

Short communication

Comparison of Two Techniques for Assessing Denitrification in Terrestrial Ecosystems <sup>☆</sup>

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In an effort to quantify denitrification in a terrestrial ecosystem we have adapted the gas chromatographic techniques as outlined by Barbaree & Payne (1967) and Payne (1973). The preliminary results presented here compare the rates of denitrification (as determined by the conversion of nitrous oxide to atmospheric nitrogen) to the standard most probable number (MPN) procedure as described by Alexander (1965). While the latter technique can provide only enumeration of denitrifying cells, the gas chromatographic procedure allows for the calculation of actual transfer rates.

Samples were collected at varying time intervals from soil under three vegetation covers at the Eastern Deciduous Forest Biome (US-IBP) Coweeta intensive study site. The samples (contained in 220 ml screw top jars whose lids were fitted with serum stoppers) were flushed with N<sub>2</sub>O. After incubation, at 18°C for 24 and 48 hours, 0.2 or 0.5 ml of the gas atmosphere over each sample was removed and injected into a F and M model 700 gas chromatograph connected to an Infotronics integrator and Honeywell strip chart recorder. The column (19.5 feet x 1/8 inch stainless steel packed with Porapak Q), was operated at ambient temperature. Detection was accomplished by a thermal conductivity detector operating at 250°C and 150 mA. The reaction products were identified by their retention times as compared to those of standard samples (N<sub>2</sub>:96 sec; N<sub>2</sub>O:480 sec).

Table 1 compares the results from a typical series of samples. The MPN analysis always shows that the highest denitrifying population occurs in soil from under the grass cover. However, the reverse is true when assays of denitrifying activity of identical samples are made using the gas chromatographic technique. In this instance, N<sub>2</sub> evolution from the mixed deciduous soil is twice that of the pine and four times that noted for the grassland soil. The application of gas chromatography will provide a rapid and accurate analysis of denitrification in soils.

Table 1 Denitrification in soils

Vegetation cover	MPN "Denitrifiers"/g soil (dry wt.)	μl N <sub>2</sub> evolved/g soil (dry wt.)/hour
Fescue grass	1.7 x 10 <sup>4</sup>	32.36 ± 2.14
White pine plantation	2.6 x 10 <sup>2</sup>	65.25 ± 4.20
Mixed deciduous forest	2.8 x 10 <sup>3</sup>	115.69 ± 30.27

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### References

- Alexander M. 1965. Denitrifying bacteria. In: Methods of soil analysis: 2: 1484-1486. Eds. C.A. Black et al., American Society of Agronomy, Madison, Wisconsin.
- Barbaree J.M. & Payne W.J. 1967. Products of denitrification by a marine bacterium as revealed by gas chromatography. Marine Biology 1: 136-139.
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### Discussion

- F. Taub: Having shown that denitrification occurs in soils, have you considered the effects of grass on tree growth in nitrogen limited systems? Might trees excrete inhibitors to denitrifiers?
- R.L. Todd: I am certain that microbes in soil under certain conditions can compete with the plant for available nitrate and the end product may be atmospheric nitrogen. It has been shown that certain grass species excrete inhibitors of nitrification into the rhizosphere. The same may be true for forest soils and denitrification. However, I know of no data on this subject.