

Total N difference method and ¹⁵N isotope dilution method – A comparative study on N-fixation

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Aim

The aim of this study was to compare the total N difference method (Munroe & Davies, 1974) with the ¹⁵N isotope dilution method (Fried & Middleboe, 1977; Warembourg, 1993) for the measurement of N-fixation by selected green manure plants in an organic crop rotation.

Method

Field experiments were located at the Agricultural Research Centre Aarslev at the isle of Funen (Denmark) in wet temperate climate (Mueller & Thorup-Kristensen, 2001). The randomised block design consisted of three replicates with a plot size of 2.5×3 m.

20.9 kg ha⁻¹ (NH₄)₂SO₄ enriched with 10.7 atom% ¹⁵N were applied after seed bed preparation. The study included 5 different leguminous green manure plants (Fig. 1) and Italian ryegrass as non-fixing control plant. As an important prerequisite, measured soil mineral N, and rooting depths of legumes and control plants indicated comparable patterns of root growth and N-uptake (Thorup-Kristensen, 2001).

As shown in box 1 for the total N difference method, N derived from the atmosphere (N_{dfa}), was calculated from N in legumes (N_{fix}) and N in non fixing control plants (N_{contr}).

For the ¹⁵N Isotope dilution method, N_{dfa} was calculated as shown in box 2.

Results

No significant differences in N_{dfa} between the ¹⁵N isotope dilution method and the total-N difference method (Fig. 1 and 2) could be observed.

Box 1: Calculation of N_{dfa} with the total N difference method:

$$N_{dfa} [\text{kg ha}^{-1}] = N_{fix} - N_{contr}$$

Box 2: Calculation of N_{dfa} with the ¹⁵N isotope dilution method:

$$N_{dfa} [\% \text{ total plant N}] =$$

$$1 - \frac{{}^{15}\text{N atom\% excess (fixing plant)}}{{}^{15}\text{N atom\% excess (nonfixing control plant)}} \times 100$$

$$N_{dfa} [\text{kg ha}^{-1}] = \frac{N_{dfa} [\% \text{ total plant N}]}{100\%} \times N_{yield} [\text{kg ha}^{-1}]$$

Discussion

Due to comparable patterns of root growth and N-uptake by legumes and control plants, both methods delivered the same results.

Taking into account uncertainties due to pool substitution (¹⁴N↔¹⁵N) and uneven distribution of ¹⁵N in the soil profile, it can be assumed that results obtained with the total-N difference method are more reliable than those obtained with the ¹⁵N isotope dilution method. This is true at least for annual crops under conditions with low plant-availability of soil N, which implies small amounts of total N in the non-fixing control plants.

If the only purpose of an experiment is to estimate N-fixation under such conditions, it must be questioned whether it is worth to invest the additional financial resources, energy and manpower required for the ¹⁵N isotope dilution method.

Literature

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for their ability to reduce soil nitrate-N content, and how can this be measured? *Plant and Soil*, **230**, 185-195.

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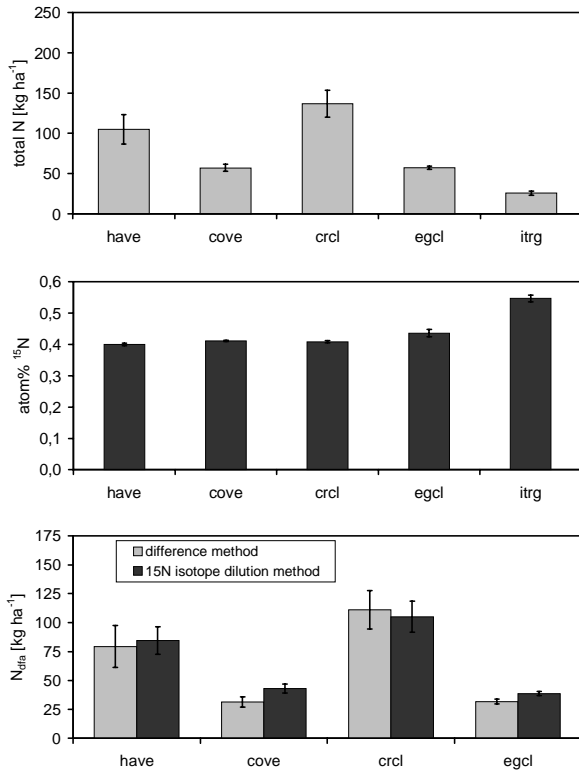


Figure 1: Above ground total shoot N, atom% ¹⁵N, and fixed N derived from the atmosphere (N_{dfa}) in different leguminous plants and in non-fixing control plants in November 1996. Bars show standard errors. have = hairy vetch (*Vicia villosa*), cove = common vetch (*Vicia sativa*), crcl = crimson clover (*Trifolium incarnatum*), egcl = Egyptian clover (*Trifolium alexandrinum*), itrgr = Italian ryegrass (*Lolium multiflorum*).

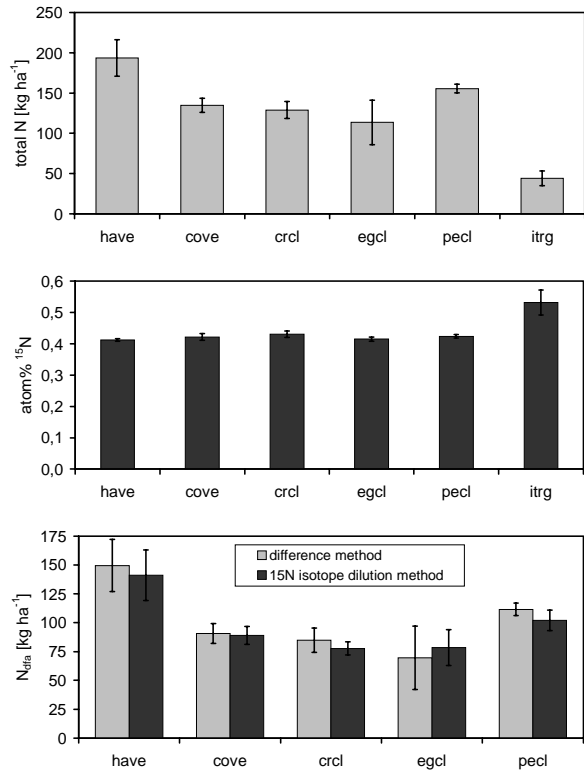


Figure 2: Above ground total shoot N, atom% ¹⁵N, and fixed N derived from the atmosphere (N_{dfa}) in different leguminous plants and in non-fixing control plants in November 1997. Bars show standard errors. have = hairy vetch (*Vicia villosa*), cove = common vetch (*Vicia sativa*), crcl = crimson clover (*Trifolium incarnatum*), egcl = Egyptian clover (*Trifolium alexandrinum*), pecl = Persian clover (*Trifolium resupinatum*), itrgr = Italian ryegrass (*Lolium multiflorum*).