Gross N turnover and soil solution chemistry as influenced by fluctuations of soil water potential and water table in a Podzol and a fen soil

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Yao-Te Chen

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Action dean:	Prof. Dr. Beate Lohnert
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Prof. Dr. Egbert Matzner	1st reviwer
Prof. Dr. Gerhard Gebauer	2nd reviewer
Prof. Dr. Bernd Huwe	Chairman
Prof. Dr. Stefan Peiffer	
Prof. Dr. Harold L. Drake	

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Summary

Given the climate scenarios, the higher frequency of drying/rewetting cycles of soils in the future can be expected. These changes of the meteorological conditions likely result in an increasing frequent and intensive drought periods in summer, causing irregular and extreme drought stress in forest soils or a drawdown of water table in wetland ecosystems, which may influence the turnover of nutrients in soils to a larger extend than previously thought.

The question arises how these climate changes will influence N and C turnover in forest and fen soils. A growing number of laboratory studies on drying/rewetting of soils have been published during past decades, but many studies used either disturbed soil samples or intact soil cores in laboratory. Although soil drying is a frequent phenomenon in the field, the long-term effects of drying/rewetting and irrigation on *in situ* fluxes and concentrations of solutes in forest and fen soils are unclear. Several studies have investigated the influence of soil water content on net N turnover rather than gross rates. Net ammonification and nitrification include two major processes: gross ammonification and gross nitrification on the one side and microbial immobilization on the other side. To identify the response of specific processes to soil drying, gross rates need to be measured.

This thesis focused on the impact of changing water potential or water table level on gross N turnover rates and soil solution chemistry in two different ecosystems in South-Eastern Germany.

In a Norway spruce forest, the effects of decreasing water potential and prolonged periods of summer drought on soil gross N turnover were investigated by laboratory and field experiments. Soil solutions and throughfall were collected and the cumulative *in situ* fluxes of DIN, DON and DOC with forest floor percolates were calculated. In a minerotrophic fen, we studied the response of N and C mineralization and soil solution chemistry to water table fluctuations in a laboratory experiment. In the field, we collected the soil pore water in 3 depths to clarify the long-term effects of water table level on the concentrations of solutes.

Homogenized soil samples of the Oi+Oe, Oa and EA horizons were taken and adjusted to 6 different water potentials in the laboratory. In the field experiment, throughfall exclusion and irrigation plots were established to simulate different precipitation patterns of a dry and wet growing season. Gross N turnover rates were determined in undisturbed soil cores from Oi+Oe and Oa+EA horizons during the drying period and after rewetting.

Soil drying decreased gross ammonification rates in the O horizon. The lowest rates were found at the throughfall exclusion plots but the differences to the irrigation and control plots were not statistically significant. A substantial ammonification rate of 14 mg N kg^{-1} soil day^{-1} was observed at 3.2 MPa (pF 4.5). The laboratory study showed that gross nitrification decreased with decreasing water potential and was more sensitive to drying than ammonification in the Oa horizon; however, this was not found in the field experiment. The latter might result from the low rates and huge spatial variation, indicating the difference between disturbed samples and intact soil cores. No rewetting pulse of gross ammonification was observed, probably due to its short duration or due to the slow changes of the water potential during the natural rewetting. Although the *in situ* fluxes of DIN increased at the throughfall exclusion plots after rewetting, the cumulative DIN flux at the throughfall exclusion plots did not significantly exceed that at the control plots. The lowest fluxes of DON and DOC were observed at the throughfall exclusion plots because of the reduction of input with throughfall. In the studies presented here, extended drought periods caused a reduction of gross N turnover in forest soils but gross ammonification continued at considerable rates at low water potential. The hypothesis of increased N turnover and fluxes of DIN, DON and DOC as a consequence of drying/rewetting was not confirmed.

In the fen site, undisturbed soil cores were taken and divided to two treatments of water table: permanently flooded and fluctuated. The later was subjected to flooding, drawdown and re-flooding. The permanently flooding enhanced gross ammonification after a lag phase of about 30 days while CO_2 emissions were constantly low. The water table drawdown also increased gross ammonification, but again after a lag phase of about 30 days. The first peak of CO_2 emissions appeared immediately after water table drawdown, followed by a decrease and a second peak. The ratio of CO_2 emission/gross ammonification were close to 2 under anoxic condition which seems to be caused by fast N turnover in the microbial biomass-N pool and low rates of CO_2 production. The changes induced by water table drawdown on the N and C turnover were found reversible after re-flooding. Drainage increases SO_4^{2-} but decrease Fe, DON and DOC concentrations and *vice versa* when the soils were flooded. Release of DON and DOC was inhibited by increasing SO_4^{2-} concentrations. Under field conditions, neither drainage nor flooding had an effect on dissolved inorganic N due to the low concentration, indicating the rapid consumption of mineralized N in the field. In the absence of plant uptake and runoff in the laboratory experiment, however, NH_4^+ increased during the flooding period.

Soil desiccation affects the upper soil layers with largest rates of N turnover. While gross N turnover is reduced by soil desiccation, a substantial rate of ammonification was observed even at low water potentials. Nitrification was found more sensitive to desiccation than ammonification which might change the NH_4/NO_3 ratio of available N under dry conditions. Rewetting of dry soil does not induce a pulse of N turnover and fluxes of DIN, DON and DOC. Overall, an increasing frequency of drying/rewetting cycles seem to have only moderate effect on the N turnover and on N solute fluxes in forest soils.

Fluctuations of water table play an important role for the organic matter mineralization, soil solution chemistry and inorganic N availability in minerotrophic fen soils. Acidification by oxidation of S to SO_4^{2-} can be expected after water table drawdown, causing inhibition of DON and DOC release. The effect of drainage and flooding on gross mineralization and solute concentrations is reversible within a month period. The effect of changing water table regime on N and C turnover in fen soils seems to depend largely on the time scale of the fluctuations. Short term fluctuations at a daily scale will have little effect on N turnover as compared to longer term changes on a monthly scale, while short term changes seem to trigger C losses by CO_2.

1. Introduction

1.1 Motivation

Climate models predict less precipitation in the future in Europe during the next decades, whereas other regions in the earth will likely receive more precipitation (IPCC, 2007). Extreme meteorological events like drying and subsequent intensive rainfalls affect many biological, chemical and physical processes in soil (Schimel et al., 2007), but little is known about the relevance of these events for the gross N turnover and solutes in soils. Hence, there is a need to improve our knowledge about the effects of changing precipitation pattern on dynamics of soil N cycling and soil solution chemistry in temperate ecosystems. Research on this topic may contribute to identify underlying processes and mechanisms of these environmental changes.

1.2 Soil water potential

Water potential is an essential concept in pedogenic studies, which is widely accepted and applied for quantifying the energy state of soil water (Jury and Horton, 2004). Water potential is the potential energy in a point of system relative to free water, without external forces acting on it, as a reference or standard state. The water potential of pure water is given in zero. Hence, the intensity of soil water potential implies the amount of energy which is required to remove per unit quantity of water isothermally from one location or state to another in soil. Soil water flows from an area of higher water potential to an area within a lower water potential. Water potential is usually assigned in unit of pressure Pa (Pascal) or the negative logarithm value of hPa which is the pF.

The total water potential is composed of some components identifiable with the forces that retain or act on the water and affect its energy state (Parr et al., 1985). These components include matric potential, osmotic potential, gravitational potential, pressure potential and overburden potential. The matric and osmotic components contribute more significantly to the soil water potential than others and exert a greater effect on water flow and availability. Although the water potential in dry soil is dominated by matric forces, there is also an osmotic component, since the soil solution contains a wide variety of dissolved compounds (Stark and Firestone, 1995).The osmotic potential is significant in saline soils and certain soil

amended with organic wastes or fertilizer.

Soil water potential rather than water contents should be used to compare the effect of water availability on microbes and turnover of N in soil (Davidson et al., 1998; Gleeson et al., 2008). Hydrophobicity of soil considerably slows down the increase in water potential following rewetting (Doerr et al., 2007), may inhibit the recovery of soil microorganisms during rewetting and thus might prevent the rewetting effect (Mataix-Solera et al., 2007). A slow increase in water potential gives microorganisms more time to equilibrate with their environment and to restore their metabolism including re-assimilation of solutes (Schimel et al., 2007). Since the water potential influences growth and survival of the soil microbes, a shift of soil microbial community can be expected when the soil drying comes to extreme by climate change (Bliss et al., 2004).

1.3 Nitrogen cycle in soils

N in soil mainly exists in organic form. Only a small part of mineralized N contributes to the inorganic N pool, and N cycle processes in soil are controlled by the interactions of microbes and surrounding environment (Ambus et al., 1992; Bechtold and Naiman, 2006; Jamieson et al., 1998; Zak and Grigar, 1991). The balances between soil processes, mainly N_2 fixation, ammonification, nitrification, immobilization, dissimilatory reduction and denitrification which determine cooperatively the net availability of inorganic N in soil (Fig. 1).

N mineralization is an enzymatic process by which the organic N compound is liberated as inorganic N into soil solution, including ammonification and heterotrophic nitrification. Ammonification denotes the process by which soil organic N is transformed to NH_4^+ as a final product. NH_4^+ is either utilized by living organisms to sustain their N requirement or for NO_3^- production or may just accumulate in soil. Nitrification is the microbial oxidation of reduced forms of N which is performed by nitrifiers. Besides the oxidation of NH_4^+ to NO_3^- by autotrophic nitrification, production of nitrate directly from organic N can occur by fungi and heterotrophic bacteria.

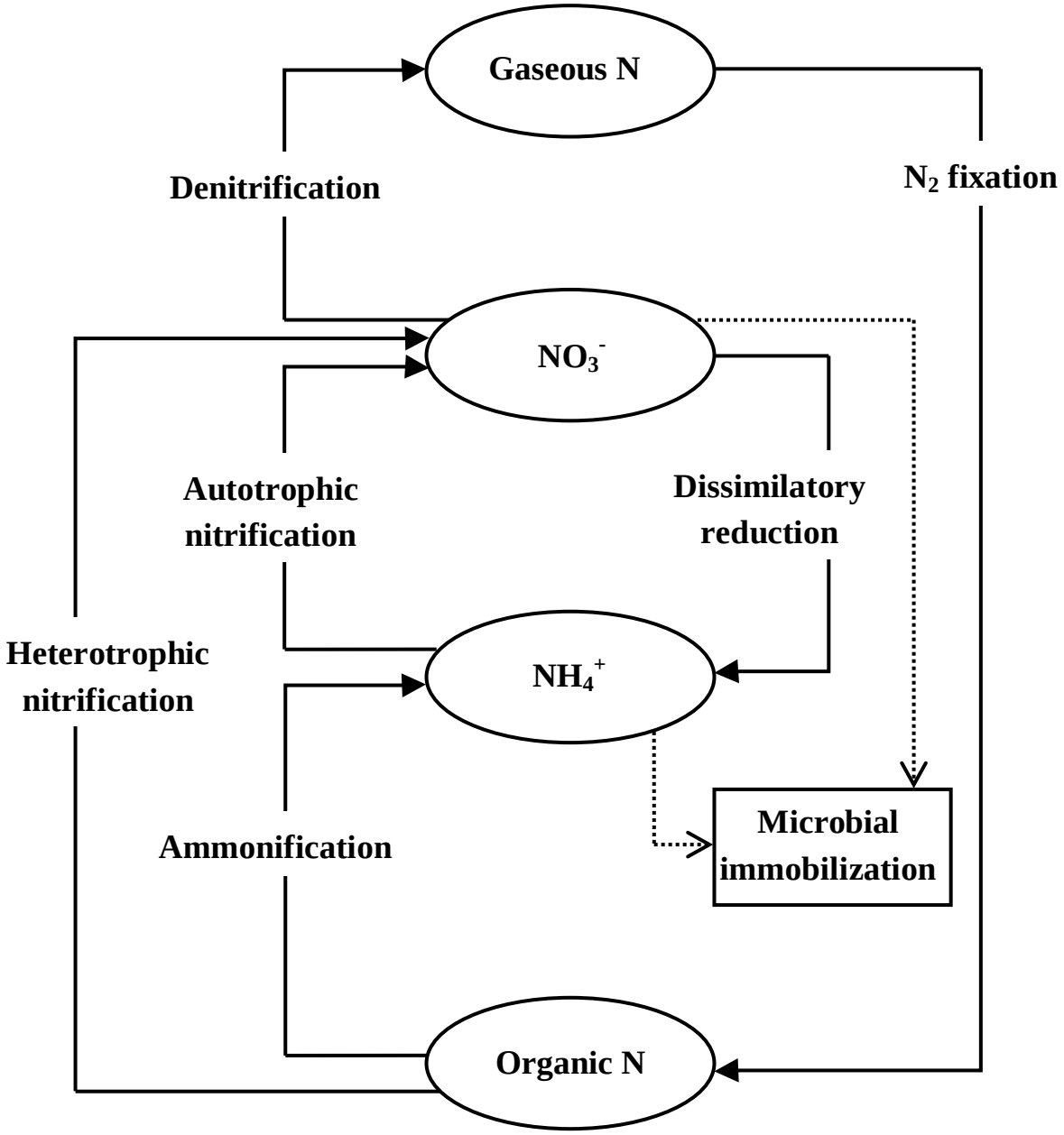

Fig. 1 Schematic representation of the processes in the soil N cycle.

Utilization of NH_4^+ and NO_3^- by microorganism and converted into biomass is commonly named immobilization. Uptake of NH_4^+ and NO_3^- by plants, the inorganic N is called assimilation.

Microbes seem to be the main consumers of NH_4^+ and NO_3^- even when plants are growing rapidly which results from the ubiquitous distribution of microbes in the soil (Schimel et al., 1989). The energetic advantages of using a reduced form of N for microbial growth may favor immobilization of NH_4^+ over NO_3^- (Corre and Lamersdorf, 2004; Schimel et al., 1989). Greater availability of NH_4^+ is likely to decrease the immobilization of NO_3^- by microbes. When NH_4^+ and NO_3^- are both available in soil, microorganism immobilize first or exclusively the NH_4^+ pool, NO_3^- will be immobilized after NH_4^+ is limited or unavailable (Cayuela et al., 2009; Davidson et al., 1992; Recous et al., 1990; Templer et al., 2008).

Dissimilatory nitrate reduction to ammonium (DNRA) is one of the microbial activities that enlarge the NH_4^+ pool (Brüggemann et al., 2005) and can be a fast process in N turnover in soil (Pandey et al., 2009; Yin et al., 2002). This pathway becomes dominate when NO_3^- concentrations are elevated in aerobic soils (Pett-Ridge et al., 2006). This reaction can be the predominant NO_3^- consumption process in an ecosystem where N availability is low and rainfall rates are high.

Denitrification is an anaerobic process by which NO_3^- is reduced to the N gases NO, N_2O and N_2 and emitted to the atmosphere (Khalil et al., 2004; Kresović et al., 2009; Liu et al., 2007). Denitrification can be easily stimulated in an aerobic soil by removing O_2 and is inhibited by drying or aerating (Aulakh et al., 2000). The most important factor controlling the denitrification was soil moisture, followed by soil temperature and NO_3^- concentration (Pinay et al., 2007).

The dynamics of the inorganic N pool in soils is therefore the outcome of the interaction between production and consumption. Gross ammonification is the total production of NH_4^+ and gross nitrification is the total production of NO_3^-. Any consumption of NH_4^+ and NO_3^- can reduce the pools and lead to low net rates. Several authors demonstrated that net N turnover rates underestimate gross rates in soils (Campbell and Gower, 2000; Verchot et al., 2001; Zaman and Chang, 2004) and the relationship between net and gross rates is poor (Burton et al., 2007; Corre and Lamersdorf, 2004, Stark and Hart, 1997).

1.4 Effect of soil drying/rewetting on N turnover

Soil drying changes the physical structure of soil, induces hydrophobicity (Doerr et al., 2007), physiological dehydration stress for microbial communities (Schimel et al., 2007) and limitation of substrates supply for microorganisms (Ford et al., 2007). The hydrophobicity of soils is likely to be an important mechanism that particularly diminishes soil N cycling. Cumulative mineralization rates theoretically decrease with increasing duration and intensity of drying (Borken and Matzner, 2009; Hentschel et al., 2007; Mikha et al., 2005). Thus, soil with thick organic horizons may prolong the period of reduced net N mineralization (Tietema et al., 1992 - Plant and soil). Nitrification seems to be even more sensitive to drought stress than ammonification (Hentschel et al., 2007; Xiang et al., 2008). Rainfall intensity might have only a limited effect on N mineralization while the duration of the rewetting period will influence the cumulative mineralization rates (Borken and Matzner, 2009).

In most cases, higher water content favors N turnover rates in soils (Matejek et al., 2008; Nishio et al., 1985; Yan et al., 2009). N turnover rates were lower in the dry season compared to the wet season (Gelfand and Yakir, 2008). Rewetting of drought soil could cause a pulse of net ammonification and nitrification (Birch 1958, Ford et al., 2007; McIntyre et al., 2009; Xiang et al., 2008). Hence, the highest N turnover rate was observed during the transition period from dry to wet season in forests (Breuer et al., 2002; Kiese et al., 2002; Yan et al., 2008). This can be due to several reasons: Drought stress of microorganisms leads to an accumulation of substrates in soil (Breuer et al., 2002; Kiese et al., 2002), which is easily available for the surviving microorganisms after rewetting (Gelfand and Yakir, 2008; Mikha et al., 2005). Additionally, an increase of substrate availability can take place by desorption from the soil matrix (Seneviratne and Wild, 1985) as well as by breakdown of soil aggregates during drought and the following rewetting, exposing physically protected organic material and NH_4^+ (Adu and Oades, 1978; Lundquist et al., 1999). The size of N mineralization pulses upon rewetting increased with the frequency of drying and rewetting cycles (Xiang et al., 2008), although this was not always seen (Gleeson et al., 2008; Hentschel et al., 2007). In fact, rewetting of dry soils increased the N mineralization rates but the observed rates were similar to the control level or only exceed the control level for few days (Muhr et al., 2010). In forest stands, simulated summer droughts and subsequent wetting did even not induce a NO_3^- pulse, suggesting that nitrification was not severely stimulated by rewetting (Tietema et al., 1997).

1.5 Effects of soil drying/rewetting on DON and DOC in forest soil

Production of dissolved organic N (DON) and C (DOC) may play an important role in many soil processes and in the turnover of organic matter in terrestrial ecosystems. The dissolved organic matter (DOM) originates from plant litter, soil humus, microorganisms and root exudates. The main source of DOM in forest soils is the forest floor and the DOC infiltration into the mineral soil represents a significant contribution to the soil C cycle and to the C pool in deeper soil horizons (Kalbitz and Kaiser, 2008; Michalzik et al., 2001). Xiang et al. (2008) showed that drying/rewetting cycles caused an increase of DOC release of grassland soils. Lundquist et al. (1999) suggested that several processes could increase DOC after soil drying/rewetting: (i) reduced microbial utilization of DOC in dry periods, (ii) enhanced turnover of microbial biomass after rewetting and (iii) drying/rewetting cycles disrupted soil aggregates thereby making previously stored C more available as DOC.

1.6 Effects of water table fluctuations on fen soil

N turnover in wetland soils is thought to be highly sensitive to fluctuations of water table and O_2 supply (Pal et al., 2010). Nitrification under anaerobic conditions is generally low (Bayley et al., 2005; Bowden 1986; Hefting et al., 2004; Neil, 1995), while ammonification can occur under both aerobic and anaerobic conditions (Hefting et al., 2004; Pinay et al., 2002). Since only 25% of NH_4^+ originating from ammonification was nitrified and up to 80% of NO_3^- was denitrified, ammonification provides the major inorganic N source in peatlands (Ambus et al., 1992). Soil aeration associated with water table drawdown can lead to higher N mineralization and increased inorganic N content in wetlands (Keller et al., 2004; Kieckbusch and Schrautzer, 2007; Venterink et al., 2002). Many studies have shown that a drawdown of water table increases the O_2 penetration and the CO_2 emissions in peatland soils (Danevčič et al., 2010; Oechel et al., 1998; Silvola et al., 1996). In contrast, Knorr et al. (2008a) and Muhr et al. (2011) reported no changes of CO_2 emissions from a minerotrophic fen after water table drawdown.

The concentrations of DON and DOC are especially important for surface waters draining from peatlands. However, the response of DON and DOC to water table fluctuations is debated and variable between the sites (Strack et al., 2008). Decreasing concentrations have been observed after water table drawdown (Clark et al., 2005; Fenner et al., 2005; Scott et al.,

1998) while others found increasing concentrations (Driscoll et al., 1989; Tipping et al., 1999) or no response (Blodau et al., 2004).

Water table drawdown in peat soils resulted in the production of SO_4^{2-} because of the oxidation of reduced S, causing episodic acidification of soil pore water (Clark et al., 2005; Scott et al., 1998). Even a small water table drawdown of 10 cm was sufficient to promote the oxidation of reduced S to SO_4^{2-} (Schiff et al., 2005). After rewetting or flooding, electron acceptors were consumed subsequent to depletion of oxygen (Peters and Conrad, 1996), resulting in reduction of SO_4^{2-}.

The level of the water table influences the concentration of Fe largely. When the peat soils become aerated, dissolved Fe^{2+} is re-oxidized to Fe^{3+} (Knorr and Blodau, 2009). The authors found that Fe^{2+} concentration of peat soils decreased to around zero during 50 days of drainage with water table at 0.55 m below the surface and increased to >100µmol L^{-1} within 2 weeks after rewetting. With a water table at 0.12 m below the surface for 70 days, a maximum concentration 5000 µmol L^{-1} of Fe^{2+} was detected at 0.1 m depth.

2. Objectives of this study

To address the uncertainties in current understanding of gross N turnover under extreme meteorological conditions and to clarify the effects of soil drying/rewetting and irrigation on soil solution chemistry, this study conducted laboratory and field experiments in a forest (study 1 and 2) and a fen site (study 3). The hypotheses were:

Study 1: (1) Gross N turnover is more sensitive to drying in the Oa and EA horizon as compared to the uppermost Oi+Oe horizon. (2) Gross nitrification is more sensitive to drying than gross ammonification in a forest soil.

Study 2: (1) Enhanced soil drying leads to a decrease of gross N turnover and natural rewetting causes a pulse of gross N turnover and DIN fluxes in forest soil. (2) Soil drying/rewetting increases and irrigation decreases the *in situ* fluxes of DON and DOC.

Study 3: (1) Water table drawdown in fen soils increases the mineralization of N and C but reduces the concentrations of DON and DOC. (2) The temporal response of gross N turnover and CO_2 emissions to water table drawdown is similar. (3) The changes induced by water table drawdown are reversible after re-flooding.

3. Materials and methods

3.1 Site description

3.1.1 Forest: Coulissenhieb II

The forest site is a 140-year-old Norway spruce forest (*Picea abies* L.), located in the Lehstenbach catchment (4.2 km^2) in the Fichtelgebirge mountains (870 m a.s.l.), Germany (58°08'N, 11°52'E). Mean annual precipitation is 1160 mm and mean annual air temperature is 5.3 °C (Foken, 2003). The soil has a sandy to loamy texture and is classified as Haplic Podzol according to the FAO soil classification (IUSS, 2006). The well stratified, mor-like forest floor of about 10 cm depth comprises Oi, Oe and Oa horizons. The forest floor is almost completely covered by ground vegetation, mainly *Deschampsia flexuosa* and *Calamagrostis villosa*. The C and N contents of the Oi horizon are 46% and 1.7%, of the Oe horizon 42% and 1.8%, of the Oa horizon 21% and 1.1% and of the EA horizon 8.3% and 0.4%. The pH($CaCl_2$) of the Oa is 3.3 and of the EA is 3.4. C and N stocks of the forest floor (Oi + Oe + Oa) are 5.0 kg C m^{-2} and 0.25 kg N m^{-2}, and in the EA horizon 2.4 kg C m^{-2} and 0.12 kg N m^{-2} (Schulze et al., 2009).

3.1.2 Fen: Schlöppnerbrunnen

This minerotrophic fen is located in the Lehstenbach catchment (4.5 km^2, Fichtelgebirge, northeastern Bavaria, Germany, 58°08'N, 11°51'E). Mean annual precipitation is 1020 mm and mean annual temperature is 6.3 °C (Knorr et al., 2009). The peat thickness ranges from 30 to 120 cm. The C and N contents of the top 10 cm are 31.1% and 1.8%. Bulk density is 0.29 g cm^{-3} and porosity is 85.5%. The soil is moderately acidic (pH 3.5 to 5.5) and rich in iron and sulfur (Goldberg et al., 2008; Knorr et al., 2008b; Paul et al., 2006). The water table level at the field site fluctuates from +0.5 cm at water saturation to -50 cm under summer drought conditions. The vegetation of the fen site comprises mainly *Nardus stricta, Agrostis sp.*,

Molinia coerulea, *Eriophorum vaginatum*, *Sphagnum fallax*, *Brachythecium rivulare*, *Atrichum undulatum* and *Galium hercynicum* (Knorr et al., 2009). Vegetation is concentrated on the hummocks while the hollows are mostly free of vegetation.

3.2 ^{15}N pool dilution technique

The ^{15}N pool dilution technique was introduced by Kirkham and Bartholomew (1954). Numerous studies have used this method and related isotope pool dilution techniques to determine the soil N turnover in the last decades (Barraclough, 1991; Barraclough and Puri, 1995; Booth et al., 2005; Corre et al., 2007; Davidson et al., 1991; Hart et al., 1997; Murphy et al., 2003; Watson et al., 2000; Westbrook and Devito, 2004).

The benefits of this technique are (i) the product pool is labeled with ^{15}N rather than the substrate pool, (ii) easy operation and (iii) N turnover can be determined within a short-term incubation. This method is based on the fact that the input flow of N from the pool with natural ^{15}N abundance leads to a dilution of the labeled pool, while the output flow uses the isotopes at the given proportion and consequently does not change the enrichment of the labeled pool. Therefore, to measure the isotopic composition and the size of different N pools during a period of incubation allows the quantification of gross N turnover rates.

The application of $^{15}NH_4^+$ allows the measurement of gross ammonification and the application of $^{15}NO_3^-$ enables the measurement of gross nitrification. Gross ammonification is measured by initial enriching the soil NH_4^+ pool, which comprises ^{15}N at natural abundance levels, with the ^{15}N-enrichment above natural abundance by adding ^{15}N. The dilution of ^{15}N-enrichment in the pool, and change in the size of NH_4^+ pool is then traced through incubation as soil organic matter is ammonified, releasing in natural $^{15}NH_4^+$ abundance. Likewise, gross nitrification is measured by first enriching the soil NO_3^- pool, which contains ^{15}N at natural abundance levels, with ^{15}N to increase the ^{15}N-enrichment above natural abundance.

However, the ^{15}N pool dilution technique has a number of assumptions and limitations which may result in large errors in the calculated gross rates if the technique is not tested and applied correctly (Murphy et al., 2003): (1) Uniform distribution of added ^{15}N. (2) No discrimination of living organisms between ^{14}N and ^{15}N. (3) No re-mineralization of added ^{15}N and constant process rates during the incubation.

The equation of Kirkham and Bartholomew (1954) is:

$$m = \frac{M_0 - M_1}{t} \times \frac{\log(\frac{H_0 \times M_1}{H_1 \times M_0})}{\log(\frac{M_0}{M_1})} \quad (1)$$

where *m* is gross ammonification rate per unit mass of soil per unit time (mg N kg^{-1} soil d^{-1}); *M*, stands for the NH_4^+ total mass of tracing plus non tracing NH_4^+-N pool per unit mass of soil (mg N kg^{-1} soil); *H*, stands for the NH_4^+ tracer mass of tracing NH_4^+-N pool per unit mass of soil (mg N kg^{-1} soil); *t*, time interval, refers to the unit of days between the initial (M_0, H_0) and post-incubation (M_1, H_1) soil analysis; log, logarithm, to base 10.

Gross nitrification rate (*n*) can also be calculated by this equation after the labeled NO_3^- is applied into the indigenous pool and the nitrification of soil NH_4^+ and organic N at natural abundance leads to a dilution in the ^{15}N-enrichment of the NO_3^- pool (Davidson et al., 1991; Murphy et al., 2003; Watson et al., 2000).

There are 3 ways for ^{15}N application to the soil: (1) by solutions; (2) by solid salt and; (3) by gaseous N compounds (Murphy et al., 2003). The application by solutions is mostly used since this is easier to prepare, apply and carry out than application by gases or solids. However, addition of solution might change the rates of N transformations by the added water (Murphy et al., 1999, Willison et al., 1998). Hence, if dry soil is investigated a proceeding test is required to define if the application of ^{15}N solution is acceptable to determine gross N turnover.

To define a suitable ^{15}N application amount is a compromise between increasing the pool sizes unrealistically and achieving sufficient enrichment to follow the ^{15}N pool dilution with precision (Murphy et al., 2003). Although the product pool is labeled with ^{15}N rather than the substrate pool using this technique, it is generally recommended that as little ^{15}N as possible should be applied to avoid stimulating microbial activities that consume N.

It is necessary to conduct an initial soil extraction to ascertain the proportion of applied ^{15}N that is actually involved in ^{15}N pool dilution. The required incubation time is not constant

since the properties vary between different soil types. In previous studies, t_0 and t_1 differed to a large extent from 10 min to 48 h (t_0) and from 24 h to 7 days (t_1) (Bjarnason, 1988; Campbell and Gower, 2000; Christenson et al., 2009; Corre et al., 2007; Davidson et al., 1991; Grenon et al., 2004; Luxhøi et al., 2005; Murphy et al., 1997; Willison et al., 1998). Defining the time span between t_0 and t_1 is a compromise between fast interactions of the ^{15}N label with soil and the reasonable incubation time (t_1-t_0) allowing the dilution of ^{15}N label. If the incubation time is too short, the dilution may not be measurable, whereas at long incubation times the re-mineralization of immobilized ^{15}N will cause an underestimation of the gross turnover rates and/or the dilution of the ^{15}N label results in near natural abundances.

3.3 Experimental design

3.3.1 Laboratory incubation of forest soils

Homogenized soil samples of the Oi+Oe, Oa and EA horizons were taken from Coulissenhieb II forest. Six different water potentials ranging from field capacity to about -1.0 MPa were adjusted by air drying at room temperature. Gross rates of ammonification and nitrification were determined in 3 replicates with the ^{15}N pool dilution technique at a t_0 of 1 h and a t_1 of 49 h. All experiments were done at 15 °C.

For calculation of gross rates, ^{15}N abundances and concentrations of three t_0 samples were randomly pairwise related to three of the t_1 samples, resulting in 3 values for gross rates. Arithmetic means and standard errors were calculated using n = 3 using the software SIGMAPLOT 10.0 as shown in our figures.

3.3.2 Field experiment in forest site

Nine study plots, 3 control (C), 3 throughfall exclusion (TE) and 3 irrigation plots (I) of 20 m × 20 m each were established in a Norway spruce forest to simulate different precipitation patterns of a dry and wet growing seasons. Five undisturbed cores of the Oi+Oe and Oa+EA horizons were taken from each of the sampling points resulting in a total number of 270 cores per sampling date (3 sampling points at each treatment plot, 5 cores, 2 horizons). We sampled at 5 dates: 1× before (May), 2× during (Jul and Aug) and 2× after treatment (Sep and Oct). One of the 5 cores was used for the determination of soil water content. Calculation of matric

potentials from volumetric water contents was carried out using the van Genuchten model (van Genuchten, 1980). The parameters for this soil were taken from Zuber (2007). The other 4 cores were used for the determination of the gross N turnover rates (2 for ammonification and 2 for nitrification).

Throughfall was collected biweekly by 9 samplers (1 sampler per experimental plot). Forest floor percolates were collected below the Oa horizon (O) at each plot by 3 suction plates. In 20 and 90 cm soil depth, 3 ceramic suction cups were installed per plot. All soil solutions were collected every 4 weeks. Samples from the 3 suction plates were mixed to 1 sample per plot and per date. The volume, conductivity and pH of solution was measured and filtered for chemical analysis. Water fluxes with forest floor percolates for each sampling date were estimated based on the volume of water collected in the suction plates, the throughfall and irrigation amounts.

We used relative changes in gross N turnover rates and water potential values for data analysis. To calculate the relative changes, we determined the initial median rates of gross ammonification and nitrification at each treatment and subtracted these median rates. This procedure guarantees that the pre-treatment rates in May have zero median and that the spreading of the data inside the treatments remains unchanged. In our statistical analysis, we took this sampling design into account by using mixed-effects ANOVA (Pinheiro and Bates, 2000). We used sampling time and the interaction between treatment and sampling time as fixed-effects. All statistical analyses were done in R (R Development Core Team, 2010) using the packages nlme (Pinheiro et al., 2009) and stats (R Development Core Team, 2010).

3.3.3 Laboratory incubation of fen soils

To measure gross N turnover, 288 intact soil cores (with a height of 10 cm and a diameter of 5.6 cm) were taken from the fen site. In addition, another 10 large intact soil cores of 17.1 cm diameter from the top 10 cm were taken for measuring CO_2 emissions and soil solution chemistry.

Two regimes of water table were established. The water table in the permanently flooded cores was maintained at +5 cm for 117 days while the fluctuated regime comprised a change of the water table from +5 cm (flooded from day 0 to 24). The water table drawdown was initiated quickly to -8 cm within a few minutes and lasted from day 25 to 70. After that, the

water table was established again within few minutes to +5 cm (re-flooded from day 71 to 117).

Gross N turnover rates were determined with 3 replicates at 12 dates during the manipulation period. The CO_2 emissions of the 5 large cores of both treatments were monitored continually and soil solutions were collected at 6 dates. All experiments were done at 15 °C.

For calculation of gross rates, ^{15}N abundances and concentrations of three t_0 cores were randomly pairwise related to three of the t_1 cores, resulting in 3 values for gross rates. Arithmetic means and standard errors were calculated using n = 3 using the software SIGMAPLOT 10.0 as shown in our figures.

3.3.4 Field experiment in fen site

Six study plots, 3 control and 3 treated plots of 7 m x 5 m each, were established in summer 2005. From 2006 to 2008, drainage was induced by installing roofs to exclude precipitation and by additional pumping of groundwater from the drainage tiles at the 3 treated plots. During the first manipulation in 2006, tile drains were evacuated manually every 2 or 3 days by a submersed pump. The system was automated in 2007 and 2008 to keep the water table constantly at a lower level. At the end of the drainage experiments, regeneration of water table levels was partly achieved by natural precipitation and lateral water flow. In 2006 and 2007, we further applied artificial rainfall by distilled water. In 2008, no rewetting was needed due to the rapid rise of water table level by natural precipitation.

In 2009 and 2010, the 3 treated plots were changed from drainage to flooding. To keep the water table level above the soil surface, the surrounding of plots were trenched with PVC plates and irrigated with the stream water nearby. The water flow of whole system was propelled by gravity and the irrigation rate was about 8.3 mm h^{-1}.

Soil pore water was collected every 4 weeks in 10, 20 and 40 cm depth at each plot. In each depth, 3 replicate porous ceramic suction cups were installed. Soil pore water from the 3 suction cups were mixed to 1 sample per plot and per date in the laboratory, resulting in a sample size n = 3 for each treatment. The irrigation water for the flooded treatment in 2009 and 2010 was also collected from the stream directly every 1 week (2009) or 2 weeks (2010). The volume, conductivity and pH of solution was measured and filtered for chemical analysis.

3.4 Analytical techniques

Soil samples for gross N turnover were extracted with 1 M KCl solution. The ratio of soil/solution was 1:10 for the organic horizon and fen soils and 1:5 for the mineral soil. Filtered KCl extracts were frozen at -20°C and sent to the Helmholtz Centre for Environmental Research (UFZ, Halle) for analysis of ^{15}N abundance and the concentrations of NO_3^- and NH_4^+ using the SPINMAS technique (**S**ample **P**reparation unit for **I**norganic **N**itrogen and **MA**ss **S**pectrometer) (Stange et al., 2007). The SPINMAS comprises a coupling of a specially developed sample preparation device with a continuous flow-quadrupole mass spectrometer (QMS GAM 400, InProcess Instruments, Germany). The detection limits for NH_4^+ and NO_3^- with SPIMAS are 140 and 4.0 µM which are much less than the concentrations in our most extracts.

The soil solutions were measured for pH, electric conductivity, dissolved organic C (DOC, Elementar, high-TOC), total N (tN, Elementar, high-TOC), ammonium (NH_4^+, MLE, FIA-LAB), nitrate (NO_3^-, DIONEX, DX500 Chromatography system), sulfate (SO_4^{2-}, DIONEX, DX500 Chromatography system) and cations (Varian, ICP-OES). Concentration of DON was calculated as the difference between total N and inorganic N ($NH_4^+ + NO_3^-$).

The CO_2 emissions of fen soils were measured by an automated system BINOS 100 IRGA (Fisher-Rosemount, formerly Leybold Heraeus, max. detection of 1000 ppm with 50 ppm error). The air collected from the head space of the cores was dried (Drierite®, 8 mesh with indicator) and then pumped at constant rate of 1.5 L min^{-1} for 5 min (flooded period) or 1 min (water table drawdown period) with CO_2 concentration being logged automatically in 10 s intervals. Gas fluxes were calculated from the observed change of concentration over time by using Eq. 2:

$$F_{gas} = \left(\frac{dc}{dt}\right) \times \left(\frac{V_H \times M_W \times 60\,\text{min} \times 24\text{h}\ d^{-1}}{M_v \times A_H \times 1000\,ppm}\right) \times \left(\frac{P_a}{P_N \times (1 + 0.00366 \times T_a)}\right) \quad (2)$$

where F_{gas} represents the gas flux of the measured gas in mg m^{-2} d^{-1}, *dc/dt* is the change of concentration over time measured in the column in ppm min^{-1}, V_H is the volume of the column in liter, M_W is the molecular weight of C, M_V is the molecular volume of measured gas in L

mol^{-1}, A_H is the surface area of soil inside the column in m^2, Pa is the measured air pressure in hPa, P_N the standard air pressure, which is 1013 hPa, and T_a is the measured air temperature in °C with the factor 0.00366 originating from 1/273.15 due to the conversion from K to °C. This value is divided by 29 kg m^{-2} (top 10 cm) to transfer the unit as mg C kg^{-1} soil d^{-1}.

4. Synthesis and discussion of the results

4.1 Effects of decreasing water potential and rewetting on gross N turnover in forest soil

The laboratory results showed that the highest gross N turnover rates were observed at field capacity and soil drying decreased rates in the O horizon (Fig. 2). Surprisingly, gross ammonification measured at -1.2 MPa was similar to that at -0.8 MPa in the Oi+Oe horizon. In the Oa horizon, gross ammonification rates decreased linearly when water potential dropped from field capacity to -0.6 MPa but did not further decrease when water potential dropped to -0.8 MPa. In the EA horizon, gross ammonification was generally low and decreased from field capacity to -0.25 MPa, but there was no further response to decreasing water potential.

Low et al. (1997) reported that the reduction of gross ammonification in a pasture soil was best fitted by an exponential function when osmotic potentials decreased from 0 to -0.5 MPa. They observed that gross ammonification did not respond to a further drop in water potential from -0.5 to -1.75 MPa, which is similar to the surprising lack of response of gross ammonification at water potentials < -0.6 MPa in the O horizons. Using soil from our site, Muhr et al. (2010) observed a reduction in total CO_2 emissions by about 65% when water potential dropped from field capacity to -1.2 MPa, supporting the presence of substantial microbial activity even under severe soil drying.

In the field experiment, the differences of gross ammonification were not statistically significant between the treatments during the manipulation (Jul and Aug) (Fig. 3). However, the rates of gross ammonification were lowest at the TE plots at pF 4.5 (-3.2 MPa) in the Oi+Oe and at pF 3.8 (-0.6 MPa) in the Oa+EA horizons in August. A substantial ammonification rate of 14 mg N kg^{-1} soil day^{-1} was still observed at pF 4.5. The natural rewetting after soil drying did not cause pulses in gross ammonification (Sep and Oct).

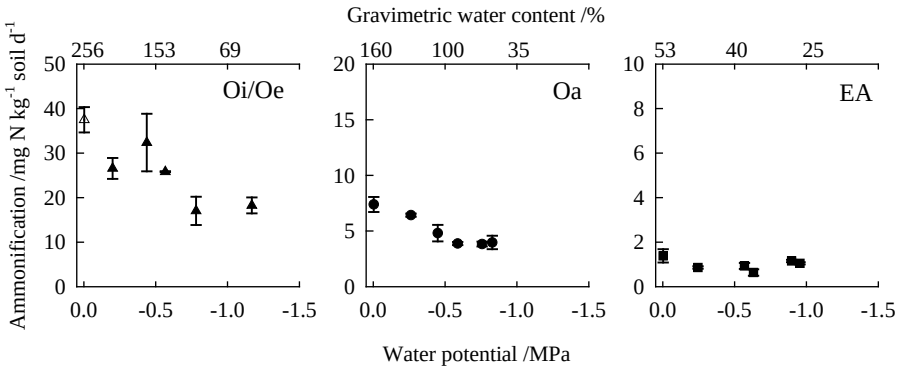

Fig. 2 Effect of water potential on gross ammonification rates in the Oi+Oe, Oa and EA horizons in a spruce forest (mean±SE, n=3).

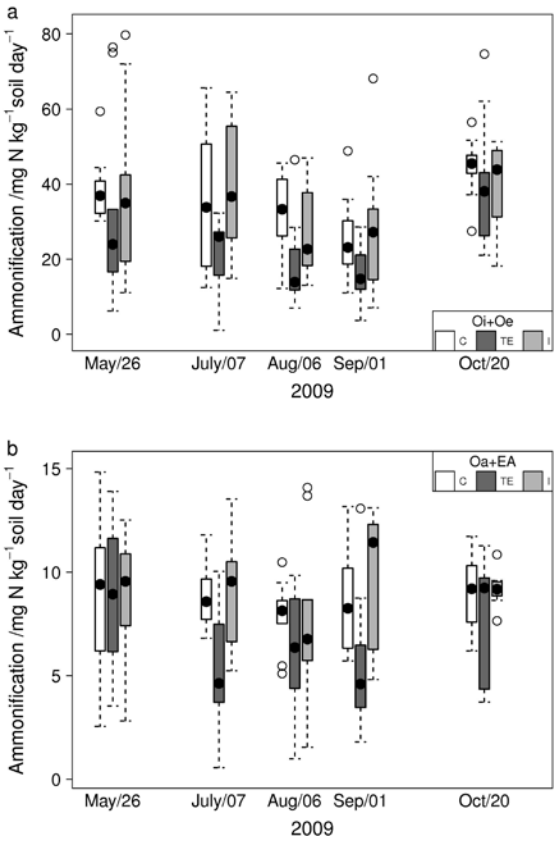

Fig. 3 Box plots of the median gross ammonification rates (●) at 5 sampling dates in (a) the Oi+Oe horizon and in (b) the Oa+EA horizon. The top and bottom of the box display the largest and smallest observations within the interquartile range (25 - 75% of our data) (n = 9). The hollow circle (○) stands for outliers over this range.

High efficiency of N mineralization due to low N requirements of fungi may contribute to the relatively high net N mineralization in acid soils (Kooijman et al., 2009). Therefore, the reason for the lack of response to low water potentials might be the contribution of less drought sensitive fungi in relation to bacterial ammonification. Another reason for the small response of gross ammonification might be the adaption of ammonifiers to frequent soil drying in the uppermost soil horizons. These interpretations remain speculative since the relative contributions of fungi and bacteria to gross ammonification at low water potential are unknown.

Several factors can explain the absence of a drying/rewetting effect under field conditions. First, soil drying to pF 4.5 (-3.2 MPa, equal to 50% w/w) was not severe enough compared to other studies (Pulleman and Tietema, 1999; Saetre and Stark, 2005). In our study, the soil microorganisms were not inhibited completely by pF 4.5 (-3.2 MPa) as indicated by a substantial ammonification rate of 13.9 mg N kg^{-1} soil day^{-1} in August. Second, the change of water potential from pF 3.2 to 2.1 (-3.2 to -0.01 MPa, 50% to 150% w/w) in 75 days was not rapid enough to induce a wetting effect. Soil microbes accumulate solutes to reduce their internal water potential to avoid dehydration and drying during the drought period. When the soil is rewetted, microbes must dispose these osmolytes immediately until the water potential equilibrates with that of the surrounding water or the water will flow into the cell and potentially cause cell rupture (Schimel et al., 2007). The peak of microbial activities might have been rather short after rewetting. Overall, the rewetting effect on ammonification might be rather small and short lived and we likely missed such an effect due to our long sampling interval. A laboratory experiment on drying and rewetting with the same soil also found no significant excess net N-mineralization after a 40 days rewetting period following severe drying (Muhr et al., 2010).

In case of gross nitrification, the laboratory results showed that the rates were highest in the Oi+Oe horizon and much lower in the Oa horizon but not detectable in the EA horizon (Fig. 4). Gross nitrification rates at -1.2 MPa were similar to that at -0.8 MPa in the Oi+Oe horizon but decreased linearly to almost zero when water potential dropped from field capacity to -0.8 MPa in the Oa horizon.

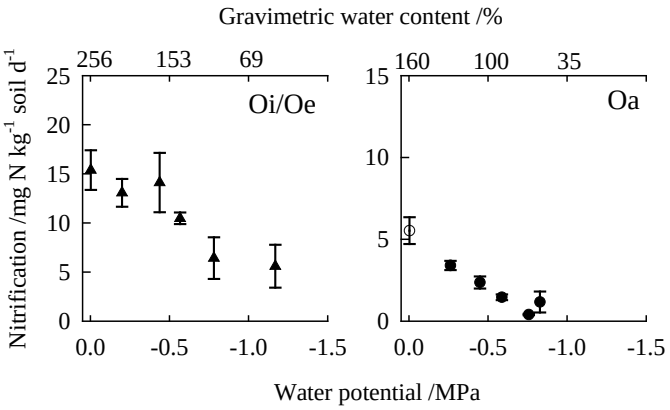

Fig. 4 Effect of water potential on gross nitrification rates in the Oi+Oe and Oa horizons in spruce forest (mean±SE, n=3).

In the field experiment, gross nitrification was very low and no effect of soil drying was observed due to the low rates and huge spatial variation (Fig. 5). No response of gross nitrification to decreasing water potential was observed, contradicting the results from disturbed samples. Gross nitrification rates were larger in disturbed soils than in intact soil cores. This finding is in agreement with Luxhøi and Jensen (2005) who found two folds greater gross nitrification rates in disturbed soils than in intact soil cores from an arable land. It seems that the mixing of soil improved the NH_4^+ supply of nitrifiers and enhanced gross nitrification rates.

The reasons for the lack of response were the generally small rates of nitrification in the undisturbed samples and the huge spatial variation among the replicates. Besides the spatial variation of microbial biomass (Matejek et al., 2010a), different proportions of Oi, Oe, Oa and A horizon material and gradients in the water contents cause great variation among the undisturbed soil cores. We did not observe a rewetting effect on gross nitrification, the reasons being similar to those discussed above for gross ammonification. In another coniferous forest, simulated summer droughts and subsequent rewetting did not induce a pulse of gross nitrification either, indicating that nitrifiers were not stimulated by rewetting (Tietema et al., 1997).

Furthermore, our laboratory results showed that the relative decrease of gross nitrification in relation to gross ammonification was similar in the Oi+Oe horizon, but the decrease of

nitrification was steeper in the Oa horizon (Fig. 6), indicating that nitrification was more sensitive to drying than ammonification in the Oa horizon. Gross nitrification almost ceased at -0.8 MPa, while ammonification was still considerable great. Hence, substrate limitation cannot explain the cease of nitrification in the Oa horizon. It might be the sensitivity of autotrophic nitrifying bacteria to drying, whereas ammonification, which is driven by a large variety of bacteria and drought tolerant fungi, is less sensitive. In contrast, nitrification in the Oi+Oe horizon continued with a considerable rate at -1.2 MPa, suggesting that nitrifiers are more tolerant to drying in the uppermost soil horizon than in the Oa horizon. It remains an

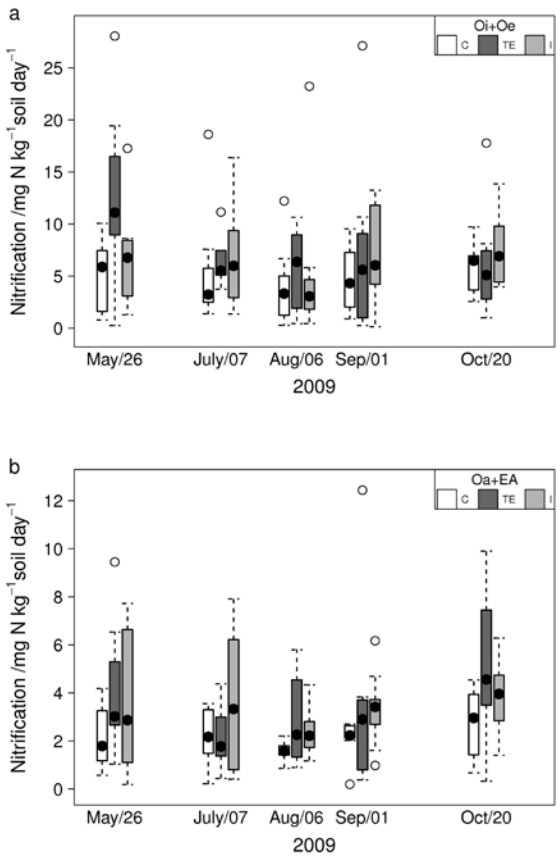

Fig. 5 Box plots of the median gross nitrification rates (●) at 5 sampling dates in (a) the Oi+Oe horizon and in (b) the Oa+EA horizon. The top and bottom of the box display the largest and smallest observations within the interquartile range (25 - 75% of our data) (n = 9). The hollow circle (○) stands for outliers over this range.

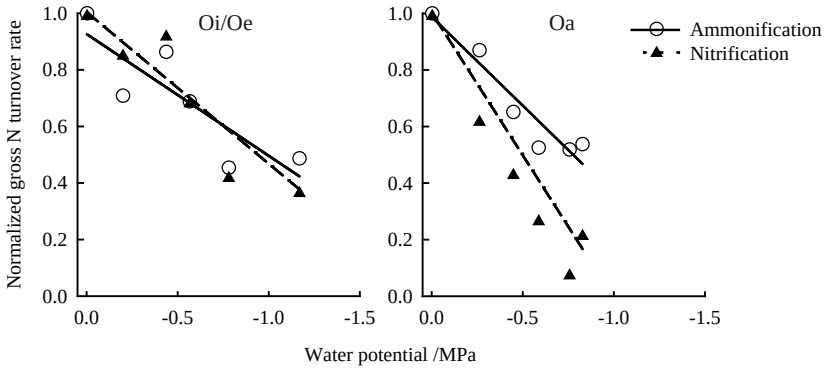

Fig. 6 Effect of water potential on normalized gross rates of ammonification and nitrification in the Oi+Oe and Oa horizons in a spruce forest.

open question if this is caused by a larger proportion of heterotrophic, drought resistant fungi in relation to bacterial autotrophic nitrification in the Oi+Oe horizon. This interpretation is supported by the observation of higher relative fungal biomass in the uppermost soil layers (Fierer et al., 2003; Fritze et al., 2000; Schmitt et al., 2008). Also, Scheu and Parkinson (1994) observed a significant reduction in bacterial biomass in forest floor horizons by soil drying whereas the fungal biomass was less affected.

4.2 Effects of soil drying/rewetting and irrigation on *in situ* DIN, DON and DOC fluxes in forest soil

The total cumulative flux of DIN with throughfall at the C plots was about 39 kg N ha^{-1} for the period from January 2009 to December 2010 (Fig. 7). Because of throughfall exclusion the N flux with throughfall was reduced to 31 kg ha^{-1} at the TE plots. On average, NO_3-N comprised about 90% of the DIN fluxes in forest floor percolates, but only about 54% in throughfall. During the period of throughfall exclusion, the solute DIN fluxes of the TE plots were smaller than those of the C and I plots.

The *in situ* fluxes of DIN with forest floor percolates corroborate the findings of gross N turnover from the soil cores: Although an increase of DIN flux occurred at the TE plots in winter, the cumulative DIN flux at the TE plots did not significantly exceed that at the C plots. Furthermore, the DIN flux at the TE plots was already slightly higher in the pre-treatment

period compared to the C and I plots. A laboratory study using the same soil also did not find an increase in N fluxes with soil solution after rewetting Muhr et al. (2010). Our results are in agreement with findings of Lamersdorf et al. (1998) and contradict our initial hypothesis. Drying/rewetting under field conditions did not enhance total DIN fluxes in forest floor percolates.

The total cumulative fluxes of DON and DOC with throughfall at the C plots were about 15 and 230 kg N ha^{-1} for the period from January 2009 to December 2010 (Fig. 8 and Fig. 9). At the TE plots, DON and DOC fluxes with throughfall were reduced to 11 and 160 kg ha^{-1}, respectively. The lowest fluxes at the TE plots were due to the reduction of throughfall. The hypothesis of increased DON and DOC fluxes by soil drying/rewetting was not confirmed. Our previous laboratory experiments also showed that cumulative DOC fluxes did not significantly increase by the drying/rewetting cycles (Hentschel et al., 2007). Because of reduced water fluxes, DOC concentrations were mostly significant higher as compared to the control. Borken et al. (1999) also did not found any increase in annual DOC flux at 10 cm mineral soil depth due to a strong reduction in annual water flux owing to drying/rewetting of a Norway spruce stand.

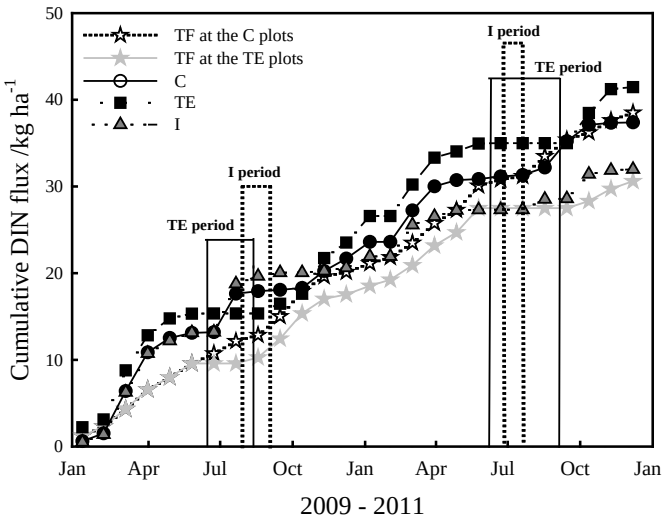

Fig. 7 Cumulative DIN (NH_4-N + NO_3-N) flux with throughfall (TF) at the C (☆) and TE (★) plots and forest floor percolates at the C (●), TE (■) and I (▲) plots.

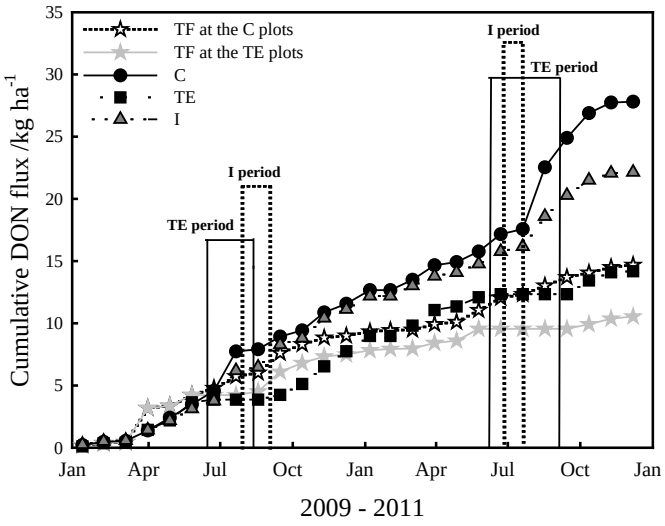

Fig. 8 Cumulative DON fluxes with throughfall (TF) at the C (☆) and TE (★) plots and forest floor percolates at the C (●), TE (■) and I (▲) plots.

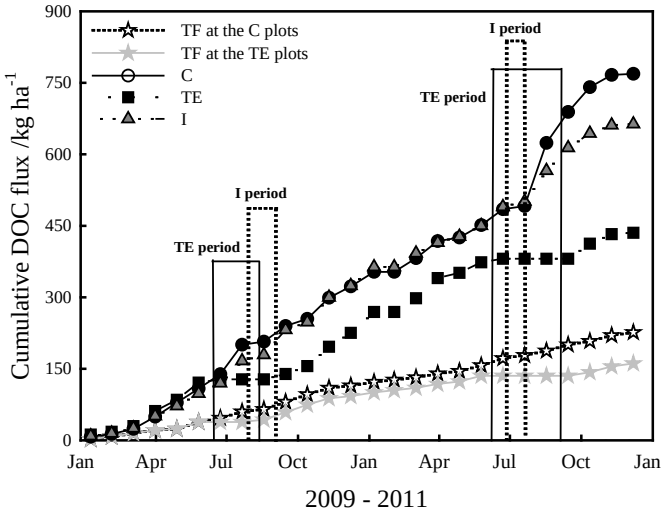

Fig. 9 Cumulative DOC fluxes with throughfall (TF) at the C (☆) and TE (★) plots and forest floor percolates at the C (●), TE (■) and I (▲) plots.

In summary, soil drying decreased gross N turnover in forest floors but the substantial rates of gross ammonification still occurred at low water potentials, indicating the adaption of ammonifiers to frequent soil drying in the O horizons. The difference between the disturbed samples and intact soil cores suggested that nitrification had a huge spatial heterogeneity and is potentially stimulated by soil disturbance. Extended drought periods caused a reduction in gross N turnover in forest soils but no rewetting effect was found. The hypothesis of increased fluxes of DIN, DON and DOC as a consequence of drying/rewetting was not confirmed.

4.3 Dynamics of N and C mineralization in a fen soil following water table fluctuations

4.3.1 Response of N turnover to water table fluctuations

Under permanently flooded and re-flooded conditions, gross ammonification increased after a lag phase of about 30 days (Fig. 10). The response of gross ammonification to flooding was matched by the increasing NH_4^+ concentrations in soil solutions (Fig. 11), indicating a similar response of net N mineralization to flooded conditions. In the fluctuated cores, gross ammonification also increased after 30 days of water table drawdown.

The cumulative gross ammonification rates were 2840 and 1940 mg N kg^{-1} soil in the permanently flooded and in the fluctuated cores, respectively (Fig. 12). Gross nitrification rates were much smaller than gross ammonification. In the permanently flooded cores NH_4^+ concentrations increased and then remained constantly high. The concentrations of NH_4^+ decreased after the water table drawdown and increased when the soil was re-flooded. The NO_3^- concentrations were always lower than NH_4^+.

The increase of gross ammonification in the permanently flooded cores can be due to changes of substrate availability, substrate pools, changes in enzymatic and microbial activities (Corstanje and Reddy, 2004; Kraigher et al., 2006; Mentzer et al., 2006). After water table drawdown and aeration, gross ammonification also increased to similar rates as in the permanently flooded cores. The increase was not instantaneous, but occurred after a lag phase of about 30 days, coinciding with the second peak of CO_2 emissions (Fig. 13). The latter indicates an enhanced activity of aerobic microorganisms after aeration with a lag phase of about 30 days for physiological adaptation of microbial activity (Blodau et al., 2004). The decrease of the NH_4^+ concentrations in soil solutions also points to the enhanced

immobilization by growing microbial biomass.

Under flooded conditions, the lack of O_2 might be the most likely reason for the low gross nitrification rates (Bayley et al., 2005; Bowden, 1986), but also low pH-values are known to reduce autotrophic nitrification. Net nitrification rates were often close to zero in peat soils (Bayley et al., 2005; Hefting et al., 2004; Neill, 1995; Wray and Bayley, 2008), but an increase of net nitrification was observed after water table drawdown or drainage

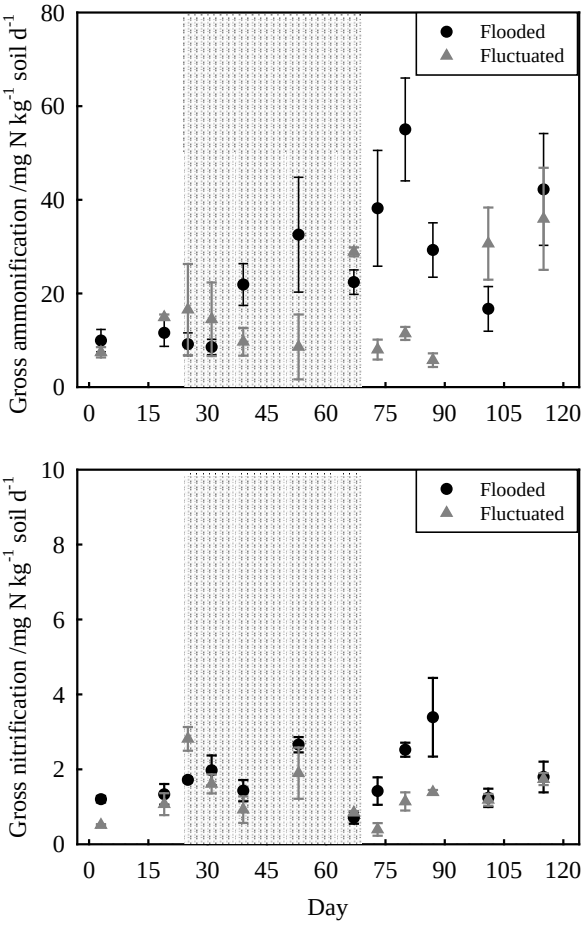

Fig. 10 Gross ammonification and nitrification rates (mean ± SE, n = 3) in the permanently flooded (●) and the fluctuated (▲) treatments. The gray bar indicates the water table drawdown period in the fluctuated treatment.

(Münchmeyer et al., 2000; Neill, 1995). Yu and Ehrenfeld (2009) found that net nitrification increased within 2 weeks when the water content decreased from 100% to 30% of the water holding capacity.

Under the field conditions, NH_4^+ concentrations were always lower than 0.3 mg L^{-1} and NO_3^- concentrations were almost zero at 10 cm depth. The low concentrations of DIN might be due to the plant uptake. Moreover, the field site is subjected to permanent lateral water flow removing huge amounts of DIN, which was not the case in the laboratory experiment.

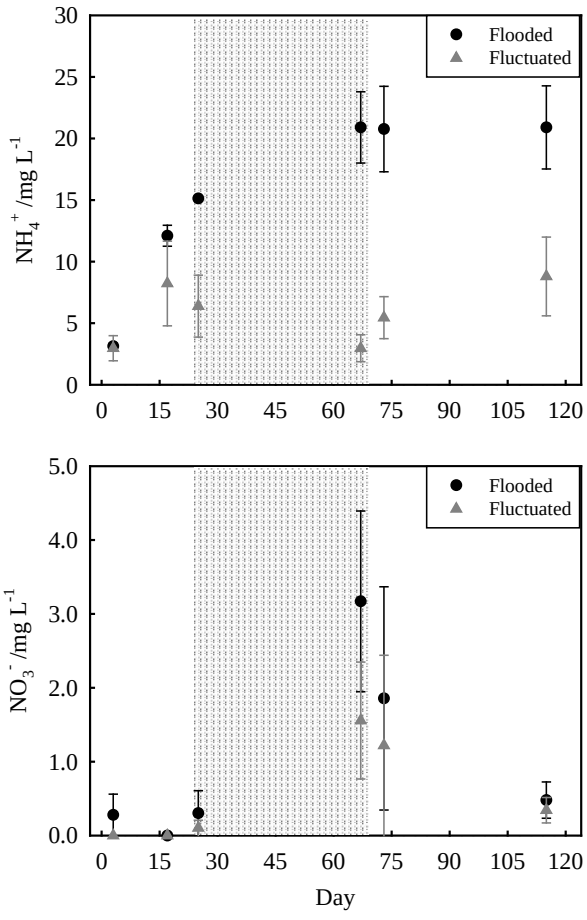

Fig. 11 NH_4^+ and NO_3^- concentrations (mean ± SE, n =3) in the permanently flooded (●) and the fluctuated (▲) treatments. The gray bar indicates the water table drawdown period in the fluctuated treatment.

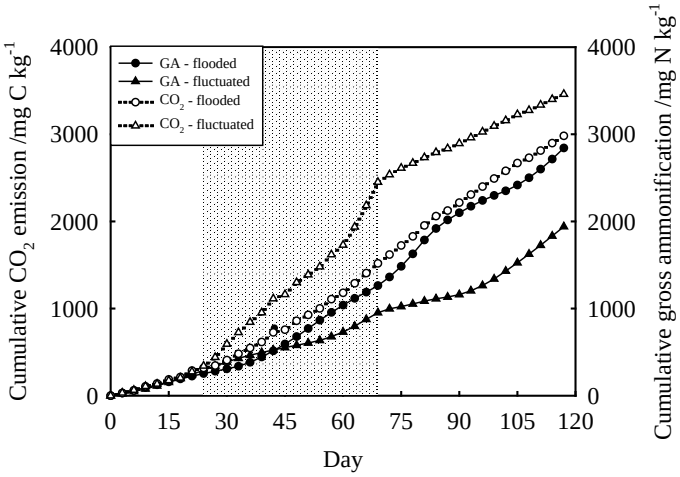

Fig. 12 Cumulative gross ammonification rate (GA) and CO_2 flux in the permanently flooded and the fluctuated treatments. The gray bar indicates the water table drawdown period in the fluctuated treatment.

4.3.2 Response of C mineralization to water table fluctuations

CO_2 emission was constantly low under the permanently flooded and O_2 limited conditions (Fig. 13). Also when the aerated soil was re-flooded, a rapid decline of CO_2 flux within 1 day to the level of the permanently flooded cores was found, indicating quick response of aerobic microorganisms to the lack of O_2. The cumulative CO_2 emission over the whole experimental period was 2980 mg C kg^{-1} soil in the permanently flooded cores and 3460 mg C kg^{-1} soil in the fluctuated cores (Fig. 12).

The first peak was probably due to the stimulating effect of aeration. Obviously a pool of easily degradable substrates was quickly mineralized causing a sharp increase in CO_2 emissions at an hourly to daily time scale. In the following 3 - 5 days, the easily degradable substrate depleted. After a lag phase of about 30 days, the growth and activity of aerobic microorganisms (Blodau et al., 2004; Jaatinen et al., 2008; van Dijk et al., 2009) might be the reason to explain the second peak of CO_2 emissions. This assumption is supported by the increased gross ammonification and decreased NH_4^+ concentration. In summary, the initial peak of CO_2 obviously consumed easily available substrates and the second peak seems to be the mineralization of more recalcitrant substrates.

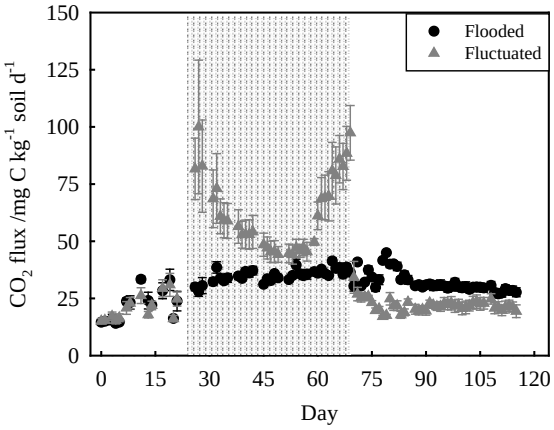

Fig. 13 CO_2 emissions in the permanently flooded (●) and the fluctuated (▲) treatments (mean ± SE, n = 5). The gray bar indicates the water table drawdown period in the fluctuated treatment.

4.3.3 C mineralization in relation to gross ammonification

The ratios of CO_2 emissions to gross ammonification (C and N mineralization in mol) were close to 2 under flooded conditions and increased to about 6 after water table drawdown (Fig. 14). Low rates of C mineralization are typically found under anoxic conditions in peat soils and C is partly reduced to CH_4 or metabolized to soluble (Fenner et al., 2005; Freeman et al., 2004; Pastor et al., 2003) or to insoluble metabolites.

Our calculations of the gross N turnover rates revealed that most of the NH_4^+ from gross ammonification is consumed by immobilization by the microbial biomass and that the net rates are much less than the gross rates (data not shown). This indicates a very fast turnover of N in the microbial biomass of our soil. Thus, low CO_2 production and high turnover rate of microbial biomass-N might explain the low C/N ratio of mineralization.

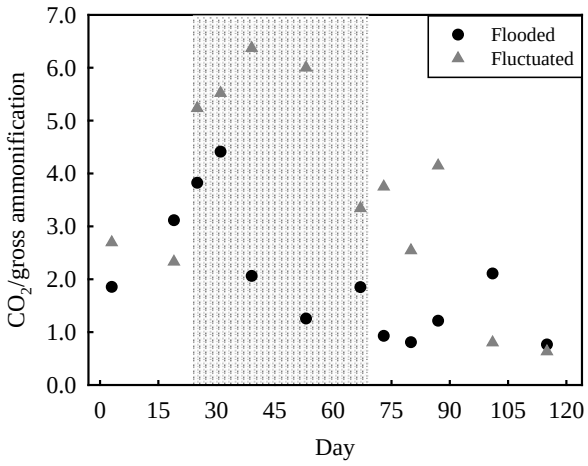

Fig. 14 Ratios of CO_2 emission/ gross ammonification in the permanently flooded (●) and the fluctuated (▲) treatments. The gray bar indicates the water table drawdown period in the fluctuated treatment.

4.3.4 Response of DON and DOC to water table fluctuations

The overall range of DON and DOC concentrations were very high with maxima of around 15 mg DON L^{-1} and 500 mg DOC L^{-1} (Fig. 15). Under field conditions, concentrations in our fen soil in 10 cm depth are much less (maximum around 2.0 mg DON L^{-1} and 60 mg DOC L^{-1} at 10 cm depth). The reasons for the high concentrations of DON and DOC in the laboratory might be the higher temperatures in our laboratory study (under field conditions soil temperature is around 10°C in the growing season). Furthermore, the field site is subjected to permanent lateral water flow removing huge amounts of dissolved organic matter, which was not the case in the laboratory experiment.

The initial flooding increased the DON and DOC concentrations in both sets of cores. Concentrations remained on a high level in the permanently flooded cores, while the water table drawdown induced a strong decrease of the concentrations. After re-flooding, concentrations increased again. During the flooding, C is less likely to be completely metabolized to CO_2 and instead, dissolved organic compounds may preferentially be formed as end products as suggested by Fenner et al. (2005), Freeman et al. (2004) and Pastor et al. (2003). One reason for decreasing concentrations of DOC might be the aerobic conditions associated with the water table drawdown, favoring CO_2 rather than DOC as the major end

product of decomposition. In our study, the average DOC pool in the soil decreased by 590 mg kg^{-1} soil. In the same time period the cumulated CO_2 emissions were 2100 mg kg^{-1} soil. This indicates that the decline of the DOC pool after water table drawdown can be explained by mineralization of DOC. A second reason for the decline in DOC after water table drawdown might be the decreased solubility of C compounds due to increased H^+ concentration and ionic strength associated with SO_4^{2-} production (Clark et al., 2005; Driscoll et al., 1989; Hruška et al., 2009).

The concentrations of DON were generally in the same range as those of NH_4^+, emphasizing the role of DON for solute N transport in fen soils. The response of DON to water table fluctuations was similar to DOC as indicated by the almost constant DOC/DON ratios.

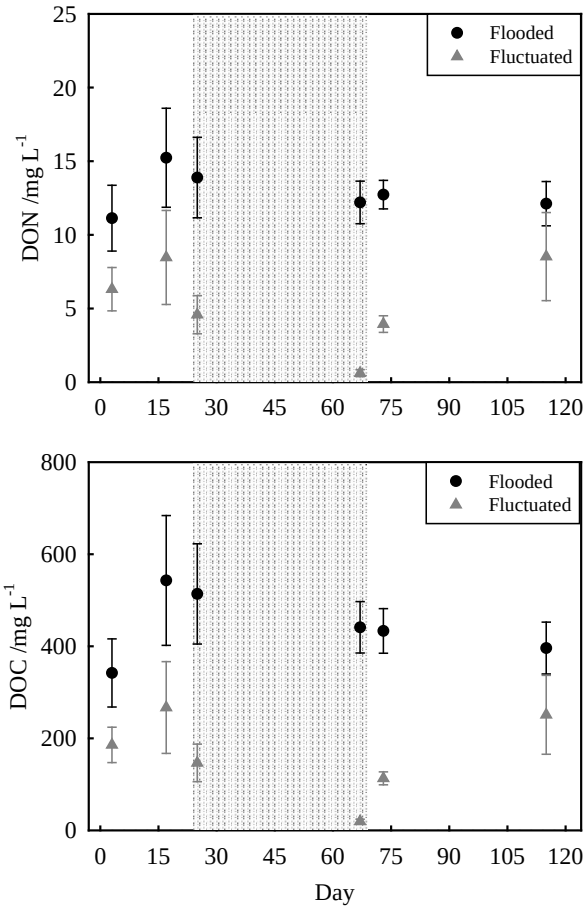

Fig. 15 DON and DOC concentrations (mean ± SE, n = 3) in the permanently flooded (●) and the fluctuated (▲) treatments. The gray bar indicates the water table drawdown period in the fluctuated treatment.

4.4 Effects of drainage and flooding on *in situ* pore water chemistry in a fen soil

Drainage increased the concentrations of SO_4^{2}-S in pore waters under field conditions and high concentrations persisted for several months in 2006 and 2008 (Fig. 16). Oxidation of reduced S compounds stored within the soil during dry periods has been noted as the main source of SO_4^{2-} in peat soils (Eimers et al., 2003; Schiff et al., 2005). The lack of response to the drainage in 2007 may be due to the high precipitation. Although the water table at the treated plots reached to -0.25 m, the effect of aeration induced by drainage was eliminated by the high precipitation during the manipulation period. In 2009 and 2010, the effect of flooding on SO_4^{2}-S was not noticeable.

The concentrations of Fe were lower in all depths at the treated plots (Fig. 17), pointing to more oxic conditions during the drainage period (Knorr and Blodau, 2009). Even after the rewetting, Fe concentrations at the treated plots were never higher than the control plots in all depths, indicating that the effect remained for few months. The largest difference in 2007 between the two treatments might be due to the high precipitation. In summer 2008, the concentrations of Fe at the control plots decreased to the level similar to the treated plots resulting from the lower water table level compared to 2006 and 2007. However, the concentrations increased again because of the high precipitation after the manipulation period. Flooding increased Fe concentrations in 2009. In 2010, the reason for similar concentrations of Fe at both treatments might be the high precipitation.

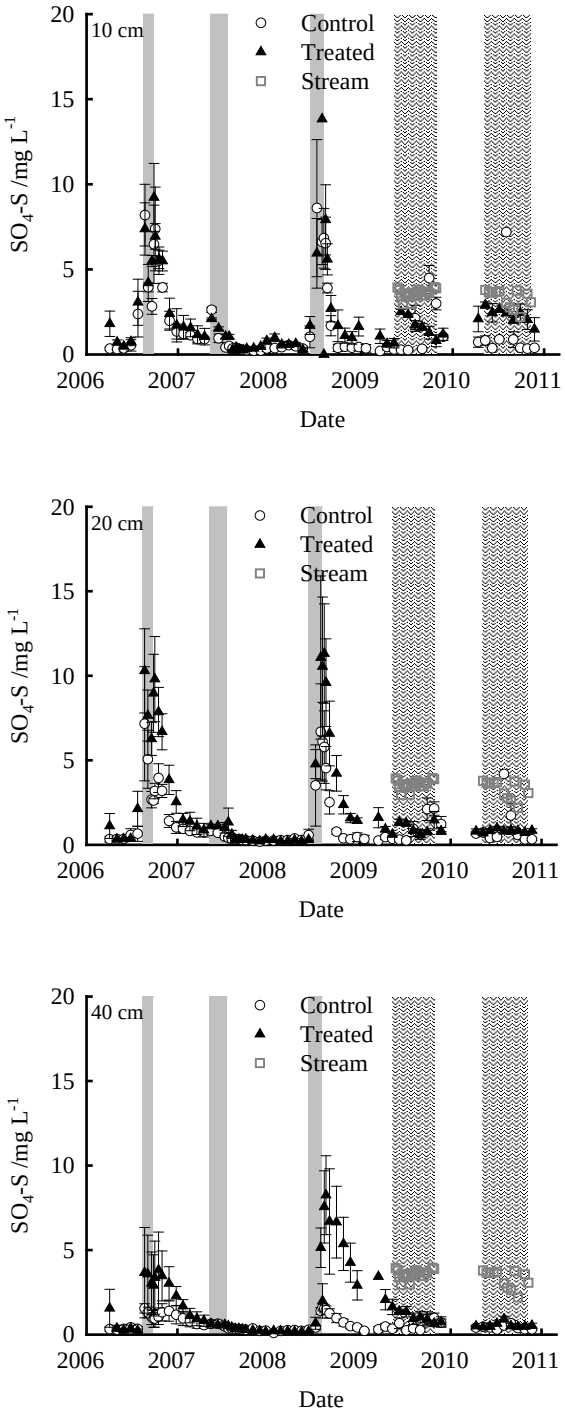

Fig. 16 Mean concentrations of SO_4^{2-}-S in 10, 20 and 40 cm depth at control (○) and treated (▲) plots (±SE; n=3). The concentrations of stream water are showed as hollow squares (□; n=1). The gray solid bars indicate the drainage period in 2006, 2007 and 2008. The grid bars indicate the flooding period in 2009 and 2010.

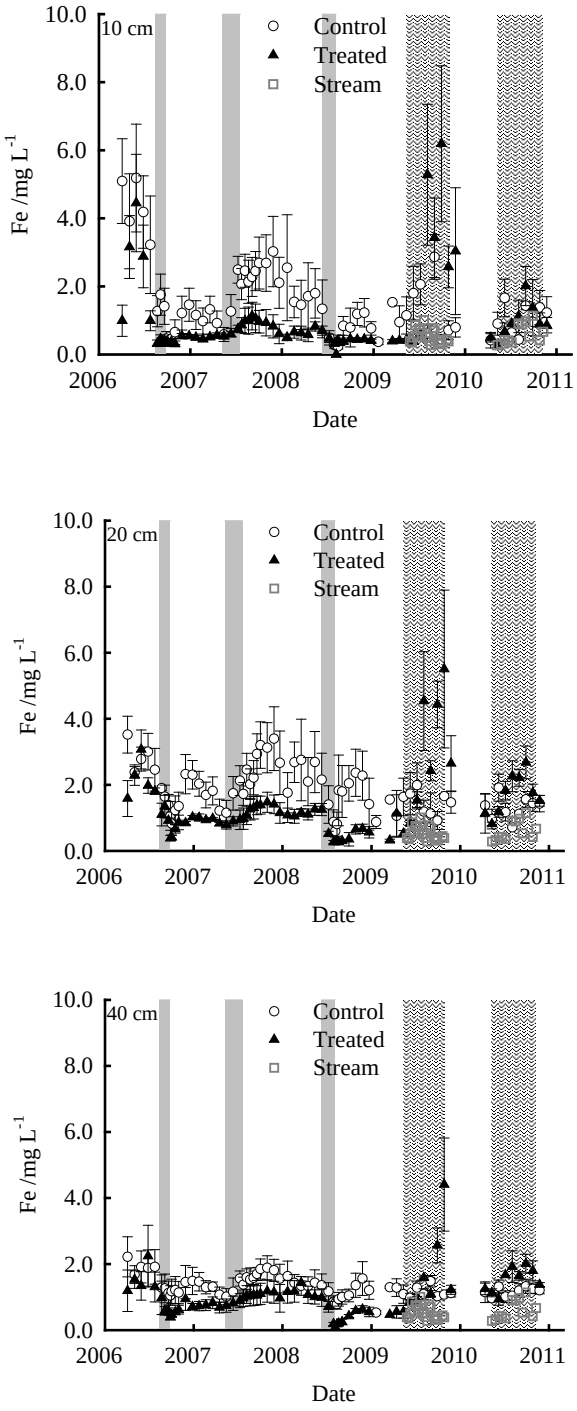

Fig. 17 Mean concentrations of Fe in 10, 20 and 40 cm depth at control (○) and treated (▲) plots (±SE; n=3). The concentrations of stream water are showed as hollow squares (□; n=1). The gray solid bars indicate the drainage period in 2006, 2007 and 2008. The grid bars indicate the flooding period in 2009 and 2010.

Drainage decreased and flooding increased the concentrations of DON (Fig. 18) and DOC (Fig. 19). Aerobic conditions associated with water table drawdown, favor CO_2 rather than DOM as the major end product of decomposition (Freeman et al., 2004). The decline of DON and DOC concentrations after the drainage might be due to the changed activity, biomass and community of microorganisms after aeration (Briones et al., 1997; Peltoniemi et al., 2009; van Dijk et al., 2009). Besides, increased H^+ concentration and ionic strength associated with increased SO_4^{2-}, hinder the dissociation of organic acids and decrease their solubility, which also leads to a decline in DON and DOC concentrations (Driscoll et al., 1989; Hruška et al., 2009). When the concentration of SO_4^{2-} > 10 mg L^{-1}, DON was always lower than 1.0 mg $L^{-1}$and DOC was never higher than 20 mg L^{-1} (data not shown).

During the flooding, DOM is less likely to be completely metabolized to CO_2 and instead, DOM may preferentially be formed as end products as suggested by Fenner et al. (2005), Freeman et al. (2004) and Pastor et al. (2003).

Neither drainage nor flooding had an effect on NH_4^+-N. Concentrations were very low with high spatial variation (Fig. 20). The microbial immobilization (Schimel et al., 1989; Stark and Hart, 1997) and vegetation uptake (Templer et al., 2008) of NH_4^+ can be the reasons to explain the lack of response to water table fluctuations. The NO_3^--N concentrations were always low and unaffected by the fluctuated water table (Fig. 21). This may be due to the limitation of oxygen (Bayley et al., 2005), inhibition of nitrification by low pH (Aciego Pietri and Brookes, 2008; Tietema et al., 1992), rapid denitrification (Hefting et al., 2004) or coupling of its reduction with the SO_4^{2-} production (Burgin and Hamilton, 2008).

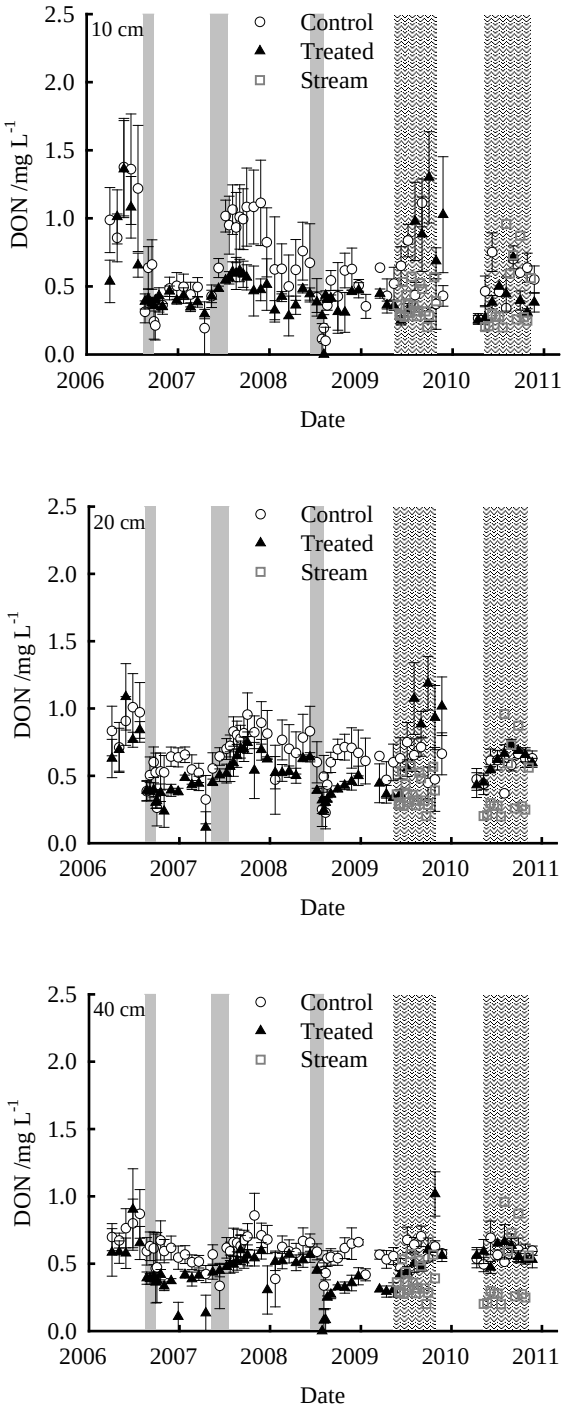

Fig. 18 Mean concentrations of DON in 10, 20 and 40 cm depth at control (○) and treated (▲) plots (±SE; n=3). The concentrations of stream water are showed as hollow squares (□; n=1). The gray solid bars indicate the drainage period in 2006, 2007 and 2008. The grid bars indicate the flooding period in 2009 and 2010.

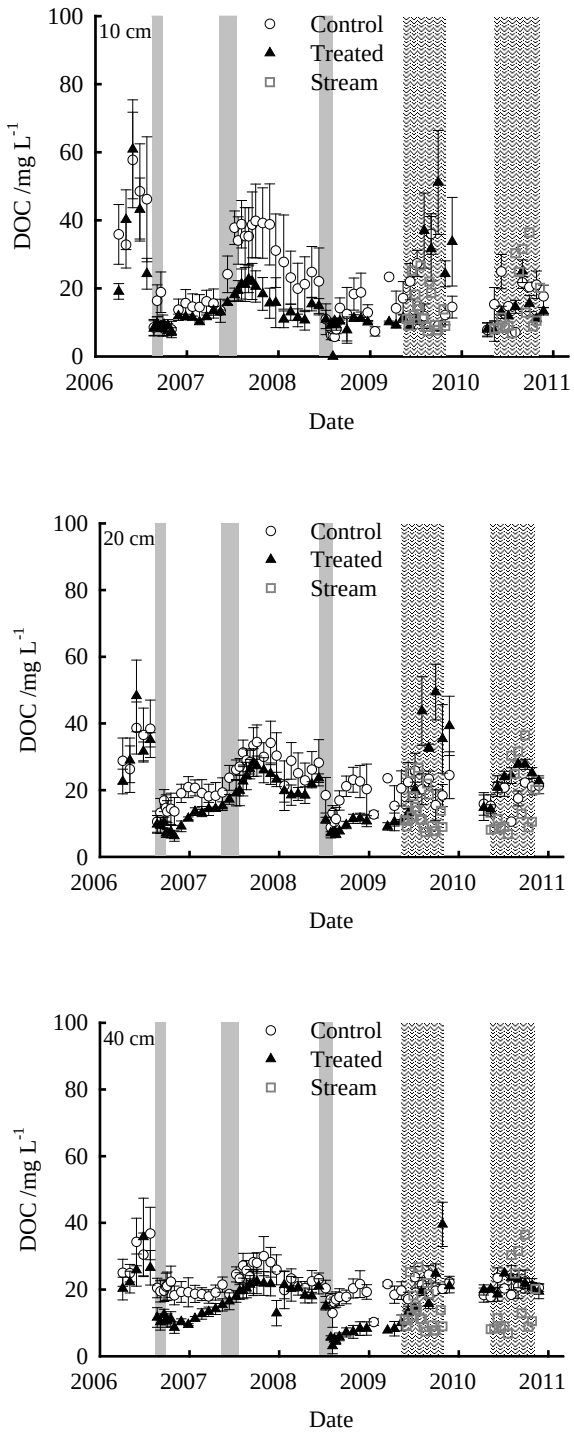

Fig. 19 Mean concentrations of DOC in 10, 20 and 40 cm depth at control (○) and treated (▲) plots (±SE; n=3). The concentrations of stream water are showed as hollow squares (□; n=1). The gray solid bars indicate the drainage period in 2006, 2007 and 2008. The grid bars indicate the flooding period in 2009 and 2010.

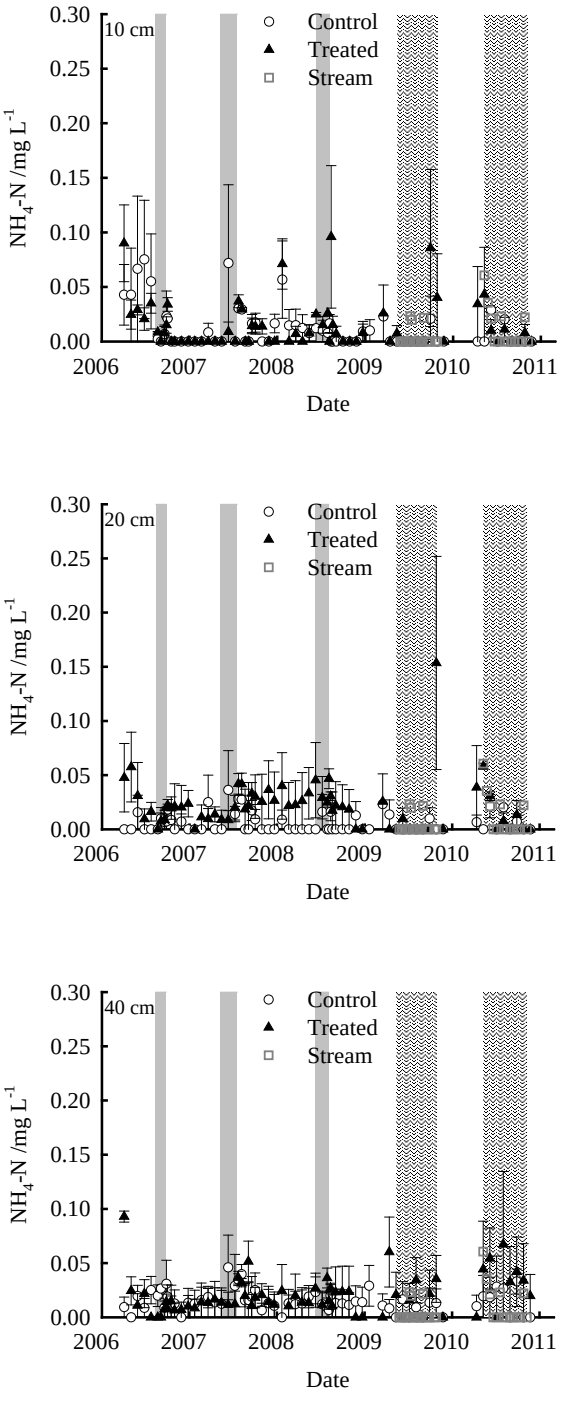

Fig. 20 Mean concentrations of NH_4^+-N in 10, 20 and 40 cm depth at control (○) and treated (▲) plots (±SE; n=3). The concentrations of stream water are showed as hollow squares (□; n=1). The gray solid bars indicate the drainage period in 2006, 2007 and 2008. The grid bars indicate the flooding period in 2009 and 2010.

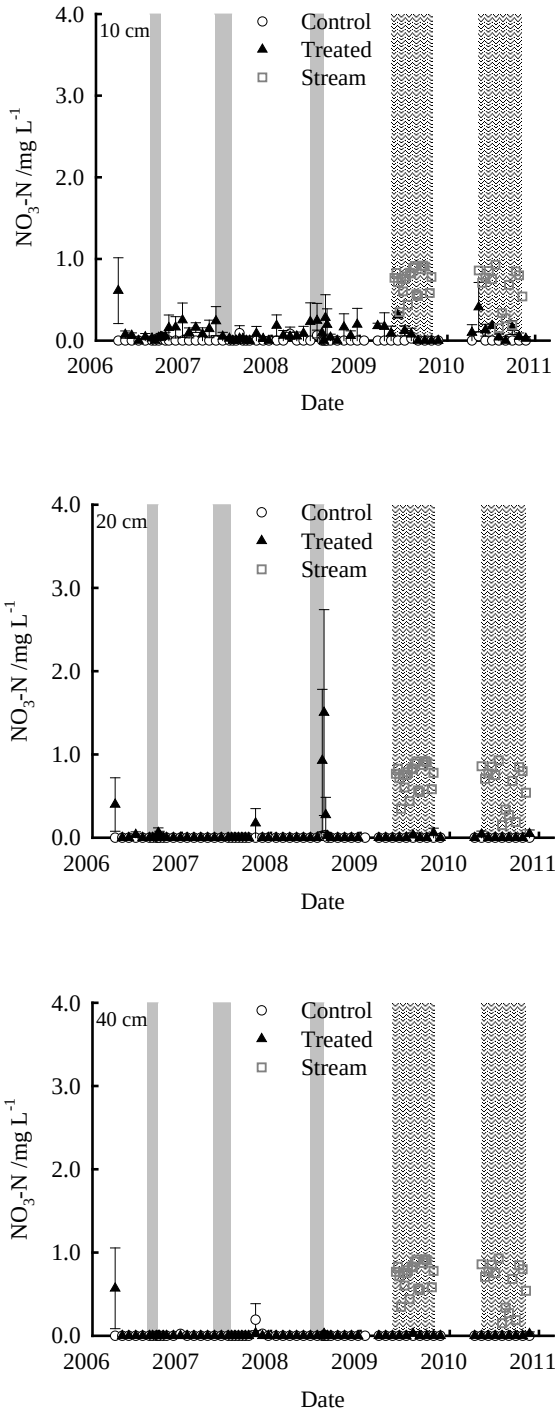

Fig. 21 Mean concentrations of NO_3^--N in 10, 20 and 40 cm depth at control (○) and treated (▲) plots (±SE; n=3). The concentrations of stream water are showed as hollow squares (□; n=1). The gray solid bars indicate the drainage period in 2006, 2007 and 2008. The grid bars indicate the flooding period in 2009 and 2010.

In summary, the fluctuations of water table influenced the organic matter mineralization, soil solution chemistry and inorganic N availability in fen soils. Mineralization of N and C was sensitive to water table fluctuations in the top 10 cm. Both flooding and water table drawdown increased gross ammonification after a lag phase of about 30 days. Flooding inhibited C mineralization and water table drawdown induced two peaks of CO_2 emissions. The first peak appeared immediately after water table drawdown, followed by the second peak after about 30 days. The low ratio of CO_2 emission/gross ammonification under flooded condition seems to be caused by fast turnover of the microbial biomass-N and low CO_2 production. Water table drawdown increased SO_4^{2-} but decrease Fe, DON and DOC concentrations in soil pore water. Productions of DON and DOC were inhibited by increasing SO_4^{2-} concentrations. The effect of drainage on solutes concentrations is reversible within a month period when the soil is flooded. Under field conditions, neither drainage nor flooding had an effect on dissolved inorganic N due to the low concentrations, indicating the rapid consumption of mineral N. Contradictory, NH_4^+ increased during the flooding period in the absence of plant uptake and runoff in the laboratory experiment.

5. Conclusion

In the forest soil, soil drying has negative effects on gross ammonification and nitrification but a substantial rate of ammonification can be expected even at low water potential. During the natural rewetting no rewetting pulse of gross ammonification or nitrification was observed. . Rewetting of dry soil did not induce any pulse of DIN, DON and DOC fluxes in the soil. Overall, an increasing frequency of drying/rewetting cycles seem to have only moderate effect on the N turnover and on N solute fluxes in forest soils.

The response of fen soils to fluctuations of water table is element specific and characterized by different time scales. Drainage increases SO_4^{2-} but decrease Fe^{2+}, DON and DOC concentrations. Acidification of fen soils by oxidation of S to SO_4^{2-} can be expected when the water table dropped. The ratios of CO_2 emission/gross ammonification are very low under anoxic conditions which seem to be caused by the fast N turnover in the microbial biomass pool and low rates of CO_2 production. The effect of drainage on solutes concentrations and gross N turnover in fen soils are mostly reversible within a month period when the soil is flooded. Short term fluctuations at a daily scale will have little effect on N turnover as

compared to longer term changes on a monthly scale, while short term changes seem to trigger C losses by CO_2.

References

Aciego Pietri, J.C., and P.C. Brookes. 2008. Nitrogen mineralisation along a pH gradient of a silty loam UK soil. Soil Biology and Biochemistry 40:797-802.

Adu, J.K., and J.M. Oades. 1978. Physical factors influencing decomposition of organic materials in soil aggregates. Soil Biology and Biochemistry 10:109-115.

Ambus, P., A. Mosier, and S. Christensen. 1992. Nitrogen turnover rates in a riparian fen determined by ^{15}N dilution. Biology and Fertility of Soils 14:230-236.

Aulakh, M.S., T.S. Khera, and J.W. Doran. 2000. Mineralization and denitrification in upland, nearly saturated and flooded subtropical soil - I. Effect of nitrate and ammoniacal nitrogen. Biology and Fertility of Soils 31:162-167.

Barraclough, D. 1991. The use of mean pool abundances to interpret ^{15}N tracer experiments. Plant and Soil 131:89-96.

Barraclough, D., and G. Puri. 1995. The use of ^{15}N pool dilution and enrichment to separate the heterotrophic and autotrophic pathways of nitrification. Soil Biology and Biochemistry 27:17-22.

Bayley, S.E., M.N. Thormann, and A.R. Szumigalski. 2005. Nitrogen mineralization and decomposition in western boreal bog and fen peat. Ecoscience 12:455-465.

Bechtold, J.S., and R.J. Naiman. 2006. Soil texture and nitrogen mineralization potential across a riparian toposequence in a semi-arid savanna. Soil Biology and Biochemistry 38:1325-1333.

Birch, H.F. 1958. The effect of soil drying on humus decomposition and nitrogen availability. Plant and Soil 10:9-31.

Bjarnason, S. 1988. Calculation of gross nitrogen immobilization and mineralization in soil. Journal of Soil Science 39:393-406.

Bliss, C.M., N.B. Comerford, and R.M. Muchovej. 2004. Determination of microbial phosphorus *K*p factors in a spodosol: influence of extractant, water potential, and soil

horizon. Soil Biology and Biochemistry 36:1925-1934.

Blodau, C., N. Basiliko, and T.R. Moore. 2004. Carbon turnover in peatland mesocosms exposed to different water table levels. Biogeochemistry 67:331-351.

Booth, M.S., J.M. Stark, and E. Rastetter. 2005. Controls on nitrogen cycling in terrestrial ecosystems: a synthetic analysis of literature data. Ecological Monographs 75:139-157.

Borken, W., and E. Matzner. 2009. Reappraisal of drying and wetting effects on C and N mineralization and fluxes in soils. Global Change Biology 15:808-824.

Borken, W., Y.J. Xu, R. Brumme, and N. Lamersdorf. 1999. A climate change scenario for carbon dioxide and dissolved organic carbon fluxes from a temperateforest soil drought and rewetting effects. Soil Science Society of America Journal 63:1848-1855.

Bowden, W.B. 1986. Nitrification, nitrate reduction, and nitrogen immobilization in a tidal fresh-water marsh sediment. Ecology 67:88-99.

Brüggemann, N., P. Rosenkranz, H. Papen, K. Pilegaard, and K. Butterbach-Bahl. 2005. Pure stands of temperate forest tree species modify soil respiration and N turnover. Biogeosciences Discussions 2:303-331.

Breuer, L., R. Kiese, and K. Butterbach-Bahl. 2002. Temperature and moisture effects on nitrification rates in tropical rain-forest soils. Soil Science Society of America Journal 66:834-844.

Briones, M.J.I., P. Ineson, and T.G. Piearce. 1997. Effects of climate change on soil fauna; responses of enchytraeids, Diptera larvae and tardigrades in a transplant experiment. Applied Soil Ecology 6:117-134.

Burgin, A.J., and S.K. Hamilton. 2008. NO_3^--driven SO_4^{2-} production in freshwater ecosystems: Implications for N and S cycling. Ecosystems 11:908-922.

Burton, J., C. Chen, Z. Xu, and H. Ghadiri. 2007. Gross nitrogen transformations in adjacent native and plantation forests of subtropical Australia. Soil Biology and Biochemistry 39:426-433.

Campbell, J.L., and S.T. Gower. 2000. Detritus production and soil N transformations in old-growth eastern hemlock and sugar maple stands. Ecosystems 3:185-192.

Cayuela, M.L., T. Sinicco, and C. Mondini. 2009. Mineralization dynamics and biochemical

properties during initial decomposition of plant and animal residues in soil. Applied Soil Ecology 41:118-127.

Christenson, L.M., G.M. Lovett, K.C. Weathers, and M.A. Arthur. 2009. The influence of tree species, nitrogen fertilization, and soil C to N ratio on gross soil nitrogen transformations. Soil Science Society of America Journal 73:638-646.

Clark, J.M., P.J. Chapman, J.K. Adamson, and S.N. Lane. 2005. Influence of drought-induced acidification on the mobility of dissolved organic carbon in peat soils. Global Change Biology 11:791-809.

Corre, M.D., and N.P. Lamersdorf. 2004. Reversal of nitrogen saturation after long-term deposition reduction: impact on soil nitrogen cycling. Ecology 85:3090-3104.

Corre, M.D., R. Brumme, E. Veldkamp, and F.O. Beese. 2007. Changes in nitrogen cycling and retention processes in soils under spruce forests along a nitrogen enrichment gradient in Germany. Global Change Biology 13:1509-1527.

Corstanje, R., and K.R. Reddy. 2004. Response of biogeochemical indicators to a drawdown and subsequent reflood. Journal of Environmental Quality 33:2357-2366.

Danevčič, T., I. Mandic-Mulec, B. Stres, D. Stopar, and J. Hacin. 2010. Emissions of CO_2, CH_4 and N_2O from Southern European peatlands. Soil Biology and Biochemistry 42:1437-1446.

Davidson, E.A., S.C. Hart, and M.K. Firestone. 1992. Internal cycling of nitrate in soils of a mature coniferous forest. Ecology 73:1148-1156.

Davidson, E.A., E. Belk, and R.D. Boone. 1998. Soil water content and temperature as independent or confounded factors controlling soil respiration in a temperate mixed hardwood forest. Global Change Biology 4:217-227.

Davidson, E.A., S.C. Hart, C.A. Shanks, and M.K. Firestone. 1991. Measuring gross nitrogen mineralization, immobilization, and nitrification by ^{15}N isotopic pool dilution in intact soil cores. Journal of Soil Science 42:335-349.

Doerr, S.H., C.J. Ritsema, L.W. Dekker, D.F. Scott, and D. Carter. 2007. Water repellence of soils: new insights and emerging research needs. Hydrological Processes 21:2223-2228.

Driscoll, C.T., R.D. Fuller, and W.D. Schecher. 1989. The role of organic acids in the

acidification of surface waters in the Eastern U.S. Water, Air, and Soil Pollution 43:21-40.

Eimers, M.C., P.J. Dillon, S.L. Schiff, and D.S. Jeffries. 2003. The effects of drying and re-wetting and increased temperature on sulphate release from upland and wetland material. Soil Biology and Biochemistry 35:1663-1673.

Fenner, N., C. Freeman, and B. Reynolds. 2005. Hydrological effects on the diversity of phenolic degrading bacteria in a peatland: implications for carbon cycling. Soil Biology and Biochemistry 37:1277-1287.

Fierer, N., J.P. Schimel, and P.A. Holden. 2003. Variations in microbial community composition through two soil depth profiles. Soil Biology and Biochemistry 35:167-176.

Foken, T. 2003. Lufthygienisch-Bioklimatische Kennzeichnung des oberen Egertales. Bayreuther Forum Ökologie, 100, 1-118.

Ford, D.J., W.R. Cookson, M.A. Adams, and P.F. Grierson. 2007. Role of soil drying in nitrogen mineralization and microbial community function in semi-arid grasslands of north-west Australia. Soil Biology and Biochemistry 39:1557-1569.

Freeman, C., N. Fenner, N.J. Ostle, H. Kang, D.J. Dowrick, B. Reynolds, M.A. Lock, D. Sleep, S. Hughes, and J. Hudson. 2004. Export of dissolved organic carbon from peatlands under elevated carbon dioxide levels. Nature 430:195-198.

Fritze, H., J. Pietikäinen, and T. Pennanen. 2000. Distribution of microbial biomass and phospholipid fatty acids in Podzol profiles under coniferous forest. European Journal of Soil Science 51:565-573.

Gelfand, I., and D. Yakir. 2008. Influence of nitrite accumulation in association with seasonal patterns and mineralization of soil nitrogen in a semi-arid pine forest. Soil Biology and Biochemistry 40:415-424.

Gleeson, D.B., A.M. Herrmann, S.J. Livesley, and D.V. Murphy. 2008. Influence of water potential on nitrification and structure of nitrifying bacterial communities in semiarid soils. Applied Soil Ecology 40:189-194.

Goldberg, S.D., K.H. Knorr, and G. Gebauer. 2008. N_2O concentration and isotope signature along profiles provide deeper insight into the fate of N_2O in soils. Isotopes in Environmental and Health Studies 44:377-391.

Grenon, F., R.L. Bradley, and B.D. Titus. 2004. Temperature sensitivity of mineral N transformation rates, and heterotrophic nitrification: possible factors controlling the post-disturbance mineral N flush in forest floors. Soil Biology and Biochemistry 36:1465-1474.

Hart, S.C., D. Binkley, and D.A. Perry. 1997. Influence of red alder on soil nitrogen transformations in two conifer forests of contrasting productivity. Soil Biology and Biochemistry 29:1111-1123.

Hefting, M., J.C. Clément, D. Dowrick, A.C. Cosandey, S. Bernal, C. Cimpian, A. Tatur, T.P. Burt, and G. Pinay. 2004. Water table elevation controls on soil nitrogen cycling in riparian wetlands along a European climatic gradient. Biogeochemistry 67:113-134.

Hentschel, K., W. Borken, and E. Matzner. 2007. Leaching losses of inorganic N and DOC following repeated drying and wetting of a spruce forest soil. Plant and Soil 300:21-34.

Hruška, J., P. Krám, W.H. McDowell, and F. Oulehle. 2009. Increased dissolved organic carbon (DOC) in central European streams is driven by reductions in ionic strength rather than climate change or decreasing acidity. Environmental Science and Technology 43:4320-4326.

IPCC, 2007. Climate Change 2007: Synthesis report. Contribution of Working Groups I, II and III to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. IPCC, Geneva, Switzerland.

IUSS Working Group WRB, 2006. World reference base for soil resources 2006. 2nd edition. World Soil Resources Report, 103, FAO, Rome.

Jaatinen, K., R. Laiho, A. Vuorenmaa, U. del Castillo, K. Minkkinen, T. Pennanen, T. Penttilä, and H. Fritze. 2008. Responses of aerobic microbial communities and soil respiration to water-level drawdown in a northern boreal fen. Environmental Microbiology 10:339-353.

Jamieson, N., D. Barraclough, M. Unkovich, and R. Monaghan. 1998. Soil N dynamics in a natural calcareous grassland under a changing climate. Biology and Fertility of Soils 27:267-273.

Jury, W.A., and R. Horton. 2004. Soil Physics. 6 ed. John Wiley and Sons, Inc., Hoboken, New Jersey, America.

Kalbitz, K., and K. Kaiser. 2008. Contribution of dissolved organic matter to carbon storage in forest mineral soils. Journal of Plant Nutrition and Soil Science 171:52-60.

Keller, J.K., J.R. White, S.D. Bridgham, and J. Pastor. 2004. Climate change effects on carbon and nitrogen mineralization in peatlands through changes in soil quality. Global Change Biology 10:1053-1064.

Khalil, K., B. Mary, and P. Renault. 2004. Nitrous oxide production by nitrification and denitrification in soil aggregates as affected by O_2 concentration. Soil Biology and Biochemistry 36:687-699.

Kieckbusch, J.J., and J. Schrautzer. 2007. Nitrogen and phosphorus dynamics of a re-wetted shallow-flooded peatland. Science of the Total Environment 380:3-12.

Kiese, R., H. Papen, E. Zumbusch, and K. Butterbach-Bahl. 2002. Nitrification activity in tropical rain forest soils of the Coastal Lowlands and Atherton Tablelands, Queensland, Australia. Journal of Plant Nutrition and Soil Science 165:682-685.

Kirkham, D., and W.V. Bartholomew. 1954. Equations for following nutrient transformations in soil, utilizing tracer data. Soil Science Society of America Proceedings 18:33-34.

Knorr, K.H., and C. Blodau. 2009. Impact of experimental drought and rewetting on redox transformations and methanogenesis in mesocosms of a northern fen soil. Soil Biology and Biochemistry 41:1187-1198.

Knorr, K.H., M.R. Oosterwoud, and C. Blodau. 2008a. Experimental drought alters rates of soil respiration and methanogenesis but not carbon exchange in soil of a temperate fen. Soil Biology and Biochemistry 40:1781-1791.

Knorr, K.H., B. Glaser, and C. Blodau. 2008b. Fluxes and ^{13}C isotopic composition of dissolved carbon and pathways of methanogenesis in a fen soil exposed to experimental drought. Biogeosciences 5:1457-1473.

Knorr, K.H., G. Lischeid, and C. Blodau. 2009. Dynamics of redox processes in a minerotrophic fen exposed to a water table manipulation. Geoderma 153:379-392.

Kooijman, A.M., J.M. van Mourik, and M.L.M. Schilder. 2009. The relationship between N mineralization or microbial biomass N with micromorphological properties in beech forest soils with different texture and pH. Biology and Fertility of Soils 45:449-459.

Kraigher, B., B. Stres, J. Hacin, L. Ausec, I. Mahne, J.D. van Elsas, and I. Mandic-Mulec. 2006. Microbial activity and community structure in two drained fen soils in the Ljubljana Marsh. Soil Biology and Biochemistry 38:2762-2771.

Kresović, M., M. Jakovljević, S. Blagojević, and S. Maksimović. 2009. Specific transformations of mineral forms of nitrogen in acid soils. Journal of the Serbian Chemical Society 74:93-102.

Lamersdorf, N.P., C. Beier, K. Blanck, M. Bredemeier, T. Cummins, E.P. Farrell, K. Kreutzer, L. Rasmussen, M. Ryan, W. Weis, and Y.-J. Xu. 1998. Effect of drought experiments using roof installations on acidification/nitrification of soils. Forest Ecology and Management 101:95-109.

Liu, X.J., A.R. Mosier, A.D. Halvorson, C.A. Reule, and F.S. Zhang. 2007. Dinitrogen and N_2O emissions in arable soils: Effect of tillage, N source and soil moisture. Soil Biology and Biochemistry 39:2362-2370.

Low, A.P., J.M. Stark, and L.M. Dudley. 1997. Effects of soil osmotic potential on nitrification, ammonification, N-assimilation, and nitrous oxide production. Soil Science 162:16-27.

Lundquist, E.J., L.E. Jackson, and K.M. Scow. 1999. Wet-dry cycles affect dissolved organic carbon in two California agricultural soils. Soil Biology and Biochemistry 31:1031-1038.

Luxhøi, J., and L.S. Jensen. 2005. Gross N mineralization-immobilization rates in heterogeneous intact soil cores can be estimated without marked error. Biology and Fertility of Soils 41:280-283.

Luxhøi, J., S. Recous, I.R.P. Fillery, D.V. Murphy, and L.S. Jensen. 2005. Comparison of $^{15}NH_4^+$ pool dilution techniques to measure gross N fluxes in a coarse textured soil. Soil Biology and Biochemistry 37:569-572.

Münchmeyer, U., R. Russow, and J. Augustin. 2000. Net and gross nitrogen mineralization in drained and reflooded fen soils. Isotopes in Environmental and Health Studies 36:79-98.

Mataix-Solera, J., V. Arcenegui, C. Guerrero, A.M. Mayoral, J. Morales, J. González, F. García-Orenes, and I. Gómez. 2007. Water repellency under different plant species in a calcareous forest soil in a semiarid Mediterranean environment. Hydrological Processes 21:2300-2309.

Matejek, B., M. Kohlpaintner, R. Gasche, C. Huber, M. Dannenmann, and H. Papen. 2008. The small-scale pattern of seepage water nitrate concentration in an N saturated spruce forest is regulated by net N mineralization in the organic layer. Plant and Soil 310:167-

179.

Matejek, B., C. Huber, M. Dannenmann, M. Kohlpaintner, R. Gasche, A. Göttlein, and H. Papen. 2010. Microbial nitrogen-turnover processes within the soil profile of a nitrogen-saturated spruce forest and their relation to the small-scale pattern of seepage-water nitrate. Journal of Plant Nutrition and Soil Science 173:224-236.

McIntyre, R.E.S., M.A. Adams, D.J. Ford, and P.F. Grierson. 2009. Rewetting and litter addition influence mineralisation and microbial communities in soils from a semi-arid intermittent stream. Soil Biology and Biochemistry 41:92-101.

Mentzer, J., R. Goodman, and T. Balser. 2006. Microbial response over time to hydrologic and fertilization treatments in a simulated wet prairie. Plant and Soil 284:85-100.

Michalzik, B., K. Kalbitz, J.-H. Park, S. Solinger, and E. Matzner. 2001. Fluxes and concentrations of dissolved organic carbon and nitrogen– a synthesis for temperate forests. Biogeochemistry 52:173-205.

Mikha, M.M., C.W. Rice, and G.A. Milliken. 2005. Carbon and nitrogen mineralization as affected by drying and wetting cycles. Soil Biology and Biochemistry 37:339-347.

Muhr, J., J. Franke, and W. Borken. 2010. Drying-rewetting events reduce C and N losses from a Norway spruce forest floor. Soil Biology and Biochemistry 42:1303-1312.

Muhr, J., J. Höhle, D.O. Otieno, and W. Borken. 2011. Manipulative lowering of the water table during summer does not affect CO_2 emissions and uptake in a minerotrophic fen in South-eastern Germany. Ecological Applications.

Murphy, D.V., I.R.P. Fillery, and G.P. Sparing. 1997. Method to label soil cores with $^{15}NH_3$ gas as a prerequisite for ^{15}N isotopic dilution and measurement of gross N mineralization. Soil Biology and Biochemistry 29:1731-1741.

Murphy, D.V., A. Bhogal, M. Shepherd, K.W.T. Goulding, S.C. Jarvis, D. Barraclough, and J.L. Gaunt. 1999. Comparison of ^{15}N labelling methods to measure gross nitrogen mineralisation. Soil Biology and Biochemistry 31:2015-2024.

Murphy, D.V., S. Recous, E.A. Stockdale, I.R.P. Fillery, L.S. Jensen, D.J. Hatch, and K.W.T. Goulding. 2003. Gross nitrogen fluxes in soil: Theory, measurement and application of N-15 pool dilution techniques. Advances in Agronomy 79:69-118.

Neill, C. 1995. Seasonal flooding, nitrogen mineralization and nitrogen utilization in a prairie marsh. Biogeochemistry 30:171-189.

Nishio, T., T. Kanamori, and T. Fujimoto. 1985. Nitrogen transformations in an aerobic soil as determined by a $^{15}NH_4^+$ dilution technique. Soil Biology and Biochemistry 17:149-154.

Oechel, W.C., G.L. Vourlitis, S.J. Hastings, R.P. Ault, and P. Bryant. 1998. The effects of water table manipulation and elevated temperature on the net CO_2 flux of wet sedge tundra ecosystems. Global Change Biology 4:77-90.

Pal, L., B. Stres, T. Danevčič, S. Leskovec, and I. Mandic-Mulec. 2010. Transformations of mineral nitrogen applied to peat soil during sequential oxic/anoxic cycling. Soil Biology and Biochemistry 42:1338-1346.

Pandey, C.B., R.C. Srivastava, and R.K. Singh. 2009. Soil nitrogen mineralization and microbial biomass relation, and nitrogen conservation in humid-tropics. Soil Science Society of America Journal 73:1142-1149.

Parr, J.F., W.R. Gardner, and L.F. Elliott. 1985. Water potential relations in soil microbiology Soil science society of America, Madison, Wisconsin, America.

Pastor, J., J. Solin, S.D. Bridgham, K. Updegraff, C. Harth, P. Weishampel, and B. Dewey. 2003. Global warming and the export of dissolved organic carbon from boreal peatlands. Oikos 100:380-386.

Paul, S., K. Küsel, and C. Alewell. 2006. Reduction processes in forest wetlands: Tracking down heterogeneity of source/sink functions with a combination of methods. Soil Biology and Biochemistry 38:1028-1039.

Peltoniemi, K., H. Fritze, and R. Laiho. 2009. Response of fungal and actinobacterial communities to water-level drawdown in boreal peatland sites. Soil Biology and Biochemistry 41:1902-1914.

Peters, V., and R. Conrad. 1996. Sequential reduction processes and initiation of CH_4 production upon flooding of oxic upland soils. Soil Biology and Biochemistry 28:371-382.

Pett-Ridge, J., W.L. Silver, and M.K. Firestone. 2006. Redox fluctuations frame microbial community impacts on N-cycling rates in a humid tropical forest soil. Biogeochemistry 81:95-110.

Pinay, G., J.C. Clément, and R.J. Naiman. 2002. Basic principles and ecological consequences of changing water regimes on nitrogen cycling in fluvial systems. Environmental Management 30:481-491.

Pinay, G., B. Gumiero, E. Tabacchi, O. Gimenez, A.M. Tabacchi-Planty, M.M. Hefting, T.P. Burt, V.A. Black, C. Nilsson, V. Iordache, F. Bureau, L. Vought, G.E. Petts, and H. Décamps. 2007. Patterns of denitrification rates in European alluvial soils under various hydrological regimes. Freshwater Biology 52:252-266.

Pinheiro, J.C. and Bates, D.M. 2000. Mixed-Effects Models in S and S-Plus. Springer, New York.

Pinheiro, J., Bates, D., DebRoy, S., Sarkar, D. and The R Core team 2009. nlme: Linear and Nonlinear Mixed Effects Models. R package version 3.1-96.

Pulleman, M., and A. Tietema. 1999. Microbial C and N transformations during drying and rewetting of coniferous forest floor material. Soil Biology and Biochemistry 31:275-285.

R Development Core Team 2010. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna.

Recous, S., B. Mary, and G. Faurie. 1990. Microbial immobilization of ammonium and nitrate in cultivated soils. Soil Biology and Biochemistry 22:913-922.

Saetre, P., and J.M. Stark. 2005. Microbial dynamics and carbon and nitrogen cycling following re-wetting of soils beneath two semi-arid plant species. Oecologia 142:247-260.

Scheu, S., and D. Parkinson. 1994. Changes in bacterial and fungal biomass-C, bacterial and fungal biovolume and ergosterol content after drying, remoistening and incubation of different layers of cool temperate forest soils. Soil Biology and Biochemistry 26:1515-1525.

Schiff, S.L., J. Spoelstra, R.G. Semkin, and D.S. Jeffries. 2005. Drought induced pulses of SO_4^{2-} from a Canadian shield wetland: use of $\delta^{34}S$ and $\delta^{18}O$ in SO_4^{2-} to determine sources of sulfur. Applied Geochemistry 20:691-700.

Schimel, J., T.C. Balser, and M. Wallenstein. 2007. Microbial stress-response physiology and its implications for ecosystem function. Ecology 88:1386-1394.

Schimel, J.P., L.E. Jackson, and M.K. Firestone. 1989. Spatial and temporal effects on plant-

microbial competition for inorganic nitrogen in a california annual grassland. Soil Biology and Biochemistry 21:1059-1066.

Schmitt, A., B. Glaser, W. Borken, and E. Matzner. 2008. Repeated freeze-thaw cycles changed organic matter quality in a temperate forest soil. Journal of Plant Nutrition and Soil Science 171:707-718.

Schulze, K., W. Borken, J. Muhr, and E. Matzner. 2009. Stock, turnover time and accumulation of organic matter in bulk and density fractions of a Podzol soil. European Journal of Soil Science 60:567-577.

Scott, M.J., M.N. Jones, C. Woof, and E. Tipping. 1998. Concentrations and fluxes of dissolved organic carbon in drainage water from an upland peat system. Environment International 24:537-546.

Seneviratne, R., and A. Wild. 1985. Effect of mild drying on the mineralization of soil nitrogen. Plant and Soil 84:175-179.

Silvola, J., J. Alm, U. Ahlholm, H. Nykänen, and P.J. Martikainen. 1996. CO_2 fluxes from peat in boreal mires under varying temperature and moisture conditions. Journal of Ecology 84:219-228.

Stange, C.F., O. Spott, B. Apelt, and R.W.B. Russow. 2007. Automated and rapid online determination of ^{15}N abundance and concentration of ammonium, nitrite, or nitrate in aqueous samples by the SPINMAS technique. Isotopes in Environmental and Health Studies 43:227-236.

Stark, J.M., and M.K. Firestone. 1995. Mechanisms for soil moisture effects on activity of nitrifying bacteria. Applied and Environmental Microbiology 61:218-221.

Stark, J.M., and S.C. Hart. 1997. High rates of nitrification and nitrate turnover in undisturbed coniferous forests. Nature 385:61-64.

Strack, M., J.M. Waddington, R.A. Bourbonniere, E.L. Buckton, K. Shaw, P. Whittington, and J.S. Price. 2008. Effect of water table drawdown on peatland dissolved organic carbon export and dynamics. Hydrological Processes 22:3373-3385.

Templer, P.H., W.L. Silver, J. Pett-Ridge, K.M. DeAngelis, and M.K. Firestone. 2008. Plant and microbial controls on nitrogen retention and loss in a humid tropical forest. Ecology 89:3030-3040.

Tietema, A., B. Warmerdam, E. Lenting, and L. Riemer. 1992. Abiotic factors regulating nitrogen transformations in the organic layer of acid forest soils: Moisture and pH. Plant and Soil 147:69-78.

Tietema, A., C. Beier, P.H.B. deVisser, B.A. Emmett, P. Gundersen, O.J. Kjonaas, and C.J. Koopmans. 1997. Nitrate leaching in coniferous forest ecosystems: The European field-scale manipulation experiments NITREX (nitrogen saturation experiments) and EXMAN (experimental manipulation of forest ecosystems). Global Biogeochemical Cycles 11:617-626.

Tipping, E., C. Woof, E. Rigg, A.F. Harrison, P. Ineson, K. Taylor, D. Benham, J. Poskitt, A.P. Rowland, R. Bol, and D.D. Harkness. 1999. Climatic influences on the leaching of dissolved organic matter from upland UK moorland soils, investigated by a field manipulation experiment. Environment International 25:83-95.

van Dijk, J., W.A.M. Didden, F. Kuenen, P.M. van Bodegom, H.A. Verhoef, and R. Aerts. 2009. Can differences in soil community composition after peat meadow restoration lead to different decomposition and mineralization rates? Soil Biology and Biochemistry 41:1717-1725.

Venterink, H.O., T.E. Davidsson, K. Kiehl, and L. Leonardson. 2002. Impact of drying and re-wetting on N, P and K dynamics in a wetland soil. Plant and Soil 243:119-130.

Verchot, L.V., Z. Holmes, L. Mulon, P.M. Groffman, and G.M. Lovett. 2001. Gross vs net rates of N mineralization and nitrification as indicators of functional differences between forest types. Soil Biology and Biochemistry 33:1889-1901.

Watson, C.J., G. Travers, D.J. Kilpatrick, A.S. Laidlaw, and E. O'Riordan. 2000. Overestimation of gross N transformation rates in grassland soils due to non-uniform exploitation of applied and native pools. Soil Biology and Biochemistry 32:2019-2030.

Westbrook, C.J., and K.J. Devito. 2004. Gross nitrogen transformations in soils from uncut and cut boreal upland and peatland coniferous forest stands. Biogeochemistry 68:33-50.

Willison, T.W., J.C. Baker, D.V. Murphy, and K.W.T. Goulding. 1998. Comparison of a wet and dry ^{15}N isotopic dilution technique as a short-term nitrification assay. Soil Biology and Biochemistry 30:661-663.

Wray, H.E., and S.E. Bayley. 2008. Nitrogen dynamics in floating and non-floating peatlands

in the Western Boreal Plain. Canadian Journal of Soil Science 88:697-708.

Xiang, S.R., A. Doyle, P.A. Holden, and J.P. Schimel. 2008. Drying and rewetting effects on C and N mineralization and microbial activity in surface and subsurface California grassland soils. Soil Biology and Biochemistry 40:2281-2289.

Yan, E.R., X.H. Wang, J.J. Huang, G.Y. Li, and W. Zhou. 2008. Decline of soil nitrogen mineralization and nitrification during forest conversion of evergreen broad-leaved forest to plantations in the subtropical area of Eastern China. Biogeochemistry 89:239-251.

Yan, E.R., X.H. Wang, M. Guo, Q. Zhong, W. Zhou, and Y.F. Li. 2009. Temporal patterns of net soil N mineralization and nitrification through secondary succession in the subtropical forests of eastern China. Plant and Soil 320:181-194.

Yin, S.X., D. Chen, L.M. Chen, and R. Edis. 2002. Dissimilatory nitrate reduction to ammonium and responsible microorganisms in two Chinese and Australian paddy soils. Soil Biology and Biochemistry 34:1131-1137.

Yu, S., and J.G. Ehrenfeld. 2009. The effects of changes in soil moisture on nitrogen cycling in acid wetland types of the New Jersey Pinelands (USA). Soil Biology and Biochemistry 41:2394-2405.

Zak, D.R., and D.F. Grigal. 1991. Nitrogen mineralization, nitrification and denitrification in upland and wetland ecosystems. Oecologia 88:189-196.

Zaman, M., and S.X. Chang. 2004. Substrate type, temperature, and moisture content affect gross and net N mineralization and nitrification rates in agroforestry systems. Biology and Fertility of Soils 39:269-279.

Study 1 - Effects of decreasing water potential on gross ammonification and nitrification in an acid coniferous forest soil

Yao-Te Chen[1)], Werner Borken[1)], Claus Florian Stange [2)] and Egbert Matzner[1)]

[1)] Soil Ecology, Bayreuth Center of Ecology and Environmental Research (BayCEER), University of Bayreuth, D-95440 Bayreuth, Germany

[2)] Soil Physics, Helmholtz Centre for Environmental Research, UFZ, D-06120 Halle/Saale, Germany

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Abstract

Changes in the soil water regime, predicted as a consequence of global climate change, might influence the N cycle in temperate forest soils. We investigated the effect of decreasing soil water potentials on gross ammonification and nitrification in different soil horizons of a Norway spruce forest and tested the hypotheses that i) gross rates are more sensitive to desiccation in the Oa and EA horizon as compared to the uppermost Oi/Oe horizon and ii) that gross nitrification is more sensitive than gross ammonification. Soil samples were adjusted by air drying to water potentials from about field capacity to around -1.0 MPa, a range that is often observed under field conditions at our site. Gross rates were measured using the ^{15}N pool dilution technique. To ensure that the addition of solute label to dry soils and the local rewetting does not affect the results by re-mineralization or preferential consumption of ^{15}N, we compared different extraction and incubation times.

T_0 times ranging from 10 to 300 min and incubation times of 48 h and 72 h did not influence the rates of gross ammonification and nitrification. Even small changes of water potential decreased gross ammonification and nitrification in the O horizon. In the EA horizon, gross nitrification was below detection limit and the response of the generally low rates of gross ammonification to decreasing water potentials was minor. In the Oi/Oe horizon gross ammonification and nitrification decreased from 37.5 to 18.3 mg N kg^{-1} soil d^{-1} and from 15.4 to 5.6 mg N kg^{-1} soil d^{-1} when the water potential decreased from field capacity to -0.8 MPa. In the Oa horizon gross ammonification decreased from 7.4 to 4.0 mg N kg^{-1} soil d^{-1} when the water potential reached -0.6 MPa. At such water potential nitrification almost ceased, while in the Oi/Oe horizon nitrification continued at a rather high level. Hence, only in the Oa horizon nitrification was more sensitive to desiccation than ammonification. Extended drought periods that might result from climate change will cause a reduction in gross N turnover rates in forest soils even at moderate levels of soil desiccation.

1. Introduction

The frequency and intensity of drought and soil desiccation are likely to increase in many areas as a result of global climate change (IPCC, 2007) which might influence the N turnover and fluxes of soils and ecosystems (Borken and Matzner, 2009). Several studies have investigated the influence of soil water on net N turnover (Owen et al., 2003; Tietema et al., 1992; Vernimmem et al., 2007). Net N ammonification and nitrification include two major processes: gross ammonification and gross nitrification on the one and microbial immobilization on the other side. To identify the response of specific processes to soil desiccation, gross rates need to be measured. Furthermore, several authors demonstrated that net N transformation rates considerably underestimate gross rates in forest soils (Campbell and Gower, 2000; Verchot et al., 2001; Zaman and Chang, 2004) and the response of net and gross rates to soil desiccation might be different. Many studies report the effect of soil water content rather than of water potential which makes it difficult to compare different soil horizons and different studies. Soil water potential is a better indicator for the water availability to soil microorganisms (Davidson et al., 1998; Gleeson et al., 2008). Decreasing microbial activity at low water potentials might result from dehydration of microorganisms (Ford et al., 2007) and from limited supply of substrates because of lower diffusion rates in the decreasing water filled pore space. Furthermore, Schimel et al. (2007) suggested that physiological stress resulting from decreasing water potentials forces microbes to accumulate osmolytes resulting in changing C and N fluxes between soil and microbes. In case of net nitrification, substrate supply seems to be the limiting factor at water potentials of > -0.6 MPa, whereas at water potentials of < -0.6 MPa, cellular dehydration of the microorganisms seem to be most important (Stark and Firestone, 1995). For N turnover, a third factor might be involved, the production of N rich osmolytes (Csonka, 1989) at low water potential. The intracellular NH_4^+ production might be used for osmolytes rather than NH_4^+ being released from the cell and measured as gross ammonification.

Only few studies dealt with soil moisture effects on gross N turnover rates. In forest floors the optimum gravimetric water content for gross N turnover was between 170 and 328% (Brüggemann et al., 2005, Stange 2007). The highest gross nitrification rate in the forest floor of two tropical rain forests was at a water filled pore space of 65% (Kiese et al., 2008). In a pasture soil, the optimum water potential for gross N mineralization and nitrification was -0.01 MPa (Zaman et al., 1999). After desiccation of a pasture soil, gross ammonification was

close to zero (<0.2 mg N kg^{-1} soil d^{-1}) at water potentials of -1.5 MPa (Murphy et al., 1997). Gross N mineralization in grassland soils reached very low rates (< 1.0 mg N kg^{-1} soil d^{-1}) at water potentials of -1.0 MPa (Jamieson et al., 1998).

Forest floors and mineral soils comprise different horizons, inhabited by specific microbial communities which cause horizon specific N turnover rates (Matejek et al., 2010). The ratio of fungi/bacteria decreases with soil depths in coniferous forest soils (Fritze et al., 2000; Schmitt et al., 2008). The reasons for that might be substrate quality in the different horizons and the competition between the microbial communities. Bacteria are known to be more sensitive to soil desiccation than fungi (Berg et al., 1998; Harris 1985; Schnürer et al., 1986). Therefore any response of microbial communities to soil desiccation should be horizon specific.

Bacteria and fungi both contribute to nitrification since nitrification in acid forest soils can be autotrophic and heterotrophic (Šantrůčková et al., 2009). The relative importance of acid-tolerant autotrophic ammonium-oxidizing bacteria to nitrification in relation to heterotrophic nitrification ranged from 8% to 100% (De Boer et al., 1992; Grenon et al., 2004; Venterea et al., 2003). Several studies revealed a positive relation between gross nitrification rates and gross ammonification rates, indicating that autotrophic nitrification is more dominant than heterotrophic nitrification in forest floors (Barraclough and Puri, 1995; Christenson et al., 2009; Corre et al., 2007). As a consequence, nitrification should be more sensitive to desiccation than ammonification, the latter being also realized by fungi. In fact, a higher sensitivity of net nitrification to soil desiccation is reported from experimental studies with soils of different land use (Hentschel et al., 2007; Xiang et al., 2008; Yahdjian and Sala, 2008), but information about gross rates is rare, especially for forest soils.

The ^{15}N pool dilution technique (Kirkham and Bartholomew, 1954) is widely used to assess gross N turnover rates in soils (e.g. Barraclough and Puri, 1995; Burton et al., 2007; Murphy et al., 1999). The method is based on two soil extractions and the measured changes in concentrations of NH_4^+ and NO_3^- and ^{15}N abundances. The extraction at T_0 is shortly after addition of the ^{15}N label to account for immediate N losses, abiotic reactions of applied ^{15}N and the initial mixture with $^{14}NH_4^+$ or $^{14}NO_3^-$ in the soil. After a specific incubation time the soil is extracted again (T_1) and the dilution of the solute ^{15}N pools is measured. When investigating N turnover of dry soils, the increasing water potential after application of the solute label might cause artifacts by rising N turnover rates as shown by Willison et al. (1998)

and Murphy et al. (1999). Hence, the definition of T_0 and T_1 is critical to ensure that the rates are constant after the application of the label.

Here we investigated the effect of decreasing soil water potentials on gross ammonification and nitrification in the O and EA horizons of an acid spruce forest soil. Nitrogen turnover in forest soils is higher in these horizons as compared to deeper soil horizons. Furthermore, the upper soil is subjected and possibly adapted to frequent desiccation.

Our goals were to investigate the influence of soil desiccation on gross N turnover and to test the hypothesis i) that gross rates are more sensitive to desiccation in the Oa and EA horizon as compared to the uppermost Oi/Oe horizon and ii) that gross nitrification is more sensitive to desiccation than gross ammonification. Lastly our goal was to clarify the influence of different extraction and incubation times on gross N turnover in desiccated soil samples.

2. Materials and methods

2.1 Study site

The Coulissenhieb II site is a mature 140-year-old Norway spruce forest (*Picea abies* L.) and located in the Lehstenbach catchment (4.2 km^2) in the Fichtelgebirge area, Germany (58°08'N, 11°52'E). Mean annual precipitation is 1160 mm and mean annual air temperature is 5.3 °C. The soil has a sandy to loamy texture and is classified as Haplic Podzol according to the FAO soil classification (IUSS, 2006). The well stratified, mor-like forest floor has a thickness of 7-10 cm, comprising Oi, Oe and Oa horizons. The litterfall in 2008 was 290 g m^{-2} y^{-1}. The C and N content of the Oi horizon is 46% and 1.7%, of the Oe horizon 42% and 1.8%, of the Oa horizon 21% and 1.1 % and of the EA horizon 8.3% and 0.4%. The pH($CaCl_2$) of the Oa is 3.3 and of the EA 3.4. Carbon and N stocks of the forest floor (Oi/Oe + Oa) are 5.0 kg C m^{-2} and 0.25 kg N m^{-2}, and in the EA horizon 2.4 kg C m^{-2} and 0.12 kg N m^{-2} (Schulze et al., 2009).

2.2 Influence of T_0 and T_1 on gross rates

Soil samples were taken from the Oi/Oe horizon in October 2008 and stored for 1 week at 2 °C before incubation. Samples were homogenized by hand; twigs, roots, branches and other

pieces of woody particles were removed. Gross ammonification and nitrification rates were determined at two different water potentials: high (field moist, -0.12 MPa) and low (about -1.0 MPa). The low water potential was adjusted by air drying at room temperature and measured by a dew point potentiometer (WP4-T, Decagon Devices, USA; accuracy of ±0.1 MPa). Gravimetric water contents of soils were determined by drying at 60 °C for 12 h.

Gross rates of ammonification and nitrification were determined by using the ^{15}N pool dilution technique (Kirkham and Bartholomew, 1954). To about 150 g fresh weight of soil, which was pre-incubated at 15°C for 2 days, 1.8 ml solution comprising in total 0.8 mg NH_4^+-^{15}N (98.0 at%) or 0.03 mg NO_3^--^{15}N (99.2 at%) was added by spraying the solution onto the soil. The soil was subsequently mixed by shaking in the polyethylene bottle. Following a T_0 of 10, 30, 60, 180 and 300 min after ^{15}N addition, subsamples of 5 g fresh weight were extracted in 100 ml polyethylene bottles with 1.0 M KCl solution at a soil/solution ratio of 1:10. All experiments were done in threefold replication at 15 °C. The KCl extractions were shaken for 1 h and the supernatant filtered (cellulose folded filters 595½, 4-7 µm, Whatman, Germany). Labelled samples were incubated for 48 h and 72 h (T_1) at 15 °C and then extracted in the same way as for T_0.

2.3 Effects of water potential on gross rates

Samples of the Oi/Oe, Oa and EA horizon were taken from Coulissenhieb II in December 2008 and adjusted to field capacity and homogenized. Six different water potentials ranging from field capacity to about -1.0 MPa were adjusted by air drying at room temperature (about 3 weeks) and measured by either tensiometer (> -0.2 MPa; Tensiometer T5x, UMS, Germany) or by dew point potentiometer (< -0.2 MPa; WP4, Decagon Devices, USA; accuracy of ±0.1 MPa). Gravimetric water contents were determined after 12 h drying at 60 °C in case of Oi/Oe and Oa samples and after 24 h drying at 105 °C in case of EA samples. Gross rates of N ammonification and nitrification were determined as described above with a T_0 of 1 h and a T_1 of 49 h. All experiments were done in threefold replication at 15 °C.

2.4 Sample analysis and gross rate calculation

Filtered KCl extracts were frozen at -20 °C and sent to the Helmholtz Centre for

Environmental Research (UFZ, Halle) using the SPINMAS technique (**S**ample **P**reparation unit for **I**norganic **N**itrogen and **MA**ss **S**pectrometer) for analysis of ^{15}N abundance and the concentrations of NO_3^- and NH_4^+ (Stange et al., 2007). The SPINMAS comprises a coupling of a specially developed sample preparation device with a continuous flow-quadrupole mass spectrometer (QMS GAM 400, InProcess Instruments, Germany). Based on pool sizes of NH_4^+, NO_3^- and their ^{15}N abundances, gross N ammonification and nitrification rates were calculated using the equation from Kirkham and Bartholomew (1954). For calculation of gross rates, ^{15}N abundances and concentrations one of the three T_0 were randomly pairwise related to one of the three T_1 values, resulting in 3 values for gross rates. Arithmetic means and standard errors were calculated using n = 3 using the software SIGMAPLOT. Differences between treatments were tested with a paired t-test using the software STATISTICA.

3. Results

3.1 Influence of T_0 and T_1 on gross rates

Gross ammonification rates were not affected by the different T_0 and incubation times. At -0.12 MPa, gross ammonification rates in the Oi/Oe horizon were about 39 mg N kg^{-1} soil d^{-1} (Fig. 1a). As expected, gross ammonification rates decreased at -1.0 MPa, to rates of 13.5 - 17.2 mg N kg^{-1} soil d^{-1} (Fig. 1b). At -1.0 MPa, gross ammonification rates determined at T_1 = 72 h were slightly smaller than those at T_1 = 48 h (Fig. 1b). The variation of gross nitrification rates among the replicates was very high as indicated by the large standard errors. However, rates were generally not influenced by the different T_0 and T_1 (Fig. 1c and 1d). Gross nitrification rates ranged from 5.6 to 13.6 mg N kg^{-1} soil d^{-1} at -0.12 MPa and from 5.2 to 11.8 mg N kg^{-1} soil d^{-1} at -1.0 MPa, differences not being statistically significant at the $p < 0.05$ level.

3.2 Effects of water potential on gross rates

As expected, the highest gross ammonification rates were observed at field capacity (Fig. 2). Maximum rates were found in the Oi/Oe horizon reaching 38 mg N kg^{-1} soil d^{-1}. Gross ammonification rates were much lower in the Oa horizon with a maximum of 7.4 mg N kg^{-1} soil d^{-1}. Lowest rates were observed in the EA horizon with a maximum of 1.4 mg N kg^{-1} soil

d^{-1}. Soil desiccation caused an immediate decrease of the gross rates in the Oi/Oe horizon. Surprisingly, gross ammonification measured at -1.2 MPa was similar to that at -0.8 MPa. Gross ammonification in the Oa horizon decreased linearly when water potential dropped from field capacity to -0.6 MPa. Rates did not further decrease when water potential dropped to -0.8 MPa. In the EA horizon gross ammonification was generally low. Rates decreased from field capacity to -0.25 MPa, but there was no further response to decreasing water potential.

The highest gross nitrification rates were also observed at field capacity (Fig. 3). Maximum rates were found in the Oi/Oe horizon reaching 15.4 mg N kg^{-1} soil d^{-1}. Gross nitrification rates were much lower in the Oa horizon with a maximum of 5.5 mg N kg^{-1} soil d^{-1}. Gross nitrification was not detectable in the EA horizon. Soil desiccation caused an immediate decrease of nitrification in the Oi/Oe horizon. Rates at -1.2 MPa were similar to that at -0.8 MPa. Gross nitrification in the Oa horizon decreased linearly to almost zero when water potential dropped from field capacity to -0.8 MPa.

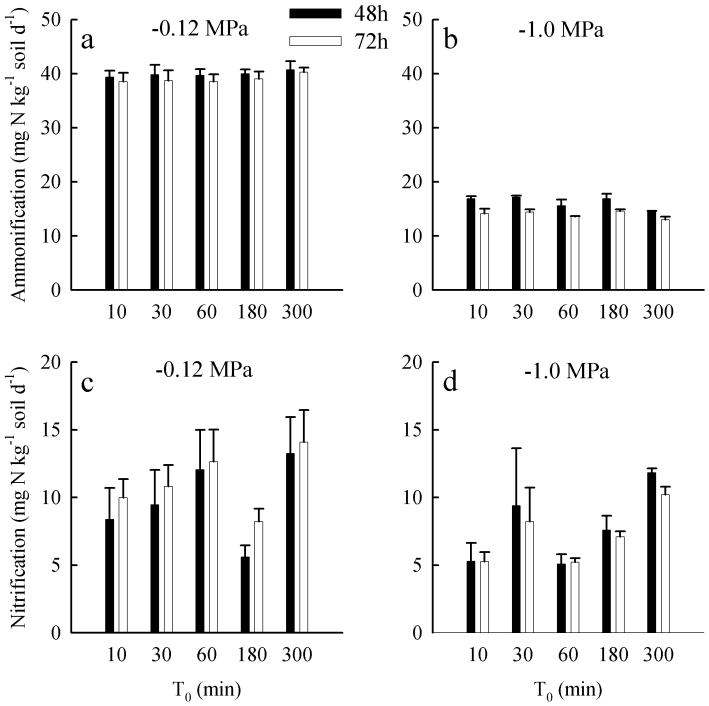

Fig.1 Gross rates of ammonification (a, b) and nitrification (c, d) measured in the Oi/Oe horizon at -0.12 MPa and -1.0 MPa (mean±SE, n=3).

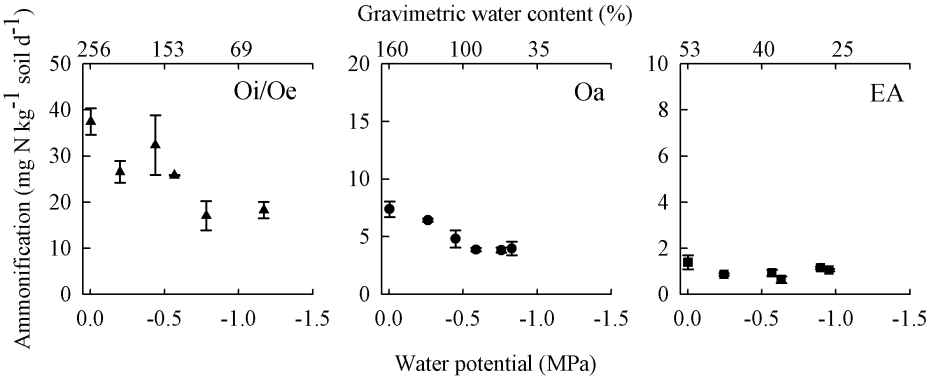

Fig. 2 Effect of water potential on gross rates of ammonification in the Oi/Oe, Oa and EA horizons in spruce forest (mean±SE, n=3).

The relative decrease of gross nitrification in relation to gross ammonification was similar in the Oi/Oe horizon, but the decrease of nitrification was steeper in the Oa horizon (Fig. 4). There was a strong linear relation between the rates of gross ammonification and nitrification over the whole range of water potentials (Fig. 5). Nitrification rates were always lower than ammonification rates.

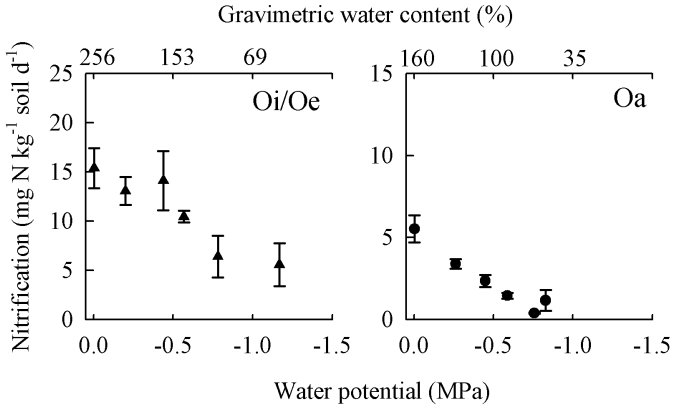

Fig. 3 Effect of water potential on gross rates of nitrification in the Oi/Oe and Oa horizons in spruce forest (mean±SE, n=3).

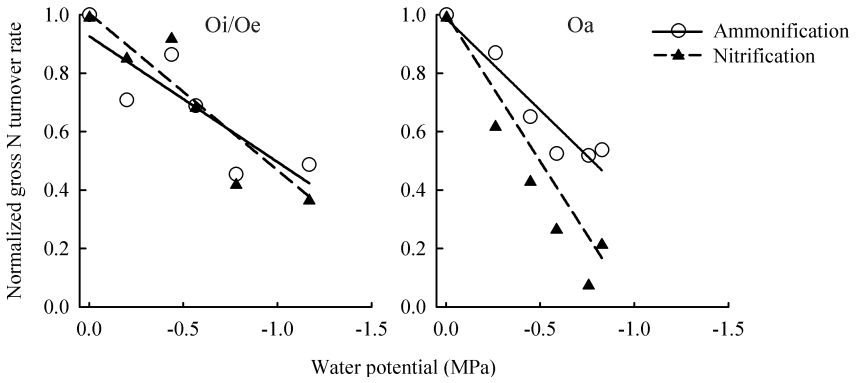

Fig. 4 Effect of water potential on normalized gross rates of ammonification and nitrification in the Oi/Oe and Oa horizons in spruce forest (mean±SE, n=3).

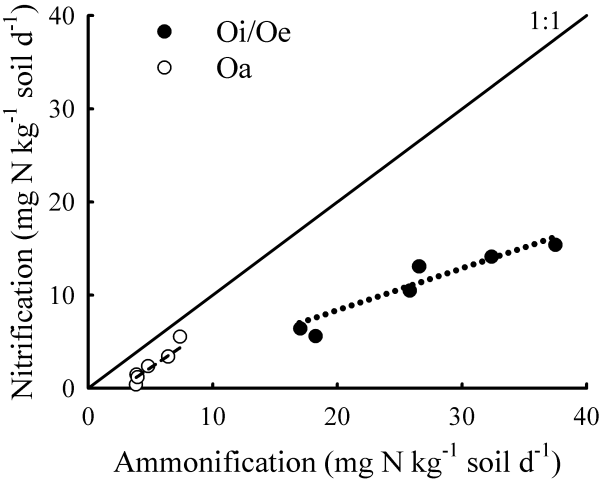

Fig. 5 Relationship between gross ammonification and nitrification in the Oi/Oe and Oa horizons.

4. Discussion

Gross ammonification and nitrification at both water potentials were generally not influenced by the different T_0 and T_1 indicating that the abiotic immobilization of ^{15}N does not play a role in the period from 10 to 300 min and no measureable re-mineralization occurs within 72 h incubation. The application of solute label to soils of low water potential could stimulate the microbial activity and might cause initial effects on the calculated gross rates at a time scale of hours to days (Murphy et al., 1997, Willison et al., 1998). Thus, a relatively higher gross turnover rate can be expected at 72 h than at 48 h incubation time. However, the application of small amounts of solution in our study did not change the water potential and no effect of different T_0 and T_1 on the gross rates was observed.

The addition of small amounts of solute label to soil of low water potential may result in an inhomogeneous distribution of the label, despite efforts to homogenize the soil. As a result, high concentrations of $^{15}NH_4^+$ occur in those parts that received the label. The diffusion of the added $^{15}NH_4^+$, its even distribution and mixing with the existing soil exchangeable $^{14}NH_4^+$ pool is supposed to be a fast process that should have finished between the time of label addition and the T_0 sampling. If the diffusion of label is limited by low water potentials, the equilibration with the soil $^{14}NH_4^+$ may take longer than at high water potential, which finally might cause an overestimation of the calculated gross ammonification rate. In case of acid forest soils this potential artefact of the ^{15}N pool dilution method should be more important for gross ammonification than for gross nitrification because of the larger soil pool of exchangeable $^{14}NH_4^+$ as compared to $^{14}NO_3^-$. Such artefacts should manifest themselves by increasing gross rates with increasing T_0 and T_1 times. Our results clearly show that this is not the case, at least at soil water potentials > -1.0 MPa.

Our experiments, using different soil horizons and water potentials, resulted in a wide range of N turnover rates. In general, gross ammonification in the O horizons was in a similar range as reported from other studies in forest soils (Brüggemann et al., 2005; Christenson et al., 2009; Corre et al., 2007). The same is true for gross nitrification (Burton et al., 2007; Sotta et al., 2008; Stange, 2007).

Gross N turnover rates might be larger in disturbed soils compared to intact soil cores because of the better substrate supply in disturbed samples (Luxhøi and Jensen, 2005). Our study was done with disturbed soil samples in order to avoid excessive problems with natural spatial

heterogeneity that is related to the use of undisturbed soil samples. We recently investigated effects of changing water potential on gross N turnover rates in a field experiment using intact soil cores (Chen et al. unpublished). Our results show that gross rates do not differ between disturbed soils and intact soil cores. The responses of gross rates to the decreasing water potential were also similar between these two techniques.

Even small changes in the water potential directly affected the gross N turnover rates. In agreement with our study (Fig. 4), an almost linear decrease of rates with decreasing water potential has also been found for net ammonification and net nitrification in a coniferous forest floor (Tietema et al., 1992). Low et al. (1997) reported that the reduction of gross ammonification in a pasture soil was best fitted by an exponential function when osmotic potential decreased from 0 to -0.5 MPa. They observed that gross ammonification did not respond to a further drop in water potential from -0.5 to -1.75 MPa, which is similar to the surprising lack of response of gross ammonification at water potentials < -0.6 MPa in the Oi/Oe and Oa horizons in our study. Using soil from our site, Muhr et al. (2010) observed a reduction in total CO_2 emissions by about 65% when water potential dropped from field capacity to -1.2 MPa, supporting the presence of substantial microbial activity at such water potentials.

High efficiency of N mineralization due to low N requirements of fungi may contribute to the relatively high net N mineralization in acid soils (Kooijman et al., 2009). Therefore, the reason for the lack of response to water potentials < -0.6 MPa might be the contribution of less drought sensitive fungi in relation to bacterial ammonification. This interpretation remains speculative since the relative contributions of both groups to gross ammonification at low water potentials are not clear.

According to Stark and Firestone (1995), cellular desiccation of microorganisms and the need to produce osmolytes should occur at water potentials of < -0.6 MPa. Bacterial osmolytes are often N rich compounds (Csonka, 1989). If their intracellular production results from the preferential immobilization of $^{15}NH_4$ from the added label over $^{14}NH_4$ (e.g. by better local availability), the ^{15}N label is diluted to greater extent than expected. Consequently, the calculated rates of gross ammonification would be overestimated. We have no proof for the existence of such an artefact, but were it to exist, it could potentially explain the constant rates of ammonification we observed at water potentials < -0.6 MPa, and would also create a potential shortcoming for using the ^{15}N pool dilution technique at low water potentials.

Gross ammonification was generally very low in the EA horizon with only a minor response to desiccation. This suggests a general substrate limitation in this horizon caused by the old and humified soil organic matter which is largely stabilized against enzymatic attack by associations with minerals (Schulze et al., 2009).

Like with gross ammonification, gross nitrification decreased with decreasing water potential in both horizons. But only in the Oa horizon nitrification was more sensitive to desiccation than ammonification. Gross nitrification almost ceased at -0.8 MPa, while ammonification was still considerable great (4.0 mg N kg^{-1} soil d^{-1}). Hence substrate limitation does not explain the cease of nitrification in the Oa horizon. The reason for the higher sensitivity of gross nitrification to desiccation might be the sensitivity of autotrophic nitrifying bacteria to desiccation, whereas ammonification, which is driven by a large variety of bacteria and drought tolerant fungi, is less sensitive. In contrast, nitrification in the Oi/Oe horizon continued with a considerable rate at -1.2 MPa, suggesting that nitrifiers are more tolerant to desiccation in the uppermost soil horizon than in the Oa horizon. It remains an open question if this is caused by a larger proportion of heterotrophic, drought resistant fungi in relation to bacterial autotrophic nitrification in the Oi/Oe horizon. This interpretation is supported by the observation of higher relative fungal biomass in the uppermost soil layers (Fierer et al., 2003; Fritze et al., 2000; Schmitt et al., 2008). Also Scheu and Parkinson (1994) observed a significant reduction in bacterial biomass in forest floor horizons by soil drying whereas the fungal biomass was less affected.

5. Conclusion

Our results show that even moderate soil desiccation has strong negative effects on gross N turnover rates. Moreover, soil desiccation effects on gross N turnover are horizon specific, probably caused by substrate quality and by different contributions of fungi and bacteria to ammonification and nitrification. The higher sensitivity of gross nitrification to desiccation in the Oa than in the Oi/Oe horizon might be due to a larger proportion of autotrophic nitrification in the Oa horizon. The implications of our findings for ecosystem functioning can be seen in a less N availability for plants and microorganisms in periods of moderate soil desiccation. Furthermore, if the duration and frequency of desiccation periods increase in a future climate, the changes of N cycling might reduce inorganic N availability for plants and

microorganisms.

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References

Barraclough, D., Puri, G., 1995. The use of ^{15}N pool dilution and enrichment to separate the heterotrophic and autotrophic pathways of nitrification. Soil Biology and Biochemistry 27, 17-22.

Berg, M.P., Kniese, J.P., Verhoef, H.A., 1998. Dynamics and stratification of bacteria and fungi in the organic layers of a Scots pine forest soil. Biology and Fertility of Soils 26, 313-322.

Borken, W., Matzner, E., 2009. Reappraisal of drying and wetting effects on C and N mineralization and fluxes in soils. Global Change Biology 15, 808-824.

Brüggemann, N., Rosenkranz, P., Papen, H., Pilegaard, K., Butterbach-Bahl, K., 2005. Pure stands of temperate forest tree species modify soil respiration and N turnover. Biogeosciences Discussions 2, 303-331.

Burton, J., Chen, C., Xu, Z., Ghadiri, H., 2007. Gross nitrogen transformations in adjacent native and plantation forests of subtropical Australia. Soil Biology and Biochemistry 39, 426-433.

Campbell, J.L., Gower, S.T., 2000. Detritus production and soil N transformations in old-growth eastern hemlock and sugar maple stands. Ecosystems 3, 185-192.

Christenson, L.M., Lovett, G.M., Weathers, K.C., Arthur, M.A., 2009. The influence of tree species, nitrogen fertilization, and soil C to N ratio on gross soil nitrogen transformations. Soil Science Society of America Journal 73, 638-646.

Corre, M.D., Brumme, R., Veldkamp, E., Beese, F.O., 2007. Changes in nitrogen cycling and retention processes in soils under spruce forests along a nitrogen enrichment gradient in Germany. Global Change Biology 13, 1509-1527.

Csonka, L.N., 1989. Physiological and genetic responses of bacteria to osmotic stress. Microbiological Reviews 53, 121-147.

Davidson, E.A., Belk, E., Boone, R.D., 1998. Soil water content and temperature as independent or confounded factors controlling soil respiration in a temperate mixed hardwood forest. Global Change Biology 4, 217-227.

De Boer, W., Tietema, A., Gunnewiek, P.J.A.K., Laanbroek, H.J., 1992. The chemolithotrophic ammonium-oxidizing community in a nitrogen-saturated acid forest soil in relation to pH-dependent nitrifying activity. Soil Biology and Biochemistry 24, 229-234.

Fierer, N., Schimel, J.P., Holden, P.A., 2003. Variations in microbial community composition through two soil depth profiles. Soil Biology and Biochemistry 35, 167-176.

Ford, D.J., Cookson, W.R., Adams, M.A., Grierson, P.F., 2007. Role of soil drying in nitrogen mineralization and microbial community function in semi-arid grasslands of north-west Australia. Soil Biology and Biochemistry 39, 1557-1569.

Fritze, H., Pietikäinen, J., Pennanen, T., 2000. Distribution of microbial biomass and phospholipid fatty acids in Podzol profiles under coniferous forest. European Journal of Soil Science 51, 565-573.

Gleeson, D.B., Herrmann, A.M., Livesley, S.J., Murphy, D.V., 2008. Influence of water potential on nitrification and structure of nitrifying bacterial communities in semiarid soils. Applied Soil Ecology 40, 189-194.

Grenon, F., Bradley, R.L., Titus, B.D., 2004. Temperature sensitivity of mineral N transformation rates, and heterotrophic nitrification: possible factors controlling the post-disturbance mineral N flush in forest floors. Soil Biology and Biochemistry 36, 1465-1474.

Harris, R.F., 1985. Effect of water potential on microbial growth and activity, in Parr, J.F., Gardner, W.R., Elliott, L.F. (Eds.), Water potential relations in soil microbiology. American Society of Agronomy, Madison, pp. 23–95.

Hentschel, K., Borken, W., Matzner, E., 2007. Leaching losses of inorganic N and DOC following repeated drying and wetting of a spruce forest soil. Plant and Soil 300, 21-34.

IPCC, 2007. Climate Change 2007: Synthesis report. Contribution of Working Groups I, II and III to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. IPCC, Geneva, Switzerland.

IUSS Working Group WRB, 2006. World reference base for soil resources 2006. 2nd edition. World Soil Resources Report No. 103, FAO, Rome

Jamieson, N., Barraclough, D., Unkovich, M., Monaghan, R., 1998. Soil N dynamics in a natural calcareous grassland under a changing climate. Biology and Fertility of Soils 27, 267-273.

Kiese, R., Hewett, B., Butterbach-Bahl, K., 2008. Seasonal dynamic of gross nitrification and N_2O emission at two tropical rainforest sites in Queensland, Australia. Plant and Soil 309, 105-117.

Kirkham, D., Bartholomew, W.V., 1954. Equations for following nutrient transformations in soil, utilizing tracer data. Soil Science Society of America Proceedings 18, 33-34.

Kooijman, A.M., van Mourik, J.M., Schilder, M.L.M., 2009. The relationship between N mineralization or microbial biomass N with micromorphological properties in beech forest soils with different texture and pH. Biology and Fertility of Soils 45, 449-459.

Low, A.P., Stark, J.M., Dudley, L.M., 1997. Effects of soil osmotic potential on nitrification, ammonification, N-assimilation, and nitrous oxide production. Soil Science 162, 16-27.

Luxhøi, J., Jensen, L.S., 2005. Gross N mineralization-immobilization rates in heterogeneous intact soil cores can be estimated without marked error. Biology and Fertility of Soils 41, 280-283.

Matejek, B., Huber, C., Dannenmann, M., Kohlpaintner, M., Gasche, R., Göttlein, A.,Papen, H., 2010. Microbial nitrogen-turnover processes within the soil profile of a nitrogen-saturated spruce forest and their relation to the small-scale pattern of seepage-water nitrate. Journal of Plant Nutrition and Soil Science 173, 224-236.

Muhr, J., Franke, J., Borken, W., 2010. Drying-rewetting events reduce C and N losses from a Norway spruce forest floor. Soil Biology and Biochemistry 42, 1303-1312.

Murphy, D.V., Fillery, I.R.P., Sparing, G.P., 1997. Method to label soil cores with $^{15}NH_3$ gas as a prerequisite for ^{15}N isotopic dilution and measurement of gross N mineralization. Soil Biology and Biochemistry 29, 1731-1741.

Murphy, D.V., Bhogal, A., Shepherd, M., Goulding, K.W.T., Jarvis, S.C., Barraclough, D., Gaunt, J.L., 1999. Comparison of ^{15}N labelling methods to measure gross nitrogen mineralisation. Soil Biology and Biochemistry 31, 2015-2024.

Owen, J.S., Wang, M.K., Wang, C.H., King, H.B., Sun, H.L., 2003. Net N mineralization and nitrification rates in a forested ecosystem in northeastern Taiwan. Forest Ecology and Management 176, 519-530.

Šantrůčková, H., Tahovská, K., Kopáček, J., 2009. Nitrogen transformations and pools in N-saturated mountain spruce forest soils. Biology and Fertility of Soils 45, 395-404.

Scheu, S., Parkinson, D., 1994. Changes in bacterial and fungal biomass-C, bacterial and fungal biovolume and ergosterol content after drying, remoistening and incubation of different layers of cool temperate forest soils. Soil Biology and Biochemistry 26, 1515-1525.

Schimel, J., Balser, T.C., Wallenstein, M., 2007. Microbial stress-response physiology and its implications for ecosystem function. Ecology 88, 1386-1394.

Schmitt, A., Glaser, B., Borken, W., Matzner, E., 2008. Repeated freeze-thaw cycles changed organic matter quality in a temperate forest soil. Journal of Plant Nutrition and Soil Science 171, 707-718.

Schnürer, J., Clarholm, M., Boström, S., Rosswall, T., 1986. Effects of moisture on soil microorganisms and nematodes: A field experiment. Microbial Ecology 12, 217-230.

Schulze, K., Borken, W., Muhr, J. Matzner, E., 2009. Stock, turnover time and accumulation of organic matter in bulk and density fractions of a Podzol soil. European Journal of Soil Science 60, 567-577.

Sotta, E.D., Corre, M.D., Veldkamp, E., 2008. Differing N status and N retention processes of soils under old-growth lowland forest in Eastern Amazonia, Caxiuanã, Brazil. Soil Biology and Biochemistry 40, 740-750.

Stange, C.F., 2007. A novel approach to combine response functions in ecological process modelling. Ecological Modelling 204, 547-552.

Stange, C.F., Spott, O., Apelt, B., Russow, R.W.B., 2007. Automated and rapid online determination of ^{15}N abundance and concentration of ammonium, nitrite, or nitrate in aqueous samples by the SPINMAS technique. Isotopes in Environmental and Health Studies 43, 227-236.

Stark, J.M., Firestone, M.K., 1995. Mechanisms for soil moisture effects on activity of nitrifying bacteria. Applied and Environmental Microbiology 61, 218-221.

Tietema, A., Warmerdam, B., Lenting, E., Riemer, L., 1992. Abiotic factors regulating nitrogen transformations in the organic layer of acid forest soils: Moisture and pH. Plant and Soil 147, 69-78.

Venterea, R.T., Groffman, P.M., Verchot, L.V., Magill, A.H., Aber, J.D., Steudler, P.A., 2003. Nitrogen oxide gas emissions from temperate forest soils receiving long-term nitrogen inputs. Global Change Biology 9, 346-357.

Verchot, L.V., Holmes, Z., Mulon, L., Groffman, P.M., Lovett, G.M., 2001. Gross vs net rates of N mineralization and nitrification as indicators of functional differences between forest types. Soil Biology and Biochemistry 33, 1889-1901.

Vernimmen, R.R.E., Verhoef, H.A., Verstraten, J.M., Bruijnzeel, L.A., Klomp, N.S., Zoomer, H.R., Wartenbergh, P.E., 2007. Nitrogen mineralization, nitrification and denitrification potential in contrasting lowland rain forest types in Central Kalimantan, Indonesia. Soil Biology and Biochemistry 39, 2992-3003.

Willison, T.W., Baker, J.C., Murphy, D.V., Goulding, K.W.T., 1998. Comparison of a wet and dry ^{15}N isotopic dilution technique as a short-term nitrification assay. Soil Biology and Biochemistry 30, 661-663.

Xiang, S.R., Doyle, A., Holden, P.A., Schimel, J.P., 2008. Drying and rewetting effects on C and N mineralization and microbial activity in surface and subsurface California grassland soils. Soil Biology and Biochemistry 40, 2281-2289.

Yahdjian, L., Sala, O.E. (2008) Do litter decomposition and nitrogen mineralization show the same trend in the response to dry and wet years in the Patagonian steppe? Journal of Arid Environments 72, 687-695.

Zaman, M., Chang, S.X., 2004. Substrate type, temperature, and moisture content affect gross and net N mineralization and nitrification rates in agroforestry systems. Biology and

Fertility of Soils 39, 269-279.

Zaman, M., Di, H.J., Cameron, K.C., Frampton, C.M., 1999. Gross nitrogen mineralization and nitrification rates and their relationships to enzyme activities and the soil microbial biomass in soils treated with dairy shed effluent and ammonium fertilizer at different water potentials. Biology and Fertility of Soils 29, 178-186.

Study 2 - Minor response of gross N turnover and N leaching to drying, rewetting and irrigation in the top soil of a Norway spruce forest

Yao-Te Chen[1)], Christina Bogner[2)], Werner Borken[1)], Claus Florian Stange [3)] and Egbert Matzner[1)]

[1)] Soil Ecology, Bayreuth Center of Ecology and Environmental Research (BayCEER), University of Bayreuth, D-95440 Bayreuth, Germany

[2)] Ecological Modelling, Bayreuth Center of Ecology and Environmental Research (BayCEER), University of Bayreuth, D-95440 Bayreuth, Germany

[3)] Soil Physics, Helmholtz Centre for Environmental Research, UFZ, D-06120 Halle/Saale, Germany

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Summary

Forest floors in the temperate climate zone are frequently subjected to strong changes in soil moisture, but the consequences for the soil N cycle are barely known. In a field experiment we tested the hypotheses that soil drying leads to a decrease of gross N turnover and that natural rewetting causes a pulse of gross N turnover and an increase of N leaching from the forest floor. A further hypothesis was that optimal water availability induced by irrigation causes maximum N turnover and N leaching. Replicated control, throughfall exclusion and irrigation plots were established in a Norway spruce forest to simulate different precipitation patterns during a growing season. Gross N turnover rates were determined in undisturbed soil cores from Oi+Oe and Oa+EA horizons by the ^{15}N pool dilution technique. Forest floor percolates were periodically collected by suction plates.

After 142 mm throughfall excluded, the median soil water potential at the throughfall exclusion plots increased from pF 1.9 to 4.5 in the Oi+Oe horizon and from pF 1.8 to 3.8 in the Oa+EA horizon. Gross ammonification ranged from 14 to 45 mg N kg^{-1} soil day^{-1} in the Oi+Oe horizon and from 4.6 to 11.4 mg N kg^{-1} soil day^{-1} in the Oa+EA horizon. Gross ammonification of both horizons was smallest in the throughfall exclusion plots during the manipulation, but the differences between all treatments were not statistically significant. Gross nitrification in both horizons was very small ranging from 1.6 to 11.1 mg N kg^{-1} soil day^{-1}. No effects of decreasing water potential and rewetting on gross nitrification rates were observed due to the small rates and huge spatial variations. The irrigation had no effect as the differences in soil water potential to the control remained small. N leaching from the forest floor was not affected by the treatments. Our findings suggest that ammonification in forest floors continues at considerable rates even at rather small water potential. The hypotheses of increased N turnover and N leaching following rewetting of dry forest floor or by irrigation were not confirmed.

1. Introduction

An increasing frequency of droughts and heavy rainfalls is expected in the future in Europe as a consequence of global climatic changes (IPCC, 2007). The question arises how these climate changes will influence the turnover and the fluxes of N in soils. In forest soils, the N turnover is concentrated in the forest floor and in the A horizon, both being subjected to frequent drying and rewetting.

Drying causes changes in the physical structure of soils, induces hydrophobicity (Doerr *et al.*, 2007) and physiological stress to microorganisms (Schimel *et al.*, 2007). Decreasing microbial activity under dry conditions results from limited supply of substrates or from dehydration of microorganisms (Ford *et al.*, 2007). Rewetting of dry soils may trigger a pulse of net ammonification and nitrification in soils (Xiang *et al.*, 2008) for several reasons. First, drought stress of microorganisms leads to an accumulation of substrates in the soil (Kiese *et al.*, 2002), which are available for surviving microorganisms after rewetting. Second, substrates may become available by disruption of soil aggregates following rewetting, thereby exposing physically protected organic matter and NH_4^+ (Lundquist *et al.*, 1999). Third, hydrophobicity of desiccated soil is most pronounced in organic matter rich horizons and may hamper the recovery of soil microorganisms during rewetting (Mataix-Solera *et al.*, 2007). The overall effect of drying and rewetting on N mineralisation depends on the duration and intensity of desiccation (Borken & Matzner, 2009).

Several studies investigated the influence of soil moisture (Tietema *et al.*, 1992; Vernimmem *et al.*, 2007) and drying/rewetting (Hentschel *et al.*, 2007; Gleeson *et al.*, 2008) on net N mineralisation rates. Net rates of ammonification and nitrification mainly result from the difference of two processes: gross production minus microbial immobilization of NH_4^+ and NO_3^-. Net rates are useful as an index of the soil inorganic N availability (Verchot *et al.*, 2001). Several authors demonstrated that net N transformation rates underestimate gross rates in forest soils by far (Stark & Hart, 1997). In addition, the response of net and gross rates to soil drying/rewetting and irrigation might be different. Rosenkranz *et al.* (2010) reported that changing soil moisture and temperature were the main drivers for seasonal dynamics of gross ammonification and nitrification in a spruce forest. The maximum gross nitrification in the forest floor of tropical rain forests occurred at a water filled pore space of 65% (Kiese *et al.*, 2008). Brüggemann *et al.* (2005) founded that the optimum gravimetric water content for

gross N turnover was between 170 and 330% in temperate forest floors. Stange (2007) observed that gross nitrification rates were reduced by 80% in the organic layer of forest soil when the soil water content decreased from 300 to 130% (w/w). In a laboratory study, Pulleman & Tietema (1999) found a pulse of gross ammonification after rewetting of a dry forest floor whereas gross nitrification did not respond to rewetting.

Increased ammonification and nitrification after rewetting could increase the fluxes of inorganic N in soil solutions, provided that roots do not take up additional N. The effects of drying/rewetting on solute N fluxes in forest soils have only rarely been studied under field conditions. Lamersdorf *et al*. (1998) did not observe a pulse of mineral N in soil solutions following rewetting of a dry Norway spruce soil.

In a laboratory study using disturbed forest soil samples, Chen *et al*. (2011) found that even moderate soil desiccation reduced gross N turnover rates in forest floors. They observed that gross nitrification was more sensitive to soil drying than gross ammonification. The results of laboratory experiments with disturbed soil samples need to be verified under field conditions at different moisture regimes and using undisturbed soil. Rates measured in disturbed samples may differ from those in undisturbed soil cores as the substrate availability is often improved in disturbed samples (Luxhøi & Jensen, 2005).

Here we present results from a field study on the effects of drying/rewetting and irrigation on gross N turnover in a forest soil using the ^{15}N pool dilution technique and undisturbed soil cores. Additionally, we investigated the effect of different soil moisture regimes on mineral N leaching from the forest floor. Our hypotheses were that soil drying (i) leads to a decrease of gross N turnover, (ii) natural rewetting causes a pulse of gross N turnover and N leaching, and (iii) optimal water potential through irrigation increases the N turnover and N leaching from the forest floor.

2. Materials and methods

2.1 Study site

The Coulissenhieb II site is a 140-year-old Norway spruce forest (*Picea abies* L.) located in the Lehstenbach catchment (4.2 km^2) in the Fichtelgebirge mountains (870 m a.s.l.), Germany (58°08’N, 11°52’E). Mean annual precipitation is 1160 mm and mean annual air temperature

is 5.3 °C (Foken, 2003). The soil has a sandy to loamy texture and is classified as Haplic Podzol according to the FAO soil classification (IUSS, 2006). The well stratified, mor-like forest floor of about 10 cm thickness comprises Oi, Oe and Oa horizons. The forest floor is almost completely covered by ground vegetation, mainly *Deschampsia flexuosa* and *Calamagrostis villosa*. Properties of the soil were described by Schulze *et al*. (2009). The C and N contents of the Oi horizon are 46% and 1.7%, of the Oe horizon 42% and 1.8%, of the Oa horizon 21% and 1.1% and of the EA horizon 8.3% and 0.4%. The pH($CaCl_2$) is 3.3 in the Oa horizon and 3.4 in the EA horizon. Organic C and total N stocks are 5.0 kg C m^{-2} and 0.25 kg N m^{-2} in the forest floor (Oi + Oe + Oa horizons), and 2.4 kg C m^{-2} and 0.12 kg N m^{-2} in the EA horizon. The C/N ratios of about 20 point to a considerable N enrichment in this forest by chronic atmospheric N deposition.

2.2 Experimental design

Within an area of 1 ha, three control (C), three throughfall exclusion (TE) and three irrigation plots (I) each of 20 m × 20 m were established in 2006 (C, TE) and 2009 (I). At the TE plots, roof constructions were installed below the forest canopy with a height of 3 m. The roofs were covered with transparent polycloride (PVC) to prevent natural rainfall on 15th June in 2009. The TE plots were trenched to a mineral soil depth of 30 cm in 2006 to prevent lateral movement of soil water into the plots. The TE plots were rewetted by natural rainfall after the roof had been removed on 11th August in 2009. In total, 142 mm throughfall were excluded during 57 days from 15th June to 11th August in 2009 (Fig.1). The annual rainfall in 2009 was 970 mm and throughfall was 820 mm.

To optimise water availability in the top soil, six sprayers were installed at each I plot with a height of 50 cm above the ground. The soil matric potential in the Oa horizon was monitored by a tensiometer. When the matric potential decreased to pF 2.0, the plots were automatically irrigated with de-ionised water at night. The maximum irrigation rate was set to 3 mm per night. In the growing season of 2009, the I plots were not irrigated before the second soil sampling due to the frequent natural precipitation. Irrigation was induced only at 14 events from July 29th to September 1st because of frequent rainfall in spring and early summer. The total amount of irrigated water was 26 mm.

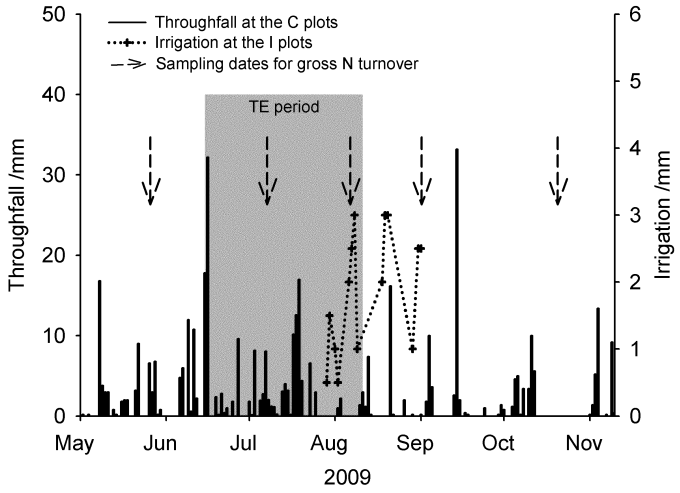

Fig. 1 Daily throughfall (solid bar) and irrigation (dotted line) during the treatment period. The dash arrows indicate the 5 sampling dates and the gray bar is the throughfall exclusion period.

2.3 Soil cores sampling and ^{15}N addition

Three sampling subplot of 1 m × 1 m were established at each of the three replicated experimental plots. The rate of gross N turnover was measured for each sampling subplot. Hence, the sampling design resulted in n = 9 replicates for each treatment and sampling date. Five undisturbed cores of the Oi+Oe and Oa+EA horizons were taken from each of the sampling subplots yielding in total 270 cores per sampling date (nine replicates x three treatments x five cores x two horizons). We sampled at five dates (1× before, 2× during and 2× after the treatments) on 26th May, 7th July, 6th August, 1st September and 20th October in 2009. The cores had a volume of 100 cm^3 with a height of 4 cm and a diameter of 5.6 cm. All intact soil cores were acclimated in a 15 °C chamber for one week prior to determination of gross N turnover.

From each sampling subplot and horizon, one of the five cores was used for the determination of soil water content and soil matric potential. Calculation of matric potentials from volumetric water contents was carried out using the van Genuchten model (van Genuchten, 1980). The parameters for this soil were taken from Zuber (2007). As a measure for the water potential we use the pF value which is the negative logarithm of the water potential in hPa.

The other four cores were used for the determination of the gross N turnover rates (two for ammonification and two for nitrification). Gross rates of ammonification and nitrification were determined by the ^{15}N pool dilution technique (Kirkham & Bartholomew, 1954). A multiple injection system comprising nine hypodermic needles (1.2 mm Ø) with side-pores was used to apply the ^{15}N (Monaghan, 1995; Luxhøi *et al.*, 2004) to ensure a uniform distribution of the label in the soil cores. For gross ammonification, one ml of 5 mM $(^{15}NH_4)_2SO_4$ (98.0 at%) was added to the cores of both horizons. For gross nitrification, one ml of 20 mM $K^{15}NO_3$ (99.2 at%) and 10 mM $K^{15}NO_3$ (99.2 at%) were added to the Oi+Oe and Oa+Ea horizons, respectively. The addition of the ^{15}N solutions increased the soil water content by about 1% (v/v).

We added the same amount of label to all cores, despite differing initial NH_4^+ and NO_3^- pools. When comparing all sampling dates, the initial NH_4^+ pools ranged from 11-170 mg N kg^{-1} and the NO_3^- pools from 1-170 mg N kg^{-1}. Hence, the enrichment of the product pools ranged from 2 - 25% for NH_4^+ and from 7 - 90 % for NO_3^-. We have chosen constant addition rates because of several reasons. (1) The use of undisturbed soil samples makes it impossible to determine the pre-existing NH_4^+ and NO_3^- pools in each core before the ^{15}N application.

Because of the heterogeneity and the large number of samples it is unreasonable to apply individual levels of ^{15}N solution to each soil core. (2) The addition of equal amounts of solution guarantees that the changes of the water content by the ^{15}N addition is similar in all soil cores. (3) Different enrichment ratios of the product do not affect the gross N turnover rates (Murphy *et al.*, 1997; Luxhøi *et al.*, 2003). The incomplete distribution of added ^{15}N was the major factor that led to the error on the estimation (Davidson *et al.*, 1991; Monaghan, 1995; Luxhøi *et al.*, 2003).

One hour after ^{15}N addition (T_0), two of the four cores were homogenized and a subsample of 5 g dry weight extracted with 1 M KCl solution. The other two cores were sealed by flexible film (Parafilm M, Alcan Packaging, USA) and incubated for 48 hours before extraction (T_1). To permit gas exchange during incubation, the film was perforated. The ratio of soil/solution during extraction was 1:10 for the Oi+Oe horizon and 1:5 for Oa+EA horizon. The KCl extractions were shaken for one hour and the supernatant filtered (cellulose folded filters 595½, 4-7 µm, Whatman, Germany). All incubations and extractions were performed at 15 °C.

Filtered KCl extracts were frozen at -20 °C and sent to the Helmholtz Centre for

Environmental Research (UFZ, Halle) for analysis of NO_3^- and NH_4^+ concentrations and the respective ^{15}N abundance using the SPINMAS technique (**S**ample **P**reparation unit for **I**norganic **N**itrogen and **MA**ss **S**pectrometer) (Stange *et al*., 2007). The SPINMAS comprises a coupling of a specially developed sample preparation device with a continuous flow-quadrupole mass spectrometer (QMS GAM 400, InProcess Instruments, Germany). The detection limits for NH_4^+ (140 µM) and NO_3^- (4.0µM) with SPIMAS are much less than the concentrations in our extracts. Gross N ammonification and nitrification rates were calculated using the equation from Kirkham & Bartholomew (1954).

2.4 Soil solutions and throughfall

Throughfall at the site was collected fortnightly by nine samplers (one sampler per experimental plot, upper diameter of 20.2 cm). Throughfall fluxes were calculated as an average for all experimental plots. Forest floor percolates were collected every four weeks below the Oa horizon at each plot by three suction plates with a surface area of 176 cm^2 to which a suction of -10 kPa was applied for one minute every five minutes. The suction plates were made of plastic bowls with a 50 µm pore-size polyethylene membrane on top. The volume of solution was measured and solutions were filtered with 0.45 µm cellulose acetate filter (Whatman, Germany) and stored at 2 °C until chemical analysis. Samples from the three suction plates were mixed to one sample per plot and date. The concentration of NH_4^+ was determined by flow-injection analyzer (MLE, FIA-LAB), and NO_3^- by an ion chromatograph (DIONEX, DX500 Chromatography system). The *in situ* fluxes of NH_4-N + NO_3-N (dissolved inorganic N = DIN) were calculated by multiplying the concentration of NH_4-N + NO_3-N with the respective water fluxes at each sampling date. Water fluxes with forest floor percolates for each sampling date were estimated based on the volume of water collected in the suction plates, the throughfall and irrigation amounts. Using this information, a water budget of the forest floor was first established for the four seasons and second for the monthly samplings. Equal water fluxes were used for the three replicated plots of each treatment. Hence, the spatial variation of N leaching in each treatment is based only on variations in N concentrations.

2.5 Statistical analysis

We used relative changes in gross ammonification and nitrification rates and pF values for data analysis because there were pre-treatment differences between the treatments in May. These differences are not due to the manipulation and have to be considered to estimate the experimental effects. To calculate the relative changes we determined the pre-treatment median rates of gross ammonification and nitrification at each treatment and subtracted these median rates. This procedure ensures that the pre-treatment rates in May have zero median and that the spreading of the data inside the treatments remains unchanged. The pre-treatment nitrification rate in the Oi+Oe horizons at the TE plots was unusually great with large spatial variation probably due to random differences between the soil cores. Therefore, we used the pre-treatment rates measured at the C and I plots in May to estimate the pre-treatment rates at the TE plots.

Soil samples were not taken randomly, but were grouped by plots and sampling points. This grouping might induce dependencies in data, i.e. soil samples that are close together might be similar while samples further apart might be more different. In our statistical analysis, we took this sampling design into account by using mixed-effects ANOVA. This type of statistical model allows for random-effects due to grouping and for fixed-effects due to treatment or sampling time (Pinheiro & Bates, 2000). We used sampling time and the interaction between treatment and sampling time as fixed-effects. Comparisons of treatment effects refer to the relative gross ammonification and nitrification rates and pF values on C plots in May.

All statistical analyses were done in R (R Development Core Team, 2010) using the packages nlme (Pinheiro *et al*., 2009) and stats (R Development Core Team, 2010).

3. Results

3.1 Soil matric potential

In the Oi+Oe horizon the pre-treatment median pF in May were similar at all treatments (Fig. 2a). The median pF at the TE plots increased from 1.9 in May to 4.5 in August. The statistical model revealed that the relative changes of the pF in July and August compared to May were significant ($p<0.003$). The median pF at the C and I plots were similar and ranged from 1.3 to

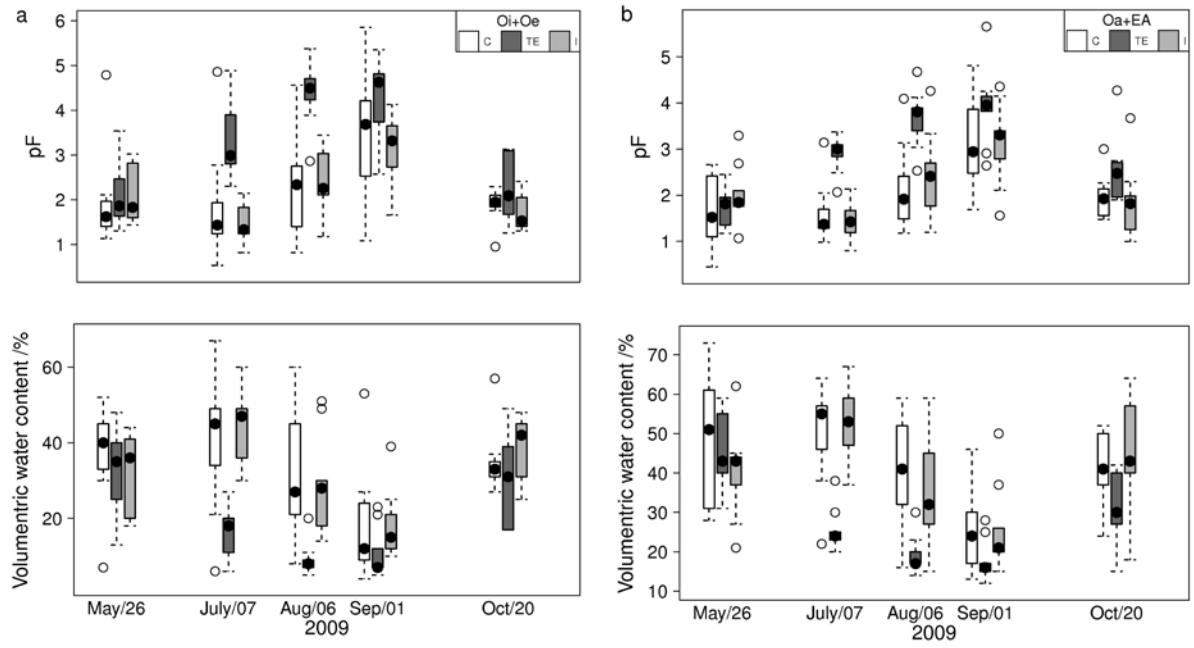

Fig. 2 Box plots of the median pF (●) and volumetric water content at 5 sampling dates in the Oi+Oe horizon (a) and the Oa+EA horizon (b). The top and bottom of the box display the largest and smallest observations within the interquartile range (25 - 75%) (n = 9). The hollow circle (○) stands for outliers over this range.

3.7 from May to September. After the rewetting of the TE plots by 117 mm natural precipitation from 11th August to 20th October, the median pF dropped to 2.1, a level similar to May. In October the pF was similar in all treatments.

In the Oa+EA horizon, the pre-treatment median pF in May were similar for all treatments (Fig. 2b). The median pF at the TE plots increased from 1.8 in May to 3.8 in August. The statistical model revealed that the relative changes of the pF in July and August compared to May were significant ($p<0.003$). The median pF at the C and I plots from May to September were similar and ranged from 1.4 to 3.3. After rewetting, the median pF at the TE plots recovered to about 2.5. In October the pF at all treatments were similar.

3.2 Gross ammonification rates

In the Oi+Oe horizon, the median rates of gross ammonification ranged from 14 to 45 mg N kg^{-1} soil day^{-1} during the whole period (Fig. 3a). Sampling time affected the rates: Compared to may, the relative changes of gross ammonification in August, September and October were significant ($p < 0.03$). No significant differences between the treatments were observed, although the lowest rates (in absolute values) were observed at the TE plots.

In case of the Oa+EA horizon, the median gross ammonification rates were less than in the Oi+Oe horizon and ranged from 4.6 to 11.4 mg N kg^{-1} soil day^{-1} (Fig. 3b). At the TE plots, the

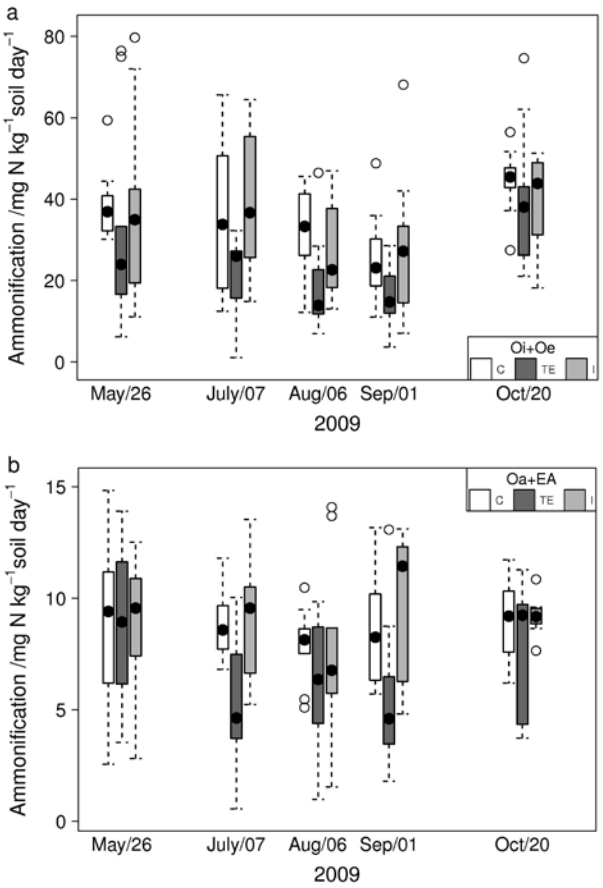

Fig. 3 Box plots of the median gross ammonification rates (●) at 5 sampling dates in the Oi+Oe horizon (a) and in the Oa+EA horizon (b). The top and bottom of the box display the largest and smallest observations within the interquartile range (25 - 75% of our data) (n = 9). The hollow circle (○) stands for outliers over this range.

relative decrease of gross ammonification in July and September compared to May, was significant ($p<0.05$). The rates at the other treatments did not change with time relative to May. Moreover, no differences between treatments were observed in August, despite the fact that the pF in the TE plots reached a maximum. After rewetting, the rates determined in October were similar in all treatments and at the same level as in May.

3.3 Gross nitrification rates

Gross nitrification rates were much less than gross ammonification in both horizons (Fig. 4). In the Oi+Oe horizon, the median gross nitrification was between 3.1 and 6.9 mg N kg^{-1} soil

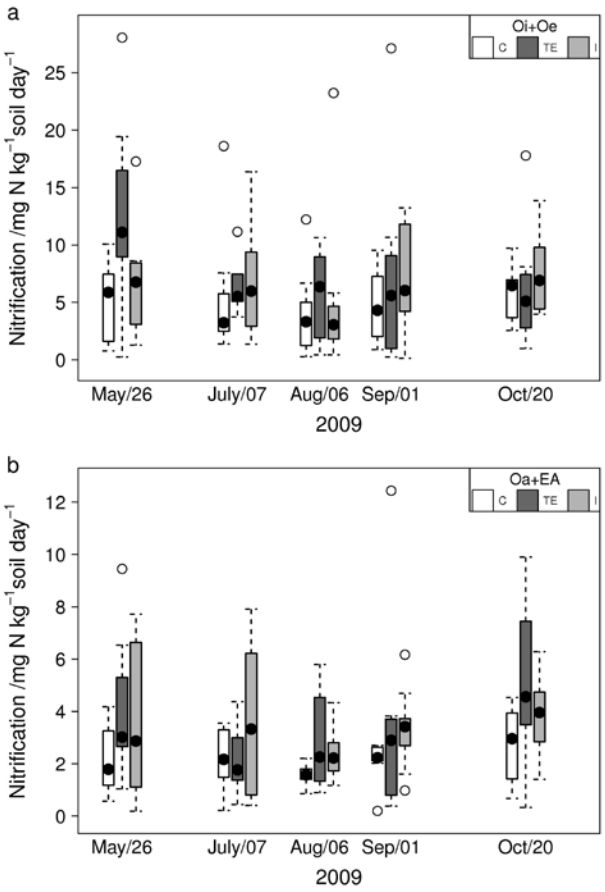

Fig. 4 Box plots of the median gross nitrification rates (●) at 5 sampling dates in the Oi+Oe horizon (a) and in the Oa+EA horizon (b). The top and bottom of the box display the largest and smallest observations within the interquartile range (25 - 75% of our data) (n = 9). The hollow circle (○) stands for outliers over this range.

day^{-1} at all plots, with the exception of 11.1 mg N kg^{-1} soil day^{-1} at the TE plots in May (Fig. 4a). The latter is considered as a random variation because of the large spatial heterogeneity at that sampling date in comparison to the C and I plots. The relative changes of gross nitrification rates during the treatment period were not significantly different between the treatments. After rewetting in October the rates were also similar in all treatments and did not exceed the pre-treatment rates in May. In the Oa+EA horizon, the median rates of gross nitrification were between 1.6 and 4.6 mg N kg^{-1} soil day^{-1} at all treatments and only about 50% of those in the Oi+Oe horizon (Fig. 4b). No effect of the throughfall exclusion, rewetting or irrigation was observed.

3.4 DIN fluxes with forest floor percolates and throughfall

From January 2009 to June 2010, the water flux with throughfall was 1219 mm at the C plots (Fig. 5). Water flux with forest floor percolates at the C, TE and I plots were estimated at 1018, 858 and 1033 mm, respectively. The total cumulative flux of DIN with throughfall at the C and I plots was about 31 kg N ha^{-1} for the period from January 2009 to June 2010 (Fig. 6). Because of throughfall exclusion the N flux with throughfall was reduced to 28 kg ha^{-1} at the TE plots. Solute DIN fluxes from the forest floor were generally in the same order of magnitude compared to throughfall. On average, NO_3-N comprised about 90% of the DIN fluxes in forest floor percolates, but only about 54% in throughfall. During the period of throughfall exclusion, the solute DIN fluxes of the TE plots were smaller than those of the C and I plots. After rewetting, the fluxes of the TE plots increased and slightly exceeded those of the C and I plots. After rewetting, DIN concentrations in the forest floor percolates in the TE plots were somewhat higher than in the controls causing the fluxes in the TE plots to increase. However, the differences in cumulative fluxes were not statistically different between the treatments at the end of the observation period. Furthermore, prior to the throughfall removal, the DIN fluxes with forest floor percolates at the TE plots were already slightly greater compared to the other treatments. No systematic changes were observed in the NO_3/NH_4 ratios of forest floor percolates as a consequence of the treatments (data not shown).

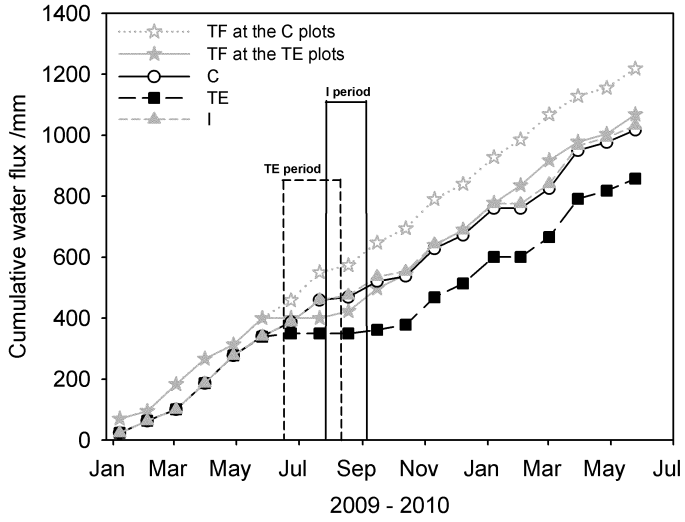

Fig. 5 Cumulative water flux with throughfall (TF) at the C (☆) and TE (★) plots and forest floor percolates at the C (○), TE (■) and I (▲) plots.

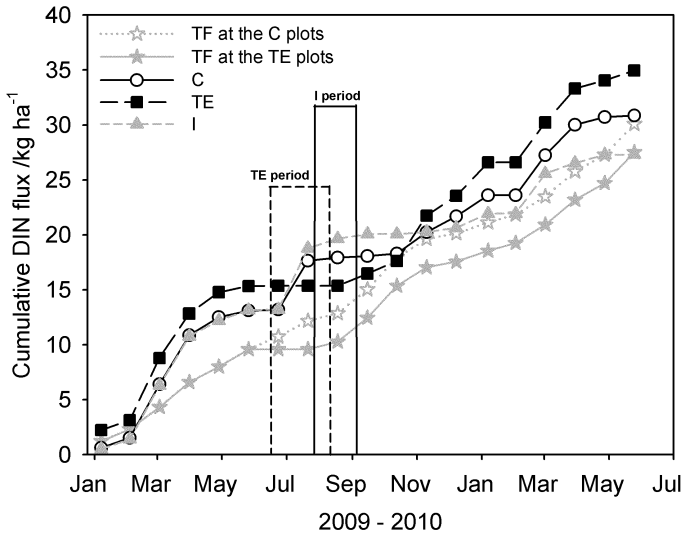

Fig. 6 Cumulative DIN (NH_4-N + NO_3-N) flux with throughfall (TF) at the C (☆) and TE (★) plots and forest floor percolates at the C (○), TE (■) and I (▲) plots.

4. Discussion

4.1 Effects of soil drying/rewetting and irrigation on gross ammonification

In both soil horizons, the rates of gross ammonification were smallest at the TE plots with pF reaching 4.5 in the Oi+Oe and 3.8 in the Oa+EA horizons in August. However, a substantial ammonification rate of 13.9 mg N kg^{-1} soil day^{-1} was still observed at pF 4.5 (58% of that at pF 1.9). The response of gross ammonification to soil drying observed in this study was similar to results from a laboratory study with disturbed soil samples from the same stand: here a rate of 18.3 mg N kg^{-1} soil day^{-1} was determined at pF 4.1 (Chen *et al.*, 2011). This finding is supported by Low *et al.* (1997) who reported substantial gross ammonification rates at pF 4.2 in a pasture soil. The response of gross ammonification and C mineralisation to soil drying is similar. C mineralisation rates in the organic layer from our site were reduced to 72%, 52% and 43% of a constantly moist control, when the matric potentials dropped to pF 3.8, 5.9 and 6.6, respectively (Muhr *et al.*, 2010). It seems that both C and N mineralisation of soil organic matter continue even at extreme soil drought.

One reason for the moderate response of gross ammonification to decreasing water availability in the Oi+Oe horizon might be the greater contribution of drought resistant fungi to ammonification compared to bacteria. This interpretation is supported by the relative greater fungal to bacterial biomass in the uppermost soil layers (Fierer *et al.*, 2003). Also Scheu & Parkinson (1994) observed a significant reduction in bacterial biomass in forest floor horizons by drying, whereas the fungal biomass was less affected. Another reason for the small response of gross ammonification might be a physiological adaption of ammonifiers to frequent soil drying in the uppermost soil horizons. These interpretations remain speculative since the relative contributions of fungi and bacteria to gross ammonification at high pF are unknown.

In both soil horizons of the TE plots, gross ammonification rates in September and October did not exceed the rates of the other treatments and no rewetting effect occurred. The response of gross ammonification to drying/rewetting has been barely studied. In one of the few studies, rewetting of dry soil from gravimetric water contents of 5.2% to 21% in a semi-arid shrubland caused a short-lived wetting pulse that lasted only the first day after rewetting (Saetre & Stark, 2005). Pulleman & Tietema (1999) reported the maximum gross ammonification of 123 mg N

kg^{-1} soil day^{-1}, during the first one to six days after rewetting of the Oe horizon in a Douglas fir stand, which was dried to 10% (w/w) and rewetted to 340% (w/w). The maximum rate after rewetting was 1.4 times of the continuously moist control. After 15 days of rewetting, the gross ammonification rate was even smaller than the one of the continuously moist control.

According to the literature, a rather short-lived pulse of net and gross ammonification might be expected following an intensive rewetting of a severely drying soil. In our field experiment, the first rainfall in September had only a minor effect on the pF and gross ammonification at the TE plots. The substantial rainfalls at the end of September and beginning of October dropped the pF to 2.1. Hence, several factors can explain the absence of a drying/rewetting effect under field conditions: First, soil drying to pF 4.5 (equal to 50% w/w) was not severe enough compared to other studies (Pulleman & Tietema 1999; Saetre & Stark, 2005). In our study, the soil microorganisms were not inhibited completely by pF 4.5 as indicated by a substantial ammonification rate of 13.9 mg N kg^{-1} soil day^{-1} in August. Second, the rewetting of the soil under natural precipitation was likely delayed by hydrophobicity. Many soil microbes accumulate solutes to reduce their internal water potential to avoid dehydration and drying during the drought period. When the soil is rewetted, microbes must dispose these osmolytes immediately until the water potential equilibrates with that of the surrounding water or the water will flow into the cell and may cause cell rupture (Schimel *et al.*, 2007). This mechanism, ascribed for mineral soils, is likely ineffective for dry organic soil horizons as hydrophobicity and preferential flow prevent a rapid and homogenous rewetting of organic matter surfaces (Mataix-Solera *et al.*, 2007). The occurrence of preferential flow paths and inhomogeneous pattern of water contents in the top soil following rewetting was shown in our site by Bogner *et al.* (2010).

We assume that the rewetting effect on ammonification was small and short-lived and that we missed such an effect as our sampling interval was too long. A laboratory experiment also found no significant increase in net N mineralisation after a 40 days rewetting period of dry organic layer from our study site (Muhr *et al.*, 2010). Our results from the field experiment question the validity of laboratory studies on rewetting effects on N turnover, which are observed after rapid and homogenous rewetting of the dry soil.

4.2 Effects of soil drying/rewetting and irrigation on gross nitrification

Gross nitrification rates were always smaller than gross ammonification rates indicating that NH_4^+ is the major substrate for autotrophic nitrification in our site. No response of gross nitrification to increasing pF was observed, contradicting findings from disturbed samples (Chen *et al*., 2011).

The median gross nitrification rates in the Oi+Oe horizon (3.1 - 6.9 mg N kg^{-1} soil d^{-1}) were smaller than the range of 6.4 - 13.1 mg N kg^{-1} soil d^{-1} observed in disturbed soils from our site at soil water potentials from pF 3.1 to 3.9 (Chen *et al.*, 2011), suggesting that gross nitrification rates are larger in disturbed soils than in intact soil cores. This finding is in agreement with Luxhøi & Jensen (2005) who found two times greater gross nitrification rates in disturbed soils than in intact soil cores from arable land. It seems that the mixing of soil improved the NH_4^+ supply of nitrifiers and enhanced gross nitrification rates.

The reasons for the lack of response were the generally small rates of nitrification in the undisturbed samples and the huge spatial variation among the replicates. Besides the spatial variation of microbial biomass (Matejek *et al*., 2010a), different proportions of Oi, Oe, Oa and A horizon material and gradients in the water contents cause great variation among the undisturbed soil cores.

In our study, the coefficient of variation (CV) for gross nitrification ranged from 44 to 102 % (mean = 75%, n = 9) in the Oi+Oe horizon and from 32 to 62 % (mean = 48, n = 9) in the Oa+EA horizon. Using the same method, CVs of 70 % in the organic horizon and 89 % in the EA horizon (n = 6) were observed in a spruce forest (Corre & Lamersdorf, 2004). Even with homogenized soils (n = 23 or 20), Matejek *et al*. (2010a; 2010b) found that the CVs of gross nitrification were 59 - 86% in the organic horizons. Based on our data from July, the sample size analysis revealed a minimum of 20 core pairs (T_0 and T_1) to detect a difference of 3 mg N kg^{-1} soil day^{-1} and a minimum of 43 pairs to detect a difference of 2 mg N kg^{-1} soil day^{-1} between control and treatment plots (power = 80%, $\alpha = 0.05$). Our sample size n = 9 pairs was much less than this level. Hence, given the huge analytical efforts of the ^{15}N pool dilution technique, the use of undisturbed cores cannot universally be recommended for experimental studies in forests.

We did not observe a rewetting effect on gross nitrification, the reasons being similar to those

discussed above for gross ammonification. In another coniferous forest, simulated summer droughts and subsequent wetting did also not induce a pulse of gross nitrification, indicating that nitrifiers were not stimulated by rewetting (Tietema *et al*., 1997).

4.3 Effects of drying/rewetting on solute fluxes

The *in situ* solute fluxes of DIN with forest floor percolates represent the interaction between throughfall fluxes, microbial N immobilization, denitrification and N uptake by roots. Thus, no direct relation between the *in situ* fluxes and the gross N turnover rates can be expected (Matejek *et al*., 2010a). However, in our study, N leaching from the forest floor corroborates the findings from the soil cores. Although an increase of DIN flux occurred at the TE plots in November, the cumulative DIN flux at the TE plots did not significantly exceed that at the C plots. Furthermore, the DIN flux at the TE plots was already slightly greater in the pre-treatment period compared to the C and I plots. A laboratory study without N uptake by roots also did not find an increase in N leaching after rewetting (Muhr *et al.*, 2010). Our results are in agreement with findings of Lamersdorf *et al*. (1998) and do not confirm our hypothesis. Drying/rewetting under field conditions did not enhance total DIN fluxes in forest floor percolates.

5. Conclusion

Our findings suggest that gross ammonification is sensitive to soil drying, but continues at considerable rates even at small water potentials in forest floors. Given the experimental conditions and the intensity of drying and rewetting in our field experiment, no rewetting pulse of gross ammonification was observed, probably due to its short duration or due to the slow changes of the water potential during the natural rewetting. No significant effects of drying/rewetting on gross nitrification were observed because of the small rates and huge spatial variation. Also the *in situ* DIN fluxes with forest floor percolates did not support the hypothesis of increased solute N fluxes following rewetting of drying forest soils. Our results suggest that increasing frequency of drying/wetting cycles will have little effect on microbial N turnover in forest floors.

Acknowledgements

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References

Borken, W. & Matzner, E. 2009. Reappraisal of drying and wetting effects on C and N mineralization and fluxes in soils. *Global Change Biology*, **15**, 808-824.

Brüggemann, N., Rosenkranz, P., Papen, H., Pilegaard, K. & Butterbach-Bahl, K. 2005. Pure stands of temperate forest tree species modify soil respiration and N turnover. *Biogeosciences Discussions*, **2**, 303-331.

Chen, Y.-T., Borken, W., Stange, C.F. & Matzner, E. 2011. Effects of decreasing water potential on gross ammonification and nitrification in an acid coniferous forest soil. *Soil Biology and Biochemistry*, **43**, 333-338.

Corre, M.D. & Lamersdorf, N.P. 2004. Reversal of nitrogen saturation after long-term deposition reduction: impact on soil nitrogen cycling. *Ecology*, **85**, 3090-3104.

Davidson, E.A., Hart, S.C., Shanks, C.A. & Firestone, M.K. 1991. Measuring gross nitrogen mineralization, immobilization, and nitrification by ^{15}N isotopic pool dilution in intact soil cores. *Journal of Soil Science*, **42**, 335-349.

Doerr, S.H., Ritsema, C.J., Dekker, L.W., Scott, D.F. & Carter, D. 2007. Water repellence of soils: new insights and emerging research needs. *Hydrological Processes*, **21**, 2223-2228.

Fierer, N., Schimel, J.P. & Holden, P.A. 2003. Variations in microbial community composition through two soil depth profiles. *Soil Biology and Biochemistry*, **35**, 167-176.

Foken, T. 2003. Lufthygienisch-Bioklimatische Kennzeichnung des oberen Egertales. *Bayreuther Forum Ökologie*, **100**, 1-118.

Ford, D.J., Cookson, W.R., Adams, M.A. & Grierson, P.F. 2007. Role of soil drying in

nitrogen mineralization and microbial community function in semi-arid grasslands of north-west Australia. *Soil Biology and Biochemistry*, **39**, 1557-1569.

Gleeson, D.B., Herrmann, A.M., Livesley, S.J. & Murphy, D.V. 2008. Influence of water potential on nitrification and structure of nitrifying bacterial communities in semiarid soils. *Applied Soil Ecology*, **40**, 189-194.

Hentschel, K., Borken, W. & Matzner, E. 2007. Leaching losses of inorganic N and DOC following repeated drying and wetting of a spruce forest soil. *Plant and Soil*, **300**, 21-34.

IPCC, 2007. Climate Change 2007: Synthesis report. *Contribution of Working Groups I, II and III to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change*. IPCC, Geneva, Switzerland.

IUSS Working Group WRB, 2006. World reference base for soil resources 2006. 2nd edition. *World Soil Resources Report*, **103**, FAO, Rome.

Kiese, R., Papen, H., Zumbusch, E. & Butterbach-Bahl, K. 2002. Nitrification activity in tropical rain forest soils of the Coastal Lowlands and Atherton Tablelands, Queensland, Australia. *Journal of Plant Nutrition and Soil Science*, **165**, 682-685.

Kiese, R., Hewett, B. & Butterbach-Bahl, K. 2008. Seasonal dynamic of gross nitrification and N_2O emission at two tropical rainforest sites in Queensland, Australia. *Plant and Soil*, **309**, 105-117.

Kirkham, D. & Bartholomew, W.V. 1954. Equations for following nutrient transformations in soil, utilizing tracer data. *Soil Science Society of America Proceedings*, **18**, 33-34.

Lamersdorf, N.P., Beier, C., Blanck, K., Bredemeier, M., Cummins, T., Farrell, E.P. *et al*. 1998. Effect of drought experiments using roof installations on acidification/nitrification of soils. *Forest Ecology and Management*, **101**, 95-109.

Low, A.P., Stark, J.M. & Dudley, L.M. 1997. Effects of soil osmotic potential on nitrification, ammonification, N-assimilation, and nitrous oxide production. *Soil Science*, **162**, 16-27.

Lundquist, E.J., Jackson, L.E. & Scow, K.M. 1999. Wet-dry cycles affect dissolved organic carbon in two California agricultural soils. *Soil Biology and Biochemistry*, **31**, 1031-1038.

Luxhøi, J., Nielsen, N.E. & Jensen, L.S. 2003. Influence of $^{15}NH_4^+$-application on gross N turnover rates in soil. *Soil Biology and Biochemistry*, **35**, 603-606.

Luxhøi, J., Nielsen, N.E. & Jensen, L.S. 2004. Effect of soil heterogeneity on gross nitrogen mineralization measured by ^{15}N-pool dilution techniques. *Plant and Soil*, **262**, 263-275.

Luxhøi, J. & Jensen, L.S. 2005. Gross N mineralization-immobilization rates in heterogeneous intact soil cores can be estimated without marked error. *Biology and Fertility of Soils*, **41**, 280-283.

Mataix-Solera, J., Arcenegui, V., Guerrero, C., Mayoral, A.M., Morales, J., González, J. *et al.* 2007. Water repellency under different plant species in a calcareous forest soil in a semiarid Mediterranean environment. *Hydrological Processes*, **21**, 2300-2309.

Matejek, B., Huber, C., Dannenmann, M., Kohlpaintner, M., Gasche, R., Göttlein, A. *et al.* 2010a. Microbial nitrogen-turnover processes within the soil profile of a nitrogen-saturated spruce forest and their relation to the small-scale pattern of seepage-water nitrate. *Journal of Plant Nutrition and Soil Science*, **173**, 224-236.

Matejek, B., Huber, C., Dannenmann, M., Kohlpaintner, M., Gasche, R. & Papen, H. 2010b. Microbial N turnover processes in three forest soil layers following clear cutting of an N saturated mature spruce stand. *Plant and Soil*, **337**, 93-110.

Monaghan, R. 1995. Errors in estimates of gross rates of nitrogen mineralization due to non-uniform distributions of ^{15}N label. *Soil Biology and Biochemistry*, **27**, 855-859.

Muhr, J., Franke, J. & Borken, W. 2010. Drying-rewetting events reduce C and N losses from a Norway spruce forest floor. *Soil Biology and Biochemistry*, **42**, 1303-1312.

Murphy, D.V., Fillery, I.R.P. & Sparing, G.P. 1997. Method to label soil cores with $^{15}NH_3$ gas as a prerequisite for ^{15}N isotopic dilution and measurement of gross N mineralization. *Soil Biology and Biochemistry*, **29**, 1731-1741.

Pinheiro, J.C. & Bates, D.M. 2000. *Mixed-Effects Models in S and S-Plus*. Springer, New York.

Pinheiro, J., Bates, D., DebRoy, S., Sarkar, D. & The R Core team 2009. *nlme: Linear and Nonlinear Mixed Effects Models*. R package version 3.1-96.

Pulleman, M. & Tietema, A. 1999. Microbial C and N transformations during drying and rewetting of coniferous forest floor material. *Soil Biology and Biochemistry*, **31**, 275-285.

R Development Core Team 2010. *R: A Language and Environment for Statistical Computing*.

R Foundation for Statistical Computing, Vienna.

Rosenkranz, P., Dannenmann, M., Brüggemann, N., Papen, H., Berger, U., Zumbusch, E. *et al*. 2010. Gross rates of ammonification and nitrification at a nitrogen-saturated spruce (*Picea abies* (L.) Karst.) stand in southern Germany. *European Journal of Soil Science*, **61**, 745-758.

Saetre, P. & Stark, J.M. 2005. Microbial dynamics and carbon and nitrogen cycling following re-wetting of soils beneath two semi-arid plant species. *Oecologia*, **142**, 247-260.

Scheu, S. & Parkinson, D. 1994. Changes in bacterial and fungal biomass-C, bacterial and fungal biovolume and ergosterol content after drying, remoistening and incubation of different layers of cool temperate forest soils. *Soil Biology and Biochemistry*, **26**, 1515-1525.

Schimel, J., Balser, T.C. & Wallenstein, M. 2007. Microbial stress-response physiology and its implications for ecosystem function. *Ecology*, **88**, 1386-1394.

Schulze, K., Borken, W., Muhr, J. & Matzner, E. 2009. Stock, turnover time and accumulation of organic matter in bulk and density fractions of a Podzol soil. *European Journal of Soil Science*, **60**, 567-577.

Stange, C.F. 2007. A novel approach to combine response functions in ecological process modelling. *Ecological Modelling*, **204**, 547-552.

Stange, C.F., Spott, O., Apelt, B. & Russow, R.W.B. 2007. Automated and rapid online determination of ^{15}N abundance and concentration of ammonium, nitrite, or nitrate in aqueous samples by the SPINMAS technique. *Isotopes in Environmental and Health Studies*, **43**, 227-236.

Stark, J.M. & Hart, S.C. 1997. High rates of nitrification and nitrate turnover in undisturbed coniferous forests. *Nature*, **385**, 61-64.

Tietema, A., Warmerdam, B., Lenting, E. & Riemer, L. 1992. Abiotic factors regulating nitrogen transformations in the organic layer of acid forest soils: Moisture and pH. *Plant and Soil*, **147**, 69-78.

Tietema, A., Beier, C., deVisser, P.H.B., Emmett, B.A., Gundersen, P., Kjonaas, O.J. *et al*. 1997. Nitrate leaching in coniferous forest ecosystems: The European field-scale manipulation experiments NITREX (nitrogen saturation experiments) and EXMAN

(experimental manipulation of forest ecosystems). *Global Biogeochemical Cycles*, **11**, 617-626.

Van Genuchten, M.T. 1980. A Closed-form equation for predicting the hydraulic conductivity of unsaturated soils. *Soil Science Society of America Journal*, **44**, 892-898.

Verchot, L.V., Holmes, Z., Mulon, L., Groffman, P.M. & Lovett, G.M. 2001. Gross vs net rates of N mineralization and nitrification as indicators of functional differences between forest types. *Soil Biology and Biochemistry*, **33**, 1889-1901.

Vernimmen, R.R.E., Verhoef, H.A., Verstraten, J.M., Bruijnzeel, L.A., Klomp, N.S., Zoomer, H.R. *et al*. 2007. Nitrogen mineralization, nitrification and denitrification potential in contrasting lowland rain forest types in Central Kalimantan, Indonesia. *Soil Biology and Biochemistry*, **39**, 2992-3003.

Xiang, S.R., Doyle, A., Holden, P.A. & Schimel, J.P. 2008. Drying and rewetting effects on C and N mineralization and microbial activity in surface and subsurface California grassland soils. *Soil Biology and Biochemistry*, **40**, 2281-2289.

Zuber, T. 2007. *Untersuchungen zum Wasserhaushalt eines Fichtenwaldstandorts unter Berücksichtigung der Humusauflage*. Doctoral dissertation, University Bayreuth, Bayreuth.

Study 3 - Dynamics of nitrogen and carbon mineralization in a fen soil following water table fluctuations

Yao-Te Chen[1)], Werner Borken[1)], Claus Florian Stange [2)] and Egbert Matzner[1)]

[1)] Soil Ecology, Bayreuth Center of Ecology and Environmental Research (BayCEER), University of Bayreuth, D-95440 Bayreuth, Germany

[2)] Soil Physics, Helmholtz Centre for Environmental Research, UFZ, D-06120 Halle/Saale; present address: Federal Institute for Geosciences and Natural Resources, Geozentrum D-30655 Hannover, Germany

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Abstract

Changes of the water table level and oxygen supply affect the N and C cycles of fen soils. We studied the response of N and C mineralization and soil solution chemistry to water table fluctuations in an acidic minerotrophic fen. In a laboratory study lasting 117 days, undisturbed soil cores were treated by a) permanently flooded or b) the water table was manipulated by flooding, water table drawdown and re-flooding.

In the permanently flooded cores the CO_2 emissions were constantly low, but gross ammonification increased after a lag phase of about 30 days. Water table drawdown increased gross ammonification after a lag phase of about 30 days. Emission of CO_2 peaked immediately after water table drawdown, followed by a decrease and a second maximum after about 30 days. Following re-flooding, gross ammonification first decreased and, after about 30 days, recovered to the level of the permanently flooded cores while the CO_2 emissions decreased immediately and permanently. The cumulative gross ammonification during the 117 days was larger in the permanently flooded cores than in the fluctuated cores. The ratio of CO_2 emission/gross ammonification were close to 2 under anoxic condition which seems to be caused by fast N turnover in the microbial biomass pool and low rates of CO_2 production.

In fen soils, the response to water table fluctuations is different for N and C turnover. Furthermore, short term (few days) water table drawdown will have different effects as compared to drawdown on longer time scales (> 1 month).

1. Introduction

The role of wetlands with respect to N and C sequestration, emission of greenhouse gases, and the quality of ground- and stream water is widely acknowledged (Bowden 1987; Mitsch et al. 2001; Scott et al. 2008; Knorr et al. 2008a; Dinsmore et al. 2009). The elemental cycling in wetlands depends on the hydrological status, and is likely to be affected by climate change. Small changes of precipitation or water table can have large effects on the moisture and O_2 status of the soil. The current climate change scenarios predict a more dynamic change of the water table in the future because of shifts in the precipitation regime (IPCC 2007).

N turnover in wetland soils is thought to be sensitive to fluctuations of water table and O_2 supply (Pal et al. 2010). Nitrification under anaerobic conditions is generally low (Bowden 1986), while ammonification can occur under both aerobic and anaerobic conditions (Pinay et al. 2002; Hefting et al. 2004). In wetlands, contradicting results on water table effects on ammonification have been reported in field or laboratory experiments. In a slurry experiment with fen soils, gross ammonification decreased under anaerobic conditions (Ambus et al. 1992). When comparing drained and re-flooded fen sites similar rates were found (Münchmeyer et al. 2000). However, Wray and Bayley (2008) found higher gross ammonification rates in non-flooded than in flooded peatlands. Net N mineralization was larger in flooded marsh soils than in non-flooded (Neill 1995) and water table drawdown in the flooded marsh decreased net N mineralization rates, while net nitrification rates increased. In some cases, aeration associated with water table drawdown also led to higher N mineralization and increased inorganic N content in wetlands (Venterink et al. 2002; Keller et al. 2004; Kieckbusch and Schrautzer 2007). Many studies have shown that a drawdown in water table increases the O_2 penetration and the CO_2 emissions in peatland soils (Silvola et al. 1996; Oechel et al. 1998; Danevčič et al. 2010). In contrast, Knorr et al. (2008a) and Muhr et al. (2011) reported no changes of CO_2 emissions from a minerotrophic fen after water table drawdown. CH_4 production did not recover after re-flooding quickly because of the suppression of methanogenesis by SO_4^{2-} being produced during water table drawdown (Freeman et al. 1993; Blodau and Moore 2003).

The concentrations and fluxes of dissolved organic C (DOC) and N (DON) are important for the quality of surface waters draining from peatlands. The response of DOC and DON in peat soils to water table fluctuations is unclear. Decreasing concentrations have been observed after water table drawdown (Scott et al. 1998; Clark et al. 2005; Fenner et al. 2005) while

others found increasing concentrations (Driscoll et al. 1989; Tipping et al. 1999) or no response (Blodau et al. 2004; Freeman et al. 2004).

The overall effect of water table fluctuations on the C and N turnover of fen soils will depend on the dynamics of the response. If, e.g., the response of N and C mineralization to changes in the O_2 regime is slow (several weeks), short-term fluctuations on a daily to weekly time scale will probably have little effect and *vice versa*. Deppe et al. (2010) found only minor impact of short term changes (1 day to 1 week) of the water table on CO_2 emissions.

Our goal was to investigate the dynamics of N and C mineralization in a fen soil in response to the water table drawdown and re-flooding by comprehensively studying gross N mineralization, CO_2 emissions and DON/DOC release. Our hypotheses were: 1) Water table drawdown increases the mineralization of N and C. 2) The temporal response of gross N turnover and CO_2 emissions to water table drawdown is similar. 3) Water table drawdown reduces the concentrations of DON and DOC. 4) The changes induced by water table drawdown are reversible after re-flooding.

2. Materials and methods

2.1 Site description

This study was conducted at the Schlöppnerbrunnen fen site, located in the Lehstenbach catchment (4.5 km^2, Fichtelgebirge, northeastern Bavaria, Germany, 58°08'N, 11°51'E). Mean annual precipitation is 1020 mm and mean annual temperature is 6.3 °C (Knorr et al. 2009). The peat thickness ranges from 30 to 120 cm. The C and N contents of the top 10 cm are 31.1% and 1.8%. Bulk density is 0.29 g cm^{-3} and porosity is 85.5%. The soil is moderately acidic (pH 3.5 to 5.5) and rich in iron and sulfur (Paul et al. 2006; Knorr et al. 2008b; Goldberg et al. 2008). The water table level at the field site fluctuates from +0.5 cm at water saturation to -50 cm under summer drought conditions. The vegetation of the fen site comprises mainly *Nardus stricta*, *Agrostis sp.*, *Molinia coerulea*, *Eriophorum vaginatum*, *Sphagnum fallax*, *Brachythecium rivulare*, *Atrichum undulatum* and *Galium hercynicum* (Knorr et al. 2009). Vegetation is concentrated on the hummocks while the hollows are mostly free of vegetation.

2.2 Soil sampling and experimental design

To measure gross N turnover, 288 intact soil cores (with a height of 10 cm and a diameter of 5.6 cm) without living vegetation were taken from the hollows in June 2010. In addition, another 10 intact soil cores of 17.1 cm diameter and 10 cm height were taken from the hollows. These larger cores were used for measuring CO_2 emissions, O_2 and CH_4 concentrations and soil solution chemistry. The soil cores were sealed with lids or plastic bags immediately after sampling. All soil cores were immediately transported into a 15°C room and flooded with de-ionized water to +5 cm above the soil surface within 24 hours after sampling. All cores were stored for 7 days before any measurement started.

Two regimes of water table were established: permanent flooding (C) and fluctuation (F). The water table in the permanently flooded cores was maintained at +5 cm for 117 days while the fluctuation comprised a change of the water table from +5 cm (flooding from day 0 to 24), to -8 cm. The water table drawdown was initiated quickly within a few minutes and lasted from day 25 to 70. After that, the water table was raised again to +5 cm within few minutes (re-flooding from day 71 to 117). For re-flooding, the water that was extracted previously for water table drawdown was recycled.

Each treatment comprised 144 small soil cores for gross N turnover and 5 large soil cores for measuring CO_2 emissions, concentrations of O_2 and CH_4 in the soil and soil solution chemistry. Gross N turnover rates were determined at 12 dates during the manipulation period. The CO_2 emissions of the 5 large cores of both treatments were monitored continually. Additionally, in 2 of the 5 large cores, concentrations of CH_4 and O_2 were measured at 12 dates. Soil solution was collected at 6 dates from the other 3 of the 5 large cores. All experiments were done at 15 °C.

2.3 Gross N turnover rates

Gross rates of ammonification and nitrification were determined by the ^{15}N pool dilution technique (Kirkham and Bartholomew 1954) with 3 replicates. This adds up to the use of 12 cores per sampling date (each 6 for ammonification and nitrification (3 for t_0, 3 for t_1). A multiple injection system comprising 9 hypodermic needles (1.2 mm Ø) with side-pores (Monaghan 1995; Luxhøi et al. 2004) was used to apply 2.6 mL with either 2.5 mM $(^{15}NH_4)_2SO_4$ (98.0 at%) or 2.0 mM $K^{15}NO_3$ (99.2 at%) to the cores.

Following 1 hour after ^{15}N addition (t_0), 6 t_0 soil cores (3 with $(^{15}NH_4)_2SO_4$ and 3 with

$K^{15}NO_3$) were homogenized and a subsample of 5 g dry weight extracted with 1 M KCl solution. To avoid gas exchange during incubation in the flooded samples of both treatments, the 6 t_1 soil cores were sealed by flexible film (Parafilm M, Alcan Packaging, USA) and lids and then incubated for 48 hours before next extraction (t_1). The ratio of soil/solution during extraction was 1:10. The KCl extracts were shaken for 1 hour and then supernatant was filtered (cellulose folded filters 595½, 4-7 µm, Whatman, Germany).

Filtered KCl extracts were frozen at -20°C and sent to the Helmholtz Centre for Environmental Research (UFZ, Halle) for analysis of ^{15}N abundance and the concentrations of NO_3^- and NH_4^+ using the SPINMAS technique (**S**ample **P**reparation unit for **I**norganic **N**itrogen and **MA**ss **S**pectrometer) (Stange et al. 2007). The SPINMAS comprises a coupling of a specially developed sample preparation device with a continuous flow-quadrupole mass spectrometer (QMS GAM 400, InProcess Instruments, Germany).

Gross ammonification and nitrification rates were calculated using the equation from Kirkham and Bartholomew (1954). The ^{15}N abundances and concentrations of 3 t_0 cores were randomly pairwise related to 3 of the t_1 cores, resulting in 3 values for gross rates. Arithmetic means and standard errors were calculated using n = 3 using the software SIGMAPLOT 10.0 as shown in our Fig.s.

2.4 CO_2, O_2 and CH_4

The CO_2 emissions of the large cores were measured by an automated system BINOS 100 IRGA (Fisher-Rosemount, formerly Leybold Heraeus, max. detection of 1000 ppm with 50 ppm error) (Muhr et al. 2008). The air collected from the head space of the cores was dried (Drierite®, 8 mesh with indicator) and then pumped at constant rate of 1.5 L min^{-1} for 5 min (flooding period) or 1 min (water table drawdown period) with CO_2 concentration being logged automatically in 10 s intervals. Gas fluxes were calculated from the observed change of concentration over time by using Eq. 1:

$$F_{gas} = \left(\frac{dc}{dt}\right) \times \left(\frac{V_H \times M_W \times 60\,\text{min} \times 24\text{h}\ d^{-1}}{M_v \times A_H \times 1000\,ppm}\right) \times \left(\frac{P_a}{P_N \times (1 + 0.00366 \times T_a)}\right), \quad (1)$$

where F_{gas} represents the gas flux of the measured gas in mg m^{-2} d^{-1}, *dc/dt* is the change of concentration over time measured in the column in ppm min^{-1}, V_H is the volume of the column in liter, M_W is the molecular weight of C, M_V is the molecular volume of measured gas in L mol^{-1}, A_H is the surface area of soil inside the column in m^2, *Pa* is the measured air pressure in hPa, P_N the standard air pressure, which is 1013 hPa, and T_a is the measured air temperature in °C with the factor 0.00366 originating from 1/273.15 due to the conversion from K to °C. This value is divided by 29 kg m^{-2} (dry mass stock in the top 10 cm) to transfer the unit as mg C kg^{-1} soil d^{-1}.

For measuring concentrations of O_2 and CH_4, gas samples were collected from horizontally inserted silicon samplers (10 mm diameter, 1 mm wall) at 2 and 8 cm depths installed in the large cores. The concentrations of O_2 and CH_4 were determined with Hewlett-Packard Co. (Palo Alto, Calif.) 5980 series II gas chromatographs. O_2 and CH_4 were separated with a molecular sieve (Alltech, Unterhaching, Germany) column (length 2 m, inner diameter 3.2 mm) and analyzed with a thermal conductivity detector (carrier gas was argon at a flow rate of 33 mL min^{-1}); the temperatures of injector, column, and detector were 150, 60, and 175°C, respectively.

2.5 Soil solution

Soil solution was sampled by Rhizon samplers (Eijkelkamp, AN Wageningen, Netherlands) of hydrophilic porous polymer (100 mm length, 2.5 mm diameter, 1.5 mm wall, 0.15 µm pore size). The samplers were installed vertically in the middle of the large cores. The soil solution was collected in a 50 mL vacuum glass bottle at -70 kPa. The soil solutions were stored at 2°C and measured for pH, electric conductivity, dissolved organic C (DOC, Elementar, high-TOC), total N (tN, Elementar, high-TOC), ammonium (NH_4^+, MLE, FIA-LAB), nitrate (NO_3^-, DIONEX, DX500 Chromatography system) and sulfate (SO_4^{2-}, DIONEX, DX500 Chromatography system). Concentration of dissolved organic N (DON) was calculated as the difference between total N and inorganic N ($NH_4^+ + NO_3^-$).

3. Results

3.1 Redox conditions

In the permanently flooded cores, the O_2 concentration of the soil air ranged from 0.5 to 8.0% (Fig. 1). The water table drawdown increased the O_2 concentration in the soil air at -2 cm depth to more than 15% after 7 days in core F2 and after 28 days in core F1. At -8 cm depth, the O_2 concentrations increased only slightly and were in the range of 5 - 10% even after 42 days of water table drawdown (day 67). When the soil was re-flooded, O_2 concentrations at -2 cm depth dropped down rapidly within 3 days (day 73) to concentrations similar to those of the permanently flooded cores.

In the permanently flooded cores, the concentrations of CH_4 in soil air increased linearly at both depths from 10% to about 40% in the first 30 days of flooding (day 0 to 30) (Fig. 2).

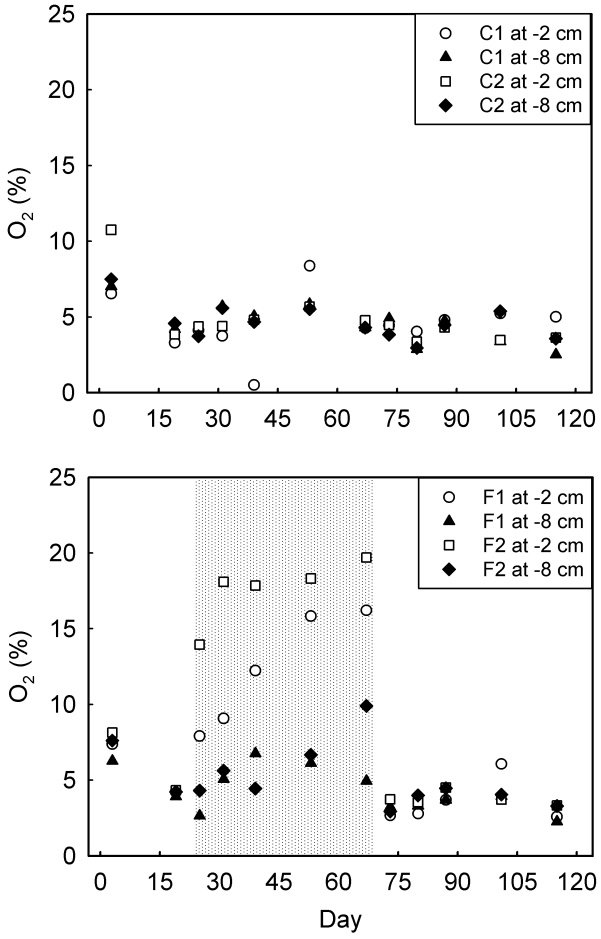

Fig. 1 O_2 concentrations in the permanently flooded (C) and the fluctuated (F) treatments (n = 2). The gray bar indicates the water table drawdown period in the fluctuated treatment.

Subsequently, concentrations increased slowly to 50% - 65% during the post-half period of flooding (day 67 to 115). Before manipulation the concentrations of CH_4 were similar in both treatments. After 7 days of water table drawdown (day 32), the CH_4 concentrations were almost close to 0% at -2 cm depths, whereas concentrations at -8 cm depths were close to 0% after 28 days of water table drawdown (day 53). At both depths, the concentrations of CH_4 increased linearly from 0% to about 40% when the soil was re-flooded for 45 days (day 115).

The concentrations of SO_4^{2-} in soil solutions were generally below 1.0 mg L^{-1}. Only in the final stage of the water table drawdown period, concentrations increased strongly to about 19 mg L^{-1} (Fig. 3).

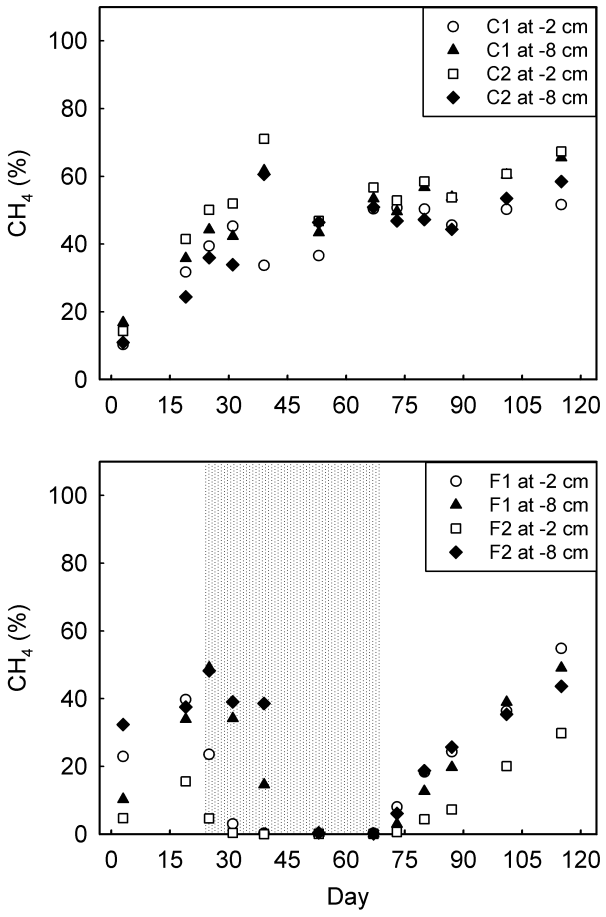

Fig. 2 CH_4 concentrations in the permanently flooded (C) and the fluctuated (F) treatments (n = 2). The gray bar indicates the water table drawdown period in the fluctuated treatment.

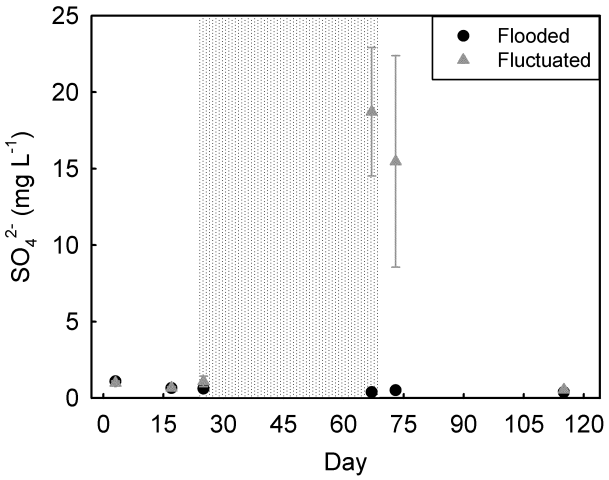

Fig. 3 SO_4^{2-} concentrations in the permanently flooded and the fluctuated treatments (mean ± SE, n = 3). The gray bar indicates the water table drawdown period in the fluctuated treatment.

3.2 Gross N turnover

Before the water table drawdown (day 0 to 24), gross ammonification was similar in both sets of cores, ranging from 7 to 15 mg N kg^{-1} soil d^{-1} (Fig. 4). In the permanently flooded cores, gross ammonification strongly increased after 30 days of flooding and peaked at 55 mg N kg^{-1} soil d^{-1} in day 80. Subsequently, the rates decreased slightly to the range observed from day 40 to 70, but were still much higher than at the beginning.

In the fluctuated cores, gross ammonification increased after 30 days of water table drawdown from 9 to 29 mg N kg^{-1} soil d^{-1} in day 67. After re-flooding, the rates decreased within 3 days to the level obtained before the water table drawdown. After 30 days of re-flooding, the rates increased again to the level of the permanently flooded cores. The dynamic of gross ammonification after re-flooding (day 70 to 115) in the fluctuated cores was similar to that in the permanently flooded cores from day 0 to 50, both showing an increase after a lag phase of about 30 days.

During the incubation of 117 days, the cumulative gross ammonification rates were 2840 and 1940 mg N kg^{-1} soil in the permanently flooded and in the fluctuated cores, respectively (Fig. 5). The water table fluctuations reduced the cumulative gross ammonification by about 30% compared to the permanently flooded cores.

In general, gross nitrification rates were much smaller than gross ammonification and ranged from 0.4 to 3.4 mg N kg^{-1} soil d^{-1} in both treatments. Gross nitrification in the fluctuated cores exceeded the rates of the permanently flooded cores only in the first phase of water table drawdown (Fig. 4).

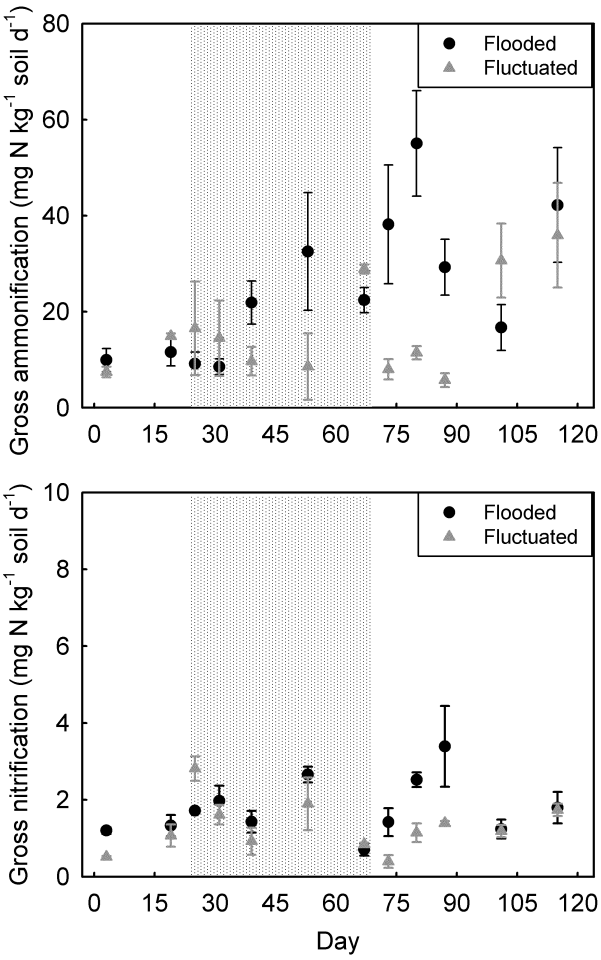

Fig. 4 Gross ammonification and nitrification rates (mean ± SE, n = 3) in the permanently flooded (●) and the fluctuated (▲) treatments. The gray bar indicates the water table drawdown period in the fluctuated treatment.

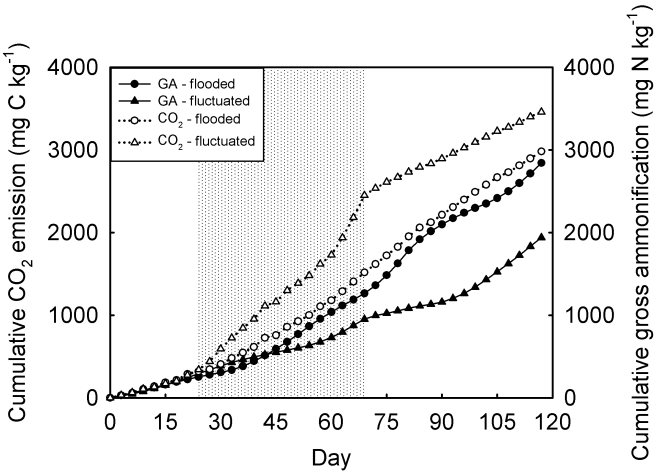

Fig. 5 Cumulative gross ammonification rate (GA) and CO_2 flux in the permanently flooded and the fluctuated treatments. The gray bar indicates the water table drawdown period in the fluctuated treatment.

3.3 CO_2 emissions

The initial CO_2 emissions did not differ between the permanently flooded cores (21 ± 10 mg C kg^{-1} soil d^{-1}, mean ± SE) and the fluctuated cores (20 ± 8 mg C kg^{-1} soil d^{-1}, mean ± SE) (Fig. 6). The CO_2 emissions from the permanently flooded cores were constant throughout the experimental period.

The response of CO_2 emissions to water table drawdown was similar in the 5 replicated cores but with different levels. The CO_2 emissions increased immediately after water table drawdown and the highest emissions were found after 1 day of water table drawdown. In the following 3 -5 days, CO_2 emissions decreased exponentially and reached a steady rate after 20 days of water table drawdown (day 45). A second increase was observed after 30 days of water table drawdown (day 55 to 70). When the soil was re-flooded (day 71), the CO_2 emissions decreased rapidly within 1 day to levels less than those of the permanently flooded cores. At the end of the experimental period the CO_2 emissions from the re-flooded cores were similar to those of the permanently flooded cores. The cumulative CO_2 emission from the fluctuated cores during the period of water table drawdown was about 1.6 fold of that from the permanently flooded cores.

The cumulative CO_2 emission over the whole experimental period was 2980 mg C kg^{-1} soil in the permanently flooded cores and 3460 mg C kg^{-1} soil in the fluctuated cores (Fig. 5). In the 117 days, about 1.0% and 1.1% of the total C pool of the fen soil was mineralized in the permanently flooded and fluctuated treatments, respectively.

3.4 Relation of CO_2 emission to gross ammonification

The ratio of CO_2 emission/gross ammonification (C and N mineralization in mol) decreased after 30 days of flooding in the permanently flooded cores and remained constant at about 2 (Fig. 7). In the fluctuated cores, the ratio increased from 3 to 6 during the water table drawdown and the ratios were always higher than those in the permanently flooded cores. After the re-flooding for 45 days, the ratios dropped to similar ratios as in the permanently flooded cores.

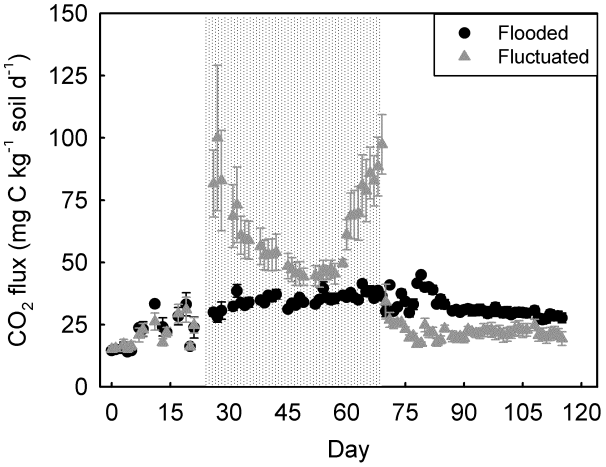

Fig. 6 CO_2 emissions in the permanently flooded (●) and the fluctuated (▲) treatments (mean ± SE, n = 5). The gray bar indicates the water table drawdown period in the fluctuated treatment.

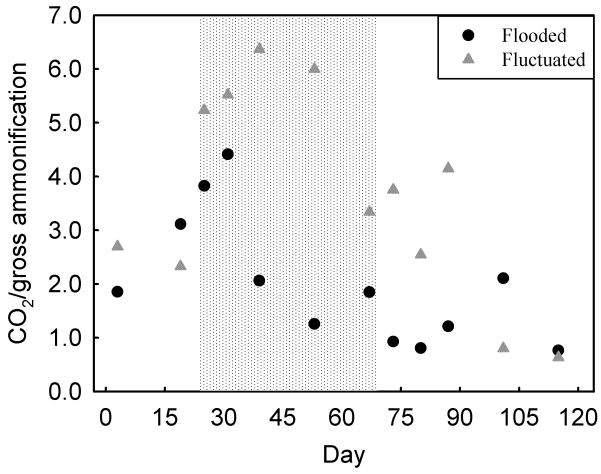

Fig. 7 Ratios of CO_2 emission/ gross ammonification (C/N ratio of mineralization in mol) in the permanently flooded (●) and the fluctuated (▲) treatments. The gray bar indicates the water table drawdown period in the fluctuated treatment.

3.5 Soil solution

NH_4^+ concentrations in soil solutions increased in both treatments after initial flooding from day 0 to 24 (Fig. 8). In the permanently flooded cores NH_4^+ concentrations increased further to 21 mg L^{-1} until day 67 and then remained constantly high.

After the water table drawdown, the concentrations of NH_4^+ decreased from 6.4 to 3.0 mg L^{-1} and increased from 3.0 to 8.8 mg L^{-1} when the soil was re-flooded. The NO_3^- concentrations were generally lower than 0.5 mg L^{-1} except from day 67 to 73 (1.2 and 3.2 mg L^{-1}), but these values had a huge spatial variation.

Concentrations of DON and DOC were higher in the permanently flooded cores than in the fluctuated cores already at the beginning. The concentrations of DOC and DON were very high, concentrations ranging from about 200 to 300 mg DOC L^{-1} and 7 to 12 mg DON L^{-1} (Fig. 9). The initial flooding further increased the concentrations in both treatments. After the water table drawdown, concentrations decreased strongly to a minimum of 19 mg L^{-1} DOC and 0.6 mg L^{-1} DON. After 45 days re-flooding (day 115), the concentrations of DOC and DON increased again to similar levels as in the permanently flooded cores.

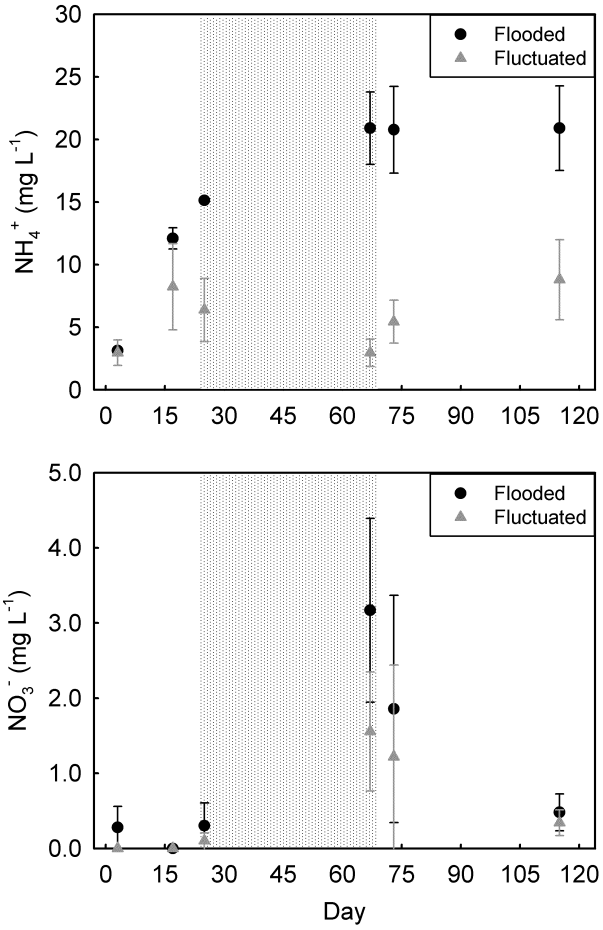

Fig. 8 NH_4^+ and NO_3^- concentrations (mean ± SE, n =3) in the permanently flooded (●) and the fluctuated (▲) treatments. The gray bar indicates the water table drawdown period in the fluctuated treatment.

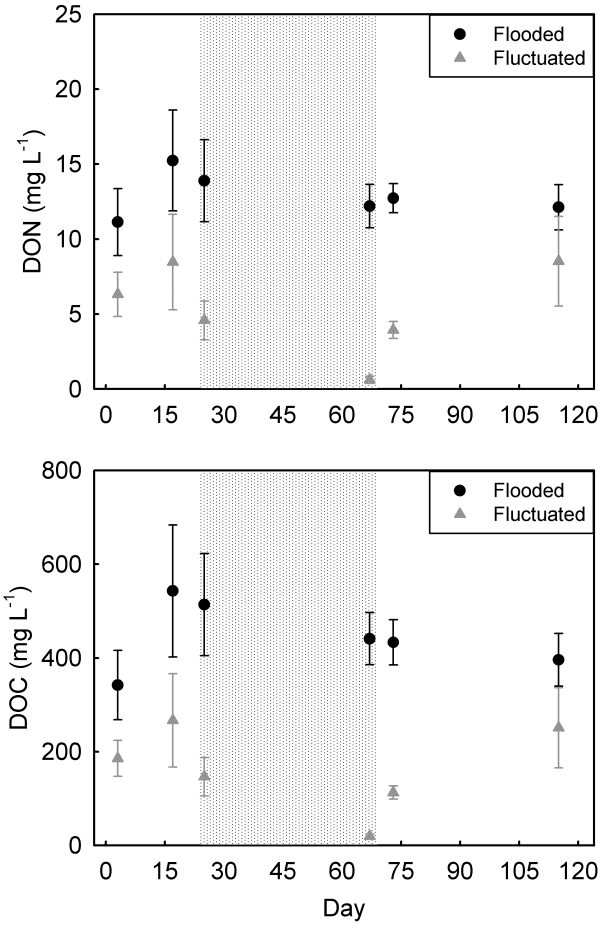

Fig. 9 DON and DOC concentrations (mean ± SE, n = 3) in the permanently flooded (●) and the fluctuated (▲) treatments. The gray bar indicates the water table drawdown period in the fluctuated treatment.

4. Discussion

4.1 Response of N turnover to water table fluctuations

Under permanently flooded and re-flooded conditions, gross ammonification increased after a lag phase of about 30 days. The response of gross ammonification to flooding was matched by the increasing NH_4^+ concentrations in soil solutions, indicating a similar response of net N mineralization to flooded conditions.

The response of gross N turnover to fluctuated water table in wetland soils has been barely studied and conflicting results were reported. In a slurry experiment, gross ammonification rates of fen soils under anaerobic incubations were only 25 - 30% of those observed in aerobic incubation (Ambus et al. 1992). In field studies, Wray and Bayley (2008) found higher gross ammonification rates in non-flooded than in flooded peatlands, however, Münchmeyer et al. (2000) observed similar gross ammonification rates in drained and re-flooded fen soils. Zak and Gelbrecht (2007) reported continuously increasing NH_4^+ concentrations during 60 weeks of re-flooding. An increase in NH_4^+ concentration was also found by Münchmeyer et al. (2000) in re-flooded fen soils.

In general, the increase of gross ammonification in the permanently flooded cores can be due to changes of substrate availability, substrate pools, changes in enzymatic and microbial activities (Corstanje and Reddy 2004; Kraigher et al. 2006; Mentzer et al. 2006). After water table drawdown and aeration, gross ammonification also increased to similar rates as in the permanently flooded cores. The increase was not instantaneous, but occurred after a lag phase of about 30 days, coinciding with the second peak of CO_2 emissions. The latter indicates an enhanced activity of aerobic microorganisms after aeration with a lag phase of about 30 days for physiological adaptation of microbial activity (Blodau et al. 2004). The decrease of the NH_4^+ concentrations in soil solutions also points to the enhanced immobilization by growing microbial biomass.

Rates of gross nitrification in our fen soil were very low, similar to those in ephemeral wetlands and fens (Ambus et al. 1992; Bedard-Haughn et al. 2006) and we did not find an effect of water table fluctuations. Gross nitrification in wetland soils has been rarely studied. Under flooded conditions, the lack of O_2 might be the most likely reason for the low rates (Bowden 1986; Bayley et al. 2005), but also low pH-values are known to reduce autotrophic nitrification. Net nitrification rates were often close to zero in peat soils (Neill 1995; Hefting

et al. 2004; Bayley et al. 2005; Wray and Bayley 2008), but an increase of net nitrification was observed after water table drawdown or drainage (Neill 1995; Münchmeyer et al. 2000). Yu and Ehrenfeld (2009) found that net nitrification increased within 2 weeks when the water content decreased from 100% to 30% of the water holding capacity. Under oxic conditions and after addition of 500 mg NH_4^+-N kg^{-1} soil to peat soils, nitrifiers oxidized NH_4^+-N completely after 8 days of incubation (Pal et al. 2010). Jones and Hood (1980) found that nitrifiers isolated from a wetland had a steep response to increasing NH_4^+ concentrations in the range of 0 - 500 mg NH_4^+ L^{-1} in pure cultures. Hence, the lack of response of nitrification after 42 days of water table drawdown in our study might be due to low NH_4^+ availability (0.8 - 11 mg NH_4^+ L^{-1} equal to 4 - 55 mg NH_4^+-N kg^{-1} soil).

4.2 Response of C mineralization and CH_4 concentrations to water table fluctuations

CO_2 emission was constantly low under the permanently flooded and O_2 limited conditions. Also when the aerated soil was re-flooded, a rapid decline of CO_2 flux within 1 day to the level of the permanently flooded cores was found, indicating quick response of aerobic microorganisms to the lack of O_2. A similar pattern of response was found by Freeman et al. (1993).

The emission of CO_2 had a first peak immediately after the water table drawdown which was probably due to the stimulating effect of aeration. Obviously a pool of easily degradable substrates was quickly mineralized causing a sharp increase in CO_2 emissions at an hourly to daily time scale. In the following 3 - 5 days, the easily degradable substrate depleted. This initial peak of CO_2 emissions contributed 390 mg C kg^{-1} soil in the first 5 days of water table drawdown while only 160 mg C kg^{-1} soil was emitted from the permanently flooded treatment. After a lag phase of about 30 days, a second peak of CO_2 emissions occurred in the fluctuated cores which contributed 1090 mg C kg^{-1} soil in a 20-day period, compared to 690 mg C kg^{-1} soil in the permanently flooded cores. The growth and activity of aerobic microorganisms (Blodau et al. 2004; Jaatinen et al. 2008; van Dijk et al. 2009) might be the reason for the second peak of CO_2 emissions. This assumption is supported by the increased gross ammonification and decreased NH_4^+ concentration. In summary, the initial peak of CO_2 obviously consumed easily available substrates and the second peak seems to be the

mineralization of more recalcitrant substrates.

An increase in CO_2 emissions after water table drawdown in wetland soils has often been reported. The water table drawdown enhanced C mineralization to CO_2 as the major product of microbial metabolism (Kechavarzi et al. 2007) and CO_2 emissions were often about doubled (Freeman et al. 1993; Silvola et al. 1996; Chimner and Cooper 2003), similar to our own findings. Even a 4.5 fold increase of CO_2 emissions after a water table drawdown was found (Blodau et al. 2004). In some cases, however, there was no response of CO_2 emissions to water table drawdown (Deppe et al. 2010; Muhr et al. 2011). Muhr et al. (2011) attributed the lack of a CO_2 response to water table drawdown to the generally low water table (-8 cm) of the active fen layer. A further decrease of the water table did not cause increased CO_2 emissions.

The initial strong peak of CO_2 emissions in our study after the rapid water table drawdown was not reported elsewhere, due to the experimental conditions and the time resolution of the measurements. In our experiment we changed the water table rapidly from flooded conditions (+5 cm) to -8 cm. This water table drawdown may be unrealistic under field conditions. The time resolution of other studies on CO_2 emissions after changes in water table is often weekly to monthly and the quick consumption of labile substrates after oxygenation might have been missed.

Even a small water table drawdown by 10 cm in fen soil is sufficient to promote the oxidation of reduced sulfur to SO_4^{2-} (Schiff et al. 2005). It is well known that SO_4^{2-} can suppress methanogenesis (Freeman et al. 1997; Dettling et al. 2006), and thus its presence might prevent the CH_4 production after the re-flooding (Blodau and Moore 2003). Hence, the enhanced SO_4^{2-} after water table drawdown in the fluctuated cores can explain the slow increase of CH_4 concentrations after the re-flooding.

4.3 C mineralization in relation to gross ammonification

The increase of gross ammonification was not accompanied by an increase of CO_2 emissions in the permanently flooded cores from day 0 to 80 and after re-flooding in the fluctuated cores. The ratios of CO_2 emissions to gross ammonification (C/N ratios of mineralization) were close to 2 under flooded conditions and increased to about 6 after water table drawdown. Relations of CO_2 emissions to gross ammonification have not been reported for peat soils so

far. In a long term laboratory incubation with an oxic forest soil, Hart et al. (1994) observed that C mineralization and gross ammonification were well correlated with a ratio of about 5, corroborating the ratios observed in our study after water table drawdown.

Several processes might explain the low C/N ratio of mineralization:

First, low rates of C mineralization are typically found under anoxic conditions in peat soils and C is partly reduced to CH_4 or metabolized to soluble (Pastor et al. 2003; Freeman et al. 2004; Fenner et al. 2005) or to insoluble metabolites. While the anoxic conditions reduce the oxidation of C, the hydrolytic cleavage of aminogroups may still continue.

Second, the mean residence times (MRT, calculated as pool/turnover) of N and C in the soil are very different: The total C and N content of our bulk soil are 311 and 18 g kg^{-1}, respectively. When taken the cumulative gross ammonification and CO_2 emission, the MRT of organic N is 2 years in the permanently flooded cores and 3 years in the fluctuated cores. The MRT of organic C is 33 years in the permanently flooded cores and 29 years in the fluctuated cores. The faster turnover of N than C cannot be explained by the mineralization of the bulk soil organic matter with a C/N of 17. Our calculations of the gross N turnover rates revealed that most of the NH_4^+ from gross ammonification is consumed by immobilization by the microbial biomass and that the net rates are much less than the gross rates (data not shown). This indicates a very fast turnover of N in the microbial biomass of our soil. In case of oxic forest soils, Hart et al. (1994) gave MRT of microbial biomass-N of 7 to 40 days in the initial phase of a long term incubation, while 43 days was reported by Davidson et al. (1992). Hence, the short MRT of microbial biomass-N is another explanation for the low C/N ratios of mineralization. The MRT of N in the microbial biomass seems to be even lower under flooded conditions since the C/N ratio of mineralization decreased to about 1.

4.4 Response of DON and DOC to water table fluctuations

Because we determined only concentrations, we are unable to assess whether concentration changes of DON and DOC result specifically from changes in production or consumption. The overall range of DON and DOC concentrations in our experimental setup were very high with maxima of around 15 mg DON L^{-1} and 500 mg DOC L^{-1}. Under field conditions, concentrations in our fen soil at 10 cm depth are much less (maximum around 2.0 mg DON L^{-1} and 60 mg DOC L^{-1} at 10 cm depth, unpublished data). The reasons for the high

concentrations of DON and DOC in the laboratory might be the higher temperatures in our laboratory study (under field conditions soil temperature is around 10°C in the growing season). Furthermore, the field site is subjected to permanent lateral water flow removing huge amounts of dissolved organic matter, which was not the case in the laboratory experiment.

The initial flooding increased the DON and DOC concentrations in both sets of cores. Concentrations remained on a high level in the permanently flooded cores, while the water table drawdown induced a strong decrease of the concentrations. After re-flooding, concentrations increased again. Increased concentrations of DOC in peat soils subjected to flooding or re-flooding were observed in other studies (Chow et al. 2006; Zak and Gelbrecht 2007), the reasons remaining speculative. During the flooding, C is less likely to be completely metabolized to CO_2 and instead, dissolved organic compounds may preferentially be formed as end products as suggested by Pastor et al. (2003), Freeman et al. (2004) and Fenner et al. (2005).

After 42 days of the water table drawdown (day 67), the DOC concentrations decreased to the minimum values in our study. Some studies report similar findings (Scott et al. 1998; Clark et al. 2005; Fenner et al. 2005) while others report increasing concentrations (Driscoll et al. 1989; Tipping et al. 1999) or no difference (Blodau et al. 2004; Freeman et al. 2004) after water table drawdown.

One reason for decreasing concentrations of DOC might be the aerobic conditions associated with the water table drawdown, favoring CO_2 rather than DOC as the major end product of decomposition. In our study, the average DOC pool in the soil decreased by 590 mg kg^{-1} soil. In the same time period the cumulated CO_2 emissions were 2100 mg kg^{-1} soil. This indicates that the decline of the DOC pool after water table drawdown can be explained by mineralization of DOC. A second reason for the decline in DOC after water table drawdown might be the decreased solubility of C compounds due to increased H^+ concentration and ionic strength associated with SO_4^{2-} production (Driscoll et al. 1989; Hruška et al. 2009). Clark et al. (2005) suggested that a rise of 0.5 pH units would result in a 60% increase in DOC. Changes in pH were not observed in our study and correlations between SO_4^{2-} /pH/conductivity and DOC were generally poor (data not shown), however, SO_4^{2-} reached highest concentration after 42 days of water table drawdown (day 67) coinciding with the lowest DOC concentrations.

The concentrations of DON were generally in the same range as those of NH_4^+, emphasizing the role of DON for solute N transport in fen soils. The response of DON to water table fluctuations was similar to DOC as indicated by the almost constant DOC/DON ratios.

5. Conclusion

Our results showed that the response of fen soils to rapid water table fluctuation is element specific and characterized by different time scales: The permanent flooding enhanced gross ammonification after a lag phase of about 30 days. The water table drawdown decreased concentrations of DON and DOC but also increased gross ammonification, the latter after a lag phase of about 30 days. Contrary to N, the emission of CO_2 peaked immediately after water table drawdown, followed by a decrease and a second peak after 30 days of water table drawdown. It seems that about 30 days are required for the microorganisms to adapt to the changes of oxic/anoxic conditions. The ratios of CO_2 emission/gross ammonification were very low under anoxic conditions which seem to be caused by the fast N turnover in the microbial biomass pool and low rates of CO_2 production. The observed changes of the N and C turnover induced by water table drawdown were reversible after re-flooding within a one month period.

Short term (few days) water table drawdown will have different effects on N and C turnover as compared to drawdown on longer time scales (> 1 month).

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References

Ambus P, Mosier A, Christensen S (1992) Nitrogen turnover rates in a riparian fen determined by ^{15}N dilution. Biology and Fertility of Soils 14: 230-236.

Bayley SE, Thormann MN, Szumigalski AR (2005) Nitrogen mineralization and decomposition in western boreal bog and fen peat. Ecoscience 12: 455-465.

Bedard-Haughn A, Matson AL, Pennock DJ (2006) Land use effects on gross nitrogen mineralization, nitrification, and N_2O emissions in ephemeral wetlands. Soil Biology and Biochemistry 38: 3398-3406.

Blodau C, Moore TR (2003) Experimental response of peatland carbon dynamics to a water table fluctuation. Aquatic Sciences 65: 47-62.

Blodau C, Basiliko N, Moore TR (2004) Carbon turnover in peatland mesocosms exposed to different water table levels. Biogeochemistry 67: 331-351.

Bowden WB (1986) Nitrification, nitrate reduction, and nitrogen immobilization in a tidal fresh-water marsh sediment. Ecology 67: 88-99.

Bowden WB (1987) The biogeochemistry of nitrogen in fresh-water wetlands. Biogeochemistry 4: 313-348.

Chimner RA, Cooper DJ (2003) Influence of water table levels on CO_2 emissions in a Colorado subalpine fen: an in situ microcosm study. Soil Biology and Biochemistry 35: 345-351.

Chow AT, Tanji KK, Gao SD, Dahlgren RA (2006) Temperature, water content and wet-dry cycle effects on DOC production and carbon mineralization in agricultural peat soils. Soil Biology and Biochemistry 38: 477-488.

Clark JM, Chapman PJ, Adamson JK, Lane SN (2005) Influence of drought-induced acidification on the mobility of dissolved organic carbon in peat soils. Global Change Biology 11: 791-809.

Corstanje R, Reddy KR (2004) Response of biogeochemical indicators to a drawdown and subsequent reflood. Journal of Environmental Quality 33: 2357-2366.

Danevčič T, Mandic-Mulec I, Stres B, Stopar D, Hacin J (2010) Emissions of CO_2, CH_4 and N_2O from Southern European peatlands. Soil Biology and Biochemistry 42: 1437-1446.

Davidson EA, Hart SC, Firestone MK (1992) Internal cycling of nitrate in soils of a mature coniferous forest. Ecology 73: 1148-1156.

Deppe M, Knorr KH, McKnight DM, Blodau C (2010) Effects of short-term drying and irrigation on CO_2 and CH_4 production and emission from mesocosms of a northern bog and an alpine fen. Biogeochemistry 100: 89-103.

Dettling MD, Yavitt JB, Zinder SH (2006) Control of organic carbon mineralization by alternative electron acceptors in four peatlands, central New York State, USA. Wetlands 26: 917-927.

Dinsmore KJ, Skiba UM, Billett MF, Rees RM (2009) Effect of water table on greenhouse gas emissions from peatland mesocosms. Plant and Soil 318: 229-242.

Driscoll CT, Fuller RD, Schecher WD (1989) The role of organic acids in the acidification of surface waters in the Eastern U.S. Water, Air, and Soil Pollution 43: 21-40.

Fenner N, Freeman C, Reynolds B (2005) Hydrological effects on the diversity of phenolic degrading bacteria in a peatland: implications for carbon cycling. Soil Biology and Biochemistry 37: 1277-1287.

Freeman C, Lock MA, Reynolds B (1993) Fluxes of CO_2, CH_4 and N_2O from a Welsh peatland following simulation of water table draw-down: Potential feedback to climatic change. Biogeochemistry 19: 51-60.

Freeman C, Liska G, Ostle NJ, Lock MA, Hughes S, Reynolds B, Hudson J (1997) Enzymes and biogeochemical cycling in wetlands during a simulated drought. Biogeochemistry 39: 177-187.

Freeman C, Fenner N, Ostle NJ, Kang H, Dowrick DJ, Reynolds B, Lock MA, Sleep D, Hughes S, Hudson J (2004) Export of dissolved organic carbon from peatlands under elevated carbon dioxide levels. Nature 430: 195-198.

Goldberg SD, Knorr KH, Gebauer G (2008) N_2O concentration and isotope signature along profiles provide deeper insight into the fate of N_2O in soils. Isotopes in Environmental and Health Studies 44: 377-391.

Hart SC, Nason GE, Myrold DD, Perry DA (1994) Dynamics of gross nitrogen transformations in an old-growth forest - The carbon connection. Ecology 75: 880-891.

Hefting M, Clément JC, Dowrick D, Cosandey AC, Bernal S, Cimpian C, Tatur A, Burt TP, Pinay G (2004) Water table elevation controls on soil nitrogen cycling in riparian wetlands along a European climatic gradient. Biogeochemistry 67: 113-134.

Hruška J, Krám P, McDowell WH, Oulehle F (2009) Increased dissolved organic carbon (DOC) in central European streams is driven by reductions in ionic strength rather than climate change or decreasing acidity. Environmental Science and Technology 43: 4320-4326.

IPCC (2007) Climate Change 2007: Synthesis report. Contribution of Working Groups I, II and III to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. IPCC, Geneva, Switzerland.

Jaatinen K, Laiho R, Vuorenmaa A, del Castillo U, Minkkinen K, Pennanen T, Penttilä T, Fritze H (2008) Responses of aerobic microbial communities and soil respiration to water-level drawdown in a northern boreal fen. Environmental Microbiology 10: 339-353.

Jones RD, Hood MA (1980) Effects of temperature, pH, salinity, and inorganic nitrogen on the rate of ammonium oxidation by nitrifiers isolated from wetland environments. Microbial Ecology 6: 339-347.

Kechavarzi C, Dawson Q, Leeds-Harrison PB, Szatyłowicz J, Gnatowski T (2007) Water-table management in lowland UK peat soils and its potential impact on CO_2 emission. Soil Use and Management 23: 359-367.

Keller JK, White JR, Bridgham SD, Pastor J (2004) Climate change effects on carbon and nitrogen mineralization in peatlands through changes in soil quality. Global Change Biology 10: 1053-1064.

Kieckbusch JJ, Schrautzer J (2007). Nitrogen and phosphorus dynamics of a re-wetted shallow-flooded peatland. Science of the Total Environment 380: 3-12.

Kirkham D, Bartholomew WV (1954) Equations for following nutrient transformations in soil, utilizing tracer data. Soil Science Society of America Proceedings 18: 33-34.

Knorr KH, Oosterwoud MR, Blodau C (2008a) Experimental drought alters rates of soil respiration and methanogenesis but not carbon exchange in soil of a temperate fen. Soil Biology and Biochemistry 40: 1781-1791.

Knorr KH, Glaser B, Blodau C (2008b) Fluxes and ^{13}C isotopic composition of dissolved

carbon and pathways of methanogenesis in a fen soil exposed to experimental drought. Biogeosciences 5: 1457-1473.

Knorr KH, Lischeid G, Blodau C (2009) Dynamics of redox processes in a minerotrophic fen exposed to a water table manipulation. Geoderma 153: 379-392.

Kraigher B, Stres B, Hacin J, Ausec L, Mahne I, van Elsas JD, Mandic-Mulec I (2006) Microbial activity and community structure in two drained fen soils in the Ljubljana Marsh. Soil Biology and Biochemistry 38: 2762-2771.

Luxhøi J, Nielsen NE, Jensen LS (2004) Effect of soil heterogeneity on gross nitrogen mineralization measured by ^{15}N-pool dilution techniques. Plant and Soil 262: 263-275.

Mentzer J, Goodman R, Balser T (2006) Microbial response over time to hydrologic and fertilization treatments in a simulated wet prairie. Plant and Soil 284: 85-100.

Mitsch WJ, Day JW, Gilliam JW, Groffman PM, Hey DL, Randall GW, Wang NM (2001) Reducing nitrogen loading to the Gulf of Mexico from the Mississippi River Basin: Strategies to counter a persistent ecological problem. Bioscience 51: 373-388.

Monaghan R (1995) Errors in estimates of gross rates of nitrogen mineralization due to non-uniform distributions of ^{15}N label. Soil Biology and Biochemistry 27: 855-859.

Muhr J, Goldberg SD, Borken W, Gebauer G (2008) Repeated drying-rewetting cycles and their effects on the emission of CO_2, N_2O, NO, and CH_4 in a forest soil. Journal of Plant Nutrition and Soil Science 171: 719-728.

Muhr J, Höhle J, Otieno DO, Borken W (2011) Manipulative lowering of the water table during summer does not affect CO_2 emissions and uptake in a minerotrophic fen in South-Eastern Germany. Ecological Applications 21: 391-401.

Münchmeyer U, Russow R, Augustin J (2000) Net and gross nitrogen mineralization in drained and reflooded fen soils. Isotopes in Environmental and Health Studies 36: 79-98.

Neill C (1995) Seasonal flooding, nitrogen mineralization and nitrogen utilization in a prairie marsh. Biogeochemistry 30: 171-189.

Oechel WC, Vourlitis GL, Hastings SJ, Aul, RP, Bryant P (1998) The effects of water table manipulation and elevated temperature on the net CO_2 flux of wet sedge tundra ecosystems. Global Change Biology 4: 77-90.

Pal L, Stres B, Danevčič T, Leskovec S, Mandic-Mulec I (2010) Transformations of mineral nitrogen applied to peat soil during sequential oxic/anoxic cycling. Soil Biology and Biochemistry 42: 1338-1346.

Pastor J, Solin J, Bridgham SD, Updegraff K, Harth C, Weishampel P, Dewey B (2003) Global warming and the export of dissolved organic carbon from boreal peatlands. Oikos 100: 380-386.

Paul S, Küsel K, Alewell C (2006) Reduction processes in forest wetlands: Tracking down heterogeneity of source/sink functions with a combination of methods. Soil Biology and Biochemistry 38: 1028-1039.

Pinay G, Clément JC, Naiman RJ (2002) Basic principles and ecological consequences of changing water regimes on nitrogen cycling in fluvial systems. Environmental Management 30: 481-491.

Schiff SL, Spoelstra J, Semkin RG, Jeffries DS (2005) Drought induced pulses of SO_4^{2-} from a Canadian shield wetland: use of $\delta^{34}S$ and $\delta^{18}O$ in SO_4^{2-} to determine sources of sulfur. Applied Geochemistry 20: 691-700.

Scott JT, McCarthy MJ, Gardner WS, Doyle RD (2008) Denitrification, dissimilatory nitrate reduction to ammonium, and nitrogen fixation along a nitrate concentration gradient in a created freshwater wetland. Biogeochemistry 87: 99-111.

Scott MJ, Jones MN, Woof C, Tipping E (1998) Concentrations and fluxes of dissolved organic carbon in drainage water from an upland peat system. Environment International 24: 537-546.

Silvola J, Alm J, Ahlholm U, Nykänen H, Martikainen PJ (1996) CO_2 fluxes from peat in boreal mires under varying temperature and moisture conditions. Journal of Ecology 84: 219-228.

Stange CF, Spott O, Apelt B, Russow RWB (2007) Automated and rapid online determination of ^{15}N abundance and concentration of ammonium, nitrite, or nitrate in aqueous samples by the SPINMAS technique. Isotopes in Environmental and Health Studies 43: 227-236.

Tipping E, Woof C, Rigg E, Harrison AF, Ineson P, Taylor K, Benham D, Poskitt J, Rowland AP, Bol R, Harkness DD (1999) Climatic influences on the leaching of dissolved organic matter from upland UK moorland soils, investigated by a field manipulation experiment.

Environment International 25: 83-95.

van Dijk J, Didden WAM, Kuenen F, van Bodegom PM, Verhoef HA, Aerts R (2009) Can differences in soil community composition after peat meadow restoration lead to different decomposition and mineralization rates? Soil Biology and Biochemistry 41: 1717-1725.

Venterink HO, Davidsson TE, Kiehl K, Leonardson L (2002) Impact of drying and re-wetting on N, P and K dynamics in a wetland soil. Plant and Soil 243: 119-130.

Wray HE, Bayley SE (2008) Nitrogen dynamics in floating and non-floating peatlands in the Western Boreal Plain. Canadian Journal of Soil Science 88: 697-708.

Yu S, Ehrenfeld JG (2009) The effects of changes in soil moisture on nitrogen cycling in acid wetland types of the New Jersey Pinelands (USA). Soil Biology and Biochemistry 41: 2394-2405.

Zak D, Gelbrecht J (2007) The mobilisation of phosphorus, organic carbon and ammonium in the initial stage of fen rewetting (a case study from NE Germany). Biogeochemistry 85: 141-151.

Appendix

Own contributions of the candidate

Study 1 - Effects of decreasing water potential on gross ammonification and nitrification in an acid coniferous forest soil

Chen, Yao-Te, Borken, Werner, Stange, Claus Florian Stange and Matzner, Egbert

Published in Soil Biology and Biochemistry (2011) 43, 333-338.

Y.-T. Chen:	50%	concepts, laboratory and field works, data analysis, manuscript preparation
W. Borken:	15%	concepts, discussion of results, manuscript preparation
C. F. Stange:	10%	discussion of results
E. Matzner:	25%	interpretation and discussion of results, manuscript preparation

Study 2 - Minor response of gross N turnover and N leaching to drying, rewetting and irrigation in the top soil of a Norway spruce forest

Chen, Yao-Te, Christina Bogner, Borken, Werner, Stange, Claus Florian and Matzner, Egbert

European Journal of Soil Science, revision submitted in June.

Y.-T. Chen:	40 %	concepts, laboratory and field works, data analysis, manuscript preparation
C. Bogner	15%	statistical modeling, discussion of results, manuscript preparation
W. Borken:	10 %	concepts, discussion of results, manuscript preparation
C. F. Stange:	5 %	discussion of results
E. Matzner:	30%	interpretation and discussion of results, manuscript preparation

Study 3 - Dynamics of nitrogen and carbon mineralization in a fen soil following water table fluctuations

Yao-Te, Borken, Werner, Stange, Claus Florian and Matzner, Egbert

Submitted to Wetlands in June.

Y.-T. Chen:	50%	concepts, laboratory and field works, data analysis, manuscript preparation
W. Borken:	15%	concepts, discussion of results, manuscript preparation
C. F. Stange:	5%	discussion of results
E. Matzner:	30%	interpretation and discussion of results, manuscript preparation

Publications

Chen, Y.-T., Borken, W., Stange, C.F. and Matzner, E. 2011. Effects of decreasing water potential on gross ammonification and nitrification in an acid coniferous forest soil. Soil Biology and Biochemistry 43, 333-338

Chen, Y.-T., Bogner C., Borken, W., Stange, C.F. and Matzner, E. 2011. Minor response of gross N turnover and N leaching to drying, rewetting and irrigation in the top soil of a Norway spruce forest. European Journal of Soil Science, revision submitted in June.

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Declaration/Erklärung

I hereby declare that this PhD thesis is entirely my own work, and that I did not use any other sources or auxiliary means other than those referenced. Moreover, I declare that no parts of this thesis have previously been submitted for the purpose of obtaining a PhD degree at any other scientific institution, and I have not yet conclusively failed any doctoral examination procedure.

Hiermit erkläre ich, dass ich die vorliegende Arbeit selbständig verfasst und keine anderen als die angegeben Quellen und Hilfsmittel benutz habe. Ich versichere weiterhin, dass ich diese Arbeit an keiner anderen wissenschaftlichen Einrichtung zum Zwecke der Promotion eingereicht habe, sowie, dass ich noch kein Promotionsverfahren endgültig nicht bestanden habe.

Bayreuth, den

01.01.2012

Yao-Te Chen