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Origin of Nitrogen in Reforested Lignite-Rich Mine Soils Revealed by Stable Isotope Analysis

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Restoration of the nitrogen cycle is an important step in the recovery of an ecosystem after mining. Carbon and nitrogen in rehabilitated lignite containing mine soils can be derived from plant material as well as from lignite inherent to the parent substrate. We assessed the use elemental and stable carbon and nitrogen isotope measurements to trace the orgin of soil nitrogen and applied these techniques to elucidate the origin of mineral N in the soil and the soil solution. The conceptual approach of this study included physical fractionation in addition to sampling of vegetation and soil from a lignite-containing mine site rehabilitated in 1985 with *Pinus Nigra*. We studied the elemental and isotopic composition of bulk samples as well as isolated fractions and soil solution. Our data indicate that the stable carbon and nitrogen isotopic composition of the soil samples are the result of mixing between plant material and substrate inherent lignite. δ^{15}N isotopes may be used as indicators of nitrogen contribution from plants to solid samples as well as soil solution. N-isotope composition of ammonia shows low spatial and interannual variability, despite strong concentration changes. Plant-derived nitrogen contributes in higher amounts to the soil solution compared to the bulk mineral soil.

Introduction

Open-cast lignite mining in the Lusatian mining district results in large areas of overburden dumps containing various amounts of pyrite and geogenic organic matter (GOM) from Tertiary sediments. These areas are predominantly restored by afforestation with pine after initial amelioration by addition of CaO and NPK fertilization (*1*). During the early stage of mine soil development the element budgets of these sites are controlled by pyrite oxidation releasing large amounts of iron and sulfuric acid, inducing enhanced weathering of primary minerals and precipitation of secondary minerals (*2, 3*).

Restoring N cycling is one of the major goals of ecosystem recovery in environments disturbed by mining (*1*). The mine sites contain high amounts of geogenic organic carbon and nitrogen in the form of lignitic fragments, which may serve as nutrient sources for plant regrowth. It was shown, for example, that this GOM is decomposed by the microbial activity in mine soils even at the very early stages of soil development (*4, 5*). Lignite was also shown to contain high amounts of exchangeable NH_4^+—N (*6*), and the lignite and pyrite containing substrates may release NH_4^+—N into soil solution (*7*). Much of the nitrogen is leached as NH_4^+—N beneath the rooting zone from these sites (*2*). At young mine sites (<40 years), this output was found to be disconnected from input by litterfall or atmospheric deposition and could only be explained by nitrogen release from GOM (*8*). In the main rooting zone of these mine soils, N fluxes were found to be low, probably as a result of plant uptake (*2*). However, it is not clear to what extent plants are using the nitrogen released from lignitic fragments.

Stable isotopes are widely used to trace the origin of soil organic matter. The stable carbon isotope ratio (δ^{13}C) of plants is determined by the δ^{13}C of the source C as modified by photosynthetic discrimination against ^{13}C in leaves. During decomposition, soil microbes discriminate against the heavier isotope and as a consequence ^{13}C is usually enriched with soil depths (*9*). This discrimination is usually assumed to be of the same magnitude for different organic matter types, and therefore, the distinct δ^{13}C ratio of C3- and C4-type vegetation allows us to follow carbon turnover at sites of vegetation change (*10*). Even if the differences of δ^{13}C for C3-type organic matter are smaller, stable isotopes of carbon can be used to trace the input of vegetation-derived carbon at young sites on lignite mine dumps (*11, 12*).

The use of the stable nitrogen isotope ratio (δ^{15}N) to study the origin of nitrogen compounds is less appropriate in most natural soils as the relation between sources and sinks of δ^{15}N in a plant–soil system is influenced more by cycling of N than its sources (*13, 14*). Similarly to δ^{13}C, δ^{15}N may become enriched during microbial decomposition and increase with profile depth (*15*−*17*). However, in soils, which have an unusual source of δ^{15}N, the source can influence the δ^{15}N of soils and plants (*18*). One example for such an unusual δ^{15}N source is fertilizer N applied on agricultural (*19*) or forested ecosystems (*20*). For soils at rehabilitated lignite mining sites, we hypothesized that GOM may be such an unusual N source with possibly a distinct δ^{15}N signature compared to plant litter. We made this hypothesis, because a distinct δ^{13}C signature of GOM could be identified and used to trace lignite C contribution in soils (*11*). Moreover, the rehabilitated mine sites are young and show strong acid pH, which leads to relatively few processes with respect to N isotope fractionation. Therefore, it may be possible to study the origin of N and NH_4^+—N in mine soils by the analysis of their stable isotope composition.

In this study, we sampled pine needles, the litter layer, different soil depths, as well as the soil solution from a mine soil, which was rehabilitated in 1985. Bulk material and size fractions were analyzed for stable carbon and nitrogen isotope signatures. The aims of the study were to

- elucidate the potential of stable nitrogen isotopes to trace the origin of nitrogen in lignite containing mine soils.
- estimate plant-derived N contribution to soil N and soil solution N.

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TABLE 1. Elemental and Isotopic Composition of Plant Material, Litter Layers, Mineral Soil (<2 mm) and GOM

	C_{org}	N		$\delta^{13}C$	$\delta^{15}N$
	g kg^{-1}	g kg^{-1}	C/N	‰	‰
GOM					
lignite ss	182 ± 0	0.3 ± 0.0	60 ± 1	−22.5 ± 0.1	3.1 ± 0.1
xylite	541 ± 0	1.4 ± 0.0	386 ± 1	−23.7 ± 0.0	2.4 ± 0.1
aboveground plant litter					
green needles	481 ± 0	7.6 ± 0.0	63 ± 1	−26.0 ± 0.7	−2.9 ± 0.2
brown needles	490 ± 0	5.6 ± 0.1	88 ± 1	−27.2 ± 0.1	−3.1 ± 0.1
forest floor					
L	486.9 ± 0.2	9.3 ± 0.1	52	−27.4 ± 0.0	−4.67 ± 0.01
L/LF	361.4 ± 0.3	6.8 ± 0.1	53	−28.0 ± 0.1	−4.21 ± 0.01
F	263.5 ± 0.2	13.6 ± 1.1	19	−28.0 ± 0.1	−3.86 ± 0.30
H	92.4 ± 0.1	3.7 ± 1.1	25	−26.2 ± 0.2	−0.33 ± 0.00
mineral soil					
0−5 cm	60.0 ± 0.5	2.0 ± 0.9	30	−25.0 ± 0.0	0.57 ± 0.01
5−10 cm	51.0 ± 1.4	1.0 ± 0.8	51	−24.5 ± 0.1	1.34 ± 0.01
10−15 cm	59.0 ± 0.2	1.0 ± 0.1	59	−24.4 ± 0.0	1.60 ± 0.00
15−20 cm	53.6 ± 0.1	1.3 ± 0.1	41	−24.5 ± 0.0	1.86 ± 0.13
20−40 cm	56.7 ± 1.0	0.9 ± 0.1	63	−24.2 ± 0.1	2.18 ± 0.01
40−70 cm	31.1 ± 1.3	0.7 ± 0.1	44	−24.4 ± 0.1	1.97 ± 0.13
70−90 cm	27.3 ± 1.0	0.6 ± 0.2	46	−24.4 ± 0.1	1.59 ± 0.08
90−110 cm	25.0 ± 1.4	0.6 ± 0.1	42	−24.7 ± 0.0	2.32 ± 0.11

Materials and Methods

Study Site. The study was carried out at a rehabilitated mine site in the Lusatian mining district (Supporting Information (SI) Figure 1), in the eastern part of the Federal Republic of Germany (51°49′N, 14°28′E; 99 m above sea level). The mine spoil was afforested with Black pine (*Pinus nigra*) in 1985. The top 0–40 cm were ameliorated using high amounts of liming material (190 Mg CaO/ha) in form of powerplant ash (*21*). The soil (SI Figure 2) represents an initial stage of mine soil development. The parent substrate for soil development is sandy (64–73% of sand) Tertiary overburden sediment. After afforestation, plant-derived material accumulated at the soil surface and was incorporated into the mineral soil. Humic substances formed during the decomposition of plant litter could not be distinguished by macro−morphological observation from lignitic material inherent to the parent substrate. ^{14}C activity analysis suggested that geogenic organic carbon contributed to the forest floor and the 0–5 and 5–10 cm layers of the mineral soil (*22*).

Sampling and Sample Pretreatment. Three replicate samples were taken from pine needles attached to trees on September 2003 (green needles and brown needles), the forest floor horizons (L, F, H-layers), and the mineral soil in eight depth increments. In the L horizon of the forest floor, plant litter freshly fallen from the trees (in this case pine needles) can be found. The material in the F horizon is partly decomposed pine needles, which are not strongly altered in their overall morphology. The L/F horizon is a transition horizon between both. In the H horizon, completely humified material is present. Additionally, two different GOM types were sampled from the lignite seam. Lignite, ss, is of black color and may contain some mineral material, whereas xylite is buried wood which survived the coalification process. Lignite ss is the most common lignite type in the overburden material. All samples were airdried. The soil samples were divided into three size fractions (>7, 2–7, and <2 mm) by dry sieving to separate fine soil containing only lignitic dust from larger fragments of GOM. Stones and large roots were separated so the fragments >2 mm only consist of lignite. We used this fractionation approach in order to obtain fractions composed only of GOM. The soil solution was sampled using ceramic cups (P80 material) in three soil depths (20–40, 70–90, and 90–110 cm) with a continuously applied low pressure (<30 kPa). Three replicated ceramic cups were sampled in each depth (SI Figure 2) during January 2002 and February 2003. The soil solutions were filtered (0.45 μm) and afterward analyzed for NO_3 and NH_4concentration as well as $\delta^{15}N{-}NH_4$ (see below). NO_3 was analyzed with an ion chromatograph (Dionex120 IC) and NH_4 with a rapid flow analyzer (Perstorp Analytical 7050).

As we did not collect soil solution from the first 5 cm of the mineral soil (just below the forest floor) for technical reasons, we assessed the NH_4^+ concentration and $\delta^{15}N$ of NH_4^+ extracted during a batch experiment with fresh soil (1:8 soil/water were shaken for three hours and filtered through 0.45 μm filter). We present these data as mean and standard deviation ($n = 3$).

Elemental and Isotope Analysis. Organic carbon (OC) and nitrogen (N) contents were determined by the dry combustion method using a CHN elemental-analyzer (CHN NA 1500, Carlo Erba). Analytical precision was ±0.1 mg g^{-1} for OC and ±0.05 mg g^{-1} for N content.

Carbon and nitrogen isotope ratios of solid samples were determined with an elemental analyzer (EA, Eurovector) coupled to an isotope ratio mass spectrometer in continuous flow mode (Isoprime GV instruments). Prior to the determination of nitrogen isotope ratios of ammonium, NH_4 was collected using the ammonium diffusion technique (*23*). Under basic conditions, 150 μg of ammonium nitrogen was transformed into ammoniac. The ammoniac was kept on an acidified glass fiber filter (H_2SO_4) and transformed into ammonium sulfate. After one week of incubation, the filter was freeze-dried, put into tin cups, and thermally decomposed in the elemental analyzer (EA, Eurovector) coupled to an isotope ratio mass spectrometer in continuous flow mode (Isoprime GV instruments) and $\delta^{15}N$ of generated $N_2$was determined.

Carbon and nitrogen isotope ratios were defined using the delta notation:

$$\delta_{\text{sample}}\ (‰) = [(R_{\text{sample}} - R_{\text{standard}})/R_{\text{standard}}] \times 1000$$

where R is the isotopic ratio $^{13}C/^{12}C$ and $^{15}N/^{14}N$ for the sample and the reference gas. The isotopes ratios were calculated relative to Pee Dee Belemnite standard (PDB) for carbon

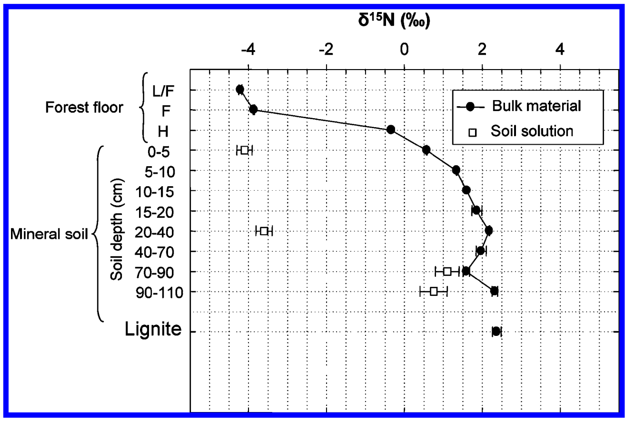

FIGURE 1. Stable nitrogen isotope values ($\delta^{15}N$) in the soil profile and soil solution ($\delta^{15}N-NH_4^+$) as well as in the precursor material of soil organic matter.

and relative to atmospheric N_2 for nitrogen (*24, 25*). Analytical precision for isotope measurements was ±0.1‰ for carbon and 0.2‰ for nitrogen isotopes.

Estimation of Plant Derived Nitrogen in Lignite Containing Mine Soils. We used the following equations to estimate plant-derived nitrogen contribution to soil by isotopic mass balance:

$$100\ \delta^{15}N_S = \delta^{15}N_P \times x + \delta^{15}N_L \times y$$

and

$$100 = x + y$$

where $\delta^{15}N_S$ is the N isotope signature of soil or soil solution, $\delta^{15}N_P$ is the isotope signature of plant-derived organic matter, $\delta^{15}N_L$ is the isotope signature of GOM, and x, y is the relative contribution of plant and GOM derived N, respectively.

To carry out this calculation, we hypothesized the ^{15}N signature of lignite ss to be the GOM-derived endmember because this is the most common lignite form occurring in soils. xylite does occur only occasionally within soil profiles. We took the ^{15}N signature of the litter layers (L and F) as the second plant-derived endmember to cover the range of possible isotopic fractionation occurring during decomposition of organic matter. With these data we calculated mean and standard deviation. This was assumed to be correct because highly humified material is not present at this young mine site, which has not yet reached the same state of humification of plant litter as mature sites (*26*).

Results and Discussion

Elemental and Isotopic Composition of Plant Material and GOM. The carbon content of litter samples ranged between 481 and 490 g kg^{-1} (Table 1). Nitrogen content was reduced in brown needles with respect to green needles by 2 g kg^{-1}, which led to an increase of the C/N ratio. The decrease in nitrogen corresponds to a nitrogen recycling by the tree, which is usually observed before the needles fall to the ground (*27*). This recycling was not accompanied by an isotopic fractionation, as illustrated by similar nitrogen isotope composition of green and brown needles (Table 1). In contrast, a depletion in ^{13}C was noticed in the brown needles as compared to the green ones. The two GOM samples were very different with regards to the carbon and nitrogen content and C/N ratio. Lignite ss had lower carbon content, but a C/N ratio which was much lower than for xylite material. Compared to the contrasting elemental data, the isotopic signatures of both GOM types were in a similar range. $\delta^{15}N$ was 2.4 and 3.1‰ for xylite and lignite ss, respectively; $\delta^{13}C$ was −23.7 and −22.5‰ (Table 1).

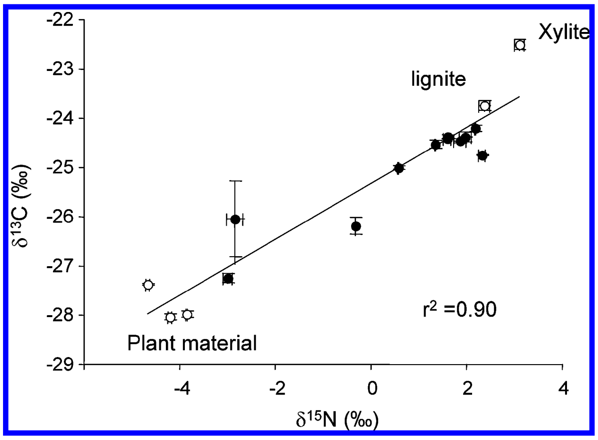

FIGURE 2. Relationship between stable carbon and nitrogen isotope values ($\delta^{15}N$, $\delta^{13}C$) in the soil profile, GOM and plant material from litter layers.

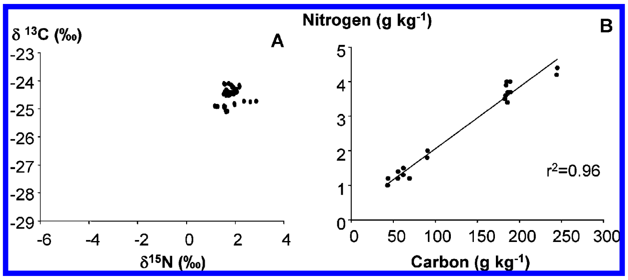

FIGURE 3. Relationship between stable carbon and nitrogen istotope values ($\delta^{15}N$, $\delta^{13}C$) as well as carbon and nitrogen content for GOM (particle size fractions >2 mm).

Strong variation in elemental composition of GOM may be related to the fact that lignite is a very diverse set of material (*28*). It is formed from different vegetation types, and as lignite, is a very early stage of coalification, the geochemical conditions were not intense enough to counterbalance the variation of the source material (*29*). In the Lusatian mining district, where GOM had been formed in the tidal region of an ancient sea, lignite ss may contain high mineral contributions. The $\delta^{13}C$ of GOM is similar to terrestrial plants and does not change significantly with age or maturity. It has been demonstrated that the variation of carbon isotope ratios in different coals is limited to a range of about 5‰ between $\delta^{13}C$-22 and -27‰ (*30*). The relatively large enrichment of $\delta^{13}C$ in GOM of 3–5‰ compared to the plants growing at the sites may not be explained solely by atmospheric $\delta^{13}C$ decrease during the last 150 years as a result of fossil fuel burning (*31*). Therefore, we suggest, that GOM enrichment in $\delta^{13}C$ may be partly a result of the intensive decomposition process prior to geochemical processes during the early phase of diagenesis (*30*). Microbial respiration discriminates against ^{13}C, leading to an enrichment of microbial products compared to plant material (*32, 33*). This enrichment is somewhat counterbalanced by the rapid degradation of ^{13}C enriched material such as cellulose and selective preservation of recalcitrant plant litter compounds, such as lipids and lignin, which are depleted in ^{13}C (*34*). However, conditions in the tidal area may have led to extensive degradation of the most stable compounds (*11*). Similarly to ^{13}C, $\delta^{15}N$ of GOM does not change during coalification even if the C/N ratio increases dramatically (*35*). $\delta^{15}N$ of plant material usually shows negative values (*36*). The enrichment of ^{15}N of GOM may be explained by isotopic fractionation during nitrification/denitrification reaction occurring prior to the coalification process. Given the differences of stable isotope ratios of plant material and GOM, we hypothesized that it may be possible to trace the origin of nitrogen in different soil depths.

TABLE 2. pH, NH_4^+ Concentrations and $\delta^{15}N$ of Soil Depth of Soil Solutions in 2002 and 2003

		2002			2003	
soil depth	pH	NH_4^+		pH	NH_4^+	
cm	mg L^{-1}	‰	$\delta^{15}N$	mg L^{-1}	‰	$\delta^{15}N$
0–5	5.1 ± 1.3	0.8 ± 0.2	−4.01 ± 0.3	4.8 ± 0.6	0.9 ± 0.1	−3.88 ± 0.5
20–40	4.8 ± 1.5	0.5 ± 0.3	−3.67 ± 0.2	4.3 ± 1.3	0.6 ± 0.2	−3.69 ± 0.3
70−90	2.6 ± 0.3	16.8 ± 3.1	0.92 ± 0.2	2.5 ± 0.4	44.2 ± 3.1	1.11 ± 0.2
90−110	2.5 ± 0.4	24.6 ± 2.7	0.75 ± 0.3	2.4 ± 0.2	62.7 ± 4.2	0.65 ± 0.2

[a] Note that all nitrate concentrations are below the detection limit <0.5 mg L^{-1}). ± SD; $n = 3$.

Elemental and Isotopic Composition of Litter Layers and Mineral Soil. The carbon content of the forest floor horizons ranged between 486 and 92 g kg^{-1} (Table 1). The C/N ratio decreased from the L to the F layer from 52 to 19 and afterward increased slightly in the H horizon (Table 1). The stable carbon isotope ratio ($\delta^{13}C$) ranged in the litter layers from −26 to −28‰. L and F horizons were depleted in ^{13}C with regards to the aboveground plant material. In the H horizon, this trend is inversed and organic matter becomes further enriched in ^{13}C in the mineral soil horizons, where $\delta^{13}C$ ranges between −24 and −25‰ (Table 1). These values are similar to those found in a number of forest soils (*37*). In the lignite-containing mine soils, the ^{13}C enrichment of the mineral soil was around 2‰ higher than that recorded for other forest soils. At the same time, the carbon content, which ranged between 25 and 60 g kg^{-1} in the mineral soil, is much higher than recorded for other forest soils (Table 1). The nitrogen content of the forest floor ranged between 3.7 and 13.6 g kg^{-1} and the $\delta^{15}N$ ratio was depleted in the L and L/F and F layer with regards to the plant material. In the H horizon, organic matter is enriched in ^{15}N. The litter layers of the forest floor are eventually incorporated into the mineral soil to form soil organic matter. The isotopic differences observed between plant litter sampled on the tree and the litter layers of the forest floor clearly show that the use of the isotopic signature of plants may not be appropriate as an endmember for the calculation of the contribution of plant-derived organic matter in lignite rich mine soils. Such a calculation has to take into account organic matter transformations occurring in the course of biodegradation. Therefore, we decided to take as endmembers the isotopic signature of the L (partly degraded plant material) and the F (completely humified plant material) horizon to calculate a range of plant litter contribution (see Material and Methods). We could not use the isotopic ratio of the H horizon as endmember, as this horizon contains GOM in form of lignite dust (*26*).

The $\delta^{15}N$ of the mineral soil ranged between 0.5 and 2.3‰ and increased with soil depth (Table 1; Figure 1). A ^{15}N enrichment of organic matter with increasing soil depth is a usual pattern (*16, 17*). Increasing $\delta^{15}N$ was found to be correlated with decreasing C/N ratio, and thus increasing the degree of decomposition, consistent with N loss through volatilization and/or nitrate leaching from net mineralization (*38, 39*). In lignite-rich mine soils, such a correlation could not be observed. In contrast, $\delta^{15}N$ ratio was correlated to the $\delta^{13}C$ ratio (Figure 2). A first-order linear correlation between $\delta^{13}C$ and $\delta^{15}N$ is often observed in marine sediments and interpreted as a mixing trend between terrestrial and marine sources of organic matter providing depleted and enriched isotope ratios, respectively (*40, 41*). For lignite-rich mine soils, we hypothesized that the mixture of plant-derived organic matter and GOM is leading to the correlation of $\delta^{13}C$ and $\delta^{15}N$ in the mineral soil.

To strengthen this hypothesis, we tried to obtain similar relationships for the 2–7 mm and >7 mm fraction of the mine soil. These two fraction contain GOM as the only organic matter type. They showed a contrasting behavior compared to the rest of the mineral soil. A correlation between $\delta^{13}C$ and $\delta^{15}N$ was thus absent for GOM (Figure 3A) despite a strong coupling of C and N in the two fractions (Figure 3B). These data may be interpreted by the presence of only one organic matter type, i.e. GOM, with different stages of transformation. They also support the evidence that isotopes may be used as tracers of the two organic matter types present in the <2 mm fraction of lignite-rich mine soils (*11*). We suggest, that this is not only true for carbon but may also concern nitrogen.

N-Isotope Composition of the Soil Solution. Nitrogen in the soil solution was almost exclusively composed of ammonium. Data of NH_4^+ concentration as well as $\delta^{15}N{-}NH_4$ of soil solution sampled in two consecutive years are presented in Table 2. NH_4^+ concentrations ranged between 0.5 and 62.7 mg L^{-1}. The NH_4 concentrations were much lower in 0–5 and 20–40 cm soil depth compared to 70–90 and 90–110 cm. They were more than doubled in 70–90 cm and 90–100 cm depth in 2003 compared to 2002. The $\delta^{15}N{-}NH_4$ recorded for the two years ranged between −4.01 and 1.11‰. In 0–5 cm and 20–40 cm of the soil, negative $\delta^{15}N$ were recorded, whereas below this depth, $\delta^{15}N$ was positive. At the two sampling dates similar $\delta^{15}N{-}NH_4$ was recorded, despite the higher NH_4 concentrations in some soil layers. Since various N transformations occur simultaneously in soil, variations in $\delta^{15}N{-}NH_4$ should depend on the kind and extent of reactions in which NH_4 participates. Possible N processes occurring in soil with the extent of N fractionation in the studied ecosystem can be found in SI Figure 3. A striking observation was the isotopic fractionation occurring between pine needles attached on the tree and pine needles collected from the L layer of the forest floor (Table 1). This fractionation led to depletion of ^{15}N isotope. Further down the forest floor, decomposition processes resulted in ^{15}N enrichment (Table 1, SI Figure 3). Kinetic isotopic fractionation during N transformation always results in ^{15}N enrichment of the substrate because the molecules bearing the lighter isotope would react somewhat faster than those of the heavier isotope (*42*). In soils, nitrification is the most important process which could lead to $\delta^{15}N{-}NH_4$ enrichment (*16*). But due to the low pH (Table 2) this process is quite null in young lignite-containing mine soils (*8*). The nitrate concentrations in the present study were below the detection limit i.e. <0.5 mg L^{-1}. Microbial immobilization, the other process affecting nitrogen transformations in soils produces less significant fractionation compared to nitrification processes (*43*). In young lignite-rich mine soils under pine, biological activity is low and this process affect N from both organic matter types: GOM and plant-derived organic matter (*5*). Therefore, $\delta^{15}N{-}NH_4$ in the soil solution of mine soils reflects the isotopic composition of the nitrogen in the source material, i.e. degraded plant litter and GOM. Our data thus indicate that NH_4 in 0–5 and 15–20 cm of soil depth may be derived from degraded plant litter, whereas below this depth it may originate from GOM present in the mineral matrix (Figure 1).

Plant Derived Nitrogen in Lignite Containing Mine Soils. Isotopic mass balance calculations showed that the highest contribution of plant derived nitrogen to the mineral soil

TABLE 3. Nitrogen Contribution from Plants in Mineral Soil and Soil Solution Calculated by Isotopic Mass Balance

soil depth	soil-N	solution—NH_4
cm	% of total N	% of total N
H horizon	40.8 ± 3.5	n.d.
0–5 cm	27.2 ± 2.3	94.1 ± 8.1
5–10 cm	15.5 ± 1.3	n.d
10–15 cm	11.6 ± 1.0	n.d
15–20 cm	7.7 ± 0.6	8.17 ± 0.7
20–40 cm	2.8 ± 0.2	n.d
40–70 cm	6.0 ± 0.5	n.d
70–90 cm	11.8 ± 1.0	21.9 ± 1.9
90–110 cm	0.7 ± 0.1	27.8 ± 2.4

[a] Note that the value of the % solution—NH_4 of the 5 cm soil depth is from the batch experiment.

can be observed in the top 0–10 cm, amounting to values between 15 and 27% of the total nitrogen present at this depth (Table 3). Below, the contribution of plant derived nitrogen is between 2 and 11%, whereas it is almost absent in 110 cm depth. ^{14}C activity measurements indicated a plant-derived C contribution of 59 and 16% of total C in the H horizon of the forest floor and the 0–5 cm layer of the mineral soil, respectively (*26*). GOM contribution to the H horizon is due to deposition of lignite-rich airborne contamination (*44*). In 100 cm depth, similar to plant-derived N, contribution of plant-derived C was absent (*26*). Using these data, we calculated a C/N ratio of plant-derived SOM between 24 and 17 in the H and 0–5 cm layers, which are values commonly observed for *Pinus* ecosystems (*45*) and indicate that the use of stable N isotopes may be adequate to calculate the contribution of plant-derived N.

NH_4—N sampled from soil solution is almost exclusively derived from plant material in 0–5 cm depth (Table 3). In the 20–40 cm depth, which corresponds to a good portion of the rooting zone of the plants, the lowest contribution of plant-derived N to NH_4—N was recorded. This may be explained either by preferential uptake of plant-derived N or by isotopic fractionation of nitrogen during its uptake. Even in the lowest soil horizons, plant-derived N still represents more than 20% of soil solution NH_4—N despite the absence of plant-derived N in the solid phase.

Acknowledgments

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Supporting Information Available

A map of the study area (SI Figure 1), a picture of the soil profile and sampled horizons (SI Figure 2), and a figure with possible nitrogen processes and possible fractionation associated with these processes in lignite-rich mine soils (SI Figure 3). This material is available free of charge via the Internet at http://pubs.acs.org.

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