# From N<sub>2</sub> fixation to N<sub>2</sub>O emission in a grass-clover mixture

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## Introduction

In organic farming, biological N<sub>2</sub> fixation in grass-legume swards provides a major N input to the system, but knowledge is sparse regarding the amount of fixed N<sub>2</sub> lost from the grasslands as N<sub>2</sub>O. Nitrifying and denitrifying bacteria are the main contributors to the N<sub>2</sub>O production in soils. Currently, no contribution from biological N<sub>2</sub> fixation in grass-legume swards is included in the national N<sub>2</sub>O inventories, partly because of uncertainties in quantifying the N<sub>2</sub> fixation in the grasslands (Mosier et al, 1998). According to the guidelines issued by The Intergovernmental Panel on Climate Change (IPCC), inventories for N<sub>2</sub>O emissions from agricultural soils should be based on the assumption that 1.25 % of the added N is emitted as N<sub>2</sub>O (IPCC, 1997). The standard N<sub>2</sub>O emission factor of 1.25 % could be considerably unrepresentative for biologically fixed N<sub>2</sub> for two reasons. First, only a part of the fixed N is mineralised during the lifetime of the crop, and second, the release of inorganic N into the soil occurs slowly following degradation of organic residues. Therefore, a <sup>15</sup>N<sub>2</sub>-tracer-experiment was initiated on grass-clover to assess the contribution of recently fixed N<sub>2</sub> as a source of N<sub>2</sub>O and the translocation of N from clover to companion grass.

### Materials and methods

A mixture of white clover (*Trifolium repens* L. cv. Klondike) and perennial ryegrass (*Lolium perenne* L. cv. Fanda) was sown in pots using topsoil from an organic crop rotation. The <sup>15</sup>N-labelling approach consisted of introducing <sup>15</sup>N<sub>2</sub> into both the above- and below-ground atmosphere to trace the biological N<sub>2</sub> fixation. A minimum-volume gastight growth cabinet was developed, which could host 12 pots of 15 cm × 15 cm size. In this growth cabinet, three incubations were conducted with grass-clover mixtures at 4, 6 and 8 months of age. At each incubation event the pots were situated in the growth cabinet for 14 days during which period the atmosphere was enriched in <sup>15</sup>N<sub>2</sub> to 0.4 atom% excess. After the labelling period, half of the grass-clover pots were sampled. The N<sub>2</sub> fixation during the labelling period was established by relating the excess <sup>15</sup>N content of the plant material to the <sup>15</sup>N enrichment of the atmospheric N<sub>2</sub>. During the following seven days, emission of <sup>15</sup>N<sub>2</sub>O was measured from the remaining half of the pots using a static chamber method.

### Results

At 4 months, N<sub>2</sub> fixation measured in grass-clover shoots and roots constituted 339 mg N m<sup>-2</sup> d<sup>-1</sup> (Fig. 1). This is three to 13 times larger than daily means of N<sub>2</sub> fixation determined in harvested shoot material in the field (Høgh-Jensen & Schjoerring, 1997; Vinther & Jensen, 2000), probably because of optimal growth conditions at this stage of the experiment. Following a severe aphid attack, N<sub>2</sub> fixation dropped dramatically at 6 months. Translocation of fixed N from clover to grass shoots was observed at 6 and 8 months and represented 1 mg N m<sup>-2</sup> d<sup>-1</sup>. Emission of N<sub>2</sub>O-N derived from fixed N<sub>2</sub> was detected at 4 months only, and accounted for 3 ppm  $\pm$  0.5 ppm of the fixed N<sub>2</sub>. The results are preliminary; since fixed N present in the soil has not yet been estimated.

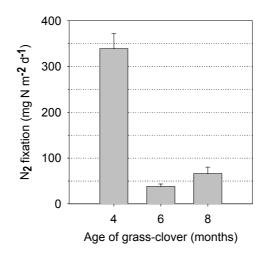


Figure 1. Biological  $N_2$  fixation measured in grass-clover shoots and roots; n = 4, means  $\pm$  SE.

### Conclusions

Biological N<sub>2</sub> fixation plays an important role as N input to the grass-clover system. The aphid attack on the clover component led to translocation of fixed N to companion grass, which agrees with the view that N transfer is indirect, *i.e.* caused by turnover of organic clover residues. Emission of N<sub>2</sub>O-N derived from recently fixed N<sub>2</sub> was not detected at 6 and 8 months, probably because the <sup>15</sup>N enrichment of the clover rhizodeposition was too low. In conclusion, results at 4 months indicate that only a small proportion of the fixed N is lost as N<sub>2</sub>O over the course of a few weeks. Even if a longer turnover time for clover N is considered, we find it unlikely that the N<sub>2</sub>O emission factor for biologically fixed N<sub>2</sub> in grass-clover mixtures would reach the standard emission factor of 1.25 % suggested by IPCC.

#### Acknowledgements

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#### References

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