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Microbial Growth and Immobilization/Mineralization of N in the Rhizosphere*

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Abstract

The release of organic materials from growing roots represents a significant energy input to the soil microbial community. If utilized efficiently for microbial growth, this release of root-C would result in a substantial microbial N-immobilization. The stimulation of the microorganisms by the growing roots may therefore result in a reduction of available mineral N to the plant, and a negative overall effect of plant roots on the net N-mineralization in the soil (planted versus unplanted soil).

However, comparisons of rhizosphere and non-rhizosphere soil have demonstrated that the "rhizosphere effect" on the net N-mineralization from soil organic matter is positive in most cases (Hart et al., 1979; Rosswall and Paustian 1984; Clarholm 1985; Haider et al., 1987, 1988). On this background, the microbial N-immobilization through growth on root-released organic C seems to be counter-balanced by a stimulation of the N-mineralization by the plant roots.

One possible explanation for an increased net N-mineralization in the rhizosphere (Wang and Bakken 1989) is that the roots successfully compete with the microflora for mineral N. Hence, an apparent stimulation of N-mineralization by plant roots does not necessarily reflect a real enhancement of the microbial decomposition of soil organic materials, but may simply be due to a reduction of the N-immobilization rate.

I will summarize and discuss some of the experiments in our laboratory, designed to isolate the two opposite effects of plant roots: The N-immobilization due to the release of organic materials from growing roots, and the restriction of microbial growth and N-immobilization in the rhizosphere due to root-uptake of mineral N.

Experiment 1

In the first experiments (Breland and Bakken 1990), we focused on the N-immobilization in the rhizosphere due to carbon supply from roots. Plants (barley, ryegras and clover) were grown in subsoil silt with an extremely low organic matter content (0.16% organic C, 0.012% organic N), hence the mineralization of soil organic

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N did not significantly influence the N budgets. The soil was supplied with ample amounts of mineral-N (NO_3). The low levels of organic matter and microbial biomass in the soil made it possible to measure the plant-induced increase of microbial biomass (Acridine orange direct count, AODC) with appreciable precision, and to estimate the total microbial N immobilization as a difference (in total soil organic N) between planted and unplanted soil (the percentage differences between planted and unplanted soil was 6–16% for soil organic N, and 100–250% for the microbial biomass).

12–17% of the fertilizer-N was recovered as an increase in soil organic N in planted pots (planted–unplanted). This was attributed to microbial growth and N-immobilization based on organic materials released from the growing roots (the silt was easily separated from roots by very gentle washing, hence losses of root material was negligible). The net increase in biomass-N (AODC-N) accounted only for $\frac{1}{5}$–$\frac{1}{4}$ of the accumulation of soil organic N. This was not unexpected, since the storage of N in biomass is only transient due to a rapid biomass turnover in the rhizosphere. Similar recoveries of immobilized N in the biomass pool have been obtained in other experiments (Schnürer and Rosswall 1987).

The turnover of the rhizosphere biomass would result partly in remineralization (and uptake by the plant roots) and partly in accumulation of non-biomass organic N. Empirical growth yield coefficients in soil (based on ^{14}C-studies) are much lower (around 0.2) than theoretical coefficients based on energetic considerations and pure culture studies (0.4-0.6). The reason is the short halflife of newly formed biomass in soil; the empirical growth yields are result of build-up, decay and reutilization of substrates released from decaying organisms.

On this background, it is likely that the amount of fertilizer nitrogen recovered as an increase in soil organic N represents only a fraction of the fertilizer nitrogen which has passed through the microbial biomass pool. Thus, the result indicates that the microbial biomass serves as an important but transient nitrogen sink, which partly delivers its nitrogen to the plants through remineralization, and partly transforms the nitrogen into more or less stable soil organic compounds. This conclusion is supported by the observations by Jackson et al. (1989) who found that around 50% of the added ^{15}N-fertilizer (annual grassland) was taken up by the microflora, provided that the uptake was measured shortly (24 hrs) after the ^{15}N was added (to prevent remineralization etc.)

As a conclusion, a microbial N-immobilization in the rhizosphere is a potentially significant sink for mineral nitrogen.

Experiment 2

A mechanism of counteracting this rhizosphere N-immobilization seems desirable from the plants point of view, and it probably exists since several experiments have

demonstrated that plant roots give an apparent stimulation of the net N-mineralization (net rates in planted versus unplanted soil).

As mentioned earlier, a successful competition with N-immobilizing microorganisms could be one way for the plants to increase their N-input. We believed this competition to be strongly dependent on the heterogeneity of the soil with respect to C- and N-rich microsites. In order to test this hypothesis, we produced growth media for plants consisting of subsoil silt to which we added finely ground clover leaves and barley straw (200 g silt per pot plus 5 g straw and 2.5 g clover leaves). The straw layers served as C-rich sites (C/N-ratio = 71) with a potential for N-immobilization. No mineral N was added, and the amount of straw was sufficient to ensure a complete immobilization of the N mineralized during decomposition of the N-rich clover leaves (C/N-ratio = 10).

The straw and clover leaves were placed at varying distance in the soil (either mixed completely with soil, or added as discrete layers with varying distance, see Table 1). Half of the pots in each treatment were planted and the other half were left unplanted and incubated at the same temperature and moisture regime as the planted ones.

Table 1. Nitrogen mineralized after four weeks: Effect of plants and spatial distance between N-source (clover leaves) and C-source (straw).

Distance between straw and clover	N mineralized (mg/pot) (plant-N + mineral-N in soil)	
	Planted	Unplanted
0 mm (mixture)	5.7	0.9
9 mm	31.4	4.0
16 mm	27.1	5.3
27 mm	29.4	16.3

The results demonstrated clearly the ability of the plants to outcompete the heterotrophic microorganisms, and this competition was strongly dependent on the distance between clover and straw: In the treatment where straw and clover were mixed completely, the plants showed N-deficiency symptoms early, and took up much less nitrogen than in the other treatments. However, the comparison of the estimated net N-mineralization (N-uptake in plants plus mineral N in soil) in the planted and unplanted pots (Table 1) showed that even in the "mixture"-treatments the plants got hold of a substantial amount of mineralized nitrogen which would otherwise (in unplanted) be immobilized by the microorganisms.

The mineral N-accumulation in the unplanted soil increased gradually with increasing distance between the clover and straw, whereas planted pots reached a maximum already at the lowest distance (9 mm). The mineral N content of the planted pots was low through the whole period (data not shown).

The data demonstrates how plant root under such rather artificial conditions may greatly increase the apparent net N-mineralization, not necessarily by stimulating the microbial activity ("priming action"), but rather by reducing the microbial immobilization rate. The extrapolations of these observations to natural conditions may be questioned. The mechanism might work, provided that natural soil has a sufficient spatial heterogeneity with respect to available C and N. More knowledge about this spatial heterogeneity and the occurrence of possible "hot spots" for N-immobilization (such as straw particles) in natural soil is therefore of great interest.

A significant depression of the microbial growth by the presence of plants was demonstrated both for bacteria (AODC and plate counts) and for fungi (fluorescence microscopy after staining with fluorescein isothiocyanate).

Experiment 3

Barley plants were grown in the same type of growth medium (layers of straw, clover and soil) as in the second experiment (Wang and Bakken, unpublished), but smaller increments for the distances between straw and clover layers were used, and several new parameters were measured.

The reduction in total organic C in the soil was measured, to check if there was any significant "priming" effect of plant roots on the C-mineralization. The measurement of C-mineralization was possible, since the fresh plant material (straw plus clover) represented the major part of the total C in the soil (85% at the beginning of the incubation). A substantial reduction in total C (25% after 42 days) was observed, but there were negligible differences between planted and unplanted pots.

The net N-mineralization (plant-N plus mineral N) in planted and unplanted soil confirmed the result of the former experiment, and demonstrated a gradual increase in the nitrogen mineralization rate in planted pots when the distance increased from 3 to 9 mm.

The N-immobilization/mineralization potential in the straw and clover layers was measured several times during the growth of the plants (measured in slurries with mineral N added, incubated aerobically (shaken) for 7 days at 21°C). It was found that the potential N-immobilization in slurries from the straw layers was higher in planted than in unplanted soil. This was expected, since the presence of plants reduced the mineral N delivery (from clover layers) to the straw decomposing microflora considerably, and at the same time the presence of plant roots represents an additional source of C for the microflora. A significant inhibition of microbial growth (bacteria and fungi) by plant roots was demonstrated in the straw-layers. In clover layers the plant roots had an insignificant effect on microbial biomass.

Concluding Remarks

Our knowledge about the regulating function of mineral N-availability on the

microbial metabolism of C and N in soil is scarce. In unplanted soil, N-limitation of microbial growth is transient, and its effects unclear (Scott Smith et al., 1989). However, the present investigation demonstrates that in the rhizosphere, N-limitation of microbial growth may be severe and persistent. The effect of this possible N-limitation on microorganisms and their transformation of C and N is an interesting aspect of the "rhizosphere" which deserves the attention of soil microbiologists.

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