Response of Total Nitrogen and Microbial Biomass Nitrogen in Brown Soils of Different Fertility Levels to Exogenous Maize Residue Nitrogen Postprint

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Abstract

Using soils with high and low fertility levels developed from a 27-year long-term continuous maize cropping experiment on brown soil as the research subjects, and employing 15N-labeled maize plants as experimental materials, maize roots, stems, and leaves were separately added to the two soils (comprising a total of 8 treatments). Laboratory incubation simulation and 15N isotopic tracing techniques were utilized to elucidate the variation patterns of total nitrogen content and microbial biomass nitrogen in soils of different fertility following the addition of maize roots, stems, and leaves. The results demonstrated: (1) Following the addition of maize roots, stems, and leaves, the increments in total nitrogen content in low-fertility brown soil were 5.75%, 4.77%, and 3.75% higher than those in high-fertility brown soil, respectively, while the contribution rates of exogenous new nitrogen were 3.54%, 3.28%, and 2.49% higher than those in high-fertility brown soil, respectively. These findings indicate that soils of different fertility levels exhibit differential responses to maize residue addition, with low-fertility brown soil demonstrating greater sensitivity in response to exogenous new nitrogen input and possessing stronger immobilization capacity. (2) During the 56-day incubation period following maize residue addition, microbial biomass nitrogen in low-fertility brown soil increased by an average of 0.83-0.98 fold, whereas that in high-fertility brown soil increased by an average of 0.87-1.56 fold. This observation reveals that the addition of different maize plant parts significantly promotes the accumulation of soil microbial biomass nitrogen, suggesting that exogenous organic matter input constitutes a crucial factor stimulating soil microbial quantity and activity, with the stimulating effect being more pronounced in high-fertility soil. Moreover, in high-fertility soil, microbial biomass nitrogen in stem and leaf addition treatments was significantly higher

than that in the root addition treatment; however, in low-fertility soil, no significant differences in microbial biomass nitrogen were observed among root, stem, and leaf addition treatments. The contribution of exogenous organic nitrogen input to the soil nitrogen pool is intimately associated with soil fertility levels and the inherent material composition characteristics of different residue parts.

Full Text

Preamble

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Effect of Maize-Derived Nitrogen Supplementation on the Total and Microbial Biomass Nitrogen of Brown Earths with Different Fertility Levels

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Abstract

This study investigated the response of total nitrogen (TN) and microbial biomass nitrogen (MBN) in brown earths of contrasting fertility to exogenous maize residue nitrogen. Using soils from a long-term (27-year) maize monoculture experiment with low-fertility (LF) and high-fertility (HF) treatments, we conducted an incubation experiment with ^{1} N-labeled maize roots, stems, and leaves. Our results showed that: (1) The addition of maize residues increased TN content in LF soil by 5.75%, 4.77%, and 3.75% for roots, leaves, and stems respectively—significantly higher than in HF soil. The contribution of maize-derived N to total N was also greater in LF soil, with MBN increasing by 3.54%, 3.28%, and 2.49% for roots, stems, and leaves respectively, compared to HF soil. This indicates that low-fertility soil is more sensitive to organic input and has a greater capacity to sequester exogenous N. (2) During the 56-day incubation, MBN in LF soil increased by 0.83–0.98 times, while HF soil MBN increased by 0.87–1.56 times, demonstrating that maize residue addition stimulates microbial biomass, particularly in HF soils. No significant differences in MBN were observed among different maize parts in LF soil, whereas in HF soil, stem and leaf additions had more pronounced effects than root addition. We conclude that the contribution of exogenous organic N to the soil N pool depends on both initial soil fertility and the biochemical composition of plant residues.

Keywords: soil fertility; maize residue; N value; soil total nitrogen; microbial biomass nitrogen

Introduction

Nitrogen is one of the most active and critical factors in soil fertility and a major limiting factor in agricultural production. Consequently, nitrogen fertilizer application has long been the most important measure for increasing crop yields. Crop residues represent the primary source of soil organic carbon in agroecosystems and also serve as a significant nitrogen pool. Returning crop residues to fields is therefore crucial for promoting soil nitrogen cycling, reducing chemical fertilizer use, and maintaining farmland productivity. Previous studies have shown that straw return effects on soil nutrients and biological properties are highly complex processes. Straw decomposition is a biochemical process driven by soil microorganisms, and promoting microbial nitrogen immobilization represents an effective pathway for reducing nitrogen losses. Microorganisms play a dominant role in the assimilation of exogenous nitrogen and transformation of organic matter. Thus, understanding the response of microbial biomass nitrogen is essential for evaluating the bioavailability of crop residue nitrogen.

Different parts of crop residues exhibit significant compositional differences, particularly in organic carbon and nitrogen content, which affect their decomposition processes in soil. Research has found that rice straw-derived nitrogen enters different soil nitrogen pools during decomposition, and that equal amounts of straw are more effective than root stubble for replenishing labile carbon and nitrogen. Xie et al. demonstrated that different maize residue parts play key roles in soil organic carbon sequestration. However, the distribution and contribution of nitrogen from different crop residue parts in soils of varying fertility levels remain poorly understood. This study utilized a long-term maize monoculture system with two fertility levels of brown earth, combined with ^{1} N stable isotope tracing and laboratory incubation methods, to investigate the dynamic changes in total nitrogen and microbial biomass nitrogen after adding different maize residue parts. Our objective was to explore the allocation and contribution of exogenous new nitrogen during straw decomposition under different soil fertility levels, providing a theoretical basis for developing rational straw return and fertilization practices.

1. Test Soils

Soils were collected from the long-term experimental station of brown earth at Shenyang Agricultural University. The soil is a loamy brown earth developed on loess parent material, with maize as the long-term crop sown in late May each year. The low-fertility brown earth (LF) was from a plot receiving no fertilizer for 27 years, while the high-fertility brown earth (HF) was from a plot receiving high amounts of organic manure (equivalent to 270 kg N ha ^{1} yr ^{1}). Detailed experimental layout and fertilization treatments are described in An et al. After

maize harvest in autumn 2014, topsoil samples (0–20 cm) were collected, air-dried at room temperature, and roots and debris were removed. The basic physicochemical properties of the test soils are shown in Table 1.

Basic physicochemical characteristics of brown earths with different fertility levels (2014)

2. Test Organic Materials

The test material was ^{1} N-labeled maize residues. Labeling began at the jointing stage by applying (^{1} NH) SO solution (0.2 mol L ^{1}) to roots. After harvest, residues were separated into roots, stems, and leaves, crushed with a straw grinder, and sieved (mesh size not specified). The basic physicochemical properties of the ^{1} N-labeled maize residues are shown in Table 2.

Basic characteristics of ^{1} N-labeled maize residues (2014)

3. Research Methods

This experiment used laboratory constant-temperature incubation. The treatments included: LF soil with maize roots, LF soil with maize stems, LF soil with maize leaves, HF soil with maize roots, HF soil with maize stems, HF soil with maize leaves, and bare soil controls for both fertility levels. For each treatment, 120 g of air-dried soil (equivalent to oven-dry weight) was weighed, distilled water was added to adjust moisture content, and the soil was thoroughly mixed with maize residues at a ratio of 1:100 (residue to oven-dry soil). Samples were placed in a 120 g capacity incubator for constant-temperature cultivation. Soil moisture was adjusted to 60% of field water holding capacity. At 1, 7, 28, 56, and 180 days after incubation, fresh subsamples were taken for MBN determination, while other portions were air-dried, ground with a mortar, and sieved for TN and ^{1} N analysis.

4. Measurement Methods

Microbial Biomass Nitrogen: Determined using the chloroform fumigation-extraction method. Fresh soil equivalent to 10 g oven-dry weight was placed in a culture dish and put in a vacuum desiccator with purified chloroform. After vacuuming for 5 minutes, the desiccator was incubated at 25°C for 24 hours. Fumigated and non-fumigated controls were extracted with 100 mL of 0.5 mol L ^{1} K SO (soil:solution ratio 1:4) by shaking for 30 minutes. Extracts were filtered through 0.45 m membranes and total nitrogen content was measured using a High-TOC II analyzer (Elementar II, Germany). MBN was calculated as: MBN = E_N / 0.54, where E_N is the difference in total nitrogen between fumigated and non-fumigated samples.

Total Nitrogen and ^{1} N: Determined using an elemental analyzer-stable isotope mass spectrometer (EA-IRMS, IsoPrime100, Germany). Samples were

combusted at 920°C (combustion tube) and 600°C (reduction tube). After CO removal, gases entered the mass spectrometer via a diluter. ^{1}N values were calculated relative to atmospheric N standard (^{1}N = 0.0036765).

5. Calculation Methods

The contribution rate of exogenous new nitrogen to soil total nitrogen was calculated as:
Contribution rate (%) = [(^{1}N_sample - ^{1}N_background) / (^{1}N_maize - ^{1}N_background)] × 100
where ^{1}N_sample is the ^{1}N value of soil with maize residues, ^{1}N_background is the ^{1}N value of bare soil control, and ^{1}N_maize is the ^{1}N value of added maize residues.

6. Data Processing and Analysis

Data were processed and plotted using Origin 8 software. SPSS 19.0 was used for ANOVA, with Duncan' s multiple comparison test at $P < 0.05$ significance level.

Results

1. Changes in Soil Total Nitrogen Content

Total nitrogen content changed significantly after maize residue addition in both fertility levels. The average TN contents of control LF and HF soils were (1.16 ± 0.03) g kg^{1} and (1.85 ± 0.02) g kg^{1} respectively. Residue addition significantly increased TN content, with greater enhancement in LF soil (increases of 9.88%, 10.21%, and 9.56% for roots, stems, and leaves respectively) compared to HF soil (increases of 5.81%, 4.13%, and 5.44%). No clear temporal patterns were observed during decomposition, and no significant differences existed among residue parts overall.

Dynamic changes of total nitrogen in different fertility soils added with ^{1}N-labeled maize residues

2. Changes in Soil Total Nitrogen ^{1}N Values

The ^{1}N values of control soils remained stable throughout incubation (data not shown). After residue addition, soil TN ^{1}N values increased significantly and showed greater fluctuations in root treatments compared to stem and leaf treatments during early incubation, stabilizing thereafter. In both fertility levels, ^{1}N values were generally higher in stem treatments than in root and leaf treatments. At 360 days, no significant differences existed among the three residue parts.

[Figure 1: see original paper] Dynamic changes of ^{1}N values in total nitrogen of different fertility brown earths added with ^{1}N-labeled maize residues

3. Changes in Contribution Rate of Exogenous New Nitrogen

The contribution rate of exogenous new nitrogen to soil TN, reflected by the proportion of maize-derived N, showed similar trends to ^{1}N values over time. Mean contribution rates were 9.09%, 9.87%, 8.62%, 5.55%, and 6.59% for LF soil, and 6.13% for HF soil. Root treatments showed greater fluctuations than stem and leaf treatments during early incubation. The contribution rate was significantly higher in LF soil than in HF soil. In LF soil, the contribution rates after root, stem, and leaf addition were 10.26%, 11.29%, and 8.49% respectively at 360 days, while in HF soil they were 6.13%, 6.74%, and 7.34%.

[Figure 2: see original paper] Dynamic changes of exogenous new nitrogen contribution rates to total nitrogen in different fertility soils added with ^{1}N-labeled maize residues

4. Dynamic Changes in Soil Microbial Biomass Nitrogen

Soil microbial biomass nitrogen increased gradually in early incubation, peaked at 28 days, then decreased. MBN was significantly higher in residue-amended soils than in controls. Over the entire incubation period, LF soil MBN increased by 0.83–0.98 times, while HF soil MBN increased by 0.87–1.56 times. Except for root treatments at certain times, MBN was consistently higher in HF than LF soils ($P < 0.05$). In HF soils, MBN increased significantly in early stages, peaking at 7–28 days, then declined and stabilized. In LF soils, MBN showed a trend of initial increase, slight decrease at 28 days, then increase again. The decline amplitudes were 32.98%, 11.75%, and 16.64% for root, stem, and leaf treatments respectively in LF soil, and 22.26%, 23.13%, and 16.81% in HF soil during 28–56 days.

[Figure 3: see original paper] Dynamic changes of microbial biomass nitrogen in different fertility soils added with ^{1}N-labeled maize residues

Discussion

1. Changes in Soil Total Nitrogen and ^{1}N Values

The addition of ^{1}N-labeled maize residues significantly increased soil TN content and ^{1}N values, consistent with previous studies. Our study further highlights the influence of fertility level and residue part. The more pronounced TN increase in LF soil indicates differential decomposition between fertility levels, affecting N supplementation. The ^{1}N fluctuations suggest that exogenous N entered the soil rapidly during early decomposition of labile compounds. The higher ^{1}N values in LF soil throughout incubation indicate greater N sequestration capacity, possibly due to higher clay content providing more surfaces for organo-mineral complexation. The overall pattern of stem > leaf > root in ^{1}N values reflects differences in biochemical composition, with stems having the lowest C/N ratio and roots containing more recalcitrant lignin. By 360 days,

differences among residue parts diminished, suggesting that long-term decomposition reduces initial compositional effects.

2. Changes in Exogenous New Nitrogen Contribution Rate

The contribution rate of residue N to soil TN was higher in LF than HF soil, reflecting the lower background TN in LF soil and greater relative impact of added residues. The increasing contribution over time indicates progressive N release from decomposing residues. The contrasting trends between fertility levels (stem > leaf > root in LF; leaf > stem > root in HF) likely reflect differences in microbial communities and priming effects. The stem' s high contribution rate in both soils suggests microbial communities are most responsive to stem N addition, possibly due to its favorable C/N ratio and labile carbon content.

3. Changes in Soil Microbial Biomass Nitrogen

Crop residue decomposition is a microbially-driven process that alters the soil microenvironment and nutrient availability. All residue parts stimulated microbial growth and N assimilation. The positive correlation between MBN and soil fertility was evident, with HF soils showing greater absolute MBN and stronger responses to residue addition. The earlier MBN peak in HF soils (7–28 days) compared to LF soils reflects greater microbial activity and N availability. The different temporal patterns between fertility levels may result from combined effects of soil fertility and microenvironment changes induced by residue addition. The 2-month period after residue incorporation represents the peak phase of MBN dynamics, with both fertility level and residue composition influencing the feedback mechanisms.

Conclusion

Low-fertility brown earth is more responsive to exogenous new nitrogen, showing greater increases in total nitrogen content. High-fertility brown earth exhibits more sensitive microbial biomass nitrogen responses to crop residue input and amplifies differences among residue parts. Maize stem and leaf return is more effective for increasing soil microbial biomass nitrogen. This study examined exogenous new nitrogen distribution in bulk soil; further research is needed on the fate and allocation mechanisms of different maize residue parts within soil aggregate fractions.

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Note: Figure translations are in progress. See original paper for figures.