



Persistence of biological nitrogen fixation in high latitude grass-clover grasslands under different management practices

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Biological nitrogen fixation (BNF) can substantially contribute to N supply in permanent grasslands, improving N yield and forage quality, while reducing inorganic N inputs. Among the factors critical to the performance of BNF in grass-legume mixtures are selected grass and legume species, proportion of legumes, the soil-climatic conditions, in particular winter conditions, and management practices (e.g. fertilization and compaction). In high latitude grasslands, low temperatures can reduce the performance of BNF by hampering the legume's growth and by suppressing N₂ fixation. Estimation of BNF in field experiments is not straightforward. Different methods have been developed providing different results. In the present study, we evaluated the performance of BNF, in a newly established field experiment in North Norway over four years. The grassland consisted of white clover (*Trifolium repens* L.) and red clover (*Trifolium pratense* L.) sown in three proportions (0, 15 and 30% in total) together with timothy (*Pheum pratense* L.) and meadow fescue (*Festuca pratensis* L.). Three levels of compaction were applied each year (no tractor, light tractor, heavy tractor) together with two different N rates (110 kg N/ha as cattle slurry or 170 kg N/ha as cattle slurry and inorganic N fertilizer). We applied two different methods, the ¹⁵N natural abundance and the difference method, to estimate BNF in the first harvest of each year. Overall, the difference method overestimated BNF relative to the ¹⁵N natural abundance method. BNF in the first harvest was compared to winter survival of red and white clover plants, which decreased with increasing age of the grassland. However, winter conditions did not seem to affect the grassland's ability to fix N in spring. The fraction of N derived from the atmosphere (NdfA) in white and red clover was close to 100% in each spring, indicating no suppression of BNF. BNF increased the total N yield of the grasslands by up to 75%, mainly due to high N-yields in red clover. However, the total biomass and N yield of red clover decreased dramatically throughout the following years, reflecting the negative cumulative effect of N fertilization and compaction. Overall, BNF by clover can contribute substantially to N supply to northern grasslands, but better cultivation strategies are needed to improve the persistence of clover.