

## **Legume Futures Report 1.5**

### **Biological nitrogen fixation (BNF) by legume crops in Europe**

**Compiled by:**

**J.A. Baddeley, S. Jones, C.F.E. Topp and C.A. Watson**

**Scotland's Rural College**

**J. Helming**

**Wageningen University and Research Centre; and**

**F.L. Stoddard**

**University of Helsinki**

February 2014



## **Legume Futures**

Legume-supported cropping systems for Europe (Legume Futures) is an international research project funded from the European Union's Seventh Programme for research, technological development and demonstration under grant agreement number 245216. The Legume Futures research consortium comprises 20 partners in 13 countries.

## **Disclaimer**

The information presented here has been thoroughly researched and is believed to be accurate and correct. However, the authors cannot be held legally responsible for any errors. There are no warranties, expressed or implied, made with respect to the information provided. The authors will not be liable for any direct, indirect, special, incidental or consequential damages arising out of the use or inability to use the content of this publication.

## **Copyright**

© The Authors 2014. Reproduction and dissemination of material presented here for research, educational or other non-commercial purposes are authorised without any prior written permission from the copyright holders provided the source is fully acknowledged. Reproduction of material for sale or other commercial purposes is prohibited.

## **Citation**

Please cite this report as follows:

Baddeley, J.A., Jones, S., Topp, C.F.E., Watson, C.A., Helming, J. & Stoddard, F.L. 2013. Biological nitrogen fixation (BNF) by legume crops in Europe. Legume Futures Report 1.5. Available from [www.legumehub.eu](http://www.legumehub.eu)

## CONTENTS

<b>ABBREVIATIONS</b> .....	<b>4</b>
<b>INTRODUCTION</b> .....	<b>5</b>
<b>METHODS</b> .....	<b>7</b>
FORAGE LEGUMES .....	7
GRAIN LEGUMES .....	7
<b>RESULTS</b> .....	<b>9</b>
<b>DISCUSSION</b> .....	<b>14</b>
APPROACH .....	14
FORAGE LEGUMES .....	14
GRAIN LEGUMES .....	15
N BALANCE .....	15
BNF IN EUROPE .....	16
PROBLEMS AND LIMITATIONS .....	16
<b>CONCLUSIONS</b> .....	<b>17</b>
<b>SUPPLEMENTARY TABLES</b> .....	<b>21</b>

## ABBREVIATIONS

BNF	Biological nitrogen fixation
$k_{\text{fix}}$	country-specific biological fixation coefficient for forage legumes
Ndfa	Proportion of nitrogen derived from the atmosphere

## INTRODUCTION

Biological nitrogen fixation (BNF) is the distinguishing feature of a legume in a cropping system. Most legume species are able to form a symbiosis with alpha- or beta-proteobacteria, collectively called rhizobia, that use solar energy captured by the plant to break the bond in inert atmospheric dinitrogen and form reactive N species, initially ammonium ( $\text{NH}_4^+$ ). As a result of this symbiosis, the legume crop requires little or no input of N fertilizer and makes little demand on soil N reserves. Since the manufacture of synthetic fertilizer consumes fossil fuel, thereby releasing  $\text{CO}_2$ , and the transport and spreading of organic and synthetic N fertilizers consumes further fuel, the use of legumes in cropping systems has immediate environmental benefits arising from reduced fossil fuel use. Nitrates from fertilizers and soil N reserves may also leach through the soil column into groundwater, and the denitrification of nitrates from synthetic or organic sources is the primary source of nitrous oxide ( $\text{N}_2\text{O}$ ), a powerful greenhouse gas, from agricultural soils (Philippot and Hallin 2011). Hence maintaining the reactive N within the plant, as happens in a symbiotic legume in the growing season, avoids some potential for environmental damage.

Crop residues of legumes contain some of the N that they have fixed, and this becomes available to subsequent crops. These residues are considered just as likely to contribute to leaching or  $\text{N}_2\text{O}$  release as any other crop residue (see the Legume Futures report on  $\text{N}_2\text{O}$ ), but a portion of the fixed N remains in the soil/plant system reducing N fertilizer needs in subsequent crops.

Depending on the genetic constitution of the rhizobium, hydrogen may be released during N fixation (Golding & Dong 2010). This gas stimulates the growth of hydrogen-fixing bacteria in the rhizosphere, and these compete successfully for living space with other rhizosphere organisms, including many pathogens. Some of the hydrogen bacteria have plant growth-promoting properties (Maimaiti et al. 2007). Thus the use of BNF leads to positive changes in the structure of the soil microbiological community.

Large, continental- or global-scale estimates of BNF have been attempted. Yang et al. (2010) used a range of national databases to compile an estimate of BNF in Canada. An Australian estimate also used national databases, and noted that below-ground residues were not included (Unkovich et al. 2010). A global estimate factored in below-ground residues and highlighted some of the other uncertainties in the process (Herridge et al. 2008). Chief amongst these uncertainties is the amount of N fixed by crops. A recent attempt to produce a high-resolution assessment of global N flows in cropland cited one of the main limitations of their method as the lack of crop-specific N fixation data (Liu et al., 2010a). This was highlighted by their use of N fixation figures from Smil (1999), who had commented on their own use of unreliable data on average annual BNF rates of important species and cultivars.

For these reasons, we set out to develop an improved estimate for BNF across Europe. BNF is usually quoted on a  $\text{kg ha}^{-1}$  basis from experiments conducted in a limited area and time, and datasets from a wide range of crops and growing conditions show a wide range in amounts fixed, depending especially on biomass and available soil N. Previous attempts to estimate N fixation have tended to follow a common approach in that they calculate the amount of N fixed as the as the product of the land area cultivated and fixation per area. The figures for the former are obtained from published databases such as FAOstat and, subject to the level of detail made available about data collection methods, are taken at face value. In contrast, data on N fixation range hugely for each legume species depending on growing conditions, in particular biomass production and available soil nitrogen. Yields range more than 10-fold across species, countries and years. Thus we took the approach that it is not desirable simply to use legume areas for each country and convert these to overall fixed N using an average figure for BNF. Hence we attempted to take a stepwise approach using the most detailed available data and robust scientific logic.

Most legume crops maintain their N concentration within a relatively narrow physiological range. This is true across a 10-fold range of overall biomass yields, for annual grain legumes (Evans et al., 1989; Pilbeam et al., 1997; Unkovich et al., 2010) and for perennial forage legumes (Carlsson and Huss-Danell 2003). In most circumstances, a legume with its rhizobia can fix its own N, so growth of the crop is unlikely to be limited by N supply. Rather, crop N yield is likely to reflect overall crop yield as limited by other factors, including rainfall and temperature.

Grain yields of the grain legumes can be used to estimate biomass by using data on harvest index and root:shoot ratio, and sufficient data are available on N concentrations in seeds, straw and roots of the main species to allow N yields to be calculated. Further factors to be considered are rhizodeposition, the deposition of N in the root zone from dead cells, root exudates, and shed fragments of roots (Fustec et al. 2010) and the amount of N derived from fixation.

In contrast, data on yields of fodder/forage legumes and legume-grass systems (hereafter “forage legumes”), and their legume content, are not available in any systematic way. Furthermore, the moisture content of the yields that are published on FAOstat is not clear, while that in Eurostat is stated as 65%. The CAPRI model allows some estimation of biological N fixation based on Eurostat data on the areas of forages per country (Britz and Witzke, 2012). Yields of grazed pastures are not estimated in any database.

Thus we have adopted different methodologies for estimating BNF by grain and forage legumes.

## METHODS

### Forage legumes

BNF of forage legumes was estimated for each country in the EU27 as the product of crop area, N retention, and biological fixation coefficient. Crop areas were obtained from Eurostat. N retention (N in plant biomass) was obtained from the Common Agricultural Policy Regionalised Impact (CAPRI) model, an economic model for agriculture focussed on Europe, which uses input data mainly compiled from official Eurostat data (Britz and Witzke, 2008). The latest complete dataset within CAPRI is from 2009, so datasets from that year were used. The biological fixation coefficients ( $k_{\text{fix}}$ ) are determined by the N fixed per hectare and the proportion of the grassland that is assumed to include a legume component.

The current version of CAPRI accounts for the share of forage legumes in all grasslands, but after an initial round of calculations, the predicted N fixation values were considered to be too high. Consequently, the proportions of forage legumes in these systems were re-estimated in collaboration with colleagues in the Legume Futures network and checked, as far as possible, with Eurostat data. This process, which follows the approach recommended by Eurostat to improve estimates of BNF in grasslands (Eurostat, 2013) led to the derivation of revised figures for  $k_{\text{fix}}$  that are used in this study.

### Grain legumes

Data on areas and yields of grain legumes were taken from FAOstat (FAOstat 2014), as this differentiates more individual crops than Eurostat. Only classes that represent agricultural production of grain legumes harvested for dry grains were included (defined in Table 1), because production of horticultural legumes such as vining pea is generally high-input and seldom relies on BNF. Cowpea was excluded as annual production in EU27 is less than 200 t and there is very little available data on N partitioning. There are some inconsistencies between FAOstat and Eurostat in the major grain legume crops, especially a misallocation of UK faba bean to "pulses" instead of "broad beans, dry", so the areas of pea, faba bean, lupins and soya were taken from Eurostat, while those of lentil, chickpea, common bean and vetches were taken from FAOstat.

The production data generally represent mass at commercial moisture content, which we taken as 14%. To convert these production figures to above-ground biomass production, the harvest index of each crop was used. This value is fairly consistent within each species, although extreme values can be found in exceptional growing conditions. Above-ground biomass was then converted into above-ground N yield by first converting the widely available data on average grain protein concentration to grain N concentration

using the standard conversion factor of 5.60 (Sosulski and Holt, 1980; Mariotti et al., 2008). This, divided by the N harvest index, gives the above-ground N yield.

Accurate determination of root biomass production is notoriously difficult as it is hard to quantitatively separate roots from field soils, and root tissues tend to die as an annual crop reaches harvest maturity. Although some previous studies have assumed root biomass to be a constant proportion of above-ground biomass, here we adopted a species-specific approach using root:shoot ratios that are obtainable for many species based on mid-season growth when living root mass is at its maximum. From above-ground biomass and root:shoot ratio, root biomass was calculated. Root N yield was then calculated using root biomass and values of root N concentration from the literature, where available. While root N yield is lower than above-ground N yield, our calculations show that it is important.

Table 1. Definitions of grain legume categories used by FAO and included in this study. Cowpea and horticulturally produced vegetable legumes were excluded from the analysis.

Name	FAO Category	Genus and species covered
Beans	Beans, dry	<i>Phaseolus</i> spp. and most <i>Vigna</i> spp.
Faba bean	Broad beans, horse beans, dry	<i>Vicia faba</i>
Chickpea	Chick pea	<i>Cicer arietinum</i>
Lentil	Lentil	<i>Lens culinaris</i>
Lupins	Lupins	<i>Lupinus</i> spp. ( <i>L. albus</i> , <i>L. angustifolius</i> , <i>L. luteus</i> )
Pea	Peas, dry	<i>Pisum sativum</i>
Soya bean	Soybeans	<i>Glycine max</i>
Vetches	Vetches	<i>Vicia</i> species other than <i>V. faba</i> (mostly <i>V. sativa</i> )

Rhizodeposition is a relatively recent addition to the legume N budget, and covers biomass left in the soil from root exudates, leachates, root-cap cells, and detached or dead root fragments. Although it is hard to determine experimentally there are a few estimates in the literature, usually given as proportional rhizodeposition, the proportion of plant N in rhizodeposits relative to total or above-ground N. Here we use species-specific values where available, otherwise the default of 0.15 (Mahieu et al., 2007; Wichern et al., 2008).

From this sequence of calculations, the total N production of a grain legume crop was calculated, relative to its grain production. Converting this to an estimate of N fixation required one more factor, namely the proportion of N derived from the atmosphere (Nd<sub>fa</sub>). This figure may be as low as zero, when there is plentiful mineral N in the soil, the appropriate symbiont is not available, or the legume-rhizobium symbiosis is ineffective. It may also exceed 90% in the opposite circumstances. Thus Nd<sub>fa</sub> is the most sensitive



factor in estimating overall European BNF. Here we used values extracted from the literature for the most typical agronomic conditions available, i.e., no or low levels of N fertilizer addition. This gave a mean value of 63%, which is comparable with that used in other studies on this scale (*e.g.* 57%, Herridge et al., 2008) and is much lower than the 75% used in CAPRI.

These figures, on a per-tonne-harvested basis, were then converted to totals for the EU27 countries, based on the grain production statistics for 2009, to be comparable with the forage legume data.

## RESULTS

Grain legumes in Europe were calculated to fix about 13 and 20 times more N per hectare than temporary or permanent pastures respectively (Table 2). Although permanent pastures had the lowest fixation rate per hectare, they were responsible for nearly half of the total 811 Gg of N fixed across Europe (Figure 1). Total N fixation by grain legumes was comparable with that of the temporary grasslands, although the area of grain legumes grown is very much smaller (78 and 1.6 M ha respectively).

Across countries, the yields of N fixed in both temporary and permanent grassland systems were smallest in Cyprus and highest in Denmark (Figure 1, with supporting data in Supplementary Table 1). However, the total amounts of N fixed tended, not surprisingly, to be determined by cultivated land areas. In temporary systems, Malta had the least N fixation (9 t) and Italy the greatest (27.8 Gg). In permanent systems, the least N fixation was in Cyprus (20 t) and the greatest in France (62.3 Gg).

Total N yield of the different grain legume crops ranged from 63.6 kg t<sup>-1</sup> for pea to 106.9 kg t<sup>-1</sup> for soya bean (Table 3; full calculations shown in Supplementary Table 2). The amounts of N fixed by each crop ranged from 28.4 kg t<sup>-1</sup> for beans to 68.9 kg t<sup>-1</sup> for lupins. Finally, the N balance for each crop ranged from -9.4 kg t<sup>-1</sup> for beans to 23.9 kg t<sup>-1</sup> for faba bean.

Table 2. Area-weighted mean N fixation (with standard deviation) and total N fixed in temporary and permanent pastures and by grain legumes in the EU27 in 2009.

	Temporary grassland	Permanent grassland	Grain legumes	Total
Mean N fixation (kg ha <sup>-1</sup> )	10.1 (0.5)	6.8 (0.3)	133 (10)	
Total N fixed (Gg)	172	414	225	811

Table 3. Calculated quantities of total N production and N fixed, and overall N balance (N fixed – N offtake) by grain legume crops EU27 in 2009. All figures are kg of N per tonne of grain production. Full calculations are shown in Supplementary Table 2.

	Common bean	Faba bean	Chick-pea	Lentil	Lupins	Pea	Soya bean	Vetches
Total N production	58.7	81.1	83.2	82.5	76.1	57.5	96.5	87.7
N fixed	26.0	62.4	41.6	57.7	62.4	40.2	50.2	63.2
N balance	-7.9	22.2	11.6	18.1	13.3	5.8	-4.6	23.2

The calculated total amounts of N fixed by each grain legume crop across EU27 in 2009 are presented in Figure 2. In common with the results for forage legumes (Table 2), the figures are largely a reflection of the areas of each crop grown, with the most N fixed by pea and the least by chickpea. The total fixation of 225 Gg was dominated by 3 crops, pea, faba bean and soya bean, which between them were responsible for about three quarters of all N fixation. A large proportion of the total N was fixed by a fourth category of crop, “pulses”, but this is an amalgamation of many minor grain legume species together with varied reporting of some of the more common species.

The potential extent by which the N in cropping systems is being replenished or depleted after the growth of grain legume crops is shown by the N balance data (Figure 3). Common bean and soya bean have negative N balances of -1.1 and -4.6 Gg respectively, indicating a net loss of N from these systems. All other grain legumes have positive N balances with the greatest being for 18.4 Gg for faba bean.

The total amounts of N fixed by grain and forage legume systems per country show a range of three orders of magnitude from 120 t in Malta and Cyprus to 151 Gg in France (Figure 4). Nearly half of all N fixed in the EU27 is in three countries, France, Italy and UK. Figure 4 also shows the large variation in the relative amounts N fixed by forage and grain legumes in each country. In Ireland and Lithuania almost all fixation is by forage legumes whereas in Austria, France and Latvia about 40% of fixation is by grain legumes.

Legume-supported cropping systems for Europe

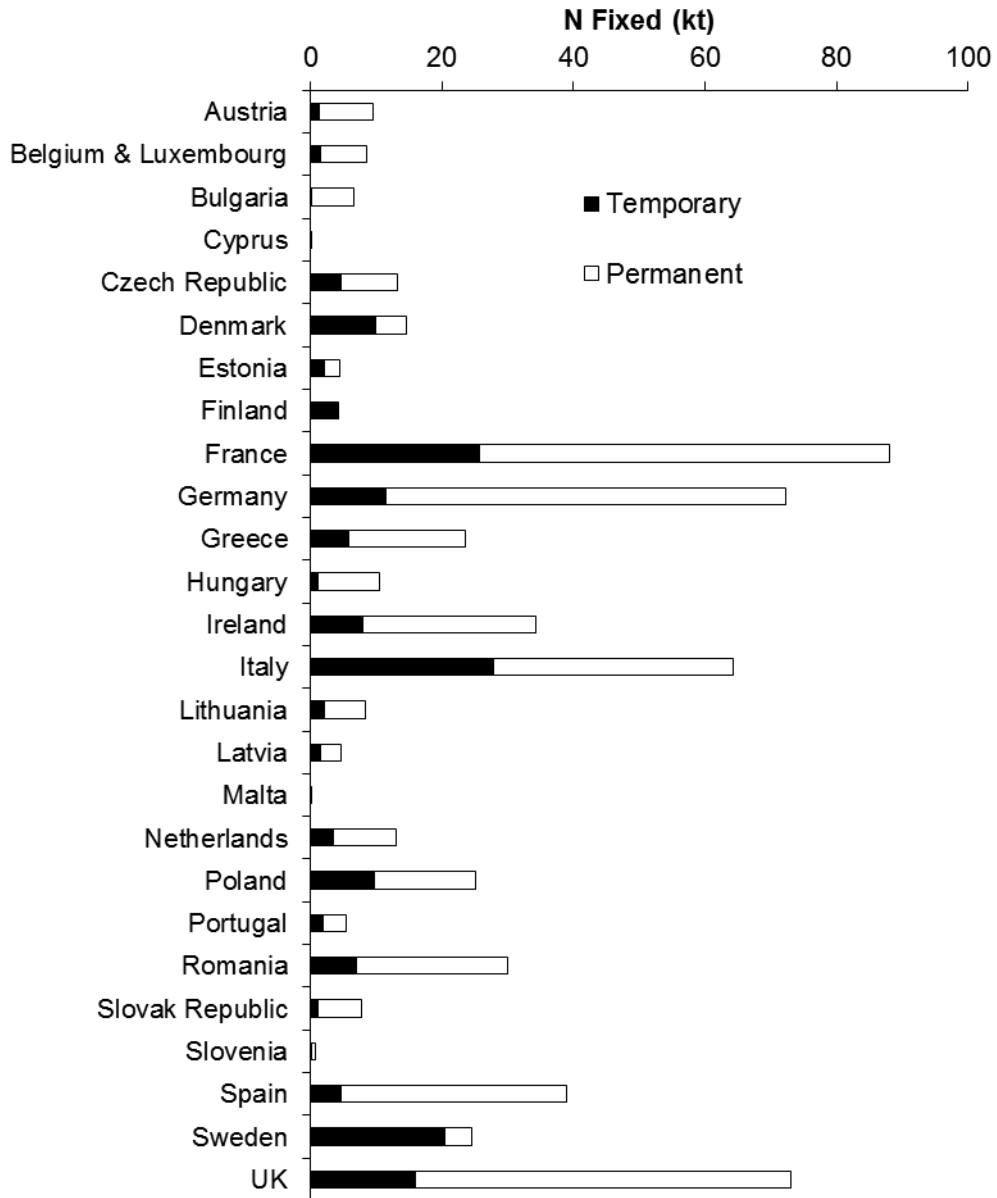


Figure 1. Calculated quantities of N fixed by forage legumes in temporary and permanent pastures in the EU27 countries in 2009. Details are given in Supplementary Table 1.

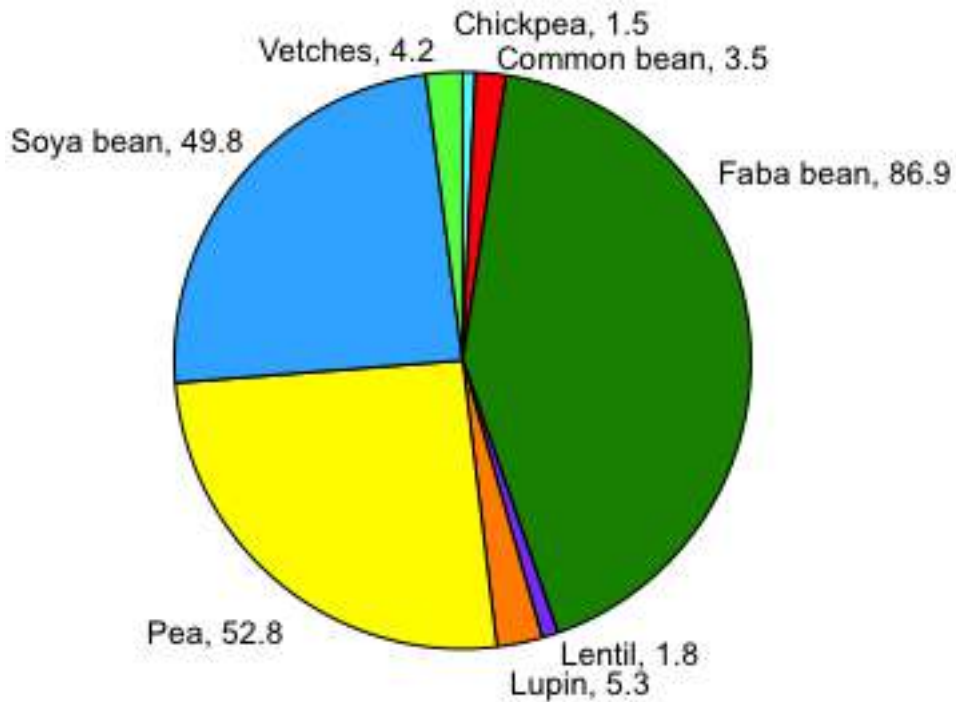


Figure 2. Calculated quantities of total N fixed (Gg) by grain legume crops across the EU27 countries in 2009. Details are given in Supplementary Table 4.

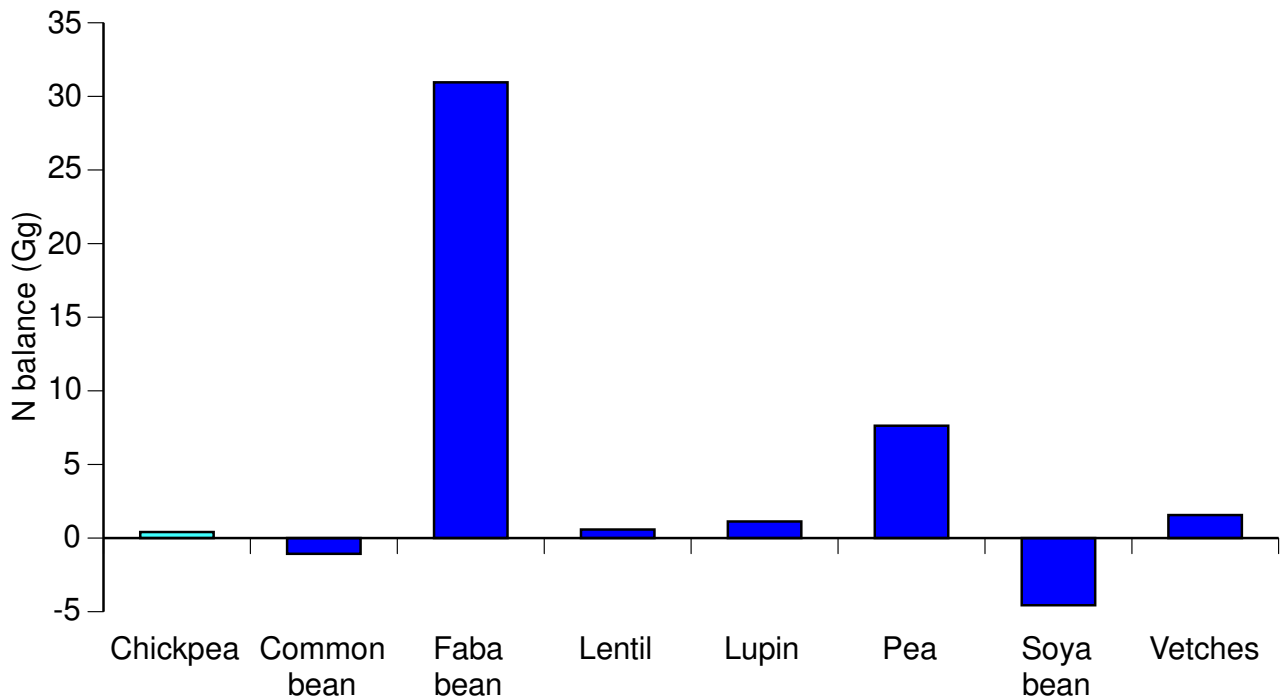


Figure 3. Calculated N balance (N fixed – N grain offtake) for grain legume crops across the EU27 countries in 2009. Overall N balance was 36.6 Gg

## Legume-supported cropping systems for Europe

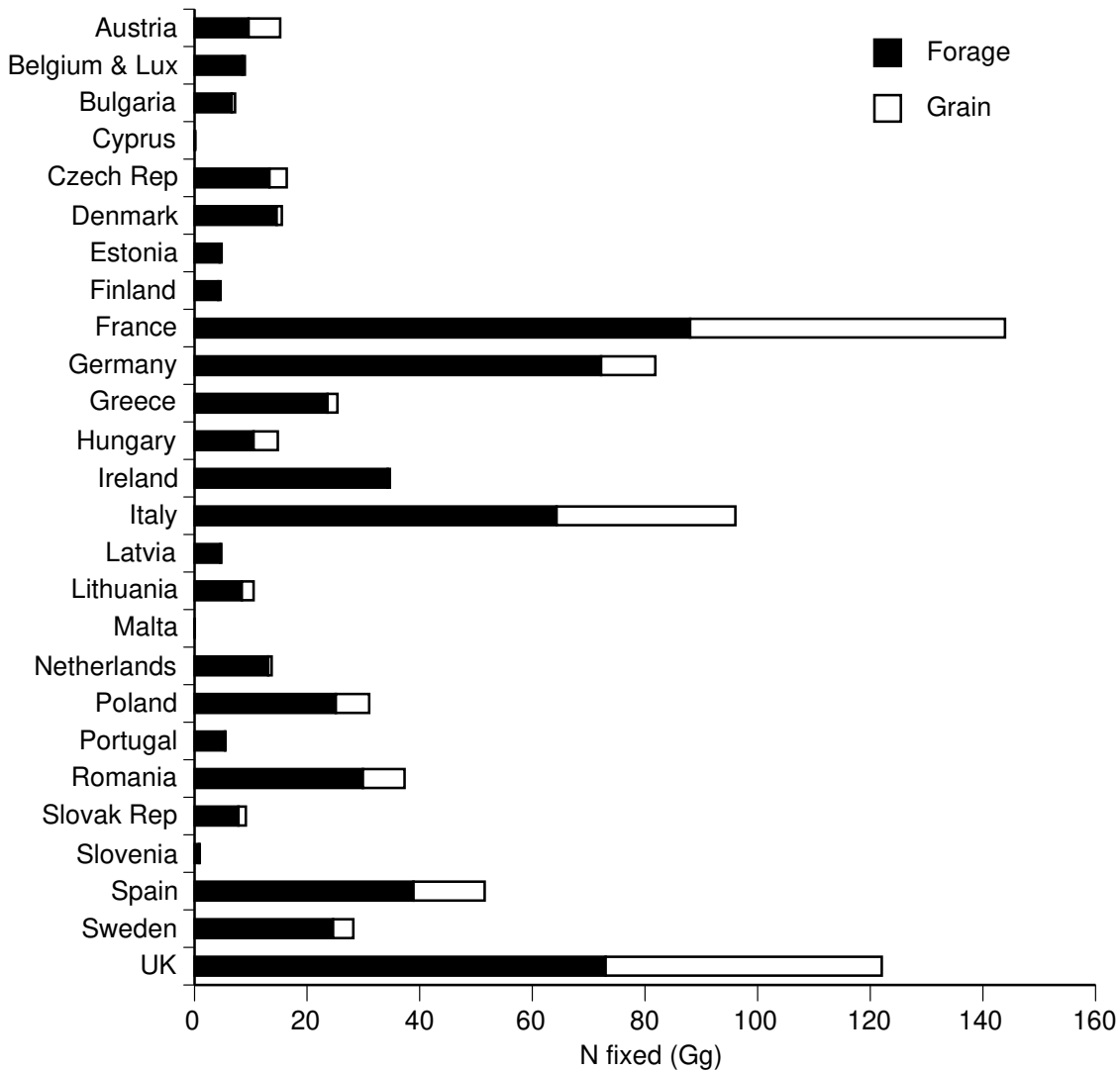


Figure 4. Calculated quantities of N fixation by forage and grain legumes in EU27 countries in 2009. Detailed figures are provided in Supplementary Table 3.

## DISCUSSION

### Approach

Any study that attempts to present an integrated estimate of agricultural N fixation will be faced by the substantial difficulty of how to deal with the contribution from legumes in the various types of forage systems. We took the basis for this element of the analysis from outputs by the CAPRI model as it is an integral part of agricultural planning in the EU. Until now, it has been difficult to derive reliable N fixation estimates from CAPRI as it has required the use of databases that are not publicly accessible. To overcome this issue and to make our estimates more accurate we, in conjunction with partners in the Legume Futures network, derived new, country-specific fixation coefficients for temporary and permanent pastures. This is an important modification in that it includes permanent pastures, hitherto not included in the CAPRI model. This decision was vindicated by the finding that these systems made the greatest contribution to total agricultural N fixation across the EU, even though the per-hectare fixation was low. The decision to use a species-specific model for the grain legumes was supported by the wide range of Ndfa values found, in contrast to the fixed value of 75% in the CAPRI model. We can therefore recommend that species-specific values be used in further developments of the CAPRI model, at least for the major grain legumes that are separately itemized in the Eurostat database, namely pea, faba bean, lupins and soya bean.

### Forage legumes

Our fixation coefficients give slightly higher results than the figure of 5 kg ha<sup>-1</sup> used in three models that deal with N budgets in European agriculture (MNP, 2006; MITERRA, Velthof et al 2009; INTEGRATOR, De Vries et al, 2011a). The difference is likely to be because the figures we used accounted for the areas devoted to the growth of near-monocultures of clovers and lucerne, which can fix considerable amounts of N. There is no indication of how these are accounted for in other models. An attempt to estimate N fixation in Canadian agriculture assumed N fixation of 4 kg ha<sup>-1</sup> in improved and unimproved pastures, but dealt with lucerne and hay (a mixture of clovers, lucerne and grasses) systems separately (Yang et al., 2010). This approach was not practicable in the present study because of the different ways that data are gathered in Canada and the EU. Data on yields and areas of alfalfa, clovers and "other forage legumes" are available, at a stated moisture content of 65% in Eurostat and an unknown moisture content in FAOstat. Data on yields and areas of mixed forages such as grass-clover pastures, and their legume content, are not available in any systematic way. Thus significant improvements in the ability to predict N fixation in the permanent and temporary pasture and fodder systems that are responsible for the majority of N fixed by European agriculture will only be possible if more detailed and reliable production statistics become available.

## Grain legumes

Grain legume crops are grown almost exclusively as monocultures, so the issue of estimating proportions, as alluded to above for forage legumes, is not a consideration. This allowed the construction of a simple model of N partitioning in the main agricultural grain legume species grown in Europe. This refines the concept of Peoples et al. (2009) in that it relates fixation to readily available data on grain production, and it incorporates the most recently available data on below-ground N partitioning and rhizodeposition.

The figures for N fixation by this novel approach compare closely with those used in other studies of similar scope. Four European N budget models use amounts of N fixed between 0.75 to 2.0 times the amount of harvested N for all grain legumes (2.0 in IMAGE, MNP, 2006; 0.75 in IDEAg, Leip et al., 2008; 1.0 in MITERRA, Velthof et al., 2009; 1.2-1.3 in INTEGRATOR, De Vries et al., 2011a). The values calculated in this study ranged from 0.75 for beans to 1.53 for vetches and faba bean. A wide-ranging review concluded that globally crop legumes fix an average of 20 kg of N per tonne of shoot biomass (Peoples et al., 2009), which is in line with our values of 15 – 39 kg t<sup>-1</sup>.

## N balance

Detailed prediction of the N partitioning in each grain legume crop also allowed the prediction of N balance, the difference between N fixed and N removed in grain. This is not the same as the N remaining after cropping, as it does not take into account mineral N in the soil before cropping (N not derived from the atmosphere), and the processing of crop residues varies widely in different systems. Nevertheless, positive values give a useful indication of the potential input of atmospheric N to a system, whereas negative values indicate an overall mining of soil N. As with the other values from the N partitioning model, those for N balance compare well with experimental studies. As examples: for faba bean the N balance related to shoot biomass was reported as 19 kg t<sup>-1</sup> (Peoples et al., 2009) and 9 kg t<sup>-1</sup> (Vinther and Dahlmann-Hansen, 2005), compared to our value of 14 kg t<sup>-1</sup>. For lentils the N balance related to grain production was 18 kg t<sup>-1</sup> (Kurdali et al., 1997) and 25 kg t<sup>-1</sup> (Schmidtke et al., 2004), compared to our value of 19 kg t<sup>-1</sup>. A meta-analysis of 637 soya bean datasets found a partial N balance (based on aboveground N only) of -17 kg t<sup>-1</sup> of grain, compared with our value of -18 kg t<sup>-1</sup>.

While the growth of the majority of grain legumes results in potential N gains, negative values were calculated for beans and soya bean, driven by low N fixation in the former and high grain N content in the latter. The extent to which a negative balance is a problem in practice is, however, questionable. Many experimental studies have found that growth of both beans and soya bean can be beneficial to a following crop (e.g. Hesterman et al., 1987; Bergersen et al., 1992; Green and Blackmer, 1995). Given that the growth of beans and soya bean does not result in a net N increase in the system, it is likely that these

reported benefits are a result of other processes. These include promotion of the mineralisation of soil organic matter, the so-called priming effect (Jenkinson et al, 1985; Kuzyakov et al, 2000), and nitrate sparing, where the legume takes up less of the soil nitrate than would a non-legume crop (Herridge et al 1995), as well as other legume-specific and non-specific break crop effects (Legume Futures report 1.6). Thus while in well managed, diverse rotations, the calculated negative N balance is probably sustainable, monoculture of beans or soya bean is likely to be as unsustainable as any other monoculture.

## **BNF in Europe**

Overall, the calculated figure of 0.81 Mt of N fixed in EU27 by agricultural legumes in 2009 is broadly comparable with the mean estimate of 1.12 Mt from four European N budget models (De Vries et al., 2011b) and the value of 1.1 Mt submitted to the United Nations Framework Convention on Climate Change (EEA, 2008). Most of the difference occurs because the N budget models allow for  $\sim 5 \text{ kg ha}^{-1}$  of N fixation by free-living microbes in all non-legume arable land, in contrast to our focus on legumes.

## **Problems and limitations**

The greatest challenge to this exercise was to estimate BNF in forage systems, where the yield is uncertain (especially in grazed systems), the legume content is unknown and management is variable. Hence, we refined the use of area-based figures for N fixation as far as possible by the derivation of new, country-specific constants for N fixation based on expert opinion. Significant improvements to this would need more detailed data on land areas under cultivation, their management, the proportion of legumes present and their temporal variation. The collection of such data would require wide-ranging changes to the way current agricultural statistics are collected, and this need has been recognised on the Eurostat website: “The comparability and transparency of the estimation of BNF in forage/fodder legumes and legume-grass mixtures could be improved if a set of common guidelines on the estimation method and update frequency were established” (Eurostat, 2013).

In terms of the calculations of N fixed by grain legumes, the most sensitive figure is Ndfa. The approach adopted here was to select literature values from a wide range of sources constrained, where possible, to agronomic conditions that were appropriate for Europe. Nevertheless, published values of Ndfa vary widely and depend on a complex set of interacting factors such as existing and applied mineral N, presence of suitable rhizobia, and efficiency of the plant-rhizobium symbiosis (e.g., Hardarson and Atkins, 2003; Herridge et al, 2008). A further refinement to the present method would be to include variables that are known to modify N fixation, such as soil N status, soil moisture, soil pH



and temperature, that are available from national institutions in Europe, and to use constants that relate them to N fixation as they become available (Liu et al., 2010b).

## CONCLUSIONS

The amount of N fixed by forage legumes and legume-grass systems was predicted by a combination of outputs from the CAPRI model and improved, country-specific N fixation coefficients. For grain legumes, the higher quality of available data made it possible to construct a detailed model based on N partitioning. Both approaches predicted quantities of N fixed that were broadly comparable with previously published estimates. Combining these figures with published crop production statistics allowed the production of detailed crop- and country-specific figures for N fixation by grain legumes that, for the first time, took into account the large differences in yields across Europe. The results also showed that while the amount of atmospheric N fixed into farming systems is likely to increase with increasing cultivation of many species of grain legumes, this is unlikely for beans and soya bean which apparently mine soil N reserves. Only minor adjustment to the Ndfa of soya bean, through management or breeding, is required to make it a net contributor to the N balance.

The results confirm that reliable estimates of agricultural N fixation in Europe require accurate crop production and yield statistics. Estimating N fixation would be greatly facilitated by changes to some of the information that is currently collected. This would result in more accurate figures for N fixed and N balance, which are crucial for reliable calculations of N losses such as nitrate leaching and emissions of the greenhouse gas N<sub>2</sub>O. These are vital for refined predictions of the effects of strategic changes in the use of legumes in farming systems. Ultimately this will lead to the development of better policies to reduce the environmental impacts of European agriculture.

## REFERENCES

- Bergersen, F.J., Turner, G.L., Gault, R.R., Peoples, M.B., Morthorpe, L.J. & Brockwell, J. 1992. Contributions of nitrogen in soybean crop residues to subsequent crops and to soils. *Australian Journal of Agricultural Research* 43: 155-169.
- Britz, W. & Witzke, H.P. 2012. CAPRI Model Documentation 2012. [www.capri-model.org/docs/capri\\_documentation.pdf](http://www.capri-model.org/docs/capri_documentation.pdf) (accessed Feb 2014).
- Carlsson, G. & Huss-Danell, K. 2003. Nitrogen fixation in perennial forage legumes in the field. *Plant and Soil* 253: 353-372.
- De Vries, W., Kros, J., Reinhardt, D.R., Wieggers, R., Velthof, G., Oudendag, D., Oenema, O., Nabuurs, G.J., Schelhaas, M.J., Perez Soba, M., Rienks, W., de Winter, W., van den Akker, J., Bakker, M., Verburg, P., Eickhout, B. & Bouman, L. 2011a. INTEGRATOR: A modelling tool for European-wide assessments of nitrogen and greenhouse gas fluxes in response to changes in land cover, land management and climate. Calculation procedures, application methodology and examples of scenario results. Alterra, Wageningen.
- De Vries, W., Leip, A., Reinds, G.J., Kros, J., Lesschen, J.P. & Bouwman, A.F. 2011b. Comparison of land nitrogen budgets for European agriculture by various modeling approaches. *Environmental Pollution* 159: 3254-3268.
- EEA 2008. Annual European Community Greenhouse Gas Inventory 1990-2006 and Inventory Report 2008. UNFCCC Secretariat, European Environment Agency.
- Eurostat. 2014. Agriculture – agricultural production – crops products. <[epp.eurostat.ec.europa.eu/portal/page/portal/agriculture/data/database](http://epp.eurostat.ec.europa.eu/portal/page/portal/agriculture/data/database)>.
- Evans, J., O'Connor, G.E., Turner, G.L., Coventry, D.R., Fettell, N., Mahoney, J., Armstrong, E.L. & Walsgott, D.N. 1989. N<sub>2</sub> fixation and its value to soil N increase in lupin, field pea and other legumes in south-eastern Australia. *Australian Journal of Agricultural Research* 40: 791-805.
- FAOstat. 2014. Crop production statistics. <[faostat.fao.org](http://faostat.fao.org)>.
- Fustec, J., Lesuffleur, F., Mahieu, S. & Cliquet, J.-B. 2010. Nitrogen rhizodeposition of legumes. A review. *Agronomy for Sustainable Development* 30: 57-66.
- Golding, A.-L. & Dong, A. 2010. Hydrogen production by nitrogenase as a potential crop rotation benefit. *Environmental Chemistry Letters* 8: 101-121.
- Green, C.J. & Blackmer, A.M. 1995. Residue decomposition effects on nitrogen availability to corn following corn or soybean. *Soil Science Society of America Journal* 59: 1065-1070.
- Hardarson, G. & Atkins, C. 2003. Optimising biological N<sub>2</sub> fixation by legumes in farming systems. *Plant and Soil* 252: 41-54.
- Herridge, D.F., Marcellos, H., Felton, W.L., Turner, G.L. & Peoples, M.B. 1995. Chickpea increases soil-N fertility in cereal systems through nitrate sparing and N<sub>2</sub> fixation. *Soil Biology & Biochemistry* 27: 545-551.
- Herridge, D.F., Peoples, M.B. & Boddey, R.M. 2008. Global inputs of biological nitrogen fixation in agricultural systems. *Plant and Soil* 311: 1-18.

- Hesterman, O.B., Russelle, M.P., Sheaffer, C.C. & Heichel, G.H. 1987. Nitrogen-utilization from fertilizer and legume residues in legume-corn rotations. *Agronomy Journal* 79: 726-731.
- Jenkinson, D.S., Fox, R.H. & Rayner, J.H. 1985. Interactions between fertilizer nitrogen and soil-nitrogen - the so-called priming effect. *Journal of Soil Science* 36: 425-444.
- Kurdali, F., Kalifa, K., AlShamma, M. 1997. Cultivar differences in nitrogen assimilation, partitioning and mobilization in rain-fed grown lentil. *Field Crops Research* 54: 235-243.
- Kuzyakov, Y., Friedel, J.K., Stahr, K. 2000. Review of mechanisms and quantification of priming effects. *Soil Biology & Biochemistry* 32: 1485-1498.
- Leip, A., Britz, W., Weiss, F. & De Vries, W. 2011. Farm, land, and soil nitrogen budgets for agriculture in Europe calculated with CAPRI. *Environmental Pollution* 159: 3243-3253
- Liu, J., You, L., Amini, M., Obersteiner, M., Herrero, M., Zehnder, A.J. & Yang, H. 2010a. A high-resolution assessment on global nitrogen flows in cropland. *Proceedings of the National Academy of Sciences of the United States of America* 107: 8035-8040.
- Liu, Y., Wu, L., Baddeley, J.A. & Watson, C.A. 2010b. Models of biological nitrogen fixation of legumes: A review. *Agronomy for Sustainable Development* 31: 155-172.
- Mahieu, S., Fustec, J., Faure, M. L., Corre-Hellou, G., Crozat, Y. 2007. Comparison of two N-15 labelling methods for assessing nitrogen rhizodeposition of pea. *Plant and Soil* 295: 193-205.
- Maimaiti, J., Zhang, Y., Yang, J., Cen, Y.-P., Layzell, D.B., Peoples, M. & Dong, Z. 2007. Isolation and characterization of hydrogen-oxidizing bacteria induced following exposure of soil to hydrogen gas and their impact on plant growth. *Environmental Microbiology* 9: 435-444.
- Mariotti, F., Tome, D. & Mirand, P.P. 2008. Converting nitrogen into protein - beyond 6.25 and Jones' factors. *Critical Reviews in Food Science and Nutrition* 48: 177-184.
- MNP. 2006. Integrated modelling of global environmental change. An overview of IMAGE 2.4. (Eds Bouwman, A.F., Kram, T. & Klein Goldewijk, K.) Netherlands Environmental Assessment Agency (MNP), Bilthoven, Netherlands.
- Peoples, M.B., Brockwell, J., Herridge, D.F., Rochester, I.J., Alves, B.J.R., Urquiaga, S., Boddey, R.M., Dakora, F.D., Bhattarai, S., Maskey, S.L., Sampet, C., Rerkasem, B., Khan, D.F., Hauggaard-Nielsen, H. & Jensen, E.S. 2009. The contributions of nitrogen-fixing crop legumes to the productivity of agricultural systems. *Symbiosis* 48: 1-17.
- Philippot, L. & Hallin, S. 2011. Towards food, feed and energy crops mitigating climate change. *Trends in Plant Science* 16: 476-480.
- Pilbeam, C.J., Wood, M. & Jones, M.J. 1997. Proportion of total nitrogen and fixed nitrogen in shoots of lentil and chickpea grown in a Mediterranean-type environment. *Experimental Agriculture* 33: 139-148.
- Schmidtke, K., Neumann, A., Hof, C., Rauber & R. 2004. Soil and atmospheric nitrogen uptake by lentil (*Lens culinaris* Medik.) and barley (*Hordeum vulgare* ssp. *nudum* L.) as monocrops and intercrops. *Field Crops Research* 87: 245-256.

- Smil, V. 1999. Nitrogen in crop production: An account of global flows. *Global Biogeochemical Cycles* 13: 647-662.
- Sosulski, F.W. & Holt, N.W. 1980. Amino-acid-composition and nitrogen-to-protein factors for grain legumes. *Canadian Journal of Plant Science* 60: 1327-1331.
- Unkovich, M.J., Baldock, J. & Peoples, M.B. 2010. Prospects and problems of simple linear models for estimating symbiotic N<sub>2</sub> fixation by crop and pasture legumes. *Plant and Soil* 329: 75-89.
- Velthof, G. L., Oudendag, D., Witzke, H.R., Asman, W.A.H., Klimont, Z. & Oenema, O. 2009. Integrated assessment of nitrogen losses from agriculture in EU-27 using MITERRA-EUROPE. *Journal of Environmental Quality* 38: 402-417.
- Vinther, F.P. & Dahlmann-Hansen, L. 2005. Effects of ridging on crop performance and symbiotic N<sub>2</sub> fixation of fababean (*Vicia faba* L.). *Soil Use and Management* 21: 205-211.
- Wichern, F., Eberhardt, E., Mayer, J., Joergensen, R.G. & Müller, T. 2008. Nitrogen rhizodeposition in agricultural crops: Methods, estimates and future prospects. *Soil Biology and Biochemistry* 40: 30-48.
- Yang, J.Y., Drury, C.F., Yang, X.M., De Jong, R., Huffman, E.C., Campbell, C.A. & Kirkwood, V. 2010. Estimating biological N<sub>2</sub> fixation in Canadian agricultural land using legume yields. *Agriculture, Ecosystems and Environment* 137: 192-201.

## SUPPLEMENTARY TABLES

Supplementary Table 1. Areas (Eurostat, 2013), N retention, N fixation coefficients  $k_{\text{fix}}$  and N fixed by temporary and permanent grasslands in EU27 in 2009.

Country	Area (1000 ha)		N retention (kg ha <sup>-1</sup> )		N fixation coefficient	N fixed (kg ha <sup>-1</sup> )		Total N fixed (Gg)		
	Temp.	Perm.	Temp.	Perm.	$k_{\text{fix}}$	Temp.	Perm.	Temp.	Perm.	Total
Austria	156	1708	152	80	0.06	9.1	4.8	1.4	8.2	9.6
Belgium & Luxembourg	103	580	257	198	0.06	15.4	11.9	1.6	6.9	8.5
Bulgaria	107	1906	30	42	0.08	2.4	3.3	0.3	6.3	6.6
Cyprus	28	14	39	30	0.05	2.0	1.5	0.05	0.02	0.08
Czech Republic	687	982	48	63	0.14	6.7	8.8	4.6	8.6	13.3
Denmark	475	216	149	157	0.14	20.9	21.9	9.9	4.7	14.6
Estonia	139	256	104	61	0.15	15.6	9.2	2.2	2.4	4.5
Finland	621	58	129	72	0.05	6.5	3.6	4.0	0.2	4.2
France	3433	9614	107	93	0.07	7.5	6.5	25.8	62.3	88.0
Germany	905	4784	159	159	0.08	12.8	12.7	11.5	60.7	72.2
Greece	281	1078	108	87	0.19	20.5	16.5	5.8	17.8	23.6
Hungary	174	1000	39	55	0.17	6.6	9.4	1.2	9.4	10.5
Ireland	586	3272	274	161	0.05	13.7	8.0	8.0	26.3	34.3
Italy	2333	3978	74	57	0.16	11.9	9.2	27.8	36.5	64.3
Lithuania	483	908	37	57	0.12	4.4	6.9	2.1	6.2	8.4
Latvia	402	714	54	62	0.07	3.8	4.3	1.5	3.1	4.6
Malta	4	0	45	43	0.05	2.3	2.2	0.009	0.000	0.009
Netherlands	196	848	364	225	0.05	18.2	11.2	3.6	9.5	13.1
Poland	1531	3420	90	64	0.07	6.3	4.5	9.7	15.4	25.1
Portugal	420	1264	92	54	0.05	4.6	2.7	1.9	3.4	5.4
Romania	801	4640	86	50	0.10	8.6	5.0	6.9	23.0	29.9
Slovak Republic	174	720	49	65	0.14	6.9	9.1	1.2	6.6	7.8
Slovenia	31	294	76	44	0.05	3.8	2.2	0.1	0.6	0.8
Spain	804	8464	84	58	0.07	5.9	4.0	4.8	34.1	38.9
Sweden	1096	452	169	84	0.11	18.6	9.3	20.4	4.2	24.6
UK	1159	9844	275	116	0.05	13.8	5.8	15.9	57.1	73.0

Supplementary Table 2. Constants (bold) and calculated values used to derive estimates of fixed N and N balance for FAO classes of grain legumes. No constants were available for the Pulses category, for which production-weighted means of the other categories were used (82.15 for Total N, 53.85 for Fixed N and 7.71 for N balance). All calculated quantities are relative to one tonne of grain production.

	Common bean	Faba bean	Chick-pea	Lentil	Lupins	Pea	Soya bean	Vetches
Moisture content (g g <sup>-1</sup> )	<b>0.140</b>	<b>0.140</b>	<b>0.140</b>	<b>0.140</b>	<b>0.140</b>	<b>0.140</b>	<b>0.140</b>	<b>0.140</b>
Grain protein content (g g <sup>-1</sup> )	<b>0.25</b>	<b>0.29</b>	<b>0.22</b>	<b>0.29</b>	<b>0.36</b>	<b>0.25</b>	<b>0.40</b>	<b>0.29</b>
Protein to N	<b>6.25</b>	<b>6.25</b>	<b>6.25</b>	<b>6.25</b>	<b>6.25</b>	<b>6.25</b>	<b>6.25</b>	<b>6.25</b>
Grain N production (kg)	<b>33.85</b>	<b>40.18</b>	<b>30.00</b>	<b>39.63</b>	<b>49.12</b>	<b>34.40</b>	<b>54.76</b>	<b>39.90</b>
Harvest index	<b>0.480</b>	<b>0.490</b>	<b>0.310</b>	<b>0.415</b>	<b>0.440</b>	<b>0.507</b>	<b>0.519</b>	<b>0.340</b>
N harvest index	<b>0.830</b>	<b>0.675</b>	<b>0.805</b>	<b>0.650</b>	<b>0.840</b>	<b>0.729</b>	<b>0.730</b>	<b>0.790</b>
Above ground biomass (t)	1.792	1.755	2.774	2.072	1.955	1.696	1.657	2.529
Above-ground N production (kg)	40.78	59.52	37.26	60.97	58.48	47.19	75.02	50.51
Root:shoot ratio	<b>0.265</b>	<b>0.230</b>	<b>0.440</b>	<b>0.370</b>	<b>0.282</b>	<b>0.110</b>	<b>0.200</b>	<b>0.350</b>
Root biomass production (t)	0.475	0.404	1.221	0.767	0.551	0.187	0.331	0.885
Root N content (g g <sup>-1</sup> )	<b>0.022</b>	<b>0.022</b>	<b>0.014</b>	<b>0.014</b>	<b>0.012</b>	<b>0.022</b>	<b>0.017</b>	<b>0.029</b>
Root N production (kg)	10.30	8.88	17.09	10.73	6.51	4.10	5.70	25.76
Proportional rhizodeposition	<b>0.150</b>	<b>0.185</b>	<b>0.530</b>	<b>0.150</b>	<b>0.171</b>	<b>0.120</b>	<b>0.195</b>	<b>0.150</b>
Rhizodeposition (kg)	7.66	12.66	28.81	10.76	11.11	6.16	15.74	11.44
Total N production (kg)	58.75	81.06	83.16	82.46	76.10	57.45	96.46	87.71
Proportion of N derived from atmosphere, Ndfa	<b>0.442</b>	<b>0.770</b>	<b>0.500</b>	<b>0.700</b>	<b>0.820</b>	<b>0.700</b>	<b>0.520</b>	<b>0.720</b>
N fixed (kg)	26.0	62.4	41.6	57.7	62.4	40.2	50.2	63.2
N balance (kg)	<b>-7.9</b>	22.2	11.6	18.1	13.3	5.8	<b>-4.6</b>	23.3

Supplementary Table 3. Calculated quantities of N fixed (t) by grain legume crops in EU27 in 2009.

Country	Chickpea	Common bean	Faba bean	Lentil	Lupins	Pea	Soya bean	Vetches
Austria			418		37	1395	3576	201
Belgium & Lux		19	169			253		
Bulgaria	114	44	106	93		201	20	23
Cyprus	4	6	31	1		4		6
Czech Rep			162		137	2087	682	6
Denmark						901		
Estonia		3	6			302		
Finland						450		
France		143	27295	775	512	21683	5508	
Germany			2965		0	6671		
Greece	112	570	156	162	44	109		613
Hungary		24	6	1	37	647	3591	2
Ireland		400						
Italy	320	308	6104	84		893	23485	537
Latvia		62				105		6
Lithuania		93	318		661	917		95
Malta		10						46
Netherlands		114	393			101		
Poland		740	967		3557	314	10	324
Portugal	27	52						
Romania	3	580	1392			1206	4228	
Slovakia	6	6	12	19		470	772	15
Slovenia		16	44			56	10	
Spain	897	342	1741	687	281	5948	140	2588
Sweden		20	1629			1966		
United Kingdom			42955			6080		
Total	1484	3554	86872	1821	5267	52760	42024	4459

Supplementary Table 4. Crop production (FAO, 2013), and calculated quantities of total N production, N fixed and the N balance for grain legume crops in the EU27 in 2009. All figures in Gg.

	Common bean	Faba bean	Chickpea	Lentil	Lupins	Pea	Soya bean	Vetches	Total
Crop production	136	1392	35	32	84	1313	992	67	4051
N production	8.0	112.8	2.9	2.6	6.4	75.4	95.7	5.9	310
N fixed	3.5	86.9	1.5	1.8	5.3	52.8	49.8	4.2	206
N balance	-1.1	31.0	0.4	0.6	1.1	7.6	-4.6	1.6	37

Supplementary Table 5. Sources of constants reported in Supplementary Table 2.

	Common bean	Faba bean	Chickpea	Lentil	Lupins	Pea	Soya bean	Vetches
Moisture correction <sup>a</sup>	default	default	default	default	default	default	default	default
Grain protein content	1	1, 2	1	1	1, 2	1, 2	1, 3	4
Protein to N	5, 6	5, 6	5, 6	5, 6	5, 6	5, 6	5, 6	5, 6
Harvest index	7, 8, 9, 10, 11	12, 13, 14	15, 16	17	18, 19	20, 21, 22	23	4, 24
N harvest index	8	25, 26, 27	27, 28	28	29	30, 31	3	4, 32
Root:shoot ratio	33, 34	19, 35, 36, 37	38	39	19	31, 40, 41, 42	43, 44	32, 45
Root N content	33, 46	47, 48	28	28	49	50, 51	23	32, 52
Proportional rhizodeposition <sup>b</sup>	default	54	27, 54	default	54	21, 31, 54	55	default
Ndfa	56, 57	58, 59	59, 60	59, 61, 62, 63, 64, 65	66	58, 59	3, 60, 23, 67	68, 69

<sup>a</sup> Default value for grains harvested dry was based on the typical industry reporting figure of 0.14.

<sup>b</sup> Default value of 0.15 based on figures in <sup>53</sup> and <sup>54</sup>.

1. U.S. Department of Agriculture, A. R. S. 2013. USDA National Nutrient Database for Standard Reference, Release 26. Nutrient Data Laboratory Home Page.
2. Annicchiarico, P. 2008. Adaptation of cool-season grain legume species across climatically-contrasting environments of southern Europe. *Agronomy Journal* 100, 1647-1654
3. Salvagiotti, F. et al. 2008. Nitrogen uptake, fixation and response to fertilizer N in soybeans: A review. *Field Crops Research* 108, 1-13
4. Larbi, A., Abd El-Moneim, A.M., Nakkoul, H., Jammal, B. & Hassan, S. 2011. Intra-species variations in yield and quality determinants in *Vicia* species: 3. Common vetch (*Vicia sativa* ssp. *sativa* L.). *Animal Feed Science and Technology* 164, 241-251
5. Mariotti, F., Tome, D. & Mirand, P.P. 2008. Converting nitrogen into protein - beyond 6.25 and Jones' factors. *Critical Reviews in Food Science and Nutrition* 48, 177-184



6. Sosulski, F.W. & Holt, N.W. 1980. Amino-acid-composition and nitrogen-to-protein factors for grain legumes. *Canadian Journal of Plant Science* 60, 1327-1331
7. Gebeyehu, S., Simane, B. & Kirkby, R. 2006. Genotype x cropping system interaction in climbing beans (*Phaseolus vulgaris* L.) grown as sole crop and in association with maize (*Zea mays* L.). *European Journal of Agronomy* 24, 396-403
8. Araujo, A.P. & Teixeira, M.G. 2012. Variability of nutrient harvest indices in common bean genotypes and their relationship with grain yield. *Revista Brasileira de Ciencia do Solo* 36, 137-146
9. Builes, V., Porch, T. & Harmsen, E. 2011. Genotypic differences in water use efficiency of common bean under drought stress. *Agronomy Journal* 103, 1206-1215
10. del Mar Alguacil, M., Roldan, A., Salinas-Garcia, J.R. & Ignacio Querejeta, J. 2011. No tillage affects the phosphorus status, isotopic composition and crop yield of *Phaseolus vulgaris* in a rain-fed farming system. *Journal of the Science of Food and Agriculture* 91, 268-272
11. Zafar, M. et al. 2011. Influence of integrated phosphorus supply and plant growth promoting rhizobacteria on growth, nodulation, yield and nutrient uptake in *Phaseolus vulgaris*. *African Journal of Biotechnology* 10, 16793-16807
12. Brereton, J.C., McGowan, M. & Dawkins, T.C.K. 1986. The relative sensitivity of spring barley, spring field beans and sugar beet crops to soil compaction. *Field Crops Research* 13, 223-237
13. Muñoz-Romero, V., López-Bellido, L. & López-Bellido, R.J. 2011. Faba bean root growth in a Vertisol: Tillage effects. *Field Crops Research* 120, 338-344
14. Confalone, A., Lizaso, J.I., Ruiz-Nogueira, B., Lopez-Cedron, F.X. & Sau, F. 2010. Growth, PAR use efficiency, and yield components of field-grown *Vicia faba* L. under different temperature and photoperiod regimes. *Field Crops Research* 115, 140-148
15. Siddique, K.H.M. & Sedgley, R.H. 1986. Chickpea (*Cicer arietinum* L), a potential grain legume for southwestern Australia - seasonal growth and yield. *Australian Journal of Agricultural Research* 37, 245-261
16. Malik, S.R., Bakhsh, A., Asif, M., Iqbal, U. & Iqbal, S. 2010. Assessment of genetic variability and interrelationship among some agronomic traits in chickpea. *International Journal of Agriculture and Biology* 12, 81-85
17. Zakeri, H. et al. 2012. Lentil performance in response to weather, no-till duration, and nitrogen in Saskatchewan. *Agronomy Journal* 104, 1501-1509
18. Sandaña, P. & Calderini, D.F. 2012. Comparative assessment of the critical period for grain yield determination of narrow-leaved lupin and pea. *European Journal of Agronomy* 40, 94-101
19. Gregory, P.J. 1998. Alternative crops for duplex soils: growth and water use of some cereal, legume, and oilseed crops, and pastures. *Australian Journal of Agricultural Research* 49, 21-32
20. Lecoeur, J. & Sinclair, T.R. 2001. Harvest index increase during seed growth of field pea. *European Journal of Agronomy* 14, 173-180
21. Arcand, M.M., Knight, J.D. & Farrell, R.E. 2013. Estimating belowground nitrogen inputs of pea and canola and their contribution to soil inorganic N pools using N-15 labeling. *Plant and Soil* 371, 67-80
22. Annicchiarico, P. 2007. Lucerne shoot and root traits associated with adaptation to favourable or drought-stress environments and to contrasting soil types. *Field Crops Research* 102, 51-59
23. Schweiger, P., Hofer, M., Hartl, W., Wanek, W. & Vollmann, J. 2012. N<sub>2</sub> fixation by organically grown soybean in Central Europe: Method of quantification and agronomic effects. *European Journal of Agronomy* 41, 11-17
24. Firincioglu, H.K., Sabahaddin, Ü., Erbehtas, E. & Dogruyol, L. 2010. Relationships between seed yield and yield components in common vetch (*Vicia sativa* ssp. *sativa*) populations sown in spring and autumn in central Turkey. *Field Crops Research* 116, 30-37
25. Brunner, H. & Zapata, F. 1984. Quantitative assessment of symbiotic nitrogen-fixation in diverse mutant lines of field bean (*Vicia, faba, minor*). *Plant and Soil* 82, 407-413
26. Li, C.J. et al. 2011. Crop nitrogen use and soil mineral nitrogen accumulation under different crop combinations and patterns of strip intercropping in northwest China. *Plant and Soil* 342, 221-231
27. Lopez-Bellido, L., Iteiz-Vega, J., Garcia, P., Redondo, R. & Lopez-Bellido, R.J. 2011. Tillage system effect on nitrogen rhizodeposited by faba bean and chickpea. *Field Crops Research* 120, 189-195
28. Gan, Y. et al. 2010. Nitrogen accumulation in plant tissues and roots and N mineralization under oilseeds, pulses, and spring wheat. *Plant and Soil* 332, 451-461

29. Ayaz, S., McKenzie, B.A., Hill, G.D. & Mcneil, D.L. 2004. Nitrogen distribution in four grain legumes. *Journal of Agricultural Science* 142, 309-317
30. Lecoeur, J. & Sinclair, T.R. 2001. Nitrogen accumulation, partitioning, and nitrogen harvest index increase during seed fill of field pea. *Field Crops Research* 71, 87-99
31. Mahieu, S. et al. 2009. The influence of water stress on biomass and N accumulation, N partitioning between above and below ground parts and on N rhizodeposition during reproductive growth of pea (*Pisum sativum* L.). *Soil Biology and Biochemistry* 41, 380-387
32. Ozpinar, S. & Baytekin, H. 2006. Effects of tillage on biomass, roots, N-accumulation of vetch (*Vicia sativa* L.) on a clay loam soil in semi-arid conditions. *Field Crops Research* 96, 235-242
33. Fernández-Luqueño, V. et al. 2010. Effect of different nitrogen sources on plant characteristics and yield of common bean (*Phaseolus vulgaris* L.). *Bioresource Technology* 101, 396-403
34. Andrews, M., Sprent, J.I., Raven, J.A. & Eady, P.E. 1999. Relationships between shoot to root ratio, growth and leaf soluble protein concentration of *Pisum sativum*, *Phaseolus vulgaris* and *Triticum aestivum* under different nutrient deficiencies. *Plant, Cell & Environment* 22, 949-958
35. Vinther, F.P. & Dahlmann-Hansen, L. 2005. Effects of ridging on crop performance and symbiotic N<sub>2</sub> fixation of fababean (*Vicia faba* L.). *Soil Use and Management* 21, 205-211
36. Rengasamy, J.I. & Reid, J.B. 1993. Root system modification of faba beans (*Vicia faba* L.), and its effects on crop performance. 1. Responses of root and shoot growth to subsoiling, irrigation and sowing date. *Field Crops Research* 33, 175-196
37. Crawford, M.C., Grace, P.R., Bellotti, W.B.D. & Oades, J.M. 1997. Root production of a barrel medic (*Medicago truncatula*) pasture, a barley grass (*Hordeum leporinum*) pasture, and a faba bean (*Vicia faba*) crop in southern Australia. *Australian Journal of Agricultural Research* 48, 1139-1150
38. Gan, Y. & Liang, B. 2010. Ratios of carbon mass in nodules to other plant tissues in chickpea. *Plant and Soil* 332, 257-266
39. Hobson, K., Armstrong, R., Nicolas, M., Connor, D. & Materne, M. 2006. Response of lentil (*Lens culinaris*) germplasm to high concentrations of soil boron. *Euphytica* 151, 371-382
40. McPhee, K. 2005. Variation for seedling root architecture in the core collection of pea germplasm. *Crop Science* 45, 1758-1763
41. Baddeley, J.A. & Henderson, T.J. 2011. Characterisation of key above- and below-ground parameters of different pea cultivars. *Aspects of Applied Biology* 109, 161-164
42. Gan, Y., Liang, B., Liu, L., Wang, X. & McDonald, C. 2011. C:N ratios and carbon distribution profile across rooting zones in oilseed and pulse crops. *Crop & Pasture Science* 62, 496-503
43. Yang, J.Y. et al. 2010. Estimating biological N<sub>2</sub> fixation in Canadian agricultural land using legume yields. *Agriculture Ecosystems & Environment* 137, 192-201
44. Gerardo, R., Gutierrez Boem, F.H. & Fernandez, M.C. 2013. Severe phosphorus stress affects sunflower and maize but not soybean root to shoot allometry. *Agronomy Journal* 105, 1283-1288
45. Mariotti, M., Masoni, A., Ercoli, L. & Arduini, I. 2009. Above- and below-ground competition between barley, wheat, lupin and vetch in a cereal and legume intercropping system. *Grass and Forage Science* 64, 401-412
46. Mortimer, P., Perez-Fernandez, M. & Valentine, A. 2009. Arbuscular mycorrhizae affect the N and C economy of nodulated *Phaseolus vulgaris* (L.) during NH<sub>4</sub><sup>+</sup> nutrition. *Soil Biology & Biochemistry* 41, 2115-2121
47. Lopez-Bellido, F.J., Lopez-Bellido, R.J., Redondo, R. & Lopez-Bellido, L. 2010. B value and isotopic fractionation in N<sub>2</sub> fixation by chickpea (*Cicer arietinum* L.) and faba bean (*Vicia faba* L.). *Plant and Soil* 337, 425-434
48. Mauromicale, G. et al. 2005. Root nodulation and nitrogen accumulation and partitioning in legume crops as affected by soil solarization. *Plant and Soil* 271, 275-284
49. McNeill, A.M. & Fillery, I.R.P. 2008. Field measurement of lupin belowground nitrogen accumulation and recovery in the subsequent cereal-soil system in a semi-arid Mediterranean-type climate. *Plant and Soil* 302, 297-316
50. Ludidi, N.N. et al. 2007. Genetic variation in pea (*Pisum sativum* L.) demonstrates the importance of root but not shoot C/N ratios in the control of plant morphology and reveals a unique relationship between shoot length and nodulation intensity. *Plant Cell and Environment* 30, 1256-1268
51. Voisin, A.S., Salon, C., Munier-Jolain, N.G. & Ney, B. 2002. Effect of mineral nitrogen on nitrogen nutrition and biomass partitioning between the shoot and roots of pea (*Pisum sativum* L.). *Plant and Soil* 242, 251-262

52. Pederson, G.A., Brink, G.E. & Fairbrother, T.E. 2002. Nutrient uptake in plant parts of sixteen forages fertilized with poultry litter: Nitrogen, phosphorus, potassium, copper, and zinc. *Agronomy Journal* 94, 895-904
53. Mahieu, S., Fustec, J., Faure, M.L., Corre-Hellou, G. & Crozat, Y. 2007. Comparison of two N-15 labelling methods for assessing nitrogen rhizodeposition of pea. *Plant and Soil* 295, 193-205
54. Wichern, F., Eberhardt, E., Mayer, J., Joergensen, R.G. & Müller, T. 2008. Nitrogen rhizodeposition in agricultural crops: Methods, estimates and future prospects. *Soil Biology & Biochemistry* 40, 30-48
55. Laberge, G., Franke, A., Ambus, P. & Høgh-Jensen, H. 2009. Nitrogen rhizodeposition from soybean (*Glycine max*) and its impact on nutrient budgets in two contrasting environments of the Guinean savannah zone of Nigeria. *Nutrient Cycling in Agroecosystems* 84, 49-58
56. Chagas, E., Araujo, A.P., Alves, B.J.R. & Teixeira, M. G. 2010. Seeds enriched with phosphorus and molybdenum improve the contribution of biological nitrogen fixation to common bean as estimated by N-15 isotope dilution. *Revista Brasileira de Ciencia do Solo* 34, 1093-1101
57. Hardarson, G. et al. 1993. Genotypic variation in biological nitrogen-fixation by common bean. *Plant and Soil* 152, 59-70
58. Peoples, M.B. et al. 2009. The contributions of nitrogen-fixing crop legumes to the productivity of agricultural systems. *Symbiosis* 48, 1-17
59. Rennie, R.J. & Dubetz, S. 1986. N-15 - determined nitrogen-fixation in field-grown chickpea, lentil, fababean, and field pea. *Agronomy Journal* 78, 654-660
60. Hardarson, G. & Atkins, C. 2003. Optimising biological N<sub>2</sub> fixation by legumes in farming systems. *Plant and Soil* 252, 41-54
61. Schmidtke, K., Neumann, A., Hof, C. & Rauber, R. 2004. Soil and atmospheric nitrogen uptake by lentil (*Lens culinaris* Medik.) and barley (*Hordeum vulgare* ssp. *nudum* L.) as monocrops and intercrops. *Field Crops Research* 87, 245-256
62. Hafeez, F.Y., Shah, N.H. & Malik, K.A. 2000. Field evaluation of lentil cultivars inoculated with *Rhizobium leguminosarum* bv. *viciae* strains for nitrogen fixation using nitrogen-15 isotope dilution. *Biology and Fertility of Soils* 31, 65-69
63. Haynes, R.J., Martin, R.J. & Goh, K.M. 1993. Nitrogen-fixation, accumulation of soil-nitrogen and nitrogen-balance for some field-grown legume crops. *Field Crops Research* 35, 85-92
64. Kurdali, F., Kalifa, K. & AlShamma, M. 1997. Cultivar differences in nitrogen assimilation, partitioning and mobilization in rain-fed grown lentil. *Field Crops Research* 54, 235-243
65. Shah, Z., Shah, S.H., Peoples, M.B., Schwenke, G.D. & Herridge, D.F. 2003. Crop residue and fertiliser N effects on nitrogen fixation and yields of legume-cereal rotations and soil organic fertility. *Field Crops Research* 83, 1-11
66. Unkovich, M.J., Pate, J.S., Armstrong, E.L. & Sanford, P. 1995. Nitrogen economy of annual crop and pasture legumes in southwest Australia. *Soil Biology & Biochemistry* 27, 585-588
67. Bohm, G.M.B. et al. 2009. Glyphosate- and imazethapyr-induced effects on yield, nodule mass and biological nitrogen fixation in field-grown glyphosate-resistant soybean. *Soil Biology & Biochemistry* 41, 420-422
68. Mueller, T. & Thorup-Kristensen, K. 2001. N-fixation of selected green manure plants in an organic crop rotation. *Biological Agriculture and Horticulture* 18, 345-363
69. Papastylianou, I. & Danso, S.K.A. 1991. Nitrogen-fixation and transfer in vetch and vetch-oats mixtures. *Soil Biology & Biochemistry* 23, 447-452