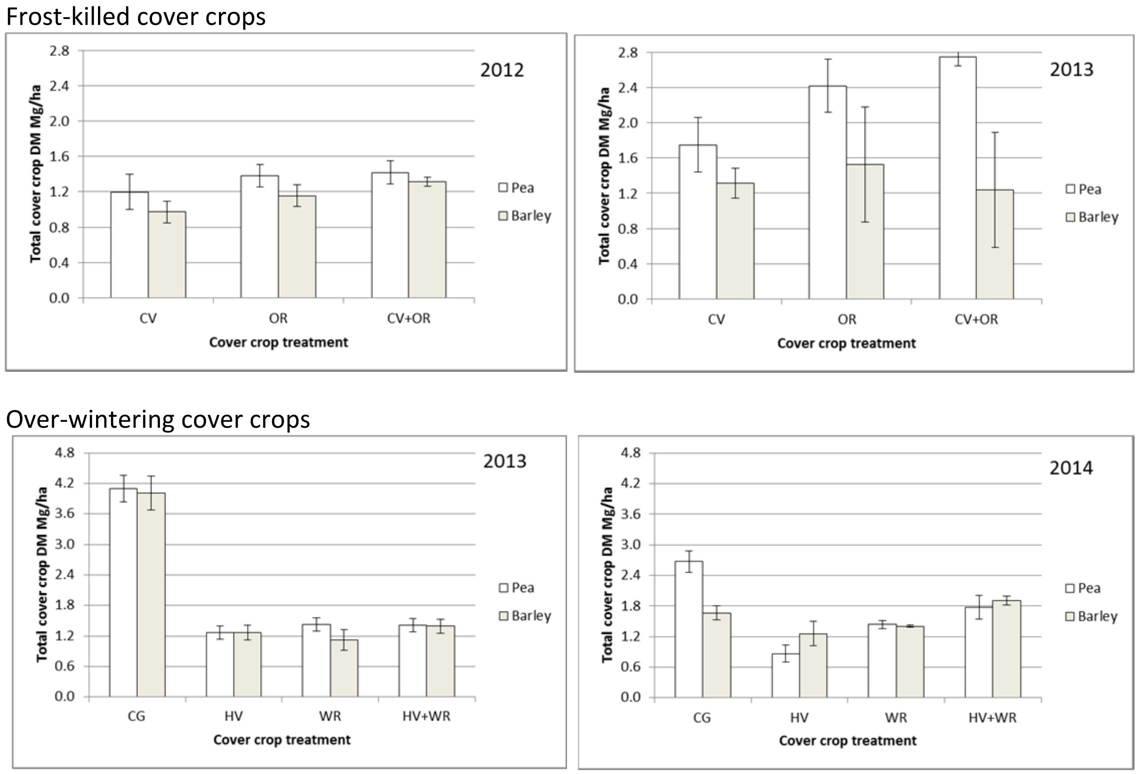



**Figure 2.** Analysis of dry matter production of frost-killed CCs in late October – early November in 2012 and 2013 (upper row) and of over-wintering CCs in April in 2013 and 2014 (lower row) after main crops pea and barley. Values are means of four replicates ± standard error.

It was not possible to find support for the hypothesis IV that the proportion of non-legume biomass in the CC mixtures was greater after pea than after barley, due to higher levels of soil mineral N after the pea (Table 4). As expected the C/N ratio of the total biomass was lowest in the common vetch sole crop CC and lower after pea than after barley, probably due to a greater N availability leading to higher N concentrations in non-legume CCs (Table 4).

4.2.2 Over-wintering CC

Dry matter production, N-concentration and C/N ratios of over-wintering CCs are shown in Table 5 and Figure 2. In the grass-clover mixture the white clover biomass was greater after barley than after pea and the reverse was the case for the ryegrass (Table 5). This was also observed for the mixture of hairy vetch and winter rye in both years. However, the total biomass of the hairy vetch+ winter rye mixture was not significantly different for pre-crops pea and barley (Figure 2). Thus the hypothesis IV can be confirmed for the CG and the HV+WR mixtures. The total HV+WR biomass was 29 and 73% greater than the sole crop WR and HV biomasses, respectively in 2014. The difference between the mixture and HV was significant ($P<0.05$). No significant differences were found in 2013 for the HV and WR CCs, but the GC had a significantly greater production than the other CCs. The Hypothesis I must be rejected in both years.

The grass-clover mixture had the highest C/N ratios of 23-27, with the lower C/N ratios after pea due to the greater N concentration in the grass after the pea crop (Table 5). The hairy vetch had C/N ratios around 11, winter rye 14-20 and the mixture of vetch and rye intermediate of 12-14 (Table 5). The N concentration in the non-legume CCs were generally much greater after pea than after barley.

**Table 5.** DM production, nitrogen concentration and C/N ratio of cover crops harvested in the spring. GC: Grass clover CC, HV: Hairy vetch, WR: Winter rye. fix: $N_2$-fixing species, nfix: non-$N_2$-fixing species. Means ± standard error (n=4, 2013, n=3, 2014)

| Pre-crop | Cover crop | DM fix (Mg/ha) | DM nfix (Mg/ha) | % N fix | % N nfix | C/N CC weighed |
|---|---|---|---|---|---|---|
| **2013 Over-wintering cover crops** | | | | | | |
| Pea | GC | 0.12 ± 0.06 | 3.99 ± 0.21 | 3.27 ± 0.17 | 1.88 ± 0.09 | 23.2 ± 0.99 |
| Barley | GC | 0.54 ± 0.34 | 3.30 ± 1.25 | 3.10 ± 0.20 | 1.69 ± 0.08 | 24.7 ± 2.06 |
| Pea | HV | 1.27 ± 0.13 | NA | 3.60 ± 0.26 | NA | 12.4 ± 0.94 |
| Barley | HV | 1.26 ± 0.14 | NA | 4.17 ± 0.14 | NA | 10.6 ± 0.35 |
| Pea | WR | NA | 1.43 ± 0.13 | NA | 3.20 ± 0.07 | 13.8 ± 0.30 |
| Barley | WR | NA | 1.12 ± 0.20 | NA | 3.26 ± 0.10 | 13.6 ± 0.46 |
| Pea | HV+WR | 0.43 ± 0.04 | 0.97 ± 0.14 | 3.77 ± 0.09 | 3.61 ± 0.27 | 12.3 ± 0.84 |
| Barley | HV+WR | 0.92 ± 0.18 | 0.46 ± 0.12 | 4.04 ± 0.20 | 3.55 ± 0.33 | 12.0 ± 0.46 |
| **2014 Over-wintering cover crops** | | | | | | |
| Pea | GC | 0.15 ± 0.09 | 2.52 ± 0.29 | 2.65 ± 0.37 | 1.75 ± 0.19 | 25.0 ± 2.35 |
| Barley | GC | 0.25 ± 0.12 | 1.42 ± 0.15 | 3.05 ± 0.24 | 1.52 ± 0.09 | 26.7 ± 1.34 |
| Pea | HV | 0.32 ± 0.04 | 0.55 ± 0.15 | 4.78 ± 0.10 | 3.82 ± 0.35 | 11.1 ± 0.97 |
| Barley | HV | 0.95 ± 0.14 | 0.31 ± 0.11 | 4.46 ± 0.08 | 3.36 ± 0.24 | 10.8 ± 0.35 |
| Pea | WR | NA | 1.43 ± 0.08 | NA | 2.69 ± 0.13 | 16.4 ± 0.82 |
| Barley | WR | NA | 1.40 ± 0.01 | NA | 2.26 ± 0.08 | 19.5 ± 0.71 |
| Pea | HV+WR | 0.34 ± 0.08 | 1.43 ± 0.15 | 4.22 ± 0.07 | 3.04 ± 0.24 | 13.9 ± 0.77 |
| Barley | HV+WR | 0.96 ± 0.01 | 0.95 ± 0.07 | 4.71 ± 0.16 | 2.47 ± 0.15 | 13.7 ± 0.45 |

### *4.3 Nitrogen acquisition of cover crops*

4.3.1 Frost-killed cover crops

Total N acquisition in frost-killed CCs in 2012 varied between 33 and 56 kg N $ha^{-1}$ and in 2013 between 35 and 78 kg N $ha^{-1}$ (Figure 3). The higher accumualtion of 64-78 kg N $ha^{-1}$ in 2013 was due to the regrowth of pea seed lost during the harvest, contributing c. 30-37 kg N $ha^{-1}$. In sole crop oil radish or the CV+OR the total N accumulation in oil radish and non-legume weeds were from 4 to 14 kg N $ha^{-1}$ greater after pea than after barley (Table 6). The amount of $N_2$ fixation was greater after pea as pre-crop due to the pea plant from lost seeds fixing $N_2$ in addition to the common vetch plants (Table 6).

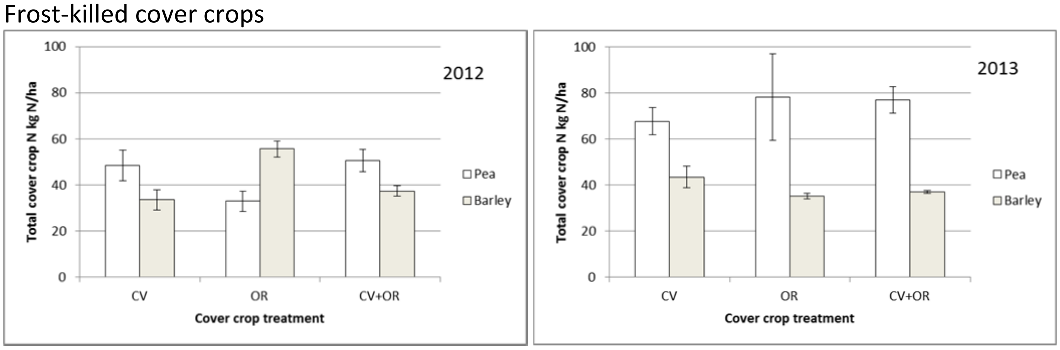
Common vetch fixed a greater proportion of their N from the atmosphere after barley than after pea, when grown as a sole crop and in mixture in 2012, but it was not the case in 2013. Total N fixed in vetch and pea was greater after pea than after barley in both years. In 2013 the reason was mainly due to the germinated pea plants (Table 6). In 2012 the total soil N accumulation in the CCs was significantly ($p< 0.033$ and $p<0.002$, in 2012 and 2013, respectively) greater after pea, on average 28 and 37 kg N $ha^{-1}$ after barley and pea, respectively and in 2013 30 and 46 kg N $ha^{-1}$ respectively (Table 6). There was a significant ($p< 0.016$) effect of CC in 2012 only. This was due to a greater soil N accumulation in the oil

**Table 6.** Nitrogen acquisition in frost-killed cover crops harvested in the late autumn. N fix: N accumulation in vetch, N nfix: N accumulation in non-$N_2$ fixing species. N pea: N accumulation in pea; %Ndfa: % of N in vetch and pea derived from $N_2$-fixation. Ndfa: N derived from $N_2$ fixation. Ndfs: N derived from soil N. Means ± standard error (n=4).

| Year Precrop | Cover crop | N fix (kg/ha) | N nfix (kg/ha) | N pea (kg/ha) | %Ndfa fix | %Ndfa pea | Ndfa fix+ pea (kg/ha) | Ndfs fix + pea (kg/ha) | Ndfs CC (kg/ha) |
|---|---|---|---|---|---|---|---|---|---|
| **2012 Frost-killed cover crops** | | | | | | | | | |
| Pea | CV | 39 ± 7.0 | 10 ± 1.3 | NA | 72 ± 12.0 | NA | 26 ± 6.3 | 13 ± 6.1 | 23 ± 5.6 |
| Barley | CV | 14 ± 2.1 | 19 ± 2.8 | NA | 76 ± 7.3 | NA | 11 ± 2.4 | 3 ± 0.6 | 22 ± 3.2 |
| Pea | OR | 10 ± 2.3* | 41 ± 2.5 | NA | 82 ± 6.8 | NA | 8 ± 2.4 | 2 ± 0.7 | 43 ± 2.6 |
| Barley | OR | NA | 34 ± 3.4 | NA | NA | NA | NA | NA | 34 ± 3.4 |
| Pea | CV+OR | 13 ± 4.0 | 43 ± 5.2 | NA | 87 ± 9.0 | NA | 11 ± 4.4 | 1 ± 0.4 | 44 ± 5.4 |
| Barley | CV+OR | 9 ± 1.2 | 29 ± 1.9 | NA | 100 ± 0.5 | NA | 9 ± 1.2 | 0 ± 0.0 | 29 ± 1.9 |
| **2013 Frost-killed cover crops** | | | | | | | | | |
| Pea | CV | 12 ± 3.5 | 22 ± 3.5 | 30 ± 6.7 | 65 ± 6.4 | 73 ± 8.2 | 28 ± 3.3 | 15 ± 3.7 | 39 ± 5.0 |
| Barley | CV | 24 ± 2.8 | 20 ± 3.5 | NA | 73 ± 5.3 | NA | 17 ± 2.4 | 7 ± 1.5 | 26 ± 4.4 |
| Pea | OR | NA | 45 ± 7.5 | 33 ± 6.2 | NA | 79 ± 3.3 | 26 ± 3.9 | 8 ± 2.4 | 53 ± 9.8 |
| Barley | OR | NA | 35 ± 1.3 | NA | NA | NA | NA | NA | 35 ± 1.2 |
| Pea | CV+OR | 7 ± 2.1 | 32 ± 6.4 | 37±10.5 | 83 ± 3.8 | 70 ± 7.8 | 30 ± 10.1 | 16 ± 8.1 | 47 ± 4.7 |
| Barley | CV+OR | 9 ± 0.5 | 28 ± 0.6 | NA | 75 ± 3.7 | NA | 7 ± 0.5 | 2 ± 0.5 | 30 ± 0.7 |

*Pea seed emerged in autumn

Frost-killed cover crops




Over-wintering cover crops

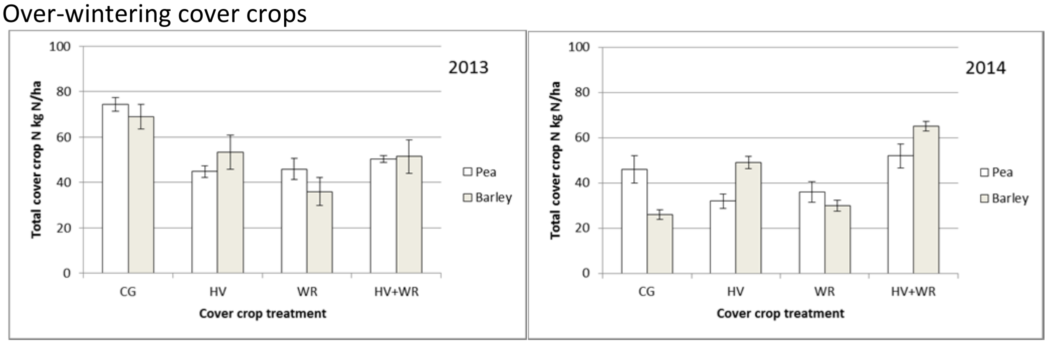



**Figure 3**. Total nitrogen in above-ground plant part of cover crops as affect by pre-crop and year. Top: Frost-killed cover crops harvested late October-early November. Lower part: over-wintering cover crops. CV: Common vetch, OR: Oil radish, CG: White clover-Perennial ryegrass mixture, HV: Hairy vetch; WR: winter rye. Means of four (2012 and 13) or three (2014) replicates±standard deviation.

radish and the mixture of CV and OR compared to the vetch sole crop (Table 6). Thus, the hypothesis II must be rejected, since the CV+OR mixture had not greater uptake of soil N than the sole CCs.

Table 7 Nitrogen acquisition in cover crops harvested in the spring. For explanation see Table 6. GC= Grass clover cc, HV: Hairy vetch, WR: Winter rye. Means ± standard error (n=4 2012, n=3 2013).

| Year and pre-crop | Cover crop | N fix (kg N/ha) | N nfix (kg N/ha) | %Ndfa fix | Ndfa (kg N/ha) | Ndfs fix (kg N/ha) | Ndfs CC (kg N/ha) |
|---|---|---|---|---|---|---|---|
| **2013 Over-wintering cover crops** | | | | | | | |
| Pea | GC | 3 ± 1.7 | 72 ± 6.0 | 55 ± 2.4 | 1 ± 0.6 | 1 ± 0.4 | 73 ± 3.0 |
| Barley | GC | 7 ± 4.0 | 62 ±9.2 | 58 ± 4.3 | 3 ± 2.0 | 3 ± 2.0 | 66 ± 7.2 |
| Pea | HV | 45 ± 2.6 | NA | 14 ±11.3 | 6 ± 4.8 | 39 ± 5.6 | 39 ± 5.6 |
| Barley | HV | 53 ± 7.5 | NA | 67 ± 9.6 | 36 ± 7.3 | 17 ± 5.3 | 17 ± 5.2 |
| Pea | WR | NA | 46 ± 4.8 | NA | NA | NA | 46 ± 4.9 |
| Barley | WR | NA | 36 ± 6.2 | NA | NA | NA | 36 ± 6.2 |
| Pea | HV+WR | 16 ± 1.6 | 34 ± 2.1 | 53 ± 9.8 | 9 ± 1.9 | 8 ± 1.6 | 42 ± 1.1 |
| Barley | HV+WR | 36 ± 4.8 | 15 ± 2.3 | 82 ± 9.4 | 29 ± 1.7 | 8 ± 4.4 | 23 ± 2.7 |
| **2014 Over-wintering cover crops** | | | | | | | |
| Pea | GC | 4 ± 2.6 | 42 ± 8.5 | 51 ± 5.9 | 2 ± 1.7 | 2 ± 0.9 | 43 ± 7.6 |
| Barley | GC | 8 ± 4.1 | 18 ± 2.0 | 63 ± 2.3 | 5 ± 2.8 | 3 ± 1.3 | 21 ± 1.6 |
| Pea | HV | 14 ± 1.9 | 18 ± 3.4 | 75 ± 3.0 | 10 ±1.4 | 4 ± 0.7 | 22 ± 3.2 |
| Barley | HV | 39 ± 6.2 | 9 ± 3.3 | 80 ± 4.3 | 32 ±6.8 | 7± 0.8 | 16 ± 2.8 |
| Pea | WR | NA | 36 ± 4.5 | NA | NA | NA | 36 ± 4.5 |
| Barley | WR | NA | 30 ± 2.3 | NA | NA | NA | 30 ± 2.2 |
| Pea | HV+WR | 13 ± 3.2 | 39 ± 2.0 | 72 ± 4.1 | 10 ± 2.7 | 3 ± 0.7 | 42 ± 2.6 |
| Barley | HV+WR | 43 ± 1.1 | 22 ± 1.2 | 86 ± 3.8 | 37 ± 2.8 | 6 ± 1.5 | 28 ± 1.1 |

3.3.2 Over-wintering cover crops

The total N acquisition in the grass-clover was on average 70 kg N ha$^{-1}$ in 2013, but only 46 and 26 kg N ha$^{-1}$, after pea and barley respectively in 2014 (Fig. 3). The total N acquisition was mainly determined by the grass N accumulation, although white clover fixed twice as much N after barley than after pea (Table 7), due to less competition from the grass. However, symbiotic $N_2$ fixation was only 1-5 kg in white clover (Table 7). The hairy vetch and the winter rye CCs had total N acquisitions between 40 and 60 kg N ha$^{-1}$ (Figure 3). Hairy vetch SC had greater N accumulation after barley than pea, whereas the opposite was the case for the winter rye (Figure 3). The mixture of the CCs had similar N-accumulation in 2013, but it was greater after pea than after barley for the CC mixture in 2014 (Fig 3).

Hairy vetch fixed a much greater proportion of its N after barley than after pea (Table 7) and after barley it fixed from 30 to 37 kg N ha$^{-1}$ (Table 7). It can be concluded that hairy vetch fixed significantly more $N_2$ in mixture than the white clover, especially after barley (Table 7). The same was the case for common vetch in mixture in with oil radish in 2012, which fixed 11 and 9 kg N ha$^{-1}$ after pea and barley, respectively as compared to 1 and 3 kg N ha$^{-1}$ after pea and barley, respectively in white clover spring 2013. Common vetch in mixture with oil radish in the autumn 2013 fixed only 5 and 7 kg N $ha^{-1}$ after pea and barley (data not shown) as compared to 2 and 5 kg N $ha^{-1}$ in white clover in spring 2014 (Table 7). Thus the hypothesis III can be accepted for hairy vetch, whereas there is less difference between white clover and common vetch grown in mixtures.

The total accumulation of soil N in the CC crops was between 6 and 22 kg N $ha^{-1}$ greater after pea compared to after barley (Table 7). For the frost-killed CCs this variability was between 1 and 17 kg $ha^{-1}$ (Table 6). In both year the effect of CC was significant (2013: $P<0.000$ and 2014: $P<0.011$) as were the effect of precrop (2013: $P< 0.003$ and 2014: $P<0.001$), whereas no significant interaction were observed between precrop and CC treatment. In 2013 the significant CCs effect was mainly caused by the significantly greater soil N accumulation in the grass-clover CC (Table 7). In 2014 the significant effect of CC was mainly due to a lower soil N accumulation in the hairy vetch sole crop (Table 7). Thus, the hypothesis II is rejected, since the hairy vetch and winter rye CC mixture did not recover more soil N than the grass-clover mixture or the sole crop winter rye CC. Only in the spring 2014 the HV+WR CC mixture recovered more soil N than the sole crop hairy vetch (Table 7).

### *3.3 Subsequent spring wheat crop*

Spring wheat was sown 10. and 29. April in 2013 and 2014 respectively. The later sowing and earlier harvest in 2014 was probably the reason for the lower yields in 2014, 2.6 Mg $ha^{-1}$, compared with 2013 (3.2 Mg $ha^{-1}$) (Figure 4a and 4b). The cover crop treatments significantly affected the spring wheat grain and straw yields (Table 8 and Figure 4).

**Table 8.** Two-way analysis of variance of selected spring wheat variables. P values and $LSD_{0.95}$

| Spring wheat variable and year | Pre-crop effect P value | Cover crop effect P value | Pre-crop × CC P value | $LSD_{0.95}$ |
|---|---|---|---|---|
| **2013** | | | | |
| Grain yield, Mg $ha^{-1}$ | 0.799 | 0.000 | 0.174 | 0.73 |
| Straw yield, Mg $ha^{-1}$ | 0.616 | 0.000 | 0.488 | 1.05 |
| % protein in grain | 0.183 | 0.000 | 0.154 | 0.69 |
| Total N, kg N $ha^{-1}$ | 0.932 | 0.000 | 0.094 | 19.0 |
| Weeds, kg $ha^{-1}$ | 0.000 | 0.000 | 0.897 | 480 |
| **2014** | | | | |
| Grain yield, Mg $ha^{-1}$ | 0.004 | 0.005 | 0.095 | 0.73 |
| Straw yield, Mg $ha^{-1}$ | 0.047 | 0.003 | 0.152 | 0.98 |
| % protein in grain | 0.044 | 0.851 | 0.349 | 1.01 |
| Total N, kg N $ha^{-1}$ | 0.002 | 0.001 | 0.048 | 22.1 |
| Weeds, kg $ha^{-1}$ | 0.013 | 0.482 | 0.570 | 890 |

The greater grain yields were obtained after the two vetch sole crops, oil radish sole crop, and the two mixtures of vetch and non-legume CCs (Figure 4). The grass-clover CC reduced the spring wheat yield compared to no CC in 2013, but had no effect in 2014. The straw yields generally mirrored the grain yields (Figure 4 a, c and b, d). The pre-crop did not significantly affect the dry matter yield in 2013, but both grain and straw yields were significantly affected in 2014. The grain yields were greater in most cases after pea (Figure 4b and 4d).

The grain protein concentration of spring wheat was significantly affected by the CCs in 2013, but not in 2014 (Table 8), because the over-winter hairy vetch and winter rye and the mixture of the two CCs slightly increased the protein concentration, which on average only was c. 11% (Figure 4e). In 2014 the protein concentration was significantly ($P<0.05$) after the pea precrop (Figure 4f). In 2014 the protein concentration did not vary much and was closed to 14%.

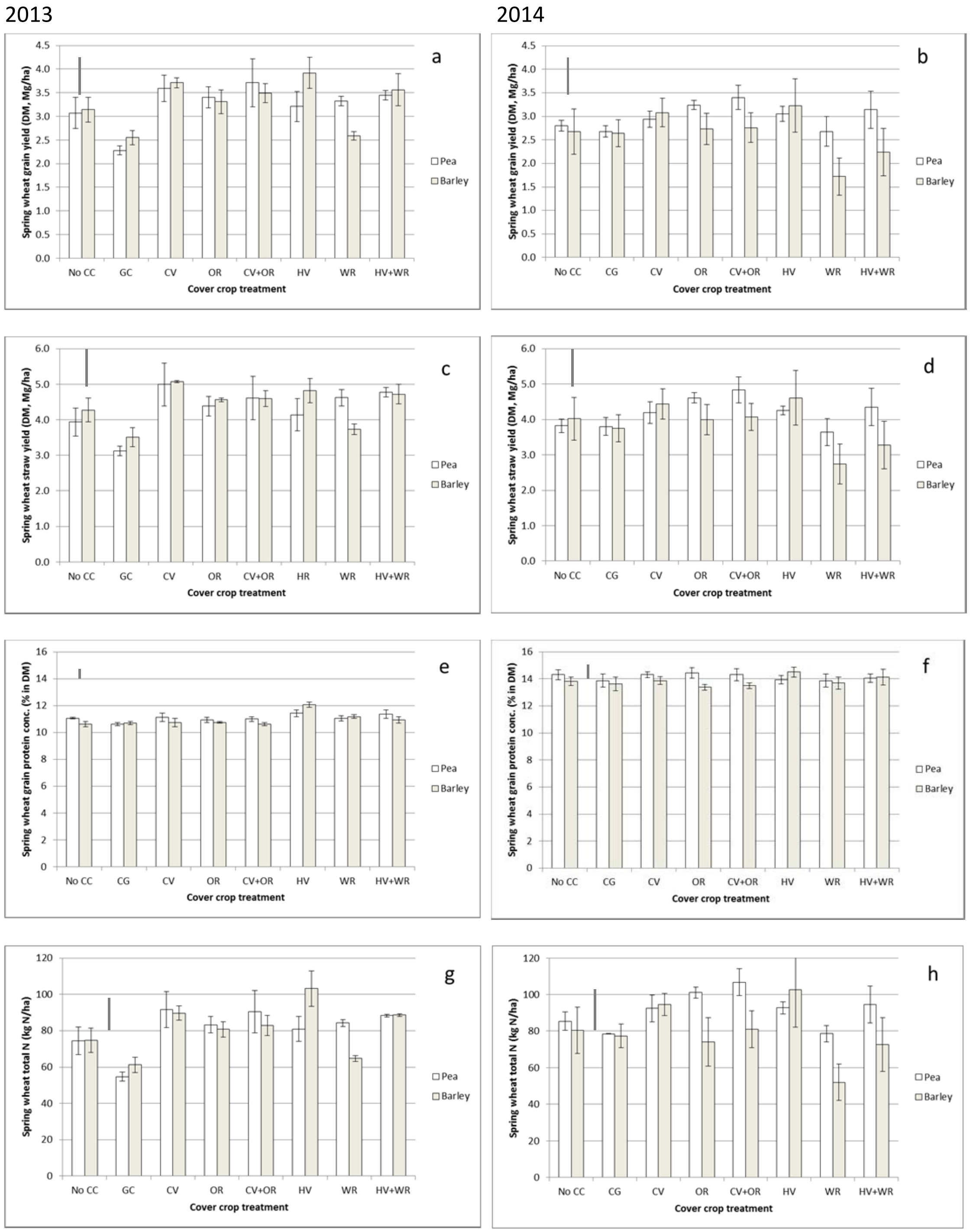



**Fig. 4** Spring wheat dry matter grain and straw yield, grain protein concentration and total aboveground N in 2013 and 2014 following precrops pea and barley and eight cover crop treatments. No CC: No cover crop, GC: Perennial ryegrass-white clover CC undersown main crops pea and barley in spring, CV: Common vetch, OR: Oil radish, CV+OR: Common vetch and oil radish, HV: Hairy vetch; WR: Winter rye, HV+WR: Hairy vetch and winter rye. Single bar represent $LSD_{0.95}$. Values are means± standard error (n=4).

The total N accumulation in spring wheat was highly significantly (P< 0.001) affected by the CC treatment and in 2013 by the precrop (Table 8 and Figure 4 g, 4h). The NO CC treatment, the GC treatment and the winter rye sole crop resulted in the lowest total aboveground N uptake. The total N uptake was on average greater in 2014 than in 2013, which also explain the greater grain protein concentration in 2014 (Figure 4 e and 4f). The total N accumulation in spring wheat was not affected significantly by precrop in 2013, but 2014 the pea precrop led to greater N accumulation in spring wheat.

At harvest of spring the amount of weeds were determined (Fig. 5). In both years regrowth of perennial ryegrass was significant in the GC treatment. The precrop significantly affected the weed biomass in both years, but the precrop effect was opposite in the two years (Figure 5). In 2013 the CC treatment significantly affected the weed biomass, probably due to the heavy regrowth of perennial ryegrass.

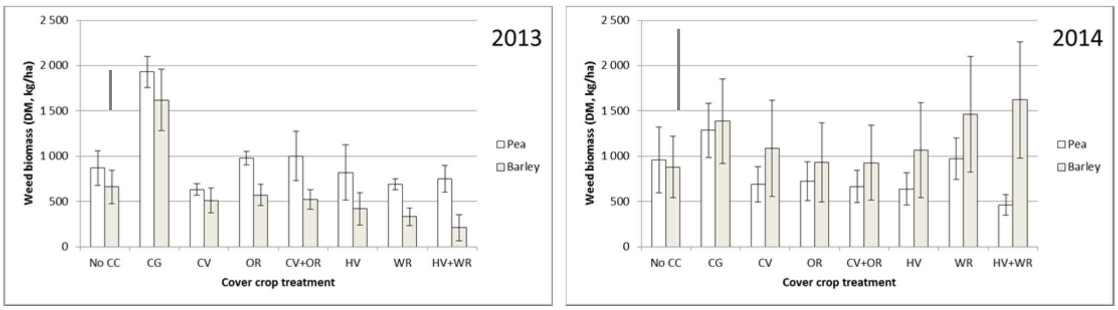



**Fig. 5** Weeds at harvest of spring wheat in 2013 and 2014 following precrops pea and barley and eight cover crop treatments. No CC: No cover crop, GC: Perennial ryegrass-white clover CC undersown main crops pea and barley in spring, CV: Common vetch, OR: Oil radish, CV+OR: Common vetch and oil radish, HV: Hairy vetch; WR: Winter rye, HV+WR: Hairy vetch and winter rye. Single bar represent $LSD_{0.95}$. Values are means ± standard error (n=4).

## 5. Discussion

Cover crops are unique tools for supply multiple ecosystem services in agroecosystems (Schipanski et al., 2014) This study aimed at determining the multifunctionality of sole and multispecies cover crops in rotational sequences with two different main crops (pea and barley), followed by different cover crops (sole and multispecies) during the autumn and winter and the third component of the sequences was a spring wheat crop.

Two agroecosystems services/functions will be studied here: Firstly, the ability of CCs to acquire soil mineral N during autumn and winter and consequently reduce soil mineral N concentration and the risk of $N_2O$ emission and N-leaching: “the catch crop function”. The second function is “the green manuring function” of the cover crop, due to the release of N from the incorporated catch materials, the N being derived from recovered soil mineral N or symbiotic $N_2$ fixation in legume CCs. In relation to both function the carbon-to-nitrogen ration is essential. It has been shown that CCs with too narrow C/N ratios may result in too rapid turnover if frost-killed and the risk of remineralization and subsequent loss of N (Thorup-Kristensen et al., 2003). A too wide C/N ratio( > 20) as often observed with perennial grass-clover result in net immobilization of N and can cause reduction of the subsequent crop N availability (Hauggaard-Nielsen et al, 2012). Additional relevant ecosystem services would be the effect of the cover crops on soil organic matter, on weeds and pests, on soil erosion and the potential value of the cover crop when harvested for use as feed or feedstock for biorefinery. Only the effect on weeds was investigated here.

The overall hypothesis was that a cover crop consisting of an $N_2$ fixing and a non-fixing species will have the plasticity to regulate the growth of the components according to the soil mineral N level and then by this ecological mechanism regulate the balance between the catch and the green manure functions. A principle elaborated by Jensen et al. (2015) and termed “ecological precision farming”. Thus, in a CC mixture after a precrop resulting in a low level of soil mineral N in the autumn the $N_2$ fixing CC species would be in favour, whereas the opposite would be the case after a crop resulting in high amounts of soil mineral N in the autumn (Hauggaard-Nielsen et al., 2012), such as is sometimes the case with grain legumes grown in sole crop (Jensen and Haahr, 1990). In addition the mixture of two species may use the available resources for plant growth more efficiently than the sole crops and produce a higher yield (Vandermeer, 1989). Using a meta-analysis approach and 11 published studies Poffenbarger and Mirsky (2014) showed that hairy vetch-rye CC mixture on average produced 50% more than the hairy vetch sole crop and 30% more than the rye sole crop. The N accumulation was similar in the mixture and the hairy vetch sole crop.

Although the level of soil mineral N was only slightly greater after harvest of pea than after barley shortly after harvest the two crops had differential effects on the subsequent cover crops and the main crop.

### *3.3.1 Biomass production and composition of CCs*

CC biomass yields varied between 1.0 and 4.1 Mg $ha^{-1}$ and were in most cases less than 2 Mg $ha^{-1}$. Under-sown grass-clover mixtures generally produced more biomass than the annual CC species. It was hypothesized (Hypothesis I) that the biomass production of mixtures of vetches and non-legume would be greater than the respective sole crops. However, it was observed that the mixtures had higher biomass yields than the sole crop vetches, but mixture dry matter yields were similar to the oil radish or winter rye sole crop yield. Consequently the hypothesis I had to be rejected.

Hypothesis IV was formulated on the effect of residual soil mineral N from pre-crop on the composition of the CC mixture. A positive relation between soil mineral N content and the proportion of non-legume was hypothesized. It was assumed that the pea precrop would cause a greater soil mineral N content in the autumn as compared to the barley crop. The difference just after harvest was only rather few kg of soil mineral N to the 50 cm depth. It was not possible to find support for the hypothesis in the frost-killed CCs, but the hypothesis was accepted for the grass-clover and the hairy vetch-winter rye mixtures.

### *3.3.2 Symbiotic $N_2$ fixation and the nitrate catch crop function*

Hairy vetch was the most efficient $N_2$ fixing crop fixing almost 40 kg N $ha^{-1}$ in mixture with winter rye. Common vetch fixed much less. The hypothesis III can be accepted since hairy vetch fixes much more N than white clover. However, common vetch was not that efficient and different from clover. The pea precrop and probably the residual soil mineral N had a marked negative effect on symbiotic $N_2$ fixation in hairy vetch.

The catch crop function of CCs was estimated by determining the soil N accumulation in legumes and non-legumes of the CCs, by the use of stable nitrogen isotope methodology (Unkovich et al 2008). In the frost-killed CCs the accumulation of soil N was similar in oil radish and the mixture of oil radish and common vetch, but greater in the mixture than in common vetch. For over-wintering CCs the hairy vetch and winter rye CC mixture did not recover more soil N than the grass-clover mixture or the sole crop winter rye CC. Only in the

2014 the hairy vetch and winter rye CC mixture recovered more soil N than the sole crop hairy vetch. Consequently, the hypothesis II has to be rejected. The symbiotic $N_2$ fixation by the vetch component in the CC mixture did not result in a greater total N content of the mixture than of oil radish. However, in the over-wintering hairy vetch-winter rye mixtures the average total N content after pea and barley were 51 and 59 kg N $ha^{-1}$ in 2013 and 2014, respectively, whereas it were 41 and 33 kg N $ha^{-1}$ after winter rye. Thus, the over-wintering CC mixture of hairy vetch and winter rye provided the same efficiency in its catch crop function and in addition to this 10 to 26 kg N $ha^{-1}$ extra in its green manure function.

### *3.3.3 Balancing the catch crop and green manure function*

The biomass C-to-N ratios were generally lower than 20, which are considered the threshold for net N mineralization (Vigil and Kissel, 1991). With greater ratios it is likely that, at least for some time, there will net immobilization of N. The GC biomass had C/N of 23 to 27. The sole crop vetches had C/N of 11-14 and the oil radish and winter rye 14-19. The mixtures had intermediate C/N of 11-16, which would be more optimal for net N mineralization, but restricting net release and leaching more than the sole crop vetches with C-to-N ratio of 11-14. The hypothesis V is accepted since there also appeared to be a rather significant negative relationship between the CC biomass C/N and the subsequent spring wheat yield.

The greatest spring wheat grain yields and total N accumulation in the short term were obtained for the vetch cover crops and the mixtures of vetches and non-fixing species oil radish and winter rye. Common vetch and hairy vetch sole crop CCs on average increased the spring wheat yield 15% over the non-CC treatment; after barley hairy vetch increased the spring wheat yield with 21-25% in both years. The common vetch-oil radish and the hairy vetch-winter rye increased the grain yield by 14 and 5%, respectively. Oil radish enhanced the yield by 9%.The undersown grass-clover mixture contributed with the greatest amount of carbon, which is important in the long-term for soil organic matter building, but the net immobilization of N in the spring and regrowth of perennial ryegrass in the spring wheat, probably led to the low spring wheat grain yields. The grass-clover and winter rye CCs reduced the spring wheat yield with 12% relative to the non-CC control.

## 6. Conclusions

This pluri-annual field experiment in a rotational sequence from a stockless arable organic cropping system successfully showed the potential of multifunctional cover crop mixtures.

- The mixtures of vetch and non-legume had higher biomass yields than the sole crop vetches, but the mixture dry matter yields were similar to the oil radish or winter rye sole crop yield.
- A positive relation between soil mineral N content after harvest of main crops (greater after pea than after barley) and the proportion of the non-legume component was found for or the grass-clover and the hairy vetch-winter rye mixtures.
- Hairy vetch was the most efficient $N_2$ fixing crop, fixing almost 40 kg N $ha^{-1}$ in mixture with winter rye. Common vetch and white clover fixed much less $N_2$.
- In the frost-killed CCs the accumulation of soil N was similar in oil radish and the mixture of oil radish and common vetch, but greater in the mixture than in common vetch. For over-wintering CCs the hairy vetch and winter rye CC mixture did not recover more soil N than the grass-clover mixture or the sole crop winter rye CC. However, the over-wintering hairy vetch-winter rye mixtures provided the same efficiency in catch crop function and in addition to this 10 to 26 kg N $ha^{-1}$ extra N in its green manure function, due to symbiotic $N_2$ fixation.

- The biomass C-to-N ratios were generally lower than 20. With wider ratios it is likely that, at least for some time, there will net immobilization of N. The grass-clover CC biomass had C/N of 23 to 27. The sole crop vetches had C-to-N ratios of 11-14 and the oil radish and winter rye 14-19. The mixtures had intermediate C-to-N ratios of 11-16, which would be more optimal for net N mineralization, but restricting net release and leaching more than the sole crop vetches with C-to-N ratios of 11-14.
- The highest subsequent spring wheat grain yields and total N accumulation in the short term were obtained after the vetch cover crops; the average (over years and precrops) yield increase was 15% over non-CC control and the mixtures of vetches and non-fixing species oil radish and winter rye, 14% and 5%, respectively.
- The undersown grass-clover mixture contributed with the greatest amount of carbon, which is important in the long-term for soil organic matter building, but the net immobilization of N in the spring and regrowth of perennial ryegrass in the spring wheat, probably led to the low spring wheat grain yields.
- Establishment of a cover crop mixture of hairy vetch and winter rye is an efficient was of maximizing the catch crop and green manure functions of a cover crop in a stockless organic crop rotation.

## 7. Acknowledgements

The funding from the SLU EkoForsk program 2011-2013 is greatly acknowledged. Thanks to the staff of SITES Lönnstorp experimental station, especially research leader Erik Rasmusson for managing the experiments and technician Fatih Mohammad for the skilled technical assistance in sample preparation.

## 8. References


Beck-Friss, B., Lindén, B., Marstorp, H. and Hendriksson, L. 1994. Nitrogen in soil and crops in cropping systems with catch crops. Report 193. Dept. Soil Sciences, SLU. Uppsala, Sweden.

Campiglia, E., Mancinelli, R., Radicetti, E. and Caporali, F. 2010. Effect of cover crops and mulches on weed control and nitrogen fertilization in tomato (*Lycopersicon esculentum Mill.).* Crop Protection 29, 354-363.

Carter, M.R., Andrews, S.S. and Drinkwater, L.E. 2004. System approaches for improving soil quality. In Managing Soil Quality – Challenges in Modern Agriculture (eds. Schjønning, P., Elmholt, S. and Christensen, B. T.), Chapt. 15, pp. 261-281, CAB International, Wallingford, UK.

Eriksen J. and Høgh-Jensen, H. 1998. Variations in the natural abundance of 15N in ryegrass/ white clover shoot material as influenced by cattle grazing. Plant and Soil 205, 67–76.

Hauggaard-Nielsen, H., Holdensen, L., Wulfsohn, D. and E.S. Jensen. 2009. Spatial variation of N-fixation in field pea (*Pisum sativum* L.) at the field scale determined by the 15-N natural abundance method. Plant and Soil 327, 167-184.

Hauggaard-Nielsen, H., Mundus, S. & Jensen, E.S. (2012). Grass-clover undersowing affects nitrogen dynamics in a grain legume–cereal arable cropping system. *Field Crops Research, 136*, 23-31. http://dx.doi.org/10.1016/j.fcr.2012.07.001

IAASTD, 2009: International Assessment of Agricultural Knowledge, Science and Technology for Development. – Executive summary of synthesis report from Intergovernmental Plenary in Johannesburg, 2008

Jensen, E.S 1991a. Nitrogen accumulation and residual effects of nitrogen catch crops. Acta Agric. Scand. 41, 333-344.

Jensen, E.S. 1991b. The release and fate of nitrogen from catch crop material decomposing under field conditions. J. Soil Sci. 43, 335-345.

Jensen, E.S. and Haahr, V. 1990. The effect of pea cultivation on succeeding winter cereal and winter oilseed rape nitrogen nutrition. App. Agric. Res. 5: 102-107

Jensen, E.S., Bedoussac, Carlsson,G., Journet, E.-P, Justes, E. & Hauggaard-Nielsen, H. 2015. Enhancing Yields in Organic Crop Production by Eco-Functional Intensification. Sustainable Agricultural Research (In press).

Knudsen, MT, Hauggaard-Nielsen, H. and Jensen, E.S. 2004. Pea, faba bean and lupin intercropped with spring barley – performance and $N_2$ fixation at two temperate locations. Journal of Agricultural Science 142, 617-627.

Knudsen, M. T. et al. 2006. Estimated leaching losses for organic and conventional farming in Denmark. J. Agric. Sci. 144, 135-149.

Laloy, E and Bielders, C.L. 2010. Effect of intercropping period management on ruoff an erosion in a maize cropping system. J. Environ. Qual. 39, 1001-1008.

Niggli, U., Slabe, A., Schmid, O., Halberg, N., & Schluter, M. 2008. *Vision for an Organic Food and Farming Research Agenda to 2025. Organic Knowledge for the Future,* Bruxelles, TP Organics

Papendick, R.I and Elliot, L.F. 1984. Tillage and cropping systems for erosion control and efficient nutrient utilization. In Organic farming – Current Technology and its role in sustainable agriculture. Chapter 6. ASA Special publication no. 46. pp. 69-81, ASA, Madison, WI.

Poffenbarger, H. and Mirsky, S. (2014) Biomass and nitrogen accumulation of hairy vetch-cereal rye cover crop mixtures: a meta-analysis. Abstract. The Agronomy meeting, Long Beach, December 2014.

Sainju et al. 2002. Long-term effects of tillage, cover crops and nitrogen fertilization on organic C and N concentrations in sandy loam soils in Georgia, USA. Soil Till. Res. 63, 167-79.

Schipanski, M.E., Barbercheck, M., Douglas, M.R., Finney, D.M., Haider, K., Kaye, J.P., Kemanian, A.R., Mortensen, D. a., Ryan, M.R., Tooker, J., White, C., 2014. A framework for evaluating ecosystem services provided by cover crops in agroecosystems. Agric. Syst. 125, 12–22.

Schröder, J.J et al. 1998. Reduction of nitrate leaching the role of cover crops. Final report of EU Concerted action, Wageningen University.

Stevenson, F.C., Knight, J.D., Van kessel, C. 1995. Dinitrogen fixation in pea: controls at the landscape and microscale. Soil Sci. Soc Am. J 59, 1603-1611.

Stokholm, E. 1979. The influence of green manure on yield and soil structure. Tidsskrift for Planteavl 83, 543-549.

Thomsen, I.K. 1993. Nitrogen uptake in barley after spring incorporation of $^{15}N$ –labelled Italian ryegrass into sandy soils. Plant and Soil 150, 193-201.

Thorup-Kristensen et al. 2003. Catch crop and green manures as biological tools in nitrogen management in temperate zones. Adv. Agron. 79, 227-302.

Unkovich, M., Herridge, D., Peoples, M., 2008. Measuring plant-associated nitrogen fixation in agricultural systems . Australian Centre for International Agricultural Research, Canberra.

Vandermeer, J. 1989. The ecology of intercropping. Cambridge University Press, Cambridge.

Vigil, M.F., and D.E. Kissel. 1991. Equations for estimating the amount of nitrogen mineralized from crop residues. *Soil Science Society of America Journal* 55: 757–76.

Wortman, S.E. et al 2012. Optimizing cover crop benfits with diverse mixtures and an alternative termination method. Agronomy Journal, 104, 1425-1435.